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# Use of soil transfer for reforestation on abandoned mined lands in Alaska

## II. Effects of soil transfers from different successional stages on growth and mycorrhizal formation by *Populus balsamifera* and *Alnus crispa*

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**Abstract.** Soil transfers from an intermediate successional site and a mature forest site were applied to *Populus balsamifera* L. cuttings and *Alnus crispa* (Ait.) Pursh seedlings placed on an abandoned mined site in south central Alaska to improve plant establishment. Mycorrhizal fungi in the soil transfers from the two successional stages were hypothesized to have different effects on plant species that colonize disturbed sites at different times or on different substrates. The site consisted of coarse, dry, low-nutrient spoils and was naturally colonized by scattered *P. balsamifera* but not *A. crispa*, although seed sources for both were adjacent to the site. Physical dimensions of the transplanted seedlings and cuttings were measured at the beginning and end of each growing season. Selected plants were harvested at the end of the 2-year study and examined for mycorrhizal formation, current growth, and leaf tissue nutrient concentrations. Both plant species were taller when treated with the soil transfers from the mature forest than with soils from the intermediate site although the increase for *A. crispa* was greater. Physical dimensions, current growth, and nutrient concentrations were greater when *A. crispa* was treated with the mature soil transfer compared with the intermediate soil transfer. Mycorrhizae which infected *Alnus* were predominantly a brown woody type, while other types accounted for greater relative mycorrhizal infection percentage on *Populus*. Insufficient quantities of mycorrhizal inoculum of suitable species, as well as low moisture and low nutrient conditions, may be factors limiting *A. crispa* colonization on primary disturbed sites in south central Alaska.

**Key words:** Soil transfer - Reforestation - *Populus* - *Alnus* - Boreal forest

### Introduction

Mycorrhizae and soil amendments may be needed to insure rapid establishment of tree seedlings to revegetate disturbed sites (Bjorkman 1970; Mikola 1973; Amaranthus and Perry 1987; Perry et al. 1987). Growth responses and mycorrhizal status of plants on disturbed sites can be improved by transferring soil containing mycorrhizal inoculum from adjacent vegetation communities to the rooting zone of transplanted cuttings or seedlings (Amaranthus and Perry 1987, 1989; Helm and Carling 1993). This soil transfer is presumed to contain indigenous fungi adapted to local environmental conditions, although not necessarily to conditions specific to the disturbed site. It may also contain other microorganisms that may facilitate mycorrhizal formation and improve plant growth.

The choice of native vegetation sites from which to collect soil transfers for placement in the rooting zone of young plants placed on disturbed sites may be critical. Site selection depends at least partially on the species composition of the plant community established on the soil to be transferred (Amaranthus and Perry 1989). The effectiveness of the soil transfer may also depend on the successional status of the vegetation, since the quantity and species of mycorrhizal fungi change with plant succession (Reeves et al. 1979; Janos 1980 a, b; Last et al. 1984, 1987; Amaranthus and Perry 1989).

Early successional communities are expected to have the mycorrhizal fungi most appropriate for revegetation of disturbed lands (Danielson and Visser 1988; Marx and Cordell 1988). In fact, mycorrhizal fungi have been classified according to early-, multi-, or late-stage based on whether they infect plants during early, multiple, or late stages of succession (Danielson and Visser 1988). An early-stage fungus such as *Pisolithus tinctorius* can readily form mycorrhizae with the roots of conifer seedlings in the southeastern United States (Marx and Cordell 1988). However, late-stage fungi may be unable to infect roots in an early stage of development (Danielson and Visser 1988; Marx and Cordell 1988).

Mycorrhizal fungi on roots of individual plants also change over time. Succession of ectomycorrhizal (ECM) fungi on *Betula pendula* has been inferred by observations of fruiting bodies (Deacon et al. 1983; Dighton et al. 1986; Fleming 1983; Mason et al. 1983; Last et al.

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1984; Fleming et al. 1986; Last et al. 1987). Litter that accumulates during plant community development is believed to affect mycorrhizal composition (Rose et al. 1983; Last et al. 1987).

