

Chapter 4

TREE SPECIES EFFECTS ON NITROGEN CYCLING AND RETENTION: A SYNTHESIS OF STUDIES USING ^{15}N TRACERS

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ABSTRACT

The forests of the Catskill Mountains in New York receive some of the highest rates of nitrogen (N) deposition in the northeastern United States, and many watersheds are beginning to show signs of N saturation. The watershed export of N is variable, despite relatively uniform N deposition. I tested the hypothesis that tree species vary in their influence on forest N retention and loss. A laboratory study showed that tree species influenced soil microbial biomass and organic soil C:N, but that variation in these properties cannot explain differences in potential net mineralization, net nitrification, or microbial uptake of ^{15}N . A greenhouse experiment showed that sugar maple seedlings take up more NH_4^+ than NO_3^- , while beech seedlings take up more NO_3^- than NH_4^+ . Results from a 300-day ^{15}N experiment in the field showed that most of the N deposited onto forests is retained within the forest floor, and the magnitude of N retention varied among tree species. Sugar maple stands retained the least N, and red oak stands retain the most. A fertilization experiment showed that red oak stands are most likely to have the greatest decrease in N retention if availability increases in the future. Sugar maple stands currently export substantially more N than the other forests, in part due to a limited capacity to take up NO_3^- . Tree species composition can be a strong regulator of forest N retention, but differences among species may depend on N inputs. Future research needs to address the interacting effects of increased N inputs and changes in tree species composition, as non-linear effects may likely occur in biogeochemical cycles.

INTRODUCTION

Humans have dramatically increased the amount of fixed nitrogen (N) in the terrestrial biosphere through the production of N fertilizers, fossil fuel combustion, and the use of N-fixing species in agriculture and forestry (Galloway et al. 1995). This has led to increasing amounts of N being deposited onto terrestrial ecosystems. Increased N inputs to forest ecosystems can lead to N saturation, the syndrome of responses in which excess N supply to forests leads to nitrate (NO_3^-) leaching into groundwater and streams and other alterations of forest nutrient cycling (Agren and Bosatta 1988; Aber et al. 1989; Stoddard 1994; Peterjohn et al. 1996). Symptoms of N saturation and excess N leaching have been observed in temperate forests of the United States (Johnson et al. 1991; Driscoll and Van Dreaseon 1993; Gilliam et al. 1996) and Europe (Gundersen et al. 1998, MacDonald et al. 2002). Despite these signs of nitrogen saturation syndrome, most forests in the United States continue to retain a large proportion of N deposition, often 90% (Peterjohn et al. 1996; Lovett et al. 2000; see chapters in this volume by Rothe, Butterbach-Bahl and Kiese, and Papen and coauthors for truly N saturated forests in Bavaria.)

Forest fertilization and ^{15}N tracer experiments in the United States have shown that of the N deposited onto forests, the majority is retained within the soil, with the vegetation accounting for a minority of the N retention (Johnson 1992, Fenn et al. 1998; Nadelhoffer 1999; Magill et al. 1997). Even if forests retain a large amount of deposited N, N leached in the form of NO_3^- can cause essential cations such as calcium (Ca) and potassium (K) to be leached out of the forest soil, and some have suggested this could lead to nutrient imbalances in trees (Friedland et al. 1988; Schulze 1989). Nitrogen leaching can also lead to acidification of stream water (Vitousek et al. 1997), eutrophication of estuaries and coastal areas (Howarth 1988) and changes in species composition (Vitousek et al. 1997).

The Catskill Mountains in southeastern New York receive among the highest inputs of N deposition in the northeastern United States (Ollinger et al. 1993; Stoddard 1994), with wet N deposition rates of 4 to 9 kg N ha⁻¹ yr⁻¹ from 1986 through 2003 (National Atmospheric Deposition Program 2004). The inclusion of dry deposition of N raises the total N deposition estimate to about 11 kg N ha⁻¹ yr⁻¹ (Lovett and Reuth 1999). Concentrations of nitrate (NO_3^-) in stream water have increased in the past 25 years (Murdoch and Stoddard 1993), but these increases have not been consistent across all watersheds in the Catskill Mountains. Nitrogen retention ranges between 49-90% of atmospheric input, and stream NO_3^- concentrations vary by 17-fold, even among watersheds that are completely forested and have similar rates of N deposition (Lovett et al. 2000).

The mechanisms behind the variation in stream NO_3^- concentration remain unclear despite extensive research on potential factors such as hydrology (Burns et al. 1998; West et al. 2001), N deposition and topography

(Lovett et al. 2000; Weathers et al. 2000). Some of the variation may relate to tree species composition; NO_3^- loss relates inversely to soil C:N, which in turn relates with species composition of the forests (Lovett et al. 2002). Mixed-species stands dominated by sugar maple (*Acer saccharum* Marsh) have low C:N and high nitrate leaching, whereas forests dominated by red oak (*Quercus rubra* L.) have higher C:N and lower nitrate losses (Lovett et al. 2002). These observations led to the hypothesis that tree species composition may influence N retention and thereby influence the amount of NO_3^- reaching streams (Lovett et al. 2000, Lovett et al. 2002).

Plant species can affect the localized movement of N through ecosystems via indirect effects on soil chemical properties and microbial activity (Vitousek et al. 1982; Zak et al. 1986; Finzi et al. 1998). Tree species differ in leaf litter quality (e.g. lignin:N or C:N ratio of litter; see Prescott and Vesterdal, this volume), providing a range of organic matter quality for microbial communities (Pastor and Post 1986). Therefore, soil microbes associated with different tree species may have variable rates of organic matter decomposition (Melillo et al. 1982), N mineralization (Vitousek et al. 1982; Zak and Pregitzer 1990; Zak et al. 1986), and nitrification (Lovett and Rueth 1999; Robertson 1982; Zak et al. 1986).

Plant species can also affect forest N cycling rates through differences in uptake and sequestration of N (Gharbi and Hipkin 1984; Horsley 1988; Crabtree and Bazazz 1993; Nadelhoffer et al. 1995). For example, plant species differ in their capacity to take up NH_4^+ or NO_3^- as their primary N source in part because of physiological trade-offs in taking up either form of N (Crabtree and Bazazz 1993; Horsley 1988; Stewart et al. 1992). While NO_3^- is readily available because of its high solubility in soil water, plants must use energy to reduce the NO_3^- prior to incorporation into their amino acids. With this constraint, it may be more efficient for plants to take up NH_4^+ because it can be immediately incorporated into amino acids. However, NH_4^+ is not always available to plants for uptake because it is tightly associated with soil exchange sites and does not move readily through the soil in solution.

