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WESTERN LARCH SEED - CONTAMINATING FUNGI AND TREATMENTS TO REDUCE INFECTION AND IMPROVE GERMINATION

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ABSTRACT

Seventeen seedlots of western larch from the Idaho Department of Lands (IDL) were assayed for fungal populations on seed. The most prominent group of fungi encountered were *Penicillium* spp. Potentially pathogenic fungi, found at low levels, included species of *Fusarium*, *Phoma*, *Cladosporium*, *Botrytis cinerea*, *Alternaria alternata*, and *Trichothecium roseum*. Another seedlot of IDL western larch, designated 1988, was extensively contaminated with *Fusarium* spp. (77-90 percent of seed infected). Treatments with high bleach concentrations did not reduce *Fusarium* levels, but did reduce other fungi, particularly *Penicillium* spp. Immersion in microwave-heated water for 80-100 seconds significantly reduced *Fusarium* levels and other fungi. Unfortunately, some bleach and hot water treatments adversely affected seed germination. Western larch seedlots extensively contaminated with *Fusarium* will perform poorly and may not be effectively treated to reduce contamination without causing seed toxicity.

INTRODUCTION

Western larch (*Larix occidentalis* Nutt.) is an important component of mixed-conifer forest stands in the inland Pacific Northwest. The species is highly prized for its rapid growth (Schmidt and Shearer 1995), superior wood quality (Keegan and others 1995), and tolerance to important root disease-causing organisms that often plague other conifer species in mixed-conifer ecosystems (Carlson and others 1995; Hagle and Shaw 1991). Therefore, demand for western larch seedlings for reforestation is high. Bareroot and container-grown larch seedlings are produced at several nurseries in Idaho and Montana (Dumroese and Wenny 1995; James and others 1995).

Unfortunately, one important problem affecting nursery seedling production is lack of sufficient high quality seed (James and others 1995). Abundant western larch seed crops are cyclic (Schmidt and Shearer 1995); weather-related cone damage can be extensive (Shearer and

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Theroux 1986). Insects and pathogens may also limit seed production (James 1988b; Shearer 1984). Therefore, when abundant seed crops occur, extensive collections are usually made, replenishing seed stocks. In this way, one specific seedlot may be used in nursery seedling production for several years.

The Idaho Department of Lands (IDL) collects western larch cones as an important initial step in reforestation of their lands. Cones are sent to nurseries and/or processing companies for seed extraction. Seeds are used to produce seedlings under contract for sale to the IDL.

In the past, some western larch seedlots were of very poor quality, resulting in low germination and production of poor quality seedlings which were more susceptible to disease (James 1986c, 1988a, 1990). There is a direct correlation between occurrence and extent of certain microorganisms on seed and eventual seedling performance in nurseries (James 1987a, 1987b; James and others 1995). Although most conifer seed routinely carries fungi externally, such organisms may or may not adversely affect seed quality (Anderson 1986). Certain organisms are more capable of causing problems with germination and seedling establishment than others. Therefore, it is important that seed mycoflora be identified as well as quantified if effects on seed quality are to be reliably predicted (Neergaard 1977).

Compared with other conifer species, information on fungi commonly contaminating western larch seed is less readily available (Anderson 1986; James 1986a, 1987b; James and Genz 1982). Also, little is known about efficacy of standard seed treatments to reduce fungal contamination and enhance larch seed performance. Treatments that work well for other, larger seeded, conifer species may be inappropriate for larch seed. Therefore, an evaluation was conducted to quantify and characterize fungal populations on selected IDL western larch seedlots. In addition, efficacy of seed sorting and pre-germination treatments to reduce fungal contamination was

evaluated on a specific IDL seedlot with relatively high levels of potentially pathogenic fungi.

