

# FINAL REPORT

## **Community-specific biogeochemical responses to atmospheric nitrogen deposition in subalpine ecosystem of Mount Rainier National Park**

TASK AGREEMENT P14AC01577 Mod 03

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06-01-16



Subalpine ecosystem of Paradise, Mount Rainier National Park

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## **Acknowledgements**

National Park Service staff provided advice on experimental design, site selection, and logistical assistance that were critical to the implementation of this project, including Gina Rochefort, Tonnie Cummings, Tamara Blett, Barbara Samora, Louis Whiteaker, and Steve Fradkin. Ben Harlow and Debbie Bikfasy at the Washington State University (WSU) Stable Isotope Core Laboratory (SICL) and Jenny Carlson at the Northwest Sustainable Agroecosystem Research Unit helped to complete biogeochemical and stable isotope laboratory analyses. Jennifer Adam and Julian Reyes at WSU and Naomi Tague at University of California, Santa Barbara assisted with ecological modeling. Field work was completed with the assistance of Lauren Kelly.

## Acronyms and Isotopic Terminology

|                       |  |
|-----------------------|--|
| ANOVA                 | Analysis of Variance   |
| $\delta^{13}\text{C}$ | Stable isotope composition of carbon                           |
| $\delta^{15}\text{N}$ | Stable isotope composition of nitrogen                         |
| $\delta^{17}\text{O}$ | Stable isotope composition of oxygen-17                        |
| $\Delta^{17}\text{O}$ | Difference between expected and observed $\delta^{17}\text{O}$ |
| $\delta^{18}\text{O}$ | Stable isotope composition of oxygen-18                        |
| Early                 | Early snow release vegetation community                        |
| IAEA                  | International Atomic Energy Agency                             |
| Mid                   | Mid snow release vegetation community                          |
| MORA                  | Mt. Rainier National Park                                      |
| Late                  | Late snow release vegetation community                         |
| NADP                  | National Atmospheric Deposition Program                        |
| $\text{NO}_3^-$       | Nitrate  |
| $\text{NH}_4^+$       | Ammonium   |
| NPS                   | National Park Service  |
| NRCS                  | Natural Resources Conservation Service                         |
| O                     | Oxygen   |
| RCP                   | Representative Concentration Pathway                           |
| RHESSys               | Regional Hydro-Ecologic Simulation System                      |
| SICL                  | Stable Isotope Core Laboratory                                 |

## Executive Summary

Atmospheric nitrogen (N) deposition occurs when N emissions are released into the atmosphere by automobile or industrial emissions or by the volatilization of fertilizer applied to farmland. These emissions deposit out of the atmosphere through precipitation, known as wet deposition, or by attaching to airborne particles of dust and pollen in dry deposition. Chronic N deposition has been shown to decrease vegetation biodiversity, reduce abundance of N sensitive species, alter biogeochemical cycling, and increase acidification and nutrient enrichment of montane watersheds (Aber et al., 1989; Williams and Tonnessen, 2000; Bowman et al. 2014). The deposition rate at which these detrimental changes occur is known as the critical load (Bobbink et al., 2002). Increases in N deposition could exceed these critical loads for Mount Rainier and high-elevation ecosystems throughout the Cascades. We examined the input of N deposition to Mount Rainier National Park (MORA) and evaluated critical loads for subalpine ecosystem N loss under elevated deposition rates and climate change.

Snowpack storage and processing of N deposition has important implications for informing N emission regulations as well as ecosystem response to N deposition. We measured rates and N forms of N deposition in subalpine snowpack at Mount Rainier National Park (MORA). Measured ambient deposition rates at MORA (3.0 to 3.7 kg N ha<sup>-1</sup> yr<sup>-1</sup>) could exceed rates that are detrimental to lichen and diatom communities and cause montane watershed N enrichment and acidification (Pardo et al., 2015). Ammonium (NH<sub>4</sub><sup>+</sup>) deposition, predominantly produced from agricultural emissions, was the dominant form measured in snowpack. Deposition of NH<sub>4</sub><sup>+</sup> has higher retention than nitrate (NO<sub>3</sub><sup>-</sup>) in subalpine ecosystems, which may reduce N leaching (Williams and Tonnessen, 2000). However, microbial nitrification in snowpack may convert snow NH<sub>4</sub><sup>+</sup> to NO<sub>3</sub><sup>-</sup>, increasing N loss to watersheds.

Using manipulated N addition and isotopic tracers, we examined the fate of N deposition in the ecosystem and critical loads of subalpine ecosystem N loss at MORA. Soil was the major fate of the applied deposition while leaching into watersheds was the dominant loss of N from the ecosystem. Leaching losses exceeded plant uptake, indicating that deposition may be bypassing soils and plant uptake to directly enter montane watersheds. Ecosystem N losses due to soil nitrous oxide (N<sub>2</sub>O) emissions and N leaching were sensitive to increased deposition rates, with significantly higher losses at 3 kg N ha<sup>-1</sup> yr<sup>-1</sup> above ambient rates. Thus, N deposition critical loads for soil N<sub>2</sub>O emissions and leaching may occur between 3 to 6 kg N ha<sup>-1</sup> yr<sup>-1</sup>. While soils and vegetation were not significantly impacted by these short-term N deposition treatments, chronic N deposition may saturate these systems (Aber et al., 1989).

We examined the combined impacts of N deposition and climate change on ecosystem response to deposition at MORA using ecological modeling. Elevated temperatures under climate change severely reduced snowpack, lengthening the vegetation growing season significantly (Mote et al., 2010). This snowpack loss shifted the subalpine N cycle, with increased N losses to leaching and soil N<sub>2</sub>O emissions under current snow-covered months and increased N retention during the growing season. Elevated N deposition magnified these fluxes, but did not alter N cycling behavior or timing. Thus, climate change may exacerbate ecosystem N losses under increased N deposition, resulting in elevated N leaching and soil N<sub>2</sub>O emissions throughout the Cascades.

# Rates and Forms of Snowpack Nitrogen Deposition at MORA

## Context

Global anthropogenic nitrogen (N) fixation now exceeds natural fixation rates, with N emissions being released into the atmosphere from urban and agricultural areas and depositing at elevated rates (Howarth et al., 2002; Matson et al., 2002). In the Northwest, emissions from vehicular and industrial combustion may contribute the majority of inorganic N deposition as nitrate ( $\text{NO}_3^-$ ) while ammonium ( $\text{NH}_4^+$ ) deposition, which largely arises from agricultural fertilizer application, may have less of an impact (Laird et al., 1983; Kendall et al., 2007; NPS, 2014). Elevated rates of N deposition can increase greenhouse gases emissions, acidification of watersheds, and eventual deterioration of sensitive vegetation communities (Aber et al., 1989; Fenn et al., 2003). Subalpine and alpine ecosystems are thought to be particularly sensitive to increases in N deposition (Fenn et al., 2003; Williams and Tonnesen, 2000).

Snowpack is often considered a reservoir of atmospheric N deposition, but it is not a static pool. In subalpine ecosystems of the Rockies and Sierra Nevada, snowpack contains 14% to 101% of estimated total atmospheric N inputs and is often the dominant source of N input for snow-covered catchments (Williams and Melack, 1991; Bowman, 1992; Brooks and Williams, 1999). Within the snowpack, melting and refreezing processes have been shown to influence the concentrations and forms of inorganic N with snowpack depth (Bowman, 1992; Brooks and Williams, 1999). Microbial activity in snowpack has been shown to influence N dynamics through  $\text{N}_2$ -fixation, nitrification, and denitrification in sites with permanent snowpack with low accumulation (Amoroso et al., 2009; Larose et al., 2013).

