

# Influence of changes in land use and earthworm activities on carbon and nitrogen dynamics in a steepland ecosystem in Northern Vietnam

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**Abstract** This manuscript focuses on the effects of land-use change and earthworm activities on the dynamics of dissolved nutrients (carbon and mineral nitrogen) in a steep slope ecosystem in Northern Vietnam. We investigated the properties of soil aggregates (casts and surrounding soils) sampled in different agrosystems: a plantation of *Bracharia ruziziensis* (BRA), a fallow (FAL) and a plantation of *Acacia mangium* and *Venicia montana* (FOR), following a cultivation of cassava (CAS), and a fallow after a forest of Eucalyptus (EUC). Soil physical, chemical and biological properties were determined on the sampling date and dissolved organic C (DOC), and mineral N ( $N_{\min}$ ) contents were followed during a 21-day incubation period. CAS, BRA and FOR were characterised by a high rate of N mineralisation, followed by a rapid loss of  $N_{\min}$ . Conversely, FAL and EUC were more interesting in terms of soil fertility because these systems had higher soil  $N_{\min}$  content that could become available to plants. Lost of C through DOC leaching was very low with values ever less than

0.5% of the total soil C content. The greatest lost of C through leaching was in FAL, EUC and FOR. The impacts of earthworms on the soil seemed site-specific. The protection of organic matter in earthworm casts varied with the initial substrate soil and agroecosystem management. Casts were characterised by greater enzymatic activities, except for alkaline phosphatase, than the surrounding soil. While SOM mineralisation was not affected in casts collected in FAL, EUC and FOR, the leaching of  $N_{\min}$  was increased over the surrounding soil. Conversely, mineralisation of SOM and nitrification activity were less in BRA-casts than in the surrounding soil, causing a greater retention of  $N_{\min}$  in soil. While the DOC leached from casts increased in BRA, it was reduced in FAL and was similar in the other sites. Our study indicates that while land-use change occurred only 3 years ago, the extent of leaching was mostly regulated by the type of agroecosystem and by earthworm activities.

**Keywords** Cassava · Land-use change ·  
Dissolved organic carbon · Mineral nitrogen · Earthworms

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## Introduction

Erosion can be regarded as a major type of environmental damage in Southeast Asia with dramatic consequences to soil fertility and water quality (Lal 1998; Magliano and Leslie 2001). Due to rapid human population growth, the cropping areas have expanded to more marginal lands such as mountains, and the fallow periods have been shortened or even abandoned (Clement et al. 2006). In Northern Vietnam, much of the moist forest was lost during the 1970s, and the trend continues (Sharma 1992; Teck

AwBeng 1997; Castella et al. 2006). Forests were cut to expand cultivated cassava, arrowroot, taro, maize and eucalyptus cropping on the uplands. Due to decreases in soil fertility in the uplands, farmers have progressively replaced annual crop cultivation by fallowing, by using it as a common grazing land, or by tree plantations (mainly acacias; Tran Duc et al. 2004; Clement et al. 2006).

Aggregation is of major importance in all soils, controlling porosity, water storage and oxygen diffusion, and therefore the mineralization of soil organic matter (SOM) and the retention or the leaching of mineral nutrients (Brady and Weil 1999; Chaplot et al. 2005). Through their feeding and burrowing activities, earthworms play a key role in soil aggregate processing within the top layers of soil (Lee and Foster 1991; Lavelle et al. 1992; Lavelle and Spain 2001). It has been suggested that earthworms, rather than plant roots, initiate aggregation mainly through the production of casts (Lee and Foster 1991; Marashi and Scullion 2003). Earthworms can contribute greatly to the creation of macro-aggregates that drastically affect soil structural characteristics, such as microbial activity and diversity, SOM sequestration and mineralisation, in temperate regions (Jongmans et al. 2001; Pulleman et al. 2005; Aira et al. 2005; Bossuyt et al. 2005, 2006; Giertz et al. 2005) and tropical ecosystems (Lavelle 1988; Fragoso et al. 1997; Blanchart et al. 1999; Decaëns et al. 1999). It has also been shown that surface casts are potential sources of particulate and dissolved nutrients in surface runoff in temperate ecosystems (Sharpley and Syers 1976; Sharpley et al. 1979; Le Bayon and Binet 1999, 2001, 2006). Surprisingly, the impacts of earthworms on soil erosion and nutrient losses in surface and sub-surface water runoff and the impact of land-use practices on earthworm distribution and activity have been only poorly studied in sloping lands of the tropics, although this geographical area has some of the most active biogeochemical cycling in the world (Koch et al. 1995).

