



Influence of the phenolic compound bearing species *Ledum palustre* on soil N cycling in a boreal hardwood forest

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Abstract

The effects of the understory shrub *Ledum palustre* on soil N cycling were studied in a hardwood forest of Interior Alaska. This species releases high concentrations of phenolic compounds from green leaves and decomposing litter by rainfall. Organic and mineral soils sampled underneath *L. palustre* and at nearby non-*Ledum* sites were amended with *L. palustre* litter leachates and incubated at controlled conditions. We aimed to know (i) whether *L. palustre* presence and litter leachate addition changed net N cycling rates in organic and mineral soils, and (ii) what N cycling processes, including gross N mineralization, N immobilization and gross N nitrification, were affected in association with *L. palustre*. Our results indicate that N transformation rates in the surface organic horizon were not affected by *L. palustre* presence or leachate addition. However, mineral soils underneath *L. palustre* as well as soils amended with leachates had significantly higher C/N ratios and microbial respiration rates, and lower net N mineralization and N-to-C mineralization compared to no *Ledum* and no leachates soils. No nitrification was detected. Plant presence and leachate addition also tended to increase both gross N mineralization and immobilization. These results suggest that soluble C compounds present in *L. palustre* increased N immobilization in mineral soils when soil biota used them as a C source. Increases in gross N mineralization may have been caused by an enhanced microbial biomass due to C addition. Since both plant presence and leachate addition decreased soil C/N ratio and had similar effects on N transformation rates, our results suggest that litter leachates could be partially responsible for plant presence effects. The lower N availability under *L. palustre* canopy could exert negative interactions on the establishment and growth of other plant species.

Introduction

The presence of plants has been shown to influence small-scale patterns of nutrient availability in various ecosystems by changing the quantity and quality of organic matter in the nearby soil (Chen and Stark, 2000; Smith et al., 1994). These effects are species-specific since plants differ in litter production quantity (Hobbie, 1992) and litter tissue chemical composition

(Bonan, 1990). Thus, soil underneath plants can have more organic matter and soil microbial activity compared to bare ground (Aguilera et al., 1999; Hook et al., 1991; Vinton and Burke, 1995) although in some cases plant presence had no effect on soil organic matter distribution (Jackson and Caldwell, 1993), or variation in organic matter was not related to variation in microbial activity (Chen and Stark, 2000; Jackson and Caldwell, 1993). Litter chemical traits such as C:N, lignin:N, and phenolic compound concentrations have been inversely related to decomposition, and thus to the release of inorganic nutrients by mi-

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crobes (Hättenschwiler and Vitousek, 2000; Hobbie, 1992).

Phenolic compounds may play a dominant role in controlling many aspects of plant–soil interactions, especially those related to organic matter dynamics and nutrient cycling (Kuiters, 1990; Northup et al., 1998; Schimel et al., 1996). One of the most characteristic properties of phenolic compounds, including the high molecular weight condensed tannins, is their capacity to form recalcitrant complexes with proteins and thus to alter the pool and form of nutrients (Hättenschwiler and Vitousek, 2000). When these complexes occur, mainly during senescence, leaf N is initially immobilized and gradually released in the course of decomposition (Kuiters, 1990). For instance, phenolic concentrations have been correlated with slower soil organic matter decomposition and turnover rates (Horner et al., 1988; Nicolai, 1988; Palm and Sanchez, 1990), higher N immobilization rates in litter (Gallardo and Merino, 1992) and lower N mineralization rates in litter and organic soil (Fox et al., 1990; Northup et al., 1995; Palm and Sanchez, 1990, 1991; Schimel et al., 1996). Phenolic compounds also have been shown to stimulate microbial population growth, microbial respiration and N immobilization (Blum, 1998; Blum and Shafer, 1988; Boufalis and Pellissier, 1994; Shafer and Blum, 1991; Sparling et al., 1981; Sugai and Schimel, 1993), and to inhibit microbial respiration (Boufalis and Pellissier, 1994; Schimel et al., 1996) and nitrification (Rice, 1984). The presence and importance of each of these effects seems to depend on the molecular size, structure and concentrations of phenolic compounds (Boufalis and Pellissier, 1994; Schimel et al., 1996). Thus, condensed tannins seem more involved in slowing degradation processes when they form stable complexes with proteins and inorganic N (Northup et al., 1995; Palm and Sanchez, 1990), whereas lower molecular weight phenolic acids are easily degraded by microbes (Blum, 1998; Blum and Shafer, 1988; Sugai and Schimel, 1993). Because the overall effects of phenolic compounds on N cycling are a decrease in net N mineralization, either by decreasing gross mineralization or nitrification, or by increasing immobilization, the use of ^{15}N pool dilution techniques for estimating gross N transformation rates is a useful way to elucidate what processes are being affected (Hart et al., 1994).

