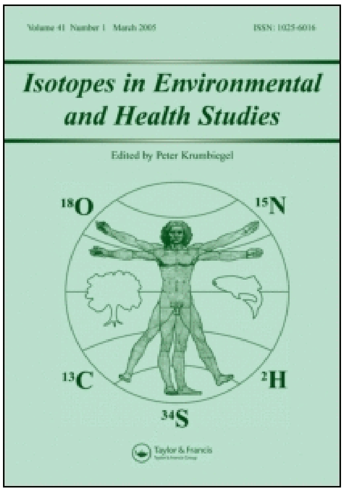

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^{15}N natural abundance of non-fixing woody species in the Brazilian dry forest (caatinga)

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Foliar δ^{15}N values are useful to calculate N_2 fixation and N losses from ecosystems. However, a definite pattern among vegetation types is not recognised and few data are available for semi-arid areas. We sampled four sites in the Brazilian caatinga, along a water availability gradient. Sites with lower annual rainfall (700 mm) but more uniform distribution (six months) had δ^{15}N values of 9.4 and 10.1‰, among the highest already reported, and significantly greater than those (6.5 and 6.3‰) of sites with higher rainfall (800 mm) but less uniform distribution (three months). There were no significant differences at each site among species or between non-fixing legume and non-legume species, in spite of the higher N content of the first group. Therefore, they constitute ideal reference plants in estimations of legume N_2 fixation. The higher values could result from higher losses of ^{15}N depleted gases or lower losses of enriched ^{15}N material.

Keywords: Brazil; forest; isotope ecology; legumes; nitrogen-15; nitrogen cycle; rainfall; tropical vegetation

1. Introduction

Measurements of plant natural δ^{15}N have two important applications: (1) estimation of atmospheric N fixation by single plants [1]; and (2) evaluation of the openness of the N cycle in a given site [2]. Both depend on the δ^{15}N signals of non-fixing plants. In the first case, they are used as reference values and, since atmospheric N has a δ^{15}N of 0, the higher they are the greater is the precision of calculation of the proportion of N fixed by leguminous plants. In the second case, higher δ^{15}N values indicate higher ^{14}N losses from the system and, therefore, a more open N cycle in the site.

In the last decade, ^{15}N natural abundance determinations have revealed considerable variation among different vegetation types [2–8]. In a globally derived data set for site-averaged foliar δ^{15}N, Handley *et al*. [5] registered values from −8.0 to +8.1‰, and even higher values (up to 12.8‰) have been reported for single-species samples [9]. In general, areas with lower mean

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annual rainfall (MAR) have higher δ^{15}N than areas with higher MAR [2,5] and tropical forests have higher values than temperate ones [3]. However, many exceptions of these general trends are found [2,5,10], probably because both characterisations include a broad range of environmental conditions. Therefore, more site data are necessary before a clearer picture can be delineated.

No information is available on δ^{15}N values of plants in the large tropical semi-arid caatinga vegetation of Northeast Brazil. In the same country, non-fixing plants in the Amazonian tropical rainforest have higher δ^{15}N values [8,11] than those of the drier tropical savanna (cerrado central), which are low and variable [12,13] enough to preclude their use as reference plants in N_2 fixation measurements. In the absence of data for the caatinga, it is not known if its N cycle is more or less open than those of the forest and the savanna and if the ^{15}N technique can be used to estimate fixation in the region. As a consequence, there are also no data on N_2 fixation of native plants in the region. Since this estimation is important for several ecological and agronomic reasons, a project was established to determine the δ^{15}N of non-fixing plants in sites chosen in a systematic way to form two pairs, one pair one latitudinal degree apart from the other, along an east–west water availability gradient. The first objective was to determine if the δ^{15}N values were high and similar enough to allow reasonable calculations of N_2 fixation by legume plants. The second objective was to evaluate if there was any spatial and climatic pattern in the δ^{15}N values. If the first hypothesis was validated, the ^{15}N technique could be used to estimate N_2 fixation by leguminous species in the caatinga vegetation. Information related to the second objective would help in designing a proper data gathering methodology.

