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## PREPLANT SOIL TREATMENT EFFECTS ON PRODUCTION OF DOUGLAS-FIR SEEDLINGS AT THE USDA FOREST SERVICE NURSERY, COEUR D'ALENE, IDAHO

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# ABSTRACT

Six pre-plant soil treatments were evaluated for their effects on soil populations of potentially-pathogenic *Fusarium* and *Pythium* spp. and saprophytic *Trichoderma* spp. and production of bare root Douglas-fir seedlings at the USDA Forest Service Nursery, Coeur d'Alene, Idaho. Incorporating green manure crops of *Brassica juncea* and composted sewage sludge resulted in large increases of *Fusarium* spp. Resulting seedling production was not improved over either dazomet fumigation or fallowing by incorporating *Brassica* green manure crops. Steam treatment of soil reduced potential pathogen populations, but did not result in improved seedling production when compared to fallowing and dazomet fumigation. Addition of the biocontrol agent *Trichoderma harzianum* following incorporation of *Brassica* green manure crops did not affect either pathogen populations or seedling production. Plastic tarps to reduce volatile losses of decomposition products of *Brassica* residues only reduced soil *Pythium* levels and did not improve seedling production. Results of this evaluation indicated that the *Brassica* cultivar tested did not provide effective disease control and could not be used as a viable alternative to either dazomet fumigation or fallowing for producing high-quality conifer seedlings at the Coeur d'Alene Nursery.

### INTRODUCTION

Bare root forest seedling production in the United States has routinely depended on preplant soil fumigation with chemical biocides to ensure production of high-quality stock for reforestation. Most nurseries have relied on methyl bromide/chloropicrin (MBC) mixtures for fumigation (Boone 1988; Boyd 1971; Ibarbia 1995; James 1989). However, methyl bromide is currently being phased out and scheduled for elimination as a soil fumigant in January 2005, primarily because it significantly contributes to the deterioration of ultraviolet light-protective stratospheric ozone (Shaheen 1996; World Meterological Association 1995). Methyl bromide has been a very effective soil fumigant for many years and, as a result, many nurseries have become reliant on this chemical. However, evaluations of possible alternatives to methyl bromide and/or pre-plant soil fumigation have been conducted at some nurseries (Chapman 1992; James et al. 1996; Linderman et al. 1994)

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The USDA Forest Service Nursery in Coeur d'Alene, Idaho has a history of using pre-plant soil fumigants to control soilborne plant pathogens and weeds. MBC had been the fumigant of choice, but was recently replaced with dazomet (Basamid granular®) because it was as effective as MBC and had less potential adverse environmental consequences (James et al. 1990, 1996). Dazomet is usually applied in the late summer or early fall prior to sowing the following spring. It is applied topically, cultivated into the soil and activated/sealed with overhead irrigation. The chemical becomes volatile when wetted and does not require tarping with plastic polyethylene like MBC (James and Beall 1999; James et al. 1996; Miller and Norris 1970).

Although dazomet is effective at the Coeur d'Alene Nursery (James et al. 1990, 1996), it is expensive, requires expert applicators, and still presents potential environmental risks at the nursery. Therefore, growers have encouraged development of possible alternatives to all chemical soil fumigation. A series of tests have been conducted at the nursery to evaluate costeffective, efficacious alternatives to chemical soil fumigation. This report summarizes findings of tests involving six different soil treatments on the of Douglas-fir (Pseudotsuga production menziesii Franco var. glauca [Mayr.] Sudw.) seedlings at the nursery.

#### MATERIALS AND METHODS

Tests were initiated during the summer of 1996. Each treatment was replicated five times; treatment blocks were located within a section of Field 9 in a complete randomized block design. Each treatment block was 50 ft (15.5 m). in length and one seedling bed width, with the exception of the dazomet treatments which were two bed widths because of the application machinery requirements. The six treatments

were: (1) standard dazomet soil fumigation (300 lbs./acre; 335 kg/hectare); (2) bare fallowing with periodic cultivation; (3) mustard (Brassica juncea - variety Pacific Gold) green manure crop that was incorporated into the soil (figure 1) after 6 weeks' growth and covered with a 2 mil clear polyethylene tarp; (4) mustard green manure crop without tarp; (5) topical applications of composted sewage sludge to a depth of 2.5 cm (figure 3)and then tilled into soil; (6) steam treatment of soil with a machine fabricated by the USDA Forest Service Technology Development Center (Missoula, Montana)(figure 4). Soil temperatures from steam treatments varied with depth and time after treatment. Maximum temperatures reached about 68°C at 13 cm just after treatment. Temperatures exceeding 63°C were detected below 20 cm from the soil surface. To evaluate potential effects of a biological control agent on these treatments, a formulation of Trichoderma harzianum Rifai (strain ThzID1) was added to a portion of the dazomet, fallow, and mustard (no tarp) treatments just prior to sowing in the spring of 1997.

