Biol Fertil Soils (2012) 48:51–66
DOI 10.1007/s00374-011-0607-8

ORIGINAL PAPER

Carbon and nitrogen mineralization dynamics in different soils of the tropics amended with legume residues and contrasting soil moisture contents

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Received: 1 May 2011 / Revised: 13 July 2011 / Accepted: 17 July 2011 / Published online: 29 July 2011

Abstract Seasonal drought in tropical agroecosystems may affect C and N mineralization of organic residues. To understand this effect, C and N mineralization dynamics in three tropical soils (Af, An_1, and An_2) amended with haricot bean (HB; *Phaseolus vulgaris* L.) and pigeon pea (PP; *Cajanus cajan* L.) residues (each at 5 mg g^{-1} dry soil) at two contrasting soil moisture contents (pF2.5 and pF3.9) were investigated under laboratory incubation for 100–135 days. The legume residues markedly enhanced the net cumulative CO_2–C flux and its rate throughout the incubation period. The cumulative CO_2–C fluxes and their rates were lower at pF3.9 than at pF2.5 with control soils and also relatively lower with HB-treated than PP-treated soil samples. After 100 days of incubation, 32–42% of the amended C of residues was recovered as CO_2–C. In one of the three soils (An_1), the results revealed that the decomposition of the recalcitrant fraction was more inhibited by drought stress than easily degradable fraction, suggesting further studies of moisture stress and litter quality interactions. Significantly ($p<0.05$) greater NH_4^+–N and NO_3^-–N were produced with PP-treated (C/N ratio, 20.4) than HB-treated (C/N ratio, 40.6) soil samples. Greater net N mineralization or lower immobilization was displayed at pF2.5 than at pF3.9 with all soil samples. Strikingly, N was immobilized equivocally in both NH_4^+–N and NO_3^-–N forms, challenging the paradigm that ammonium is the preferred N source for microorganisms. The results strongly exhibited altered C/N stoichiometry due to drought stress substantially affecting the active microbial functional groups, fungi being dominant over bacteria. Interestingly, the results showed that legume residues can be potential fertilizer sources for nutrient-depleted tropical soils. In addition, application of plant residue can help to counter the N loss caused by leaching. It can also synchronize crop N uptake and N release from soil by utilizing microbes as an ephemeral nutrient pool during the early crop growth period.

Keywords CO_2–C flux · Labile pool · Legume residues · Litter quality · N immobilization · N mineralization · Moisture stress

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Introduction

The carbon input to soil through plant residues is an important source of labile C substrate that drives the growth and activity of heterotrophic microorganisms, which is a key linkage between soil C and N cycling (Hooker and Stark 2008). These inputs in turn are the sources of soil organic matter (SOM), thus influencing both soil structure and fertility. Additionally, the ability of soil to retain C has also become increasingly important for noncropping reasons, mainly for greenhouse gas mitigation strategies (Thomsen et al. 2008). Depletion of SOM is a serious threat to agricultural production and food security in many tropical regions; it leads to a decline in agricultural and biomass productivity, poor environmental quality, soil degradation and nutrient depletion, and finally, food insecurity (Lal 2004). Soil degradation is a serious problem

in most developing countries especially in Sub-Saharan Africa (Mulumba 2004).

As a practical means to improve soil productivity, amendment of local organic residues has been gaining worldwide support (Huang et al. 2004). Meanwhile, organic resources constitute a major source of nutrient input to both soils and livestock in smallholder tropical production systems (Shepherd et al. 2003). Incorporation of crop residues provides readily available C and N to soils. Legumes are vital sources of N that could offset the nutrient depletion resulting from continued cereal cropping. Ethiopia has a wide diversity of legumes and correspondingly harbors huge residents of rhizobial strains that fix atmospheric N_2 (Beyene et al. 2004; Wolde-meskel et al. 2005). Hence, incorporation of plant residues, mainly legumes, to agricultural soils has huge benefits. It is useful for sustaining SOM content, enhancing biological activity, improving physical properties, increasing nutrient availability, improving water infiltration, decreasing evaporation, and increasing water-holding capacity of soils (Kumar and Goh 2000; Palm et al. 2001).

The amount of N that recycles into agricultural fields through residues may add 25–100 Tg N $year^{-1}$ into agricultural soils, mainly from crop residues (Mosier and Kroeze 1998). Indeed, legume residues and animal manures are potentially important source of nutrients for crop production in smallholder agriculture of Sub-Saharan Africa. However, the decomposition rate of residues is often regulated by environmental factors, such as temperature and soil moisture, and quality factors and their interaction (Heal et al. 1997). For instance, legume residue decomposition and nutrient release rates are influenced by legume quality parameters such as N, polyphenol, and lignin contents and their ratios (Palm and Sanchez 1990; Tian et al. 1992; Giller and Cadisch 1997; Heal et al. 1997).

In low-input farming systems, the primary sources of mineral N to crops is that produced by microbial mineralization of N in crop residues and SOM. Synchronization of net N mineralization with plant/crop growth is desirable to maximize N delivery for the crop and minimize losses by leaching. One of the "instruments" to achieve synchronization is the amendment of soils with plant residues of different qualities. The application of residues with high C/N ratio results in immediate net N immobilization, while the application of residues with low C/N ratio results in net N mineralization.

Under tropical conditions, temporal variations in moisture have a more pronounced effect on microbial activity and decomposition than temperature, which remains relatively constant except during the dry season (Cornejo et al. 1994). Therefore, temperature is not a constraint in most tropical climates; rather moisture could be a predominant factor that impedes organic matter (OM) decomposition and N mineralization. On the other hand, the response of microbes to sporadic rainfall events and then to subsequent decreasing water availability as soils dry will vary with taxon. Thus, with decreasing water availability, a smaller proportion of the microbial community is likely to be active. As such, the composition of the active microbial community may also become more similar (converge) with decreases in water availability (Ford et al. 2007). Soil moisture content influences microbial activity by modifying substrate availability and affecting osmotic potential. Consequently, water potential alters the rate of OM mineralization. Rates of microbial processes are generally most rapid near field capacity (−10 kPa) and then linearly decline as soil water potential becomes more negative (Linn and Doran 1984; Paul and Clark 1996).

Most decomposition studies conducted elsewhere have dealt with litter quality, mainly of forest litters and green manures, in field conditions during optimum soil moisture availability. However, information on the effects of soil moisture and residue quality on decomposition and N mineralization of residues are scarce. Therefore, the objective of the present study was to understand the decomposition and N mineralization potential of legume residues of different qualities in three tropical soils and with two moisture contents at constant temperature (15°C) under laboratory condition.

Materials and methods

Soils sampling sites

The composite soil samples were collected from Awassa, Wondo Genet, and Yirgalem, located in southern Ethiopia. The first two soils are Andosols (both loam), hereafter referred as An_1 and An_2, respectively, while the soil of Yirgalem is an Alfisol (clay), hereafter referred as Af. At the time of sampling, the An_1 soil was being cropped with maize, whereas the soils of Wondo Genet and Yirgalem were under perennial crop (coffee). The surface soil of Af had 39.0 g kg^{-1} OM, 24.4 g kg^{-1} total C, 22.6 g kg^{-1} organic C, 1.53 g kg^{-1} total N, and 1.2 mg kg^{-1} available P. The surface soil of An_1 contained 22.4 g kg^{-1} OM, 13.0 g kg^{-1} total C, 12.6 g kg^{-1} organic C, 0.92 g kg^{-1} total N, and 67 mg kg^{-1} available P. Similarly, the An_2 surface soil had 54.5 g kg^{-1} OM, 32.3 g kg^{-1} total C, 31.6 g kg^{-1} organic C, 2.40 g kg^{-1} total N, and 13.6 mg kg^{-1} available P. The Af soil contains 55%, 26%, and 19% of clay, silt, and sand, respectively, while An_1 had 21%, 42%, and 37% clay, silt, and sand, respectively. The An_2 also had 19%, 37%, and 36% clay, silt, and sand, respectively, which is very similar to the content of An_1. The initial soil pH (H_2O) of the three soils is 5.5, 7.3, and

6.2, for Af, An_1, and An_2, respectively. The long-term climate data for the soil (Af, An_1, and An_2) sampling sites are given in Fig. 1.

Each soil sample was collected from 10 to 15 points at a depth of 0–15 cm, immediately transported to Hawassa University, and then air dried under ambient temperature. The air-dried samples were transported to Norwegian University of Life Sciences, Norway in August 2007 where the study was conducted. Large aggregate samples were ground and passed through a 2-mm sieve for the determination of selected soil characteristics.

Both respiration and N mineralization experiments were set separately in factorial combination of the treatments: 3×2×2, soil types, legume residues, and soil moisture, respectively. The finely ground residues were applied at 5 mg g^{-1} dry soil after adjusting soil moisture to the required levels. The treatments were laid out in completely randomized design in triplicates under laboratory condition.

Plant residues sampling and selection of legume species

Legume residues of cowpea (CP; *Vigna unguiculata* (L.) Walp.), haricot bean (HB; *Phaseolus vulgaris* L.), pigeon pea (PP; *Cajanus cajan*), and soybean (SB; *Glycine max*) were sampled at the time of maturity from research and farmers' fields. The residues collected were similar to what farmers usually leave in the field after harvesting grains. The aboveground biomass including stems, petioles, and leaves were collected and employed for the study. However, since the PP stems were woody, they were not included. The average nutrient concentrations were 42.8% C and 2.0% N in CP, 40.6% C and 1.0% N in HB, 44.4% C and 2.2% N in PP, and 41.7% C and 1.0% N in SB. Based on their distinct C/N ratio variation, both HB and PP were finally selected for this particular study. The selection of these two species of legumes was also based on their practical merits, i.e., HB is produced extensively, is highly productive, and is an export commodity more than others, while PP has a wide agroecological coverage and is highly adapted to moisture stress condition.

