TREE SPECIES AND MACROFAUNA INFLUENCE ON SOIL AGGREGATES AND CARBON IN EASTERN DEMOCRATIC REPUBLIC OF CONGO †

[INFLUENCIA DE LAS ESPECIES DE ÁRBOLES Y LA MACROFAUNA EN LOS AGREGADOS DEL SUELO Y EL CARBONO EN EL ESTE DE LA REPÚBLICA DEMOCRÁTICA DEL CONGO]

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SUMMARY

Background. Soil aggregates represent a major indicator of soil fertility as they are important components of soil C protection. However, in the Democratic Republic of Congo, the choice of tree species being integrated into crop farms influence the type of soil macrofauna beneath these agroforestry trees with effect on soil aggregates and carbon fractions. **Objective**. To assess the combined effect of diverse tree species alongside earthworms and termites on soil aggregation and soil organic C. **Methodology**. Soil samples were collected from five agroforestry systems comprising *Eucalyptus saligna*, *Grevillea robusta* and *Ficus benghalensis* (an indigenous tree) woodlots, an agricultural farm (beans) and a natural forest in four different locations. Soil aggregates and soil organic C were analysed in the laboratory following standard procedures. The analyses of variance and correlations were carried out using R programming software. **Results**. A significantly higher proportion of large macroaggregates was recorded in soil under natural forest (13.1 %) whereas microaggregates were significantly low in soils under natural forest. For all the categories of aggregate-associated C, natural forest recorded the highest values while beans had the lowest values. The same trends were observed for labile C with 22.9 g kg^{-1} in soil under natural forest and 9.7 g kg^{-1} in soil under beans. **Implication**. The high levels of soil aggregates as well as carbon fractions recovered from natural forest clearly show the significance of a well-diversified tree-based farm. **Conclusion**. Therefore, it is advisable to consider a large diversification of tree species on farms to sustain and maintain a better soil quality.
Key words: soil aggregates; soil carbon; termites; earthworms; *Eucalyptus saligna*; *Grevillea robusta*.

RESUMEN

Antecedentes. Los agregados del suelo representan un indicador importante de la fertilidad del suelo, ya que son componentes importantes de la protección del C del suelo. Sin embargo, en la República Democrática del Congo, la elección de las especies de árboles que se integran en las granjas de cultivos influye en el tipo de macrofauna debajo de estos árboles agroforestales y, por lo tanto, en los agregados del suelo y las fracciones de carbono. **Objetivo**. Evaluar el efecto combinado de diversas especies de árboles junto con lombrices y termitas sobre la agregación del suelo y el C orgánico del suelo. **Metodología**. Se tomaron muestras de suelo de cinco sistemas agroforestales que comprenden bosques de *Eucalyptus saligna*, bosques de *Grevillea robusta* y bosques de *Ficus benghalensis* (un árbol autóctono), una finca agrícola (frijoles) y un bosque natural en cuatro lugares diferentes. Los agregados del suelo y el C orgánico del suelo se analizaron en laboratorio siguiendo procedimientos estándar. Los análisis de varianza y correlaciones se realizaron mediante el software de programación R. **Resultados**. Una proporción significativamente mayor de macroagregados grandes se registró en suelos bajo bosque natural (13.1%), mientras que los microagregados fueron significativamente bajos en suelos bajo bosque natural. Para todas las categorías de C agregado asociado, el bosque natural registró los valores más altos mientras que los valores más bajos se encontraron bajo el frijol. Se observaron las mismas tendencias para el C lábil con 22.9 g kg^{-1} en suelo bajo bosque natural y 9.7 g kg^{-1} en suelo bajo frijol. **Implicación**. Los grandes niveles de agregados del suelo, así como de fracciones de carbono recuperadas de los bosques naturales, muestran claramente la importancia de una explotación agrícola bien diversificada.

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Conclusión. Por lo tanto, es aconsejable considerar una gran diversificación de especies de árboles en las granjas para sustentar y mantener una mejor calidad del suelo.
Palabras clave: agregados del suelo; carbono del suelo; termitas; lombrices de tierra; *Eucalipto saligna*; *Grevillea robusta*.

