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Nitrogen isotope tracer acquisition in low and tall birch tundra plant communities: a 2 year test of the snow–shrub hypothesis

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Abstract Deciduous shrub density and landcover are increasing across many areas of the Arctic. Shrub growth may be promoted by a snow–shrub feedback whereby relatively tall shrubs accumulate deeper snow, raising winter soil temperature minima, increasing microbial activity, and enhancing soil solution nitrogen (N). Although there is good evidence for the above components of the hypothesis, it has not yet been determined if shrubs can access the elevated N pool generated by deepened snow. We added isotopic N tracer (^{15}N) in late summer to control and snowfenced low birch hummock tundra to test the influence of deepened snow on N cycling. Furthermore, tracer was added to tall birch tundra to compare N cycling in low and tall shrub ecosystems that have the same species composition. Experimentally deepened snow in low birch tundra did not significantly affect ^{15}N uptake by shrubs or any other species 2 years after the tracer addition. However, there were strong differences between the low and tall birch ecosystems, with the deciduous shrubs and graminoids accumulating more ^{15}N than the evergreen shrubs in the relatively productive tall shrub site, and vice versa in the relatively infertile low birch site. The greater ^{15}N acquisition by birch in the more fertile site, together with the absence of a deepened snow effect on ^{15}N acquisition by any species in the low birch hummock ecosystem, suggest that climate-change induced increases in birch shrub growth and expansion across the landscape will tend to occur most rapidly in and around existing tall birch shrub patches.

Keywords Arctic · Increased snow · Isotope ^{15}N · Shrub expansion · Snow–shrub feedback hypothesis · Winter processes

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Introduction

Recent satellite imagery indicates that a vegetation transition coinciding with climate warming may be occurring across the Arctic (Goetz et al. 2005; Forbes et al. 2010; Beck and Goetz 2011; Xu et al. 2013). Repeat aerial photography, plot-level monitoring, and experimental manipulations suggest that increased growth and expansion of the deciduous shrubs alder, willow, and birch is a major component of this vegetation transition (Chapin et al. 1995; Bret-Harte

et al. 2001; Tape et al. 2006; Myers-Smith et al. 2011; Tremblay et al. 2011; Elmendorf et al. 2012a; Elmendorf et al. 2012b; Ropars and Boudreau 2012; Tape et al. 2012). Increased shrub cover is expected to reduce both winter and summer albedo, thereby providing a positive feedback to regional warming (Chapin et al. 2005; Sturm et al. 2005). Furthermore, increased shrub cover and associated changes in litter quality may shift tundra ecosystem carbon balance, resulting in either a positive or negative feedback to atmospheric carbon dioxide concentrations depending on whether net primary production or ecosystem respiration is most affected (Shaver et al. 1992; Hobbie et al. 2000).

The Snow–Shrub Feedback Hypothesis predicts that relatively tall dense shrub areas preferentially accumulate snow, resulting in higher soil temperature minima, increased microbial decomposition of soil organic matter (SOM), and enhanced nutrient supply to those shrubs (Sturm et al. 2001; 2005). Since fertilization studies indicate that plant growth (especially birch) in many low arctic sites is strongly nutrient-limited (Jonasson et al. 1999; Mack et al. 2004; Zamin and Grogan 2012) the snow–shrub feedback may contribute to the observed increases in growth and expansion of deciduous shrubs. Field studies support many components of the hypothesis. Shrub tundra communities accumulate deeper snow than lower stature vegetation (Sturm et al. 2001, 2005), and experimentally deepened snow elevates soil temperature minima (Schimel et al. 2004; Nobrega and Grogan 2007; Buckeridge and Grogan 2008), enhancing winter soil microbial activity (Schimel et al. 2004; Nobrega and Grogan 2007; Grogan 2012) and N mineralization rates (Schimel et al. 2004). Furthermore, in concurrent research in the same snow manipulation plots as used here, Buckeridge and Grogan (2010) demonstrated that deeper snow increases late thaw pulses of organic and inorganic nitrogen (N) and phosphorus that occur towards the end of snowmelt when soils are water-saturated and very close to 0 °C. Thus, the applicability of the Snow–Shrub Feedback Hypothesis now rests on determining if deciduous shrubs can preferentially acquire the additional nutrients that are made available in the soil as a result of deepened snow.

Deepened snow alone may not be the only mechanism contributing to enhanced N availability in the Snow−Shrub Feedback Hypothesis. Apart from preferential snow accumulation, tall shrub vegetation may also have distinct biological and chemical characteristics that promote its own growth. For example, the more easily decomposable and larger quantities of litter associated with tall versus low shrub vegetation communities may be as important as snow depth in determining organic matter decomposition and N mobilization rates over winter (Buckeridge and Grogan 2008; Hobbie 1992; Nadelhoffer et al. 1991).

Ultimately, as the arctic climate warms and soil N availability is enhanced, the extent to which low stature and relatively scarce deciduous shrubs rise to biomass dominance within tundra plant communities will depend on their ability to preferentially acquire and utilize N for growth (McKane et al. 2002). Chronic high level fertilization can clearly promote transitions from low to tall shrub tundra (Chapin et al. 1995; Mack et al. 2004), but differing *naturally occurring* levels of soil fertility may also influence plant community responses to increased N availability. Species differ in physiological capacity for nutrient uptake (Grogan and Jonasson 2003), in timing of nutrient uptake (Larsen et al. 2012), in affinity for different N forms (McKane et al. 2002) and in rooting biomass and depth. These differences, which reflect their adaptation to particular fertility niches, may affect their relative abilities to compete for N. Thus site N availability may be an important determinant of success in N acquisition by the various deciduous, evergreen, and graminoid plant species within low and tall shrub vegetation communities. In particular, the ability of deciduous shrubs to outcompete other species for N under differing *naturally occurring* levels of soil fertility has not been investigated despite its importance to understanding and predicting the potential for shrub expansion across the low Arctic.

Here, we investigated the impact of deepened snow on ecosystem partitioning of an added isotopic N (^{15}N) tracer among soil and plant species pools in low birch hummock tundra. Specifically, the tracer was added in late summer to control and snowfenced low birch hummock tundra plots to test the influence of deepened snow on microbial ^{15}N turnover and soil ^{15}N availability over one winter, and on plant and microbial ^{15}N acquisition after 2 years. We also added an equivalent amount of tracer to tall birch (unfenced) tundra plots to compare species ^{15}N acquisition in low and tall shrub ecosystems that have the same species

composition but that differ in site N availability. We tested the following hypotheses:

I. Deepened snow in low birch hummock tundra significantly affects soil microbial biomass N, soluble N, and tracer ^{15}N pools in early spring (i.e. over a single winter following tracer addition).
II. Deepened snow in low birch hummock tundra enhances deciduous shrub ^{15}N acquisition 2 years after tracer addition.
III. Species dominance within the low and tall birch plant communities correlates with ^{15}N acquisition, and differs according to site N availability.

Methods

Site description

This study was conducted in low arctic tundra near the tundra ecological research station (TERS) at Daring Lake (64°52′N, 111°35′W) in the Coppermine River watershed, ~300 km northeast of Yellowknife from late summer 2006–2008. Local climate records (1996–2009; Bob Reid, Indian and Northern Affairs Canada, *unpublished data*) indicate daily mean air temperatures ranging from −40 °C in winter to 22 °C in summer, with a mean annual rainfall of 142 mm (Standard error (SE) ± 14). Snow is present for ~200 days (mid-October–June), with snow depth <10 cm until November, by which time soil temperatures are below freezing (Buckeridge and Grogan 2008). The research area is located in a gently sloping valley bordered by an esker to the north and Canadian Shield bedrock outcrops to the south, and contains a variety of different vegetation types underlain by continuous permafrost with a soil active layer ranging from 0.3 to 2 m (Obst 2008). These vegetation types generally occur along toposequences that extend from exposed ridges where dry heath is common down to mesic low birch hummock and ultimately wet sedge at the base. Tall birch patches are found scattered across the landscape in areas close to obvious seasonal surface water flow, but also in apparently mesic locations including relatively high elevation topographic areas protected from the prevailing wind where snow preferentially accumulates (Obst 2008).