Sources and processing of N deposition can be determined through analysis of the  $\delta^{15}\text{N}$ ,  $\Delta^{17}\text{O}$ , and  $\delta^{18}\text{O}$  values of  $\text{NO}_3^-$  in snowpack. The  $\delta^{15}\text{N}$  of  $\text{NO}_3^-$  in precipitation and snowpack can differentiate between biogenic and combustion N sources (Pichlmayer et al., 1998; Elliott et al., 2007). Snow  $\text{NO}_3^-$   $\Delta^{17}\text{O}$  and  $\delta^{18}\text{O}$  can indicate whether the  $\text{NO}_3^-$  was produced by atmospheric or biological sources (Michalski et al., 2003; Elliott et al., 2007). Snow  $\text{NO}_3^-$   $\delta^{15}\text{N}$ ,  $\Delta^{17}\text{O}$ , and  $\delta^{18}\text{O}$  can also provide insight into microbial activity in snowpack (Kendall et al., 1998; Amoroso et al., 2009). Microbial nitrification in snowpack has been shown to increase snow  $\text{NO}_3^-$   $\delta^{15}\text{N}$  and significantly reduce  $\Delta^{17}\text{O}$ , removing the atmospheric signal (Amoroso et al., 2009).

## Objective

We examined forms, rates, and stable isotope composition of snowpack N deposition in subalpine ecosystems at MORA to characterize deposition patterns and inform ecosystem response to N deposition. We hypothesized that  $\text{NO}_3^-$  would be the dominant form of N deposition, as consistent with N emission projections and measurements from nearby NADP sites (NADP, 2015). Additionally, we hypothesized that  $\text{NO}_3^-$   $\delta^{15}\text{N}$  and  $\Delta^{17}\text{O}$  could be used to differentiate different atmospheric N deposition sources.

## Approach

The study was located in the subalpine ecosystems of Paradise in MORA at an elevation of 1930 m (N 46°47', W 121°43'). Snow sampling was conducted in six plots located randomly over the field site in the winters of 2012-2013 and 2013-2014. Each plot was sampled using a soil

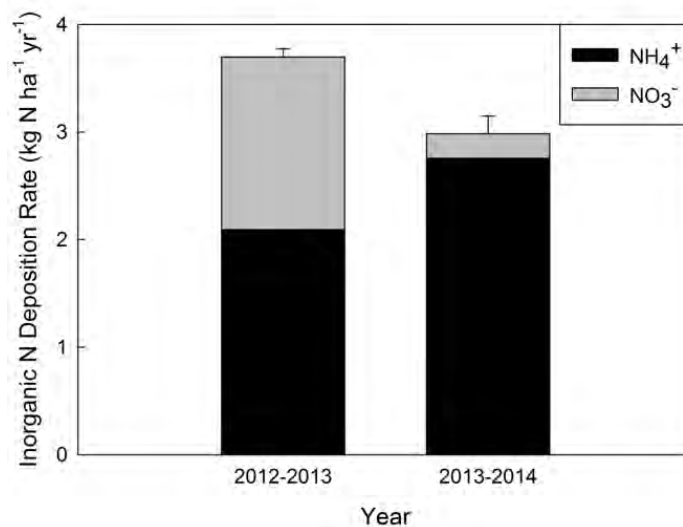
auger at every 25 cm depth increment to 3 m deep into the snowpack. Snow samples were tested for density, total N content,  $\text{NO}_3^-$  and  $\text{NH}_4^+$  content, the  $\text{NO}_3^-$   $\delta^{15}\text{N}$ ,  $\Delta^{17}\text{O}$ , and  $\delta^{18}\text{O}$ , and the total N  $\delta^{15}\text{N}$ .

Subsamples of  $\text{NO}_3^-$  in the melted snow were converted into  $\text{N}_2\text{O}$  gas using the denitrifier method, which conserves the  $\delta^{15}\text{N}$ ,  $\delta^{17}\text{O}$ , and  $\delta^{18}\text{O}$  values of the  $\text{NO}_3^-$  through conversion to  $\text{N}_2\text{O}$  by *Pseudomonas aureofaciens* (Casciotti et al., 2002). The produced  $\text{N}_2\text{O}$  was then analyzed for  $\delta^{15}\text{N}$ ,  $\delta^{17}\text{O}$ , and  $\delta^{18}\text{O}$  using a Finnigan™ Gas Bench II (Thermo Electron Corp., Waltham, MA, USA) coupled to a Delta V™ Isotope Ratio Mass Spectrometer (Thermo Electron Corp., Waltham, MA, USA) (Kaiser et al., 2007). Total N analysis was conducted by converting all N forms to  $\text{NO}_3^-$  using a persulfate oxidizing reagent in an autoclave at 121 °C for 45 minutes. These resulting solutions were allowed to cool and then neutralized to 6 to 7 pH (Knapp et al., 2005). The neutralized samples were then converted into  $\text{N}_2\text{O}$  gas using the denitrifier method to analyze the total N  $\delta^{15}\text{N}$  as for the snow  $\text{NO}_3^-$  above (Casciotti et al., 2002).

Results of these measurements were tested for statistical differences between snowpack depths and sampling years using an analysis of variance (ANOVA) design. Interactions between sampling year and snowpack depth were considered. A paired t-test was conducted to determine differences between snow  $\text{NH}_4^+$  and  $\text{NO}_3^-$  concentrations during the same years and at the same depths. Additionally, correlations between snow density,  $\text{NH}_4^+$  and  $\text{NO}_3^-$  concentrations,  $\text{NO}_3^-$   $\delta^{15}\text{N}$ ,  $\text{NO}_3^-$   $\Delta^{17}\text{O}$ , total N  $\delta^{15}\text{N}$ , and residual N  $\delta^{15}\text{N}$  were analyzed via Spearman and Pearson tests.

## Findings

- 1. Total annual inorganic N deposition in snowpack was 3.74 kg N ha<sup>-1</sup> yr<sup>-1</sup> in 2012-2013 and 2.99 kg N ha<sup>-1</sup> yr<sup>-1</sup> in 2013-2014 (Figure 1). These rates exceed critical loads for lichen and diatom community changes as well as alpine lake enrichment and acidification in the Rocky Mountains.**



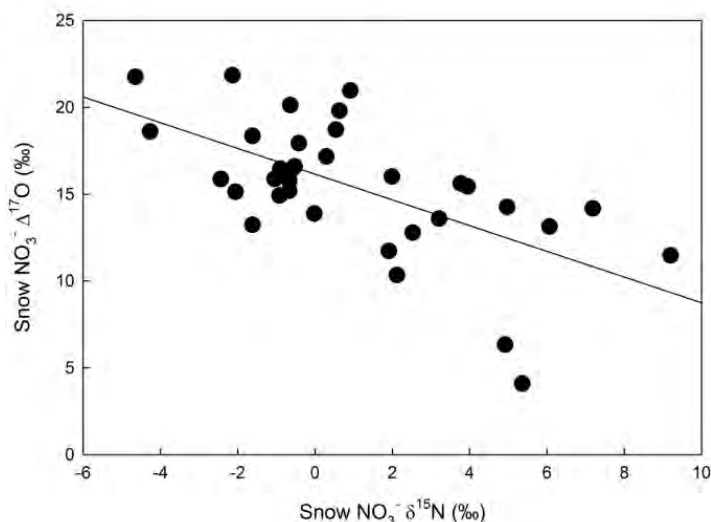
**Figure 1.** Snowpack inorganic N rates were 3.74 and 3.0 kg N ha<sup>-1</sup> yr<sup>-1</sup> for the 2012-2013 and 2013-2014, respectively.

