Stable nitrogen isotopes in plant and soil processes

Natural ¹⁵N abundance

Terrestrial N cycle



Modified after D. Robinson



- Plant N uptake and ¹⁵N discrimination
- Plant-mycorrhiza associations
- Symbiotic N₂ fixation (legumes)

Plant N uptake and ¹⁵N discrimination



 ϵ (- Δ) of **ammonium nutrition** by two rice cultivars ϵ Range -20 to -55‰ f value .. % residual N left in solution Yoneyama et al. (2001) Plant Cell Environ 24: 133 ϵ of **nitrate nutrition**

Koshihikari: $+0.6 \pm 1.8\%$ Kanto 168: $-2.5 \pm 1.5\%$

Plant N uptake and ¹⁵N discrimination



Högberg et al. (1999) New Phytol 142: 569

Substrate consumption (%)

Plant N uptake and ¹⁵N discrimination



Fig. 5. Kinetic isotope fractionation during log phase for *T.* pseudonana growth on nitrate (data from Fig. 1), nitrite, ammonium, and urea (data from the first log phase, i.e. from 0 to 55 h, presented in Fig. 3). *F* is related to the fraction of unconsumed dissolved N, *f*, by the relationship $F = [-f/(1 - f)] \ln f$. The solid line represents the least-squares fit of the $\delta^{13}N_{\rm FN}$, the dashed lines are the 95% confidence intervals, and the dotted lines are the $\delta^{15}N$ of the initial N source. The regression is shown for each substrate.

Isotope discrimination during N assimilation differs between N forms

- Uptake/Influx F_{IN}
- Assimilation F_{RN} discriminates by b
- Efflux F_{EN}
- Net uptake $F_{NN} = F_{IN} F_{EN}$



$$\begin{array}{l} \delta_{\mathsf{plant}} = \\ \delta_{\mathsf{NO3}} - \mathsf{F}_{\mathsf{RN}}/\mathsf{F}_{\mathsf{NN}}^* \\ (\mathsf{1}\text{-}\mathsf{F}_{\mathsf{NN}}/\mathsf{F}_{\mathsf{IN}})^* b \end{array}$$

Figure 1. A simplified model of steady-state nitrate uptake, based on the conceptual model of Comstock (2001). F_{ln} is the influx of nitrate from the soil solution into root cells; F_{En} is the efflux of nitrate from root cells back to the soil solution; F_{Rn} is the assimilation flux of nitrate from root cells into the xylem. Nitrate loaded into the xylem is carried to the leaf in the transpiration stream, where it can also be assimilated into organic molecules. Because nitrate is not exported from leaves in the phloem, the flux of nitrate into the xylem in the root is equal to the assimilation flux in the shoot in the steady state. The discrimination against ¹⁵N during nitrate reduction in both the root and the shoot is defined as *b*, which is a constant. $F_{In'}$, $F_{En'}$ and F_{Xn} are assumed to proceed without discrimination against ¹⁵N.

Effect of mycorrhiza on plant ¹⁵N discrimination



Fig. 2 Mean $\delta^{15}N$ (±SE) of plant species without mycorrhiza (NON), with ectomycorrhizal (ECM) fungi or with ericoid mycorrhizal (ERI) fungi at the four heath and forest tundra sites in Fig. 1. The means within each functional group and site are based on the means of the species presented in Fig. 1; n is the number of replicates (plant species)

N transfer in plant-myc associations



Fig. 5 Schematic diagram of hypothesized interactions among vegetation, soil available N, and mycorrhizal fungi under conditions of rapid N cycling (a), or slow N cycling (b)

Hobble et al. (2000) Oecologia 122: 273

Natural ¹⁵N method for estimating the proportion of N derived from N₂ fixation



%Ndfa=100*($\delta^{15}N_{ref}-\delta^{15}N_{leg}$)/($\delta^{15}N_{ref}-B$) B-value: $\delta^{15}N$ of legume totally independent on soil N (-1.0 to -2.5‰)

Peoples et al. (1989) ACIAR Series

Symbiotic N₂ fixation (legumes) and N availability



Unkovich et al. (2000) Aust J Plant Physiol 27: 921

Fig. 1. An example of the use of natural ¹⁵N abundances (δ¹⁵N) to estimate the proportion of plant N derived from the atmosphere by symbiotic N₂ fixation⁹. The tree legume Chamaecytisus proliferus was grown in sand containing different amounts of added NO₂with a $\delta^{15}N$ value of +3.5‰. δ^{15} N of N, is, by definition (Box 1), 0‰. Whole-plant δ¹⁵N values (circles) varied within these limits, depending on how much N was derived from N, and NO,-, becoming more ¹⁵N enriched as the availability of source NO₂increased. From these data, the proportion of plant N derived from N₂ (squares) was estimated using a mixing model (Box 2).