This research tested the hypothesis that N cycling and retention varies significantly among forested stands dominated by different tree species in the Catskill Mountains. I expected N retention to be low in stands dominated by sugar maple, high in stands dominated by red oak, and intermediate in beech (*Fagus grandifolia* Ehrh.) stands. This pattern is based on differences in litter quality and soil chemistry characteristics associated with each species. For example, laboratory (Lovett and Rueth 1999; Templer et al. 2003; Lovett et al. 2004) and field rates (Finzi et al. 1998; Lawrence 2000) of net nitrification were higher in stands of sugar maple compared to the other three tree species. Higher net nitrification rates in sugar maple stands may increase NO_3^- leaching and lower forest N retention compared to forest stands dominated by other tree species. Lower rates of net nitrification in red oak stands may lead to greater N retention within these stands compared to other tree species.

This paper summarizes three experiments that used ^{15}N isotope methods to:

- (1) examine the effect of tree species on soil microbial biomass, determining if microbial biomass strongly influences microbial processes that affect retention or loss of N;
- (2) compare NH_4^+ and NO_3^- uptake and sequestration by dominant tree species, and
- (3) quantify the fate of deposited N within stands of the dominant tree species, and determine the impact of N fertilization on forest N retention.

MATERIALS AND METHODS

Site Description

The Catskill Mountains are a range of low, mostly flat-topped mountains in southeastern New York. The bedrock is composed mostly of sandstone, shale, and conglomerate (Stoddard and Murdoch 1991), covered by glacial till that ranges from 0 to > 30 m in depth (Kudish 2000). The Inceptisol soils have moderate to high acidity (Stoddard and Murdoch 1991) and are well drained and moderately steep. Soils average 60% sand, 30% silt and 10% clay content (Kudish 2000). Vegetation between 500-1100 m elevation is dominated by northern hardwood forests common throughout the northeastern United States, including sugar maple, American beech, red oak, and eastern hemlock (*Tsuga Canadensis* L.; Kudish 1971; McIntosh 1972). About 80-90% of the original forest of the Catskill Mountains was subject to some level of harvesting by the end of the nineteenth century, although most of the cutting was selective harvest rather than clear-cutting. The Catskill Forest Preserve was created in 1885 with most of the land within its current borders added by the 1930's (Kudish 2000). The Catskill Park covers approximately 285,500 hectares, with about 41% of that protected in the Catskill Forest Preserve (Van Valkenburgh 1996). Mean annual temperature is 4.3 °C and mean annual precipitation is 153 cm at the 808 m elevation weather station on Slide Mountain.

Objective 1: Tree species effects

A laboratory study examined microbial biomass, potential net mineralization, potential net nitrification, and microbial uptake of added $^{15}\text{NH}_4$ in soils collected in August 1997 from plots of the five dominant tree species inhabiting watersheds of the Catskill Mountains, NY. We chose two 12 m diameter plots that were dominated by mature individuals of each of the

4 target species: American beech, red oak, sugar maple, and yellow birch (*Betula alleghaniensis* Britton). These stands were similar in age, had no recent disturbance, and were surrounded by mixed species stands. Litter of the target tree species represented 67-84% of the total litter at each plot. Soils under beech, sugar maple and yellow birch were collected from the upper Rondout watershed, and red oak soils were collected from the Kanape Brook watershed (see Lovett et al. 2000 for watershed characteristics). After brushing aside the litter layer (Oi), we collected 4 samples of the rest of the O horizon (two samples from each plot, including Oe and Oa where present). Each sample was a composite of four soil cores (6 cm diameter; 12 cm depth maximum each). Composited samples were sieved (8 mm sieve) to remove larger roots, woody fragments and stones, then analyzed for total C and N (Carlo-Erba NA 1500 C-N analyzer), soil organic matter content (loss on ignition), extractable N (KCl extraction), microbial biomass N (MBN, chloroform fumigation-extraction), microbial uptake of added ^{15}N , and potential net mineralization and nitrification for 5 incubation periods (0, 1, 3, 10 or 28 days; see Templer et al. 2003 for more detail).

Two-way analyses of variance (ANOVA) tested for the effects of tree species and incubation period for MBN, soil NO_3^- and NH_4^+ using SAS JMP software (Version 3.2.5, 1999). One-way ANOVA tested for the main effects of species on other variables. We also examined correlations between soil C, N, C:N, field condition soil moisture and microbial properties and processes to examine potential relationships among factors. The effects of species were confounded with any differences that existed between the sites in other factors, but for simplicity the effects will be referred to simply as species effects. In all experiments, a P value of 0.05 was used to avoid Type I errors in our hypothesis tests.

Objective 2: Ammonium versus nitrate uptake

A greenhouse study in 1998 examined N uptake by American beech, red oak, and sugar maple seedlings. We collected 14 seedlings of each species, and their native Oe and Oa horizons, which were randomly selected from mixed species stands of the Kanape Brook watershed in the Catskill Mountains, NY in November 1997. Seedlings retained their own soil and were kept moist and cool while being transported from the field. To maintain the dormancy normally experienced in the field, we over-wintered them at 2.2 °C. The plants were moved into a greenhouse (25-28 °C) on April 3, 1998 to break dormancy. Most leaves reached full expansion by May 5, 1998, when we added 99 atom% ^{15}N - NH_4 (5 ml of 12 mg N/L as $^{15}\text{NH}_4\text{-NO}_3$) to six of the seedlings of oak and maple, and 99 atom% enriched ^{15}N - NO_3 (5 ml of 12 mg N/L as $\text{NH}_4\text{-}^{15}\text{NO}_3$) to six others to examine uptake of NH_4^+ and NO_3^- . Two individuals of each tree species served as controls, receiving no N addition and providing the natural abundance of ^{15}N for each tree species. The same

experimental design was used for beech, except ^{15}N tracer was added to only 10 individuals because 2 died during the winter. Plants were harvested 4 days after ^{15}N addition to minimize the impact of microbial activity on N transformations of NH_4^+ and NO_3^- in the soil; this was a long enough time to detect enriched ^{15}N in the plant biomass. We analyzed C, N and ^{15}N on roots, stem and leaves of each seedling and extractable NH_4^+ and NO_3^- in the soil of each pot using the methods described in Templer and Dawson (2004).

We calculated plant N uptake as:

$$N_{\text{up}} = B^{-1} * \frac{(N_{\text{plt}})(\text{atom } \% ^{15}\text{N}_{\text{plt}}) - (N_{\text{plt}})(\text{atom } \% ^{15}\text{N}_{\text{NA}})}{\text{atom } \% ^{15}\text{N}_{\text{S}}}$$

where N_{up} is the amount of N taken up by the plant; B = biomass of the plant at the end of the experiment; $\text{atom } \% ^{15}\text{N}_{\text{plt}}$ = atom percent ^{15}N of the plant at the end of the experiment; N_{plt} = mass of N in the plant at the end of the experiment; $\text{atom } \% ^{15}\text{N}_{\text{NA}}$ = atom percent ^{15}N of the reference plants (seedlings that did not receive ^{15}N tracer); $\text{atom } \% ^{15}\text{N}_{\text{S}}$ = atom percent ^{15}N of soil NO_3^- or NH_4^+ at the end of the experiment. We calculated uptake of N per unit plant biomass for a measure of plant demand for N that accounts for differences in plant biomass among tree species. We also calculated uptake of N per total soil NH_4 and NO_3 to account for differences in availability of the two forms of N.

