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Response of soil organic and inorganic nutrients in alpine soils to a 16-year factorial snow and N-fertilization experiment, Colorado Front Range, USA

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Abstract

Alpine ecosystems are thought to be particularly sensitive to small environmental changes in climate and other parameters due to the plants and soil organisms being on the edge of environmental tolerances. Snow distribution is critical to microclimate in the alpine, affecting soil temperature, growing season duration, and nutrient cycling. Moreover anthropogenic nitrogen (N) deposition over the past half century has had a detrimental impact on temperate ecosystems, resulting in soil acidification and a reduction in plant biodiversity. Here we used a snowfence experiment combined with an N-fertilization experiment, at the NSF-funded Niwot Ridge (NWT) Long-Term Ecological Research (LTER) site (3528 m ASL), to increase our understanding of how changes in snow properties and N deposition may affect soil processes. The snowfence used in this manipulation resulted in a consistent pattern of snow accumulation, from deep snowpacks near the fence to a shallow snowpack away from the snowfence. As measured after 16 years of the experiment, the amount, timing, and duration of snow cover appears to affect soil properties. Under moderate snow cover and without N addition, the total soil organic carbon (TOC) and total nitrogen (TN) were significantly greater than either under deep or shallow snow. Nitrogen amendments in general worked in the opposite direction of snowpack controls on soil processes. The N addition caused a significant increase under the shallow snow treatments for TOC and TN, while there was a significant decrease of these properties under the moderate snow treatment. In the latter case the N addition didn't cause any significant effect on the inorganic N forms but was correlated with a decline of soil pH, and a consequent increase of exchangeable Al and a reduction of exchangeable base cations, which may have influenced soil microbial biomass found in this study. Our results demonstrate how long-term changes in snow properties and N deposition may interact in affecting alpine soil characteristics, with an important response of soil nutrients.

Keywords: Snow, soil, nitrogen, tundra

Introduction

Nutrient availability is an important factor for the functioning of an ecosystem, and is especially critical in extreme environments. Alpine ecosystems are thought to be particularly sensitive to small environmental changes in climate and other parameters due to the plants and soil organisms being on the edge of environmental tolerances (Williams et al., 1998a, 2002). Snow redistribution by wind action is critical to microclimate in the alpine, affecting soil temperature (Walker et al., 1993; Williams et al., 2009), soil moisture (Taylor and Seastedt, 1994), decay rates (O'Lear and Seastedt, 1994), plant productivity (Walker et al., 1994), organic matter accumulation (Burns and Tonkin, 1982), species diversity (Litaor et al., 2008), soil genesis (Schaetzl, 1990; Holtmeier and Broll, 1992) and fluxes of trace gases such as CO_2, N_2O, and CH_4 (Brooks et al., 1997; West et al., 1999, Filippa et al., 2009; Liptzin et al., 2009). It also influences microbial processes, which control gross N mineralization and N immobilization among plant communities (Fisk et al., 1998, Litaor et al., 2002; Edwards et al., 2007).

Most studies of environmental change in the alpine have focused on two key features: precipitation and atmospheric deposition of dissolved inorganic nitrogen in wetfall (DIN = NH_4^+ + NO_3^-) (Williams et al., 1998b). In areas with previously stable snow conditions, climate change may cause a reduction in the depth, duration, and stability of the snow cover (Williams et al., 1998a). Milder winters with thinner or less permanent snow cover are likely to affect the occurrence, depth, and duration of soil frost and are associated with midwinter snowmelts (Austnes et al., 2008). Nevertheless, even in a warming climate, some areas may experience deeper snowpacks and longer snow duration and other areas may have lower snow cover and lower snow duration (Billings, 1968; Williams et al., 2009). Precipitation, much of which occurs in the form of snow, has significantly increased at the Niwot Ridge in the Colorado Front Range since measurements began in the 1950s (Williams et al., 1996). Moreover at the mesotopographic scale there are large differences in snow accumulation driven by the interaction of the snowfall with topography and wind (Figure 1). Changes in wind speed could affect the historical patterns of snow distribution. In particular several regional studies looking at the United States, Australia, China and parts of Europe have shown decreasing wind speeds just above the planet's surface. Climate change, afforestation and urban development had been suggested as possible causes (Vautard et al., 2010). Any changes in the type, timing and total amounts of winter deposition have been shown to potentially feedback on carbon (C) and nitrogen (N) dynamics (Liptzin et al., 2009, Monson et al., 2006; Brooks and Williams, 1999). Atmospheric deposition of DIN has increased three to four-fold at Niwot Ridge over the past decades (Williams and Tonnessen, 2000) (Figure 2). Measures to reduce nitrogen emission have

begun to take effect, and deposition has a downward trend at least in some areas (Kelly et al., 2002). But the deposition is still high; for instance the average deposition in Western Europe is approximately 18 kg ha^{-1} N, whereas the estimated background deposition is approximately 3 kg ha^{-1} N (Galloway et al., 1984).
Anthropogenic DIN deposition over the past half century has had a detrimental impact on temperate ecosystems in Europe and North America, resulting in soil acidification and a reduction in plant biodiversity. During the acidification process, soils release base cations, such as calcium and magnesium, neutralizing the increase in acidity. Once these base cations have been depleted, aluminium is released from the soils, and can potentially reach toxic levels (Bowman et al., 2008).

Present levels of atmospheric deposition at Niwot Ridge and the Green Lakes Valley in the Colorado Front Range are sufficient to cause N-saturation of surface waters (Williams et al., 1996; Williams and Tonnessen, 2000; Elser et al. 2009). Experimental additions of N to alpine tundra have caused changes in individual species abundance, increases in the soil solution concentrations and leaching of nitrate, and increased net nitrification (Bowman et al., 2006; Bobbink et al., 2010). The reduction in species richness due to experimental N addition occurred across all of the tundra communities studied on Niwot Ridge (Seastedt and Vaccaro, 2001). Moreover, the fertilized plots exhibited higher nutrient concentrations in both above- and below-ground plant material compared to controls (Litaor et al., 2008). It has been hypothesized that these changes in plant characteristics may best be explained by changes in soil chemistry that resulted directly or indirectly from the N additions (Seastedt and Vaccaro, 2001).

Snow amounts have decreased in some areas of the western USA (Monson et al., 2006), while in other areas an increase of snow deposition has been found, particularly in the Arctic (Sturm et al., 2005). There, increases in snow depth and duration were driven in part by increases in air temperature and shrub growth. At Niwot Ridge there is also an expansion of shrubs, which may be due to an increase of snow deposition (Williams et al., 1996). Several researchers have tried to simulate the effect of changes in the seasonal snow-cover on soil properties. Experimental snow-removal, as a simulation of a lack of snowcover, has been carried out world-wide in the last decade (Groffman et al., 2001; Decker et al., 2003; Freppaz et al., 2008), while other researchers have used snowfences to experimentally manipulate snow accumulation (Williams et al., 1998; Nobrega and Grogan, 2007), or grooming to change its density (Rixen et al., 2008). A decrease in winter precipitation may result in shorter winter seasons, in more pronounced and more frequent freeze/thaw cycles, and in more days with the soil temperature well below 0°C during winter. The experiments sometimes indicate as a consequence of these phenomena a faster mineralization of N (Panikov and Dedysh, 2000; Grogan et al., 2004; Freppaz et al., 2007), higher N_2O emissions related to freeze thaw cycles (Sharma et al., 2006), and a reduction of respiration rates (Mariko et al., 1994; Melloh and Crill, 1996; Brooks et al., 1997; Welker et al., 2000; Nobrega and Grogan, 2007). At the same time, a lower mineralization of N (Walker et al., 1999; Schimel et al., 2004), lower N_2O emissions (Goldberg et al., 2008), an increase in respiration rates (Nielsen et al., 2001; Goldberg et al., 2008), were also indicated as the consequence of comparable experimental simulations, making it considerably difficult to derive any general conclusion (Henry, 2007). In particular it's still uncertain if changes in snow cover may cause the same changes in soil properties as increasing N deposition. Brooks and Williams (1999) had proposed that increasing snow depth and duration result in increasing mineralization up to a maximum snow depth. Moreover it's still unknown what the potential interaction may be between changes in snow depth and duration and N deposition, with respect to soil properties such as N availability and soil acidification.

