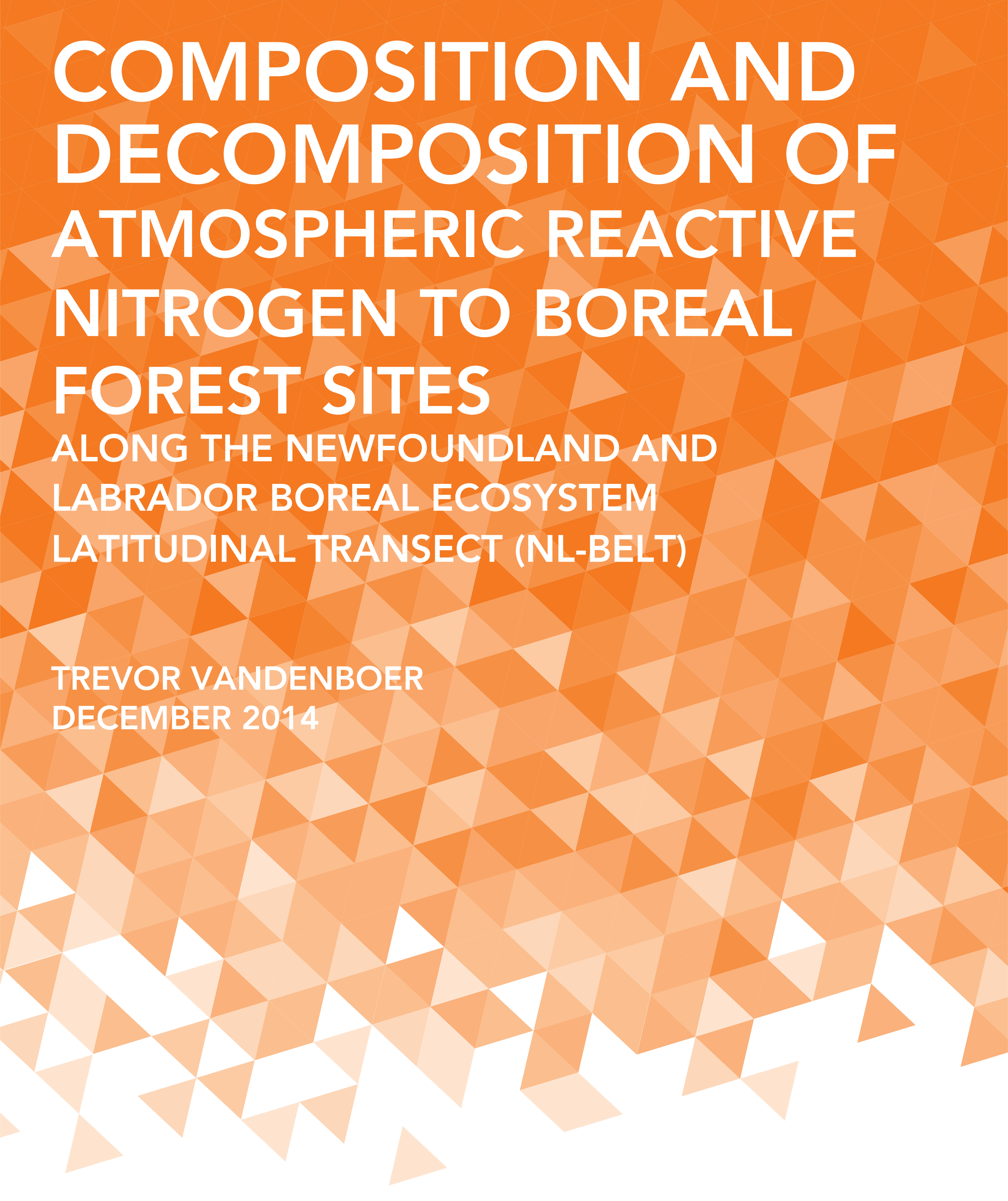

COMPOSITION AND DECOMPOSITION OF ATMOSPHERIC REACTIVE NITROGEN TO BOREAL FOREST SITES

ALONG THE NEWFOUNDLAND AND LABRADOR BOREAL ECOSYSTEM LATITUDINAL TRANSECT (NL-BELT)

TREVOR VANDENBOER
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THE HARRIS CENTRE
Memorial University

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Executive Summary

Increased nitrogen fixation by human activity has mobilized this element to the greatest extent in known history on Earth. Nitrogen released from burning fossil fuels and inefficient fertilizer use transfers reactive-nitrogen compounds to the atmosphere which can be transported to remote ecosystems. Productivity of remote ecosystems may change due to nitrogen-additions by a fertilizing effect, depending on the form deposited, until a tipping-point is reached, which can result in deleterious effects such as soil acidification, algal blooms in aquatic systems, and loss of ecosystem plant and animal diversity. Nitrogen-additions and exchange alters the use and loss rates of this element in affected ecosystems, and also use and loss of carbon, potentially affecting climate processes we know are linked to carbon cycling. These feedbacks make study of single ecosystems challenging and, because of its potential significance in climate change, increasing our understanding should be seen as a pressing issue.

Climate transects, such as the four regions of the Newfoundland and Labrador Boreal Ecosystem Latitudinal Transect (NL-BELT), provide a unique platform to unravel the impact of reactive-nitrogen from climate measurements, leading to a better understanding of ecosystem changes that are expected to occur across the Canadian Boreal region with a changing future climate. In this work, I proposed to evaluate the forms, and their quantity, of reactive-nitrogen from field samples collected across this climate transect in Atlantic Canada to determine the possible impact of atmospheric reactive-nitrogen inputs on the forest ecosystems of the NL-BELT.

Overall, this work highlights the utility of a unique Canadian resource, NL-BELT, in the international research community, aiding in the development of regional and national mitigation strategies for the impacts of climate change on communities dependent on the forestry industry, and to help develop preventative strategies against loss of biodiversity for the protection of our natural ecosystems.

In this work, we undertook two projects. First, we have established infrastructure and preliminary measurements of the reactive nitrogen gases ammonia (NH_3), nitrogen dioxide (NO_2) and nitrogen monoxide (NO). Supporting funds from the Newfoundland and Labrador Department of Natural Resources Centre for Forestry Science and Innovation allowed us to construct, test, and collect preliminary measurements of reactive nitrogen deposited to the NL-BELT in precipitation.

Gases

The preliminary data collected through the Harris Centre Applied Research Fund has allowed us to establish remote monitoring infrastructure across the NL-BELT and to collect preliminary data toward understanding the potential transport, deposition, and exchange of reactive nitrogen in these terrestrial environments. We have successfully made quantitative measurements of NH_3 and NO_2 at levels of tens of parts per trillion, but were unable to quantify NO. With these preliminary measurements in hand, it is now possible to refine and improve the quality and quantity of data collected by this infrastructure over a much longer timeframe to

elucidate statistically robust seasonal and annual trends in the biogeochemical cycling processes of nitrogen in the NL-BELT forests. Our current data suggest that NH_3 levels are similar across the latitudinal transect of forest sites, and will effectively deposit to these ecosystems on a continuous basis. Future work will give us better information on soil composition so that flux quantities can be calculated. Conversely, our measurements of NO_2 show declining levels with increasing latitude, consistent with known long range transport of man-made pollution, but other processes such as soil emissions cannot be ruled out. Future measurements of NO_2 vertical gradients and chemical transport modelling will rigorously test which contributions are the most significant and if these lead to net uptake or release of reactive nitrogen from these ecosystems.

Precipitation

All hardware has been designed, tested, and deployed for data collection to the NL-BELT field sites. Sample collection and analyses were performed with static precipitation samplers from October 2013 – October 2014, while automated samplers have undergone field testing and collected preliminary precipitation samples between August and October 2014. Total nitrogen (N) fluxes to two of the NL-BELT field locations span 1.30±0.71 to 2.7±0.97 kilograms per hectare per year, with significantly more N deposited in the south (Grand Codroy) than further north (Salmon River). These were the only two sites for which annual deposition values were possible to calculate. We caution that uncertainty in sample collection procedures may negate the significance between sites found in this preliminary work since in excess of 100 % variation was found between replicate precipitation samples. Seasonal trends in deposition were not consistent across the sites and times of maximum deposition differed between the locations. The design and preliminary results from the project have been disseminated at provincial, national, and international conferences. In addition, three highly qualified personnel have been trained in precipitation sampler design and construction, sample collection, and sample analysis. A summer student has been trained in the detection and quantitation of key tracers, amino sugars, in tracking the movement of reactive-N in NL-BELT soils affected by N inputs. A field technician has been trained in the collection and processing of samples across the NL-BELT sites. A PhD student, John MacInnis, from collaborator Dr. Cora Young in the Department of Chemistry at MUN has been trained on all components of the precipitation collection program and been awarded a Northern Scientific Training Program award to continue this work under my supervision for the 2014 NL-BELT field season. Collaboration with Dr. Cora Young has led to a new collaboration with Dr. Amila De Silva at the Canadian Centre for Inland Waters facility of Environment Canada to track the transport of persistent pollutants to remote regions, which can be determined from the same samples used for the reactive-N analysis. This work and the established collaborations currently represent three publications in preparation and the long term datasets of the continuing project will generate more robust measures of the interannual variability and seasonal trends in atmospheric reactive N transport to these boreal ecosystems.

Matching Funds and Knowledge Mobilization

- All passive gas sampling hardware has been installed and samples have been collected on a monthly basis from May – November since May 2013, with samples collected over the duration of the 2013-2014 winter from direct and in-kind support provided by the Harris Centre at MUN and the Canadian Forest Service located in Corner Brook, respectively.

- Passive gas sampling results and precipitation sampler design, upon which future data will build, have been or are scheduled for presentation at the following conferences provincially, nationally, and internationally:
  - Canadian Institute of Forestry, Corner Brook, NL; September 2013
  - Canadian Society for Chemistry Annual Meeting, Vancouver, BC; June 2014
  - American Geophysical Union Fall Meeting, San Francisco, CA; December 2013
  - American Chemical Society Spring Meeting, Denver, CO; March 2015

- Collaboration with Dr. Cora Young (Department of Chemistry, Memorial University) has led to a new collaboration with Dr. Amila De Silva at the Canadian Centre for Inland Waters facility of Environment Canada to track the transport of persistent pollutants to remote regions, which facilitates assistance with sample collection. These additional measurements can be made using the same samples that are collected for reactive-N in precipitation.

- All data collected from this project is shared amongst the international collaborators of the NL-BELT project and will be made available through GeoKIRV (Geospatial Knowledge Identification, Retrieval, and Visualization).

Glossary

Accuracy ≡ the degree to which the result of a measurement conforms to the correct value

Certified Reference Material ≡ a measurement standard used to check the quality of analytical measurement methods and instrumentation

Charged compound ≡ a molecule that carries either a net positive or negative charge due to its chemical structure

Chromatography ≡ the separation of a mixture by passing it in solution or suspension or as a vapor through a medium in which the components move at different rates

Compensation point ≡ equilibrium exchange value for a given compound between soils and the overlying atmosphere, where an atmospheric amount below the compensation point results in emission of it from the soil and vice versa

Conductivity ≡ the ability of a material to conduct electricity

Column ≡ an open cylinder packed with beads of a specific material that allows for the separation of a complex mixture introduced at one end in a liquid phase

Detection Limits ≡ the lowest quantity of a substance that can be distinguished from the absence of that substance, within a general confidence limit of 1 %

Elution ≡ the process of separating one material from another on a column by washing it with a solvent

Litterfall ≡ includes plant, leaf, tree, and soil materials which have fallen to the ground

Ion ≡ an atom or molecule with a net electric charge due to the loss or gain of one or more electrons

Ion Chromatography ≡ a process that allows the separation of a mixture of ions and polar molecules based on their affinity for an ion exchanger in a column

Isocratic ≡ a chromatographic method involving a mobile phase whose composition is kept constant and uniform

Isotope ≡ each of two or more forms of the same element that contain the same number of protons but different numbers of neutrons in their nuclei, which differ in relative mass but not in their chemical properties

Matrix ≡ components of a sample other than the analyte of interest

Mixing Ratio ≡ in a sample, the ratio of analyte molecules in relation to the total number of molecules

Openfall ≡ direct precipitation inputs to a collection vessel without passing through any intervening materials (e.g. trees)

Parts per Billion ≡ ratio of molecules of analyte to per billion molecules in a sample

Parts per Trillion ≡ ratio of molecules of analyte to per trillion molecules in a sample

Precision ≡ refers to the closeness of two or more measurements to each other

Product ≡ substances formed during a chemical change

Stoichiometry ≡ the relationship between the relative quantities of substances taking part in a reaction or forming a compound, typically a ratio of whole integers

Throughfall ≡ indirect precipitation inputs to a collection vessel by passing through intervening materials (e.g. trees)

$\delta^{15}N$ ≡ Stable isotopic signature of nitrogen

‰ ≡ ‘per mil’ describing the change in stable isotope composition of a measurement relative to a standard value (e.g. atmospheric dinitrogen has a stable isotopic signature of 0 per mil for the isotope of nitrogen-15)

Acronyms

C ≡ Carbon

C:N ≡ Ratio of carbon atoms to nitrogen atoms

CAPMoN ≡ Canadian Air and Precipitation Monitoring Network

ER ≡ Eagle River Watershed

GC ≡ Grand Codroy Watershed

HR ≡ Humber River Watershed

N ≡ Nitrogen

N_2 ≡ Dinitrogen

NH_3 ≡ Ammonia

NH_4^+ ≡ Ammonium

NL ≡ Newfoundland and Labrador

NL-DNR-CFSI ≡ Newfoundland and Labrador – Department of Natural Resources –Centre for Forestry Science and Innovation

NO ≡ Nitrogen monoxide

NO_2 ≡ Nitrogen dioxide

NO_x = Sum of nitrogen monoxide and nitrogen dioxide

NO_3^- ≡ Nitrate

N_{org} ≡ Organic Nitrogen

N_r ≡ Reactive Nitrogen

NL-BELT ≡ Newfoundland and Labrador Boreal Ecosystem Latitudinal Transect

PANs ≡ Peroxyalkylnitrates

PB ≡ Pynn's Brook Watershed

ppb ≡ parts per billion

ppt ≡ parts per trillion

SR ≡ Salmon River Watershed

Introduction

1. Project Background

Coupled C-N dynamics critical to assessing climate feedbacks in boreal forest landscapes. Nitrogen has traditionally been viewed as a limiting nutrient on ecosystem productivity, and is implicated as a fertilizing agent in remote forests[1,2]. Through its controls on productivity and microbial processes, N may also play a significant role in regulating mechanisms central to climate feedbacks in forests[3-6]. Such feedback mechanisms are of particular interest in boreal landscapes where predicted warming and soil organic matter reservoirs are globally significant[7-9]. The cycling and fate of boreal forest soil organic matter is likely linked to the availability and form of N[10,11]. Nitrogen can enter remote ecosystems in two ways: i) through dinitrogen (N_2) fixing microbes or ii) by transport from other locations in media such as rain water or atmospheric gases (Figure 2). The increased mobilization of N_r from human activity has increased ecosystem inputs of nitrogen via transportation process[12], in some cases leading to nitrogen saturation of sensitive ecosystems[13]. Thus, to understand how boreal forest ecosystem C-cycling and sequestration will respond to a changing climate, measurements of N_r input quantities and chemical identities of N_r compounds must be made to contrast its significance and potential impact against the magnitudes of the terrestrial and aquatic N-cycles at these locations.