2. Snowpack  $\text{NH}_4^+$  content was significantly higher than  $\text{NO}_3^-$ , which may indicate that agricultural emissions of ammonia ( $\text{NH}_3$ ) have a larger influence on deposition at MORA than nitrogen oxide emissions ( $\text{NO}_x$ ) from combustion (Table 1).

**Table 1.** Mean snow N deposition ammonium ( $\text{NH}_4^+$ ) and nitrate ( $\text{NO}_3^-$ ) concentrations and  $\text{NO}_3^-$  and total N stable isotope composition across all depths for the sampling years 2013 and 2014.  $\text{N}_{\text{residual}}$  is the components of total N that are not  $\text{NO}_3^-$ . Superscripts *a* and *b* indicate significant difference between years. Numbers in parentheses indicate standard error of the mean.

| Year | $\text{NH}_4^+$<br>( $\text{kg N ha}^{-1}$ ) | $\text{NO}_3^-$<br>( $\text{kg N ha}^{-1}$ ) | $\text{NO}_3^- \delta^{15}\text{N}$<br>(‰) | $\text{NO}_3^- \Delta^{17}\text{O}$<br>(‰) | $\text{NO}_3^- \delta^{18}\text{O}$<br>(‰) | Total N $\delta^{15}\text{N}$<br>(‰) |
|------|--|--|--|--|--|--------------------------------------|
| 2013 | 2.1 (0.01) <sup>a</sup>                      | 1.6 (0.04) <sup>a</sup>                      | 1.4 (0.84)                                 | 14.9 (0.97)                                | 24.9 (2.4)                                 | 3.2 (0.72)                           |
| 2014 | 2.8 (0.09) <sup>b</sup>                      | 0.2 (0.01) <sup>b</sup>                      | 1.1 (0.74)                                 | 15.6 (0.95)                                | 28.1 (2.9)                                 | 3.3 (0.56)                           |

3. Snow  $\text{NO}_3^- \delta^{15}\text{N}$  and  $\Delta^{17}\text{O}$  were significantly negatively correlated, indicating that microbial nitrification may be converting deposited  $\text{NH}_4^+$  to  $\text{NO}_3^-$  in the snowpack (Figure 2). This conversion to  $\text{NO}_3^-$  could promote inorganic N leaching to watersheds as  $\text{NO}_3^-$  is water-soluble and more mobile in ecosystems than  $\text{NH}_4^+$ .



**Figure 2.** Snowpack  $\text{NO}_3^- \delta^{15}\text{N}$  and  $\Delta^{17}\text{O}$  were significantly negatively correlated ( $p < 0.001$ ;  $R = 0.72$ ).

## Synthesis

Our findings indicate that rates of inorganic N deposition match elevation-adjusted network measurements at low elevation (NADP, 2015). Rates of snowpack inorganic N deposition at MORA ( $3.7 \text{ kg N ha}^{-1} \text{ yr}^{-1}$  in 2012-2013 and  $3.0$  in 2013-2014) were similar to those measured at nearby high-elevation sites (Agren et al., 2012) but exceeded those measured at a nearby National Atmospheric Deposition site ( $0.76$  to  $2.5 \text{ kg N ha}^{-1} \text{ yr}^{-1}$ ) at lower elevation (424 m) (NADP, 2015). With the PRISM adjustment, NADP interpolations of total inorganic N deposition across the Cascades, which range from  $2$  to  $10 \text{ kg ha}^{-1} \text{ yr}^{-1}$  were similar to measured snowpack inorganic N deposition rates at MORA (NADP, 2015). Ecosystem management based on network measurements should incorporate elevational effects to avoid underestimation of



deposition rates in these high-elevation ecosystems (Weathers et al., 2000; Latysh and Wetherbee, 2012).

Critical loads, a concept used to quantify the threshold at which ecological impacts of N deposition occur, are often determined using network deposition measurements due to lack of deposition data at high elevations (Bobbink et al., 2002; NPS, 2014). Thresholds set using low elevation network data may underestimate deposition rates in these high-elevation ecosystems, establishing critical loads that are too sensitive (NPS, 2014). The N deposition rates measured in this study would exceed critical loads for sensitive thresholds, including lichen chemistry and biodiversity, lake diatom community change, and N enrichment and acidification of montane watersheds (Pardo et al., 2011). Management for subalpine critical loads should measure N deposition rates at high elevations to properly constrain the thresholds at which biogeochemical and community responses are significantly affected.

Snowpack deposition forms and microbial communities may regulate ecosystem response to N deposition. Snowpack N deposition had a greater proportion of  $\text{NH}_4^+$  than  $\text{NO}_3^-$ , which may indicate that agricultural N emissions have a larger role in deposition than previously expected (Kendall et al., 2007). Ammonium is less mobile than  $\text{NO}_3^-$  in soil as it is not water soluble and often binds to negatively charged soil particles (Chapin et al., 2002). However, the significant negative correlation between  $\delta^{15}\text{N}$  and  $\Delta^{17}\text{O}$  was indicative of the fractionation that occurs in  $\delta^{15}\text{N}$  with microbial nitrification (Robinson, 2001; Frey et al., 2009). Thus, microbial nitrification may be converting the greater concentrations of snow  $\text{NH}_4^+$  into  $\text{NO}_3^-$ , which can increase its mobility in the ecosystem (Williams and Tonnessen, 2000). Alpine vegetation may have a higher affinity for  $\text{NH}_4^+$  than  $\text{NO}_3^-$ , which can increase plant uptake (Miller and Bowman, 2003). Additionally,  $\text{NO}_3^-$  has lower retention than  $\text{NH}_4^+$  in subalpine and alpine ecosystems due to its water solubility (Williams and Tonnesson, 2000; Miller and Bowman, 2003). Thus, microbial nitrification in snowpack could increase rates of N loss in subalpine ecosystems of MORA even if N deposition rates remain stable.

# Critical Loads for Subalpine Biogeochemical Cycling

## Context

The critical load concept has been widely utilized to quantify the impacts of N deposition across a variety of ecosystems (Pardo et al., 2015). A critical load is the threshold of a pollutant at which a significant harmful effect begins to occur (Bobbink et al., 2002; Porter et al., 2005). Critical loads within high-elevation ecosystems have been used to quantify impacts of N deposition on lake diatom communities (Baron et al., 2000), vegetation composition (Bowman et al., 2006), soil biogeochemistry (Bowman et al., 2006), and N leaching into watersheds (Williams and Tonnessen, 2000; Sickman et al., 2001).

Application of synthetically-enriched  $^{15}\text{N}$  has been commonly used as a method of tracing biogeochemical fluxes across many ecosystems, including studies of N deposition (Dawson et al., 2002). An enriched  $^{15}\text{N}$  label allows an isotopic signal to be traced as it moves between ecosystem pools and fluxes. This tracer enables research that determines the impact and fate of the applied N (Dawson et al., 2002; Templer et al., 2012).

## Objective

We examined ecosystem N storage and loss under manipulated rates of N deposition to determine biogeochemical critical loads in three subalpine vegetation communities of the Cascades. We hypothesized that ecosystem N losses through soil  $\text{N}_2\text{O}$  emissions and inorganic N leaching to watersheds would be more sensitive to increased N deposition than N storage in soils or vegetation. Additionally, we utilized an isotopic tracer recovery study to examine N deposition partitioning in subalpine ecosystems.

