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RESEARCH ARTICLE

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Kev Points:

- Soils often do not freeze under snowpack on forested catchments
- Significant soil denitrification beneath a snowpack with nitrogen gas
 released.
- Represents an important (often unmonitored) nitrogen flux in catchment budgets

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Snow-covered soils produce N₂O that is lost from forested catchments

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Abstract The magnitude of net soil nitrous oxide (N_2O) production from a snow-covered catchment in a northern temperate forest was investigated. There was considerable net soil N_2O -N production and consumption through the snowpack, ranging from -6.6 to 26.2 g-N ha⁻¹ d⁻¹. There was no difference in net N_2O production among topographic positions despite significant variation in soil moisture, reduction-oxidation conditions, and pore water dissolved organic carbon and nitrate. Soil temperatures did not vary among topographic positions, suggesting that temperatures at or above the freezing point allow N_2O production to proceed under the snowpack. Redox conditions were lower at wetland positions compared to lowlands and uplands, suggesting that the biogeochemical pathway of N_2O production varies with topography. Over the entire nongrowing season, 1.5 kg of N_2O -N was exported to the atmosphere from the 6.33 ha catchment, representing 31% of the growing season N_2O -N production. These results suggest that winter is an active time for gaseous N production in these forests and that N_2O production under the snowpack represents an often unmonitored flux of N from catchments.

1. Introduction

There is a long history of atmospheric nitrogen (N) deposition in the northern temperate forest biome [Galloway et al., 2008]. Increased reactive N promotes N cycling processes [Aber et al., 1989; Stoddard, 1994], including nitrification and denitrification, which produce nitrous oxide (N_2O), a powerful greenhouse gas [Smith, 1997]. Production of N_2O is dependent on the reduction-oxidation (redox) potential of the soil environment [Gambrell and Patrick, 1978]. During the growing season, catchments with substantial wetland areas with low redox potential have greater potential for high rates of denitrification [Morse et al., 2015]. However, during the nongrowing season that includes the snow period, saturation of surface soils may expand areas of low redox potential to other parts of the catchment. This paper investigates net soil N_2O production underneath the snowpack to determine if it is an important loss of N from forested ecosystems.

Many northern temperate forests have a significant snow period throughout the nongrowing season, during which the fate of reactive N has often been ignored due to challenges of conducting research during this time. Climatic variability results in reduced or increased depth of snow cover, but these changes have not yet led to an increase in soil freezing days in Canadian landscapes [Henry, 2008]. Snow-covered soils often do not freeze in temperate forests because they are insulated by the overlying snowpack [Hardy et al., 2001], and biological processes in soils underneath the snowpack can contribute substantially to N cycling transformations [e.g., Brooks and Williams, 1999]. Climate change projections suggest an increase in winter precipitation and larger snowpacks in some areas of the northern limits of the temperate forest biome [Colombo et al., 2007], which means that winter soils may become warmer for longer in the future.

Topography underneath the snowpack may influence the physical and chemical controls on biological N transformations within forest soils. N_2O formed under the snowpack may be released as gas to the atmosphere [Groffman et al., 2009], consumed within the soil [Billings, 2008; Chapuis-Lardy et al., 2007; Schlesinger, 2013], or flushed into streams as dissolved N_2O [Heincke and Kaupenjohann, 1999], similar to the NO_3^- flushing hypothesis [Creed et al., 1996]. N_2O is an intermediate product of both nitrification and denitrification. Nitrification is the oxidation of NH_4^+ to NO_3^- , and as such requires redox potentials of above 100 mV [Kralova et al., 1992]. In contrast, denitrification is an anaerobic metabolic process, where NO_3^- is sequentially reduced to N_2O and finally to N_2 , and requires lower redox potentials (-100 to +100 mV [Kralova et al., 1992]). Both processes can persist in soils beneath the snowpack [Casson et al., 2014; Morse et al., 2015; Spoelstra et al., 2001], but the dominance of one process over another will depend on the availability of substrate and the appropriate redox conditions being present. There are also other pathways for



production of gaseous N products, including chemodenitrification [*Davidson*, 1992], fungal codentrification [*Laughlin and Stevens*, 2002], and dissimilatory nitrate reduction to ammonium [*Rutting et al.*, 2011]. The relative importance of these pathways will also vary depending on environmental conditions such as pH, redox, and the presence of other electron acceptors [*Burgin and Hamilton*, 2007].

Improved understanding of topographic controls on net soil N_2O production underneath the snowpack will contribute to improved catchment N budgets in temperate forests. Previous work investigated topographic controls on dissolved N export, including NO_3^- , ammonium, and dissolved organic nitrogen [*Creed and Beall*, 2009; *Creed and Band*, 1998a; *Creed and Band*, 1998b]. A related study investigated the physical and chemical controls on biological N transformations within forested catchments during the growing season and observed that precipitation triggered changes in redox conditions in wetland soils that resulted in net soil N_2O production events [*Enanga et al.*, 2016]. This study builds on this work by providing new insights into gaseous N exports during the nongrowing season including the snow period. We hypothesized that snow insulates soils and creates optimal redox conditions at the soil surface for N_2O production across the entire catchment as long as there is an adequate energy source, N pool, and nonfreezing soil temperatures. This hypothesis was tested in the forested landscape of the Great Lakes-St. Lawrence forest region at the northern edge of the temperate forest biome of North America.

2. Materials and Methods

2.1. Study Area

The Turkey Lakes Watershed is a long-term experimental watershed located approximately 50 km north of Sault Ste. Marie in the Algoma Highlands of central Ontario (47°03′N and 84°25′W). It comprises an area of 10.5 km² and has a continental climate that is strongly influenced by its proximity to Lake Superior [Jeffries et al., 1988]. Average total precipitation is 1189 mm, and average annual temperature is 4.6°C (1981–2010). A snowpack generally persists from November to April. Peak discharge occurs in April–May during spring snowmelt and again in October–November during autumn storms.

The watershed rests on bedrock primarily of greenstone with small outcrops of granite. The topographic relief of the watershed is 400 m from its summit at Batchawana Mountain (644 m above sea level (asl)) down to the outlet (244 m asl) at the mouth of the Batchawana River. There is a thin discontinuous glacial till deposited on the bedrock that ranges from less than 1 m at high elevations to 1–2 m at lower elevations and is occasionally as deep as 65 m in bedrock depressions. Podzolic soils are typically thin and undifferentiated near ridges, gradually thickening, differentiating, and increasing in organic content on topographic benches and near the stream with highly humified organic deposits in wetlands [Creed et al., 2002].