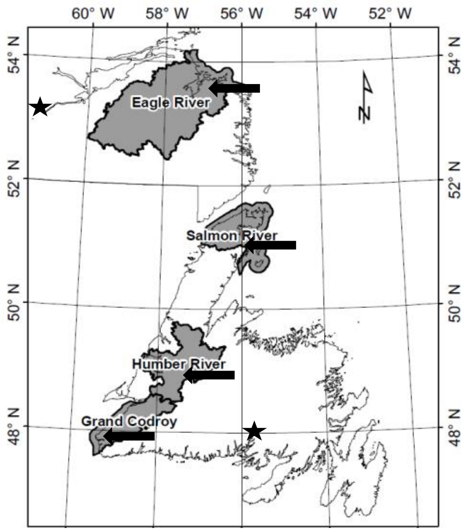
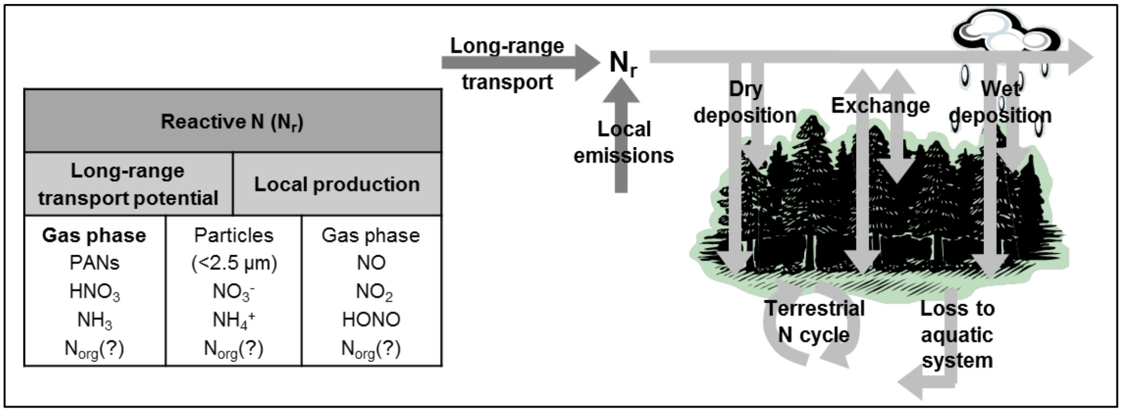


Figure 1. *Locations of CAPMoN sites (stars) for atmospheric inputs and NL-BELT instrumented sites (arrows) in Atlantic Canada. Shaded areas represent National Hydrological Network units associated with NL-BELT.*

Atmosphere-ecosystem exchange of reactive N (N_r) species. Exchange of N_r across the atmosphere-ecosystem boundary has been explored extensively for NH_3[14,15] and lower order nitrogen oxides, such as NO and NO_2[16]. However, effective parameterizations for exchange processes based on soil properties such as nutrient content, temperature, water content, and pH, have not been developed to describe the exchange of these compounds in the NL-BELT ecosystems. Thus, it represents a critical new undertaking to constraining transport of N_r through collaboration across soil science, atmospheric chemistry, and aquatic-terrestrial biogeochemistry[17].

Filling in the N source gaps along a boreal forest climate transect. The impact of climate change on the cycling and fate of soil organic carbon reservoirs in boreal forest ecosystems is currently under investigation by the research group of Dr. Susan Ziegler. She and her colleagues have established the NL-BELT - a climate transect of four forested watersheds with established terrestrial and stream sites along the west coast of Newfoundland and southern Labrador (Figure 1). Currently, the forms and quantities of atmospheric N_r inputs are not constrained for the NL-BELT forest sites (Figure 2). Only two Canadian Air and Precipitation Monitoring Network (CAPMoN) sites are measuring nitrate (NO_3^-) and ammonium (NH_4^+) in NL precipitation at locations a considerable distance from NL-BELT (Figure 1). Measured quantities of this N_r subsection in precipitation from 2007 through 2011 by CAPMoN have shown rain water to be a consistent source of N_r to the southern region, consistent with chemical transport model predictions for long-range transport of reactive-N from the central and northeastern US[18,19]. This historical data suggests nearly equal importance of NO_3^- (60 %) and NH_4^+ (40 %) in precipitation inputs, but the sparse data coverage cannot be assumed comparable for the NL-BELT locations. Additionally, precipitation is only one mechanism of N import to ecosystems via the atmosphere and NO_3^- and NH_4^+ are only some of the possible N_r-compounds that may be transported (Figure 2). Organic N (N_{org}), in particular, is poorly understood, yet being consistently reported as significant and sometimes the dominant form of N-input to remote locations[10,13,20,21]. Therefore, measures of atmospheric N-inputs represent a critical knowledge gap in developing a predictive understanding of forest ecosystem productivity and biogeochemistry with changing climate.



Reactive N (N_r)		
Long-range transport potential		Local production
Gas phase	Particles (<2.5 μm)	Gas phase
PANs	NO_3^-	NO
HNO_3	NH_4^+	NO_2
NH_3	N_{org}(?)	HONO
N_{org}(?)		N_{org}(?)

Figure 2. Definition of reactive nitrogen (N_r) to be collected at NL-BELT and a schematic of N_r atmospheric biogeochemical cycling.

Elemental and stable isotopic signatures of soils. Measurements of the N stoichiometry and stable N-isotope ($\delta\ ^{15}N$) values in soils provides some evidence for two possible phenomena relevant to the interpretation of climate effects and biogeochemistry of NL-BELT forests: i) atmospheric N-inputs vary in quantity and composition with latitude along the transect; and/or ii) more rapid recycling of N at the most southern site currently reduces tree N-limitation relative to cooler regions due to increased N-cycling with climate warming.

Litterfall inputs, mosses and soil exhibit increasing $\delta^{15}N$ values from north (ER) to south (GC) along the transect. Coniferous needle litterfall, the dominant soil input in these forests, increases by more than 4 ‰ from north to south (Figure 3) which could reflect increased N-cycling in the warmer climate forests. Chemical transport models[18,22], while not projecting as far as NL along the Atlantic coast, and observations suggest that atmospheric N_r should become more $\delta^{15}N$ deplete to the levels observed at the northern end of NL-BELT when transported long distances from within North America[12,23]. Litterfall C:N for the brown litterfall from NL-BELT decreased from 87 in the north to 64 in the south signifying potential increase in tree N-availability with climate warming. The decrease in difference between brown and green needle litterfall C:N potentially indicates a decrease in tree N-resorption from north to south. This data suggests that potential increases in N availability, and perhaps N-cycling, may occur with warming in these forests (Figure 3). Constraining ecosystem N sources, particularly atmospheric N_r, is critical to uncovering potential controls on forest productivity, diversity, and function with climate warming.

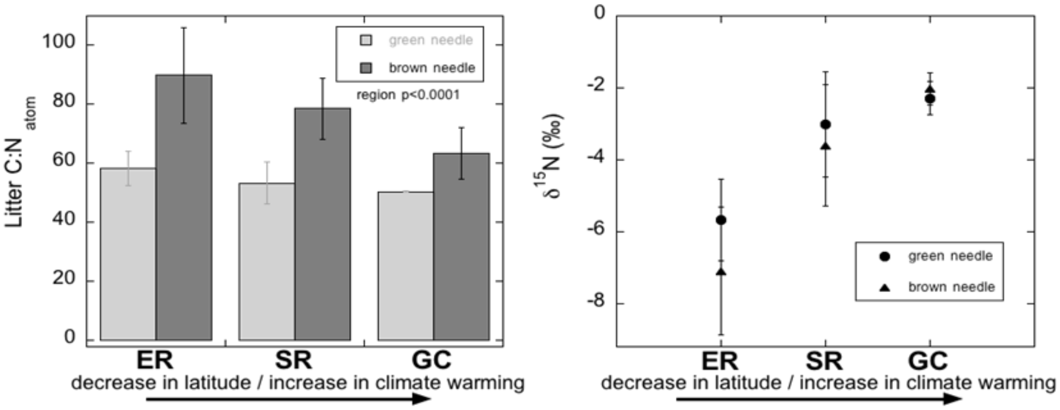


Figure 3. *Regional trends in N from north (ER) to south (GC). (**Left)** Litterfall carbon to nitrogen ratios sorted by litter source type and region. (**Right)** Litterfall $\delta^{15}N$ sorted by litter source type and region.*

Constraining N_r at the biosphere-atmosphere interface. The quantities of atmospheric-N gases, N-containing particulate matter, and organic-N species deposited is unknown (Figure 2), but likely significant in relation to the total quantity of N deposited to these remote systems, as has been found for many locations[15,20,24-27]. In particular, NH_3, particles and organic-N compounds like peroxyalkylnitrates (PANs) have long atmospheric lifetimes (weeks to months)[28,29], which allows them to be transported over great distances before being removed by dry or wet deposition (Figure 2). Thus, the atmospheric sources of N_r to ecosystems is complex and requires monitoring of many different compounds to produce a complete picture of the biogeochemical cycle at the biosphere-atmosphere interface. Furthermore, the isotopic signatures carried by atmospheric N_r have not been measured at these sites, but could allow us to discern between whether inputs exhibit decreasing $\delta^{15}N$ with transport, or if ecosystem recycling processes are dominating the signatures observed at these sites.

2. Objectives

A pair of projects were proposed to be undertaken over a one year period starting in May 2013 that would preliminarily identify and quantify atmospheric some forms of N_r at the NL-BELT sampling regions The goals of Project 1 (funded by the Harris Centre) were to provide measurements and speciation of the quantity of atmospheric-N as NO, NO_2, and NH_3 on monthly and annual timescales. From these field measurements of atmospheric gases, the possibility for deposition would be explored. The goals of Project 2 (funded by the NL-DNR-CFSI) were to build 24 fully automated precipitation samplers and to collect preliminary data on total and speciated nitrogen inputs to the NL-BELT. Taken together, this research could link atmospheric-N isotopic signatures to the litterfall and soil $\delta^{15}N$ and C:N observations to further the understanding of other nutrient fluxes in research conducted by collaborators at NL-BELT. In the case that these inputs and cycling are minor contributors to the N-cycle at NL-BELT, it will rule out one source contributor to the $\delta^{15}N$ signatures measured and allow nutrient budgets to be better constrained. It will also provide a key insight into the relative role of atmosphere-biosphere exchange, where presumably nutrient limited ecosystems will be impacted by even small exchanges of N_r. This project, under an extended and expanded sampling framework, will allow results to be upscaled from the sites of NL-BELT to a broader representation of global N-cycling at the biosphere-atmosphere interface.

3. Research Methodology and Approach

Project 1 - Atmospheric gaseous N_r species and their quantities along NL-BELT

Establishing monitoring infrastructure for the collection of the reactive nitrogen gases NO, NO_2 and NH_3 was completed by procuring 15 Ogawa® passive diffusion samplers for each region within the NL-BELT (Figure 1). Concentrations of NO are determined by collecting both NO and NO_2 (*aka* NO_x) and subtracting the independent NO_2 measurement from co-located passive samplers. The Ogawa® samplers were installed in groups of five (5) at three (3) sites within each region, colocated with instrumentation recording atmospheric temperature, atmospheric relative humidity, soil temperature, and soil water content. Each sampler collects duplicate samples, such that each group of five (5) Ogawa® samplers produced statistical replication for each gas (n=3) within and between the four NL-BELT sampling regions (Figure 4). Field blanks (n=2 per analyte) and reagent blanks (n=1) were also collected at each site over the same duration of sample collection or during sample processing, respectively. This was done to track inadvertent collection of target analytes during transport of equipment to field locations, and through subsequent sample extractions and analysis. Samples for each reactive nitrogen gas were collected on a monthly basis from May through October 2013 and once from October 2013 to May 2014 across NL-BELT.

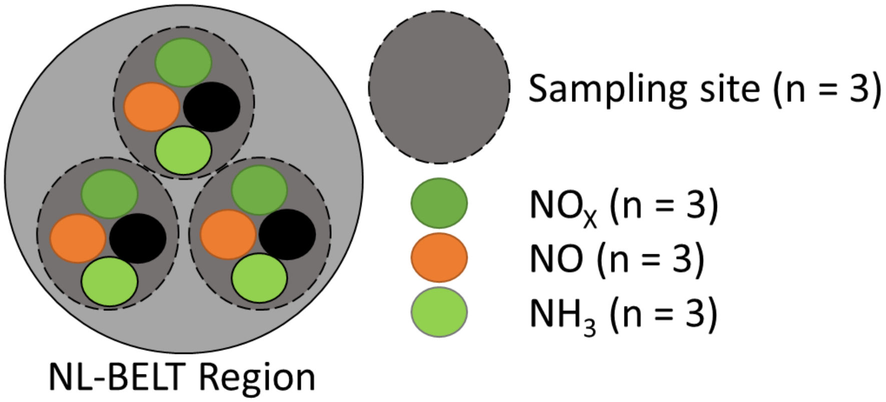


Figure 4. *Passive sampling experimental design of nested triplicates.*

For each type of passive sampler, the target gas is collected by diffusion followed by reaction on a chemically impregnated filter. The resultant reaction product is stable on the filter and recovered by extraction in 8 mL of ultrapure deionised water by intermittent shaking over 60 minutes. This procedure yields an analyte quantitatively collected in solution which carries a charge. In the case of NH_3, the charged product is NH_4^+ and in the case of both NO and NO_2, the charged product is NO_2^-. These charged molecules (*aka* ions) were analyzed using an ion chromatography instrument to separate them from other ions present in the same sample as part of the matrix. The target ions were detected by conductivity after separation by an isocratic elution program.

To separate NH_4^+ from the matrix in the NH_3 passive samples, an isocratic separation was performed using a Dionex CS12A analytical and guard column assembly in conjuction with a trace cation concentration column (TCC-ULP1, Dionex Corp., Sunnyvale, CA). The eluent was composed of 20 mM H_2SO_4 at a flow of 2 mL min^{-1} and room temperature. The eluent was passed through a CSRS-3000 suppressor run in recycle mode to remove the eluent ions from the flow exiting the columns to improve analyte detection. Analyte NH_4^+ was detected by a conductivity detector set with a sensitivity of 30 µS.

