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Nitrogen release dynamics of *Erythrina abyssinica* and *Erythrina brucei* litters as influenced by their biochemical composition

Abebe Abay

Central Ethiopian Environment and Forest Research Centre, P. O. Box 31037, Addis Ababa, Ethiopia.

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Litter mineralization is a crucial process in providing nutrients through decomposition to plants, which also depends in the biochemical composition of the litter and soil properties as well. Decomposition rate of *Erythrina abyssinica and Erythrina brucei* in Luvisol was investigated in relation to their nutrient release dynamics such as NH_4^+-N and NO_3^--N in relation to their initial concentrations of lignin, acid detergent fiber, cellulose and total polyphenol content and their ratios. The dynamic was followed in an incubation pot experiment, complete randomized design in replication. *E. abyssinica* has an average of 4.05, 9.7 and 2.04% TN, lignin and total polyphenol content respectively. *E. brucei* has also an average of 3.05, 12.63 and 1.05% content of TN, lignin and total polyphenol respectively. The samples of *E. abyssinica* and *E. brucei* were ground and incorporated with Luvisol in pots. To determine the amount of ammonium and nitrate released each treatment and control were sampled and analyzed on weekly basses. The lignin and total polyphenol was significantly positively correlated with the release of NH_4^+-N, while the NO_3^--N showed significant negative correlations with the release of ammonium. From the experiment, it was observed that the *E. abyssinica* with lower content of lignin and high in TN has released the nutrients faster whereas *E. brucei* with high lignin and low total polyphenol content released slowly. In general, these leguminous trees released NH_4^+-N and NO_3^--N easily because of their high total nitrogen content and low lignin, ADF, cellulose and total polyphenol content. They attained their half-life within 2-3 weeks. Therefore, *E. abyssinica* and *E. brucei* bears fast mineralization as a result they can be used for fast-term correction of crop nutrient demand. However, more detailed researches are needed to synchronize and verify laboratory results with field measurements of their effect on crop production and synchronization of soil nutrient availability and crop demand in different agro ecology and soil types.

Key words: Incubation, lignin, luvisol, total polyphenol, nitrate, ammonium.

INTRODUCTION

Incorporation of agroforestry legumes trees to the soil have been identified as important management practice to increase soil fertility. Moreover, the influences of litter quality in determining decomposition rate have been

E-mail: abebeabayt@yahoo.com. Tel: +25191146 7175.

recognized in the tropics since agricultural practice commenced (Yamoah et al., 1986; Swarup, 1987). Plant litter mineralization is biological decomposition of litter by microorganisms and transfer of organic and mineral compounds to the soil (Loranger et al., 2002). Studies on Ethiopian soil and forest revealed that the very wide range of climate, topography, parent material and microorganisms' and the local conditions of the specific areas have assisted in development of different soil types as well as over 6,600 higher plant species including indigenous leguminous and non-leguminous trees (Mesfin, 1998; MoA, 2000; Mohammed, 2003; Bekele, 2007). According to MoA (2000), Luvisol is one of the soil types estimated to cover 64,063.5 km^2 or 5.8% of the country. In the future agricultural system, application of organic farming systems is important in order to lessen impacts of climate change, decrease soil erosion (Reganold et al., 1987), improve biodiversity (Hole et al., 2005), and enhance soil fertility (Watson et al., 2002).

Organic farming incorporates researched outputs with ecology and cultural agricultural practices based on naturally occurring microbial processes (Shi, 2013). Among the potentially valuable plant species used for organic farming, *Erythrina abyssinica* and *Erythrina brucei,* are leguminous trees with immense use. *E. abyssinica* fixes nitrogen in the roots infected by Rhizobia nodulate (Legesse, 2002) whereas *E. brucei* fixes nitrogen in the leaves with leaf symbiotic N-fixing characteristics (Legesse, 2002; Orwa et al., 2009). The application of these organic nutrient resources can also be directly incorporation in to the soil and allow it to decompose and release nutrients. However, litter decomposition rates are also governed by the initial biochemical composition as lignin, carbon, total nitrogen, cellulose, polyphenol and phosphorus, potassium concentrations and ratios of carbon/nitrogen (C/N), lignin/N, lignocellulose /N, lignin: N.