Here we used a long-term snowfence experiment, combined with a N-fertilization experiment, at the Niwot Ridge Long-Term Ecological Research (NWT LTER) site, to increase our understanding of how long-term changes in snow properties and N deposition may affect soil processes. The large snowfence used in this experimental manipulation resulted in a consistent pattern of snow accumulation, from early-accumulation and deep snowpacks near the fence to late accumulating and a shallow snowpack away from the snowfence. The snowfence allowed us to investigate three different pedoenvironments: a) a deep snow-wet meadow close to the snowfence, b) a shallow snow-dry meadow some distance from the snowfence and c) a moderate snow-moist meadow in between (Figure 1, 3). The snowfence was erected in 1993 and our soil measurements were made in 2009, providing a 16-year time frame for soil processes to differ. The main goals of this research were to 1) characterize the soil properties under different snow cover conditions and 2) to examine the reaction of the same parameters to N addition under different snow cover depths and duration.

Material and Methods

The study site

The influence of changes in snowpack timing, duration, and depth on an alpine ecosystem was part of an ongoing study at Niwot Ridge, Colorado. The Niwot Ridge LTER site (3528 m ASL) is designed to incorporate continuing studies over many years to assess the effects of changes in snow pack properties on alpine ecology and biogeochemical cycles (Bowman and Seastedt, 2001). The climate is continental, with a mean annual air temperature (MAAT) equal to -3.8°C and the mean annual precipitation equal to 1006 mm (Williams et al., 1996). Daily mean air temperatures during the winter are often below –20°C (Williams et al., 1998). Snowfall contributes about 80% to the annual precipitation (Caine, 1996). Average wind speed is 10-13 m s^{-1} during the winter, and westerly winds prevail at all seasons (Blanken et al., 2009). Due to redistribution of snow, soil moisture is locally much higher (or much lower) than expected given the recorded amount of precipitation (Litaor et al., 2008). Niwot Ridge is a gently undulating interfluve that did not experience Pleistocene Glaciation and therefore this area has more extensive soil development than many other Colorado Alpine areas (Burns, 1980).

Prior to the construction of the snowfence, the area was an ecotone separating moist meadow and dry meadow (Walker et al., 2001), underlain by Pergelic Cryumbrept soils (Burns, 1980). The depth of the A horizon varied from about 0.2 to 0.4 m, overlying unconsolidated granite parent material (Burns, 1980). Soil pH ranged between 5.3 and 5.4 (Seastedt and Vaccaro, 2001), soil total organic carbon (TOC) and total nitrogen (TN) (10-cm depth) ranged respectively between 130-200 g kg^{-1} and 9-15 g kg^{-1}. The original vegetation cover was dominated by the sedge, *Kobresia myosuroides.*with patches of the forb *Acomastylis (= Geum) rossii)* (Walker et al., 1993).

The snowfence

The saddle of NWT is the site of a long-term snowfence study designed to assess the effects of potential climate change on alpine ecology and biogeochemical cycles (Williams et al., 1998a). In October 1993, a 2.6-m tall × 60-m long snowfence of a composite Centaur® polymer wire rail with a density of 50%, was installed in the saddle’s dry alpine meadow perpendicular to the prevailing westerly winds. The presence of the snowfence creates a gradient of snow depth and, consequently, of snow onset, duration, and date of meltout with distance from the snowfence. Here, we characterized the snow gradient into three snow depth sections (Figure 3): deep snow cover (D), moderate snow cover (M), and shallow snow cover (S).

During the first winter of the experiment, the deep snow cover section increased 100 to 200% in depth, and snow duration increased an average of 90 days compared to the pre-fence measurements (Williams et al., 1998a). The increase in snow depth, almost constant through the years, because of the aerodynamics of the snowfence, and snow duration resulted in underlying soil temperatures in January increasing from about -15°C before installation of the snowfence to -5°C after installation of the snowfence (Brooks et al., 1997). Because the wire rail of the fence can create localized summer wind regime changes affecting the microclimate behind the fence, the wire rail is removed each June for the non-snow season and re-installed each subsequent October.

N addition plots

Eight plots were placed in each snow depth sector in 1993. Each plot was 2 m × 2 m; specific treatments of control (deep snow, control (DC), moderate snow, control (MC), and shallow snow, control (SC)) and N additions (deep snow, nitrogen (DN), moderate snow, nitrogen (MN) and shallow snow, nitrogen (SN)) were randomly selected. Each snow depth sector therefore contained a two-factor factorial experiment for nutrient additions with four replicates per treatment. In the summers of 1993, 1994, and 1995, 20 g m^{-2} N was added as ammonium nitrate (NH_4NO_3 was 25% of added N) and ammonium sulfate ($(NH_4)_2SO_4$ was 75% of added N). Beginning in 1996, plots were fertilized using 10 g m^{-2} N as $(NH_4)_2SO_4$. The logic for using ammonium as the dominant form of inorganic N was to minimize leaching of nitrate. Plots were subsequently fertilized at 1996 rates for 1997, and 1998, and thereafter at these rates every other year. The plots were last fertilized before measurements were made in 2008.

Soil sampling and analysis

The field campaign for this study was conducted in 2009. Soil samples were collected in 24 plots using a standard soil corer during the snow free season (September 2009) on the leeward side of the fence corresponding to deep, moderate and shallow snow pack depths. The soil colour was determined in the laboratory after drying (dry) by the Munsell Soil Colour Charts. In the laboratory, samples were dried and passed through a 2-mm sieve in order to separate the fine earth fraction from the rocks (> 2mm). Soil moisture content was determined gravimetrically after drying at $105^{o}C$. Topsoil (A horizon) and subsoil (AB horizon) were analyzed for TOC and TN using a LECO CHN-1000 CHN Analyzer, pH in water and exchangeable cations (Ca, Mg, Al). Additionally, topsoil samples were processed for NH_4^+ and NO_3^- pools, within 12 hours of returning from the field. Fresh soils were sieved and homogenized using a 2-mm sieve. Subsamples of this soil were extracted with K_2SO_4 0.5M (1:5, weight: volume) by shaking at 250 rpm for 60 min and allowing to sit at room temperature for 18 hours. These extracts were filtered through pre-rinsed (300 mL distilled water) Whatman #1 filter paper, and aliquots were analyzed on the Lachat autoanalyzer. NH_4^+ was determined colorimetrically on a Lachat flow injection analyzer using a phenolate reaction enhanced by nitroprusside. NO_3^- was analyzed using a sulphanilamide reaction following reduction to nitrite on a cadmium column. To measure microbial biomass C (C_{mic}) and N

(N_{mic}), a set of samples was fumigated overnight with chloroform and extracted with K_2SO_4 0.5 M in parallel with a set of unfumigated samples (Brookes et al., 1985).

For dissolved organic compounds, subsamples (10 g fresh weight) were shaken with 100mL 0.5M K_2SO_4 for 1 h and the suspension filtered at 0.45 µm under suction. Total dissolved N (TDN) in the extracts was measured as NH_4^- after oxidation of aliquots of extracts with alkaline persulfate.

We compare some of these results to soil cores that were collected before construction of the snowfence on the Niwot Ridge Saddle in the autumn of 1993. These 75 samples were analyzed for organic matter content, TOC, and TN using the same methods as above. These 1993 samples are representative of the baseline (pre-snowfence) soil conditions; raw values are available at http://culter.colorado.edu/exec/.extracttoolA?soilorgm.ts.

The effects of snow and N addition on soil properties in the topsoil and subsoil were tested with a two-factor factorial ANOVA with Tukey Post Hoc Test. Analysis included Spearman's rho non parametric correlation analysis. Data analysis was performed using the SPSS statistical software. Graphs have been produced using R statistical software (R Development Core Team, 2010).

Results

Soil characteristics

The organic matter content was always higher in the topsoil than in the subsoil (p=0.001), with values ranging from 8.1% in the subsoil of SC to 23% in the topsoils of MC and SN (Table 1). The predominant soil colour was very dark brown in the topsoil, while in the subsoil it was more variable (e.g. dull yellowish brown in DC). The C/N ratio ranged between 12.9 in DC and MN topsoils and 14.5 in DC subsoil. Gravimetric water content at the sampling time (September 2009) ranged from 10.3% (subsoil SC) to 36.7% (topsoil MC), and was marginally higher in the topsoil (p=0.06).

In the snow manipulation plots without N addition (control plots), the TOC and TN concentration in the topsoil was significantly higher under the moderate snow cover (Figure 4) than under the deep and shallow snow covers (p<0.05). In the subsoil the TOC concentration was higher under the moderate snow cover, intermediate under the deep snow cover and the lowest under the shallow snow cover (Figure 5). In the deep and shallow control plots the TOC and TN content was significantly lower than the values recorded in 1993, before the snowfence setup, while a slight increase was recorded under the moderate snow cover plots (Figure 6 and 7). In the N addition plots the topsoil TOC and TN content was significantly higher under the shallow snow cover, while no significant difference was found between the deep and moderate snow cover (Figure 4). The N addition plots exhibited reduced amounts of TOC and TN both in the topsoil and subsoil under the moderate snow cover, while an opposite trend was observed under the shallow snow cover. No significant effect was found under the deep snow cover (Table 2). In the N addition plots the TOC and TN content were significantly lower than the values recorded in 1993 in the deep and moderate snow cover plots (Figure 6 and 7).