To separate NO_2^- from the matrix in the NO_x and NO_2 passive samples, an isocratic separation was optimized and performed on a Dionex AS9-SC analytical and guard column assembly in conjunction with a trace anion concentration column (TAC-ULP1, Dionex Corp., Sunnyvale, CA). For the NO_2 passive samples the eluent was composed of 1.8 mM Na_2CO_3 and 1.7 mM $NaHCO_3$ and for the NO_x passive samples the eluent was composed of 0.425 mM Na_2CO_3 and 0.45 mM $NaHCO_3$. Both separations were done at a flow of 2 mL min^{-1} and room temperature. The eluent was passed through an ASRS-3000 suppressor run in recycle mode to remove the eluent ions from the flow exiting the columns to improve analyte detection. Analytes were detected by a conductivity detector set with a sensitivity of 30 µS.

In both ion chromatography analyses, the amount of the analyte is determined from instrumental calibration. The reliability of these analyses was investigated in detail from an analytical perspective where accuracy and precision were determined for the entire method. Subsequent conversion of the analyte concentration to an atmospheric mixing ratio during the sampling period (see the corresponding sections of Project Details and Results below).

Project 2 – Atmospheric precipitation Nr deposition, speciation, and quantitation along NL-BELT

Supporting funds were provided by NL-DNR-CFSI for the design and construction of automated precipitation collection units for quantifying total, inorganic and organic N inputs directly from the atmosphere and from precipitation falling through the forest canopy (n = 24 per month). Determining N_r quantities and speciation relevant to ecosystem processes was performed from October 2013 through October 2014 in conjunction with the availability of these funds through the former Office of Research Services at Memorial University.

In order to compare fluxes of these N_r species to factors controlling those fluxes (e.g. soil moisture, temperature) sample collection must be made over two or more years[10]. Thus, these supporting funds were used to build functional precipitation collectors, establish baseline methodology tests, and conduct preliminary measurements.

For the full duration of the October 2013 to October 2014 sample collections, passive samplers were deployed across the four NL-BELT regions to collect total rainfall inputs for subsequent analysis. No microbial killing agents were used in these samplers as their design required field confirmation of being leak-free to be sure hazardous materials would not be released into the environment. The assembly, setup, repair, and maintenance of these samplers was optimized over the collection period. More detail on these samplers can be found in Appendix ‘A’.

The design and construction of the 24 automated precipitation samplers, their associated sensing electronics, and solar panel array were completed in August 2014. Prior to field deployment, functionality of each sampler and their control electronics was tested for 7 days. Six (6) automated samplers were deployed at each region of the NL-BELT with three (3) collecting incident precipitation (i.e. atmosphere to sampler with nothing intervening, *aka* openfall) and three (3) collecting precipitation passing through the canopy (*aka* throughfall). From August through October 2014 preliminary tests were carried out to determine: i) openfall and throughfall collected volume reproducibility, ii) differences in total N_r deposited per unit area, iii) differences in total N_r deposited from openfall and throughfall, and iv) the necessity of microbial preservation agents for stabilizing precipitation samples over the month-long collection periods. More detail on these samplers and their associated tests and results can be found in Appendix ‘A’.

4. Project Details and Results

4.1. Passive Sampling - Nitrogen Dioxide (NO_2)

Separations and Matrix Effects

Nitrite (NO_2^-) was found to elute at a retention time of 8.20±0.02 minutes under these separation conditions. The detector response to NO_2^- was linear ($R^2 > 0.99$) across a calibrated range of 0.003 to 0.12 mg L^{-1}, representative of the concentration range expected in real samples. There were no discernable interferences from the coextracted ions in the sample matrix. This was determined by comparing instrument signal for calibration standards prepared in ultrapure deionised water and in the presence of the passive sampler reactive pad extract matrix on at least 5 different days. There was no statistically significant difference between the slopes or intercepts of the lines generated within and between days.

Detection Limits

Routine determinations of the calibration factors allowed the instrument accuracy and precision to be determined as ≤ 15 % and ≤ 20 %, respectively, over n = 5 calibrations. These were determined through the use of check standards with known concentrations of NO_2^- made from a certified mixed anion standard (Dionex 7-anion mixed standard) and standards prepared from pure $NaNO_2$ dissolved in ultrapure deionised water. The upper limit on these values was reached as analyte concentrations approached the limit of detection. The detection limits, when applied to the passive sample extracts, were determined as three times the instrument signal at the retention window of the target NO_2^- and had a value below a part per trillion in the atmosphere, much lower than known ambient mixing ratios. Thus the entire analytical method was demonstrably suitable for the detection of atmospheric NO_2.

Calculations

To convert the mass of detected NO_2^- in a sample extract to an atmospheric mixing ratio of NO_2 in parts per billion, Equation 1 was used:

$$NO_2\ (ppb) = \frac{\alpha_{NO2} \cdot mNO_2^{\,-}}{t} \qquad \text{(E1)}$$

Where mNO_2^- is the mass of NO_2^- in a sample extract, t is the sampling duration in minutes, and α_{NO2} is a conversion coefficient in ppb·min·ng^{-1} determined from collocated atmospheric measurements of temperature and relative humidity (Equation 2)

$$\alpha_{NO_2} = \frac{10000}{(0.677 \times P \times RH) + (2.009 \times T) + 89.8} \qquad \text{(E2)}$$

Where P is a temperature corrected water vapour pressure in mmHg, RH is the relative humidity in percent, and T is the temperature in degrees Centigrade.

These calculations were carried out on all samples collected across the NL-BELT, which were pooled together for each region at each collection time point (Figure 5). The variability in the measurements for any given collection was calculated as one standard deviation from the mean.

Measurements

Nitrogen dioxide measurements over the observation period (Figure 5) show a clear gradient with atmospheric concentrations decreasing from south to north sampling locations. The observed mixing ratios were, generally, on the order of tens of parts per trillion (ppt). Grand Codroy, the most southerly location consistently had the highest measured NO_2 mixing ratios near 30 ppt, with an increase to 50 ppt in August 2013. The reason for this increase is not clear and requires continued observations (see Recommendations and Future Research). Mixing ratios decreased to 15-20 ppt over the observation period in Salmon River and the two samples from Eagle River show very low mixing ratios (5 ppt) in July through August, increasing to 15 ppt in the period from September through October.

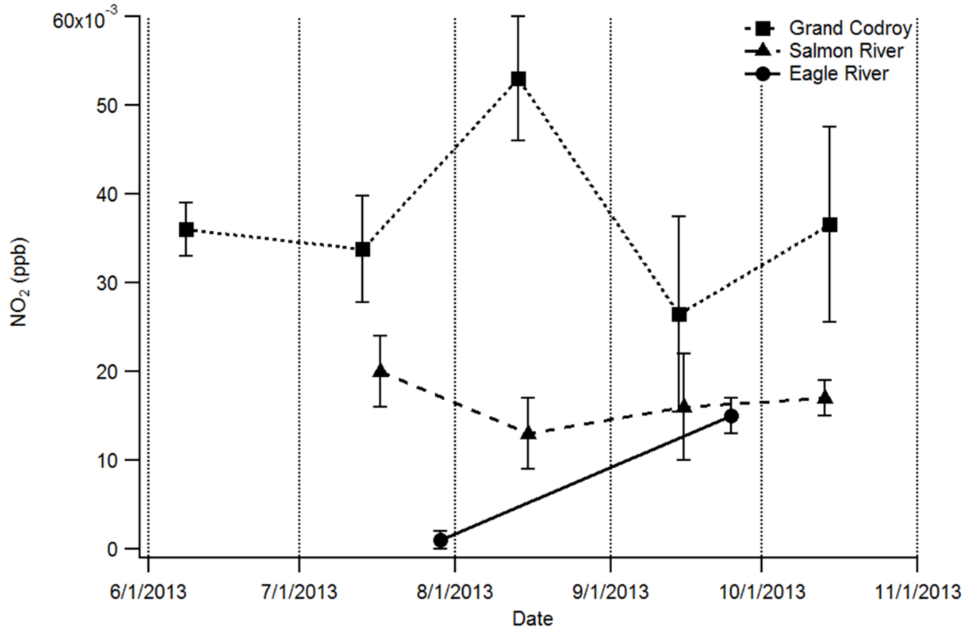


Figure 5. *Seasonality and concentrations of $NO_{2(g)}$ across the NL-BELT showing a latitudinal gradient in atmospheric mixing ratios, which increase moving from the most northerly site to the most southerly. Data points are plotted at the midpoint of the sampling period. For each plot within the region sampling period averages (± 1 s.d.) are shown for Eagle River, Salmon River, and Grand Codroy.*

Peroxyacetic nitric anhydride (PAN) has been measured as a significant reservoir species for the transport of N_r to remote regions, such as eastern Canada[30,31]. These aircraft studies over Newfoundland and Labrador have suggested that transport of nitrogen oxides, as NO_2, to remote locations like the NL-BELT is driven primarily by the reservoir compound PAN that is exported from source regions (e.g. urban locations in the US northeast or along the St. Lawrence seaway). However, the seasonality of PAN infiltration to the lower atmosphere is not well understood[32]. PAN and other reactive nitrogen compounds have been modeled to be diluted as they undergo transport away from source regions and are lost due to chemical and physical processes[18], consistent with our observations. These data suggest that this may be the case for the NL-BELT sites and that collection of PAN data during sampling periods at these locations could have high utility.

Alternatively, NO and NO_2 can be released from soils due to microbial production of these gases. The emission of NO will lead to the subsequent production of NO_2 in the atmosphere, since it is an oxidizing environment. To determine whether soil emissions are significant contributors to these observations will require measurements made at least at two heights above the forest floor, with one closer and further away from the soil surface (see Recommendations and Future Research). Thus, determining the net deposition or emission of nitrogen dioxide (NO_2) from the NL-BELT regions requires further research.

4.2. Passive Sampling - Nitrogen Monoxide (NO) = NO_x – NO_2

Separations and Matrix Effects

Nitrite (NO_2^-) was found to elute at a retention time of 9.70±0.02 minutes under the optimized separation conditions for the more complex matrix of these reactive pads. The detector response to NO_2^- was linear ($R^2 > 0.99$) across a calibrated range of 0.003 to 0.12 mg L^{-1}, representative of the concentration range expected in real samples. Initially, when the ion chromatography conditions for NO_2^- from the NO_2 samples were used, there was a significant interferences from the coextracted ions in the sample matrix (Figure 6). This was determined by comparing instrument signal for calibration standards prepared in the presence of the passive sampler reactive pad extract matrix on 2 different days. There was statistical difference between the slopes and intercepts of the lines generated within and between days indicative of 33 % loss in signal for the same quantity of nitrite in a NO_x sample compared to that of NO_2. There was also increased uncertainty in the slope of the line for the NO_x matrix-matched calibrations with an intercept equal to the second highest standard under complete separation. Given that the quantity of NO is determined by the difference in NO_2^- measured in the NO_x sample and the NO_2 sample, increased uncertainty in the NO_x measurement could invalidate the ability to detect a statistically significant difference between the passive sample extracts. Overall, the results of this analytical work indicated that the NO_x extract matrix interference was variable and capable of masking NO_2^- present in a real sample. Thus, improved separation of the NO_2^- from the matrix was required and optimized.

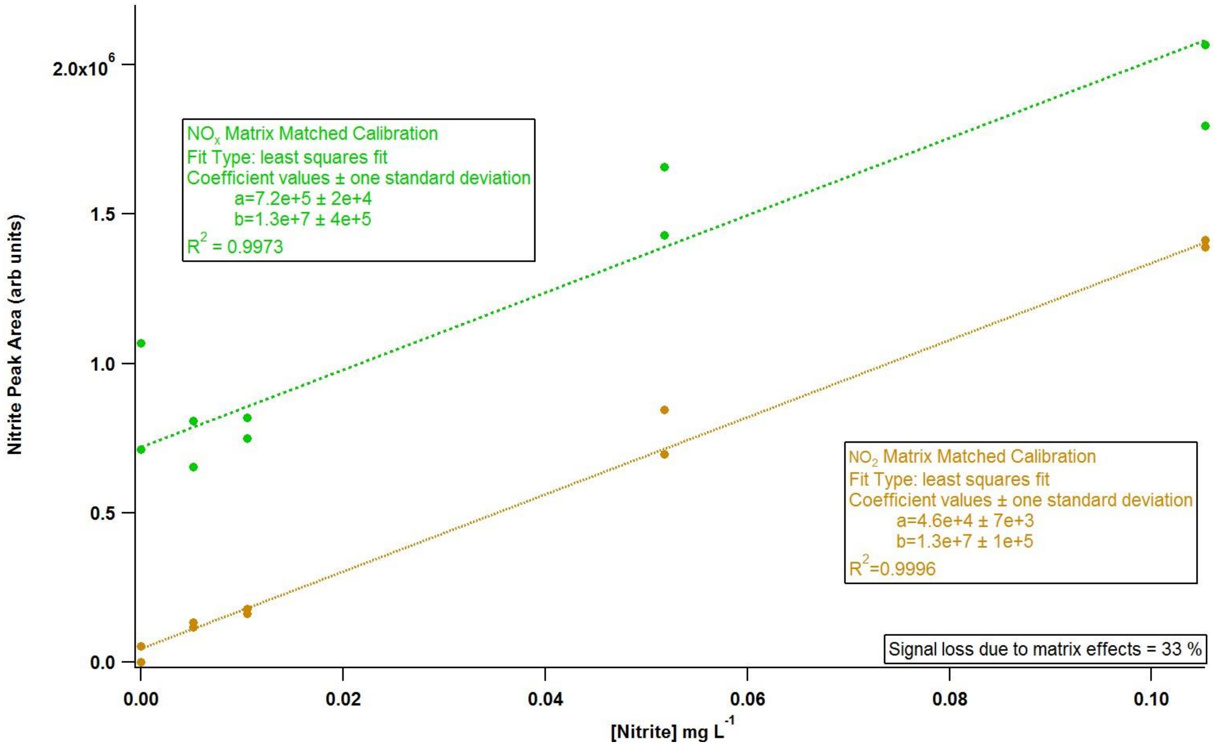


Figure 6. *Comparison of absence of matrix effects in NO_2 sample extracts (orange) and presence of matrix effects in NOx sample extracts (green) with known amounts of nitrite added when both are analyzed by the optimized chromatography method for NO_2 extracts.*

Detection Limits

Routine determinations of the calibration factors allowed the instrument accuracy and precision to be determined as ≤ 15 % and ≤ 20 %, respectively, over n = 5 calibrations. These were determined through the use of check standards with known concentrations of NO_2^- made from a certified mixed anion standard (Dionex 7-anion mixed standard) and standards prepared from pure $NaNO_2$ dissolved in ultrapure deionised water. The upper limit on these values was reached as analyte concentrations approached the limit of detection. The detection limits, when applied to the passive sample extracts, were determined as three times the instrument signal at the retention window of the target NO_2^- and had a value below a part per trillion in the atmosphere, much lower than known ambient mixing ratios. Thus the entire analytical method was demonstrably suitable for the detection of atmospheric NO_2.