One of the major decomposition retarding chemical composition of litter is lignin. It represents nearly 30% of the carbon sequestered in plant materials annually (Boerjan et al., 2003). Lignin is a complex organic polymer which is used for structural materials in the support tissues of vascular plants and some algae as well as it is important in the formation of cell walls (Martone et al., 2009; Lebo et al., 2001). The second mineralization retarding chemical constituents are phenolic compounds. These compounds constituting up to 60% of plant dry mass (Cates and Rhoades, 1997). These consist of more than one aromatic ring, bearing one or more hydroxyl functional groups. Once integrated into the soil, it controls below-ground processes, including soil organic matter decomposition and nutrient cycling (Rovira and Vallejo, 2002*;* Toberman et al., 2010). As outlined by Zhang et al. (2008) the litter chemicals compositions and their ratio are strong predictors of litter decay, which accounts for over 73% of the variation in litter decomposition rates. At large, the reset parameters that can affect the litter degradation depends on environmental factors, season and soil types and properties, such as soil pH, temperature, soil moisture, oxygen content, bulk density and particle size.

In order to manage and predict the nutrients released from organic residues for crop uptake there is need to understand the N mineralization dynamics of the litter in relation to their major biochemical composition under specific soil type. Leaf litter of different plant species has diverse nutrient release patterns, which are related to quality, season, and environmental factors (Arunachalam et al., 2003; Abiven et al., 2005).

In order to comprehend the effects of litter quality on N release and how the mineralization rate is affected by biochemical composition of *E. abyssinica* and *E. bruce*i, in this study litter decomposition were studied though an incubation experiment. The experiment was designed to determine the amount of NH_4^+ and NO_3^- release in Luvisol and correlate with biochemical constituents of both litter types. Moreover, the study investigates the influence of difference in geographical location on the biochemical composition (N, lignin and polyphenol) of the present leguminous tree species.

MATERIALS AND METHODS

Description of the Sampling Sites

Sidama and Wolaita zones are found in southern Ethiopian. Sidama covers 6972.1 km^2 and lies between 6°14' to 7° 18' N and 37° 92' to 39°19' E. It has an elevation ranging from 502 to 3000 m.a.s.l. The annual mean temperature of the zone ranges between 10.5 to 27.1°C and the annual mean rainfall ranges from 801 to 1600 mm (HMD, 2015). Wolaita covers an area of 4471.3 km^2. The zone lies on an elevation ranging from 1200 to 2950 m.a.s.l. with annual average temperature of 15.0 to 28.0°C. The area has a bimodal rainfall pattern, with an average annual rainfall of 1300 to 2000 mm distributed over 8 to 9 months (HMD, 2015).

Leaf and soil sampling

Green leaves samples of *E. abyssinica* and *E. brucei* were collected from ten different randomly selected but geo-referenced locations of Sidama and Wolaita zones of southern Ethiopia biased on soil colour (Tables 1 and 2). A composited Dystric Luvisol sample 0-20 cm in depth was collected from Sidama.

Selected soil physical analysis

Hydrometer method was used for the soil particle size analysis (Bouyoucos, 1951). The bulk density of the sample was determined from sample collected using core ring sampler. The sample was dried at 105° for 24 h, then weighed and calculated accordingly. The plant available soil water holding capacity was determined after determining the field capacity (FC) and permanent wilting point (PWP) of the Luvisol as described by Hillel 1980).

Selected soil chemical analyses

Soil pH and electrical conductivity were measured using an extract

Table 1. *E. abyssinica* sampling sites.

Site	N	E	masl
Aleta Wondo	06° 35'48.2''	38° 25'36.6''	1941
Aleta Wondo	06° 34'53.2''	38° 26'39.3''	2234
Aleta Wondo	06° 34' 56.7''	38° 23'53.7 ''	2483
Titecha	06° 33' 28.2''	38° 31'28.5 ''	2686
Hula	06° 29'26.1''	038° 30'45.3''	2767

Table 2. *E. brucei* sampling sites.

