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PATHOGENIC CHARACTERISTICS OF *FUSARIUM SOLANI* ISOLATED FROM INLAND PACIFIC NORTHWEST FOREST NURSERIES

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ABSTRACT

Forty-seven isolates of *Fusarium solani* obtained from the roots of diseased and healthy conifer seedlings and forest nursery soil were tested for pathogenicity on young Douglas-fir germinants under controlled laboratory conditions. Isolate virulence varied widely; a few were highly virulent whereas many were classified as non-pathogenic. Isolates from the roots of conifer seedlings were generally more virulent than soil isolates; no significant differences in virulence between isolates obtained from diseased roots and healthy roots were found. We concluded that although some *F. solani* isolates commonly inhabiting forest nurseries have the potential to elicit disease, most are probably only slightly virulent or non-pathogenic.

INTRODUCTION

Fusarium solani (Mart.) Appel & Wollenw. (teleomorph: *Nectria haematocca* Berk. & Br.) is an important plant pathogenic fungal species that attacks a wide range of plants (Nelson et al. 1983). It is primarily soil borne and naturally within a wide range of agricultural and grassland soils (Joffe and Palti 1977, Kommendahl et al. 1975; Summerell et al. 1993). *Fusarium solani* is most frequently isolated from warmer (Burgess 1981; Hancock 1985; Joffe and Palti 1977) and wetter (Summerell et al. 1993) soils; it survives for long periods in soil as chlamydospores (Alexander et al. 1966; Bourret et al. 1968; El-Ani 1988). Forest nursery soils typically contain populations of this potentially important fungus (James and Beall 1999, 2000; James and Perez 1999a; James et al. 1990, 1996).

Fusarium solani usually induces cortical root decay following infection of susceptible plants (Price 1984). This fungus causes important economic losses to a wide range of agricultural plants, particularly peas (Chakraborty and Charkraborty 1989; Cook and Flentje 1965), several types of beans (Adams et al. 1968; Baker and Nash 1965; Reddy et al. 1994; Schuerger and Mitchell 1992), alfalfa (Elliott et al. 1969; Graham et al. 1979), sweet potatoes (Duarte and Clark 1993), squash (Samac and Leong 1989), cucumbers (Snyder et al. 1975), red clover (Smith and Peterson 1966), and different types of cereals (Francis and Burgess 1975; Gordon and Sprague 1941; Sturz and Bernier 1991; Warren and Kommedahl 1973).

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Fusarium solani has periodically been associated with forest tree seedling diseases in nurseries. For example, it can cause damping-off on a wide range of conifer seedlings (Hartley et al. 1918; James et al. 1989; Vaartaja and Cram 1956), the most important being Scots pine (Kulakova 1984) and red pine (Pomerleau 1934). The fungus has routinely been isolated from root diseased bareroot (James 1983a; James et al. 1989) and container-grown (James 1989, 1990; James and Gilligan 1985; James et al. 1989) conifer seedlings. It has also been associated with decline and mortality of oak seedlings in forest nurseries (Ragazzi et al. 1993) and may even cause stains in maple trees (Kabir 1965). *Fusarium solani* is routinely isolated from the roots of healthy-appearing conifer seedlings in nurseries (James and Beall 1999; James and Gilligan 1988; James et al. 1989, 1996). In addition to its common occurrence within forest nursery soils, *F. solani* is often isolated from conifer seeds (James 1987; James et al. 1989). Examples include Douglas-fir (James 1983b, 1984), ponderosa pine (James and Genz 1982), and different species of spruce (*Picea* spp.)(James 1985). Seed-borne inoculum may be an important way potentially pathogenic isolates of *F. solani* are introduced into both container and bareroot growing areas (James 1985, 1986, 1987).

The role of *F. solani* as an important pathogen on forest nursery seedlings remains questionable. Although often isolated from nursery soil and diseased seedlings, this fungus may not necessarily be pathogenic under all environmental conditions. However, its reputation as an important pathogen on numerous agricultural crops has been firmly established (Nelson et al. 1983; Price 1984). Therefore, as a step toward elucidating the importance of *F. solani* as a forest nursery pathogen, investigations were conducted to determine its ability to elicit disease on conifer seedlings under controlled laboratory conditions.