INTRODUCTION

Sustainable maintenance of soil quality on a long term has always been a worldwide challenge in agricultural ecosystems (Powlson *et al.*, 2011). Soil aggregates and soil organic carbon (SOC) have been identified as key components in determining soil quality such that deterioration of soil structure, among other factors, results in soil degradation (Mahesh *et al.*, 2017b). Soil aggregates stability represents a significant indicator of soil fertility, and may influence agronomic productivity (Bronick and Lal, 2005). Stable soil aggregates maintain soil fertility by playing a key role in soil processes such as carbon sequestration (Dhaliwal *et al.*, 2017). In addition, soil aggregates improve soil infiltration reducing surface water run-off, thus preserving the integrity of soil surface as well as improving root development (Franzluebbers *et al.*, 2000; Lawal, 2013). Furthermore, the carbon (C) content present in tree litter and crop residues has been documented to be sequestered within soil aggregates after their decomposition (Unger, 1997; Zeng *et al.*, 2010). Therefore, soil aggregates and soil organic C are intimately related. In fact, soil organic matter (SOM) is acknowledged to be important in the process of soil structure formation and stabilization as it stimulates soil aggregation through binding of soil mineral constituents and in turn, soil aggregates offer protection to SOM from decomposer microorganisms (Dhaliwal *et al.*, 2017; Elliott and Coleman, 1988). However, different types of land use or land cover impact differently on soil structure which may lead to changes in both soil aggregation and soil carbon content (Conant *et al.*, 2004; Six *et al.*, 2002). For instance, integrating trees into agricultural crop farms has been shown to modify soil differently when compared to farms with few or no tree cover. This is due to the fact that soils beneath trees receive improved SOM input from litter fall and root turnover processes which leads to improved soil aggregation and carbon storage (Barrios *et al.*, 2011; Kamau *et al.*, 2020). Likewise, it is important to acknowledge the role played by the soil biological component (soil biota) in the dynamics of SOC and the aggregation process. Soil biota rely on the soil C content for energy, while their activities contribute to modification of the soil structure (Coleman *et al.*, 2004). Soil macrofauna such as termites and earthworms, for example, can ingest significant quantity of organic matter and integrate them into the soil through their excretions thus offering physical protection to SOM against microbial degradation. Hence, due to their significant churning of the soil, these groups of macrofauna are said to be ecosystem engineers (Ayuke *et al.*, 2011; Six *et al.*, 2004).

A particular pattern of trees in farms can determine a specific configuration of soil aggregates and C content in a given environment. For instance, comparing the influence of diverse land-use systems on soil aggregates, Dhaliwal *et al.* (2017) found the lowest (5.8%) values of large macroaggregates in soils under the maize-wheat system and the highest (67.3%) in soils under agroforestry system (comprised of *Populus deltoids* trees intercropped with *Sorghum bicolor* during summer and with wheat during winter), whereas the values of soil micro-aggregates were highest in maize-wheat systems (69.4%) compared to those in agri-horticulture and agroforestry systems (45.2% and 21.4% respectively). Dhaliwal *et al.* (2017) results are consistent with those obtained by Morlue *et al.* (2021). On the other hand, SOC is said to be more stable and preserved in systems producing more and diversified organic material (Anderson and Domsch, 1989). For example, Yusheng *et al.* (2009) observed higher labile C in soils under natural forest in comparison to the homogeneous tree species farms in China.

In the Democratic Republic of Congo, farmers intercrop trees with annual crops. In North Kivu province Geert *et al.* (2013) have documented that *Grevillea robusta* and *Eucalyptus saligna* are the most grown trees on farms, given their beneficial role in charcoal and fire wood production, provision of timber, and integration with crop production to improve agricultural productivity (Geert *et al.*, 2013; He *et al.*, 2015; Nabunya, 2017). In the region, few studies have been carried out regarding the influence of tree species on soil macrofauna and soil chemical properties (Kataka *et al.*, 2023), and tree species ability to stock carbon (Katembo, 2017). This study therefore evaluated the influence of common agroforestry trees (*Grevillea robusta*, *Eucalyptus saligna* and ficus sp.) and earthworms and termites, on soil aggregates and soil C. Natural forest and beans farms were used as undisturbed and highly disturbed reference ecosystems, respectively.