Soil pH and Al

The pH of the shallow control plot in the topsoil was about 5.6, significantly higher ($p=0.041$) compared to that measured under the deep and moderate snow covers (Figure 8). In the subsoil the pH under all controls was similar to that of the shallow control plot in the topsoil, near 5.6 (Figure 8). In the control plots, both in the topsoil and subsoil, a significant and negative correlation was found between the pH and the TOC concentration ($r = -0.56$; $p=0.02$). The N addition plots exhibited lower values of pH for the moderate and shallow snow covers and for both topsoil ($p=0.001$) and subsoil ($p=0.001$) (Figure 8). The change in pH was most striking for the moderate snow cover, which was about 4.4 for the topsoil and 4.7 in the subsoil. Using all results, a significant and negative correlation was found between the pH and the DOC concentration ($r = –0.686$; $p=0.007$).

In the control plots, the exchangeable Al concentration was less than 0.2 $\mathrm{cmol_c\ kg^{-1}}$, with no significant differences among the different snow treatments, both in the topsoil and subsoil (Figure 9). Conversely, the N addition plots had increased values of of exchangeable Al, particularly evident under the moderate snow cover (Figure 9), with values greater than 3.3 $\mathrm{cmol_c\ kg^{-1}}$ for both the topsoil and the subsoil. Considering both the control and N addition plots, a significant correlation was found between pH and exchangeable Al in the topsoil ($r=-0.903$, $p<0.01$) and subsoil ($r=-0.933$, $p<0.01$).

Soil exchangeable base cations

The exchangeable base cations in the control plots ranged widely, from about 10 to more than 15 $\mathrm{cmol_c\ kg^{-1}}$ for Ca and 1.2 to 2.4 $\mathrm{cmol_c\ kg^{-1}}$ for Mg (Figures 10 and 11). The concentration of Ca and Mg in the control plots was not significantly different among snow treatments. The exchangeable Ca and Mg were higher in the upper horizon than in the subsoil ($p<0.001$). The N addition caused a significant decrease of exchangeable Ca and Mg (Figures 10 and 11) in the topsoil and subsoil under moderate snow cover, while no significant effect was found under the deep and shallow snow covers, though in the latter case a small increase was measured. Moreover the N addition caused a significant reduction of exchangeable Mg in the subsoil under the deep snow treatment.

Topsoil N and C forms

In the control plots the NH_4^+ and NO_3^- concentrations were significantly greater under deep and moderate snow cover than under the shallow snow cover (Table 3). The K_2SO_4-extractable DOC and DON concentrations in the control plots were significantly greater under the moderate snow cover (Table 3, p = 0.001). Similarly, N_{micr} and C_{micr} concentrations were all significantly greater under the moderate snow cover (Table 3).

The N addition plots exhibited increased NH_4^+ and NO_3^- concentrations only under the shallow snow cover and of NH_4^+ under the deep snow cover (Table 3). The N plots showed increased DOC concentrations under the shallow snow cover (Table 3). In contrast to the control plots, the N addition plots exhibited decreased N_{micr} and C_{micr} concentrations under the moderate snow cover, while no significant effect was recorded under the deep and shallow snow covers (Table 3). The C_{micr}/N_{micr} ratio ranged between 8.01 in DC and 9.17 in MN (Table 3), and significantly decreased in the N addition plots under the shallow snow cover ($p<0.05$).

Results from the general linear model (2-way ANOVA) support the observation that the snow manipulation resulted in a significant effect on almost all C and N variables, excluding NH_4^+ ($p=0.577$) and NO_3^- ($p=0.667$). In the latter cases a significant interaction between the snow depth and N addition was found ($p<0.001$; $p<0.05$). Among the tested variables, only N_{micr} ($p=0.067$) and C_{micr} ($p=0.061$) did not show a relationship with N addition (Table 4 and 5), while a significant interaction between snow depth and N addition was found ($p<0.001$).

Discussion

The soils at Niwot Ridge appear similar to tundra soils in Europe and at high latitudes, revealing a good incorporation of organic matter in these organo-mineral horizons. The TOC content ranged from 47 to 134 g kg^{-1}, slightly higher than values reported for example in the A horizons of tundra soils in the Italian Alps (Freppaz et al., 2010), These values are comparable to the values recorded in mountain grassland soils of the Pyrenees (Garcia-Pausas et al., 2007), at elevations ranging from 1845 m ASL to 2900 m ASL, and to the values reported for Arctic and Alpine tundra by McGuire et al. (1997), without considering the most recalcitrant fractions.

Over the 16 years of the snowfence experiment, the amount, timing, and duration of snow cover appear to affect soil TOC and TN content. The soil under the moderate snow cover, without N addition, revealed a significantly greater TOC and TN content than either under deep or shallow snow (Figure 4 and 5). Brooks and Williams (1999) developed a conceptual model to understand how the seasonal snowpack may control under-snow microbial activity and the release/retention of nitrate. That model may help explain the differences in soil nutrients that we report for differences in snow accumulation. Our moderate snow cover corresponds to zone III of Brooks and Williams (1999), where the snow cover develops early in the season and soils typically do not experience severe freeze/thaw events. Here free water is available throughout the winter and heterotrophic activity continues through the winter and N retention is relatively high. By the comparison with the TOC and TN content in 1993, before the snowfence setup, under the shallow (S) and deep (D) snow cover there was a significant reduction of both TOC and TN content. At sites with a very short duration snow cover (zone I in Brooks and Williams (1999) and sites S in our study) the soil remains frozen through much of the winter, there is very little free water available, and over-winter heterotrophic activity is very low. Consequently, there is a very weak N sink and N leachate is high. Here the vegetation is sparse and consequently we expect low production of OM. In zone IV of Brooks and Williams (1999) (D in our study) snow cover is present for much of the year, occasionally never melting in large snow years. Microbial activity under snow in zone IV is reduced because there is very little primary production during the growing season to provide carbon substrate. In these areas there is a weak N sink during snowmelt and NO_3^- stored in the seasonal snowpack contributes directly to snow melt runoff. These limiting factors could result in a lower concentration of TN in the upper horizons, in comparison to the moderate snow cover area, where the N retention is relatively high.

Moderate snow depths may lead to an ideal combination of moister and warmer soil conditions that result in substantially increased C accumulation relative to deeper or shallower snow depths. Williams et al. (1998a) have shown that there is an increased rate of litter decomposition (up to 50%) under deeper and earlier snowpacks at the Saddle site on Niwot Ridge that can result in increased rates of C and N mineralization with earlier and deeper snow. Our results and those of Williams et al. (1998a) are consistent with previous litter decomposition experiments by O'Lear and Seastedt (1994) on Niwot Ridge that have shown that alpine litter does exhibit significant decay under snow. The snowpack enhances moisture content and increases winter soil surface temperatures, stimulating surface decomposition. The reduction of soil organic matter under the deepest snowpacks is consistent with Webber et al. (1976), who speculated that excessive snow cover is expected to ultimately reduce decomposition. Thus, these results are consistent with Zone

III of Brooks and Williams (1998), with the addition that moderate snow depths result in an increase in microbial biomass, higher rates of mineralization and decomposition, which in turns results in higher soil C, N and organic matter content.

Soil conditions under alpine snow packs can be very favourable to microbial growth (Brooks et al., 1998; Lipson et al., 1999) and the highest populations of microbes on an annual basis occur during the winter in alpine tundra soils (Lipson et al., 1999, Schadt et al., 2003). Our results show that microbial biomass C and N concentrations were significantly greater under the moderate snow depths when compared to deep and shallow control snowpacks. Microbial biomass contained a relatively constant proportion of TOC (0.8-1.2%). These percentages are lower than what reported for example by Cheng and Virginia (1993), across seven Alaskan tundra sites (2.5-2.7%). The same authors reported that the N incorporated in the soil microorganisms was about 7% of the total soil N. In our study the values are lower, ranging from 1.3 to 1.8%, with percentages slightly higher in the control plots than in the N addition plots.

