

## Nitrogen use in mixed tree crop plantations with a legume cover crop

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## Abstract

In a multi-strata agroforestry system in central Amazonia, we studied the nitrogen (N) use of two indigenous fruit tree species, Theobroma grandiflorum Willd. (ex Spreng.) K. Schum. (cupuaçu) and Bactris gasipaes Kunth. (peachpalm) for heart of palm production, and a legume cover crop, Pueraria phaseoloides Roxb. (Benth.) (pueraria).  $^{15}$ N was applied at a rate of 1 kg ha<sup>-1</sup> twice at the beginning and at the peak of the rainy season, in a split plot design under either cupuaçu, peachpalm or pueraria together with fertilizer N usually applied (95.4 and 42.4 g N tree<sup>-1</sup> for cupuaçu and peachpalm, respectively). Plant and soil <sup>15</sup>N content and total <sup>15</sup>N uptake were measured for 1 year. The highest N uptake by the trees occurred from areas underneath their canopy being more than 70% of their total N uptake. During the dry season, pueraria also took up most of its N (more than 70%) from the area underneath its own canopy. During the rainy season, however, pueraria utilized N from the area under cupuaçu (27–40%) and peachpalm (34–47% of the total N uptake by pueraria). Cupuaçu took up between 12 and 26% of its N from the area covered by pueraria, peachpalm slightly less with 10 to 18% (significant only at the end of the rainy season; P<0.05). Competition for N uptake between the trees was negligible. The above-ground recovery was highest in cupuaçu (15% of the applied <sup>15</sup>N), followed by pueraria (11%) and peachpalm (3%). Pueraria proved to be very important for the N cycling in the mixed tree cropping system recovering most (31%) of the applied <sup>15</sup>N in plant and soil in comparison to cupuaçu (20%) and peachpalm (21%). However, the natural  $^{15}$ N abundance of the tree leaves did not show a significant transfer of biologically fixed N<sub>2</sub> from pueraria to the trees (P > 0.05) and the cover crop did not improve tree N nutrition. The investigated fruit trees did not benefit from biologically fixed  $N_2$ of the legume cover crop due to their low lateral root activity and the high available soil N contents largely being an effect of the amount and placement of mineral fertilizer.

## Introduction

The Ferralsols in the central Amazonian lowlands are highly weathered and have very low available nutrient contents (Cravo and Smyth, 1997). In annual cropping systems, high amounts of nutrients have to be supplied to maintain soil fertility at an acceptable level. Land use systems with perennial crops may be superior in this environment, as soil fertility decline through soil organic matter decomposition and nutrient leaching may be lower than in annual cropping systems (Van Wambeke, 1992). However, considerable soil fertility decline was reported even in tree plantations such as peachpalm monocultures compared to pasture and cocoa in Costa Rica (Fernandes and Sanford, 1995). The combination of different tree species may offer a better possibility for nutrient conservation. But in some of these systems, soil fertility may decrease as with cocoa under different shade trees in Ghana (Ahenkorah et al.,

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1987). The nutrient use by trees in mixed cropping systems is not fully known, and its further understanding would help to design land use systems in the humid tropics by minimizing losses of applied fertilizer and optimizing nutrient use for crop production.

The use of a legume ground cover may improve soil N contents through atmospheric N<sub>2</sub>-fixation and on the long term provide additional N nutrition to associated trees. However, the proportion of N uptake by the trees from the area between the rows, where the legume is growing, has been rarely determined. Some results exist from root activity experiments with various monocropped, tropical fruit trees where nutrient uptake was compared at discrete distances from the stem (IAEA, 1975). N uptake from whole areas in mixed cropping systems has not been reported so far.

In this study, we addressed the following questions: (i) What is the area from where trees take up their N in a mixed cropping system with a legume cover crop? and (ii) How important is the legume cover crop for the N cycle of the whole cropping system?

## Materials and methods

The study was carried out in the humid rainforest region of the central Amazon in 1997 and 1998. The rainfall distribution is unimodal with a maximum between December and May and a mean annual precipitation of 2503 mm (1971–1993). The soils are classified as Xanthic Ferralsols (FAO, 1990). They are deep and clayey, with low pH (in H<sub>2</sub>O; 4.0–4.5), medium levels of organic C (28.4 mg g<sup>-1</sup>) and N (2.3 mg g<sup>-1</sup>) but low in P content (6.1 mg g<sup>-1</sup>), cation exchange capacity (4.9 mmol<sub>c</sub> kg<sup>-1</sup>) and base saturation (33%).

