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Organic matter manipulations have little effect on gross and net nitrogen transformations in two temperate forest mineral soils in the USA and central Europe☆

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Abstract

Soil nitrogen transformations are intricately linked to carbon transformations. We utilized two existing organic matter manipulation sites in western Oregon, USA and Hungary to investigate these linkages. Our questions were: (1) what effect does the quantity and quality of organic matter have on net and gross N processing in mineral soil? and (2) do these effects vary across sites with very different climate, soils, vegetation, and N deposition status? The organic matter manipulations had small if any effects on gross and net N cycling rates. Gross N cycling rates under low N deposition increased with increasing soil C and N, but C:N ratio had no correlation with gross N cycling rates. Soil ammonium concentrations under high N deposition, however, were higher in the organic matter manipulation plots without roots and lower in plots with double litter, indicating a tree root effect and a litter immobilization effect, respectively, but did not differ significantly under low N deposition. Net ammonium production was lowest in the litter removal and root removal plots and highest in the litter addition plots at both sites. Gross and net N cycling rates and mineral nitrogen pool sizes were generally higher under higher N deposition and lower C:N ratio soil, which was consistent with past studies over gradients of N deposition. By looking at organic matter manipulations in two very

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different sites we gained some insight into the role that C:N ratio as well as total C and total N have in controlling N and C cycling in forest soils.

Keywords: ^{15}N; Carbon; Gross nitrogen mineralization; Litter addition; Litter removal; DIRT

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1. Introduction

Many studies have examined the effects that individual carbon (C) or, nitrogen (N) compounds as well as various complex organic matter additions have on net N cycling rates yet less is known about the effects of C and N manipulations on gross N cycling. Gross rates measure the one-way transformations of N, while net rates are determined by changes in pool size over time and reflect the summation of many gross rates. In some studies, especially in high N deposition sites, gross N mineralization has been negatively correlated with C:N ratios (Gundersen et al., 1998; Ross et al., 2004), while others have found no effects (Fisk and Fahey, 2001) and still others have found increasing gross rates with increasing C:N ratio (Parfitt et al., 2003). This discrepancy indicates that the C:N ratio might not always be a consistent controlling factor of gross N cycling rates. Perhaps other factors such as the total amount of organic matter can influence N cycling.

The Detritus Input and Removal Treatments (DIRT) project is an international network of field-based organic matter manipulations that were designed to assess how rates and sources of plant litter inputs control the accumulation and dynamics of organic matter and nutrients in soils (Nadelhoffer et al., 2000). Treatments consist of doubling leaf litter, doubling woody litter, removing all litter, trenching to remove roots, and removing litter combined with removing roots. As applied research plots, these treatments could provide indication of potential results of forest management practices or future climate change scenarios, but fundamentally these manipulations provide an excellent opportunity to examine some factors controlling N cycling in forest soils. We had two primary questions: (1) what effect does the quantity and quality of organic matter have on net and gross N processing in mineral soil? and (2) do these effects vary across sites with very different climate, soils, vegetation, and N deposition status?

Nitrogen transformations in soil are closely linked to the C content and the C quality of the soil. The influence that C has on N cycling, and vice versa, is derived primarily from the complementary role that these nutrients play in supporting soil microbial populations. Microbes need both C and N for growth and metabolism and a scarcity of either will limit their activity. If microbes are C limited, soil mineral N pools should be large and when available C increases (e.g. following addition of labile C or high C:N material) then mineral N pools should decrease as microbes immobilize N (on a net basis). If N is added (as mineral N or low C:N material) to C limited microbes mineral N pools should increase because of the lack of demand for N. If microbes are instead N limited, soil mineral N pools should start out small and any increase in available C should not affect available N concentrations. Adding N to N limited soil microbes should increase microbial activity, but mineral N pools should remain small until the microbial demand for N eases. Additions of C to soil normally lead to net N immobilization or at least lower net N mineralization rates (e.g. Schimel et al., 1992; Magill and Aber, 2000), giving some indication that soils are generally available-C limited. Carbon-limited microbes might be overwhelmingly common especially in agricultural soils, but at least some soil microbial populations show signs of N limitation especially when net N cycling rates are examined (Schimel and Weintraub, 2003).