2. Material and methods

2.1. *Site description*

Four municipalities were selected in the states of Pernambuco and Paraíba, one in the Agreste and one in the Sertão zone of each state (Table 1). Both Agreste municipalities (Remígio in Paraíba and Caruaru in Pernambuco) are located about 100 km from the Atlantic coast in the Borborema Mountain Range. Remígio is approximately 150 km north of Caruaru. The two Sertão municipalities (Santa Teresinha in Paraíba and Serra Talhada in Pernambuco) are at about the same latitudes as the Agreste ones but 300–400 km from the coast, within the large interplanaltic depression zone.

The two Agreste municipalities have total MAR close to 700 mm while the two Sertão municipalities have MAR around 800 mm (Table 1). In spite of the relatively small difference of only 10–15 % lower MAR, the Agreste sites have a higher water availability throughout the year than the Sertão sites. In fact, MAR alone is not a good measure of the rainfall regime, especially in

Table 1. General characteristics of the four chosen municipalities, in the States of Paraíba (PB) and Pernambuco (PE), Brazil.

	Municipality (state)			
Characteristics	Remígio (PB)	Santa Teresinha (PB)	Caruaru (PE)	Serra Talhada (PE)
Coordinates	6°52′S 35°47′W	07°03′S 37°29′W	08°15′S 35°57′W	07°59′S 38°18′’W
Altitude (m)	596	380	545	500
Soil type	Regolithic Neosol	Litholic Neosol	Yellow Argisol	Luvisol
Annual rainfall (mm)	700	824	696	768
Months with water deficit	4–5	9–10	4–5	6–7
Average temperature (°C)	22	26	24	24

the Sertão sites, because of the large interannual variation. In a 30-year time series, total annual rainfall varied from 187 to 1522 mm in the Sertão sites but only from 465 to 1004 mm in the Agreste sites. Rainfall is concentrated mostly in three months (February–April) in the Sertão sites (average of 73 % of the annual rainfall) and the month of highest rainfall represents 41 % of the annual rainfall. In contrast, in the Agreste sites, rainfall is reasonably well distributed over five months, from March to July (67 % of the annual rainfall), and the month of highest precipitation represents only 16 % of the annual rainfall. Average temperatures vary little in the region but Remígio is slightly cooler and Santa Teresinha slightly hotter than the other areas (Table 1).

The more uniform rainfall distribution in the Agreste is reflected in its vegetation. Descriptions of vegetation composition and structure have been published for the sampling sites of Caruaru [14], Serra Talhada [15] and Remígio [16]. The number of species and the height and diameter of plants are higher in the Agreste sites than in Serra Talhada. There is no published description of vegetation in the Santa Teresinha site but unpublished data from this site show a slightly lower number of species and height and diameter of plants than in Serra Talhada, most likely reflecting an even lower water availability.

2.2. *Sampling procedure and analyses*

One sampling site was chosen in each municipality, except in Remígio, where two sites were selected. At Remígio, site 1 was an area with a vegetation that has had little disturbance over the last 60 years while site 2 was a caatinga forest fragment about 100 m from the border of site 1 and subjected to a higher degree of human impact [17]. The pair was intended to evaluate the influence of management under the same environmental conditions. The sampling sites at Caruaru and Serra Talhada were also caatingas that have had little impact in the last decades. The Santa Teresinha site, although located in the experimental station of Universidade Federal de Campina Grande, is subject to a degree of disturbance similar to that of the forest fragment in Remígio (presence of cattle and occasional firewood gathering by the local population).

Two groups of plants were selected for sampling: non-nodulating Leguminosae species [18–21] and species belonging to other families. The Leguminosae species were: *Acacia glomerosa* Benth, *Amburana cearensis* (Allemão) A.C. Smith, *Bauhinia cheilantha* (Bong.) Steud, *Caesalpinia ferrea* Mart. ex Tul, *Caesalpinia pyramidalis* Tul., *Senna macranthera* (Collad.) H.S. Irwin and Barneby and *Senna spectabilis* (DC.) H.S. Irwin and Barneby. The species belonging to other families were: *Aspidosperma pyrifolium* Mart., *Commiphora leptophloeos* Mart. J.B. Gillett, *Croton sonderianus* Muell. Arg. *and Tabebuia impetiginosa* (Mart. ex DC.) Standl. They are all tree or large shrub species commonly found in caatinga areas and their presence in each site, except Santa Teresinha, was known from previous studies in the same areas [14–16]. The two groups were selected because the legume species, in spite of being non-nodulating, tend to have higher leaf N content than plants belonging to other families [12,22].

