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Nitrogen recovery by alley-cropped maize and trees from ^{15}N-labeled tree biomass in the subhumid highlands of Kenya

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Abstract The effectiveness of tree-leaf biomass as a source of N to crops in agroforestry systems depends on the rate at which crops can obtain N from the biomass. A study was conducted to determine the fate of ^{15}N labeled, soil-applied biomass of two hedgerow species, *Calliandra calothyrsus* Meissner (calliandra) and *Leucaena leucocephala* (Lam.) de Wit (leucaena), in the subhumid highlands of Kenya. Labeled biomass obtained from ^{15}N fertilized trees was applied to microplots in an alley cropping field and maize planted. N uptake and recovery by maize and hedgerow trees was periodically determined over a 20-week period during the short rain (1995) and the long rain (1996) growing seasons. In maize crop from treatments that received leucaena biomass, higher N uptake and recovery were recorded than in maize from the plots that received calliandra biomass. However, N uptake and recovery were higher in calliandra tree hedges than in leucaena hedges, indicating differences in N uptake by the two tree species. The largest fraction (55–69%) of N in the applied tree biomass was left in the soil N pool, 8–13% recovered by maize, 2–3% by tree hedges, and 20–30% could not be accounted for. Some of the unaccounted for N may have been left in the wood and root portions of the tree hedges and in the bulk soil below the 20-cm depth. The study shows that only a small fraction of the N contained in the N-rich biomass that is applied to the soil is taken up by the current season's crop, suggesting that a major benefit may be in the build-up of the soil N store.

Key words Agroforestry · *Calliandra* · *Leucaena* · Nitrogen uptake · Soil nitrogen pool

Introduction

Incorporation of tree-leaf biomass (prunings) into the soil is a well-known agricultural practice for enhancing soil fertility and sustaining crop production (Fu et al. 1987). The practice is more prevalent in the tropics, where use of inorganic fertilizers by small holder farmers is limited (Kang 1987; FAO 1989). Though application of tree biomass to the soil has been shown to improve soil chemical and physical properties and to sustain and increase crop yield (Kang et al. 1990), N contribution from the biomass to the associated crop has been low, representing a N recovery rate of less than 20% in most cases (Mulongoy and Meersh 1988; Xu et al. 1993; Giller and Cadisch 1995; Palm 1995). The low N recovery rate from tree biomass is probably due to lack of synchronization between N demand by the crop and that released by the biomass, or N losses through volatilization, immobilization and leaching (Myers et al. 1994; Mugendi et al. 1999b). It is also possible that decomposition of biomass may lead to retention of N in soil organic forms that are resistant to rapid mineralization (Haggar et al. 1993). Few studies have quantified the fate of soil-applied tree biomass N, especially the transfer of N from biomass to crops and tree hedges.

The objective of this study was to trace the path of N in ^{15}N-labeled biomass of *Calliandra calothyrsus* Meissner (calliandra) and *Leucaena leucocephala* (Lam.) de Wit (leucaena) when the biomass was soil-incorporated into a crop field in an alley cropping situation.

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Materials and methods

The study was conducted at the Embu Regional Research Centre, Eastern Province, Kenya. The centre is located in the central highlands of Kenya on the south eastern slopes of Mt. Kenya at 0°30′S, 37°30′E and an altitude of 1480 m. The soils are Typic Palehumults (Humic Nitisols according to FAO-UNESCO) derived from basic volcanic rocks. They are deep, well weathered with friable clay texture with moderate to high inherent fertility. Total annual average rainfall is approximately 1200–1500 mm received in two distinct rainy seasons: the long rains (LR) from mid-March to June with historical average precipitation of 650 mm and the short rains (SR) from mid-October to December with an average of 450 mm. The average monthly maximum temperature is 25 °C and the minimum 14 °C. The long-term monthly average is 19.5 °C.