The growth of some plant species is improved when infected with certain species of mycorrhizal fungi but not with other mycorrhizal fungal species (Molina and Chamard 1983). However, only a few fungal species form ECM on *Alnus rubra* (Molina 1979; Molina and Trappe 1982). In contrast, some species of *Populus* can be both VAM (vesicular-arbuscular mycorrhizal) and ECM while other species can be VAM or ECM (Vozzo and Hacskaylo 1974; Rothwell and Vogel 1982; Lodge and Wentworth 1990). ECM fungi can displace VAM fungi from *Populus deltoides* Bartr. under certain moisture conditions (Lodge and Wentworth 1990). Whether *Populus* and *Alnus* would more likely form VAM or ECM when planted in or inoculated with soil transfers from early or late successional communities may be important in determining the rate of succession or the pathways that may be followed.

Species of *Populus* and *Alnus* are common colonizers of disturbed lands in the boreal forest zone but differ in terms of the type or the time since disturbance of areas they colonize. *Populus balsamifera* L. is one of the most important colonizing species on dry disturbed sites, steep slopes, and river floodplains in south central Alaska. Species such as *P. balsamifera* and *Salix alaxensis* (Anderss.) Cov. may colonize during the first year after the disturbance (Helm et al. 1985). Species of *Alnus* more commonly colonize secondary disturbances in south central and interior Alaska. *A. tenuifolia* Nutt. does not usually appear until the second wave of colonizers on river floodplains (Helm et al. 1985; Grubb 1986; Walker et al. 1986) and on recently deglaciated areas. *Alnus* is rarely found on dry, coarse soils or steep slopes that characterize many disturbed lands. *Alnus* requires greater moisture levels (Mitchell and Mitchell 1980) or possibly some additional factors associated with more developed soils. Greater densities of appropriate species of mycorrhizal fungi present in developed soils may be important for *Alnus* establishment on a site. Danielson and Visser (1988) reported that *Alnus* seedlings became nodulated within 2 year of outplanting while mycorrhizal colonization was still low after 2 years in soils reconstructed from oil sands and peat.

Amaranthus amd Perry (1989) have demonstrated that some soil transfers are more effective than others when used as mycorrhizal inoculum on *Pseudotsuga menziesii* in Oregon. These results depended on the plant species present. Our objective was to determine whether the successional stage of the soil transfer being used as a mycorrhizal inoculum (soil transfer) affected growth and mycorrhizal responses of plant species. *P. balsamifera* and *A. crispa* were selected since they become established at different times during succession.

## Materials and methods

### *Study area*

The study area consisted of overburden on coal-mined lands near Jonesville, Alaska (61°44'N 148°56'W) that were abandoned over 20 years ago. Scattered individuals of young *P. balsamifera* and *Betula papyrifera* Marsh. (paper birch) had naturally colonized the mine spoil although many of these plants were stunted. *A. crispa* had not colonized the site but it did occur in the surrounding draws where moisture was greater. Seed sources were abundant adjacent to the study plots. The intermediate successional vegetation community selected as one source for soil transfer was located adjacent to the plots and consisted of *P. balsamifera* and *A. crispa* with *Equisetum* spp. (horsetails) in the herbaceous layer. This is the same source used in Helm and Carling (1993) and will be referred to as "intermediate soil transfer."

Mature forest communities in the area were dominated by combinations of *B. papyrifera, P. balsamifera,* and *P. tremuloides* Michx. (aspen). Understory plant species included *A. crispa* and *Calamagrostis canadensis* (Michx.) Beauv. (bluejoint reedgrass), *Heracleum lanatum* Michx. (cow parsnip), *Echinopanax horridum* [J. E. Smith] Decne. & Planch (devil's club), and *Viburnum edule* (Michx.) Raf. (highbush cranberry). Soil representative of a later successional stage was collected from this mature forest and will be referred to as the "mature soil transfer." Individuals of *A. crispa* were scattered throughout both stands, so differences in density of *Alnus* were not expected to be a major factor. Both sites were selected after a preliminary survey of local plants for mycorrhizal infection (Helm and Carling 1990).