#### Experimental setup

The site was cleared from secondary forest in 1992 in order to establish several cropping systems mainly with fruit trees. In the study presented, two fruit tree species of high regional importance, *Bactris gasipaes* Kunth. (peachpalm or pupunha, Arecaceae) and *Theobroma grandiflorum* Willd. (ex Spreng.) K. Schum. (cupuaçu, Sterculiaceae), and a cover crop of *Pueraria phaseoloides* Roxb. (Benth.) (pueraria, Fabaceae) were investigated in a mixed cropping system. The studied agroforestry system also contained rows of rubber trees of the same age as the fruit trees located between experimental units described here, which



*Figure 1.* Layout of the experiment; circles show positions of cupuaçu in area CP and peachpalm in area PP;  $^{15}$ N was applied to CP, PP or PU area (CP cupuaçu, PP peachpalm, PU pueraria); distances in [m].

were not investigated further (142 plants  $ha^{-1}$ ). In December 1992, cupuaçu and peachpalm were planted in rows with 5 m distance, leaving 6 m between cupuaçu and 2 m between peachpalm within the rows (100 and 467 plants  $ha^{-1}$ , respectively). The peachpalm was managed for palmito production (heart of palm) and cut every 4-5 months. Pueraria was sown between the trees and manually cut under the tree canopy every 3-4 months. The residues were pulled from underneath the canopy but remained in the field. Fertilizer was applied according to local recommendations at the beginning of the rainy season in December and towards the end of the rainy season in May using 95.4 and 42.4 g N (as ammonium sulfate), 77 and 11 g P (as TSP) per plant and year for cupuaçu and peachpalm, respectively. Lime was broadcast on the soil surface at a rate of 1.9 Mg  $ha^{-1}$  in 1992. By the start of the presented experiment, the trees were 5-years old and in full production.

Within the main plots of  $48 \times 32$  m, three microplots of  $3 \times 5$  m were randomly chosen with two peachpalms and one cupuaçu tree (Figure 1). <sup>15</sup>N enriched ammonium sulfate was added to each of the three microplots in different concentrations either in an area under cupuaçu (CP: 1.5 g<sup>15</sup>N m<sup>-2</sup>), peachpalm (PP:  $0.5 \text{ g}^{15}\text{N m}^{-2}$ ) or pueraria (PU:  $0.14 \text{ g}^{15}\text{N}$  $m^{-2}$ ), in order to compensate for the differing sizes of the areas  $(1, 3 \text{ and } 11 \text{ m}^2, \text{ respectively; Figure 1})$ . The dimension of the application area under the trees (CP and PP) was determined according to the area where fertilizer was usually applied leaving the space between the trees covered by pueraria (PP area Figure 1, also referred to as 'alley' in the text). In contrast to mineral fertilizer applications, the three <sup>15</sup>N application areas were rectangular approximations of the meaningful areas 'where fertilizer is applied' and 'under the pueraria' due to a calculation of uptake per unit area. At times of regular fertilization, at the beginning of December 1997 (end of the dry season) and at the end of April 1998 (peak of the wet season), the <sup>15</sup>N was applied in aqueous solution with 10 atom% excess <sup>15</sup>N using a manual sprayer. The amount of <sup>15</sup>N tagged ammonium sulfate applied to the trees (15 and 5 g N to cupuaçu and peachpalm, respectively) was not an effective fertilization (being only 15 and 13% of the N fertilizer applied), but a labeling of different soil surface areas of the cropping system in order to determine root activity distribution. The main plots (application to the different plants) were arranged in a randomized complete block design with three replicates, the subplots (plant species within one microplot) were considered as a split plot (Little and Hills, 1978). Assigning variances in the described manner, it was possible to compute the effects of each plant species (the trees and the cover crop) on  $^{15}$ N uptake.

#### Plant and soil sampling

The N isotope composition was measured in the three plant species on February 15, April 15, May 14, June 1 and November 14, 1998. From February to June 1998, the youngest fully developed leaves were sampled, the middle leaflets of peachpalm and the whole leaves of cupuaçu and pueraria. No significantly different N isotope composition existed between the youngest leaves and the whole plant of pueraria, therefore only the results of the latter are shown here. In November 1998, a mixture of young and old leaves and branches as well as stem samples of the two tree species were obtained separately. Random sampling across the whole PU area was done for pueraria, weighted samples were taken from both peachpalms in the PP area (Figure 1). In each of the three areas (Figure 1) labeled with <sup>15</sup>N composite soil samples from four subsamples were collected at 0-0.1, 0.1-0.3, 0.3-0.5 and 0.5-1.0 m depths on April 15, 1998, using a purkhauer auger (diameter 30 mm).

# Calculation of ${}^{15}N$ uptake and biomass determinations

From the <sup>15</sup>N contents in excess of natural abundance, the relative uptake from one area was calculated in relation to the total uptake from all three areas (Figure 1). In September 1998, total above-ground biomass was determined using allometric relationships for cupuaçu (biomass: stem = volume<sub>stem</sub> × 0.4997, n = 7; branches =  $\sum 11.8 \times \text{dia}_{\text{branch}}^{3.02}$  n = 39; leaves  $= \sum 23.4 \times \text{dia}_{\text{branch}}^{2.39}$ , n = 36; all P < 0.05 for independent samples; Wolf, 1997) in order to calculate total N uptake and <sup>15</sup>N recovery. Additional destructive sampling of the whole plant was made for peachpalm and pueraria. The peachpalm shoots were dried at 70°C until constant weight and reweighed. Pueraria was harvested in three subplot areas of one square meter each. Uptake of applied <sup>15</sup>N was calculated from total above-ground biomass and <sup>15</sup>N contents of leaves, branches and stem. Recovery of <sup>15</sup>N was determined as the ratio of uptake to application. For peachpalm, both individual plants were sampled and uptake per plant was calculated as a mean plant occupying the space of both plants together (PP area). The uptake per unit area was calculated per land use system summing up both peachpalm plants. The recovery of <sup>15</sup>N by peachpalm and pueraria was calculated with the <sup>15</sup>N uptake determined by direct biomass measurements, the recovery of cupuaçu by allometric relationships. The biomass and uptake per unit areas was computed assuming that the agroforestry system only consisted of the presented peachpalm-cupuaçu arrangement not considering the rows of interplanted rubber trees outside the experimental units. Recovery of <sup>15</sup>N in below-ground biomass was not accounted for. It may constitute a large part of total plant recovery, more for peachpalm (root-shoot ratio > 0.86) than for cupuaçu (0.3–0.5; Haag, 1997).

