

Contrasting nitrogen and phosphorus dynamics across an elevational gradient for subarctic tundra heath and meadow vegetation

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Abstract

Aims This study explores soil nutrient cycling processes and microbial properties for two contrasting vegetation types along an elevational gradient in subarctic tundra to improve our understanding of how temperature influences nutrient availability in an ecosystem predicted to be sensitive to global warming.

Methods We measured total amino acid (Amino-N), mineral nitrogen (N) and phosphorus (P) concentrations, in situ net N and P mineralization, net Amino-N consumption, and microbial biomass C, N and P in both heath and meadow soils across an elevational gradient near Abisko, Sweden.

Results For the meadow, NH_4^+ concentrations and net N mineralization were highest at high elevations and microbial properties showed variable responses; these

variables were largely unresponsive to elevation for the heath. Amino-N concentrations sometimes showed a tendency to increase with elevation and net Amino-N consumption was often unresponsive to elevation. Overall, $\text{PO}_4\text{-P}$ concentrations decreased with elevation and net P immobilization mostly occurred at lower elevations; these effects were strongest for the heath.

Conclusions Our results reveal that elevation-associated changes in temperature can have contrasting effects on the cycling of N and P in subarctic soils, and that the strength and direction of these effects depend strongly on dominant vegetation type.

Keywords Nutrient availability · Mineralization · Immobilization · Microbial biomass · Amino acids · Temperature

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Introduction

Rising nutrient availability due to temperature-induced increases in mineralization has been suggested as a likely driver of increased plant productivity in arctic tundra (Dormann and Woodin 2002; Chapin et al. 2005; Hill and Henry 2011). However, some studies have found experimental warming in tundra to increase net nutrient mineralization rates (Schmidt et al. 2002), while others have shown it to lead to net nutrient immobilization (Jonasson et al. 1993; Rinnan et al. 2007). Further, little is known about how organic nitrogen (N) in the form of amino acids is affected by increasing temperatures, despite this being an important N source

for plants and microorganisms in high latitude and high altitude ecosystems (Chapin et al. 1993; Persson and Näsholm 2001; Averill and Finzi 2011). While results from experimental warming studies can inform on how changes in nutrient availability can occur in the space of a few years (Hartley et al. 1999; Schmidt et al. 2002), they have generally ran for insufficient time to capture the longer-term effects of warming on nutrient availability and microbial communities (Rinnan et al. 2007). Elevational gradients have the potential to complement experimental manipulations in understanding the effects of temperature on ecosystem processes (Körner 2007), partly because they have been in place for sufficient time for longer term temperature effects to be manifested across sites (Dunne et al. 2004; Fukami and Wardle 2005; Sundqvist et al. 2013). However, while the effect of elevation on mineral and organic nutrient cycling has been studied in some forested systems (e.g. Groffman et al. 2009; Unger et al. 2010; Loomis et al. 2006; Averill and Finzi 2011), little is known about responses of these processes to elevational gradients in treeless tundra ecosystems.

In sub-arctic ecosystems, nutrient availability and net mineralization are known to differ greatly amongst vegetation types (Hobbie et al. 2002; Björk et al. 2007; Chu and Grogan 2010). In the Fennoscandian tundra, two co-dominant vegetation types typically occur at all elevations, i.e., heath dominated by woody dwarf shrub species, and meadow dominated by herbaceous species (Nilsson 1991; Molau and Alatalo 1998). These vegetation types differ in several above- and belowground properties. For example, fungal-to-bacterial ratios are lower, while pH and available concentrations of NH_4^+ -N are higher in the meadow (Björk et al. 2007; Eskelinen et al. 2009). Meanwhile available PO_4 -P concentrations can be higher in the heath (Sundqvist et al. 2011a; Giesler et al. 2012). Recent work has also shown that most belowground properties are more unidirectionally responsive to elevation for the heath (Sundqvist et al. 2011a), with the exception of available soil P which declines with elevation for both heath and meadow (Vincent et al. 2014). In contrast, plant species composition and plant functional leaf and litter traits are more responsive to elevation for the meadow than the heath (Sundqvist et al. 2011a, b, 2012). Notably, while foliar and leaf litter N:P ratios increase with elevation for both vegetation types, this increase is most pronounced for leaf litter from the meadow

(Sundqvist et al. 2011b). Moreover, microbial N and phosphorus (P) pools comprise a large part of the total labile N and P in subarctic ecosystems (Schmidt et al. 2002) and can act as source of nutrients to plants, as well as a nutrient sink through immobilizing mineral nutrients (Achat et al. 2010) and consuming organic N in the form of amino acids (Nordin et al. 2004). However, little is known about how the microbial-driven processes that regulate soil N and P availability respond to elevation for these two contrasting tundra vegetation types.

In this study, we use a well-established elevational gradient across both meadow and heath vegetation in subarctic tundra in northern Sweden (Sundqvist et al. 2011a, b, 2012; Milbau et al. 2013; Vincent et al. 2014) to test three hypotheses. First, we hypothesise that during the growing season, an increase in elevation (i.e. a decrease in temperature) will be associated with a decrease in net N and P mineralization and Amino-N release, due to the negative influence that elevation-associated declines in temperatures can have on microbial activity and nutrient mineralization (Sveinbjörnsson et al. 1995; Averill and Finzi 2011). Second, we hypothesise that these patterns will be more pronounced for the heath than the meadow vegetation, consistent with previous findings that most belowground properties such as soil N availability and fungal:bacterial ratios are more unidirectionally responsive to elevation for the heath (Sundqvist et al. 2011a). Third, because soil microbial nutrient stoichiometry could also inform on microbial nutrient limitation patterns in ecosystems (Cleveland and Liptzin 2007) we hypothesise that variation across the gradient and across vegetation types in N and P dynamics are also reflected in the stoichiometry of microbial carbon (C), N and P. By addressing these hypotheses we aim to advance our understanding on how elevation-associated changes in temperature affect the dynamics of labile N and P pools in the soil for two fundamentally different vegetation types that co-occur in the same landscape.