The presence and concentration of plant phenolic compounds, and thus the potential effects of their release to the nearby soil, depends both on the plant species and the abiotic environment (Bryant et al.,

1983; Hamilton et al., 2001; Peñuelas and Estiarte, 1998). High amounts of phenolic compounds can be released by rainfall from green foliage and decomposing litter (Gallet and Pellissier, 1997; Harborne, 1997; Inderjit and Mallik, 1996a; Kuiters and Sanrink, 1986), thus affecting nutrient cycling. The ecological relevance of phenolic compounds can be of especial interest in N-limited ecosystems with slow decomposition, such as boreal ecosystems, where slow growing species with high concentrations of carbon-based secondary compounds predominate. However, it is still not clear whether changes in N cycling related to plant phenolic compounds can be found in natural conditions since these effects have been mainly tested in laboratory experiments with individual compounds.

Ledum palustre (Labrador tea) is a late successional evergreen shrub widely distributed in boreal ecosystems that readily leaches high concentrations of soluble phenolic compounds into water. We selected this species in order to study whether leaf leachates could be responsible for the effects of plant canopy on N cycling under natural conditions. Organic matter content, soil respiration and net N mineralization were measured in organic and mineral soil horizons and gross N mineralization was also measured in mineral soils sampled underneath *L. palustre* in a hardwood forest dominated by *Populus tremuloides* and *Betula neoalaskana*. Soils were amended with *L. palustre* litter leachates and incubated in the laboratory. Because mineral soil is also influenced by the overlying organic horizon we also characterized some simple indices of the organic horizon carbon quality (lignin, cellulose and condensed tannins) to look for differences in likely decomposability.

Our objectives were (i) to determine whether *L. palustre* presence and *L. palustre* leachates addition changed soil N availability, and (ii) to determine the specific N cycling processes that were affected, including changes in mineralization, nitrification or immobilization. We wanted to know whether *L. palustre* leachates could be a mechanism through which plant presence would impact N cycling. Besides affecting site chemical quality, plant secondary compounds of some ericaceous species, including *Ledum groenlandicum*, have been shown to impede the establishment and growth of other plants species (Inderjit and Mallik, 1996b). In this study, we also discuss whether the chemical interactions between *L. palustre* and other plants could be related to decreases in soil N availability for plant uptake or otherwise to direct effects through allelopathic processes.

Materials and methods

Site description

The study sites were located in a south-facing mixed hardwood forest in the Caribou Poker Creeks Research Watershed near Fairbanks, Alaska, USA (lat. 65.16 N, long. 147.5 W). Soils are coarse-silty well drained with no permafrost, mixed, superactive (CEC/clay % >0.6), frigid (mean annual temperature at 50 cm >0 °C but <8 °C, and typic Dystrycyrypt belonging to the Olnes series). Dominant species are aspen (*Populus tremuloides*), alaskan paper birch (*Betula neoalaskana*) and black spruce (*Picea mariana*). *Ledum palustre* is a late successional evergreen shrub that forms moderately dense patches in relatively open areas of the forest. During summer 1999 we randomly selected five *L. palustre* patches and five adjacent non-*Ledum* plots. A 1-m² quadrat was located in the middle of each *L. palustre* patch in order to avoid edge effects, and in the selected non-*Ledum* sites. We avoided areas near black spruce trees because they can also leach phenolic compounds into rainfall. Annual new growth biomass production of *L. palustre* was estimated by sampling shoot new growth within the 1-m² quadrat. Average density (annual production by area) of *L. palustre* was 35.58 ± 6.64 g m⁻² and it ranged from 18.75 to 59.91 g m⁻² in the different plots although no significant differences were detected among sites. Biomass of other understory species in both *Ledum* and non-*Ledum* sites was determined by sampling all rooted plants in three 20-cm² quadrats in each plot. Feathermoss (*Hylocomium splendens* and *Pleurozium schreberi*), *Polytricum* sp and *Vaccinium vitis-idaea* were the dominant understory species, followed by *Lycopodium annotinum*, *Lycopodium complanatum*, *Cornus canadensis*, *Epilobium angustifolium* and *Equisetum silvaticum*. Total understory biomass other than *Ledum palustre* (mean and std error, $n = 5$) was 49.57 ± 6.63 g m⁻² for *Ledum* sites and 28.49 ± 14.95 g m⁻² for non-*Ledum* sites. There were no statistical differences between *Ledum* and non-*Ledum* sites for total biomass (Table 1) as well as for biomass of each individual species (data not shown).

Preparation of *L. palustre* leachates

In order to study the global effects on nutrient dynamics of the high variety of phenolics and other carbon-based compounds present in plants, we used

foliage leachates instead of purified phenolic compounds. We collected leachate from litter rather than green leaves because release of soluble phenolic compounds from litter has been documented to be larger than from leaves (Kuiters, 1990). Standing dead leaf litter of *L. palustre* corresponding to the previous year's production was sampled at the study sites described above. Leachate was obtained by shaking fresh litter (25 g equivalent DW) in 1 L distilled water at room temperature for 24 h (Zackrisson and Nilson, 1992) and filtered through Whatman 42 filter paper. Distilled water was used as a control. Leachates were analyzed for total phenolics by the Folin-Ciocalteu method (Marigo, 1973) using gallic acid as a standard (767 mg L⁻¹), and for dissolved organic carbon (DOC) (1222 mg L⁻¹) (Shimadzu TOC 5000).