Low birch hummock tundra is characterized by hummocks 10–30 cm high and deciduous dwarf birch (*Betula glandulosa* (Michx.), shrubs that range from 10–40 cm tall. The vegetation is dominated by mosses and lichens and the short evergreen shrubs mountain cranberry (*Vaccinium vitis-idaea* L.) and labrador tea [*Rhododendron subarcticum* (Harmaja) (formerly *Ledum decumbens*)], with bog blueberry (*Vaccinium uliginosum* L.), bog rosemary (*Andromeda polifolia* L.), cloudberry (*Rubus chamaemorus* L.), and graminoids (mainly *Carex* spp.) (Nobrega and Grogan 2007) (Table S1). Tall birch tundra is dominated by dense cover of *B. glandulosa* (Michx.) shrubs ~80 cm high. Species composition in this ecosystem was almost identical to the birch hummock vegetation (Table S1), except that occasional tall willow shrubs (*Salix* spp.) are present and *A. polifolia* is absent.

An experimentally deepened snow treatment was established in replicate patches of low birch hummock tundra in summer 2004 using snow fences in a staggered formation ~30 to 60 m apart from one another, and control plots ($n = 5$) located alongside in nearby patches of similar vegetation composition and topography (Nobrega and Grogan 2007). Five plots were randomly located along a downslope transect within a large patch (~40 × 130 m) of tall birch tundra located on a gentle slope without obvious signs of preferential water flow in the same valley as the low birch hummock sites (Buckeridge et al. 2010b). Seasonal soil moisture in this shrub patch is comparable to mesic shrub tundra in Toolik, Alaska (Weintraub and Schimel 2005), but is 2–5 times dryer than 'moist acidic' tussock tundra (Clemmensen et al. 2008; Sorensen et al. 2008; DeMarco et al. 2011). Soil active layer depths late in the growing season tend to be ~10 cm deeper in the tall birch site (e.g. 62 ± 2 cm; $n = 15$; August 12, 2008) than is typical in the birch hummock site (e.g. 48 ± 5 cm; August 14, 2004), but the snowfence treatment there has not significantly affected thaw depth (Nobrega and Grogan 2008; Grogan 2012).

Soil temperature and soil water

Soil temperatures at 2, 5, and 10 cm depth were recorded every 4 h in the control and snowfenced low birch hummock plots ($n = 2$ pairs of probes in each of two plots of each treatment for each depth) and in tall birch plots ($n = 3$ probes at each depth) using thermocouple probes (with an in situ precision coefficient of variation 1.7–7.4 %) connected to CR10X

dataloggers (Campbell Scientific). Soil moisture was measured gravimetrically on all soil samples, and continuously as volumetric water content in two plots at each site using 30 cm long dielectric permittivity probes (Campbell Scientific) inserted at an angle to measure moisture within the top 5 cm of soil, and connected to the same dataloggers that recorded soil temperatures.

^{15}N isotope injections

We labeled three sub-plots within each of the five replicate plots at each site to permit multiple destructive ^{15}N harvests for this study, and longer term investigations in future years. Each ^{15}N injection sub-plot was centered over a randomly selected single representative discrete birch plant. Sub-plots ($n = 3$) were located 2–5 m apart within each of the five birch hummock control plots, and 1–2 m to the north side of each the five snowfences, and within each of the five tall birch plots. We injected each sub-plot with solutions containing 98 % enriched $^{15}NH_4Cl$ (Cambridge Isotope Laboratories, Andover, MA) between August 22–24, 2006 to achieve a common label addition of 50 mg ^{15}N m^{-2} in all sites (equivalent to 4.3, 3.4, and 3.0 μg ^{15}N g dw soil^{-1} in the birch hummock, snowfenced low birch hummock, and tall birch sites respectively). Injections (5 ml each) were made using a syringe with a terminally blocked 8.5 cm needle that had a pair of opposite holes at 8 cm depth and an orthogonal pair of holes at 7.3 cm depth. The needle was inserted to 8 cm depth below the green–brown moss transition and then slowly withdrawn as the syringe was depressed to distribute the solution laterally through the soil profile up to just below the soil surface. Over 80 % of the roots in these ecosystems occur in the top 5 cm of the organic soil layer (Churchland et al. 2010; Grogan, pers. observation), and so we anticipate that most of the root biomass would have had access to the label. The birch hummock and snowfenced low birch hummock ^{15}N additions were made using a 35 × 35 cm injection grid with 8 injections (each 1.66 mM $^{15}NH_4Cl$) 5 cm apart, resulting in a total labeled area of 40 × 40 cm in each of the sub-plots. The size, canopy cover, and associated root biomass of the individual birch plants in the tall birch site were much larger than in the birch hummock sites. In order to account for this plant size difference, we labeled the soil beneath each tall birch shrub using the same number of injections as was used in the low birch sites, but at a larger spatial interval to account for the larger ground area occupied by that plant. Therefore, the ^{15}N additions in the tall birch site were made using a larger injection grid of 70 × 70 cm with eight injections (each 6.66 mM $^{15}NH_4Cl$) each 10 cm apart, resulting in a total labeled area of 80 × 80 cm for each sub-plot. We acknowledge that the different spacing of injections at the low birch and tall birch ecosystems could result in confounding differences in uptake by small-sized species whose root distributions are not similarly distributed at the two grid spacings used (25 and 100 cm^2). However, this concern does not apply to our comparison of tracer uptake in the snowfenced and control low birch vegetation sites. Vascular plants and/or green mosses were gently pushed aside at each injection location so that the label could be delivered directly into the soil. Isotope distribution was predominantly in the organic layer but variability in its depth within each of the sites often resulted in the needle penetrating down into the mineral layer at full insertion.

Sample collection

To determine initial label distribution, intact samples of soil plus associated vegetation (~10 × 10 cm area) were removed to a standard depth of 10 cm (measured from the green–brown moss transition) with a serrated knife from a corner of one sub-plot within each of the five replicate plots at the three sites on August 28, 2006. Only vegetation that was rooted within the sampling area was included. Organic layer depths were measured at the four corners of each sample in the field, and all soil samples were stored at ~5 °C prior to processing (within 2 days). Where the soil samples contained some mineral soil at the base, the depth of each soil type was noted before separating the two types for all subsequent analyses. To determine changes in ^{15}N total distribution over the first winter after labeling (Hypothesis I), similar samples were removed from different corners of the same sub-plots 10 months later (June 21–24, 2007).

To determine the 2 year pattern of ^{15}N distribution among plant species and soil N pools (Hypotheses II and III), we removed an entire ^{15}N labeled sub-plot within each tall birch and birch hummock replicate plot (40 × 40 cm or 80 × 80 cm as appropriate; all to

10 cm soil depth) just prior to leaf senescence (between August 9–23, 2008). To calculate species and growth form biomass per 1 m^2 area, we extrapolated from the sampled biomasses within each sub-plot. However, since each sub-plot was deliberately centered on a birch shrub and the nearest neighbour distance for *B. glandulosa* in the low birch hummock sites clearly exceeded the sub-plot dimensions, we calculated its overall areal biomass in those sites using a previously determined density of two birch plant ramets per m^2 (Zamin and Grogan 2012).