Materials and methods

Study site

The study was conducted on the north-east facing slope of Mt Suorooaivi (1,193 m), located approximately 20 km south-east of Abisko, Sweden (68°21'N, 18°49'E)

as described by Sundqvist et al. (2011a). The underlying bedrock is dominated by salic igneous rocks (mainly granite and syenite) with potentially some influence of quartic and phyllitic hard schists at the higher elevations (SGU 1965). Mountain birch (*Betula pubescens* ssp. *czerepanovii*) forms the treeline that is situated at an elevation of 500–600 m at the study site. In this system there are two co-dominant field-layer vegetation types that occur in a mosaic over short distances at all elevations, namely heath (dominated by deciduous and evergreen dwarf-shrubs) and meadow (dominated by herbs, graminoids and sedges) with the meadow commonly found in shallow depressions. The differences in vegetation types across the gradient are independent of bedrock and likely to be driven by local scale topography and hydrochemistry and variation in soil pH (Björk et al. 2007; Sundqvist et al. 2011a; Giesler et al. 2012). As such, there is consistently a higher soil pH and Ca content in the meadow than in the heath (Sundqvist et al. 2011a, Appendix 1). These two vegetation types not only differ in several above- and belowground properties, but also in how these properties respond to elevation (Appendix 1; see also Sundqvist et al. 2011a, b, 2012). In September 2007, four replicate square plots (each 2×2 m) were established on each of the two vegetation types, at every 100 m along an elevational gradient ranging from 500 to 1,000 m, yielding a total of 48 plots. Within each elevation, the mean distance of each plot to the next nearest plot was ~15 m, and the mean distance between the two most distant plots was ~100 m, to ensure that pseudoreplication was minimized; because of high spatial heterogeneity over short distances in microtopography, hydrology and soil fertility in these plant communities (Björk et al. 2007), it is expected that this distance among plots is sufficient to ensure adequate independence among them. Plots at the 500 m elevation were situated in open birch forest, plots at 600 m were located immediately above the forest line, and plots from 700 to 1,000 m were devoid of trees. Daily mean air temperatures in July and August 2009 at 500, 700 and 1,000 m at the study site are given in Appendix 2. The average decrease in air temperature with elevation (July–August 2009) was 0.5 °C for every 100 m elevational increase (Appendix 2). The mean annual precipitation in the area (Abisko Scientific Research Station) for the period 1913–2000 was 310 mm; the highest mean monthly precipitation was in July (51 mm) and the lowest in April (12 mm) (Kohler et al. 2006). Further details on the study system are given by Sundqvist et al. (2011a).

Incubation for measuring mineralization

We used the buried-bag technique for determining net N and P mineralization and net Amino-N consumption, as described by Eno (1960) and Adams et al. (1989). Soil sampling took place on three occasions during the growing season in 2009, i.e., July 6, August 4 and September 1. From each plot, several (≥ 4) soil samples were taken from the humus layer using a 45 mm diameter soil corer. The depth of the humus layer in each core was then measured. Sufficient cores were collected to ensure ~ 0.5 L of humus; all cores within a plot were bulked and sieved (2 mm) in the field, after which each sample from each plot was divided into two equally sized subsamples. Each subsample was put into a semi-permeable polyethylene bag. One subsample from each plot was brought back to the laboratory on the same day as sampling and the other subsample was buried in the plot to the full depth of the humus layer and left to incubate in situ until the next sampling occasion (28–29 days later), after which the sample was collected and brought to the laboratory. Soil buried in the humus layer on September 1 was collected on September 29. All samples were kept cold (+2 °C) for a maximum of 24 h before further analysis.

Soil measurements

For all samples brought back to the laboratory, soil pH was measured in a soil:water suspension (50 mL de-ionized water and 6 g fresh soil) after shaking it overnight and leaving it to sediment for 1 h. Soil moisture content was determined after oven-drying (105 °C, 24 h) and soil organic matter (OM) content was determined by loss on ignition in a muffle furnace (550 °C, 4 h). A subsample of 5 g fresh weight humus was extracted in 80 ml 1 M KCl, and the concentrations in the extract were then determined for $\text{NH}_4^+\text{-N}$, $\text{NO}_3^-\text{-N}$, $\text{PO}_4\text{-P}$, and total Amino-N by colorimetry on an AutoAnalyser III (SEAL Analytical, Kontram OmniProcess AB, Sweden). Total Amino-N was analyzed using ninhydrin and lithium acetate as reagents and leucine as a standard (Moore 1968), and total Amino-N concentrations are thus expressed as leucine equivalent concentrations. All nutrient concentrations are expressed as mg g^{-1} OM. For each of the three soil incubations, net mineralization for N (including both $\text{NH}_4^+\text{-N}$ and $\text{NO}_3^-\text{-N}$) and P, as well as the net

release of Amino-N (leucine equivalent) was calculated as the difference between its concentration after the incubation (time t_1) and at the start of the incubation (time t_0). As such, negative values represent net immobilization of N and P, or net consumption Amino-N, respectively. Concentrations of NO_3^- -N were below the detection limit for 93 % of the samples, but when measurable were included in the calculations for net N mineralization.