### *Plant material preparation and preliminary surveys*

Rooted cuttings of *P. balsamifera* were prepared as described in Helm and Carling (1993). The *Alnus* seeds were obtained from the Alaska Division of Forestry Nursery and started on 21 April 1988. Germinaton was 60% at room temperature (21°C). Seeds were planted in a sterilized mixture of coarse sand and potting soil on 3 May. Seedlings and cuttings were hardened outdoors for at least 1 week before planting on 13 July, 1988.

Spoil samples were analyzed the same as in Helm and Carling (1993) for the following parameters: extractable $NH_4^+$, $NO_3^-$ (2-N KCl); total N (digestion in $H_2SO_4$-$H_2SeO_3$); extractable P (Olsen's); total P (digestion in $H_2SO_4$-$H_2SeO_3$); K, Ca, Mg, Na (Mehlich 3; $NH_4OAc$) cation exchange capacity ($NH_4OAc$); organic carbon (Walkley-Black); and pH (1:1 and paste with water). Roots were collected from the proposed sources for the soil transfer to verify presence of mycorrhizal fungi.

The base levels of fertilizer applied were based on pre-experiment spoil sampling described in Helm and Carling (1993). Because of the low levels of N and P in the overburden, it was assumed that a base rate of fertilizer would be applied in any commercial reclamation program.

The experimental design consisted of a randomized factorial complete block design with two treatments at two levels each replicated three times: plant species (*P. balsamifera, A. crispa*) and inoculum source (intermediate successional soil transfer, mature forest soil transfer). Plots were sized 2 m × 5 m and were arranged in two columns such that the four plots within a block filled an area 4 m × 10 m. Treatments were randomly assigned to each plot.

### *Soil transfer and planting*

Soil transfers were collected from the top 5–10 cm of soil in the feeder-root zone of the intermediate successional stand and from the mature forest. The presence of spores, hyphae, and infected

root fragments in these soil transfers was confirmed during a preliminary survey and an analysis of the soil transfer used in the field. Collected soils were homogenized by hand and applied the same day.

A 250-ml subsample of soil including roots was obtained from the soil transfer sample. Roots in the sample were washed, oven-dried, and weighed to estimate the root mass in a soil transfer unit. Root mass combined with infection levels were used to indicate mycorrhizal inoculum density.

Spacing between outplanted individuals was 70 cm, a spacing equivalent to the average distance between volunteer plants on the site. Each of the ten *P. balsamifera* and nine *A. crispa* individuals per plot were treated with 250 ml of soil transfer and 100 mg N/kg spoil and 100 mg P/kg spoil.

### *Estimates of plant growth*

Field measurements included plant height, crown length and width, basal stem diameter, and twig length and diameter (Helm and Carling 1990, 1993). Crown length and width were combined into an area estimate using the formula for an ellipse. Categorical observations were made for vigor and phenology. Measurements were made at the beginning and end of each of the two growing seasons. Plant heights were measured to the top of the highest living point on the plant.

An average of three plants per plot were harvested at the end of year 2 to estimate levels of mycorrhizal formation on roots, leaf and twig growth, and leaf nutrient concentrations. Current twig and leaf biomass were separated from each plant, oven-dried, and weighed. Leaves from each plot were composited to determine the nutrient concentration of N, P, K, Ca, and Mg in a $H_2SO_4$-$H_2SeO_3$ digestion.

Ectomycorrhizal formation levels were determined by counting root tips in the following categories: white wooly, white smooth, tan, brown wooly, black smooth, or not infected according to Zak's (1973) classification. Estimates of VAM and ECM formation were obtained by examining roots cleared and stained by methods of Phillips and Hayman (1970) and Kormanik et al. (1980) and described by Helm and Carling (1993). VAM spores were separated from the rhizosphere soil using differential centrifugation (Allen et al. 1979) and counted on a gridded petri dish under a dissecting microscope (×40).