The watershed is covered by an uneven-aged mature hardwood forest. The canopy is dominated (90%) by sugar maple (*Acer saccharum* Marsh), with white pine (*Pinus strobus* L.), yellow birch (*Betula alleghaniensis* Britton), ironwood (*Ostrya virginiana* (Mill.) K. Koch), white spruce (*Picea glauca* Moench Voss.), and red oak (*Quercus rubra* L.) in upland areas and eastern white cedar (*Thuja occidentalis* L.), balsam fir (*Abies balsamea* (L.) Mill.), and black ash (*Fraxinus nigra* Marsh.) in wetland areas [*Wickware and Cowell*, 1985]. The forest has remained largely undisturbed since the 1950s, although several of the catchments (c31, c33, and c34) were experimentally harvested in 1996. Further details about the Turkey Lakes Watershed are provided in *Jeffries et al.* [1988].

2.2. Experimental Design

Located in the Turkey Lakes Watershed, catchment 38 (c38) is a catchment with a total area of 6.33 ha, including a single 1.58 ha wetland. A hillslope transect was established with sampling sites at four topographic positions: the raised center of the wetland (inner wetland (IW)), the outer ring of the wetland (outer wetland (OW)), the flat to gently sloped area at the base of the hillslope (lowlands (LOW)), and the steep area at the middle and top of the hillslope (uplands (UP); Figure 1). Each sampling site was instrumented for sampling of gaseous net soil N_2O production from the soil surface and through the snowpack. Sampling equipment for gas sampling under a snowpack was installed prior to snowpack formation. Samples were collected during the warmest hours of the day between 10:00 h and 14:00 h, daily during snowmelt periods, and every 2 weeks otherwise using different methods with and without a snowpack from October 2006 to May 2007. All instruments were accessed from boardwalks to prevent soil disturbance during sampling.



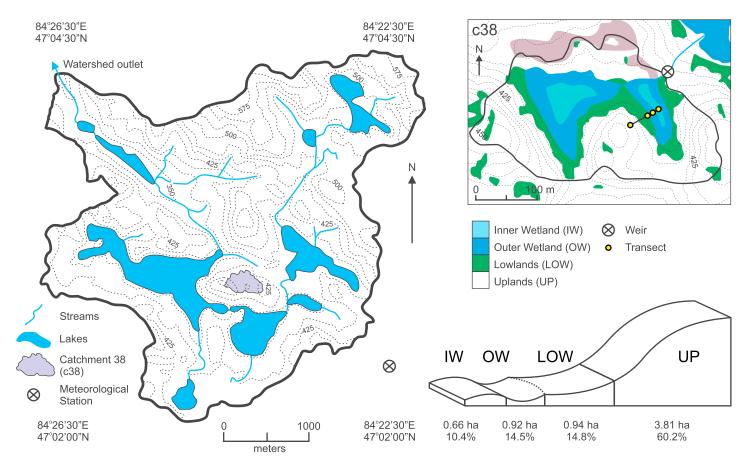


Figure 1. Map of the Turkey Lakes Watershed near Sault Ste. Marie, ON, Canada. (top right) Map of topographic positions within c38 and (bottom right) proportion of area of each topographic position within c38.

2.3. Data Collection

2.3.1. Catchment Hydrology

Air temperature and precipitation were continuously monitored at the Canadian Air and Precipitation Monitoring Network (CAPMON) station just outside the Turkey Lakes Watershed (47°02′06"N, 84°22′52"W) operated by Environment Canada since 1981. Catchment snow depth and density data were collected daily at each topographic position during snowmelt and every 2 weeks otherwise. Snow depth was measured after digging through the snowpack to expose the ground surface, and snow density was measured using a snow cutter (volume 78.5 cm³) at depths of 5, 10, 25, 50, 75, and 100 cm from the snowpack surface. Catchment water table depth was continuously monitored at the center of the wetland using a water-level logger (WT-HR Water Height Data Logger, TruTrack Inc., Christchurch, NZ). Catchment daily discharge data were continuously monitored using stream stage at a V notch weir (Figure 1).

2.3.2. Defining Catchment Topographic Positions

The topographic positions of catchment c38 (IW, OW, LOW, and UP) were defined using a 5 m digital elevation model (DEM) interpolated from light detection and ranging data (horizontal accuracy of 0.15 m under open canopy and 0.30 m under closed canopy). Wetland areas were defined using a probabilistic approach to determine the likelihood of a DEM grid cell being flat or in a depression [see Lindsay and Creed, 2006]. In wetlands, depth to bedrock varied considerably, with the wetland edge having depths to bedrock of about 70 cm, after which the depth to bedrock fell precipitously to over 5 m. A ground-based survey was used to define the portion of the wetland with peat depths less than 70 cm (the OW), with the rest of the wetland defined as the IW. LOW and UP topographic positions were classified using a method described by Webster et al. [2011]. Five topographic attributes were derived at each DEM grid cell location: (a) percent height relative to local pits and peaks calculated from the DEM with single pits removed; (b) percent height relative to local channels and divides from the DEM with depressions removed; (c) slope curvature and



(d) slope gradient both calculated on a 3×3 moving window; and (e) topographic wetness index [Beven and Kirkby, 1979], calculated using the infinite direction (Dinf) flow algorithm [Tarboton, 1997]. For each topographic position, topographic attributes were converted to fuzzy membership scores between 0 (no probability of being in a given position) and 1 (full probability) based on expert knowledge of scientists working in the watershed. Fuzzy scores were then combined in weighted attribute models to determine the probability of being a topographic position, and grid cells were assigned to LOW or UP positions according to the highest probability.

2.3.3. Soil Physical and Chemical Properties

Soil temperature, moisture, and redox potential were continuously monitored at a depth of 5 cm into the mineral soil profile. Soil temperature was measured using a thermocouple wire embedded into a 0.63 cm internal diameter copper tube with epoxy, soil moisture was measured using water content reflectometers (CS 616, Campbell Scientific Canada Corp., AB, Canada), and soil redox potential was determined by measuring the voltage between a platinum electrode and potassium chloride reference electrode. The thermocouples, water content reflectometers, and electrodes were wired to Campbell Scientific CR10X data loggers with AM16/32 relay multiplexors and powered by batteries that were charged by 30 W solar panels. Soil environmental data were recorded every 5 min and averaged every 30 min by data loggers. Water content reflectometer output was converted to volumetric water content based on calibration equations from the manufacturer for the UP and LOW positions and from *Yoshikawa et al.* [2004] for the wetland soils. Once the continuous measurements were averaged to daily measurements, 243 samples existed for each topographic position throughout the nongrowing season that included the snow period.