MATERIALS AND METHODS

Isolates of *F. solani* were obtained during investigations of diseased and apparently healthy conifer seedlings at several forest nurseries in the inland Pacific Northwest. Many isolates were also obtained from routine analyses of populations in forest nursery soil. In general, isolates were obtained from either roots or soil dilutions on a selective agar medium for *Fusarium* and closely related fungi (Komada 1975). Isolates were typically transferred from the selective medium, single-spored, and grown on carnation leaf agar (Fisher et al. 1982) and potato dextrose agar (PDA) to facilitate their identification using the taxonomy of Nelson et al. (1983). *Fusarium* isolates were stored for long periods as spore suspensions either in sterile soil or on carnation leaf substrates within sterile water. Under these conditions, isolates did not mutate and remained viable for at least 10 years.

A total of 12 isolates from roots of conifer seedlings (six each from diseased and healthy seedlings) and 35 from forest nursery soil (total of 47 isolates) were tested for pathogenicity. Pathogenicity tests were conducted using procedures outlined in James (1996). The basic approach was to expose young Douglas-fir (*Pseudotsuga menziesii* var. *glauca* [Beissn.] Franco) germinants to isolates of *F. solani* and record production of disease symptoms. Cornmeal-perlite inoculum was prepared using the techniques of Miles and Wilcoxin (1984). Perlite, an inert, inorganic, siliceous rock of volcanic origin commonly used in potting mixtures, was the matrix for fungal growth. 150 g of yellow cornmeal was moistened with 300 ml warm 1 percent PDA, to which 75 g of perlite were added. The mixture was placed into glass vials (23 ml capacity) to about two-thirds capacity, which were then autoclaved for 60 minutes at 121°C. After cooling, vials were inoculated with about 10 ml spore suspension of the test isolate (produced by adding sterile, distilled water to 14-day-old cultures grown on PDA). Vial caps were left loose to allow aeration. Vials were incubated in the dark for at least 21 days, after which the fungal isolates had thoroughly colonized the perlite/cornmeal mixture. After incubation, inoculum was removed from vials and dried in open petri plates within a cabinet. Inoculum dried within 5-7 days and was not contaminated with other organisms because the food base was completely colonized by the inoculated test isolate. Once dry, inoculum was stored in plastic vials and refrigerated until needed.

Twenty-four vials (23 ml capacity each) were used to test each fungal isolate. Each vial was filled to about two-thirds capacity (about 2.5 g) with dried coconut-vermiculite media (Grace/Sierra Horticultural Products, Milpitas, CA) and autoclaved at 121°C for 60 minutes. Vial lids were placed loosely on vials before sterilization.

One Douglas-fir seedlot with high germination capacity (designated Flat Creek 78-10, Moscow Mountain – courtesy of the University of Idaho Forest Research Nursery) was used throughout pathogenicity tests. Seeds were soaked in a 2-part bleach and 3-part water solution for 10 minutes (Wenny and Dumroese 1987), rinsed 48 hours in running tap water, and stratified 21 days at 2-3°C. After stratification, seeds were placed on filter paper moistened with sterile water, in petri plates. Seeds were incubated under 12-hour diurnal fluorescent light cycles at about 24°C and monitored daily for germination. Seeds were considered germinated when their primary root was at least 3 mm long.

For each tested isolate, perlite/cornmeal inoculum was ground to a fine powder with mortar and pestle and 0.05 g of the powder was added to each vial containing dried media. This resulted in an approximate 1:50 w/w mixture of inoculum to media. Inoculum was distributed throughout the media by shaking. One recently germinated seed (germinant) was placed in each vial with its radicle placed downward into the media. Four ml of sterile water was added to each vial with caps replaced loosely to allow aeration. Adding water activated inoculum (Miles and Wilcoxin 1984). At least 20 of the 24 vials must have had germinants with normally extending radicles for a valid test (occasionally, a germinant's radicle would abort or decay because of infection by seed-borne microorganisms). One set of 24 control vials was included; each vial contained non-inoculated perlite instead of fungal inoculum.

