





Microbial Properties of Soil in Different Coverages in the Colombian Amazon

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ABSTRACT

Changes in coverage affect the activity of soil's microbial communities, affecting the carbon and nitrogen cycle. The variability of biochemical properties in different coverages (native forest, forest plantation, silvopastoral system and pasture) located in the northwest of the Colombian Amazon was evaluated. Edaphic properties were determined as: organic carbon (OC), total nitrogen (TN), microbial biomass carbon (MBC) and microbial biomass nitrogen (MBN) using the fumigation method. A significant effect was found to land use × depth interaction for the variables moisture, pH, CO and MBC/OC ratio ($p < 0.05$), while MBC and dCO_2 showed differences only for land use and NT, MBN and MBN/TN ratio at depth ($p < 0.05$). In general, when modifying the soil cover, changes were made in the chemical properties that affected the microbial activity.

Keywords: fumigation, CO₂ emissions, multivariate analysis, structural equations modeling.

1. INTRODUCTION AND OBJECTIVES

Changes in coverage affect different soil properties (Jia et al., 2005), such as microbials (Krauss & Allen, 2003), which act as early signs of degradation or soil improvement (Gichangi et al., 2016; Vallejo et al., 2012). These signs of variation are different between disturbed and undisturbed systems (Lagerlöf et al., 2014), which directly influence the stability of the ecosystem and soil fertility (Yang et al., 2010).

One of the main attributes to assess changes in soil properties caused by crops or changes in vegetation cover is the microbial biomass carbon (MBC) (Lopes et al., 2011). This responds quickly to the effect of soil disturbance or recovery (Rangel-Vasconcelos et al., 2015), since it maintains the living component of the soil's organic matter (SOM) (Araújo et al., 2013).

Fluctuations in the size of the MBC during the growing season is considered an important factor in controlling the carbon and nitrogen rotation of the soil (Yang et al., 2010). Likewise, the microbial biomass of the soil represents the lowest percentage of total nitrogen (1 to 5%) of the soil, responsible for the labile reserve, the nutrient cycle and the decomposition of the SOM, being a good indicator of soil together with soil nitrogen (Fernandes et al., 2011; Jackson et al., 2003).

In this sense, the loss of organic matter and nutrients from the soil reduces the biomass and the microbial activity of the soil (Nunes et al., 2012); since the larger the MBC, the greater the temporary immobilization of C, N and other nutrients and then the lower the loss of nutrients from the soil/plant system (Moreira & Malavolta, 2004; Wang et al., 2003). The transition from forest to pasture in the Amazon region has shown that MBC is reduced three years after pasture establishment but shows higher levels in old pastures with forest-like content (Cenciani et al., 2009).

On the other hand, it has been reported that tree species in a system have a different influence on soil C labile content (Silberman et al., 2015), thus influencing the structure and functionality of soil microbial communities (Anriquez et al., 2016; Silberman et al., 2016). Likewise, it is known that silvopastoral systems (SPS) are capable of improving the quality and function of the soil by reflecting increases in the C and N content of the soil microbial biomass, enzymatic activities and edaphic respiration (Cubillos et al., 2016;

Silberman et al., 2016; Vallejo et al., 2012). Therefore, the variability of microbial properties under different coverings in the Colombian Amazon was evaluated, hoping to find greater microbial activity of the soil in less intervened coverings such as the forest.

2. MATERIALS AND METHODS

2.1. Study area

The study was carried out in the Northwest of the Colombian Amazon, Southwest of Caquetá Department, between the municipalities of Morelia, Belén de los Andaquíes, Albania and San José del Fragua, cataloged in the life zone as a tropical rainforest. The climate is characterized by an average temperature of 24.1 °C in the period of highest precipitation (April-August), and 25 °C in the dry season (November-March) with an annual rainfall between 2,500 and 4,000 mm, its average relative humidity is 84% and the average height above sea level is 250 m (IGAC, 2014).