Soil moisture regulation and moisture treatment setup

The moist soil samples were prepared by equilibrating to pF=2 (−10 kPa) with a pressure plate apparatus. The gravimetric soil moisture content of the moist and air-dried soil samples for proportional mixing were determined by oven drying at 100°C for 24 h. Thus, the two moisture levels, pF2.5 and pF3.9, were attained by mixing moist and air-dried soils samples for the incubation experiments. The two soil moisture levels were selected based on the previous glutamate mineralization study (Girma et al. 2009, unpublished). After adding and mixing 10 g (dry weight basis) moist and dry soils into 120 mL serum flasks, the legume residues were amended at 5 mg g^{-1} dry soil. The flasks were immediately sealed with butyl rubber septa to avoid moisture loss for respiration measurement. For the N mineralization experiment, 10 g (dry weight basis) moist and dry soils were added into 50-mL tubes and the tubes were immediately capped to avoid moisture loss. The amended soil samples were rolled on a horizontal roller bed for 1 min to homogenize soil moisture and uniformly distribute the legume residues. The respiration and N mineralization kinetics studies were conducted by incubating in water bath system at constant temperature (15°C). Both the flasks and tubes were regularly aerated after each sampling to maintain aerobic condition.

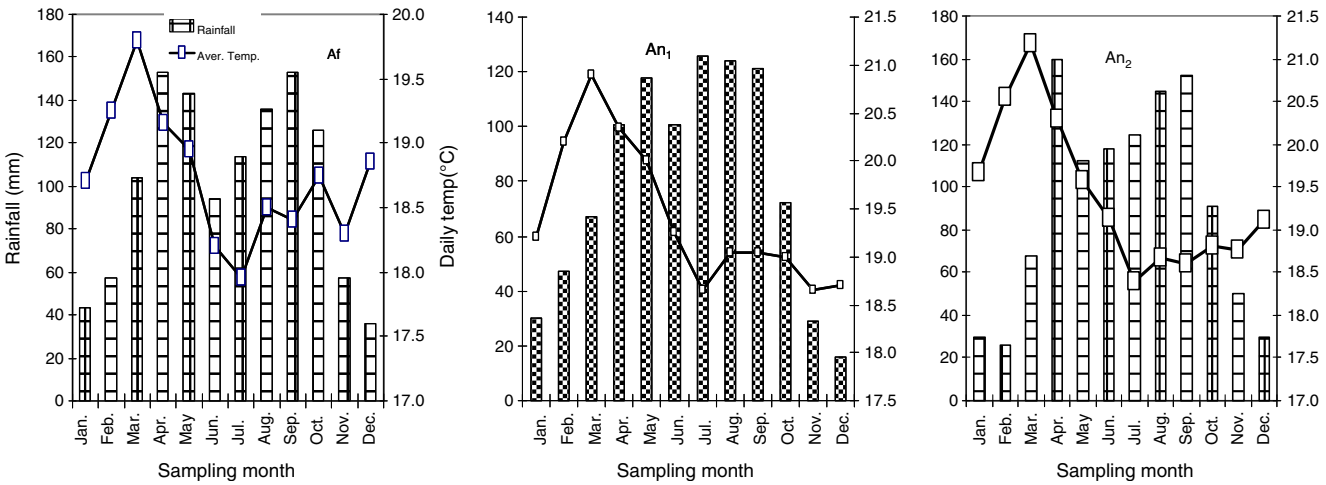

Fig. 1 Long-term (more than 20 years) average daily air temperature (in degrees Celsius) and annual rainfall (in millimeters) of Af, An_1, and An_2 soil sampling sites

Soil and plant tissue analyses

The total and organic C contents were determined by dry combustion according to the Allison method (Nelson and Sommers 1982). Total N was determined by the Dumas method (Bremner and Mulvaney 1982). Extractable P was determined with the Bray II method (Bray and Kurtz 1945). Soil pH was determined with a pH electrode at a soil/water ratio of 1:2.5 (pH H_2O). The concentrations of NH_4^+–N and NO_3^-–N were determined by extracting ~10 g air-dried soil with 25 mL of 2 M KCl solution. The soil slurries were shaken in 50-mL tubes for 30 min with reciprocal shaker at 180 strokes per minute and centrifuged for 10 min. The slurries were filtered with Whatman #42 filters. The NH_4^+–N content was measured by colorimetric method (NSF 1975), while NO_3^-–N was measured with continuous flow injection analysis method (EPA 1979).

Respiration and N mineralization

Respiration (CO_2–C flux) was measured using a robotized gas chromatography (Molstad et al. 2007) with and without legume residue amendment at two contrasting soil moisture contents (pF2.5 and pF3.5). The high gas standard, air, and low standards were employed for calibration, dilution, and leakage estimation that might happen through the peristaltic pump during automatic sampling. The respiration measurement was performed at different sampling dates: 3, 7, 15, 30, 50, 75, and 100 days of incubation. Meanwhile, O_2 concentration was inspected at each sampling date. The level of O_2 concentration was reduced from about 20% to 10% at the end of the incubation period, suggesting sufficient O_2 concentration was maintained for aerobic respiration throughout the incubation period. The net cumulative flux and rates of CO_2–C fluxes due to legume residue amendment were calculated as the difference between the amended and non-amended (control) soils, assuming that the priming effect was negligible.

Nitrogen mineralization was assessed based on NH_4^+–N and NO_3^-–N contents. The soil samples for initial NH_4^+–N and NO_3^-–N contents were immediately frozen after the required soil moisture regulation. The soils incubated for N mineralization were destructively sampled at 0, 3, 7, 15, 30, 50, 75, 105, and 135 days of incubation. The net NH_4^+–N and NO_3^-–N content at each sampling date was determined by subtracting the initial values from the final values (sampling dates). Similarly, the net N released from legume residue-amended soil was determined by subtracting the values of controls. Both respiration and N mineralization were measured in triplicates and incubation was carried out at constant temperature of 15°C.

Statistical analyses

The effects of soil moisture and legume residue amendment on C and N mineralization dynamics were assessed by one-way and two-way analyses of variance using the Minitab software program (Minitab 14). The means that were found different were declared statistically significant at $p \leq 0.05$. The correlation matrix was calculated to investigate the relationships of C and N mineralization. The CO_2–C flux from legume residue-amended and control soils were subjected to different regression models to identify the best fits and to calculate the rate of CO_2–C flux.

Results

Carbon mineralization

Legume residue amendment apparently stimulated the net cumulative flux and rate of CO_2–C flux over 100 days of incubation (Fig. 2 and Table 1). The net cumulative CO_2–C flux of legume residue-amended soil samples was 3–14 times greater than the control samples. The CO_2–C flux was greater with PP than HB amendment in most of the incubation periods. The trends and degree of C mineralization appeared to be very similar among the three soils when legume residues were amended (Fig. 2). The cumulative CO_2–C flux greatly varied in response to soil moisture effect with the control soil samples of Af (Fig. 2). The CO_2–C flux was relatively low at lower soil moisture content (pF3.9) with control soils. By contrast, the variation in CO_2–C flux was marginal in response to soil moisture effect when legume residue was amended, except with An_1 (Fig. 2). The C mineralization of legume residue was explained by logarithmic function (R^2=0.96–0.99), whereas the native SOM–C mineralization was displayed by linear function (R^2=0.93–0.98) during the 100-day incubation period.

The rate of CO_2–C flux varied among soil types in response to legume residue amendment and soil moisture levels (Table 1). The highest rate was recorded in An_2, while the lowest rate was recorded in An_1 averaged over organic sources and soil moisture contents. The rate of flux was nonsignificantly different between the two moisture levels in Af soil amended with legume residues, while An_1 and An_2 soil samples revealed significant variation between the two moisture levels with legume residue amendment. The An_1 soil CO_2–C flux was greater at pF2.5 with HB and PP, while in An_2, it was relatively greater with PP at both pF2.5 and pF3.9 than HB. The rate of flux of control samples of An_2 soil was high, followed by Af at pF2.5 soil moisture content. However, the flux rate showed a declining trend with all soils at low soil moisture content

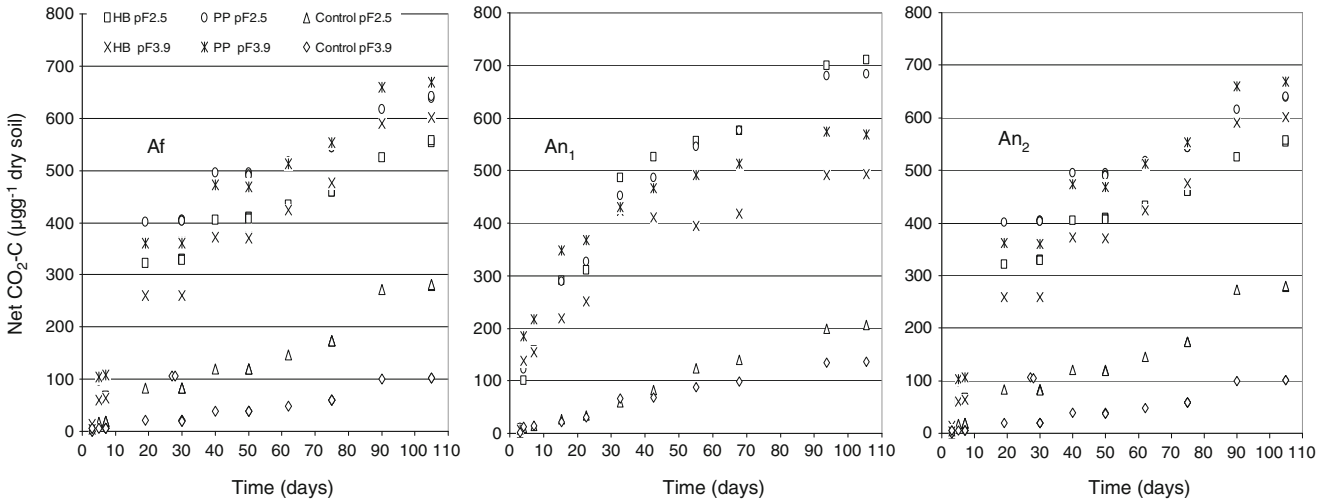

Fig. 2 Net cumulative CO_2–C efflux of Af, An_1, and An_2 soils in legume residue-amended and non-amended conditions in response to contrasting soil moisture contents (pF2.5 and pF3.9). *HB* haricot bean residue, *PP* pigeon pea residue

(Table 1). The respiration rate of An_2 soil was greater with and without legume residue amendment at both soil moisture levels than other soils (Table 1).