### *Statistical analysis*

All quantitative parameters (plant height, crown area, basal diameter, twig length and diameter, biomass, nutrient concentrations) measured at the end of the 2-year study were analyzed using analysis of variance with a blocking factor. Field estimates of size parameters were analyzed with initial height as a covariate. Physical dimensions (height, crown, and twig dimensions) were analyzed separately for the two species because of the inherent differences in initial sizes for rooted *Populus* cuttings and *Alnus* seedlings. Quantitative data were analyzed using a general linear model program (GLM), while categorical data (vigor, VAM infection) were analyzed using a categorial modeling program (CATMOD) (SAS 1985). Linear contrasts were used to test specfic comparisons. Type III hypothesis testing was used.

## Results

Spoils on this experimental site contained 52% gravel and had a pH of 8.3. Levels of extractable nutrients were low: 4 mg/kg $NH_4$-N, <0.5 mg/kg $NO_3$-N, P<2 mg/kg, K 77 mg/kg, Ca 1330 mg/kg, Mg 563 mg/kg, and Na 22 mg/kg. Cation exchange capacity was 10 meq/100 g and organic carbon 2.16%.

Soil transfer from the mature site contained 0.49±0.08 g (standard error) oven-dried roots per 250 ml, while soil transfers from the intermediate site contained 0.41±0.16 g roots per 250 ml soil. These sources had been evaluated for mycorrhizal formation in an initial study in June 1988 (Helm and Carling 1990).

Survival of plants over the first 2 years averaged nearly 90% for the *P. balsamifera* and 80% for the *A. crispa*. Survival of *P. balsamifera* was not affected by the soil-transfer source, but more *A. crispa* survived on the plots treated with the intermediate soil transfer (90% survival) compared with plants treated with the mature soil transfer (70% survival) ($P<0.09$). However, the surviving *A. crispa* were more vigorous when mature soil transfer was used compared with the intermediate soil transfer ($P<0.06$).

Plant heights of both *P. balsamifera* ($P<0.05$) and *A. crispa* ($P<0.01$) were significantly greater after two growing seasons when treated with the mature soil transfer compared with the intermediate soil transfer (Table 1). *P. balsamifera* was almost 25% taller and *A. crispa* almost 30% taller when treated with the mature soil transfer compared to the intermediate soil transfer.

**Table 1.** Above-ground dimensions and current growth of *Populus balsamifera* and *Alnus crispa* when treated with soils from different successional stages at end of year 2, August 1989. SE, Standard error of the mean: values in parentheses at the top of each column are the probabilities that the means do not differ between soil-transfer sources within each plant species. NA, Not available: twig lengths were not measured on *A. crispa* because of the relatively large number of twigs

| Soil transfer source | *n* | Plant height (cm) | | Crown area ($cm^2$) | | Twig length (cm) | | Leaf mass (g) | | Twig mass (g) | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | | Mean | SE | Mean | SE | Mean | SE | Mean | SE | Mean | SE |
| *P. balsamifera* | | | (0.05) | | (0.97) | | (0.25) | | (0.47) | | (0.77) |
| Intermediate | 25 | 28.9 | 1.8 | 799.7 | 81.6 | 7.7 | 1.4 | 2.43 | 0.41 | 0.71 | 0.17 |
| Mature | 23 | 35.2 | 2.9 | 753.1 | 103.2 | 9.6 | 1.4 | 1.93 | 0.50 | 0.78 | 0.24 |
| *A. crispa* | | | (0.01) | | (0.21) | | | | (0.01) | | (0.03) |
| Intermediate | 25 | 28.0 | 2.3 | 951.4 | 152.2 | NA | | 2.87 | 0.50 | 0.92 | 0.26 |
| Mature | 19 | 40.2 | 2.7 | 1478.3 | 331.3 | NA | | 5.52 | 0.90 | 2.10 | 0.45 |