Vials containing germinants were incubated at about 24°C on a lab bench, providing fluorescent light for 8-12 hours daily. Each test ran for 14 days. After 3 days, germinants were first checked for disease. Germinants were then checked for disease daily throughout the 14-day test period. Diseases were categorized as either standard post-emergence damping-off with fungal growth appearing just above the ground line or root decay in which fungal growth at or above the ground line was lacking, but the radicle was decayed while growing within the media. When germinates appeared diseased, they were carefully removed with forceps, their stems and root washed thoroughly, and placed on Komada's medium for re-isolation of inoculated isolates. After 14 days, surviving germinants were examined to determine if their roots were diseased (decayed or with necrotic lesions) and had grown to the bottom of the vial. Roots were then washed and plated on Komada's medium for re-isolation of inoculated isolates.

A numerical rating system for isolate comparisons was used. This system awarded points based on duration of germinant survival within inoculated vials, occurrence and type of disease, re-isolation of inoculated fungal isolates, and root growth within the vial. The range of possible points was 3-23, with higher point values reflecting less aggression by the tested isolates. To convert points to a score in which higher numbers represented greater virulence, a reciprocal rating was devised. In this system, the maximum score (germinants killed within 3 days by the test isolate) was 100 and the minimum score (indicating germinants were unaffected by the test isolate within 14 days) was zero. Based on previous experience (James 1996; James and Perez 1999b; James et al. 1995, 1997), highly virulent isolates exhibited scores of 80-100, moderately virulent isolates from 60-80, isolates with low virulence from 40-60, and isolates with average scores below 40 were considered non-pathogenic.

The average rating and average number of days' survival (without noticeable disease) were used to compare isolates. These averages were compared using one-way analysis of variance; comparisons were made among isolate sources (soil vs. healthy roots vs. diseased roots). Significant means (P=0.05) were separated using Tukey's HSD test.

RESULTS AND DISCUSSION

Fusarium solani isolates obtained from the roots of conifer seedlings were generally more virulent on Douglas-fir germinants in our tests than isolates from forest nursery soil (Table 1). However, isolates from roots of diseased and apparently healthy seedlings did not significantly differ in their virulence on Douglas-fir. There were wide ranges of isolate virulence from all three locations (diseased roots, healthy roots, and nursery soil). Isolates classified as highly virulent (above the average score of 80) were obtained from roots of both healthy and diseased seedlings. However, no highly virulent isolates were obtained from nursery soil (individual virulence ratings of tested isolates are summarized in Tables 3, 4, and 5 in the Appendix). Many of the soil isolates were classified as non-virulent in our tests.

Another important measure of aggressiveness of tested isolates in our pathogenicity tests was the number of days' survival of inoculated Douglas-fir germinants. Generally, the longer the germinants survived exposure to the test fungus, the less virulent the fungus. Average germinant survival when exposed to *F. solani* isolates from roots of diseased and healthy seedlings and nursery soil is summarized in Table 2 (individual survival averages for all tested isolates are summarized in Tables 6, 7, and 8 in the Appendix). No significant differences in average length of germinant survival were found among isolates from seedling roots or nursery soil.

These results indicated that although a few *F. solani* isolates encountered during investigations of forest nursery diseases were capable of eliciting diseases on Douglas-fir germinants, most displayed either low virulence or were non-pathogenic, at least under the conditions of our tests. Previous reports (Hartley et al. 1918; Reddy et al. 1994) have verified wide-ranging pathogenic variability of this fungal species on different host plants. In some cases, *F. solani* may comprise a relatively significant proportion of the *Fusarium* isolates obtained from nursery soil (James and Beall 1999, 2000; James et al. 1990, 1996). Populations tend to decrease naturally in soil over time in the absence of susceptible host plants (Hall and Phillips 1992) or sufficient organic matter (Parkinson et al. 1989; Snyder et al. 1959). Based on our pathogenicity test results, we would expect that most of these isolates provide little threat to conifer seedlings being grown in infested soil.

This is in contrast with *F. oxysporum* Schlecht., which is much more common within forest nursery soil and on the roots of bareroot conifer seedlings (Bloomberg 1976; James et al. 1991). Although the taxon *F. oxysporum* comprises both pathogenic and saprophytic isolates (Gordon and Martyn 1977; Gordon and Okamoto 1992), we suspect that a greater proportions of those from soil are pathogenic than we found for *F. solani* (James et al. 2000). *Fusarium oxysporum* isolates from roots of diseased seedlings are generally more virulent than those from healthy seedling roots (James et al. 2000), although large variations in disease-causing abilities may occur within soil-borne populations. In any event, we would expect much more disease-causing potential from nursery populations of *F. oxysporum* than from populations of *F. solani*.