We conducted one-way analyses of variance (ANOVA) with tree species as the main effect to examine plant biomass, plant N, as well as soil NH_4^+ and NO_3^- . To examine preference for NH_4^+ or NO_3^- , we conducted an ANOVA within each tree species with form of N (NH_4^+ vs NO_3^-) as the main effect. All of the data were log-transformed prior to statistical analysis. A Kruskal-Wallis test was performed if the data were not normally distributed after the log transformation. The effects of species were again confounded with any differences that covaried with the locations where the seedlings had established.

Objective 3: Fate of N deposition

A 300-day field ^{15}N experiment examined soil and plant pools in the field, tracing N movement and retention times within various pools. If retention within different pools differed among tree species, changes in tree species composition in the future could alter the location and retention of N within forests. A fertilization treatment was used to examine the potential effect of higher N inputs on N retention. Methods are described in detail in Templer et al. (2005).

In 1999-2000, we examined the fate of N in 6 sets of 12-m diameter paired plots for sugar maple, American beech and red oak (12 plots per

species total). The plots were located within mixed-species forest and were composed of clusters of the target tree species. The subsequent measurements showed that litter of the target tree species represented 67-84% of the total litter at each plot, with non-target litter resulting primarily from understory trees and trees outside the plot. The 6-paired plots were distributed across three watersheds for each of the tree species. Because of the distribution of tree species within the Catskill Mountains, we could not easily locate all three species within each watershed. One plot of each pair received only ambient levels of N deposition (approximately 11.2 kg N/ha/yr), while the other had been fertilized with an additional 50 kg N/ha/year (as granular NH_4NO_3 in four doses per year-- June, July, August and November of each year) for the 1.5 years prior to and during this experiment; thus the total N enhancement in the fertilized plots by the end of this experiment was 112.5 kg N / ha. This enabled us to compare the movement of N within plots that received ambient levels of N to those that received a higher level of N. To each of the paired plots, we added trace amounts of 99 atom % enriched $^{15}\text{NH}_4\text{Cl}$ to the inner 8 m of each plot during July, August and October 1999 (3 additions of 10 mg $^{15}\text{NH}_4\text{Cl-N} / \text{m}^2$ each; dissolved in 5 L deionized water for each plot). We added ^{15}N three times, including one dose in the fall after leaf drop, rather than as a single pulse, for a more natural simulation of N availability throughout the year. In July and August of 1999, ^{15}N was added approximately 1.5 weeks following the N fertilization additions. We added the ^{15}N isotope as NH_4 to trace the large flux of N that is mineralized from organic matter, as opposed to the smaller flux of N received from ambient deposition, which occurs primarily as NO_3 . The tracer solution was applied to an 8-m diameter subplot using a backpack sprayer to ensure even distribution. For this experiment, the plot size was reduced to 8 m diameter because that area was adequate to detect ^{15}N tracer additions and because of the significant cost of ^{15}N addition.

To determine natural abundance background ^{15}N , roots and soil (Oe and Oa horizons) samples were collected from each plot in June 1999. Surface litter layer (Oi) samples were collected from areas outside, and directly adjacent to, each plot during August 1999. Wood and bark samples were collected from areas outside and directly adjacent to each plot during May 2000. To track the fate of the added ^{15}N in the soil, we collected 4 samples of the litter layer (Oi), organic (Oe and Oa horizons) and mineral soil and fine root samples from each plot on three dates. The first sampling occurred 2 days following the first ^{15}N addition in July 1999. The second sampling (day 90 after initial ^{15}N addition) occurred just prior to the third ^{15}N addition in October 1999. The third sampling (day 300) occurred during May 2000, prior to budbreak of the deciduous tree species. In the calculations of ^{15}N recovery percentages, we used only ^{15}N inputs occurring before the sampling date (see Templer et al. 2005 for details).

We also measured total N content and ^{15}N concentrations of aboveground plant pools as described in Templer et al. (2005). We collected the outer 2 cm

of wood and bark from three individuals of the target tree species within each plot during May 2000 (prior to budbreak) to examine how much of the N was moved to long-term sinks within the trees. We used allometric equations for each tree species (Tritton and Hornbeck 1982) to calculate total aboveground tree biomass of each plot (4 m radius). Litterfall samples were collected biweekly during leaf fall in 1998 and 1999. Sun-lit foliage samples were collected from each plot during the peak-growing season (August) of 1998, 1999 and 2000 and were composited by plot and year prior to ^{15}N analysis.

We calculated ^{15}N recovery using N mass, the amount of ^{15}N added, and the atom % ^{15}N enrichment of the various ecosystem pools:

$$\% ^{15}\text{N}_{\text{rec}} = 100 * \frac{m_{\text{pool}} * (\text{atom}\% ^{15}\text{N}_{\text{pool}} - \text{atom}\% ^{15}\text{N}_{\text{ref}})}{^{15}\text{N}_{\text{tracer}}}$$

where $\% ^{15}\text{N}_{\text{rec}}$ = percent of ^{15}N tracer recovered in the labeled N pool; m_{pool} = N mass of the labeled pool; $\text{atom}\% ^{15}\text{N}_{\text{pool}}$ = atom percent ^{15}N in the labeled pool; $\text{atom}\% ^{15}\text{N}_{\text{ref}}$ = atom percent ^{15}N of the reference (non-labeled) plots; $^{15}\text{N}_{\text{tracer}}$ = amount of ^{15}N added to each plot prior to sample collection (Buchmann et al. 1996). We use ^{15}N recovery as an estimate of net retention of N in a given pool at a given point in time.

We averaged the mass and N pool size for soil and plant samples over time for each plot, and conducted two-way analyses of variance (ANOVA) testing the effects of tree species and N treatment (ambient vs. fertilized). Each plot was nested within tree species because of the paired-plot design. Data that were not normally distributed were log-transformed prior to statistical analysis. We conducted linear contrasts of the means with Tukey-Kramer post hoc tests to examine the effect of tree species on forest ^{15}N recovery and to compare ambient and fertilized plots. Only the results from day 300 (May 2000) are presented. The effect of tree species was again confounded with any other factors that may have differed among the plots.

Enriched ^{15}N samples were analyzed on a Europa Integra, which is a combined sample combustion unit and isotope ratio mass spectrometer. Natural abundance ^{15}N samples were analyzed on a Europa 20-20 mass spectrometer after combustion in a Europa ANCA-GSL combustion unit. The standard used was 0.36679 atom% ^{15}N calibrated against IAEA N1, an International Atomic Energy Agency standard.