We hypothesized that mineral N pools would decrease as organic matter additions increase and net rates would tend towards more N immobilization because new available C would drive heterotrophic microbes to take up mineral N. Likewise, removing litter and roots should cause an increase in the mineral N pool sizes over time as the lack of available C eases heterotrophic demand for N, and because plants no longer take up N. We hypothesized that gross N mineralization would increase as more organic substrate was added and decrease as substrate was removed, because when more organic material is present more substrate should be available to be colonized and decomposed and thus more N should be mineralized assuming similar litter chemistry. Gross ammonium consumption should also follow a similar trend because as more (or less) C is available there should be more (or less) demand for N by microbes. All of these hypotheses assume that litter quality is similar, there just would be more or less of the same kind of material.

2. Materials and methods

2.1. Site descriptions

The Síkfőkút Experimental Forest International LTER site (SIK) is near Eger, Hungary (47°90′N, 20°46′E). Total atmospheric N deposition at a nearby site was 19 kg N ha^{-1} y^{-1} in 1996–1998 (Horváth, 2004), and has been as high at 22.62 kg N ha^{-1} y^{-1} in the mid-1970's (Jakucs, 1985). SIK is dominated by *Quercus petraea* (Mattuschka) Liebl. (sessile oak) and *Q. cerris* L. (turkey oak) and is 320–340 m in elevation. Mean annual (1973–1978) temperature was 9.7 °C. Average annual (1973–1978) precipitation, primarily as rain, was 68.2 cm. Soils at this site have been described as a brown earth with Cambisol feature and brown forest soils with clay illuviation and Luvisol features. Bulk density of surface soils at this site average 1.4 Mg m^{-3} (Jakucs, 1985). Additional related site data can be found in Jakucs (1985).

The H.J. Andrews Experimental Forest LTER site (HJA) is near Blue River, Oregon, USA (44°14′N, 122°14′W). Atmospheric N deposition at HJA is less than 2 kg-N ha^{-1} y^{-1} (Sollins et al., 1980). This old-growth, mid-elevation (550 m) site is dominated by large *Pseudotsuga menziesii* (Mirb.) Franco (Douglas-fir) with mid-size *Tsuga heterophylla* (Raf.) Sarg. (western hemlock) and *Thuja plicata* Donn ex D. Don (western red cedar) in the overstory. Mean annual (1980–2000) temperature is 9.2 °C. Average annual (1980–2000) precipitation was 224 cm (S.E. = 10 cm), primarily as rain or intermittently melting snow. Soils at this site have been classified recently as coarse loamy mixed mesic Typic Hapludands. Bulk density of the surface soils at this site averaged 0.8 Mg m^{-3} (Holub and Lajtha, 2004). Additional related site data can be found in Henshaw et al. (1998).

These two sites were selected for two primary reasons: (1) both sites had active litter manipulation experiments and (2) the sites differed in a variety of ways, including N deposition, soil type, vegetation type, etc. so where similarities were observed they

Table 1
Explanation of treatments

Treatment name	HJA manipulation (Mg ha^{-1} y^{-1})	SIK manipulation (Mg ha^{-1} y^{-1})[a]	Description
2x wood	+2.7[c]	+0.8	The total amount of annual woody litter input was doubled
2x litter	+2.6[d]	+3.7	The total amount of annual non-woody litter (leaf and needle) input was doubled
Control	0	0	No manipulation
No litter	−5.3[e]	−4.6	All above ground inputs were removed[b]
No roots[f]	−1.3[g]	−1.2	Trenched and lined outside boarder of plots to 1 m depth to exclude root ingrowth
No inputs	−6.5[h]	−5.8	Combined no litter and no roots treatment

[a] Based on annual averages from Jakucs (1985).

[b] Because of the nature of the fine litter (needles at HJA and broadleaves at SIK), plots at HJA were covered with fine screen and litter fall was swept while plots At SIK were raked. Woody litter was also removed.

[c] Regional average coarse woody inputs (Sollins, 1982).

[d] Measured on site.

[e] Sum of fine and woody litter.

[f] Mass values do not include mycorrhizal fungi or root exudates that would increase total below ground allocation, thereby leading to a larger biomass removal when roots are excluded.