Calculations

To convert the mass of detected NO_2^- in a sample extract to an atmospheric mixing ratio of NO in parts per billion, Equations 3 and 4 were used:

$$mNO_2^-{}_{(NO)} = mNO_2^-{}_{(NOx)} - mNO_2^-{}_{(NO2)} \qquad (E3)$$

$$NO\ (ppb) = \frac{\alpha_{NO} \cdot mNO_2{}^{-}{}_{(NO)}}{t} \qquad \text{(E4)}$$

Where $mNO_2{}^{-}{}_{(NO)}$ is the mass of $NO_2{}^{-}$ in a sample extract in nanograms derived from NO, t is the sampling duration in minutes, and α_{NO} is a conversion coefficient in $ppb \cdot min \cdot ng^{-1}$ determined from collocated atmospheric measurements of temperature and relative humidity (Equation 5)

$$\alpha_{NO_2} = \frac{10000}{(-0.78 \times P \times RH) + 220} \qquad \text{(E5)}$$

Where P is a temperature corrected water vapour pressure in mmHg, and RH is the relative humidity in percent.

These calculations were not possible to carry out on the samples collected across the NL-BELT, due to additional matrix effects not captured by the preceding analysis by standard addition to the extracted NO_x matrix.

Measurements

The NOx, and therefore the NO, measurements were all affected by matrix degradation products of the reactive pads. The interferences were namely acetic and formic acids generated by the hydrolysis of the cellulose filter that retains the reactive compound for collecting the target analytes. In these analyses, the cellulose filters were not removed from the extracts after shaking and degraded up until the time of analysis by ion chromatography. As such, when $NO_2{}^{-}$ was measurable, it was often in the presence of very high background signals from this interference, resulting in unreliable data. More often than this, the interference was large enough to make $NO_2{}^{-}$ completely immeasurable. Strategies to overcome this limitation in future work are discussed in detail below (see Recommendations and Future Research).

4.3. Passive Sampling – Ammonia (NH_3)

Separations and Matrix Effects

Ammonium ($NH_4{}^{+}$) was found to elute at a retention time of 8.80 ± 0.02 minutes under the separation conditions used. There were no discernable interferences from the coextracted ions in the sample matrix. This was determined by comparing instrument signal for calibration standards prepared in ultrapure deionised water and in the presence of the passive sampler reactive pad extract matrix on at least 5 different days. There was no statistically significant difference between the curves generated within and between days. The detector response to $NH_4{}^{+}$ was non-linear, as determined in previous work for weak bases[33], which was best fit with Equation 6:

$$y = -\frac{A}{2} + \sqrt{\left(\frac{A}{2}\right)^2 + C[TN]} \qquad \text{(E6)}$$

Where A and C are fitting coefficients related to the acid dissociation constant of NH_4^+ and the conductivity detector sensitivity, y is the measured peak area by ion chromatography, and [TN] is the amount of NH_4^+ in a sample of unknown concentration.

Accuracy, Precision, and Detection Limits

Routine determinations of the calibration factors allowed the instrument accuracy and precision to be determined as ≤ 15 % and ≤ 1.6 %, respectively, over n = 8 calibrations (Table 1). These were determined through the use of check standards and the certified reference material AES-05 rain water provided by Environment Canada. The upper limit on these values was reached as analyte concentrations approached the limit of detection. The detection limits, when applied to the passive sample extracts, were determined as three times the instrument signal at the retention window of the target NH_4^+ and had a value below a part per trillion in the atmosphere, much lower than known ambient mixing ratios. Thus the entire analytical method was demonstrably suitable for the detection of atmospheric NH_3.

Table 1. *Quality assurance and quality control results for IC analytical method applied to NH_3 passive sampling extracts over n = 8 calibrations.*

QA/QC Sample	**$[NH_4^+]_{known}$ ($mg\ L^{-1}$)**	**Accuracy (%)**	**Precision (%)**
Certified Reference Material (AES-05)	0.243±0.024	15	0.9
Certified Reference Material (AES-05) (x10 dilution)	0.0243	7.5	1.3
Check Standard 1	0.1	9.1	1.1
Check Standard 2	0.02	1.5	1.6

Calculations

Using the calibration curve derived using Equation 6, the concentration and total amounts of NH_4^+ in nanograms in the aqueous extracts were determined. The total amounts could then be converted to an atmospheric mixing ratio in parts per billion using Equation 7:

$$NH_3(ppb) = \frac{\alpha_{NH3} \cdot mNH_4{}^+}{t} \qquad \text{(E7)}$$

Where mNH_4^+ is the mass of NH_4^+ in a sample extract in nanograms, t is the sampling duration in minutes, and α_{NH3} is a conversion coefficient of 43.8 ppb·min·ng^{-1}.

The potential for the exchange of NH_3 between the atmosphere and biosphere was calculated using the concept of a compensation point (Equation 8) between the NH_4^+ found in the soils of the NL-BELT and the mixing ratios of NH_3 observed in the atmosphere. The compensation point calculation relies on measurements of soil physical properties and atmospheric temperature to predict the movement of NH_3 between the two reservoirs. The value of the compensation point is determined as an atmospheric mixing ratio.

$$\chi_{NH_{3(g)}} = (1.01 \times 10^{-11}) \times \frac{[NH_4{}^+]}{[H^+]} exp\left(-10378\left(\frac{1}{T} - \frac{1}{298.15}\right)\right) \quad \text{(E8)}$$

Where $[NH_4^+]$ is the concentration of ammonium in soil pore water, $[H^+]$ is the acidity of the soil as determined from a pH measurement, and T is temperature in degrees Kelvin. The compensation point was calculated for soils with sufficient characterization of ammonium content and pH, and with collocated atmospheric temperature measurements.

If atmospheric observations of NH_3 are greater than the compensation point, then NH_3 will deposit continuously to the soils. If NH_3 observed is less than the compensation point, emission occurs. Figure 8 shows the concept of compensation point schematically.

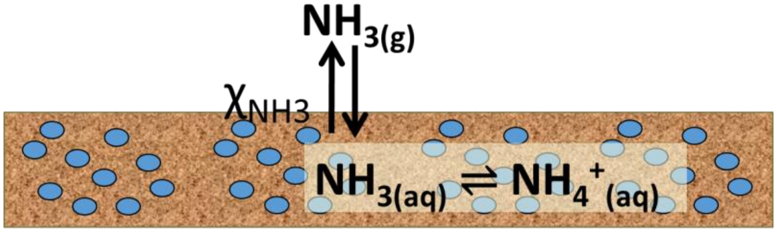


Figure 8. *Chemical schematic of the exchange of NH_3 with soil pore water, where the NH_3 enters the aqueous phase and can transform to NH_4^+. The process is dynamic and therefor reversible. Exchange processes depend on soil pH, water content, and the amount of NH_3 in the overlying atmosphere. The net flux of NH_3, F_{NH3}, can be calculated from the parameterized compensation point by: $F_{NH3} = V_{ex}([NH_{3(g)}] - \chi_{NH3})$.*

The exchange velocity, V_{ex} in cm s^{-1}, of NH_3 depends on three processes that can be conceptualized as the sum of three resistances in series (Equation 9):

$$V_{ex} = \frac{1}{R_a + R_b + R_c} \quad \text{(E9)}$$

Where, R_a is the aerodynamic resistance representative of atmospheric mixing, R_b is the laminar boundary layer resistance that represents elements present at the surface that affect diffusivity of gases to the surface (e.g. trees), and R_c is the residual resistance toward the exchange of a gas with the surface reservoir (e.g. soil, water, etc.). As is the case for NH_3, compounds with high affinity for surfaces or high water solubility can be more generally described with an exchange velocity on the order of 1 -2 cm s^{-1}. These calculations and concepts were used to understand the potential atmosphere-biosphere exchange of ammonia.

Measurements

There was no observed gradient in NH_3 across the NL-BELT, but some seasonal trends were observed with NH_3 declining from the summer into the fall (Figure 9). Observed mixing ratios ranged as high as 200 ppt, but were generally in the 30 – 70 ppt range. Using the concept of a compensation point (i.e. equilibrium exchange of NH_3 in soils with the overlying atmosphere), we found that NH_3 will deposit continuously to all of the NL-BELT ecosystems excepting an area in Grand Codroy which had an isolated soil pH value reaching 6.0 (Figure 9, upper green dotted line). However, these calculations are preliminary and based on a small survey of soil pH and $[NH_4^+]$ measurements and prevent us from being able to quantify the total amount of deposited NH_3 to the NL-BELT ecosystems (see Recommendations and Future Research).

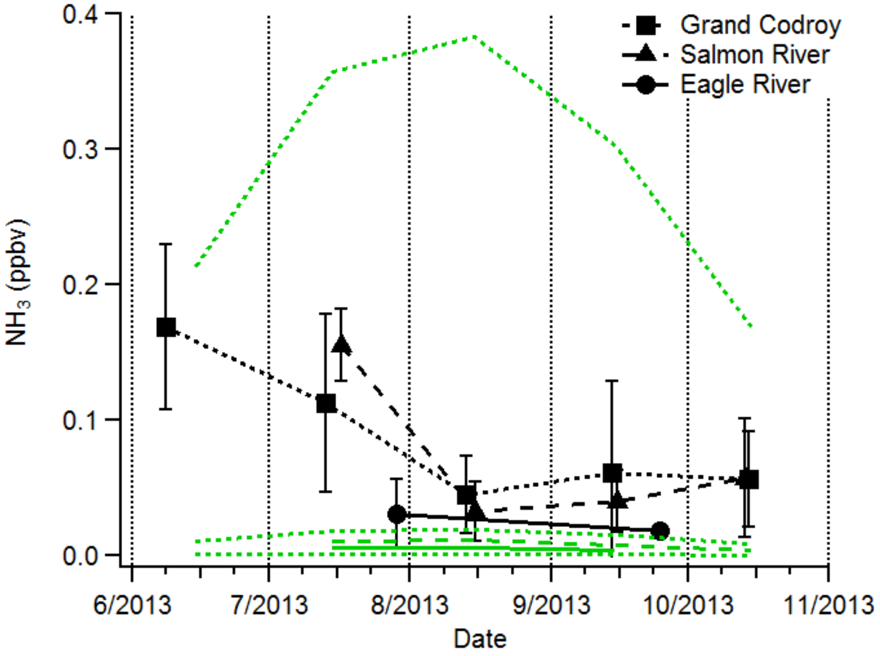


Figure 9. Seasonality and mixing ratios of passive samplers for $NH_{3(g)}$. Data points are plotted at the midpoint of the sampling period. Within site variability for each plot within the region and sampling period averages (± 1 s.d.) are shown for Eagle River, Salmon River, and Grand Codroy. When combined with the natural gradient in soil temperatures, and soil nutrient loadings and pH across the NL-BELT, the potential for biosphere-atmosphere exchange can be assessed by compensation point calculation (green lines with corresponding dash types). For Grand Codroy, the range of measured soil pH values was modeled (green dotted lines) for the upper (6.0), median (4.4), and lower limits (3.2). Measurements greater than the compensation point denote a potential for deposition.

4.4. Precipitation

(see submitted CFSI funding report in Appendix ‘A’)

5. Recommendations on Project Implementation and Future Research

Future research will undertake longer collection periods for all the N_r species targeted from this preliminary project. Extended observations are essential to elucidating seasonal and annual trends in the nitrogen cycle across NL-BELT and, ideally, another 3-5 years of observations will be made to support these first observations. This will help us to understand why sudden increases in gases, such as NO_2 in Grand Codroy in August 2013, occur. More observations of these occurrences will lend itself to a higher likelihood that the physical and/or chemical processes, such as soil emissions, that drive these observations can be determined. One way in which this will be accomplished is by expanding our sampling infrastructure numbers or relocating some samplers closer to the ground surface to determine if NL-BELT soils are sources or sinks of the reactive nitrogen gases we are observing in our passive diffusion samples.

One significant shortfall of this work was an inability to quantify NO from the difference between nitrite detected in our NO_x and NO_2 passive samples. This was due to an unforeseen matrix effect from the degradation of the cellulose filters in the extraction solution despite having made significant analytical inroads in the detection of nitrite in the presence of the NOx passive sample matrix. These degradation products further confounded our ability to make an accurate measurement as they overloaded the ion chromatography system and we did not have a mechanism to account for the degree of analyte loss. Thus, we will implement the immediate removal of the cellulose reactive pad from the aqueous extracts after 60 minutes to reduce the quantity of interfering compounds that can be produced in our samples. We will also include an internal standard in the form of lithium bromide to track other losses in any of our passive diffusion samples. The lithium ion will be used in our analyses of NH_3 and the bromide ion in our analyses of nitrite from NO_x and NO_2 samples.

Finally, these results will be incorporated into a longer observational dataset to determine the total fluxes of gases between the biosphere and atmosphere. In the case of NH_3, we will collect soil samples from nearby the sampling locations at the three sites within each NL-BELT region on a quarterly basis to determine how dynamic the amount of NH_4^+ in the soils is and to capture any variability in soil pH as well. This will then allow us to model the amount of NH_3 deposition into the NL-BELT regions using our compensation point calculations. For NO_x, NO_2 and NO, we will use the long-term observational data to drive a vertically resolved chemical transport model in order to describe the potential net exchange processes of these gases.

6. Conclusions

The preliminary data collected through the Harris Centre Applied Research Fund has allowed us to establish remote monitoring infrastructure across the NL-BELT and to collect preliminary data toward understanding the potential transport, deposition, and exchange of N_r in these terrestrial environments. We have successfully made quantitative measurements of the N_r gases ammonia and nitrogen dioxide at levels of tens of parts per trillion, but were unable to quantify the amount of nitrogen monoxide. With these preliminary measurements in hand, it is now possible to refine and improve the quality and quantity of data collected by this infrastructure over a much longer timeframe to elucidate statistically robust seasonal and annual trends in the biogeochemical cycling processes of nitrogen in the NL-BELT forests.