Sample processing

Intact vegetation from each sub-plot was gently teased out from the soil to collect as much of the attached root system as possible, and then sorted to species and growth forms. Birch shrubs were sorted into shoot new growth (all leaves plus apical stem tissue produced in the most current growing season as determined by colour change and complete budscars), shoot old growth (stem biomass from all previous years including secondary growth (shoot thickening)); belowground stems (>5 mm diameter), coarse roots (2–5 mm diameter), and fine roots (<2 mm diameter). Birch belowground stems and coarse roots were not observed in the mineral layer of any sub-plot.

Other vascular plant shoots were separated into shoot new growth (apical stem tissue in the current growing season, and leaves and inflorescences/fruits), shoot old growth, and fine roots (all were <2 mm diameter). All evergreen species leaves were included with stems in the shoot new apical growth category to restrict sorting time and analytical costs while still allowing us to determine the effect of the snowfence treatment on ^{15}N uptake and allocation to photosynthetic tissues over the 2 years. *Salix* spp. were rare but had distinctive 'tap' roots extending >20 cm down into the mineral layer. Graminoid species were sorted as a group into shoot new growth (leaves and inflorescences), shoot old growth (standing dead tissue, stem bases and rhizomes), and fine roots (all <2 mm diameter). The remaining material above the organic soil layer was sorted into mosses, lichens, and surface litter (including leaf fragments, small twigs and dead moss tissue). Organic and mineral soil layer volumes were measured separately on 4–5 sub-samples of each sub-plot (10 × 10 cm to a 10 cm depth) to determine bulk densities. Afterwards, the sub-samples were cut into small chunks ($\sim$27 cm^3), and a random selection was sorted into 'unidentified fine roots' (<2 mm diameter) and 'bulk soil' (i.e. with roots removed). Only living roots (on the basis of tissue turgor and colour) were included in the former category. Roots were rinsed in water to remove attached soil and then immersed in 0.005 M K_2SO_4 for 5–10 min to remove ^{15}N label adhering to exterior surfaces (Grogan and Jonasson 2003).

Soil microbial biomass C and N (MBC and MBN) contents were determined by the chloroform-fumigation direct-extraction using fresh soil (10 g) from which roots had been removed and 50 ml 0.5 M K_2SO_4 for each extraction (Brookes et al. 1985). Fumigation lasted 24 h in a darkened vacuum desiccator jar. Separate non-fumigated (10 g) samples were extracted with 50 ml 0.5 M K_2SO_4 immediately after sorting. All extracts were manually shaken several times over >1 h, left to settle for 30 min, and then filtered through a 1.2 μm glass fibre filter (G4; Fisher Scientific) and frozen for future analyses. Two blanks without sample were included to detect contamination during extraction and filtration each day. All shoot and root samples as well as organic and mineral soil sub-samples and bulk samples were dried at 65 °C for 2–5 days.

Chemical analyses

Dissolved total N (DTN) and total organic C (TOC) in the extracts were determined by oxidative combustion and infrared and chemiluminescence analyses, respectively (TOC-TN autoanalyzer, Shimadzu Japan). NH_4 and NO_3–N were determined colorimetrically, using automated flow analysis and the indophenol and sulphanilamide methods respectively (Bran–Leubbe Autoanalyzer III, Germany). Dissolved organic N (DON) was calculated by subtracting inorganic N forms from the DTN in the non-fumigated samples. MBC and MBN contents were calculated as the differences between fumigated and non-fumigated extractable C and N samples. Our interpretations of the microbial C, N, and ^{15}N data assume that the chloroform fumigation efficiency did not vary between soil types or sampling times. No K_n or K_c correction factor was used in the MBN or MBC calculations (Brookes et al. 1985) because values specific to the site soil-types have not been determined. All extract concentrations were corrected for initial soil moisture contents.

^{15}N isotopic and total N and C analyses

Oven dried shoot tissues, roots, and organic and mineral soil were ground for ~1 min to a fine powder with a centrifugal (plant organic tissues), or ball mill (organic and mineral soils) (Retsch, Germany), weighed out (~1 mg and ~10 mg for organic and mineral samples respectively) into tin capsules (2 mm, Elemental Microanalysis Limited, UK) and sent for ^{15}N and total N analyses at the University of Waterloo Environmental Isotope Lab. We also analyzed total N (and C) in larger samples of the organic and mineral soils (~0.3 g and ~1.2 g) using an elemental analyzer (LECO CNS 2000, USA). Total N concentrations were very closely correlated between the two analyses (IRMS values = 1.01 × elemental analyzers values + 0.006; $r^2 = 0.99$) indicating that the samples were well homogenized and suggesting high analytical accuracy. DT^{15}N and MB^{15}N were determined by drying 3 ml of the non-fumigated and fumigated extracts at 65 °C for 4–6 days (Dijkstra et al. 2006). Dried salt samples were ground using a mortar and pestle and weighed (~120 mg) into tin capsules (5 mm, as above) and sent for analysis to the Colorado Plateau Stable Isotope Laboratory (University of Northern Arizona).

^{15}N calculations

^{15}N natural abundances were determined in shoot new growth, old growth, and root tissues of each species, as well as from mosses, lichens, surface litter, and soils sampled from 5–6 random locations >5 m away from the plots in the low birch hummock and tall birch sites during the 2008 harvest. Replicate tissue samples for each species shoot new growth, old growth, and roots were pooled and ground into a composite sample representing each species growth category from its respective site. Natural abundance ^{15}N atom percentages in plants ranged from 0.3645 % (*V. vitis-idaea;* new growth) to 0.3666 % (graminoids; old growth). Natural abundance ^{15}N atom percentages were 0.3668 and 0.3669 % in organic and mineral soils respectively, and 0.3655 % in the organic soil microbial biomass. Paired t tests across the species and soil pools indicated no differences in natural abundances between the low birch and tall birch sites. For calculating label uptake, we assumed that ^{15}N natural abundance levels in the control and snowfenced low birch sites were equivalent at the start of the experiment, and that any changes due to the snowfences were minimal in magnitude compared to tracer enrichment levels. Supporting this assumption, the mean isotopic increase as a result of label addition was 0.09 %, while the range of variation in isotopic concentration among sample categories used to determine natural abundance levels was always considerably lower (<0.003 %). ^{15}N pool enhancement (i.e. added ^{15}N accumulated) in the plant and microbial pools was calculated as (atom% $^{15}N_{measured}$ − atom% $^{15}N_{natural\ abundance}$) X total N pool size. Note that we report our enhancement data on an areal basis (mg $^{15}N\ m^{-2}$) so that plant and soil pools can be directly compared. Since the same amount of label was added to all plots (50 mg m^{-2}), the percent recovery of the added label can be calculated from these data, and the results from the statistical analyses of both variables are identical. Furthermore, site differences in tracer dilution in either the soil or plants caused by differing N cycling rates among sites should not affect our ^{15}N results since we are comparing tracer acquisition in absolute amounts on an areal basis. However, accordingly, interpretation of ^{15}N accumulation as an indicator of species overall N accumulation assumes that those microbial and soil pools most affecting available N to plants have been turned over enough times in the study's 2 years that any impacts of site differences in N turnover rate (and therefore label dilution) have long passed. The rapid label turnover in the soil solution N pool within days of its addition (see "Results" section) supports this assumption. Tracer in the soil that was not present in either root, microbial, or soil solution pools was calculated by subtracting (MB^{15}N+DT^{15}N) from the total label ^{15}N in the root-removed soil samples and termed fixed SOM^{15}N. Note that since no Kn factor (Brookes et al. 1985) was applied, our fixed SOM estimates are relatively high and would have been reduced from ~80 to 60 % of the total amount added had we used a K_n of 0.4 (Jonasson et al. 1999).

