Biogeochemistry (2009) 95:355–371
DOI 10.1007/s10533-009-9328-6

Controls of nitrogen isotope patterns in soil profiles

Erik A. Hobbie · Andrew P. Ouimette

Received: 12 January 2009 / Accepted: 24 April 2009 / Published online: 26 May 2009

Abstract To determine the dominant processes controlling nitrogen (N) dynamics in soils and increase insights into soil N cycling from nitrogen isotope (δ^{15}N) data, patterns of ^{15}N enrichment in soil profiles were compiled from studies on tropical, temperate, and boreal systems. The maximum ^{15}N enrichment between litter and deeper soil layers varied strongly with mycorrhizal fungal association, averaging 9.6 ± 0.4‰ in ectomycorrhizal systems and 4.6 ± 0.5‰ in arbuscular mycorrhizal systems. The ^{15}N enrichment varied little with mean annual temperature, precipitation, or nitrification rates. One main factor controlling ^{15}N in soil profiles, fractionation against ^{15}N during N transfer by mycorrhizal fungi to host plants, leads to ^{15}N-depleted plant litter at the soil surface and ^{15}N-enriched nitrogen of fungal origin at depth. The preferential preservation of ^{15}N-enriched compounds during decomposition and stabilization is a second important factor. A third mechanism, N loss during nitrification and denitrification, may account for large ^{15}N enrichments with depth in less N-limited forests and may account for soil profiles where maximum δ^{15}N is at intermediate depths. Mixing among soil horizons should also decrease differences among soil horizons. We suggest that dynamic models of isotope distributions within soil profiles that can incorporate multiple processes could provide additional information about the history of nitrogen movements and transformations at a site.

Keywords Nitrogen isotopes · Soil horizons · Isotopic fractionation · Modeling · Mycorrhizal fungi · Soil mixing · Denitrification

Introduction

Many processes in the nitrogen (N) cycle of terrestrial soils can alter the ^{15}N:^{14}N ratios of source and sink pools (Table 1). These ratios (expressed as δ^{15}N values) in soils can accordingly be used to understand dominant sources, sinks, and processes of N in different biogeochemical reservoirs. Because surface litter and fresh belowground inputs are relatively labile compared to organic matter in deeper soil horizons, differences in ^{15}N between litter layers and deeper horizons reflect the sum total of N transformations and N movements over decades to centuries. δ^{15}N usually increases with depth in forest and grassland soils (Fig. 1a; Nadelhoffer and Fry 1988; Högberg 1997), although occasionally maximum δ^{15}N is at an intermediate depth followed by a subsequent decline at greater depths (Fig. 1b).

Electronic supplementary material The online version of this article (doi:10.1007/s10533-009-9328-6) contains supplementary material, which is available to authorized users.

E. A. Hobbie (✉) · A. P. Ouimette
Complex Systems Research Center, University of New Hampshire, Durham, NH 03824, USA
e-mail: erik.hobbie@unh.edu

Table 1 Fractionations against ^{15}N associated with different soil N processes

Process	Fractionation (‰)	Source
N_2 fixation	−2 to 2	(1)
Assimilation	−1 to 1.6	(2)
Nitrification	12 to 35	(3), (1)
Denitrification	0 to 33, 26	(1), (4)
Ammonia volatilization	20 to 27	(1)
Mineralization	−1 to 1	(2)
Ion exchange	−1 to −8	(5)
Enzymatic hydrolysis	10 to 24	(6)
N transfer, ECM fungi to plant host	8 to 10	(6)
N transfer, AM fungi to plant host	0 to 3.5[a]	(7)

Positive values indicate that the reactant is enriched in ^{15}N (e.g., NH_4^+ in nitrification) and the product is depleted in ^{15}N (e.g., NO_3^- in nitrification). Sources: (1) Högberg (1997), (2) Kendall (1998), (3) Shearer and Kohl (1986), (4) Pörtl et al. (2007), (5) Hübner (1986), (6) Hobbie and Colpaert (2003), (7) Handley et al. (1999b)

ECM ectomycorrhizal, *AM* arbuscular mycorrhizal

[a] Based on maximum difference between nonmycorrhizal and mycorrhizal plants

Several mechanisms probably contribute to these patterns.

Studying nitrogen dynamics and isotopic patterns in the soil together may reveal which processes control nitrogen isotope patterns in the soil. However, the labile pools of ammonium, nitrate, and amino acids turn over quickly (Jones and Kielland 2002; Schimel and Bennett 2004) and therefore a snapshot of isotopic composition may poorly represent the long-term average signal. In addition, the intrinsic fractionation factors (Δ) for specific reactions of labile N are imperfectly known and the expression of that fractionation depends on the relative sizes of the product and reactant pools as well as competing biogeochemical reactions (Robinson 2001). As a consequence, the causes of soil $\delta^{15}N$ patterns are still quite uncertain.

Both the average $\delta^{15}N$ of bulk soil and the specific pattern of $\delta^{15}N$ within a soil profile may provide useful information on soil N cycling. As pointed out by Amundson et al. (2003), the $\delta^{15}N$ of bulk soil at equilibrium should reflect the $\delta^{15}N$ of inputs and the net fractionation against ^{15}N during losses. In contrast, patterns of $\delta^{15}N$ within a soil profile can reflect many different processes operating simultaneously. In this review, we focus on mechanisms

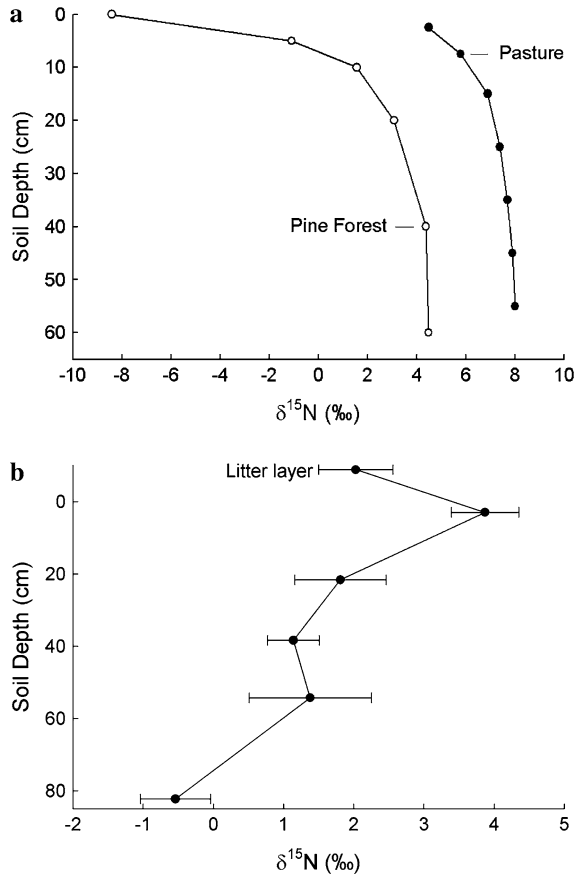

Fig. 1 **a** Representative soil profiles of $\delta^{15}N$ versus depth under pasture (AM-dominated) and pine forest (ECM-dominated) showing a consistent decline in $\delta^{15}N$ with depth. Pasture from Ledgard et al. (1984), pine forest from the United States Geological Survey (USGS unpublished); **b** Representative soil profile with maximum $\delta^{15}N$ at intermediate depths. Such profiles are common in arbuscular mycorrhizal systems and in sites of higher nitrogen availability. Data from Schuur and Matson (2001) for a Hawaiian forest dominated by *Metrosideros polymorpha*

influencing $\delta^{15}N$ distribution within soil profiles, although discussions of $\delta^{15}N$ distributions cannot be completely separated from bulk soil $\delta^{15}N$ values. Mechanisms of N export from soil and isotopic fractionation against ^{15}N during processes creating exported compounds will control isotopic signatures of bulk soil.

Three potentially important mechanisms influencing $\delta^{15}N$ values within soil profiles are given below:

1. Creation of ^{15}N-depleted N by mycorrhizal fungi and transfer of that ^{15}N-depleted N to plants (Hobbie et al. 2000). ^{15}N-depleted litter then

accumulates at soil surfaces through litterfall, and ^{15}N-enriched N derived from mycorrhizal fungi accumulates at depth (Högberg et al. 1996).
2. Creation of ^{15}N-depleted N as products of enzymatic hydrolysis (Silfer et al. 1992), ammonification, nitrification, or denitrification followed by leaching, gaseous losses, or uptake of that ^{15}N-depleted N by plants (Nadelhoffer and Fry 1988; Handley and Raven 1992; Houlton et al. 2007).
3. Mixing of soil N among different layers by bioturbation, windthrow, or other mechanical processes (Gabet et al. 2003).

Temperature and precipitation patterns can influence vegetation type and microbial activity, and therefore indirectly influence litter quality, the prevalence of different mycorrhizal associations (Read 1991), and soil nitrogen dynamics. These climatic factors can therefore influence ^{15}N patterns of bulk soil and vegetation (Handley et al. 1999a). For example, foliar $\delta^{15}N$ values have been correlated with climate across a wide range of study sites, with foliar $\delta^{15}N$ (and bulk soil $\delta^{15}N$) lower in colder, wetter areas compared to warmer, drier sites, and lower at temperate than at tropical sites (Handley et al. 1999a, Martinelli et al. 1999; Amundson et al. 2003). We might expect climatic factors to have a similarly large influence on ^{15}N patterns in soil profiles.

The two main types of mycorrhizal fungi, ectomycorrhizal fungi and arbuscular mycorrhizal fungi, differ in their spatial extent (Coleman et al. 2004), effects on plant $\delta^{15}N$ (Hobbie and Hobbie 2008), and enzymatic capabilities (Chalot and Brun 1998). They are generally adapted to different N dynamics, with arbuscular mycorrhizal fungi primarily taking up soluble nutrients whereas ectomycorrhizal fungi often possess enzymes to degrade complex organic compounds that arbuscular mycorrhizal fungi lack (Olsson et al. 2002). Partitioning of nitrogen isotopes has been reasonably well-studied in ectomycorrhizal systems and studied very little in arbuscular mycorrhizal systems.