Table 1. Comparisons of average virulence scores of *Fusarium solani* isolates obtained from roots of healthy and diseased conifer seedlings and soil at nurseries in the inland Pacific Northwest.

Isolate Source	No. Isolates Tested	Virulence Rating	
		Average[1]	Range
Nursery Soil	35	36.2 B	20.0-76.1
Healthy Roots	6	46.3 A	26.0-85.0
Diseased Roots	6	47.4 A	25.5-97.1
All Isolates	47	39.0	20.0-97.1
Control	-	19.8	19.8

[1] Within this column, means followed by the same capital letter are not significantly different (P=0.05) using Tukey's HSD.

Table 2. Comparisons of average number of days' survival of young Douglas-fir germinants inoculated with *Fusarium solani* isolates obtained from roots of healthy and diseased conifer seedlings and soil at nurseries in the inland Pacific Northwest.

Isolate Source	Number of Isolates Tested	Average Survival[1]
Nursery Soil	35	12.8 A
Healthy Roots	6	11.0 A
Diseased Roots	6	10.7 A
All Isolates	47	12.3
Control	-	14.0

[1] Maximum survival of 14 days (length of pathogenicity tests); within this column, means followed by the same capital letter are not significantly different (P=0.05) using Tukey's HSD.

LITERATURE CITED

Adams, P.B., J.A. Lewis and G.C. Papvizas. 1968. Survival of root-infecting fungi in soil. IX. Mechanisms of control of Fusarium root rot of bean with spent coffee grounds. Phytopathology 58:1603-1608.

Alexander, J.V., J.A. Bourret, A.H. Gold and W.C. Snyder. 1966. Induction of chlamydospore formation by *Fusarium solani* in sterile soil extracts. Phytopathology 56:353-354.

Baker, R. and S.M. Nash. 1965. Ecology of plant pathogens in soil. VI. Inoculum density of *Fusarium solani* f. sp. *phaseoli* in bean rhizosphere as affected by cellulose and supplemental nitrogen. Phytopathology 55:1381-1382.

Bloomberg, W.J. 1976. Distribution and pathogenicity of *Fusarium oxysporum* in a forest nursery soil. Phytopathology 66:1090-1092.

Bourret, J.A., A.H. Gold and W.C. Snyder. 1968. Effect of carbon dioxide on germination of chlamydospores of *Fusarium solani* f. sp. *phaseoli*. Phytopathology 58:710-711.

Burgess, L.W. 1981. General ecology of the Fusaria. *In*: Nelson, P.E., T.A. Toussoun and R.J. Cook (eds.). *Fusarium*: Diseases, Biology & Taxonomy. The Pennsylvania State University Press, University Park. pp. 225-235.

Chakraborty, U. and B.N. Chakraborty. 1989. Interaction of *Rhizobium leguminosarum* and *Fusarium solani* f. sp. *pisi* on pea affecting disease development and phytoalexin production. Canadian Journal of Botany 67:1698-1701.

Cook, R.J. land N.T. Flentje. 1965. Influence of soil water on growth and survival of *Fusarium solani* f. sp. *pisi* in the rhizosphere of pea. Phytopathology 55:1051-1085.

Duarte, V. and C.A. Clark. 1993. Interaction of *Erwinia chrysanthemi* and *Fusarium solani* on sweet potato. Plant Disease 77:733-735.

El-Ani, A.S. 1988. The chlamydospore in *Fusarium solani*. Mycologia 80:885-887.

Elliott, E.S., R.E. Baldwin and R.B. Carroll. 1969. Root rots of alfalfa and red clover. West Virginia Agricultural Experiment Station Bulletin 585T. 32p.

Fisher, N.L., L.W. Burgess, T.A. Toussoun and P.E. Nelson. 1982. Carnation leaves as a substrate and for preserving cultures of *Fusarium* species. Phytopathology 72:151-153.

Francis, R.G. and L.W. Burgess. 1975. Surveys of Fusaria and other fungi associated with stalk rot of maize in eastern Australia. Australian Journal of Agricultural Research 26:801-807.

Gordon, T.R. and R.D. Martyn. 1977. The evolutionary biology of *Fusarium oxysporum*. Annual Review of Phytopathology 35:111-128.