[g] Estimated as about one half of aboveground fine litter.

[h] Sum of no root and no litter (before rounding to one decimal place).

might be expected to apply across many forest ecosystems.

2.2. Experimental procedure

At both sites litter and root inputs were manipulated (Table 1) on three replicate plots for each manipulation at each site. Plots at SIK were 7 m by 7 m and were implemented in 2000. Plots at HJA were 10 m by 15 m or equivalent area and were implemented in 1997.

A composite sample of approximately 500 g of 0–10 cm A-horizon mineral soil (excluding all fresh litter and organic horizon) was collected from 4 to 5 locations in each plot at SIK on 24 April, 2002, and each plot at HJA on 30 April, 2002 and homogenized. We elected to use homogenized composite soil from each plot rather than intact cores, because our primary interest was in treatment and site differences and not in N cycling rates of undisturbed soil. Compositing and homogenization allowed for a single representative sample to be collected from each plot, which should reduce sample variation and the need for a large number of samples. Only shallow mineral soil was used because that was where we expected to see the greatest influence from surface litter manipulations due to its close proximity to the manipulated organic matter, as opposed to deeper soil.

While on site a 50–100 g subsample of the composite soil to be used for final net ammonium production and net nitrification rates was placed in a plastic bag, sealed, and buried in one of the sampling holes in each plot ($n = 18$ per site). The remaining soil from SIK was brought to the University of Debrecen and soil from HJA was brought to Oregon State University and placed in a refrigerator (4 °C) overnight prior to further preparation and analysis beginning the following day.

Initial soil ammonium and nitrate concentrations, and the initial values for net ammonium production and net nitrification rates were determined the following morning by extracting soil with 2 M KCl (5:1 solution:soil mass). Gravimetric soil moisture was determined on a subsample of each soil by drying at 60 °C to a constant mass. After 26 days of field incubation the soil that was buried in the field was collected and analyzed in the same way (Robertson et al., 1999).

Gross N mineralization and gross ammonium consumption rates were determined in the remaining originally-collected soil the day following collection. Once the soil was allowed to warm to room temperature (22 °C), a subsample of approximately 200 g dry-soil-equivalent composite soil from each plot was added to a plastic bag, which served as the incubation container for the gross N mineralization measurements. A 6 ml aliquot of a 100 mg-N L^{-1} solution of 98 at.% ^{15}N-ammonium chloride ($^{15}NH_4Cl$) was added to the soil in each bag which equaled about 3 mg-^{15}N kg soil^{-1}. The soil was mixed to distribute the added ^{15}N evenly and allowed to incubate at room temperature. An 'initial' sample was collected from each bag after 1.0 hr of incubation and was extracted immediately with 2 M KCl. An additional subsample was taken at this time for soil moisture. Twenty-four hours after the initial extraction a 'final' subsample from each bag was extracted with 2 M KCl.

All KCl extracts were filtered through pre-rinsed Whatman 1 filter paper, returned to Oregon State University, and frozen for storage prior to ammonium, nitrate, and ^{15}N-ammonium analyses. Ammonium concentrations were determined on all KCl extracts using a modified Berthelot reaction on an Orion Scientific AC 100 colorimetric autoanalyzer. Nitrate concentrations were determined using a copper/cadmium reduction column with sulfanilamide and N-1-naphthyl-ethylenediamine on an Orion Scientific AC 100 colorimetric autoanalyzer. Solution volume on all extracts was corrected for soil moisture content.

Extractable ammonium was prepared for ^{15}N analysis using ammonia diffusion to Teflon-enclosed acid traps as described by Stark and Hart (1996). Diffusion of all of the dissolved N forms was carried out for 7 days. Following drying over desiccant and concentrated sulfuric acid, the acid traps were wrapped in tin cups and submitted for ^{15}N and total N analysis at the U.C. Davis Stable Isotope Facility, Davis California using a Europa Scientific Integra continuous flow mass spectrometer.