In each site, five to six sampling plots were established. The centre of each plot was established in an area where the largest diversity of the previously defined sampling species was found. Plants belonging to these species were sampled usually within 10 m distance from the centre point but occasionally up to 20 m if no other plant of one species had been found at a shorter distance. Plots were at least 50 m distant from each other. Even when extending the plot size to include a nearby plant, not all selected species were sampled in each plot and for most species we did not obtain the five replicates originally intended.

All plants selected for sampling had at least 5 cm diameter at the base of the stem and 3 m of total height. Approximately 100–200 g of dry mass of fully expanded green leaves from each selected plant were cut, oven dried at 60 °C, ground and analysed for their total N and ^{15}N abundance contents. Total N was analysed by the micro Kjeldahl procedure at DEN Laboratory in Recife, Brazil. For determination of ^{15}N abundance, plant sub-samples were sent to the Mass

Spectrometry Laboratory of the Institute of Tropical Agriculture at the University of Göttingen, Germany, where they were analysed using a Finnegan delta plus MAT 251 with ConFlo and an internal standard of acetanilide. The results are expressed as a 'delta' notation:

$$\delta = \left(\frac{R_{\text{sample}}}{R_{\text{standard}}} - 1\right) \times 1000,$$

where R_{sample} and R_{standard} are the ratio ^{15}N:^{14}N of the sample and the standard (air), respectively.

The data were subjected to usual statistical analysis, comparing different plant groups. Using individual plant values as replicates, comparisons were made of: (1) averages of all species of a same site; (2) averages of the group of all non-fixing legumes versus all non-legume species of a same site; (3) averages of non-fixing legume, non-legume and all species of the four sites; and (4) averages of non-fixing legume, non-legume and all species of the two Remígio sites (little disturbed and forest fragment). Considering the absence of significant differences among species of a same site, values of different species were used as replicates to compare plot averages of a same site. Significant differences were established considering the 0.05 probability level.

3. Results

There were no significant differences between averages of $\delta^{15}N$ values of the two Agreste sites (9.4 and 10.1 ‰) nor between the two Sertão sites (6.7 and 6.3 ‰) but those of the Agreste were significantly higher than those of the Sertão (Table 2). The average of the disturbed forest fragment at Remígio (10.2 ‰) was not significantly different from that of the little disturbed caatinga at the same site (10.1 ‰).

Within each site, the similarity of $\delta^{15}N$ of all plants was remarkable. The averages of the legume group were not different from the non-legume group in any of the sites (Table 2), despite differences in the leaf N content (Table 3). The leaf N contents of the legume plants were higher than those of the non-legume group but the differences were only significant in the Agreste sites, which also tended to have higher values than the Sertão sites.

Table 2. Leaf $\delta^{15}N$ of plants belonging to non-N_2 fixing species of Leguminosae or of other families, collected at four different sites in the semi-arid caatinga of Paraíba and Pernambuco States, Brazil.

Species	Remígio		Caruaru (undisturbed)	Serra Talhada (undisturbed)	Santa Teresinha (disturbed)
	Undisturbed	Disturbed			
Leguminosae					
A. glomerosa	(1) 10.63	(2) 11.37 ± 1.40	(4) 9.39 ± 0.51	–	–
A. cearensis	(5) 10.07 ± 0.54	–	–	(4) 5.12 ± 0.97	–
B. cheilantha	(4) 9.92 ± 0.72	(1) 7.63	–	(5) 6.30 ± 1.56	(5) 5.87 ± 0.63
C. ferrea	–	(1) 11.58	–	(2) 6.76 ± 0.25	–
C. pyramidalis	(4) 10.03 ± 0.48	(4) 10.01 ± 1.76	(5) 9.48 ± 1.06	(5) 6.54 ± 1.28	(3) 6.76 ± 0.67
S. macranthera	(3) 10.61 ± 050	–	–	(3) 5.59 ± 0.44	–
S. spectabilis	–	(2) 10.29 ± 1.98	–	(1) 8.34	–
Average	10.19 ± 0.60	10.19 ± 1.71	9.73 ± 0.96	6.01 ± 1.24	6.16 ± 0.72
Other families					
A. pyrifolium	(5) 9.71 ± 0.68	(5) 11.14 ± 1.40	–	(5) 7.75 ± 1.62	(2) 7.20 ± 0.97
C. leptophloeos	(6) 10.65 ± 0.65	–	–	(2) 6.10 ± 0.61	–
C. sonderianus	(6) 9.84 ± 0.83	(5) 9.22 ± 1.79	(4) 8.46 ± 2.55	(5) 5.73 ± 1.20	(2) 8.27 ± 1.37
T. impetiginosa	–	–	–	–	(1) 6.08
Average	10.08 ± 0.80	10.21 ± 1.79	8.46 ± 2.55	6.64 ± 1.58	7.40 ± 1.23
Overall average	10.12 ± 0.71 A	10.22 ± 1.72 A	9.44 ± 1.49 A	6.34 ± 1.37 B	6.71 ± 1.27 B