Site	N	E	m.a.s.l
Delbo Atwero	06° 54' 34.2''	37°49' 04.0''	2236
Doga	06° 58' 26.0''	37°52' 25.7''	1975
Gacheno	07° 02' 37.7''	37°55' 33.0''	1884
Kokote	06° 52' 37.7''	37° 35' 33.0''	2154
Shone	07° 09' 34.3''	37° 57' 25.5''	1996

Figure 1. Partial view of the incubation experiment set up in greenhouse.

1:2.5 (soil: water) as described by Reeuwijk, (2002). Soil organic carbon content and available P were determined using Walkley and Black (1934) chromic acid wet oxidation method and Olsen and Sommer (1982) based on alkaline extraction by 0.5N $NaHCO_3$ methods respectively. The total N content in the soil was determined according to Reeuwijk, (2002). Mineral N content (NH_4^+ and NO_3^-) was extracted at a ratio of 1:4 (soil: 2*M* KCl) and determined according to Keeney and Nelson (1982).

Determination of lignin and cellulose via acid detergent fiber (ADF)

Klasson method referenced in Browning (1967) was selected for the analysis of lignin and cellulose via ADF. ADF is prepared from organic nutrient sources material by boiling with sulphuric acid solution of cetyltrimethyl ammonium bromide (CTAB) under controlled condition. The CTAB dissolves nearly all nitrogenous constituents, and the acid hydrolyses the starch to residue containing lignin, cellulose and ash as outlined in Clancy and Wilson (1966). The 72% H_2SO_4 destroys cellulose; lignin is then determined upon ashing by weight-loss.

Incubation of soil samples

From the composite soil sample two hundred gram for each pot was weighed and mixed with 0.127 g and 0.169 g of *E. abyssinica* and *E. brucei* respectively followed by fully homogenizing in complete random design in replication (Figure 1). Then watering to field capacity was made every day or two until the end of the experiment. On weekly basses, analyses were done to determine, the amount of ammonium and nitrate released from each treatment and control. The analysis were conducted in soil chemistry laboratory using the standard methods.

Statistical

The data obtained from the laboratory analyses of litters and

Table 3. Selected soil chemical and physical characteristics of Luvisol.

Depth cm	$pH\text{-}H_2O$ 1:2.5	Av.P mg kg^{-1}	TN %	OC	C/N	BD Mg m^{-3}	FC V/V %	PWP	Sand %	Clay	Silt	Textural class
0-20	4.98	5.32	0.16	1.76	11	1.23	46.20	31.55	14	32	54	Clay

Table 4. Major chemical constituents of *E. abyssinica* and *E. brucei*.

Plant type	TN (%)	K (%)	P(%)
E. abyssinica	4.05^{a} (3.16-5.16)	2.02^{b} (1.94-2.08)	0.39^{a} (0.36-0.43)
E. brucei	3.36^{b} (2.70-3.93)	2.61^{a} (2.54-2.68)	0.31^{b} (0.30-0.32)
LSD (0.05)	0.70	0.36	0.05
CV (%)	13.01	6.32	8.01

*Note: Means in a column followed by the same superscript letters are not significantly different.
**Values in brackets shows range.

Table 5. *E. abyssinica's* sampling sites and their total nitrogen content.

Site	Altitude (m.a.s.l)	***E. abyssinica*** **TN (%)**
Aleta Wondo	1941	5.13^{a} (5.16-5.08)
Aleta Wondo	2234	3.26^{d} (3.16-3.31)
Aleta Wondo	2483	4.21^{b} (3.85-4.31)
Titecha	2686	3.67^{c} (3.62-3.70)
Hula	2767	3.98^{b} (3.85-4.08)
LSD (0.05)		0.29
CV (%)		3.97

*Note: Means in a column followed by the same superscript letters are not significantly different.
**Values in brackets shows range

mineralization were subjected to analysis of variance (ANOVA) using statistical analysis software version 9.3 (SAS Institute, 2003). The least significant difference (LSD) was worked to separate means at $p \leq 0.05$ using Duncan Multiple Range Test. To measure release of nutrients (Ammonium and Nitrate) in soil, simple correlation analysis (at $p \leq 0.05$) was carried out.