The index ratio (amended over non-amended samples) of the rate of CO_2–C flux was calculated to understand the drought stress effect on microbial activity. Accordingly, the index ratios for Af were 2.4 and 6.1, while 2.4 and 2.9 for An_1 and 2.5 and 4.6 for An_2 at pF2.5 and pF3.9, respectively. In other words, the rate of CO_2–C respiration was 2.4 times higher when legume residues were amended than the control Af at pF2.5 and six times higher at pF3.9. Likewise with An_1 soil, CO_2–C respiration was 2.4 times higher at pF2.5 and 2.9 times higher at pF3.9. The same trends were also observed with An_2, where the CO_2–C respiration was 2.5 times higher at pF2.5 and 4.6 times higher at pF3.9 when legume residues were amended than the control samples. In general, the index ratio of CO_2–C flux was greater at lower soil moisture content with all soils (Table 1).

The index ratio of the rate of CO_2–C flux in control soil samples at pF2.5 over pF3.9 was also estimated to understand the moisture stress effect on microbial activity. Hence, Af, An_1, and An_2 soil samples showed 2.4, 1.5, and 1.6 times higher rate of respiration, respectively, at pF2.5 than at pF3.9 (Table 1).

As a result of the decomposition of legume residues, the pH in Af and An_2 soil samples were increased compared to their respective controls, while soil pH in An_1 soil sample was reduced. Similarly, at low soil moisture content (pF3.9), the decomposition of legume residue resulted in increased soil pH. Soil types and legume residues interaction showed highly significant variation ($p \leq 0.001$) on soil pH (Table 2).

Carbon recovery

Carbon recovery from legume residue amendment varied among the soil types and between soil moisture contents

Table 1 Rate of net CO_2–C flux (in micrograms per gram dry soil) in tropical soils amended with legume residues at contrasting soil moisture contents during 100 days of incubation

Treatment	Af		An_1		An_2	
	pF2.5	pF3.9	pF2.5	pF3.9	pF2.5	pF3.9
C	2.2	0.9	2.0	1.3	2.9	1.8
PP	5.3	5.7	5.6	3.9	7.7	8.6
HB	5.1	5.3	5.9	3.7	7.0	8.1

R^2 =0.70–0.98

HB haricot bean residue, *PP* pigeon pea residue, *C* control (non-amended soil)

Table 2 Effect of mineralization of legume residues on soil pH after 100 days of incubation

Soil	Soil moisture	Soil pH		
		HB	PP	C
Af	pF2.5	5.65±0.02	5.75±0.05	5.19±0.20
	pF3.9	5.79±0.09	5.79±0.08	5.57±0.06
An_1	pF2.5	7.07±0.19	7.16±0.02	7.30±0.16
	pF3.9	7.46±0.04	7.27±0.04	7.64±0.12
An_2	pF2.5	5.81±0.05	5.86±0.07	5.67±0.07
	pF3.9	6.20±0.05	6.58±0.12	5.92±0.05

Each value is followed by the standard deviation

HB haricot bean residue, *PP* pigeon pea residue, *C* control (non-amended soil)

(Fig. 3). The average C recovery of legume residue-amended soils ranged from 21% to 32% at pF2.5 and from 20% to 23% at pF3.9 after 50 days of incubation. The C recovery increased to 35–42% and 32–35% at pF2.5 and pF3.9, respectively, after 100 days of incubation. The soil moisture stress effect on C recovery was high with Af and An_1 soils amended with legume residues. After 50 days of incubation, An_1 samples showed pronounced variation in C recovery in response to soil moisture. However, the An_2 soil revealed marginal difference in C recovery between the two soil water contents (Fig. 3).

Nitrogen mineralization

Effect of legume residue and soil moisture on ammonification

The NH_4^+–N accumulation varied among soils and between soil moisture levels and days of incubation. However, the way the data presented can conceal the results. To clearly reveal each effect, we have presented the data in at least two tables (Tables 3 and 4). Ammonification was significantly affected by different organic N sources. The average net NH_4^+–N accumulation was relatively high in acidic Af soil than the rest with or without legume residue amendment (Tables 3 and 4). Similarly, a greater amount of NH_4^+–N accumulated with PP amendment than with HB of Af soils. By contrast, An_2 showed relatively high NH_4^+–N immobilization irrespective of the amendment, while An_1 soil showed net ammonification and immobilization with or without residue amendment, respectively. The results revealed that net ammonification was related with soil moisture content. However, the effect of soil moisture varied based on soil organic N sources, soil types, and incubation period (Tables 3 and 4). Accordingly, average NH_4^+–N accumulation was superior with Af at relatively low soil moisture content (pF3.5). On the other hand, NH_4^+–N accumulation of An_1 and An_2 soils was unaffected by the two moisture contents (Table 3). Indeed, though the levels varied among soil types and moisture levels at respective sampling dates, all soils showed either periodical mineralization and immobilization or periodical immobilization and remineralization when legume residues were amended (Table 4). At both soil moisture contents, Af non-amended (control) soil showed episodic immobilization and mineralization of ammonium (Table 4). On the other hand, An_1 and An_2 soils completely immobilized ammonium at respective sampling times throughout the incubation period. The interaction effect of organic N sources with soil moisture was also significant ($p<0.03$) in affecting NH_4^+–N content of Af soils, but not with others (Table 3).

Effect of legume residue and soil moisture on nitrification

The net nitrification varied between soils, organic N sources, and soil moisture levels during 135 days of incubation (Tables 3 and 5). The average net nitrification was superior with An_2 control soils to An_1 and Af control soils. The average net nitrification of Af soil was greater with PP amendment irrespective of soil moisture content than An_1. In contrast, An_2 soil showed either complete immobilization of NO_3^-–N or very low rate of nitrification

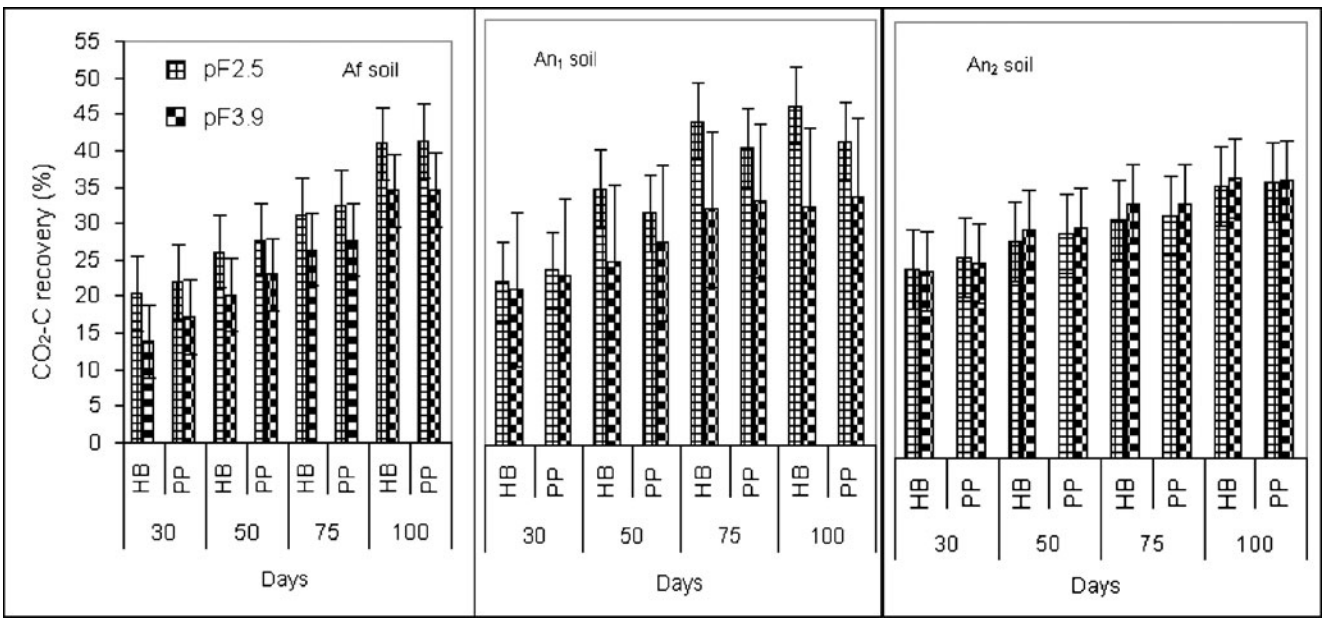

Fig. 3 Average C recovery (in percent) of tropical soils (Af, An_1, and An_2) amended with legume residues (*HB* haricot bean, *PP* pigeon pea) and contrasting soil moisture contents. *Vertical bars* indicate standard error