This study will examine patterns of $\delta^{15}N$ with depth in terrestrial soils and compare those patterns with data on precipitation, temperature, nitrification rates, and mycorrhizal association. We will then discuss the main direct controls over soil ^{15}N patterns, such as fractionation against ^{15}N during creation of mycorrhizal transfer compounds, leaching of ^{15}N-depleted nitrate or dissolved organic nitrogen down soil profiles, mixing among soil layers by windthrow (Kramer et al. 2004) or bioturbation (Kaste et al. 2007), or gaseous losses via denitrification. Finally, we will synthesize the factors influencing N movement and ^{15}N fractionation within soil profiles into a conceptual model.

Methods

Small natural variations in the ratio of ^{15}N to ^{14}N have proven useful as markers of N cycling processes in soil profiles. To express these variations in a tractable form, ^{15}N:^{14}N ratios for samples are referenced against a universal standard and calculated as $\delta^{15}N$ values, defined as $\delta^{15}N_{sample} = [(^{15}N/^{14}N)_{sample}/(^{15}N/^{14}N)_{standard} - 1] \times 1{,}000$, with the standard the ^{15}N:^{14}N ratio of atmospheric N_2. The isotopic difference between two values is often expressed as $\varepsilon = \delta^{15}N_{sample1} - \delta^{15}N_{sample2}$. The discrimination against the heavy isotope (^{15}N) during chemical or physical processes is defined as $\Delta = (\delta^{15}N_{reactant} - \delta^{15}N_{product})/(1 + \delta^{15}N_{reactant})$.

For our analyses, data on $\delta^{15}N$ patterns in soil profiles were collected from published and unpublished sources. $\delta^{15}N$ data from the litter layer, organic horizon, and mineral horizon were necessary to be considered a profile. Data were obtained directly from authors or digitized from published figures. If available, data on mean annual precipitation, mean annual temperature, nitrification rates, and soil %N were also collected. The potential mycorrhizal association for the dominant vegetation of each site was determined from the available literature (e.g., Molina et al. 1992). To provide additional insight on the mechanisms causing soil ^{15}N profile patterns, data on isotopic patterns by soil particle size, density, or chemistry were also collected. Data from 88 soil profiles were collected from the literature, with sites in the USA (21 profiles), Europe (26 profiles), and Brazil (18 profiles) contributing much of the data.

Several previous studies have used the ^{15}N enrichment of deep soil relative to either plant foliage or surface litter as an integrated measure of nitrogen cycling processes in the soil profile (Garten 1993; Emmett et al. 1998; Amundson et al. 2003). Here, the maximum ^{15}N enrichment (ε_{soil}) from the litter layer

to 50 cm soil depth was calculated. This ^{15}N enrichment with depth was then compared to climatic factors, nitrification rates, and to the potential mycorrhizal association of the dominant vegetation at each site. Because litter layers were occasionally absent (particularly in the tropics), the maximum ^{15}N enrichment from foliage to 50 cm soil depth was also calculated. Multiple regressions of ^{15}N enrichment versus temperature and precipitation were done separately for sites dominated by ectomycorrhizal or arbuscular mycorrhizal symbioses.

Analytical and computational solutions to ^{15}N enrichment patterns in soil profiles

Several approaches have been developed that provide some insight into the pattern of ^{15}N enrichment in soil profiles. In a simple analytical approach to estimating the isotopic composition of soil and plants (expressed as ratios (R) $^{15}N/^{14}N$), Amundson et al. (2003) calculated that at steady-state the bulk soil isotopic ratio (R_s) could be expressed as the average isotopic ratio of all inputs (R_{inputs}) divided by the fractionation factor (α_{ex}) during N losses, with α_{ex} defined as the rate of a process for a heavy isotope (k_H) divided by the rate of a process for a light isotope (k_L).

$$R_s = R_{inputs}/\alpha_{ex}$$

This is strictly equivalent to $\delta^{15}N_{soil} = \delta^{15}N_{inputs} + \Delta_{ex}\,(1 + \delta^{15}N_{inputs})$, and for values of $\delta^{15}N_{inputs}$ close to 0‰ can be expressed as:

$$\delta^{15}N_{soil} = \delta^{15}N_{inputs} + \Delta_{ex}$$

If we assume that soil N is the ultimate source for plant N, then the isotopic ratio of plant N (R_p) can be simply expressed as reflecting the fractionation (α_p) going from the soil to the plant (Amundson et al. 2003).

$$R_p = \alpha_p R_s$$

This is equivalent to $\delta^{15}N_{plant} = \delta^{15}N_{soil} - \Delta_p$

These simple isotopic mass balance formulations are important to keep in mind when considering nitrogen isotope patterns in plants and soil. However, these equations do not provide of themselves any mechanistic insight into processes controlling isotopic composition. Fractionation factors in these equations must be considered as being averages for multiple processes.

An alternate approach has applied a Rayleigh equation to patterns of %N and $\delta^{15}N$ in soil profiles. The Rayleigh equation was first applied to soils by Mariotti et al. (1981) to study isotopic fractionation during denitrification but has primarily been used to examine unspecified N loss processes within soil profiles. In the Rayleigh equation, soil is treated as a semi-closed pool which can lose nitrogen as one moves down the soil profile but can not gain nitrogen. By plotting the log of the nitrogen concentration against the $\delta^{15}N$ in soils, a fractionation factor (Δ) can be calculated for the nitrogen loss, according to Eq. (1). The value of Δ (e.g., Table 2) may provide insight into the dominant mechanism of nitrogen loss. However, the assumptions of the Rayleigh equation make it difficult to interpret in many cases, as it requires: (1) a semi-closed system, (2) a single removal mechanism, and (3) declines in soil %N only from removal and not from dilution (e.g., admixture of additional mineral soil). With f defined as the fraction of N at depth relative to the surface N, the Rayleigh equation can be expressed as

$$\delta^{15}N_{depth} = \delta^{15}N_{surface} - \Delta \ln f. \quad (1)$$

We propose here an additional analytical approach incorporating two main factors, the discrimination against ^{15}N during creation of transfer compounds by mycorrhizal fungi and the discrimination against ^{14}N during formation of stable soil organic matter. The basis for this approach is an equation that integrates $\delta^{15}N$ patterns among plants, mycorrhizal fungi, and available N (Hobbie et al. 2000), in which Δ_f is the fractionation during creation of transfer compounds within mycorrhizal fungi and T_r is the proportion of N assimilated by mycorrhizal fungi that is transferred to host plants. If all plant N is derived from mycorrhizal fungi then Eq. (2) results, if only a fraction (f) is derived from mycorrhizal fungi, then Eq. (3) results.

$$\delta^{15}N_{plant} = \delta^{15}N_{available\,nitrogen} - \Delta_f \times (1 - T_r) \quad (2)$$

$$\delta^{15}N_{plant} = \delta^{15}N_{available\,nitrogen} - \Delta_f \times (1 - T_r) \times f \quad (3)$$

The equation for the mycorrhizal fungus is:

$$\delta^{15}N_{fungi} = \delta^{15}N_{available\,nitrogen} + \Delta_f \times T_r \quad (4)$$

We can combine Eqs. (3) and (4) to calculate the isotopic difference between fungi and plant

Table 2 Fractionation factors (Δ) estimated from soil $\delta^{15}N$ patterns

Environment	Method	Δ (‰)	Comments	Source
Ectomycorrhizal				
Pinus (7.5–15 cm)	Rayleigh	5.5	Time-series	Billings and Richter (2006)
Pinus (35–60 cm)	Rayleigh	9.7	Time-series	Billings and Richter (2006)
Pinus (soil)	Rayleigh	2.2 ± 0.4		Lindahl et al. (2007)
Pinus	Rayleigh	4.7 ± 0.3		USGS (unpublished)
Alnus/*Picea*	Modeling	8–10	Mycorrhizal transfer	Hobbie et al. (2000)
Nothofagus	Rayleigh	4.7, 7.9, 4.3		Boeckx et al. (2005)
Spruce	Rayleigh	3.4 ± 0.7		Mariotti et al. (1980)
Forests	Rayleigh	2.4, 5.2	High N deposition	Vervaet et al. (2002)
Pinus	Rayleigh	2.9	Plantation	Nadelhoffer et al. (2004)
Hardwood	Rayleigh	3.8		Nadelhoffer et al. (2004)
Arbuscular mycorrhizal				
Fields	Modeling	0 to −3.6	Active to stabilized	Baisden et al. (2002a)
Fields	Modeling	3.3 to −7.4	Active to passive	Baisden et al. (2002a)
Meadows	Rayleigh	2.5 ± 0.8		Mariotti et al. (1980)
Forests	Rayleigh	3.1 ± 1.1		Wada et al. (1984)
Cryptomeria	Rayleigh	1.7–3.3		Koba et al. (1998)
Hardwoods	Rayleigh	2.7 ± 0.3		USGS (unpublished)
Pasture	Rayleigh	1.3	Mineralization	Ledgard et al. (1984)
Nonmycorrhizal				
Hardwood litter	Rayleigh	2.6 ± 0.8	Inorganic N losses	Nadelhoffer and Fry (1988)
Desert soil	Rayleigh	8	NH_3 volatilization	Evans and Belnap (1999)
Arbuscular-ectomycorrhizal				
Araucaria-Nothofagus				
Unburned	Rayleigh	6.2		Boeckx et al. (2005)
Burned	Rayleigh	4.3		Boeckx et al. (2005)

Rayleigh fractionation factors either provided in reference or estimated from the following equation in regression analyses, $\delta^{15}N_{depth} - \delta^{15}N_{surface} = -\Delta \ln (\%N_{depth}/\%N_{surface})$

$$\delta^{15}N_{fungi} - \delta^{15}N_{plant} = \Delta_f \times (T_r + f - T_r \times f) \quad (5)$$

If we assume that $^{15}N_{plant} = \delta^{15}N_{litter}$, then

$$\delta^{15}N_{fungi} - \delta^{15}N_{litter} = \Delta_f \times (T_r + f - T_r \times f). \quad (6)$$