Due to unexpected delays in the release of supporting funds from NL-DNR-CFSI through the former Office of Research Services at Memorial University, the deployment of static precipitation samplers occurred in October 2013 and automated precipitation samplers the following field season of 2014. Despite this limitation, robust automated precipitation samplers have been designed and deployed across NL-BELT from August to October 2014. Preliminary tests and sample collection have allowed us to estimate a lower limit of N_r wet deposition inputs to the NL-BELT forests, suggestive that greater inputs are seen in the southern region of Grand Codroy compared to the more northern region of Salmon River. We have also elucidated the necessity in utilizing antimicrobial agents in our samples to ensure robust sample preservation over the month-long collection periods.

References

1 Bobbink, R. *et al.* Global assessment of nitrogen deposition effects on terrestrial plant diversity: a synthesis. *Ecological Applications* **20**, 30-59 (2010).
2 De Frenne, P. *et al.* Latitudinal gradients as natural laboratories to infer species' responses to temperature. *Journal of Ecology* **101**, 784-795, doi:doi: 10.1111/1365-2745.12074 (2013).
3 Sokolov, A. P. *et al.* Consequences of Considering Carbon–Nitrogen Interactions on the Feedbacks between Climate and the Terrestrial Carbon Cycle. *Journal of Climate* **21**, 3776-3797 (2008).
4 Bonan, G. B. Forests and Climate Change: Forcings, Feedbacks, and the Climate Benefits of Forests. *Science* **320**, 1444-1449 (2008).
5 Ollinger, S. V. *et al.* Canopy nitrogen, carbon assimilation, and albedo in temperate and boreal forests: Functional relations and potential climate feedbacks. *PNAS* **105**, 19336-19341 (2008).
6 Zaehle, S. & Friedlingstein, P. Terrestrial Nitrogen Feedbacks May Accelerate Future Climate Change *Geophysical Research Letters* **37** (2010).
7 Gorham, E. Northern Peatlands: Role in the Carbon Cycle and Probable Responses to Climatic Warming. *Ecol Appl* **1**, 182, doi:doi:10.2307/1941811 (1991).
8 Hobbie, S. E., Schimel, J. P., Trumbore, S. E. & Randerson, J. R. Controls over carbon storage and turnover in high-latitude soils. *Glob Change Biol* **6**, 196-210, doi:doi:10.1046/j.1365-2486.2000.06021.x (2000).
9 Soja, A. J., Tchebakova, N. M. & French, N. Climate-induced boreal forest change: Predictions versus current observations. *Earth and Planetary Change* (2007).
10 Korhonen, J. F. J. *et al.* Nitrogen balance of a boreal Scots pine forest. *Biogeosciences* **10**, 1083-1095 (2013).
11 Li, J., Ziegler, S. E., Lane, C. S. & Billings, S. Legacies of native climate regime govern responses of boreal soil microbes to litter C:N and temperature. *Soil Biology and Biochemistry* **66**, 204-213 (2013).
12 Holtgrieve, G. W. *et al.* A coherent signature of anthropogenic nitrogen deposition to remote watersheds of the northern hemisphere. *Science* **334**, 1545-1548 (2011).
13 Sutton, M. A. in *Nature* Vol. 472 159-161 (Macmillan Publishers Ltd, 2011).
14 Ellis, R. A. *et al.* The influence of gas-particle partitioning and surface-atmosphere exchange on ammonia during BAQS-Met. *Atmospheric Chemistry and Physics* **11**, 133 - 145 (2011).
15 Flechard, C. R. *et al.* Dry deposition of reactive nitrogen to European ecosystems: a comparison of inferential models across the NitroEurope network. *Atmospheric Chemistry and Physics* **11**, 2703-2728 (2011).
16 Canfield, D. E., Glazer, A. N. & Falkowski, P. G. The evolution and future of Earth's nitrogen cycle. *Science* **330**, 192-197 (2010).
17 Sutton, M. A. *et al.* Challenges in quantifying biosphere-atmosphere exchange of nitrogen species. *Environmental Pollution* **150**, 125-139 (2007).
18 Lamarque, J.-F. *et al.* Multi-model mean nitrogen and sulfur deposition from the Atmospheric Chemistry and Climate Model Intercomparison Project (ACCMIP): evaluation of historical and projected future changes. *Atmospheric Chemistry and Physics* **13**, 7997-8018, doi:10.5194/acp-13-7997-2013 (2013).
19 Sanderson, M. G. *et al.* A multi-model study of the hemispheric transport and deposition of oxidised nitrogen. *Geophysical Research Letters* **35**, doi:10.1029/2008GL035389 (2008).

20 Cape, J. N., Cornell, S. E., Jickells, T. D. & Nemitz, E. Organic nitrogen in the atmosphere — Where does it come from? A review of sources and methods. *Atmospheric Research* **102**, 30-48 (2011).

21 Nyaga, J. M., Cramer, M. D. & Neff, J. C. Atmospheric nutrient deposition to the west coast of South Africa. *Atmospheric Environment* **Accepted 14/09/2013**, doi:doi: 10.1016/j.atmosenv.2013.09.021 (2013).

22 Elliott, E. M. *et al.* Dual nitrate isotopes in dry deposition: Utility for partitioning NO_x source contributions to landscape nitrogen deposition. *Journal of Geophysical Research* **114**, doi:10.1029/2008JG000889 (2009).

23 Altieri, K. E., Hastings, M. G., Gobel, A. R., Peters, A. J. & Sigman, D. M. Isotopic composition of rainwater nitrate at Bermuda: The influence of air mass source and chemistry in the marine boundary layer. *Journal of Geophysical Research* **118**, 1-13, doi:doi:10.1002/jgrd.50829 (2013).

24 Committee, E. S. A. B. I. N. Reactive nitrogen in the United States: An analysis of inputs, flows, consequence, and management options. 172 (United States Environmental Protection Agency, 2011).

25 Zhang, L. *et al.* Nitrogen deposition to the United States: distribution, sources, and processes. *Atmospheric Chemistry and Physics* **12**, 4539-4554, doi:10.5194/acp-12-4539-2012 (2012).

26 Zhang, L. *et al.* Dry deposition of individual nitrogen species at eight Canadian rural sites. *Journal of Geophysical Research* **114**, doi:10.1029/2008JD010640 (2009).

27 Zhang, L. *et al.* Measurements of reactive oxidized nitrogen at eight Canadian rural sites. *Atmospheric Environment* **42**, 8065-8078 (2008).

28 Finlayson-Pitts, B. J. & Pitts, J. N., Jr. *Chemistry of the upper and lower atmosphere: Theory, experiments, and applications*. (Academic Press, 2000).

29 Seinfeld, J. H. & Pandis, S. N. in *Atmospheric Chemistry and Physics: From Air Pollution to Climate Change* Ch. 7, 299 (John Wiley & Sons, Inc., 2006).

30 Singh, H. B. *et al.* Summertime distribution of PAN and other reactive nitrogen species in the northern high-latitude atmosphere of eastern Canada. *Journal of Geophysical Research* **99**, 1821-1835 (1994).

31 Singh, H. B. *et al.* Reactive nitrogen distribution and partitioning in the North American troposphere and lowermost stratosphere. *Journal of Geophysical Research* **112**, D12S04, doi:10.1029/2006JD007664 (2007).

32 Fischer, E. V. *et al.* Atmospheric peroxyacetyl nitrate (PAN): a global budget and source attribution. *Atmospheric Chemistry and Physics* **14**, 2679-2698, doi:10.5194/acp-14-2679-2014 (2014).

33 VandenBoer, T. C. *et al.* Ion chromatographic separation and quantitation of alkyl methylamines and ethylamines in atmospheric gas and particulate matter using preconcentration and suppressed conductivity detection. *J Chromatogr A*, 12, doi:10.1016/j.chroma.2012.06.062 (2012).

Appendices

Appendix ‘A’

Summary for Centre for Forest Science and Innovation

Synopsis

All hardware has been designed, tested, and deployed for data collection to the NL-BELT field sites. Sample collection and analyses were performed with static precipitation samplers from October 2013 – October 2014, while automated samplers have undergone field testing and collected preliminary precipitation samples between August and October 2014. Total nitrogen (N) fluxes to two of the NL-BELT field locations span 1.30±0.71 to 2.7±0.97 kilograms per hectare per year, with significantly more N deposited in the south (Grand Codroy) than further north (Salmon River). These were the only two sites for which annual deposition values were possible to calculate. We caution that uncertainty in sample collection procedures may negate the significance between sites found in this preliminary work since in excess of 100 % variation was found between replicate precipitation samples. Seasonal trends in deposition were not consistent across the sites and times of maximum deposition differed between the locations. The design and preliminary results from the project have been disseminated at provincial, national, and international conferences. In addition, three highly qualified personnel have been trained in precipitation sampler design and construction, sample collection, and sample analysis. A summer student has been trained in the detection and quantitation of key tracers, amino sugars, in tracking the movement of reactive-N in NL-BELT soils affected by N inputs. A field technician has been trained in the collection and processing of samples across the NL-BELT sites. A PhD student, John MacInnis, from collaborator Dr. Cora Young in the Department of Chemistry at MUN has been trained on all components of the precipitation collection program and been awarded a Northern Scientific Training Program award to continue this work under my supervision for the 2014 NL-BELT field season. Collaboration with Dr. Cora Young has led to a new collaboration with Dr. Amila De Silva at the Canadian Centre for Inland Waters facility of Environment Canada to track the transport of persistent pollutants to remote regions, which can be determined from the same samples used for the reactive-N analysis. This work and the established collaborations currently represent three publications in preparation and the long term datasets of the continuing project will generate more robust measures of the interannual variability and seasonal trends in atmospheric reactive N transport to these boreal ecosystems.

Timeline of work completed

October 2013 – January 2014

- Design, development and manufacture of prototype precipitation collection cluster custom electronics control board.
- Static precipitation samplers deployed to collect snow throughout winter. These collect all inputs from the atmosphere, not just rain when sensed by a sensor.
- Collaborator (Dr. Cora Young, Chemistry) graduate student (John MacInnis, PhD Student) trained on collection of static precipitation samples.

January 2014 – March 2014

- Testing of electronics control board functionality and durability, precipitation sensor, and energy consumption.
- Design and procurement of precipitation collection cluster hardware and weather resistant hardware.

March 2014 – May 2014

- PhD student John MacInnis earned a Northern Scientific Training Program award to continue training in field precipitation collection and analysis across the NL-BELT, facilitating construction of automated precipitation samplers and additional field work capacity through his assistance.
- Construction and testing of prototype precipitation collection cluster (6 units):
    - Integrated electronics control board, solar panels, sensors, and weather resistant collection hardware.
- Static precipitation samples collected and repairs made to compromised units.

June 2014

- Trained field technician in collection of field samples.
- Began training of summer student (Rachel Hems, B.Sc. Hons., University of Guelph) in detection of amino sugar marker compounds for N-cycling in soils impacted by precipitation reactive-N inputs.
- Trained students Rachel Hems and John MacInnis in the construction and testing of precipitation collection clusters.
- Replication and testing of first precipitation collection cluster (6 additional units).
- Static precipitation samples collected and repairs made to compromised units.

July 2014

- Deployment of precipitation collection clusters at NL-BELT test sites located at Grand Codroy (Doyles, NL) and Pynn's Brook.
- Static precipitation samples collected and repairs made to compromised units.

August – October 2014

- Production and testing of remaining precipitation collection clusters (12 additional units).
- Confirmed functionality of test site precipitation collectors and optimized collection strategy.
- Deployment of precipitation collection clusters at remaining NL-BELT sites located at Eagle River (Cartwright, NL) and Salmon River (Main Brook, NL).
- Trained student John MacInnis in field precipitation collection of automated precipitation samplers.
- Preliminary precipitation samples collected for analysis from automated samplers and static precipitation sampling continued.

- Summer student Rachel Hems' amino sugar project completed and data in preparation for publication.

September – December 2014

- Development of precipitation sample processing methods for analysis of total N and reactive-N.
- Collection and analysis of collected static and automated precipitation samples for reactive nitrogen wet deposition fluxes.

Matching Funds and Knowledge Mobilization

- All passive gas sampling hardware has been installed and samples have been collected on a monthly basis from May – November since May 2013, with samples collected over the duration of the 2013-2014 winter from direct and in-kind support provided by the Harris Centre at MUN and the Canadian Forest Service located in Corner Brook, respectively.
- Passive gas sampling results and precipitation sampler design, upon which this precipitation data will build, have been or are scheduled for presentation at the following conferences provincially, nationally, and internationally:
    - Canadian Institute of Forestry, Corner Brook, NL; September 2013
    - Canadian Society for Chemistry Annual Meeting, Vancouver, BC; June 2014
    - American Geophysical Union Fall Meeting, San Francisco, CA; December 2013
    - American Chemical Society Spring Meeting, Denver, CO; March 2015
- Collaboration with Dr. Cora Young has led to a new collaboration with Dr. Amila De Silva at the Canadian Centre for Inland Waters facility of Environment Canada to track the transport of persistent pollutants to remote regions, which facilitates assistance with sample collection. These additional measurements can be made using the same samples that are collected for reactive-N.
- All data collected from this project is shared amongst the international collaborators of the NL-BELT project and will be made available through GeoKIRV (Geospatial Knowledge Identification, Retrieval, and Visualization).