## Approach

The study site was located the subalpine ecosystems of the Paradise area in MORA at an elevation of 1890 to 1960 m (N 46°47', W 121°43'). This study focused on three subalpine meadow communities that are distinguished by their date of snow release (for a full description of these communities, see Henderson, 1973). The Early snow release community, where snow release typically occurs in mid-July, the Mid community, where snow release occurs in late July, and the Late community where snow release occurs in early to mid August.

Five patches, located at least 20 m apart in areas of similar topography and elevation, were selected for each vegetation community, with four plots (4 m<sup>2</sup>) in each patch for a total of 60 plots. Each plot was separated by a 2 m buffer zone. The four plots within each patch were randomly selected for one of the following N application treatments: control, +3, +5, and +10 kg N ha<sup>-1</sup> yr<sup>-1</sup>. These treatments were applied as an enriched  $^{15}\text{N}$  (10 atom %)  $^{15}\text{NH}_4^+$ - $^{15}\text{NO}_3^-$  solution, which was added evenly across the melting snowpack in each appropriate plot (Bilbrough et al., 2000; Inselsbacher et al., 2012). The proportion of enriched ammonium ( $^{15}\text{NH}_4^+$ ) and enriched nitrate ( $^{15}\text{NO}_3^-$ ) added matched ambient N deposition proportions in snowpack (60%  $\text{NH}_4^+$  and 40%  $\text{NO}_3^-$ ) (Poinsatte and Evans, *in prep.*).

Measurements of soil temperature and moisture, soil N, microbial biomass N, plant N uptake,  $\text{N}_2\text{O}$  emissions, and N leaching were conducted throughout the 2014 growing season. Collection occurred at 0, 10, 30, 50, and 70 days after snow release in each community type, respectively.

All of the biomass, gaseous, and resin extract samples were analyzed for their  $^{15}\text{N}$  enrichment and total N content. The percentage of each flux from the applied N deposition was calculated using the relative  $^{15}\text{N}$  of the flux to the 10 atom% of the tracer. Tracer recovery was measured as the proportion of  $^{15}\text{N}$  in each flux or pool divided by the total amount of  $^{15}\text{N}$  added (Inselsbacher et al., 2012; Templer et al., 2012).

Soil cores were collected from 0 to 0.2 m depth. Samples were sieved, dried, and pulverized to a fine powder to prepare for analysis. The prepared soil samples were analyzed for C and N concentration and stable isotopic composition using an elemental analyzer coupled to a continuous flow isotope ratio mass spectrometer at the WSU SICL. Isotope composition was determined by comparing sample  $^{15}\text{N}$  to known international standards (IAEA, 2009).

Microbial biomass N concentrations ( $\text{g N g}^{-1}$  soil) and stable isotope composition were determined using the chloroform fumigation method (Jin and Evans, 2007; Dijkstra et al., 2013). Two sets of 5 cm depth soil were collected, with one set subjected to chloroform fumigation. Samples were extracted and analyzed for N concentration and stable isotopic composition. Microbial biomass N content and  $^{15}\text{N}$  atom percent was calculated as the difference between the fumigated and unfumigated sets (Jin and Evans, 2007; Dijkstra et al., 2013).

Changes in plant C and N content ( $\text{g m}^{-2}$ ) and stable isotope composition were measured to determine plant growth and N uptake at each collection date (Billings et al., 2002). The percent canopy coverage of each species in each plot was determined by visual estimation. Aboveground biomass was collected for  $0.01 \text{ m}^2$  of the most dominant species in each plot. Samples were prepared and analyzed for C and N concentration and stable isotope composition as for soils.

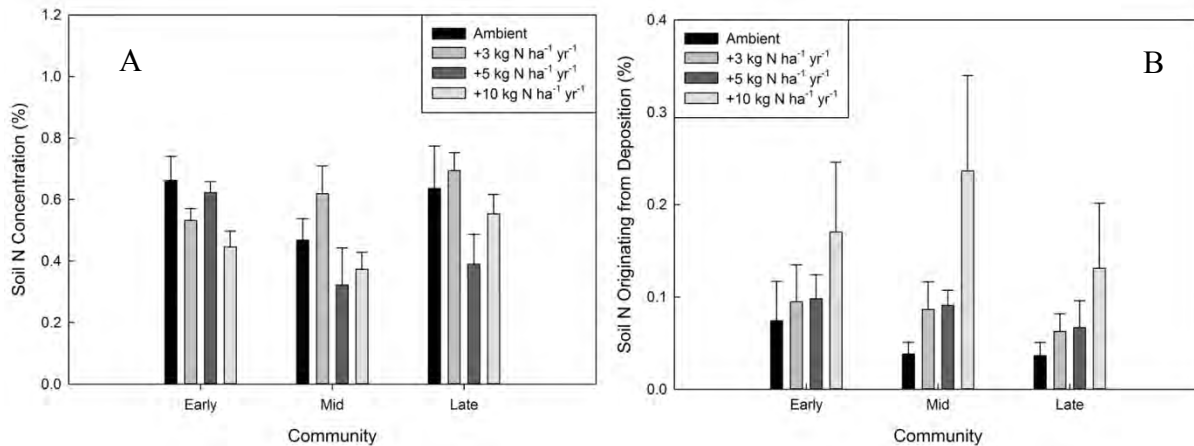
Soil nitrous oxide ( $\text{N}_2\text{O}$ ) emissions were measured at each collection date using a static chamber approach (Billings et al., 2002). Gas samples were collected from the chambers with sampling intervals at 0, 15, and 30 minutes to determine  $\text{N}_2\text{O}$  flux rates. Additional gas samples were sent to the University of California Davis Stable Isotope Facility to analyze  $\text{N}_2\text{O}$   $^{15}\text{N}$ .

Soil inorganic leaching rates ( $\text{g N m}^{-2} \text{ yr}^{-1}$ ) were determined using the soil resin core method (Billings et al., 2002). Polyvinylchloride columns were installed into the soil, with resin bags at the top and bottom to capture  $\text{NH}_4^+$  and  $\text{NO}_3^-$  ions throughout the year. Resin extracts were analyzed for  $\text{NH}_4^+$  and  $\text{NO}_3^-$  concentrations. The  $^{15}\text{N}$  atom percent of the total N in the extracts were measured through persulfate digestion and the denitrifier method (Casciotti et al., 2002).

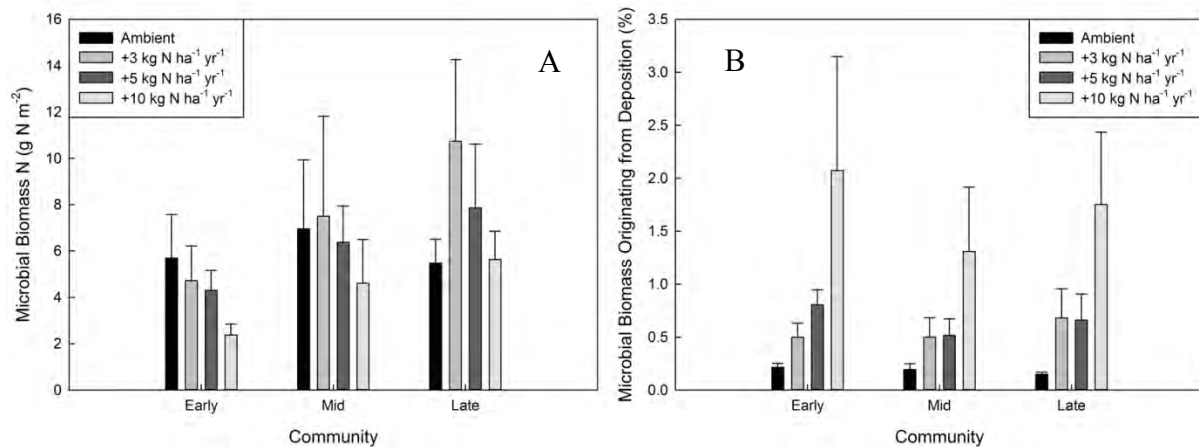
The experiment utilized a mixed effect complete block design, where vegetation patches were treated as blocks for all statistical analyses. Results of these measurements were analyzed for significant differences between community types, treatment, and collection dates using a repeated measures ANOVA design. Interaction effects between the community, treatment, and collection date were considered. Additionally, correlations between soil conditions and C and N fluxes were analyzed.