This study was in part of the Vietnamese watershed of the project Management of Soil Erosion Catchment (MSEC) from the International Water Management Institute (IWMI; Maglinao and Leslie 2001; Maglinao et al. 2003), which is examining the effects of land use changes on soil erosion and fertility at small watershed levels, on steep slopes, on a regional scale in five Southeastern Asia countries. Nitrogen has been considered as a limiting factor of grass and crop productivity. The dynamic of N within ecosystems is regulated mainly through biological activities, in contrary to phosphorus that is regulated mainly through physical processes (Bardgett 2005). Thus, it is of primary interest to consider how humans, through land-use management, and earthworms, through their engineering activities (*sensu* Jones et al. 1994), can influence N dynamics in soils. Nitrogenous compounds are subject to

dissolution and transport out of agroecosystems via water runoff in tropical steepland ecosystems, or when they are leached below the crop-rooting zone. As a consequence, leachates contaminated with N compounds can contaminate surface waters and ground water reserves (Wilkinson and Blevins 1999). This study focuses on the dynamic of N in soil aggregates sampled in different Vietnamese agro-systems. Soil physical, chemical and biological properties were determined on the sampling date, and changes in dissolved organic carbon (DOC) and mineral N ( $N_{\min}$ ) contents were followed during a 21-day incubation period.

## Materials and methods

### Study site and soil sampling

The experimental watershed of the MSEC project of Dong Cao (50 ha) is located in the northeast Vietnam, approximately 60 km southwest of Hanoi (20°57'N, 105°29'E). The Dong Cao watershed is surrounded by hills with a general slope of more than 40%, sometimes reaching 100%. The annual rainfall ranges from 1,500–1,800 mm, of which 80–85% of total rainfall is concentrated in the rainy season from April to October. The humidity is always high, between 75 and 80%. The mean daily temperature varies from 15 to 25°C. January and February are the coldest months (Tran Duc et al. 2004).

The dominant soil type in the landscape is an Acrisol (World Reference Base for Soil Resources 1998) or Ultisol (Soil Survey Staff 1999). Soils derived from the weathering of volcano-sedimentary schists of Mesozoic age. Soils are over 1.0 m deep but with marked variation in thickness. They have more than 50% clay content, and are very porous with a bulk density of 1,000 kg.m<sup>-3</sup>. They have a homogenous brown colour 10YR4/4 to 7.5 YR 4/6, and a weak differentiation. The clay is almost exclusively kaolinite with a low CEC; the pH is low, usually below 5.0. Cambisols, Fluvisol and Leptosols are also present on a restricted area (Tran Duc et al. 2004).

Before the 1960s, the region was covered by a dense primary forest. Deforestation and conversion to agricultural land has led to the disappearance of this primary forest. From the mid 1970s until very recently, villagers have cultivated cassava, taro and maize on the uplands. Since 1998, the watershed studied was covered mainly by cassava crop with some area of eucalyptus. From 2002, practices changed very quickly because the soil fertility decreased. Finally, five agrosystems were set up in 2005: (1) a young fallow after the last cassava planting in 2001 (FAL), (2) a plantation of *Acacia mangium* and *Venicia montana* planted in 2002 after a cassava plantation (FOR), (3) a fodder plantation with *Bracharia ruzziensis* planted in 2003 after

cassava (BRA), (4) a very young fallow associated with regrowth of eucalyptus trees, following a forest cut in 2003 (EUC), and (5) a small surface crop of cassava (CAS). These five land uses represent the diversity of managements in this region of Southeast Asia.

The influence of land-use types on the diversity and abundance of earthworms has been considered in another study (Mathieu et al., submitted for publication). The soil in EUC is covered by functional and old earthworm casts that form a typical granular horizon. The macro-porosity and rugosity of soil are important, and infiltration is improved by earthworms. The quantity of casts collected on the surface of the soil in August 2005 has been estimated as approximately 20, 8, 2, 2 and 0 kg.m<sup>-2</sup> respectively in EUC, FOR, BRA, FAL and CAS. *Pheretima leucocirca*, an epi-anecic earthworm species, is responsible for this accumulation of casts on the soil surface (Jouquet, personal observation).

Soil samples were collected randomly in three plots within each land use in January 2005. This period corresponds to the winter season in North Vietnam. Casts were considered as aged and uninfluenced by recent earthworm activity. For each plot, approximately 30 g surface casts produced by *P. leucocirca* and 40–50 g of surrounding soil (0–3 cm depth) with no trace of earthworm activities (considered as control) were sampled and air dried before analyses.

#### Soil incubations

Earthworm casts and control soils (100 g D.W.) were transferred into microcosms made from plastic cylindrical funnels 250 ml deep, covered with a perforated cap to restrict evaporation and to allow air diffusion. Soil samples were watered carefully with distilled water after 0, 1, 3, 7 and 21-day incubation and stored at 25°C.

#### Analyses

Soil samples were analysed at the beginning of the experiment and after the 21-day incubation period. Solutions leached from soil samples were analysed after 1, 3, 7 and 21 days.