**Table 2.** Concentrations (%) of nutrients in leaf tissues of *P. balsamifera* and *A. crispa* when treated with soils from different successional stages at end of year 2, August 1989

| Soil transfer source | N | | P | | K | | Ca | | Mg | |
|---|---|---|---|---|---|---|---|---|---|---|
| | Mean | SE | Mean | SE | Mean | SE | Mean | SE | Mean | SE |
| *P. balsamifera* | | (0.60) | | (0.25) | | (0.46) | | (0.91) | | (0.89) |
| Intermediate | 1.28 | 0.07 | 0.14 | 0.00 | 0.65 | 0.04 | 1.17 | 0.06 | 1.10 | 0.04 |
| Mature | 1.23 | 0.07 | 0.15 | 0.01 | 0.71 | 0.06 | 1.18 | 0.07 | 1.09 | 0.05 |
| *A. crispa* | | (0.01) | | (0.08) | | (0.83) | | (0.48) | | (0.01) |
| Intermediate | 2.04 | 0.08 | 0.16 | 0.01 | 0.72 | 0.04 | 0.75 | 0.04 | 0.60 | 0.01 |
| Mature | 2.44 | 0.04 | 0.20 | 0.01 | 0.73 | 0.02 | 0.80 | 0.06 | 0.70 | 0.02 |

Crown areas were not affected by the source of the inoculum (Table 1). Basal diameters of *P. balsamifera* averaged 7.9 mm and were slightly greater for plants treated with the soil transfer from the mature forest ($P<0.10$). Basal diameters for *A. crispa* were sligthly greater at the end of two growing seasons when the seedlings were treated with the mature soil transfer (4.9 versus 4.1 mm, $P<0.07$).

Current leaf ($P<0.08$) and twig biomass ($P<0.04$) as well as first year twig production ($P<0.007$) (Helm and Carling 1990) were almost twice as great for *A. crispa* when treated with mature soil transfer than when treated with the intermediate soil transfer (Table 2). Twig and leaf growth by *P. balsamifera* did not differ when treated with the two different soil transfers.

*P. balsamifera* root system dimensions did not vary in response to inoculum sources but averaged 19.4 cm in depth. However, the width of the root system of *A. crispa* was greater when treated with the mature soil transfer (13.9 cm) compared with root systems of *A. crispa* treated with intermediate soil transfer (10.6 cm) ($P<0.01$). Depths of roots of *A. crispa* averaged 17.9 cm.

One of the largest responses to soil-transfer source was the leaf nutrient concentrations for *A. crispa* although similar differences did not occur for *P. balsamifera*. The plant species were expected to have different nutrient compositions because *A. crispa* is an N-fixing plant species. Concentrations of N ($P<0.0001$) and P ($P<0.002$) were greater in *A. crispa* leaves than in *P. balsamifera* while leaf concentrations of Ca and Mg were greater in *P. balsamifera* ($P<0.0001$) compared to *A. crispa* (Table 2). Potassium levels did not vary with species or source of soil transfer (Table 2). Nitrogen ($P<0.003$) and phosphorus ($P<0.02$) concentrations were about 20% greater in *A. crispa* treated with mature soil transfer compared with intermediate soil transfer (Table 2). Concentrations of K and Ca did not differ in response to the source of the soil transfer. Foliar Mg concentrations were greater in *Alnus* when the mature soil transfer was used (Table 2).

Soil-transfer source did not affect phenological development in either species. However, *A. crispa* broke bud earlier than *P. balsamifera* in the spring (Helm and Carling 1990).

More ECM formed on *A. crispa* than on *P. balsamifera* ($P<0.0001$), but the soil-transfer source did not affect the total percentage of mycorrhizal tips for either plant species (Fig. 1). Evenness of mycorrhizal types, based on surface morphological characteristics, was greater on *P. balsamifera* than on *A. crispa*. A brown wooly type of ECM fungus dominated the *Alnus* roots while several types (brown wooly, white wooly, and tan types) of mycorrhizae were about equally represented on the *P. balsamifera* roots (Fig. 1). The white wooly type was about three times as abundant on the *P. balsamifera* compared with the *A. crispa* ($P<0.02$). The brown wooly mycorrhizae were about four times as prevalent on *A. crispa* compared with *P. balsamifera* ($P<0.0001$). Differing morphological characteristics do not necessarily indicate different fungal species. In particular, some light-colored mycorrhizae may become dark-colored with age.