We assume that soil N can be conceptualized as consisting of fractions deriving from either plant N ($1 - f_f$) or mycorrhizal fungal N (f_f), and that further processing and loss results in an enrichment (ε_d) of the soil N found at depth relative to its ultimate origin.

$$\delta^{15}N_{soil} = f_f \times \delta^{15}N_{fungi} + (1 - f_f) \times \delta^{15}N_{plant} + \varepsilon_d \quad (7)$$

$$\delta^{15}N_{soil} - \delta^{15}N_{litter} = f_f \times \delta^{15}N_{fungi} + (1 - f_f) \times \delta^{15}N_{litter} - \delta^{15}N_{litter} + \varepsilon_d \quad (8)$$

$$\delta^{15}N_{soil} - \delta^{15}N_{litter} = f_f \times \left(\delta^{15}N_{fungi} - \delta^{15}N_{litter}\right) + \varepsilon_d \quad (9)$$

$$\delta^{15}N_{soil} - \delta^{15}N_{litter} = f_f \times \Delta_f \times (T_r + f - T_r \times f) + \varepsilon_d. \quad (10)$$

If $f = 1$, and ^{15}N enrichment during decomposition is relatively constant in deep soil horizons, then the relative contribution of plant-derived versus mycorrhizal-derived N to deep soil horizons can be

estimated from $\delta^{15}N$ patterns by the following equation:

$$\delta^{15}N_{soil} - \delta^{15}N_{litter} = f_f \times \Delta_f + \varepsilon_d. \quad (11)$$

The quantity f_f is the fraction of soil N derived from mycorrhizal fungal litter, Δ_f the fractionation during transfer of N from mycorrhizal fungi to host plants, and ε_d the cumulative enrichment in ^{15}N of stable soil organic N due to decomposition and loss processes. The use of a constant enrichment factor ε_d is one underlying assumption in this equation. The lack of large shifts in soil $\delta^{15}N$ with increasing depth beyond the first 30 cm or so in many cases is one argument that turnover of soil N and accompanying isotopic fractionation may slow down dramatically with declines in labile N. For the special case of f equal to one, Eq. (11) can be rearranged to estimate the relative importance to deep soil horizons of N derived from either mycorrhizal fungi (f_f) or plants ($1 - f_f$).

$$(1 - f_f) = \left(\delta^{15}N_{soil} - \delta^{15}N_{litter} - \varepsilon_d\right)/[(1 - T_r) \times \Delta_f] \quad (12)$$

$$f_f = \left(\delta^{15}N_{soil} - \delta^{15}N_{litter} - \varepsilon_d\right)/\Delta_f. \quad (13)$$

The value for Δ_f is constrained by estimates from computer and analytical modeling in field studies and culture studies to 8–10‰ (Hobbie et al. 1999, 2000, Hobbie and Colpaert 2003). The value for ε_d is less well-constrained. We use here a value of 3‰ based on the calculated enrichment factor during litter decomposition of 2.6 ± 0.8‰ (Nadelhoffer and Fry 1988).

Results

Sites were classified by the dominant mycorrhizal type of the vegetation, with 49 ectomycorrhizal sites, 33 arbuscular mycorrhizal sites, and six sites dominated by both ectomycorrhizal and arbuscular mycorrhizal plants. About 40% of the ectomycorrhizal sites were coniferous, 40% deciduous, and 20% mixed coniferous/deciduous. From the sites with typically arbuscular mycorrhizal associations, 64% were classified as broadleaf-evergreen, 18% mixed broadleaf-evergreen/coniferous, 9% coniferous, and 9% grassland. The sites with both ectomycorrhizal and arbuscular mycorrhizal vegetation were mainly from savannas (*cerrado*) in Brazil. The mean annual precipitation ranged from 600 to 7,000 mm/year and mean annual temperature ranged from 4.5 to 27.0°C, with higher temperature and precipitation for arbuscular mycorrhizal sites (average of 17.7°C and 2,230 mm) than for ectomycorrhizal sites (average of 10.6°C and 1,540 mm) (Fig. 2). Median precipitation was 2,250 mm/year for arbuscular mycorrhizal sites and 1,100 mm/year for ectomycorrhizal sites.

The climatic factors of mean annual precipitation and mean annual temperature were not strongly related to the maximum ^{15}N enrichment in soils (Figs. 3, 4). Temperature and precipitation together accounted for 16% of the variability in ^{15}N enrichment between litter and deep soil, and for 8% of the variability in ^{15}N enrichment between foliage and deep soil. In both cases temperature was the stronger control. Splitting the data by mycorrhizal type revealed that ^{15}N enrichment at arbuscular mycorrhizal-dominated sites was weakly and negatively correlated with precipitation ($r^2 = 0.17$, $p = 0.047$), with no other significant regressions (Table 3).

Across all studies, differences in $\delta^{15}N$ between the surface and deeper soil layers were uncorrelated with nitrification rates (Fig. 5) ($r^2 = 0.05$). This pattern contrasts with ^{15}N enrichment between foliage and deeper soil decreasing with increasing nitrification in several prior studies (Garten 1993; Garten and Van Miegroet 1994; Emmett et al. 1998; Pardo et al. 2002).

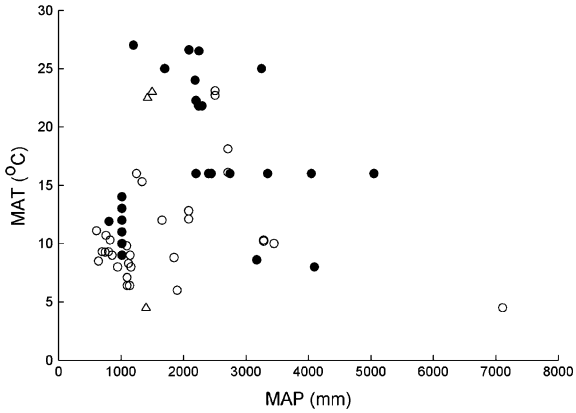

Fig. 2 Mean annual precipitation (MAP) versus mean annual temperature (MAT) for soil profiles studied. Symbols: *filled circles*, AM-dominated sites; *clear circles*, ECM-dominated sites; *clear triangles*, mixed AM/ECM sites. Studies are listed in the Appendix in Electronic Supplementary Material

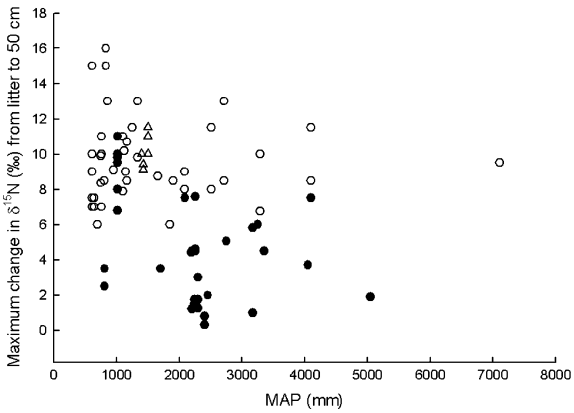

Fig. 3 Mean annual precipitation (MAP) does not correlate with the maximum change in δ^{15}N from the litter layer to 50 cm for soil profiles. Symbols: *filled circles*, AM-dominated sites; *clear circles*, ECM-dominated sites; *clear triangles*, mixed AM/ECM sites. Studies are listed in the Appendix in Electronic Supplementary Material

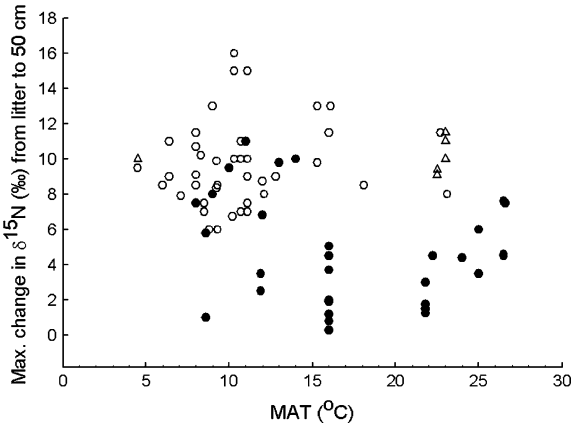

Fig. 4 Mean annual temperature (MAT) does not correlate with the maximum change in δ^{15}N from the litter layer to 50 cm for soil profiles. Symbols: *filled circles*, AM-dominated sites; *clear circles*, ECM-dominated sites; *clear triangles*, mixed AM/ECM sites. Studies are listed in the Appendix in Electronic Supplementary Material

Table 3 Statistics on a multiple regression of ^{15}N enrichment (ε_{soil}) versus climatic factors of mean annual precipitation (MAP) and temperature (MAT)

Mycorrhizal type	*n*	r^2	*P* (MAP)	*P* (MAT)
Arbuscular mycorrhizal	32	0.17	0.047	0.266
Ectomycorrhizal	45	0.02	0.984	0.347

The correlation (r^2) for the multiple regression is given, as well as the significance level (*P*) for the two independent variables

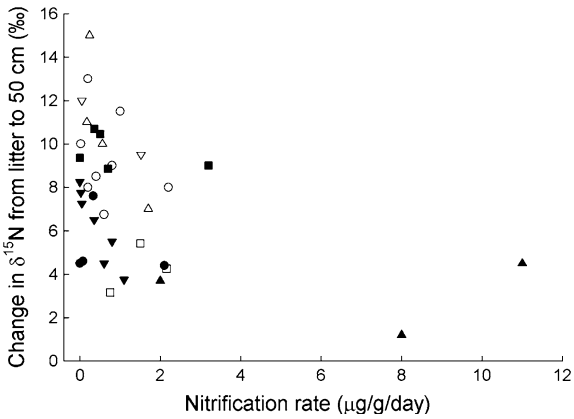

Fig. 5 Nitrification rate versus changes in ^{15}N from litter to 50 cm soil depth from four tropical and temperate sites. *Clear circles*, Kitayama and Iwamoto (2001); *clear upright triangles*, Vervaet et al. (2002); *clear upside-down triangles*, Pörtl et al. (2007); *clear squares*, Silver et al. (2000); *filled circles*, Nardoto (2005); *filled upright triangles*, Schuur and Matson (2001); *filled upside-down triangles*, Garten (1993); *filled squares*, Pardo et al. (2007). Two other studies compared δ^{15}N patterns with different units for nitrification rates; those results are given in the Appendix in Electronic Supplementary Material (e.g., Emmett et al. 1998)

In contrast to the small effects of nitrification and climate on δ^{15}N profiles, ^{15}N enrichment within soil profiles differed significantly between sites dominated by ectomycorrhizal versus arbuscular mycorrhizal symbioses (Fig. 1; Appendix in Electronic Supplementary Material). Sites with ectomycorrhizal plants were consistently (47 of 48) enriched in ^{15}N with increasing depth, whereas only about 60% of the sites dominated by arbuscular mycorrhizal plants were enriched in ^{15}N with depth. The other 40% of arbuscular mycorrhizal sites attained maximum δ^{15}N at some shallow depth and then declined in δ^{15}N in deeper soil layers. The ^{15}N enrichment from the litter layer to 50 cm was greater in ectomycorrhizal sites (9.6 ± 0.4‰) than in arbuscular mycorrhizal sites (4.6 ± 0.5‰).