RESULTS AND DISCUSSION

Phsico-chemical property of Soil used for mineralization

The soil particle size analysis of experiment soil sample was found to be clayey. The critical bulk density value for agricultural use according to Hillel (1980) is 1.4 g cm^{-3}, implying that there is no excessive compaction and restriction to root development. The bulk density of the Luvisol was 1.23 g cm^{-3}. As described by Werner (1997) the soils in this range possesses good porosity for aerobic microorganisms' activities. According to Landon (1996) rating the OC and TN contents of the soil fall in the "very low" and "low" categorized respectively. The soil is also rated as "low" in available P according to Havlin et al. (2010) rating. Since the soil is at low pH, it possesses phosphorus fixation as well (Table 3).

Litter quality rating of *E. abyssinica* and *E. brucei with respect to altitude*

In this study the analyses of variance for *E. abyssinica and E. brucei* showed significant variation in TN content with altitude and sampling site. Sangiga and Woomer (2009) indicated organic nutrients sources with higher than 2.5% TN content are considered high quality. Thus, the average TN content of *E. abyssinica* (4.05%) and *E. brucei* (3.36%) are categorized with the highest N_2 fixing plants (Table 4). The highest mean value in TN content for *E. abyssinica* was obtained at 1941 m.a.s.l (5.16%) and the lowest mean value (3.26%) was found at 2234 m.a.s.l (Table 5). Whereas the highest mean value of TN content (3.93%) of *E. brucei* was obtained at 2154 m.a.s.l

Table 6. *E. brucei's* sampling sites and their total nitrogen content.

Site	Altitude (m.a.s.l)	***E. brucei*** **TN(%)**
Delbo Atwero	2236	3.85[a] (3.77-3.93)
Doga	1975	3.38[b] (3.31-3.47)
Gacheno	1884	2.85[c] (2.70-3.00)
Kokate	2154	3.93[a] (3.91-3.92)
Shone	1996	2.77[c] (2.70-2.85)
LSD (0.05)		0.17
CV (%)		2.71

*Note: Means in a column followed by the same superscript letters are not significantly different.
**Values in brackets shows range

Table 7. Chemical characterization of *E. abyssinica* and *E. brucei*.

Litter	TPP	ADF	Lignin	Cellulose	TN	C:N	ADF : N	L:N	TPP:N
				(%)					
E. abyssinica	2.04[a]	30.57[b]	9.70[b]	1.10[b]	4.15[a]	10.44	12.07	2.34	0.49
E. brucei	1.05[b]	34.37[a]	12.63[a]	1.80[a]	3.35[b]	12.26	10.25	3.77	1.24
CV (%)	0.83	1.83	1.77	12.53	1.50				
LSD(0.05)	0.017	1.17	0.34	0.29	0.10				

Where: L–Lignin, TPP- Total Polyphenol, Cellulose– C, TN- Total Nitrogen, ADF- Acid Detergent Fiber.

and the lowest mean value (2.77%) at 1996 (Table 6). As shown on Tables 5 and 6 the variation in TN content of *E. brucei* and *E. abyssinica* can be due to cumulative contribution of micro agroclimatic factors: soil fertility, temperature, and microorganism, in addition to the differences in altitude. The existence of different soil orders might have contributed to the difference in TN content.

E. abyssinica is much better nitrogen fixers as compared to the common organic nutrient sources as referred by Mugendi et al. (2011) which are *Leuceana diversifolia* 3.9%, *Crotalaria juncea* 3.9%, *Vicia benghalensis* 3.7%, *Mucuna pruriens* 4.0%, *Calliandra calothyrsus 3*.4%, *Crotalaria ochroleuca* 4.5%.