2.2. Soil sampling

Four constant land uses were studied in their vegetation cover: forest, forest plantation, silvopastoral system and pasture (Table 1). In each cover with a minimum area of one hectare, a diagonal transect was made in the direction of the slope. In this transect, three main sampling points with a diameter of 25 m were taken. At each point, a composite sample was made from four sub-samples at two depths (0-10 and 10-20 cm, respectively). For the chemical analysis, the samples were dried at room temperature, then passed through a 2 mm sieve and stored in plastic bags. For the biochemical analysis, the samples were treated wet and were passed through a 2 mm sieve, stored in hermetic plastic bags and refrigerated at 4 °C until further analysis.

2.3. Edaphic properties

To measure the water content of the soil, the ProCheck equipment adapted to a 5TE sensor was used. The pH was determined by the potentiometer in water. The organic carbon (OC) content of the soil was determined by the Walkley-Black method (Li et al., 2017).

The total nitrogen (TN) of the soil was measured by the micro-Kjeldahl method. The carbon of the MBC

Table 1. Description of the coverages evaluated in the Colombian Amazon.

Coverage	Definition
Native forest	Conservation area with low human intervention, diversified vegetation coverage and in different states of natural succession.
Forest plantation	Area in recovery process, with vegetal cover of pasture in natural regeneration and 2-year-old timber trees (<i>Cariniana pyriformis</i> , <i>Cedrella odorata</i>).
Silvopastoral system	Area dedicated to bovine livestock farming, with diversified plant cover, divided into three strata: high timber tree stratum (<i>Gmelina arborea</i>), middle stratum species for browsing (<i>Tithonia diversifolia</i>) and stratum under the grass.
Pasture	Area dedicated to bovine livestock farming, with vegetal cover in monoculture of grasses.

and the nitrogen of the microbial biomass (MBN) were determined according to Vance et al. (1987) with extraction by K_2SO_4 of fumigated and non-fumigated soils with $CHCl_3$.

Soil CO_2 emissions were measured using the CIRAS-3 Portable Photosynthesis System infrared gas analyzer (PP Systems Inc., Amesbury, MA, USA) equipped with a soil respiration chamber (SRC-1). Registered CO_2 emissions included autotrophic respiration of plant roots and heterotrophic respiration of soil organisms. The total readings of CO_2 emissions from the soil shown by the CIRAS-3 were recorded and the results are presented as grams of carbon dioxide per square meter per second ($g\ CO_2\ m^{-2}\ s^{-1}$).

2.4. Statistical analysis

A mixed linear model (MLM) was adjusted to predict the differences of soil microbial properties, and coverage and depth were included as fixed factors. The farm where the coverage was plotted and the plot considering the repetition within the coverage were included as random factors. The means comparisons were made using the Fisher LSD test ($p < 0.05$). The assumptions of normality and homogeneity of variance were evaluated through exploratory residue analysis. Likewise, a multivariate analysis was carried out to determine similarity between coverage-depths and to explore the relationship between variables through a principal components analysis (PCA), determining the similarity between coverage-depths by Monte Carlo test (1,000 permutations). Likewise, to determine the causal relationships between the multiple variables that interact and influence the microbial properties of the soil, they were adjusted in a structural equation model (SEM).

The MLM settings were carried out using the *lme* function of the *nlme* package (Pinheiro et al., 2015), the PCAs were developed in the package *Ade4* (Dray & Dufour, 2007) and the SEM was adjusted with the function *cfa* of the SEM package (Fox, 2006). The visualization of the adjusted SEM was performed using the *semPaths* function of the *semPlot* package (Epskamp, 2014). All the statistical analyses carried out were developed using the R language version 3.4.0 (R Development Core Team, 2017) through the interface of the statistical software InfoStat (Di Rienzo et al., 2017).

3. RESULTS AND DISCUSSION

When analyzing the effect of coverage and depth on soil microbial properties, significant differences were found for soil moisture, pH, OC and the MBC/OC ratio ($p < 0.05$). For MBC and dCO_2 differences were presented only for coverage and for TN, MBN and the MBN/TN ratio in depth ($p < 0.05$). Likewise, differences were found for the interaction coverage \times depth for the C/N ratio ($p < 0.05$) (Table 2).