Researchers have used several approaches to quantify fractionation factors (Δ) from ^{15}N patterns in soil profiles or during litter decomposition, of which the most common is the Rayleigh equation (Table 2). An alternative analytical framework to the Rayleigh equation was developed in the "Methods" section that stressed the role of mycorrhizal fungi in controlling δ^{15}N in soil profiles. In N-limited temperate and boreal forests dominated by ectomycorrhizal plants, fungal colonization of fine roots is high,

with most absorptive surfaces covered by a fungal sheath (Taylor and Alexander 2005; Guo et al. 2008). Under these conditions, most plant N is probably derived from mycorrhizal fungi and not from direct plant uptake (Wallenda et al. 2000; Hobbie and Hobbie 2008), so that Eq. (13) can be applied to estimate the relative importance of mycorrhizal fungi to soil profile development in $\delta^{15}N$. However, this approach will not be successful at high N availability if fractionations against ^{15}N during transformations of inorganic N are quantitatively important in determining N distributions within soil profiles. With values of 3 and 10‰ used for ε_d and Δ_f respectively, and values of 9.6 and 4.6‰ applied for the quantity $\delta^{15}N_{soil} - \delta^{15}N_{litter}$ for ectomycorrhizal and arbuscular mycorrhizal systems, respectively, then the proportion of deep soil N derived from mycorrhizal fungi (f_f) is about 70% for ectomycorrhizal systems and ~20% for arbuscular mycorrhizal systems.

Processes causing fractionation between the soil surface and deeper soils include loss of ^{15}N-depleted inorganic N from soil via nitrate leaching or ammonia volatilization, denitrification, or transfer of ^{15}N-depleted N from mycorrhizal fungi to host plants. Since no nitrogen is lost from the soil-plant system in the last mechanism, the overall signature of the soil-plant system will not change. If a single mechanism dominates removal, then insights into specific mechanisms of nitrogen removal are theoretically possible by applying the Rayleigh equation because fractionation against ^{15}N differs for the above mechanisms. Other approaches explaining $\delta^{15}N$ patterns within soil profiles have incorporated additional opportunities for fractionation, such as fractionation against ^{15}N during N movement between organic pools differing in lability (Baisden et al. 2002a). Values for Δ are generally higher in ectomycorrhizal sites (average ~5‰) than in arbuscular mycorrhizal sites (average ~2–3‰) (Table 2). A modeling study in ectomycorrhizal forests estimated a fractionation against ^{15}N during ectomycorrhizal transfer of 8–10‰ (Hobbie et al. 2000), whereas a modeling study in arbuscular mycorrhizal fields estimated fractionation during movement among different soil fractions of 0 to −7‰ (Baisden et al. 2002a).

Different disturbance processes can also alter $\delta^{15}N$ patterns in soil profiles. For example, burning surface layers often eliminates the most ^{15}N-depleted portion of the soil profile (Fig. 6), and other disturbances

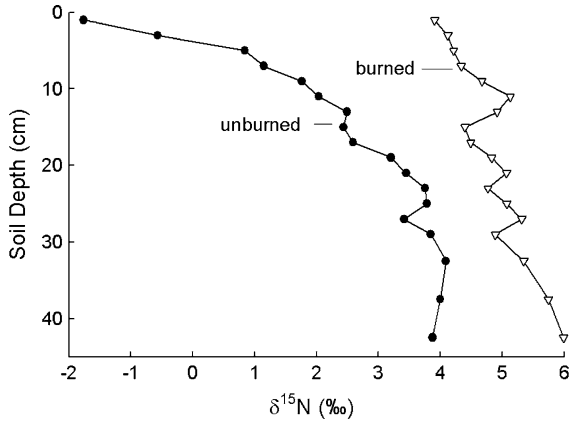

Fig. 6 The $\delta^{15}N$ soil profile at an unburned (*filled circles*) and burned (*open circles*) site in an old-growth *Araucaria-Nothofagus* in Chile. Disturbances such as burning, clear-cutting, and blowdowns appear to decrease the ^{15}N enrichment between the litter and deeper soil layers. Redrawn from Boeckx et al. (2005)

such as windthrow or clearcuts can also alter $\delta^{15}N$ patterns in soil profiles (Table 4).

Isotopic patterns in soil particles of different densities, sizes, surrounding vegetation, or chemical properties such as aliphaticity can also potentially reveal the causes of ^{15}N enrichment in soil profiles (Table 5). Grassland or arbuscular mycorrhizal sites sometimes have smaller increases in $\delta^{15}N$ with increasing density than ectomycorrhizal sites. In an early study in pastures, Ledgard et al. (1984) reported that sand, silt, and clay fractions differed in ^{15}N content, with clay about 4 and 3‰ enriched in ^{15}N relative to sand and silt, respectively. In this study, fractions increased on average 1.4‰ with depth. About half of the increase of bulk soil with depth arose from the 1.4‰ increase in $\delta^{15}N$ of individual fractions and half arose from large increases in the proportion of total soil N in the clay fraction with depth. Soils with more clay-sized particles are generally higher in $\delta^{15}N$ than sandy soils (Silver et al. 2000; Baisden et al. 2002b; Quideau et al. 2003).

Discussion

Researchers have proposed various direct (e.g., loss of ^{15}N-depleted nitrate) and indirect (e.g., climate) controls over $\delta^{15}N$ patterns in soil horizons. Indirect controls influence $\delta^{15}N$ patterns by affecting the

Table 4 Disturbance alters ^{15}N enrichment with depth, with ε_{soil} defined as $\delta^{15}N_{deep\ soil} - \delta^{15}N_{litter}$

Disturbance	ε_{soil} without disturbance (‰)	ε_{soil} with disturbance (‰)	Reference
Burning	5.8	1.0	Boeckx et al. (2005)
Windthrow	8.5	3.0[a], 6.7[b]	Kramer et al. (2003)
Clear-cut	10	6.5	Pardo et al. (2002)

[a] Major windthrow

[b] Moderate windthrow

Table 5 ^{15}N fractionation patterns among density and size fractions for various studies

Coniferous forest[1]		Coniferous forest[2]		Pasture[3]		Other sites	
$\delta^{15}N$ (‰)	Density	$\delta^{15}N$ (‰)	Density	$\delta^{15}N$ (‰)	Size class	ε (‰)	Soil fractions
0.03	1.65	5.01 ± 0.27[a]	<1.65	3.0–6.0	Sand	0–2	HF–LF[4]
2.67	1.85	5.68 ± 0.16[a]	>1.65	4.5–6.3	Silt	1–3	HF–floatable[4]
4.19	2.00	6.58 ± 0.25[b]	<1.65	7.6–9.0	Clay	5	Stabilized N–sand[5]
4.80	2.28	7.18 ± 0.29[b]	>1.65			3	HF–LF[6]
5.42	2.55	2.8 ± 1.73[c]	<1.65			1.6	<0.5 to >0.5 mm[7]
4.83	<2.55	6.65 ± 0.22[c]	>1.65				

Either isotopic values ($\delta^{15}N$) or ^{15}N enrichment (ε) values are given for specific fractions

[1] Coniferous forest in Alaska, Sollins et al. (2006)

[2] Coniferous forest in Alaska, Kramer et al. (2004). Disturbance level: [a]Heavy. [b]Moderate. [c]None

[3] Improved and native New Zealand pasture, across three depths (0–5, 5–10, and 50–60 cm), Ledgard et al. (1984), $\delta^{15}N_{clay} - \delta^{15}N_{sand} = 4‰$, $\delta^{15}N_{clay} - \delta^{15}N_{silt} = 3‰$

[4] Four agricultural California sites from 3 kyr to 4 Myr old, Baisden et al. (2002b). *HF* heavy fraction, <2.22 g cm^{-3}; *LF* light fraction, >2.22 g cm^{-3}

[5] Native and cultivated prairie, Tiessen et al. (1984). Stabilized N ≥ 12‰, sand 5‰ lighter, native and cultivated prairie

[6] Prairie and forest in Kansas, USA. Density is LF < 1.3 g cm^{-3} < HF. Billings (2006)

[7] Disturbed grasslands, humic fractions, greater and less than 0.5 mm, Kerley and Jarvis (1997)

factors directly controlling $\delta^{15}N$. Partitioning of ^{15}N within soil profiles, compound classes, or organisms will only influence patterns of $\delta^{15}N$ among soil horizons if ^{15}N-enriched or ^{15}N-depleted nitrogen can then preferentially move up or down soil profiles. In the following sections, we discuss the main potential controls over soil $\delta^{15}N$ patterns, including climate, mycorrhizal fungi, differential preservation of organic matter, nitrification, denitrification, disturbance, and bioturbation.