Soil and ecosystem N dynamics

- Soil ¹⁵N enrichment (bulk and inorganic N)
- Nitrification, Denitrification
- Ecosystem N losses and global patterns

Isotopic signatures of bulk soil N



Fig. 1. Frequency distribution of $\delta^{13}N$ values of total soil N in 124 surface soils from 20 States of the U.S.A. Mean $\delta^{13}N$ was 9-92. Samples were collected near the soil surface. Modified from Shearer *et al.* (1978).

Bulk soil ¹⁵N enriched compared to atm. N₂

$\delta^{15}N$ of mineral N across soil profiles



δ^{15} N of soil mineral N [**I**•] (‰) 5 10 15 0 25 (b) O^{CL} 25 Soil depth (cm) 50 75 100 □■ Site 3 125 O● Site 4 20 10 30 40 50 Concentration of soil mineral N [□0] (mgN/kg soil)

Soil ¹⁵N enrichment due to cumulative losses

Figure 3. Changes in concentration and ¹⁵N abundance of mineral N (NH₄⁺+NO₃⁻) in the profile of different cropping and perennial pasture systems where the δ^{15} N of soil mineral N is either (A) relatively similar or (B) changes down the profile (data from Peoples et al., 1995b).

Concentration and $\delta^{15}N$ of mineral N in cropping and perennial pasture systems in New Zealand Boddey et al. (2001) Nutr Cycl Agroecosystems 57: 235

Soil N processes and ¹⁵N discrimination

Anhydrous ammonia 15applied Ammonium 10 Nitrate ς Ω -10 200 r 150 soil Ammonium N kg⁻¹ 100 bu 50 Nitrate M Å Months in 1972

Fig. 2.3 The conversion of NH_4^+ to NO_3^- and changes in the $\delta^{15}N$ values of these to nitrog forms following application of anhydrous NH_3 fertilizer to an agricultural field. (From Feigin *et al.*, 1974b.)

Microbial NITRIFICATION (ammonium oxidation to nitrate)

(
$$\Delta \sim 15-50$$
‰)

Feigin et al. (1974)

Soil N processes and ¹⁵N discrimination



Microbial DENITRIFICATION (dissimilatory reduction of nitrate to nitrite, NO, N_2O and N_2)

$$(\Delta \sim 0 - 35\%)$$

Mariotti et al. (1981) Plant Soil 62: 413

Ecosystem N losses and foliar $\delta^{15}N$



Fig. 1. The relation between the fractional loss of added N from plots fertilized with N or NPK and the change in ¹⁵N abundence of current needles of Scots pine during the experiment at Norrliden 1970–1989. **a**, plots fertilized with NH₄NO₃; **b**, plots fertilized with urea (cf. Table 1). Each data point represents the mean for 2–3 plots.

Högberg & Johannisson (1993) Plant Soil 157: 147

Rates of soil N cycling and foliar $\delta^{15}N$



FIG. 11. Relationship between mean enrichment factors $(\delta^{15}N_{leaf} - \delta^{15}N_{soil})$ (A) or mean foliar ¹⁵N abundance (B) in deciduous trees and net nitrification potential per unit of soil dry mass in laboratory incubations of surface mineral soil. Data are means ± 1 SE. Numbers next to each point designate the following data sets: red maple and dogwood trees from (1) SW-facing slopes, (2) ridges, (3) NE-facing slopes, and (5) valley bottoms in the summer of 1989 on Walker Branch Watershed; mixed deciduous tree species from (4) ridge and (6) valley-bottom sampling sites on Walker Branch Watershed in the summer of 1991; and (7) sycamore and sweet gum trees from a plantation at the Oak Ridge National Laboratory 0800 Area.

Higher net nitrification = higher N losses by leaching or denitrification

The higher the N losses the higher the residual inorg. N and plant $\delta^{15}N$ values

Garten (1993) Ecology 74: 2098

Comparison of foliar δ¹⁵N between temperate and tropical forests



Figure 1. Histogram of $\delta^{15}N$ (‰) for tree leaves collected in tropical and temperate forests. Solid bars are tropical sites and hatched bars are temperate sites. (b) Plot of $\delta^{15}N$ (‰) for tree leaves collected in tropical and temperate forests. Error bars represent one standard-deviation.

Ecosystem N cycling and foliar δ^{15} N: influence of mean annual precipitation



Handley et al. (1999) Aust J Plant Physiol 26: 185

Strong seasonality: Litterfall and decomposition provide pulses of N when vegetation is inactive \rightarrow large N losses, open N cycle

Comparison of foliar δ¹⁵N between temperate and tropical forests



Figure 2. (a) Estimated geographical distribution of soil $\delta^{15}N$ values to 50 cm and (b) estimated geographical trends in $\Delta \delta^{15}N_{\text{plant-soil}}$. Global mean annual temperature and precipitation (0.5 × 0.5 degree grids) data are obtained from *Willmott and Matsuura* [2000].

 Low ¹⁵N enrichment in Northern ecosystems
High ¹⁵N enrichment in tropical ecosystems

+ DIN and gaseous N (strongly ¹⁵N depleted) in tropics + low losses from northern ecosystems (pref. ¹⁵N enriched DON)