Total C and N concentrations of oven-dried 0–10 cm mineral soil from all of the plots at both sites were determined. HJA soil was analyzed by the University of Georgia Stable Isotope Facility on a Carlo Erba Strumentazione, Milan NA1500 C/H/N Analyzer. SIK soil was analyzed by the Agiricultural Facility at the University of Debrecen, Hungary, using an

Table 2
Total C and N concentrations in 0–10 cm mineral soil

Site[a]		No inputs	No roots	No litter	Control	2x litter	2x wood	Site average
HJA	Total N (%)	0.22	0.20	0.18	0.18	0.22	0.26	0.21
	Standard error	0.01	0.02	0.02	0.01	0.04	0.08	0.02
SIK	Total N (%)	0.26	0.27	0.28	0.31	0.28	0.29	0.28
	Standard error	0.01	0.01	0.02	0.01	0.01	0.03	0.01
HJA	Total C (%)	7.6	5.5	4.4	5.0	6.7	7.7	6.2
	Standard error	0.9	0.6	0.7	0.5	1.2	3.2	0.6
SIK	Total C (%)	3.1	3.3	3.4	3.8	3.5	3.5	3.4
	Standard error	0.2	0.1	0.3	0.2	0.0	0.3	0.1
HJA	C:N ratio	33.0	26.9	24.6	26.5	30.2	26.1	28.0
	Standard error	2.3	0.2	1.8	1.8	0.8	3.0	1.0
SIK	C:N ratio	12.0	12.0	12.1	12.3	12.3	12.3	12.2
	Standard error	0.1	0.1	0.1	0.1	0.1	0.2	0.1

Total N was marginally lower at HJA than at SIK ($p = 0.07$). Total C and C:N ratio were significantly higher at HJA than at SIK ($p = 0.0002$ and $p < 0.0001$ respectively). Within sites treatments had no significant effects. See also Table 4.

[a] HJA: H.J. Andrews Experimental Forest, OR, USA; SIK: Síkfőkút Experimental Forest, Hungary, $n = 3$.

Elementar Analysensysteme GmbH, Vario EL. Different labs were used to eliminate the need for international transport of soil. As a quality assurance measure, results for both sites were compared with other published data (SIK, Jakucs, 1985; HJA, Holub and Lajtha, 2004) and found to be consistent.

2.3. Calculations and statistics

The net ammonium production and net nitrification rates were calculated using standard equations (Robertson et al., 1999) after 26 days of field incubation. Gross N mineralization rates and gross ammonium consumption rates were determined using pool dilution equations from Kirkham and Bartholomew (1954) and Davidson et al. (1991). Gross ammonium consumption can be overestimated by this method of calculation because the substrate pool is increased as a result of the ^{15}N addition required for the calculation. Consumption is the sum of all of the processes that remove NH_4^+ including microbial uptake, nitrification, volatilization and etc. The gross N mineralization rates more accurately represent un-amended rates because they rely on the pool dilution principle.

A general linear model (GLM) was used (SAS version 8.02) to test for differences between sites

Table 3
Initial concentrations of mineral N (mg N kg^{-1}) in 0–10 cm mineral soil

Site[a]	Concentration	No inputs	No roots	No litter	Control	2x litter	2x wood
HJA	NH_4^+ mean	1.5	0.9	0.9	1.0	1.1	0.7
	Standard error	0.3	0.7	0.3	0.4	0.2	0.4
SIK	NH_4^+ mean	11.8ab	16.7a	9.9b	2.7c	1.0c	2.1c
	Standard error	2.2	2.3	0.7	0.3	0.2	0.5
HJA	NO_3^- mean	0.9	1.4	0.37	0.4	0.3	0.4
	Standard error	0.2	1.0	0.03	0.2	0.3	0.2
SIK	NO_3^- mean	6.6	6.3	6.5	5.6	4.7	3.8
	Standard error	1.0	1.4	1.6	0.8	1.6	1.0

SIK > HJA for both ($p < 0.05$ GLM). Within sites, values that are different have different letters. See also Table 4.

[a] HJA: H.J. Andrews Experimental Forest, OR, USA; SIK: Síkfőkút Experimental Forest, Hungary, $n = 3$.

(categorical) and among organic matter manipulation treatments (categorical) including the manipulation by site interaction. When the GLM revealed significant differences among manipulations it was followed by Tukey's HSD to determine which manipulations differed from others within each site.