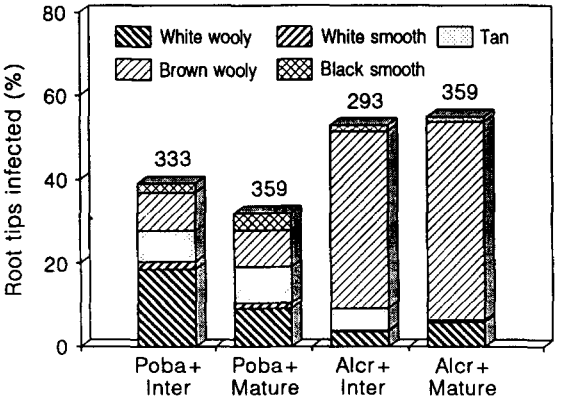


**Fig. 1.** Percent of root tips of *Populus balsamifera* cuttings (*Poba*) and *Alnus crispa* seedlings (*Alcr*) infected by ectomycorrhizae when treated with soil from different successional stages. *Inter*, Soil transferred from intermediate successional vegetation dominated by *P. balsamifera* and *A. crispa*. *Mature*, Soil transferred from soil under mature forest containing *P. balsamifera, Betula papyrifera, P. tremuloides*, and *A. crispa*. *Numbers* above each bar are mean number of root tips examined per plant. Standard error of the mean for percent tips infected was 5% for *P. balsamifera* and 4% for *A. crispa*

Both VAM and ECM fungi were found on both plant species. VAM infection was greater with the intermediate soil transfer compared with the mature soil transfer for *P. balsamifera* ($P>0.09$), but the source of soil transfer made no difference to VAM infection levels for *A. crispa* ($P>0.37$) (Fig. 2). However, *A.crispa* tended to have more root segments with some VAM infection

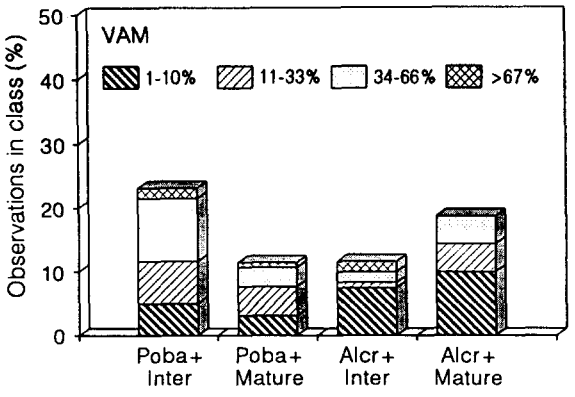


**Fig. 2.** Percent of observations in each infection percentage class for VAM in harevested plants at end of year 2. Estimates were made on stained samples using a compound microscope, at magnification ×100

with the mature soil transfer compared with the intermediate soil transfer ($P<0.15$). When an analysis was performed for presence or absence of VAM infection on both species, neither the species ($P>0.62$) nor the soil-transfer source ($P>0.59$) main effects were significant. However, the interaction of species and soil-transfer source was significant ($P<0.007$). Most microscope fields of *A. crispa* roots contained low levels (category 1, 1–10%) of VAM infection if they were infected at all (Fig. 2).

The number of spores extracted ranged from 0.1 to 4.5 spores/g soil transfer and averaged 1.5 spores/g soil transfer. The number of spores did not differ between inocula for *P. balsamifera*. Numbers of VAM spores collected from the rooting zone of the *A. crispa* plants were also very variable, but tended to be higher than those of the *P. balsamifera*. Values ranged from 0.2 to 34.7 spores/g soil transfer and averaged 3.3 spores/g soil transfer. Most *A. crispa* root systems contained fewer than 10 spores/g soil transfer.