Gordon, T.R. and D. Okamoto. 1992. Population structure and the relationship between pathogenic and nonpathogenic strains of *Fusarium oxysporum*. Phytopathology 82:73-77.

Gordon, W.L. and R. Sprague. 1941. Species of *Fusarium* associated with root rots of the Gramineae in the northern Great Plains. Plant Disease Reporter 25:168-180.

Graham, J.H., D.L. Stuteville, F.I. Frosheiser and D.C. Erwin. 1979. Compendium of alfalfa diseases. The American Phytopathological Society, St. Paul, MN. 65p.

Gregory, P.H., E.J. Guthrie and M.E. Bunce. 1959. Experiments of splash dispersal of fungus spores. Journal of General Microbiology 20:328-354.

Hancock, J.G. 1985. Fungal infection of feeder rootlets of alfalfa. Phytopathology 75:1112-1120.

Hall, R. and L.G. Phillips. 1992. Effects of crop sequence and rainfall on population dynamics of *Fusarium solani* f. sp. *phaseoli* in soil. Canadian Journal of Botany 70:2005-2008.

Hartley, C., T.C. Merrill and A.S. Rhoades. 1918. Seedling diseases of conifers. Journal of Agricultural research 15:521-558.

James, R.L. 1983a. Fusarium root disease of western white pine seedlings at the Coeur d'Alene Nursery, Idaho. USDA Forest Service, Northern Region, Forest Pest Management. Report 83-19. 5p.

James, R.L. 1983b. Occurrence of *Fusarium* on Douglas-fir seed from the Coeur d'Alene Nursery. USDA Forest Service, Northern Region, Forest Pest Management. Nursery Disease Notes No. 4. 11p.

James, R.L. 1984. Fungi colonizing Douglas-fir seed at the Champion Timberlands Nursery, Plains, Montana. USDA Forest Service, Northern Region, Forest Pest Management. Report 84-13. 3p.

James, R.L. 1985. Pathogenic *Fusarium* on spruce seed from the Towner Nursery, North Dakota. USDA Forest Service, Northern Region, Forest Pest Management. Report 85-23. 9p.

James, R.L. 1986. Diseases of conifer seedlings caused by seed-borne *Fusarium* species. In: Shearer, R.C. (compiler). Proceedings: Conifer Tree Seed in the Inland Mountain West Symposium. USDA Forest Service, Intermountain Research Station, General Technical Report INT-203. pp. 267-271.

James, R.L. 1987. Occurrence of *Fusarium* spp. on conifer tree seed from Northern Rocky Mountain nurseries. *In*: Landis, T.D. (technical coordinator). Proceedings: Combined Western Nursery Council and Intermountain Nursery Association Meeting. USDA Forest Service, Rocky Mountain Forest and Range Experiment Station, General Technical Report RM-137. pp. 104-114.

James, R.L. 1989. Mortality of young, container-grown Pacific silver fir seedlings – North Woods Nursery, Elk River, Idaho. USDA Forest Service, Northern Region, Forest Pest Management. Nursery Disease Notes No. 95. 4p.

James, R.L. 1990. Needle-tip necrosis of container-grown western white pine seedlings – University of Idaho Research Nursery. USDA Forest Service, Northern Region, Forest Pest Management. Nursery Disease Notes No. 106. 4p.

James, R.L. 1996. Technique for quantifying virulence of *Fusarium* and *Cylindrocarpon* spp. on conifer germinants. USDA Forest Service, Northern Region, Forest Pest Management. Nursery Disease Notes No. 132. 8p.

James, R.L. and K. Beall. 1999. An evaluation of the effects of dazomet on soil-borne diseases and conifer seedling production - USDA Forest Service Lucky Peak Nursery, Boise, Idaho. USDA Forest Service, Northern Region, Forest Health Protection. Report 99-9. 15p.

James, R.L. and K. Beall. 2000. Effects of fallowing on *Fusarium*-associated root diseases and production of bare root ponderosa pine seedlings at the USDA Forest Service Lucky Peak Nursery, Boise, Idaho. USDA Forest Service, Northern Region, Forest Health Protection. Report 00-3. 13p.

James, R.L., R.K. Dumroese and D.L. Wenny. 1989. Occurrence, characteristics, and descriptions of *Fusarium* isolates from Douglas-fir seed and seedlings. USDA Forest Service, Northern Region, Forest Pest Management. Report 90-4. 23p.