Microbial carbon, nitrogen and phosphorus

For determination of microbial biomass C, N and P, we determined chloroform-labile C and N and hexanol-labile P on subsamples of each t_0 soil sample collected on August 4 and September 1, 2009, following sample storage at +2 °C for 1–2 days (P) and 4–5 days (C and N). Chloroform-labile C and N (hereafter referred to as microbial C and N) were determined using the CHCl_3 fumigation-extraction procedure (Brookes et al. 1985; Vance et al. 1987). Fumigated (24 h) and corresponding unfumigated soil subsamples of each soil were extracted with 0.5 M K_2SO_4 and filtered (Munktell 00H filter paper, pore size approx. 1 μm ; Grycksbo, Sweden); blanks contained 0.5 M K_2SO_4 alone. Organic C and N in the extracts were measured after a 50-fold dilution by automated combustion (TOC-Vcph analyzer; Shimadzu, Tokyo, Japan). Hexanol-labile P (hereafter referred to as microbial P) for each soil was determined using a simultaneous hexanol fumigation and extraction method with anion exchange resins (BDH # 55164 2S, BDH laboratory supplies, Poole, England) in bicarbonate form for 16 h (Kuono et al. 1995), using hexanol instead of chloroform as a fumigant (McLaughlin et al. 1986). Resins were eluted with 0.1 M NaCl/HCl . Phosphorus in the extracts was measured by molybdate colorimetry after a 5-fold dilution for unfumigated samples or a 15-fold dilution for fumigated samples. Microbial C, N, and P for each soil were each estimated as the difference in the concentration of a given element in fumigated and non-fumigated subsamples. We did not use extractability factors to convert microbial C, N and P into actual biomass values, because these factors are known to be soil-specific and therefore vary considerably among different soils (e.g. Oberson et al. 1997; Turner et al. 2002). Instead we present them as relative figures, i.e. the difference between the fumigated and unfumigated fractions.

Statistical analysis

To analyze the effect of each of our main factors, i.e., month, vegetation type and elevation, and their interactions, on each response variable, we used repeated measures ANOVA. Differences among months were compared using a Bonferroni-test with confidence interval adjustment. Whenever a significant effect of elevation was found, differences among elevations within each vegetation type for each month were analyzed using Tukey's honestly significant difference (h.s.d.) at $P=0.05$. When required, data were transformed to conform to the assumptions of parametric tests. For all statistical analyses we used SPSS Statistics 17.0.

Results

Nutrient concentrations

Overall, concentrations of NH_4^+ -N and total Amino-N were significantly higher in the meadow than in the heath vegetation, while the overall concentrations of PO_4 -P were higher in the heath than in the meadow (Table 1, Fig. 1a–c). Concentrations of NH_4^+ -N, PO_4^- -P and Amino-N were all affected by month, and NH_4^+ -N was also affected by the interactive effect of month and vegetation type (Fig. 1a, Table 1). This is because NH_4^+ -N concentrations were overall significantly higher in July and August than in September for the heath, while it was significantly higher in July than in August and September for the meadow. For both vegetation types, overall Amino-N concentration decreased during the growing season and overall PO_4 -P concentrations were highest in July (Fig. 1b–c, Table 1).

Elevation, as well as its interaction with month and with vegetation type, consistently affected concentrations of NH_4^+ -N, PO_4^- -P and Amino-N, and there was also a significant three-way interaction between elevation, vegetation type and month on NH_4^+ -N concentrations (Fig. 1a–c, Table 1). As such, elevation never had an effect on NH_4^+ -N concentration for the heath, while for the meadow NH_4^+ -N was always highest at 900 m (Fig. 1a). For both vegetation types, concentrations of PO_4 -P were overall highest at the lowest elevation and often lowest at the highest elevation, and these effects were more pronounced in the heath than in the meadow (Fig. 1b). Meanwhile, Amino-N concentrations

Table 1 Effect of month, vegetation type and elevation, and their interactions, as revealed by repeated measures ANOVA (*F* values, with *P* in parentheses), on concentrations of NH_4^+ -N, PO_4 -P andAmino-N (leucine equivalent) in humus, and on net mineralization of N (including NH_4^+ -N and NO_3^- -N) and P as well as net Amino-N consumption (mg g^{-1} OM)

Variables	Month (M)	Within-subjects effects			Between-subjects effects		
		M×Vegetation type (V)	M×Elevation (E)	M×E×V	V	E	V×E
NH_4^+ -N _{conc}	30.4 (<0.001)	32.0 (<0.001)	4.0 (<0.001)	5.5 (<0.001)	395.2 (<0.001)	23.7 (<0.001)	18.3 (<0.001)
PO_4^- -P _{conc}	10.3 (0.003)	0.1 (0.814)	4.7 (0.002)	0.813 (0.549)	24.5 (<0.001)	32.9 (<0.001)	10.6 (<0.001)
Amino-N _{conc}	36.1 (<0.001)	1.1 (0.352)	2.4 (0.019)	1.9 (0.063)	137.0 (<0.001)	4.6 (0.003)	3.1 (0.021)
N _{min}	1.8 (0.173)	0.3 (0.723)	3.1 (0.003)	2.2 (0.029)	5.4 (0.027)	7.5 (<0.001)	7.2 (<0.001)
P _{min}	35.2 (<0.001)	0.2 (0.805)	2.5 (0.011)	1.9 (0.061)	4.7 (0.037)	15.0 (<0.001)	4.1 (0.005)
Amino-N _{cons}	14.7 (0.001)	4.9 (0.033)	2.0 (0.098)	2.3 (0.068)	158.2 (<0.001)	4.3 (0.004)	3.8 (0.008)

Degrees of freedom for nutrient concentration are 2,72 for M, 2,72 for M×V, 10,72 for M×E and 10,72 for M×V×E, 1,36 for V, 5,36 for E and 5,36 for V×E. Degrees of freedom for net mineralization of nutrients are 2,68 for M, 2,68 for M×V, 10,68 for M×E, 10,68 for M×V×E, 1,34 for V, 5,34 for E and 5,34 for V×E. Values in boldface indicate statistical significance at $P \leq 0.05$

conc concentration; *min* mineralization; *cons* consumption

generally increased with elevation in July and August for the heath, while they were significantly (but not unidirectionally) related to elevation in September for the meadow (Fig. 1c).