## Discussion

*P. balsamifera* and *A. crispa* represent species that occur under different successional conditions. These two species responded differently to the two soil transfers from different successional stages. Although height for both species was greatest with the soil transfer from the mature vegetation, the increase for *A. crispa* was greater than that for *P. balsamifera*. Twig and leaf masses increased for *A. crispa* treated with mature soil transfer whereas no significant increase occurred for *P. balsamifera*. *P. balsamifera* is one of the first colonizers in primary succession on floodplains, glacial outwash plains, and mined lands in the boreal forest zone of south central Alaska. *A. crispa* colonizes sites somewhat later and seems more dependent on adequate moisture or factors associated with more developed soil. Prior to the experiment, the intermediate successional soil transfer was expected to provide the best results overall because it would have more fungi appropriate for colonizing disturbed areas. However, the soil transfer from the mature forest improved height growth of both species, especially *A. crispa*, compared with the intermediate soil transfer. Biomass of roots, a major criterion for determining mycorrhizal fungal inoculum density in the soil transfers, was not greatly different for the two soil transfers.

Past studies have shown that plant responses to inoculation depedend on the soil-transfer source. Different growth responses for two plant species were reported on four overburden types in Alberta (Danielson et al. 1983). ECM developed evenly in all four, while VAM developed slowly in peat (Danielson et al. 1983). Medve et al. (1977) also reported different responses to different inocula. The source of soil transfer resulted in different growth responses of *Pseudotsuga menziesii* (Schoenberger and Perry 1982; Pilz and Perry 1984; Amaranthus and Perry 1987, 1989). Specifically, Amaranthus and Perry (1989) demonstrated different responses of *P. menziesii* to soil transfers from the same soil series but different forest types.

Most plant growth measurements of *A. crispa* were greater on plants treated with the mature soil transfer compared to those treated with the intermediate soil transfer. Ectomycorrhizal formation percentages varied with the plant species, but not the soil-transfer source. A brown wooly ECM type dominated on *A. crispa* roots. A white wooly type of ECM was more common on the *P. balsamifera* roots, which also had a more even dispersal of ECM types.

Responses of the plant species to the soils transferred from different successional stages could have resulted from physical, chemical, or biological properties of the soils or interactions. Most likely, interactions among several factors were responsible for these differences. If the increased growth resulted only from greater nutrient levels, then growth of *Populus* rather than *Alnus* would have increased more in response to greater soil transfer nutrient levels as it has in other studies (Chapin et al. 1983, 1986; Heilman 1990). *P. balsamifera* is one of the fastest growing boreal forest tree species on high-nutrient sites. Chapin et al. (1983) attributed different responses of *A. crispa* and *P. balsamifera* to P as adaptations to their normal environments. *P. balsamifera* dominates early successional floodplain sites and south-facing upland sites where higher temperatures increase nutrient mineralization and availability. *Alnus* spp. occurs on cooler upland sites where mineralization is lower. Increased water-holding capacity associated with the soil transfers might be expected to favor *A. crispa* slightly. It is believed that biological properties may be at least partially responsible.

Interactions of these biological, chemical, and physical properties may have been responsible for the different results. Part of the reason for the different responses among species may be attributable to the brown wooly mycorrhiza dominating *A. crispa* compared with the more even distribution of types on *P. balsamifera*. The brown wooly type may have been more efficient at absorbing nutrients from the soil transferred from the mature forest community, although this is speculation.

Since total percentage infection of ECM formation did not differ between the two soil-transfer sources, biological factors besides mycorrhizae, such as *Frankia,* may have played a role. However, Danielson and Visser (1988) found nodules, but frequently no mycorrhizae, on *Alnus* growing in nurseries in Alberta. *Alnus* seedlings were nodulated within 1 year of outplanting on soils reconstructed from sands and peat, but mycorrhizal infection was still low during the second year (Danielson and Visser 1988). This would imply that infection by mycorrhizal fungi may be more difficult to achieve than nodule formation by the bacteria *Frankia.* An established stand of *A. crispa* was about 15 m uphill from the plots.

The presence of relatively few types of ECM on *Alnus* and more types on *Populus* is consistent with past studies. *A. rubra* and probably other *Alnus* species are colonized by only a few ECM fungal species (Molina 1979; Danielson and Visser 1988) even though species within the *Alnus* genus may be both VAM and ECM. Most plants in our study had substantial numbers of nodules although nodules were not counted.