MATERIALS AND METHODS

To characterize fungi on a diverse range of western larch seedlots, 17 IDL seedlots processed by the USDA Forest Service Nursery in Coeur d'Alene, Idaho were evaluated for the quantity and identity of fungi commonly contaminating seed. Seeds were obtained from bulk storage (pre-stratification). Two hundred seeds per seedlot were aseptically placed on an agar medium selective for *Fusarium* spp. and closely related fungi (Komada 1975). Plates were incubated for 7-10 days at about 24°C, under diurnal cycles of cool, fluorescent light. Selected fungi emerging from seed were transferred to potato dextrose agar and carnation leaf agar (Fisher and others 1982) to aid identification. Morphological characteristics of sporulation were the major means of identification using several taxonomic treatises (Barnett and Hunter 1972; Domsch and others 1980; Ellis 1971; Nelson and others 1983; Raper and Thom 1949). When possible, fungal isolates were identified to species. Percent of sampled seed colonized with specific fungi were calculated for each seedlot.

IDL seedlot (designated 1988) had a history of poor performance. This was a large seedlot and managers were concerned that its continued use would be necessary because other seedlots were unavailable. Seeds from this seedlot were initially sorted using the incubation drying separation (IDS) procedure (Simak 1984). Seeds were soaked in distilled water for 24 hours at 22°C. They were then spread in a 1-seed-deep layer on moist paper towels, enclosed in plastic bags, and incubated 3 days at 15°C. After incubation, seeds were dried 12 hours at 15°C, then poured into a larger beaker of distilled water. This technique separates seeds based on their ability to either sink or float in solution. Theoretically, seeds that sink contain normal endosperms and are viable, whereas those that float may not be fully developed, are empty, or contain decayed endosperms. Under normal seed processing,

"floaters" would be discarded and not sown. Twenty-two treatments were conducted (Table 1). Selected seeds in the three sorting categories (untreated, sinkers, floaters) were treated with three solutions of bleach: 50 percent, 75 percent, and 100 percent (2.63 percent, 3.94 percent and 5.25 percent aqueous sodium hypochlorite, respectively). Treatments were for 10 min., after which seeds were rinsed in sterile water, blotted dry and placed on the selective agar medium. In addition, selected seeds in each of the three categories were also treated with hot water. Water, into which seeds were placed, was heated with microwaves in a commercial oven (1,400 watts heating power; 2450 MHz) 60, 80, or 100 seconds, resulting in final water temperatures of 45, 56 and 62°C, respectively. After hot water treatment, seeds were blotted dry and placed on selective agar media. One group of unsorted seed were treated with 5 percent aqueous sodium metabisulfite for 30 seconds, rinsed in sterile water, blotted dry and placed on selective agar media. Controls consisted of untreated seeds in the three sorting categories: unsorted, sinkers, and floaters. Pieces of debris associated with seeds (wings, cone scales, and aggregations of resin) were randomly selected from the seedlot and exposed to the same treatments as seeds. All agar plates with seeds or debris were incubated as described above. Two hundred fifty seeds were analyzed for each control and 300 seeds analyzed in the sodium metabisulfite treatment. All other treatments consisted of 100 seeds. Most emerging fungi were identified to genus and effects of treatments on percentage colonization by groups of fungi calculated.

Treatment effects were analyzed with a one-way analysis of variance (Snedecor and Cochran 1989). Significant treatment means were separated using Tukey's HSD. All percentages underwent arc-sin conversion prior to analysis.

RESULTS AND DISCUSSION

Assaying seeds for fungi presents unique problems because of the numerous organisms carried on or within seed (Neergaard 1977). If non-selective

Table 1. Treatments to sort and surface sterilize western larch seeds - 1988 Idaho Department of Lands seedlot

Treatm No.	Treatment Description ¹
1	Control #1 - unsorted, untreated (from bulk)
2	Control #2 - floaters, untreated
3	Control #3 - sinkers, untreated
4	Unsorted - 50% bleach solution
5	Unsorted - 75% bleach solution
6	Unsorted - 100% bleach solution
7	Floaters - 50% bleach solution
8	Floaters - 75% bleach solution
9	Floaters - 100% bleach solution
10	Sinkers - 50% bleach solution
11	Sinkers - 75% bleach solution
12	Sinkers - 100% bleach solution
13	Unsorted - 60 second hot water
14	Unsorted - 80 second hot water
15	Unsorted - 100 second hot water
16	Floaters - 60 second hot water
17	Floaters - 80 second hot water
18	Floaters - 100 second hot water
19	Sinkers - 60 second hot water
20	Sinkers - 80 second hot water
21	Sinkers - 100 second hot water
22	Unsorted - sodium metabisulfite solution