Soil populations of potential pathogens in the genera *Fusarium* and *Pythium* and potential antagonists in the genus *Trichoderma* were assayed three times during the test. The first sample was taken in July, 1996 (pre-treatment); the second in October, 1996 (post-treatment) and the third in late April, 1997 (pre-sowing). Samples were collected from within each replicate plot; three cores from near the center of each plot were collected and mixed together to represent a single sample. Each collection consisted of a soil core taken to a depth of about 8 inches (20 cm). Soil was placed in plastic bags, kept refrigerated, and transported to the laboratory for analysis



Figure 1. *Brassica juncea* green manure crop flowering after being grown for 6 weeks at the USDA Forest Service Nursery, Coeur d'Alene, Idaho



Figure 2. Incorporation of *Brassica juncea* green manure crop into soil after chopping - USDA Forest Service Nursery, Coeur d'Alene, Idaho

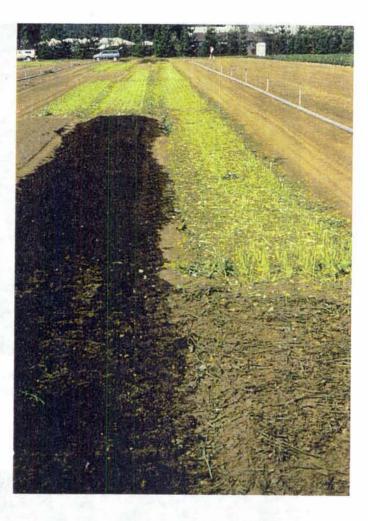


Figure 3. Topical application of sewage sludge compost on a replicate-plot - USDA Forest Service Nursery, Coeur d'Alene, Idaho. Compost was incorporated into soil after application.



Figure 4. Injecting steam into soil to control soilborne pathogens - USDA Forest Service Nursery, Coeur d'Alene, Idaho. Standard soil dilutions (Hildebrand and Dinkel 1988; James et al. 1990, 1996; Stone et al. 1995) were conducted to estimate populations of Fusarium, Trichoderma, and Pythium spp. Soil from each sample was initially sieved (2-mm sieve) to remove rocks, pieces of organic matter, and soil aggregates. From each sample, an approximate 5-g subsample was oven-dried at about 100°C for at least 24 h until sample weight stabilized. Oven-dry weight was then calculated to provide a standard for sample comparison. For assays of Fusarium and Trichoderma populations, 0.05 g of field-moist soil was combined with 10 ml of 0.3% water agar (WA) and thoroughly mixed. One milliliter of solution was placed on each of three plates of selective agar medium for Fusarium and closely-related fungi (Komada 1975) and spread uniformly. Trichoderma propagules were also enumerated on Komada's medium which readily supports growth of this fungus unless the medium is amended with benomyl or lithium chloride. Plates were incubated at least 7 days at about 24°C under diurnal cycles of cool, fluorescent light. Fusarium and Trichoderma colonies were identified by their morphology on the selective medium: populations were expressed as number of colony-forming units (cfu) per gram of ovendried soil (it was assumed that each fungal colony originated from one propagule). Selected Fusarium isolates were transferred to carnation leaf agar (Fisher et al. 1982) and potato dextrose agar (PDA) for species identification using the taxonomy of Nelson et al. (1983). Ratios of Trichoderma to Fusarium populations were calculated for each treatment; these ratios may indicate a very rough estimate of potential disease suppressiveness of the soil since Trichoderma spp. are known antagonists of a wide range of soilborne plant pathogens, including Fusarium spp. (Papavizas 1985).

