

Salal Cedar Hemlock Integrated Research Program

Research Update #1: December 1996

Edited by

C.E. Prescott

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Cover photo: 14-year-old plantation of western red cedar on a cedar-hemlock (CH) cutover 2 years after application of fish-wood compost.



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SALAL CEDAR HEMLOCK INTEGRATED RESEARCH PROGRAM (SCHIRP)

Research Update #1: December 1996

Introduction

Cedar-hemlock (CH) cutovers in the Coastal Western Hemlock very wet maritime (CWHvm) biogeoclimatic subzone on northern Vancouver Island show poor regeneration about 8 years after clearcutting. The problem is indicated by foliar chlorosis and growth stagnation in planted Sitka spruce (*Picea stichensis* (Bong.) Carr.), and naturally regenerated western hemlock (*Tsuga heterophylla* (Raf.) Sarg.). Western redcedar (*Thuja plicata* Donn. ex D. Don) is less affected than the other species. These indications of nutrient deficiency on CH sites coincide with the vigorous reinvasion of the ericaceous shrub salal (*Gaultheria shallon* Pursh). Trees established on adjacent hemlock-amabilis fir (*Abies amabilis* (Dougl.) Forbes) (HA) cutovers do not experience this "growth check" (Weetman *et al.* 1989).

Prior to clearcutting, forests on CH sites are relatively open, uneven-aged old-growth stands with dense salal understories. The soils are typically undisturbed, compact, and imperfectly to moderately well-drained gleyed, humoferric podzols. In contrast, HA forests are dense, even-aged second-growth stands with minimal salal cover. Disturbed, friable humoferric podzols characterize these stands which originated from windthrow events in 1906 (Weetman *et al.* 1990). Lewis (1982) classified the two forest types as distinct phases of a single "salal-moss" (S1) ecosystem, and hypothesized that regeneration and site growth conditions typical of the HA phase could be achieved through silvicultural management of CH sites.

Foliar analyses indicated that conifers on CH cutovers were deficient in nitrogen and phosphorus. These low levels of N and P exist in CH sites prior to harvesting (Prescott *et al.* 1993), and are attributed to the immobilization of N in humus, resulting from excessive moisture, low soil faunal activity, cedar litter and tannins from salal. Additions of N and P fertilizers, in chemical forms (Weetman *et al.* 1989) or in organic wastes (McDonald *et al.* 1994), alleviate the chlorosis

significantly improve conifer growth on CH cutovers. Following harvesting, the poor nutrient supply on CH sites is exacerbated by rapid reinvasion of salal. Salal competes with young conifers for water and nutrients, produces tannins which inhibit N mineralization and uptake (deMontigny and Weetman 1990), and interferes with the ectomycorrhizae of hemlock (Xiao 1986).

Results of research conducted prior to 1994 were summarized in the first synthesis report (Prescott and Weetman 1994). The purpose of this report is to update the synthesis, providing results of research conducted since that time. Studies presented are:

- 1) a trial comparing effects of species, planting density, scarification and fertilization at planting
- 2) a trial comparing salal eradication and fertilization
- 3) trials with sewage sludge and fish silage
- 4) additional fish silage fertilization trials
- 5) a trial with fish-wood compost and straw amendments
- 6) studies of N fixation and denitrification in CH and HA cutovers
- 7) studies of condensed tannins in salal and humus
- 8) studies of N cycling and fertilizer N fate with ¹⁵N
- 9) studies of P forms in CH and HA forests and cutovers.

Influence of Density, Scarification and Fertilization at Planting on Growth of Cedar and Hemlock on CH and HA Sites

Jessica Pratt, Leandra Blevins and Cindy Prescott

Introduction

The current strategy for regenerating CH sites is to reach crown closure as soon as possible, thereby shading out the salal. Several silvicultural techniques might be applied to achieve this goal, alone or in combination. Planting trees at high densities should hasten crown closure, unless it results in slower growth of individual trees. Fertilization with N and P accelerates tree growth on CH sites, and typically increases crown size (Binkley 1986), which would further hasten crown closure. Scarification may also increase early growth rates by providing more planting spots and by mechanically disrupting the salal. Lewis (1982) suggested that mixing organic and mineral soil on CH sites by scarifying would also improve nutrient availability in a manner similar to that suggested for windthrow disturbance (Ugolini *et al.* 1990). The effects of these three silvicultural treatments on rates of growth of cedar and hemlock on CH and HA sites were compared in a large field trial. Planting density varied from 500 to 2500 stems/ha. Unlike previous trials in which fertilizer applied after growth rates had declined, trees in this trial were fertilized at the time of planting and 6 years later. Some of the plots at the highest density were also scarified prior to planting.

Methods

The trial was established in 1987 on a cutover that formerly supported both CH and HA forests. The area had been logged in 1986 and broadcast burned in the spring of 1987. There were 128 plots established, 64 on CH sites and 64 on HA sites. Each plot contained 64 trees; plot size varied with tree density. One-half (32) of the the plots on each site were planted with cedar, the other half with hemlock. One-half (16) of the plots of each species on each site were fertilized, the other half were not. Of these 16 plots, 4 plots were planted at 500 stems/ha and 4 at 1500 stems/ha. The remaining 8 were

planted at 2500 stems/ha; 4 of these plots were scarified prior to planting.

Thirty-two plots were scarified in January 1988 with a 215 Cat Excavator backhoe with a 3-tined rake attachment. Container stock seedlings (1-P415) of cedar and hemlock were planted in February 1988. Sixty-four seedlings were planted in each plot at spacings of 4.5, 2.6 or 2.0 m, corresponding to 500, 1500 and 2500 stems/ha. Sixty g of Nutricoat™ controlled-release fertilizer was applied and raked into the ground within a 15 cm radius of each tree in the fertilized plots. This provided 10 g N, 2.5 g P and 5 g K to each tree. Loading rates on an areal basis varied with planting density, with a maximum loading of 25 kg N, 6.25 kg P and 12.5 kg K /ha. Plots were refertilized in 1993; at this time N and P were broadcast to the entire plot at rates of 225 kg N and 75 kg P per hectare.

A subsample of 10 trees in each of the 64 plots planted at 2500 stems/ha were measured at the time of planting in February 1988. Average values for each species-by-site combination were used in the analyses. The height and root collar diameter of each of the 8,192 trees were remeasured in the fall or winter of 1988, 1989, 1990, 1992 and 1994.

Separate statistical analyses were conducted for each of the four species by site combinations (Cw on CH, Cw on HA, Hw on CH, Hw on HA). The effects of density and fertilization were tested with two-way ANOVA, excluding the scarified plots. Scarification and fertilization were compared using the values from the 2500 stems/ha plots only.

Results and Discussion

The effects of fertilizing at time of planting for the three tree densities are shown in Figure 1. Height responses are shown; the same trends were apparent in diameter and individual-tree volume responses. Cedar growth was similar in unfertilized plots on both CH and HA

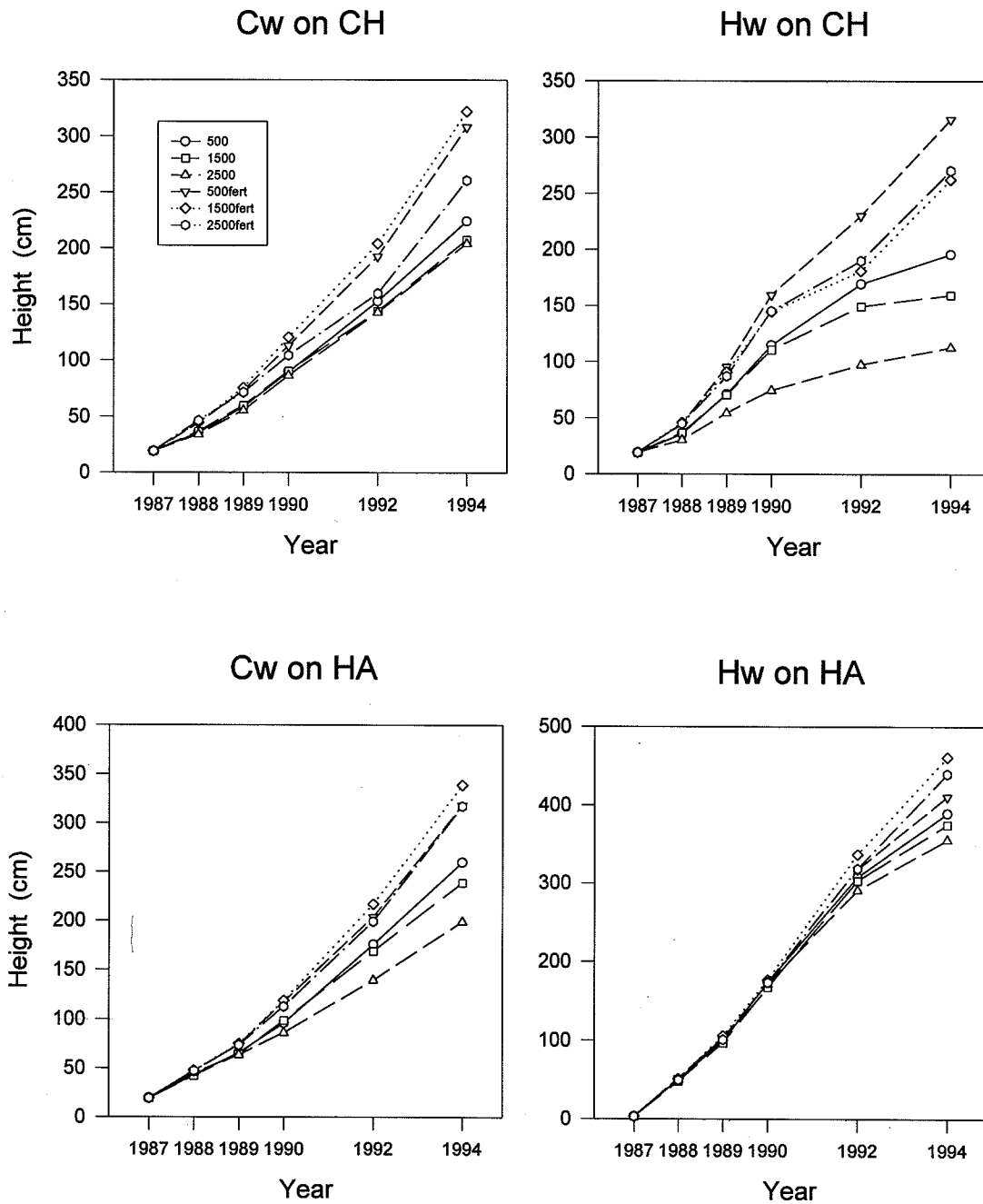


Figure 1. Height in cm of trees planted at densities of 500, 1500 and 2500 stems/ha with or without N+P fertilization at time of planting and in 1993.

sites, and it responded to fertilization similarly on both sites. Hemlock grew much more slowly on CH sites than HA sites, and responded much more to fertilization on CH sites. In both cedar and hemlock on both CH and HA sites, trees were larger in fertilized plots than in unfertilized plots of the same density. The similar growth of cedar and hemlock on fertilized CH sites contrasts with earlier studies in which N and P were applied 8-10 years after planting, in which hemlock growth was consistently greater than cedar. This suggests that cedar responds better when fertilized at time of planting than later. Greater response of cedar than hemlock to fertilization with teabags and briquettes at the time of planting has been observed on a salal-cedar site on northern Vancouver Island (S. Chambers, personal communication). In both of these trials, the fertilizer was placed very near the tree, in contrast to earlier trials in which it was broadcast across the plot. This would improve access to the fertilizer for the tree, and limit its uptake by salal, which may be particularly beneficial for cedar. Other trials have shown cedar to be particularly responsive to removal of salal (see next report), so cedar may also be more affected by increased growth of salal following broadcast fertilization. Therefore, the response of cedar may be attributable to the distribution of the fertilizer, rather than the timing. More research on the nutrition and rooting habits of cedar is necessary to better understand its response to fertilization at planting.

The significant growth response to fertilization during the first 6 years was unexpected, given that this is considered to be the period of high nutrient availability following clearcutting. Earlier studies documented a gradual decline in N availability on CH cutovers during the first 8 years after clearcutting and slashburning (Prescott and Weetman 1994). Results from the fertilization at planting trial indicate that the elevated levels of N on CH sites early after disturbance are still insufficient for maximum conifer growth. Individual tree fertilization is preferable to broadcast fertilization at time of planting for two reasons. First, tree roots would not yet be extensive enough to capture nutrients applied more than 1 m from the stem, so much of the fertilizer would be wasted. Second, competing vegetation would respond more to broadcast fertilizer, which would reduce conifer responses.

Lewis (1982) suggested that the high productivity characteristic of HA sites could be achieved on CH sites through silvicultural intervention. In the current trial, this goal was at least temporarily achieved through

fertilization at planting. Fertilized cedar on CH sites were larger than unfertilized cedar at corresponding densities on HA sites, indicating that fertilization at planting had improved growth of cedar trees on CH sites to a level comparable to HA sites. However, the fertilized hemlock on CH sites were not as large as unfertilized hemlock on HA sites.

Average tree sizes of both species planted at the highest density (2500 stems/ha) were smaller than those planted at the lower densities on both CH and HA sites. Responses to fertilization were also smaller in trees planted at the highest density. It is unlikely that competition with neighbouring trees would be significant in trees of this size. A more probable explanation is the lack of good planting sites on these cutovers which are characterized by mounds, depressions, airpockets in humus, and heavy slash accumulations. Many of the additional planting sites at the highest density were likely less suitable, resulting in poorer growth of many of the trees in this treatment. Growth and fertilization response was generally as good or better at densities of 1500 stems/ha compared to 500. Total stand volumes after 7 years were greatest at 2500 stems/ha, except in cedar on CH sites where volume was greatest at 1500 stems/ha (Figure 2).

Growth responses of both species planted at 2500 stems/ha to fertilization, scarification, or both, are shown in Figure 3. On HA sites, fertilization with or without scarification provided the greatest responses in both cedar and hemlock. Cedar appeared to be more responsive to fertilization than hemlock on HA sites. Responses to scarification were much smaller on HA sites. On CH sites, both species responded most to the combined treatment, and responses to either fertilization or to scarification alone were much smaller. Both treatments are therefore recommended to maximize growth on CH sites. Scarification is commonly carried out on CH sites as an alternative to slashburning; these results suggest that scarification may be beneficial in addition to slashburning. The additional effects of scarification are probably due to disruption of salal by this treatment, by removing the rhizomes. Salal resprouts rapidly after burning, which removes only the aboveground portions of the plant. Other trials in which salal has been removed manually have demonstrated prolonged growth responses of trees (see following report). An increase in nutrient availability following scarification is unlikely, since Keenan *et al.* (1994) found that N availability was not higher in mixed soil than in unmixed soil on these sites, and earlier trials

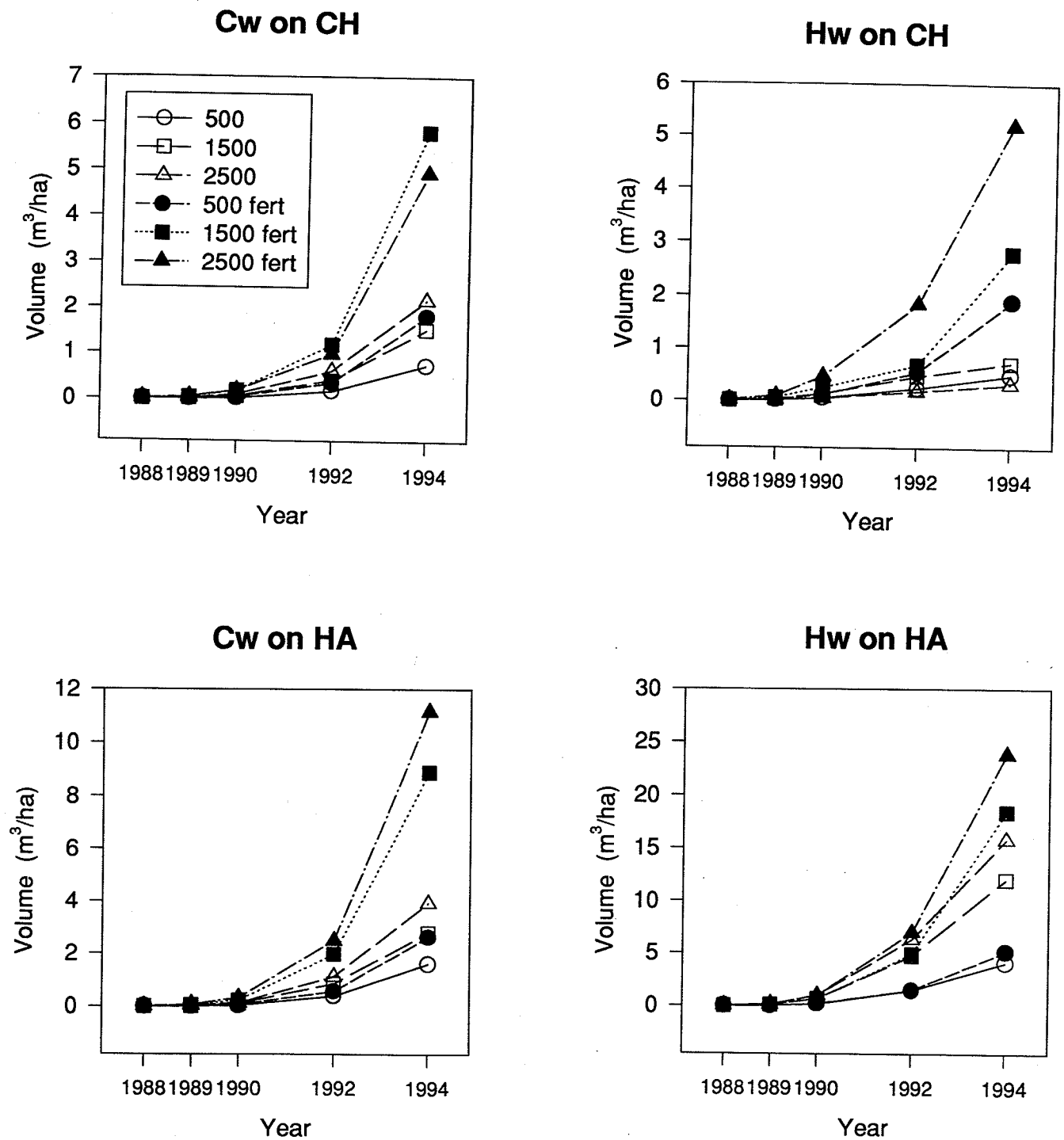


Figure 2. Volume in m^3/ha of trees planted at densities of 500, 1500 and 2500 stems/ha with or without N+P fertilization at time of planting and in 1993.