Mineralization, immobilization and Amino-N consumption

Net mineralization/immobilization rates of N and P and net release/consumption of Amino-N were all significantly affected by vegetation type (Table 1, Fig. 2a–c). The largest values of both net mineralization and immobilization for N were for the meadow, while the largest values for P generally occurred for the heath. There was always a net consumption of Amino-N, and this was highest for the meadow (Table 1, Fig. 2c). Month had a significant effect on net P mineralization/immobilization because for both vegetation types net mineralization generally occurred in August and net immobilization usually occurred in July and September (Table 1, Fig. 2). Net Amino-N consumption was affected by both month and its interaction with vegetation type, because these values were lowest in September for the heath and in August and September for the meadow. This pattern was related to changes over time in initial (pre-incubation) concentrations of Amino-N, which declined during the growing season for both vegetation types (Figs. 1c and 2c). Net N

mineralization did not vary significantly across months for either vegetation type (Table 1).

Net N and P mineralization/immobilization and net release/consumption rates of Amino-N were significantly affected by elevation and by its interaction with vegetation type (Table 1, Fig. 2a–c). Additionally, the effect of elevation on net N and P mineralization/immobilization also varied significantly among months, and there was a significant three-way interaction of elevation, month and vegetation type on net N mineralization (Table 1). As such, net N mineralization was always responsive to elevation for the meadow, with a large variation across lower elevations over the study period but with the greatest net mineralization always occurring at the highest elevation. For the heath this process was only significantly responsive to elevation in August when it was highest at the 600 m elevation (Fig. 2a). Net P immobilization was highest at the lowest elevation for all 3 months for the heath and in July and September for the meadow. Higher elevations generally had low net P immobilization or low net mineralization, with the exception for the heath in August where net mineralization increased with elevation from 600 m (Fig. 2b). Net Amino-N consumption for the heath increased with elevation from 600 m in July (Fig. 2c). For the meadow, this process increased with elevation from 600 m in August and showed an idiosyncratic response to elevation in September. Generally, net consumption of Amino-N was high when the initial Amino-N concentration was also high (Figs. 1c and 2c).

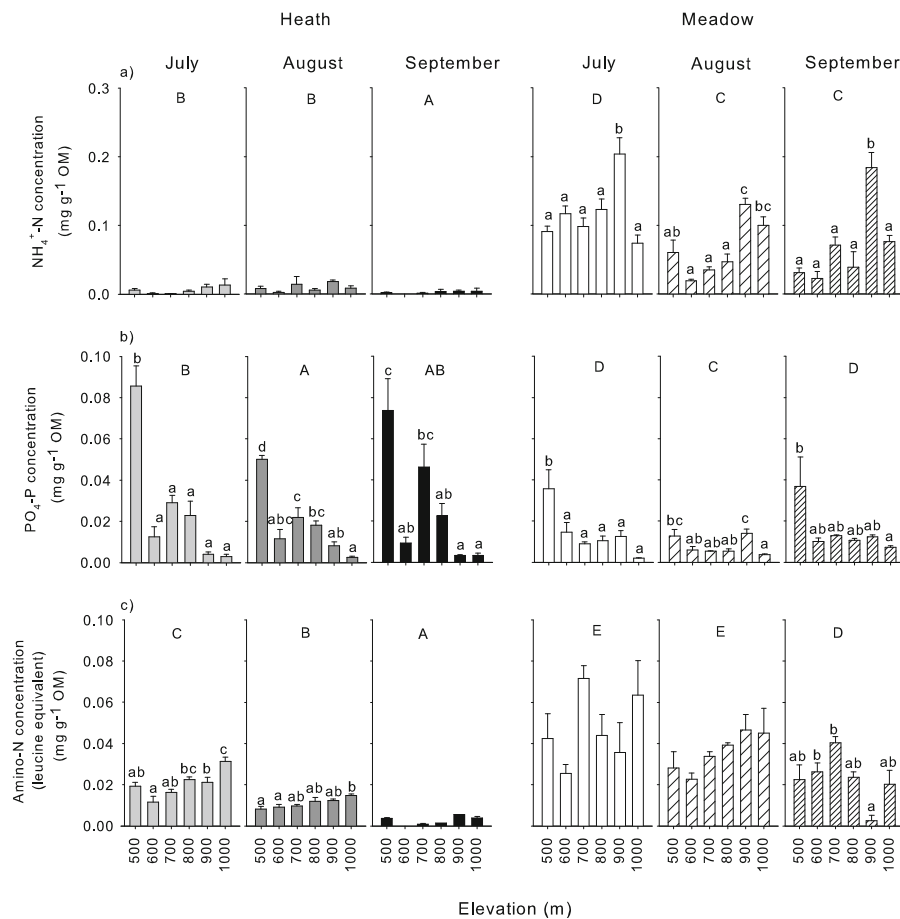


Fig. 1 Initial (t_0) concentrations of $\text{NH}_4^+\text{-N}$, $\text{PO}_4\text{-P}$ and total Amino-N (leucine equivalent) in humus, for heath and meadow vegetation across an elevational gradient sampled in early July, August and September 2009. Error bars=SE ($N=4$). Within each row (i.e., groups of 6 panels), panels topped with the same capital

letters are not significantly different (Bonferroni-test). Within each panel, bars topped with the same lower case letters are not significantly different at $P < 0.05$; Tukey's h.s.d (ANOVA results shown in Table 1)

Microbial variables

Total microbial C, N and P were all significantly higher for the meadow than for the heath, and all three variables were significantly higher in September than in August for the meadow but did not differ between months for the heath (Table 2, Fig. 3a–c). Elevation also had a significant effect on all three variables, and there was an interactive effect of elevation and vegetation type for microbial C and N, and of elevation and month for microbial P (Table 2). For the meadow, microbial C and N were unresponsive to elevation in August and highest at 600 m and lowest at 1,000 m in September (Fig. 3a–b). Microbial P was highest at the 500 m elevation and lowest at the 1,000 m elevation in August, and unresponsive to elevation in September (Fig. 3c). For the heath, microbial

C was highest at 800 m and lowest at 500 m in August, and unresponsive to elevation in September, while microbial N was unresponsive to elevation. Meanwhile, microbial P was unresponsive to elevation in August and highest at 900 m and lowest at 700 m in September.