RESULTS

Objective 1: Tree species effects

The N content of microbial biomass and rates of N transformation in the laboratory clearly differed among soils collected from sites dominated by

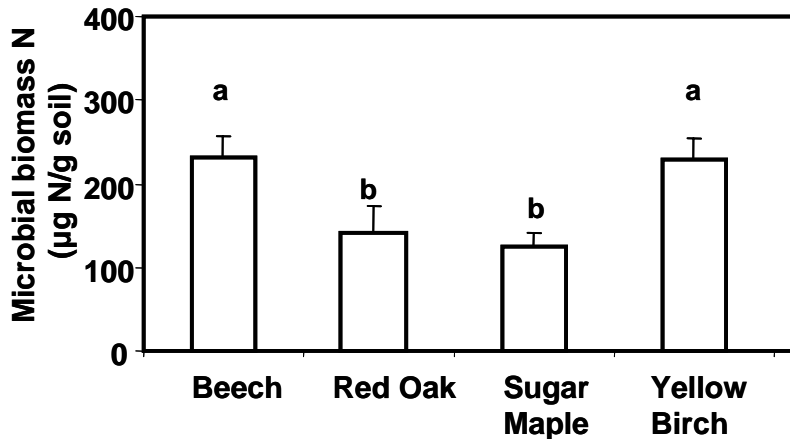


Figure 1. Microbial biomass N among tree species (error bars represent standard error; $n=4$ for each tree species; each bar represents the mean of four sample replicates averaged across the following 5 sampling periods: 0, 1, 3, 10, 28 days). From Templer et al. 2003.

different tree species, but the patterns among parameters was not consistent. Soil MBN ranged 2-fold, with lower values in stands of red oak and sugar maple than beech and yellow birch (Figure 1). Sugar maple soils consistently had larger soil NO_3^- pools (Table 1) and greater rates of potential net nitrification (Figure 2) than red oak soils, despite similar soil MBN. Short-term recovery (day 3 minus day 0) of ^{15}N recovery per unit MBN varied among tree species with greater values in soils of red oak (0.023) stands compared to sugar maple stands (-0.024). The uptake of ^{15}N per unit MBN in 28 days was similar to the 3-day trend, but the differences among tree species were not significant ($p=0.6$). Soil moisture accounted for 22% of the variation in MBN and 36% of the variation in net mineralization. The C:N ratio of the soil samples differed among species, with lowest values in sugar maple soils and the greatest values in red oak soils (Table 1). However, C:N did not correlate with rates of net N mineralization, net nitrification, or MBN.

Objective 2: Ammonium versus nitrate uptake

Total plant uptake of $\text{NH}_4\text{-N}$ was 5-fold greater (per unit plant biomass) than uptake of $\text{NO}_3\text{-N}$ in sugar maple seedlings (Figures 3 and 4). The red oak seedlings contained about 3 times more N derived from $^{15}\text{NH}_4^+$ than $^{15}\text{NO}_3^-$, but the power of the experiment could not show that this difference was significant. In contrast to the other three tree species, beech took up 5 times more N as $^{15}\text{NO}_3^-$ than $^{15}\text{NH}_4^+$.

When total soil $\text{NH}_4\text{-N}$ and $\text{NO}_3\text{-N}$ were taken into account, beech seedlings still took up 10 times more N as $\text{NO}_3\text{-N}$ than $\text{NH}_4\text{-N}$ ($p=0.022$; Figure 4). Sugar maple seedlings again took up almost 5 times more N as

Table 1. Organic horizon properties by tree species with standard error in parentheses (n=4 for each tree species). NH_4^+ , NO_3^- , dissolved organic N (DON) and total dissolved N (TDN) were averaged across days 0, 1, 3, 10 and 28 for each tree species. Different letters above values represent statistically significant differences at $p < 0.05$. From data in Templer et al. 2003.

Species	Moisture	Organic Matter	Carbon	Nitrogen	C:N	NH_4^+ N	NO_3^- N	DON	TDN
		------(%)-----		-----		-----	-----	-----	-----
Beech	47 (15)	79 (7)	34.3 (7.6)	1.88 (0.41)	18.4 ^{ac} (0.9)	69.4 (15.6)	11.0 ^a (2.5)	67.7 (8.4)	147 (18)
Red Oak	18 (6)	78 (5)	35.9 (3.5)	1.74 (0.20)	20.7 ^b (0.7)	56.4 (11.1)	1.3 ^a (0.3)	59.1 (9.4)	113 (15)
Sugar Maple	32 (9)	84 (2)	42.7 (1.4)	2.49 (0.09)	17.3 ^c (1.0)	51.2 (14.0)	53.6 ^b (19.1)	62.4 (27.6)	148 (37)
Yellow Birch	48 (8)	86 (5)	43.5 (3.8)	2.23 (0.19)	19.5 ^{abc} (0.1)	81.0 (13.4)	8.0 ^a (2.3)	71.2 (8.3)	157 (13)

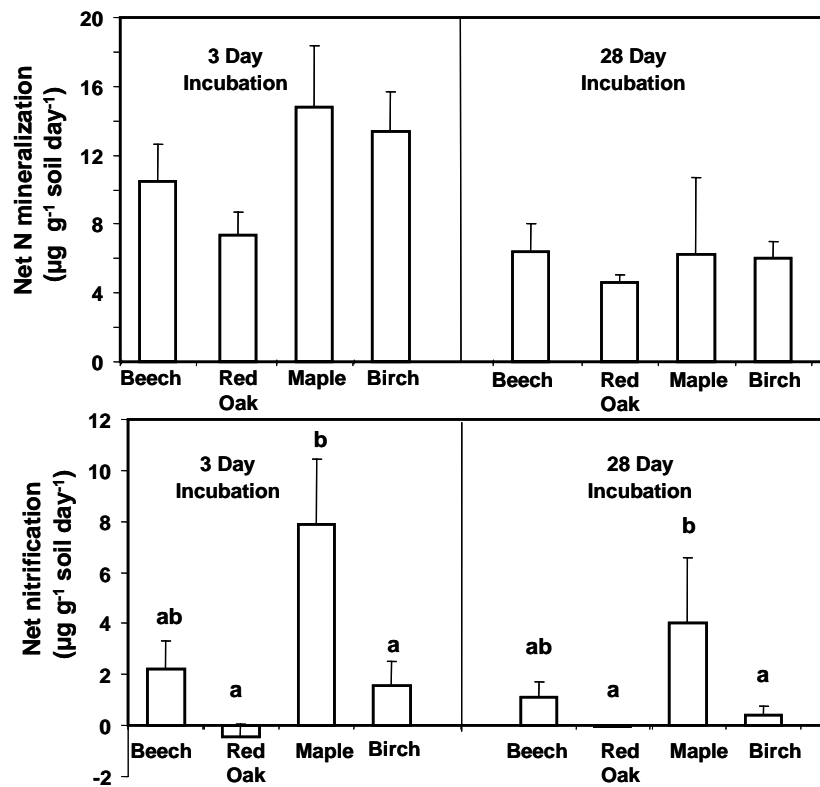


Figure 2. Potential net mineralization (upper) and potential net nitrification (lower) measured after 3 and 28 day laboratory incubations (error bars represent standard error; $n=4$ for each tree species). Different letters above bars indicated differences at $p<0.05$ (after Templer et al. 2003).