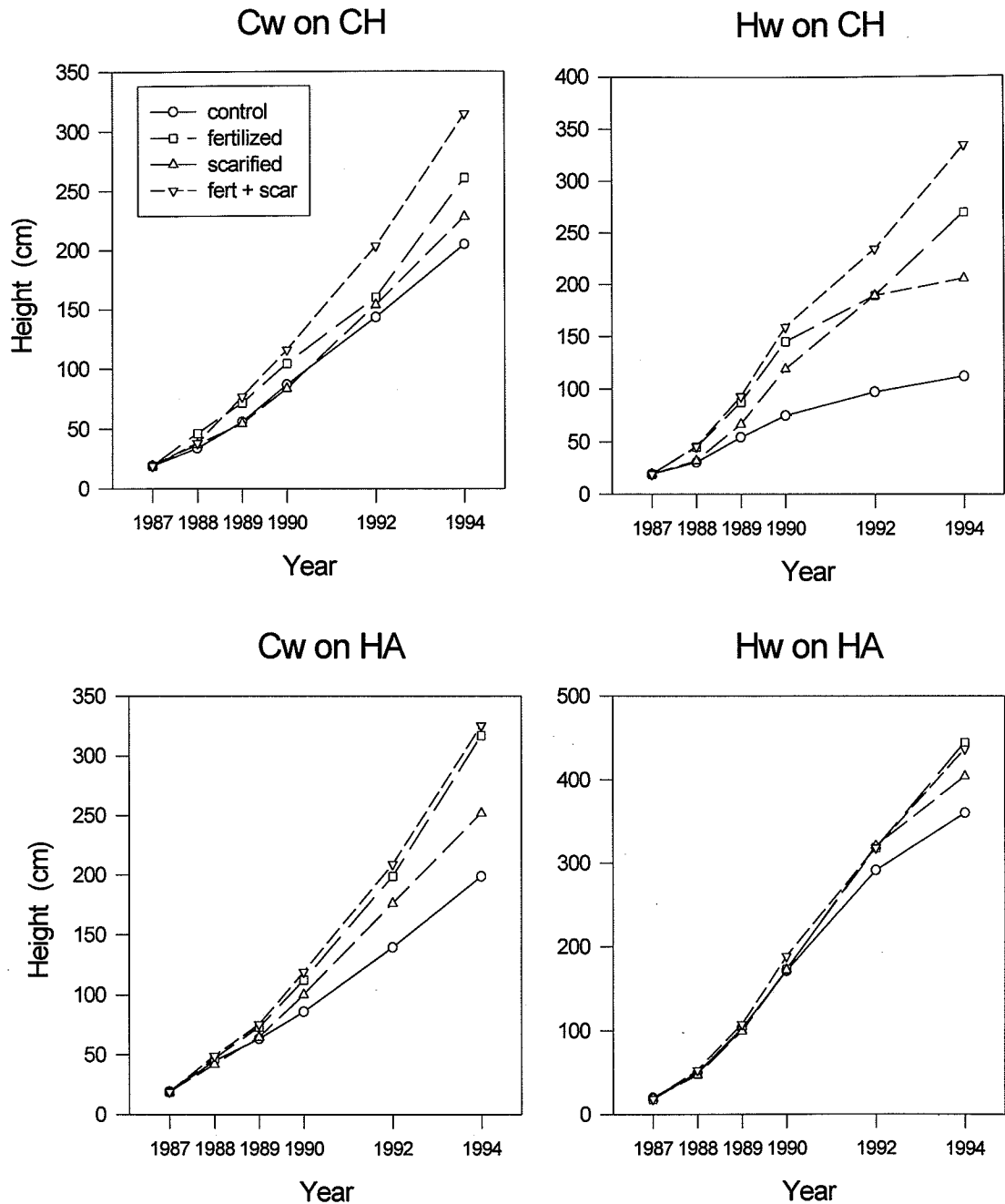


Figure 3. Height in cm of trees planted at 2500 stems/ha in plots that were scarified, fertilized at time of planting, both (fert + scar), or neither (control).

have indicated little response to scarification. Mechanical site preparation is used on other competitive sites as alternative to slashburning and herbicides to reduce competition from minor vegetation.

Rapid early growth of planted seedlings is essential on nutrient-poor, competitive sites such as CH cutovers. In this trial, mechanical site preparation in combination with individual tree fertilization at planting significantly

accelerated crown closure on these sites. This combined treatment may be particularly effective on other sites dominated by ericaceous shrubs, such as *Kalmia* in eastern Canada or *Calluna* in the U.K. Increasing pressure to hasten early plantation growth to achieve "green-up", or to shorten rotation lengths to avoid local shortages in wood supply, may make these treatments economically attractive on other types of sites.

Fertilization and Salal Removal for Improving Conifer Regeneration on CH Sites

Jennifer Bennett

Introduction

A salal eradication research trial was established on CH sites in 1984 to test the efficacy of salal removal and N and P fertilization for controlling salal competition and improving the growth rates of conifers. It was hypothesized that reduced salal cover should result in both an increased nutrient availability to trees and a sustained higher level of productivity. Similarly, fertilization with N and P was hypothesized to reduce salal biomass by hastening crown closure and decreasing the amount of light in the understorey. Messier *et al.* (1989) demonstrated that salal biomass on CH sites was significantly reduced when the crown closure reached approximately 80%. The salal eradication trial was remeasured in 1995 to determine the duration and magnitude of changes in tree growth in response to the treatments, the effects of the treatments on salal cover, crown closure and nutrient availability, and whether crown closure alleviates growth problems through salal depression or improved nutrient availability. Questions asked included: have significant tree growth responses been achieved through salal removal and fertilization; are these responses sustained; do the dominant tree species vary in response to the treatments; have the treatments successfully advanced the stands towards crown closure, reduced salal cover and more efficient nutrient cycling; and have the treatments resulted in a significant alteration of the nutritional status of the soils and the trees?

Methods

The trial consisted of five replicate blocks located on five CH sites harvested and burned in either 1969, 1971, 1972 (2 sites) or 1975. Following burning, Sitka spruce seedlings were planted and western redcedar, western hemlock and shorepine (*Pinus contorta* var. *contorta*) regenerated naturally. The

trial was established in 1984 and consisted of a 2×3×2 factorial treatment system of: salal removal; N fertilization (250 kg N/ha of ammonium nitrate (NH₄NO₃) or urea); P fertilization (100 kg P/ha of triple superphosphate). In 1985 and 1989, Garlon 4E™ was applied to all treatment plots to inhibit salal re-establishment.

In this study, only four treatments in each of the five blocks were measured; control, salal removal, N (NH₄NO₃) and P fertilization, and combined (salal removal plus N and P fertilization) treatments. In each plot, the diameters, (at 1.3 m) of the 30 largest trees were measured to compare treatment responses. Height, dbh, age (at dbh), and annual radial increment of the 10 largest individuals of cedar, hemlock, and spruce were also measured to examine species-specific responses. The selected trees were representative of the harvestable component of each stand. Average crown closures and salal covers were visually estimated in each plot. All trees taller than 1.3m were counted to estimate stand densities, and in each plot one soil pit was dug to determine soil characteristics and aid ecosystem classification.

Five samples of the forest floor "F" layer horizon were collected randomly from each plot and were combined to produce one composite sample per plot. From each of the five composite samples, one 5 g (dry weight) sub-sample was analyzed for concentrations of total N, P and S. Nitrogen and P were analyzed with an Alpchem RFA 300 auto-analyzer, following sulphuric acid – hydrogen peroxide digestion (Parkinson and Allen 1975). Concentrations of total S were measured by combustion in a Leco furnace. A second sub-sample was dried at 70°C for 72 hours to determine the moisture content of each composite forest floor sample. A third 20 g (fresh weight) sub-sample was extracted with Bray's solution and concentrations of PO₄-P were determined using the auto-analyzer (McKeague 1978).

A fourth fresh sub-sample from each plot was incubated for 21 days at approximately 20°C. After incubation, concentrations of NH₄-N and NO₃-N were measured with the auto-analyzer following extraction with 50 mL of 2M KCl (Page *et al.* 1982).

In August, 1995, hemlock foliar litter was collected from 10–15 trees in each plot and was oven-dried for 72 hours at 70°C. These samples were then digested in sulphuric acid and hydrogen peroxide, and total N and P concentrations were determined using the auto-analyzer. Green hemlock foliage was collected from the upper crowns of five individuals in each plot during the initial stages of tree dormancy in September 1995. These samples were dried at 70°C for 48 hours and concentrations of total N, P and S were measured as described above. All analyses were conducted at the MacMillan Bloedel Woodland Services Laboratory in Nanaimo.

Statistical analyses

Differences in arithmetic mean tree diameters were initially tested using two-factor analysis of variance to assess the effects of treatment and block, and interactions between the two factors. The results showed that tree diameters (the 30 largest trees in each plot) from each block should be analyzed separately. One-way analyses of variance were then conducted on the tree diameters to establish significant differences in the treatments within each block.

The ages of the three tree species varied as a result of the different harvest years of the five blocks and the different times of establishment; spruce were planted and hemlock and cedar naturally regenerated. Therefore, a direct statistical comparison of the

responses of the three species was not possible. Within-species covariance analyses were necessary to determine the influence of tree age as a covariant ($p < 0.0001$) of tree height and diameter. As a result, individual trees of each species were grouped into age classes composed of average ages that were not significantly different prior to comparing height and diameter responses to the treatments in each block. These comparisons were made on graphical and numerical bases.

The forest floor and the hemlock foliar nutrient concentrations from each block were combined by treatment and analyzed through one-way analysis of variance to establish any significant treatment effects on site nutrition. Annual radial growth increments were averaged and combined (all five blocks) to determine species-specific and treatment-specific diameter growth responses over time.

Results

The mean diameter of the 30 largest trees in each plot was significantly greater in the combined (fertilization plus salal removal) than in the control treatments (except in block 5) (Figure 4). Tree growth response to the salal removal and the fertilization treatments varied between blocks, which can be attributed to the differences in the species composition of the 30 largest trees in each of the blocks. To compare the diameter and height responses in the treatments, each species in each block was grouped into similar age classes. Blocks were compared independently; overall trends are presented in Table 1. Diameter and height response were highly

Table 1. Diameter and height of trees of each species in each treatment expressed as a percentage relative to the control.

	control (%)	fertilization (%)	salal removal (%)	combined (%)
Diameter				
spruce	100	143.4	124.2	200.2
hemlock	100	139.6	110.3	171.4
cedar	100	95.1	137.2	114.8
Height				
spruce	100	142.2	122.4	184.0
hemlock	100	132.3	111.3	159.0
cedar	100	104.9	126.5	113.8

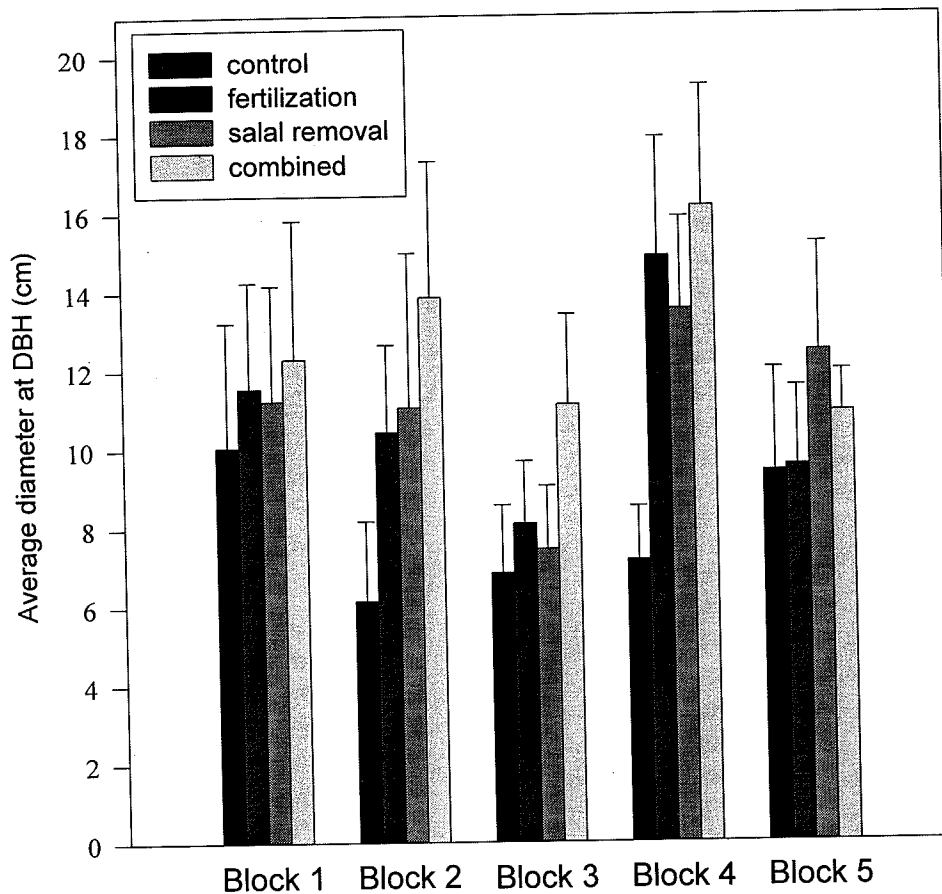


Figure 4. Average diameters of the 30 largest trees in each plot 10 years after fertilization, salal eradication or combined treatment.

species-specific. Spruce and hemlock growth was greatest in the combined treatments, followed by the fertilization treatments. Cedar growth was greatest in the salal removal and combined treatment plots.

The mean annual radial increments of each species over time are shown in Figure 5. In the control plots, cedar trees had the highest annual growth and the cedar radial increments generally increased through time. In the fertilization treatments, spruce and hemlock showed substantial increases in annual radial increments, but the increases declined within five years. Cedar did

not show a large response to fertilization. Salal removal resulted in gradual and sustained increases in annual increment in all three species, however, cedar showed the greatest increases. In the combined treatments, patterns of annual radial increment growth of all three species suggested that the effects of the individual treatments may be additive, as indicated by the initial dramatic increase in annual growth (fertilization response) which was relatively sustained (salal removal response). The 1995 annual increments of hemlock and cedar in the combined treatments, however, did show a lower level of radial