## Findings

- 1. Soil N storage was not significantly impacted by elevated N deposition treatments due to the relative size of the N deposition inputs to the large total soil N pools in MORA subalpine soils (Figure 3). Microbial biomass was not significantly affected by deposition, indicating that it may not buffer against ecosystem N loss (Figure 4).**

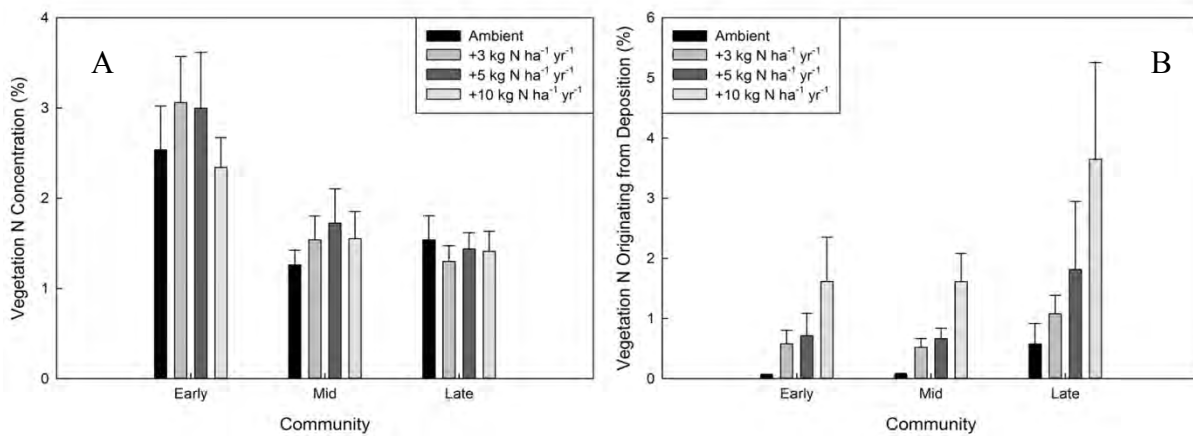


**Figure 3.** Average total soil nitrogen (N) concentrations (A) were not significantly different between the Early, Mid, and Late subalpine vegetation communities or between the ambient, +3, +5, and +10 kg N ha<sup>-1</sup> yr<sup>-1</sup> treatments. However, the average percentage of soil N that originated from the applied N deposition treatment (B), as determined by the soil <sup>15</sup>N, was significantly higher in the +10 kg N ha<sup>-1</sup> yr<sup>-1</sup> than all other treatments in each community. Error bars indicate the standard error of the mean.

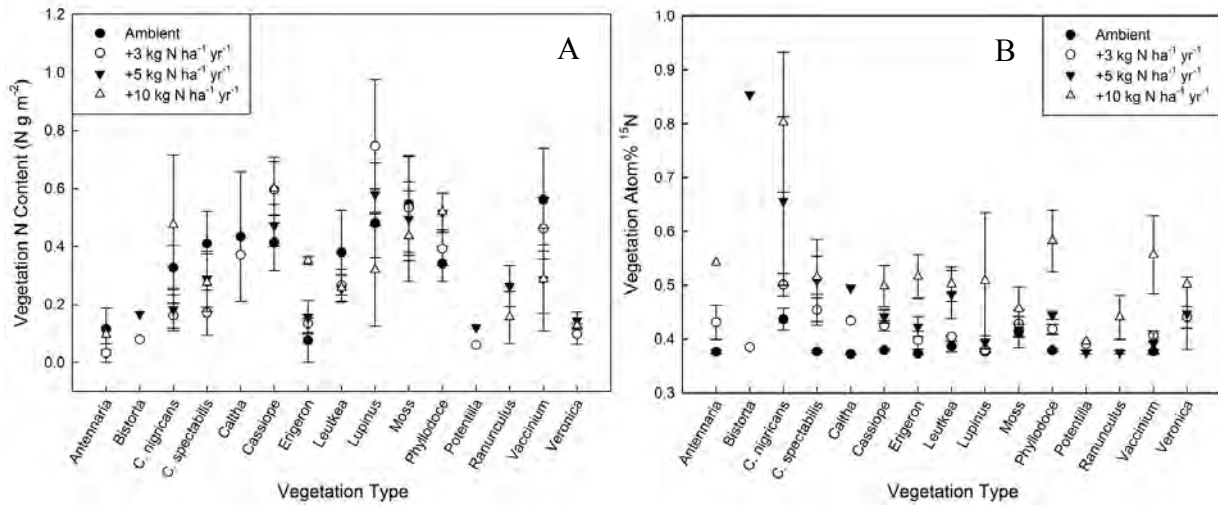


**Figure 4.** Average microbial N biomass (A) was not significantly different between the Early, Mid, and Late subalpine vegetation communities or between the ambient, +3, +5, and +10 kg N ha<sup>-1</sup> yr<sup>-1</sup> treatments. The average percentage of microbial N biomass from the treatment (B) was significantly different higher in the +10 kg N ha<sup>-1</sup> yr<sup>-1</sup> than all other treatments in each community. Error bars indicate the standard error of the mean.

- 2. Deposition replaced rather than supplement vegetation N sources, with no significant increases in plant biomass or N content but plant <sup>15</sup>N indicating that uptake of deposition did occur (Figure 5). All vegetation species studied were able to utilize the applied N deposition, with *Carex nigricans* and *C. spectabilis* having significantly higher uptake than all other species (Figure 6).**

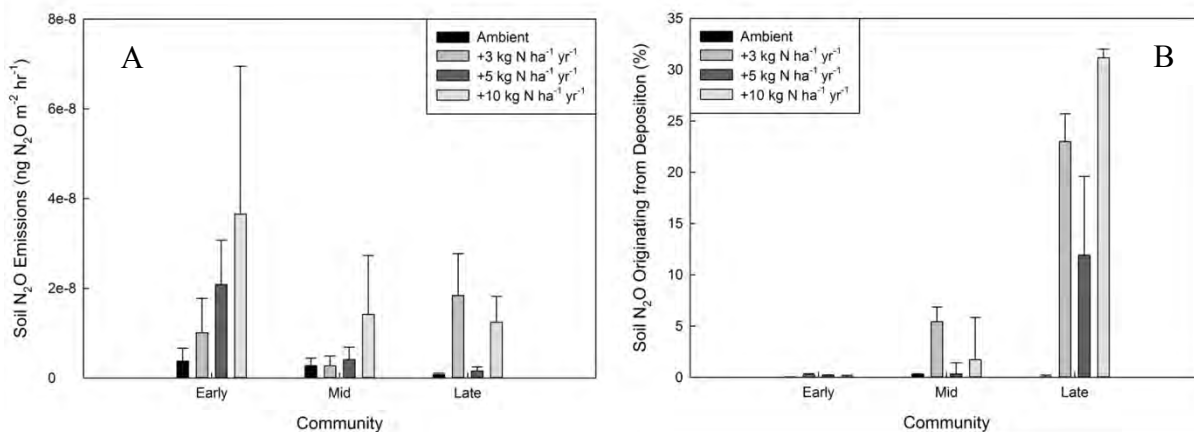


**Figure 5.** Average vegetation nitrogen (N) concentrations (A) were not significantly different between the ambient, +3, +5, and +10 kg N ha<sup>-1</sup> yr<sup>-1</sup> treatments. However, the vegetation was capable of utilizing N deposition inputs as the percentage of vegetation N from deposition increased significantly with the deposition treatment strength (B). Error bars indicate the standard error of the mean.