Note: Numbers in parentheses refer to replicates.

Table 3. Average leaf nitrogen concentrations of plants belonging to non-N_2 fixing species of Leguminosae or of other families, collected at five different localities in the semi-arid caatinga of Paraíba and Pernambuco States, Brazil.

Site	Leguminosae	Other families	Average
Remígio (undisturbed area)	3.08 ± 0.42Aa	2.29 ± 0.34Ba	2.70 ± 0.54a
Remígio (disturbed area)	3.09 ± 0.52Aa	2.29 ± 0.26Ba	2.69 ± 0.58a
Caruaru	2.50 ± 0.58Ab	1.81 ± 0.20Bab	2.31 ± 0.60ab
Serra Talhada	2.24 ± 0.45Ab	1.75 ± 0.24Ab	2.04 ± 0.41b
Santa Teresinha	1.99 ± 0.54Ab	1.62 ± 0.15Ab	1.86 ± 0.48b
Average	2.58 ± 0.63A	2.04 ± 0.38B	

Note: Averages followed by the same capital letter in the line and small letter in the column are not significantly different at the 0.05 level.

There were also no significant differences in δ^{15}N among species in any of the sites (Table 2). The largest absolute differences between averages of two species, within the same site, varied from 0.85 (Remígio) to 2.4 δ^{15}N units (Santa Teresinha). In the two Sertão sites, the lowest absolute values were for two of the legume species (*B. cheilantha* in Santa Teresinha and *S. macranthera* in Serra Talhada) and in the two Agreste sites for a non-legume species (*C. sonderianus*). The opposite happened with the highest values: in the two Agreste sites they were for two legume species (*A. glomerosa* in Remígio and *Acacia paniculata* in Caruraru) and in the two Sertão sites for non-legumes (*C. sonderianus* in Santa Teresinha and *A. pyrifolium* in Serra Talhada). However, in all sites there were legume and non-legume species with very similar values.

Considering the similarity in δ^{15}N averages of species, the values of all plants in each plot (data not shown) were used as replicates to test for spatial differences within each site. In any of the sites, there were no significant differences among plots. The absolute difference values varied from 1.06 (Remígio) to 2.24 ‰ (Serra Talhada).

4. Discussion

The site-averages of foliar δ^{15}N reported in this paper for the Agreste sites (9.4–10.1 ‰) were among the highest reported in the literature [5,10,23]. The highest average previously reported went up to about 11 ‰ [23] but the large review of Handley *et al*. [5] showed less than 10 sites around the world having averages above 7.0 ‰. Most of these sites are in tropical dry regions, in Africa and Australia, and were the basis of the confirmation by Handley *et al*. [5] of the tendency proposed by Austin and Vitousek [2] of increasing δ^{15}N values in decreasing MAR sites. The fact that our high δ^{15}N caatinga values were obtained under semi-arid conditions seems to confirm the hypothesis proposed by those authors and corroborated by some others [9,10].