Soil sampling and incubations

The organic horizon (Oe and Oa) and the top 7 cm of mineral soil were sampled in three different locations inside the 1-m² quadrat, and samples were bulked together by horizon within each site. Organic and mineral horizons were sieved through 5.6- and 2-mm mesh, respectively, and kept at 4 °C before incubating for 1 week. pH of mineral soil (4.9) was not significantly different between *Ledum* and non-*Ledum* sites.

Soils sampled under *L. palustre* and in nearby non-*Ledum* sites were incubated in the dark at 15 °C for 30 days. Organic (20 g) and mineral soil (50 g fresh weight) were placed into 250-mL jars fitted with septa to allow headspace samplings. Samples were amended with litter leachate or distilled water (4 and 6 mL for organic and mineral soils, respectively) just before starting the incubation. Additional distilled water was added to the soils to bring moisture to field capacity, which had been determined on a subsample using a modified procedure from Tan (1996). Soil respiration was measured every week by analyzing CO₂ accumulated in the jars by a Gas Chromatograph (Shimadzu GC-14A) fitted with a methanizer and flame ionization detector. After each measurement, jars were opened, ventilated and re-wetted with distilled water to maintain constant weight. Potential net N mineralization rates were calculated as the difference between initial and final KCl extractable ammonium and nitrate concentrations. Soils (15 g FW) were shaken with 75 mL of 2 M KCl for 1 h and filtered. Filtrates were analyzed for ammonium and nitrate using a modified Technicon AutoAnalyzer II system. Net N mineraliz-

Table 1. Means \pm SE and *t*-test comparing understory biomass other than *L. palustre*, depth of organic horizon and organic soil chemical analyses sampled in non-*Ledum* sites and *Ledum* sites (under *L. palustre*). *N* = 20 for organic horizon depth and *n* = 5 for the other variables ($\alpha=0.05$). No significant differences were found for any variable. nd = not detectable

	Non- <i>Ledum</i> site (Mean \pm SE)	<i>Ledum</i> site (Mean \pm SE)	<i>F</i>	<i>P</i>
Understory biomass (g m ⁻²)	28.49 \pm 14.95	49.57 \pm 6.63	2.56	0.19
Depth organic horizon (cm)	5.06 \pm 0.31	4.97 \pm 0.31	0.06	0.8
Neutral detergent fiber (NDF) (%)	61.9 \pm 1.1	66.08 \pm 3.14	1.34	0.3
Acid detergent fiber (ADF) (%)	49.86 \pm 1.37	53.83 \pm 2.9	0.99	0.37
Hemi-cellulose (%)	12.04 \pm 0.69	12.26 \pm 0.48	0.13	0.73
Cellulose (%)	12.51 \pm 1.36	13.89 \pm 1.58	0.24	0.64
Lignin (%)	21.82 \pm 1.04	22.05 \pm 1.65	0.01	0.92
Condensed tannins (mg g ⁻¹ DM)	nd	nd	–	–

ation rates were expressed per unit of soil dry weight and per unit organic C. Total C and N were analyzed at the end of incubation using a CNS analyzer (LECO Model 2000). Subsamples of the organic horizon were dried at 65 °C, ground and analyzed at the University of Alaska Palmer Research Station for cellulose, hemicellulose and lignin using the Van Soest procedure (Goering and Van Soest, 1970). Condensed tannin concentrations were analyzed by the proanthocyanidin method (Waterman and Mole 1994).

Gross N transformation rates

Gross rates of N mineralization and ammonium consumption in mineral soils sampled under *L. palustre* and at the non-*Ledum* sites were determined by ¹⁵NH₄⁺ isotope dilution (Hart et al., 1994). A solution containing 6 mL distilled water and 3 mL of ¹⁵NH₄⁺ (containing 0.05 mg ¹⁵N 99 atom %) were added to 50 g of soil sampled at the *Ledum* sites and at the non-*Ledum* sites. For the soils sampled at the non-*Ledum* sites an additional treatment replacing the distilled water by *L. palustre* leachate was conducted. Five replicates per type of soil and treatment were used. In all samples, additional distilled water was added until field capacity when necessary. Samples were homogenized for 5 min and thereafter 5 g of soil was immediately extracted with 2 M KCl in order to obtain initial inorganic N concentrations and ¹⁵N recovery. The rest of the sample was incubated at 15 °C and extracted after 24 h. Initial and final times of incubation were recorded. Inorganic N was transferred

from KCl extracts to a cut filter paper Whatman 3 using the ammonia diffusion procedure (Holmes et al., 1998). A disk of acidified filter paper wrapped using a Teflon membrane was added to the KCl extracts together with MgO. The MgO makes the solution basic, causing the NH₃ vapor to be released and captured by the acidified filter disk. After 6 days of diffusion the filter disks were dried in a desiccator over concentrated H₂SO₄ and analyzed for ¹⁵N atom % in a continuous flow isotope ratio mass spectrometer (PDZ Europa Geo 20/20, Cheshire, UK). Gross N transformation rates were calculated from changes in inorganic N concentrations and changes in ¹⁵N atom % during the incubation following the methods and equations in Kirkham and Bartholomew (1954). We assumed the background ¹⁵N enrichments to be 0.37% atom % ¹⁵N (Hart et al., 1994). Since no net nitrification was detected during the incubations, we assumed that ammonium consumption represented ammonium immobilization.