Soil pH was determined in 1:2 soil/water suspension and SOM was assessed by total C and N content with an elemental analyser (NA 1500 Series 2, Fisons). Clay particles (<2 µm) were separated by sedimentation (AFNOR, NFX 31107).

Ammonium was determined by the colourimetric Nessler method according to the HACH method (Anonymous 1994). Nitrate was determined by the colourimetric phenoldisulphonic method (Bremner 1965) and DOC contents through the chemical oxygen demand, with the HACH method and reagents (Anonymous 1994).

The bioavailability of C (C available) was assessed by the ratio of mineralised C versus total soil C content. The mineralisation of C was followed by measuring the rate of CO<sub>2</sub> production from 50 g of soil in a closed flask after a 24-h incubation period. CO<sub>2</sub> concentrations were measured with an infrared CO<sub>2</sub> analyser Dimarsol® (IRD).

Enzyme activities were used as a broad-spectrum indicator of soil biological activity (Mora et al. 2005). Enzyme activity measurements were made on samples with 75% of their water holding capacity and incubated at 26°C for 24 h to reactivate the microflora. Acid (pH 4) and alkaline (pH 9) phosphatase activities were determined by the method described by Tabatabai (1982). The α- and β-glucosidase activities were determined according to a method adapted from Hayano (1973).

#### Nitrogen cycling

Mineralisation of N was assessed by measuring of the overall production of  $N_{\min}$  during the 21-day incubation experiment. It was calculated by the following formula:

$$\left[ (N_{\min} \text{ in soil})_{\text{day 21}} + (N_{\min} \text{ leached from soil})_{\text{day 1,3,7,21}} \right] - \left[ (N_{\min} \text{ in soil})_{\text{day 0}} + (N_{\min} \text{ leached from soil})_{\text{day 0}} \right]$$

where  $N_{\min}$  is the sum of N – NH<sub>4</sub><sup>+</sup> and N – NO<sub>3</sub><sup>-</sup> in µg.g<sup>-1</sup> soil.

Nitrification was assessed by the N – NO<sub>3</sub><sup>-</sup>/N – NH<sub>4</sub><sup>+</sup> ratio from  $N_{\min}$  mineralised during the incubation period.

The retention of  $N_{\min}$  in soil was assessed by measuring the proportion of  $N_{\min}$  remaining in soil for comparison with the overall  $N_{\min}$  produced during the incubation experiment.

#### Statistical analyses

Treatments were replicated three times and for each sample. Analyses were done in triplicate.

Before analyses, data were inspected for homogeneity of variance using the Levene's test and log-transformed when required. 'Land use' and 'earthworm' effects on soil properties were tested through an analysis of variance, and differences between means were tested with the LSD test. All statistical calculations were carried out using Statistica 6.0 for Windows™.

A principal component analysis (PCA) was done by R (R Development Core Team 2004) using a matrix of 27 samples: 15 variables. A permutation test was used to test the significance of the groupings suggested by PCA.

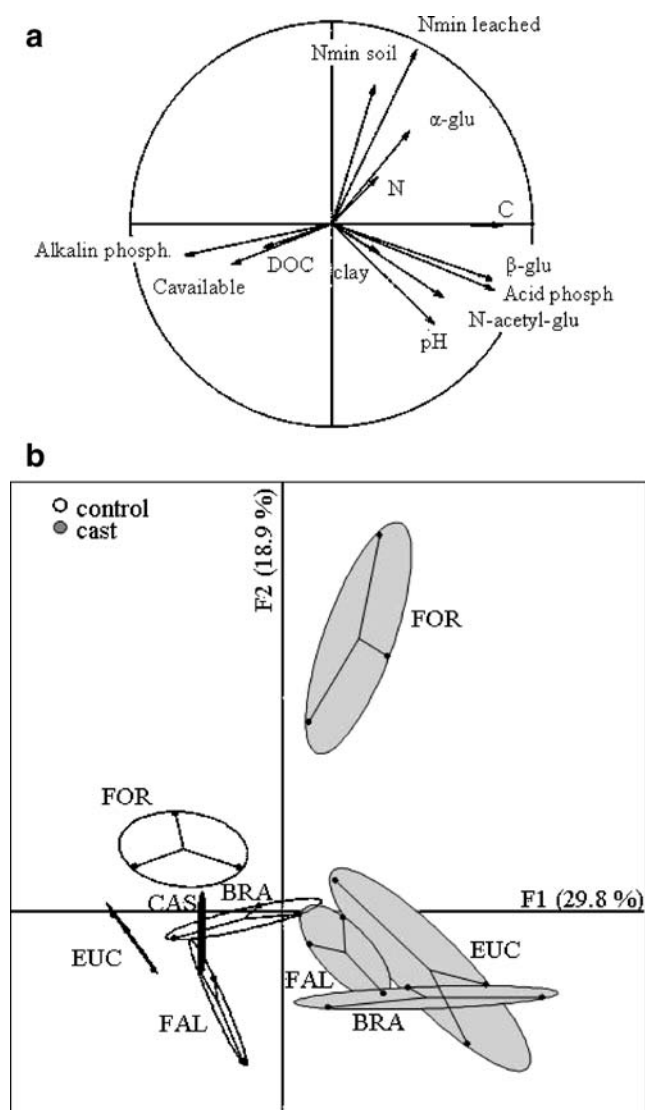
Differences were considered significant only when *P* values were lower than 0.05.