For assays of *Pythium* populations, 0.5 g of soil was combined with 10 ml of 0.3% WA. One milliliter of solution was placed on each of three plates containing another selective medium consisting of V-8 juice agar amended with the antibiotics pimaricin, rifamycin, and ampicillin and the fungicide pentachloronitrobenzene (James et al. 1990, 1996; Stone et al. 1995). Plates were incubated in the dark at about 24°C for 3 days. *Pythium* colonies were identified on

the basis of their diameter after 3 days (15-20 mm), feathery margin, and growth within rather than superficially on the agar surface. Populations were expressed as cfu/g of ovendried soil. Selected *Pythium* isolates were transferred to PDA for identification using the taxonomy of Waterhouse (1968)

*Brassica juncea* tissues were obtained immediately prior to incorporating the crop into soil by randomly selecting four plants from the center of each replicate plot. The plants were stored on ice for transport to the laboratory where they were frozen, freeze-dried, and ground to a fine powder. Glucosinolate analysis of plant tissues was performed using GC-MS (Gardiner et al. 1999).

All plots were sown during early May, 1997 with the same seedlot of Douglas-fir using standard procedures covered nurserv and with hydromulch. After complete seedling emergence (mid-July), three sampling sub-plots (0.5 m<sup>2</sup>) were installed within each replicate-plot; these subplots were located approximately equidistant from each other within the center of each replicate-plot. Seedling emergence and postemergence damping-off were determined in each sub-plot in July; selected damped-off seedlings were collected for laboratory analysis of associated pathogens. At the end of the first growing season (October 1997), seedling density and disease were determined within each sub-plot. Selected diseased seedlings were again collected for laboratory analysis, which included thoroughly washing roots and incubating them on Komada's medium and identifying associated organisms as described above.

At the end of the second growing season (November 1998), seedling density within each replicate-plot was determined using standard nursery density measurements (number of seedlings per 0.32 m with bed widths of 1.12 m and 7 rows of seedlings). After density estimates, sample seedlings were carefully from beds extracted for morphology measurements. Sample seedlings were located within inner seedling rows to eliminate edge effects. Fifty "average" seedlings were collected from each replicate plot. Seedlings were

transported to the laboratory for measurement. Seedling height (from basal cotyledon scar to the tip of the terminal bud), diameter (just above the groundline) and root mass (oven-dry weight of all roots) were determined for each sample seedling. Seedling heights, diameters, and root masses were compared among the six treatments with an analysis of variance. Significant differences (P=0.05) in these three morphology categories were located using the LSD procedure.

#### RESULTS

The only soil treatments that resulted in reduced populations of Fusarium by the time of sowing dazomet fumigation were and steam applications (table 1). Adding organic matter in the form of incorporated Brassica crops and composted sewage sludge resulted in extensive increases in Fusarium populations; such increases were extremely large in the Brassica green manure plots by the time of sowing. The vast majority of the Fusarium population in all plots was comprised of isolates of F. oxysporum Schlecht. (table 2). Six other Fusarium species [F. solani (Mart.) Appel & Wollenw., F. equiseti (Corda) Sacc., F. avenaceum (Fr.) Sacc., F. acuminatum Ell. & Ev., F. sambucinum Fuckel and F. culmorum (W.G. Smith) Sacc.] were isolated infrequently (table 2).

Soil populations of *Trichoderma* spp. were also greatly reduced by dazomet fumigation (table 3). *Trichoderma* spp. were lower in plots treated with *Brassica* green manure crops compared to pre-treatment levels. However, populations increased in fallowed plots and those treated with sewage sludge and steam. The ratios of *Trichoderma* to *Fusarium* populations indicated the most desirable values (highest numbers) for the fallow and steam treated plots (table 4). In general, the higher the ratio, the more potential for populations of *Trichoderma* to limit disease development by resident *Fusarium* populations. *Pythium* populations were greatly reduced by dazomet fumigation and somewhat reduced by steam treatment and the plastic tarping of a *Brassica* green manure crop (table 5). The only treatments that resulted in population levels of concern (near 100 cfu/g) by the time of sowing were the sewage sludge and *Brassica* green manure crop without tarping.

Glucosinolate concentrations in *B. juncea* tissue samples did not differ among the treatments (table 6). The dominant glucosinolate contained in plant tissues was 2-propenyl.

The lowest first-year seedling density was consistently found in plots treated with composted sewage sludge (table 7); much of the lack of seedling establishment may have been due to pre-emergence damping-off since little post-emergence mortality was noted in these plots. All the other treatments resulted in approximately equal 1-0 seedling densities; highest first-year mortality was obtained in plots treated with *Brassica* green manure crops (table 7).