Statistical analyses

T-tests were conducted to test differences in understory biomass, depth of organic horizon and organic soil chemical analyses between non-*Ledum* sites and *Ledum* sites. A repeated measures ANOVA using absence or presence of *Ledum* as a repeated variable and treatment (water or leachate) as a factor was conducted for organic C, organic N, C/N ratio, net N mineralization and N-to-C mineralization. Repeated measures ANOVA was chosen because each of the five

non-*Ledum* sites was paired with a *Ledum* site. A two-trial repeated measures ANOVA was conducted for C mineralization using absence or presence of *Ledum* and date of sampling as repeated variables, and treatment (water or leachate) as a factor (see table legends for more details). *L. palustre* biomass density (annual production by area) was used as a covariant when indicated. $\alpha = 0.05$ was used. Data were analyzed for normality and no transformations were necessary in any case. All analyses were performed using Statistica '99 Edition (Statsoft Inc., Tulsa, USA).

Results

Organic horizons sampled underneath *L. palustre* had 3.6 and 7.9% lower organic C (soils unamended and amended with *L. palustre* leachates, respectively) than non-*Ledum* soils (Figure 1). No differences in neutral detergent fiber (NDF), acid detergent fiber (ADF), lignin, hemicellulose or cellulose were found between organic horizons under or away from *L. palustre*. Condensed tannins were not present in any site (Table 1). No differences were found for total organic C and N in mineral soils underneath *L. palustre* and non-*Ledum* sites. However, C/N ratio was significantly higher in the soils under *L. palustre* and those amended with leachate (Figure 1).

No nitrification was detected in organic or mineral soils. Neither the presence of *L. palustre* in the field nor leachate addition in the lab had any significant effects on net N mineralization of organic soils (Figure 2, Table 2). In mineral horizons, net N mineralization on a dry weight basis was 73.4 and 264.3% significantly lower (soils unamended and amended with *L. palustre* leachate, respectively). Leachate addition decreased net N mineralization in soils from non-*Ledum* plots (by 109.7%) and from plots underneath *L. palustre* (234.2%). The effects were similar in both soils as no significant interactions were found between treatment and plant presence (Figure 2, Table 2). Total CO₂ production was 27.9 and 27.8% higher in soils sampled under *L. palustre* compared to non-*Ledum* sites although this trend was marginally significant (Figure 2, Table 2). Although no effect of leachate addition was found for mineral soil respiration during the incubation period, the significant interaction between treatment and incubation time showed that leachate addition increased soil respiration by 48.1 and 71.5% control and *L. palustre* soil, respectively, during the first week of incubation (Figure 3, Table 2). Both

soils sampled under *L. palustre* and in non-*Ledum* sites had similar responses to leachate addition for net N mineralization, C mineralization and N-to-C mineralization.

L. palustre sites had 310.8% higher marginally significant gross N mineralization rates and 110.6% higher gross ammonium immobilization rates compared to non-*Ledum* sites ($P < 0.1$) (Figure 4). When leachates were added to non-*Ledum* site soils, and ammonium consumption increased 151.5% compared to unamended soils ($p < 0.05$). No significant differences were found for leachate addition in ammonium consumption rates (Figure 4). We found no differences between the effects of plant presence and leachate addition on either gross N mineralization or ammonium consumption (Figure 4).

Discussion

Effects of L. palustre presence and leachate addition on N cycling

The results show that *L. palustre* canopy and litter leachate addition changed N cycling in mineral soils. Although we found no differences in organic matter content between mineral soils sampled under *L. palustre* and soils sampled in nearby non-*Ledum* sites, both plant presence and leachate addition increased soil C/N and microbial respiration, and decreased net N mineralization. Gross N mineralization and ammonium consumption generally tended to be more rapid in soils associated with *L. palustre* compared to non-*Ledum* and no leachates soils. However, the only statistically discernible effects were those of plant presence on gross N mineralization and leachate addition on ammonium consumption. Changes in mineralization were entirely explained by changes in ammonium concentrations as nitrate pools were always low. *L. palustre* presence as well as leachate addition produced no detectable changes in net N mineralization and microbial respiration in the organic horizon, probably because of high variability within sites and small sample size. Since the broad indices of chemical composition of organic soil C compounds (fibers, lignin, hemicellulose and cellulose), depth of organic horizon and understory biomass were similar between *Ledum* and non-*Ledum* sites, it is likely that the described effects on mineral soil are caused mainly by direct effects of *L. palustre* presence.