MATERIALS AND METHODS

Description of the study site

This study was carried out in North Kivu province, Democratic Republic of the Congo. The North Kivu province has a surface area of 60,000 km^2 and is situated between latitude 0°58’ North and 02°03’

South and longitude 27°14’ and 29°58’ East. The Eastern part borders Rwanda and Uganda (South-East), the North-west by the Ituri province, the South-West by the Maniema province, and the South by the South-Kivu province (Muvunga, 2019). North Kivu province lies at an altitude ranging from 800 m to 2,500 m a.s.l. Temperatures vary from 23 *°C* in lower altitudes (less than 1000 m) to about 15 *°C* in higher altitudes (2000 m), with rainfall varying from 1000 to 2000 mm per year. Following the INEAC classification proposed by Sys *et al.* (1961), the soils range from Kaolisols to Ferralsols while others are alluvial soils or Lithosols and Xerokaolisols (Vyakuno, 2006). Agriculture in North Kivu is largely subsistence and employs over 80% of the working population which is mostly from rural and poor regions which cultivate marginal soils. The fields are generally vulnerable to erosion, due to lack of control measures. Slash-and-burn with little or no fallow period is a dominant practice, which has led totremendous decline in soil fertility thus giving poor yields and increasing the pressure on natural forest reserves (Emilie *et al.*, 2015; Emilie and Subira, 2015). Farmers have adopted Taungya system where crops are grown during the first three years of tree plantation, after which the trees are left alone to grow in the fields up to five years before harvesting for timber and then restart the cycle. Within farms, trees are planted in rows at a spacing varying from two to three meters according to the farmers' will. Study farms were selected randomly from four different locations near Butembo city. They are Lukanga, Musienene, Malende and Bunyuka (Figure 1).

Selection of tree species and experimental design

The selection of tree species for the study was guided by extensive research on documentated literature and information and in situ observations. *Eucalyptus saligna* and *Grevillea robusta* were selected for this study because they are the most grown trees on farms in the study region. In addition, three control farms were also considered: one negative control consisting of a piece of agricultural land cropped with beans and with no tree species influence, and two positive controls, one set as a natural forest (constituted with ten main tree species: *Syzigium guineense, Macaranga neomildbraediana, Bridelia ferruginea, Rapanea melanophloeus, Bellucia axinanthera, Piptadeniastrum africanum, Sephonia globeliflolia, Musanga cecropioides, Miriantus arboreus and Polisia vulvae*) and another cropped with an indigenous tree species, *Ficus benghalensis*.

The criteria for selecting the tree species farms were: (1) the size of farms: any selected farm had a minimum area of 0.5 ha, since 0.5 ha is the common size of woodlots owned by farmers in the study region; (2) the tree attributes: all trees within the different farm replicates had comparable characteristics in terms of shape, height, and age of their plantation; (3) the dominance: samples were collected only in farms having woodlots of the tree species of interest. The experimental layout was a complete randomized design. The five different land cover constituting different plant species as described in table 1 represented the different treatments for this study. At each location, five farms for each treatment were identified for sample collection. The different tree species represented the controlled factors and each farm within a site represented the replications.

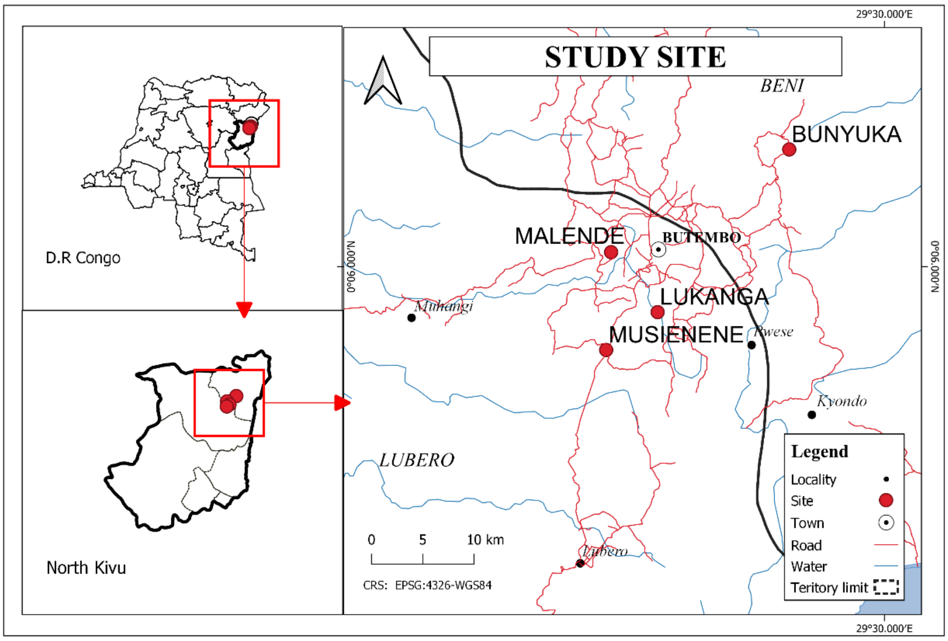

Figure 1. Study site map (Kataka *et al.*, 2023).