The lower seedling density in composted sewage sludge treatments extended into the second seedling growing season (table 8). Highest seedling densities were found in the dazomet fumigated plots, but the other treatments, particularly the fallow, steam and *Brassica* green manure/no tarp treatments, were only slightly less.

Seedling size was directly proportional to density, i.e., low-density stands (composted sewage sludge treatment) resulted in significantly larger seedlings (table 9). Taller seedlings were also produced in dazomettreated plots; significantly taller seedlings were produced in fallow and steam-treated plots compared to those produced in some of the *Brassica* green manure plots. Table 1. Effects of selected pre-plant soil treatments on soil populations of *Fusarium* at the USDA Forest Service Nursery, Coeur d'Alene, Idaho.

Treatment	Colony-forming Units/g Oven-dry soil				
	Pre-	Freatment	Pos	t-Treatment	Pre-Sowing
Dazomet	287	[68-685]	7	[0-68]	13 [0-133]
Fallow/Cultivation	382	[68-819]	352	[68-819]	495 [67-1203]
Mustard/No Tarp	335	[68-755]	7583	[1489-19210]	10170 [5283-12572]
Mustard/Tarp	830	272-1904]	7579	[1912-17480]	9593 6413-12025]
Sewage Sludge	371	[275-412]	326	[68-952]	1244 [134-2675]
Steam	408	[204-749]	287	[409-2866]	280 [0-1069]

<sup>1</sup> Values in bold are means; ranges are in brackets; means are from 5 replicate plots per treatment.

Table 2. *Fusarium* species isolated from soil during assays of populations at the USDA Forest Service Nursery, Coeur d'Alene, Idaho.

· 清楚· 道:"是要要要	Number of Colonies Assayed					
Fusarium Species	Pre-Treatment	Post-Treatment	Pre-Sowing	All Samples		
F. oxysporum	253 [95.8]	1725 [97.4]	2408 [99.1]	4386 [98.2]		
F. solani	1 [0.4]	0	6 [0.2]	7 [0.2]		
F. equiseti	0	31 [1.7]	0	31 [0.7]		
F. avenaceum	9 [3.4]	11 [0.6]	0	20 [0.4]		
F. acuminatum	1 [0.4]	1 [0.1]	5 [0.2]	7 [0.2]		
F. sambucinum	0	3 [0.2]	5 [0.2]	8 [0.2]		
F. culmorum	0	0	6 [0.2]	6 [0.1]		
Totals	264	1771	2430	4465		

<sup>1</sup> Percentages are within brackets.

Table 3. Effects of selected pre-plant soil treatments on soil populations of *Trichoderma* at the USDA Forest Service Nursery, Coeur d'Alene, Idaho.

Treatment	Colony-forming Units/g Oven-dry soil <sup>1</sup>					
a 聖話の詩 「論」現為noon Palalana	Pre	-Treatment	Pos	t-Treatment	100 B	re-Sowing
Dazomet	2550	[953-6855]	14	[0-136]	7	[0-67]
Fallow/Cultivation	3429	[546-6494]	3904	[1422-6987]	6316	[1537-12028]
Mustard/No Tarp	1990	[1156-4395]	3085	[815-8349]	428	[67-1939]
Mustard/Tarp	1442	[748-3808]	464	[341-683]	508	[0-1269]
Sewage Sludge	2486	[1236-5287]	2584	[1224-3672]	3450	[1471-7757]
Steam	1606	[544-2927]	1515	[409-2866]	2044	[200-3406]

<sup>1</sup> Values in bold are means; ranges are in brackets; means are from 5 replicate plots per treatment.

Table 4. Effects of selected pre-plant soil treatments on ratios of *Trichoderma* to *Fusarium* populations at the USDA Forest Service Nursery, Coeur d'Alene, Idaho.

Treatment	Trichoderma/Fusarium Ratio <sup>1</sup>					
	Pre-Treatment	Post-Treatment	Pre-Sowing			
Dazomet	8.95	2.0	0.50			
Fallow/Cultivation	8.97	12.24	13.77			
Mustard/No Tarp	5.95	0.52	0.04			
Mustard/Tarp	1.74	0.06	0.05			
Sewage Sludge	6.70	7.93	2.77			
Steam	3.94	5.28	7.30			

<sup>1</sup> The higher the value the more potential the soil has for disease suppressiveness.