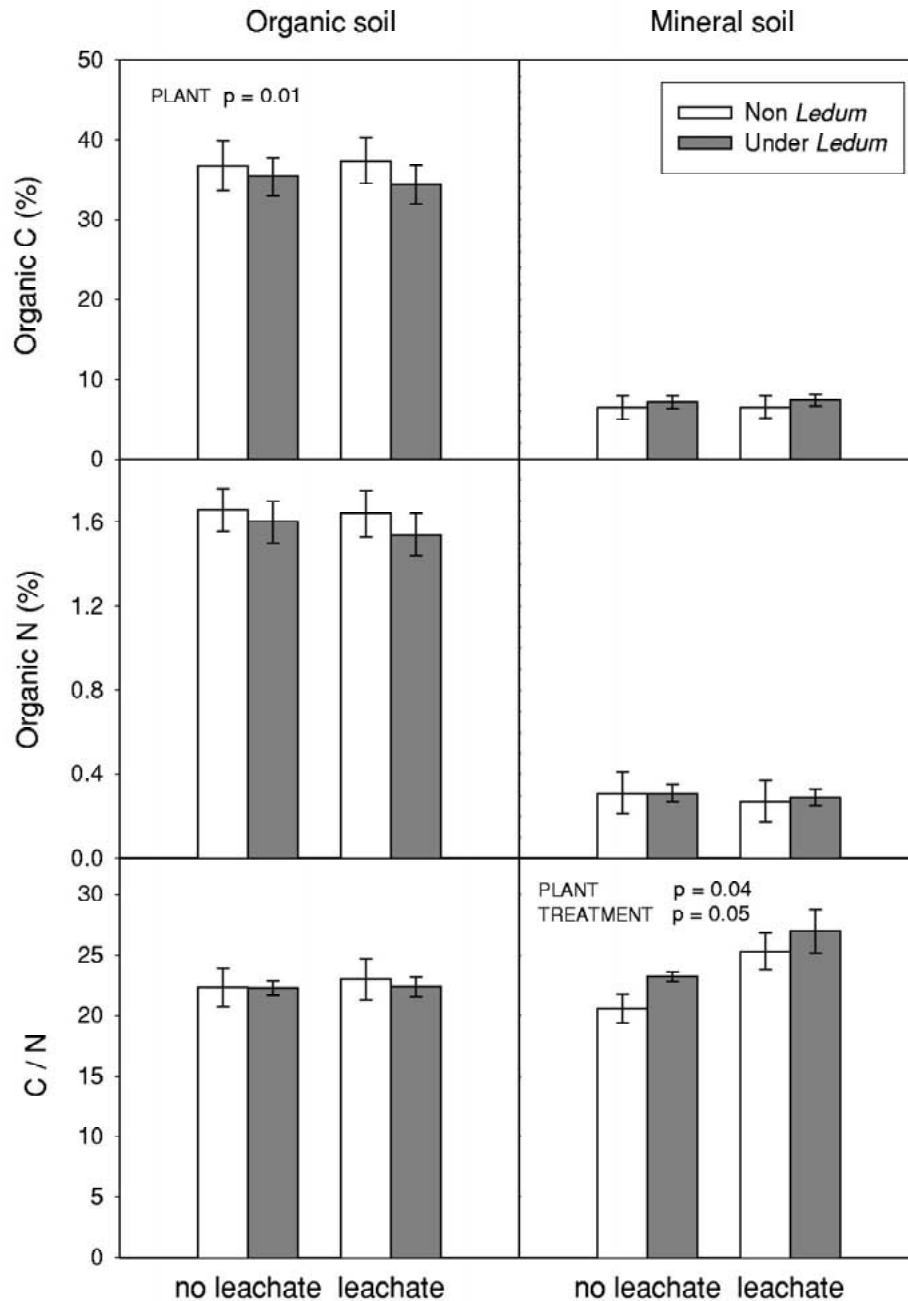


Figure 1. Effects of *L. palustre* presence and leachate addition on organic C, organic N and C/N ratio of organic and mineral soils. Values represent means and SE. We tested statistical differences for plant presence or absence ('PLANT') and amendments with leachate or distilled water ('TREATMENT') conducting a repeated measures ANOVA for PLANT ($n = 5$). *L. palustre* biomass density was used as a covariant. Significant P levels are shown.

The higher rates of ammonium immobilization and C mineralization and lower ratio of N-to-C mineralization suggest that addition of C compounds from *L. palustre* canopy and leachates stimulated microbial

activity when microbes used them as a C substrate. Since a lower ratio of N-to-C mineralization indicates a low supply rate of N to the microbial community (Schimel et al., 1996), soils associated with