E. abyssinica and *E. brucei* had diverse TN content and the data obtained in this study showed that no direct or inverse relationship was found between total nitrogen content and altitude.There was significant difference in TP content between EB and EA. The results revealed that the percentage TP content in EA was higher and significantly different from EB. This difference may be due to the P extracting ability of EA. Both of the organic sources were significantly different with respect to K content (P < 0.05) too (Table 4).

Total polyphenol content

The highest total polyphenol (TPP) content was found in *E. abyssinica* (2.04%) and the lowest in *E. brucei* (1.06%) (Table 7). The study conducted by Palm and Sanchez (1990) and Anis et al. (2012) also indicated that the primary factors that determines the rate of mineralization and activity of decomposers are the litter quality parameters, especially N, lignin and polyphenol content by inhibiting function of the decomposer organisms by chemically binding to proteins. The study conducted by Grubeˇsi´c et al. (2005) and Srisuda et al. *(*2008) indicated that the TPP content of pigeon pea leaves and leaves of *Pouteria altissima* were 2.31%, 4.55 % respectively. The TPP contents in different parts of *Plantago holosteum* were: leaves 10.15%, stems 4.13 %, and flowers 3.91% which are higher than the present study spps that is, *E. abyssinica* and *E. brucei*. Therefore, based on their low TPP content it can be said that these plants can undergo in fast decomposition rate.

Total lignin content

E. brucei litters contained the highest (12.63%) lignin content whereas *E. abyssinica* had the lowest (9.70%) of both. Leaf quality have generally been described as high-quality materials in terms of high N and low-lignin contents (Sakala et al., 2000), high ADF and (lignin + polyphenol): N ratio. In general, increasing lignin concentration reduces the residue decomposition rate as outlined by Tian et al. (1992). However, many researches

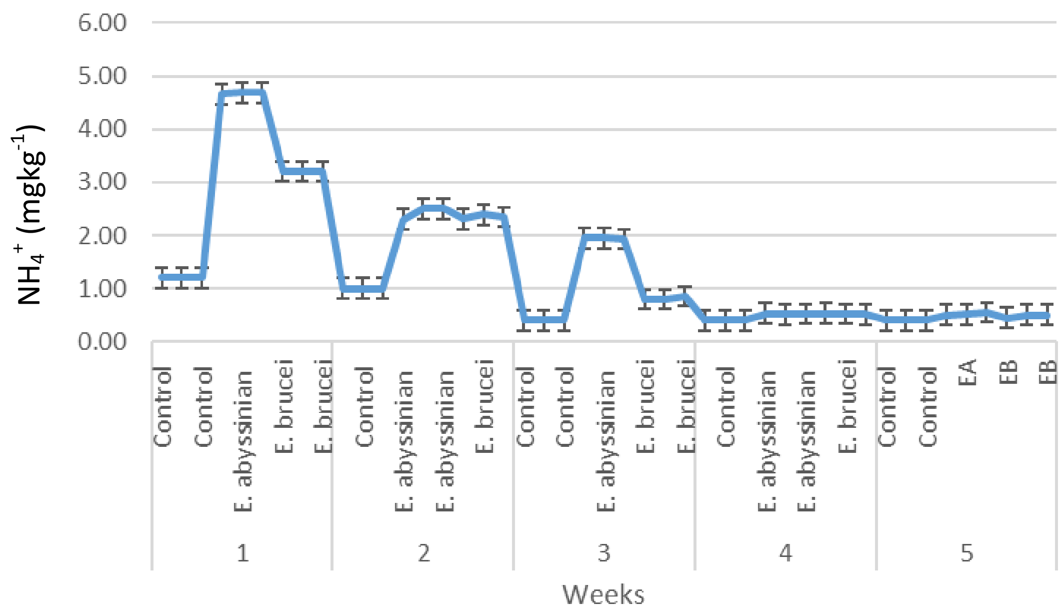

Figure 2. Weekly NH_4^+-N mineralization of *E. abyssinica* and *E. brucei* in Luvisol.