A correlation matrix was prepared for each site separately to test for relationships among organic matter manipulation (numerical, from Table 1); total C, total N, and C:N ratio (Table 2); ammonium concentration and nitrate concentration (Table 3); gross N mineralization rate; gross ammonium consumption rate; net ammonium production rate; net nitrification rate; and soil moisture content. Spearman's rank correlation coefficients were calculated for correlations involving organic matter manipulation because organic matter manipulation values are effectively discrete treatments. The remaining correlations were calculated using Pearson's correlation coefficients.

3. Results

3.1. Organic matter manipulation effects

Gross N mineralization rates and gross ammonium consumption rates were not significantly affected by organic matter manipulations at either site ($p > 0.05$, GLM, Fig. 1A and B). Nor were gross N transformation rates and the amount of organic matter added or removed correlated at either site (Table 4).

Neither net ammonium production nor net nitrification were significantly affected by the organic matter manipulations at either site ($p > 0.05$, GLM, Fig. 2A and B), but net ammonium production and the amount of organic matter added or removed were positively correlated at HJA (Table 4). A negative correlation between net nitrification and the amount of organic matter added or removed at SIK was revealed (Table 4). At HJA the sum of net ammonium production and net nitrification were often negative indicating net immobilization of mineral N.

Soil ammonium concentrations differed by treatment at SIK ($p < 0.0001$), but not at HJA ($p = 0.82$, Table 3). There was also a significant negative correlation between ammonium concentration and the amount of organic matter added or removed at SIK, which supports the results of the GLM, but this relationship was not significant at HJA (Table 4). Soil nitrate concentrations did not differ by treatment at either site ($p = 0.42$, GLM), but at SIK there was a marginally significant negative correlation between nitrate concentration and the amount of organic matter added or removed across manipulations; there was no such correlation at HJA (Table 4).

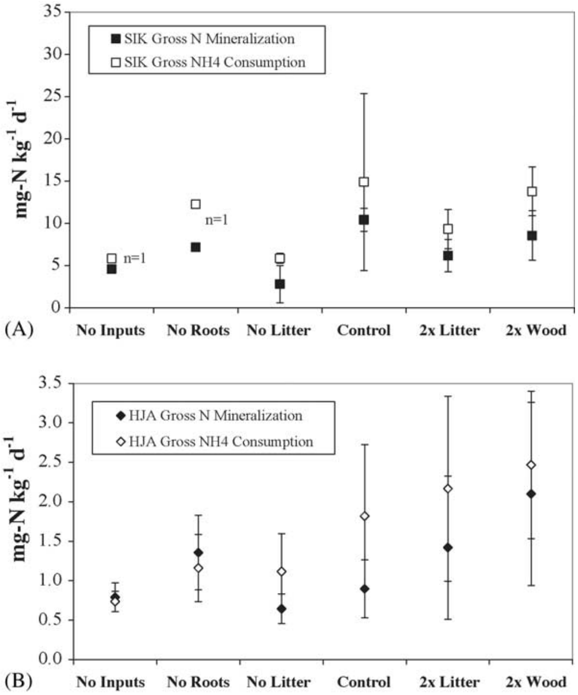

Fig. 1. Gross N cycling rates in 0–10 cm mineral soil from (A) SIK and (B) HJA. See Table 1 for treatment definitions. Note change in scale between sites. SIK > HJA for both ($p < 0.0001$, GLM). Within sites treatments not different ($p > 0.05$, GLM). See also Table 4; $n = 3$ per treatment unless noted. Error bars represent plus and minus one standard error.

Soil moisture was not significantly different across organic matter manipulations at HJA ($p = 0.2$, GLM) or with regression versus the amount of organic matter added or removed (Table 4). At SIK, however, there were differences among organic matter manipulations with the wetter soil occurring in treatments that excluded tree roots ($p = 0.007$, see Fig. 3), but the