James, R.L., R.K. Dumroese and D.L. Wenny. 1991. *Fusarium* diseases of conifer seedlings. *In*: Sutherland, J.R. and S.G. Glover (eds.). Proceedings of the first meeting of IUFRO Working Party S2.07.09 (Diseases and Insects in Forest Nurseries). Forestry Canada, Pacific and Yukon Region. Information Report BC-X-331. pp. 181-190.

James, R.L., R.K. Dumroese and D.L. Wenny. 1995. *Fusarium proliferatum* is a common, aggressive pathogen of container-grown conifer seedlings. Phytopathology 85:1129.

James, R.L., R.K. Dumroese and D.L. Wenny. 1997. Pathogenicity of *Fusarium proliferatum* in container-grown Douglas-fir seedlings. *In*: James, R.L. (editor). Proceedings of the third meeting of IUFRO Working Party S7.03-04 (Diseases and Insects in Forest Nurseries). USDA Forest Service, Northern Region, Forest Health Protection. Report 97-4. pp. 26-33.

James, R.L. and D. Genz. 1982. Evaluation of fungal populations on ponderosa pine seed. USDA Forest Service, Northern Region, Forest Pest Management. Report 82-22. 21p.

James, R.L. and C.J. Gilligan. 1985. Containerized Engelmann spruce seedling diseases at the USDA Forest Service Nursery, Coeur d'Alene, Idaho. USDA Forest Service, Northern Region, Forest Pest Management. Report 85-17. 15p.

James, R.L. and C.J. Gilligan. 1988. Occurrence of *Fusarium* on the roots of nondiseased bareroot Douglas-fir seedlings - USDA Forest Service Nursery, Coeur d'Alene, Idaho. USDA Forest Service, Northern Region, Forest Pest Management. Report 88-12. 4p.

James, R.L., S. Metzger and C.J. Gillian. 1990. Effects of soil fumigation on conifer seedling production at the USDA Forest Service Nursery, Coeur d'Alene, Idaho. USDA Forest Service, Northern Region, Forest Pest Management. Report 90-11. 18p.

James, R.L., D.S. Page-Dumroese, S.K. Kimball and S.Omi. 1996. Effects of *Brassica* cover crop, organic amendment, fallowing and soil fumigation on production of bareroot Douglas-fir seedlings - USDA Forest Service Nursery, Coeur d'Alene, Idaho. USDA Forest Service, Northern Region, Forest Health Protection. Report 96-5. 16p.

James, R.L. and R. Perez. 1999a. Fungal colonization of residual conifer seedling roots in soil - USDA Forest Service Lucky Peak Nursery, Boise, Idaho. USDA Forest Service, Northern Region, Forest Health Protection. Report 99-10. 13p.

James, R.L. and R. Perez. 1999b. Pathogenic characteristics of *Fusarium sporotrichioides* isolated from inland Pacific Northwest forest nurseries. USDA Forest Service, Northern Region, Forest Health Protection. Report 99-8. 11p.

James, R.L., R. Perez, R.K. Dumroese and D.L. Wenny. 2000. Virulence of *Fusarium oxysporum* on Douglas-fir germinants: comparisons of isolates from nursery soil and roots of healthy and diseased seedlings. In: Lilja, A. (ed.). Proceedings of the fourth meeting of IUFRO Working Party S7.03-04 (Diseases and Insects in Forest Nurseries). The Finnish Forest Research Institute (In press).

Joffe, A.Z. and J. Palti. 1977. Species of *Fusarium* found in uncultivated desert type soils in Israel. Phytoparasitica 5:119-121.

Kabir, A.K. 1965. The Columbian timber beetle and its associated microorganisms in soft maple. Review of Applied Mycology 44:161-162.

Komada, H. 1975. Development of a selective medium for quantitative isolation of *Fusarium oxysporum* from natural soil. Review of Plant Protection Research (Japan) 8:114-125.

Kommedahl, T., C.S. Windels and D.S. Lang. 1975. Comparisons of *Fusarium* populations in grasslands of Minnesota and Iceland. Mycology 67:38-44.

Kulakova, N.M. 1964. [Preventing damping-off of Scots pine seedlings](In Russian). Lesn. Khov. 17:41-44.