Statistical analyses

We tested for significant effects of site, year, and their interaction on each of the soil microbial and solution C and N pools, ^{15}N label concentrations, and enhancement pools in organic and mineral soil layers, using separate repeated measures two-way analyses of variance (ANOVAs). Multiple comparisons were

conducted using the Tukey–Kramer HSD test to identify significant differences between all pairwise combinations (JMP 7.0.2, SAS Institute). Datasets were tested for normality, and log transformed as necessary (all soil ^{15}N data but not total elemental pools). Site effects on ^{15}N concentrations and enhancements for each species growth component were tested using separate one-way ANOVAs. All plant species ^{15}N concentrations were log transformed to meet normality tests prior to analyses. Tukey–Kramer HSD tests were not applied to the ^{15}N pool data by plant species because the distributions could not be readily transformed to make them normal. Mean concentrations of N and ^{15}N label in all plant and organic soil variables were calculated based on the actual number of replicate plots in which the component was present in each site. By contrast, mean ^{15}N pool enhancements were based on five replicates. Means and statistical analyses for mineral soil variables were based on the number of plots in which mineral soil was present within the sampling depth of ($n = 3$, 4 and 5 for the low birch hummock, snowfenced low birch hummock and tall birch sites, respectively).

Results

Site differences in soil ^{15}N concentrations and N and C pools across years

Soil organic layer MB^{15}N tracer concentrations declined from the initial sampling within 4–6 days of injection through the following winter and on to the full 2 year harvest (Table 1). Although initial label immobilization at the control low birch hummock tended to be substantially higher than in either of the other sites, MB^{15}N concentrations were similar among all sites 2 years later (Table 1). Note that the pattern for MB^{15}N pool sizes was statistically identical to the concentrations because variation in isotope concentration was much larger than variation in soil bulk density (Table S2; Fig. S1). Soil microbes released ~75 % of the acquired label over the first winter after labeling (i.e. from Fall 2006 to Spring 2007) in the control low birch hummock and tall birch sites, but MB^{15}N in the snowfenced plots changed relatively little over that same period (Table 1; Fig. S1). Mineral soil MB^{15}N label concentrations and enhancement pools 2 years after labeling were ~10 % of the overlaying organic layer values, and did not differ among sites (Table S3). ^{15}N label concentrations within the dissolved total N (DTN) pool of the organic layer were ~2 % of the concentration added within 4–6 days of injection, indicating rapid turnover in this pool (Table 1; "Methods" section). Similar to MB^{15}N, DT^{15}N concentrations declined over the first winter, but in this case there were no site differences.

The soil organic layer ammonium (NH_4–N), dissolved organic N (DON), and dissolved organic C (DOC) pools were generally at least two times larger in the tall birch site than in the birch hummock site (especially during the 2007 and 2008 sampling times), but there were no effects of the snowfence treatment (Table 1). Correspondingly, the organic layer microbial biomass N (MBN) pool was ~18 % larger in the tall birch site than in either of the birch hummock sites, in particular during the latter two sampling times. By contrast, microbial biomass C (MBC) was similar across sites and years. Microbial biomass and soil solution pools in the uppermost mineral soil underlying the organic layer in 2008 were <1/3 of the overlaying organic layer and did not differ statistically among sites (Table S3).

Site differences in ^{15}N label partitioning among plant species components

Shoot new growth ^{15}N enhancement (i.e. the amount of added ^{15}N label accumulated in new shoot apical growth) differed significantly among the low birch and tall birch sites for many of the plant species, but there were no significant snowfence treatment effects (Fig. 1A). Generally, the deciduous shrub *B. glandulosa* dominated new shoot ^{15}N enhancement in the tall birch, while the evergreens (*R. subarcticum* and *V. vitis-idaea)* dominated in the birch hummock sites. New apical shoots of *B. glandulosa* accumulated at least sixteen times more ^{15}N in the tall birch than at the birch hummock sites ($F_{2,12} = 73.5$, $P = <0.001$; Fig. 1A), partly as a result of 2–4 times larger ^{15}N label concentrations (Table 2), and therefore the remainder of this effect being due to larger biomass. The other common deciduous shrub species *V. uliginosum* accumulated two times as much ^{15}N in the tall birch compared to both birch hummock sites ($F_{2,12} = 5.3$, $P = 0.004$), with no corresponding differences in ^{15}N label concentrations (Table 2).

Table 1 Soil solution and microbial N and C pools (g m^{-2}) and ^{15}N label concentrations (μg/g dw soil) in the organic soil layer of the three sites in late summer 2006, early spring 2007 and late summer 2008

		Birch Hummock		Snowfenced Birch Hummock		Tall Birch		Statistical analyses
NH_4-N	Late summer 2006	0.07^{abc}	(0.01)	0.08^{abc}	(0.02)	0.12^{ab}	(0.03)	Site: $F_{2,12} = 7.20$, $P = 0.0088$; Year: $F_{2,24} = 8.31$, $P = 0.0018$
	Early spring 2007	0.02^{bc}	(<0.01)	0.03^{bc}	(<0.01)	0.13^{a}	(0.04)	
	Late summer 2008	$<0.01^{c}$	(<0.01)	0.02^{c}	(<0.01)	0.08^{abc}	(0.01)	
DON	Late summer 2006	0.82^{ab}	(0.09)	0.98^{a}	(0.20)	1.24^{a}	(0.26)	Site: $F_{2,12} = 10.99$, $P = 0.0019$; Year: $F_{2,24} = 24.68$, $P < 0.0001$; Site×Year: $F_{4,24} = 3.44$, $P = 0.0232$
	Early spring 2007	0.30^{c}	(0.03)	0.42^{bc}	(0.08)	0.87^{ab}	(0.14)	
	Late summer 2008	0.29^{c}	(0.06)	0.30^{bc}	(0.07)	1.01^{a}	(0.12)	
DOC	Late summer 2006	10.40^{b}	(0.82)	13.29^{ab}	(2.07)	14.93^{ab}	(3.57)	Site: $F_{2,12} = 8.54$, $P = 0.0049$;Year: $F_{2,24} = 13.47$, $P = 0.0001$; Site×Year: $F_{4,24} = 3.99$, $P = 0.0127$
	Early spring 2007	4.49^{c}	(0.34)	6.47^{bc}	(1.08)	9.47^{bc}	(1.37)	
	Late summer 2008	6.00^{bc}	(1.21)	7.12^{bc}	(0.78)	22.48^{a}	(4.68)	
MBN	Late summer 2006	3.83^{a}	(0.27)	4.20^{a}	(0.62)	3.78^{a}	(0.50)	Site: $F_{2,12} = 6.93$, $P = 0.010$; Year: $F_{2,24} = 5.73$, $P = 0.0093$
	Early spring 2007	2.88^{ab}	(0.20)	3.58^{ab}	(0.59)	4.82^{a}	(0.74)	
	Late summer 2008	1.98^{b}	(0.16)	2.72^{ab}	(0.18)	4.18^{a}	(0.62)	
MBC	Late summer 2006	46.04^{a}	(2.93)	54.43^{a}	(8.53)	44.36^{a}	(7.02)	No significant differences
	Early spring 2007	42.00^{a}	(1.42)	50.51^{a}	(7.27)	47.50^{a}	(6.01)	
	Late summer 2008	36.95^{a}	(4.92)	45.36^{a}	(4.75)	63.00^{a}	(8.18)	
$MB^{15}N$	Late summer 2006	2.35^{a}	(0.62)	1.15^{ab}	(0.42)	0.75^{abc}	(0.22)	Site: $F_{2,12} = 8.15$, $P = 0.0058$; Year: $F_{2,24} = 21.68$, $P = 0.0001$; Site×Year: $F_{4,24} = 3.83$, $P = 0.016$
	Early spring 2007	0.58^{abc}	(0.18)	0.76^{abc}	(0.21)	0.14^{d}	(0.10)	
	Late summer 2008	0.24^{bcd}	(0.01)	0.37^{bcd}	(0.09)	0.27^{cd}	(0.12)	
$DT^{15}N$	Late summer 2006	0.08^{a}	(0.02)	0.05^{ab}	(0.01)	0.08^{a}	(0.04)	Year: $F_{2,24} = 79.07$, $P = <0.0001$
	Early spring 2007	$<0.01^{c}$	(<0.01)	$<0.01^{c}$	(<0.01)	0.01^{c}	(<0.01)	
	Late summer 2008	$<0.01^{c}$	(<0.01)	$<0.01^{c}$	(<0.01)	0.01^{bc}	(<0.01)	