The PCA for soil microbial properties explains 43.3% of the total variability of the data with the first two axes (Figure 1). Axis 1 associates the highest contents of MBC, MBC/MBN and MBC/OC with the SPS coverage (Figure 1). Axis 2 expresses a gradient of higher pH and TN, which are not clearly associated with a coverage (Figure 1). The results show a high coverage effect on soil microbial properties (p-value: 0.001) (Figure 1). There are also significant differences between depths for soil microbial properties (p-value: 0.001) (Figure 1). The depth 0-10 cm was characterized by having the highest contents of C/N, MBN/N, MBN, OC, SM, dCO_2 , MBC, MBC/MBN and MBC/OC (Figure 1).

Table 2. Mean values (\pm SE) of the edaphic properties, studied under different coverages in the Colombian Amazon.

Variable	Native forest		Forest plantation		Silvopastoral system		Pasture		p-value		
	0-10		10-20		0-10		10-20		Coverage	Depth	Cob*Prof
	Average	SE	Average	SE	Average	SE	Average	SE			
SM (%)	54.62 \pm 2.51	47.73 \pm 0.91	56.99 \pm 2.66	45.61 \pm 2.17	39.39 \pm 1.92	34.69 \pm 0.94	41.8 \pm 1.67	34.57 \pm 1.15	<0.0001	<0.0001	
pH	4.54 \pm 0.15	4.49 \pm 0.12	4.5 \pm 0.07	4.64 \pm 0.09	4.33 \pm 0.06	4.56 \pm 0.06	4.36 \pm 0.06	4.64 \pm 0.06	0.0107	0.0146	
OC (%)	2.9 \pm 0.15	2.01 \pm 0.13	3.54 \pm 0.2	2.1 \pm 0.11	3.55 \pm 0.11	1.88 \pm 0.08	3.37 \pm 0.11	1.8 \pm 0.06	0.0491	<0.0001	
TN (%)	0.74 \pm 0.01	0.58 \pm 0.06	0.72 \pm 0.03	0.67 \pm 0.03	0.69 \pm 0.03	0.69 \pm 0.01	0.73 \pm 0.02	0.7 \pm 0.01		<0.0001	
MBC (mg/kg)	765.2 \pm 120.8	654 \pm 103	1002.5 \pm 286.9	1729.8 \pm 391.6	2376.9 \pm 566.9	2252 \pm 440	1707.1 \pm 311.3	1734.6 \pm 371.1	0.0191		
MBN (mg/kg)	39.18 \pm 9.22	28.46 \pm 4.3	34.45 \pm 6.81	26.27 \pm 4.04	42.55 \pm 6.34	27.77 \pm 8.88	56.77 \pm 8.03	22.51 \pm 2.64		0.0077	
dCO ₂ (g CO ₂ m ⁻² s ⁻¹)	0.6 \pm 0.16	0.66 \pm 0.3	1.27 \pm 0.3	1.12 \pm 0.31	0.85 \pm 0.16	0.35 \pm 0.09	0.65 \pm 0.11	0.58 \pm 0.13	0.0085		
C/N	3.95 \pm 0.23	5.54 \pm 1.84	5.37 \pm 0.58	3.71 \pm 0.66	6.24 \pm 0.74	2.78 \pm 0.16	4.93 \pm 0.35	2.57 \pm 0.08		0.001	0.0044
MBC/MBN	34.57 \pm 8.36	34.48 \pm 9.17	68.64 \pm 21.49	114.31 \pm 28.61	199.38 \pm 99.27	205.3 \pm 59.02	93.91 \pm 28.97	83.03 \pm		20.1	
MBC/OC	270.5 \pm 43.34	345 \pm 62.1	281.94 \pm 74.35	777.06 \pm 140.2	769.5 \pm 198.1	1522 \pm 355.6	565.42 \pm 107.8	1108.8 \pm 267.5	0.0351	0.0343	
MBN/N	54.79 \pm 13.57	56.73 \pm 11.2	55.27 \pm 13.01	41.42 \pm 6.35	62 \pm 8.23	41.2 \pm 12.63	78.53 \pm 10.42	31.77 \pm 3.5		0.0251	