The microbial C:N ratio was significantly higher for the heath than for the meadow vegetation, and microbial C:P and N:P ratios did not differ among vegetation types (Fig. 4a–c, Table 2). For both vegetation types, the microbial C:N ratio was overall higher in September compared to August, while ratios of C:P did not differ among months for any vegetation type, and ratios of N:P were highest in August than in September only for the meadow. The microbial C:N, C:P and N:P ratios were all affected by elevation and by its interaction with month (Table 2). For both vegetation types, elevation

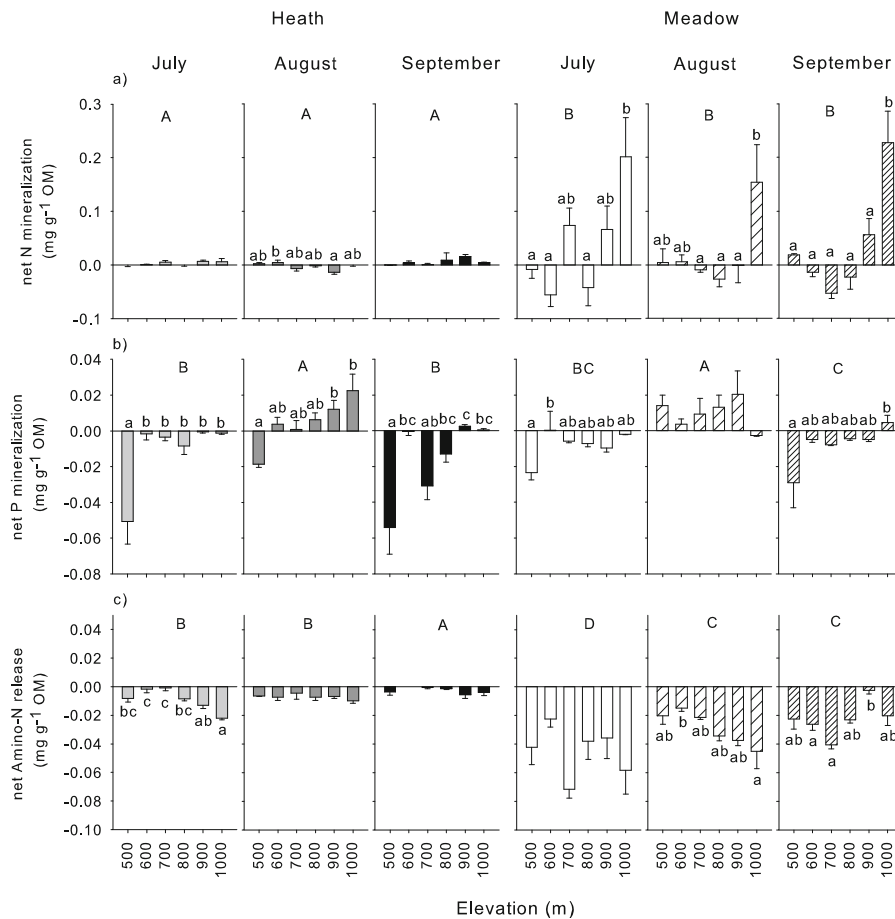


Fig. 2 Net mineralization/immobilization of N (including NH_4^+ -N and NO_3^- -N), PO_4 -P and net Amino-N (leucine equivalent) release in humus, for heath and meadow vegetation across an elevational gradient, after in situ incubations in July, August and September. Error bars=SE ($N=4$, except for 500 and 900 m in

heath in August where $N=3$). Within each row (i.e., groups of 6 panels), panels topped with the same capital letters are not significantly different (Bonferroni-test). Within each graph, bars topped with the same letters are not significantly different at $P<0.05$; Tukey's h.s.d (ANOVA results shown in Table 1)

had a significant but non-unidirectional effect on microbial C:N ratios in August but no effect in September (Fig. 4a). Further, for both vegetation types, the microbial C:P ratio was lowest at the lowest elevation in August while in September it was instead lowest at the highest elevation for the meadow and unresponsive to elevation for the heath (Fig. 4b). The microbial N:P ratio generally increased with elevation in August and was maximal at 600 m in September for the meadow, while this ratio was unresponsive to elevation for the heath (Fig. 4c).

Discussion

Our results revealed that heath and meadow vegetation differed greatly in terms of how N availability, N cycling

and microbial properties responded to elevation, but not in the manner we hypothesized. However, and partly consistent with our hypothesis, we found both PO_4 -P concentrations and net P mineralization/immobilization to respond more strongly to elevation for heath than for meadow. Below, we discuss possible causes for these findings.

Contrasting responses for soil N cycling to elevation for tundra heath and meadow

Inconsistent with our predictions, net N mineralization, net Amino-N consumption rates and NH_4^+ -N concentrations were mostly unaffected by elevation for the heath while these variables often varied for the meadow; here the effects were mostly non-unidirectional except

Table 2 Effect of month of sampling (August and September), vegetation type, elevation, and their interaction as revealed by repeated measures ANOVA (*F* values, with *P* in parentheses) onrelative measures of microbial C, N and P (mg g^{-1} OM) (i.e. the difference between the fumigated and unfumigated fractions) and their ratios