NH_4–N ammonium, *DON* dissolved organic nitrogen, *DOC* dissolved organic carbon; *MBN* microbial biomass nitrogen; *MBC* microbial biomass carbon. Parentheses indicate standard errors ($n = 5$). NO_3–N was below detection limit. For each variable, mean differences among sites and years are indicated by different superscript letter(s) (ranging from a–d, $P < 0.05$)

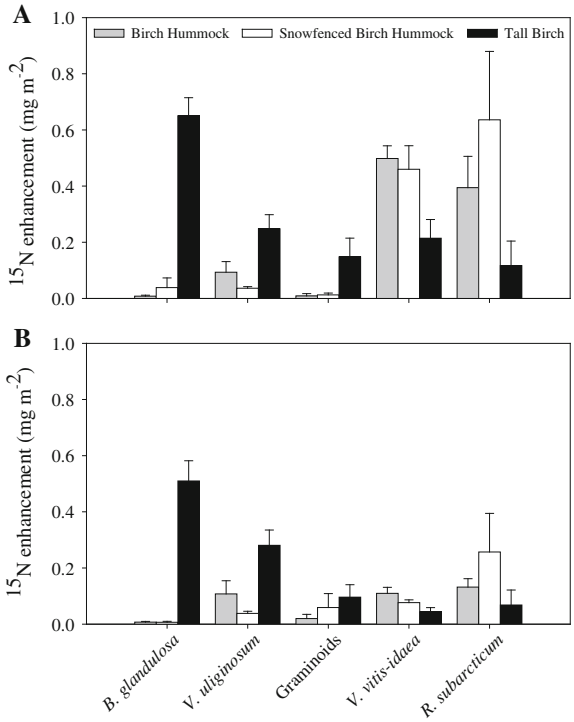

Fig. 1 ^{15}N enhancement (mg m^{-2}) in plant shoot new growth (**A**) shoot old growth (**B**) of the major vascular species at each site 2 years after the tracer ^{15}N addition (late August 2008) raw means are displayed ($n = 5$) and *error bars* denote standard error

Graminoid new shoots also accumulated substantially more ^{15}N in the tall birch site than at the other sites ($F_{2,12} = 4.2$, $P = 0.04$) (Fig. 1A), but in this case the effect was driven primarily by the at least six times larger tissue ^{15}N concentrations (Table 2). By contrast, *R. subarcticum* and *V. vitis-idaea* accumulated half as much ^{15}N in the tall birch site compared to the birch hummock sites ($F_{2,12} = 3.9$, $P = 0.05$; $F_{2,12} = 5.3$, $P = 0.02$, respectively) (Fig. 1A), with no differences in ^{15}N label concentration (Table 2).

Similar to shoot new tissues, ^{15}N enhancement in old shoots was larger at the tall birch site than at either of the low birch sites, and this effect was primarily due to tracer accumulation by *B. glandulosa* ($F_{2,12} = 49.0$, $P = < 0.001$; Fig. 1B). Once again, there were no significant snowfence effects on old shoot ^{15}N concentration or enhancement of any species in the birch hummock ecosystem (Table 2; Fig. 1B). The strong ^{15}N enhancement of old shoots in *B. glandulosa* at the tall birch site must have been due to its larger biomass there since the shoot ^{15}N label concentrations did not significantly differ across sites (Table 2). Likewise, *V. uliginosum* old shoot ^{15}N enhancement was higher at the tall birch site ($F_{2,12} = 8.9$, $P = 0.004$; Fig. 1B; Table 2). By contrast, *V. vitis-idaea* old shoot ^{15}N enhancement was higher in the low birch hummock sites compared to the tall birch ($F_{2,12} = 4.2$, $P = 0.04$; Fig. 1B), but there was no significant analogous site effect on *R. subarcticum* old shoots.

As expected from the old shoot results, ^{15}N enhancements in belowground stems, as well as coarse and fine root components of *B. glandulosa* were higher at the tall birch site compared to the low birch sites (Table S4), and these effects were due to larger biomass rather than site differences in ^{15}N concentrations (Table 2). Similarly, *V. uliginosum* root ^{15}N enhancement was also substantially higher at the tall birch site for the same reason (Tables 2, S4). The large proportions of organic layer fine roots that could not be identified to species had similar ^{15}N label concentrations across sites (Table 2), but nevertheless total enhancement was ~60 % larger in the tall birch ecosystem (Table S4). Root ^{15}N label concentrations within the underlying mineral soil were similar among sites (Table 2), but were generally at least twice those of organic layer roots. Therefore, although organic layer fine roots altogether accounted for 80–97 % of the total ^{15}N enhancement within all fine roots measured, some label was clearly available to, or translocated to, fine roots in the mineral layer. Once again, there were no snowfence effects on belowground ^{15}N concentration or enhancement of any species in the birch hummock ecosystem (Tables 2, S4).

Overall ^{15}N label partitioning among ecosystem components

^{15}N label distribution among the ecosystem components 2 years after its addition indicated that total vascular shoot ^{15}N accumulation (across all species combined) was similar among the sites (Fig. 2). Surface litter ^{15}N pools were about half of the total shoot pools, and also did not differ among sites. By contrast, total belowground plant tissues ^{15}N accumulation was about twice as high in the tall birch as compared to the birch hummock sites. Overall, total ^{15}N total accumulation in vascular plants (above- and below-ground) matched total soil microbial accumulation within the top 10 cm of soil. The sum totals of

Table 2 ^{15}N label concentrations ($\mu g^{15}N/g$ dw tissue) in vascular shoot new growth, vascular shoot old biomass, mosses, lichens, surface litter, belowground stems (>5 mm diameter), coarse roots (2–5 mm), and fine roots (<2 mm) of the major plant species at each site in late August 2008