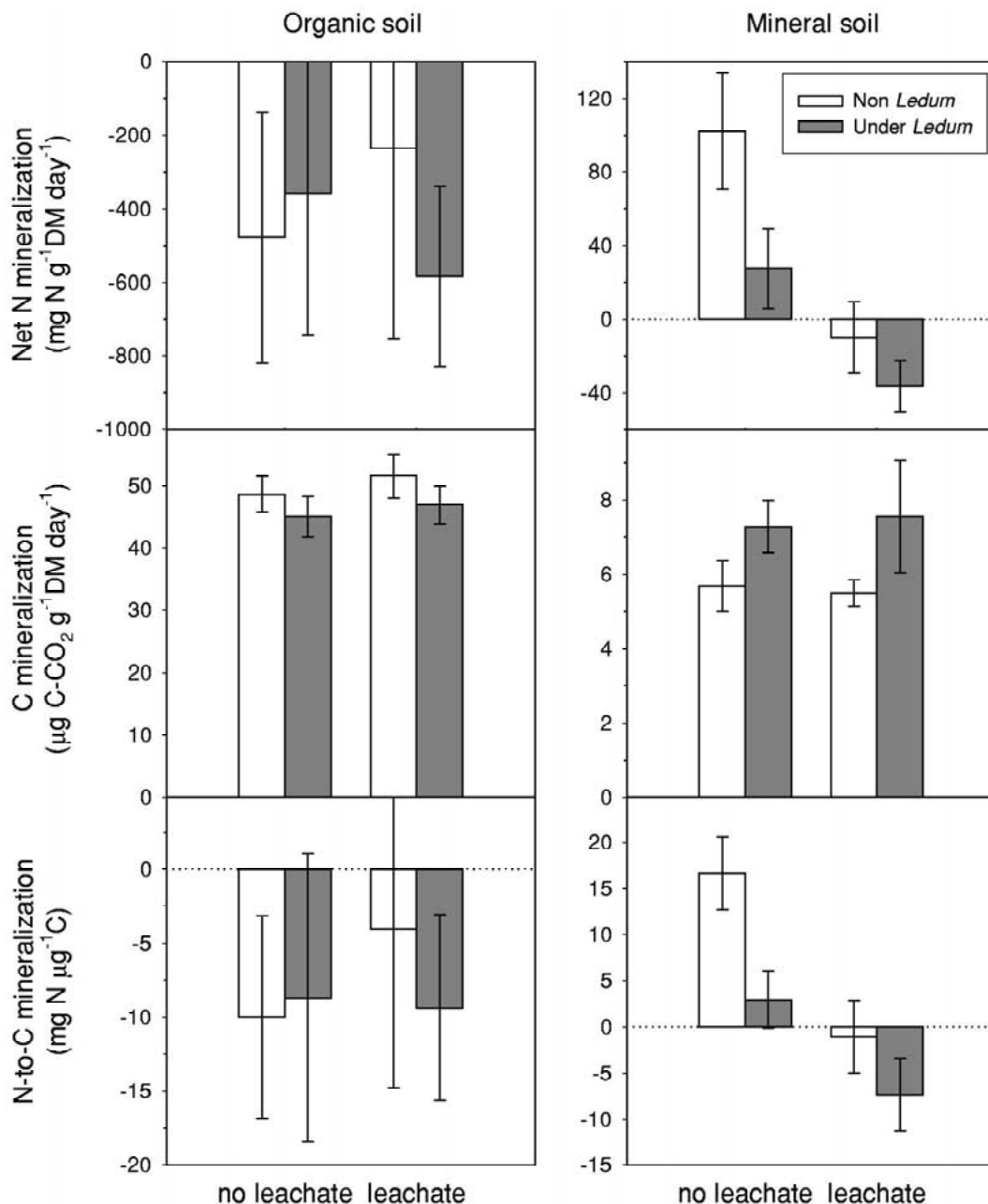


Figure 2. Net N mineralization, soil respiration and N-to-C mineralization of organic and mineral soils sampled under *L. palustre* and in nearby control sites, and amended with distilled water (control) or *L. palustre* leachate. Values represent means and SE ($n = 5$).

L. palustre would enhance inorganic N immobilization. Several studies have found that low molecular weight phenolic compounds can be readily metabolized by microbes, thus stimulating soil respiration and microbial growth (Blum and Shafer, 1988; Bouffalis and Pellissier, 1994; Schimel et al., 1996; Sparling et al., 1981). Some low molecular weight compounds

are metabolized as fast as with sugars such as glucose (Sugai and Schimel, 1993). Our results partially support the lability of compounds present in leachates because soil respiration increased only during the first week following leachate addition. Increases in N immobilization induced by other C compounds released from the canopy such as carbohydrates (Magill and

