## Performance of Outplanted Western Hemlock (*Tsuga heterophylla* (Raf.) Sarg.) Seedlings Inoculated With *Cenococcum geophilum*<sup>1</sup>

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In the first of two experiments, container-grown seedlings of western hemlock (Tsuga heterophylla (Raf.) Sarg.) inoculated with Cenococcum geophilum Fr. had significantly (P = 0.05) better top growth 2 years after outplanting than did noninoculated controls. Seedling survival was unaffected by mycorrhizal inoculation.

In the second experiment, western hemlock seedlings inoculated with Cenococcum -bearing rotten wood and noninoculated western hemlock seedlings planted in either rotten wood or mineral soil on a clearcut in western Oregon survived and grew equally well during the first two growing seasons. Survival was not affected by mycorrhizal inoculation but significantly (P = 0.05) fewer seedlings survived in rotten wood than in mineral soil because of the higher second-season mortality in rotten wood. (Tree Planters' Notes 36(4):13-16; 1985)

Seedling responses to mycorrhizal inoculation can vary with different combinations of site and fungi. For example, pines grew and survived better on coal spoils with *Pisolithus tinctorius* (Pers.) Coker & Couch than with other fungi (10). Increased growth and survival on less stressful sites are-sometimes obtained with other fungi; for example, pine seedlings inoculated with the mycorrhizal fungus *Paxillus involutus* (Batsch: Fr.) Fr. (7).

Inoculation with mycorrhizal fungi has occasionally decreased seedling growth. On certain soils, *Leccinum scabrum* (Bull.:Fr.) Gray decreases growth of birch (8). Shemakhanova (14) reports variable growth response of pine and oak seedlings to mycorrhizal inoculation. In some cases, the height of inoculated seedlings increased and in other cases it decreased when compared to that of noninoculated seedlings. Response varied with soil characteristics and with the fungus used.

Responses of western hemlock (*Tsuga heterophylla* (Raf.) Sarg.) seedlings to inoculation with ectomycorrhizal fungi have been little studied. Trappe (16) reports that dry weight of container-grown western hemlock increased by inoculation with several mycorrhizal fungi. No data have been published on the performance of mycorrhizal versus nonmycorrhizal western hemlock seedlings after planting.

Along the Pacific coast, western hemlock commonly regenerates in mature forests with heavy to moderate shade, on rotten logs or stumps rather than on the forest floor (3, 11). This species also regenerates on rotten wood in clearcuts (1) and can colonize brushy sites by establishing on the relatively brush-free wood residue (13).

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> MacBean (9) suggest that survival of germinants on rotten wood or thick humus requires a mild, moist sum mer climate.

Kropp (4) shows that nonmycorrhizal, container-grown western hemlock seedlings outplanted onto rotten wood or onto mineral soil survived equally well the first year. Some seedlings on rotten wood had less growth than did those on mineral soil, but all appeared vigorous and had formed mycorrhizae by the end of the first growing season. How western hemlock seed lings perform on rotten wood is important for foresters to learn, especially for sites on mineral soil where brush competition is severe.

Two experiments were designed to determine how mycorrhizal inoculation affects western hemlock survival and growth in plantations and whether inoculation affects seedling performance in mineral soil versus rotten wood.

## **Materials and Methods**

**Experiment 1.** Cenococcum geophilum (Isolate A-145) was isolated from surface-sterilized sclerotia (15), and vegetative inoculum was prepared as described by Molina (12).

Noninoculated, 1-year-old container-grown western hemlock seedlings were obtained from the Crown Zellerbach Corp. container nursery near Aurora, Oregon, in January 1978 and kept at 3 to 4 °C until inoculation in early March. The seedlings, originally grown in 98-cubic-centimeter plastic contain-

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ers, were transplanted during inoculation into 656-cubic-centimeter plastic containers filled with pasteurized peat and vermiculite (1:1). Seedlings were extracted from the original containers and placed (along with about 16 cubic centimeters of inoculum) into a hole pressed into the medium in the larger containers. Noninoculated controls were similarly transplanted but without inoculum.

Seedlings were grown in a lath house under natural light and temperature conditions until the following January, when they were placed in storage at 3 to 4 °C. In early April 1979, they were outplanted on six separate sites selected for a variety of aspect, slope, vegetation, and soil types. Three sites were in the moist coastal zone near Seaside. OR. at an elevation of 183 meters with an average annual precipitation of 200 to 250 centimeters. The remaining three sites were on Mary's Peak in the Siuslaw National Forest in western Oregon at an elevation of 183 meters with 178 centimeters of precipitation annually.

Three treatments of 30 seedlings each were planted in a completely randomized design within each plot. Treatments were a) noninoculated seedlings, b) well-colonized mycorrhizal seedlings (about 67 square centimeters of the plug surface was colonized), and c) moderately colonized mycorrhizal seedlings (about 32 square centimeters of the plug surface was colonized). After planting, each seedling was protected with plastic tubing to minimize animal damage.

At the end of the first and second years after outplanting, survival and current year's leader growth were measured for each seedling. An analysis of variance was done to test for treatment differences. Treatment means were separated by Tukey's test.

**Experiment 2.** The site, at an elevation of 870 meters on Mary's Peak in the Siuslaw National Forest, was originally a Douglas-fir stand. The stand was clearcut in winter 1977, and the slash was burned in autumn 1978. It receives an average annual precipitation of 178 centimeters, with 13 centimeters falling from May to August. The soil is a deep, well-drained gravelly loam in the Slickrock series.