$\text{NH}_4\text{-N}$ than $\text{NO}_3\text{-N}$ ($p=0.0005$). Red oak seedlings averaged one-third higher uptake of NO_3 than NH_4 , but this difference was again not significant.

In addition to variation in composition of N uptake, tree species also varied in the magnitude of $\text{NH}_4\text{-N}$ and $\text{NO}_3\text{-N}$ uptake per unit plant biomass. For example, sugar maple took up over 6 times more $\text{NH}_4\text{-N}$ than red oak, and over 180-times more $\text{NH}_4\text{-N}$ than beech seedlings ($p<0.05$; Figure 3). Sugar maple and red oak seedlings had similarly high amounts of $\text{NO}_3\text{-N}$ uptake into their biomass.

Objective 3: Fate of N deposition

Total plot ^{15}N recovery (sum of the O horizon, fine roots, and any mineral soil to a depth of 12 cm, and aboveground biomass) did not vary among tree species ($p=0.7$), but the addition of extra N in fertilizer reduced the quantitative recovery of the added ^{15}N (Figure 5). Recovery of added ^{15}N

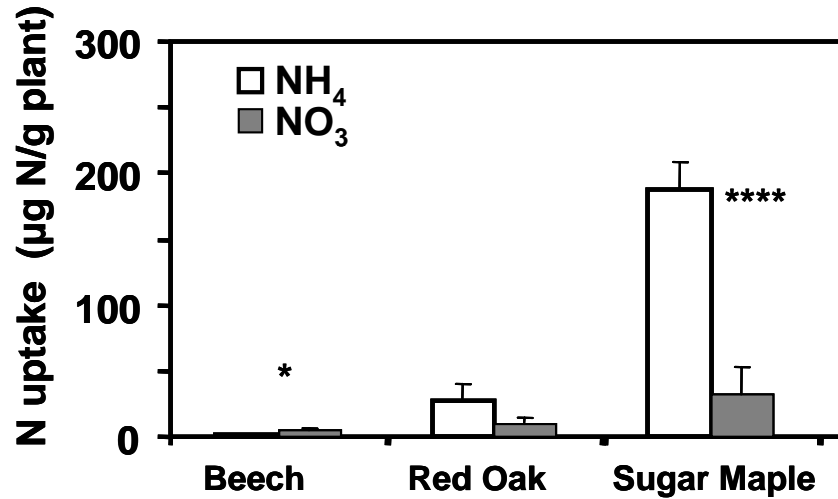


Figure 3. Plant uptake and sequestration of NH₄-N and NO₃-N per g plant biomass (n=4 for beech and n=5 for red oak, and sugar maple seedlings). Within a tree species, differences in NH₄-N vs NO₃-N uptake are denoted by the following: * p<0.10, ** p<0.05, *** p<0.01, and **** p<0.005, after Templer and Dawson 2004.

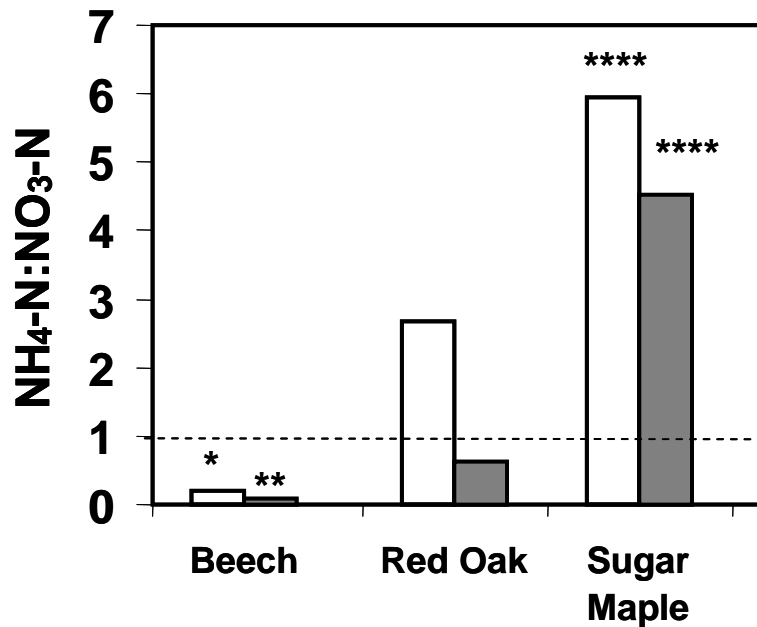


Figure 4. Ratio of N taken up as NH₄ vs NO₃ by plants. Open bars are the ratio of total plant uptake of NH₄-N:NO₃-N per g plant biomass (n=4 for beech and n=5 for red oak and sugar maple seedlings). Solid black bars show ratio of total plant uptake of NH₄-N:NO₃-N per g plant biomass per total µg NH₄-N/NO₃-N in pot. Within a tree species, differences in NH₄ vs NO₃ uptake are denoted by the following: *p<0.1, **p<0.05, ***p<0.01, and ****p<0.005. Dashed line denotes equal uptake of NH₄-N and NO₃-N, after Templer and Dawson 2004.

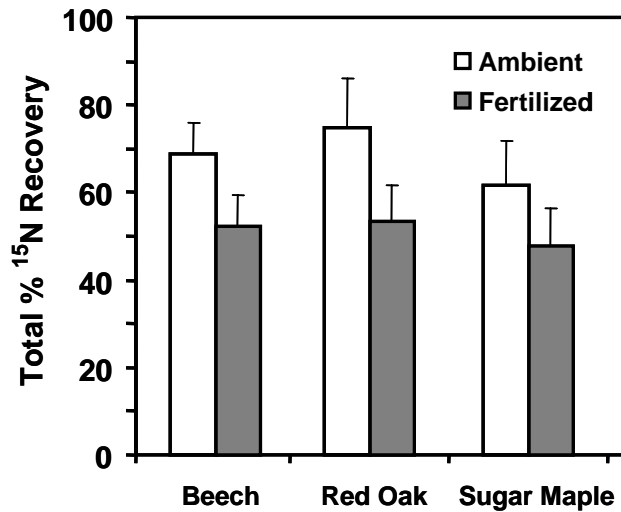


Figure 5. Fertilization reduced total % ^{15}N retention ($p=0.02$) at day 300. Includes surface O horizon, fine roots, any mineral soil with 12 cm depth and aboveground biomass (means with standard errors, after Templer et al. 2005).