Soil pore water was collected between 5 and 10 cm to analyze dissolved organic carbon (DOC) and NO_3 . Soil pore water was collected using suction lysimeters (model 1900, Soil Moisture Corp., CA, USA) at least every 3 days during snowmelt periods and every 2 weeks otherwise. Triplicate shallow lysimeters were installed at each sampling site 5 to 10 cm in the soil. Prior to installation, the lysimeters were acid washed once using 10% hydrochloric acid and rinsed with distilled water 30 times and deionized water 10 times. After installation, the first liter of soil pore water flushed through the lysimeters was discarded. The lysimeters used in this study had been installed the previous year, so had ample time to equilibrate in the soil prior to sample collection. Lysimeters were prepared by applying a 50 kPa vacuum 24 h prior to collection. Samples were collected into acid-washed polycarbonate bottles. In the laboratory, each sample was filtered through a 0.45 μm polysulfone membrane filter (Supor 450, Pall Gelman Science, MI, USA) and were then frozen for transport and storage until analysis. Dissolved organic carbon was measured following acidification (pH < 2, 1 N HCl) using high-temperature catalytic oxidation and infrared detection (TOC 5000A, Shimadzu Corp., Tokyo, Japan). Nitrate concentrations were analyzed on a Technicon AutoAnalyzer II (Technicon Corp., NY, USA) using the cadmium reduction and ammonium concentrations from the sodium nitroprusside methods [Keeney and Nelson, 1982]. The water chemical analyses were conducted at the Water Chemistry Laboratory of the Great Lakes Forestry Centre in Sault Ste. Marie, Ontario.

2.3.4. Net Soil N₂O Production

Samples for N₂O production measurements were collected biweekly during nonmelt periods and daily during snowmelt at each position. When there was a snowpack, samples were taken at various depths from the soil surface within the snowpack [Filippa et al., 2009; Sommerfeld et al., 1996]. Three 1.25 m tall PVC tubes with six sampling ports attached to each at heights of 5, 10, 25, 50, 75, and 100 cm (measured from the bottom of the PVC tube) were installed vertically at the soil surface at each position (Figure 2). The PVC tubes were held in place by two reinforcement bars (rebars) that had been driven firmly into the ground and adjacent to the PVC tubes. The PVC tubes were secured onto the reinforcement bars using zip ties. One end of a 4 m of microbore polytetrafluoroethylene (PTFE) tubing was attached to each of the sampling ports with a gas-tight Swagelok© fitting. The other end of each PTFE tubing was fitted with a Swagelok© fitting with a septum injector nut to provide needle access. The exposed PTFE tubing was enclosed in copper pipes to prevent animals from damaging the tubing. The copper pipes were in turn enclosed in insulating foam. The septum end of the PTFE tubing was attached by bungee cord to a nearby tree at a height of approximately 1.5 m above ground to allow for sampling when the snowpack was greater than 1 m.

Gas samples were collected by first inserting a needle through both the septum and the Swagelok© fitting and into the tube. Five milliliter of gas was pulled from the tubing into a 10 mL syringe to purge the line of old air, and then 25 mL of snowpack air was pulled from the tubing into a 30 mL syringe and injected into

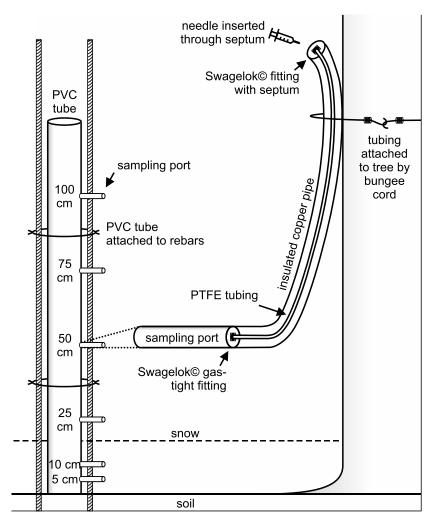


Figure 2. Gas sampling design via PVC tube under snowpack conditions.

a 12.1 mL evacuated exetainer. Each exetainer contained 0.1 g of magnesium perchlorate to absorb any moisture from the gas samples.

When there was no snowpack, N_2O was sampled using the static chamber method (Figure 3) [Livingston and Hutchinson, 1995]. Three round PVC collars (30 cm ID), each enclosing a soil area of $0.07\,\mathrm{m}^2$, were inserted level into the soil at each position. The volume of air in each collar over the soil surface was determined on each sampling date by measuring the distance from the top of the collar to the soil (or water in flooded collars). Vented PVC flux chambers (14 L) [Hutchinson and Livingston, 2001] with flexible tubing attached to sampling ports were placed on top of the collars. Gas samples were collected from valves attached to the end of the tubing for 1 h at 0, 15, 30, 45, and 60 min intervals using a 30 mL syringe. Prior to each sample being taken, the sampling tube was purged 5 times with 30 mL of air from the syringe to ensure mixing of the contents of the chamber. A 30 mL sample was then pulled, and a needle was fitted to the syringe. Five milliliter of air was flushed through the needle, and the remaining 25 mL sample was injected into a 12.1 mL evacuated exetainer. As with the exetainers for snowpack gas sampling, each contained 0.1 g of magnesium perchlorate to eliminate moisture from the gas samples.

 N_2O concentrations were determined using an SRI 8610C (SRI Instruments, CA, USA) gas chromatograph equipped with an electron capture detector with P5 (5% methane and 95% argon) or P10 (10% methane and 90% argon) used for the makeup gas and helium for the carrier gas. All samples were analyzed by the gas chromatograph within 2 months of the day they were collected. Samples were withdrawn from the exetainers by flushing a 10 mL syringe and needle with helium and allowing the sample to flow freely out



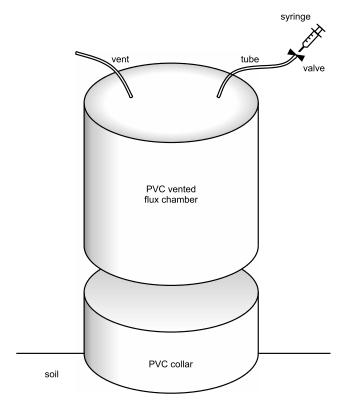


Figure 3. Chamber and collar gas sampling design when no snowpack occurs.

of the overpressurized exetainer. Five milliliter of each sample was manually injected into the gas chromatograph, where N2O concentrations were determined using a one-point calibration of 5 ppm N₂O standard:

$$c = \frac{A}{R} \tag{1}$$

where c is the N₂O concentration of the sample (ppm), A is the peak area from the gas chromatograph, and R is the response of the standard:

$$R = \frac{\mu}{a} \tag{2}$$

where μ is the mean peak area of the standards run before and after the sample and a is the actual concentration of the standard (5 ppm).