## Uptake of biologically fixed N

Using the natural <sup>15</sup>N abundance technique, the utilization of biologically fixed N from pueraria by cupuaçu and peachpalm was compared between trees intercropped with the legume cover crop at a low and high abundance. The treatment with high pueraria abundance (P1) was the identical one used for the enrichment experiment described above but with a sufficient distance between plots with and without <sup>15</sup>N enrichment (>50 m). The low pueraria abundance (P2) treatment consisted of the same experimental units (Figure 1), planting history and management as the experiment described above. However, instead of the rubber trees, Brazil nut (Bertholletia excelsa Humb. & Bonpl.) and annatto (Bixa orellana L.) were interplanted with peachpalm and cupuaçu. The lower pueraria abundance was most likely caused by a higher shading intensity due to the large Brazil nut trees planted outside the experimental units (Figure 1). Production of cupuaçu and peachpalm, however, were not

affected by the two different cropping systems at the time of measurement (Macêdo, unpubl. data). Therefore, a measured N isotope difference of the trees between cropping systems can be attributed to pueraria abundance. Pueraria biomass amounted to 5 and 3 Mg ha<sup>-1</sup>, for high and low abundance, respectively (Silva Jr. and Lehmann, unpubl. data). The youngest fully developed leaves were taken from cupuaçu and pueraria, and the middle leaflets from the youngest fully developed leaves of peachpalm. The foliar N isotope composition was measured in the different plants at the end of the dry season in November 1997, and at the end of the wet season in June 1998. The experiment was replicated three times.

## N isotope analyses

The leaf samples were dried at 70 °C for 48 h. The soil samples were air-dried and finely ground with a ball mill. The <sup>15</sup>N-enriched samples were analyzed using an Elemental Analyzer (Carlo Erba NA 1500; for Dumas combustion) connected to an isotope mass spectrometer (FINNIGAN MAT delta E) via a split interface. <sup>15</sup>N natural abundance was determined with an elemental analyzer (Fisons 1108) coupled via a ConFlo II Interface to a delta S isotope mass spectrometer (FINNIGAN MAT).

## Statistical analyses

The analyses of variance were computed using a split plot design (STATISTICA Version 5). For the analysis of the foliar <sup>15</sup>N contents, the logarithmic values were used due to inhomogeneity of variances. In case of significant main or subplot effects or interactions, individual cell means for the respective level were compared using LSD at P<0.05 (Little and Hills, 1978).

## Results

## Foliar <sup>15</sup>N dynamics

The foliar N contents of pueraria (46.2 g kg<sup>-1</sup>) were higher than those of peachpalm (39.2 g kg<sup>-1</sup>) and cupuaçu (17.7 g kg<sup>-1</sup>) (means of all analyses, part of them shown in Table 1). After application of <sup>15</sup>N at the end of the dry season, December 1997, the  $\delta^{15}$ N values of cupuaçu were significantly higher than those of pueraria and peachpalm (Figure 2). After the second <sup>15</sup>N application at the peak of the rainy season, May



*Figure 2.*  $\delta^{15}$ N values and relative uptake of  $^{15}$ N applied to soil at the three areas depicted in Figure 1 (CP, PP, PU) by cupuaçu, peachpalm and pueraria; means and standard errors (n=3).

1998, pueraria contained the highest proportions of  $^{15}$ N and cupuaçu the lowest (Figure 2). At the end of the next dry season, November 1998, it was again cupuaçu that showed higher  $\delta^{15}$ N values than the other two species (Table 1 and Figure 2). The relative uptake of  $^{15}$ N by the trees from the areas where they were growing (e.g. uptake by cupuaçu from underneath the cupuaçu; Figure 1) was always higher than from the area of pueraria or the associated tree species. Pueraria, however, took up more  $^{15}$ N from the soil underneath the trees (PP and CP area) than from the alley (PU area) towards the end of the rainy season while showing the same pattern as the trees at the end of the dry season until the beginning of the rainy season (Figure 2).