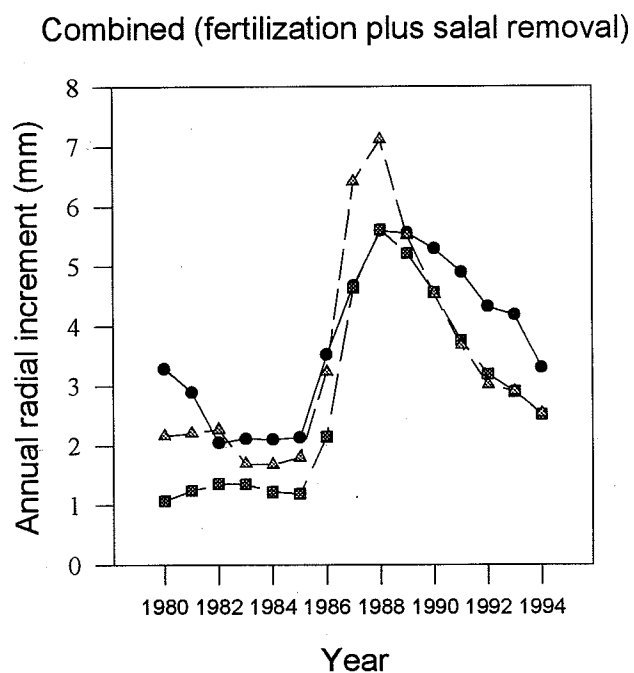
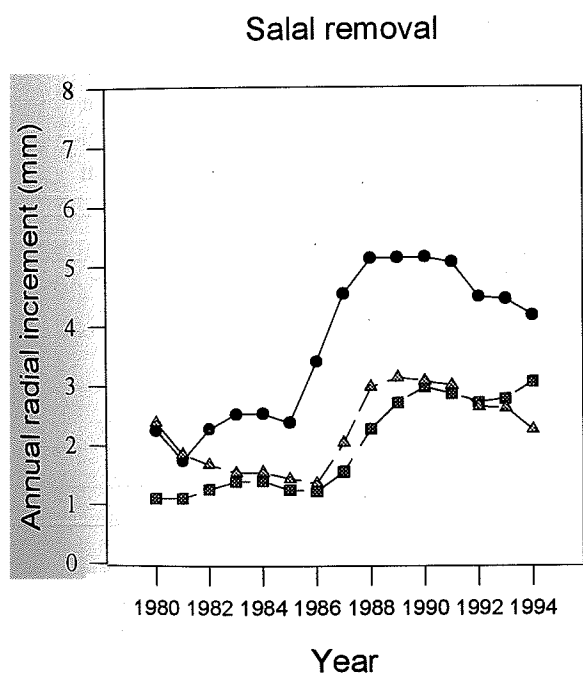
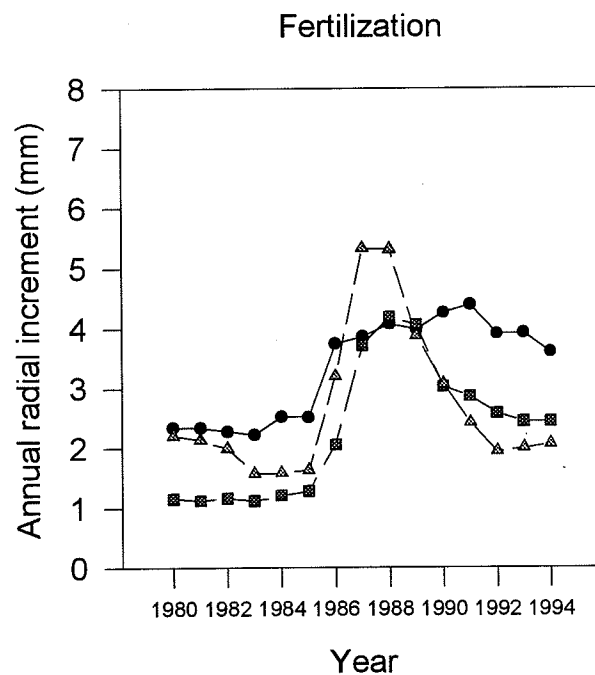
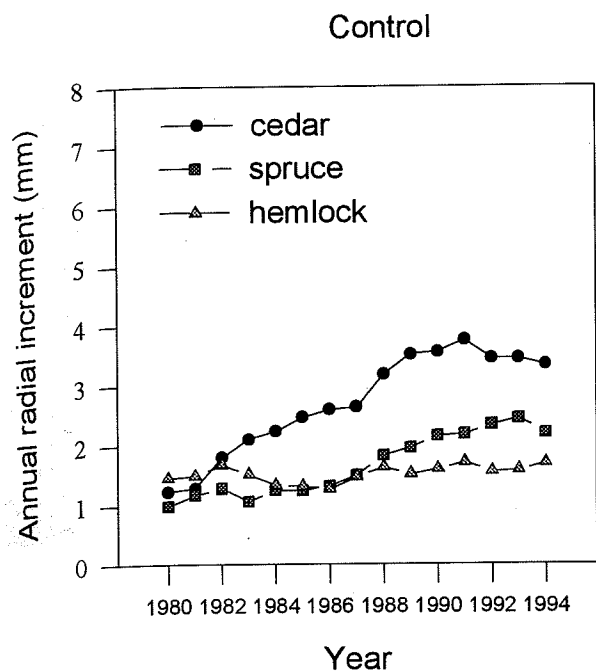


Figure 5. Annual radial increment of cedar, hemlock and spruce trees in plots receiving fertilization, salal removal or combined treatment in 1985.

growth than those in the salal removal plots. Over a ten-year period (1983-1993), changes in increment growth for all three species were greatest in the combined treatment followed by salal removal (Table 2). Of the three species, cedar, in the combined treatments, showed the greatest change in annual increment growth over the ten years.

Salal cover, as estimated in 1995, was lowest in the combined (5% cover) and salal removal (35% cover) treatments and highest in the fertilization (87% cover) and control (93% cover) treatments. A direct relationship between crown closure and salal cover was not found. In the fertilization treatments both were high, and in the combined treatments both were low.

Total N concentrations in the hemlock litter and foliage and total S concentrations in the green foliage did not differ significantly among treatments (Table 3). Total foliar P concentrations, however, were greater in the fertilization and combined treatments. Significantly higher total P concentrations were also found in the forest floors of the combined and fertilization treatments, but there were no differences in forest floor N or S concentrations (Table 4). Mineralized and nitrified N concentrations did not differ significantly, but a greater proportion of the mineralized N was nitrified in the salal removal

treatment (Table 5). There were no significant differences in the amount of extractable P in the "F" layers of the three treatments and the control.

Discussion

In general, the heights and diameters of each tree species were consistently larger in the three treatments than in the controls. These responses, however, varied among the species. Similar to responses reported in previous trials (Prescott and Weetman 1994), spruce and hemlock showed the greatest response to N and P fertilization. Fertilization caused an immediate (within one year) response, but growth declined within five years of fertilization. This large response to fertilization shows that low nutrient availability limits the growth of these two species on CH sites and that they are more sensitive than cedar to low nutrient supply. Hemlock and spruce also responded to salal removal, but cedar showed the largest growth response to this treatment. Salal removal resulted in gradual but sustained increases in cedar radial growth increment. This suggests that growth of cedar is inhibited more by the presence of salal than by a low N and P supply. For all three tree species, the removal of salal may reduce the competition for available N, to varying degrees, and result in sustained higher N availability.

Table 2. Average net annual radial increments of each species in each of the four treatments.

Treatment	Tree species	Average 1983 radial increment (mm × 10 ⁻²)	Average 1993 radial increment (mm × 10 ⁻²)	Difference in radial increment from 1983-1993 (mm × 10 ⁻²)
control	Cw	210	345	135
	Hw	139	245	106
	Ss	152	158	7
fertilization	Cw	223	393	169
	Hw	113	244	132
	Ss	158	200	42
salal removal	Cw	253	445	192
	Hw	139	278	139
	Ss	154	262	109
fertilization plus salal removal	Cw	212	419	207
	Hw	135	290	154
	Ss	170	291	120

Table 3. Concentrations of N, P and S in senesced and live foliage of hemlock in each treatment.

Treatment	Senesced Foliage		Live Foliage		
	% N	% P	% N	% P	% S
control	0.28 (0.02) a	0.03 (0.01) b	0.80 (0.15) a	0.09 (0.01) b	0.984 (0.012) a
fertilized	0.24 (0.06) a	0.15 (0.09) a	0.78 (0.13) a	0.23 (0.08) a	0.980 (0.010) a
salal removal	0.27 (0.02) a	0.03 (0.01) b	0.97 (0.12) a	0.09 (0.01) b	0.121 (0.026) a
fertilized + salal removal	0.26 (0.02) a	0.14 (0.05) a	0.78 (0.12) a	0.18 (0.06) ab	0.099 (0.006) a

Mean and standard deviation in brackets; values followed by the same letters are not significantly different.

Table 4. Concentrations of N, P and S in the forest floor "F" layers of each treatment.

Treatment	% N	% P	% S
control	1.03 (0.04) a	0.08 (0.01) b	0.12 (0.02) a
fertilized	1.15 (0.10) a	0.14 (0.01) a	0.12 (0.01) a
salal removal	1.07 (0.11) a	0.08 (0.01) b	0.11 (0.01) a
fertilized + salal removal	1.13 (0.62) a	0.12 (0.02) a	0.12 (0.01) a

Mean and standard deviation in brackets; values followed by the same letters are not significantly different.

Table 5. Extractable N ($\mu\text{g/g}$) after incubation and extractable P concentrations in the forest floor "F" layers of each treatment.

Treatment	Mineralized N ($\text{NH}_4 + \text{NO}_3$)	Nitrified ($\text{N} - \text{NO}_3$)	Proportion Nitrified ($\text{NO}_3 / \text{NH}_4 + \text{NO}_3$)	Extractable P
control	3.38 (1.23) a	1.78 (0.55) a	0.63 (0.36) ab	58.88 (5.81) a
fertilized	8.08 (6.72) a	2.24 (0.99) a	0.40 (0.22) b	64.02 (3.73) a
salal removal	3.04 (0.70) a	2.94 (0.79) a	0.96 (0.09) a	55.58 (7.37) a
fertilized + salal removal	3.10 (1.82) a	2.08 (1.11) a	0.80 (0.34) ab	58.96 (2.98) a

Mean and standard deviation in brackets; values followed by the same letters are not significantly different.

Crown closure was highest in the plots receiving the fertilization and combined treatments. This higher degree of closure probably resulted from the higher tree density in combination with the denser crowns in the fertilization treatments. Salal re-establishment was lowest in the combined treatment plots, likely a result of the reduced levels of light accompanying the increased canopy closure in this treatment. In the control plots, there was abundant salal and relatively open canopies. Unlike the combined treatment and the control, the fertilization and salal removal treatments did not follow the crown closure-salal cover inverse relationship. In the fertilized plots, high salal cover (87%) was sustained despite the increase canopy closure. The salal in these plots, however, had taller, lankier stalks and larger leaves, indicative of low light intensities (Messier *et al.* 1989). In the salal removal plots, both crown closure and salal cover were relatively low, as were the tree densities. Damage or stress to tree root systems during salal removal may have accounted for the low densities. Salal abundance was low despite the open canopy, suggesting that low light intensity is not the sole factor in determining salal re-establishment.

Foliar and forest floor nutrient analyses were used as indications of long-term changes in tree and site nutrient status resulting from the treatments. Concentrations of P in needle litter, green foliage and the forest floor were significantly higher in both the fertilized and combined treatments, suggesting that the single application of 100 kg P/ha resulted in a high site retention of P. Total N concentrations in both needle litter and green foliage indicated that no long-term changes in the N status of the trees had occurred as a result of the treatments.

Rates of mineralization and nitrification represent the amount of N potentially available to the trees. No patterns in response to the treatments were evident in measures of the mineralizable N, but the proportions of the two forms of mineralized N (NO_3 and NH_4) differed among the treatments. Forest floor NO_3 concentrations were the highest in the salal removal and combined treatments. Such increased NO_3 levels in the forest floor may have indirectly resulted from the removal of salal and the increases in nitrifier populations in the forest floor with reduced salal competition. Salal is able to access NH_4 and organic

N (Xiao 1994), and with its extensive rhizome systems and mycorrhizal associations and may be competitively superior, keeping nitrifier populations (and tree growth) low. Furthermore, the ability of salal to use organic forms of N further "short-circuits" the N cycle, by reducing the amount of organic N available for mineralization (Chapin 1995). Removal of salal might therefore increase both tree growth and nitrification.

The presence of salal appears to be the factor most limiting the growth of cedar. This growth limitation, however, does not appear to be solely due to competition for nutrients with salal, because fertilization did not cause a dramatic increase in cedar growth. The greater NO_3 proportions in the forest floor in the salal removal plots may be the factor accounting for the larger growth response of cedar. Cedar is generally associated with NO_3 - rich sites (Turner and Kranz 1985), and Krajina *et al.* (1972) demonstrated a preference of cedar for NO_3 in a seedling study. Cedar has also been shown to create and perpetuate conditions which favour higher nitrifier populations due to the high pH and base content of cedar litter (Prescott and Preston 1994, Turner and Kranz 1985). Other explanations for the relatively high growth performance of cedar when salal is removed are possible and warrant further research.

The changes in the proportions of available N (NO_3 and NH_4) in the forest floor of the salal removal treatments could also have influenced re-establishment of salal. Nitrate may be unavailable for salal use because like other ericaceous species, salal may not produce nitrate reductase (Smirnov *et al.* 1984), and so may not be able to take up N in the NO_3 form. With a higher proportion of NO_3 in the forest floor following salal removal, salal may be at a competitive disadvantage with species that are able to synthesize nitrate reductase required for nitrate assimilation, and may not be able to re-establish vigorously. Higher NO_3 concentrations were not found in the fertilization treatments, and salal was abundant in these plots. The ability of salal to establish and maintain growth may, therefore be limited by both higher concentrations of forest floor NO_3 and reduced understorey light following crown closure.

Conclusions

Both fertilization and salal removal increased tree diameter and height growth, with the greatest response to a combination of these two treatments. Fertilization resulted in an immediate, temporary burst of growth which accelerated the stands towards crown closure. Within five years of fertilization, however, annual growth was reduced to pre-treatment levels. In contrast, salal removal resulted in a gradual but sustained growth increases. Responses varied among the three tree species; spruce and hemlock responded most to fertilization, and cedar responded most to salal removal. The response of cedar to salal removal may have been related to the higher NO_3 concentrations in the forest floors in these plots, which may have also slowed the re-establishment of salal in the removal plots.

There were no indications of sustained higher N availability in response to the treatments, as N concentrations in foliage, litter and forest floors were similar in all treatments. However, with increased crown closure and tree size, the rate of litter input into the system probably increased, although N concentrations did not. The absence of a significant alteration in N concentrations suggests that N availability did not exceed tree demand in any treatment, and therefore still limited tree growth. Phosphorus availability, however, was still elevated the fertilized and combined treatment plots, suggesting that there had been a long-term (11-year) increase in P availability in fertilized plots.

Long-term Growth Response of Conifers on CH Sites to Organic Fertilizers

Susann Brown and Cindy Prescott

Introduction

Municipal sewage sludge contains organic matter, nitrogen, phosphorus and most other nutrients, which should make useful as a fertilizer for nutrient-poor sites. Growth responses have been reported following fertilization with sewage sludge in many species of conifers (McDonald *et al.* 1994). Volume growth tripled in Douglas-fir stands treated with sewage sludge in southwestern Oregon 5 to 12 years after application (Gessel *et al.* 1990). In organic fertilizers most of the nutrients are bound in an organic matrix from which they are released over time (Cole *et al.* 1984). In chemical fertilizers the nutrients are mostly in inorganic forms and are quickly released and are available primarily during the first growing season after application (Binkley 1986). The longer duration of nutrient release from organic fertilizers should result in a sustained growth response in trees.

Substantial growth responses occurred on northern Vancouver Island in plantations of western red cedar, western hemlock and amabilis fir treated with inorganic fertilizer, sewage sludge, and fish silage (McDonald *et al.* 1994, Weetman *et al.* 1993). Trees treated with inorganic fertilizer (ammonium nitrate and triple super phosphate) grew more rapidly during the first two growing seasons than those treated with organic fertilizers (Weetman *et al.* 1989). However, if the organic fertilizers release nutrients over a longer period of time than inorganic fertilizers, the growth responses thereafter should be greater in the trees treated with organic fertilizers. We tested whether or not organic fertilizers provided a longer growth response by examining height increments of trees treated with organic and inorganic fertilizer from year 1 to year 3 and from year 3 to year 5 after treatment application. Growth responses to inorganic fertilizers on these sites have been shown to decline 3 years after fertilization (Weetman *et al.* 1989).

Methods

Study site

Both trials were situated on a single cedar-hemlock (CH) cutover which was clearcut, burned, and planted with conifers. The site was described in more detail by McDonald *et al.* (1994) and Weetman *et al.* (1993).

■ Trial One

Sewage sludge, sewage sludge mixed with pulp sludge, fish silage mixed with wood ash, silage and ash mixed with pulp sludge, wood ash alone, and inorganic fertilizer (ammonium nitrate and triple super phosphate) were applied to a 9-year-old plantation of western red cedar in December 1990. Organic wastes were applied at 500 kg N/ha and inorganic fertilizer was applied at 225 kg N/ha and 75 kg P/ha. Pulp sludge was added to the sewage sludge and fish silage to increase the C:N ratio of the fertilizers. The trial was replicated in three blocks, each containing six treatments and one control (untreated) plot which were randomly distributed within each block. Tree heights were measured in December of 1990 through 1993 and 1995. The height increments from 1991 to 1993 and from 1993 to 1995 were calculated for all trees (22-38) in each plot and the means were compared among treatments by univariate analysis of repeated measures (SPSS 1992). The mean values were adjusted for original tree height (1990) before fertilizer application.

■ Trial Two

Sewage sludge or inorganic fertilizer (ammonium nitrate and triple super phosphate) were applied to 9-year-old plantations of western red cedar, western hemlock, and amabilis fir. There were 3 blocks; each block received one treatment. Each block contained four 15m x 15m plots of each tree species randomly distributed. There were 36 plots in total, including 12 control (unfertilized) plots. Tree heights were measured at the same time as in Trial One. For each species, the total height and height increments from

1991 to 1993 and from 1993 to 1995 were calculated for 50-84 trees in each plot and the means were compared among treatments by univariate analysis of repeated measures (SPSS 1992).

Results

■ Trial One

There were significant differences in height growth of western red cedar among treatments. Height increments were greater in fertilized plots than in control plots or plots treated with wood ash during both intervals (Figure 6). The height increments of all fertilized trees were smaller during the second interval than during the first interval, despite greater height growth in control trees and in trees treated with wood ash during the second interval. The greatest responses were to inorganic fertilizer and the two fish silage treatments. During the second interval, increments were still greater in all fertilized treatments than in the controls, but there was little difference between the various organic and inorganic treatments. Heights of trees in the sewage sludge treatments were similar to those in control plots after 5 years. Wood ash suppressed height growth relative to controls.

■ Trial Two

Sewage sludge and N+P fertilizer dramatically increased the height growth of all three species (Figure 7 a-c), but there was little difference between the two treatments. Height increments of all three species in fertilized plots during the second interval were much less than during the first interval but were still greater than in control plots. During the second interval, growth increments were similar in plots treated with sewage sludge and inorganic fertilizer in all 3 species. Height increments in control plots were similar during the two intervals, increasing slightly in amabilis fir and decreasing slightly in western hemlock and western red cedar.

Discussion

All of the organic amendments used in these trials increased foliar nutrient concentrations and growth of conifers on the CH cutover. Growth responses to the organic amendments were similar to those from the conventional N+P fertilizer, both in magnitude and duration. In theory, organic fertilizers should provide available N in small amounts over time while inorganic fertilizers provide a large amount of available N primarily during the first growing season. However, it

appears that the organic fertilizers released N shortly after treatment application, resulting in larger height increments from year 1 to year 3 than from year 3 to year 5. The sewage sludge used in the trials was anaerobically digested and stockpiled before application. Therefore, much of the mineralizable N may have been released before the sludge was applied. This available N would account for the large growth response during the first two years after sewage sludge application (McDonald *et al.* 1994, Weetman *et al.* 1993). Most of the remaining nitrogen in the sludge was probably in recalcitrant forms, from which mineralization would be slow. The net effect would be a flush of N immediately after treatment application, but little release thereafter, so the nutritional effects of sewage sludge would resemble those of chemical fertilizers.