SE: standard error; SM: soil moisture; pH: acidity coefficient; OC: organic carbon; TN: total nitrogen; MBC: microbial biomass carbon; MBN: microbial biomass nitrogen; dCO₂: carbon dioxide delta; C/N: ratio of carbon and nitrogen; MBC/MBN: ratio of microbial biomass carbon and microbial biomass nitrogen; MBC/OC: ratio microbial biomass carbon and organic carbon; MBN/N: ratio of microbial biomass nitrogen and nitrogen

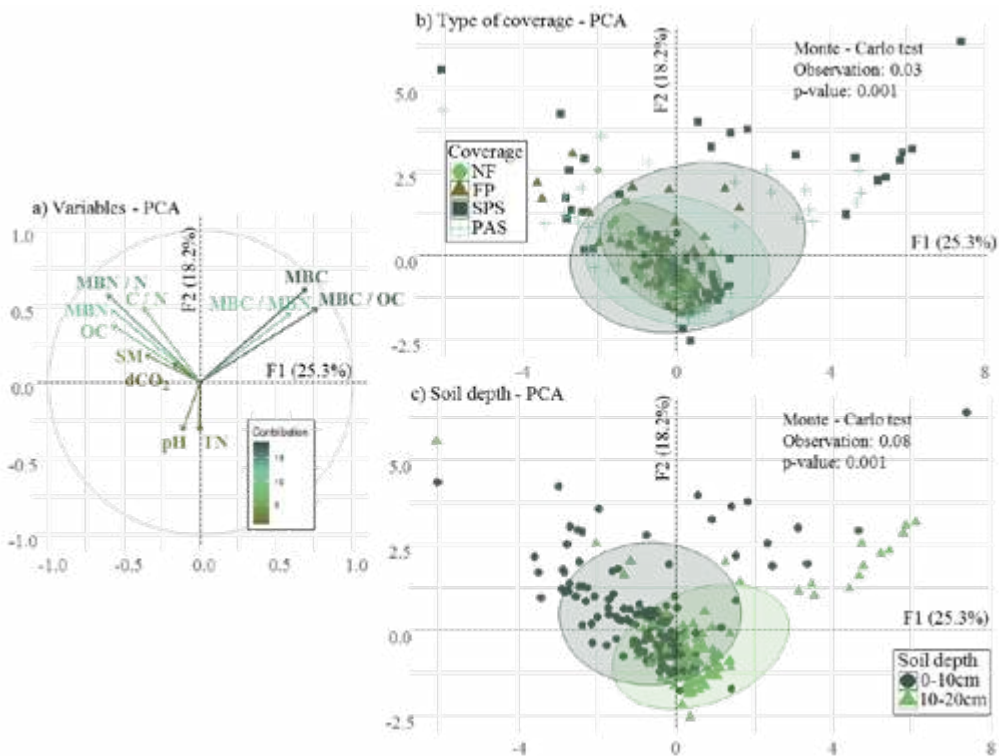


Figure 1. Projection in the factorial plane F1/F2 of the principal components' analysis of edaphic properties, and the samples points group depending on the type of coverage or the soil depth. a) Circle of correlations. b) Ordination of the coverage included in the sample on the factorial plane of a PCA. c) Ordination of the depths included in the sample on the factorial plane of an PCA. SM: soil humidity; pH: coefficient of acidity; OC: organic carbon; TN: total nitrogen; MBC: microbial biomass carbon; MBN: microbial biomass nitrogen; MBC/OC: relation of microbial biomass carbon and organic carbon; MBN/N: relation of microbial biomass nitrogen and nitrogen; NF: native forest, FP: forest plantation; SPS: silvopastoral system; y PAS: pasture.