The pH of the shallow control plot in the topsoil of about 5.6 is on the lower edge of soil pH values reported by Burns (1980) for Niwot Ridge. Here, the large additions of inorganic N as well as the addition of sulphate in the fertilizer amendments beginning in 1996 likely created conditions for soil acidification. The fact that pH declined in fertilizer treatments should not be surprising. However, the fact that the various treatments exhibited variable responses in soil pH show that the snow amounts and subsequent community response altered rates of soil acidification. The significantly lower pH values under the moderate snow depths reflect increased mineralization rates related to the snow conditions. Higher rates of mineralization under the deeper snow may move the system towards net nitrification, which would result in lower soil pH. Higher mineralization rates and lower soil pH is consistent with the increased amounts of soil microbial biomass that we report under the moderate snow depths. Exchangeable aluminium values in the control plots show little variation and are consistent with the relatively high soil pH values (Bowman et al., 2008).

The elevated calcium and magnesium values in the topsoil of the moderate and deep snowpacks relative to the shallow snowpack may reflect aeolian deposition. At the NWT LTER site, we know that aeolian deposition is an important source of base cations (Litaor, 1987; Rhoades et al., 2010). The majority of annual dust deposition occurs as dust on snow events, with much less dust deposition during the summer. Dust is entrained in snow, and dust deposition in this region during the period has been large (Corey et al., 2009), maybe buffering the higher leaching due to the greater snow depth. Regions with large annual inputs of loess, such as Prudhoe Bay, Alaska, have nonacidic snowbeds and support relatively rich plant communities (Walker et al., 2001b). For example, the amount of dust trapped over the winter and spring of 1997-1998 in the seasonal snowpack at the NWT LTER site was 43 kg ha^{-1}, compared to a summer value of 7.7 kg ha^{-1} (Ley et al., 2004). The Ca^{2+} and Mg^{2+} concentrations of a dust event snowfall in the Southern Rocky Mountains in February 2006 were respectively 35 and 9-fold higher than previous snow (Rhoades et al., 2010). Most likely, the deeper the snowpack, the more aeolian deposition of dust. Thus, there may be higher amounts of calcium and magnesium loading from dryfall under the moderate and deep snowpacks compared to the shallow snowpack. Aeolian deposition as an important source of exchangeable calcium and magnesium is also consistent with higher values in the topsoil compared to the subsoil. The felsic crystalline rocks that underlie the soils on Niwot Ridge (Williams et al., 2006) weather slowly and release only small amounts of calcium and magnesium relative to aeolian inputs.

Nitrogen amendments in general worked in the opposite direction of snowpack controls on soil processes. For both the topsoil and the subsoil, the N addition caused a significant increase under the shallow snow treatments for soil organic matter, and TN, while there was a significant decrease of these values under the moderate snow treatment (Table 2). Similarly, while there was a significant decrease in microbial biomass C and N under the moderate snowpack with N fertilization, the N-amendments resulted in either no change or an increase in these variables for deep and shallow snowpacks.

Brooks et al. (1996, 1997, 1998) have shown that carbon and nitrogen mineralization along with microbial biomass are inhibited under shallow snowpacks when compared to deeper snowpacks because consistent snow cover insulated the soil surface from extreme air temperatures and allowed heterotrophic activity to continue through much of the winter, while under shallow snowpacks the soil remained frozen and production did not begin until snowmelt. However, Seastedt and Vaccaro (2001) showed that N-fertilization at the shallow snow cover sites enhanced net primary production. Moreover, we show that the increase of inorganic N forms due to the N addition in shallow snow sites was significant. These results are consistent with Schmidt et al. (2004) in dry meadow tundra soils at Niwot Ridge who found a significant increase both of ammonium and nitrate after N fertilization. Under the shallow snowpack, the pH of 5.6 for topsoil and 5.4 for subsoil for controls were both reduced to a pH of 5.0 after N-fertilization. The net production of hydrogen ions is consistent with N-fertilization enhancing mineralization activities under shallow snowpacks. Thus, for shallow snowpacks (and possibly to some extent under deep snowpacks), N-fertilization may compensate for the lack of inorganic N production from under-snow mineralization, resulting in an increase in net primary productivity, soil organic matter, and TN, as well as inorganic N pools.

An intriguing question is why the levels of soil organic matter, TN, and microbial biomass were lower in soils from fertilized plots under moderate snowpacks than in soil from control plots. In the moderate snow cover areas, nitrogen fertilization has allowed for a grass (*Deschampsia* sp) to replace a forb (*Geum* sp; Bowman et al., 1995). This grass produces litter with a higher decomposition rate (Stelzer and Bowman, 1998), which may have resulted in an overall soil organic matter decline.

Microbial biomass and activity have been shown to be significantly suppressed by mid- to long-term (> 1 year) additions of nitrogen in a number of different studies (Fisk and Fahey, 2001; Prescott et al., 1992; Söderström et al., 1983). Nitrogen fertilization not only increases levels of nitrogen in the soil but also can decrease soil pH, even in already acidic soils (Aerts and de Caluwe, 1999; Fisk and Schmidt, 1996). This was the case in the present study because the pH under the moderate snow cover averaged 4.3 in plots receiving N compared to 5.3 for control plots. It is not clear if this sharp pH change alone could be the cause of the reduced microbial biomass recorded in this study. The effect of fertilization could be higher in the summer, when soils were sampled, than in the winter and this is consistent with recent findings that there is a profound microbial community shift from summer to winter in tundra soils (Lipson et al., 2002). The C:N ratio of the chloroform-labile microbial biomass ranged from 8 under deep snow cover without N addition and 9.1 under shallow snow cover without N addition and under moderate snow cover with N addition. A significant reduction of the microbial C/N ratio due to the N addition was recorded only under the shallow snow cover, as reported also by Schimdt et al. (2004), who in alpine tundra soils reported a C/N ratio of 8.3 in the control plots and of 5.0 in the fertilized plots, indicating an increased capacity to immobilize N in the fertilized plots.

The reduction of exchangeable base cations due to N addition was more significant under the moderate snow cover, lowering their availability as nutrients and decreasing their potential contribution to soil buffering capacity. Consequently a lower plant biomass production could be expected, with a decrease of organic matter input into the soil which could contribute to explain the reduced soil organic matter content recorded under the moderate snow cover after N addition. In the same experimental plots, the N addition strongly reduced the plant species richness (Seastedt and

Vaccaro, 2001). Bowman et al. (2008) in the Western Tatra Mountains of Slovakia reported how the above-ground plant biomass decreased with increasing inputs of inorganic N in the study plots. Multiple decades of elevated anthropogenic N deposition may have alleviated any pre-existing N limitation of production, and simultaneously exacerbated plant P limitation through higher P occlusion with increasing soil acidification, as reported by Bowman et al., (2008).

From these results it appeared how any changes in the snow distribution pattern could strongly influence the soil properties, altering the mineralization processes, and consequently with an important response of soil nutrients. The N addition in general worked in the opposite direction of snowpack controls on soil processes, resulting in an interaction between these factors, with significant effect on high elevation seasonally snow covered pedoenvironments.

Conclusions

A greater content of organic matter and microbial biomass occurred under the moderate snow cover than under the other snow treatments. Under the moderate snow cover N additions caused a sharp decrease of soil pH, with a significant increase of exchangeable Al and a reduction of exchangeable base cations, which limit the plant growth and reduce the microbial biomass. Consequently a reduction of the soil TOC and TN content was observed in the N addition plots.

Under the shallow snow cover a lower content of organic matter, microbial biomass and inorganic N forms than the other snow treatments was found. Under the shallow snow cover the N addition caused a decrease of soil pH, which reached values higher than values measured in the N addition plots under moderate snow cover, causing only a slight increase of exchangeable Al, mainly in the subsoil, but no significant effect on exchangeable base cations. Here the N addition caused a great increase of TOC and TN both in the topsoil and subsoil, and of DOC and inorganic N forms in the topsoil.

Under the deep snow cover the organic matter and total nitrogen content was slightly greater than under the shallow snow cover, but lower than under the moderate snow cover, revealing how in this zone the soil properties are affected by the significant snow accumulation. Under the deep snow cover the N addition caused a slight decrease of soil pH and consequently the exchangeable Al only slightly increased. The TOC, TN, and inorganic N content slightly increased due to the N addition, while the microbial biomass was unchanged.

These results demonstrate how long-term changes in snow properties and excessive N deposition may interact, sometimes with contrasting effects, in affecting alpine soil characteristics, with an important response of soil nutrients. The curvilinear responses often observed here argue that biotic and soil changes in response to multiple drivers such as climate and N enrichment are produced by the interaction of the factors rather than by linear combinations of these drivers.

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Caption of tables and figures

Table 1 Mean soil characteristics. Organic matter (OM), Soil colour, C:N ratio (C/N), Water content (WC).