¹Seeds separated using IDS procedure: floaters = seeds that floated in solution; sinkers = seeds that did not float in solution. Bleach treatments: 50%, 75% and 100% bleach solution = 2.63%, 3.95%, and 5.25% aqueous sodium hypochlorite solution, respectively (treated for 10 minutes and rinsed in sterile water). Hot water treatments = immersion in water heated by microwaves for either 60, 80, or 100 seconds (water temperatures: 45, 56, and 62°C, respectively). Sodium metabisulfite treatment = 5% solution soaked for 30 seconds and rinsed in sterile water.

media are used, the fastest-growing organisms may obscure presence of others that may be present at greater concentrations. For example, many mucoraceous fungi colonize seeds of many plants, including forest trees (Neergaard 1977).

These fungi grow extremely rapidly on standard nutrient agar, often overgrowing any other fungi present. Unless a medium that helps restrict development of these fungi is used, it is impossible to ascertain presence of other seed-colonizing organisms, particularly those that might be potential plant pathogens. Therefore, the selective medium formulated by Komada (1975) was used because it repressed growth of most mucoraceous fungi while allowing limited growth of other fungi colonizing seed, particularly those that may cause damping-off and root disease (James and others 1991).

Many different fungal taxa were isolated from the 17 IDL larch seedlots. By far, the most prevalent genus was *Penicillium* (Table 2). Nine different *Penicillium* species were found on seed, the most common being *P. chrysogenum* Thom, *P. glabrum* (Wehner) Westling, and *P. oxalicum* Currie and Thom. Species isolated less frequently included *P. citrinum* Thom, *P. expansum* Thom, *P. fuscum* Westling, *P. patulum* Bain., *P. rubrum* Stoll. and *P. viridicatum* Westling. Several of these species are common inhabitants of plant seeds in general (Neergaard 1977) and conifer seed in particular (Anderson 1986; James and Genz 1982).

Penicillium chrysogenum, *P. glabrum* and *P. oxalicum* are all common contaminants of ponderosa pine seed (James and Genz 1982). Effects of these fungi on performance of conifer seed (germination capacity and seedling establishment) may be variable (Neergaard 1977; Urosevic 1961). *Penicillium* spp. may elicit limited seed decay and adversely affect germination under certain conditions (Mason and Van Arsdel 1978; Schubert 1960; Urosevic 1961).

Penicillium spp. are very common on seedcoats of conifers (Anderson 1986; James and Genz 1982), although they are unlikely to be detrimental. On the other hand, their colonization of seeds may exclude other fungi that could adversely affect seed performance (James and Genz 1982).

Fusarium spp. are very important pathogens of conifer seedlings in nurseries (James and others 1991) and are often carried on and within seeds (James 1986a; 1987b). However, these fungi were found at relatively low levels on the IDL western larch seedlots assayed (Table 3). Only 11 of the sampled seedlots were infected with *Fusarium* spp., and these had very low infection. Four species were isolated: *F. acuminatum* Ell. & Ev., *F. oxysporum* Schlecht., *F. sambucinum* Fuckel, and *F. sporotrichioides* Sherb. Most of these *Fusarium* spp. were previously isolated from other western larch seedlots (James 1986b, 1986c, 1987a, 1988a, 1990). Generally, *Fusarium* infection of less than 10 percent in a particular seedlot is considered normal (James 1987b).

Four *Phoma* spp. were located on seeds (Table 3) and found at higher levels than *Fusarium*. They included *P. eupyrena* Sacc., *P. glomerata* (Cda.) Wollenw. & Hochapf., *P. herbarum* Westend., and *P. pomorum* Thum. Although these species have been implicated in seedling diseases (James 1983a, 1983b, 1985; James and Hamm 1985), their importance as seed pathogens needs further investigation.