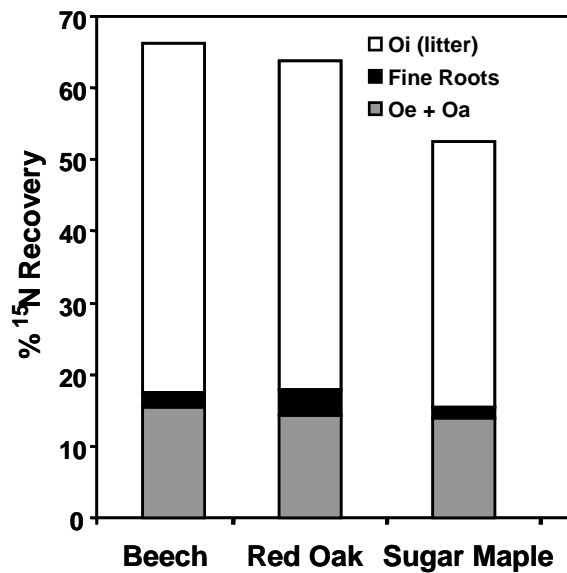


Figure 6. Recovery of ^{15}N within the forest floor (Oi, Oe+Oa, and fine roots) of ambient plots (no N fertilizer) at day 300 (means May 2000). Beech and red oak differed from sugar maple ($p<0.05$, after Templer et al. 2005).

ranged from 62-75% in the unfertilized ambient plots to 48-61% in the fertilized plots.

The O horizon (Oi+Oe+Oa, with fine roots included) was the dominant sink for ^{15}N in all plots, and recovery of ^{15}N in the O horizon differed among the species (Figure 6). Sugar maple plots retained 53% of the tracer N, compared with 63 to 66% retention in soils from the other species.

DISCUSSION

Soils dominated by different tree species varied in capacity to retain N in these Catskill Mountain forests, both indirectly through their effect on microbial processing of N, and directly through plant uptake and sequestration (Figure 7). Soils beneath sugar maple cycle soil N rapidly (Frehlich et al. 1993, Templer et al. 2003, Lovett et al. 2004), with a substantial proportion of N leaching from the soil (Fitzhugh et al., *in preparation*). Some of this loss results from high rates of net nitrification, and low rates of plant uptake of NO_3^- . The most surprising result from these experiments was that although red oak forest stands retained a large portion of added N, they had the lowest capacity to retain larger additions of N (from fertilization). We need to understand not only how tree species currently retain N, but how they may respond to increases in N availability in the future.

Tree species influenced the size of the soil microbial biomass pool and soil C:N, but the patterns in these properties did not account for patterns in microbial processes and N transformations. Sugar maple and red oak had low microbial biomass compared to the other beech and birch, but this similarity in microbial biomass did not translate into similar rates of N transformation. Indeed, soils from under sugar maple had lower C:N, higher pools of NO_3^- and rates of potential net nitrification than the other three species. Microbes in sugar maple soils took up the highest amount of ^{15}N immediately and released it, whereas soil microbes from beneath the other four species continued to immobilize added ^{15}N throughout the experimental period. Properties other than soil C:N and microbial biomass need to be examined to better understand the factors controlling microbial N transformations in these forest soils.

The greenhouse experiment showed that sugar maple seedlings took up much more NH_4^+ than NO_3^- , and beech seedlings showed the opposite pattern. The results of the laboratory and greenhouse studies suggest that there is a combination of soil and plant processes that can help explain why sugar maple stands may export more N than other forested stands such as red oak. Net nitrification and soil pools of NO_3^- are highest within sugar maple soils. However, sugar maples are not able to take up a significant amount of NO_3^- , the form of N that is produced in the largest amount in their stands and is most likely to be leached. We also note that both sugar maple and beech are

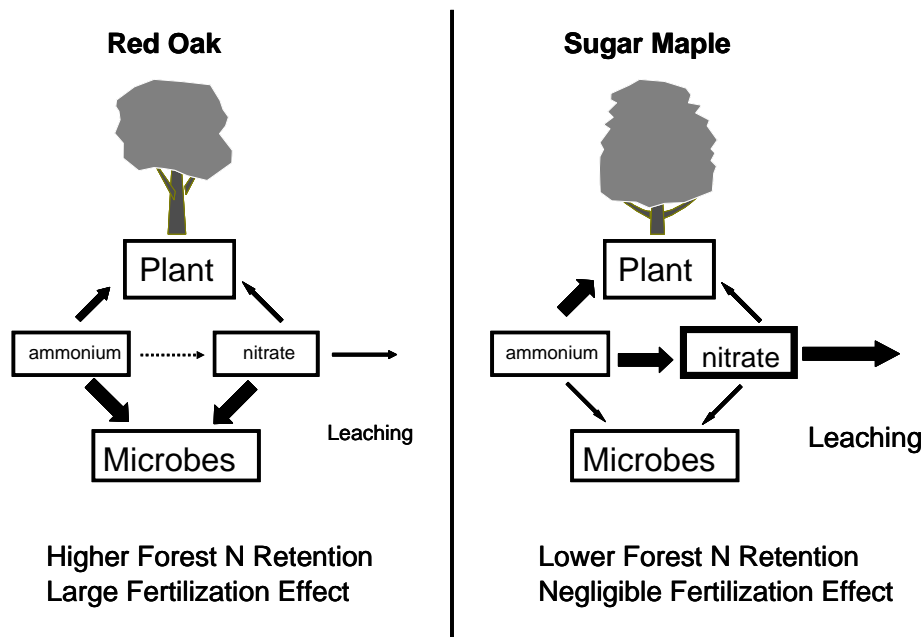


Figure 7. Conceptual model demonstrating potential mechanisms leading to greater forest N retention within red oak than sugar maple stands. Thicker arrows (indicating fluxes), boxes (indicating pools) and fonts indicate larger pool size or fluxes. Dashed lines represent non-detectable amounts of a particular flux. Sugar maple stands had higher nitrification rates, larger soil nitrate pools, less microbial uptake of N, six times greater plant uptake of ammonium relative to nitrate, and larger leaching losses of N (summary from data presented in this chapter, and cited references).

considered dominant components of mature (climax) northern hardwood forests, yet they each rely primarily on a different form of N; generalizations about patterns in form of N usage with succession would not be supported.

These differences among species in uptake of NO_3^- versus NH_4^+ may also be important in other forests in this region. For example, clearcutting northern hardwood forests in New Hampshire increases leaching losses of NO_3^- , and these losses decline sharply as forest regeneration develops (Likens et al. 1970, Pardo et al. 1995). Regeneration often includes a dominant component of pin cherry (*Prunus pennsylvanica* L.; Marks 1974, Bormann and Likens 1979), with rapid rates of N accumulation. Intriguingly, pin cherry has very low rates of NO_3^- reductase activity, indicating that its role in retaining ecosystem N probably comes from rapid uptake of NH_4^+ and not NO_3^- (Truax et al. 1994a, 1994b).

Similar to the laboratory and greenhouse experiments, tree species varied in forest retention of added N in a 300-day field ^{15}N experiment. Most of the added N was retained within the forest floor, but the magnitude of N retention varied among the soils from beneath different tree species. Sugar maple stands retained the least amount of N with current levels of N deposition,

while red oak stands retain the most. However, the fertilized treatment of this study shows that red oak stands may be more likely to have decreased forest N retention if availability increases in the future.