do not agree on Tian generalization's. They had tried to show by conducting different researched evidences. Among these scholars Stump and Binkley (1993) referred to the lower lignin/N ratio as rate mineralization determining step rather than lignin or nitrogen alone. Thus, according to Tian et al. (1992) study the decomposition order of the litters under investigation shall be *E.brucei* first followed by *E. abyssinica.* However according to Stump and Binkley (1993) the order of mineralization will be *E. abyssinica* first followed by *E. brucei.*

Furthermore, others researchers concluded that it was not the only ratio (lignin/N) that regulates mineralization but other ratios also regulate mineralization (such ratios are shown in Table 7). Handayanto et al. (1997) noted that the initial lignin+polyphenol:N ratios was a good predictor strongly for N mineralization rates or N accumulation when dealing with complex materials. Following the conclusions of these authors, the order of mineralization for the spps under investigations shall be *E. abyssinica* followed by *E. brucei*. However, other researchers such as Probert et al. (2004) and Teklay et al. (2007) concluded that initial N concentration or C:N ratio of residues was the main factor controlling decomposition and nutrient release. These ratios are commonly used to determine the quality of the ONS, that is, the higher the ratio the lower the quality. Moreover, other chemical constituents like potassium (Zaharah and Bah, 1999), phosphorus (Goya et al., 2008) of the litter has a great role on the decomposition rate. From this study, we can infer that the high TN content, low TPP and lignin content of *E. abyssinica* and *E. brucei* as compared to other spps helps to undergo through fast mineralization.

Mineral N release from *E. abyssinica* and *E. brucei* on luvisol

NH_4^+-N release

NH_4^+-N is the initial by-product of organic N mineralization. Through the incubation experiment, the highest release NH_4^+-N (4.68 mg kg^{-1}) and the lowest release (0.48 mg kg^{-1}) was recorded from *E. abyssinica* and *E. brucei*, respectively. However, the laboratory results revealed that there was general decreasing trend in the release of NH_4^+-N. During the decomposition process each of the litters had showed significant difference with respect to NH_4^+-N content and the change in NH_4^+-N was significantly influenced by time of incorporation ($r = -0.90784$; $p < 0.0001$). The NH_4^+-N released in Luvisol differed significantly at each sampling period (Figures 2 and Table 8). The levels of accumulated NH_4^+-N released from *E. abyssinica* and *E. brucei* were significantly similar ($r = 0.9374$, $p < 0.5150$) at each week of incubation. The probable reasons for the difference in mineralization of these spps could be the difference in residue quality, total nitrogen content and microbial activity. The results are also in line with that of Schomberg et al. (2009) who stated that the decomposition rate of residues is often influenced by temperature, soil moisture, legume quality parameters such as N, polyphenol, and lignin contents and their ratios regulated by environmental factors. As outlined in Giller and Cadisch (1997) physical accessibility for microbes may also be an important determinant of decay rate, as was observed by increase in microbial activity or decomposition once litter is ground.

Table 8. Interaction effect of Luvisol, *E. abyssinica,* and *E. brucei* and weeks on NH_4^+-N release (mg kg^{-1}).

Week	Control.	*E. abyssinica*	*E. brucei*
1	1.200^{e}	4.680 ^{a}	3.200 ^{b}
2	1.000 ^{f}	2.433 ^{c}	2.350 ^{c}
3	0.400 ^{i}	1.943 ^{d}	0.813 ^{g}
4	0.400 ^{i}	0.520 ^{h}	0.520 ^{h}
5	0.400 ^{i}	0.520 ^{h}	0.483 ^{h}
LSD	(0.05)	0.033	
CV (%)	2.49		

Table 9. Interaction effect of Luvisol, *E. abyssinica, E. brucei* and weeks incubation on NO_3^--N release (mg kg^{-1}).