	Birch Hummock		Snowfenced Birch Hummock		Tall Birch		Statistical analyses
Shoot new growth							
Betula glandulosa	0.99^{y}	(0.21)	1.99^{y}	(1.28)	4.48^{x}	(0.49)	$F_{2,12} = 6.43$, $P = <0.0127$
Vaccinium uliginosum	17.08^{x}	(1.12)	13.17^{x}	(1.98)	19.11^{x}	(2.76)	
Graminoids	0.69^{x}	(0.14)	4.49^{x}	(2.40)	26.74^{x}	(7.01)	$F_{2,9} = 3.56$, $P = 0.0729$
Vaccinium vitis-idaea[a]	10.26^{x}	(1.68)	8.25^{x}	(1.15)	10.41^{x}	(2.22)	
Rhododendron subarcticum[a]	11.17^{x}	(1.89)	12.43^{x}	(2.93)	14.19^{x}	(5.17)	
Shoot old growth							
Betula glandulosa	0.28^{x}	(0.07)	0.40^{x}	(0.18)	0.54^{x}	(0.08)	
Vaccinium uliginosum	3.52^{x}	(1.03)	3.38^{x}	(0.64)	4.38^{x}	(0.47)	
Graminoids	1.96^{x}	(0.53)	1.40^{x}	(0.42)	5.60^{x}	(1.79)	
Vaccinium vitis-idaea	5.23^{x}	(0.08)	3.87^{x}	(0.50)	4.17^{x}	(0.84)	
Rhododendron subarcticum	2.20^{x}	(0.57)	2.49^{x}	(0.58)	2.89^{x}	(0.74)	
Mosses	0.97^{x}	(0.16)	0.99^{x}	(0.23)	0.60^{x}	(0.13)	
Lichens	0.76^{x}	(0.33)	0.49^{x}	(0.07)	0.37^{x}	(0.08)	
Surface litter	1.27^{x}	(0.37)	1.12^{x}	(0.16)	0.94^{x}	(0.10)	
Belowground stems							
Betula glandulosa	0.33^{x}	(0.12)	0.65^{x}	(0.14)	0.69^{x}	(0.15)	$F_{2,10} = 3.61$, $P = 0.0661$
Coarse roots							
Betula glandulosa	0.65^{x}	(0.10)	0.79^{x}	(0.20)	1.46^{x}	(0.32)	
Fine roots (<2 mm)							
Betula glandulosa	1.91^{xy}	(0.43)	1.09^{y}	(0.35)	4.13^{x}	(0.91)	$F_{2,12} = 8.69$, $P = 0.0046$
Vaccinium uliginosum	2.88^{x}	(0.77)	3.09^{x}	(0.32)	3.30^{x}	(0.37)	
Graminoids	3.02^{x}	(2.10)	4.35^{x}	(2.33)	8.57^{x}	(1.98)	
Vaccinium vitis-idaea	3.93^{x}	(1.20)	3.29^{x}	(0.34)	3.13^{x}	(0.75)	
Rhododendron subarcticum	1.93^{x}	(0.27)	2.24^{x}	(0.45)	2.57^{x}	(0.84)	
Unidentified roots (organic soil)	2.80^{x}	(0.46)	2.55^{x}	(0.42)	2.94^{x}	(0.32)	
Mineral soil roots[b]	5.93^{x}	(1.09)	7.72^{x}	(5.31)	10.56^{x}	(4.34)	

Means within rows for each component are not significantly different if they share the same superscript letters (ranging from x–z; $P < 0.05$). Parentheses indicate standard errors ($n = 3$–5)

[a] New growth in evergreen species includes current year's shoot growth (stem and leaves) plus all other leaves

[b] Only the uppermost 3–4 cm of the mineral layer were sampled

the added ^{15}N label in all measured plant, microbial, and soil solution pools within the organic and sampled mineral soils (Fig. 2) were 8.1 (SE ± 0.7), 10.5 (SE ± 0.7), and 12.9 (SE ± 2.2) mg ^{15}N m^{-2} at the birch hummock, snowfenced low birch hummock, and tall birch sites, respectively. In other words, ~20 % of the added label was recovered in these pools, and there were no significant differences among sites. These data together with our fixed SOM ^{15}N label estimates in the soil (29.3 (SE ± 5.3), 44.2 (SE ± 6.4), and 74.5 (SE ± 29.4) mg ^{15}N m^{-2} in the birch hummock, snowfenced birch hummock, and tall birch soils, respectively) suggest that most of the added tracer (50 mg ^{15}N m^{-2}) was retained within the plots (to 10 cm soil depth) at each site over the 2 years.

Site differences in soil environmental and edaphic characteristics

As expected, the deeper snow in the snowfenced low birch hummock and tall birch sites resulted in strong thermal insulating effects in both winters (Fig. 3). The snowfence treatment effect on soil temperature did not become evident until late December in winter 2006/2007, and early February in winter 2007/2008, suggesting an interannual difference in the timing of snow accumulation. By contrast, soil cooling patterns in the tall birch site were very similar in the 2 years, and soil temperatures were generally ~5 °C warmer than the control low birch hummock site from early December to May in both years. Snowfenced low birch

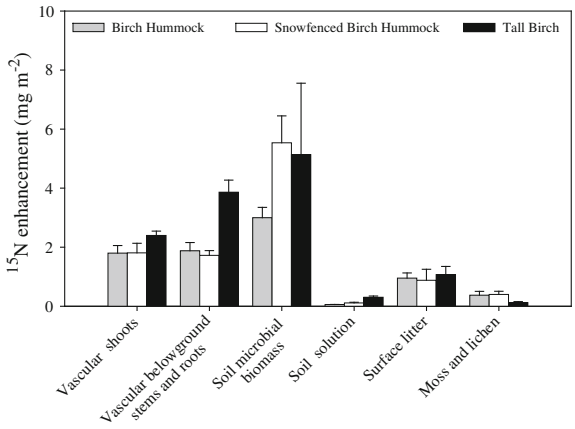

Fig. 2 ^{15}N enhancement (mg m^{-2}) in the principal ecosystem component pools for each site 2 years after the tracer ^{15}N addition (late August 2008) Raw means are displayed ($n = 5$) and *error bars* denote standard error. Microbial and soil solution pools are calculated to 10 cm soil depth which includes the organic layer and some underlying mineral soil

hummock plots were generally 3–5 °C warmer than the control plots from January to April in both years. Mean snow depths in the low birch hummock, snowfenced low birch hummock, and tall birch plots in late winter were 29 (SE ± 2), 87 (SE ± 3), and 52 (SE ± 3) cm on April 8th 2007, and 29 (1), 96 (3) and 62 (5) cm on May 05, 2008 respectively. Soil temperatures on those dates were >1.5 °C warmer in the tall birch site compared to the snowfence site (Fig. 3) indicating that the taller vegetation type provided better thermal insulation even though the snow depth was ~1/3 less than that of the snowfenced plots.

Soil organic layer moisture was at least 25 % drier over the growing season in the tall birch site compared to the two birch hummock sites as indicated by the continuous volumetric water content datalogger records (Fig. S2) and the gravimetric data after the June snowmelt was complete (Table S2). Soil organic layer bulk density was ~50 % higher in the tall birch site and C concentrations were correspondingly ~30 % lower, but there were no significant differences in soil N concentration, or organic layer depth among sites (Table S2). Finally, there were no significant site differences in moisture content, bulk density, C or N concentrations, or sampling depth in the uppermost mineral soil underlying the organic layer in each ecosystem (Table S2).