Table 2 TOC, TN concentration in the control plots (DC, MC, SC) and N addition plots (DN, MN, SN) in the topsoil and subsoil. In the column Δ are reported the differences between the N addition and the control plots. * indicate significant differences between means ($p<0.05$).

Table 3 Mean (± standard deviation) N forms (topsoil) in the control and N addition plots under deep (D), moderate (M) and shallow snow covers (S). In brackets % of TN for N forms and % of TOC for C forms. Different letters in the same columns indicate significant differences between means ($p<0.05$).

Table 4 Results of ANOVA for snow depth and N addition on NH_4^+ (a) and NO_3^- (b)

Table 5 Results of ANOVA for snow depth and N addition on N_{micr} (a) and C_{micr} (b)

Figure 1 Alpine mesotopographic gradient at Niwot Ridge (3528 m ASL).

Figure 2 Annual inorganic nitrogen (DIN) wet depositions at Niwot Ridge (data from the NADP program).

Figure 3 Snowfence at Niwot Ridge. In brackets the mean snow depths in the different sectors. Data (n= 24) collected at about maximum snow accumulation in 1993 (4/21/1993, gray line) and 1994 (4/18/94, black line).

Figure 4 TOC and TN concentration (g/kg) in the topsoil under the different snow cover treatments. Different letters indicate significant differences between means ($p<0.05$).

Figure 5 TOC and TN concentration (g/kg) in the subsoil under the different snow cover treatments. Different letters indicate significant differences between means ($p<0.05$).

Figure 6 Total organic carbon **(**TOC) content (gkg^{-1}) in the topsoil of the control and N addition plots under deep, moderate and shallow snow cover in 2009 and 1993 (pre-snowfence).

Figure 7 Total nitrogen **(**TN) content (g/kg) in the topsoil of the control and N addition plots under deep, moderate and shallow snow cover in 2009 and 1993 (pre-snowfence).

Figure 8 pH of the topsoil and subsoil under deep, moderate and shallow snow cover, in the control, and in the N addition plots. Different letters indicate significant differences between means ($p<0.05$).

Figure 9 Exchangeable Al of the topsoil and subsoil under deep, moderate and shallow snow cover, in the control and in the N addition plots. Different letters indicate significant differences between means ($p<0.05$).

Figure 10 Exchangeable Ca^{2+} of the topsoil and subsoil under deep, moderate and shallow snow cover, in the control and in the N addition plots. Different letters indicate significant differences between means ($p<0.05$).

Figure 11 Exchangeable Mg^{2+} of the topsoil and subsoil under deep, moderate and shallow snow cover, in the control and in the N addition plots. Different letters indicate significant differences between means ($p<0.05$).

Tables

Table 1 Mean soil characteristics. Organic matter (OM), Soil colour, C:N ratio (C/N), Water content (WC).

Site	**Horizon**	**OM**	**Soil colour**	**C/N**	**WC**
		%	dry		%
	topsoil				
DC		16.2	10YR2/2	12.9	24.4
DN		17.7	10YR2/2	14.2	33.1
MC		22.9	10YR2/2	13.5	36.7
MN		18.6	10YR2/2	12.9	15.5
SC		14.8	10YR2/2	13.9	20.4
SN		23.0	10YR3/2	13.0	25.6
	subsoil				
DC		9.8	10YR4/3	14.5	15.1
DN		11.2	10YR2/2	13.9	24.9
MC		14.6	10YR2/1	13.5	21.1
MN		10.1	10YR3/3	13.8	17.3
SC		8.1	10YR3/2	13.6	10.3
SN		10.8	10YR3/2	13.8	18.3

Table 2 TOC, TN concentration in the control plots (DC, MC, SC) and N addition plots (DN, MN, SN) in the topsoil and subsoil. In the column Δ are reported the differences between the N addition and the control plots. * indicate significant differences between means ($p<0.05$).

Horizon			**DC**	**DN**	**Δ**	**MC**	**MN**	**Δ**	**SC**	**SN**	**Δ**
topsoil	**TOC**	gkg^{-1}	94	103	+9	134	108	-26*	86	134	+48*
	TN	gkg^{-1}	7.4	7.3	+0.1	9.9	8.4	-1.5*	6.2	10.2	+4.0*
subsoil	**TOC**	gkg^{-1}	56.9	64.9	+8.0	84.8	59.4	-25.4*	46.8	62.7	+15.9*
	TN	gkg^{-1}	3.9	4.7	+0.8	6.3	4.3	-2.0*	3.4	4.5	+1.1*

Table 3 Mean (± standard deviation) N forms (topsoil) in the control and N addition plots under deep (D), moderate (M) and shallow snow covers (S). In brackets % of TN for N forms and % of TOC for C forms. Different letters in the same columns indicate significant differences between means ($p<0.05$).

		NH_4^+		**NO_3^-**		**DON**		**DOC**		**C_{micr}**		**N_{micr}**		**C/N_{micr}**
		$mgkg^{-1}$		$mgkg^{-1}$		$mgkg^{-1}$		$mgkg^{-1}$		$mgkg^{-1}$		$mgkg^{-1}$		
	D	4.26±0.91a	(0.06)	2.45±0.77b	(0.02)	13.05±1.90c	(0.18)	170.57±23.17b	(0.18)	1086.74±148.34b	(1.15)	135.43±15.73c	(1.84)	8.01±0.17a
Control	**M**	4.96±0.54a	(0.05)	1.82±0.55ab	(0.02)	24.04±1.70ab	(0.24)	266.62±6.19ab	(0.20)	1450.93±91.93a	(1.09)	167.05±17.17a	(1.69)	8.72±0.46bc
	S	2.52±0.68b	(0.04)	0.87±0.10a	(0.01)	19.02±3.81bc	(0.31)	177.63±36.66b	(0.21)	933.22±144.97b	(1.08)	102.71±12.41b	(1.65)	9.07±0.51c
	D	6.35±0.54c	(0.09)	2.55±0.29b	(0.03)	20.62±2.01b	(0.28)	227.05±21.52bc	(0.22)	1085.17±98.61b	(1.05)	133.34±7.38bc	(1.82)	8.13±0.44ab
Nadd	**M**	4.80±0.95a	(0.06)	3.07±1.18bc	(0.04)	31.66±5.21a	(0.38)	338.29±31.67a	(0.31)	978.12±94.80b	(0.90)	107.15±13.31bc	(1.28)	9.17±0.66c
	S	7.51±1.15c	(0.07)	3.57±0.45c	(0.03)	25.43±9.16ab	(0.25)	292.81±93.54ac	(0.22)	1102.18±153.68b	(0.82)	131.79±14.66bc	(1.29)	8.34±0.32a

Table 4 Results of ANOVA for snow depth and N addition on NH_4^+ (a) and NO_3^- (b)

a)				
Variable	**df**	**MSE**	**F**	**P**
N addition	1	31.945	46.768	0.000
Snow depth	2	0.387	0.567	0.577
N addition * Snow depth	2	13.346	19.539	0.000
error	12.295			
b)				
Variable	**df**	**MSE**	**F**	**P**
N addition	1	10.966	25.466	0.000
Snow depth	2	0.179	0.415	0.667
N addition * Snow depth	2	3.409	7.916	0.003
error	7.751			

Table 5 Results of ANOVA for snow depth and N addition on N_{micr} (a) and C_{micr} (b)

a)

Variable	**df**	**MSE**	**F**	**P**
N addition	1	721.25	3.788	0.067
Snow depth	2	927.100	4.868	0.020
N addition * Snow depth	2	4076.524	21.404	0.000
error	18			

b)

Variable	**df**	**MSE**	**F**	**P**
N addition	1	62188.785	3.978	0.061
Snow depth	2	79904.356	5.111	0.017
N addition * Snow depth	2	221003.329	14.137	0.000
error	18			

Figures

Figure 1 Alpine mesotopographic gradient at Niwot Ridge (3528 m ASL).