The organic material in the fish silage had been ground and ensiled, so would have been largely broken down prior to application. Therefore most of the N in the silage was available immediately after application, resulting in a brief growth response. In later trials with fish silage (see following report), N concentrations in foliage and soil solution returned to pretreatment levels within a year of application, suggesting that silage also releases N immediately and only for a short time. Foliar N concentrations were greater in the fish silage treatments than any other treatment during the first two years after application (McDonald *et al.* 1994), indicating a high initial availability of N.

The pulp sludge added to the sewage sludge and fish silage was expected to immobilize some of the N in the sludge and silage, and release it gradually over time. There is little evidence of immobilization in the pulp sludge, since foliar N concentrations (McDonald *et al.* 1994) and growth increments were similar in treatments with and without pulp sludge. However, growth increments during the second interval were larger in the treatments that included pulp sludge, suggesting there may be a long-term effect of mixing the sewage sludge or fish silage with pulp sludge.

In summary, these trials demonstrated that large growth responses can be achieved with organic fertilizers on CH sites. Growth of trees in plots treated with organic or inorganic fertilizers was still elevated above control plots five years after treatments. Responses to sewage sludge and fish silage were similar in magnitude and duration to responses from inorganic fertilizers. Mixing sewage sludge or fish silage with pulp sludge may extend the response period.

western red cedar

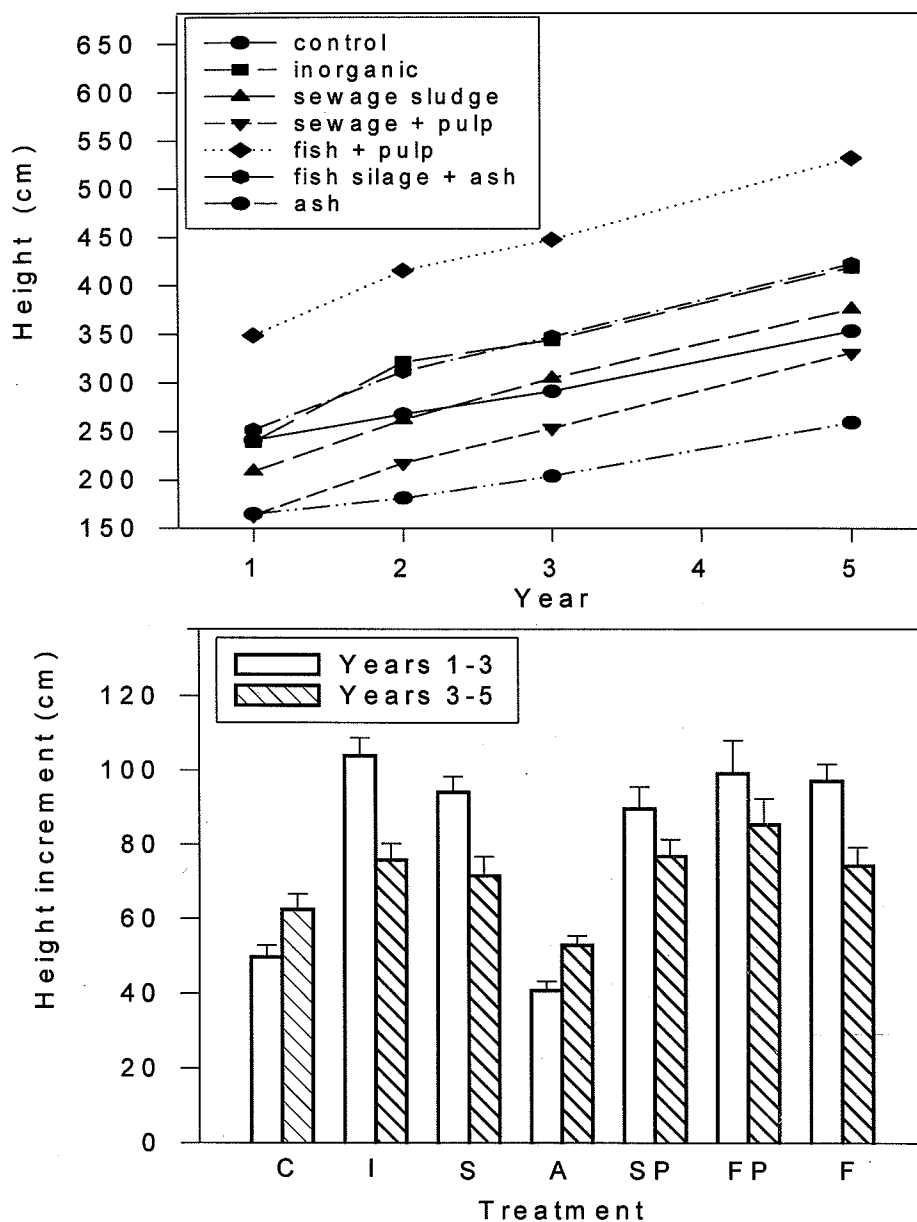


Figure 6. Height increments of western red cedar during years 1-3 (Interval 1) and years 3-5 (Interval 2) after application of organic fertilizers. C=control (untreated), I=inorganic (N+P), S=sewage sludge, A=wood ash, SP=sewage + pulp sludge, FP=fish silage + pulp sludge, F=fish silage + ash.

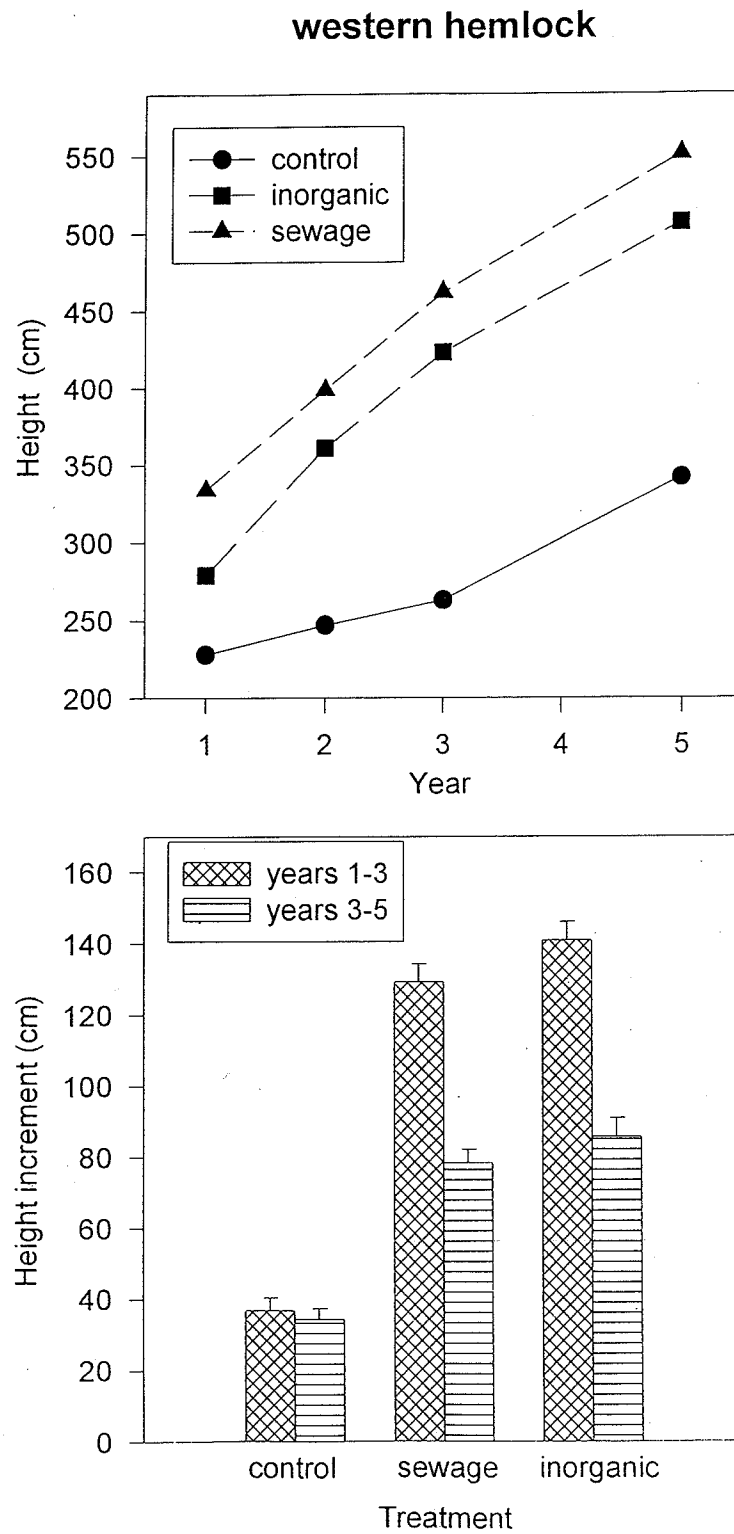


Figure 7a. Height increments of western hemlock, amabilis fir, and western red cedar during years 1-3 (Interval 1) and years 3-5 (Interval 2) after treatment with sewage sludge or inorganic (N+P) fertilizer.

amabilis fir

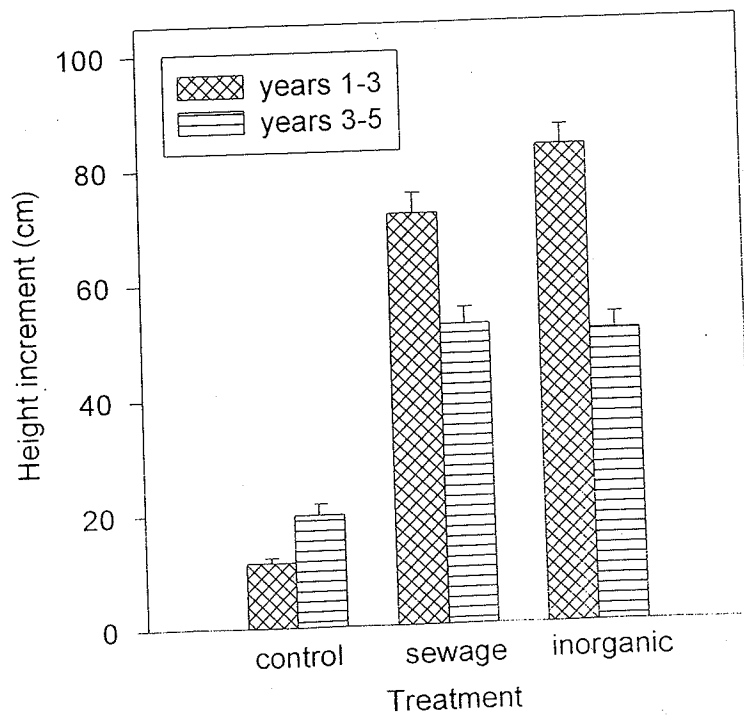
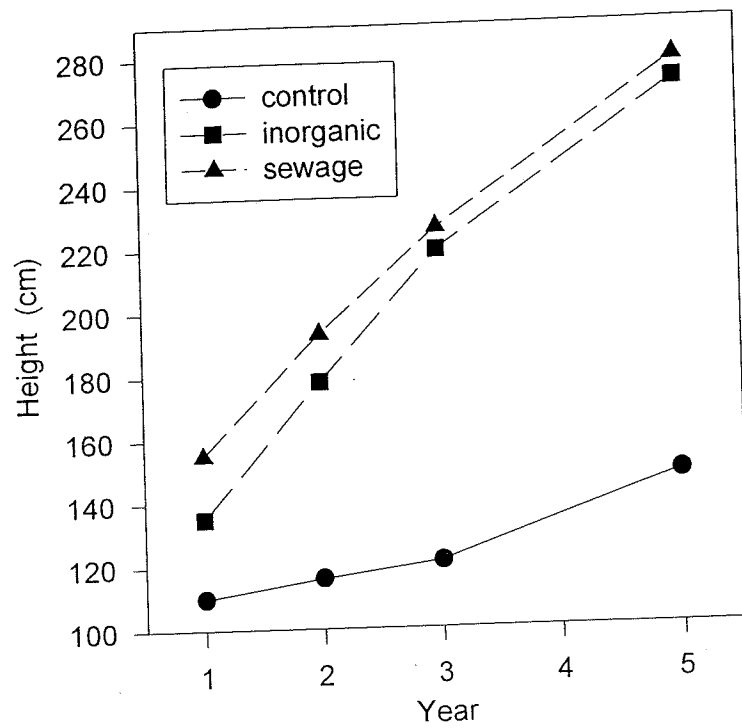


Figure 7b.

western red cedar

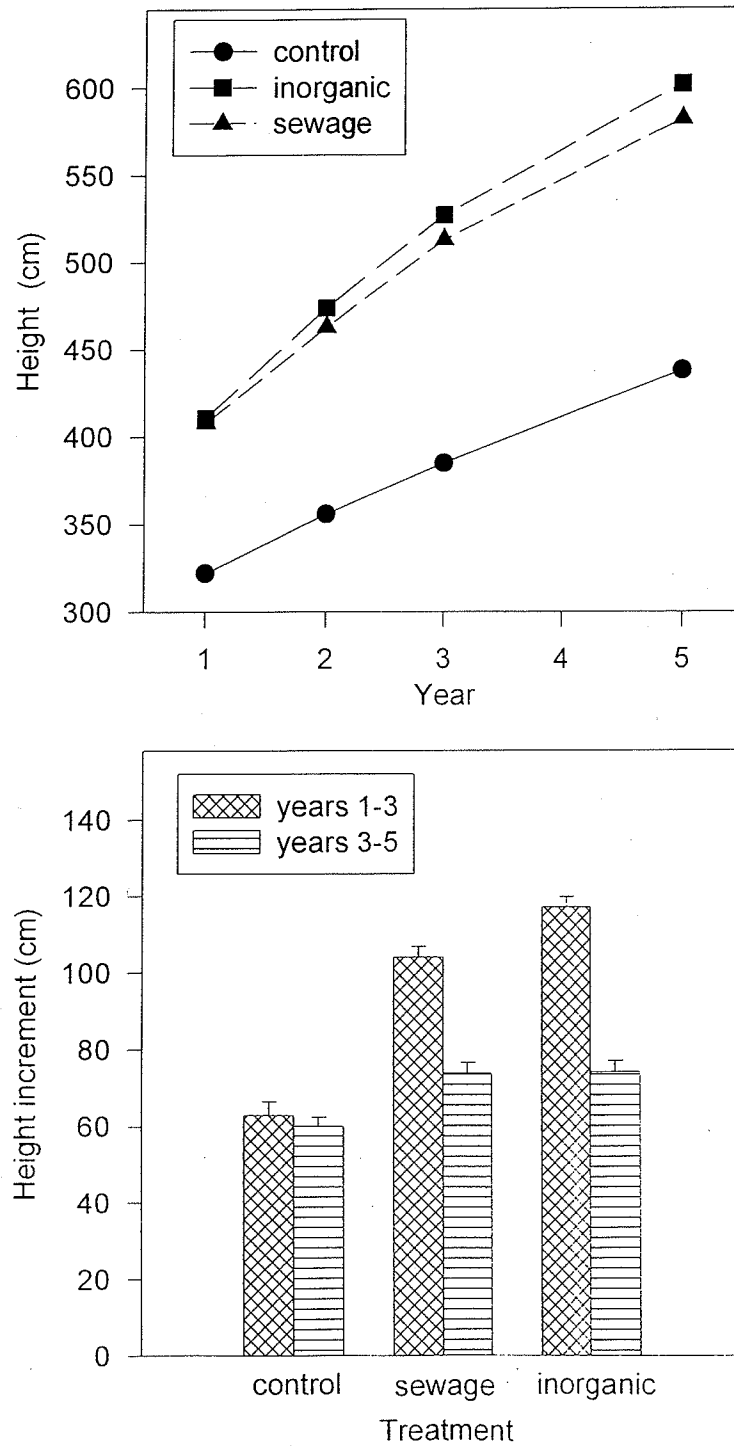


Figure 7c.

The Potential for Use of Fish Silage as a Fertilizer on CH Cutovers

Cindy Prescott

Introduction

The province of British Columbia is in need of an environmentally sound and economical means of disposing of fish waste. In 1988, aquaculture and fisheries industries in B.C. produced about 109,000 tonnes of offal and 1,600 tonnes of fish farm mortalities (morts). It was estimated that about 80,000 tonnes of this waste was disposed of in landfill sites (Holbek *et al.* 1991). Many landfills no longer accept fish waste and so producers have adopted the process of ensiling the fish waste as a means of storing salmon mortalities. In this process, morts are ground, preserved in acid (citric, formic or propionic) and stored in drums, and then transported to landfills or composting facilities. The most attractive option for disposal of fish waste is to develop end uses that will offset costs of ensilement and disposal. A possible end use of ensiled fish waste is as a forest fertilizer. Fish silage is very rich in most macronutrients essential for plants (N, P, S, K, Ca, Mg). A trial study in nutrient-deficient plantations of western red cedar on a CH cutover indicated a very favourable growth response to fish silage application (McDonald *et al.* 1994, previous report). The environmental consequences of fish silage applications are not currently known and are crucial to determining the overall feasibility of its implementation as an organic fertilizer and waste management alternative. Two possible environmental impacts of concern are leaching of nitrogen into watercourses, and acidification of the soil solution. Leaching or surface runoff into watercourses can occur after application of any nitrogenous fertilizer if more N is added than the plants and soil can take up. In coniferous forests, there may also be a longer-term effect, if the added N is sufficient to stimulate nitrification. Nitrate is usually present in very small amounts in coniferous forest soils, but often increases within a year of adding nitrogen. Nitrate is much more mobile in soil

than ammonium, and so prone to leaching loss, and also tends to acidify the soil. The silage itself might also acidify the soil water, as it has a pH of approximately 3.8. There may also be a loss of cations associated with the movement of nitrate out of the soil.