## Results

### Soil quality at the beginning of the experiment

A PCA was made (Fig. 1). Axes 1 and 2, respectively, explained 29.8 and 18.9% of the total variance. The significance test was carried out on 1,000 permutations showed that PCA was highly significant ( $P < 0.001$ ).

Control soil aggregates in BRA and FAL did not differ from those collected in CAS but a small significant difference ( $P < 0.05$ ) occurred with EUC and FOR. This difference can be explained mainly by variations in DOC and  $N_{\min}$  contents (details are given in Table 1). Soils under EUC and FAL were characterised mainly by higher



**Fig. 1** PCA on initial soil aggregate properties: clay (%), C and N (%), pH,  $N_{\min}$  in soil and in leachates ( $\mu\text{g N.g}^{-1}$ ), DOC ( $\mu\text{g C.g}^{-1}$ ), available C (%), acid- and alkaline-phosphatase,  $\alpha$ - and  $\beta$ -glucosidase and N-acetyl-glucosidase activities ( $\text{U.g}^{-1}$ ). **a** Correlation circle. **b** Ordination of the samples in the plane defined by axes 1 and 2 of the PCA

concentrations of leached-DOC than those in soils under CAS, BRA and FOR. The soil from FOR was clearly different from the other sites, mainly because of its higher  $N_{\min}$  concentrations.

Earthworm casts were clearly different in properties from their surrounding soils mainly along the first axis. We note also that the casts had greater C and N contents and less C biodegradability and leached-DOC contents than the surrounding soils (Table 1). The soil in the earthworm casts were also characterised by a higher pH and greater microbial activities, i.e. higher enzymatic activities, except for alkaline phosphatase (details in Table 2). Casts were always enriched in  $N_{\min}$  compared to control soils. These increases were particularly high in casts sampled in FOR (Table 1). Casts collected in EUC and BRA were not discriminated, and their properties were very close to those of casts sampled in FAL (Fig. 1). Casts in FOR differed from the other casts due to more mineral N contents.

### Leaching of DOC

Concentrations of DOC leached from soil aggregates during the incubation experiment (T21-T1) are summarised in Fig. 2. The percentage of C lost through leaching was low with values below 0.5%. The concentrations of DOC leached from soil were significantly lower in CAS and BRA ( $P < 0.05$ ) without any significant difference between them ( $P > 0.05$ ). DOC contents were higher for FAL, EUC and FOR ( $P < 0.05$ ). The soils in these three last land uses have a DOC content ranged from 8 to 15  $\mu\text{g.g}^{-1}$  soil/21 days.

The DOC content leached from the casts was more for BRA than from FOR, EUC and FAL ( $P < 0.05$ ). The concentrations of DOC leached from casts was similar to those from control soils for EUC and FOR ( $P > 0.05$ ) and less for FAL. DOC leaching was correlated negatively with the C content ( $y = 281.39x^{-1.87}$ ,  $r = -0.57$ ) and was not correlated with the fraction of clay ( $r = 0.15$ ).

### Mineralisation of nitrogen

The mineralisation of N is summarised in Fig. 3. No differences occurred between control soils collected in CAS, BRA and FOR ( $P > 0.05$ ). For comparison, mineralisation was reduced in FAL and EUC ( $P < 0.05$ ), and no significant differences occurred between them ( $P > 0.05$ ).

No significant differences in mineralisation occurred between casts and their surrounding soil in FAL and EUC ( $P > 0.05$ ) and FOR, although the variability was very high for the last one. Conversely, the significantly lower concentration of  $N_{\min}$  produced in casts in BRA by comparison with soil ( $P < 0.05$ ) showed that the mineralisation of N was greatly slowed down or decreased in casts.