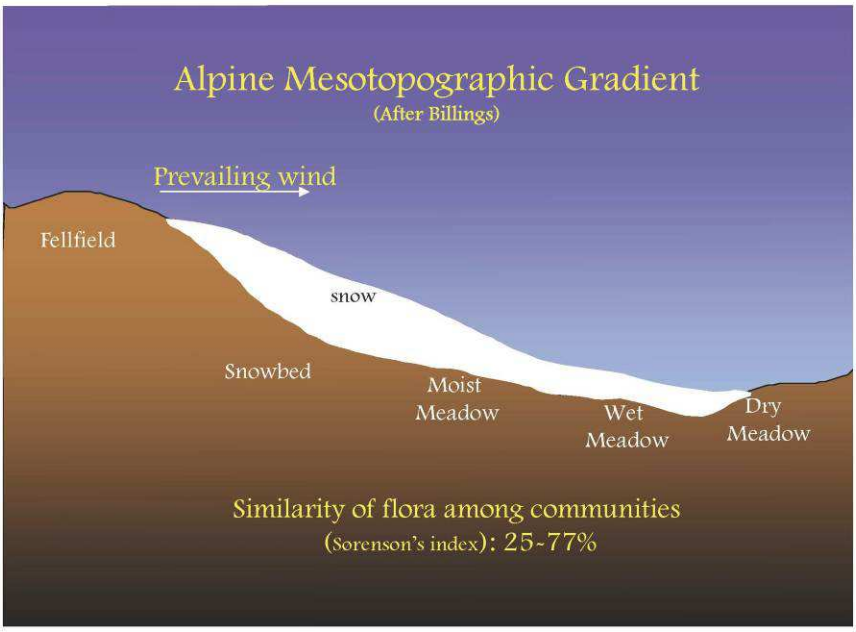

Figure 2 Annual inorganic nitrogen (DIN) wet depositions at Niwot Ridge (data from the NADP program).

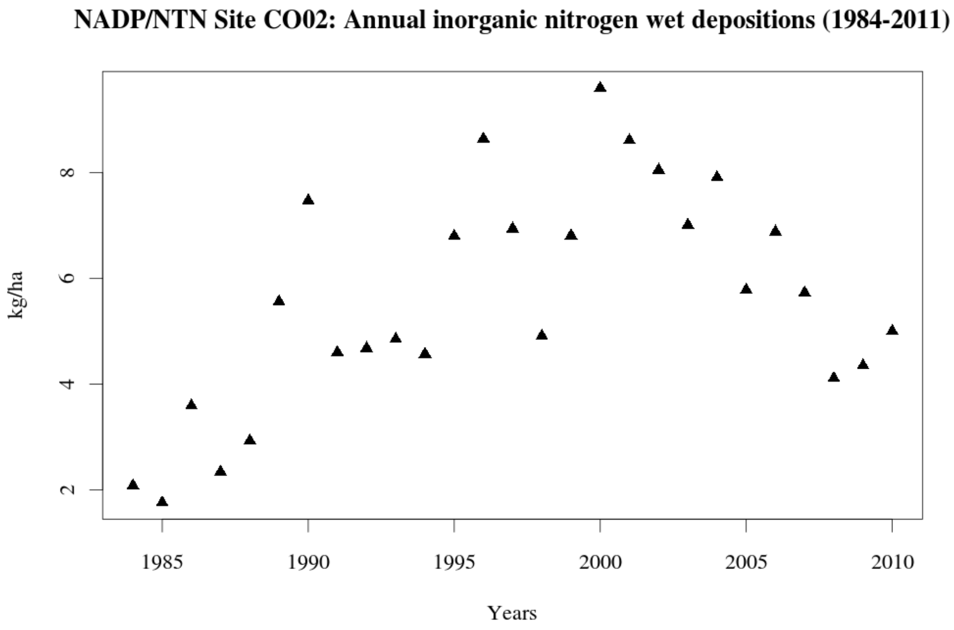

Figure 3 Snow fence at Niwot Ridge. In brackets the mean snow depths in the different sectors. Data (n= 24) collected at about maximum snow accumulation in 1993 (4/21/1993, gray line), prior to the snowfence setup, and 1994 (4/18/94, black line).

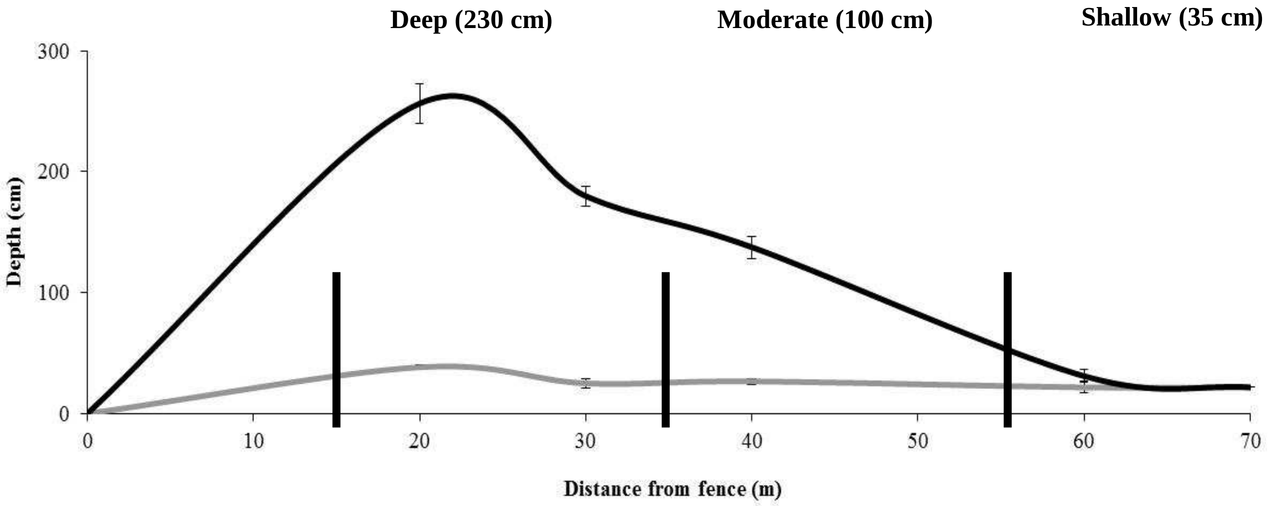

Figure 4 TOC and TN concentration (g/kg) in the topsoil under the different snow cover treatments. Different letters indicate significant differences between means ($p<0.05$).

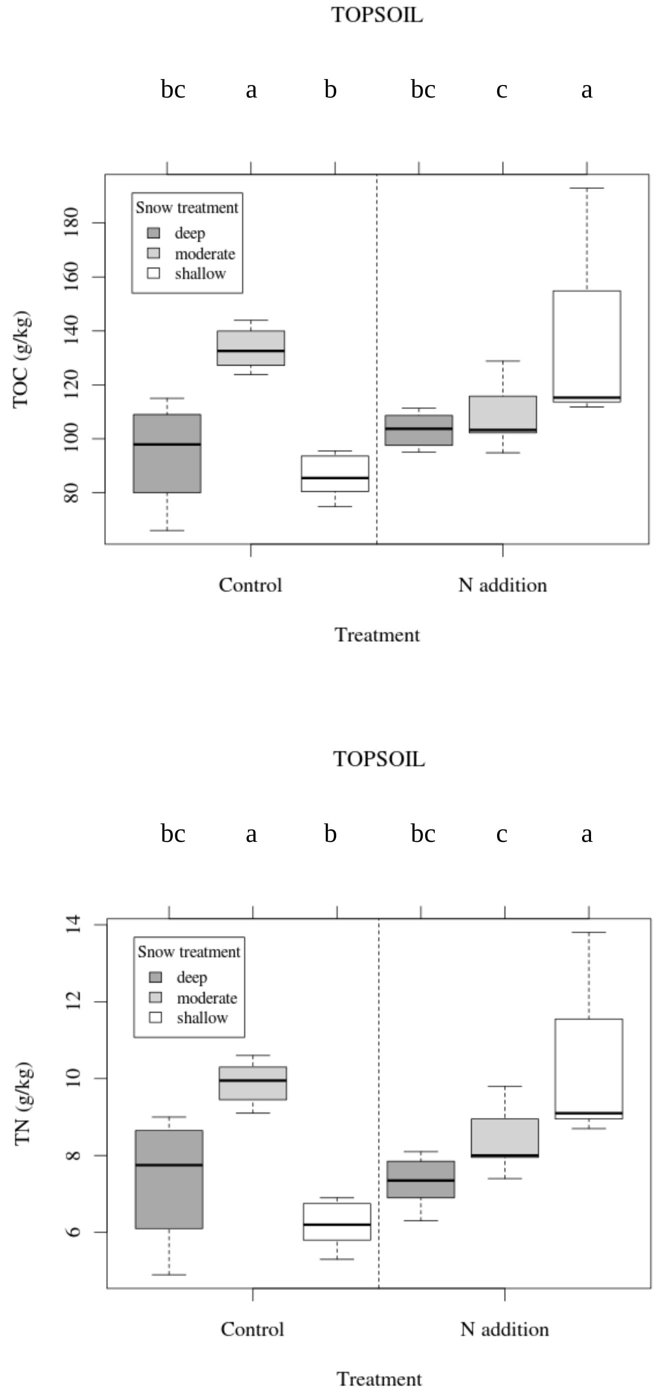

Figure 5 TOC and TN concentration (g/kg) in the subsoil under the different snow cover treatments. Different letters indicate significant differences between means ($p<0.05$).