A comparison of the Agreste and Sertão values also seems to confirm the tendency. The Sertão sites with their higher MAR had significantly lower δ^{15}N averages (6.7 and 6.3 ‰) than the Agreste sites (9.4 and 10.1 ‰). However, the differences in mean average rainfall are rather small (100 mm in averages of 700–800 mm, or less than 15 %) and their large annual variations indicate that these means are not a good measure of local precipitation. A critical examination of world values shows that the general tendency is contradicted by both types of exceptions: high MAR with high δ^{15}N and low MAR with low δ^{15}N. Values of δ^{15}N as high as those of the Sertão sites have been reported in Amazon sites with MAR around 2000 mm [8,11] and also in other tropical rain forests [3]. On the other hand, there are sites with similar to and even lower MAR than the Agreste ones and much lower δ^{15}N values. In the paper where this hypothesis was proposed [2], the site with lowest MAR (500 mm) had a δ^{15}N value of only 0.04 ‰. In Africa, several sites with

MAR below 700 mm had lower δ^{15}N values than those of the Agreste, down to values as low as 1.0 ‰ [5,9,10].

Exceptions to the MAR tendency have been observed not only at this global scale, comparing sites in different continents, as was done by Handley *et al*. [5]. They are also present at the continental level in Africa [10] and in South America, if we compare the data from our caatinga sites in the present study with those of Amazonia [4,8,11] and cerrado sites [12,13]. Among these three regions, the lowest average δ^{15}N values (0.49–1.73 ‰) were reported for cerrado sites [12] which have MAR values (around 1500 mm) intermediate between Amazonian (>2000 mm) and caatinga sites (<1000 mm).

In addition to these comparisons at the continental scale, differences in δ^{15}N that could not be explained by variations in MAR have also been reported at the local level [24,25]. The explanation for these differences was shifted to other variables affecting water availability in the soil–plant system. When larger areas are considered, MAR is probably the variable responsible for the largest variation in water availability besides being the simplest to refer and the most frequently available data. MAR was certainly used as an approximation to water availability in all regional and continental comparisons of δ^{15}N, even if not explicitly stated [2,5,9,10].

When comparing the two caatinga sites in the present study, MAR may not be the best estimator of water availability. Not only are the mean values not very different, but the differences in rainfall distribution indicate that water is more available, throughout the year, in Agreste than in Sertão, in spite of the slightly lower MAR of the former. The larger size of the vegetation and the greater number of species seem to confirm the better growing conditions in Agreste, most of which are determined by water availability since other soil characteristics are more favourable for plant growth and development in the Sertão sites [14–16]. If we consider these indications, the sites with higher water availability also had higher δ^{15}N. However, in a continental scale, comparing Amazonian, cerrado and caatinga sites, water availability has the same pattern as MAR and, in this comparison, cerrado, with its intermediate availability, has the lowest δ^{15}N, while the two extreme regions have similar ^{15}N concentrations. Clearly, MAR or water availability alone cannot explain the variations in δ^{15}N.

Martinelli *et al*. [3] proposed that tropical forests had higher δ^{15}N values than temperate ones, implying an effect of latitude. The similarity of δ^{15}N values between both the two Agreste and the two Sertão sites indicates that they were not affected by latitude. However, the differences in latitude were small: only one degree. Besides, latitude *per se* has no direct influence in the soil–plant system. Its clearer indirect influence is higher temperature at lower latitudes, a factor that is present in the broad comparison of tropical and temperate forests but not at the regional scale of our caatinga sites. Differences in temperature were small (maximum of 4 °C) and more affected by local conditions than by latitude, both the lowest and the highest temperatures at the sites of the same parallel (Remígio and Santa Teresinha). Comparing this pair of sites, the one with lowest temperature had the highest δ^{15}N value, contrary to the expected tendency of hotter areas with higher ^{15}N enrichment. Temperature has a direct effect on plant and soil microorganism metabolism and an indirect effect on water availability, higher temperatures resulting in higher potential evapotranspiration. Both higher metabolism and higher evapotranspiration may result in faster N cycling in the soil–plant system, provided other growth factors are not limited.

The faster N cycling of tropical forests compared with temperate ones has been proposed as one of the causes of their higher δ^{15}N [3]. The higher δ^{15}N values in the Agreste sites, which most likely have a faster cycle than the sites at Sertão, seem to confirm the tendency. However, the tendency does not hold at the regional or continental scales. Forests and other vegetation types in tropical dry areas have their N cycles limited by water availability and yet have the highest δ^{15}N signals. N cycles in cerrado areas are certainly faster than those in caatinga and their δ^{15}N values are much lower.