Environmental concerns related to forest fertilization have centered on drinking water quality and the effects of increased N concentrations on fish populations (Bisson *et al.* 1992). Monitoring of applications of urea and other N fertilizers during the 1960's and 1970's demonstrated that routine applications of N to coniferous forests do not result in nitrate and ammonium concentrations in excess of recommended limits for drinking water or for the protection of aquatic life (Bisson *et al.* 1992). Potentially toxic ammonium levels were detected immediately after fertilization of watersheds on Vancouver Island, but only where the streams were not protected by buffer strips (Perrin *et al.* 1984). In general, forest fertilization results in elevated organic N concentrations for a few days after fertilization, elevated ammonium concentrations for a few weeks or months, and elevated nitrate concentrations for up to a year or longer (Perrin *et al.* 1984).

The purpose of this study was to assess the environmental implications of the application of fish silage to young regenerating stands of western red cedar and western hemlock on northern Vancouver Island, B.C. Unlike previous trials, the silage was not mixed with wood ash to raise the pH, so we could assess the effects of the silage alone. Ammonium and nitrate concentrations and pH of soil water and stream water were monitored for 6 months after application, to assess the environmental impacts of fertilization. Tree heights and foliar nutrient concentrations were measured prior to fertilization and one growing season after application to determine the effect of fish silage on tree growth and nutrition.

Methods

The research plots were in a 4-year-old plantation of western red cedar on a gently sloping CH cutover. The trees were chlorotic and slow-growing and there was a dense cover of salal throughout the study area. Two 1 ha (50×200 m) plots were established adjacent to each other, with a buffer of 10 m (spray distance) between them. Both plots were located on typical terrain for the area *i.e.* sloped, and near a water source, but within the established Ministry of Environment guidelines. The plots were more than 50 m from the nearest ditch (below the plots), and almost 100 m from the nearest stream.

In each plot, 60 tension lysimeters were installed in pairs at 30 points, in a grid of 3 rows of 10. At each point, one lysimeter was installed below the organic layer (about 15 cm deep) and one below the rooting zone (about 30 cm deep). The soil solution was sampled from the lysimeters 1 month prior to application (February 17), and 1 week (March 23), 1 month (April 14), and 6 months (October 25) after the silage was applied. These dates coincided with rainfall events. There were no summer collections due to an extended drought. After collection, solutions were kept cool and transported to the Soil Science Laboratory at UBC. All samples were filtered and pH and conductivity were measured as above, and ammonium and nitrate concentrations with a Lachat autoanalyzer.

Fish silage was delivered from nearby fish farms, and stored on site in a 100,000 L tank. Nitrogen concentrations and pH of each batch of silage were estimated prior to application. Nitrogen concentrations in wet silage ranged from 3.12 – 3.75% N, and averaged 3.66% N. Nitrogen concentrations in oven-dried silage ranged from 8.35 – 11.56% N, and averaged 9.59% N. The moisture content of silage ranged from 55–73%, and averaged 62%. The average pH of the silage was 3.9. The silage was applied to the treated plot on March 17, 1994, using a tanker truck with a trash pump feeding two 1" hoses. A total of 12.96 tonnes of silage was added to one 1 ha plot, which is equivalent to 472 kg N/ha.

All sampling was conducted within an inner 30×180 m subplot in each plot. Ten samples of the forest floor and of the upper 15 cm of mineral soil were taken from each plot in December 1993. The pH was measured in mixtures of 5 g forest floor or 20 g

mineral soil and 20 mL distilled water, with an Accumet pH meter model 810.

To determine if there was an increase in pH, or ammonium and nitrate concentrations in streams as a result of fish silage application, the stream draining the plots was sampled at 5 locations upstream and immediately downstream of the plots. The ditch at the base of the plots was also sampled at 5 locations upstream and immediately downstream of the plots. Samples were collected at the same times as the lysimeter samples and similarly analyzed for pH, conductivity, ammonium and nitrate concentrations.

To determine the effect of fish silage on tree growth, 30 randomly selected trees in each plot were tagged and height was measured in December 1993 (prior to application), and 1 and 2 growing seasons after application (November 1994 and December 1995). Green foliage was collected from 5 trees in each plot and current-year foliage was separated as described by Radwan and Harrington (1986), oven-dried at 70°C, and ground in a Wiley mill. Samples were digested in sulphuric acid and hydrogen peroxide using a modification of the method of Parkinson and Allen (1975). Concentrations of N and P in each sample were measured with an auto-analyzer, and concentrations of K, Mg and Ca were measured by atomic absorption. Concentrations of S were measured by combustion in a combustion oven. All foliar analyses were done at the MacMillan Bloedel Lab, Nanaimo, B.C.

Results and Discussion

Prior to silage application, the average pH of the mineral soil was 4.68; and the range was 3.73 to 5.17. The average pH of the forest floor was 4.23 and the range was 3.66 to 5.53. These are similar to other values reported for CH forests and cutovers in this area (Germain, 1985, Prescott and Weetman 1994). The silage was not much more acidic (pH 3.9) than the forest floor onto which it was applied (pH 4.2), so it is unlikely that this treatment will directly result in soil acidification on these sites. Soil pH should be monitored occasionally on these plots to ensure that there is no long-term acidification effect of fertilization. If deemed necessary, the pH of the silage could be raised by mixing it with wood ash, as in previous trials (McDonald *et al.* 1994). The pH of soil solution in lysimeters was slightly lower in the treated plot on

all sampling occasions, including pretreatment. There was no consistent change in pH in the ditch samples taken upstream (1-2) and downstream (3-5) of the treated plot, or in the stream samples taken upstream (1-4) and downstream (5) of the treated plot. Conductivity was also greater in lysimeter samples from the treated plot, but this was also true prior to fertilization.

Concentrations of nitrate and ammonium in soil solution samples from the lysimeters were extremely low, usually below detection limits of the auto-analyzer. Prior to fertilization (in February), N concentrations within detection limits occurred in only 4 lysimeters. Two weeks after fertilization (in March), N concentrations within detection limits occurred in 7 lysimeters, 6 of which were in the treated plot. Average ammonium concentrations were greater in the treated plot, but average nitrate concentrations were greater in the control plot. Five weeks after fertilization (in April 1994), N concentrations within detection limits occurred in 14 lysimeters, 10 of which were in the treated plot. Average ammonium concentrations were greater in the treated plot, but average nitrate concentrations were greater in the control plot. Six months after fertilization (in October), nitrate concentrations were similar in control and treated plots, and ammonium was detected in only three samples.

Concentrations of nitrate in the stream and ditch samples were very low (maximum 0.083 mg/L), well below the water quality criteria of 10 mg/L for drinking water and 200 mg/L for aquatic life (Ministry of Environment, Lands and Parks 1994). The maximum concentration of ammonium in the stream and ditch was 0.063 mg/L, well below the recommended limit for the protection of aquatic life (Ministry of Environment, Lands and Parks 1994). There were no consistent differences in nitrate or

ammonium concentrations in samples from upstream and downstream of the treated plot.

The height of cedar trees in the control and treated plots were not different in December 1993, prior to silage application. In November 1994, one growing season after application, trees in the treated plot were significantly taller than in the control plot (Table 6). Height increments during the first two years after treatment were significantly greater in the treated plot (Figure 8a). Large growth responses have been observed in previous trials in which N+P fertilizer, sewage sludge or fish silage were applied to CH cutovers (Weetman *et al.* 1993, McDonald *et al.* 1994). Although these trials are not directly comparable, these findings suggest that fish silage alone is as effective in promoting tree growth as silage mixed with wood ash.

Concentrations of N, P and S in foliage of trees in the treated plot were significantly greater than in the control plot in November 1994 (Table 7). This indicates that these nutrients were sufficiently abundant in the treated plot to be taken up in excess of plant demand during the first growing season after fertilization. By December 1995, concentrations of all nutrients were similar in control and treated plots. Continued growth response is, however, anticipated in the future from these stored nutrients. Foliar concentrations of K, Ca and Mg were slightly depressed in the treated plot, which may indicate cation stripping from the soil. This contrasts with earlier trials, in which concentrations of these nutrients were greater in cedar trees amended with silage mixed with wood ash. Wood ash is known to be high in P, K and base cations (Erich 1991, Unger and Fernandez 1990). Concentrations of Ca and Mg in cedar foliage in the treated plot indicate that there is currently no deficiency of these elements; K concentrations suggest a slight-to-moderate

Table 6. Height growth (cm) of western red cedar in control and silage-treated plots one and two growing seasons after silage application.

Treatment	Height 1993	Height 1994	Height 1995	Increment 1993-94	Increment 1994-95	Increment 1993-95
Control	118.8	152.8	185.6	34.0	32.8	66.8
Treated	129.5	179.7*	232.1*	50.2*	52.4*	102.6*

*Significant ($p < 0.05$) difference between control and treated trees, based on oneway analysis of variance.

Table 7. Foliar nutrient concentrations (%) of western red cedar in control and silage-treated plots one and two growing seasons after silage application.

	% N	% P	% K	% Ca	% Mg	% S
1994						
Control	1.19	0.20	0.55	0.92	0.25	0.101
Treated	1.53*	0.25*	0.53	0.87*	0.20*	0.128*
1995						
Control	1.27	0.19	0.59	0.65	0.18	0.119
Treated	1.37	0.22	0.54	0.62	0.16	0.128

*Significant ($p < 0.05$) difference between control and treated trees, based on oneway analysis of variance.

deficiency (Ballard and Carter 1986). Foliar concentrations of these elements should be monitored annually to determine if deficiencies develop; if so, mixing the silage with wood ash would be recommended.

The very low concentrations of N in soil solution and the substantial growth response of trees in this CH cutover were not surprising, since very low rates of N mineralization and extreme N deficiency of trees have been documented on these sites (Prescott and Weetman 1994). The addition of N and other nutrients in fish silage increased N concentrations in a few lysimeters, but resulted in no detectable change in N concentrations in drainage waters. Most of the added N was probably taken up by trees or other vegetation, or immobilized in soil microbial biomass, humus or mineral soil (Binkley 1986). In addition to increased growth and N uptake measured in the trees, there was a very obvious increase in the amount of fireweed (*Epilobium angustifolium*) in treated plots during the first growing season after fertilization. Fireweed is characteristic of rich sites, and indicates sites of high N availability (Haeussler *et al.* 1990), and may also immobilize a large amount of available N in biomass (Messier and Kimmins 1991). It appears therefore that leaching of nitrogen into watercourses is unlikely to occur after fertilization of cedar-hemlock cutovers, because of the capacity of these N-deficient sites to immobilize added N.

■ Trial 2 – Hemlock

A similar trial was established in nearby areas with western hemlock in the same CH cutover. The height of 30 trees in both the control and the fertilized plot

was measured in May 1994. Silage was applied to the treated plot in June 1994. Trees were remeasured in November 1994 and December 1995, and foliage was collected for determination of nutrient concentrations. Height growth of hemlock trees was significantly greater in the silage-treated plot during the first two years after application (Figure 8b). Height increments in the treated plots in 1994 and 1995 were 24.5 and 57.5 cm, respectively, compared to 12.3 and 14.5 cm in the control plots. By the end of the second growing season after silage application, trees in the treated plot were significantly taller than those in control plots. At the end of the first growing season after silage application, foliar concentrations of N and P were substantially greater in the fertilized plots, (% N increased from 0.56 to 1.75, % P increased from 0.08 to 0.20). After 2 growing seasons, concentrations of N and P were still significantly higher in plots treated with silage, but Mg concentrations were significantly lower (Table 8).

Conclusions

The addition of 13 tonnes (472 kg N/ha) of fish silage to a cedar-hemlock cutover significantly increased growth and foliar nutrient concentrations of chlorotic cedar and hemlock trees. Elevated concentrations of $\text{NH}_4\text{-N}$ and reduced pH in soil solution were found in some lysimeters in the treated plot, but there were no detectable changes in N concentrations or pH in the stream or ditch draining the site. Application of fish silage at this rate appears to provide an effective means of improving tree growth on cedar-hemlock cutovers without affecting water quality.

Table 8. Nutrient concentrations in western hemlock foliage two growing seasons after silage application.

Treatment	% N	% P	% K	% Ca	% Mg	% S
Control	0.70	0.09	0.68	0.20	0.11	0.089
Silage	0.82*	0.12*	0.61	0.20	0.09*	0.097

*Significant ($p < 0.05$) difference between control and treated trees, based on one-way analysis of variance.

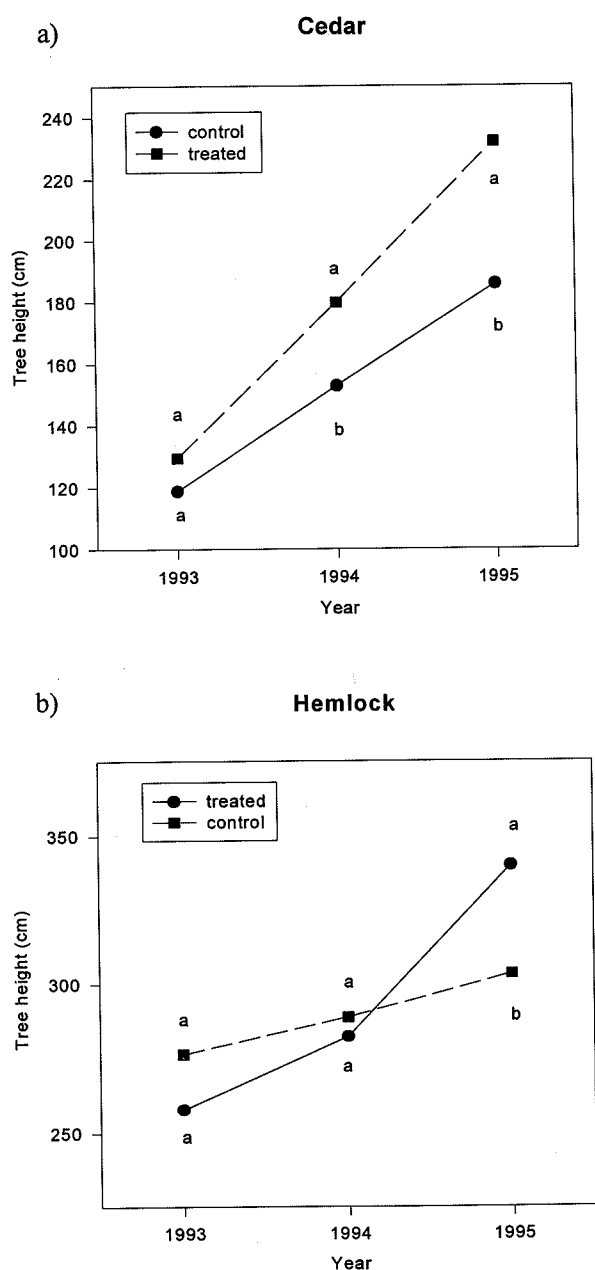


Figure 8. Height growth of cedar and hemlock trees following application of fish silage. For each year, means followed by the same letter are not significantly different.

Influence of Fish Compost and Straw on Growth and Nutrition of Cedar on CH Sites

Lisa Zabek and Cindy Prescott

Introduction

Ensiled fish waste and offal from fish farms on northern Vancouver Island and wood waste from pulp mills are composted at a new facility operated by Pacific BioWaste Recovery Society at the U.B.C. Research Farm near Campbell River. The end product is less noxious than the silage and has commercial value as a fertilizer and soil conditioner. The potential use of the product for silvicultural purposes was tested by measuring the response of nutrient deficient plantations on CH cutovers on northern Vancouver Island. Previous trials have shown these plantations to be very responsive to additions of N + P fertilizers (Weetman *et al.* 1989), and other organic wastes such as sewage sludge and fish silage (Weetman *et al.* 1993, McDonald *et al.* 1994, previous two reports). Fish-wood compost was applied to the same plantation as these other organic trials and the growth response was compared to that with the other fertilizers.

A single application of straw to a nutrient-deficient jack pine stand in Quebec smothered the ericaceous vegetation and promoted a long-term increase in tree growth and N availability (Prescott *et al.* 1995). The mechanisms for this response are not clear, but were probably the result of a reduction in shrub cover and improvement in the quality of the organic matter on the site. Cedar-hemlock cutovers on northern Vancouver Island are also very nutrient deficient and have poor quality organic matter and dense cover of the ericaceous shrub, salal. Straw was applied to these plantations to determine if the same effects were observed, and to identify the mechanisms involved in these changes.

The purposes of this trial were 1) to test the efficacy of composted fish silage and wood waste as a fertilizer for chlorotic cedars on CH cutovers, and 2) to monitor the long-term effects of straw application

on tree growth and N cycling in a cedar plantation on a CH cutover.

Methods

The trial is in an 11-year-old plantation of western red cedar on a CH cutover in TFL 25 of Western Forest Products, north of Port McNeill, B.C. Six 15×15 m plots were established in December 1992 adjacent to the existing organic fertilization trials (see earlier report by Brown and Prescott). In each plot, 25-31 trees were tagged and the height of each tree was measured in December 1992, 1993, 1994 and 1995. Foliage was collected from 5 trees in each plot in December 1992 and concentrations of N, P, K, Ca, Mg and S were measured in one composite sample from each plot.