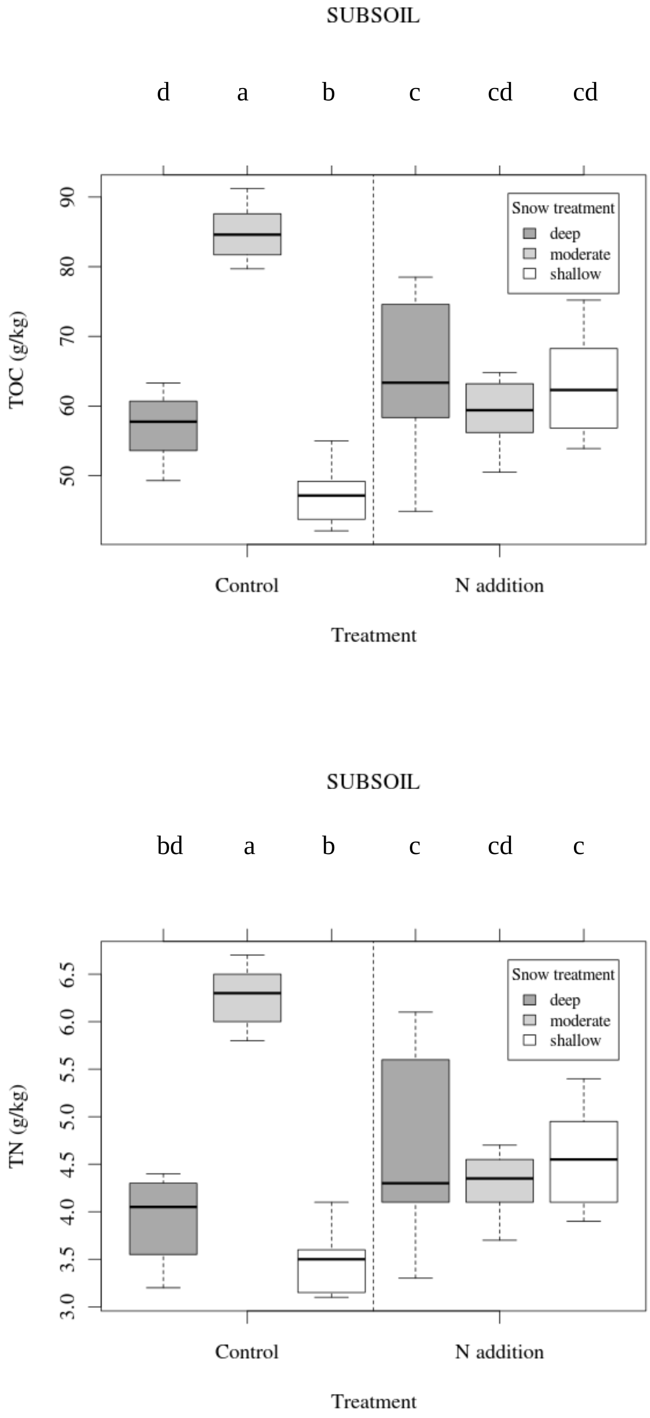

Figure 6 Total organic carbon **(**TOC) content (gkg^{-1}) in the topsoil of the control and N addition plots under deep, moderate and shallow snow cover in 2009 and 1993 (pre-snowfence).

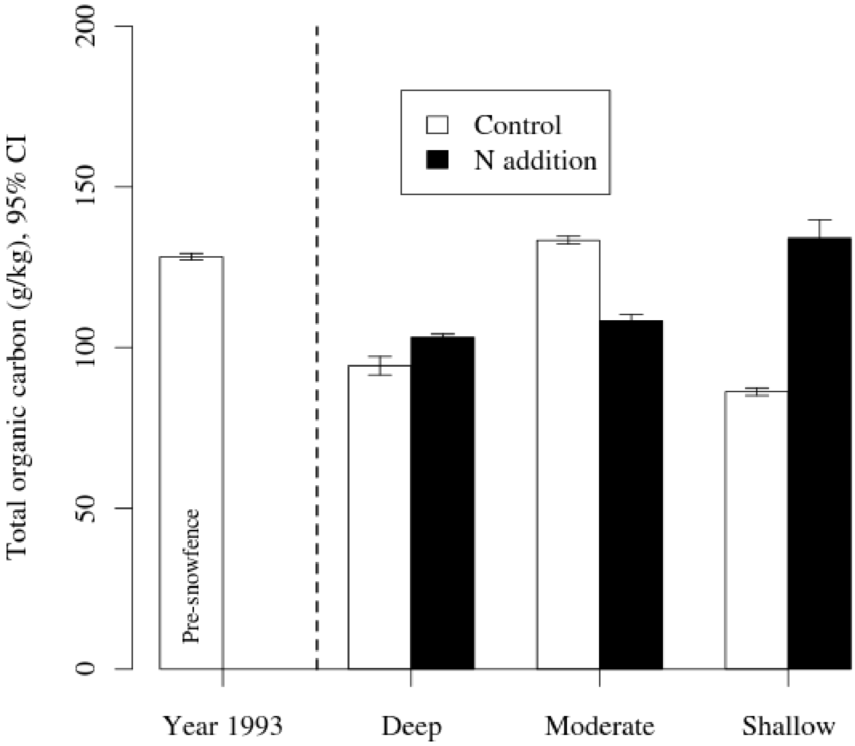

Figure 7 Total nitrogen **(**TN) content (g/kg) in the topsoil of the control and N addition plots under deep, moderate and shallow snow cover in 2009 and 1993 (pre-snowfence).

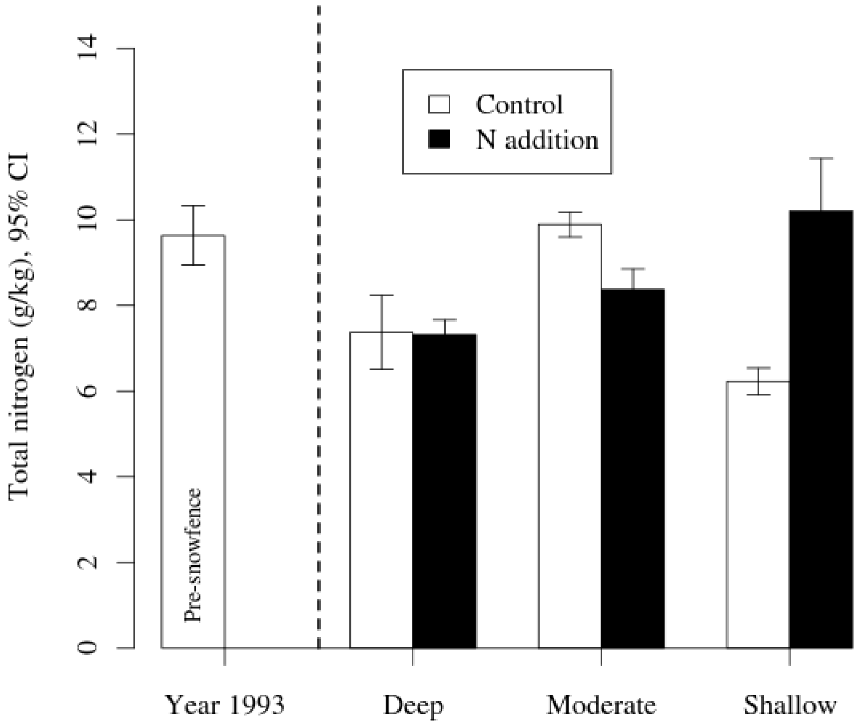

Figure 8 pH of the topsoil and subsoil under deep, moderate and shallow snow cover, in the control, and in the N addition plots. Different letters indicate significant differences between means ($p<0.05$).