Labeling of calliandra and leucaena prunings with ^{15}N

Calliandra and *Leucaena* trees that had been planted in 1992 in the external boundaries surrounding the experimental site were labeled in March 1995 using 10 atom% ^{15}N excess double-labeled fertilizer in the form of ammonium nitrate (Isotope Services, USA). One hundred plants of each tree species were selected, lopped to a height of 50 cm, and pruned clean of all the leaves. Approximately 1 g of the enriched fertilizer was dissolved in 1 l of water before being applied around the base of each tree using a watering can. Subsequent watering was done to move the fertilizer into the soil. The trees were supplied with the labeled fertilizer in a similar manner at 4-week intervals during 4 consecutive months. In early August, 1 month after the last application, the trees were pruned back to 50 cm height (initial biomass). The harvested prunings were sun dried to constant weight. The leafy biomass was separated from the small branches and twigs, and then stored in gunny bags for use in the short rain season of 1995 (SR 95).

After the initial pruning in August, the trees were allowed to sprout and grow for another 5 months (no ^{15}N fertilizer was added), after which they were pruned and the resulting biomass treated as above (second-generation biomass). This biomass was used in the long rain season of 1996 (LR 96).

Random subsamples of the dried leaf biomass from both species were recovered for determination of the level of ^{15}N enrichment in each season. The resulting atom% ^{15}N values were 0.6085 (calliandra) and 0.7280 (leucaena) for SR 95 and 0.5357 (calliandra) and 0.5228 (leucaena) during LR 96, respectively. Reference samples were also harvested from calliandra and leucaena trees that had not been labeled.

Experimental treatments

The experimental treatments for the ^{15}N study were demarcated on an existing randomized block experiment composed of ten treatments in which the test crop, maize, was grown alone or alley cropped with or without fertilizer/prunings application (Mugendi et al. 1999a). Treatments 1 and 2 (calliandra and leucaena alley cropped with maize, prunings applied) were selected for this study because we wanted to monitor nutrient uptake by both the maize and tree hedges. Microplots measuring 2 m × 9 m were demarcated in the middle of the existing plots that measured 9 m × 10 m. The microplots were positioned perpendicular to the two hedges. Ten trees (five on either side of the hedge) were included in the microplots for this purpose. The ^{15}N-labeled calliandra and leucaena biomass was soil-incorporated in these microplots using hand hoes at an equal rate of 1 kg m^{-2} before maize was planted in the two seasons of experimentation (initial biomass – SR 95, and second-generation biomass – LR 96). Unlabeled prunings from the trees in the microplots were not added to the microplots.

Sampling and sample handling

Maize plants were sampled at 4, 7, 10, 15, and 20 weeks after planting (WAP) maize, the first sampling coinciding with the thinning stage and the last with crop harvest. At 4 WAP, all the thinnings obtained from the microplots were sampled; however, at 7, 10, and 15 WAP, three maize plants were sampled from each microplot respectively, and at harvest, ten maize plants were sampled. Young leaves from calliandra and leucaena tree hedges in the microplots were also sampled during the same intervals. Except at seedling stage (4 WAP), maize leaves were always separated from stems and analyzed separately. At harvest (20 WAP), the cob was separated from the grain and stover. Maize roots and soil (top 20 cm) were also sampled at that stage. At every sampling date, reference samples (from unlabeled trees and maize plants) were taken alongside those from labeled plots. Samples were cleaned with distilled/deionized water and oven dried at 65 °C to constant weight.

All the samples were ground through a 0.5-mm sieve mill. The resulting powder was thoroughly mixed, packed in polyethylene bags and stored under dry conditions and shipped to the University of Florida (USA) for analysis.

Analysis of ^{15}N

Samples were analyzed for ^{15}N using a Carbon Nitrogen Analyzer 1500 (Carlo Erba) coupled with an isotope ratio mass spectrometer (VG 602 E). Recovery of the applied N was calculated following Westerman et al. (1972):

$$\%\ \mathrm{N\ recovery} = 100\,P(C-B)/F(A-B) \qquad (1)$$

where P = total N uptake in plant (maize or tree) or total N in soil, F = total N applied via labeled leafy-biomass (calliandra or leucaena), A = atom% ^{15}N abundance in the applied labeled biomass (calliandra or leucaena), B = atom% ^{15}N abundance in unlabeled control sample (maize, tree, or soil), C = atom% ^{15}N abundance in the sample after application of labeled biomass (maize, tree, or soil). Calculation of N recovery after the second-generation biomass was applied (LR 96) included the amount of N that had been left in the soil at the end of the SR 95 season (initial application).