Budget Summary

Date	Transaction description	Amount	Notes
30-Sep-13	Revenue - Govt of NL	16,000.00	
31-Mar-13	Salary - Vandenboer	-1,292.20	
10-Dec-13	Cdn Tire	-548.20	
24-Jan-14	Rona Chester Dawe	-208.13	
24-Jan-14	Dongguan Tsiny Moter Industrial Co.	-317.20	
24-Jan-14	The Home Depot	-45.89	
1-Mar-14	Allied Electronics Inc	-1,661.67	
1-Mar-14	Allied Electronics Inc	-2,369.03	
26-Mar-14	Cdn Tire	-3.45	
26-Mar-14	Cdn Tire	-206.12	
15-Jan-14	Customs and duties	-0.82	
15-Jan-14	Courier/freight charges	-10.96	
21-Aug-14	PDF pay	-2,707.84	
21-Aug-14	CPP	-1,982.11	JV in progress to move $1,896.67 to other payroll accounts
21-Aug-14	EI	-119.96	
21-Aug-14	Group health	-19.00	
21-Aug-14	WCC	-23.25	
21-Aug-14	Dental	-10.50	
21-Aug-14	Basic Life Insurance	-3.52	
21-Aug-14	Basic AD & D	-0.18	
21-Aug-14	Vacation Pay	-77.92	
21-May-14	Cdn Tire	-833.29	
2-Jul-14	Tech services	-1,450.07	
23-Jul-14	Conferences/seminars - CDN	-2,108.69	
	Balance as of Jan 9, 2015	0.00	

Detailed Project Summary

1. Sampling Locations and Strategy

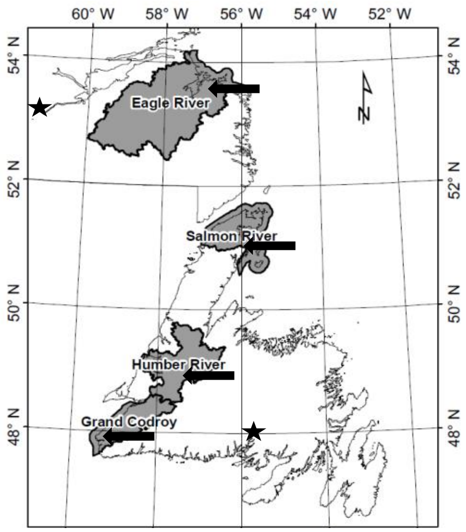


Figure 1. *Locations of CAPMoN sites (stars) for atmospheric inputs and NL-BELT instrumented sites (arrows) in Atlantic Canada. Shaded areas represent National Hydrological Network units associated with NL-BELT.*

The sampling locations for this project are at preexisting instrumented field sites within the Newfoundland and Labrador Boreal Ecosystem Latitudinal Transect (NL-BELT, Figure 1). The measurements made at the instrumented sites are essential to appropriately modeling the biosphere-atmosphere exchange processes from the long-term dataset collected for reactive nitrogen gases (Figure 2). The strategy for sample collection was monthly site visits from the finish of snow melt to the beginning of snow fall at the Eagle River site in Labrador, or roughly May through October, for both reactive nitrogen gases and precipitation. Triplicate collections allow for statistically significant analyses of collected and deposited nitrogenous compounds at each location (Figure 2).

Nested triplicates of the Ogawa© passive gas samplers were deployed within each NL-BELT region to capture potential heterogeneity across the instrumented sites since microbial processes are suspected to govern the presence of these gases, affected primarily by the local temperature and water availability (Figure 2a). The passive gas samplers were installed on standing dead wood located in clearings of at least 10 m in diameter within the forest canopy or on the edge of the experimental stand at a height of about 8 m (Figure 2b). The solar-powered, automated precipitation samplers were deployed in triplicate at the edge of the instrumented forest stands in order to capture direct inputs of reactive nitrogen in rain water in addition to that captured by the canopy, or leached from it, in the form of through-fall (Figure 2c).

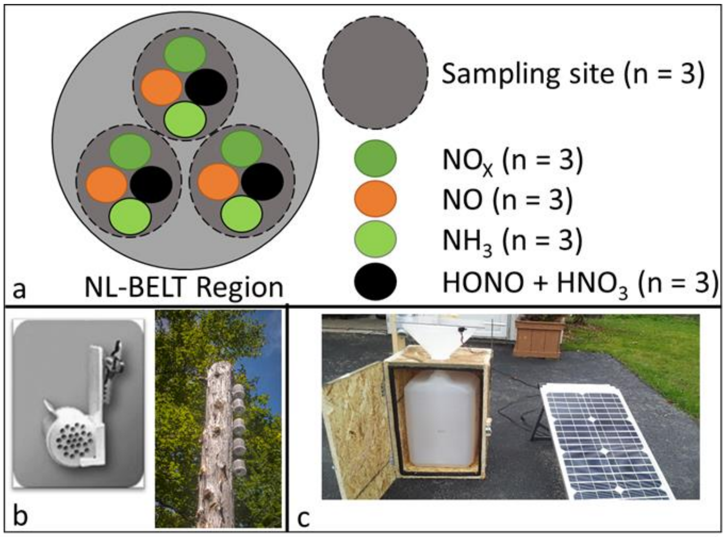


Figure 2. *Installed in each NL-BELT region with statistical replication within and between sampling sites (a) are Ogawa® passive diffusion samplers for reactive gases (b) and custom-built automated precipitation collectors, (c) for through-fall (n = 3) and open fall (n = 3) rain samples.*

2. Sampling Apparatus Details

a. Static Precipitation Samplers

Figure 3. Static precipitation sampler field setup.

The static precipitation samplers were developed to enable snowfall reactive-N inputs during the winter seasons as the automated units do not have the capacity to melt snow. These samplers are constructed from PVC sewer pipe (2.88 m long by 0.23 m diameter) stood upright with a liquid-tight end cap installed on the bottom. The top is left open to the atmosphere to continuously collect precipitation (Figure 3). The pipes are tethered to prevent tipping and their contents are collected monthly, provided the sites are accessible, using a portable 12 V battery-operated pump connected to tubing inserted from the top of the pipe. Any debris or other residues are removed between sampling periods by rinsing with ultrapure deionized water. Field blanks are collected from these washings to capture and account for any potential contamination effects between months.

Static precipitation samplers were installed at all NL-BELT field sites in October 2013 and collected monthly so long as sample and equipment integrity were determined to be sound (Table 1).

Table 1. Summary of samples collected using the static precipitation samplers.

Sample ID	**Date (mmm-yy)**	**Volume Total (mL)**	**Uncertainty (mL)**	**Notes**
GC	May-14	39615	3128	
GC	Jun-14	2600	12	Leaking cap
GC	Jul-14	0	0	Dead bat in sample
GC	Aug-14	0	0	Sampler repair and reinstall
GC	Sept-14	1762	10	
GC	Oct-14	2658	13	
HR	May-14	0	0	Sampler installation
HR	Jun-14	5423	16	
HR	Aug-14	2310	11	Leaking cap
SR	May-14	1517	10	
SR	Jun-14	0	0	4 trees blown down on sampler
SR	Aug-14	2816	14	
SR	Sept-14	0	0	Pump apparatus failure
SR	Oct-14	3555	14	
ER	Jun-14	1895	11	Leaking cap
ER	Aug-14	2216	13	Leaking cap

b. Automated Precipitation Samplers

Automation of precipitation collection was proposed to be achieved by the construction of in-house electronics (Figure 4a-b) using a heated precipitation sensor (Figure 4c) and a cluster of six (6) collection vessels with lids that open and close synchronously (Figure 4d-e). The heated precipitation sensor allows the sensor to dry itself after a precipitation event and thus detects only active precipitation and is not strongly affected by fog and dew. The precipitation sensor operates based on a conductivity measurement and so the precipitation samplers only collect polluted precipitation, characteristic of that in which the majority of nutrients are deposited, including reactive-N. The benefit of this sampling approach is that very clean precipitation is not collected, resulting in dilution of the target analytes for later analysis, facilitating easier detection and quantification. The six automated precipitation samplers are split into triplicate measurements that capture direct inputs of N from precipitation (open-fall) and altered N-inputs from the transit of precipitation through the forest canopy (through-fall) with statistically meaningful values. The entire assembly is powered by a 12 V deep-cycle marine battery that is topped up by a solar panel to maintain functional precipitation collection over a period of 4 weeks (Figure 4b and 4d).

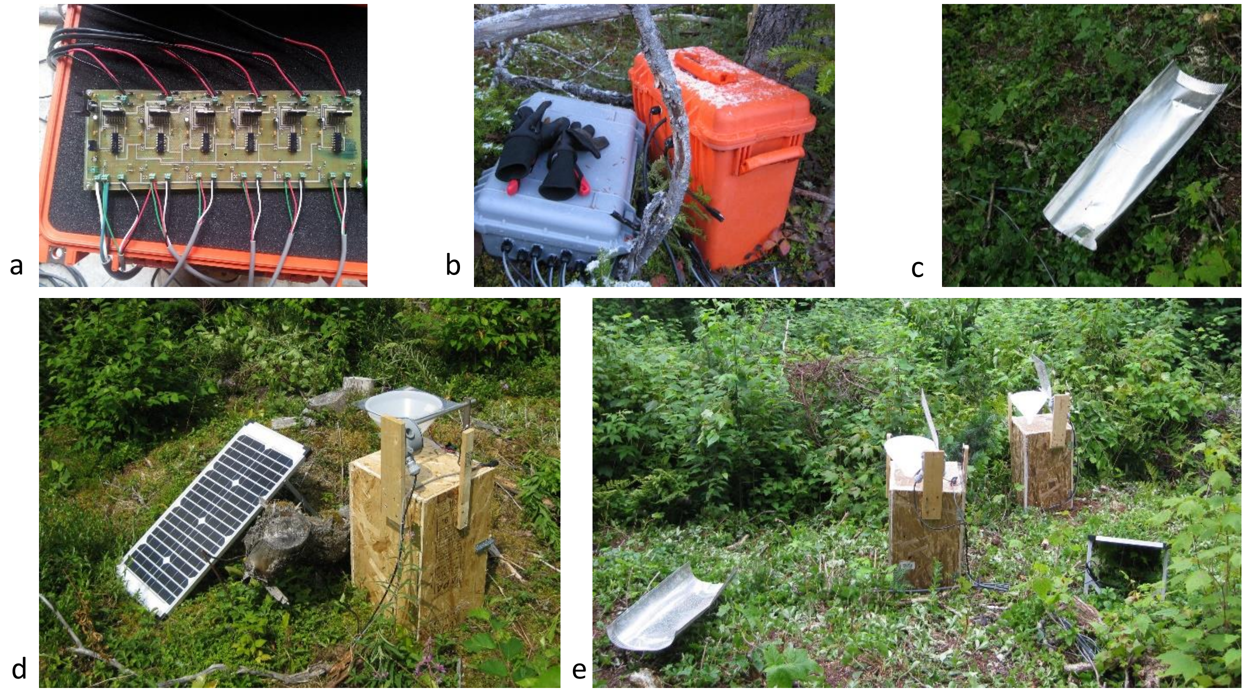


Figure 4. Automated precipitation samplers. In-house electronics design **(a)** were deployed in environmentally protected cases **(b)** to interface with the precipitation sensor **(c)** and solar panel **(d)** such that precipitation events **(e)** could be captured over a period of up to a month.

The automated precipitation samplers are deployed at the same sites across NL-BELT as the static precipitation samplers, except in the Humber River region where the automated samplers are collocated with percolated soil water collectors (lysimeters) to understand further the potential impact that precipitation reactive-N inputs have on boreal forest soil processes. The alternate site is located at

Pynn's Brook, east of Pasadena, NL and is instrumented for supporting measurements similarly to the other NL-BELT sites. Twelve (12) automated precipitation samplers were installed at the Pynn's Brook (PB) and Grand Codroy (GC) field sites in August 2014 for preliminary testing of the sensor clusters (e.g. robust, reproducible, and reliable). During this period, we were able to determine an effective sensor slope to maximize the recovered volume of precipitation (e.g. steep enough that water didn't pool on the sensor, but shallow enough that the water droplets didn't shoot across it without being sensed) .The remaining twelve automated samplers were deployed in Salmon River (SR) and Eagle River (ER) in September 2014. Samples were collected until the end of October 2014 (Table 2). The automated samplers and supporting hardware were collected and stored for the winter at the end of October 2014 to prevent damage from ice and snow.

The effects of microbial activity on the recovery of reactive-N in the precipitation samples was tested by leaving them alive or killing them with a biocide and subsequently comparing different metrics in the lab to show the potential for bias. In the case of biocide use, two options were available in mercuric chloride ($HgCl_2$) and thymol. These allowed us adaptability in our subsequent analyses of total dissolved nitrogen (TDN) and inorganic-N as nitrate and ammonium. In addition, the use of $HgCl_2$ allowed us to measure dissolved organic carbon (DOC) in support of ongoing C-budget closure for the NL-BELT field sites.

Table 2. Summary of samples collected using the automated precipitation samplers

Sample ID	Date (mmm-yy)	Alive/Thymol/ $HgCl_2$	Volume Total (mL)	Uncertainty (mL)	Notes
GC_OF1	Aug-14	Alive	395	5	Sensor slope too steep.
GC_OF2	Aug-14	Alive	253	5	Small volumes collected for August at GC
GC_OF1	Sept-14	Alive	1004	9	
GC_OF2	Sept-14	Alive	773	7	
GC_OF2	Oct-14	Alive	0	0	
GC_TF1	Aug-14	Alive	244	5	
GC_TF2+3	Aug-14	Alive	49	5	
GC_TF1	Sept-14	Alive	298	5	
GC_TF2	Sept-14	Alive	435	7	
GC_TF3	Oct-14	Alive	126	5	
PB_OF1	Aug-14	Alive	792	9	
PB_OF2	Aug-14	Alive	602	7	
PB_OF3	Aug-14	Alive	1783	12	
PB_OF1	Sept-14	Alive	119	5	
PB_OF2	Sept-14	Alive	577	7	
PB_OF2	Oct-14	Alive	527	7	
PB_TF1	Aug-14	Alive	319	5	
PB_TF2	Aug-14	Alive	336	7	
PB_TF3	Aug-14	Alive	542	7	
PB_TF1	Sept-14	Alive	15	5	
PB_TF2	Sept-14	Alive	305	5	
PB_TF1	Oct-14	Alive	215	5	

SR_OF1	Sept-14	Alive	264	5	Sampler lids on three units disconnected from motor and fully open for unknown period of time
SR_OF2	Sept-14	Alive	1616	10	
SR_TF1+2	Sept-14	Alive	34	5	
SR_OF1	Oct-14	Alive	544	7	Lid open for undetermined amount of time
SR_TF2	Oct-14	Alive	0	0	
ER_OF1	Oct-14	Alive	0	7	
ER_OF3	Oct-14	Alive	1341	10	Battery dead after 2 months of deployment
ER_TF1	Oct-14	Alive	205	7	
ER_TF3	Oct-14	Alive	2095	12	
GC_OF3	Sept-14	HgCl2	1035	9	
GC_TF3	Sept-14	HgCl2	567	7	
GC_OF	Oct-14	HgCl2	255	5	
GC_TF	Oct-14	HgCl2	144	5	
PB_OF3	Sept-14	HgCl2	155	5	
PB_TF3	Sept-14	HgCl2	6	5	
PB_OF	Oct-14	HgCl2	45	5	
PB_TF	Oct-14	HgCl2	0	5	
SR_OF3	Sept-14	HgCl2	585	7	
SR_TF3	Sept-14	HgCl2	645	5	
SR_OF	Oct-14	HgCl2	350	5	
SR_TF	Oct-14	HgCl2	220	5	
ER_OF	Oct-14	HgCl2	260	5	
ER_TF	Oct-14	HgCl2	95	5	
GC_OF	Oct-14	Thymol	260	5	
GC_TF	Oct-14	Thymol	44	5	
PB_OF	Oct-14	Thymol	35	5	
PB_TF	Oct-14	Thymol	23	5	
SR_OF	Oct-14	Thymol	460	5	
SR_TF	Oct-14	Thymol	45	5	

3. Precipitation Sampler N-Deposition Loads

Sample Analysis

Samples collected from both the static (Table 3) and automated (Table 4) precipitation samplers were refrigerated at 4 °C until required, filtered (0.45 µm, polyethersulfone), volume measured, and then analyzed for total dissolved nitrogen (TDN). Analyses of TDN were performed using a Shimadzhu TOC-vcsh instrument outfitted with a nitrogen analyzer and autosampler. The concentrations of total nitrogen were determined using calibration standards of glutamic acid (as N) spanning from 0 to 2 ppm (equivalent to units of: mg L^{-1}) and triplicate analyses of samples. Glutamic acid is an amino acid residue and used here as a proxy for organic nitrogen compounds. To ensure reasonable quantification of organic nitrogen check standards of EDTA (ethylenediaminetetraacetic acid) were also run at regular intervals during analysis. The method accuracy determined to be within 2 % of the check standard true values with a precision of ±7 %. Apparatus and method blanks accounted for carryover and background nitrogen in the sampling apparatus and were generally below the instrument detection limits or found to be negligible compared to the nitrogen contents of the precipitation samples collected. Speciation of the inorganic components (nitrate, NO_3^-; and ammonium, NH_4^+) of the precipitation samples is ongoing as the complexity of the precipitation requires significant additional analytical work to resolve and quantify.