There were differences in correlation trends between variables. OC was the soil variable that presented the highest correlation with the others, being positive with TN, MBN, dCO_2 and the C/N and MBN/N ratios ($p < 0.05$); and negative with MBC/OC ($p < 0.05$) (Table 3). The SEM analysis showed a good fit to the data (Figure 2), indicated by the non-significant value χ^2 ($p = 0.85$), high CFI (Confirmatory Fit Index 0.97),

low RMSEA (Root Mean Square Error of Approximation 0.005) and low stability index 0.23. The model suggests that the OC content strongly affects the activity of MBN, just as the concentration of TN affected at a low level. On the other hand, soil moisture content (SM) and organic carbon (OC) affected the carbon dioxide delta (dCO_2). However, the SEM shows a negative relationship between the pH and the MBC.

Table 3. Correlation coefficients between edaphic properties under different coverages in the Colombian Amazon.

Variables	SM	pH	OC	TN	MBC	MBN	dCO_2	C/N	MBC/MBN	MBC/OC	MBN/N				
SM	1														
pH	0.01	1													
OC	0.39	<0.0001	-0.16	0.013	1										
TN	0.16	0.0102	0.04	0.13	0.037	1									
MBC	-0.07	-0.2	0.002	-0.07	-0.04	1									
MBN	0.12	0.02	0.29	<0.0001	0.18	0.0043	-0.02	1							
dCO_2	0.33	<0.0001	-0.02	0.23	0.0003	0.07	0.01	0.05	1						
C/N	0.09	-0.02	0.45	<0.0001	-0.7	<0.0001	-0.05	0.02	0.02	1					
MBC/MBN	-0.01	-0.12	-0.08	-0.09	0.61	<0.0001	-0.18	0.0049	-0.02	0.01	1				
MBC/OC	-0.12	0.0683	-0.11	-0.25	0.0001	-0.06	0.9	<0.0001	-0.08	-0.06	-0.13	0.0361	0.58	<0.0001	1
MBN/N	0.08	0.01	0.27	<0.0001	-0.08	-0.02	0.95	<0.0001	0.02	0.23	0.0002	-0.18	0.0042	-0.07	1

SM: soil moisture; pH: acidity coefficient; OC: organic carbon; TN: total nitrogen; MBC: the microbial biomass carbon; MBN: microbial biomass nitrogen; dCO_2 : carbon dioxide delta; C/N: ratio of carbon and nitrogen; MBC/MBN: ratio of microbial biomass carbon and microbial biomass nitrogen; MBC/OC: ratio of microbial biomass carbon and organic carbon; MBN/N: ratio of microbial biomass nitrogen and nitrogen