Table 3 Inorganic N pools (NH_4^+–N and NO_3^-–N) in three tropical soils as influenced by legume residues and soil moisture contents after 135 days of incubation

Soil type	Soil moisture		Organic N sources			Interaction[a]
	pF2.5	pF3.9	C	PP	HB	Prob.
NH_4^+–N (mg kg^{-1})						
Af	−4.53b	6.80a	0.08b	−4.43c	−4.4c	0.032
An_1	−3.94a	−3.87a	−14.12b	1.06a	1.06a	0.520
An_2	−10.69a	−7.10a	−17.20b	−5.46a	−5.4603a	0.695
NO_3^-–N (mg kg^{-1})						
Af	15.75a	3.16b	13.68a	16.10a	1.41b	0.002
An_1	1.38a	3.65a	16.88a	7.95b	−17.28c	0.897
An_2	12.11a	1.70b	56.16a	−6.83a	−28.61c	0.203

Mean values followed by different letters in a row show statistically significant difference (otherwise nonsignificant)

HB haricot bean residue, *PP* pigeon pea residue, *C* control (non-amended soil)

[a] The interaction effect between organic N sources and soil moisture

with legume residue amendment irrespective of soil moisture content (Tables 3 and 5). The legume N sources exhibited two phases of nitrification when amended to An_2 soil at the beginning (days 0–3) and at the end (days 75–135) of the incubation period.

The Af and An_2 soils displayed greater net nitrification at pF2.5 irrespective of organic N sources at respective sampling dates. The nitrification was three to six times higher at pF2.5 than at pF3.9 for Af soil amended with PP, whereas it was double with nontreated An_2 (Tables 3 and 5). By contrast, An_1 soil exhibited greater nitrification at low soil moisture content with all organic N sources (Table 5). In general, with respect to sampling date, the result revealed cyclical nitrification and immobilization irrespective of organic N sources and soil moisture levels (Table 5).

The relationships between C and N mineralization with legume residue amendment of the tropical soils revealed that C recovery was positively and significantly correlated with ammonification (r=0.446, p<0.017) and nitrification (r=0.474, p<0.011, n=32) with Af soils, whereas C recovery was nonsignificantly and positively correlated with ammonification of An_1 (r=0.293, p<0.110, n=32) and An_2 (r=0.157, p<0.391, n=32). By contrast, C recovery was positively and significantly associated with nitrification of An_1 (r=0.544, p<0.002, n=32) and An_2 (r=0.370, p<0.037, n=32). The results indicated that most soil microorganisms get their energy and substance from organic materials. Hence, N mineralization and transformation are intimately linked to organic C decomposition (Huang et al. 2004).

Rate of N mineralization/immobilization

The rate of N mineralization/immobilization was calculated by linear regression (Fig. 4). With Af soil, the rate of N immobilization at pF3.9 was 78% for HB, 25% for PP, while for An_1, N immobilization at pF3.9 was 75% for HB and, by contrast, N mineralization of 284% took place with PP. Similarly, with An_2 soil, the rate of N immobilization at pF3.9 was 46% for HB and 51% for PP (Fig. 4). The study showed strong interaction between soil types and legume residue types.

Discussion

Carbon mineralization

The present study revealed 3–14 times greater net cumulative CO_2–C flux of legume residue-amended soil than the control (Fig. 2). Similar to our findings, greater CO_2–C flux was reported with plant residue addition to soils by some other workers, largely through microbial biomass C and N flushes compared to untreated soils (Huang et al. 2004; Xu et al. 2006; Bertrand et al. 2007; Lou et al. 2007; Hooker and Stark 2008). In accordance with our results, CO_2 production in the rape seed residue-amended soils respired nearly 10 times greater than the basal respiration (Bertrand et al. 2007). The higher CO_2–C flux in residue-treated soil suggests that soil microbial activities could be C substrate limited for energy and biomass production. Our findings also confirmed that organic amendments improved soil biology by increasing soil microbial activities (Stark et al. 2006; Vinten et al. 2002).

The C decomposition of legume residue-amended samples was rapid (Tables 6 and 7). In the early sampling date (third day), the net CO_2–C flux of residue-treated samples was five times higher than the control soils. The rapid C decomposition of legume residue-amended soil

Table 4 Net ammonification (NH_4^+–N, in milligrams per kilogram) of organic N sources amended to tropical soils in response to soil moisture contents

Days of incubation	Af						An_1						An_2					
	pF2.5			pF3.9			pF2.5			pF3.9			pF2.5			pF3.9		
	HB	PP	C	HB	PP	C	HB	PP	C	HB	PP	C	HB	PP	C	HB	PP	C
3	15a	24a	−12b	14a	17a	3b	1a	2a	−15b	−0.8a	−0.1a	−13b	−10a	−9a	−13a	−3b	0.4a	−10c
7	−2b	0.1b	5a	3b	11a	6b	−0.8a	−0.1a	−13b	1a	2a	−15b	−18a	−17a	−11a	−6b	−3a	−9c
15	−22c	−1b	10a	9b	22a	−6c	−0.1a	0.6a	−14b	0.7a	0.8a	−14b	−6a	−6a	−24b	−22c	−20b	2a
30	−23c	−3b	5a	19b	27a	−14c	2a	1a	−14b	2a	3a	−14b	0.0b	0.8a	−30c	−19b	−17b	−1a
50	−24b	−15b	7a	2b	36a	−13c	2a	2a	−14b	2a	2a	−15b	−2a	−2a	−27b	−1b	−0.8b	−16b
75	−30c	−15b	14a	−12b	8b	11a	1a	1a	−14b	2a	2a	−15b	0.1b	0.3a	−30c	−8a	−8a	−12a
105	−9b	−3a	−9b	−5b	6a	2ab	0.9a	1a	−13b	2a	2a	−14b	1.3a	0.2a	−28b	3b	5a	−20c
135	−5b	−0.6a	−14bc	3b	10a	0.7b	2a	2a	−14b	0.8a	1a	−14b	0.1a	−0.5a	−27b	3b	11a	−20c

Mean values followed by different letters in a row show statistically significant difference (otherwise, nonsignificant difference). Values are the mean of observations

HB haricot bean residue, *PP* pigeon pea residue, *C* control (non-amended soil)

Table 5 Net nitrification (NO_3^-–N, in milligrams per kilogram) of legume residue-amended tropical soils in response to soil moisture contents

Days of incubation	Af						An_1						An_2					
	pF2.5			pF3.9			pF2.5			pF3.9			pF2.5			pF3.9		
	HB	PP	C	HB	PP	C	HB	PP	C	HB	PP	C	HB	PP	C	HB	PP	C
3	−3b	16a	16a	4a	5a	6a	9b	23a	10b	10b	26a	24a	8c	17b	54a	−5c	4b	32a
7	−5b	17a	21a	4b	5b	7a	−0.3b	13a	18a	1b	16ab	30a	−3b	−2b	74a	−9c	3b	36a
15	−5c	21a	23a	3b	2b	7a	−22c	0.5b	17a	−13b	6a	25a	−30c	4b	75a	−45c	−26b	60a
30	−5b	28a	29a	6a	5a	7a	−40c	0.0b	27a	−36c	3b	29a	−81c	−42b	123a	−42c	−22b	56a
50	−4b	55a	36a	7a	10a	6a	−48c	−0.2b	36a	−43c	−0.1b	35a	−80c	−11b	132a	−64c	−36b	83a
75	−17b	53a	56a	−3b	−1b	17a	−33b	26a	26a	−46c	16b	37a	−74c	−14b	140a	−36c	5b	54a
105	−0.6b	8a	−1b	−3b	−3b	−5a	−4b	0.3a	−11c	−4b	−0.4a	−12c	1b	6a	−5c	0.5a	0.5a	−6b
135	−0.9b	8a	0.1b	−3a	−3a	−4a	−4b	0.8a	−11c	−4b	−0.2a	−12c	1a	2a	−4b	0.7a	0.5a	−6b

Mean values followed by different letters in a row show statistically significant difference (otherwise, nonsignificant)

HB haricot bean residue, *PP* pigeon pea residue, *C* control (non-amended soil)