On May 7, 1994, 13 m³ of fish compost and 178 bales of straw were transported from the UBC Research Farm to the field site. On May 12, 1994, 64 bales of straw were transported by backpack to each of two "straw" plots, and about 6 m³ of compost was transported in buckets to each of two "compost" plots. Bales were broken up and straw was applied loosely with a pitchfork to the forest floor and over the salal. The average depth of straw was 25 cm, and the salal was almost completely covered. Fish compost was spread onto the forest floor using a shovel or bucket. Conifer branches and salal were shook to remove compost from foliage, and compost on logs was brushed off. The average depth of compost was 1–2 cm. Control plots received similar traffic and shaking of vegetation. Concentrations of C, N, P, K, Ca and Mg were measured in 3 samples of the compost and straw.

The height of the 25 trees in each plot were measured annually in the winter, and foliage was collected from 3 and 5 trees per plot in 1994 and 1995, respectively for measurement of nutrient

concentrations. Differences between treatments were analyzed using oneway analysis of variance and Bonferroni's multiple range test. Samples from the 2 plots per treatment were pooled prior to each analysis. Data with non-homogeneous variances were log-transformed prior to analysis.

To establish effects of the treatments on N availability, rates of N mineralization were measured in the forest floor in each of the plots in March 1996. Five samples of the organic layer, including residual straw and compost, were collected from each plot on March 1, 1996. Live vegetation, coarse roots and wood were removed. A portion of each sample was dried at 70°C and moisture contents were determined. One 5-g dry weight equivalent subsample of each field-moist sample was extracted in 2M KCl and initial concentrations of $\text{NH}_4\text{-N}$ and $\text{NO}_3\text{-N}$ were measured on an Alpkem RFA-300 autoanalyzer. A second 5-g portion of each sample was incubated in a 1-pint jar for 29 days. The concentration of $\text{CO}_2\text{-C}$ in the headspace of each jar was measured weekly, after which the jars were opened for 15 minutes. After 29 days, the samples were extracted and final N concentrations (mineralized N) were measured as described earlier. Three bulked samples of salal leaves were also collected from each plot in March 1996, and concentrations of macronutrients were measured as described earlier.

Differences between treatments were analyzed using oneway ANOVA and Bonferroni's multiple range test. Samples from the two plots per treatment were pooled prior to analysis.

Results

The fish-wood compost had higher concentrations of total N, P, Ca and Mg than the straw, but the straw had higher K concentrations (Table 9). There was substantially more KCl-extractable $\text{NO}_3\text{-N}$ in the compost than in the straw. The C:N ratio of the compost (28) was narrower than straw (40).

Tree heights were greatest in the straw plots in 1994, prior to amendment, and increasingly so after amendment (Figure 9). Annual height increments (Table 10) were smallest in the compost plot the year prior to amendment (1992-1993). The first year after amendment, height growth was greater in both treatments than in the control plots. The second year after amendment height growth in the straw plots was greater than in the compost plots, and was twice that in the control plots.

Foliar concentrations of N, P, K and S were greater in plots amended with compost and straw at the end of the first growing season (1994). At the end of the second growing season after amendment (1995), foliar concentrations of N, P, K and S were significantly greater in the straw plots than in the control or compost plots (Table 11). Concentrations of N and P in salal leaves were also greater in straw and compost-amended plots than in control plots.

Extractable $\text{NH}_4\text{-N}$ and $\text{NO}_3\text{-N}$ concentrations after the 29-day incubation were much greater in the straw-amended forest floor than in the control or compost-amended plots (Figure 10). Rates of $\text{CO}_2\text{-C}$ evolution were also greater in straw-amended plots (6.5 ± 1.8 mg/g) than in compost-amended (3.6 ± 2.0) or control (2.5 ± 0.6) plots.

Discussion

Both of the organic amendments used in this trial increased growth of the previously chlorotic and slow-growing western red cedar trees in this plantation. This is consistent with previous trials with other organic fertilizers such as sewage sludge and fish compost (Weetman *et al.* 1993, McDonald *et al.* 1994), and chemical N and P fertilizers (Weetman *et al.* 1989). Neither the immediate effect of straw amendment, nor the greater response to straw than fish-wood compost was expected based on the nutrient concentrations of these materials. The compost had greater concentrations of all macro-

Table 9. Nutrient concentrations of compost and straw amendments. Each value is the mean of 3 samples.

	% C	% N	C:N	% P	% K	% Ca	% Mg	$\text{NH}_4\text{-N}$	$\text{NO}_3\text{-N}$
Compost	46.8	1.69	27.7	1.37	0.35	4.62	0.19	14.2	2420.0
Straw	47.1	1.19	39.6	0.14	1.37	0.35	0.11	22.6	2.6

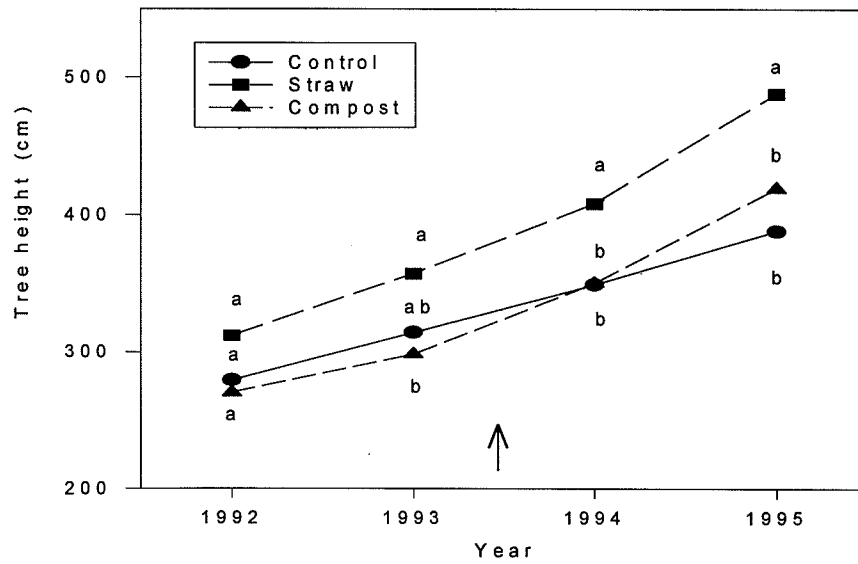


Figure 9. Height growth of cedar trees following application of fish-wood compost and straw. Arrow denotes time of application. For each year, means followed by the same letter are not significantly different.

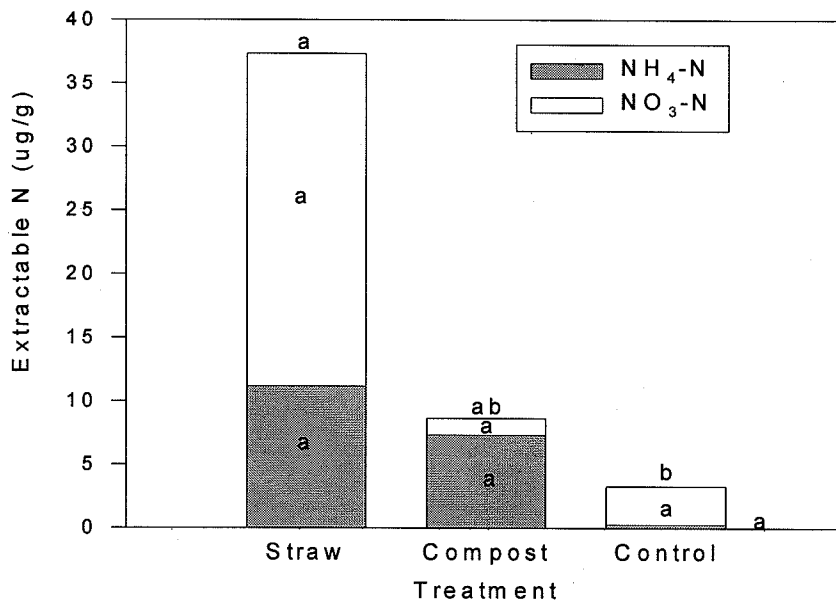


Figure 10. Concentrations of KCl-extractable N in forest floors amended with straw or compost, two years after application. Means followed by the same letter are not significantly different.

Table 10. Annual height increments of cedar trees in control and treated plots prior to and following amendment in May 1994.

Treatment	n	1992-1993	1993-1994	1994-1995
Control	58	35.3 (13.8) a	34.9 (11.5) b	39.3 (22.1) c
Compost	49	28.1 (12.8) b	52.1 (18.5) a	68.7 (19.5) b
Straw	54	45.1 (23.8) a	51.5 (23.5) a	80.2 (24.5) a

Within a column, values followed by the same letter are not significantly different ($p > 0.05$).

nutrients except K than straw, and had a narrower C:N ratio. It was therefore expected to have a greater influence on nutrient availability hence growth and foliar nutrition of trees. A long-term increase in N availability was also measured when straw was applied to a jack pine forest in Quebec (Prescott *et al.* 1995). Part of this effect was thought to be due to the reduction in the cover of the ericaceous shrub, *Kalmia* on these plots. In the present study, salal cover was not noticeably reduced by straw addition, but N availability was increased substantially. This suggests that the increased tree growth was a direct nutritional effect of the straw.

Differences in the carbon chemistry of the two amendments may account for their different effects on N availability, despite similar total N loadings (2000 kg N ha⁻¹). Compost is already partially decomposed, so it would provide little readily available C for microbial activity. After release of the extractable N, mineralization of the remaining material would be slow, so the increase in N availability would be short-lived. Straw, like other fresh residues, contains available C which stimulates microbial activity (particularly cellulolytic fungi), resulting in a larger and more active microbial biomass and faster mineralization. Turnover of the microbial biomass would prolong the period of increased N availability. Increased microbial activity was evident in the higher rates of C evolution from straw. Soil fauna were much more apparent in straw than in the compost or humus, which would further stimulate mineralization and turnover of the microbial biomass.

The response of cedar trees to straw and compost amendments can be compared with responses to other fertilizers measured in earlier trials on the same cutover (McDonald *et al.* 1994), although the trees were two years older when the present trial was

established. The height increment of trees in the compost plots during the second growing season after application (68.7 cm) was similar to the second-year response to chemical (N+P) fertilizer (65.7) and greater than responses to sewage sludge (57.8) or fish silage (57.4). The second-year height increments in the straw plots (80.2) surpassed that in any of the other treatments. The increase in foliar N concentrations in compost and straw plots during the first season after application was similar to that in plots treated with sewage sludge, and much less than that resulting from additions of chemical fertilizer or fish silage. Therefore, the higher N concentrations in cedar foliage in straw plots after the second growing season cannot be attributed to internal recycling of N taken up during the first year, and so is further evidence of sustained higher N availability in the straw plots. This supports the suggestion that there may be a long-term increase in N availability in the straw-amended plots, as was observed in the earlier trial in jack pine (Prescott *et al.* 1995). Although the application of fresh residues is more costly than conventional fertilization, it should be explored as means of promoting a long-term improvement in the productivity of CH sites.

Conclusions

Applications of fish-wood compost and wheat straw increased growth and foliar nutrition of chlorotic western red cedar on a CH cutover. Straw had a larger and longer-lasting effect than compost on foliar nutrient concentrations. Fresh residues, such as straw, which provide labile C as well as nutrients, may promote a long-term increase in N availability on CH cutovers.

Table 11. Foliar nutrient concentrations in cedar trees in control and treated plots prior to and following amendment in May 1994.

Treatment	% N	% P	% K	% Ca	% Mg	% S
1992*						
Control	1.15	0.16	0.47	0.56	.017	0.102
Compost	1.02	0.18	0.52	0.59	0.19	0.094
Straw	1.16	0.17	0.51	0.59	0.17	0.127
1994						
Control	1.06 b	0.17 b	0.45 c	0.61 b	0.18 a	0.099 b
Compost	1.63 a	0.24 a	0.81 a	0.59 b	0.16 a	0.125 a
Straw	1.61 a	0.22 a	0.73 b	0.76 a	0.18 a	0.135 a
1995						
Control	1.01 b	0.16 b	0.45 b	0.56 a	0.19 a	0.105 b
Compost	1.06 b	0.20 b	0.59 b	0.55 a	0.18 ab	0.106 b
Straw	1.39 a	0.29 a	0.95 a	0.51 a	0.15 b	0.140 a

Within a column, values followed by the same letter are not significantly different ($p>0.05$).

*There were no significant differences prior to application.

Nitrogen Fixation and Denitrification in CH and HA Forests

Andreas Brunner, Lisa Zabek and Mike Van Ham

Two hypotheses regarding the differences in N availability between CH and HA forests were investigated:

1. Greater input of N through N fixation in woody debris contributes to higher N availability in HA forests.
2. Greater loss of N through denitrification contributes to lower N availability in CH forests.

Nitrogen Fixation

Introduction

It was hypothesized that differences in rates of N fixation in coarse woody debris (CWD) between CH and HA forests are a significant factor contributing to differences in N availability on the two sites. There may be greater rates of N fixation in HA forests, resulting from both greater quantities of decomposable wood added by major windthrow events, and greater rates of fixation in the faster decomposing hemlock and amabilis fir logs than in the slower cedar logs. Rates of nitrogenase activity were measured in CWD in the two forest types, and in HA stands of different ages. Measurements of nitrogenase activity in different CWD of different decay stages, of different species, and in different positions in decomposing logs were made to assess the variation within the stands.

Methods

Study sites

Four plots were chosen: one each in HA stands of 5, 53 and 88 years of age (HA5, HA53, HA88) and one in an old-growth CH forest. The HA88 and CH stands were the SCHIRP-sites described by Keenan *et al.* (1993). The HA5 plot was in a 0.5 ha patch of the HA88 stand that was disturbed by a storm 5 years previously. About 80% of the stems in this area were

blown down and most were still suspended above ground. Natural regeneration and salal were abundant in patches. Due to the small size of the patch and shelter from remaining trees, the micro-meteorological conditions in this plot were more similar to that in a gap than in a clearcut. The HA53 stand, located 7 km northwest of the other three sites (Rupert 448 H2), originated from a storm in 1941. It was an even-aged, closed stand dominated by western hemlock. The proportion of amabilis fir in the stand had been reduced by recent stand self-thinning, as indicated by a high frequency of small fir logs on the ground. The stand had a dense, closed canopy and ground vegetation was sparse.

The investigation focused on woody debris greater than 1 cm in diameter. The five-class scheme of Sollins (1982) was used to classify the different stages of decay, based on the structural integrity of heartwood and sapwood. Decay classes I and II were combined because of the small amounts of this material and the small differences in structural integrity between these two classes. Preliminary investigations showed differences in nitrogenase activity between CWD of different species, and in material from different positions within a given log. Species differences were only investigated for decay class I/II. Material from the outer part and the core of logs were sampled separately and are referred to hereafter as sapwood and heartwood. The combination of decay class, species, and position in the log resulted in a total of 9 different substrates being sampled in each stand.

Biomass of decaying wood

Data on the biomass of decaying fallen logs were available for CH and HA88 stands from Keenan *et al.* (1993). The biomass of the HA5 patch was estimated from the standing volume of the adjacent HA88 stand (Keenan 1993). For the HA53 stand, the volume of the decaying woody biomass on the ground was

estimated using the line intersect method (Van Wagner 1982). The mass of woody biomass on the ground was calculated from these volumes using estimates for densities of wood in different decay stages based on Keenan *et al.* (1993). Decay classes III to V values were reduced by approximately 25%, which resulted in densities of 0.25, 0.17, 0.14 g/cm³ respectively for decay class III, IV and V.

Nitrogenase activity

Nitrogenase activity was determined in the field between October 3-7 1994, using an adaptation of the acetylene reduction assay (Hardy *et al.* 1973, Turner *et al.* 1980). Discs from up to five logs each of the different decay classes and species were cut with a chain saw. The discs were immediately separated into heartwood and sapwood, cut into pieces smaller than 5 cm, and transported to the lab in plastic bags. Within 12 hours, the samples were placed into 520 mL glass jars equipped with a plastic lid and a rubber septum for injection of gases. The sample jars were flushed with Argon for 30 to 60 sec prior to injection of acetylene to re-establish anaerobic conditions around the wood samples. Fifty-two mL of purified acetylene that had been scrubbed through a water trap was injected into the jars with syringes after the same volume of the jar atmosphere had been removed to maintain normal air pressure. The amount of acetylene injected was equivalent to 10% of the total jar volume. Depending on the volume of the wood sample the initial concentration of acetylene in the jar atmosphere was between 10 and 15%. The jars were incubated for 24 h on the ground in a stand of similar density and species composition as the stands under investigation, shaded with a plywood board. Minimum and maximum temperatures under the shelter during incubation were recorded (HA5: 9-13°C, HA53: 9-13°C, HA88: 4-11°C, CH: 9-14°C). Gas samples of 11 mL were taken after 24 h, and were stored in sterilized and evacuated 10 mL Vacutainers® for later analyses.

The gas samples were analyzed with a gas chromatograph model hp 5830A. Nitrogenase activity was calculated with the internal standard method (McNabb and Geist 1979). The samples were corrected for background amounts of 1 ppm of ethylene (79% of all control samples had a concentration of < 1 ppm) and 20 ppm of acetylene. Dry weight and water content of the samples were determined by drying them at 70°C for 3 days.

Results and Discussion

Mean nitrogenase activity in the 36 sampled substrates ranged from 1.3 to 19.5 nmoles C₂H₄/d/g dry weight (Table 12). Activity was measurable in all 180 samples. The lowest activity rates were mostly found in decay class I/II, and the highest in classes III and IV. Maximum activity in all four stands was in the sapwood of decay class III material. Only some of these differences are statistically significant due to the high variability in nitrogenase activity. The most significant variable influencing nitrogenase activity was water content.