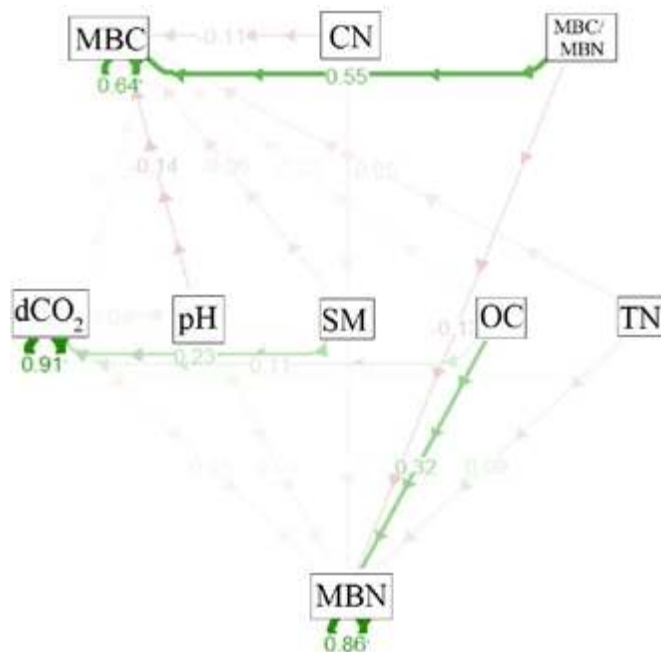


Figure 2. Direct and indirect effects between soil microbial properties with MBN and MBC. MBN: microbial biomass nitrogen; MBC: microbial biomass carbon; NT: total nitrogen; OC: organic carbon; CN: carbon/nitrogen ratio; SM: soil moisture; dCO_2 : carbon dioxide delta; pH: acidity coefficient; MBC/MBN: ratio of microbial biomass carbon and microbial biomass nitrogen. The numbers in the vectors indicate significant standardized coefficients of relationship between variables. The green line indicates positive and the red line indicates negative effects.

The OM decomposition is accelerated by the activity of the soil microorganisms (Spohn et al., 2016) which is measured by the MBC, in this way, the availability of OC depends on the characteristics of the soil, such as the depth, and not of coverage (Cubillos et al., 2016; Silva et al., 2012). Among coverages, SPS presented the highest MBC concentration (Table 1), possibly to the soil restoration that positively affected the microbial properties (Araújo et al., 2013), generating greater stability to the microorganisms (Anriquez et al., 2016) as well as the processes that they develop in the soil (Silva et al., 2012).

The high content of MBC is also associated with the radicular system of grasses that is abundant and remains active throughout the year (Nogueira et al., 2016) and with OM derived from animals (Toda et al., 2011). Therefore, land uses that guarantee the continuous entry of organic waste (Silva et al., 2012) will have a higher MBC (Nsabimana et al., 2004). By generally analyzing the relationships between different soil variables with microbial properties, associations were found with ecological functions capable of reflecting changes in land use (Jackson et al., 2003). Specifically, the important relationships are those presented between the OC and MBC, which explain the metabolic efficiency that exists under the coverage (dCO_2), that depends on the availability of substrate as organic matter.

Although in the present study there was no direct relationship between OC and MBC, it is important to mention that this relationship is directly proportional, being susceptible to changes. The greater the MBC, the greater the temporary immobilization of C, N and other nutrients, the lower the loss of nutrients from the soil-plant system (Moreira & Malavolta, 2004; Wang et al., 2003), being analyzed in different studies (Oulbachir et al., 2009).

MBC is a very small fraction of the soil; however, it is very important since it refers to the fraction of C immobilized in microorganisms (Jia et al., 2005), being an easily mineralizable fraction (Rodrigues et al., 2015). Microbial growth depends mainly on greater availability of carbon in the soil, which is responsible for its variation (Rodrigues et al., 2015) because it regulates the energy flow and the nutrient cycle. On the other hand, when modifying the coverage, soil moisture changes that affect the microbial activity are presented

(Jia et al., 2005); in this way, a highly significant and positive correlation was found between soil moisture and OC (Table 3), which suggests that soil moisture has a significant influence on the accumulation or decomposition of organic matter (Jia et al., 2005) and therefore on microbial biomass, since it depends mainly on the organic carbon of the soil.

Among the various soil properties that influence the MBC of the soil, pH is an important component (Vallejo et al., 2010), especially in humid regions with acid soils (Tian et al., 2017), since most of the microorganisms are inhibited when the pH is less than 4.5 (Chen et al., 2015), which is considered a critical factor that determines the effect of the deposition of N in microorganisms (Geisseler & Scow, 2014). In this study, it was found that the pH of the soil was negative with the MBC, due to the sensitivity of microbial activity to it (Vallejo et al., 2010). On the other hand, it was found that OC has a great influence on the activity of MBN and TN (Figure 2).

4. CONCLUSIONS

- The change in coverage affected the microbial properties of the soil, with the MBC being greater in the coverage with livestock activity such as silvopastoral system and pasture.
- The soil depth 0-10 cm exhibited higher microbial activity, as well as higher soil moisture, organic carbon and carbon dioxide delta.
- The organic carbon content in the soil affected edaphic properties, such as NT, MBN, dCO_2 , C/N, MBN/N and MBC/OC.

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