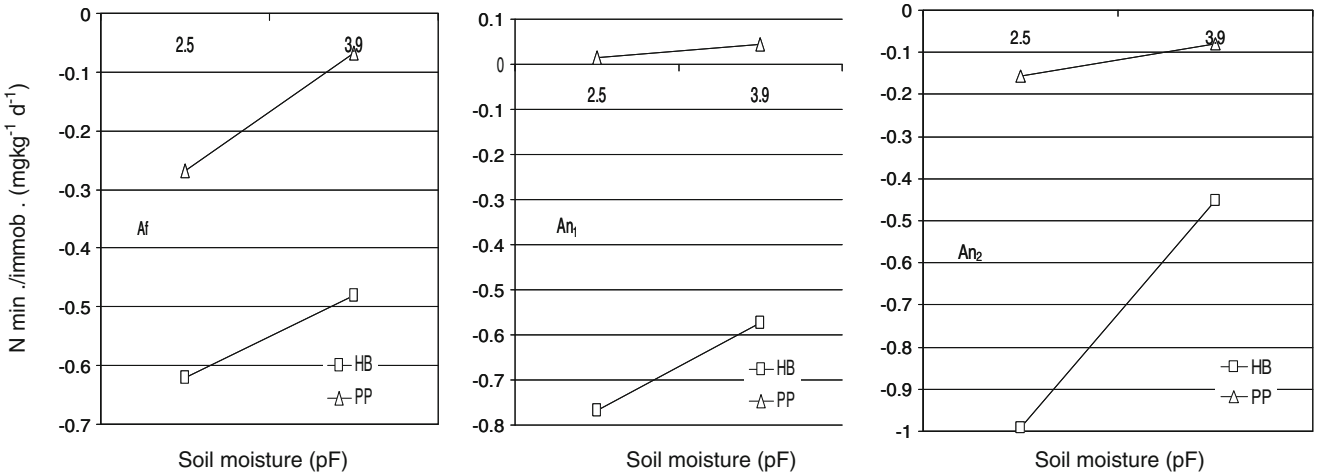

Fig. 4 The rate of N mineralization (*min.*)/immobilization (*immob.*) of HB-amended and PP-amended tropical soil samples (Af, An_1, and An_2) under contrasting soil moisture contents. *HB* haricot bean residue, *PP* pigeon pea residue

samples showed that the residues comprise soluble or labile C pool (Tables 6 and 7). Similar studies also observed that organic sources which contain labile C pool (organic acids, amino acids, and simple sugars) could be rapidly mineralized than the nonlabile pool (Rochette et al. 2000; Huang et al. 2004). In one of the three soils (An_1), the results revealed that the decomposition of the recalcitrant fraction was more inhibited by drought stress than the easily degradable fraction (Fig. 2). The CO_2–C flux was greater with legume residues in An_1 samples at both pF2.5 and pF3.9, suggesting that it is more responsive to substrate addition than the other soil samples. Correspondingly, the An_1 soil index ratio of CO_2–C flux of amended over non-amended at pF2.5 and at pF3.9 was similar, suggesting it is more substrate limited. The C substrate limitation of An_1 soil can be explained by its low organic C content (An_1= 1.26%) over other samples (for Af=2.24% and for An_2= 3.16%) and could be due to its continuous and intensive cultivation for decades. Therefore, this indicated that the application of organic amendments to soils could have more pronounced impact on biological and physicochemical properties of poor fertility soils. On the other hand, the highest index ratio of CO_2–C flux of amended over non-amended soil samples were observed with Af soil at the lowest soil moisture content (pF3.9). This implies substantial limitation in water availability of the soil samples for microbial activities attributed to its greater proportion of clay content. The CO_2–C flux was relatively low at low soil moisture content (pF3.9) for Af with control soils, implying that the availability of labile C pool and moisture stress interactively affected OM mineralization. However, the soil moisture stress effect was marginal with legume residue amendment because of the presence of soluble and easily mineralizable substrate that offsets the stresses. The lower CO_2–C flux of non-amended soils suggested the absence of sufficient labile pool, and as a result, the majority of microflora may exist in inactive condition. Several reports suggest that soil away from the rhizosphere is usually stressful for microorganisms. A majority of microorganisms exist in soils under such low-nutrient condition and may be starving (Morita 1997). In most of the cases, the CO_2–C flux was higher with PP than with HB, which could be related to its high N content and low C/N. Congruent with our results, greater CO_2–C flux was reported with the addition of organic sources with low C/N ratio by Xu et al. (2006). The present study showed that the trends of C mineralization appeared to be very similar among the three soils with legume residue amendment. In contrast, substantial variation in CO_2–C flux was elsewhere reported among the soils varying in initial pH (4.23–6.15) and organic carbon (OC) content (Xu et al. 2006; Bertrand et al. 2007).

Relative increase in soil moisture enhanced the rate of CO_2–C flux with or without legume residue amendment, perhaps through improving microbial activities. This could be attributed to easy substrate access by microorganisms due to substrate diffusion (Griffin 1981; Schjønning et al. 2003) and/or high soil moisture-facilitated mobility of microorganisms towards available substrate and optimum intracellular water potential (Csonka 1989; Killhma et al. 1993). Thus, our findings also confirm that decomposition rates usually increase with water potential, from −5,000 to −5 kPa, and then decrease in wetter conditions due to oxygen deficiency (Lomander et al. 1998) and in drier conditions due to stress and substrate limitation. The lowering of intracellular water potential alters enzyme conformation and inhibits activity (Csonka 1989; Brown 1990), or a combination of these mechanisms (Strak and Firestone 1995). On the other hand, the relative rate (amended) of CO_2–C flux was higher at lower soil moisture content with all soil samples (Table 1), suggesting that soil

Table 6 Net N mineralization (NH_4^+–N+NO_3^-–N, in milligrams per kilogram) of legume residue-amended tropical soils in response to soil moisture contents

Days of incubation	Af						An_1						An_2					
	pF2.5			pF3.9			pF2.5			pF3.9			pF2.5			pF3.9		
	HB	PP	C	HB	PP	C	HB	PP	C	HB	PP	C	HB	PP	C	HB	PP	C
3	12b	40a	3c	19a	22a	10b	11b	22a	12b	10b	23a	22a	−3b	8a	7a	−7c	5b	23a
7	−7c	17b	26a	7b	15a	13a	−1b	14a	14a	3b	16ab	27a	−22b	−19b	63a	−15c	0.3b	26a
15	−27c	20b	33a	12b	24a	0.3c	−22c	1b	18a	−12b	5ab	22a	−36c	−1b	52a	−67c	−45b	62a
30	−28b	42a	34a	25b	32a	−7c	−39c	2b	28a	−33c	2b	25a	−81c	−41b	93a	−60c	−38b	55a
50	−27b	57a	42a	9b	45a	−7b	−46c	2b	38a	−49c	−1b	32a	−82c	−12b	105a	−66c	−36b	67a
75	−47c	38b	70a	−13b	7ab	24a	−32b	27a	27a	−44c	15b	34a	−74c	−13b	110a	−44c	−3b	41a
105	−10b	5a	−10b	−8b	3a	−3ab	−3b	2a	−10c	−2a	−0.4a	−14b	3b	6a	−33c	4a	5a	−26b
135	−5b	7a	−14b	−5b	7a	−4b	−2b	2a	−8c	−3b	−0.6b	−14c	1a	1a	−31b	4b	11a	−26c

Mean values followed by different letters in a row show statistically significant difference (otherwise, nonsignificant)

HB haricot bean residue, *PP* pigeon pea residue, *C* control (non-amended soil)

moisture stress could be modified if readily mineralizable substrates are applied to soils.

The decomposition of legume residues markedly resulted in an increase of soil pH compared to the non-amended soil samples, particularly of acidic soils (Table 2). The increased pH observed in the two acidic soils following residue addition was also reported in many studies. It has been attributed to the alkalinity of plant residues (Paul et al. 2001; Sakala et al. 2004), decarboxylation of organic anions during decomposition (Xu et al. 2006), and/or ammonification of the added residue N (Bertrand et al. 2007).

Carbon recovery

During 30–75 days of incubation, Af soils C recovery was on average lower than the other two soils, An_1 and An_2. However, at the end of incubation period (100 days), its C recovery was higher over the others. This suggests that the microbial communities in this soils required certain time to adapt to the organic resources. The C recovery was 35–42% and 32–35% at pF2.5 and pF3.9, respectively, after 100 days of incubation (Fig. 3). After 50 days of incubation, An_1 samples showed pronounced variation in C recovery in response to soil moisture, indicating that the decomposition of the more recalcitrant fractions of the litter are more severely inhibited by moisture limitation (pF3.9 versus pF2.5) than the decomposition of easily degradable fractions (Fig. 2). This most likely suggests that polymer decomposition is more sensitive to moisture stress than monomer decomposition.

The variation in C recovery of soil samples in response to applied residues could be related to the level of native soil OC content and active microbial biomass. Interestingly, the highest native soil OC content of the soils led to the lowest C recovery in response to legume residue amendment. In accordance with our findings, Khalil et al. (2005) observed greater variation in C loss among different organic materials amended to different soil types. They reported significantly higher amounts of C (60–67%) loss with certain soils and relatively lower C (50–54%) loss with others, attributed to the substantial variation of the inherent C content of the soils and its pH. Similarly, Nyberg et al. (2002) observed high C loss (70–90%) using a $\delta^{13}C$ natural abundance technique for *Sesbania sesban*, while 53.8–61.2% of the added rape seed residues C loss was observed by Bertrand et al. (2007). On the other hand, the C loss from four leaves of agroforestry tree species studied in Ethiopia soils ranged from 10% to 44% (Teklay et al. 2007). The substantial variations in C recovery (loss) employing various organic resources in different investigations suggest that different factors control the whole process. Firstly, the variation could be ascribed to the

Table 7 Net cumulative N mineralization (NH_4^+–N+NO_3^-–N, in milligrams per kilogram) in response to organic N sources incubated with three soil types for 135 days irrespective of soil moisture contents

Days of incubation	Af			An_1			An_2		
	HB	PP	C	HB	PP	C	HB	PP	C
3	15.4	31.2	−6.2	10.0	9.5	3.0	−5.0	6.3	31.7
7	−0.3	15.9	19.9	0.6	0.3	10.3	−18.3	−9.1	44.6
15	−5.0	22.4	16.4	−17.1	−10.5	7.2	−51.4	−23.5	57.1
30	−1.9	36.9	13.3	−36.4	−12.9	13.3	−70.6	−39.9	74.0
50	−13.5	51.1	17.5	−43.3	−14.8	21.0	−74.0	−24.4	86.0
75	−30.5	22.2	48.5	−37.9	6.2	16.9	−59.3	−8.0	75.8
105	−7.3	3.9	−6.5	−2.8	−13.7	−24.8	3.2	5.8	−29.3
135	−3.7	7.2	−9.1	−2.5	−13.4	−24.9	2.6	6.2	−28.4
Mean	−5.8	23.8	13.3	−16.2	−6.2	2.8	−34.1	−10.8	39.0

HB haricot bean residue, *PP* pigeon pea residue, *C* control (non-amended soil)

differences in sites, soil types, soil moisture and temperature conditions, temporal resolution of the experiments, and methods used for the investigations. Secondly, the species and litter quality variations and time of incubation might have contributed to the substantial variation in C loss. Thirdly, the reduction in respiration (low C recovery) observed in the present study could be attributed to C utilization that tended to be more efficient due to C being redirected from waste respiration to microbial growth. Similar results were observed by Teklay et al. (2007). However, the stress effects such as soil acidity and contaminants could lead to more C respiration than C utilization for cell building (Xu et al. 2006).