Cupuaçu took up slightly more (12-26%) of its <sup>15</sup>N from the alley (PU area) than peachpalm (10–18%; Figure 2; only significant in May, at the end of the rainy season; *P*<0.05). The trees took up less than 7% of their total <sup>15</sup>N uptake from the opposite tree row with the respective other tree species (Figure 2 and Table 1). For pueraria and cupuaçu, the <sup>15</sup>N area

| Species     | <sup>15</sup> N application in area | Plant part  | N content<br>[mg g <sup>-1</sup> ] | <sup>15</sup> N<br>[‰] | <sup>15</sup> N content<br>[ $\mu$ g <sup>15</sup> N excess g <sup>-1</sup> ] | <sup>15</sup> N uptake<br>[%] | <sup>15</sup> N area uptake<br>[% <sup>15</sup> N uptake % area] |
|-------------|-------------------------------------|-------------|------------------------------------|------------------------|---|-------------------------------|--|
| Cupuaçu     | СР                                  | leaves      | 18.6                               | 361 a                  | 23.8 a  | 80.8 a                        | 538 bc   |
|             |                                     | branches    | 4.7                                | 235 a                  | 4.0 a   | 77.7 a                        | 518 c  |
|             |                                     | stem        | 3.8                                | 296 a                  | 4.0 b   | 64.2 a                        | 428 b  |
|             | PP                                  | leaves      | 18.5                               | 9 de                   | 0.3 c   | 1.1 c                         | 22 d   |
|             |                                     | branches    | 5.6                                | 23 c                   | 0.4 b   | 5.8 c                         | 117 d  |
|             |                                     | stem        | 3.7                                | 74 b                   | 1.0 c   | 14.0 b                        | 281 b  |
|             | PU                                  | leaves      | 18.0                               | 81 cd                  | 5.0 ab  | 18.2 b                        | 1331 b   |
|             |                                     | branches    | 4.1                                | 55 b                   | 0.9 c   | 18.4 b                        | 1205 ab  |
|             |                                     | stem        | 4.3                                | 99 b                   | 1.7 bc  | 21.8 b                        | 1599 a   |
| Dessburghes | CD                                  | 1           | 25.6                               | 0 -                    | 0.4 -   | 1.6 -                         | 11.1   |
| Peachpalm   | CP                                  | leaflets    | 35.6                               | 8 e                    | 0.4 c   | 1.6 c                         | 11 d   |
|             |                                     | rachis      | 5.8                                | 5 d                    | 0.0 c   | 1.7 c                         | lld  |
|             |                                     | stem        | 13.5                               | 8 c                    | 0.1 c   | 2.6 c                         | 18 c   |
|             | PP                                  | leaflets    | 32.0                               | 196 b                  | 21.9 a  | 85.0 a                        | 1700 b   |
|             |                                     | rachis      | 6.3                                | 121 a                  | 2.7 a   | 83.4 a                        | 1668 a   |
|             |                                     | stem        | 22.4                               | 107 b                  | 8.1 a   | 76.4 a                        | 1527 a   |
|             | PU                                  | leaflets    | 33.3                               | 36 d                   | 4.2 b   | 13.4 b                        | 982 bc   |
|             |                                     | rachis      | 5.3                                | 23 bc                  | 0.3 b   | 14.9 b                        | 1093 bc  |
|             |                                     | stem        | 18.3                               | 32 c                   | 1.6 b   | 21.0 b                        | 1543 a   |
| Dueraria    | CP                                  | whole plant | 40.3                               | 11 e                   | 0.9.0   | 320                           | 21 d   |
| i ucidilă   |                                     | whole plant | 40.5                               | 11 C                   | 0.70  | J.2 C                         | 424 a  |
|             | rr                                  | whole plant | 39.1                               | 152                    | 0.0 U   | 21.00                         | 434 C  |
|             | ΡU                                  | whole plant | 39.6                               | 155 C                  | 21.5 a  | /3.1 a                        | 5508 a   |

*Table 1.* Biomass N content, N-isotope composition and proportion of <sup>15</sup>N uptake from different areas in agroforestry with cupuaçu, peachpalm and pueraria at the end of the dry season in November 1998, 6 months after application of <sup>15</sup>N at the beginning of May 1998; values in one column followed by the same letter are not significantly different at P < 0.05 (comparison only within same plant parts; n=3)

uptake, calculated as the product of the <sup>15</sup>N uptake and the area in percent indicating the relative importance of different areas for N uptake between species, was highest under pueraria (PU; Table 1). For peachpalm, however, the values were slightly higher under the peachpalm itself.

#### **Biomass production**

Cupuaçu had a higher biomass per tree than peachpalm at time of harvest with 26.4 and 1.8 kg tree<sup>-1</sup>, respectively (Table 2; P < 0.001). Most of the biomass of cupuaçu consisted of leaves (43%), followed by branches (35%) and stem (22%; P < 0.05). Biomass of the leaflets of peachpalm equaled that of the rachis with 46 and 54%, respectively (Table 2). Aboveground biomass per unit area increased in the order peachpalm (1.4) < cupuaçu (4.4) < pueraria (8.8 Mg ha<sup>-1</sup>) being 14.6 Mg ha<sup>-1</sup> for the whole cropping system. The biomass production was not affected by the place of <sup>15</sup>N application (P > 0.05).

## Total <sup>15</sup>N uptake

The <sup>15</sup>N uptake of a single cupuaçu tree was more than one order of magnitude higher than that of peachpalm (Table 2). Most of the <sup>15</sup>N uptake was located in the leaves, only 20% in the stem and branches of cupuaçu. Ninety percent of the <sup>15</sup>N uptake of peachpalm was found in the leaflets, only 10% in the rachis. On an area basis, pueraria had a higher uptake of applied <sup>15</sup>N than peachpalm (P<0.05), but lower than cupuaçu (not significant). The <sup>15</sup>N uptake by pueraria was higher between the tree rows and lowest underneath cupuaçu. Cupuaçu took up significantly more <sup>15</sup>N from the area covered by pueraria than peachpalm (Table 2; P<0.05).