Ultimately, the causes of higher δ^{15}N in a soil–plant system have to be higher inputs of ^{15}N enriched nitrogen and/or higher losses of ^{15}N depleted nitrogen [5]. Since, in general, the balance of N inputs tends to have a δ^{15}N signal close to zero, it is assumed that sites with higher δ^{15}N have higher relative losses of ^{15}N depleted nitrogen. Therefore, high N losses have become the standard explanation for the high δ^{15}N of any site. They are referred to as the openness of the N cycle [2]. In most cases, the real losses are not known but hypotheses are created to justify their high relative level in any high δ^{15}N site. In the absence of data, they are theoretical speculations. For these reasons, measurements of N losses coupled with determination of the ^{15}N abundance in the different forms of N are strongly needed but they are difficult to make [26]. In a few measured cases, in African dry areas, the losses did not justify the δ^{15}N signals [9].

The results indicate a series of variables with little effect on the ^{15}N discrimination (Table 2). Anthropic disturbance caused little alteration in the δ^{15}N of plants in the disturbed fragment compared with those of the more preserved neighbour site at Remígio. The nature of disturbance (more frequent cattle grazing and eventual cutting of wood) is less drastic than those assumed to cause higher N losses from the system, like burning and clear cutting [27–29].

Plot averages, within each site, were not significantly different and varied in a relatively narrow range (data not shown), implying that local scale (less than 1 km distance) variations in environmental conditions had little effect in the N cycle. Similar plot comparisons were not found in the literature but comparisons of sites located at greater distances within a 1350 ha ecological reserve in the cerrado region also showed little variation, in spite of a marked difference in vegetation physiognomy [13]. Papers relating differences among sites usually sampled places further apart than our plots or these cerrado sites [8,12]. One important consequence of the absence of differences among plots is that it simplifies sampling of reference plants when trying to quantify N_2 fixation using ^{15}N natural abundance.

The δ^{15}N averages of the group of non-fixing legume plants were not different from those of the non-legume species group and species within the same site had similar ^{15}N abundances. Significant differences between the two groups of plants were found in cerrado sites, with the interesting fact that, in some places, non-legumes had higher δ^{15}N than non-nodulating legumes while in other sites, the opposite occurred [12]. Within the same ecological reserve [13], non-legumes had a slightly higher, although significant, δ^{15}N than legumes (0.52 versus −0.52 ‰) but the result may have been influenced by the absence of a clear separation of fixing and non-fixing legumes.

The similarity between the non-fixing legume and the non-legume groups is more striking because the first group had a significantly higher N content than the second one, in the more humid Agreste sites (Table 3). This superiority has been registered elsewhere [12,13,22]. McKey [30] suggested that, in general, Leguminoseae are nitrogen-demanding plants that maintain high levels of N in leaf tissue in order to maximise photosynthetic rates per unit leaf area. This, in turn, allows them to amortise the carbon costs of leaf construction and enables them to produce and drop leaves quickly in response to changing resource availability.

The absence of differences among species and plants within the same site contrasts sharply with results obtained in other vegetation types. Differences of up to 4–5 δ^{15}N units have been reported among species of a single site [3,12,13] and up to 8 δ^{15}N units among plants of the same site [8]. In the present study, the limitation of species to deciduous trees and large shrubs may have contributed to this uniformity. They are probably absorbing N from the same sources, with roots distributed in all layers of the relatively shallow soils present in all sites. The number of species was also not very high and the same ones were collected in all sites, reducing the variability of the whole sample. It is possible that other caatinga plant types (vines, epiphytes, herbaceous, etc) present different δ^{15}N, but no data are yet available.

5. Conclusions

Plants in semi-arid Northeast Brazil had high $\delta^{15}N$ values, indicating that their cycles are subjected to high losses of ^{15}N, depleted nitrogen. Plants in the Agreste sites, which have a lower mean average but a more uniform rainfall distribution along the year than the Sertão sites, had the highest $\delta^{15}N$ values, and among the highest already reported in the world.