Week	Control	*E. abyssinica*	*E. brucei*
1	1.100^{f}	1.917^{d}	1.333^{e}
2	0.950^{g}	2.560^{c}	2.447^{c}
3	0.943^{g}	3.050^{a}	2.533^{c}
4	0.950^{g}	3.080^{a}	2.550^{c}
5	0.950^{g}	2.750^{b}	2.467^{c}
LSD (0.05)		0.0284	
CV (%)		1.928	

Note: Means in a column followed by the same superscript letters are not significantly different at p<0.05.

NO_3^- -N release

During the five weeks' greenhouse mineralization experiment on Luvisol, the experiment revealed that the initial NH_4^+-N and NO_3^--N contents were affected by mineralization and nitrification process. The decomposition of *E. abyssinica* and *E. brucei* had similarities in terms of NO_3^--N release pattern and the change in NO_3^--N was influenced by both litter types (r= 0.9243; p < 0.0238), and weeks of incorporation (Table 9).

During incubation period, the nitrification of the control was at the lower rate, compared to the amended soil, where the average release of NO_3^- was in the order of *E. abyssinica* > *E. brucei* (Figure 3). Positive correlation (r = 0.6571, p < 0.0283) was found between period of incubation (weeks) and NO_3^--N release in the soil, indicating that there was good association of weeks (incubation) and release of NO_3^--N. Nitrification was significantly and negatively correlated with ammonification (r = -0.6571, p $\leq$0.784). The decline in the amount of NO_3^--N starting the end of the experiment and latter might be caused by leaching (because of its high solubility) and denitrification. The soil's cation exchange site does not retain the negative charge, NO_3^- -N, but easily lost from the root zone by leaching and inherent microbes potentially, which accelerates the process of denitrification (Rochette et al., 2000). Khalil et al. (2002) had observed the application of organic residues produced mineral N in the form of NO_3^- under neutral and slightly alkaline conditions. This study also showed an increase in NO_3^- was dependent on the nitrogen content of the litters. Thus, the increase in NO_3^--N content was in the order of *E. abyssinica* > *E. brucei* until denitrification commence (Figure 4).

Conclusion

Present results clearly indicate that these species are categorized as the fast decomposing organic nutrient resources with highest TN content regardless of sampling site. *E. abyssinica* and *E. brucei* had diverse TN content and the data obtained in this study showed that no direct or inverse relationship was found between TN content and altitude but other factors could govern the differences in TN content of the organic sources. The faster decomposition and NH_4^+ release performance of *E. abyssinica* in the Luvisol can be accounted for its highest TN content and low-ADF concentrations. The better biochemical quality characteristics of *E. abyssinica* had enhanced the decomposition rate. Incorporating *E. abyssinica* and *E. brucei* to Luvisols showed an increase