Several other fungi were isolated from the IDL western larch seedlots (Table 4). Most were probably saprophytes, although a few may occasionally be pathogenic either on seed or young seedlings (Neergaard 1977). The most probable pathogens included *Botrytis cinerea* Pers. ex Fr. (James 1984), *Trichothecium roseum* (Pers.) Link ex S. F. Gray (Neergaard 1977; Urosevic 1961), *Alternaria alternata* (Fr.) Keissler (James and Woo 1987; Neergaard 1977; Urosevic 1961), and *Cladosporium* sp. (Neergaard 1977). Occurrence of *B. cinerea* is especially noteworthy since this fungus is an important pathogen of container-grown western larch (James 1984; James and Genz 1983; James and others, 1982).

Table 2. Contamination of selected Idaho Department of Lands western larch seedlots with *Penicillium* spp.

Seedlot	Percent Colonization ¹									
	<i>P. citrinum</i>	<i>P. chrysogenum</i>	<i>P. expansum</i>	<i>P. fuscum</i>	<i>P. glabrum</i>	<i>P. oxalicum</i>	<i>P. patulum</i>	<i>P. rubrum</i>	<i>P. viridicatum</i>	All species
C051	8.5	62.5	1.0	1.0	45.0	12.0	0	0	1.0	99.5
C052	1.0	57.0	5.5	0	45.0	9.0	0	0	0.5	98.0
C053	0.5	59.0	2.5	2.5	39.0	10.5	0	0	0.5	99.0
C054	2.5	52.5	2.0	0	34.5	8.0	0	0	0	90.0
K052	0	62.5	4.0	0.5	36.0	11.0	0	0	0	92.0
K053	1.0	58.5	3.0	1.0	37.0	12.0	1.0	0.5	0.5	95.0
K054	0.5	52.0	3.5	0.5	32.0	9.0	0	0	0.5	82.5
K055	0	72.5	3.5	0.5	26.0	7.0	0.5	0	0	97.0
K057	1.0	64.0	1.5	0	32.5	9.5	0	0	0	93.5
M019	0	79.0	0.5	0	25.5	4.5	0	0	0	99.0
P057	0	59.5	0.5	0	41.5	12.5	0	0	0	97.0
P059	1.5	52.0	0.5	0.5	41.5	15.0	0	0	0	96.5
P080	1.0	51.0	3.0	0	44.0	11.0	0	0	0	98.0
P081	1.0	47.5	5.5	0	27.5	21.0	0	0	0	94.5
S053	0.5	71.0	3.5	1.0	17.5	12.5	0	0	0	97.5
S054	0	78.0	5.0	0	18.5	6.0	0	0	0	98.0
S055	0.5	73.0	4.0	0.5	17.0	10.5	0	1.0	0.5	97.0
Avg.	1.1	61.9	2.9	0.5	32.9	10.7	0.1	0.1	0.2	95.2

¹ 200 seeds sampled per seedlot; percent colonization reflects growth from seedcoat when incubated on selective agar medium.

Table 3. Contamination of selected Idaho Department of Lands western larch seedlots with *Fusarium* and *Phoma* spp.

Seedlot	Percent Colonization ¹									
	<i>Fusarium</i> spp. ²					<i>Phoma</i> spp. ³				
	<i>FACU</i>	<i>FOXY</i>	<i>FSAM</i>	<i>FSPO</i>	<i>ALL</i>	<i>PHEU</i>	<i>PHGL</i>	<i>PHHE</i>	<i>PHPO</i>	All
C051	0	0	0	0	0	3.0	3.5	2.0	0	8.5
C052	0	0.5	0	0	0.5	6.0	5.5	1.5	0	13.0
C053	0.5	0	0.5	0	1.0	5.0	7.0	2.0	0.5	14.0
C054	0	0	0	0	0	5.0	5.0	2.0	0	12.0
K052	0	0.5	0	0	0.5	4.0	3.5	1.0	0	8.5
K053	0	0.5	0	0	0.5	3.5	3.5	2.0	0.5	9.5
K054	0	0.5	0	0	0.5	3.0	0.5	3.5	0	7.9
K055	0	0	0	0	0	0.5	0	4.0	0	4.5
K057	3.0	1.0	0	0	4.0	3.5	2.0	9.0	0	14.5
M019	0	0	0	0	0	0	3.0	1.0	0	4.0
P057	0.5	0	0	0	0.5	2.5	2.5	11.5	0	16.5
P059	0.5	0	1.5	0	2.0	0.5	12.5	3.5	0	16.5
P080	0.5	0	0	0.5	1.0	0.5	2.5	0.5	0	3.5
P081	0	0	0	0	0	4.0	12.0	2.0	0	18.0
S053	0.5	0	2.0	0	2.5	1.0	6.5	0	0	7.5
S054	0	0	0	0	0	1.5	12.5	2.0	0	16.0
S055	0	0	0.5	0	0.5	9.5	8.5	2.0	0	20.0
Avg.	0.3	0.2	0.3	0.1	0.8	3.1	5.2	2.9	0.1	11.4