Statistical analysis

Data were subjected to analysis of variance (ANOVA) using SAS (1988). Means were separated by LSD procedure and declared different at $P < 0.05$ level of significance.

Results

Uptake and recovery of N by maize and tree hedges during different stages of crop growth

Total N uptake by maize increased progressively as the two seasons advanced [from about 3 kg N ha^{-1} (LR 96) at 4 WAP to about 114 kg N ha^{-1} (SR 95) at 15 WAP] (Table 1). The most rapid uptake occurred during the 4–10 WAP growth period. Maize in the plots that were treated with leucaena biomass significantly took up more total N from the soil compared to that where calliandra biomass was applied (except at 4 WAP for both seasons, and at 7 WAP for SR 95). On the other hand, calliandra tree hedges had a significantly higher N uptake compared to leucaena tree hedges at 7, 10, and 15 WAP sampling dates during the two growing sea-

Table 1 Nitrogen uptake and recovery by maize and tree hedges 7, 10, and 15 weeks after planting maize during two cropping seasons at Embu, Kenya. *WAP* weeks after planting (maize), *Trt* treatments, *SR* short rain, *LR* long rain, *CC Calliandra calothyrsus, LL Leucaena leucocephala, Mz* maize, *lvs* leaves, *stm* stems

Total N uptake (kg ha^{-1})					Recovery of N (%)			
Season	WAP	Trt	Maize	Trees	Mz (lvs)	Mz (stm)	Total (Mz)	Trees (lvs)
SR 95	4	CC	4.6 a	26.7 a	0.7 a	–	0.7 a	0.4 a
		LL	5.1 a	25.0 a	0.8 a	–	0.8 a	0.3 a
	7	CC	44.9 a	56.3 a	5.2 b	0.9 b	6.1 b	1.5 a
		LL	45.7 a	51.2 b	7.4 a	3.3 a	10.7 a	1.3 a
	10	CC	75.4 b	78.2 a	5.7 b	1.8 b	7.5 b	1.8 a
		LL	108.2 a	59.4 b	8.3 a	3.5 a	11.8 a	1.5 a
	15	CC	78.0 b	87.9 a	6.1 b	2.4 b	8.5 b	2.2 a
		LL	113.5 a	64.3 b	8.9 a	3.7 a	12.6 a	1.7 b
LR 96	4	CC	3.2 a	24.9 a	0.5 a	–	0.5 a	0.8 a
		LL	3.0 a	22.9 a	0.6 a	–	0.6 a	0.5 b
	7	CC	48.3 b	51.2 a	3.7 a	1.1 a	4.8 a	1.6 a
		LL	58.9 a	46.7 b	4.3 a	2.1 a	6.4 a	1.5 a
	10	CC	65.1 b	67.1 a	5.3 a	1.9 a	7.2 a	2.1 a
		LL	87.5 a	57.3 b	6.1 a	2.4 a	8.5 a	1.9 a
	15	CC	72.2 b	87.9 a	5.5 b	2.1 a	7.6 b	2.6 a
		LL	98.7 a	73.2 b	7.3 a	2.8 a	10.1 a	2.0 b

Means followed by the same letter within a column at a particular sampling time are not significantly different at *P*

sons (Table 1). As in the case with maize, N uptake by trees increased as the seasons progressed, though the trees' uptake was much higher at the beginning of each season (4 WAP) compared to maize (e.g., 25 kg N ha^{-1} for trees compared to 5 kg N ha^{-1} for maize during SR 95).

Maize in the treatments that received leucaena biomass recovered more N than those in the treatments that received calliandra biomass. However, only the 7, 10, and 15 (SR 95) and 15 (LR 96) WAP recovery values were significantly different from each other (Table 1). Total recovery of N increased from 0.5% (maize; LR 96) and 0.3% (trees; SR 95) at 4 WAP to the highest level of 12.6% (maize; SR 95) and 2.6% (trees; LR 96) at 15 WAP in the two seasons. Recovery values for tree hedges were generally low (compared to maize). Though calliandra tree hedges recovered more N compared to leucaena hedges, the differences were only significant at 15 WAP (SR 95) and at 4 and 15 WAP (LR 96) sampling dates (Table 1).