The total recovery of applied <sup>15</sup>N by the vegetation was 29% (Table 3) and followed the same pattern among plants as the <sup>15</sup>N uptake (Table 2): cupuaçu had significantly higher recovery of <sup>15</sup>N from the alley covered by pueraria (2.2%) than peachpalm (0.45%; P < 0.05). Pueraria had a non-significantly lower re-

| Species   | <sup>15</sup> N appl.<br>in area | Plant part               | Biomass [kg tree <sup>-1</sup> ] | Biomass<br>[Mg ha <sup>-1</sup> ] | <sup>15</sup> N uptake<br>[mg <sup>15</sup> N excess tree <sup>-1</sup> ] | <sup>15</sup> N uptake<br>[g <sup>15</sup> N excess ha <sup>-1</sup> ] |
|-----------|----------------------------------|--------------------------|----------------------------------|-----------------------------------|---|--|
| Cupuaçu   | СР                               | leaves                   | $13.3 \pm 2.8$                   | $2.22\pm0.47$                     | 305.5 ± 51.1  | $203.7 \pm 34.0$   |
|           |                                  | branches                 | $11.5\pm4.0$                     | $1.92\pm0.67$                     | $47.1 \pm 17.2$   | $31.4 \pm 11.5$  |
|           |                                  | stem                     | $7.2 \pm 1.1$                    | $1.20\pm0.18$                     | $30.0\pm~8.9$   | $20.0 \pm 5.9$   |
|           |                                  | whole plant              | $32.0\pm7.5$                     | $5.33 \pm 1.24$                   | $382.6\pm72.7$  | $255.0\pm48.5$   |
|           | PP                               | leaves                   | $10.1\pm2.6$                     | $1.69\pm0.43$                     | $3.3 \pm 1.8$   | $2.2\pm~1.2$   |
|           |                                  | branches                 | $7.5\pm1.7$                      | $1.25\pm0.28$                     | $2.4\pm~0.8$  | $1.6\pm~0.6$   |
|           |                                  | stem                     | $4.5\pm0.8$                      | $0.76\pm0.13$                     | $3.9 \pm 1.4$   | $2.6\pm~0.9$   |
|           |                                  | whole plant              | $22.2\pm4.8$                     | $3.70\pm0.81$                     | $9.6 \pm 1.7$   | $6.4 \pm 1.1$  |
|           | PU                               | leaves                   | $10.5\pm3.7$                     | $1.75\pm0.62$                     | $50.3 \pm 14.3$   | $33.5\pm9.5$   |
|           |                                  | branches                 | $8.6\pm3.0$                      | $1.43\pm0.49$                     | $8.1\pm~5.1$  | $5.4\pm~3.4$   |
|           |                                  | stem                     | $5.9\pm1.7$                      | $0.99\pm0.28$                     | $8.4\pm~2.7$  | $5.6 \pm 1.8$  |
|           |                                  | whole plant              | $25.0\pm8.3$                     | $4.16\pm1.38$                     | $66.7 \pm 19.3$   | $44.5 \pm 13.2$  |
| Peachpalm | СР                               | leaflets                 | $0.9 \pm 0.1$                    | $0.45\pm0.04$                     | $0.4 \pm 0.0$   | $0.5\pm~0.1$   |
|           |                                  | rachis                   | $1.1\pm0.1$                      | $0.54\pm0.05$                     | $0.0\pm~0.0$  | $0.1\pm~0.0$   |
|           |                                  | whole plant <sup>a</sup> | $1.9\pm0.3$                      | $0.93\pm0.17$                     | $0.4\pm~0.0$  | $0.6\pm~0.1$   |
|           | PP                               | leaflets                 | $1.3 \pm 0.4$                    | $0.63\pm0.19$                     | $31.1\pm17.5$   | $41.5\pm23.3$  |
|           |                                  | rachis                   | $1.5\pm0.4$                      | $0.75\pm0.22$                     | $4.5\pm2.5$   | $6.0 \pm 3.4$  |
|           |                                  | whole plant <sup>a</sup> | $1.4 \pm 0.3$                    | $0.69\pm0.12$                     | $18.7 \pm 9.1$  | $25.0\pm12.2$  |
|           | PU                               | leaflets                 | $1.6\pm0.3$                      | $0.79\pm0.13$                     | $6.1 \pm 4.0$   | $8.1\pm$ 5.3   |
|           |                                  | rachis                   | $1.9\pm0.3$                      | $0.94\pm0.16$                     | $0.7\pm~0.3$  | $0.9\pm~0.4$   |
|           |                                  | whole plant <sup>a</sup> | $2.1\pm0.3$                      | $1.06\pm0.12$                     | $3.8\pm~1.8$  | $5.1 \pm 2.4$  |
| Pueraria  | СР                               | whole plant              | _                                | $8.77\pm0.13$                     | _   | 8.1 ± 3.0  |
|           | PP                               | whole plant              | _                                | $8.77\pm0.13$                     | -   | $17.9 \pm 4.1$   |
|           | PU                               | whole plant              | -                                | $8.77\pm0.13$                     | -   | $186.9 \pm 14.8$   |