N₂O fluxes from the soil through the snowpack to the atmosphere were calculated using Fick's first law of diffusion:

$$J = -D_s \left(\frac{dc}{dz}\right) \tag{3}$$

where J is the flux of N_2O (μ mol cm⁻² s⁻¹), $\frac{dc}{dz}$ is the average verti-

cal concentration (c) gradient in snow (rate of change in concentration of N₂O with depth of snow (z)), and D_s (cm² s⁻¹) is the effective diffusion coefficient of gas in snow:

$$D_{s} = -\varphi \tau D_{0} \tag{4}$$

where τ is the tortuosity of the snow (equation (6)), D_0 is the diffusivity of gas in air (0.139 cm² s⁻¹ for N₂O) [Sommerfeld et al., 1993], and φ is the porosity of the snow:

$$\varphi = 1 - (\partial_{\mathsf{snow}}/\partial_{\mathsf{ice}}) \tag{5}$$

where δ_{snow} (g cm⁻³) is the density of snow determined using a snow cutter (volume 78.5 cm³) at 5, 10, 25, 50, 7 5, and 100 cm within the snowpack and δ_{ice} is the density of ice (0.9168 g cm⁻³).

$$\tau = \varphi^{1/3} \tag{6}$$

N₂O fluxes from nonsnow-covered soil were calculated by linear regression of the slope of increasing N₂O concentration in the chambers with time, scaled according to the total volume (collar and chamber) and area of the collar, and corrected for ambient pressure and temperature:

$$F_{\text{corr}} = \left(\frac{F \times V}{V_{\text{corr}} \times A}\right) \tag{7}$$

where F_{corr} is the final corrected N₂O flux (μ g m⁻² min⁻¹), F is the flux measured in the field (μ L L⁻¹ min⁻¹), Vis the volume of the chamber plus collar combined (L), A is the cross-sectional area of the collar (m²), and V_{corr} is the molar volume of N_2O corrected for temperature and pressure ($\mu L \mu mol^{-1}$):

$$V_{\text{corr}} = \frac{22.414 \times T}{273.15 \times P} \tag{8}$$

where T(K) and P(hPa) are the air temperature and pressure at the time of measurement.

 N_2O fluxes were converted to N equivalents (N_2O -N) by multiplying by $28/44\,\mathrm{g\,mol}^{-1}$ N_2O . All N_2O fluxes were used in the analysis, including 25 negative (consumptive) fluxes (-6.6 to -0.009 g-N ha⁻¹ d⁻¹)

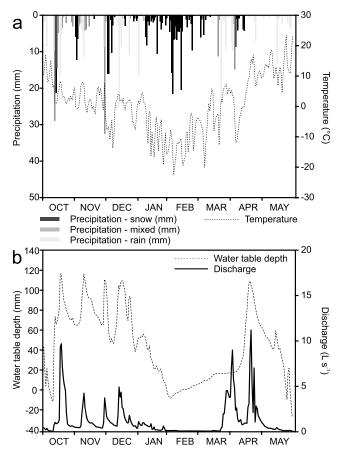


Figure 4. (a) Daily-averaged air temperature and precipitation in the Turkey Lakes Watershed and (b) daily water table depth at the center of the wetland in c38 and discharge at the outlet of c38 from October 2006 to May 2007.

out of 161 estimates. The fluxes were converted to production in units of q-N d⁻¹ by multiplying by the area of the respective topographic position within the catchment, as defined using the method from Webster et al. [2011] and described earlier in the methods (IW: 0.66 ha, OW: 0.92 ha, LOW: 0.94 ha, and UP: 3.81 ha). Net N₂O-N production for the nongrowing season at each topographic position was estimated by multiplying the median daily N2O-N production at the respective position by the number of days in the nongrowing season. Total N₂O-N production for the catchment was estimated by summing the net productions at each position. Net N₂O-N production was converted back to net N2O production by multiplying by 44/28.

The proportion of atmospheric N input that was net soil N_2O production was estimated using atmospheric N deposition measurements collected at the CAPMoN station (see *Sirois and Vet* [1988] for details on methods for atmospheric N deposition measurements; total organic N deposition was estimated as 15% of total N, based on average regional deposition of total organic N [*Dillon et al.*, 1991]).

2.4. Data Analyses

Differences in soil physical properties (temperature, moisture, and redox potential), chemical properties (DOC and NO_3^-), and net soil N_2O production among topographic positions were assessed using one-way analysis of variances on ranks, with Dunn's tests used to detect pairwise differences. Spearman's rank correlations were performed between soil physical and chemical properties and net soil N_2O production. All statistical analyses were performed using SigmaPlot 12 (Systat Software, San Jose, CA) and Microsoft Excel, and significance was assessed at p < 0.05.

3. Results

3.1. Catchment Hydrology

Air temperature steadily decreased from a high of approximately 15°C in October to a low of less than -20° C in February and rose to greater than 20° C in May (Figure 2a). The average monthly temperature from October 2006 to May 2007 was -2.2° C; this was within 1 standard deviation (1.2°C) of the mean average monthly temperature for the October to May period for 1981–2010 (-3.0° C). Precipitation during this period was a mix of snow, rain, and mixed precipitation, with fall storm events in October and November, regular snow starting in January and February, and minimal precipitation in April and May (Figure 2a). Total precipitation from October 2006 to May 2007 was 660.0 mm; this was within 1 standard deviation (94.0 mm) of the mean total precipitation for the October to May period for 1981–2010 (-692.7 mm). Peaks in discharge coincided with large precipitation events in the fall and increasing temperatures in the spring (Figure 2b). Water table depth increased from near zero in mid-October to more than 100 mm in response to fall storms, fluctuating at the beginning of the winter then gradually decreasing before gradually increasing through February and

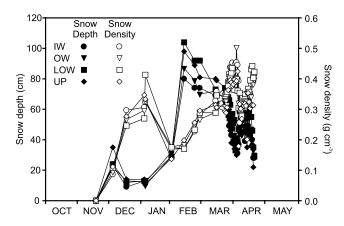


Figure 5. Daily snow depth and snow density in c38 from October 2006 to May 2007.

March until there was a sharp increase in response to snowmelt in April (Figure 4b). Snow depth generally increased from the end of November to approximately mid-February when it began to recede (Figure 5); the LOW and UP positions generally had deeper snow than the wetland positions. Snow density generally increased throughout the nongrowing season, with one notable decrease in January (Figure 5). The snow density increase beginning in February was coincident with a steady increase in water table depth during a period with sustained air temperatures below zero.

3.2. Soil Physical and Chemical Properties

Soil temperature at all topographic positions was approximately 10°C in October and rapidly declined during the month and eventually stabilized and ranged between 0°C and 2°C from December to mid-April (Figure 6, left). There was no significant variation in soil temperature among the topographic positions (Figure 6, right). The IW recorded a minimum of 0.2°C and maximum 12.7°C , the OW recorded a minimum of 0.4°C and maximum of 13.8°C , and the LOW recorded a minimum of 0.2°C and a maximum of 13.8°C . The UP was the only position that had subzero temperatures. The UP position recorded 28 days of subzero temperatures (all from January to March), but the subzero temperatures were close to zero, with a minimum temperature of -0.3°C and a maximum temperature of 12.0°C .