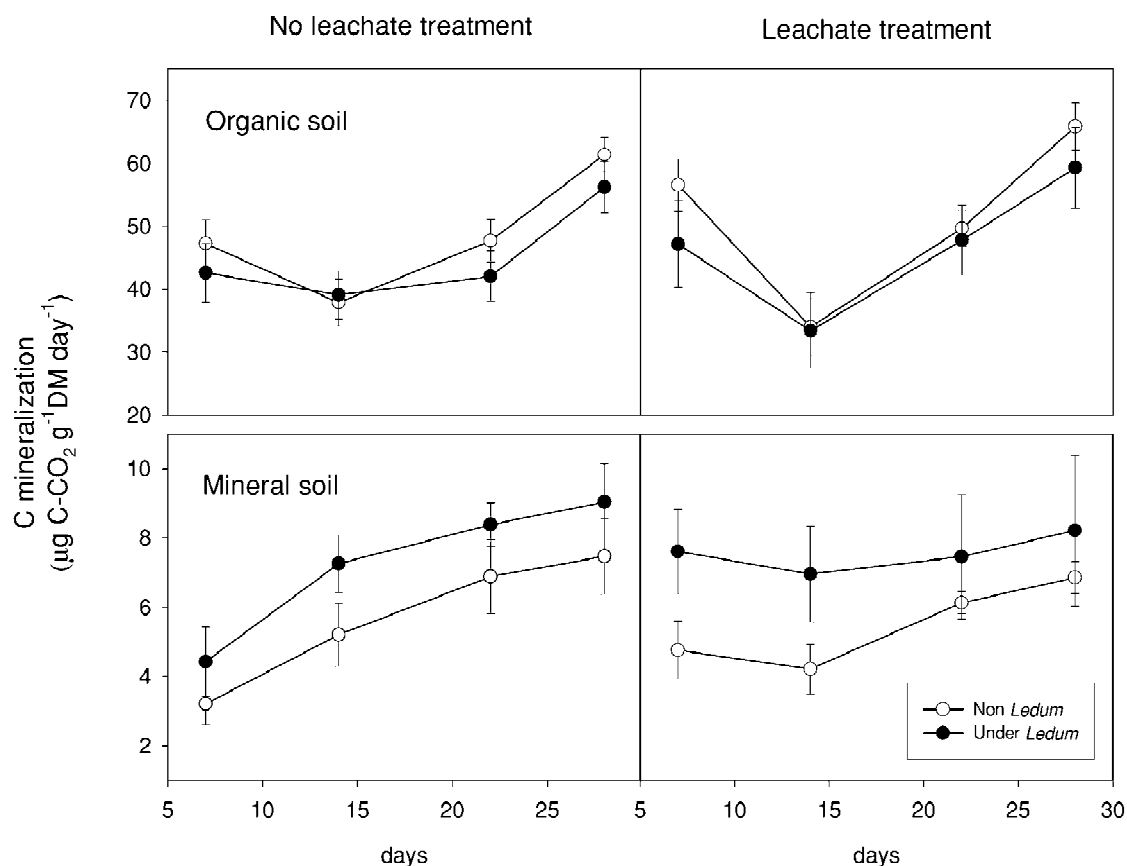


Figure 3. Changes in soil respiration during a 1-month incubation for organic and mineral soils sampled under *L. palustre* and in nearby control sites, and amended with distilled water (control) or with litter leachate. Values represent means and SE ($n = 5$).

Aber, 2000; Michelsen et al., 1995) or monoterpenes (Mackie and Wheatley, 1999; Vokou et al., 1984) cannot be dismissed. However, leaching of water-soluble compounds such as phenolics should exceed those components that are only slightly soluble in water, such as terpenes and lignin (Horner et al., 1988).

Soils associated with *L. palustre* also exhibited elevated rates of gross N mineralization. This enhancement also could be driven by the use of C compounds as a substrate by microbes. Additions of C are expected to increase microbial biomass (Bradley et al., 1997) and thus to stimulate microbial turnover increasing gross mineralization as well as immobilization rates (Clein and Schimel, 1995). While our results are consistent with an important role of foliar leachates in governing rates of N mineralization, they do not exclude the possibility that differences in chemistry and decomposition rates of foliar and root litter may also govern N mineralization rates.

The expected decreases in rates of gross N mineralization and decomposition when condensed tannins form complexes with proteins, which has been identified as the major link between phenolic compounds and nutrient cycling (Hättenschwiler and Vitousek, 2000), were not found in our study. Although we cannot conclude that this process was not taking place, it was small compared to the net positive effect of *L. palustre* presence and leachate addition on gross N mineralization. Proteins linked to phenolic compounds are likely to be less degraded by microbes than free organic N (Horner et al., 1988; Palm and Sanchez, 1990) and this may lead to conservation of N in the ecosystem by delaying decomposition and mineralization and thus slowing N losses (Northup et al., 1998). If this process enhances N conservation in the soil, higher phenolic compound concentrations would be selected during evolution in those plants growing on a N-limited soil compared to plants with no nutrient restriction (Bending and Read, 1996; Northup et al.,

Table 2. Statistical analyses for net N mineralization and respiration of organic and mineral soils sampled under *L. palustre* and in a nearby non-*Ledum* site, with PLANT as the independent variable. Soils were amended with leachate or with distilled water (TREATMENT). We conducted a repeated measures ANOVA (PLANT) for net N mineralization and N-to-C mineralization, and two-trial repeated measures ANOVA (PLANT and TIME) for soil respiration. TIME variable is referring at date of sampling along a 1-month incubation period. Plant density was used as covariate, $n = 5$ ($\alpha=0.05$), $P < 0.05$ are highlighted in bold

	Net N mineralization		C mineralization		N-to-C mineralization	
	$mg\ N\ g^{-1}\ DM\ day^{-1}$		$\mu g\ C-CO_2\ g^{-1}\ DM\ day^{-1}$		$mg\ N\ \mu g^{-1}\ C$	
	F	P	F	P	F	P
<i>Organic soil</i>						
Treatment	0.00	0.98	0.28	0.61	0.17	0.69
Plant	0.1	0.75	2.97	0.13	0.14	0.72
Time	-	-	25.98	<0.001	-	-
Treatment \times Plant	0.42	0.53	0.32	0.58	0.25	0.62
Treatment \times Time	-	-	1.77	0.18	-	-
Plant \times Time	-	-	1.47	0.25	-	-
Treatment \times Plant \times Time	-	-	0.42	0.74	-	-
<i>Mineral soil</i>						
Treatment	14.03	0.007	0.002	0.96	10.31	0.01
Plant	6.16	0.03	3.46	0.09	11.07	0.01
Time	-	-	10.78	<0.001	-	-
Treatment \times Plant	1.4	0.26	0.05	0.81	1.48	0.25
Treatment \times Time	-	-	3.86	0.02	-	-
Plant \times Time	-	-	0.56	0.64	-	-
Treatment \times Plant \times Time	-	-	0.47	0.7	-	-

1995, 1998). These processes could be important in low N availability ecosystems with high soil C/N, such as boreal ecosystems. However, conservation of N in the soil through decreases in gross N mineralization was not observed in our study.