Discussion

The influence of experimentally deepened snow on soil N and ^{15}N pools in the low birch hummock ecosystem

The snowfences did not substantially alter microbial or soil solution N or ^{15}N pools in the low birch

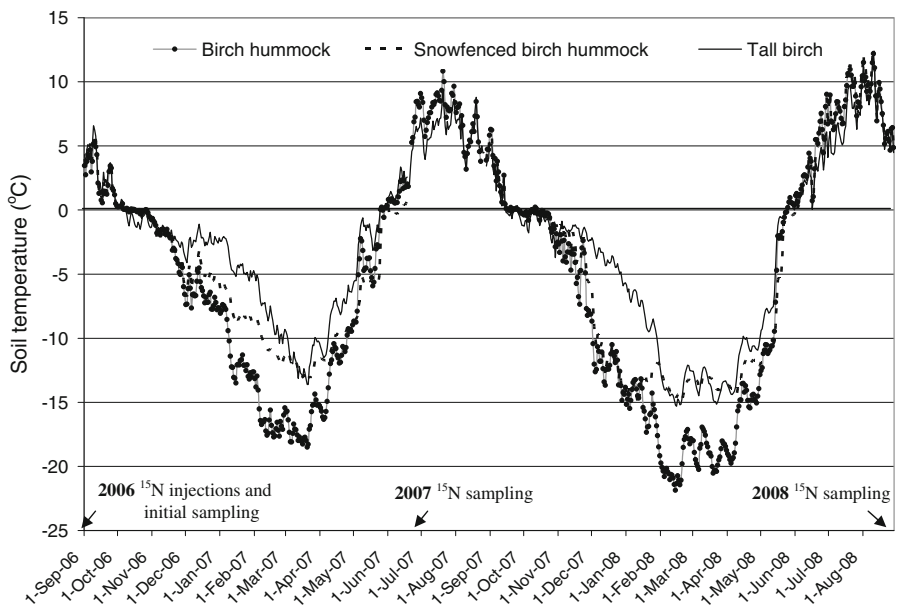

Fig. 3 Diel mean soil temperatures at the birch hummock, snowfenced low birch hummock and tall birch sites from late August 2006–2008 (probes at 5 cm depth; $n = 3$–4). Soil temperatures within each site at 2 and 10 cm soil depth were very similar to the above

hummock ecosystem in Spring 2007 at the end of the first winter after labeling (Table 1), rejecting Hypothesis I. Furthermore, there were no significant increases in ^{15}N enhancement in any plant species in the snowfence plots compared to the control plots (Fig. 1). Although all of these data are instantaneous pools sizes rather than flux rates, together they strongly suggest that the deepened snow treatment did not increase N availability to plants at least within the study's 2 year time frame.

We added the ^{15}N to all plots in late summer in a form (ammonium) that should be readily available to both plants and microbes. Our overall goal was to test for overwinter site and treatment effects on microbial N turnover that could influence the timing and amount of ^{15}N made available to plants in the following spring. Since previous analogous studies indicated strong competitive ability of microbes relative to plants for added ^{15}N in tundra (Grogan and Jonasson 2003; Larsen et al. 2012), we labeled in late summer on the assumption that root N uptake would be minimal during plant senescence and that soil microbes would immobilize most of the added tracer prior to Fall. Our data from the small subsamples taken 1 week after tracer addition in 2006 support this assumption. Microbial label uptake ranged from 8–23 mg ^{15}N m^{-2} among the sites (Fig. S1), while root uptake was at least an order of magnitude smaller (0.2–0.6 mg m^{-2}; Table S5). Furthermore, total aboveground ^{15}N enhancement (all species pooled) in spring 2007 was only 0.7 (SE $\pm$ 0.3) and 0.4 (SE $\pm$ 0.2) mg m^{-2} in the low birch hummock and snowfenced birch hummock, respectively, and did not differ between sites (no corresponding data for the tall birch site because of low number of replicates with sufficient species tissue for isotopic analyses). Note that these plant and microbial ^{15}N enhancement values are not as accurate or representative as the 2008 harvest values because the latter were determined on much larger-sized samples (the whole sub-plot) and because the 2006 sub-samples were taken from a plot corner and therefore furthest away from the central birch shrub. In any event, over the 2 years after labeling, total plant ^{15}N accumulation had increased to 4–6 mg m^{-2} (Fig. 2), while the microbial ^{15}N pool had declined by an even greater magnitude (Table 1; Fig. S1). Hence, we conclude that the plants in the final harvest had acquired most of their label via inorganic and as well as organic ^{15}N compounds (McKane et al. 2002) that were made available via substantial turnover of the microbial N pool during the 2 years since initial label addition.

Over the first winter after labeling, organic soil $MB^{15}N$ pools declined in the control low birch hummock and tall birch sites, but were not significantly altered in the snowfenced low birch hummock site (Fig. S1). This effect could be associated with a deeper snow treatment effect on wintertime biogeochemical processing, but even if so, it clearly had no impact on the ^{15}N distribution pattern 2 years after labeling (Fig. 2). Accordingly, we think it is more likely an artifact of using the same early Spring sampling date at all sites even though the soil thaw date in the snowfenced plots was 2 weeks later than for the other sites (Table S2). Frequent measurements from late winter to early spring in a concurrent study in the same plots, demonstrated that in the days immediately after soil temperatures had risen up to 0 °C, N was taken up by microbes under deepened snow while at the same time soil microbes in control plots (where the snow has already completely melted) were losing N (Buckeridge and Grogan 2010). In any event, although this issue is a concern for our interpretation of the 2007 soil data, it has no impact on the interpretation of the plant and soil data from the 2 year harvest.

The influence of experimentally deepened snow on plant species ^{15}N uptake in the low birch hummock ecosystem

The snowfences did not enhance ^{15}N acquisition by deciduous shrubs or any other plant species within our study's 2 year timeframe (Table 2; Fig. 1A, B), rejecting Hypothesis II. Accordingly, we found no evidence to support the final phase (i.e. enhanced shrub N accumulation) of the Snow–Shrub Feedback Hypothesis (Sturm et al. 2001, 2005). Nevertheless, our study period was short relative to the multi-decadal timeframe of recent shrub expansion (Tape et al. 2006,2012; Forbes et al. 2010). Although not statistically significant, we did observe trends toward relatively high $MB^{15}N$ enhancement (organic+mineral) and birch shrub ^{15}N concentration in the snowfenced low birch hummock site compared to the control 2 years after the label addition (Tables 1, 2; Fig. 2). Thus, our study may not have been sufficiently sensitive to detect snow–shrub feedbacks associated with enhanced N availability within a 2 year timeframe. We will harvest the plants

and soils in the remaining labeled shrub-plots after a decade as a longer term test of the Snow–Shrub Feedback Hypothesis.

Alternatively, there are several reasons why the enhanced shrub N accumulation phase of the snow–shrub feedback hypothesis may be incorrect. First, the additional nutrients mobilized under the deepened snow may be lost from the system before plants could acquire them. Although we know that deepened snow increases dissolved organic and inorganic nutrient pulses in our plots (Buckeridge and Grogan 2010), plant uptake of the mobilized nutrients may have been prohibited by the greater surface runoff of these nutrients that would be expected with high snowmelt water flow in the deep snow treatment. Denitrification losses are also possible, although N_2O production rates at our site in early spring were very low and unaffected by deeper snow (Buckeridge et al. 2010a). Second, plant nutrient uptake capacity at the end of snowmelt may be severely restricted. The enhanced spring pulse of dissolved nutrients in the snowfence treatment was relatively short-lived, and occurred when the snowfenced plants were first exposed above the melting snow, and while the soils were very wet and just rising above 0 °C (Buckeridge and Grogan 2010). Although some alpine plant species can actively take up N during snowmelt, arctic tundra plants do not seem to have that ability (Bilbrough et al. 2000). Furthermore, a recent tracer study in Swedish tundra suggests that deciduous shrubs are not able to resume nutrient uptake until after snowmelt is complete (Larsen et al. 2012). Interestingly, evergreen shrubs in that study were able to acquire label several weeks earlier (Larsen et al. 2012), but we found no evidence that the evergreen species at our sites were able to preferentially take advantage of snowfence-enhanced ^{15}N tracer pulses in late winter. Finally, although we found little evidence that deepened snow alone would facilitate the enhanced shrub N accumulation phase of the Snow–Shrub Feedback Hypothesis, this does not preclude other factors associated with tall shrubs from interacting with the deep snow around them to promote their own growth. For example, the tall shrubs generated a particularly effective thermal insulation of their underlying soils that cannot be attributed to deeper snow alone (Fig. 3). In addition, the relatively high productivity of deciduous tall shrubs results in greater soil litter inputs that provide the substrate for N mobilization and the relatively fast N cycling of tall birch ecosystems (Buckeridge et al. 2010b). This litter feedback alone likely promotes deciduous shrub growth, but in addition, deepened snow may contribute by facilitating decomposition of that litter in the fall, winter and spring.