Table 5. Effects of selected pre-plant soil treatments on soil populations of *Pythium* at the USDA Forest Service Nursery, Coeur d'Alene, Idaho.

Treatment	Colony-forming Units/g Oven-dry soil <sup>1</sup>					
近风,从他都能	Pre-Treatment	Post-Treatment	Pre-Sowing			
Dazomet	<b>123</b> [68-212]	0 [0]	1 [0-13]			
Fallow/Cultivation	87 [55-164]	43 [7-224]	53 [0-140]			
Mustard/No Tarp	100 [14-163]	72 [0-285]	92 [0-260]			
Mustard/Tarp	107 [54184]	32 [7-55]	44 [0-80]			
Sewage Sludge	<b>117</b> [75-144]	53 [20-82]	103 [40-194]			
Steam	101 [75-136]	25 [7-61]	31 [0-67]			

<sup>1</sup> Values in bold are means; ranges are in brackets; means are from 5 replicate plots per treatment.

Table 6. Average glucosinolate concentrations in *B. juncea* plants used as a green manure crop at the USDA Forest Service Nursery, Coeur d'Alene, Idaho.

Glucosinolate	Tissue Concentration (µmol/g)		
2-propenyl	35.24		
3-butenyl	0.46		
Phenylethyl	0.50		
3-indolymethyl	0.11		
1-methoxy-3-indolymethyl	0.07		
4-OH-3-indolylmehtyl	0.12		
4-methoxy-3-indolymethyl	trace		

Table 7. Effects of selected pre-plant soil treatments on density and mortality of 1-0 Douglas-fir seedlings at the USDA Forest Service Nursery, Coeur d'Alene, Idaho.1

Treatment	Live Seedling Density	Dead Seedling Density
Dazomet	97 [75-145]	1 [0-5]
Fallow/Cultivation	92 [51-135]	3 [0-14]
Mustard/No Tarp	90 [61-123]	10 [0-19]
Mustard/Tarp	85 [41-108]	12 [0-8]
Sewage Sludge	55 [33-108]	3 [0-8]
Steam	88 [65-128]	1 [0-8]

Values in bold are means; ranges are in brackets. Density based on number of seedlings within subplots measuring 0.5m<sup>2</sup> located within each replicate plot.

Table 8. Effects of selected pre-plant soil treatments on density of 2-0 Douglas-fir seedlings at the USDA Forest Service Nursery, Coeur d'Alene, Idaho.1

Treatment	Live Seedling Density
Dazomet	28.4 [25.1-32.2]
Fallow/Cultivation	24.9 [21.3-28.6]
Mustard/No Tarp	25.4 [19.6-30.3]
Mustard/Tarp	21.6 [16.8-23.6]
Sewage Sludge	19.1 [14.1-26.1]
Steam	24.8 [22.7-26.9]

<sup>1</sup> Values in bold are means; ranges are in brackets. Density based on number of seedlings per 0.32m with bed widths of 1.12m and 7 rows of seedlings.

Table 9. Effects of selected pre-plant soil treatments on height, diameter, and root mass of 2-0 Douglasfir seedlings at the USDA Forest Service Nursery, Coeur d'Alene, Idaho<sup>1</sup>.

Treatment	Height2	Diameter3	Root Mass4
Dazomet	23.8 A	4.7 A	1.42 A
Fallow/Cultivation	21.7 B	4.6 A	1.48 A
Mustard/No Tarp	20.1 C	4.4 A	1.25 A
Mustard/Tarp	21.6 BC	4.6 A	1.44 A
Sewage Sludge	35.6 D	6.9 B	2.76 B
Steam	21.6 BC	4.8 A	1.48 A

Based on measuring 50 seedlings per replicated plot for each treatment. Means followed by the same capital letter are not significantly different (P=0.05) using LSD.

Measured from the groundline to the tip of the terminal bud (cm).
 Measured just above the groundline (mm).

<sup>4</sup>. Based on oven-dry weight of roots at lifting (g).