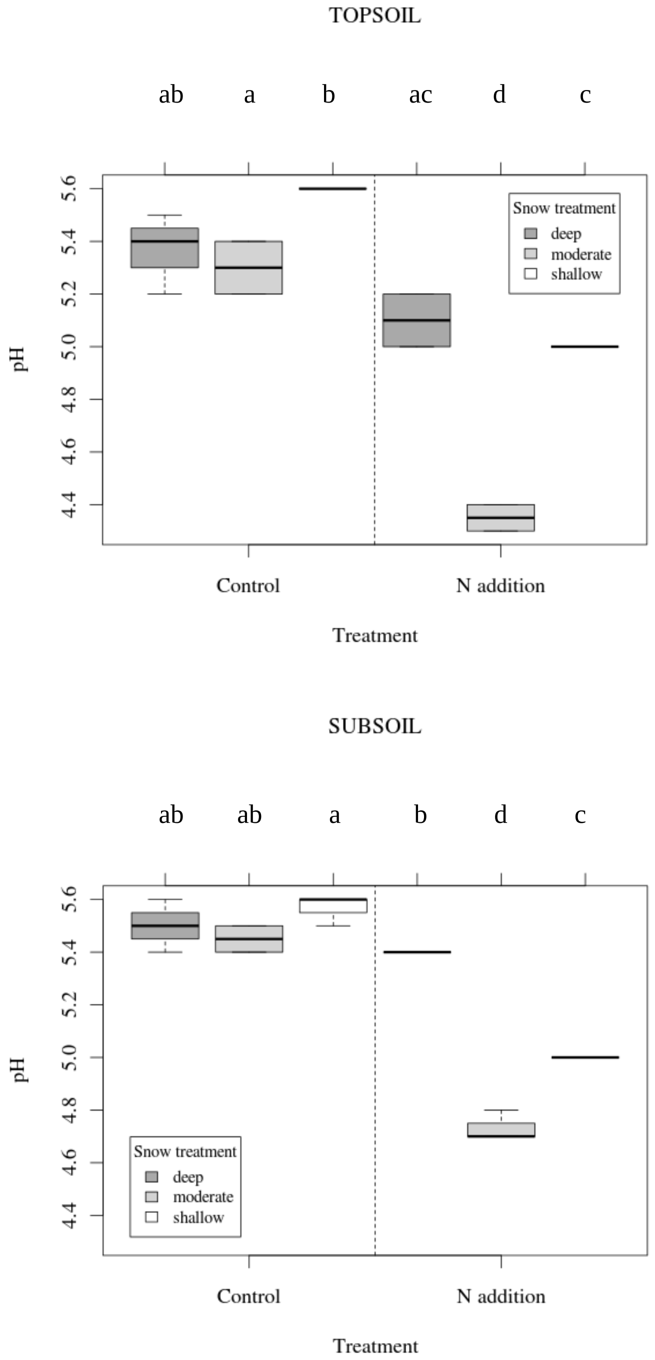

Figure 9 Exchangeable Al of the topsoil and subsoil under deep, moderate and shallow snow cover, in the control and in the N addition plots. Different letters indicate significant differences between means ($p<0.05$).

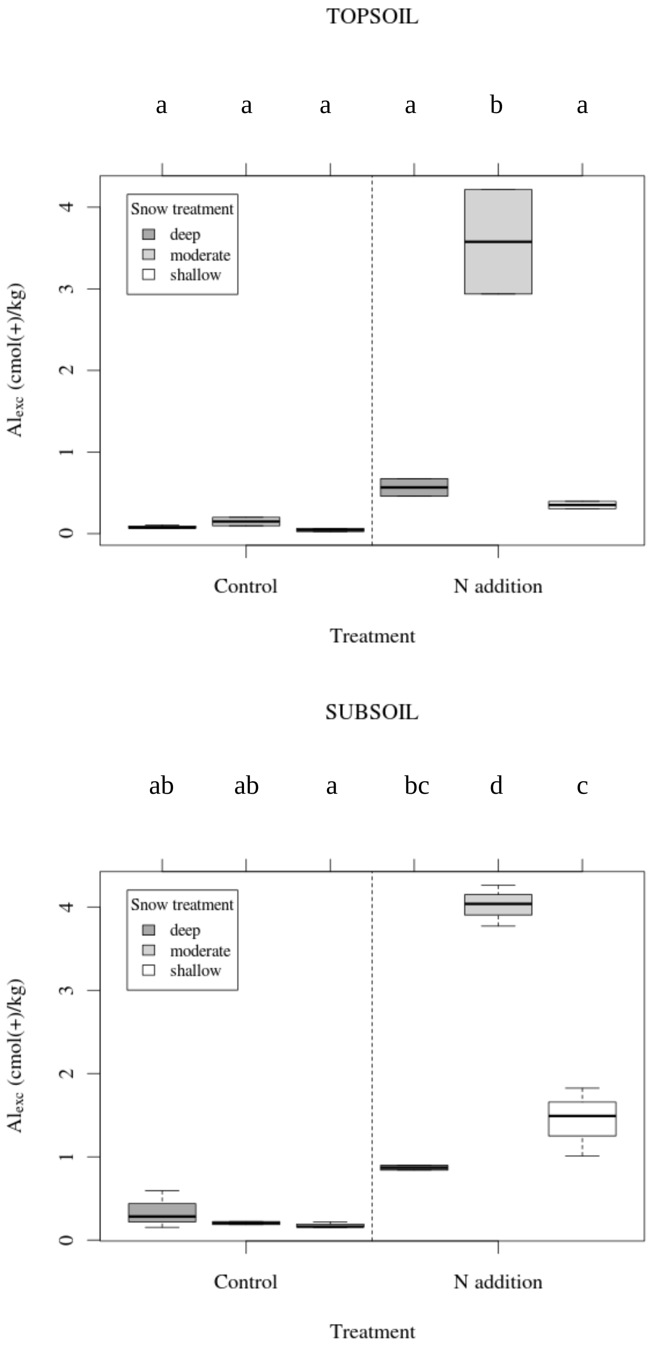

Figure 10 Exchangeable Ca^{2+} of the topsoil and subsoil under deep, moderate and shallow snow cover, in the control and in the N addition plots. Different letters indicate significant differences between means ($p<0.05$).

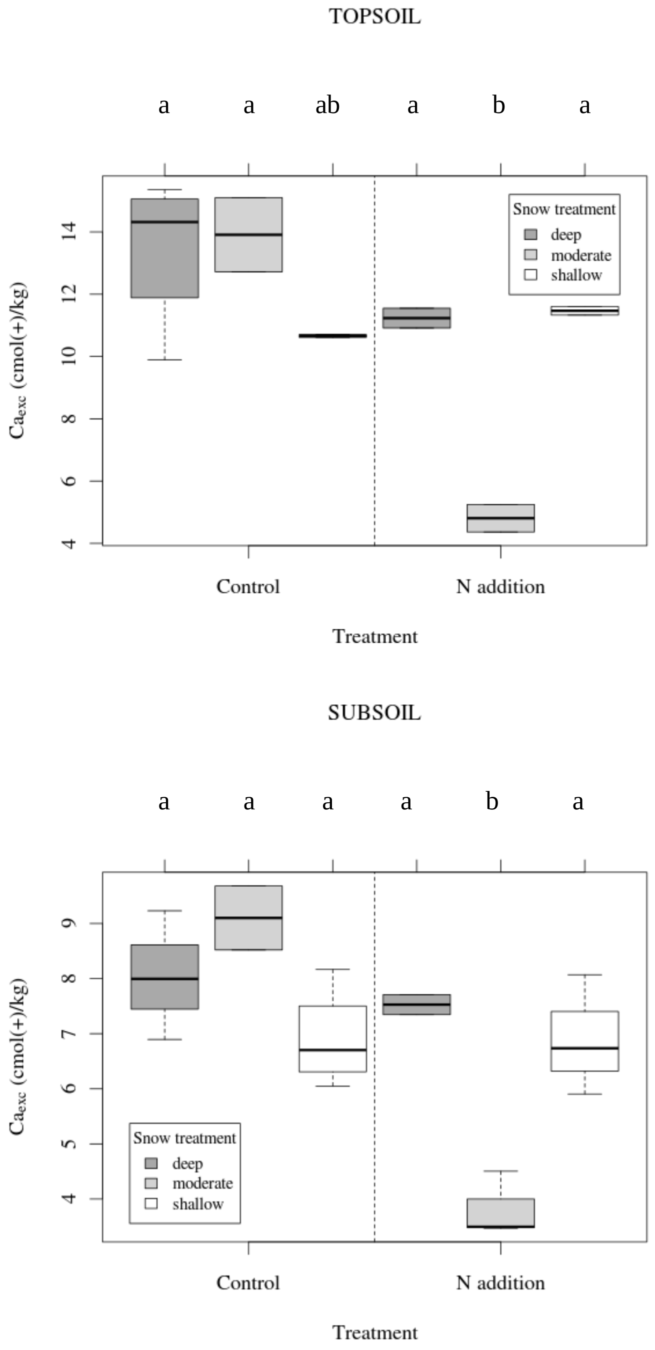

Figure 11 Exchangeable Mg^{2+} of the topsoil and subsoil under deep, moderate and shallow snow cover, in the control and in the N addition plots. Different letters indicate significant differences between means ($p<0.05$).