Table 1. Description of fields sampled.

	Tree Fields sampled				
Tree species	*Eucalyptus saligna*	*Grevilea robusta*	*Ficus benghalensis*	Natural forest	Beans
Description	Farm purely cropped with *Eucalyptus saligna*	Farm purely cropped with *Grevilea robusta*	Indigenous species (Ficus) within farms	No agricultural activities	No trees. Beans based system

Collection of earthworms and termites

Earthworms and termites were collected using soil monoliths, pitfall traps and Winkler litter extraction methods. Soil monoliths measuring 0.25 x 0.25 x 0.30 m were dug out in accordance with the Tropical Soil Biology and Fertility Program method (Anderson and Ingram, 1993). In each plot, following the method used by Mboukou-Kimbatsa and Bernhard-Reversat (2001), two transects were selected randomly. Along each transect, three monoliths were excavated making a total of six monoliths per plot. After the excavation, the soil was placed in buckets and then transferred little by little to a sorting tray for easy hand picking of soil organisms visible with naked eyes. All the soil macrofauna recovered were first conserved in 75% ethanol before being separated at the end of the collection activity. Following the method used by Catherine and Steven (2001), five pitfall traps were set and implanted into the ground to the soil surface level along a randomly chosen transect at a 5 m interval. Pitfall traps were made of a plastic cup with dimensions of 9 cm in depth and 5.5 cm in diameter and partially filled with ethanol and remained on farms for 7 days, with collection being done after every 24 hours. In the same transects, leaf litter was collected at 5 different sampling points using a quadrat of 0.25 m^2 for obtaining Winkler samples (Michael *et al.*, 2017). The whole 0.25 m^2 leaf litter was put in a bag sieve and sifted through a 10 mm mesh by vigorously shaking the bag sifter for one minute. After sieving, the obtained samples were moved into Winkler bags and were retained for 3 days for macrofauna collection (Hopp *et al.*, 2011). All the organisms were identified and grouped according to their respective taxonomic groups, and their abundance was determined as the number of individuals per square meter.

Determination of water stable aggregates

The sampling points for soil to be used in aggregate determination were located adjacent to the points where macrofauna sampling was done. The soil samples were broken along the natural plane of weakness and air-dried, after which the soil aggregates were extracted following wet sieving process as documented by Elliott (1986). Soil samples were dried and passed through a 5 mm sieve, after which 100 g of soil was soaked within the water by quick immersion. The wet samples were sieved using 2 mm, 0.25 mm, and 0.053 mm sieves. The aggregates were then classified into four classes of water-stable aggregates: large macroaggregates (> 2 mm "LM"), small macroaggregates (0.25-2 mm "SM"), microaggregates (0.053-0.25 mm "m") and silt plus clay-sized aggregates (< 0.053 mm "s + c"). After extraction, the aggregates were oven-dried overnight at 60 *°C* to a constant weight (Elliott, 1986).

Determination of soil organic carbon in aggregate fractions

Soil organic carbon content was assessed in the aggregate portions (2 to 0.25 mm, 0.25 to 0.053 mm, and < 0.053 mm) by the dichromate (wet) oxidation method as used by Lawal (2013). Accordingly, to each respective aggregate fraction, organic carbon content was categorized as Fine Particulate Organic Carbon (FPOC) for the 2 to 0.25 mm fraction, Intra-aggregate Particulate Organic Carbon (IPOC) for the 0.25 to 0.053 mm fraction, and Silt plus Clay associated Organic Carbon for the fraction lesser than 0.053 mm.