Soil moisture in the IW and OW were consistently around 70% throughout the nongrowing season that included the snow period, whereas soil moisture in the LOW and UP fluctuated more, with the LOW recording a minimum of 24% and maximum of 57% and the UP recording a minimum of 18% and a maximum of 50% (Figure 6, left). There was significant variation in soil moisture among topographic positions, with the IW being the wettest, the OW being slightly but significantly drier, and the LOW and UP significantly drier than both of the wetland positions (Figure 6, right).

Redox potential also fluctuated over the course of the nongrowing season, with the OW position most consistently in the -100 to $+100\,\text{mV}$ range (Figure 6, left). There was significant variation in soil redox potential among all topographic positions (Figure 6, right). The IW had redox potential frequently between $-100\,\text{and}\,+100\,\text{mV}$. The OW had significantly lower redox potential and was consistently in the $-100\,\text{to}\,+100\,\text{mV}$. The LOW and UP had significantly higher redox potentials; the majority of which were above $+100\,\text{mV}$.

There were no clear trends in soil pore water NO_3^- and DOC concentrations over time (Figure 6, left), but they did generally have the same trends across topographic positions (Figure 6, right). Time series of NO_3^- and DOC concentrations revealed higher concentrations coinciding with premelt with NO_3^- and DOC concentrations significantly higher at the UP position. The UP had significantly higher DOC and NO_3^- concentrations than the other positions, and the OW had the lowest DOC and NO_3^- concentrations (Figure 6, right).

3.3. Net Soil N₂O Production

There was generally a uniform distribution of net soil N_2O -N production during the nongrowing season that included the snow period (Figure 7a), although the highest variation in net soil N_2O -N production was observed during prepeak discharge period, which also coincided with changing soil moisture and redox conditions as the snowpack melted. Net soil N_2O -N production was not significantly different among the topographic positions (Figure 7b). The OW had the largest range in net soil N_2O -N production (-3.3 to 26.2 g-N ha $^{-1}$ d $^{-1}$), while the LOW position recorded the lowest values of net soil N_2O -N production (-6.6 g-N ha $^{-1}$ d $^{-1}$). There were weak but significant relationships between soil physical and chemical

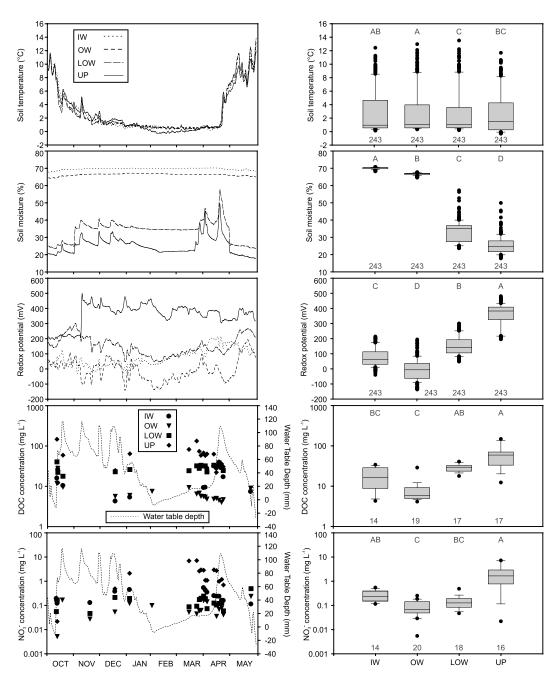


Figure 6. (left) Temporal and (right) spatial distributions of daily soil temperature, soil moisture, soil redox potential, DOC concentration, and NO_3^- concentration. The different letters above the boxes indicate the significant differences based on ANOVAs on ranks with post hoc Dunn's tests. The numbers below the boxes indicate the sample sizes (one value per day).

properties and net soil N_2O -N production (Table 1). The IW position had the highest percentage of negative N_2O flux estimates (19.5%); lower percentages were recorded at the OW, LOW, and UP positions (14.3, 15.0, and 13.2%, respectively).

Total atmospheric N input during the nongrowing period was $6.8 \, \text{kg ha}^{-1}$ or $43.2 \, \text{kg}$ for the entire catchment. Total net soil N₂O-N production was $1.5 \, \text{kg-N}$ ($1.0 \, \text{g-N ha}^{-1} \, \text{d}^{-1}$ or $0.24 \, \text{kg-N ha}^{-1}$) for the entire nongrowing period, which represents 3.6% of the atmospheric N input for the same period and 31% of the growing season N₂O-N export. Total net soil N₂O production was $2.4 \, \text{kg}$, of which $1.3 \, \text{kg}$ was produced under the snowpack.

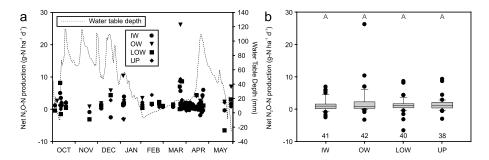


Figure 7. Net soil N_2O-N production (a) over time and (b) by topographic position. The different letters above the boxes indicate the significant differences based on ANOVAs on ranks with post hoc Dunn's tests. The numbers below the boxes indicate the sample sizes (one value per day).

4. Discussion

The purpose of this paper was to test the hypothesis that an insulating snowpack creates the conditions for significant gaseous N production. Net soil N_2O -N production ranged from -6.6 to 26.2 g-N ha⁻¹ d⁻¹ at different topographic positions during the nongrowing season that includes the snow period. Despite substantial topographic variability in soil physical and chemical properties, there was no significant difference in net soil N_2O -N production among the topographic positions, suggesting that different processes might be responsible for N_2O production or consumption at different positions.

Snowpacks insulate soils and can maintain soil temperatures above freezing despite subzero air temperatures, fostering the conditions for biological N cycling. *Groffman et al.* [2006] reported net N_2O -N production of approximately -3 to 3 g-N ha $^{-1}$ d $^{-1}$ beneath a snowpack in a different temperate sugar maple forest. Our study observed similar net N_2O -N production during the nongrowing season that included conditions beneath a snowpack (median value of 1.0 g-N ha $^{-1}$ d $^{-1}$), although our estimates were more variable, including a maximum of 26.2 g-N ha $^{-1}$ d $^{-1}$ and a minimum of -6.6 g-N ha $^{-1}$ d $^{-1}$. This suggests that we may have captured hot moments for denitrification [*McClain et al.*, 2003] that may have occurred due to a coincidence in optimal conditions for N_2O efflux (NO_3^- , DOC, redox, and moisture conditions) or consumption due to further reduction of N_2O to N_2 . *Filippa et al.* [2009] reported N_2O export during the snow period to be 18% of the annual total in Colorado, which is comparable to the values in our study, although we included the shoulder months of October and May when there was no snow cover.