The four stands showed no significant differences in nitrogenase activity (Table 12). The estimated nitrogen fixation rates for the three stands varied between 1.2 and 1.9 kg/ha/yr. Most of the N fixation occurred in the medium and advanced decayed classes that accounted for both the majority of CWD mass and the highest activity rates. Most of the variation between stands was due to differences in biomass of CWD because the differences in nitrogenase activity between stands were small. The low rate of N fixation in the CH stand was due to the small amount of CWD in this stand, which may have been anomalous. Keenan *et al.* (1993) did not find any differences in the masses of CWD in CH and HA stands. It is unlikely, therefore, that there are significant differences between CH and HA stands in rates of N fixation.

The chronosequence of the three different HA stands suggests a cycle in CWD and N fixation. Blowdowns add large masses of logs to the CWD already present in the older stand. Even if the recent material has a much lower nitrogenase activity it will contribute substantially to N fixation. As the stand develops, the supply of new CWD is low until it begins to self-thin. Consequently, the 53-year-old stand had lower CWD mass and N fixation rate than the other two stands. With continued stand development, more and larger self-thinning material would be produced, with an increase in the rate of N fixation.

Denitrification

Introduction

Earlier studies in CH forests (Prescott *et al.* 1993) determined rates of net N mineralization and nitrification to be low relative to adjacent HA forests, which could account for the low N availability after

Table 12. Estimated rates of nitrogen fixation in coarse woody debris in HA forests of different ages and an old-growth CH forest.

Stand	Decay class	Species*	Wood	Nitrogenase activity (nmoles C ₂ H ₄ /d/g DW)	Wood ratio	Species ratio	Mean nitrogen activity (nmoles C ₂ H ₄ /d/g DW)	Biomass (Mg/ha)	N-fixation (kg/ha/yr)		
HA5	I/II	fir	sap	3.50	0.33	0.6	2.35	150.0	0.66		
			heart	1.52	0.67						
	hemlock	sap	5.33	0.33	0.4	6.25				35.5	0.41
		heart	1.26	0.67							
	III	sap	8.92	0.33	0.67						
		heart	4.94	0.67							
	IV	sap	5.61	0.33	0.67		2.15	43.6	0.17		
		heart	11.12	0.67							
	V		2.15			296.2				2.41	
HA53	I/II	fir	sap	3.01	0.33						0.90
			heart	2.22	0.67						
	hemlock	sap	1.96	0.33	0.10		15.52	11.1	0.32		
		heart	2.67	0.67							
	III	sap	19.54	0.33	0.67	8.51				35.4	0.56
		heart	13.54	0.67							
	IV	sap	7.63	0.33	0.67						
		heart	8.95	0.67							
	V		4.77				102.8	1.37			
	I/II	fir	sap	5.25	0.33	0.90			4.25	11.8	0.09
			heart	3.88	0.67						
	hemlock	sap	4.70	0.33	0.10	6.62					
		heart	2.94	0.67							
	III	sap	7.42	0.33	0.67		7.91	43.6			
		heart	3.18	0.67							
	IV	sap	5.97	0.33	0.67				158.0	1.87	
		heart	6.94	0.67							
	V		7.91								
CH	I/II	cedar	sap	10.90	0.14		0.50	2.97			
			heart	2.41	0.86						
	hemlock	sap	3.10	0.33	0.50		4.56		63.0	0.54	
		heart	1.96	0.67							
	III	sap	7.53	0.20	0.80	6.20					30.1
		heart	3.82	0.67							
	IV	sap	5.57	0.33	0.67			5.77			
		heart	6.51	0.67							
	V		5.77				129.1		1.18		

*Species not determined for decay classes III – V.

clearcutting. Among the factors suggested as possibly contributing to low N availability was denitrification, *i.e.*, gaseous loss of N after conversion from nitrate. Rates of N mineralization and nitrification were measured during laboratory incubations during which there is the potential for denitrification. If there were considerable losses through denitrification, rates of mineralization and nitrification would have been underestimated. The purposes of this study were to:

- 1) measure rates of denitrification in CH and HA forest floors in the field, to determine if this is a significant route of N loss in CH forests;
- 2) measure rates of denitrification in CH and HA forest floors during aerobic laboratory incubations to determine if rates of N mineralization and nitrification were underestimated due to unmeasured gaseous losses of N;
- 3) measure rates of denitrification in decaying wood from CH and HA forests to estimate potential rates of N loss from this material.

Methods

Actual rates of denitrification were measured in the field, and potential rates were measured in the laboratory. Both procedures used the acetylene block technique to prevent reduction of N_2O to N_2 . All samples were from the same uncut CH and HA forests at the "SCHIRP" site, used for the N fixation study.

Field estimation of denitrification

In December of 1995, ten closed chambers were inserted in the forest floor of each stand to a depth of 3 cm. Each chamber was fitted with a rubber septum in the top, through which 20 mL of scrubbed acetylene was introduced to the atmosphere in the head space. After 4 hours, 5 mL of the gas in the head space was removed and injected into 4.7 mL Vacutainers[®]. The samples were transported back to Vancouver and analyzed for N_2O concentrations. One sample from each site was not injected with acetylene, to measure background concentrations of N_2O .

Laboratory incubations

Five samples of the fermentation (F) and humus (H) layers of the forest floors were taken from the CH forest and the HA forest in February, 1996. Each layer from each stand was processed separately and kept refrigerated until use. Samples were passed

through a 2 cm mesh sieve to remove large pieces of wood, roots and stones. Five gram (dry weight equivalent) samples of each forest floor were incubated in canning jars (Kerr wide mouth, 1 pint (0.6 dm³)) for 40 days (Prescott *et al.* 1993). At 5 times during the incubation period 10 mL of acetylene was injected into each jar. N_2O was allowed to evolve for 4 hours after which time 5 mL of the atmosphere in the jar was removed. After sampling, the jars were opened to the outside air for 30 minutes and the 20 samples were immediately sent for analysis. One sample of each forest floor type was not injected with acetylene, to measure background concentrations of N_2O . Concentrations of N_2O in the gas samples from the field incubations of CWD were measured in 105 samples of wood in decay classes IV and V. For each substrate, two samples from incubation without acetylene were similarly measured.

Results and Discussion

Concentrations of N_2O during the field incubations of forest floors and CWD were not significantly different from background concentrations. Moisture content of the forest floors were very high (80%), but low temperatures at this time of year might have limited rates of denitrification. However, N_2O concentrations measured in the laboratory, at about 20°C, were also similar to ambient levels throughout the 5-week incubation. This suggests that the denitrification potential of these forest floors is low, so it is unlikely that significant denitrification occurs in the field.

Conclusions

- The highest rates of N fixation in CWD were in decay class III and IV, the lowest activity rates were in the less decayed material (class I/II).
- Rates of N fixation in CWD were similar in CH and HA forests.
- Rates of N fixation in CWD in HA forests follow patterns of CWD input; peaking early in stand development following windthrow and again during self-thinning.
- Rates of denitrification from forest floors and woody debris were low in both CH and HA forests.

Background

Tannins are substances of plant origin which were originally defined by their astringent taste, and by their protein-binding ability. Their ecological role has long been a subject of interest. Tannins apparently play a role in plant defences against herbivory and may be produced in response to stress, in particular under conditions of high photosynthesis (C-fixation) coupled with nutrient (N and P) limitation. Tannins affect nutrient cycling as they enter the soil organic matter pool with litterfall. Because they bind to proteins, tannins reduce decomposition and N mineralization from litter and humus. Although substantial amounts of tannins are released from decomposing plant material, virtually nothing is known of the fate of tannins soil.

Tannin chemistry

Condensed tannins, or proanthocyanidins are based on the flavan-3-ol structure, with catechin (4) and epicatechin (5) occurring most frequently (Figure 11a). They form polymers with linkages at C4–C8, or less commonly, C6–C8 (Figure 11b). Polymers based on the tri-hydroxy B ring (6 and 7) are prodelphinidins (PD); those with the di-hydroxy B ring (4 and 5) are procyanidins (PC). Variations occur in the number of OH groups on the B ring, the occurrence of C4–C8 or C4–C6 linkages, the chain length, and other structural modifications of the basic building blocks.

Figure 12 shows solution ^{13}C NMR spectra of condensed tannins extracted from salal flower heads. The ^{13}C NMR spectrum can be used to determine the ratio of PC to PD units, the stereochemistry at C2–C3 (cis or trans), the average chain length, and information about linkages. Carbon-13 “CPMAS” NMR uses a combination of techniques (Cross-Polarization, Magic-Angle Spinning and high-power decoupling) to obtain well-resolved spectra of solid samples. A variant “dipolar-dephasing” (DD) CPMAS NMR,

shows carbons with no attached hydrogens, and also those which have some molecular motion in the solid state.

Carbon-13 CPMAS tannin spectra have considerable diagnostic potential for plant components and organic horizons. Most of the tannin signals overlap those of other biopolymers such as cellulose, lignin and cutin (Preston 1996), but the tannin peak at 145 ppm occurs in a region that is relatively clear. A second indicator is found in the DD spectrum, where a peak around 105 ppm is a special characteristic of condensed tannins.

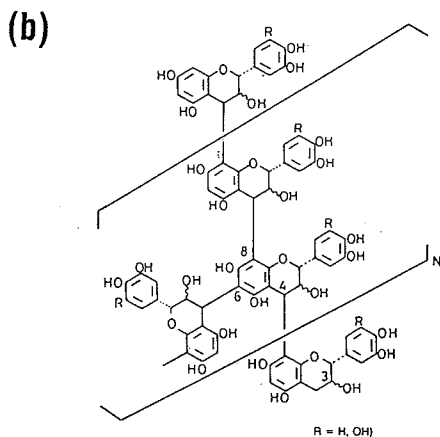
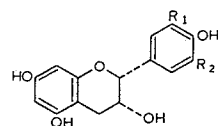
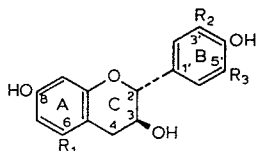
Methods for extraction and purification of tannins are shown in Figure 13, while Figure 14 outlines analytical methods. Carbon-13 CPMAS NMR can be used to screen whole plant material, but solution ^{13}C NMR is essential for characterizing extracts and monitoring purification. Two procedures are recommended for condensed tannins, the vanillin and proanthocyanidin assays. We use the latter, in which the condensed tannin is depolymerized by an acid catalyst (HCl) in *n*-butanol, to give a red anthocyanin product whose absorbance is determined at 555 nm. The best analytical standard is a purified condensed tannin from the same species.

The radial diffusion assay quantifies the protein-binding ability of tannins (Hagerman 1987). A thin layer of agar containing a small amount of protein is prepared in a Petri dish. An aliquot (10 to 40 μl) of test solution is placed in a well, and left to diffuse. Tannins interact with the protein, producing an opaque circle whose area is proportional to the concentration of tannin in the well.

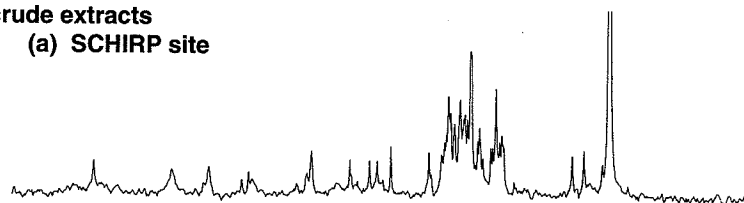
Results

Salal chemistry — Preparation and characterization of tannins from open-grown salal flowers and stalks.

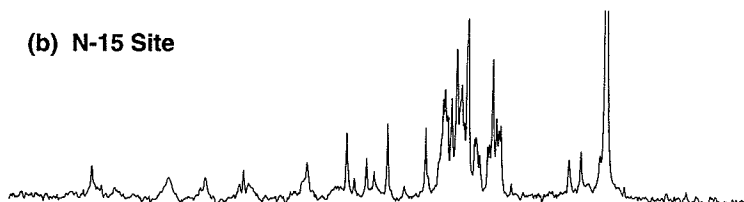
Tannins from salal flowers and stalks. Tannins were isolated from fresh, intact salal flower-heads



(a) SCHIRP site

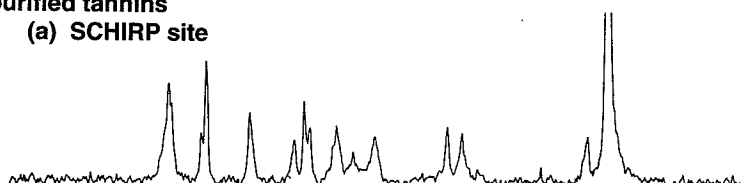


(b) N-15 Site

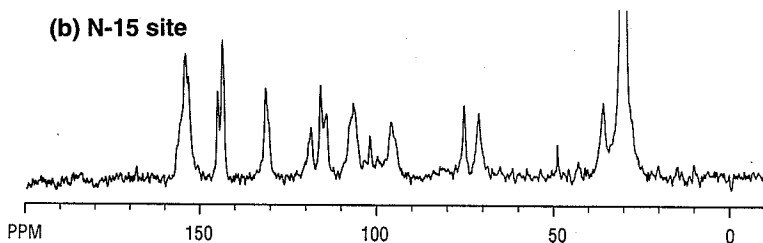


-purified tannins

(a) SCHIRP site



(b) N-15 site



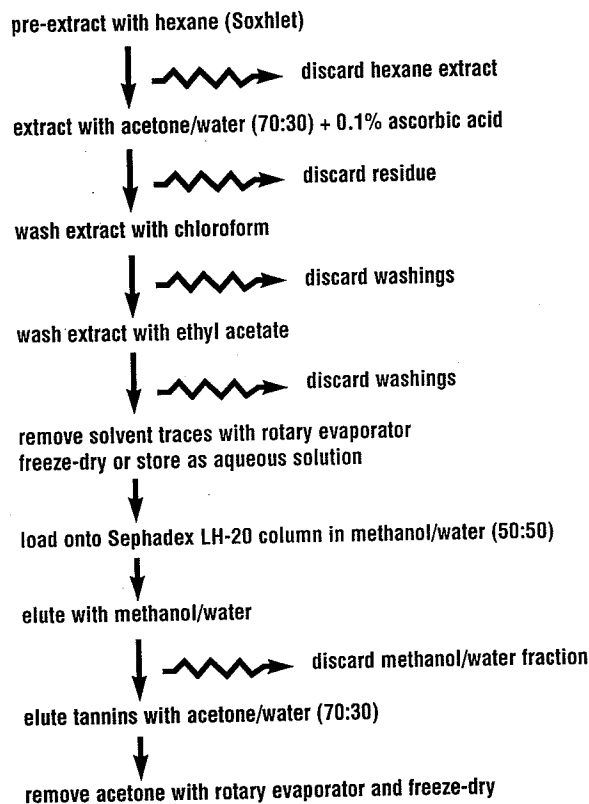


Figure 13. Procedure for preparation of condensed tannins from plant material.

1. Detection in Plant Material

-non-destructive screening by solid-state C-13 NMR

2. Quantitative Extraction and Characterization

-extract, purify, characterize by solution C-13 NMR
-use as analytical standard for assays

3. Routine Analysis of Plant Samples

-50-100 mg of dry, powdered material
-extract in screw-cap test tubes with 2 ml acetone/water and 1 ml ether; centrifuge

↓

-discard ether/acetone layer, transfer aqueous layer to clean screw-cap test tubes

↓

-repeat extraction, combine aqueous extracts
-dry at 70°

↓

-allow sample residue to air-dry for residual tannin analysis

↓

-prepare standards using purified tannin from same plant

↓

HCl/n-butanol hydrolysis
(extracts and residues)
-read absorbance at 550 nm

↓

Diffusion assay (extract only)
-pipette aliquot onto gelatin-agar plate
-develop at 30°C for 3 days
-measure ring diameter

Figure 14. General procedures for analysis of condensed tannins.

from two CH cutover sites. The ^{13}C solution NMR spectra of the purified tannins are similar and characteristic of condensed tannins (Figure 12c,d), while the crude extracts are high in carbohydrates (Figure 12a,b). The 146/146 ppm ratio indicates approximately one-third PD and two-thirds PC monomer content. The stereochemistry is almost entirely cis, as C2 is represented by a peak at 76 ppm, with barely detectable intensity at 83 ppm where trans C2 would appear. The small peak at 67 ppm from C3 in terminal units is more obvious in the SCHIRP tannin.

NMR survey of salal components. High levels of tannins in salal flowers, leaves and roots, were reported by de Montigny *et al.* (1993). We examined salal components from plots used for an ^{15}N study in a CH clearcut (Chang *et al.* 1996b). The ^{13}C CPMAS NMR spectra were in good agreement with those found previously, indicating high tannin levels in salal leaves, dead leaves, litter, roots and senesced fruits. There was much less tannin in the live and dead salal stems. The roots (rhizomes) and stems had lower levels of alkyl C than the leaves and fruits.

Characterization of humus by CPMAS ^{13}C NMR. de Montigny *et al.* (1993) previously used ^{13}C CPMAS NMR to characterize humus horizons from undisturbed CH and HA sites. We examined two sets of humus samples. One was from plots which were sampled in September 1992 as part of an ^{15}N uptake study (Chang *et al.* 1996b). This provided samples of humus from a CH site which had uncontrolled salal invasion for 8 years after clearcutting in 1979, and then either vegetation control or no control for over 5 years, from April 1987 to September 1992. The second set was part of a study of microbial biomass in a clearcut chronosequence (Chang *et al.* 1995). The overall conclusion was that the spectra of both woody and non-woody horizons up to thirteen years after clearcutting and burning (with or without 5 years of salal control) showed essentially no differences from those obtained from old-growth CH sites. All of the samples of non-woody humus showed evidence of tannins. Five years of salal control may not be enough time for large differences to develop. Despite 5 years of above-ground control, salal continued to sprout; although by 1995, the extent of salal resprouting was diminishing.