Variables	Month (M)	Within-subject effects			Between-subjects effect		
		M×Vegetation type (V)	M×Elevation (E)	M×E×V	V	E	V×E
C_{micr}	20.8 (<0.001)	13.9 (0.001)	0.936 (0.469)	1.3 (0.289)	63.7 (<0.001)	6.8 (<0.001)	4.2 (0.004)
N_{micr}	11.8 (0.002)	13.8 (0.001)	1.3 (0.291)	1.2 (0.317)	93.9 (<0.001)	4.4 (0.003)	2.9 (0.026)
P_{micr}	40.5 (<0.001)	34.2 (<0.001)	4.1 (0.005)	1.6 (0.172)	76.2 (<0.001)	4.3 (0.004)	1.5 (0.219)
C:N ratio _{micr}	32.6 (<0.001)	1.1 (0.302)	4.3 (0.003)	0.986 (0.440)	22.0 (<0.001)	5.6 (0.001)	1.4 (0.258)
C:P ratio _{micr} ^a	0.4 (0.538)	0.004 (0.949)	3.8 (0.007)	1.1 (0.376)	0.1 (0.755)	6.8 (<0.001)	1.6 (0.185)
N:P ratio _{micr} ^a	6.4 (0.016)	0.3 (0.605)	4.2 (0.004)	1.0 (0.415)	2.9 (0.096)	4.3 (0.004)	1.3 (0.268)

Degrees of freedom are 1,36 for M, 1,36 for M×V, 5,36 for M×E, 5,36 for M×V×E, 1,36 for V, 5,36 for E and 5,36 for V×E. Values in boldface indicate statistical significance at $P \leq 0.05$

micr microbial biomass

^a Analysis was performed on log-transformed data

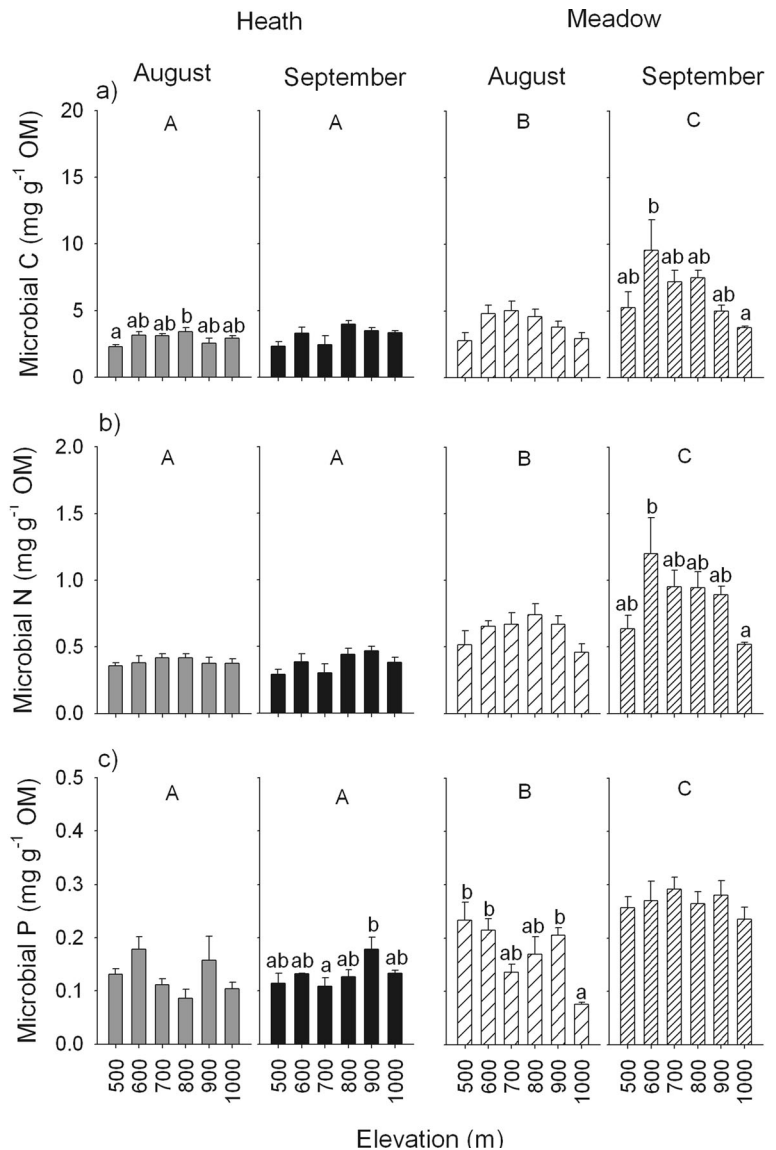
that the highest net N mineralization rates were consistently found at the highest elevation. These results contrast with findings of declining net N mineralization rates with increasing elevation elsewhere (Sveinbjörnsson et al. 1995; Averill and Finzi 2011), and also show that there can be great variation in responses of N cycling processes to elevation for contrasting vegetation types. Further, soil pH is closely linked to bedrock and vegetation patterns as well as N dynamics in these ecosystems (e.g. Björk et al. 2007; Eskelinen et al. 2009; Giesler et al. 2012), and the high mineral N concentrations we found at the 900 m elevation may at least partly be related to the higher pH at this elevation (Fig. 1, Appendix 1). However, since variation in soil pH did not correspond with the patterns we observed for Amino-N concentrations or components of the soil N cycle across the gradient for either vegetation type, our findings point to a greater importance of other factors in driving these patterns and processes. Our results suggest that a temperature change of ~ 3 °C, which is comparable to the temperature range found across our elevational gradient, does not necessarily increase soil N cycling. This is also partly in line with several warming experiments in tundra vegetation which have found only minor effects of rising temperatures on soil N cycling (Jonasson et al. 1993; Schmidt et al. 2002; Rinnan et al. 2007).