Determination of soil oxidizable organic carbon (SOC) fractions

The SOC fractions were analysed by subjecting soil to oxidizing conditions using an aqueous sulphuric acid (H_2SO_4) solution following the method described by Oliveira *et al.* (2016). Organic C fractions were determined by oxidation using 0.167 mol L^{-1} potassium dichromate ($K_2Cr_2O_7$) in an acidic medium containing various H_2SO_4 concentrations without external heating. Sulphuric acid concentrations used were 2.5, 5, and 10 mL giving final H_2SO_4 concentrations of 3, 6, and 9 mol L^{-1}, with unchanged concentrations of potassium dichromate. After determining the organic C levels through the described three acid-aqueous solutions process, the separation of SOC into the subsequent 4 fractions (F) of diminishing lability (oxidizability) was possible:

Very labile (F1): organic C (OC) oxidized below 3 mol $L^{-1}H_2SO_4$

Labile (F2): difference between OC oxidized at 3 and 6 mol L^{-1} H_2SO_4

Less labile (F3): difference between OC oxidized at 6 and 9 mol L^{-1} H_2SO_4

Recalcitrant (F4): difference between total SOC and OC oxidized at 9 mol L^{-1} H_2SO_4.

The OC found in the first fraction (F1) was taken as labile C (C_L), whereas the sum of OC in the F3 and F4 (F3+F4) fractions was considered as non-labile C (C_{NL}) (Xavier *et al.*, 2009).

Statistical analysis

R statistical software was used for analysing the variance (ANOVA) and correlations (R Core Team, 2015). Due to the fact that the earthworms and termites data did not show normality (Shapiro-Wilk test), and homogeneity of variance (Levene's test), the generalized linear mixed models (GLMM) test through the R package lme4 (Bates *et al.*, 2015) was performed in order to assess the effects of macrofauna and the tree species on soil aggregate fractions and the associated C content. Tukey’s post-hoc comparisons (HSD tests) were executed at $\alpha = 0.05$ where analysis of the variance (ANOVA) revealed significant effects. Correlation analysis was carried out to determine the relationship between soil macrofauna and soil aggregates fractions and soil C.

RESULTS

Earthworms and termites under different tree species

Earthworms (except from the *Acantodrilidae* species) and termites were both significantly influenced by the tree species. *Eudrilidae* were significantly more abundant in soil under ficus (23.3 individuals) in comparison to grevillea and beans (13.8 and 12.5 individuals respectively) and eucalyptus (3.5 individuals), while natural forest did not record any earthworms in this family. Termites recorded significant highest values under natural forest and eucalyptus (12.8 and 12.7 individuals respectively) compared to ficus, grevillea and beans (2.5; 2.3 and 0.0 individuals respectively).

Soil aggregate fractions and associated C under different tree species woodlots

Except for small macroaggregates (SM), all the classes of aggregate sizes were significantly influenced by tree species (Table 3). Significantly higher large macroaggregates (LM) were observed in soil under natural forest (13.1%) while lower values were recorded under beans and grevillea (6.1 and 7.2 % respectively). Microaggregates (m) and the silt and clay (s+c) sized aggregates were significantly lower in soils from natural forest (6.6 and 4.8 % respectively). The different tree species influenced significantly the aggregate associated carbon pools. Lower fine particulate organic carbon (FPOC), intra-aggregate particulate organic carbon (IPOC) and silt and clay associated carbon (s+c) contents were found in soils under beans, whereas natural forest yielded the highest followed by eucalyptus, grevillea and ficus.

Influence of different tree species on oxidizable organic C fractions

The bioavailable organic carbon (labile) was strongly influenced by the tree species (Table 4). Soils under natural forests had significantly greater values (22.9 g kg^{-1}) of labile carbon in comparison to eucalyptus (14.8 g kg^{-1}), grevillea (13.7 g kg^{-1}), ficus (11.1 g kg^{-1}) or beans (9.7 g kg^{-1}). Non-labile fraction did not show significant difference between the agroforestry systems.

Correlations

Termites showed significant correlations with soil aggregates sizes but the earthworm species did not show any significant correlation in this study did not (Table 5). Termites were significantly and positively correlated with large macroaggregates (LM), but negatively correlated with microaggregates (m) and silt and clay sized fraction (s + c). The organic carbon associated with FPOC, IPOC and s+c was significantly and

Table 2. Earthworms and termites as influenced by tree species (mean ± (SE)).

Tree species	Earthworms		Termites
	Eudrilidae	*Acantodrilidae*	*Termitidae*
Beans	12.5 (3.2) ^{ab}	0.25 (0.2)	0.0 (0.0) ^{b}
Eucalyptus	3.5 (3.0) ^{bc}	1.1 (0.7)	12.7 (8.2) ^{a}
Ficus	23.3 (9.9) ^{a}	0.8 (0.6)	2.5 (1.2) ^{b}
Grevillea	13.8 (7.1) ^{ab}	3.3 (6.0)	2.3 (1.9) ^{b}
Natural Forest	0.0 (0.0) ^{c}	0.8 (0.4)	12.8 (7.1) ^{a}

Within columns, means followed by different letters are significantly different at $p < 0.001$.
Mean separation by Tukey’s HSD test.