The high $\delta^{15}N$ values of all non-fixing plants within a site were very similar, both spatially and among species. This isotopic pattern is ideal for studies of biological N_2 fixation, facilitating the selection of reference plants and the sampling procedure.

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References

[1] G. Shearer and D.H. Kohl, N_2-fixation in Field Settings: Estimations Based on Natural ^{15}N Abundance, *Aust. J. Plant Physiol.* **13**, 699 (1986).
[2] A.T. Austin and P.M. Vitousek, Nutrients Dynamics on a Precipitation Gradient in Hawai'i, *Oecologia* **113**, 519 (1998).
[3] L.A. Martinelli, M.C. Piccolo, A.R. Townsend, P.M. Vitousek, E. Cuevas, W. McDowell, G.P. Robertson, O.C. Santos, and K. Treseder, Nitrogen Stable Isotopic Composition of Leaves and Soil: Tropical Versus Temperate Forests, *Biogeochemistry* **46**, 45 (1999).
[4] J.C. Roggy, M.F. Prévost, J. Garbaye, and A.M. Domenach, Nitrogen Cycling in the Tropical Rain Forest of French Guiana: Comparison of Two Sites with Contrasting Soil Types Using $\delta^{15}N$, *J. Trop. Ecol.* **15**, 1 (1999).
[5] L.L. Handley, A.T. Austin, D. Robinson, C.M. Scrimgeour, J.A. Raven, T.H.E. Heaton, S. Schmidt, and G.R. Stewart, The ^{15}N Natural Abundance ($\delta^{15}N$) of Ecosystem Samples Reflects Measures of Water Availability, *Aust. J. Plant Physiol.* **26**, 185 (1999).
[6] Z. Eshetu and P. Högberg, Effects of Land Use on ^{15}N Natural Abundance of Soils in Ethiopian Highlands, *Plant Soil* **222**, 109 (2000).
[7] S.K. Arndt, A. Kahmen, C. Arampatsis, M. Popp, and M. Adams, Nitrogen Fixation and Metabolism by Groundwater-Dependent Perennial Plants in a Hyperarid Desert, *Oecologia* **141**, 385 (2004).
[8] J.P.H.B. Ometto, J.R. Ehleringer, T.F. Domingues, J.A. Berry, F.Y. Ishida, E. Mazzi, N. Higuchi, L.B. Flanagan, G.B. Nardoto, and L.A. Martinelli, The Stable Carbon and Nitrogen Isotopic Composition of Vegetation in Tropical Forests of the Amazon Basin, Brazil, *Biogeochemistry* **79**, 251 (2006).
[9] J.N. Aranibar, L. Otter, S.A. Macko, C.J.W. Feral, H.E. Epstein, P.R. Dowty, F. Eckardt, H.H. Shugart, and R.J. Swap, Nitrogen Cycling in the Soil-plant System along a Precipitation Gradient in the Kalahari Sands, *Glob. Chang. Biol.* **10**, 359 (2004).
[10] R.J. Swap, J.N. Aranibar, P.R. Dowty, W.P. Gilhooly III, and S.A. Macko, Natural Abundance of ^{13}C and ^{15}N in C3 and C4 Vegetation of Southern Africa: Patterns and Implications, *Glob. Chang. Biol.* **10**, 350 (2004).
[11] C. Gehring and P.L.G. Vlek, Limitations of the ^{15}N Natural Abundance Method for Estimating Biological Nitrogen Fixation in Amazonian Forest Legumes, *Basic Appl. Ecol.* **5**, 567 (2004).
[12] J.I. Sprent, I.E. Geoghegan, P.W. Whitty, and E.K. James, Natural Abundance of ^{15}N and ^{13}C in Nodulated Legumes and other Plants in the Cerrado and Neighboring Regions of Brazil, *Oecologia* **105**, 440 (1996).
[13] M.M.C. Bustamante, L.A. Martinelli, L.A. Silva, P.B. Camargo, C.A. Klink, T.F. Domingues, and R.V. Santos, ^{15}N Natural Abundance in Woody Plants and Soils of Central Brazilian Savannas (Cerrado), *Ecol. Appl.* **14**, 200 (2004).
[14] F.G. Alcoforado-Filho, E.V.S.B. Sampaio, and M.J.N Rodal, Florística e fitossociologia de um Remanescente de Vegetação Caducifólia Espinhosa Arbórea em Caruraru, Pernambuco, *Acta Bot. Brasilica* **17**, 287 (2003).
[15] E.M.N. Ferraz, M.J Rodal, and E.V.S.B. Sampaio, Physiognomy and Structure of Vegetation Along an Altitudinal Gradient in the Semi-Arid Region of Northeastern Brazil, *Phytocoenologia* **33**, 71 (2003).
[16] I.M. Pereira, L.A. Andrade, M.R.V. Barbosa, and E.V.S.B. Sampaio, Composição Florística e Análise Fitossociológica do Componente Arbustivo-Arbóreo de um Remanescente Florestal no Agreste Paraibano, *Acta Bot. Brasilica* **16**, 357 (2002).
[17] I.M. Pereira, L.A. Andrade, E.V.S.B. Sampaio, and M.R.V. Barbosa, Use-History Effects on Structure and Flora of Caatinga, *Biotropica* **35**, 154 (2003).