N uptake and recovery by maize and tree hedges at maize harvest

The maize crop in the treatments where leucaena biomass was applied took up significantly higher amounts of N from the soil compared to that where calliandra biomass was applied in both seasons (116 kg and 113 kg ha^{-1} for leucaena and 79 kg and 76 kg ha^{-1} for calliandra). However, an opposite scenario was observed with the trees where calliandra tree hedges had a significantly higher N uptake than leucaena tree hedges (99 kg and 104 kg ha^{-1} for calliandra vs 73 kg and 75 kg ha^{-1} for leucaena) (Table 2).

Recovery of N in different parts of the maize crop varied, the grain accounting for the highest, followed by the stover (Table 2). Total N recovered by maize was significantly higher in the treatments that received leucaena biomass compared to those that received calliandra biomass in both seasons (13.0% and 10.8% for leucaena-biomass vs 9.3% and 8.2% for calliandra-biomass). However, calliandra tree hedges recovered significantly higher amounts of N compared to leucaena hedges though the recoveries were low (2.7% and 3.2% for calliandra and 2.1% and 2.5% for leucaena for the two seasons respectively) (Table 2).

A major part of the N applied to the soil through the biomass was left in the soil pool at the end of the two growing seasons: 55% and 61% for leucaena and 69% and 67% for calliandra for SR 95 and LR 96; and, approximately 20–22% and 25–30% could not be accounted for in the case of calliandra- and leucaena-biomass-applied treatments, respectively (Table 2).

Discussion

Treatments that received leucaena biomass produced maize that consistently showed higher total N uptake and higher N recovery than those treatments that received calliandra biomass especially during the SR 95 growing season. The reason for this could be that leucaena biomass applied initially (SR 95) had a significantly higher % content of both total N (4.0%) and ^{15}N (0.7280%) compared to calliandra biomass which had 3.7% and 0.6085% respectively. Secondly, leucaena biomass decomposed and released N faster than calliandra biomass (Mugendi et al. 1998b); hence, the released N was readily available for uptake. Thirdly, calliandra tree hedges were more competitive compared to leucaena hedges and depressed maize yields (Mugendi et al. 1999a), explaining why total N uptake (by maize) was lower in the calliandra alley-cropped treatments.

Table 2 Nitrogen uptake and recovery by maize and tree hedges at maize harvest during two cropping seasons at Embu, Kenya

	Total N uptake (kg ha^{-1})			
Season	SR 95		LR 96	
Treatment	CC	LL	CC	LL
Maize	79.0 b	116 a	75.7 b	112.6 a
Trees	99.3 a	73.0 b	103.5 a	74.9 b
	Recovery of N (%)			
Season	SR 95		LR 96	
Treatment	CC	LL	CC	LL
Grain	5.3 a	6.5 a	4.7 a	5.5 a
Cob	0.3 a	0.3 a	0.5 a	0.4 a
Stover	2.9 b	5.0 a	2.4 a	3.7 a
Roots (maize)	0.8 b	1.4 a	0.6 b	1.2 a
Total maize	9.3 b	13.0 a	8.2 b	10.8 a
Trees	2.7 a	2.1 b	3.2 a	2.5 b
Soil	69.3 a	55.0 b	66.7 a	61.4 b
Unaccounted	19.6 b	29.7 a	21.9 b	25.3 a

Means followed by the same letter within a row at a particular season are not significantly different at $P < 0.05$. For abbreviations, see Table 1