Table 3. Soil aggregates and associated C as influenced by tree species (mean ± (SE)).

Aggregate size	Beans	Eucalyptus	Ficus	Grevillea	Natural Forest
LM	6.1 (1.5) ^{b}	10.4 (4.3) ^{ab}	9.8 (1.8) ^{ab}	7.2 (2.9) ^{b}	13.1 (1.7) ^{a}
SM	51.5 (7.1)	47.8 (5.4)	54 (3.1)	50.3 (5.1)	59.1 (7.7)
m	24.9 (3.4) ^{a}	20.1 (4.4) ^{a}	18.4 (5.4) ^{a}	20.8 (5.1) ^{a}	6. 6 (1.1) ^{b}
Silt plus clay	14.1 (4.1) ^{a}	15.1 (3.6) ^{a}	13.9 (2.3) ^{a}	12.7 (2.4) ^{a}	4.8 (0.8) ^{b}
Aggregate associated C					
FPOC	22.1 (2.7) ^{b}	44.8 (6.3) ^{a}	40.1 (6.1) ^{a}	33.9 (7.9) ^{ab}	44.0 (7.3) ^{a}
IPOC	28.4 (3.3) ^{b}	40.6 (6.8) ^{b}	35.2 (4.9) ^{b}	38.0 (10.2) ^{b}	58.5 (5.1) ^{a}
Silt plus clay	33.7 (4.4) ^{c}	47.3 (7.2) ^{ab}	39.6 (1.5) ^{bc}	45.9 (8.7) ^{b}	58.7 (5.0) ^{a}

Within rows, means followed by different letters are significantly different at p < 0.001. Abbreviations: LM=large macroaggregate, SM=small macroaggregate; m=microaggregate; FPOC=fine particulate organic carbon; IPOC=intra-aggregate particulate organic carbon. Mean separation by Tukey's HSD test.

Table 4. Soil oxidizable carbon fractions from different tree species (mean ± (SE)).

Carbon fractions	Beans	Eucalyptus	Ficus	Grevillea	Natural Forest
Labile	9.7 (1.1) ^{b}	14.8 (2.4) ^{b}	11.1 (2.4) ^{b}	13.7 (4.0) ^{b}	22.9 (3.7) ^{a}
Non.Labile	15.8 (2.0)	19.4 (4.5)	17.5 (3.5)	18.7 (5.6)	22 (2.6)
C_L/SOC	0.3 (0.0) ^{ab}	0.3 (0.1) ^{a}	0.2 (0.0) ^{b}	0. 3 (0.1) ^{ab}	0.4 (0.0) ^{a}
C_{NL}/SOC	0.5 (0.0) ^{a}	0.4 (0.1) ^{ab}	0.4 (0.0) ^{b}	0.4 (0.1) ^{ab}	0.4 (0.0) ^{a}

Within rows, means followed by different letters are significantly different at $p < 0.001$. Abbreviations: SOC=soil organic carbon; C_L=labile carbon; C_{NL}= non-labile carbon. Mean separation by Tukey's HSD test.

Table 5. Pearson correlation between soil aggregates and earthworms and termites.

	Earthworms		Termites
Soil parameters	*Eudrilidae*	*Acantodrilidae*	*Termitidae*
LM	0.00	-0.02	**0.52 *****
SM	0.07	-0.01	0.16
m	-0.02	-0.07	**-0.27 ****
s+c	-0.02	-0.04	**- 0.27 ****
FPOC	-0.18	-0.05	0.04
IPOC	**- 0.39 *****	-0.11	0.06
s+c	**- 0.35 *****	-0.06	0.1
Labile C	**- 0.21 ***	-0.12	0.09
Non labile C	-0.1	-0.14	-0.03

Abbreviations: LM=large macroaggregate, SM=small macroaggregate; m=microaggregate; FPOC=fine particulate organic carbon; IPOC=intra-aggregate particulate organic carbon; C=carbon. Coefficients marked in bold indicate a significant correlation: * $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$.

negatively correlated with *Eudrilidae* abundance. The bioavailable C (labile C), on the other hand, showed negative and significant correlations with *Eudrilidae*.