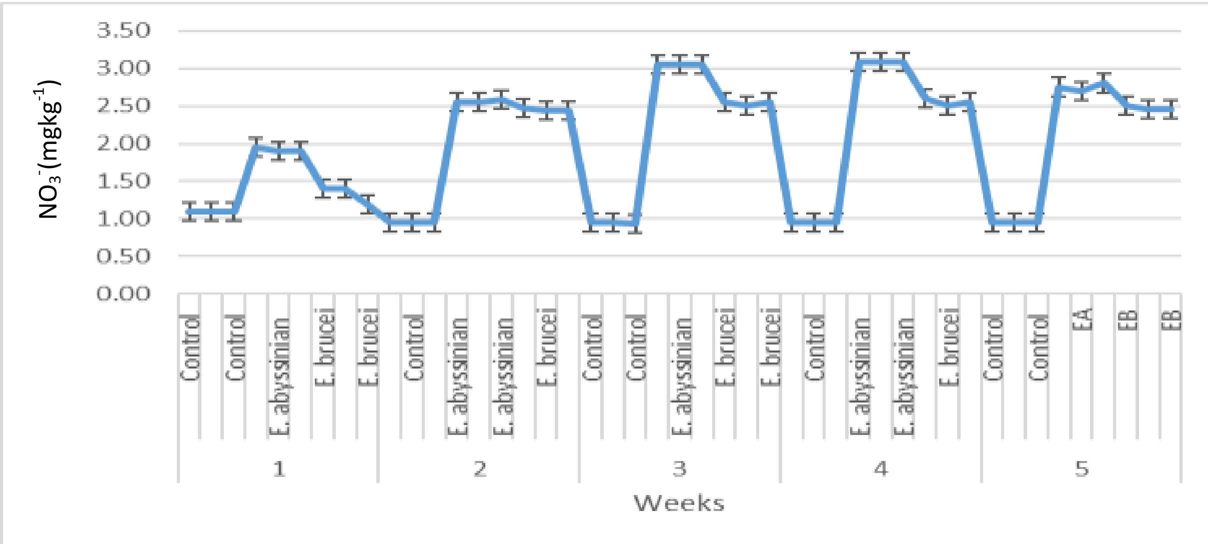

Figure 3. Weekly NO_3^--N mineralization from *E. abyssinica* and *E. brucei* in Luvisol.

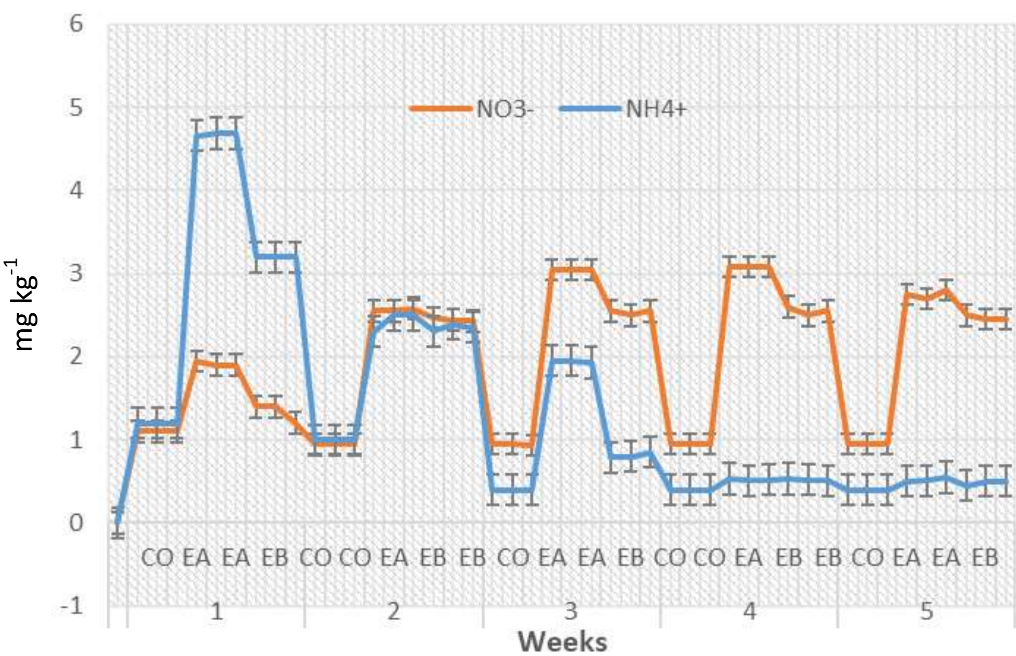

Figure 4. Release of NH_4^+-N and NO_3^--N contents in Luvisol during decomposition.
Error bars represent 1 standard deviation. Main and interaction effects were significant at $p < 0.05$.

in NH_4^+-N and NO_3^--N content of the soil as compared to their respective controls in few weeks. Based on the dynamics of release of NH_4^+-N and NO_3^--N $^-$ content, the species showed the order: *E. abyssinica* > *E. brucei*.

In general, *E. abyssinica* and *E. brucei* amended soil revealed that NO_3^--N release was negatively and significantly correlated with ammonification. During incubation period, the control was at the lower level compared to the amended soil in ammonium and nitrate concentration.

RECOMMENDATION

Researches that are more detailed are needed to synchronize and verify laboratory results with field measurements. Their effect on crop production alongside synchronization of different soil properties with crop demand is also necessary.

CONFLICT OF INTERESTS

The author has not declared any conflict of interests.

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