**Table 1** Characteristics of casts and soil surrounding casts (control): clay (%), C and N (%),  $N_{min}$  contents in soil and leached from soil ( $\mu\text{g N}\cdot\text{g}^{-1}$ ), DOC ( $\mu\text{g C}\cdot\text{g}^{-1}$ ) and C bio-available ( $\mu\text{g C}\cdot\text{h}^{-1}\cdot\text{g}^{-1}$ ; Mean  $\pm$  SE,  $n=3$ )

	Clay	C	N	pH	$N_{min}$ (soil)	$N_{min}$ (leached from soil)	DOC	C available
<b>CAS</b>								
Control	43.9 $\pm$ 1.4	2.45 $\pm$ 0.17	0.21 $\pm$ 0.01	4.5 $\pm$ 0.0	35.4 $\pm$ 2.8	26.9 $\pm$ 7.9	47.0 $\pm$ 6.5	2.07 $\pm$ 0.31
Casts	–	–	–	–	–	–	–	–
<b>BRA</b>								
Control	55.0 $\pm$ 1.3	3.01 $\pm$ 0.14	0.33 $\pm$ 0.06	4.3 $\pm$ 0.1	25.6 $\pm$ 11.8	32.2 $\pm$ 7.5	26.2 $\pm$ 4.1	2.37 $\pm$ 1.56
Casts	53.4 $\pm$ 0.4	4.04 $\pm$ 0.63	0.24 $\pm$ 0.02	5.5 $\pm$ 0.1	46.5 $\pm$ 1.1	54.2 $\pm$ 11.8	15.5 $\pm$ 3.3	2.97 $\pm$ 1.62
<b>FAL</b>								
Control	48.6 $\pm$ 1.7	3.38 $\pm$ 0.22	0.26 $\pm$ 0.02	5.3 $\pm$ 0.1	36.3 $\pm$ 5.6	1.6 $\pm$ 0.1	82.8 $\pm$ 6.5	4.27 $\pm$ 0.91
Casts	50.2 $\pm$ 0.4	4.39 $\pm$ 0.40	0.33 $\pm$ 0.02	5.7 $\pm$ 0.0	51.5 $\pm$ 7.8	26.1 $\pm$ 8.5	17.8 $\pm$ 12.1	1.67 $\pm$ 0.22
<b>EUC</b>								
Control	63.7 $\pm$ 6.5	2.23 $\pm$ 0.05	0.19 $\pm$ 0.01	4.6 $\pm$ 0.0	42.3 $\pm$ 4.4	6.4 $\pm$ 2.9	79.0 $\pm$ 16.0	3.17 $\pm$ 0.64
Casts	68.4 $\pm$ 3.0	4.99 $\pm$ 1.51	0.38 $\pm$ 0.16	5.0 $\pm$ 0.0	31.4 $\pm$ 11.4	53.5 $\pm$ 0.9	9.7 $\pm$ 5.3	1.63 $\pm$ 0.24
<b>FOR</b>								
Control	49.3 $\pm$ 9.2	3.00 $\pm$ 0.44	0.25 $\pm$ 0.06	4.1 $\pm$ 0.1	36.7 $\pm$ 1.2	44.2 $\pm$ 21.6	49.8 $\pm$ 6.9	3.63 $\pm$ 2.64
Casts	53.8 $\pm$ 6.6	4.09 $\pm$ 0.30	0.32 $\pm$ 0.02	4.5 $\pm$ 0.0	69.8 $\pm$ 13.9	241.0 $\pm$ 23.5	21.1 $\pm$ 10.9	1.60 $\pm$ 0.13

Figure 4 summarises the ratio of  $\text{N} - \text{NO}_3^- / \text{N} - \text{NH}_4^+$  in overall  $N_{min}$  content. Nitrogen was mainly in the form of  $\text{NO}_3^-$  for BRA, CAS and FOR; the highest value occurring in BRA and the lowest in FOR ( $P < 0.05$  in all cases). The  $N_{min}$  was mainly  $\text{NH}_4^+$  in FAL and EUC, with no significant differences between them ( $P > 0.05$ ). The ratio  $\text{N} - \text{NO}_3^- / \text{N} - \text{NH}_4^+$  was greater in casts than in soil in FAL, EUC and FOR ( $P < 0.05$  for the three cases) but lower in BRA ( $P < 0.05$ ).

Retention of  $N_{min}$  in soil

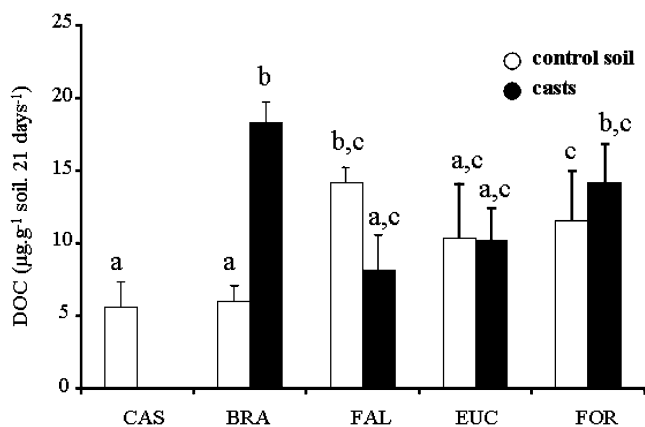
The concentration of  $N_{min}$  remaining in soil after the 21-day incubation period was significantly lower in soil under CAS and BRA (mean value:  $N_{min} = 10.39$  and  $7.9$ , SE  $3.0$  and