*Populus*, on the other hand, had more types of ECM, which was also consistent with previous studies. Different clones of hybrid *Populus* had different levels of mycorrhizal colonization and different fungal species colonized from different inocula (Schultz et al. 1983).

Inoculation with early-stage fungi or soil transfer from early successional stages has been recommended for plants being outplanted on disturbed sites (Marx and Cordell 1988). Later successional sites contain more litter and different plant and fungal species (Last et al. 1987). Results of our study indicated better responses to the more mature soil transfer, especially when the later-successional plant species was treated. Whether the fungal species were early-, multi-, or late-successional is not known. Both *P. balsamifera* and *A. crispa* grew naturally on both soils transferred to the disturbed site so mycorrhizal fungi suitable to the plant species should have been present. Densities of woody plants were not measured but *P. balsamifera* had more stems per area and more cover on the intermediate site than in the mature forest. *A. crispa* occurred as scattered individuals on both sites.

Nitrogen concentrations and growth may have been affected by $N_2$ fixation by bacteria such as *Frankia* which form nodules on *Alnus* roots and/or by free-living bacteria found in ectomycorrhizae such as reported by Amaranthus et al. (1990). Bacteria have also been reported to either enhance or suppress mycorrhizal formation (Bowen and Theodorou 1979).

*Alnus* is not usually a colonizer of primary successional sites during the earliest years in the boreal forest zone of Alaska (Helm et al. 1985; Grubb 1986; Walker et al. 1986), but is a rapid colonizer of secondary successional sites where soil is available. Lack of early colonization by *Alnus* on floodplains has usually been attributed to life history traits such as seed dispersal distance (seed size is large compared with that of Salicaceae family) and timing of dispersal (winter) (Helm et al. 1985; Walker and Chapin 1986; Walker et al. 1986). Grubb (1986) has stressed that few $N_2$-fixing plant species occur among the early colonists but are usually in the second or third wave.

However, these reasons do not help explain why *Alnus* is not colonizing abandoned mined lands but does rapidly colonize secondary disturbances where developed soil is available. One explanation is that low soil moisture levels may limit *Alnus* establishment (McVean 1956; Mitchell and Mitchell 1980). *Alnus* has colonized naturally the depressed areas adjacent to the study plots. *Alnus* spp. do appear to be among the earlist colonizers on recently deglaciated terrain in coastal Alaska where *P. balsamifera* was not present and precipitation levels were greater (personal observation). Another explanation may be the lack of sufficient, appropriate mycorrhizal inoculum. This inoculum may be present on secondary disturbances or may accumulate in the first few years of primary succession. Ectomycorrhizal development on *Alnus* roots may help reduce the nutrient and moisture stress on these sites. Lack of *Frankia* may also reduce establishment although mycorrhizae appear to be more difficult to form than nodules based on results of Danielson and Visser (1988). Sites with secondary disturbances may have the appropriate combinations of microorganisms and soil conditions that permit *Alnus* to develop there but not on dry, primary successional sites. Hence, other organisms may facilitate the establishment of *Alnus* and it, in turn, can facilitate the establishment of other plant species through additions of N to the soils.

To the best of our knowledge, this was the first field study involving comparisons of effects of soil transfers from different successional stages on plant species with different functions during colonization of disturbed sites in the boreal forest zone of Alaska. We need to understand better the functions of microorganisms, plant species, successional stage, and environmental conditions to help select the most appropriate soils for transfer to improve establishment on disturbed sites, such as mined lands. Similar studies have been performed in Oregon (Amaranthus and Perry 1987, 1989) and Alberta (Danielson and Visser 1988, 1989). These studies have also confirmed the need to correctly identify soil-transfer source for each plant species. At least three characteristics apparently must be considered when selecting a site for collection of soil to be transferred: (1) the plant species on the site to be used as a soil-transfer source, (2) the successional stage of the vegetation on that site, and (3) the plant species to be inoculated. We need to better understand the mechanisms by which soil transfers improve growth of certain plant species to select soil transfers that are best suited to specific combinations of plant species and site conditions.

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