L. palustre presence versus leachate addition

Could the lower N availability in soils underneath *L. palustre* than in non-*Ledum* soils be explained by the effects of soluble C compounds leached from litter? Since both plant presence and leachate addition increased C/N ratio and had similar effects on N transformation rates, our results suggest that litter leachates could be partially responsible for the plant presence effects. Although leachates released from *L. palustre* only stimulated microbial activity for a few days, they may have longer-term effects under field conditions. Only a small percentage (5.9 and 11.3% for non-*Ledum* and *Ledum* soils, respectively) of DOC added with the leachates was actually respired dur-

ing the first week of incubation. If we assume that one-third of the metabolized C was incorporated into microbial biomass while the rest was respired, and the increase in C mineralization of amended soils compared to no leachates soils was a result of leachate addition, 9 and 15% of DOC, in non-*Ledum* and *Ledum* soils respectively, was readily metabolized by microbes within the first week. These percentages are much lower than those found by Sugai and Schimel (1993), who estimated that 90% of two phenolic acids were metabolized within 4 h. Our low percentages could indicate the presence of more recalcitrant compounds in *L. palustre* leachates. Thus, a constant input of C compounds from *L. palustre* may not be required in order to change soil N cycling over the long term because those compounds not only affected short-term microbial activity but also increased the C/N of organic matter and thus the fate of N. Inderjit and Mallik (1997) also found changes in soil chemical properties after *Ledum groenlandicum* litter amendments, includ-

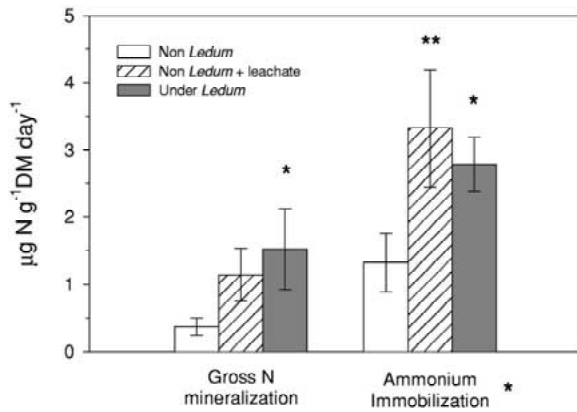


Figure 4. Gross N mineralization and ammonium immobilization rates from mineral soils sampled under *L. palustre* and in nearby control sites, and from control site soils amended with *L. palustre* leachates. Values represent means and SE. A *t*-test was conducted in order to test significant differences for treatment ($n = 5$). *L. palustre* biomass density (annual production by area) was used as a covariant. Post-hoc comparisons were conducted using a LSD-test. Significant levels: * $P < 0.1$, ** $P < 0.05$. For post-hoc comparisons significant levels are referring to differences between control and the other treatments.

ing increases in organic matter, total phenolics, K and PO_4 .

Negative interactions among plants through carbon-based secondary compounds: changes in N cycling or allelopathy?

Several arctic species have been shown to inhibit plant growth, root elongation and seed germination of other species through the release of carbon-based secondary metabolites (Inderjit and Mallik, 1996b; Nilsson and Zackrisson, 1992). There is some controversy about whether those effects are caused by toxic effects of secondary metabolites (termed 'allelopathy'), or by indirect processes through changing soil nutrient availability, or both (Michelsen et al., 1995; Wardle and Nilsson, 1997). Few studies have described the allelochemical mechanisms of action (Peñuelas et al. 1996; Whitehead et al., 1982). We found a lower net N mineralization and thus lower N availability in mineral soils under *L. palustre* canopy as a result of increasing microbial activity. Because the organic horizon had net N immobilization, as is generally found in arctic soils during the growing season (Jonasson et al., 1993; Klingensmith and Van Cleve, 1993), differences between mineral soils associated and non-associated with *L. palustre* could be important in determining plant N availability. This study, thus, supports the hypothesis that potentially

negative interactions among plants could be caused by changes in nutrient dynamics (Michelsen et al., 1995; Schmidt et al., 1997) although allelopathic effects cannot be excluded. Changes in N cycling through the release of C compounds could be a general process for many species. However, not all studies found a lower N availability under plant canopy since more organic matter may accumulate (Castells and Peñuelas, submitted). The dilemma of reduced plant growth mediated by secondary metabolites through chemical interactions among plants or through changes in soil fertility remains unresolved. Depending on the predominant processes in natural ecosystems, the ecological and evolutionary consequences would be rather different. Since allelopathy requires a species-specific coupled system of secondary metabolites and their targets, this process could be less widespread than changes in soil N availability through fueling soil microbial activity. Ecologically, the latter could have a greater potential impact on the ecosystem because changing soil quality may in turn modify plant species establishment and successional dynamics (Clein and Schimel, 1995). Further research is needed to understand the importance of direct and indirect effects of plant species interactions in ecosystems.

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