How long would it take for soils to reflect a change in overstory from red oak to sugar maple? The differences in these experiments resulted from an unknown period of soil development under the influence of each species, as well as any unknown differences in other site factors. If the effects of tree species are particularly strong, the change in soils (and N retention) should develop over a period of a few decades. Weaker species effects might take much longer to develop, and might differ substantially across site types.

These three experiments demonstrated that ecosystems dominated by different tree species in the Catskill Mountains varied in N retention, and these differences might change if N deposition changed. The species composition of these forests changed dramatically over the last 100 years in the Catskill Mountains, and these changes continue in response to climate changes, selective harvesting, and the spread of exotic pests and pathogens. Each of these environmental disturbances affects tree species differently, making it difficult to predict the future composition of these forests. For example, introduced beech bark disease (Houston et al. 1979; Griffin et al. 2003) and the hemlock wooly adelgid (Orwig and Foster 1998) have increased mortality of beech and hemlock across the northeastern United States. The dominance of sugar maple appears to be increasing in response to the decline of the other species (Griffin et al. 2003). This shift in species composition could increase N losses from forests, as beech and hemlock currently take up and retain a larger amount of N than the sugar maple stands that replace them. Other environmental changes could impair the increase in sugar maple, including climate change, acid rain and other introduced pests could (Lovett and Mitchell 2004). Future research is needed to address the interactions of multiple anthropogenic disturbances so that we can better predict which tree species will remain as dominants of northeastern United States forests. This information is necessary if we want to better manage these forests to retain the maximum amount of N and to understand the biogeochemical effects of changes in tree species composition.

REFERENCES CITED

- Aber J, Nadelhoffer K J, Steudler P and Melillo J M 1989 Nitrogen saturation in northern forest ecosystems. *BioScience* 39:378-386.
- Agren G I and Bosatta E 1988 Nitrogen saturation of terrestrial ecosystems. *Environ. Pollut.* 54:185-197.
- Bormann F H and Likens G E 1979 *Pattern and Process in a Forested Ecosystem: Disturbance, Development and the Steady State Based on the Hubbard-Brook Ecosystem Study*. Springer-Verlag, New York.

- Buchman N, Gerbauer G and Schulze E D 1996 Partitioning of ^{15}N -labeled ammonium and nitrate among soil, litter, below- and aboveground biomass of trees and understory in a 15-year old *Picea abies* plantation. *Biogeochemistry* 33:1-23.
- Burns D A, Murdoch P S, Lawrence G B and Michel R L 1998 Effect of groundwater springs on NO_3^- concentrations during summer in Catskill Mountain streams. *Water Resour. Res.* 34:1987-1996.
- Crabtree R C and Bazazz F A 1993 Tree seedling response of four birch species to simulated nitrogen deposition: ammonium vs nitrate. *Ecol. Appl.* 3:315-321.
- Driscoll C and Van Dreaseon R 1993 Seasonal and long-term temporal patterns in the chemistry of Adirondack lakes. *Water Air Soil Poll.* 67:319-344.
- Fenn M, Poth M, Aber J, Baron J, Bormann B, Johnson D, Lemly A, McNulty S, Ryan D and Stottlemeyer R 1998 Nitrogen excess in North American ecosystems: predisposing factors, ecosystem responses, and management strategies. *Ecol. Appl.* 8:706-733.
- Finzi A C, Van Breemen N and Canham C D 1998 Canopy tree-soil interactions within temperate forests: species effects on soil carbon and nitrogen. *Ecol. Appl.* 8:440-446.
- Fitzhugh R, Lovett G M, Weathers K C and Arthur M A. Canopy tree species and soil solution C, N, and P chemistry in northern hardwood forests of the Catskill Mountains, New York, USA. *In Prep.*
- Frelch L E, Calcote R R, Davis M B and Pastor J 1993 Patch formation and maintenance in an old-growth hemlock-hardwood forest. *Ecology* 74:513-527.
- Friedland A J, Hawley G J and Gregory R A 1988 Red spruce *Picea-rubens* sarg. Foliar chemistry in northern Vermont and New York USA. *Plant Soil* 105:189-194.
- Galloway J N, Schlesinger W H, Levy II H, Michaels A and Schnoor J L 1995 Nitrogen fixation: atmospheric enhancement-environmental response. *Global Biogeochem. Cy.* 9:235-252.
- Gharbi A and Hipkin C 1984 Studies on nitrate reductase in British angiosperms. I. A comparison of nitrate reductase activity in ruderal, woodland-edge and woody species. *New Phytol.* 97:629-639.
- Gilliam F S, Adams M B and Yurish B M 1996 Ecosystem nutrient responses to chronic nitrogen inputs at Fernow Experimental Forest, West Virginia. *Can. J. Forest Res.* 26:196-205.
- Griffin J M, Lovett G M, Arthur M A and Weathers K C 2003 The distribution and severity of beech bark disease in the Catskill Mountains, NY. *Can. J. Forest Res.* 33:1754-1760.
- Gundersen P, Emmett B A, Kjonaas O J, Koopmans C J and Tietema A 1998 Impact of nitrogen deposition on nitrogen cycling in forests: a synthesis of NITREX data. *Forest Ecol. Manag.* 101:37-55.
- Horsley S B 1988 Nitrogen species preference of *Prunus-serotina* Ehrh. And *Betula-Alleghaniensis* Britt. Seedlings. *Am. J. Bot.* Annual Meeting of the Botanical Society of America, Davis, California, USA, August 14-18, 1988.
- Houston D R, Parker E J and Lonsdale D 1979 Beech bark disease: patterns of spread and development of the initiating agent *Cryptococcus fagisuga*. *Can. J. Forest Res.* 9:336-344.
- Howarth R W 1988 Nutrient limitation of net primary production in marine ecosystems. *Annu. Rev. Ecol. Syst.* 19:89-110.
- Johnson D, Van Miegroet H, Lindberg S, Harrison R and Todd D 1991 Nutrient cycling in red spruce forests of the Great Smoky Mountains. *Can. J. Forest Res.* 21:769-787.
- Johnson D W 1992 Nitrogen retention in forest soils. *J. Environ. Qual.* 21:1-12.
- Kudish M 2000 *The Catskill Forest A History*. Purple Mountain Press, New York. 217p.
- Lawrence G B, Lovett G M and Baevsky Y H 2000 Atmospheric deposition and watershed nitrogen export along an elevational gradient in the Catskill Mountains, New York. *Biogeochemistry* 50:21-43.
- Likens G E, Bormann F H, Johnson N M, Fisher D W and Pierce R S 1970 Effects of Forest Cutting and Herbicide Treatment on Nutrient Budgets in the Hubbard Brook Watershed-Ecosystem. *Ecol. Mon.* 40, 23-47.
- Lovett G M and Rueth H 1999 Soil nitrogen transformations in beech and maple stands along a nitrogen deposition gradient. *Ecol. Appl.* 9:1330-1344.