¹ 200 seeds sampled per seedlot; percent colonization reflects growth from seedcoat when incubated on selective agar medium

² *FACU* = *F. acuminatum*; *FOXY* = *F. oxysporum*; *FSAM* = *F. sambucinum*; *FSPO* = *F. sporotrichioides*l.

³ *PHEU* = *P. eupyrena*, *PHGL* = *P. glomerata*; *PHHE* = *P. herbarum*, *PHPO* = *P. pomorum*.

Table 4. Contamination of selected Idaho Department of Lands western larch seedlots with miscellaneous fungi.

Percent Colonization ¹											
Fungal Contaminant ²											
	ALT	ASP	BOT	CHA	CLA	GEN	MUC	PES	RHI	TRI	TRO
CO51	0	1.0	1.5	0	0	0	0	0	0	0	0
CO52	0	0	1.5	0	0	0	26.5	0	10.5	1.0	0
CO53	0	0	2.0	0	0	0	2.5	0	0	0.5	0
CO54	0	0	4.5	0	0	0	0.5	0	0	9.5	0
KO52	0	0	0	0	0	0	3.0	0	0.5	12.5	0
KO53	0.5	2.0	2.5	0	0	1.0	0	0	0	13.5	0
KO54	0	4.5	0.5	0	0	0	1.0	0	0	34.0	2.0
KO55	0	0	0.5	0	0	0	0	0	0	3.5	0
KO57	0	0	3.0	0	0	0	8.5	0	1.5	3.5	0
MO19	0	0	0	0	0	0	0	0	0	1.5	0
PO57	0.5	0	2.0	0	0.5	0	0	1.0	0	1.0	0.52
PO59	0	3.5	0	0.5	0	0	0	0	0	0	0
PO80	0	2.0	3.5	0	0	0	0.5	0	0	10.0	0.5
PO81	1.0	0	0.5	0	0	0	0.5	0	0	3.0	0
SO53	0	0	3.5	0	0	0	2.5	0	0	2.0	0
SO54	1.5	0	3.5	0	0	0	0	0	0	0.5	0
SO55	0	0	1.5	0	0	0	0.5	0	0.5	0.5	0
Avg.	0.1	0.7	1.8	0.1	0.1	0.1	2.7	0.1	0.7	5.7	0.2

¹ 200 seeds sampled per seedlot; percent colonization reflects growth from seedcoat when incubated on selective agar medium.

² ALT = *Alternaria alternata*; ASP = *Aspergillus* spp.; BOT = *Botrytis cinerea*; CHA = *Chaetomium globosum*; CLA = *Cladosporium* sp.; GEN = *Geniculodendron pyriforme*; MUC = *Mucor* spp.; PES = *Pestalotia* sp.; RHI = *Rhizopus* spp.; TRI = *Trichoderma* spp.; TRO = *Trichothecium roseum*.

Botrytis was found on 14 of the 17 seedlots, although it occurred at relatively low levels; introducing this pathogen into greenhouses on seeds may be important (James 1984). Other potential plant pathogens occurred at very low levels, with *A. alternata* on only 4 of the seedlots, *T. roseum* on three, and *Cladosporium* on one. Occurrence of common saprophytes, *Trichoderma* spp and *Mucor* spp., was much more common. Mucoraceous fungi (*Mucor* and *Rhizopus* spp.) were regularly detected, but may have been more abundant than detected levels because the selective medium used for the assay restricted their growth.