Climate

Mean annual precipitation and mean annual temperature significantly correlated to either bulk soil $\delta^{15}N$ or foliar $\delta^{15}N$ in several review papers (Handley et al. 1999a; Amundson et al. 2003). In these studies, foliar $\delta^{15}N$ and bulk soil $\delta^{15}N$ were higher in warmer, drier tropical areas, and lower in colder, wetter regions. Although climatic factors strongly correlated with bulk soil $\delta^{15}N$ in these studies, in our study they did not correlate with ^{15}N enrichment with depth in soils. This suggests that the processes controlling foliar and soil $\delta^{15}N$ at large scales differ from the processes controlling the development of $\delta^{15}N$ patterns within soil profiles, and that nitrogen dynamics within soil profiles are not primarily controlled by climatic factors. Prior correlations of $\delta^{15}N$ with climate may reflect an underlying correlation between climate and mycorrhizal type (Read 1991). In the current study the mean climate averaged colder for ectomycorrhizal symbioses (11°C) than for arbuscular mycorrhizal symbioses (18°C). Precipitation correlated weakly with ^{15}N profile enrichment in arbuscular

mycorrhizal systems. Because climate will also covary with other system properties (e.g., forests versus grasslands), an additional possibility is that systems dominated by woody plants behave differently from systems dominated by herbaceous plants or grasslands.

Nitrification and denitrification

Because nitrification and denitrification are two common processes in soils and fractionate highly against ^{15}N, they have been occasionally examined to explain ^{15}N enrichment in soil profiles. Several studies have compared nitrification rates to enrichment factors (ε_{soil}, here defined as $\delta_{soil} - \delta_{foliage}$) between foliar $\delta^{15}N$ and soil $\delta^{15}N$ (Garten 1993; Pardo et al. 2002; Vervaet et al. 2002). In Garten (1993), nitrification rate strongly and negatively correlated with the difference between foliar $\delta^{15}N$ and mineral soil $\delta^{15}N$ in a deciduous forest in Tennessee, with nitrification rates increasing from 0 to 1.2 μg g^{-1} day^{-1} as the ^{15}N enrichment of mineral soil relative to foliage decreased from 8 to 4‰. Emmett et al. (1998) also reported that nitrification rate negatively correlated with the enrichment factor across a European transect and Pardo et al. (2002) attributed increases in $\delta^{15}N$ in surface soil horizons after disturbance to increased nitrification. Despite these occasional successes at local scales, nitrification is not a straightforward mechanism for causing enrichment in ^{15}N with depth. The high mobility of nitrate in soil and the potential for ^{15}N-depleted nitrate to be either leached, assimilated by plants and microbes, or transformed into a residual pool of ^{15}N-enriched nitrate after denitrification means that the isotopic contribution of nitrification to different soil horizons is quite difficult to assess. Given the high mobility of nitrate in soils, the most probable outcome is for ^{15}N-depleted nitrate to be transported down the soil profile and either assimilated or denitrified. Nitrification rates and ^{15}N enrichment with depth are uncorrelated (Fig. 5, $r^2 = 0.05$), and nitrate leaching followed by immobilization could plausibly produce patterns of ^{15}N depletion at depth relative to intermediate soil depth.

The loss of ^{15}N-depleted N from soils during denitrification has also been invoked to explain $\delta^{15}N$ patterns in soil profiles. Because laboratory studies indicate that denitrification discriminates from 13 to 30‰ against ^{15}N (Perez et al. 2000), the remaining, ^{15}N-enriched compounds could be assimilated by microbes or other soil fauna and over time preserved and sequestered. This scenario was suggested recently by Houlton et al. (2006) as explaining nitrogen isotope patterns across a large rainfall gradient in arbuscular mycorrhizal *Metrosideros* forests of Hawaii. At very high rainfall, once a pool of N became available for denitrification, it was completely converted to N_2 and N_2O. Since denitrification went to completion, no isotopic partitioning between reactants and products was possible. At lower rainfall, the authors suggested that incomplete denitrification accounted for ^{15}N enrichment of residual soil N.

The influence of denitrification on 15N enrichment in soil profiles will depend on the Variable Rates of denitrification vary widely within soil profiles depending on water saturation, nitrate availability, and carbon availability (Hedin et al. 1998; Goldberg et al. 2008). The location and degree of ^{15}N enrichment within soil profiles caused by denitrification should therefore also vary.

Assessing the role of denitrification in soil $\delta^{15}N$ patterns requires quantitative estimates of denitrification versus other pathways of N loss. The extremely low rates of denitrification in many soils (Barton et al. 1999; Stehfest and Bouwman 2006) mean that denitrification is quantitatively unimportant in many N-limited systems. Average denitrification rates (as N_2O) were much higher in tropical forests (0.85 kg ha^{-1} $year^{-1}$) than in deciduous forests (0.48 kg ha^{-1} $year^{-1}$), coniferous forests (0.11 kg ha^{-1} $year^{-1}$), or savannas (0.12 kg ha^{-1} - $year^{-1}$) (Stehfest and Bouwman 2006), suggesting that denitrification in tropical systems was more likely to influence $\delta^{15}N$ patterns than in other systems. One complication to applying loss rates of N_2O to estimate ^{15}N effects on soil is that the proportion of nitrogen lost as N_2 versus N_2O during denitrification is extremely difficult to determine (Gruber and Galloway 2008). In a temperate forest, Pörtl et al. (2007) used a ratio of 6:1 for N_2:N_2O flux during denitrification, and in tropical forests Houlton et al. (2006) used isotopic mass balances to calculate that denitrification to N_2 could often be much higher than denitrification to N_2O under low O_2 conditions.

In the latter study, the authors concluded that partial conversion of nitrate during denitrification

could strongly influence soil ^{15}N values by creating a pool of ^{15}N-enriched nitrate that could be subsequently reassimilated by the microbial community and ultimately increase soil $\delta^{15}N$ at depth. If this nitrate is preferentially assimilated by microbes at depth rather than by trees, then this could plausibly cause large enrichments in ^{15}N with depth in tropical forests. This mechanism may operate in some temperate forests as well with high N cycling rates, such as the beech-spruce (ECM) forest studied by Pörtl et al. (2007) in which denitrification rates were 0.7 kg ha^{-1} $year^{-1}$ as N_2O. We conclude that nitrification, denitrification, and the leaching or diffusion of the products of these two processes could contribute to soil profiles of $\delta^{15}N$, and could be particularly important for profiles with maximum $\delta^{15}N$ values at intermediate depths, such as many sites dominated by arbuscular mycorrhizal vegetation.

Mycorrhizal fractionation

Relative to bulk soil, foliar litter and fine root litter are generally depleted in ^{15}N, whereas sporocarps of mycorrhizal fungi are often enriched in ^{15}N (Pardo et al. 2006, Hobbie and Hobbie 2008). Nitrogen availability can influence the partitioning of nitrogen and nitrogen isotopes between plants and associated mycorrhizal fungi (Hobbie and Colpaert 2003) and could therefore potentially influence nitrogen isotope profiles in soil. Accordingly, inputs of ^{15}N-depleted foliar litter to the soil surface (Nadelhoffer and Fry 1988; Högberg 1997) and inputs of ^{15}N-depleted root litter and ^{15}N-enriched mycorrhizal fungi at depth could result in deep soil horizons being substantially enriched in ^{15}N relative to surface litter (Högberg et al. 1996). The degree of ^{15}N enrichment should reflect in part the relative importance of root versus mycorrhizal fungi as sources for soil organic N in stable soil organic matter. The common practice of sieving soil prior to sampling may preferentially remove root litter, potentially increasing the perceived contribution of fungal litter to total soil N.

The type of mycorrhizal symbiosis dominant on a site greatly influences soil ^{15}N enrichment patterns. The average ^{15}N enrichment from litter to 50 cm soil depth for sites dominated by ectomycorrhizal vegetation is 9.6 ± 0.4‰ and for sites dominated by arbuscular mycorrhizal vegetation is only 4.6 ± 0.5‰. Ectomycorrhizal plants are generally depleted in ^{15}N relative to arbuscular mycorrhizal plants (Michelsen et al. 1996, 1998; Schmidt and Stewart 1997; Hobbie et al. 2005), although exceptions exist (Högberg 1990; Pardo et al. 2006). Ectomycorrhizal fungi are enriched in ^{15}N relative to their host plants. The difference in $\delta^{15}N$ between ectomycorrhizal plants and ectomycorrhizal fungi arises because of a 8–10‰ fractionation during the creation of transfer compounds by ectomycorrhizal fungi (Hobbie et al. 2000; Hobbie and Colpaert 2003). The few culture studies of the effects of mycorrhizal colonization on the $\delta^{15}N$ of arbuscular mycorrhizal plants have been inconclusive (Handley et al. 1993; Azcon-Aquilar et al. 1998), but in field studies arbuscular mycorrhizal plants average 2‰ depleted in ^{15}N relative to nonmycorrhizal plants (Michelsen et al. 1996, 1998; Schmidt and Stewart 1997; J. Craine, personnel communication). This suggests that similar ^{15}N fractionation processes operate in arbuscular mycorrhizal and in ectomycorrhizal symbioses.

Additional evidence that ectomycorrhizal symbiosis could influence ^{15}N differences between litter layers and the soil comes from culture studies with nonmycorrhizal and ectomycorrhizal *Pinus sylvestris* (Hobbie and Colpaert 2003). After a short growth period, the perlite growth media was enriched in ^{15}N relative to foliage on average by 1.4‰ for nonmycorrhizal pine, 3.2‰ for *Thelephora*-colonized pine, and 4.3‰ for *Suillus*-colonized pine. In the mycorrhizal cultures, perlite N was primarily of fungal origin. If similar partitioning of ^{15}N operates in forests, then the flux of nitrogen through mycorrhizal fungi and the resulting movement of ^{15}N-depleted nitrogen from soil horizons to the plant and then into litter could explain many of the observed $\delta^{15}N$ trends with depth in soil. In this scenario, ^{15}N-depleted foliar litter contrasts with stable soil nitrogen at depth processed through mycorrhizal fungi with a 8–10‰ enrichment in ectomycorrhizal-dominated systems.