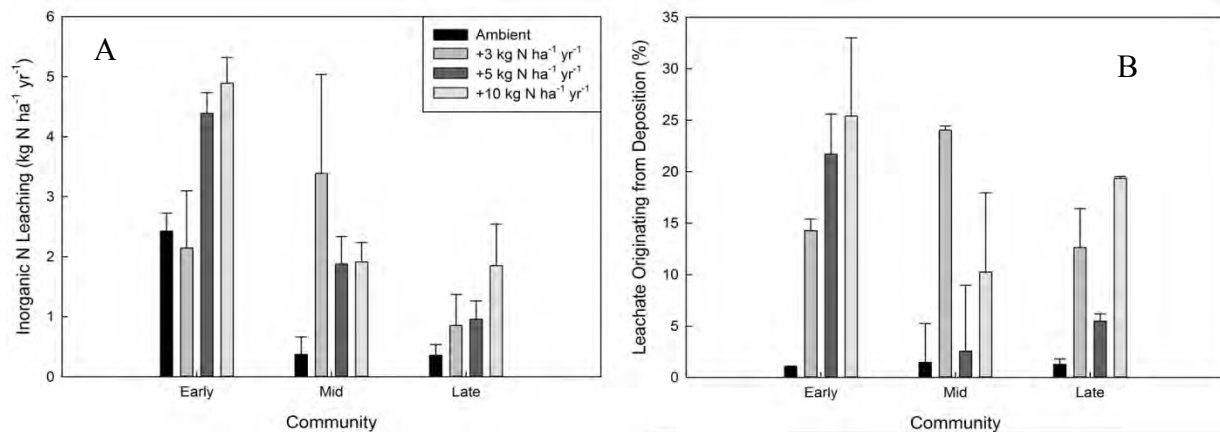


**Figure 6.** Vegetation nitrogen (N) concentrations (A) were not significantly different between the ambient, +3, +5, and +10 kg N ha<sup>-1</sup> yr<sup>-1</sup> treatments for any species. However, the vegetation <sup>15</sup>N did indicate that *Carex nigricans* and *C. spectabilis* had significantly higher uptake of N deposition than did all other studied species. Error bars indicate the standard error of the mean.

**3. Ecosystem N losses through soil N<sub>2</sub>O emissions (Figure 7) and N leaching (Figure 8) were highly sensitive to increases in N deposition, with significant increases at +3 to +10 kg N ha<sup>-1</sup> yr<sup>-1</sup>. Thus, a critical load for subalpine ecosystems at MORA may occur between 3 to 6 kg N ha<sup>-1</sup> yr<sup>-1</sup>, based on the ambient and applied deposition rates.**

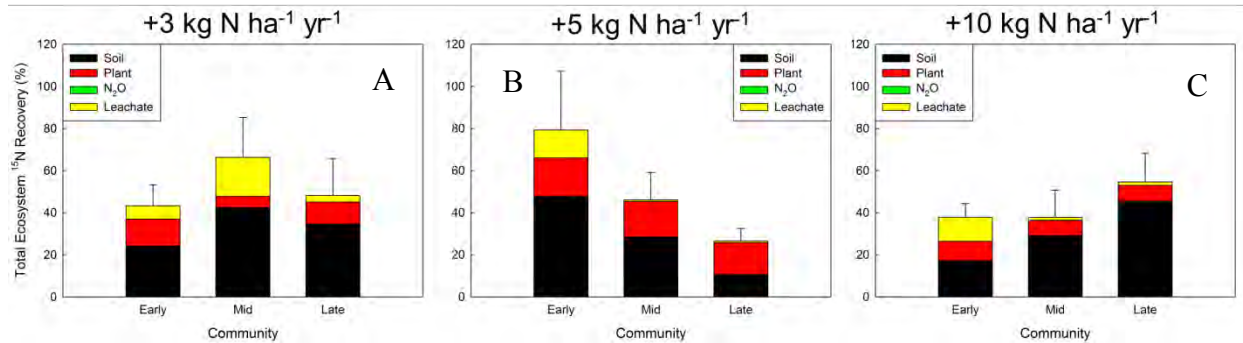


**Figure 7.** Average soil nitrous oxide (N<sub>2</sub>O) emissions (A) were significantly impacted by the N deposition treatments, with increased N<sub>2</sub>O at low N rates. The Early snow release community had the highest N<sub>2</sub>O emissions, but the lowest percentage of the N<sub>2</sub>O directly from the applied N (B). Instead, the Late community had significantly greater proportions of N<sub>2</sub>O produced using the applied N. Error bars indicate the standard error of the mean.



**Figure 8.** Average inorganic N leaching (A) significantly increased with the N deposition treatments in all communities. The Early snow release community had the highest percentage of the N leaching directly from the applied N (B), potentially due to reduced vegetation uptake in this community after snow release. Error bars indicate the standard error of the mean.

- 4. Soil was the dominant fate of N deposition in the ecosystem, with leaching as the most significant fate of ecosystem N loss (Figure 9). These results indicate that significant proportions of N deposition may be bypassing N storage in soils and vegetation, directly entering watersheds after release from snowpack.**



**Figure 9.** Average ecosystem <sup>15</sup>N recovery for the Early, Mid, and Late snow release communities under +3, +5, and +10 kg N ha<sup>-1</sup> yr<sup>-1</sup> treatments indicates the fate of the applied N deposition. Error bars indicate the standard error of the mean.

## Synthesis

We observed significant increases in soil N<sub>2</sub>O emission and inorganic N leaching between the ambient and +3 kg N ha<sup>-1</sup> yr<sup>-1</sup> treatments, indicating that a critical load for subalpine ecosystem N loss at MORA may occur within this range. Biogeochemical critical loads are often defined as the N deposition threshold at which there are significant shifts in N cycling from organic N to inorganic N systems, accompanied by elevated rates of N loss through N emissions and leaching (Fenn et al., 2003; Bowman et al., 2014; Pardo et al., 2015). Critical loads of ecosystem N loss varied between subalpine vegetation communities. Compared to leaching under the Ambient treatment, the Mid and Late communities had significantly greater N leaching at +3 kg N ha<sup>-1</sup> yr<sup>-1</sup> while the Early community had significantly greater leaching at +5 kg N ha<sup>-1</sup> yr<sup>-1</sup>. Soil N<sub>2</sub>O emissions were significantly greater than the ambient control at +3 kg N ha<sup>-1</sup> yr<sup>-1</sup> in the Early and Late communities, but were only significantly greater at +10 kg N ha<sup>-1</sup> yr<sup>-1</sup> in the Mid community. Critical loads for ecosystem N loss may occur between total inorganic N deposition rates of 3 to 6 kg N ha<sup>-1</sup> yr<sup>-1</sup>, as defined by the combined inputs of ambient deposition (3.0 to 3.7 kg N ha<sup>-1</sup> yr<sup>-1</sup>) and +3 kg N ha<sup>-1</sup> yr<sup>-1</sup> treatment.