$1.5$ , respectively;  $P < 0.05$ ), with no significant differences between them ( $P > 0.05$ ), than in the other treatments (mean value:  $N_{min} = 61.6$ ,  $45.5$  and  $28$ , SE  $12.0$ ,  $7.3$  and  $2.3$ , respectively in FAL, EUC and FOR).  $N_{min}$  content was greater in soils than in casts (mean value:  $N_{min} = 12.9$ ,  $8.9$  and  $13$ , SE  $1.9$ ,  $2.7$  and  $2.2$ , respectively in FAL, EUC and FOR casts;  $P < 0.05$ ), except under BRA land use where the  $N_{min}$  concentration in cast was particularly high (mean value:  $N_{min} = 45.6$ , SE  $16.4$ ;  $P > 0.05$ ).

The proportion of  $N_{min}$  that remains in soil is summarised in Fig. 5. About 20 and 50%  $N_{min}$ , respectively, were retained in the soils under EUC and FAL, although almost all  $N_{min}$  was lost in soil under CAS, BRA and FOR ( $P > 0.05$ ). The retention of  $N_{min}$  in casts was very variable. The retention of  $N_{min}$  was greater in casts under BRA than in the

**Table 2** Enzymatic activity ( $\text{U}\cdot\text{g}^{-1}$ ) in casts and soil surrounding casts (control; Mean  $\pm$  SE,  $n=3$ )

	Acid-phosphatase	Alkaline-phosphatase	$\beta$ -glucosidase	$\alpha$ -glucosidase	<i>N</i> -acetyl-glucosamine
<b>CAS</b>					
Control	7.12 $\pm$ 1.10	0.90 $\pm$ 0.15	0.48 $\pm$ 0.11	0.28 $\pm$ 0.06	0.87 $\pm$ 0.20
Casts	–	–	–	–	–
<b>BRA</b>					
Control	6.87 $\pm$ 2.07	1.40 $\pm$ 0.24	1.31 $\pm$ 0.60	0.47 $\pm$ 0.06	0.66 $\pm$ 0.12
Casts	11.56 $\pm$ 1.96	0.75 $\pm$ 0.09	3.41 $\pm$ 1.13	0.53 $\pm$ 0.28	1.06 $\pm$ 0.23
<b>FAL</b>					
Control	9.86 $\pm$ 2.32	0.73 $\pm$ 0.10	0.84 $\pm$ 0.31	0.30 $\pm$ 0.10	0.55 $\pm$ 0.10
Casts	11.86 $\pm$ 2.43	1.44 $\pm$ 0.04	1.18 $\pm$ 0.23	0.33 $\pm$ 0.07	0.55 $\pm$ 0.10
<b>EUC</b>					
Control	5.25 $\pm$ 0.78	1.67 $\pm$ 0.16	0.58 $\pm$ 0.13	0.43 $\pm$ 0.01	0.46 $\pm$ 0.16
Casts	12.58 $\pm$ 2.08	0.73 $\pm$ 0.17	1.32 $\pm$ 0.84	0.39 $\pm$ 0.11	0.75 $\pm$ 0.20
<b>FOR</b>					
Control	6.05 $\pm$ 0.52	0.62 $\pm$ 0.22	0.73 $\pm$ 0.12	0.21 $\pm$ 0.10	0.45 $\pm$ 0.19
Casts	7.84 $\pm$ 2.28	0.75 $\pm$ 0.14	1.05 $\pm$ 0.06	0.79 $\pm$ 0.53	0.53 $\pm$ 0.22



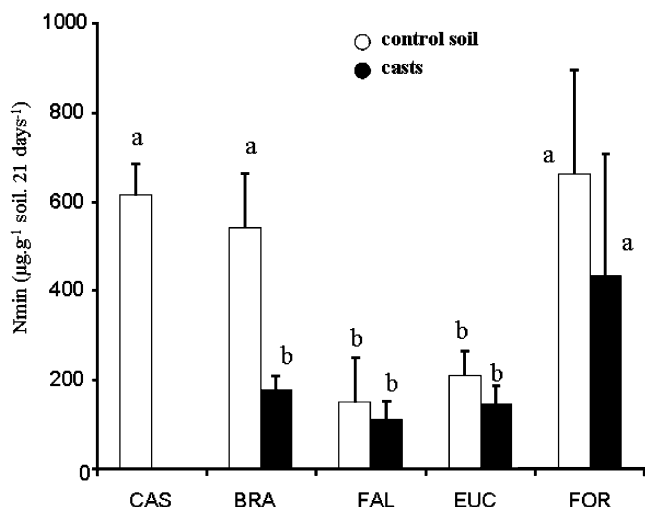
**Fig. 2** DOC ( $\mu\text{g C.g}^{-1}$  soil) leached from soil during the 21-day incubation period ( $n=3$ ; bars indicate standard errors)