Nitrogen kinetics

Ammonification

Net ammonification was relatively superior at pF3.5 than at pF2.5 with or without legume residue amendment across the sampling periods. Also, low NH_4^+–N immobilization occurred at pF3.5 than at pF2.5 (Table 5), suggesting the robustness of heterotrophic decomposers. However, it is not known whether the loss of ammonium was essentially due to immobilization by microorganism, fixation into clays or SOM, and rapid nitrification of NH_4^+ since isotopic tracer technique was not conducted in the present study.

The Af soil exhibited significant accumulation of NH_4^+–N with PP at pF3.5 across all sampling days probably due to adsorption of NH_4^+ to clay particles or due to low nitrifier activity as the soil was clay in texture and slightly acidic in pH. It is also important to note that net ammonification of Af varies with amendment, days of incubation, and moisture content (Table 4). Ammonium was rapidly produced with HB and PP residues at both moisture levels with Af and at pF2.5 with An_1 reflected with accumulation of NH_4^+–N content during the third day of incubation (Table 4). This could be because of the presence of favorable soil environment and active microflora. Another most likely explanation could be that the organic N fraction of the legume residues was composed of unstable forms (organic acids, amino acids, sugars, etc.) that could readily mineralize. In accordance with the present findings, a rapid NH_4^+–N accumulation was reported with alfalfa pellets, blood meal, and composted manure when sampled on the second day of incubation (Agehara and Warncke 2005). Verchot et al. (2001) also observed rapid turnover rate of NH_4^+–N at 2–4 days based on the pool size and gross mineralization rate among different forest soils.

Subsequently, Af soil exhibited net NH_4^+–N immobilization with HB and episodic accumulation with PP, which could be due to the variation in N content and C/N ratio of the residues. Although the magnitude was relatively low, An_1 exhibited net NH_4^+–N accumulation throughout the incubation period with HB and PP residues. However, An_2 immobilized through days 3 to 75 and remineralized through days 105 to 135 with both residues, implying rapid conversion of all NH_4^+–N to nitrate and then slow release of N afterwards (Table 4). In contrast, the three (Af, An_1, and An_2) control samples displayed ammonium immobilization at the third day of incubation, suggesting either lack of readily mineralizable substrate or the microflora that could be inactive soon after wetting the soils due to substrate starvation. Furthermore, another explanation could be that all the mineralized NH_4^+–N could be rapidly nitrified. However, Af soil showed episodic NH_4^+–N mineralization–immobilization pattern, whereas the other two soils (An_1 and An_2) displayed subsequently complete immobilization throughout the incubation period, suggesting high inherent C/N ratio of soils and the presence of active decomposing microbes and/or rapid nitrification of available NH_4^+–N. Our results support the hypothesis that soil microorganisms are efficient scavengers of inorganic N, with immobilization rates sometimes even exceeding mineralization rates

(Giblin et al. 1991; Wagener and Schimel 1998), but later, the biomass becomes a nitrogen source as the cells die and/or decompose.

Nitrification and N mineralization

Nitrate accumulation was the predominant form of inorganic N in all soil types (Tables 3 and 5). In most of the cases, nitrification was relatively greater and/or immobilization of nitrate was low at high soil moisture content (pF2.5) along with PP amendment, which is a contrast to ammonification. However, HB-treated soil samples showed inconsistent trend in response to soil moisture, possibly because the residue held more water, besides its low N content and high C/N ratio (Table 5).

Invariably, the soils exhibited cyclical nitrification and N mineralization with legume residue amendment. They manifested mineralization at the beginning, immobilization at the middle, and remineralization at the end of the incubation period (Tables 5 and 6). The nitrate immobilization observed in the present investigation could be either due to substrate limitation as reflected in the disappearance of NH_4^+–N or due to immobilization of NO_3^-–N into microbial biomass. Immobilization/mobilization of NO_3^-–N could be ascribed to high C/N ratio of the legume residues and soil samples, as moisture contents tested seems optimum for nitrifier activities. Soil moisture is an important factor influencing microbial activities; mainly, nitrifiers' activities are more inhibited than those of ammonifiers at soil moisture potential below −1.5 MPa (Singh and Kashyap 2007). The observed opposite trend of ammonification and nitrification to soil moisture contents suggest a differential sensitivity to water availability of the microorganisms responsible for the two soil N transformation processes (Yahdjian and Sala 2008). Thus, soil moisture affected the activities of nitrifying bacteria perhaps through both dehydration and substrate limitation.

Rapid ammonification and nitrification of legume residue-amended samples indicate that the residues had quite sufficient labile pool and/or most of the decomposers and nitrifier communities existed in the active state. In some cases, nitrification also occurred at the end of the incubation period, which implied that the microbial turnover rate may take a longer time. Furthermore, the observation of episodic nitrification in soils exhibited that there could be changes in the composition of nitrifying communities with those that become active at different phases or there could be differences in N pools from organic N sources that were nitrified at different periods of incubation. Some studies have reported net N release in soil after addition of detritus, notably studies of root decomposition (Seastedt et al. 1992; Parton et al. 2007). Based on their own observations and other reports, Hooker and Stark (2008) stated that two important factors regulate whether microbial consumption of OM results in N release (mineralization) or assimilation of additional inorganic N (immobilization); these are (1) the microbial growth per unit substrate consumed (substrate use efficiency [SUE]) and (2) the C/N ratio of substrate consumed. On the other hand, Parton et al. (2007) suggested that the linear release of N from grass roots over time was due to greater availability of soil moisture, labile OM, and nutrients to microbes decomposing roots versus surface leaf litter. However, all of these conditions would be expected to increase microbial SUE (Del Giorgio and Cole 1998; Six et al. 2006), and this would result in greater N immobilization rather than N mineralization (Hooker and Stark 2008). Therefore, the episodic N release and immobilization observed in the present study was most likely related with the substrate C/N ratio, since soluble components of plant detritus may be released during the early stages of decomposition, while plant polymers (such as cellulose) with higher C/N ratios are degraded more slowly during later stages of decomposition (Hooker and Stark 2008). Results from soil laboratory incubations with plant residue amendments suggest that temporal changes in C and N cycling after residue incorporation reflect differences in the C and N stoichiometry of substrates utilized by microorganisms (Trinsoutrot et al. 2000; Jensen et al. 2005; Bruun et al. 2006).

The An_2 control samples showed greater nitrification and net N mineralization than residue-treated ones. Perhaps this indicates preference of the microbial communities towards substrate types, i.e., the microorganisms might choose the freshly applied substrates for C, as soils are mainly C limited. These results in immobilization of N from soil for the stoichiometric balance for biomass build up. The other most likely explanation is that the added residues raised soils C/N ratio that could be accounted for the low rate of nitrification and N mineralization and/or net immobilization of NO_3^- and inorganic N pools. Besides, as a result of residue amendment, soil microenvironment might have become favorable for microbial growth, tending to greater immobilization of N pools.

Mineral N, mainly in the form of NH_4^+–N, is supposed to be the main source for microbial immobilization (Paul and Clark 1989). In the absence of NH_4^+–N, NO_3^-–N has been shown to be a good substrate for microbial assimilation (Recous et al. 1990, 1992). The present study revealed an average of 30 and 80 mg kg^{-1} NH_4^+–N and NO_3^-–N immobilization, respectively (Tables 4 and 5). Our results supported previous observations of high NH_4^+–N immobilization rate, when NH_4^+–N concentration in soils was high, and NO_3^-–N immobilization, when NH_4^+–N concentration is low (http://www.for.gov.bc.ca/hfd/library/documents/bib95675). Microbial growth during decomposition is also

substrate-dependent (Nicolardot et al. 1994; Trinsoutrot et al. 2000). This could be the reason "why" high apparent N immobilization occurred during legume residue decomposition. The present findings also revealed a large amount of NO_3^-–N immobilization together with NH_4^+–N immobilization (Tables 4 and 5), supporting the previous observations. These could be because of the fact that part of the microbial community might have a preference for NO_3^- (Bengtson and Bengtsson 2005) and/or the NH_4^+–N was insufficient to support microbial growth.