Table 4
Correlation coefficients from 0 to 10 cm mineral soil

	Total C	Total N	C:N ratio	NH_4^+ conc.	NO_3^- conc	Net NH_4^+ produc.	Net nitrifica.	Gross N mineraliz.	Gross NH_4^+ consu.	Soil moisture
HJA										
OM manipulation	ns	ns	ns	ns	ns	0.55*	ns	ns	ns	ns
Total C		0.95**	0.64**	0.45^{+}	ns	ns	ns	0.74**	0.40^{+}	−0.41^{+}
Total N			ns	ns	ns	ns	ns	0.76**	0.44^{+}	ns
C:N ratio				0.51*	ns	−0.55*	ns	ns	ns	ns
NH_4^+ concentration					0.51*	ns	−0.56*	ns	ns	ns
NO_3^- concentration						−0.72**	ns	ns	ns	ns
Net NH_4^+ production							ns	ns	ns	ns
Net nitrification								ns	ns	ns
Gross N mineraliz.									0.83**	ns
Gross NH_4^+ consu.										ns
SIK										
OM manipulation	ns	ns	0.43^{+}	−0.80**	−0.40^{+}	ns	−0.61**	ns	ns	ns
Total C		0.98**	ns	ns	ns	ns	ns	ns	ns	ns
Total N			ns	ns	ns	ns	ns	ns	ns	ns
C:N ratio				−0.56*	ns	ns	−0.63**	ns	ns	ns
NH_4^+ concentration					0.43^{+}	−0.44^{+}	0.45^{+}	ns	ns	0.66**
NO_3^- concentration						−0.68**	ns	ns	ns	ns
Net NH_4^+ production							ns	ns	ns	ns
Net nitrification								ns	ns	0.52*
Gross N mineraliza.									0.70*	ns
Gross NH_4^+ consu.										ns

ns = not significant, $n = 18$ except for SIK Gross mineralization and consumption where $n = 14$. Note: Correlations involving OM manipulations used Spearman's rank method, all others use Pearson's method. HJA: H.J. Andrews Experimental Forest, OR, USA; SIK: Síkfőkút Experimental Forest, Hungary.

$^{+}$ $p < 0.10$.

* $p < 0.05$.

** $p < 0.01$.

differences were not systematic across the amount of organic matter added or removed so the correlation was not significant (Table 4).

Gross N transformation rates and net N transformation rates were never significantly correlated at either site (Table 4). At both sites there were positive correlations of total C versus total N, ammonium concentration versus nitrate concentration, gross N mineralization rate versus gross ammonium consumption rate and the negative correlation between net ammonium production rate versus nitrate concentration were similar across both sites. The correlation of C:N ratio versus ammonium concentration was positive at HJA, but negative at SIK while the correlation of net nitrification rate versus ammonium concentration was negative at HJA, but positive at SIK. Gross N mineralization and gross ammonium consumption were both positively correlated to total C and total N, at HJA, but not at SIK. C:N ratio was not correlated with gross N mineralization or gross ammonium consumption at either site.

3.2. Site effects

Gross N mineralization rates and gross ammonium consumption rates at SIK were five to ten times larger than at HJA ($p < 0.0001$ and $p = 0.0002$, respectively, Fig. 1A and B). Gross N mineralization was about 6.6 times higher than net ammonium production at SIK. Net ammonium production and net nitrification were significantly higher at SIK than at HJA ($p < 0.0001$ for both, Fig. 2A and B). Soil ammonium and nitrate concentrations were significantly higher at SIK than at HJA ($p < 0.0001$ for both, Table 3). Gravimetric soil moisture at SIK was about 50% less than HJA ($p < 0.0001$, Fig. 3).