Functionally, dry boreal and alpine vegetation such as the heath vegetation is known to have an overall tight

N cycle in which gross N mineralization rates seldom exceed gross immobilization rates (Fisk et al. 1998; Högberg et al. 2006). This may explain the consistently low Amino-N and NH_4^+ -N concentrations and net N mineralization throughout the heath sites, which is in line with findings from the Alaskan tundra (Nordin et al. 2004). Further, because elevation-associated changes in temperature commonly lead to a change in plant community composition (Sundqvist et al. 2013), plant responses to elevation may indirectly influence soil N cycling processes across our study site (Wardle 2002; Laungani and Knops 2012). However, in previous research we did not observe any large changes in plant community composition for the heath vegetation along this gradient (Sundqvist et al. 2011a, Appendix 3). The weak response of net N mineralization to elevation in the heath vegetation could therefore be a consequence of the low turnover of plant species for the heath across the gradient in combination with the overall low C quality of plant litter inputs that is characteristic of tundra heath vegetation (Schmidt et al. 1999; Jonasson et al. 1999). This is further supported by previous findings for the heath across our study site, where the low turnover of plant species with elevation is associated with a low variation across the gradient in community functional litter traits, litter C quality (in the form of phenolics such as tannins) and litter decomposability (Sundqvist et al. 2011a, b, 2012).

In contrast to the heath, meadow vegetation always showed high soil NH_4^+ -N concentrations, suggesting

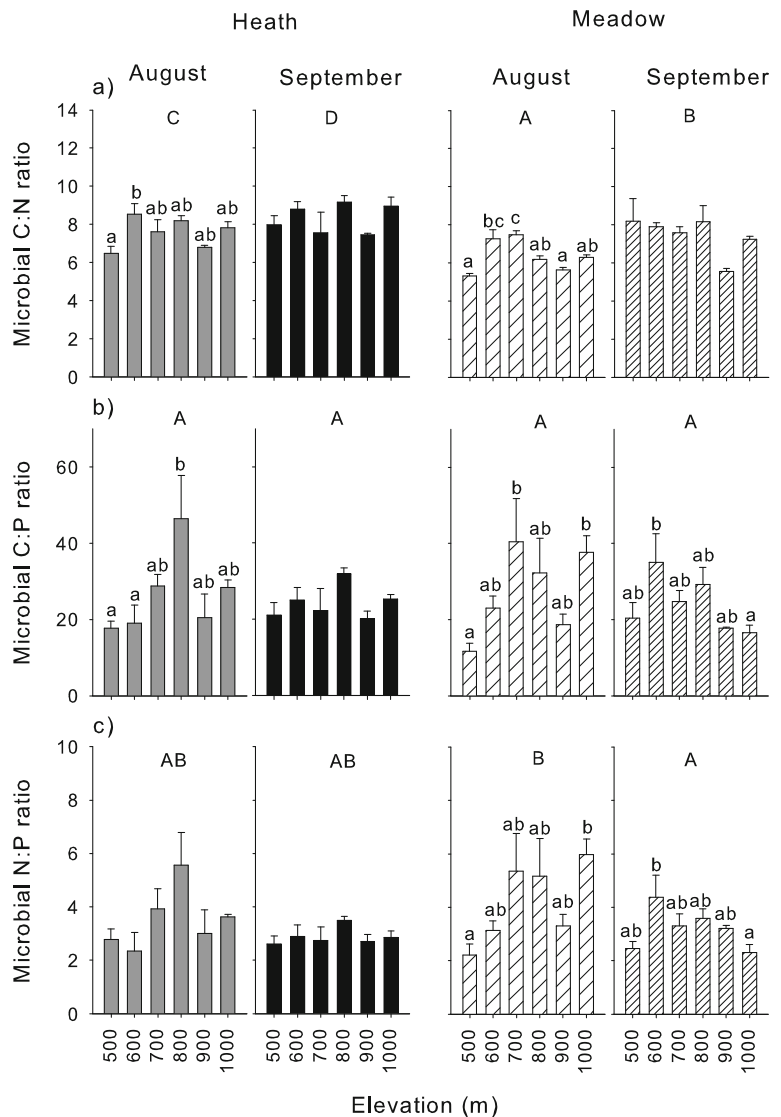
Fig. 3 Relative measures of microbial biomass C, N and P (i.e. the difference between fumigated and non-fumigated fractions) in humus, for heath and meadow vegetation across an elevational gradient sampled early August and September, 2009. Error bars=SE ($N=4$). Within each row (i.e., groups of 4 panels), panels topped with the same capital letters are not significantly different (Bonferroni-test). Within each graph, bars topped with the same letters are not significantly different at $P<0.05$; Tukey's h.s.d (ANOVA results shown in Table 2)



that the meadow vegetation has a looser N cycle than heath, consistent with what has been shown for other tundra environments (Björk et al. 2007; Chu and Grogan 2010) as well for forb dominated understory vegetation in boreal forest (Högberg et al. 2006). Our results for the meadow do, however, differ from previous studies in comparable vegetation (Björk et al. 2007; Chu and Grogan 2010) in that we found low net N mineralization or net N immobilization at all elevations except the highest one. This is in contrast to our predictions and suggests a more complex relationship between microbial N demand and gross N mineralization rate for the meadow than the soil NH₄⁺-N concentrations

indicate. First, the consistently high NH₄⁺-N concentrations in the meadow soils suggest that gross mineralization rates are higher than the rates of plant and microbial uptake (Högberg et al. 2006). Second, our results may indicate that when the influences of plants are excluded (i.e. in our soil incubations) microbial immobilization is favoured and/or gross mineralization rates are reduced. Therefore, the high net N mineralization rates at the highest elevation are likely related to a lower microbial demand for N rather than overall higher gross N mineralization rates. A low microbial demand for N at the highest elevation may be due to a lower C quality at the highest elevation as a result of a shift in the relative

Fig. 4 Microbial C:N, C:P and N:P ratios in humus, for heath and meadow vegetation across an elevational gradient sampled early August and September, 2009. Error bars=SE ($N=4$). Within each row (i.e., groups of 4 panels), panels topped with the same capital letters are not significantly different (Bonferroni-test). Within each graph, bars topped with the same letters are not significantly different at $P<0.05$; Tukey's h.s.d (ANOVA results shown in Table 2)



abundances of different plant functional groups, notably a decreasing cover of forbs and increasing cover of sedges (Appendix 3; Sundqvist et al. 2011a). This is also in line with results from our study site showing that litter decomposability is lowest at the highest elevation for meadow vegetation (Sundqvist et al. 2011b). In any case, these results overall suggest that the direct effect of temperature is not the main factor regulating N cycling in subarctic heath and meadow soils across our study site.