#### DISCUSSION

Preplant soil fumigation with wide-activity biocides used in forest seedling nurseries for many years. This has usually resulted in production of high-quality seedlings, but has also required continued reliance on this expensive procedure. Because fumigants are not selective in their target microorganisms (Boone 1988; Boyd 1971, James 1989), beneficial as well as detrimental organisms are equally affected. Re-invasion of fumigated soil by pathogens instead of saprophytes may result in greater disease severity than if no fumigation had been done (Marois et al. 1983; Vaartaja When fumigation is curtailed or 1967). terminated, it may take several years before soil is capable of producing high-quality seedling crops unless disease-suppressive amendments (composted organic amendments, biological control agents) are added (Gouin 1993; Papavizas 1985). Fallowing fields may reduce pathogen populations (Hamm and Hansen 1990; Hansen et al. 1990; James and Beall 2000; Stone et al. 1995), but will generally not eliminate them like chemical fumigation. The transition from routine pre-plant soil fumigation to non-fumigation may be difficult, particularly if fields cannot be fallowed for several years and/or the proper combination of organic amendments are not available.

Brassica spp. may help bridge the transition from fumigation to non-fumigation. Certain cultivars have shown high toxicity to common soilborne plant pathogens (Mayton et al. 1996) exhibit disease-suppressive and may characteristics under agricultural conditions (Chung et al. 2002; Mayton et al. 1996; Ramirez-Villapudua and Munnecke 1988). Glucosinolates in Brassica tissues are converted to isothiocvanates upon decomposition (Mayton et al. 1996; Rosa and Rodrigues 1999); these isothiocyanates may be toxic to certain soil high microorganisms, particularly at concentrations. However, populations of Fusarium spp. tend to increase, sometimes dramatically, whenever plant organic matter is added to soil (Hamm and Hansen 1990; Hansen et al. 1990; James et al. 1996; Wall 1984). Response to organic matter may outweigh any potential toxicity that may be introduced when Brassica crops are incorporated into soil (Hamm and Hansen 1990; Hansen et al. 1990). This apparently happened in the current evaluation. Fusarium populations increased dramatically after incorporation of the Brassica crop into soil. In addition, enough of this high population was comprised of pathogenic isolates to result it increased seedling mortality, particularly during the first growing season when most root disease mortality occurs (James 2001, 2002). By the end of the two-year crop cycle, seedling densities were less and seedlings somewhat shorter than those in fumigated plots. Therefore, it appeared that incorporating green Brassica juncea residues into soil as a biofumigant and added source of organic matter was not as effective as standard dazomet fumigation and did not improve seedling production. Perhaps other Brassica cultivars may perform better under forest nursery conditions and produce sufficient toxic metabolites to overcome stimulation of pathogen populations by the added organic matter.

We previously showed that 2-propenyl isothiocyanate was the most effective of six isothiocyanates tested against several forest nursery isolates of F. oxysporum (Smolinska et al. 2003), the most prevalent soilborne pathogen at the Coeur d'Alene Nursery (James et al. 1990, 1996). Bioassays conducted with 2propenyl isothiocyanate in volatile form inhibited mycelial growth and completely suppressed condial and chlamydospore germination of the pathogen. Therefore, a B. juncea variety with high 2-propenyl glucosinolates concentrations was selected for our studies. Lack of F. oxysporum control may have been due to inefficient release of isothiocyanate from decomposing plant tissues, a key step that requires extensive maceration of tissues and ample water (Morra and Kirkegaard 2002). Ensuring efficient isothiocyanate release from Brassica tissues is critical since organic carbon inputs without effective control of soilborne pathogens is common (Hansen et al. 1990; Hamm and Hansen 1990; James et al. 1996).

No beneficial effects were found when Trichoderma harzianum was added to some treatments. This fungus has been an effective suppressor of soilborne pathogens (Knudsen and Bin 1990; Knudsen et al. 1991) and should help reduce disease severity when incorporated into seedling management regimes. However, in our evaluation, it is possible that the large increases in potential pathogen populations resulting from incorporating organic matter into soil may have outweighed any potential benefit of adding T. harzianum inoculum. This biocontrol agent may have greater potential when applied next to sown seed, particularly in fields that have been fallowed or otherwise not amended with organic matter. Further tests should evaluate this potential.

Composted sewage sludge amendments resulted in greatly reduced seedbed densities, although surviving seedlings were much larger than those from other treatments. This type of compost has proven effective in reducing soilborne diseases in the past (Gouin 1993), although accumulation of soluble salts from such composts may result in phytotoxicity of some crops (Gouin 1977)

Steam treatment of soil has potential as an effective alternative to chemical fumigation (Karsky and Trent 2000). However, currently available application equipment requires high energy inputs and takes much too long to effectively treat sufficient volumes of soil for operational use. Further refinements will be necessary for steam applications to effectively compete with existing preplant chemical soil fumigation.