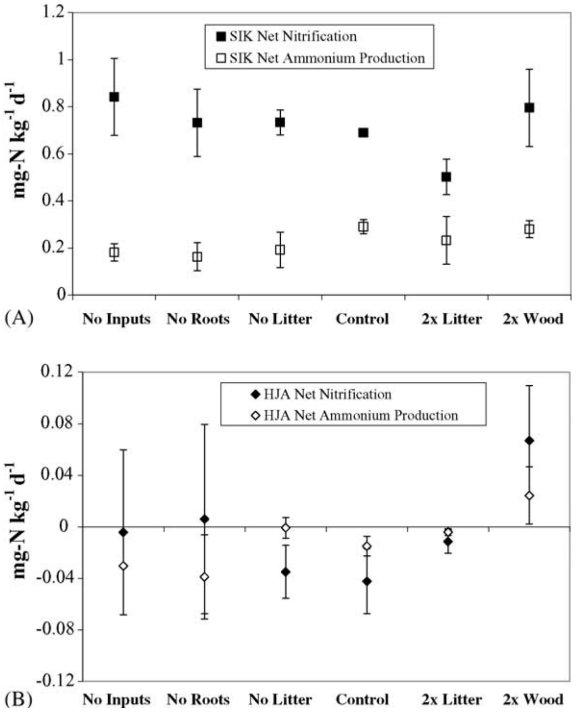

Fig. 2. Net N cycling rates in 0 to 10 cm mineral soil from (A) SIK and (B) HJA. See Table 1 for treatment definitions. Note change in scale between sites. SIK > HJA for both ($p < 0.0001$, GLM). Within sites treatments not different ($p > 0.05$, GLM). See also Table 4; $n = 3$ per treatment. Error bars represent plus and minus one standard error.

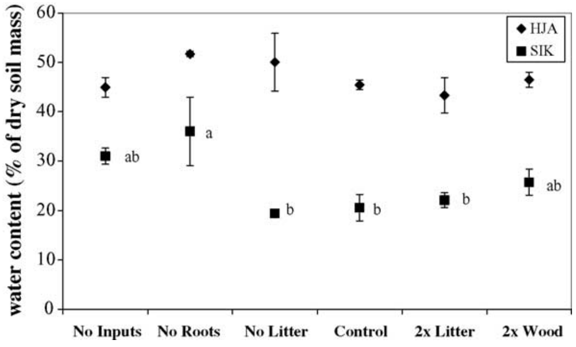

Fig. 3. Gravimetric moisture in 0 to 10 cm mineral soil from SIK and HJA. See Table 1 for treatment definitions. SIK < HJA ($p < 0.0001$). Within a site values that are different have different letters. See also Table 4; $n = 3$ per treatment. Error bars represent plus and minus one standard error.

4. Discussion

4.1. Organic matter manipulations

The weak or non-existent response in N cycling resulting from drastic litter and root manipulations indicates that N transformations in mineral soil are not strongly linked to new litter inputs at either site. Perhaps transport of new available organic matter to the mineral soil does not occur rapidly, so that the mineral soil does not ‘recognize’ the large pool of organic matter above it. Therefore organic matter present in the mineral soil probably drives the N processing in mineral soil because of the relative proximity. N cycling in the mineral soil of these two forests appears robust to changes in the amount of litter covering it. Given enough time there may be effects, as the correlations hint to, but the time scale at which litter and root manipulations affect gross and net N transformations in the mineral soil is probably on the order of decades rather then months or years.

Other studies testing effects of added N on N cycling rates often find varied effects of N addition, especially when not over a broad geographic area (Currie, 1999). Fisk and Fahey (2001) found no effect of N addition on gross N mineralization or gross ammonium consumption, but observed a reduction in net N mineralization. Scott et al. (1998) and Corre et al. (2003) observed decreases in gross N mineralization and gross ammonium immobilization rates following additional N fertilization. In both of the studies, as well as our current study, it is possible that other factors besides the amount of organic matter added to the surface of the soil are having greater impact on gross and net N transformation rates.

Results from the correlation matrices from both sites showed some similarity in their responses to litter manipulations. Significant correlations do not indicate causal relationships; they merely show associations and suggest areas for future study. The possibility for spurious correlations resulting from a third, known or unknown controlling factor exists so extreme caution should be used when interpreting results of correlation analysis. Non-significant correlations, however, do indicate a weak or non-existent relationship and therefore show that a causal relationship is probably not present. The absence of a significant correlation between any of the gross rates and any of the longer-term net rates was similar to Davidson et al. (1992) and Hart et al. (1994) and emphasizes the importance of measuring both gross and net processes, because gross rates and net rates are not equivalent measures of N cycling. Positive correlations between ammonium and nitrate concentration as well as between gross N mineralization rate and gross ammonium consumption rate were found at both sites and also by Davidson et al. (1992). These two correlations are probably common among other forest soil studies. The correlation between gross N mineralization rate and gross ammonium consumption rates is expected because they are complementary since they sum to the change in pool size over the incubation time. Net ammonium production and initial nitrate concentration were negatively correlated at both sites, but positively correlated in Davidson et al. (1992). Net nitrification and ammonium concentration were negatively correlated at HJA, but positively correlated at SIK and in Davidson et al. (1992). The reason for the discrepancy among the correlations found at the different sites and different studies is not clear.