soil ( $P<0.05$ ), but the reverse occurred under FAL and EUC ( $P<0.05$ ). The ability of casts to retain  $N_{\text{min}}$  under FOR was not significantly different from the control soil ( $P>0.05$ ).

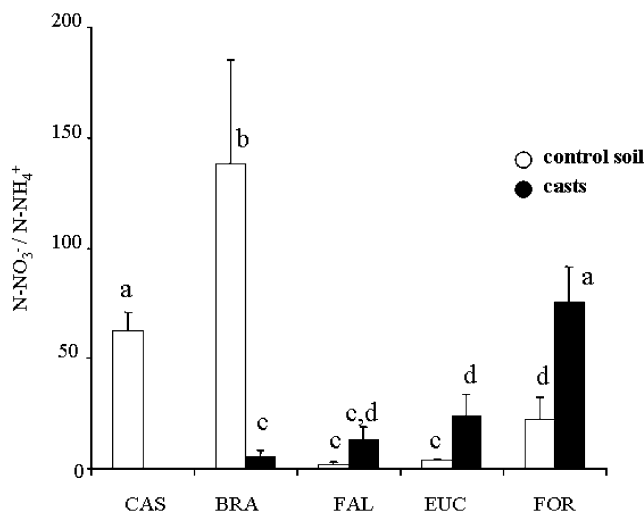
### Discussion

#### Land use changes and soil properties

From the measurements of N contents and N availability in soils, it is obvious that land use impacted microbiological activities and chemical properties of soils after only 3 years. Land-use practices affected the dynamic of C and N in soil significantly. DOC and  $N_{\text{min}}$  are transient pools, usually considered as limiting factors for the growth and activity of soil microorganisms and plants (Bardgett 2005). While  $N_{\text{min}}$  concentrations were similar in soil samples collected in the field, the incubation experiment showed that the mineralisation of N was faster under CAS, BRA and FOR than under the two fallow systems (FAL and EUC). It



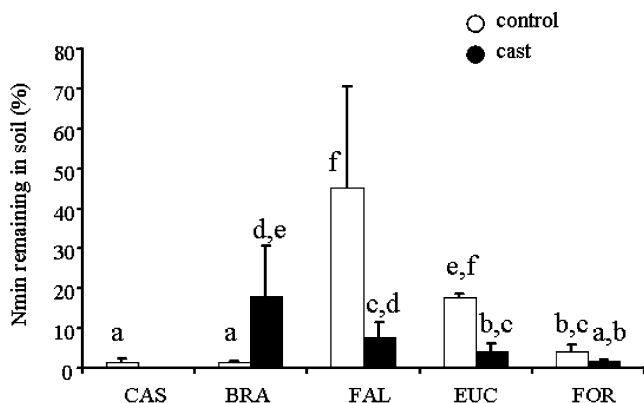
**Fig. 3**  $N_{\text{min}}$  content ( $\mu\text{g N.g}^{-1}$  soil) mineralised during the 21-day incubation period ( $n=3$ ; bars indicate standard errors)



**Fig. 4** Ratio  $N-NO_3^-/N-NH_4^+$  from values collected during the 21-day incubation period ( $n=3$ ; bars indicate standard errors)

seems that  $N_{\text{min}}$  was lost mainly through leaching (less than 1% retained in soil) under CAS, BRA and FOR. In the field, nutrients loss could have been high during the rainy season, and  $N_{\text{min}}$  would then be non-available for plants. Conversely, although mineralisation was decreased in FAL and EUC, the retention and concentration of  $N_{\text{min}}$  were greater. This higher retention of  $N_{\text{min}}$  might be explained by the lower nitrification rates (i.e. lower  $N-NO_3^-/N-NH_4^+$  ratio) in FAL and EUC. These two kinds of fallow and FOR also had the highest rates of loss of DOC through leaching. Stabilisation of dissolved organic C by sorption depends on the type and quantity of the clay minerals, as well as the chemical composition of the nutrients (Baldock and Skjemstad 2000). The negative correlations between the leaching of DOC and soil C content suggest that the type and quantity of plant litter available to microorganisms regulate DOC leaching.