Although direct measures of carbon allocation to arbuscular mycorrhizal fungi are few (Johnson et al. 2002; Gavito and Olsson 2003), the available evidence and the much smaller spatial extent of arbuscular mycorrhizal fungal hyphae relative to ectomycorrhizal fungal hyphae (e.g., arbuscular mycorrhizal hyphae extend up to 6–10 cm from roots, ectomycorrhizal hyphae extend up to several meters from roots; Coleman et al. 2004), indicate that arbuscular mycorrhizal fungi should contribute less to

the soil N pool than ectomycorrhizal fungi. This would account in part for the smaller values of ε_{soil} in arbuscular mycorrhizal-dominated systems than in ectomycorrhizal-dominated systems. The larger ^{15}N enrichment between litter and deeper soil horizons in ectomycorrhizal-dominated than in arbuscular mycorrhizal-dominated sites is consistent with isotopic partitioning between mycorrhizal fungi and their plant hosts largely driving these ^{15}N enrichment patterns.

Fractionation during decomposition

Enrichment in ^{15}N during decomposition could also influence soil $\delta^{15}N$ patterns if coupled to losses of ^{15}N-depleted inorganic or organic N. Fractionation against ^{15}N during ammonification, nitrification, or denitrification creates ^{15}N-depleted products. Subsequent assimilation, leaching, or gaseous losses could increase the ^{15}N content of the residual soil N. For example, Nadelhoffer and Fry (1988) calculated that N losses from temperate forest litter fractionated by 2.6 ± 0.8‰ against ^{15}N during a 600-day laboratory incubation. About 84% of losses were as ammonium or nitrate, with the remaining 16% attributed to denitrification.

Although fractionation during decomposition is often invoked to explain enrichment in ^{15}N with increasing soil depth, few studies have actually measured this fractionation. Melillo et al. (1989) observed minimal $\delta^{15}N$ change during a litterbag decomposition study on red pine. During the first 20 months of their study, the litter immobilized N (N% increased) and $\delta^{15}N$ actually decreased by 2 to 3‰. Over the next 6.5 years N loss was large but became only slightly enriched in ^{15}N, with a calculated enrichment factor of about 0.5‰. This is much less than the 2.6‰ enrichment factor calculated from laboratory incubations of the upper 10 cm of soil (including litter) from an oak-dominated site (Nadelhoffer and Fry 1988). Lindahl et al. (2007) recorded the relative abundance of saprotrophic and ectomycorrhizal fungi in a soil profile of a boreal forest. Soil horizon $\delta^{15}N$ increased less than 2‰ in the upper litter layers where saprotrophic fungi dominated community composition and increased about 8‰ thereafter where ectomycorrhizal fungi dominated community composition. The few studies reported suggest that the isotopic effects during decomposition of both litter and root material are relatively small, at only a few per mille. These small enrichment factors of 3‰ or less cannot fully account for the large shifts in ^{15}N in many soil profiles, since many of the profiles studied in Table 2 resulted in calculated enrichment factors of 5‰ or greater.

Soil fauna, soil foodwebs, bioturbation, and disturbance

Since soil fauna process large quantities of soil N (deRuiter et al. 1993; Moore et al. 2005), cycling of N through soil foodwebs could potentially influence soil $\delta^{15}N$ patterns. Although N can be released from litter directly during fungally or bacterially mediated decomposition (i.e., when the C:N ratio of available C and N divided by the microbial efficiency is less than the C:N of microbial biomass), much N release in soils depends on grazing of primary decomposers by higher trophic levels such as nematodes and amoebae (Clarholm 1981, 1985). $\delta^{15}N$ values increased 3.4‰ per trophic level during N transfer from litter to detritivores to predators (Ponsard and Arditi 2000); Haubert et al. (2006) found similar ^{15}N enrichment during trophic transfer of N in soil microinvertebrates (2.9 ± 2.1‰). If ^{15}N-enriched nitrogen from soil fauna is preferentially preserved and the ^{15}N-depleted excretion products are preferentially removed, then faunal processing of soil N could contribute to ^{15}N enrichment with increasing soil depth if the net N flux is down the soil profile.

In some systems, macroinvertebrates such as termites, ants, earthworms (Lee and Foster 1991), or fossorial mammals (Yoo et al. 2005) can move soil among different soil horizons, and this could obviously diminish ^{15}N shifts among those horizons. Microinvertebrates appear to have little effect on soil transport processes, however (Lee and Foster 1991), and N movement between soil horizons in terrestrial ecosystems such as pine forests without large populations of earthworms is small (Bird and Torn 2006). Because earthworms move higher volumes of soil than other invertebrates (Gabet et al. 2003), and can increase nitrogen leaching rates (Dominguez et al. 2004), they could affect patterns of soil N isotopes. In one study in temperate mixed hardwoods, sites with earthworms were about 1‰ depleted in ^{15}N throughout the soil profile relative to sites without earthworms (Bohlen et al. 2004). Although fungi are

not normally considered agents of bioturbation, they are quite important in moving N into decomposing litter from lower soil horizons (Frey et al. 2003; Caner et al. 2004).

Abiotic and biotic disturbances of soil can dramatically affect patterns of soil ^{15}N distributions. Fire can eliminate surface litter layers and may also increase reliance of plants on deeper soil horizons for N (Högberg 1997). Disturbances such as clearcutting (Pardo et al. 2002) or grazing often increase N availability by decreasing plant N demands and the flux of labile carbon into the microbial community. This may diminish the importance of mycorrhizal fungi for plant N supply and increase nitrification and denitrification rates, thereby ultimately increasing the $\delta^{15}N$ of plants and the litter layer.

Synthesizing the processes influencing soil $\delta^{15}N$ into a conceptual model

In Fig. 7, we propose a conceptual model of how various processes influence $\delta^{15}N$ patterns in soil profiles. The model has three possible inputs of N to soil profiles—leaf litter, root litter, and fungal material. Several mechanisms can potentially move nitrogen up or down a soil profile without directly changing the ^{15}N signature of the nitrogen that is being moved, including leaching, bioturbation, and fungal immobilization. However, non-fractionating processes such as leaching of ^{15}N-depleted N can alter ^{15}N profiles if the ^{15}N-depleted nitrate is reassimilated at depth. In some systems, bioturbation may diminish the ^{15}N difference within soil profiles.

Horizontal lines in Fig. 7 represent isotopically fractionating processes that can directly change the $\delta^{15}N$ signature of a soil profile. For example, mycorrhizal discrimination and supply of ^{15}N-depleted N to host plants should increase ^{15}N differences within soil profiles. This fractionation and N supply should be less important in arbuscular mycorrhizal systems than in ectomycorrhizal systems and should be less important at high N availability than at low N availability (Wallenda and Kottke 1998; Hobbie et al. 2000; Hobbie and Colpaert 2003). Gaseous losses of ^{15}N-depleted N could increase ^{15}N differences within soil profiles. Production of N_2O or leaching and subsequent reassimilation of ^{15}N-depleted nitrate at depth are suggested as plausible mechanisms for causing ^{15}N enrichment at intermediate depths relative to greater depths, as was reported for about 40% of arbuscular mycorrhizal-dominated profiles. This conceptual model includes the major processes likely to influence $\delta^{15}N$ patterns in soil profiles and may provide a useful framework for relating N dynamics to these $\delta^{15}N$ patterns in quantitative and dynamic models. Numerical

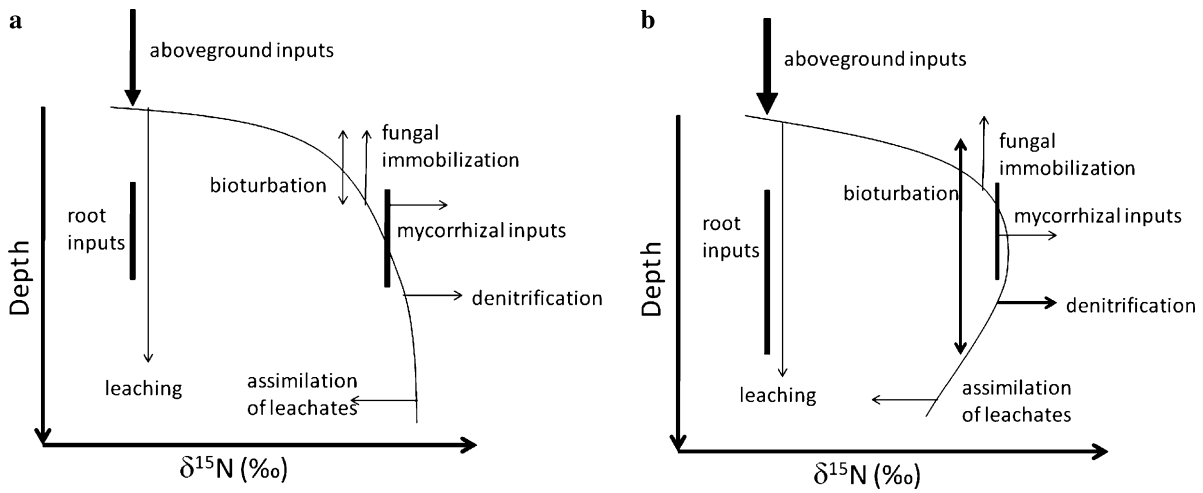

Fig. 7 Summary diagram of processes affecting soil $\delta^{15}N$ in soil profiles, with depth on the *x*-axis and $\delta^{15}N$ on the *y*-axis. Nitrogen inputs are shown as heavy dark bars, processes moving N without fractionation are shown as *vertical arrows* and processes directly causing shifts in soil $\delta^{15}N$ are shown as *horizontal arrows*. Two cases are shown: **a** N-limited system dominated by mycorrhizal transfer and organic N cycling; **b** system with less N limitation and more inorganic N cycling

simulations using this conceptual model as a template could of course be constructed as well.

Conclusion

In comparing δ^{15}N data from temperate and tropical soils, it appears that climatic factors (MAP and MAT) and nitrification do not directly affect patterns of soil δ^{15}N with depth. Also, because processing by micr-oinvertebrates such as soil mites cannot redistribute N within soil profiles, this process accordingly cannot directly affect patterns of δ^{15}N with depth in terrestrial soils. However, macroinvertebrates (particularly earthworms) and fossorial mammals can redistribute large quantities of soil and could directly influence soil N and ^{15}N distributions. Mycorrhizal association (ectomycorrhizal versus arbuscular mycorrhizal) correlated significantly with ^{15}N enrichment, with sites dominated by ectomycorrhizal vegetation displaying larger enrichments with depth. Fungal ^{15}N enrichment cannot account for all δ^{15}N patterns, and other soil processes such as ^{15}N discrimination during decomposition, the differential stabilization of different N compounds and redistribution among soil horizons must also influence these patterns. The numerous processes controlling N movement and isotopic patterns in soil profiles suggest that further insights are likely to come from applying dynamic models that can incorporate multiple competing processes.