4.2. *Site effects*

Site differences, including N deposition, soils (both types and C and N content), climate, and vegetation had a much larger effect on N cycling rates than the organic matter manipulations. SIK had higher net ammonium production, net nitrification, gross N mineralization and gross ammonium consumption rates than HJA. Higher net rates at SIK versus HJA follow the predictable and widespread pattern of decreasing net rates with increasing C:N ratio (e.g. Vestgarden et al., 2003). When C is available relative to N heterotrophic microbes can utilize N for growth, but as C levels decrease relative to N autotrophic microbes nitrify NH_4^+ (Hart et al., 1994).

The higher gross N cycling rates observed in SIK relative to HJA could be due to a lower C:N ratio, which indicates a relative abundance of N. Microbes require both C and N for growth and during organic matter decomposition both are made available. When soil C:N ratios are low microbes have enough N, but decompose organic matter to make C available leaving excess N as a by-product of decomposition. This is the process that appears to be occurring at SIK; microbes are C limited. However, when soil C:N ratios are high microbes have enough C, but decompose organic matter to make N available. This process appears to be occurring at HJA; microbes are N limited. Under C limited conditions microbes have enough N to produce decomposing enzymes and therefore decomposition and thus gross N cycling should be faster. Under N limited conditions, microbes do not necessarily have enough N to make all of the needed decomposition enzymes and decomposing and thus gross N cycling should occur more slowly. This is, of course, a simplistic model and there are other processes that can keep C limited sites from decimating soil C stores and N limited sites from halting decomposition altogether.

At HJA gross N mineralization and gross ammonium consumption increased as total C and N in the soil increased, but not as the C:N ratio increased. This was consistent with our hypothesis that as more substrate is made available more mineral N production and more mineral N consumption will occur if C:N ratio differences are small. These processes balanced each other, however, to keep net rates across organic matter manipulations near zero. This indicates that soil microorganisms are probably very N limited in the C rich forest soil at HJA. Microbes release N based on the total amount of material available to decompose if litter quality is similar. Thus total C and total N should have a larger influence on the rates in these organic matter manipulations which have more-or-less similar C:N ratios, yet vary greatly in the amount of organic matter manipulated. This was supported at HJA.

Organic matter removal at SIK led to increases in mineral N pools, although no strong differences in N cycling rates were found across any of the manipulations. High concentrations of ammonium and nitrate in the removal plots indicate that the soil microbes at SIK are strongly C limited, because as the source of C (i.e. litter and roots) was removed microbes no longer could utilize available N. However since trees roots are no longer present in the no roots and no inputs plots the absence of plant N uptake might also be affecting mineral N pools. The effect of the missing tree roots can also be seen in the moisture data where plots without roots at SIK were significantly wetter than plots with roots. The role of the trees in removing mineral N is supported by the gross N mineralization and gross ammonium consumption rates where no differences among organic matter manipulations at

SIK were found. This indicates that microbes are producing and consuming ammonium at a similar rate in all organic matter manipulations, leaving the lack of trees, and their associated root exudates and mycorrhizal symbionts, as the cause of mineral N accumulation. The higher soil moisture in SIK plots without roots further substantiates the role that roots play in removing nutrients (and water) from the soil. In a study involving N fertilization and litter removal Fisk and Fahey (2001) also found little difference in gross N mineralization and gross N consumption rates among organic matter manipulations.

5. Conclusion

Litter and root manipulations had little or no effect on gross and net N cycling and N pools in the mineral soil of the two forests examined in this study despite the large magnitude of the organic matter manipulations. Additionally the C:N ratio in the mineral soil, which is a measure of soil quality, had only small or non-existent effects on N cycling rates, because the C:N ratios of the mineral soil within each site were not drastically different. Although many of the differences observed among the DIRT treatments were not large, we anticipate the emergence of stronger patterns as the plots age and manipulation continues.

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