DISCUSSION

Soil aggregate fractions and associated carbon from different tree species woodlots

This study showed that the soil under natural forests had higher values of soil macroaggregates (both LM and SM) than those under other tree species. This might be due to the high level of disturbance in soil under the other agroforestry systems, given that they are often tilled during crop cultivation or when trees are planted. Results from previous investigations have shown that tillage reduces soil aggregate stability and that no-till practice promotes the stability of soil macroaggregates (Du *et al.*, 2015; Yoseph *et al.*, 2017; Morlue *et al.*, 2021). This is elucidated by the fact that soils not tilled tend to accumulate higher amounts and diverse quality of organic matter, which enhance organic C content. In addition to this, recurrent tillage has been associated with a decline in organic binding agents such as biomass of fungal hyphae (temporary) and polysaccharides derived from microbes (transient). The reduction in these organic binding agents lead to a decrease in the stability of soil organic matter, thus degradation of soil macroaggregates (Kamau *et al.*, 2020). Besides, fungi and bacteria produce mucilages during decomposition of organic residues which help to stabilize and sustain the macro-aggregates (Kamau *et al.*, 2020; Six *et al.*, 2000), and thus frequent cultivation could lead to a loss of these binding agents. Other studies have reported similar findings; for example, while comparing the effect of soil disturbance under cultivated farms (maize-wheat) and tree based systems (agroforestry) on soil macroaggregate, Dhaliwal *et al.* (2017) found the lowest (5.8%) values of large macroaggregates in soils under the maize-wheat system, where tillage is intense and frequent, and the highest (67.3%) in soils under agroforestry system, receiving less tillage. Similarly, Vladimir *et al.* (2013) reported a significant change in soil aggregates composition when native vegetation was converted into cropland and long term cultivation lands across three different types of soils. Higher losses of macroaggregates were found in all the three types of soil, being higher in Haplic Chernozem soil (93 %), followed by in Haplic Fluvisol (72 %) and Gleyic Vertisol (66 %).

The lowest aggregate-associated organic C in beans could be due to the continuous cultivation with no or little soil organic matter input, contrary to soil under tree species woodlots. It has been indicated that soil under tree-based systems accumulate more organic C content due to the constant organic matter from litter fall and root turnover (Hermine *et al.*, 2010; Kamau *et al.*, 2020). These results are similar to those of Vladimir *et al.* (2013) who found that on average soil aggregate-associated C concentrations were significantly higher under native vegetation and lower by 2-45 % under cropland. The results of the present research revealed that, higher SOC was interrelated to smaller aggregate fractions (<0.053) mm for all the tree species. This might be due to the composition of this fraction which, in comparison to others, contains greater content of silt and clay particles. Clay causes soil to stock more C and enhances soil's capacity to retain C because of the protective nature of clay particles against enzymatic and microbial degradation (Hermine *et al.*, 2010; Trujilo *et al.*, 1997). Furthermore, SOC plays a crucial role in soil aggregation by providing a binding force for soil particles (Dhaliwal *et al.*, 2017). Similar trends were documented by Mahesh *et al.* (2017) where higher values of aggregate-associated OC were recorded in microaggregates than in macroaggregates across all the treatments; natural forest, degraded forest, cropland and biofuel plantation (*Jatropha curcas*).

Influence of soil macrofauna on soil aggregates

Due to their intense activities, soil macrofauna are recognized to lead to changes in soil aggregation, especially regarding earthworms and termites. In this study, earthworms did not significantly correlate with the aggregate stability, unlike termites which did. The termites' significant and positive correlation with large macroaggregates might be associated to the differences in organic matter levels in soils under different tree species woodlots. For instance, the highest levels of termites were recorded in soil under natural forest. Natural forests are generally known as ecosystems that have high tree diversity which implies additional and diversified organic matter input from litter fall, and more food resource for soil macrofauna like termites. A previous study carried out in the same study region as this research linked higher lignin in litter content to higher number of termites under natural forest due to their ability to feed on recalcitrant material (Kataka *et al.*, 2023). Termites influence soil aggregation through their ability to ingest soil particles together with their food leading to the mixing of the soil organic matter with the mineral component in their gut hence altering the physical properties of soil (Jungerius *et al.*, 1999). Termites especially the humivorous type have the aptitude to construct nests enriched with organic matter impacting more on the structural stability of soil. A previous study by Ayuke *et al.* (2011) have confirmed these findings by partially attributing the stability of soil aggregates to the presence of termites.