Net inorganic N accumulation of studied soils was essentially related with net cumulative nitrification. A similar report with soils of different pH ranges, 5.05 to 8.06, revealed that the nitrification rate was almost equal to the mineralization rate, while NH_4^+–N did not accumulate (Bertrand et al. 2007). In general, the degree of immobilization was relatively higher with HB (C/N ratio, 40.6) than with PP (C/N ratio, 20.4; Table 6), indicating variation of N availability from different organic sources owing to C/N ratio and N contents. The C/N ratio has been identified as a good indicator of N availability among various organic N sources (Aulakh et al. 2000; Trinsoutrot et al. 2000; Rowell et al. 2001). Decomposition rate and N mineralization of organic resources may also be related with lignin/N and polyphenol/N ratios, as they are negatively correlated with mineralization rate (Kumar and Goh 2000). Other resource quality parameters have been highlighted as important modifiers, including tannin, soluble C, and fiber-bound N (Palm and Rowland 1997). However, the importance of anyone (or any combination) of these constituents may vary depending on the process, time frame, or type of plant material involved (Palm 1995; Palm and Rowland 1997). Furthermore, the actual rate and degree of decomposition are also moderated by the local activity of the decomposers and the environmental conditions (Giller and Cadisch 1997; Heal et al. 1997).

Soil microbial N immobilization may play an important role in regulating the soil N retention capacity and prevent nitrogen loss through leaching of NO_3^- (Bengtson and Bengtsson 2005). Singh et al. (1989) showed that soil microbial biomass (SMB) can be a source of plant nutrients in nutrient-poor tropical soils where it acted as a sink of nutrients during the dry period (high biomass, low turnover) and as a source during the monsoon period of plant growth (low biomass, high turnover). Therefore, N flux through SMB has been shown to be sufficient to supply plant N demand (Paul and Voroney 1984). In the present study, positive and marginally significant correlation of C with N mineralization was observed. This confirms the coupling of C and N in most organic compounds and that most soil microorganisms get their energy and substance from organic materials. Hence, N mineralization and transformation is intimately linked to the organic C decomposition (Huang et al. 2004).

Overall analysis of N release potential of control soils depicted that An_2 was substantially high followed by Af, reflecting the superiority of coffee land use over agriculture land use in nutrient supply ascribed to its high litter return to soils (Table 6). In general, our results revealed immobilization of inorganic N pools, both in NH_4^+–N and NO_3^-–N forms. However, we could not be able to trace back the sources of immobilization, whether it is microbial, clay, or OM. Interestingly, a study by Verchot et al. (2001) showed wide ranges of 5–50% of microbial N immobilization in different forest soils. Another study (Bengtson and Bengtsson 2005) revealed that mobilization and remineralization of microbial N is likely to occur only when conditions promote high growth rates, i.e., when microorganisms are not substrate limited and when temperature and moisture conditions are favorable.

Rate of N mineralization/immobilization

The present study showed that the rate of N mineralization was higher at pF3.9 than at pF2.5 or the rate of immobilization was low at pF3.9 than at pF2.5 irrespective of the soil types, implying that drought effect could have led to a dominance of fungi over bacteria which consequently led to the release of high C/N ratio. It is well established that microbe survival varies in response to soil water when taxon is taken into consideration (Harris 1981) and drought may change the composition of the microbial community with fungi being more tolerant of dry conditions than bacteria (Griffin 1972; Hendrix et al. 1986). The study confirms the differential sensitivity to water availability of the microorganisms responsible for the soil N transformation processes (Yahdjian and Sala 2008). Under low soil moisture condition, it is presumed that less N assimilation per mole C assimilation and/or less aggressive N immobilization during decomposition of litter with high C/N ratio and more net N mineralization per gram C during decomposition of litter with low C/N ratio. The results strongly support the C/N stoichiometry as altered by drought stress. This confirms that the N assimilation/mineralization during decomposition depends both on the C/N ratio of the substrates as well as the C/N ratio of active microorganisms. The observation also has implication for C/N biogeochemical modeling in tropical agroecosystems.

Conclusion

Relative rate of CO_2–C flux (amended/non-amended) was greater at pF3.9 than at pF2.5 when legume residues were amended, suggesting that easily mineralizable substrates could ameliorate the effect of soil moisture stress on microbial activity. The results revealed that, in one of the

three soils (An_1), the decomposition of the recalcitrant fraction was more inhibited by drought stress than the easily degradable fraction, suggesting further studies of moisture stress and litter quality interactions. Elevated N cycling (mineralization/immobilization) persisted throughout the study period in residue-treated soil samples compared to control. Nitrification was enhanced at higher soil moisture contents in contrast to ammonification. Therefore, there should be a tradeoff for these two important N transformation processes in soils.

Strikingly, both NH_4^+–N and NO_3^-–N were immobilized with all soil types at both soil moisture contents. These findings question the general belief of ammonium preference of microbes over nitrate. Greater N immobilization in residue-treated An_2 soil was observed, which showed greater microbial growth (high SUE) compared to other soil samples. Legume residues can be potential N sources for the low-input farming systems prevailing in tropical ecosystems. By contrast, immobilization of inorganic N pool with the application of legume residues with high C/N could be a good strategy to mitigate NO_3^- loss and thereby reduce environmental pollution, besides offering high C sequestration as revealed by relatively low CO_2–C loss. Therefore, the immobilization of N could also be a good approach for synchronizing the N nutrient release and crop N requirement through adjusting the C/N ratio (by mixing residues with different qualities) and by utilizing microbes as an ephemeral nutrient pool during the early crop growth period. The observation has implication for C/N biogeochemical modeling in tropical agroecosystems.

Acknowledgments The authors duly acknowledge the Norwegian Programme for Development, Research and Higher Education (NUFU) for financing this research work through the legume–rhizobia collaborative project with Hawassa University. The assistance in the laboratory analyses of plant and soil samples rendered by Mrs. Grete Bloch, Irene E. E. Dahl, and Caren Aldler is gratefully acknowledged. We would like to thank Dr. Dhyan Singh and Dr. Wakene Negassa for their help in reviewing the early draft of the manuscript.

References

Agehara S, Warncke DD (2005) Soil moisture and temperature effects on nitrogen release from organic N sources. Soil Sci Soc Am J 69:1844–1855

Aulakh MS, Khera TS, Doran JW (2000) Mineralization and denitrification in upland, nearly saturated and flooded subtropical soil. II. Effect of organic manures varying in N content and C:N ratio. Biol Fertil Soils 31:168–174

Bengtson P, Bengtsson G (2005) Bacterial immobilization and remineralization of N at different growth rates and N concentrations. FEMS Microbiol Ecol 54:13–19

Bertrand I, Delfosse O, Mary B (2007) Carbon and nitrogen mineralization in acidic, limed and calcareous agricultural soils: apparent and actual effects. Soil Biol Biochem 39:276–288

Beyene D, Kassa S, Ampy F, Asseffa A, Gebremedhin T, Berkum P (2004) Ethiopian soils harbor natural populations of rhizobia that form symbioses with common bean (*Phaseolus vulgaris* L.). J Microbiol 181:129–136

Bray RH, Kurtz LT (1945) Determination of total, organic and available forms of phosphorous in soils. Soil Science 59:39–45

Bremner JM, Mulvaney CS (1982) Total nitrogen. In: Page AL (ed) Methods of soil analysis. Part 2. Agronomy monograph no. 9, 2nd edn. ASA and SSSA, Madison, pp 595–624

Brown AD (1990) Microbial water stress physiology. Wiley, Chichester

Bruun S, Luxhoi J, Magid J, de Neergaard A, Jensen LS (2006) A nitrogen mineralization model based on relationships for gross mineralization and immobilization. Soil Biol Biochem 38:2712–2721

Cornejo FH, Varela A, Wright SJ (1994) Tropical forest litter decomposition under seasonal drought: nutrient release, fungi and bacteria. Oikos 70:183–190

Csonka LN (1989) Physiological and genetic responses of bacteria to osmotic stress. Microbiol Rev 52:121–147

Del Giorgio PA, Cole JJ (1998) Bacterial growth efficiency in natural aquatic systems. Annu Rev Ecol Syst 29:503–541

EPA (1979) Methods for chemical analysis of water and waste waters. Method 353.2

Ford DJ, CooksonWR AMA, Grierson PF (2007) Role of soil drying in nitrogen mineralization and microbial community function in semi-arid grasslands of north-west Australia. Soil Biol Biochem 39:1557–1569

Giblin AE, Nadelhoffer KJ, Shaver GR, Laundre JA, McKerrow AJ (1991) Biogeochemical diversity along a riverside toposequence in Arctic Alaska. Ecol Monogr 61:415–435

Giller KE, Cadisch G (1997) Driven by nature: a sense of arrival or departure. In: Cadisch G, Giller KE (eds) Driven by nature: plant litter quality and decomposition. CAB International, Wallingford, pp 393–399

Griffin DM (1972) Ecology of soil fungi. Chapman and Hall, London

Griffin DM (1981) Water potential as a selective factoring the microbial ecology of soils. In: Elliot LF (ed) Water potential relations in soil microbiology. SSSA special publication no. 9. SSSA, Madison, pp 141–151

Harris RF (1981) Effect of water potential on microbial growth and activity. In: Parr JF, Gardner WR, Elliot LR (eds) Water potential relations in soil microbiology. Soil Science Society of America, Madison, pp 23–95

Heal OW, Anderson JM, Swift MJ (1997) Plant litter quality and decomposition: an historical overview. In: Cadisch G, Giller KE (eds) Driven by nature: plant litter quality and decomposition. CAB International, Wallingford, pp 3–30

Hendrix PJ, Parmelee RW, Crossley DA, Coleman DC, Odum EP, Goffman PM (1986) Detritus food webs in conventional and no-tillage agroecosystems. Bio science 36:374–380