*Table 2.* Above-ground biomass and <sup>15</sup>N uptake from different areas in agroforestry with cupuaçu, peachpalm and pueraria at the end of the dry season in November 1998, 6 months after application of <sup>15</sup>N at the beginning of May 1998; means and standard errors (n=3)

<sup>a</sup> <sup>15</sup>N uptake computed by assuming equal biomass of rachis and leaflets.

covery under peachpalm (PP area) as peachpalm itself with 0.9 and 2.4%, respectively (Table 3). Summing up the uptake of all three plant species, 4% of the applied <sup>15</sup>N was recovered from the area under peachpalm, 13% from under cupuaçu and 12% from the area between the trees under pueraria. Only 3% of the applied <sup>15</sup>N was taken up by peachpalm, but 15% by cupuaçu and 11% by pueraria (Table 3). Thus, more than half of the <sup>15</sup>N taken up by all plants together was recovered by cupuaçu (53%), followed by pueraria (37%) and peachpalm (10%).

## Soil <sup>15</sup>N contents

The organic C content decreased with depth (surface soil 29 mg  $g^{-1}$ , subsoil 7 mg  $g^{-1}$ ), but similar values were found on the three areas in the cropping sys-

tem (Figure 3). The soil N contents were significantly higher in the surface soil under pueraria than under the trees (P < 0.05). The soil <sup>15</sup>N enrichment was significantly lower where pueraria was growing than under the tree rows up to 0.5 m depth. At 0.3 to 0.5 m depth, cupuaçu showed higher  $\delta^{15}$ N values than peachpalm. The recovery of applied <sup>15</sup>N in the soil from 0 to 0.3 m was significantly higher in the alley under pueraria than under the trees. In the subsoil, a lower proportion of applied <sup>15</sup>N was found under pueraria than under the two trees (not significant). The total <sup>15</sup>N recovery in the soil (0-1 m) increased from cupuaçu (6%) over peachpalm (17%) to pueraria (19%; Table 3). In plant and soil together, most (31%) of the <sup>15</sup>N was recovered where pueraria was growing (PU) in comparison to cupuaçu (20%) and peachpalm (20%) resulting in a

*Table 3.* Recovery of <sup>15</sup>N applied to different areas in mixed cropping of cupuaçu, peachpalm and pueraria at the end of the dry season in November 1998, 6 months after application of <sup>15</sup>N at the beginning of May 1998; values in one column or row followed by the same small or capital letter, respectively, are not significantly different at P<0.05; means and standard errors (n=3)

| Compartment   | Cupuaçu<br>CP  | Recovery [%] of <sup>15</sup> N<br>peachpalm<br>PP | applied to<br>pueraria<br>PU                                       | Total area<br>CP+PP+PU |
|---------------|--|--|--|------------------------|
| Cupuaçu       | $12.75 a \pm 2.42$   | $0.32~b\pm0.06$                                    | $2.22~b\pm0.66$  | $15.30 a \pm 2.89$     |
|               | А  | С  | В  |                        |
| Peachpalm     | $\begin{array}{c} 0.03 \text{ c} \pm 0.00 \\ \text{C} \end{array}$ | 2.37 a±1.33<br>A                                   | $\begin{array}{c} 0.45 \text{ c} \pm 0.25 \\ \text{B} \end{array}$ | $2.85 b \pm 1.59$      |
| Pueraria      | $\begin{array}{c} 0.40 \text{ b} \pm 0.16 \\ \text{B} \end{array}$ | 0.90 a±0.21<br>B                                   | 9.34 a ± 0.74<br>A   | $10.84 a \pm 0.84$     |
| Sum of plants | $13.18 \pm 2.36$   | $3.59 \pm 1.56$                                    | $12.02\pm1.23$   | $28.79 \pm 4.26$       |
|               | А  | В  | А  |                        |
| Soil          | $6.43\pm0.84$  | $16.99 \pm 3.44$                                   | $18.97 \pm 3.49$   | $42.38 \pm 7.32$       |
| (0–1 m)       | В  | А  | А  |                        |
| Total sum     | $19.61\pm3.19$   | $20.58 \pm 4.99$                                   | $30.99 \pm 4.02$   | $71.18 \pm 10.98$      |
|               | В  | В  | А  |                        |

total recovery in the whole cropping system of 71% (Table 3).

## Foliar natural <sup>15</sup>N abundance

The natural <sup>15</sup>N abundance of pueraria leaves was significantly lower than of cupuaçu and peachpalm (Table 4; P < 0.05). The difference was larger at the end of the wet season than at the end of the dry season. The  $\delta^{15}$ N values of the two tree species were not significantly lower in the agroforestry system with a higher abundance of pueraria (P1) than with low pueraria abundance (P2; P > 0.05). Similarly, the foliar N contents were not significantly affected by the amount of pueraria cover present (Table 4).

#### Discussion

## Dynamics of N uptake

The <sup>15</sup>N uptake suggested that cupuaçu took up a higher proportion of its N at the end of the dry season than peachpalm, indicating that it had a more active root system at that time. One reason for that may be the large amount of surface roots of cupuaçu (Haag,

1997), which can immediately take up applied nutrients from the soil surface. Another explanation is the harvest of a large part of the peachpalm shoot biomass at the end of the dry season. The removal of the aboveground biomass of peachpalm may have induced root die-back, as also shown for a tree legume in a tropical agroforestry system (Lehmann et al., 1998). In the rainy season, peachpalm was growing again and taking up more of its N from the applied N source than during the dry season. Therefore, peachpalm should not be fertilized at the beginning of the rainy season, but later when the N demand is higher due to a higher biomass production. In contrast, relative <sup>15</sup>N uptake of cupuaçu did not vary throughout the year.