Earthworms particularly *Eudrilidae* species strongly and negatively correlated with the intra-aggregate particulate organic carbon (IPOC) and the silt plus clay sized aggregate associated C (S+C). This might be linked to the digestive process of earthworms. Casts deposited by some earthworm species after feeding on the soil substrate are shown to contain considerable amounts of semi-processed organic matter. This promotes a favourable environment for proliferation of microorganisms which lead to further mineralisation of the semi-processed organic matter providing more assimilable organic matter which increases soil C mineralisation (Ruiz *et al.*, 2008). Therefore, the low levels of aggregate-associated carbon found in soils under ficus, beans and grevillea might be attributed to the greater values of earthworms. These results are in agreement with Kamau *et al.* (2020) who attributed the decrease in C content in soil aggregates under *Z. gilletii* trees to the action of earthworms, and especially the species *Nematogenia lacuum*. In accordance with these findings, Sylvain *et al.* (2007) and Desjardins *et al.* (2003) documented in their studies that the higher number of earthworms was related to a significant decrease in C content in the first 10-20 cm layer of soil. Although they didn't look at the aggregate-associated C but at the total soil C, they highlighted that earthworm activities might lead to C losses.

Oxidizable organic carbon under different tree species woodlots

Determining soil oxidizable organic carbon fractions was convenient for the assessment of the degree of soil organic C lability. This analysis provided important information on the quality of organic materials from diverse tree species which was achieved through the separation of the labile organic C forms from the non-labile. Natural forest had higher values of labile C. This might be associated with the nature and good quality of natural forest litter which is so much diversified (Anderson and Domsch, 1989). Similar findings were reported by Yusheng *et al.* (2009) where natural forest recorded higher labile C than that reported in plantations of *Castanopsis kawakamii* and *Chinese fir*. Same trends were found by Jurisandhya and Sharma (2022) with higher values of labile carbon being recovered under natural forest (2.6%) compared to that in horticulture (pineapple) and agriculture (lowland paddy) land use system (2.4% and 2.0% respectively). They attributed high labile C content in natural forest to the abundance of litter materials which increases organic substrates and microbial biomass, and the low labile C in agriculture land use system to the prolonged cultivation practices.

The higher labile C recorded under natural forest coincides with its greater values in the soil large macroaggregates. This might reflect soil carbon's binding capacities in the soil aggregation formation. It has been recognised that soil labile C is important in the process of soil aggregates formation and stabilization because it supports microbial biomass production which is in part responsible for soil aggregation improvement (Haynes, 2005). In addition, soil organic labile C has been documented to act as a binding agent in the process of soil macroaggregate formation (Dhaliwal *et al.*, 2017). Moreover, soil labile C is sensitive to soil management practices, especially to soil tillage. This might also be another reason natural forest had higher values of labile C compared to the other agroforestry systems where several operations that cause soil disturbance are carried out at the initial stages of tree growth. Soil tillage is known to disrupt soil macroaggregates, which ends up affecting soil labile C by reducing its physical protection (Bronick and Lal, 2005). Other studies have also suggested similar outcome where stability of soil aggregates was significantly associated to the quantity of soil organic C together with its labile component (Somasundaram *et al.*, 2018; Tisdall and Oades, 1982). Similarly, Clever *et al.* (2023) reported that the conversion from tillage systems to no-tillage systems showed a positive correlation between soil labile compounds and the proportion of large macroaggregates in 0–20 cm soil layer, and suggested that good maintenance of labile organic C by no-tillage practices results in improvement of soil large macroaggregates due to an accumulation of SOC.

CONCLUSION

This study was conducted to assess the combined effect of diverse tree systems, earthworms and termites on soil aggregates and carbon in order to inform on tree species selection and sustainable soil productivity. The results of this study, in reference to natural forest, showed that tree cultivation may have detrimental impacts on soil aggregates and C content due to the presence of soil disturbance from crop cultivation, where trees are intercropped with annual crops, and tree management in woodlots. Soil aggregates and carbon may have been improved in natural forest due to the higher input of diverse litter and to the mediating role of earthworms and termites through the production of casts and soil mixing hence improving soil aggregation and promoting microbial mineralization of organic matter. This study therefore suggests that a high diversification of tree species is to be considered when integrating trees into crop farms to achieve a better maintenance of soil quality.

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Data availability. data are available upon reasonable request to jeanleonkataka96@gmail.com

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