The extents of leaching, nutrient loss and retention were clearly regulated by the type of agricultural practices with



**Fig. 5** Proportion of  $N_{\text{min}}$  (%) remaining in soil aggregates at the end of the incubation experiment in control soils and in casts ( $n=3$ ; bars indicate standard errors)

large consequences on cultural yields (Shuster et al. 2002). Three types of practices were differentiated: CAS and BRA were characterised by a rapid SOM mineralisation, with a loss of  $N_{\min}$  but also less leaching of DOC. Conversely, FAL and EUC were more interesting in terms of soil fertility because these systems had more  $N_{\min}$  that could become available to plants. However, these practices were also characterised by a faster loss of C that might be latter mineralised or incorporated in sediments. Indeed, the C contents had decreased only in the soil under EUC in comparison with those in soils under CAS while the highest level of C content was in the soil under FAL. FOR had both higher losses of  $N_{\min}$  and DOC.

### Effects of earthworms

The importance of earthworms as a major factor of soil fertility was established by the pioneering work of Darwin (1881). Since that time, the influence of earthworms on nutrient cycling and hydrologic processes has received much attention (Lavelle and Spain 2001). However, the retention or loss of dissolved forms of nutrient by earthworms, with implication for nutrient management, has never been studied in steep-slope tropical ecosystems where leaching of soluble nutrients could drastically affect the fertility of soils (Norton et al. 1998).

The overall effect of earthworms on C mineralisation depends on the temporal scale as C mineralisation is stimulated initially but reduced later (Lavelle et al. 1998; Decaëns et al. 1999; McInerney and Bolger 2000). Wolters and Joergensen (1992) speculated that the decreased microbial biomass and increased specific microbial activity commonly observed in fresh earthworm casts were due to the increases in the relative importance of bacteria and actinomycetes in response to labile organic substrates in fresh casts. As casts age, labile substrates are exhausted, and fungi may become relatively more important, resulting in increased microbial immobilisation of C and N and a lower overall specific metabolic activity. We considered casts to be old when they were collected during the dry season (January 2005) and produced during the previous rainy season (May to September). The higher acid phosphatase activity in casts than in the parent material, confirmed that microbial activity was linked mainly to fungi, probably due to less bioavailability of SOM (Bardgett 2005).

Many studies have examined the controversial impacts of earthworms on C and N fluxes in soils and on plant growth (Blair et al. 1997; Bohlen et al. 1997; Bouché et al. 1997; Lavelle et al. 1997; Brown et al. 1999; Whalen and Janzen 2002). Such papers have suggested that earthworm casts have higher contents of  $N_{\min}$  than the bulk soil due to a greater microbial activity and a greater retention of labile N (e.g. Lavelle et al. 1992; Buck et al. 1999; Blair et al.

1997; Marhan and Scheu 2005). However, some experiments have shown that earthworms increase N loading in leachates and the loss of N from colloids (Decaëns et al. 1999; Shuster et al. 2002; Marhan and Scheu 2005). We showed that the influence of earthworms on N and C cycling depended on agricultural practices and on the history of casts. In our laboratory experiment, casts sampled in the field were initially enriched in  $N_{\min}$  and with a less DOC-leaching. However, the incubation of casts for 21 days showed that the quantity of N mineralised from SOM was similar in FAL, EUC and FOR or even decreased in BRA. In addition, our study also demonstrated that leaching of  $N_{\min}$  from casts was greater in FAL and EUC but less in BRA and was not different in FOR. The greater retention of  $N_{\min}$  in BRA might be explained by decreasing nitrification activity. Thus, while  $N_{\min}$  contents in FAL, EUC and FOR were less in casts than in soil at the end of the experiment, the concentrations of  $N_{\min}$  were greater in BRA-casts. The reverse occurred for DOC with a more leaching of DOC from casts in BRA, a less leaching in FAL and no significant leaching in EUC and FOR. Thus, land-use practices were of primary importance in the regulation of the effect of earthworms in the loss and retention of dissolved nutrients. Except in BRA, the loss of  $N_{\min}$  for plants was probably increased from casts during high runoff intensity events that occur during the rainy season.

The higher C and N content, lower bioavailability of C, lower amounts of DOC leached from soil and less or similar  $N_{\min}$  production suggest that SOM was protected against decomposition and mineralisation in casts. There is an evidence from other studies that organic matter in earthworm casts may be protected from decomposition (Scheu and Wolters 1991; Edwards et al. 1995; Decaëns et al. 1999) due to enclosure in stable aggregates (Shipitalo and Protz 1988; Marinissen and Dexter 1990; McInerney et al. 2001; Bossuyt et al. 2005, 2006), and that this stabilisation would depend on the quality of litter fed on by earthworms (Buck et al. 1999; Marhan and Scheu 2005). Consequently, casts may be considered as microsites of short-term mineral N production and medium-term SOM accumulation with net effects varying with initial substrate soil and agroecosystem management.

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