- Lovett G M, Weathers K C and Sobczak W V 2000 Nitrogen saturation and retention in forested watersheds of the Catskill Mountains, New York. *Ecol. Appl.* 10:73-84.
- Lovett G M, Weathers K C and Arthur M A 2002 Control of nitrogen loss from forested watersheds by soil C:N ratio and tree species composition. *Ecosystems* 5:712-718.
- Lovett G M, Weathers K C, Arthur M A and Schultz J C 2004 Nitrogen cycling in a northern hardwood forest: do species matter? *Biogeochemistry* 67:289-308.
- Lovett G M and Mitchell M J 2004 Sugar maple and nitrogen cycling in forests of eastern North America. *Front. Ecol. Environ.* 2:81-88.
- MacDonald J A, Dise N B, Matzner E, Armbruster M, Gundersen P and Forsius M 2002 Nitrogen input together with ecosystem nitrogen enrichment predict nitrate leaching from European forests. *Global Change Biol.* 8:1028-1033.
- Magill A H, Aber J D, Hendricks J J, Bowden R D, Melillo J M and Steudler P A 1997 Biogeochemical response of forest ecosystems to simulated chronic nitrogen deposition. *Ecol. Appl.* 7:402-415.
- McIntosh R P 1972 Forests of the Catskill Mountains, New York. *Ecol. Monogr.* 42:143-161.
- Melillo J M, Aber J D and Muratore J F 1982 Nitrogen and lignin control of hardwood leaf litter decomposition dynamics. *Ecology* 63: 621-626.
- Murdoch P S and Stoddard J L 1993 Chemical characteristics and temporal trends in eight streams of the Catskill Mountains, New York. *Water Air Soil Poll.* 67:367-395.
- Nadelhoffer K J, Downs M R, Fry B, Aber J D, Magill A H and Melillo J M 1995 The fate of ¹⁵N-labelled nitrate additions to a northern hardwood forest in eastern Maine, USA. *Oecologia* 103:292-301.
- Nadelhoffer K J, Downs M R and Fry B 1999 Sinks for ¹⁵N-enriched additions to an oak forest and a red pine plantation. *Ecol. Appl.* 9:72-86.
- National Atmospheric Deposition Program 2004. <http://nadp.sws.uiuc.edu/> (accessed December 10, 2004).
- Ollinger S V, Aber J D, Lovett G M, Millham S E and Lathrop R G 1993 A spatial model of atmospheric deposition for the northeastern United States. *Ecol. Appl.* 3:459-472.
- Orwig D A and Foster D R 1998 Forest response to the introduced hemlock woolly adelgid in southern New England, USA. *J. Torrey Bot. Soc.* 125:60-73.
- Pardo L H, Driscoll C T and Likens G E 1995 Patterns of nitrate loss from a chronosequence of clear-cut watersheds. *Water Air Soil Poll.* 85, 1659-1664.
- Pastor, J and Post W M 1986 Influence of climate, soil moisture, and succession on forest carbon and nitrogen cycles. *Biogeochemistry* 2: 3-27.
- Peterjohn W T, Adams M B and Gilliam F S 1996 Symptoms of nitrogen saturation in two central Appalachian hardwood forest ecosystems. *Biogeochemistry* 35: 507-522.
- Robertson G P 1982 Factors regulating nitrification in primary and secondary succession. *Ecology* 63: 1561-1573.
- Schulze E D 1989 Air pollution and forest decline in a spruce (*Picea abies*) forest. *Science* 244:776-783.
- Stewart G R, Joly C A, and Smirnov N 1992 Partitioning of inorganic nitrogen assimilation between the roots and shoots of cerrado and forest trees of contrasting plant communities of South East Brasil. *Oecologia* 91:511-517.
- Stoddard J L and Murdoch P S 1991 Catskill Mountains. In *Acidic Deposition and Aquatic Ecosystems: Regional Case Studies*. Ed. D F Charles. pp. 237-271. New York: Springer-Verlag New York.
- Stoddard J L 1994 Long-term changes in watershed retention of nitrogen. In *Environmental Chemistry of Lakes and Reservoirs*, Advance in Chemistry Series, Volume 237. Ed. L A Baker. pp. 223-284. American Chemical Society, Washington, D.C.
- Templer P, Findlay S and Lovett G 2003 Soil microbial biomass and nitrogen transformations among five tree species of the Catskill Mountains, New York, USA. *Soil Biol. Biochem.* 35:607-613.
- Templer P H and Dawson T E 2004 Nitrogen uptake by four tree species of the Catskill Mountains, New York: Implications for forest N dynamics. *Plant Soil* 262:251-261.

- Templer P H, Lovett G, Weathers K, Findlay S and Dawson T 2005 Tree species influence on forest nitrogen retention in the Catskill Mountains, New York, USA. *Ecosystems*, *in press*.
- Tritton L M and Hornbeck J W 1982 Biomass equations for major tree species of the northeast. General Technical Report NE-69. United States Department of Agriculture. Forest Service. Northeastern Forest Experiment Station.
- Truax B, Gagnon D, Lambert F and Chevrier N 1994a Nitrate assimilation of raspberry and in cherry in a recent clear-cut. *Can J Bot* 72, 1343-1348.
- Truax B, Lambert F, Gagnon D and Chevrier N 1994b Nitrate reductase and glutamine-synthetase activities in relation to growth and nitrogen assimilation in red oak and red ash seedlings – effects of N-forms, N-concentration and light-intensity. *Trees-Struct. Funct.* 9, 12-18.
- Van Valkenburgh N J 1996 Forest Preserve of New York State in the Adirondack and Catskill Mountains: A Short History. Purple Mountain Press, New York.
- Vitousek P M, Gosz J R, Grier C C, Melillo J M and Reiners W 1982 A comparative analysis of potential nitrification and nitrate mobility in forest ecosystems. *Ecol. Monogr.* 52:155-177.
- Vitousek P M, Aber J, Howarth R, Likens G, Matson P, Schindler D, Schlesinger W and Tilman D 1997 Human alteration of the global nitrogen cycle: source and consequences. *Ecol. Appl.* 7:737-750.
- Weathers K C, Lovett G M, Likens G E and Lathrop R 2000 Effect of landscape features on deposition to Hunter Mountain, Catskill Mountains, New York. *Ecol. Appl.* 10:528-540.
- West J A, Findlay S E G, Burns D A, Weathers K C and Lovett G M 2001 Catchment-scale variation in the nitrate concentrations of groundwater seeps in the Catskill Mountains, NY. *Water Air Soil Poll.* 132:389-400.
- Zak D R, Pregitzer K S and Host G E 1986 Landscape variation in nitrogen mineralization and nitrification. *Can. J. Forest Res.* 16:1258-1263.
- Zak D R and Pregitzer K S 1990 Spatial and temporal variability of nitrogen cycling in Northern lower Michigan. *Forest Sci.* 36:367-380.