Acknowledgments We thank Ben Houlton and two anonymous reviewers for useful comments on an earlier version of the manuscript. This work was suported by NSF grant DEB-0614266.

References

Amundson R, Austin AT, Schuur EAG et al (2003) Global patterns of the isotopic composition of soil and plant nitrogen. Global Biogeochem Cycles 17:1031–1041. doi:10.1029/2002GB001903

Azcon-Aquilar GR, Handley LL, Scrimgeour CM (1998) The δ^{15}N of lettuce and barley are affected by AM status and external concentration of N. New Phytol 138:19–26. doi:10.1046/j.1469-8137.1998.00883.x

Baisden WT, Amundson R, Brenner DL et al (2002a) A multiisotope C and N modeling analysis of soil organic matter turnover and transport as a function of soil depth in a California annual grassland soil chronosequence. Global Biogeochem Cycles 16:26

Baisden WT, Amundson R, Cook AC, Brenner DL (2002b) Turnover and storage of C and N in five density fractions from California annual grassland surface soils. Global Biogeochem Cycles 16:16

Barton L, McLay CDA, Schipper LA, Smith CT (1999) Annual denitrification rates in agricultural and forest soils: a review. Aust J Soil Res 37:1073–1093. doi:10.1071/SR99009

Billings SA (2006) Soil organic matte dynamics and land use change at a grassland/forest ecotone. Soil Biol Biochem 38:2934–2943. doi:10.1016/j.soilbio.2006.05.004

Billings SA, Richter DD (2006) Changes in stable isotopic signatures of soil nitrogen and carbon during 40 years of forest development. Oecologia 148:325–333. doi:10.1007/s00442-006-0366-7

Bird JA, Torn (2006) Fine roots versus needles: a comparison of ^{13}C and ^{15} N dynamics in a ponderosa pine forest soil. Biogeochemistry 79:361–382. doi:10.1007/s10533-005-5632-y

Boeckx P, Paulino L, Oyarzun C et al (2005) Soil δ^{15}N patterns in old-growth forests of southern Chile as integrator for N-cycling. Isotopes Environ Health Stud 41:249–259. doi:10.1080/10256010500230171

Bohlen PJ, Pelletier DM, Groffman PM et al (2004) Influence of earthworm invasion on redistribution and retention of soil carbon and nitrogen in northern temperate forests. Ecosystems (N Y, Print) 7:13–27. doi:10.1007/s10021-003-0127-y

Caner L, Zeller B, Dambrine E et al (2004) Origin of the nitrogen assimilated by soil fauna living in decomposing beech litter. Soil Biol Biochem 36:1861–1872. doi:10.1016/j.soilbio.2004.05.007

Chalot M, Brun A (1998) Physiology of organic nitrogen acquisition by ectomycorrhizal fungi and ectomycorrhizas. FEMS Microbiol Rev 22:21–44

Clarholm M (1981) Protozoan grazing of bacteria in soil—impact and importance. Microb Ecol 7:343–350. doi:10.1007/BF02341429

Clarholm M (1985) Interactions of bacteria, protozoa and plants leading to mineralization of soil-nitrogen. Soil Biol Biochem 17:181–187. doi:10.1016/0038-0717(85)90113-0

Coleman DC, Crossley DA Jr, Hendrix PF (2004) Fundamentals of soil ecology, 2nd edn. Academic Press, New York

DeRuiter PC, Moore JC, Zwart KB et al (1993) Simulation of nitrogen mineralization in the belowground food webs of 2 winter-wheat fields. J Appl Ecol 30:95–106. doi:10.2307/2404274

Dominguez J, Bohlen PJ, Parmelee RW (2004) Earthworms increase nitrogen leaching to greater soil depths in row crop agroecosystems. Ecosystems (N Y, Print) 7:672–685. doi:10.1007/s10021-004-0150-7

Emmett BA, Kjonaas OJ, Gundersen P et al (1998) Natural abundance of ^{15}N in forests across a nitrogen deposition gradient. For Ecol Manag 101:9–18

Evans RD, Belnap J (1999) Long-term consequences of disturbance on nitrogen dynamics in an arid ecosystem. Ecology 80:150–160

Frey SD, Six J, Elliott ET (2003) Reciprocal tansfer of carbon and nitrogen by decomposer fungi at the soil-litter interface. Soil Biol Biochem 35:1001–1004. doi:10.1016/S0038-0717(03)00155-X

Gabet EJ, Reichman OJ, Seabloom EW (2003) The effects of bioturbation on soil processes and sediment transport. Annu Rev Earth Planet Sci 21:249–273. doi:10.1146/annurev.earth.31.100901.141314

Garten CT (1993) Variation in foliar ^{15}N abundance and the availability of soil nitrogen on Walker Branch watershed. Ecology 74:2098–2113. doi:10.2307/1940855

Garten CT, van Miegroet H (1994) Relationships between soil nitrogen dynamics and natural ^{15}N abundance in plant foliage from Great Smoky Mountains National Park. Can J Res 24:1636–1645. doi:10.1139/x94-212

Gavito ME, Olsson PA (2003) Allocation of plant carbon to foraging and storage in arbuscular mycorrhizal fungi. FEMS Microbiol Ecol 45:181–187. doi:10.1016/S0168-6496(03)00150-8

Goldberg SD, Knorr KH, Gebauer G (2008) N_2O concentration and isotope signature along profiles provide deeper insight into the fate of N_2O in soils. Isotopes Environ Health Stud 41:377–391. doi:10.1080/10256010802507433

Guo DL, Li H, Mitchell RJ, Han WX, Hendricks JJ, Fahey TJ, Hendrick RL (2008) Fine root heterogeneity by branch order: exploring the discrepancy in root turnover estimates between minirhizotron and carbon isotopic methods. New Phytol 177:443-456

Handley LL, Raven JA (1992) The use of natural abundance of nitrogen isotopes in plant physiology and ecology. Plant Cell Environ 15:965–985. doi:10.1111/j.1365-3040.1992.tb01650.x

Handley LL, Daft MJ, Wilson J et al (1993) Effects of the ecto- and VA-mycorrhizal fungi *Hydnagium carneum* and *Glomus clarum* on the ^{15} N and ^{13}C values of *Eucalyptus globulus* and *Ricinus communis*. Plant Cell Environ 16:375–382. doi:10.1111/j.1365-3040.1993.tb00883.x

Handley LL, Austin AT, Robinson D et al (1999a) The ^{15}N natural abundance ($\delta^{15}N$) of ecosystem samples reflects measures of water availability. Aust J Plant Physiol 26:185–199

Handley LL, Azcon R, Lozano JMR, Scrimgeour CM (1999b) Plant $\delta^{15}N$ associated with arbuscular mycorrhization, drought and nitrogen deficiency. Rapid Commun Mass Spectrom 13:1320–1324. doi:10.1002/(SICI)1097-0231(19990715)13:13<1320::AID-RCM607>3.0.CO;2-M

Haubert D, Haggblom MM, Langel R et al (2006) Trophic shift of stable isotopes and fatty acids in Collembola on bacterial diets. Soil Biol Biochem 38:2004–2007. doi:10.1016/j.soilbio.2005.11.031

Hedin LO, von Fischer JC, Ostrom NE et al (1998) Thermodynamic constraints on nitrogen transformations and other biogeochemical processes at soil-stream interfaces. Ecology 79:684–703

Hobbie EA, Colpaert JV (2003) Nitrogen availability and colonization by mycorrhizal fungi correlate with nitrogen isotope patterns in plants. New Phytol 157:115–126. doi:10.1046/j.1469-8137.2003.00657.x

Hobbie EA, Hobbie JE (2008) Natural abundance of ^{15}N in nitrogen-limited forests and tundra can estimate nitrogen cycling through mycorrhizal fungi: a review. Ecosystems (N Y, Print) 11:815–830. doi:10.1007/s10021-008-9159-7

Hobbie EA, Macko SA, Shugart HH (1999) Interpretation of nitrogen isotope signatures using the NIFTE model. Oecologia 120:405–415. doi:10.1007/s004420050873

Hobbie EA, Macko SA, Williams M (2000) Correlations between foliar $\delta^{15}N$ and nitrogen concentrations may indicate plant-mycorrhizal interactions. Oecologia 122:273–283. doi:10.1007/PL00008856

Hobbie EA, Jumpponen A, Trappe J (2005) Foliar and fungal ^{15}N:^{14}N ratios reflect development of mycorrhizae and nitrogen supply during primary succession: testing analytical models. Oecologia 146:258–268. doi:10.1007/s00442-005-0208-z

Högberg P (1990) ^{15}N natural abundance as a possible marker of the ectomycorrhizal habit of trees in mixed African woodlands. New Phytol 115:483-486

Högberg P (1997) ^{15}N natural abundance in soil-plant systems. New Phytol 137:179–203. doi:10.1046/j.1469-8137.1997.00808.x

Högberg P, Hogbom L, Schinkel H et al (1996) ^{15}N abundance of surface soils, roots and mycorrhizas in profiles of European forest soils. Oecologia 108:207–214

Houlton BZ, Sigman DM, Hedin LO (2006) Isotopic evidence for large gaseous nitrogen losses from tropical rainforests. Proc Natl Acad Sci USA 103:8745–8750. doi:10.1073/pnas.0510185103

Houlton BZ, Sigman DM, Schuur EAG, Hedin LO (2007) A climate-driven switch in plant nitrogen acquisition within tropical forest communities. Proc Natl Acad Sci 104:8902–8906

Hübner H (1986) Isotope effects of nitrogen in the soil and biosphere. In: Fritz P, Fontes PC (eds) Handbook of environmental isotope geochemistry, vol 2. Elsevier, Amsterdam, pp 361–425