Miles, M.R. and R.D. Wilcoxin. 1984. Production of fungal inoculum using a substrate of perlite, cornmeal, and potato dextrose agar. Plant Disease 68:310.

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.

Parkinson, S.M., W. Wainwright and K. Killham. 1989. Observations on oligotrophic growth of fungi on silica gel. Mycological Research 93:529-534.

Pomerleau, R. 1934. The fungi responsible for seedling blight of conifers at the Berthierville forest nursery. Quebec Society of Plant Protection. Report 1932-43. pp. 58-61.

Price, D. 1984. *Fusarium* and plant pathology: the reservoir of infection. In: Moss, M.O. and J.E. Smith (eds.). The Applied Mycology of *Fusarium*. Cambridge University Press. Pp. 71-93.

Ragazzi, A., L. Mugnai, S. Moricca, S. Vagniluca and I. Dellavalle. 1993. Requirements and biological aspects of *Fusarium eumartii* and its possible role in oak decline in Northeastern Italian forests. European Journal of Forest Pathology 23:171-177.

Reddy, M.S., R.K. Hynes and G. Lazarovits. 1994. Relationship between in vitro growth inhibition of pathogens and suppression of preemergence damping-off and postemergence root rot of white bean seedlings in the greenhouse by bacteria. Canadian Journal of Microbiology 40:113-119.

Samac, D.A. and S.A. Leong. 1989. Disease development in *Cucurbita maxima* (squash) infected with *Fusarium solani* f. sp. *cucurbitae*. Canadian Journal of Botany 67:3486-3489.

Schuerger, A.C. and D.J. Mitchell. 1992. Effects of temperature and hydrogen ion concentration on attachment of macroconidia of *Fusarium solani* f. sp. *phaseoli* to mung bean roots in hydroponic nutrient solutions. Phytopathology 82:1311-1319.

Smith, W.H. and J.L. Peterson. 1966. The influence of the carbohydrate fraction of the root exudates of red clover, *Trifolium pratense*, on *Fusarium* spp. isolated from the clover root and rhizosphere. Plant and Soil 25:413-424.

Snyder, W.C., S.C. Georgopoulous, R.K. Webster and S.N. Smith. 1975. Sexuality and genetic behavior in the fungus *Hypomyces* (*Fusarium*) *solani* f. sp. *cucurbitae*. Hilgardia 43:161-185.

Snyder, W.C., M.T. Schroth and T. Christen. 1959. Effect of plant residues on root rot of bean. Phytopathology 49:755-756.

Sturz, A.V. and C.C. Bernier. 1991. Fungal communities in winter wheat roots following crop rotations suppressive and nonsuppressive to take-all. Canadian Journal of Botany 69:39-43.

Summerell, B.A., C.A. Rugg and L.W. Burgess. 1993. Mycogeography of *Fusarium*: survey of *Fusarium* species associated with forest and woodland communities in north Queensland, Australia. Mycological Research 97:1015-1019.

Vaartaja, O. and W.H. Cram. 1956. Damping-off pathogens of conifers and of *Caragana* in Saskatchewan. Phytopathology 46:391-397.

Warren, H.L. and T. Kommedahl. 1973. Fertilization and wheat refuse effects on *Fusarium* species associated with wheat roots in Minnesota. Phytopathology 63:103-108.

Wenny, D.L. and R.K. Dumroese. 1987. Germination of conifer seeds surface sterilized with bleach. Tree Planters' Notes 38(3):18-21.

APPENDIX

Table 3. Average virulence scores on young Douglas-fir germinants for *Fusarium solani* isolates from roots of healthy conifer seedlings within inland Pacific Northwest nurseries.

Isolate	No. Germinants	Average Score	Standard Deviation
907A	24	85.0	19.3
9048Q	24	47.7	25.6
9054P	24	41.9	25.6
9611K	22	30.2	19.2
9639I	23	45.4	28.4
9666D	24	26.0	16.9
Average	23.5	46.3	23.0

Table 4. Average virulence scores on young Douglas-fir germinants for *Fusarium solani* isolates from roots of diseased conifer seedlings with inland Pacific Northwest nurseries.