The critical load for ecosystem N storage was not attained in this study. Ecosystem N storage pools were much less sensitive to elevated N deposition rates, with no consistent response in the soil or vegetation N pools. Thus, inputs from N deposition may bypass storage in the bulk soil, microbial, and vegetation pools to be directly lost from the ecosystem as N<sub>2</sub>O emissions or leaching. However, chronic deposition within these elevated rates may attain N storage critical loads once the ecosystem becomes N saturated (Aber et al., 1989).

There were several indicators for susceptibility to N deposition that may inform management of N deposition critical loads. Generally, soils with lower C:N ratios were more susceptible to soil N<sub>2</sub>O emissions and N leaching under ambient and elevated deposition rates (Dise et al., 1998). Soil moisture was positively correlated with vegetation N uptake and soil N<sub>2</sub>O emissions, which indicates that it may be critical to drive subalpine ecosystem N cycling (Campbell et al., 2000; Williams and Tonnessen, 2000; Filippa et al., 2009). Communities with high soil moisture throughout the growing season, such as the Late snow release community, may be susceptible to N loss under elevated deposition rates. For species-specific critical loads, *Carex nigricans* and *C. spectabilis* had the only significant responses in uptake of N deposition. Although neither the biomass nor N concentration of these species changed with deposition, this may have due to the short-term nature of this study (Bowman et al., 2014). These species in particular should be investigated for susceptibility to chronic elevated N deposition rates.

# Impacts of Climate Change on Ecosystem Response to Nitrogen Deposition

## Context

Snow regime influences ecosystem N cycling in high-elevation ecosystems. Concurrently, the western US is estimated to warm between 2 to 5 °C over the next century, consistent with rising concentrations of greenhouse gases, with the Cascade Range projected to experience winter warming between 0.2 to 0.6 °C per decade (Cubasch et al., 2001). With this warming, snowpack losses are projected to accelerate, reducing high-elevation snowpack to less than half of current amounts by 2050 (Leung et al., 2004; Mote et al., 2010). Timing of snow release has been shown to determine vegetation community composition in the Cascades (Douglas and Bliss, 1979; Evans and Fonda, 1990). Additionally, snowpack is the major source of soil moisture throughout the growing season in these subalpine ecosystems (Douglas and Bliss, 1979). With loss of snowpack under climate change, species composition and the biogeochemical cycling of these communities may shift.

## Objective

Through its influence on vegetation community composition and soil abiotic factors, snow regime has a major influence on the N cycle of subalpine ecosystems. In order to understand ecosystem response to N deposition, it is necessary to understand the interaction of snow regime and the N cycle. We evaluated the combined impacts of N deposition and snowpack loss under climate change on subalpine ecosystem N cycling through modeled simulations in the Regional Hydro-Ecologic Simulation System (RHESSys) model.

## Approach

Regional Hydro-Ecologic Simulation System (RHESSys) is a process-based hydro-ecological model that simulates C, N, and water cycling (see Tague and Band, 2004 for model description). The RHESSys model (version 5.18.r3-NGAS2) was parameterized for subalpine ecosystems at MORA using geographic information system (GIS) maps and field measurements. Digital elevation models, watershed maps, and subalpine vegetation community maps were downloaded from the National Map Viewer and the National Park Service (Pacific Meridian Resources, 1992; Dollison, 2010). Field measurements, including soil depth and bulk density, vegetation phenology, rooting depth, C:N ratios of foliage and roots, and allometric relationships, were added as parameters into the RHESSys soil and vegetation library.

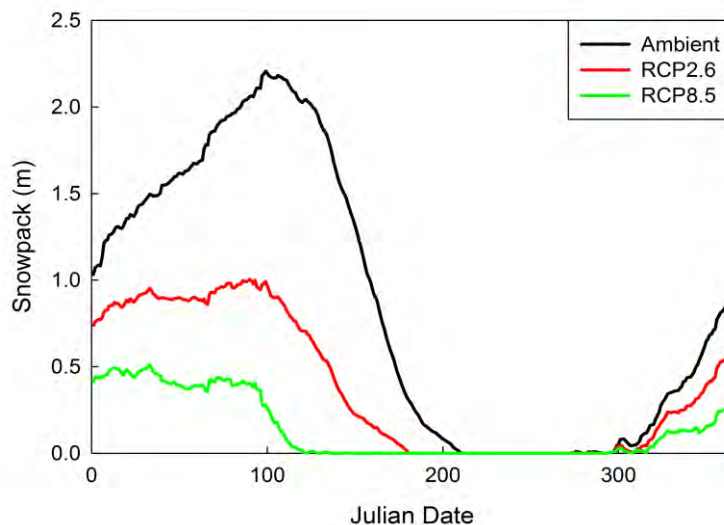
Daily records of precipitation and minimum and maximum temperature were collected from the National Climatic Data Center (NCDC) Paradise ranger station (GHCND:USS0021C35S) between January, 1980 to December, 2013. The model was simulated at the patch scale for 300 years using these repeated meteorological measurements to initialize subalpine soil and vegetation C and N pools. Comparisons of the spun-up modeled site to observed field measurements of soil and vegetation C and N content during the 2013-2014 growing seasons were used for model evaluation. Climate change simulations for 2050 in the RCP2.6 and RCP8.5 scenarios under the CMIP5 framework were performed using warming of 1.0 and 2.0 °C without any significant change in precipitation, which was applied uniformly across 30 years of historic meteorological data (Tague et al., 2008). Simulations of elevated N deposition were performed in



conjunction with the climate change scenarios by adding an additional  $5 \text{ kg N ha}^{-1} \text{ yr}^{-1}$  to the RHESSys atmospheric deposition parameter. These comparisons were conducted using the same initial model state where the climate and deposition scenarios were run for 2008-2013. Snowpack (m), plant N uptake ( $\text{g N m}^{-2} \text{ day}^{-1}$ ), soil  $\text{N}_2\text{O}$  emissions ( $\text{ng N m}^{-2} \text{ day}^{-1}$ ), and inorganic N leaching ( $\text{g N m}^{-2} \text{ yr}^{-1}$ ) output are shown as daily values averaged across 2008-2013.