In contrast to the 17 western larch seedlots described above, the 1988 IDL seedlot was extensively colonized with *Fusarium* spp. (Table 5). Approximately 60 percent of the fusaria isolated from seeds were *F. acuminatum* or *F. avenaceum* (Fr.) Sacc.; about 20 percent were *F. poae* (Peck) Wollenw. and 10 percent were either *F. equiseti* (Corda) Sacc. or *F. oxysporum*. This seedlot also contained relatively high background levels of other potentially-pathogenic fungi including *B. cinerea*, *A. alternata*, *Phoma* spp., and *Cylindrocarpon* spp. Level of *Penicillium* spp. was less than that found in the other larch seedlots (Tables 2 and 5), indicating when higher *Penicillium* occur on seeds, fewer potential pathogens may be present.

Because of extensive *Fusarium* levels on the 1988 larch seedlot, bleach treatment concentrations exceeded those normally recommended for conifer seed (Wenny and Dumroese 1987). Bleach treatments were generally ineffective in reducing seedborne *Fusarium* levels (Table 5). In some cases, bleach treatment resulted in assaying higher levels of *Fusarium* than on untreated seeds. *Fusarium* spp. are apparently less susceptible to bleach treatment than several other seed-colonizing fungi. Therefore, more *Fusarium* might be assayed because of reduced buffering capacity by other competing fungi. The most effective treatments to reduce *Fusarium* spp. were immersion in microwave-heated water for 80-100 seconds; immersion for only 60 seconds was not effective. However, even treatment in hot water for 100 seconds failed to reduce seed infection to desirable levels, i.e., below 10 percent, even though it reduced seed germination (Table 5). Microwave hot water treatment was highly effective in reducing *Fusarium* levels on Douglas-fir seed without reducing germination (Dumroese and others 1988; James and others 1988).

Other fungi responded to seed treatment differently. *Penicillium* spp. were very susceptible to bleach treatment, but less affected by hot water treatment (Table 5). However, hot water treatment, especially for 100 seconds, was effective in removing most fungi from contaminated seed (see "No fungi" column in Table 5).

Germination capacity of this seedlot was quite low; those seeds which sank in solution during sorting had higher germination than those which floated, indicating that denser seeds were more viable. All bleach and hot water treatments reduced seed germination somewhat, but extensive reduction only occurred when seeds were exposed to hot water for 100 seconds. Exposure of Douglas-fir seed to hot water (66.5°C) for 120 seconds reduced germination to zero (Dumroese and others 1988).

Treatment with sodium metabisulfite (treatment 22 - Table 5) failed to significantly reduce levels of *Fusarium* spp. or other fungi on western larch seeds. Although this chemical is effective in reducing fungal inoculum on styrofoam and plastic containers used to grow seedlings (Dumroese and others 1993), it was not effective in reducing fungi on western larch seed, at least at the concentration tested.

Seed debris (wings, cone scales, resin aggregations) from the 1988 seedlot were also extensively colonized with *Fusarium* spp. and other fungi (Table 6).

Treatments with bleach and microwave-heated water resulted in similar responses by fungi on debris as on seed (compare Tables 5 and 6). Debris carried within seedlots may be a significant source of contamination with potentially pathogenic fungi.

Table 5. Effects of sorting, bleach and hot water treatments on occurrences of *Fusarium*, *Penicillium* and other fungi and germination of western larch seeds - 1988 Idaho Department of Lands seedlot.