In conclusion, the various pre-plant soil treatments tested in this evaluation were not as effective as standard dazomet fumigation which, is routinely used at the Coeur d'Alene Nursery. Brassica crop amendments were generally ineffective in this test. Problems associated with Brassica crop production prior to incorporation into soil occurred. The crop grew quickly and produced mostly above-ground biomass and little root mass before it required incorporation into soil because of flowering. If greater root production had occurred, perhaps our results would have been different. Future tests should evaluate additional cultivars, including those produce higher levels known to of glucosinolates. Greater root development with less overall biomass production is desired to enhance the ability of Brassica crops to act as effective biofumigants.

#### LITERATURE CITED

- Angus, J.F., P.A. Gardner, J.A. Kirkegaard, J.M. Desmarchelier. 1994. Biofumigation: isothiocyanates released from Brassica roots inhibit growth of the take-all fungus. Plant and Soil 162:107-112.
- Boone, A.J. 1988. Soil fumigation in forest tree nurseries. Proceedings of the Southern Forest Nursery Association – 1988. pp. 33-38.
- Boyd, R.J. 1971. Effects of soil fumigation on production of conifer nursery stock at two northern Rocky Mountain nurseries. USDA Forest Service, Intermountain Forest & Range Experiment Station. Research Paper INT-91. 19p.
- Chapman, W. 1992. Alternative treatments to methyl bromide. Proceedings of the Southern Forest Nursery Association – 1992. pp. 96-103.
- Chung, W.C., J.W. Huang, H.C. Huang, J.F. Jen. 2002. Effect of ground Brassica seed meal on control of Rhizoctonia damping-off of cabbage. Canadian Journal of Plant Pathology 24:211-218.
- Fisher, N.L., L.W. Burgess, T.A. Toussoun, P.E. Nelson. 1982. Carnation leaves as a substrate and for preserving cultures of Fusarium species. Phytopathology 72:151-153.
- Gardiner, J.B., M.J. Morra, C.V. Eberlein, P.D. Brown, V. Borek. 1999. Allelochemicals released in soil following incorporation of rapeseed (Brassica napus) green manures. Journal of Agricultural and Food Chemistry 47:3837-3842.
- Gouin, F.R. 1977. Conifer tree seedling response to nursery soil amended with composted sewage sludge. HortScience 12:341-342.

- Gouin, F.R. 1993. Utilization of sewage sludge compost in horticulture. HortTechnology 3(2):161-163.
- Hansen, E.M., D.D. Myrold, P.B. Hamm. 1990. Effects of soil fumigation and cover crops on potential pathogens, microbial activity, nitrogen availability, and seedling quality in conifer nurseries. Phytopathology 80:698-704.
- Hamm, P.B., E.M. Hansen. 1990. Soil fumigation, cover cropping, and organic soil amendments: their effect on soil-borne pathogens and the target seedlings. In: Rose, R., S.J. Campbell and T.D. Landis (eds.). Target Seedling Symposium: Proceedings, Combined Meeting of the Western Forest Nursery Associations. USDA Forest Service, Rocky Mountain Experiment Station. General Technical Report RM-200. pp. 174-180.
- Ibarbia, E.Z. 1995. Fundamentals of fumigation. Greenhouse Grower 13(1):141-142, 144.
- James, R.L. 1989. Effects of fumigation on soil pathogens and beneficial microorganisms. In: Landis, T.D. (tech. coord.). Proceedings: Intermountain Forest Nursery Association Meeting. USDA Forest Service, Rocky Mountain Forest and Range Experiment Station. General Technical Report GTR-184. pp. 29-34.
- James, R.L. 2001. Root disease of 1-0 bareroot Douglas-fir seedlings – USDA Forest Service Lucky Peak Nursery, Boise, Idaho. USDA Forest Service, Northern Region, Forest Health Protection. Nursery Disease Notes #144. 10p.
- James, R.L. 2002. Root disease of 1-0 bare root ponderosa pine seedlings – Lone Peak Nursery, Draper, Utah. USDA Forest Service, Northern Region, Forest Health Protection. Nursery Disease Notes #147. 8p
- James, R.L., 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., 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, D.L. Wenny, 1991. Fusarium diseases of conifer seedlings In: Sutherland. J.R., S.G. Glover (eds.). Proceedings of the First Meeting of IUFRO Working Party S2.07-09 (Diseases and Forest Nurseries). Insects in Forestry Pacific Canada. and Yukon Region. Information Report BC-X-331. pp. 181-190.
- James, R.L., S. Metzger, C.J. Gilligan. 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, 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 96-6. 10p.
- Karsky, D., A. Trent. 2000. Nusery soil steam fumigation. USDA Forest Service, Technology & Development Program, Missoula, Montana. 16p.
- 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.
- Knudsen, G.R., L. Bin. 1990. Effects of temperature, soil moisture, and wheat bran on growth of Trichoderma harzianum from alginate pellets. Phytopathology 80:724-727.