Calculations of Deposition Flux and Associated Error

Where the total sample volume spanned multiple storage containers, uncertainty in the measured TDN was found by analyzing an aliquot from each container. In the remainder of the samples, where the volume was smaller than the storage container volume, the standard variation in triplicate analysis of the sample was used. Determination of the total nitrogen collected in a given sample, in micrograms, was found by dividing the concentration in ppm by the total volume of the sample, in millilitres, with the uncertainty propagated using the sum of squares approach. The total deposited flux of nitrogen and its associated error, in kilograms per hectare, was determined from the total nitrogen collected over the surface area of the precipitation sampler.

Table 3. Total dissolved nitrogen (TDN) measured in static precipitation samples in parts per million concentration (ppm), total mass of nitrogen deposited per sample in micrograms (TDN as N), and the deposition flux of nitrogen (N deposition), along with their calculated uncertainties.

Sample ID	Date (mmm-yy)	TDN (ppm)	TDN Uncertainty (ppm)	TDN as N (ug)	TDN as N Uncertainty (ug)	N deposition (kg/ha)	N deposition uncertainty (kg/ha)	Duration (days)
GC	May-14	0.022	0.004	867	161	0.211	0.043	212
GC	Jun-14	0.731		1899	9	0.463	0.041	32
GC	Jul-14							
GC	Aug-14							
GC	Sept-14	0.403	0.221	710	389	0.173	0.096	27
GC	Oct-14	0.197	0.010	524	26	0.128	0.013	30
HR	May-14							
HR	Jun-14	1.282		6952	20	1.694	0.152	48
HR	Aug-14	0.631	0.089	1459	205	0.355	0.059	30
SR	May-14	0.020	0.005	30	8	0.007	0.002	211
SR	Jun-14							
SR	Aug-14	0.147	0.018	415	50	0.101	0.015	22
SR	Sept-14							
SR	Oct-14	0.392	0.017	1393	60	0.339	0.034	30
ER	Jun-14	0.080		152	1	0.037	0.003	245
ER	Aug-14	0.163	0.004	362	10	0.088	0.008	62

Table 4. Total dissolved nitrogen (TDN) measured in automated precipitation samples in parts per million concentration (ppm), total mass of nitrogen deposited per sample in micrograms (TDN as N), and the deposition flux of nitrogen (N deposition), along with their calculated uncertainties. Preservation of samples against microbial activity was accomplished by the addition of mercuric chloride ($HgCl_2$) for TDN analyses.

Sample ID	Date (mmm-yy)	Alive/ Thymol/ $HgCl_2$	TDN (ppm)	TDN Uncertainty (ppm)	TDN as N (ug)	TDN as N Uncertainty (ug)	N deposition (kg/ha)	N deposition uncertainty (kg/ha)	Duration (days)
GC_OF1	Aug-14	Alive	0.080		31.8	0.4	0.008	0.001	24
GC_OF2	Aug-14	Alive	0.141		35.5	0.7	0.009	0.001	24
GC_OF1	Sept-14	Alive	0.247		248.3	2.1	0.065	0.006	27
GC_OF2	Sept-14	Alive	0.398		307.3	2.8	0.081	0.008	27
GC_OF2	Oct-14	Alive							30
GC_TF1	Aug-14	Alive	0.377		92.0	1.9	0.024	0.002	24
GC_TF2+3	Aug-14	Alive	8.880		435.1	44.4	0.114	0.016	24
GC_TF1	Sept-14	Alive	0.271		80.6	1.4	0.021	0.002	27
GC_TF2	Sept-14	Alive	0.226		98.4	1.6	0.026	0.002	27
GC_TF3	Oct-14	Alive	0.497		62.6	2.5	0.016	0.002	30
PB_OF1	Aug-14	Alive	0.052	0.003	41.2	2.4	0.011	0.001	22
PB_OF2	Aug-14	Alive	0.062		37.4	0.4	0.010	0.001	22
PB_OF3	Aug-14	Alive	0.313	0.023	558.1	41.4	0.147	0.017	22
PB_OF1	Sept-14	Alive	0.116		13.8	0.6	0.004	0.000	26
PB_OF2	Sept-14	Alive	0.209		120.4	1.5	0.032	0.003	26
PB_OF2	Oct-14	Alive	0.063		32.9	0.4	0.009	0.001	31
PB_TF1	Aug-14	Alive	0.273		87.1	1.4	0.023	0.002	22
PB_TF2	Aug-14	Alive	1.571		527.9	11.1	0.139	0.013	22
PB_TF3	Aug-14	Alive	0.573		310.3	4.0	0.082	0.008	22
PB_TF1	Sept-14	Alive	0.434		6.5	2.2	0.002	0.001	26
PB_TF2	Sept-14	Alive	0.580		176.8	2.9	0.047	0.004	26
PB_TF1	Oct-14	Alive	0.511		109.9	2.6	0.029	0.003	31

SR_OF1	Sept-14	Alive	0.207		54.6	1.0	0.014	0.001	27
SR_OF2	Sept-14	Alive	0.118		190.0	1.2	0.050	0.005	27
SR_TF1+2	Sept-14	Alive							27
SR_OF1	Oct-14	Alive	0.479	0.142	260.8	77.1	0.069	0.021	30
SR_TF2	Oct-14	Alive							30
ER_OF1	Oct-14	Alive	2.866		730.8	20.3	0.192	0.019	57
ER_OF3	Oct-14	Alive	0.244		326.8	2.4	0.086	0.008	57
ER_TF1	Oct-14	Alive	1.405		288.0	9.9	0.076	0.008	57
ER_TF3	Oct-14	Alive	0.216	0.003	451.5	7.3	0.119	0.011	57
GC_OF3	Sept-14	HgCl2	0.588		608.8	5.1	0.160	0.015	27
GC_TF3	Sept-14	HgCl2	1.084		614.6	7.7	0.162	0.015	27
GC_OF	Oct-14	HgCl2	1.005		256.3	5.0	0.067	0.006	30
GC_TF	Oct-14	HgCl2	3.544		510.3	17.7	0.134	0.013	30
PB_OF3	Sept-14	HgCl2	0.567		87.9	2.8	0.023	0.002	26
PB_TF3	Sept-14	HgCl2	3.174		19.0	15.9	0.005	0.004	26
PB_OF	Oct-14	HgCl2	1.142		51.4	5.7	0.014	0.002	31
PB_TF	Oct-14	HgCl2	0.000		0.0				31
SR_OF3	Sept-14	HgCl2	0.793		464.1	5.6	0.122	0.011	27
SR_TF3	Sept-14	HgCl2	1.316		848.8	6.6	0.223	0.021	27
SR_OF	Oct-14	HgCl2	1.377		482.0	6.9	0.127	0.012	30
SR_TF	Oct-14	HgCl2	1.387		305.1	6.9	0.080	0.008	30
ER_OF	Oct-14	HgCl2	1.308		340.1	6.5	0.089	0.008	57
ER_TF	Oct-14	HgCl2	4.708		447.3	23.5	0.118	0.013	57

Annual Deposition Flux Estimates and Seasonal Trends

The combined static and automated precipitation sampler datasets for the Grand Codroy and Salmon River sites provided sufficient temporal coverage to calculate annual deposition loads with a minimum of estimation (Table 5, Figure 5). Over the course of sampling from October 2013 to October 2014, the combined datasets from the static and automated samplers were only missing input measurements for July and August 2014 in Grand Codroy and June and July 2014 in Salmon River. Where both static and automated inputs from openfall were available, the results were combined and the potential error propagated to capture the potential range of input values. The static samplers have the potential to collect reactive-N compounds from dry deposition in addition to wet deposition and therefore could lead to an overestimation of wet-deposited nitrogen inputs. Where data was not available from either sampling method, the average of the upper and lower limits from the rest of the dataset were used as reasonable limits on the actual deposition values. As such, the certainty in our estimated values is low, but provides the capacity for an annual deposition value to be determined. The determined annual deposition loads, upper and lower limits, uncertainty, and bias corrected load estimates are shown in Table 5. A description of the identified biases in our sampling methodology is presented in Section 4.

The annual flux measured, for Grand Codroy was 1.22±0.44 kg ha^{-1} yr^{-1} (n = 5, range: 0.78 – 1.65), with a corrected value of 2.7±0.97 kg ha^{-1} yr^{-1}. The highest deposition loading was observed in June 2014 at 0.46±0.04 kg ha^{-1}. The remainder of the observations were reasonably consistent near 0.2 kg ha^{-1}.

The annual flux measured, for Salmon River was 0.46±0.25 kg ha^{-1} yr^{-1} (n = 7, range: 0.21 – 0.73), with a corrected value of 1.3±0.71 kg ha^{-1} yr^{-1}. The highest deposition loading was observed in the static sampler in October 2014 at 0.34±0.03 kg ha^{-1}. The remainder of the observations were at or below 0.1 kg ha^{-1}.

These results suggest that there is a potential deposition gradient of reactive nitrogen with increasing latitude across the NL-BELT (two-tailed t-test; P=0.0033). However, given the number of issues encountered with the static precipitation samplers over this period (Table 1), we advise strong caution towards the strength of this conclusion. Until October 2014, it was not possible to be sure that the static samplers deployed across the NL-BELT were free of leaks. Thus, deposition loads at any of the field sites could be higher than we have reported and our estimates should be viewed as a conservative lower limit on the total reactive nitrogen inputs to the NL-BELT ecosystems, including both dry and wet deposition since the static samplers do not discriminate between these sources. The corrected values we have determined assume that microbial processing of rain samples in the static and automated sampling reservoirs is identical and this may not be the case. Future collections of precipitation across the NL-BELT will minimize these errors by uniformly killing microbes in the static and automated samplers with $HgCl_2$ to ensure no losses due to microbial processes between the time of deposition and collection.

Table 5. Summary of estimated annual deposition loads for October 2013-2014 from combined static and automated open-fall samples. From the assumptions made on potential inputs where measurements were not available, and calculated error in measurements, upper and lower limits on the deposition loads were determined as well as the total uncertainty in our estimates. Bias correction for these inputs was made based on the ratio of TDN between automated open-fall samples collected in each region that were killed with $HgCl_2$ or left alive (see Section 4).

		Annual Openfall Deposition Loads of TDN as N (kg/ha/yr)				
Sampling Site	Collection Period	Estimated	Upper Limit	Lower Limit	Uncertainty	Bias Corrected
Grand Codroy	10/2013 - 10/2014	1.22	1.65	0.78	0.44	2.7
Humber River	10/2013 - 10/2014	-	-	-	-	-
Salmon River	10/2013 - 10/2014	0.46	0.73	0.21	0.25	1.3
Eagle River	10/2013 - 10/2014	-	-	-	-	-

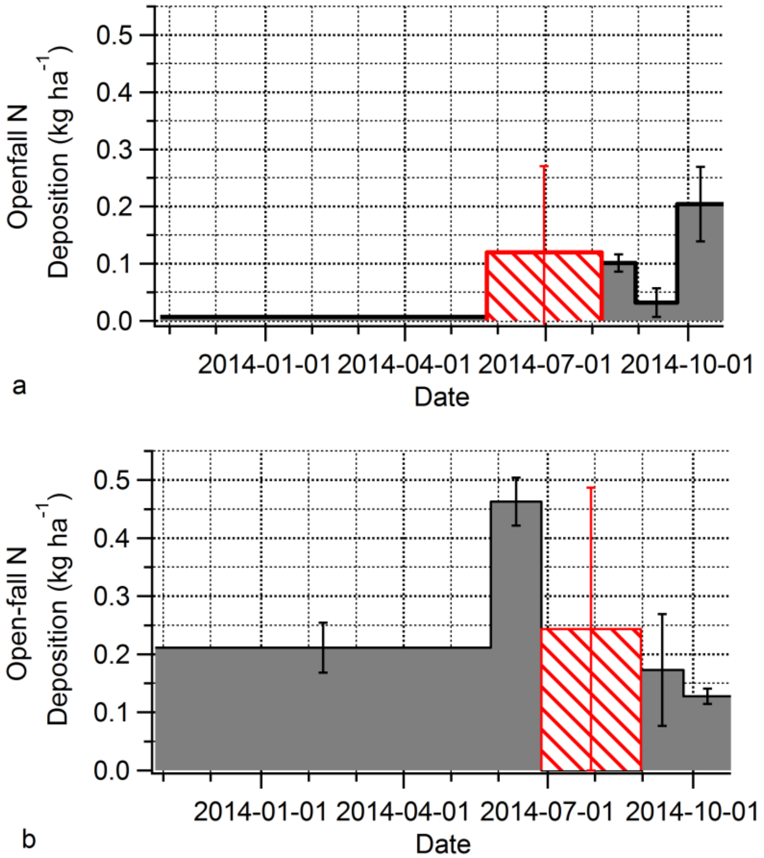


Figure 5. Seasonal trends and annual deposition loads of TDN to Salmon River (a) and Grand Codroy (b) sites of the NL-BELT. Direct measurements or combined observations from static and automated samplers are shown in black with grey fill (±1 s.d.). Estimates based on the observations for periods where no measurements were possible are denoted in red with diagonal line fill (±1 s.d.).