Johnson D, Leake JR, Read DJ (2002) Transfer of recent photosynthate into mycorrhizal mycelium of an upland grassland: short-term respiratory losses and accumulation of ^{14}C. Soil Biol Biochem 34:1521–1524. doi:10.1016/S0038-0717(02)00126-8

Jones DL, Kielland K (2002) Soil amino acid turnover dominates the nitrogen flux in permafrost-dominated taiga forest soils. Soil Biol Biochem 34:209–219. doi:10.1016/S0038-0717(01)00175-4

Kaste JM, Heimsath AM, Bostick BC (2007) Short-term soil mixing quantified with fallout radionuclides. Geology 35:243–246. doi:10.1130/G23355A.1

Kendall C (1998) Tracing nitrogen sources and cycling in catchments. In: Kendall C, McDonnell J (eds) Isotope tracers in catchment hydrology. Elsevier, Amsterdam, pp 519–578

Kerley SJ, Jarvis SC (1997) Variation in ^{15}N natural abundance of soil, humic fractions and plant materials in a disturbed and an undisturbed grassland. Biol Fert Soil 24:147–152

Kitayama K, Iwamoto K (2001) Patterns of natural ^{15}N abundance in the leaf-to-soil continuum of tropical rain forests differing in N availability on Mount Kinabalu, Borneo. Plant Soil 229:203–212. doi:10.1023/A:1004853915544

Koba K, Tokuchi N, Yoshioka T et al (1998) Natural abundance of ^{15}N in a forest soil. Soil Sci Soc Am J 62:778–781

Kramer MG, Sollins P, Sletten RS, Swart PK (2003) N isotope fractionation and measures of organic matter alteration during decomposition. Ecology 84:2021–2025. doi:10.1890/02-3097

Kramer MG, Sollins P, Sletten RS (2004) Soil carbon dynamics across a windthrow disturbance sequence in southeast Alaska. Ecology 85:2230–2244. doi:10.1890/02-4098

Ledgard SF, Freney JR, Simpson JR (1984) Variations in natural enrichment of ^{15}N in the profiles of some Australian pasture soils. Aust J Soil Res 22:155–164. doi:10.1071/SR9840155

Lee KE, Foster RC (1991) Soil fauna and soil structure. Aust J Soil Res 29:745–775. doi:10.1071/SR9910745

Lindahl BD, Ihrmark K, Boberg J et al (2007) Spatial separation of litter decomposition and mycorrhizal nitrogen uptake in a boreal forest. New Phytol 160:255–272

Mariotti A, Pierre D, Vedy JC et al (1980) The abundance of natural nitrogen 15 in the organic matter of soils along an altitudinal gradient (Chablais, Haute Savoie, France). Catena 7:293–300

Mariotti A, Germon JC, Hubert P et al (1981) Experimental determination of nitrogen kinetic isotope fractionation: some principles; illustration for the denitrification and nitrification processes. Plant Soil 62:413–430. doi:10.1007/BF02374138

Martinelli LA, Piccolo MC, Townsend AR et al (1999) Nitrogen stable isotopic composition of leaves and soil: tropical versus temperate forests. Biogeochemistry 46:45–65

Melillo JM, Aber JD, Linkins AE et al (1989) Carbon and nitrogen dynamics along the decay continuum—plant litter to soil organic matter. Plant Soil 115:189–198. doi:10.1007/BF02202587

Michelsen A, Schmidt IK, Jonasson S et al (1996) Leaf ^{15}N abundance of subarctic plants provides field evidence that ericoid, ectomycorrhizal and non- and arbuscular mycorrhizal species access different sources of soil nitrogen. Oecologia 105:53–63. doi:10.1007/BF00328791

Michelsen A, Quarmby C, Sleep D, Jonasson S (1998) Vascular plant ^{15}N natural abundance in heath and forest tundra ecosystems is closely correlated with presence and type of mycorrhizal fungi in roots. Oecologia 115:406–418. doi:10.1007/s004420050535

Molina R, Massicotte H, Trappe JM (1992) Specificity phenomena in mycorrhizal symbioses: community-ecological consequences and practical implications. In: Allen MF (ed) Mycorrhizal functioning. Chapman and Hall, New York, pp 357–423

Moore JC, McCann K, de Ruiter PC (2005) Modeling trophic pathways, nutrient cycling, and dynamic stability in soils. Pedobiologia (Jena) 49:499–510. doi:10.1016/j.pedobi.2005.05.008

Nadelhoffer KF, Fry B (1988) Controls on natural ^{15}N and ^{13}C abundances in forest soil organic-matter. Soil Sci Soc Am J 52:1633–1640

Nadelhoffer KJ, Colman BP, Currie WS et al (2004) Decadal-scale fates of ^{15}N tracers added to oak and pine stands under ambient and elevated N inputs at the Harvard Forest (USA). For Ecol Manag 196:89–107

Nardoto GB (2005) Abundância natural de 15N na Amazônia e Cerrado—implicações para a ciclagem de nitrogênio. PhD thesis, Universidade de São Paulo

Olsson PA, Jakobsen I, Wallander H (2002) Foraging and resource allocation strategies of mycorrhizal fungi in a patchy environment. In: van der Heijden MGA, Sanders I (eds) Mycorrhizal ecology. Springer, Berlin, pp 93–115

Pardo LH, Hemond HF, Montoya JP et al (2002) Response of the natural abundance of ^{15}N in forest soils and foliage to high nitrate loss following clear-cutting. Can J Res 32:1126–1136. doi:10.1139/x02-041

Pardo LH, Templer PH, Goodale CL et al (2006) Regional assessment of N saturation using foliar and root $\delta^{15}N$. Biogeochemistry 80:143–171. doi:10.1007/s10533-006-9015-9

Pardo LH, Hemond HF, Montoya JP, Pett-Ridge J (2007) Natural abundance ^{15}N in soil and litter across a nitrate-output gradient in New Hampshire. For Ecol Manag 251:217–230

Perez T, Trumbore SE, Tyler SC et al (2000) Isotopic variability of N_2O emissions from tropical forest soils. Global Biogeochem Cycles 14:525–535. doi:10.1029/1999GB001181

Ponsard S, Arditi R (2000) What can stable isotopes ($\delta^{15}N$ and $\delta^{13}C$) tell about the food web of soil macro-invertebrates? Ecology 81:852–864

Pörtl K, Zechmeister-Boltenstern S, Wanek W et al (2007) Natural ^{15}N abundance of soil N pools and N_2O reflect the nitrogen dynamics of forest soils. Plant Soil 295:79–94. doi:10.1007/s11104-007-9264-y

Quideau SA, Graham RC, Feng X, Chadwick OA (2003) Natural isotopic distribution in soil surface horizons differentiated by vegetation. Soil Sci Soc Am J 67:1544–1550

Read DJ (1991) Mycorrhizas in ecosystems. Experientia 47:376–391. doi:10.1007/BF01972080

Robinson D (2001) $\delta^{15}N$ as an integrator of the nitrogen cycle. Trends Ecol Evol 16:153–162. doi:10.1016/S0169-5347(00)02098-X

Schimel JP, Bennett J (2004) Nitrogen mineralization: challenges of a changing paradigm. Ecology 85:591–602. doi:10.1890/03-8002

Schmidt S, Stewart GR (1997) Waterlogging and fire impacts on nitrogen availability and utilization in a subtropical wet heathland (wallum). Plant Cell Environ 20:1231–1241. doi:10.1046/j.1365-3040.1997.d01-20.x

Schuur EAG, Matson PA (2001) Net primary productivity and nutrient cycling across a mesic to wet precipitation gradient in Hawaiian montane forest. Oecologia 128:431–442. doi:10.1007/s004420100671

Shearer G, Kohl DH (1986) N_2-fixation in field settings—estimations based on natural ^{15}N abundance. Aust J Plant Physiol 13:699–756

Silfer JA, Engel MH, Macko SA (1992) Kinetic fractionation of stable carbon and nitrogen isotopes during peptide bond hydrolysis: experimental evidence and geochemical implications. Chem Geol 101:211–221

Silver WL, Neff J, McGroddy M et al (2000) Effects of soil texture on belowground carbon and nutrient storage in a lowland Amazonian forest ecosystem. Ecosystems (N Y, Print) 3:193–209. doi:10.1007/s100210000019

Sollins P, Swanston C, Kleber M et al (2006) Organic C and N stabilization in a forest soil: evidence from sequential density fractionation. Soil Biol Biochem 38:3313–3324. doi:10.1016/j.soilbio.2006.04.014

Stehfest E, Bouwman L (2006) N_2O and NO emission from agricultural fields and soils under natural vegetation: summarizing available measurement data and modeling of global annual emissions. Nutr Cycl Agroecosyst 74:207–228. doi:10.1007/s10705-006-9000-7

Taylor AFS, Alexander I (2005) The ectomycorrhizal symbiosis: life in the real world. Mycologist 19:102–112. doi:10.1017/S0269915XO5003034

Tiessen H, Karamanos RE, Stewart JWB, Selles F (1984) Natural nitrogen-15 abundance as an indicator of soil organic matter transformations in native and cultivated soils. Soil Sci Soc Am J 48:312–315

Vervaet H, Boeckx P, Unamuno V et al (2002) Can $\delta^{15}N$ profiles in forest soils predict NO_3^- loss and net N mineralization rates? Biol Fertil Soils 36:143–150. doi:10.1007/s00374-002-0512-2

Wada E, Imaizumi R, Takai Y (1984) Natural abundance of 15N in soil organic-matter with special reference to paddy soils in Japan–biogeochemical implications on the nitrogen-cycle. Geochem J 18:109–123

Wallenda T, Kottke I (1998) Nitrogen deposition and ectomycorrhizas. New Phytol 139:169–187

Wallenda T, Stober C, Högbom L (2000) Nitrogen uptake processes in roots and mycorrhizas. In: Schulze E-D et al (eds) Carbon and nitrogen physiology in forest ecosystems. Springer, Berlin, pp 122–143

Yoo K, Amundson R, Heimsath AM, Dietrich WE (2005) Process-based model linking pocket gopher (*Thomomys bottae*) activity to sediment transport and soil thickness. Geology 33:917–920. doi:10.1130/G21831.1