Percent Colonization ¹					
Treatment No. ²	<i>Fusarium</i>	<i>Penicillium</i>	Other Fungi ³	No Fungi ⁴	Percent Germination
1	78 BCD	52 E	34 DE	0 E	58
2	89 AB	67 CD	48 BD	0 E	52
3	90 AB	78 BC	54 AB	0 E	72
4	61 DE	3 I	42 BCD	12 C	56
5	44 EF	29 FG	26 DE	8 C	42
6	66 CDE	8 HI	66 A	4 D	34
7	01 AB	6 I	53 AB	0 E	48
8	91 AB	4 I	39 BCD	0 E	48
9	97 A	3 I	24 DE	0 E	32
10	88 AB	0 I	30 DE	4 D	66
11	91 AB	1 I	28 DE	1 E	56
12	93 AB	1 I	30 DE	0 E	52
13	88 AB	45 EF	33 DE	0 E	60
14	17 H	98 A	21 E	1 E	62
15	16 H	22 GH	4 F	60 A	30
16	79 BCD	59 DE	57 AB	0 E	52
17	37 FG	97 A	29 DE	1 E	48
18	19 GH	54 DE	26 DE	25 B	40
19	82 BC	60 DE	29 DE	0 E	66
20	23 FGH	95 A	30 DE	0 E	52
21	15 H	97 A	24 DE	1 E	46
22	86 B	82 B	37 CD	0 E	54

¹Percent of seeds colonized with appropriate fungi. Within each column, means followed by the same capital letter are not significantly different (P=0.05) using Tukey's HSD.

²See Table 1 for description of treatments.

³Includes *Cylindrocarpon* spp., *Trichoderma* spp., *Phoma* spp., *Alternaria alternata*, and *Botrytis cinerea*.

⁴No fungi emerged from incubated seeds; many were colonized with unidentified bacteria.

Table 6. Effects of sorting, bleach and hot water treatments on occurrence of *Fusarium*, *Penicillium* and other fungi on debris from western larch seedlot 1988, Idaho Department of Lands.

Treatment No. ²	Percent Colonization ¹			
	<i>Fusarium</i>	<i>Penicillium</i>	Other Fungi ³	No Fungi ⁴
1	90	75	55	0
2	80	65	85	0
3	55	75	75	0
4	65	0	25	30
5	65	5	15	25
6	20	10	45	35
7	100	0	40	0
8	95	0	45	0
9	90	5	40	0
10	90	0	45	0
11	80	0	35	10
12	80	15	45	0
13	95	50	50	0
14	35	100	20	0
15	15	35	10	55
16	85	75	65	0
17	25	100	25	0
18	0	50	15	40
19	70	70	75	0
20	10	100	30	0
21	0	65	20	30
22	93	80	40	0

¹Percent of seed debris (wings, cone scales, resin aggregates) colonized with appropriate fungi based on 20 randomly selected pieces of debris per treatment.

²See Table 1 for description of treatments.

³Includes *Cylindrocarpon* spp., *Trichoderma* spp., *Phoma* spp., and *Alternaria alternata*.

⁴No fungi emerged from incubated seed debris; many pieces were colonized with unidentified bacteria.

CONCLUSIONS

Our evaluation showed that seed-borne fungi are common on western larch seedlots and may adversely affect seed performance in nurseries. *Fusarium* spp. are particularly of concern because they not only adversely affect germination, but can cause high levels of seedling disease if introduced on infected seed. Reducing *Fusarium* on conifer seed may be difficult, particularly for small-seeded species like western larch which may be more sensitive to chemical sterilants than larger-seeded species. Although hot water may effectively reduce *Fusarium* on seed, it is

important that thresholds (temperature - exposure time) be determined so that germination may not be reduced. In some cases, seedlots with extensive *Fusarium* contamination may have to be discarded rather than trying to reduce pathogens to acceptable levels. Treatments capable of reducing *Fusarium* sufficiently may also reduce germination too much. Poor stands of seedlings with greater than normal levels of disease would likely result if such seedlots were used.

Because of variable levels of potentially pathogenic fungi found on different seedlots, it may be necessary to establish procedures for periodically assaying specific lots for pathogens. In particular, those lots with germination problems probably should be analyzed for pathogenic fungi. If specific western larch seedlots are used to produce numerous seedling crops in nurseries, they should be assayed for pathogen levels. If abundant pathogen populations are encountered, affected seedlots must be treated to reduce pathogens or discarded if treatment is not likely to be effective or practical.

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