4. Precipitation Collection – Comparisons and Limitations

As discussed above, the role of microbial processing on our collected precipitation samples could have led to losses of measured TDN, biasing our annual deposition estimates lower than they may actually be. This was determined by comparing the TDN measured in living and HgCl2-killed samples collected with our automated samplers (Figure 6, Table 6) when it was possible to do so at each region within the NL-BELT. In addition to this bias, we identified a similar issue with our through-fall precipitation samples which also generally demonstrated TDN losses, especially in the warmer location of Grand Codroy. As mentioned above, our deposition estimates from the static precipitation samplers also includes reactive nitrogen derived from dry deposition processes. Our data show that static precipitation samplers consistently saw more TDN input than the automated samplers for the two samples we have collected.

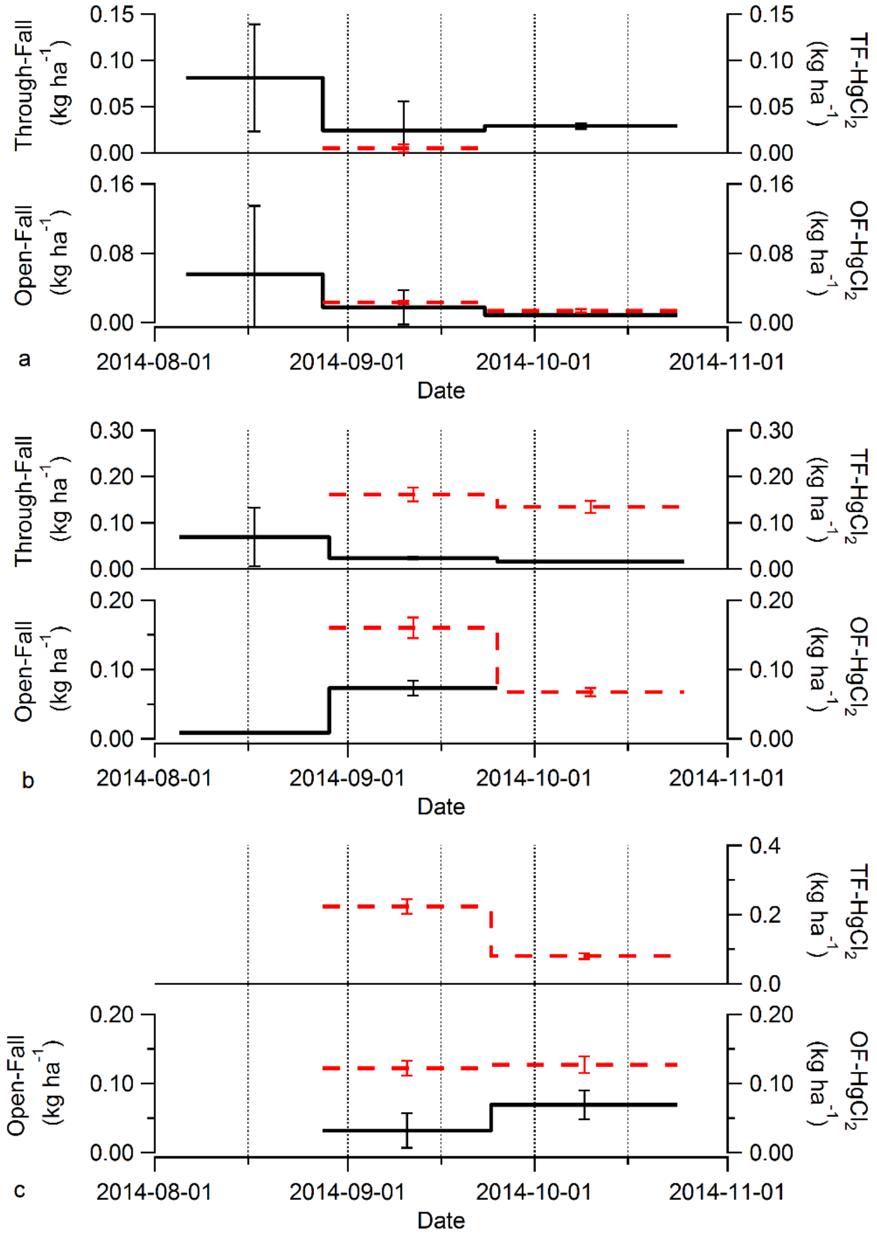


Figure 6. Comparison of NL-BELT automated open-fall (OF) and through-fall (TF) precipitation samples that were alive (black line) and killed with $HgCl_2$ (dashed red line) for Grand Codroy (a), Humber River (b), and Salmon River (c). Error bars represent ±1 s.d. in replicate samples where possible and standard deviation of the instrumental analysis where only a single sample of the given type was collected.

Table 6. Identified biases in comparisons of static and automated precipitation samplers.

		Precipitation Sampler Biases in TDN			***Alive***	***Killed***	***Autosampler Reproducibility in Alive TDN***		
Sampling Site	Mid-Date for Collection	Killed/Alive Openfall	Killed/Alive Throughfall	Static/Auto Openfall	Throughfall / Openfall	Throughfall / Openfall	Openfall (% RSD)	Throughfall (% RSD)	# Replicates (O,F)
Grand Codroy	17-08-2014	-	-	-	7.8	-	8	92	2,2
	11-09-2014	2.2	6.9	2.37	0.32	1	15	14	2,2
	10-10-2014	-	8	-	-	2	-	-	-
Humber River	17-08-2014	-	-	-	1.5	-	141	71	3,3
	10-09-2014	1.3	0.2	-	1.4	0.2	112	131	2,2
	08-10-2014	1.6	-	-	3.3	-	-	-	
Salmon River	10-09-2014	3.8	-	-	-	1.8	78	-	2
	08-10-2014	1.8	-	4.9	-	0.6	-	-	-
Eagle River	23-09-2014	0.6	1.2	-	0.7	1.3	54	31	2,2

We have not applied an additional correction to our deposition estimates based on this at this time. The reason for not doing so is because the automated samplers may not be collecting the entirety of the wet deposited reactive nitrogen due to known issues with the slope requirements of our rain sensors in addition to our marine battery lifetimes. These issues were identified during our preliminary tests at the end of the 2014 field season. To address this issue in future samples, we will be building static samplers identical to the automated samplers that are deployed in close proximity. This will allow us to better determine the total fraction of precipitation that our automated samplers are collecting. We will also be adding data logging devices to our electronic controls so that the detection and duration of collected rain events will be known for data analysis and interpretation.

We also investigated the difference between alive and killed ratios of throughfall to openfall precipitation samples. If the forest canopy collects nitrogenous compounds between rainfall events, then the precipitation collected is expected to be enhanced in these compounds. If the canopy takes up the reactive nitrogen deposited or the canopy microbes process it, then the throughfall precipitation collected could be depleted in these compounds. Our limited data on this phenomenon indicate a wide range of values depending on the site, whether the sample was killed or alive, and the collection period. Greater observation of this phenomenon with uniformly killed samples will allow inputs and canopy processing of wet deposited reactive nitrogen to be assessed. Much of this variability may be due to deposition heterogeneity in living samples and, at best, a single pairing of killed samples.

Replicate sample collections of TDN for living precipitation samples across all the NL-BELT regions were found to also range broadly, with relative standard deviations of 8 to 141 %. This suggests again that killing the precipitation samples is the best option moving forward to control for microbial processing and analyte losses and will allow us to capture the true precipitation input variability across the NL-BELT sites.

5. Supporting measurements of reactive-N gases and preliminary atmosphere-biosphere exchange estimates

Passive samplers deployed at the same sites as the static precipitation samplers across the NL-BELT were collected from May through October 2013 for the reactive nitrogen species ammonia (NH_3) and nitrogen dioxide (NO_2). There was no observed gradient in NH_3 across the NL-BELT, but some seasonal trends were observed with NH_3 declining from the summer into the fall (Figure 7). Using the concept of a compensation point (i.e. equilibrium exchange of NH_3 in soils with the overlying atmosphere), we found that NH_3 will deposit continuously to all of the NL-BELT ecosystems excepting those areas in Grand Codroy which have soil pH values reaching 6.0 (Figure 7, upper green dotted line).

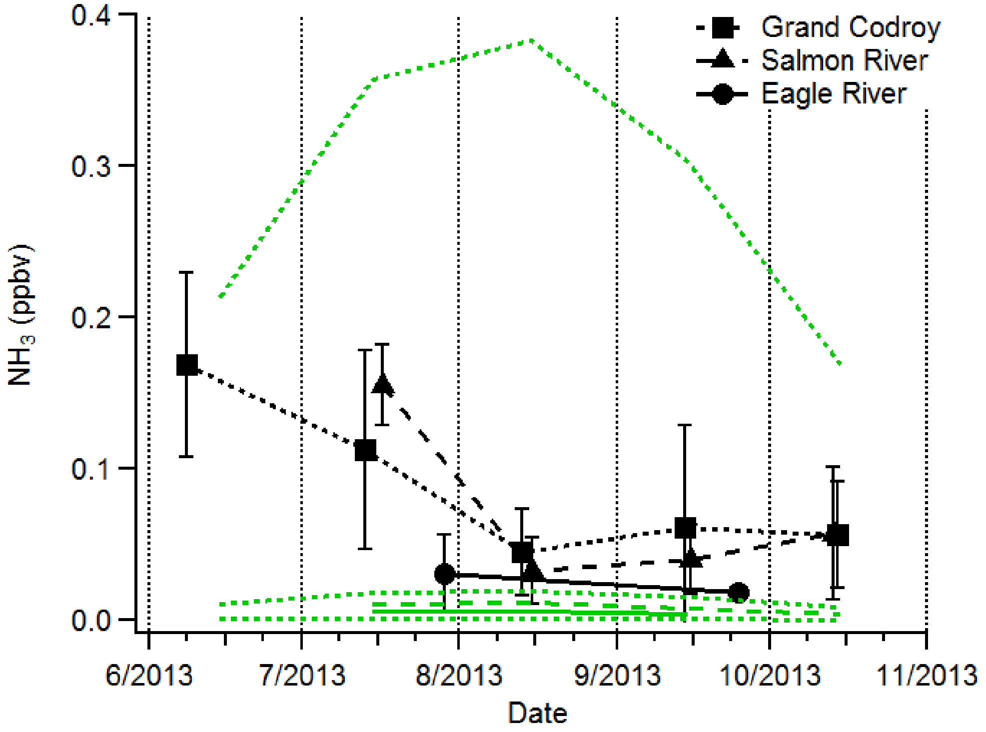


Figure 7. Seasonality and concentrations of passive samplers for $NH_{3(g)}$. Data points are plotted at the midpoint of the sampling period. Within site variability for each plot within the region and sampling period averages (± 1 s.d.) are shown for Eagle River, Salmon River, and Grand Codroy. When combined with the natural gradient in soil temperatures, and soil nutrient loadings and pH across the NL-BELT, the potential for biosphere-atmosphere exchange can be assessed by compensation point calculation (green lines). For Grand Codroy, the range of measured soil pH values was modeled (green dotted lines) for the upper (6.0), median (4.4), and lower limits (3.2). Measurements greater than the compensation point denote a potential for deposition.

Nitrogen dioxide measurements over the same period (Figure 8) show a clear gradient with atmospheric concentrations decreasing from south to north sampling locations. Previous aircraft studies over Newfoundland and Labrador have suggested that transport of nitrogen oxides, as NO_2 to such remote locations is driven primarily by the reservoir compound peroxyacetic nitric anhydride (PAN) that is exported from source regions (e.g. urban locations in the US northeast or along the St. Lawrence seaway). PAN is expected to be diluted as it undergoes transport away from source regions and is lost due to chemical and physical processes. These data suggest that this may be the case for the NL-BELT sites and show that collection of PAN data during sampling periods at these locations could have high utility.

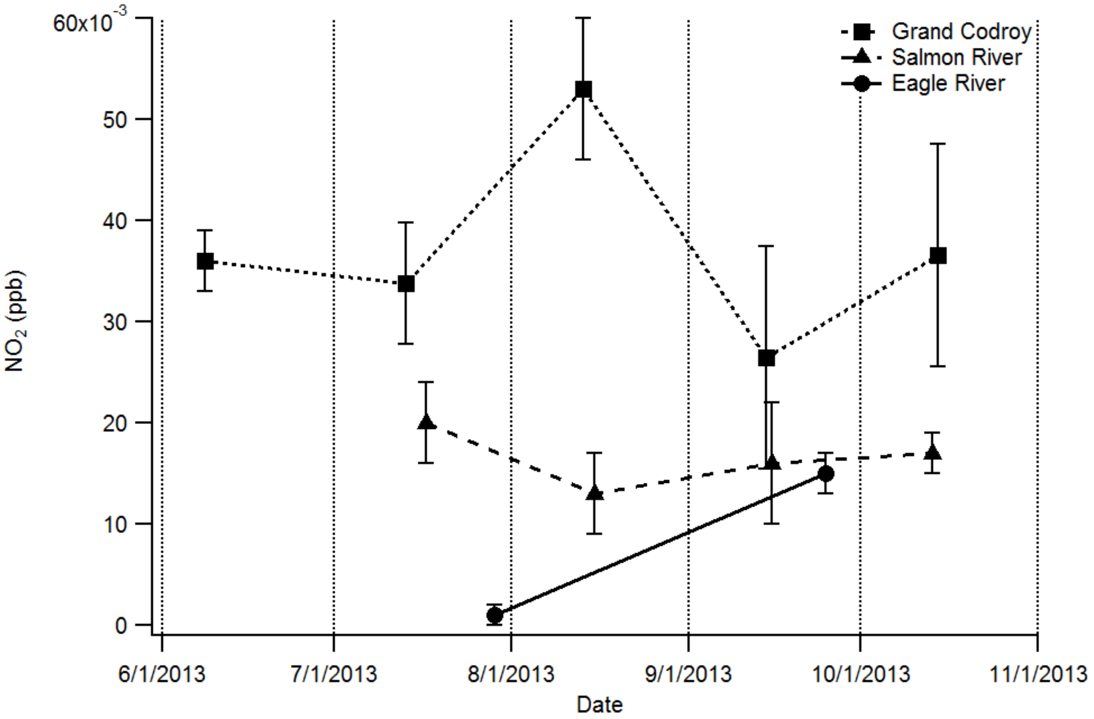


Figure 8. Seasonality and concentrations of $NO_{2(g)}$ across the NL-BELT showing a vertical gradient in atmospheric mixing ratios, which increase moving from the most northerly site to the most southerly. Data points are plotted at the midpoint of the sampling period. For each plot within the region sampling period averages (± 1 s.d.) are shown for Eagle River, Salmon River, and Grand Codroy.

Further interpretation of the NH_3 and NO_2 datasets requires explicit modeling of the vertical transport of these gases into the forest environment followed by exchange at the soil interface. This will be undertaken in collaboration with Dr. Jochen Stutz at the University of California, Los Angeles in the future such that total deposition loads from dry deposition can be compared to those found from wet deposition.