Also, pueraria seemed to take up less of its N from the applied N source during the dry season than during the rainy season. This coincides with field observations from the same site and year which showed a reduced biomass production of pueraria during the dry season in comparison to the rainy season by about 50% (Uguen, in Lehmann et al., 2000). Whereas the N uptake of pueraria was more restricted to the alley (PU area; Figure 1) during the dry season, it could use soil N from underneath the tree canopies during the rainy season when water competition was low. In contrast

| Species   | System | End of dry seas   | son           | End of wet season     |                |  |
|-----------|--------|-------------------|---------------|-----------------------|----------------|--|
|           |        | N $\delta^{15}$ N |               | Ν                     | $\delta^{15}N$ |  |
|           |        | $[mg g^{-1}]$     | [‰]           | [mg g <sup>-1</sup> ] | [‰]            |  |
| Cupuaçu   | P1     | $17.74\pm0.52$    | $4.06\pm0.56$ | $14.82\pm0.61$        | $4.08\pm0.33$  |  |
| Cupuaçu   | P2     | $17.35\pm0.74$    | $4.64\pm0.53$ | $14.10\pm0.95$        | $4.70\pm0.81$  |  |
| Peachpalm | P1     | $28.91 \pm 0.46$  | $4.90\pm0.48$ | $37.34 \pm 4.87$      | $5.02\pm0.59$  |  |
| Peachpalm | P2     | $31.80 \pm 1.61$  | $5.19\pm0.20$ | $34.85 \pm 2.47$      | $6.83\pm0.18$  |  |
| Pueraria  | P1     | $43.52\pm0.65$    | $2.16\pm0.08$ | $44.26 \pm 2.35$      | $1.72\pm0.75$  |  |
| Pueraria  | P2     | $38.71\pm0.64$    | $2.75\pm0.33$ | $37.13 \pm 2.02$      | $0.97\pm0.35$  |  |

*Table 4.* Foliar N contents and natural <sup>15</sup>N abundance of cupuaçu, peachpalm and pueraria in an agroforestry system with high (P1) and low (P2) abundance of pueraria; means and standard errors (n=3)



*Figure 3.* Soil carbon (a) and nitrogen contents (b), <sup>15</sup>N enrichment (c) and recovery (d) under cupuaçu, peachpalm and pueraria five months after <sup>15</sup>N application in November 1997; only sites where <sup>15</sup>N was applied; asterics denote significant effects at P < 0.05 \* and 0.01 \*\* (ANOVA); means and standard errors (n=3).

to pueraria, the trees did not show a higher N uptake further away from the stem in the rainy season than the dry season. In a more seasonal climate with a distinct dry season, however, coffee showed pronounced dynamics of lateral root activity in an experiment in Kenya (Huxley et al., 1974).

## Distribution of N uptake

In contrast to our study, Theobroma cacao had the highest root activity at 1.2-1.6 m and the lowest at 0.9 m distance from the stem in Ghana (Ahenkorah, 1975). The high proportion of N uptake of cupuaçu from the area under the canopy may be explained by the current fertilization practice: The mineral fertilizer was always applied around the stem, resulting in a low stimulation for the trees to grow roots further away from the area under the canopy. Furthermore, the amount of applied fertilizer was found to be sufficient for the two tree crops, since the foliar nutrient contents did not increase with higher fertilizer applications (Assis and Schroth, unpubl. data). A third reason for the low utilization of legume N by the trees may be a high soil N mineralization of more than 150 kg N  $ha^{-1}$  $yr^{-1}$  in the topsoil (0–0.1 m) under the tree canopies found in the same experiment during the preceding year (Schroth et al., unpubl. data).

The low lateral root activity of peachpalm in the alley was also an effect of shoot pruning as stated above. Additionally, the peachpalm biomass production and hence N turnover per tree were estimated to be lower than those of cupuaçu. Using our biomass determinations and the litterfall at the same site and year found by Uguen (unpubl. data) of 2.7 and 0.6 kg tree<sup>-1</sup> yr<sup>-1</sup> for cupuaçu and peachpalm, respectively, biomass production of peachpalm was 48% lower than that of cupuaçu. As a result, the rooting area (defined as the lateral zone where N was taken up) of peachpalm was more restricted to the area under the canopy than that of cupuaçu. The <sup>15</sup>N area uptake also indicated that on a landuse system level the area underneath the pueraria is more important for cupuaçu than for

peachpalm, since the values were significantly higher in the PU area than the CP area for cupuaçu, whereas it was the other way round for peachpalm.