[18] O.N. Allen and E.K. Allen, *The Leguminosae: A Source Book of Characteristics, Use and Nodulation* (University of Wisconsin Press, Wisconsin, 1981).
[19] S.M Faria, A.A. Franco, R.M. Jesus, M.S. Menandro, J.B. Baitello, E.S.F. Mucci, J. Dobereiner, and J.I. Sprent, New Nodulating Legume Trees from Southeast Brazil, *New Phytol*. **98**, 317 (1984).
[20] S.M. Faria, H.C. Lima, A.A. Franco, E.S.F. Mucci, and J.I. Sprent, Nodulation of Legume Trees from Southeast Brazil, *Plant Soil* **99**, 347 (1987).
[21] S.M. Faria and H.C. Lima, Additional Studies of the Nodulation Status of Legume Species in Brazil, *Plant Soil* **200**, 185 (1998).
[22] L.A. Martinelli, S. Almeida, I.F. Brown, M.Z. Moreira, R.L. Victoria, C.S. Filoso, A.C. Ferreira, and W.W. Thomas, Variation in Nutrient Distribution and Potential Nutrient Losses by Selective Logging in a Humid Tropical Forest of Rondônia, Brazil, *Biotropica* **32**, 597 (2000).
[23] E.D. Schulze, G. Gebauer, H. Ziegler, and O.L. Lange, Estimates of Nitrogen Fixation by Trees on an Aridity Gradient, *Oecologia* **88**, 451 (1991).
[24] C.T. Garten, Variation in Foliar ^{15}N Abundance and the Availability of Soil Nitrogen on Walker Branch Watershed, *Ecology* **74**, 2098 (1993).
[25] L.L. Handley and C.M. Scrimgeour, Terrestrial Plant Ecology and ^{15}N Natural Abundance: The Present Limits to Interpretation for Uncultived Systems with Original Data from a Scottish Old Field, *Adv. Ecol. Res.* **27**, 133 (1997).
[26] L. Högbom, U. Nilsson, and G. Örlander, Nitrate Dynamics after Clear Felling Monitored by In vivo Nitrate Reductase Activity (NRA) and Natural ^{15}N Abundance of *Deschampsia flexuosa* (L.) Trin, *For. Ecol. Manage.* **160**, 273 (2002).
[27] P. Högberg, ^{15}N Natural Abundance in Soil-Plant Systems, *New Phytol.* **137**, 179 (1997).
[28] C. Neill, M.C. Piccolo, P.A. Steudler, J. Melillo, B.J. Feigel, and C.C. Cerri, Nitrogen Dynamics in Soils of Forests and Active Pastures in the Western Brazilian Amazon Basin, *Soil Biol. Biochem.* **27**, 1167 (1995).
[29] M.C. Piccolo, C. Neil, and C.C. Cerri, Natural Abundance of ^{15}N in Soils along Forest to Pasture Chronosequences in the Western Brazilian Amazon Basin, *Oecologia* **99**, 112 (1994).
[30] D. McKey, in *Advances in Legume Systematics 5: The Nitrogen Factor*, edited by J.I. Sprent and D. McKey (Royal Botanic Gardens, Kew, UK, 1994).