Capturing hot spots and hot moments in N_2O production and redox values requires many sample replicates, although we expect N transformation processes to be more uniform with the relatively stable conditions beneath a snowpack. The hot spot and hot moment phenomena may have contributed to the considerable N_2O efflux observed in upland areas. This, coupled with the higher likelihood of consumptive processes in wetland areas [Schlesinger, 2013] due to lower redox potentials, may have contributed to the lack of significant differences in N_2O efflux between wetland and upland areas.

Snow reduction can impact N cycling, and snow removal experiments that induce soil freezing can increase N_2O production through increased levels of inorganic N in the soil associated with reduced plant

Table 1. Spearman's Rank Correlations Between Soil Physical and Chemical Properties and Net N₂O Production in the Inner Wetland (IW), Outer Wetland (OW), Lowland (LOW), and Upland (UP) Positions^a

	IW		OW		LOW		UP	
Control Variable	r	р	r	р	r	р	r	р
Water table depth (mm)	-0.270	0.088	-0.325	0.036	-0.117	0.469	-0.206	0.212
Soil temperature (°C)	0.005	0.976	-0.165	0.295	-0.020	0.900	-0.219	0.185
Moisture (%)	-0.358	0.022	0.074	0.639	0.054	0.741	-0.320	0.050
Redox potential (mV)	-0.159	0.319	-0.005	0.976	0.162	0.317	0.171	0.303
Soil pore water DOC	-0.056	0.852	0.650	0.006	-0.288	0.308	-0.161	0.558
Soil pore water NO ₃	-0.084	0.783	-0.228	0.371	0.635	0.014	-0.042	0.880

^aThe bold numbers indicate the significant correlations (p < 0.05).



uptake (0 to $10 \text{ g-N} \text{ ha}^{-1} \text{ d}^{-1} [\text{Groffman et al., } 2006])$ or NO_3^- leaching [Fitzhugh et al., 2001]. In the current study, there was a snowpack (>10 cm) over the landscape for most of the snow period. Soil temperatures were typically greater than 0°C throughout the snow period (November to April; minimum of -0.3°C and maximum of 7.2°C) as well as the shoulder months (October and May). Even when soil temperature dipped slightly below zero, soil temperatures were still higher than ranges where NO_3^- consumption and N_2O production have previously been observed [Dorland and Beauchamp, 1991; Malhi et al., 1990]. There were no significant differences in soil temperature among the topographic positions, which may have contributed to the lack of significant differences in net soil N_2O production among the topographic positions.

Even though the rates of net N₂O production were similar between the UP and LOW positions and the IW and OW positions, the significant differences in redox and soil moisture across positions suggest that different processes were responsible for N₂O production. While microsites with different redox conditions likely exist across topographic positions, the dominant patterns in redox and soil moisture were strikingly different and strongly suggest that the dominant processes varied with topography, with more preferential conditions in wetland areas for N₂O consumption processes. The UP and LOW positions had soil moisture and redox potentials in the range that would support nitrification, while the IW and OW positions were in the range that would support denitrification. Work from other hardwood forests in central Ontario has demonstrated that substantial nitrification occurs beneath the snowpack at upland soils, but redox conditions limit the process in wetland soils during the winter [Casson et al., 2014]. Isotopic work at Turkey Lakes Watershed has demonstrated that the NO₃ in snowmelt discharge waters is of biological origin (nitrification) [Spoelstra et al., 2007, 2001], suggesting active biological nitrification at upland positions during winter. Within the wetland, the OW had both lower soil moisture and redox potentials than the IW position. This suggests greater microbial activity at the OW position generating a higher electron density and lower redox [Farquharson and Baldock, 2008], which nevertheless did not result in significant differences in net N₂O production between wetland positions; this lack of difference could be partly attributed to consumption of N₂O at these positions. Despite similar (yet significantly different) moisture and redox conditions in the IW and OW, the IW may have lacked the appropriate microbial community to support N₂O accumulation [Holtan-Hartwig et al., 2000], favoring N₂O consumption instead [Billings, 2008; Schlesinger, 2013].

The soil pore water DOC and NO_3^- concentrations varied among the topographic positions, which provides further insight into the processes generating N_2O . The DOC and NO_3^- accumulated at the UP position, which may suggest that denitrification was limited by high redox conditions, as is observed during the growing season [Enanga et al., 2016; Kralova et al., 1992], but that the NO_3^- pools were maintained through nitrification. In contrast, DOC and NO_3^- did not accumulate in the wetland positions, suggesting that denitrification may be limited by nutrient conditions beneath a snowpack in the wetland. At both wetland positions, high soil moisture ensured that reducing conditions persisted over long periods of time, limiting the opportunity to replenish NO_3^- pools through nitrification [Casson et al., 2014; Wrage et al., 2001]. Another possibility is that N_2O consumption by denitrification may occur at higher rates at the wetland positions, although this is very difficult to measure in the field [Groffman et al., 2009].

It is possible that pathways other than denitrification and nitrification are being used to produce gaseous N in the Turkey Lakes Watershed. Chemodenitrification is unlikely in our system because the pH of soils in the Turkey Lakes Watershed is too low (<4 [Kesik et al., 2006]). Similarly, dissimilatory nitrate reduction to ammonium can produce N₂O as a detoxification mechanism to deal with high concentrations of NO₂⁻ [Rutting et al., 2011], but this mechanism is only favored under high soil pH (>6.5 [Stevens et al., 1998]), so while it is unlikely to be important at this site, further study would be required to distinguish N₂O production via this pathway. It is challenging to distinguish between pathways of N₂O production in the field, and generally requires using isotopic techniques [Burgin and Hamilton, 2007; Spoelstra et al., 2001], made even more difficult by the practical challenges of collecting gas samples from snowpack-covered forests. Discerning pathways of gaseous N production under snowpacks in temperate forests is an area of opportunities for future research.

Future climate scenarios for northern temperate forests predict warmer air temperatures, with some areas receiving more precipitation in the form of snow [Colombo et al., 2007]. This may result in smaller snowpacks because of corresponding increases in thaw and rain-on-snow events [Campbell et al., 2005; Casson et al., 2014]. Decreasing snowpacks may paradoxically promote soil freezing despite increasing temperatures



[Lemke et al., 2007]. Soil freezing can have a large impact on N cycling in northern catchments, and freezethaw cycles can result in bursts of N₂O production [Groffman et al., 2006], but any reduction in snowpacks caused by increased temperatures did not result in an increase in soil freezing on average [Henry, 2008]. The results of the present study suggest that N₂O production can persist in soils where the soils are cold but above 0°C. The effects of climate change on N processes in the winter are complex and an important area of further research.