Species differences in ^{15}N acquisition within and among ecosystems

The comparison of tracer ^{15}N enhancement pools and tracer ^{15}N concentrations indicates that individual vascular plant species ^{15}N acquisition differed among ecosystems not just because of differences in biomass, but also because of species differences in whole plant physiological uptake capacity and/or species differences in internal translocation of label. For example, the two evergreens *R. subarcticum and V. vitis-idaea* acquired twice as much tracer in new shoot tissue in the birch hummock sites compared to the tall birch site (Fig. 1A). There were no site differences in shoot ^{15}N concentrations for these species (Table 2), indicating that this effect was due to larger shoot biomass of each at the former site. By contrast, new apical shoots of *B. glandulosa* and graminoids accumulated 10–16 times more ^{15}N in the tall birch than at the birch hummock sites (Fig. 1A). For the shrub, this effect was due to significantly higher ^{15}N label concentrations as well as larger biomass production (Table 2; Fig 4), and for the graminoid, it was driven primarily by higher tissue ^{15}N concentrations alone (Table 2; Fig. 4). The elevated label concentrations in new and old shoots as well as roots of *B. glandulosa* and the graminoids in particular suggest enhanced ^{15}N uptake capacity per unit biomass of these species in the tall birch ecosystem which we know has greater N availability (Table 1), and faster N cycling rates than the low birch hummock ecosystem (Buckeridge et al. 2010b; Chu and Grogan 2010). Superior nutrient uptake capacity is typical of fast growing opportunist species within a plant community, and the strong birch and graminoid responses we observed here are consistent with their relatively quick responses to fertiliser additions (Chapin et al. 1995; Press et al. 1998). Together, these results suggest that *B. glandulosa* and the graminoids are better competitors for N when it becomes more available, indicating that they may have a distinct advantage over the rest of the vegetation community in sites where climate warming increases soil fertility.

Vascular plant species composition is almost identical in the birch hummock and tall birch sites, and yet

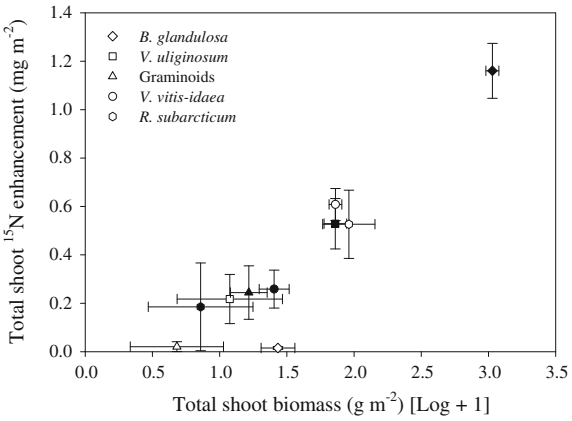

Fig. 4 Shoot ^{15}N acquisition in relation to shoot biomass for each species that occurred in both the control low birch hummock (*open symbols*) and tall birch plant communities (*filled symbols*). Sampling was 2 years after the tracer ^{15}N addition (late August 2008; $n = 5$; *bars* = SE)

species relative abundances differ greatly (Buckeridge et al. 2010b; Grogan 2012) (Table S1). Species dominance in an Alaskan tussock tundra plant community that closely resembles birch hummock tundra was positively correlated with their individual capacities to acquire N in its most available forms (McKane et al. 2002). Correspondingly, dominance by the evergreens *V. vitis-idaea* and *R. subarcticum* at our birch hummock site was matched by strong total shoot^{15}N accumulation, and likewise for *B. glandulosa* in our tall birch site (Fig. 4). However, in addition, our data can be used to compare the performance of each species within these two ecosystems that differ greatly in species relative abundances, productivity, and N cycling. Species total shoot (new plus old growth) ^{15}N accumulation 2 years after its addition correlated closely with total shoot biomass (Fig. 4), suggesting that shoot growth of all of these species at both sites is N-limited. Note that the corresponding relationship for *new* shoot growth only is very similar and even more closely correlated, but we chose here to report the total shoot data because it best indicates overall aboveground biomass (and therefore relative dominance), and because it represents total label accumulation over the 2 years of the study for all species with perennial shoot tissues. Furthermore, comparison of each species' ^{15}N accumulation between these two sites suggests that differences in soil N availability exerted a fundamental control on the outcome of each species' success in competing for N, and therefore of dominating the plant community. The evergreens *R. subarcticum* and *V. vitis-idaea* dominated the biomass and ^{15}N accumulation in the low birch hummock ecosystem, but acquired less ^{15}N (while maintaining the same ratio to biomass) in the more fertile tall birch ecosystem (Fig. 4). By contrast, *B. glandulosa* in particular, as well as the graminoids and *V. uliginosum* all accumulated substantially more ^{15}N in the tall birch ecosystem. These results together with the soil N data (Table 1) and previous studies at these sites which indicate higher N cycling rates in the tall birch ecosystem (Buckeridge et al. 2010b, Chu and Grogan 2010) support Hypothesis III that species dominance within the low and tall birch plant communities correlates with ^{15}N acquisition, and differs according to site N availability.

Conclusion

Species ^{15}N acquisition patterns at the low birch hummock and tall birch sites suggest growth form-specific responses to the enhanced soil nutrient availability expected with arctic climate warming. Low levels of N enhancement will likely favour the evergreen species in current low birch tundra plant communities because they seem to be superior competitors under infertile conditions (Fig. 1A, B), and even after limited fertilizer addition (Churchland et al. 2010). By contrast, *B. glandulosa* may outcompete these species and dominate these communities if N availability is enhanced toward levels comparable with the tall birch ecosystem. *B. glandulosa's* higher N competitive ability under relatively fertile conditions is a typical trait of species that promote favorable environments for their own growth by producing large amounts of readily decomposable litter (Hobbie 1992). Furthermore, this species can switch from producing 'short' and 'long' shoots to all 'long' shoots under increased nutrient availability (Bret-Harte et al. 2001), thereby rapidly enhancing primary production and litter fall. These traits, and the lack of a deepened snow effect on ^{15}N acquisition in low birch tundra, suggest that climate-change induced growth and expansion of birch shrubs across the landscape will tend to occur most rapidly in and around existing tall shrub patches which are already relatively fertile. This conclusion supports recent meta-analyses of pan-Arctic long-term plot-level vegetation change (Elmendorf et al. 2012b) and impacts

of moderate experimental warming (Elmendorf et al. 2012a) which both indicated that deciduous shrub growth is most responsive to warming in tundra habitats that are inherently relatively warm and that have moist-wet soils (i.e. where fertility would be expected to be relatively high).

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