When calculating the total <sup>15</sup>N uptake per unit area and the recovery of the applied <sup>15</sup>N, the lower N uptake of peachpalm than cupuaçu from the alley becomes even more evident: Peachpalm took up less than 10% of the amount of  $^{15}$ N taken up by cupuaçu. Cupuaçu also had a 40% higher <sup>15</sup>N accumulation than pueraria. However, the total <sup>15</sup>N uptake of pueraria over time was presumably much higher, because N turnover by pueraria was found to be very high. Using the litterfall data cited above, the N turnover of peachpalm calculated to 90 kg  $ha^{-1}$  yr<sup>-1</sup> and of cupuaçu to 21 kg ha $^{-1}$  yr $^{-1}$ . In contrast, N turnover of pueraria was as high as 212 kg  $ha^{-1}$  yr<sup>-1</sup> at a nearby site with an 81% lower above-ground biomass (Uguen, in Lehmann et al., 2000), suggesting that the production may be even higher in the presented experiment. The large N turnover of pueraria may also explain the high <sup>15</sup>N recovery in the topsoil under pueraria. The significantly higher total soil and plant recovery of <sup>15</sup>N by pueraria than the tree crops confirmed this observation.

Only 71% of the applied <sup>15</sup>N was recovered in plant and soil of the whole cropping system. Due to the low <sup>15</sup>N contents in the subsoil, it can be assumed that a large part of the <sup>15</sup>N not accounted for (29%) was lost by denitrification. Similarly, Alfaia (1997) reported a gaseous loss of almost 30% of <sup>15</sup>N labeled ammonium sulfate applied to a central Amazonian Ferralsol in a pot experiment. The resulting N<sub>2</sub>O evolution possesses important climatic relevance as a greenhouse gas.

## Nitrogen competition

Competition for N between the two tree species was very low, since far less than 1% of the total N applied (6% of its N uptake) to one tree species was retrieved by the other species. The use of soil N from the alley (PU area) by the trees can be considered as low generally being below 20% of the tree N uptake. The low N use from the alley (PU) by the trees was most likely an effect of the amount and placement of N fertilizer as discussed above. This is in contrast to many leguminous trees, which have been tested in mixed cropping systems and shown to compete strongly with associated plants for water and nutrients (Rao et al., 1997). Harvesting the heart of palms by pruning certainly reduced the competitiveness of the peachpalm as it was shown for a woody legume in a tropical agroforestry system (Peter and Lehmann, 2000). In our study with intercropped trees and a legume cover crop, more competition would be advantageous, since it would increase the utilization of soil N underneath the legume which may contain biologically fixed N (see below) without reducing tree crop production due to competition by neighboring trees.

In contrast, pueraria substantially competed for N with the tree crops. Approximately the same fraction of N fertilizer applied to the area under peachpalm (PP) was retrieved by pueraria and by peachpalm. However, this N uptake did not seem to be a sink for N but the N was rapidly recycled as seen from the large N turnover by pueraria and <sup>15</sup>N recovery at the topsoil under pueraria.

## Tree uptake of biologically fixed N

Biological fixation of atmospheric N2 was estimated between 9 and 37% of the pueraria N in the dry season and 23 and 45% in the wet season as determined by the <sup>15</sup>N enrichment method for the presented experiment (Silva Jr. and Lehmann, in Lehmann et al., 2000). The differences of the tree <sup>15</sup>N contents between P1 and P2, however, indicated that only a small fraction of the tree N was derived from sources associated with a higher pueraria abundance. During biological N fixation, the fraction of  ${}^{15}N$  (shown as  $\delta^{15}N$  values) decreases, because atmospheric N2 possesses lower <sup>15</sup>N-<sup>14</sup>N ratios than soil N. The N sources of plants growing in association with legumes may similarly be estimated by their <sup>15</sup>N-<sup>14</sup>N ratios (Chalk and Smith, 1994). In our experiment, the N nutrition of the trees did not increase with higher legume abundance, indicating that biologically fixed N did not significantly contribute to tree N uptake. It is not likely that the tree N nutrition was already benefiting from the low pueraria abundance. The main reason for the little utilization of legume N by the trees was their low N uptake from the area underneath the pueraria being below 20% of their total N uptake, due to the amount and placement of the fertilizer as discussed in an earlier section. A higher root activity in the alley (PU area) and, therefore, uptake of biologically fixed N may be achieved by a lower fertilizer application and a placement further away from the stem. Wahid et al. (1993) concluded for Cocos nucifera that the lateral root activity distribution can be manipulated by the place of fertilizer application. In future studies, low or

no N applications should be tested for tree crops with legume covers on central Amazonian Ferralsols.

## Conclusions

Peachpalm managed for heart of palm production and cupuaçu proved to be tree species well suited for mixed cropping systems, since both cause low competition with neighboring plants. At the same time, however, the trees did not efficiently utilize the soil N from the area between the tree crops and, therefore, the N fixed by the intercropped legume cover crop. This may have been an effect of the amount and placement of the mineral fertilizer. Due to the regular shoot pruning and harvesting, peachpalm had a smaller rooting area than cupuaçu. The low N uptake by peachpalm at the onset of the rains in contrast to cupuaçu also suggested that peachpalm should not be fertilized at the beginning of the rainy season. Management techniques should be developed which increase the utilization of biologically fixed N by cupuaçu and peachpalm, e.g. applying less fertilizer, broadcasting fertilizer to stimulate tree root growth underneath the legume or enhancing the growth of the pueraria underneath the trees. Since pueraria is a vine, this may cause damage to the trees. Therefore, pueraria has to be regularly cut, but the biomass should be left to decompose in the area where the trees take up most of their N. More information is needed about the effects of different legume cover crops in fruit tree production systems.

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