5. Conclusion

A stable snowpack created conditions that promoted the formation of N₂O in a sugar maple forest in central Ontario. While there was considerable net soil N₂O production in the nongrowing season (representing 31% of the growing season production), differences in physical and chemical soil properties did not result in significant differences in net soil N₂O production among topographic positions. Consumptive N₂O processes were marginally more preferred in the soil conditions in wetland areas under a snowpack compared to upland areas. Soil temperatures greater than 0°C allow N₂O production to persist under snowpacks, and soil redox conditions control the pathway by which the gas is produced. Investigating winter net N₂O production will lead to a better understanding of the fate of atmospherically deposited N on forested landscapes. Future work will investigate N processes during the entire year and examine the relative importance of atmospheric versus aquatic fates of N in this northern temperate forest.

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References

Aber, J. D., K. J. Nadelhoffer, P. Steudler, and J. M. Melillo (1989), Nitrogen saturation in northern forest ecosystems, BioScience, 39, 378-386. Beven, K., and M. J. Kirkby (1979), A physically based, variable contributing area model of basin hydrology, Hydrol. Sci. Bull., 24, 43-69. Billings, S. A. (2008), Nitrous oxide in flux, Nature, 456, 888-889.

Brooks, P. D., and M. W. Williams (1999), Snowpack controls on nitrogen cycling and export in seasonally snow-covered catchments, Hydrol. Proc., 13, 2177-2190.

Burgin, A. J., and S. K. Hamilton (2007), Have we overemphasized the role of denitrification in aquatic ecosystems? A review of nitrate removal pathways, Front. Ecol. Environ., 5, 89-96.

Campbell, J. L., M. J. Mitchell, P. M. Groffman, L. M. Christenson, and J. P. Hardy (2005), Winter in northeastern North America; a critical period for ecological processes, Front. Ecol. Environ., 3, 314-322.

Casson, N. J., M. C. Eimers, and S. A. Watmough (2014), Sources of nitrate export during rain-on-snow events at forested catchments, Biogeochemistry, 120, 23-36.

Chapuis-Lardy, L. Y., N. Wrage, A. Metay, J. L. Chotte, and M. Bernoux (2007), Soils, a sink for N₂O? A review, Glob. Change Biol., 13, 1-7. Colombo, S. J., D. W. McKenney, K. M. Lawrence, and P. A. Gray (2007), Climate Change Projections for Ontario: Practical Information for Policymakers and Planners, Ontario Ministry of Natural Resources, Toronto, Canada.

Creed, I. F., and L. E. Band (1998a), Exploring functional similarity in the export of nitrate-N from forested catchments: A mechanistic modeling approach, Water Resour. Res., 34, 3079–3093, doi:10.1029/98WR02102.

Creed, I. F., and L. E. Band (1998b), Export of nitrogen from catchments within a temperate forest: Evidence for a unifying mechanism regulated by variable source area dynamics, Water Resour. Res., 34, 3105-3120, doi:10.1029/98WR01924.

Creed, I. F., and F. D. Beall (2009), Distributed topographic indicators for predicting nitrogen export from headwater catchments, Water Resour. Res., 45, W10407, doi:10.1029/2008WR007285.

Creed, I. F., L. E. Band, N. W. Foster, I. K. Morrison, J. A. Nicolson, R. S. Semkin, and D. S. Jeffries (1996), Regulation of nitrate-N release from temperate forest: A test of the N flushing hypothesis, Water Resour. Res., 32, 3337-3354, doi:10.1029/96WR02399

Creed, I. F., C. G. Trick, L. E. Band, and I. K. Morrison (2002), Characterizing the spatial pattern of soil carbon and nitrogen pools in the Turkey Lakes Watershed: A comparison of regression techniques, Water Air Soil Poll.: Focus, 2, 81–102.

Davidson, E. A. (1992). Sources of nitric-oxide and nitrous-oxide following wetting of dry soil. Soil Sci. Soc. Am. J., 56, 95–102.

Dillon, P. J., L. A. Molot, and W. A. Schneider (1991), Phosphorus and nitrogen export from forested stream catchments in central Ontario, J. Env. Oual., 20, 857-864.

Dorland, S., and E. G. Beauchamp (1991), Denitrification and ammonification at low soil temperatures, Can. J. Soil Sci., 71, 293-303.

Enanga, E. M., I. F. Creed, N. J. Casson, and F. D. Beall (2016), Summer storms trigger soil N₂O efflux episodes in forested catchments, J. Geophys. Res.-Biogeo., 121, 95-108.

Farquharson, R., and J. Baldock (2008), Concepts in modelling N₂O emissions from land use, *Plant Soil*, 309, 147–167.

Filippa, G., M. Freppaz, M. W. Williams, D. Helmig, D. Liptzin, B. Seok, B. Hall, and K. Chowanski (2009), Winter and summer nitrous oxide and nitrogen oxides fluxes from a seasonally snow-covered subalpine meadow at Niwot Ridge, Colorado, Biogeochemistry, 95, 131–149.

Fitzhugh, R. D., C. T. Driscoll, P. M. Groffman, G. L. Tierney, T. J. Fahey, and J. P. Hardy (2001), Effects of soil freezing disturbance on soil solution nitrogen, phosphorus, and carbon chemistry in a northern hardwood ecosystem, Biogeochemistry, 56, 215-238.

Galloway, J. N., A. R. Townsend, J. W. Erisman, M. Bekunda, Z. Cai, J. R. Freney, L. A. Martinelli, S. P. Seitzinger, and M. A. Sutton (2008), Transformation of the nitrogen cycle: Recent trends, questions, and potential solutions, Science, 320, 889-892.

Gambrell, R. P., and W. H. Patrick Jr. (1978), Chemical and microbiological properties of anaerobic soils and sediments, in Plant Life in Anaerobic Environments, edited by D. D. Hook and R. M. Crawford, pp. 375-423, Ann Arbor Sci. Pub. Inc., Michigan.

Groffman, P. M., J. P. Hardy, C. T. Driscoll, and T. J. Fahey (2006), Snow depth, soil freezing, and fluxes of carbon dioxide, nitrous oxide and methane in a northern hardwood forest, Glob. Change Biol., 12, 1748–1760.

Groffman, P. M., K. Butterbach-Bahl, R. W. Fulweiler, A. J. Gold, J. L. Morse, E. K. Stander, C. Tague, C. Tonitto, and P. Vidon (2009), Challenges to incorporating spatially and temporally explicit phenomena (hotspots and hot moments) in denitrification models, Biogeochemistry, 93, 49-77.

Cambridge, Mass.