Recovery figures for maize reported here, though low, are consistent with the 5–20% recovery range reported in the literature for most maize varieties (Ladd et al. 1981; Haggar et al. 1993; Jensen 1994; Vanlauwe et al. 1998). Most of the N from the applied biomass was left in the soil N pool as was shown by the high amounts of N in the soil pool after the end of the season (69% and 67% for calliandra and 55% and 61% for leucaena). Calliandra biomass, with a relatively lower decomposition and N release rate than leucaena (Mugendi et al.1999b), had more of the N left in soil and less of it unaccounted for compared to leucaena biomass. Other researchers, working with labeled N, reported >70% of N applied in the biomass as having been left in the soil after the first season's crop (Ladd et al. 1981; Ng Kee Kwong et al. 1987; Haggar et al. 1993; Becker et. al. 1994a; Vanlauwe et al. 1998). It is generally assumed that the large N fraction remaining in the soil at the end of the season would decompose slowly and have some residual effects on the subsequent season's crop. Indeed, many researchers working with labeled materials have reported residual effects on succeeding crops, though in most cases these effects are generally very low (Singh et al. 1991; Ladha et al. 1992; Vanlauwe et al. 1998). The low residual effect may be attributed to the fact that organic substrates left in the soil after the more labile organic portions decompose form part of the soil humus containing components that decompose and release nutrients slowly (Becker et al. 1994b). Residual effects were difficult to assess in the present study due to application of the labeled second-generation (LR 96) prunings. However, it was observed that the recovery figures of the second season's crop were close to those of the first season with even the tree hedges showing higher recovery values in the second season compared to the first. This observation is in spite of the fact that determination of recovery values took into account the amount of N left in the soil at the end of the first season.

Recovery of N by the tree hedges (leaves) was relatively low (2–3%), but comparatively more in calliandra than in leucaena hedges. This again may be explained by the more aggressive rooting system of calliandra compared to leucaena trees (as explained in Mugendi et al. 1999a). An estimation of N recovery in the tree wood and roots (assuming that all plant parts have the same fraction of total N derived from added ^{15}N as found in the leaves) more than tripled the total amount of N recovered by calliandra and leucaena tree hedges respectively. The resulting figures seem to compare reasonably well with the recently published results of Vanlauwe et al. (1998) – the only kind available for comparison – in which *Leucaena leucocephala* hedgerows recovered 16%, 9%, 8%, and 2%, respectively, of the total N applied to the soil (as labeled prunings) following four subsequent pruning applications at IITA, south western Nigeria. On the other hand, *Dactyladenia barteri* (Hook fex Oliv.) Engl. hedgerows recovered 0.3%, 0.3%, 3.1%, and 3.4% of the total N applied during the same period. Palm (1995) stated that the biggest and perhaps the most important unknown in agroforestry systems is the amount of nutrients released from added plant material that is taken up by the hedgerow trees. More studies therefore need to be conducted to accurately determine how much of the nutrients applied in the tree-leaf biomass are eventually taken up by the tree hedges for the different species commonly used in different agroforestry systems and for the different environments.

Some of the relatively large amounts of N that were unaccounted for (20–30%) may have been left in the wood and root portions of the tree hedges (since N in the tree hedges was only determined in the tree leaves). An estimation of N recovery in the tree wood and roots that more than tripled the amount of N recovered by the tree hedges indicates that wood and roots could contain a sizable fraction of the unaccounted for N. Some more of the unaccounted N might have also been left in the bulk of the soil below the 20 cm depth since soil sampling was not determined below this depth. The rest of the N may have been lost, principally through leaching (as discussed in Mugendi et al.1999b).

The lower amount of N that was unaccounted for in the treatments that received calliandra biomass compared to those that received leucaena biomass was due to the fact that calliandra biomass decomposed and released N at a much slower rate than leucaena biomass (Mugendi et al. 1999b). Thus, leucaena leafy biomass, which released over 50% of its N approximately 2 weeks after incorporation into the soil, may have released the N too fast and too early in the season before the maize crop developed an extensive root system to take it up. Such available N in the soil is subject to loss through leaching, volatilization and denitrification or it may be immobilized into forms not readily available to plants. Calliandra's biomass slower decomposition and N release rate compared to leucaena me be explained by its high polyphenolic concentrations which are known to bind with N lowering the decomposition rate and N release (Chesson 1997).

The study shows that only a small fraction of the N contained in the N-rich biomass that is applied to the soil is taken up by the current season's crop, suggesting that a major benefit may be in the build-up of soil N store. There is a need to conduct more studies to accurately determine the amounts of nutrients released from added tree biomass that are taken up by trees in different agroforestry systems.

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