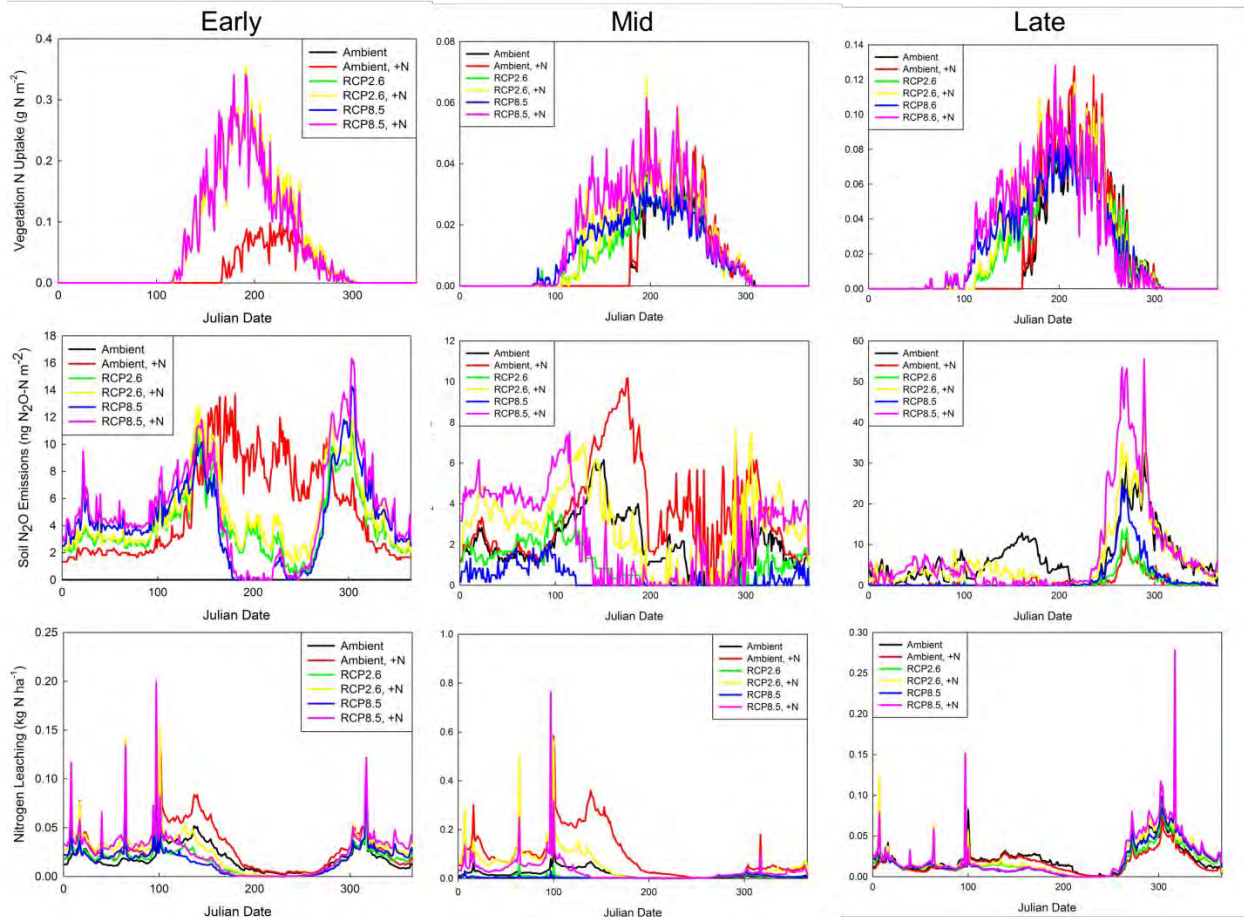
## Findings

1. Modeled snowpack was severely reduced under elevated temperatures in the climate change simulations, decreasing by 91% in the RCP8.5 simulation (Figure 10).



**Figure 10.** The RCP2.6 and 8.5 scenarios, which have  $+1.0$  and  $+2.0$  °C warmer air temperatures, respectively, decreased modeled snowpack by 91% compared to the Ambient simulation.

2. Reduced snowpack under climate change had the largest impact on ecosystem N cycling, lengthening the vegetation growing season and shifting the timing and magnitude of ecosystem N losses (Figure 11). This resulted in higher N loss from soil  $\text{N}_2\text{O}$  emissions and N leaching during winter months and greater N retention from increased plant N uptake and reduced N loss during the growing season.



**Figure 11.** RHESSys simulations of mean vegetation N uptake, soil N<sub>2</sub>O emissions, and soil inorganic N leaching for the Early, Mid, and Late community types under Ambient, RCP2.6 and 8.5 scenarios with ambient and elevated N deposition. The +1.0 and +2.0 °C warmer air temperatures of the RCP2.6 and 8.5 simulations more strongly affected the magnitude and timing of all simulated responses than the elevated N deposition.

## Synthesis

Simulations of ecosystem response to N deposition under climate change simulations indicated that winter snowpack severely decreased, with modeled snow release occurring 40 days earlier than observed in 2013. These projections match those seen in other studies of snowpack in the Cascades, which project 50 to 80% snowpack loss by 2050 under warming scenarios (Leung et al., 2004; Mote et al., 2010). This reduction in snowpack had important implications for ecosystem N cycling, with shifted timing and magnitude of N retention and loss.

Snowpack reduction lengthened vegetation growing seasons at MORA. Plant communities began their growing season up to 40 days earlier, which increased the vegetation N uptake and biomass. This is consistent with modeled studies of vegetation communities under climate change, which have indicated that plant phenology shifts to earlier in the year, lengthening the growing season and increasing productivity (Cayan et al., 2001; Christensen et al., 2008). However, this earlier release of snowpack may expose subalpine vegetation to spring frost damage, which can inhibit growth and N retention (Bannister et al., 2005). These changes in vegetation community response may be exacerbated with the expansion of lower elevation herbaceous and coniferous species into subalpine ecosystems, which has already been evident in

the Cascades (Franklin et al., 1971; Rochefort and Peterson, 1996). Thus, climate change may rapidly alter subalpine vegetation community growth and species composition at MORA.

The earlier snowmelt also induced changes in soil biogeochemistry, driving increased N<sub>2</sub>O emissions and leaching under current snow-covered periods but reduced emissions and leaching during snow-free periods. Melting snow increased soil moisture, which is directly utilized in the model algorithms of N<sub>2</sub>O emissions and leaching, and thus increased these losses as well (Dingman et al., 1994; Brooks and Williams, 1999). However, as snowpack provides the largest reservoir for soil moisture in these subalpine ecosystems, the earlier melting of the snowpack resulted in decreased soil moisture by peak growing season. These results are consistent with other projections of changing snow regimes in alpine ecosystems, where consistency of snow cover has been shown to regulate soil N<sub>2</sub>O emission rates (Brooks et al., 1997). Additionally, inorganic N leaching has been shown to be highest during snowmelt, as snowpack is often considered a reservoir of N deposition (Brooks et al., 1995; Brooks and Williams, 1999; Bilbrough et al., 2000). Thus, loss of snowpack may exacerbate ecosystem N losses during winter months while increasing N retention during the growing season.

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## Outreach

As a product of this research, undergraduate students at the WSU Murrow College of Communication joined the primary investigator to profile N deposition research and impacts in MORA through the Backpack Environmental Program. These students filmed segments of the field campaign and laboratory analyses as well as conducted interviews with researchers at WSU and National Park Service staff. Products from this activity included print publications and media, which have been hosted on the WSU Center for Environmental Outreach and Education (CEREO) [website](#) and the Murrow College of Communication [website](#).

## Products of this Award

- Poinsatte, J., Rochefort, R., and R.D. Evans. 2015. Using isotopic tracers to examine biogeochemical responses to nitrogen deposition in subalpine ecosystems. Oral presentation. Ecological Society of America Annual Meeting, Baltimore, MD.
- Nergui, T., Anderson, S.M., **Poinsatte, J.**, Reyes, J.J., Miller, C.C., Chung, S.H., Evans, R.D., Harrison, J.A., Lamb, B.K., and J.C. Adam. 2015. Understanding excess reactive nitrogen in the environment. Poster presentation. USDA-NIFA AFRI Climate Change Annual Project Director Meeting, Washington, D.C.
- Poinsatte, J., Anderson, S.M., Harlow, B., and R.D. Evans. 2015. Snowpack as a reservoir of nitrogen deposition in subalpine ecosystems of the Cascades. Poster presentation: 1<sup>st</sup> place student poster competition. 86<sup>th</sup> Annual Meeting of the Northwest Scientific Association, Pasco, WA.
- Poinsatte, J., Rochefort, R., and R.D. Evans. 2014. Community-specific biogeochemical responses to atmospheric nitrogen deposition in subalpine meadow ecosystems of the Cascade Range. Oral presentation. American Geophysical Union Fall Meeting, San Francisco, CA.
- Poinsatte, J., Reyes, J.J., Tague, C.L., and R.D. Evans. 2014. Evaluating the impacts of climate change on ecosystem response to atmospheric nitrogen deposition in subalpine meadows of the Cascades. Poster presentation. 5<sup>th</sup> Annual Pacific Northwest Climate Science Conference, Seattle, WA.