Seedlings were grown in 98-cubic-centimeter molded polyethylene containers, which were surface-disinfected by submersion for 5 minutes in a 10-percent bleach solution. Containers were filled with a 3:1 mixture of peat vermiculite and fragmented rotten wood inoculum (S). Control seedlings were grown in peat vermiculite medium alone.

Seedlings were grown for 10 months in 1978 at the Crown Zellerbach container nursery near Aurora, Oregon, and throughout 1979 in a lath house under natural light and temperature at Oregon State University, Corvallis. In the nursery, nutrients were applied twice weekly at operational rates (weeks 4 to 5, 10-52-16 fertilizer at 625 grams per 1,000 liters; weeks 6 to 16, 20-20-20 fertilizer at 500 grams per 1,000 liters; weeks 17 to 18, 0-52-34 fertilizer at 625 grams per 1,000 liters; weeks 19 to 26, 10-52-16 fertilizer at 625 grams per 1,000 liters; ferrous sulfate at 155 grams per 1,000 liters was added occasionally to all seedlings). In the lath house, fertilizer was applied once every 2 weeks at 11.34 grams, 20-20-20 (N-P-K) soluble fertilizer and 2.8 grams Fe iron chelate micronutrient per square meter of surface area. Seedlings were selected for uniformity of size and mycorrhizal colonization and outplanted in five blocks in mid-April 1980. Each block had two plots: mineral soil and well-decayed rotten wood. Each plot contained 20 seedlings: 10 mycorrhizal and 10 noninoculated controls.

Early in October 1980, seedling survival was recorded and five inoculated and five noninoculated seedlings were randomly selected and carefully excavated both from rotten wood and mineral soil. The current year's leader growth and the average length of the five longest roots growing from each excavated plug were measured in centimeters. General observations on types of mycorrhizae were recorded.

After the 1981 season, survival of remaining seedlings was recorded and all seedlings were excavated for measurements as in 1980. An analysis of variance was performed to test for treatment differences. Differences between treatment means were separated by Tukey's test at P = 0.05.

## **Results and Discussion**

Bartlett's test of homogeneity revealed no differences among the three sites at the Seaside area and also among the three sites on the Mary's Peak area, so data were pooled for further analyses. Moderately colonized and well-colonized seedlings did not differ significantly in leader growth after 2 years, so data from these two categories were pooled for further analyses. In experiment 1, inoculated seedlings had significantly more leader growth after the second growing season than did noninoculated seedlings. Leader growth of all seedlings, regardless of mycorrhizal status, was significantly greater at Seaside than at Mary's Peak after 2 years (table 1). No significant area x treatment interactions were found, so overall differences between inoculated and noninoculated seedlings on both areas were due to inoculation rather than to site differences.

The cumulative difference in height growth between inoculated and noninoculated seedlings became greater from year 1 to year 2 (table 1). If differences in height growth between the two treatments continue to increase over time, growth benefits from inoculation could be more obvious much later after outplanting than soon after. This type of continued stimulation in height growth would be particularly relevant where brush competition or animal browsing impede establishment of plantations. **Table 1**—Leader growth and survival for inoculated and noninoculated western hemlock seedlings in the first 2 years after outplanting (experiment 1)<sup>1</sup>

Location	First year		Second year (cumulative)	
	Noninoculated mean	Inoculated mean	Noninoculated mean	Inoculated mean
Leader growth (cm)				
Mary's Peak	12.55a	14.23a	24.46a	26.86a
Seaside	23.16b	23.67b	52.46b	56.77b
Pooled mean	17.86A	18.95A	38.47A	41.82B
Survival (%)				
Mary's Peak	100.0a	98.3a	77.8a	80.7a
Seaside	100.0a	100.0a	96.6b	95.0b
Pooled mean	100.0A	99.2A	87.2A	87.9A

<sup>1</sup>Treatment means within sample years not sharing a common letter differ significantly by Tukey's test (P = 0.05).

Seedling survival in the first experiment was not affected by mycorrhizal inoculation but was significantly higher at Seaside than at Mary's Peak (table 1).

In experiment 2, leader and root growth did not differ significantly among treatments. Mycorrhizal inoculation did not improve survival on either rotten wood or mineral soil either year. Seedlings on rotten wood appeared more chlorotic and generally less vigorous at the end of the first season than did those on mineral soil, although neither growth nor survival differed significantly (P = 0.05). During the second growing season, mortality of seedlings on the rotten wood was significantly lower than that of seedlings on mineral soil (84 vs. 55 percent, respectively). Seedlings on rotten wood remained chlorotic except for those with occasional roots growing into underlying mineral soil; these were green and more vigorous than seedlings with roots confined to rotten wood.

Earlier work on western hemlock (4) indicated that nonmycorrhizal, container-grown seedlings planted in rotten wood survived as well as did those in mineral soil. Seedling root growth did not differ significantly between mineral soil or rotten wood; on one clearcut, however, seedling leader growth was significantly lower on rotten wood than on mineral soil. Berntsen (2) concluded that planting Douglas-fir (Pseudotsuga menziesii (Mirb.) Franco) or Sitka spruce (Picea sitchensis (Bong.) Carr.) on rotten wood in high rainfall areas is a sound practice. Results of our study, however, indicated that planting hemlock in rotten wood on open sites cannot be recommended.

Our observations of mycorrhizae on roots of excavated seedlings in

experiment 2 showed that *Cenococcum* geophilum colonized many new roots growing out from inoculated seedlings in both rotten wood and mineral soil. Numerous fungi shown to be mycorrhizal with western hemlock (6) need to be compared with *C.* geophilum to determine whether they can improve seedling survival or more strikingly stimulate height growth.

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