Isolate	No. Germinants	Average Score	Standard Deviation
9144I	23	27.4	18.8
9221K	24	58.7	29.3
9233B	24	97.1	4.5
9306F	24	43.5	29.1
9502B	21	25.5	9.2
9502J	23	27.6	9.9
Average	23.2	47.4	16.8

Table 5. Average virulence scores on young Douglas-fir germinants for *Fusarium solani* isolates from bareroot field soil from inland Pacific Northwest nurseries.

Isolate	No. Germinants	Average Score	Standard Deviation
9226D	24	64.8	19.8
9226K	24	37.3	24.1
9301L	21	45.0	28.2
9301O	22	37.0	10.9
9312K	22	25.0	11.4
9312P	22	32.9	26.1
9313B	24	20.2	10.4
9313G	20	28.5	7.8
9313H	24	54.2	21.9
9313K	24	36.9	27.1
9313M	23	34.1	12.8
9314C	23	20.2	9.1
9320D	22	43.9	24.9
9323B	24	35.0	19.9
9323H	24	26.3	14.8
9348M	23	43.5	16.4
9447F	24	26.5	13.3
9449I	22	22.3	8.4
9538I	24	28.7	16.0
9538I	23	37.0	16.6
9539K	22	76.1	15.4
9539Q	24	45.0	19.6
9553D	22	46.4	26.3
9558G	22	48.6	22.2
9617C	23	34.8	23.5
9618B	23	27.6	12.5
9664B	22	24.5	12.1
9715D	20	34.5	23.2
9717E	21	31.9	12.4
9719D	24	39.0	17.5
9720L	22	22.0	15.3
9731N	23	20.0	8.0
9749F	24	22.5	14.5
9750E	23	27.2	16.2
9824I	24	67.3	28.4
Average	22.8	36.2	17.3

Table 6. Average number of days' survival of young Douglas-fir germinants inoculated with *Fusarium solani* isolates from roots of healthy conifer seedlings within inland Pacific Northwest nurseries.

Isolate	No. Germinants	Average Survival[1]	Standard Deviation
907A	24	5.1	3.2
9048Q	24	10.8	3.8
9054P	24	12.0	3.3
9611K	22	13.3	1.9
9639I	23	11.5	3.9
9666D	24	13.7	1.2
Average	23.5	11.0	2.9

[1] Maximum survival of 14 days (length of pathogenicity test).

Table 7. Average number of days' survival of young Douglas-fir germinants inoculated with *Fusarium solani* isolates from roots of diseased conifer seedlings with inland Pacific Northwest nurseries.

Isolate	No. Germinants	Average Survival[1]	Standard Deviation
9144I	23	13.3	2.1
9221K	24	9.2	4.4
9233B	24	3.5	0.7
9306F	24	11.2	3.8
9502B	21	14.0	0
9502J	23	13.9	0.2
Average	23.2	10.7	1.9

[1] Maximum survival of 14 days (length of pathogenicity test).

Table 8. Average number of days' survival of young Douglas-fir germinants inoculated with *Fusarium solani* isolates from bareroot soil from inland Pacific Northwest nurseries.

Isolate	No. Germinants	Average Survival[1]	Standard Deviation
9226D	24	8.8	3.4
9226K	24	12.3	3.2
9301L	21	11.7	3.7
9301O	22	13.7	1.5
9312K	22	14.0	0
9312P	22	12.5	3.0
9313B	24	14.0	0
9313G	20	14.0	0
9313H	24	11.1	3.1
9313K	24	12.0	3.4
9313M	23	13.6	1.5
9314C	23	14.0	0
9320D	22	11.9	2.8
9323B	24	13.3	2.5
9323H	24	13.7	1.4
9348M	23	12.9	2.4
9447F	24	13.8	0.4
9449I	22	14.0	0
9538I	24	13.7	1.4
9539I	23	13.3	2.2
9539K	22	7.8	3.1
9539Q	24	12.8	3.3
9553D	22	11.9	3.9
9558G	22	11.8	3.4
9617C	23	12.7	2.7
9618B	23	14.0	0
9664B	22	14.0	0
9715D	20	12.8	2.6
9717E	21	13.7	1.5
9719D	24	13.5	2.2
9720L	22	13.7	1.5
9731N	23	14.0	0
9749F	24	13.8	0.9
9750E	23	13.7	1.4
9824I	24	9.0	5.1
Average	22.8	12.8	1.9

[1] Maxi mum survival of 14 days (length of pathogenicity test).