Effect of elevation on soil P cycling

Both the PO_4^- -P concentrations and net P mineralization/immobilization were often highly responsive to elevation

for both vegetation types, and in line with our predictions, more responsive for the heath. However, while PO_4^- -P concentrations generally decreased with elevation, net P immobilization was generally highest at the lowest elevation (Figs. 1 and 2) in contradiction with our first hypothesis. Therefore, our findings of decreasing PO_4^- -P concentrations with increasing elevation could either be related to an increase in plant demand for P, or a decrease in the release of mineral P into the soil solution, with increasing elevation (Vincent et al. 2014). An increase in plant uptake of P with increasing elevation is unlikely because net primary production generally decreases with increasing elevation (e.g. Raich et al. 1997; Jansson et al. 2008), and because foliar P concentrations

often decrease with increasing elevation across our study site (Sundqvist et al. 2011b). We suggest that the low soil $\text{PO}_4\text{-P}$ concentrations at high elevations is therefore more likely due to a decrease in the overall microbial release rates of P.

It has long been hypothesized that in contrast to N, organic P mineralization is generally decoupled from that of C mineralization (McGill and Cole 1981). They proposed that N release is determined through biological mineralization driven by the microbial search for energy, whereas P release is governed by biochemical mineralization driven by the supply and specific need for P. This is because the majority of organic P compounds in soil (which serve as important substrates for P mineralization) are phosphate esters which have a C-O-P bond, meaning that the P is not directly linked to C (Condon et al. 2005); this is in contrast to organic N compounds where N is directly linked to C. Only phosphonates (which constitute ~4 % of total soil P in the study region; Turner et al. 2004) have a direct C-P bond. Our findings of a more unidirectional response of P mineralization compared to soil N cycling processes to elevation are consistent with such a decoupling of N and P mineralization, and suggest a direct effect of elevation-associated changes in temperature on processes rates for net P mineralization across our study site.

Microbial nutrient pools and stoichiometry across the elevation gradient

Our results showed consistently higher microbial C, N and P in the meadow than in the heath, indicating a higher availability of labile C and nutrients (N and P) in meadow compared to heath soil (Schmidt et al. 2002). These differences in microbial C and N are therefore likely to be related to the pattern of greater overall N availability and N cycling that we observed across the meadow compared to the heath plots. Additionally, the greater microbial nutrient pools found in September for the meadow is likely due to a greater microbial immobilization of nutrients at the end of the growing season following plant senescence (Jaeger et al. 1999). These findings further suggests that the temporal variation in microbial nutrient dynamics across the growing season is greater for forb dominated vegetation compared to dwarf-shrub dominated vegetation across our study system.

Our results revealed overall low microbial N:P ratios in comparison with global literature values (Fig. 4,

Cleveland and Liptzin 2007). These findings corroborate the view that N is the main limiting nutrient over P for plants in tundra systems (Aerts and Chapin 2000), and also suggest that N is relatively more limiting than P for microbes in these subarctic tundra soils. Further, previous results from our study site have shown an increase in N:P ratios in plant leaf and litter tissue across the elevational gradient (Sundqvist et al. 2011b), indicative of a shift in the relative importance of N versus P limitation for plant growth across our study system with increasing elevation. However, inconsistent with our third prediction, no such response to elevation was found for soil microbial N:P with the exception of the meadow in August; further, microbial C:N and C:P ratios did not respond in a consistent manner to elevation for either vegetation type. In addition, we only found a decline in microbial P with increasing elevation in the meadow in August and without any associated changes in microbial C and N being apparent (Fig. 3). Our results are, however, consistent with previous findings showing that microbial C:N ratios were lower and microbial biomass N was higher in meadow than in heath sites (Högberg et al. 2006; Chu and Grogan 2010), which is in line with the higher N availability for the meadow sites. Together, these results highlight that elevation may influence the relative importance of N versus P limitation in very different ways for plants and microbes across these two tundra vegetation types, and is consistent with recent findings showing complex interactions between N and P addition, elevation, and plant and microbial communities in these highly nutrient and temperature limited ecosystems (Sundqvist et al. 2014).

Conclusions

In contrast to our predictions, we found a great variability in responses of soil N cycling, microbial C, N and P, and microbial nutrient stoichiometry to elevation across two distinct vegetation types. In contrast, soil P availability and net P mineralization were generally responsive to elevation and displayed similar patterns for heath and meadow vegetation, although these responses were stronger for the heath. These results have several implications. First, and in support of previous findings in subarctic tundra (Jonasson et al. 1993), they indicate that a change in temperature will have contrasting effects on the cycling of N and P in subarctic tundra. They

point to temperature having a direct effect on P cycling potentially via processes that affect the release rates of P in soils, such as enzymatic activity. In contrast, we propose that temperature may influence soil N cycling and microbial nutrient properties more indirectly, potentially via driving shifts in the amount and quality of soil C, and thus the availability of C for microbes. Second, our results show that the responsiveness of soil N and P cycling to elevation can differ across two co-occurring vegetation types, and suggest that, at least for microbial properties and N cycling processes, these patterns may be driven by differences in plant community responses to elevation. Finally, our study highlights that investigating the responses of N and P cycling processes and microbial stoichiometry to elevational gradients for contrasting vegetation types can provide information on the role of climate warming on belowground processes in subarctic tundra landscapes that are relevant at larger spatial and temporal scales than can be achieved through direct experimentation (Sundqvist et al. 2013).

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