Hooker TD, Stark JM (2008) Soil C and N cycling in three semiarid vegetation types: response to an in situ pulse of plant detritus. Soil Biol Biochem 40:2678–2685

Huang Y, Zou J, Zheng X, Wang Y, Xu X (2004) Nitrous oxide emissions as influenced by amendment of plant residues with different C:N ratios. Soil Biol Biochem 36:973–981

Jensen LS, Salo T, Palmason F, Breland TA, Henriksen TM, Stenberg B, Pedersen A, Lundstrom C, Esala M (2005) Influence of biochemical quality on C and N mineralization from a broad variety of plant materials in soil. Plant Soil 273:307–326

Khalil MI, Hossain MB, Schmidhalter U (2005) Carbon and nitrogen mineralization in different upland soils of the subtropics treated with organic materials. Soil Biol Biochem 37:1507–1518

Killhma K, Amato A, Ladd JN (1993) Effect of substrate location in soil and soil pore-water regime on carbon turnover. Soil Biol Biochem 25:57–62

Kumar K, Goh KM (2000) Nitrogen release from crop residues and organic amendments as affected by biochemical composition. Commun Soil Sci Plant Anal 34:2441–2460

Lal R (2004) Soil carbon sequestration impacts on global climate change and food security. Science 304:1623–1629

Linn DM, Doran JW (1984) Effect of water-filled pore space on carbon dioxide and nitrous oxide production in tilled and non-tilled soils. Soil Sci Soc Am J 48:1267–1272

Lomander A, Kätterer T, Andrén O (1998) Carbon dioxide evolution from top- and subsoil as affected by moisture and constant and fluctuating temperature. Soil Biol Biochem 30:2017–2022

Lou Y, Ren L, Li Z, Zhang T, Inubushi K (2007) Effect of rice residues on carbon dioxide and nitrous oxide emissions from a paddy soil of Subtropical China. J Water Air Soil Poll 178:157–168

Molstad L, Dörsch P, Bakken LR (2007) Robotized incubation systems for monitoring gases (O_2, NO, N_2O, N_2) in denitrifying cultures. J Microbiol Method 71:202–211

Morita RY (1997) In bacteria in oligotrophic environments. In: Reddy CA, Chakrabarty AM, Demain AL, Tiedje JM (eds) Starvation–survival lifestyle. Chapman & Hall, New York, pp 368–385

Mosier AR, Kroeze C (1998) A new approach to estimate emissions of nitrous oxide from agriculture and its implications for the global N_2O budget. IGBP Newsletter 34:8–13, http://www.igbp.kva.se/cgi-bin/php/frameset.php

Mulumba LN (2004) Land use effects on soil quality and productivity in the Lake Victoria basin of Uganda. Ph.D. dissertation, Graduate School of the Ohio State University, pp 166

Nelson DW, Sommers LE (1982) Total carbon, organic carbon and organic matter. In: Page AL (ed) Methods of soil analysis. Part 2. Agronomy monograph no. 9, 2nd edn. ASA and SSSA, Madison, pp 539–580

Nicolardot B, Fauvet G, Cheneby D (1994) Carbon and nitrogen cycling through soil microbial biomass at various temperatures. Soil Biol Biochem 26:253–261

NSF (1975) Water analysis: determination of ammonia-nitrogen by Norwegian Standard NS4746

Nyberg G, Ekblad A, Buresh R, Högberg P (2002) Short-term patterns of carbon and nitrogen mineralization in a fallow field amended with green manures from agroforestry trees. Biol Fertil Soils 36:18–25

Palm CA (1995) Contribution of agroforestry trees to nutrient requirements of intercropped plants. Agroforst Syst 30:105–124

Palm CA, Rowland AP (1997) A minimum dataset for characterization of plant quality for decomposition. In: Cadisch G, Giller KE (eds) Driven by nature: plant litter quality and decomposition. CAB International, Wallingford, pp 379–392

Palm CA, Sanchez PA (1990) Decomposition and nutrient release patterns of the leaves of three tropical legumes as affected by their lignin polyphenolic contents. Soil Biol Biochem 22:330–338

Palm CA, Gachengo CN, Delve RJ, Cadisch G, Giller KE (2001) Organic inputs for soil fertility management in tropical agroecosystems: application of an organic resource database. Agric Ecosyst Environ 83:27–42

Parton W, Silver WL, Burke IC, Grassens L, Harmon ME, Currie WS, King JY, Adair EC, Brandt LA, Hart SC, Fasth B (2007) Global-scale similarities in nitrogen release patterns during long-term decomposition. Science 315:361–364

Paul EA, Clark FE (1989) Soil microbiology and biochemistry. Academic, London, p 273

Paul EA, Clark FE (1996) Soil microbiology and biochemistry, 2nd edn. Academic, New York

Paul EA, Voroney RP (1984) Field interpretations of microbial biomass and activity measurements. In: Klug MJ, Reddy CA (eds) Current perspectives in microbial ecology. American Society for Microbiology, Washington, pp 509–514

Paul KI, Black AS, Conyers MK (2001) Effect of plant residue return on the development of surface soil pH gradients. Biol Fertil Soils 33:75–82

Recous S, Mary B, Faurie G (1990) Microbial immobilization of ammonium and nitrate in cultivated soils. Soil Biol Biochem 22:913–922

Recous S, Machet JM, Mary B (1992) The partitioning of fertilizer-N between soil and crop: comparison of ammonium and nitrate applications. Plant Soil 144:101–111

Rochette P, Angers DA, Cote D (2000) Soil carbon and nitrogen dynamics following applications of pig slurry for the 19th consecutive years: I. Carbon dioxide fluxes and microbial biomass carbon. Soil Sci Soc Am J 64:1389–1395

Rowell DM, Prescott CE, Preston CM (2001) Decomposition and nitrogen mineralization from biosolids and other organic materials: relationship with initial chemistry. J Environ Qual 30:1401–1410

Sakala GM, Rowell DL, Pilbeam CJ (2004) Acid–base reactions between an acidic soil and plant residues. Geoderma 123:219–232

Schjønning P, Thomsen IK, Moldrup P, Christensen BT (2003) Linking soil microbial activity to water- and air-phase contents and diffusivities. Soil Sci Soc Am J 67:156–165

Seastedt TR, Parton WJ, Ojima DS (1992) Mass loss and nitrogen dynamics of decaying litter of grasslands: the apparent low nitrogen immobilization potential of root detritus. Can J Bot 70:384–391

Shepherd KD, Palm CA, Gachengo CN, Vanlauwe B (2003) Rapid characterization of organic resource quality for soil and livestock management in tropical agroecosystems using near-infrared spectroscopy. Agron J 95:1314–1322

Singh JS, Kashyap AK (2007) Variations in soil N-mineralization and nitrification in seasonally dry tropical forest and savanna ecosystems in Vindhyan region. India Trop Ecol 48:27–35

Singh JS, Raghubanshi AS, Singh RS, Srivastava SC (1989) Microbial biomass acts as a source of plant nutrients in dry tropical forest and savanna. Nature 338:449–500

Six J, Frey SD, Thiet RK, Batten KM (2006) Bacterial and fungal contributions to carbon sequestration in agroecosystems. Soil Sci Soc Am J 70:555–569

Stark C, Condron LM, Stewart A, Di HJ, O'Callaghan M (2006) Effects of past and current management practices on crop yield and nitrogen leaching—a comparison of organic and conventional cropping systems. New Zeal J Crop Hort 34:207–216

Strak JM, Firestone MK (1995) Mechanisms for soil moisture effects on activity on nitrifying bacteria. Appl Environ Microbiol 61:218–228

Teklay T, Nordgren A, Nyberg G, Malmer A (2007) Carbon mineralization of leaves from four Ethiopian agroforestry species under laboratory and field conditions. Appl Soil Ecol 35:193–202

Thomsen IK, Petersen BM, Bruun S, Jensen LS, Christensen BT (2008) Estimating soil C loss potentials from the C to N ratio. Soil Biol Biochem 40:849–852

Tian G, Kang BT, Brussaard L (1992) Biological effect of plant residues with contrasting chemical compositions under humid tropical conditions—decompositions and nutrient release. Soil Biol Biochem 24:1051–1060

Trinsoutrot IS, Recous B, Bentz M, Lineres D, Nicolardot B (2000) Biochemical quality of crop residues and carbon and nitrogen mineralization kinetics under non-limiting nitrogen conditions. Soil Sci Soc Am J 64:918–926

Verchot LV, Holmes Z, Mulon L, Groffman PM, Lovett GM (2001) Gross vs net rates of N mineralization and nitrification as indicators of functional differences between forest types. Soil Biol Biochem 33:1889–1901

Vinten AJA, Whitmore AP, Bloem J, Howard R, Wright F (2002) Factors affecting N immobilization/mineralization kinetics for cellulose-, glucose- and straw amended sandy soils. Biol Fertil Soils 36:190–199

Wagener SM, Schimel JP (1998) Stratification of soil ecological processes: a study of the birch forest floor in the Alaskan taiga. Oikos 81:63–74

Wolde-meskel E, Terefework Z, Frostegård Å, Lindström K (2005) Genetic diversity and phylogeny of rhizobia isolated from agroforestry legume species in southern Ethiopia. Int J Syst Evol Microbiol 55:1439–1452

Xu JM, Tang C, Chen ZL (2006) Chemical composition controls residue decomposition in soils differing in initial pH. Soil Biol Biochem 38:544–552

Yahdjian L, Sala OE (2008) Do litter decomposition and nitrogen mineralization show the same trend in the response to dry and wet years in the Patagonian steppe? J Arid Environ 72:687–695