Preparation of a protein-binding fraction from humus. A humus sample was extracted using conventional methods for tannins, except that the sample was extracted directly with acetone:water without a preliminary hexane extraction. A fraction which eluted with 50% methanol/water was high in carbohydrate. Two fractions had protein-binding ability and gave a response in the proanthocyanidin assay. The yields as a percentage of the original humus were 0.83% for fraction 3 and 0.22% for fraction 4.

Analytical procedures

Proanthocyanidin assay. The procedure was used with 5% HCl, 12 mg/L Fe^{2+} and 4.7% H_2O . [Work in progress indicates that the optimum iron concentration is around 200 mg Fe^{2+} /L.] Five mL of the reagent is added to the sample in 0.5 mL methanol. The tube is heated at 95 – 100°C for 1 h, and absorbance is measured at 550 nm.

Radial diffusion assay. This test provided reasonably reproducible and consistent results, although its precision is limited. It provides a valuable counterpoint to the proanthocyanidin and NMR assessments, because it is responsive to both hydrolysable and condensed tannins.

Recommendations for Further Study

- Prepare tannin from salal foliage; compare structure with that from the flower-heads.
- Optimize the hydrolysis conditions for salal tannins.
- Optimize conditions for routine extraction and analysis of foliage.
- Survey tannin levels in salal foliage and other components under different conditions of nutrition and shade; compare stems, roots and rhizomes.
- Continue to develop an extraction procedure for tannins from humus.
- Survey tannins in humus of old-growth CH and HA and clearcuts, including the salal eradication plots which have now achieved crown closure.
- if suitable equipment can be obtained, develop protocols for running ^{15}N CPMAS NMR spectra of ^{15}N labelled humus and tannin-protein complexes.

The full report is available from Caroline Preston,
Pacific Forestry Centre, Victoria (26 figures, 4 tables,
2 appendices of spectra, and 123 references).

Acknowledgements

Others have made huge contributions to this work:

UBC PhD students Louise de Montigny and Scott Chang, co-op students Myles Bos, Tim Coram, Shelley Duquette, Andrew Preston, Derek Wong, and Andrea Ellis, and PFC staff Kevin McCullough, Ann van Niekerk, Junning Niu and Brian Titus. Brian Sayer (McMaster University) ran some of the earlier CPMAS spectra.

Fertilizer N Efficiency and Incorporation and Soil N Dynamics

Scott Chang

Summary of PhD thesis, Department of Forest Sciences, UBC, 1996

To better understand the nutritional problems affecting conifer regeneration on western red cedar – western hemlock (CH) cutover sites, the purpose of this study was to:

- 1) investigate the effect of salal competition on tree growth and nutrition, particularly the fate of fertilizer N;
- 2) determine the distribution and availability of residual fertilizer N in forest soils; and
- 3) study if soil microbial competition for nutrients is also a factor limiting early conifer growth on CH cutover sites.

One experiment had single tree plots planted to western red cedar, western hemlock, and Sitka spruce, either with understory removed or remaining. Plots were fertilized with 200 kg N ha⁻¹ (¹⁵N labeled) in spring 1991 and destructively sampled in fall 1992. Treatments had significant ($p < 0.1$) effects on height growth in 1992. Height growth between 1987 and 1989, and total height measured in early October 1992 were significantly greater in the treated plots than in the control plots. No treatment-by-species interactions were observed for any of the parameters measured, which means that treatment had similar effects on tree height growth of all species studied. Understory removal also increased root collar diameter growth. The diameters of cedar, hemlock and spruce, respectively, measured in early October 1992, were 38, 88, and 65% greater in the treated plots than in the control plots. Diameter growth of cedar in the summer of 1992 was 111% greater in the treated plots than in the control plots, but was not different for hemlock and spruce. Better growth in the understory removal treatment was a result of reduced uptake and immobilization of nutrients by the competing vegetation. The differences between species were generally not significant and no

treatment-by-species interactions were found. Prolonged exclusion of understory competing vegetation resulted in a significant increase in biomass of trees. Biomass was more reliable than height and diameter growth as an indicator of the effect of understory competition on tree growth (Chang *et al.* 1996a).

In the study of fertilizer ¹⁵N (Chang *et al.* 1996b), the majority of the ¹⁵N in the trees one year after fertilization was in the current-year needles. The second largest proportion of the ¹⁵N recovery was in branches older than 3 years, followed by 2-year-old needles, except for ¹⁵N in the treated western hemlock plots. From 49 to 74% of the ¹⁵N recovered in the trees was in the needles. Between 84 to 94% of the ¹⁵N in the trees was in the above-ground tree components, the remainder was in below-ground components including the stump. No difference was induced by salal-removal for ¹⁵N distribution in all of the tree components studied. The pattern of ¹⁵N distribution was determined by the distribution pattern of biomass in various tree components. However, tree species significantly affected the distribution of ¹⁵N in 1-year-old needles, 2-year-old needles, older than 3-year-old branches, stumps, and coarse and medium roots. The distribution of ¹⁵N followed that of total N very closely, indicating either a proportionate uptake of applied N and native N by tree components or a rapid turnover and redistribution of ¹⁵N within the tree. When ¹⁵N was expressed in mg per plot, salal removal significantly increased the storage of ¹⁵N in all of the above-ground tree components. Salal-removal treatment apparently reduced ¹⁵N abundance in the above-ground tree components, regardless of the species used, with the exception of 1-year-old spruce branches. This was a dilution effect, *i.e.* the greater ¹⁵N uptake could not compensate for the greater biomass accumulation in the treated plots.

From 7.7 to 17.8% of the applied fertilizer was recovered in the trees in the treated plots, compared to only 2 to 5% in the control plots. If the ^{15}N recovered in the litter and standing dead biomass is added to that in the understory, the amount of ^{15}N labeled fertilizer immobilized by understory vegetation was 4.8, 15.9 and 3.3 times as much as that taken up by trees in the control plots. Treatment effects on ^{15}N recoveries in above- and below-ground tree and total tree components were significant in hemlock ($p < 0.05$). Differences in ^{15}N recovery among species were observed only between cedar and hemlock in the understory removal treatment. The greatest total recovery of applied ^{15}N in the soil-plant system was in the treated hemlock plots (87.3%), while the least was in the treated spruce plots (56.8%). After two growing seasons, most of the recovered ^{15}N was in the soil component. Total recovery of ^{15}N in the soil was as high as 66.8% of added N in the treated hemlock plots.

Some of the other observations made from this study: (1) salal was a persistent competitor for ^{15}N , i.e., six years after understory removal, from 1.4 to 3.1% of the applied ^{15}N was recovered in the understory of treated plots; (2) removal of salal understory increased ^{15}N incorporation into soil; (3) total recovery tended to be higher in the plots with understory present, because the understory took up more applied N shortly after application, thus reducing N loss from the system; and (4) accumulation of ^{15}N in the understory biomass, litter and standing dead material greatly reduced the availability of fertilizer N to crop trees. The abundant understory vegetation in the control plots perhaps resulted in more ^{15}N being taken up initially by plants in the plot and might also have encouraged re-mineralization (because plants can increase substrate availability to soil micro-organisms) and uptake by plants of the ^{15}N immobilized by the soil organic matter, thus reducing the amount of ^{15}N in the soil. Results clearly showed that trees competed poorly with the understory and soil microbial population for the applied N. Despite low fertilizer recovery efficiencies, fertilization of those sites has been shown to be effective in improving plantation growth.

A second experiment examined the transformations and extractabilities of the residual N in forest floor 24 hours, 7 months, and 31 months after application. Net mineralization of total and applied N in a 42-day aerobic incubation was greatest in the

samples from the 24-h treatment followed by those from the 31-month treatment, indicating that immobilized ^{15}N was more remineralizable in the samples with ^{15}N labeled for a short term. The percentage of applied N found in the total net N mineralized ranged from 76.6 to 87.4%, 13.1 to 42.0% and 10.6 to 14.0% in samples from the 24-h and 7- and 31-month treatments, respectively, showing reduced availability of applied N with increased residence time.

The flush of N after chloroform fumigation is the result of N released from microbial cells killed by the fumigant. The flush of N was in the order of 24-h > 7-month > 31-month treatments for both total and applied N. The fumigation-extraction result showed that applied N was quickly immobilized by the microbial population in the soils. The N fertilizer was immobilized within hours after application by the microbial biomass. The N immobilized in soil organic matter had a low mineralization potential after one growing season.

The extractability of the residual ^{15}N was studied using 2 N KCl, 0.5 M K_2SO_4 , autoclaving, and acidic permanganate (of different strength) extractions and fumigation-extraction methods. Regardless of the extraction method used, a greater percentage of the total soil N was extracted from the 24-h than from the 7- and 31-month treatments ($p < 0.05$). However, there was no difference in the percent of total soil N extracted between the 7- and 31-month treatments. The amount of applied N extracted followed fairly closely that of the total N extracted. Regardless of the extraction method used, the only difference was between the 24-h and the other two treatments. There was consistently a greater percentage of applied N in the total N extracted for the 7-month than for the 31-month treatment. The greatest percentage of applied N extracted was in the 24-h treatment. The extractability ratio increased from samples with ^{15}N labeled for longer periods to samples with recent ^{15}N labeling. Extractability ratios were all greater than 1, indicating that the extracted fractions were always more enriched with ^{15}N than the bulk soil.

The incorporation of ^{15}N into the classical fulvic (FA), humic (HA) and humin fractions was also studied. In the FA fraction, a significantly greater percentage of the total recovered ^{15}N was in the 24-h treatment than in the other two treatments. However, there was no difference in ^{15}N distribution among the treatments for HA. In the humin fraction, a

significantly lower percentage of applied N was recovered in the 24-h treatment than in the other two treatments which was opposite to the ^{15}N distribution in the FA fraction. Most of the recovered ^{15}N was in the humin fractions for the 7-month and 31-month treatments, while for the 24-h treatment, most of the recovered ^{15}N was almost evenly distributed in the FA and humin fractions, with little in the HA fraction.

The results agree with the mineralization studies and showed that the extractability of residual ^{15}N was quickly reduced with increased residence time due to its incorporation into stable humin fraction. However, residual ^{15}N remained more extractable than the bulk soil N regardless of residence time.

A third experiment studied the dynamics of microbial biomass and N in old-growth CH forests, and in 3- and 10-year-old western red cedar plantations (Chang *et al.* 1995). Three forest floor layers: F, woody F (Fw) and H (humus) were sampled in May, July, August, and October of 1992. Microbial biomass C and N were relatively constant throughout the sampling period. Microbial C content was in the order: old-growth forests > 10-year-old plantations > 3-year-old plantations. Microbial N content was significantly greater in the old-growth forest than in the young plantations, but was not different between the plantations. Therefore, the hypothesis that the microbial biomass acted as a net sink in the 10-year-

old plantations by immobilizing N into the microbial N pool was rejected. Microbial C/N ratios were greater in the 10-year-old plantations than in the old-growth forests and in the 3-year-old plantations in H and F material, suggesting that the microbial biomass was also deficient in N. As a result, microbial competition for N may be more pronounced in 10-year-old plantations and may be a factor in the growth decline on CH sites. Extractable C and N, and mineralizable N were generally higher in the old-growth forests than in the 3-year-old plantations and higher in the 3-year-old than in the 10-year-old plantations. Tree and understory foliage in the 3-year-old plantations had higher N concentrations and lower C/N ratios than in the 10-year-old plantations.

Conclusions

- 1) Salal strongly competed for fertilizer N on CH cutovers.
- 2) Salal competition greatly reduced tree growth.
- 3) Incorporation of N into soil organic matter further reduced the availability of fertilizer N.
- 4) Microbial competition for N may contribute to the nutrient supply problems on CH cutovers.

Phosphorus forms of podzolic soils and their use by western red cedar

Barbara Cade-Menun

Summary of PhD thesis, Department of Soil Science, UBC, 1995

Soil chemistry, particularly P forms, were studied in uncut CH and HA stands, and on CH sites 0, 5 and 10 years after clearcutting and slashburning. There were 3 locations per forest type (CH, HA), and age post-burn (0,5,10), and 3 soil pits per site. LF, H and Bf horizons of the soil were sampled separately in each pit.

In the uncut CH and HA stands, there were no significant differences in moisture content, total C, total N, C:N, and extractable Ca, Mg, Al, Fe and Mn. The pH of the LF and H horizons was higher in CH soils than in HA. There were no significant differences between CH and HA forests in available P, total P, organic P, or C:P. There were no differences in Fe-, Al- or Ca-phosphates. The ^{31}P NMR spectra were also quite similar. The diversity of P forms in all horizons, and the persistence of the relatively labile orthophosphate diesters (which include phospholipids, RNA and DNA) down to the Bf horizon, were typical of soils with slow or restricted decomposition.

CH sites 10 years, 5 years and 2 months after slashburning were sampled and compared with uncut CH forests. There were no significant differences among the ages in any horizon for total C, total N or C:N. Field moisture was significantly higher in the uncut stands in the LF horizon, but there were no differences in the lower horizons. A significant increase in pH was seen in the LF horizon of the 2-month post-burn stands, the H horizon of the 2-month and 5-year sites, and the Bf horizon of the 10-year site. There were no significant differences in extractable Ca, Al or Fe. The 0, 5 and 10-year postburn sites had higher concentrations of extractable Mg in the LF and H horizons than the uncut stands. There was a significant decrease in

organically-complexed Fe and Al in the Bf horizon 10 years after burning.

Available P was significantly increased in the LF horizon of the 2-month sites and in the H horizon of the 0- and 5-year sites. Total P, C:P, and Fe-, Al- and Ca-phosphates were not different among the ages. There was a decrease in organic P in the Bf horizons 10 years after burning. The ^{31}P NMR spectra revealed a shift to predominantly inorganic forms in the LF horizon of the 2-month sites, but no changes in the lower horizons. In the 10-year samples, the spectra of the surface horizons resembled those of the uncut forests. However, there was so little P in the Bf horizons that reliable spectra could not be obtained.

The results of this study suggest that slashburning produces an immediate release of nutrients such as P and Mg, and an increase in pH. However, the destruction of organic matter in the surface horizons interrupts illuviation, reducing the transport of organic matter into the mineral horizons. This potential problem will be alleviated once a new organic layer is formed under the regenerated stand. In the old-growth stands, the mean depth to the Bf horizon is 25.4 cm, and the feeder roots are found mainly in the surface horizons. On the 10-year sites, the mean depth to the Bf horizon is 11.9 cm. The feeder roots may be going into mineral soils on these sites, where the organic material has been depleted.

A greenhouse study was also conducted to determine if western red cedar could use organic forms of P. Although there were problems with N enrichment from a fungicide used to produce mycorrhiza-free controls, the results clearly indicated that the cedar seedlings were obtaining P from organic sources. Phosphatase production was observed, but it was not possible to separate the

enzymes produced by the trees from those produced by mycorrhizae and other rhizosphere organisms.

The effect of extractants on P determination by ^{31}P NMR spectroscopy was also examined using five forest floor samples (Cade-Menun and Preston 1996). The extractants used were: 0.25 M NaOH, 1:6 soil to Chelex in water, 1:6 soil to Chelex in 0.25 M NaOH, and a 1:1 mix of 0.5 M NaOH and 0.1 M EDTA. Chelex + NaOH extracted 23 to 35% of the total P,

Chelex in water extracted 10 to 13%, NaOH alone extracted 22 to 34%, and NaOH + EDTA extracted 71 to 90%. There were also differences among extractants in the diversity of P forms detected and the sharpness of peaks. The results suggested that care must be taken when interpreting studies of P cycling in soils using ^{31}P NMR spectroscopy and when comparing studies using different extractants.

Conclusions

- Both cedar and hemlock on CH cutovers responded to scarification and to individual-tree fertilization at planting with NPK. The greatest response was to the combined treatment.
- Individual tree volumes were lower at 2500 stems/ha than at 1500. In fertilized plots, total tree volume was greatest at 2500 stems/ha in hemlock, and at 1500 stems/ha in cedar.
- Mixed conifer stands on CH cutovers that received salal removal (manual and chemical) and N+P fertilization responded greatly and have reached crown closure. Hemlock and spruce responded most to fertilization; cedar responded most to salal removal.
- There was no evidence that N availability was greater in closed stands with little salal cover.
- Concentrations of P in foliage and litter were still greater in fertilized plots 10 years after application.
- Applications of organic fertilizers (sewage sludge, fish silage, fish-wood compost and straw) increased height growth of conifers on CH cutovers. The magnitude and duration of the growth response was generally similar to that resulting from addition of chemical fertilizers. The largest responses (greater than responses to chemical fertilizer) followed additions of straw or a mixture of fish silage and pulp sludge.
- There was no evidence of N leaching into water-courses after application of fish silage, but there may have been losses of base cations (K, Mg, Ca) associated with nitrate movement in the soil.
- Rates of nitrogen fixation in woody debris are similar in CH and HA forests.
- Rates of denitrification are low in CH and HA forest floors and woody debris.
- There are high levels of tannins in leaves, litter, roots and senesced fruits of salal.
- Tannins are still present in CH humus after 5 years of salal removal.
- There is little change in the carbon chemistry of CH humus and woody debris (examined with ^{13}C NMR) after clearcutting and slashburning.
- Availabilities of P and Mg and pH increase after clearcutting and slashburning.
- There is little difference between CH and HA forests in P forms and availability.
- Cedar appears to use organic forms of P.
- Only 2-5% of fertilizer N is taken up by trees; this increases to 8-18% when salal is removed.
- Fertilizer N is quickly immobilized in the soil microbial biomass and is progressively incorporated into the stable fractions of humus.
- The C/N ratio of the soil microbial biomass increases after clearcutting and burning, indicating that these organisms also become N-deficient and may compete with plants for available N.

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