- Hardy, J. P., P. M. Groffman, R. D. Fitzhugh, K. S. Henry, A. T. Welman, J. D. Demers, T. J. Fahey, C. T. Driscoll, G. L. Tierney, and S. Nolan (2001), Snow depth manipulation and its influence on soil frost and water dynamics in a northern hardwood forest, *Biogeochemistry*, 56, 151–174. Heincke, M., and M. Kaupenjohann (1999), Effects of soil solution on the dynamics of N₂O emissions: A review, Nut. Cyc. Agroecosyst., 55,
- Henry, H. A. L. (2008), Climate change and soil freezing dynamics: Historical trends and projected changes, Clim. Change, 87, 421-434. Holtan-Hartwig, L., P. Dörsch, and L. R. Bakken (2000), Comparison of denitrifying communities in organic soils: Kinetics of NO₃⁻ and N₂O reduction, Soil Biol. Biochem., 32, 833-843.
- Hutchinson, G. L., and G. P. Livingston (2001), Vents and seals in non-steady-state chambers used for measuring gas exchange between soil and the atmosphere, Eur. J. Soil Sci. 52, 675-682.
- Jeffries, D. S., J. R. M. Kelso, and I. K. Morrison (1988), Physical, chemical, and biological characteristics of the Turkey Lakes Watershed, central Ontario, Canada, Can. J. Fish. Aquat. Sci., 45, 3-13.
- Keeney, D. R., and D. W. Nelson (1982), Nitrogen-inorganic forms, in Methods of Soil Analysis Part 2, vol. 9, 2nd ed., edited by A. L. Page, R. H. Miller, and D. R. Keeney, pp. 643-698, Madison, Wis
- Kesik, M., S. Blagodatsky, H. Papen, and K. Butterbach-Bahl (2006), Effect of pH, temperature and substrate on N₂O, NO and CO₂ production by Alcaligenes faecalis p, J. App. Microbiol., 101, 655-667.
- Kralova, M., P. H. Masscheleyn, C. W. Lindau, and W. H. Patrick Jr. (1992), Production of dinitrogen and nitrous oxide in soil suspensions as affected by redox potential, Water Air Soil Poll., 61, 37-45.
- Laughlin, R. J., and R. J. Stevens (2002), Evidence for fungal dominance of denitrification and codenitrification in a grassland soil, Soil Sci. Am. J., 66, 1540-1548.
- Lemke, P., et al. (2007), Observations: Changes in snow, ice and frozen ground, in Climate Change 2007: The Physical Science Basis. Contribution of Working Group I to the Fourth Assessment Report of the Intergovernmental Panel on Climate Change, edited by S. Solomon et al., Cambridge University Press, New York.
- Lindsay, J., and I. F. Creed (2006), Distinguising actual and artefact depressions in digital elevation data, Comp. Geosci., 32, 1192-1204. Livingston, G. P., and G. L. Hutchinson (1995), Enclosure-based measurement of trace gas exchange: Applications and sources of error, in Biogenic Trace Gases: Measuring Emissions from Soil and Water, edited by P. A. Matson and R. C. Harriss, pp. 14-51, Blackwell Science,
- Malhi, S. S., W. B. McGill, and M. Nyborg (1990), Nitrate losses in soils: Effects of temperature, moisture and substrate concentration, Soil Biol. Biochem, 22, 733-737.
- McClain, M. E., et al. (2003), Biogeochemical hot spots and hot moments at the interface of terrestrial and aquatic ecosystems, Ecosystems, 6.
- Morse, J. L., J. Durán, F. Beall, E. M. Enanga, I. F. Creed, I. Fernandez, and P. M. Groffman (2015), Soil denitrification fluxes from three northeastern North American forests across a range of nitrogen deposition, Oecologia, 177, 17-27.
- Rutting, T., P. Boeckx, C. Muller, and L. Klemedtsson (2011), Assessment of the importance of dissimilatory nitrate reduction to ammonium for the terrestrial nitrogen cycle, Biogeosciences, 8, 1779–1791.
- Schlesinger, W. H. (2013), An estimate for the global sink for nitrous oxide in soil, Glob. Change Biol., 19, 2929–2931.
- Sirois, A., and R. Vet (1988). Detailed analysis of sulphate and nitrate atmospheric deposition estimates at the Turkey Lakes Watershed. Can. J. Fish. Aquat. Sci., 45, 14-25.
- Smith, N. K. (1997), The potential for feedback effects induced by global warming on emissions of nitrous oxide by soils, Glob. Change Biol., 3,
- Sommerfeld, R. A., A. R. Mosier, and R. C. Musselman (1993), CO₂, CH₄ and N₂O flux through a Wyoming snowpack and implications for global budgets, Nature, 361, 140-142.
- Sommerfeld, R. A., W. J. Massman, R. C. Musselman, and A. R. Mosier (1996), Diffusional flux of CO2 through snow: Spatial and temporal variability among alpine-subalpine sites, Global Biogeochem. Cy., 10, 473-482.
- Spoelstra, J., S. L. Schiff, R. J. Elgood, R. G. Semkin, and D. S. Jeffries (2001), Tracing the sources of exported nitrate in the Turkey Lakes Watershed using ¹⁵N/¹⁴N and ¹⁸O/¹⁶O isotopic ratios, *Ecosystems*, 4, 536–544.
- Spoelstra, J., S. L. Schiff, P. W. Hazlett, D. S. Jeffries, and R. G. Semkin (2007), The isotopic composition of nitrate produced from nitrification in a hardwood forest floor, Geochim. Cosmochim. Ac., 71, 3757-3771.
- Stevens, R. J., R. Laughlin, and J. Malone (1998), Soil pH affects the processes reducing nitrate to nitrous oxide and di-nitrogen, Soil Biol. Biochem., 30, 1119-1126.
- Stoddard, J. L. (1994), Long-term changes in watershed retention of nitrogen, in Environmental Chemistry of Lakes and Reservoirs, edited by L. A. Baker, American Chemical Society, Washington, D. C.
- Tarboton, D. G. (1997), A new method for the determination of flow directions and upslope areas in grid digital elevation models, Water Resour, Res., 33, 309-319, doi:10.1029/96WR03137.
- Webster, K. L., I. F. Creed, F. D. Beall, and R. A. Bourbonnière (2011), A topographic template for estimating soil carbon pools in forested catchments, Geoderma, 160, 457-467.
- Wickware, G. M., and D. W. Cowell (1985), Forest ecosystem classification of the Turkey Lakes Watershed, Ontario: A research contribution, No. 18, Environment Canada, Lands Directorate, Ottawa, Canada
- Wrage, N., G. L. Velthof, M. L. van Beusichem, and O. Oenema (2001), Role of nitrifier denitrification in the production of nitrous oxide, Soil Biol. Biochem., 33, 1723-1732.
- Yoshikawa, K., P. P. Overduin, and J. W. Harden (2004), Moisture content measurements of moss (Sphagnum spp.) using commercial sensors, Permafrost Periglac. Proc., 15, 309-318