- Knudsen, G.R., D.J. Eschew, L.M. Dandurand, Z.G. Wang. 1991. Method to enhance growth and sporulation of pelletized biocontrol fungi. Applied and Environmental Microbiology 57:2864-2867.
- Linderman, R., W. Dixon, S. Fraedrich, R.S. Smith, Jr. 1994. Alternatives to methyl bromide: assessment of research needs and priorities for forestry, nursery, and ornamental crops. Tree Planters' Notes 45:43-47.
- Marois, J.J., M.T. Dunn. G.C. Papavizas. 1983. Reinvasion of fumigated soil by Fusarium oxysporum f.sp. melonis. Phytopathology 73:680-684.
- Mayton, H.S., C. Oliviker, S.F. Vaughn, R. Loria. 1996. Correlation of fungicidal activity of Brassica species with allyl isothiocyanate production in macerated leaf tissue. Phytopathology 86:267-271.
- Miller, W.O., M.G. Norris. 1970. A new review of soil fumigation practices for use in forest nurseries. Down to Earth 26(3):9-12.
- Morra, M.J., J.A. Kirkegaard. 2002. Isothiocyanate release from soil-incorporated Brassica tissues. Soil Biology & Biochemistry 34:1683-1690.
- Nelson, P.E. T.A. Toussoun, 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. Annual Review of Phytopathology 23:23-54.
- Ramirez-Villapudua, J., D.E. Munnecke. 1988. Effect of solar heating and soil amendments of cruciferous residues on Fusarium oxysporum f.sp. conglutinans and other organisms. Phytopathology 78:289-295.
- Rosa, E.A.S., P.M.F. Rodrigues. 1999. Towards a more sustainable agriculture system: The

effect of glucosinolates on the control of soilborne diseases. Journal of Horticultural Science & Biotechnology 74:667-674.

- Shaheen, L. 1996. Potential loss of methyl bromide to prompt changes in Clean Air Act. Pest Control 64(5):68,74.
- Smith, R.S., Jr., R.V. Bega. 1966. Root disease control by fumigation in forest nurseries. Plant Disease Reporter 50:245-248.
- Smolinska, U., M.J. Morra, G.R. Knudsen, R.L. James. 2003. Isothiocyanates produced by Brassicaceae species as inhibitors of Fusarium oxysporum. Plant Disease 87:407-412.
- Stone, J.K., D.M. Hildebrand, R.L. James, S.M.
  Frankel, D.S. Germandt. 1995. Alternatives to methyl bromide for control of soil-borne diseases in bare root forest nurseries. In: Proceedings: Annual International Research Conference on Methyl Bromide Alternatives and Emissions Reductions. November 6-8, 1995, San Diego, CA. Methyl Bromide Alternatives Outreach, US Environmental Protection Agency and US Department of Agriculture. pp. 77-1 – 77-4.
- Vaartaja, O. 1967. Reinfestation of sterilized nursery seedbeds by fungi. Canadian Journal of Microbiology 13:771-776.
- Wall, R.E. 1984. Effects of recently incorporated organic amendments on damping-off of conifer seedlings. Plant Disease 68:59-60.
- Waterhouse, G.M. 1968. The genus Pythium Pringsheim. Commonwealth Mycological Institute, Kew, Surrey, England. Mycological Papers No. 110. 70p.
- World Meterological Association. 1995. Scientific assessment of ozone depletion: 1994 executive summary. Global Ozone Research and Monitoring Project Report No. 37. Global Ozone Observing System, Geneva, Switzerland. 36p.

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