Microbiological and faunal soil attributes of coffee cultivation under different management systems in Brazil

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Abstract

Brazil is the biggest coffee producer in the world and different plantation management systems have been applied to improve sustainability and soil quality. Little is known about the environmental effects of these different management systems, therefore, the goal of this study was to use soil biological parameters as indicators of changes. Soils from plantations in Southeastern Brazil with conventional (CC), organic (OC) and integrated management systems containing intercropping of *Brachiaria decumbens* (IB) or *Arachis pintoi* (IA) were sampled. Total organic carbon (TOC), microbial biomass carbon (MBC) and nitrogen (MBN), microbial activity (C-CO₂), metabolic quotient (*q*CO₂), the enzymes dehydrogenase, urease, acid phosphatase and arylsulphatase, arbuscular mycorrhizal fungi (AMF) colonization and number of spores and soil fauna were evaluated. The greatest difference between the management systems was seen in soil organic matter content. The largest quantity of TOC was found in the OC, and the smallest was found in IA. TOC content influenced soil biological parameters. The use of all combined attributes was necessary to distinguish the four systems. Each management presented distinct faunal structure, and the data obtained with the trap method was more reliable than the TSBF (Tropical Soils) method. A canonic correlation analysis showed that *Isopoda* was correlated with TOC and the most abundant order with OC. *Isoptera* was the most abundant faunal order in IA and correlated with MBC. Overall, OC had higher values for most of the biological measurements and higher populations of *Oligochaeta* and *Isopoda*, corroborating with the concept that the OC is a more sustainable system.

Keywords: Coffea arabica L., soil macrofauna, soil metabolism.

Atributos microbiológicos e da fauna do solo com diferentes sistemas de cultivo de cafeeiro no Brasil

Resumo

O Brasil é o maior produtor mundial de café e diferentes sistemas de manejo têm sido aplicados para melhorar a sustentabilidade e a qualidade do solo. Pouco se conhece sobre os efeitos ambientais desses sistemas de manejo, assim, o objetivo desse estudo foi utilizar parâmetros biológicos do solo como indicadores de mudanças nos sistemas. Foram amostrados, na região sudeste do Brasil, solos com cultivo convencional (CC), orgânico (OC) e sistema integrado de cultivo consorciado com $Brachiaria\ decumbens$ (IB) ou com $Arachis\ pintoi$ (IA) na entrelinha. Foram avaliados o carbono orgânico total (TOC), carbono e nitrogênio da biomassa microbiana (MBC e MBN), atividade microbiana (C-CO₂), quociente metabólico (qCO₂), as enzimas desidrogenase, urease, fosfatase ácida e arilsulfatase, a colonização e número de esporos de fungos micorrízicos arbusculares (AMF) e a fauna do solo. A maior diferença entre os sistemas de manejo foram verificadas no teor de matéria orgânica do solo. O maior teor de TOC foi encontrado no OC, e o

menor teor encontrado no sistema IA. O teor de TOC influenciou os parâmetros biológicos e a diferenciação da fauna do solo. O uso combinado de todos os atributos foi necessário para diferenciar os quatro sistemas de cultivo. Cada manejo apresentou estruturas diferentes de fauna, e dados obtidos com o método de armadilhas tipo *pitfall* foi mais confiável do que o método TSBF (Solos Tropicais). A análise de correlação canônica mostrou que *Isopoda* foi correlacionado com TOC e a ordem mais abundante em OC. *Isoptera* foi a ordem da fauna mais abundante em IA e foi correlacionada com MBC. Em geral, OC apresentou os maiores valores para a maioria dos atributos biológicos, inclusive para abundância de indivíduos de *Oligochaeta* e *Isopoda*, corroborando com o conceito de que OC é um sistema mais sustentável.

Palavras-chave: Coffea arabica L., macrofauna do solo, metabolismo do solo.

1. Introduction

Coffee is one of the most important tropical crops in the world, with an estimated exports trade of US\$ 15.4 billion in 2012/2013, according to the International Coffee Organization (ICO, 2014). The world total production of coffee beans was more than 145 million bags (8.7 teragrams) in 2013 (ICO, 2014). With 2.2 million hectares of cultivated area, Brazil is the largest producer in the world (Brasil, 2014). In the year 2013/2014, Brazil had an estimated production of 53.7 million bags (3.2 teragrams) of coffee, which corresponds to approximately one third of the world production (Brasil, 2014).

During the last decades, many farmers adopted alternative management systems such as coffee being produced under organic farming. The main driver for this change was the higher prices paid for products of organic origin, even without considering the technical and environmental aspects. Information about best management practices for sustainable crop production is scarce, and the priority should focus on which management systems can sustain or improve the physical, chemical and biological attributes of the soil (Bhardwaj et al., 2011; Lima et al., 2013; Doran and Zeiss, 2000). Evaluating soil quality in the field, where contrasting production methods are used, will help with the understanding of how sustainable these practices are, and the development of soil management strategies (Bhardwaj et al., 2011; Garrigues et al., 2012).

Current understanding of soil biological processes is not comprehensive enough to use them solely as soil quality (SQ) indicators, even though a great effort is being made in this respect. However, the importance of the biological soil attributes for the soil sustainability is consensual (Chaer et al., 2009; Rutgers et al., 2012). Influencing several biological attributes, the soil organic matter (SOM) is as a key indicator of SQ. Besides fulfilling the basic requirement of being sensitive to soil management changes, the SOM influences water infiltration and retention, soil susceptibility to erosion, and, additionally, its mineralization is an important source of nutrients for plants (Blanco-Canqui et al., 2013; Chaer et al., 2009; Velasquez et al., 2007).

Several biological attributes have potential to be used as SQ. The microbial activity may be used to understand the soil mineralization processes, the intensity of the energy flow in the soil, and to monitor SOM decomposition (Anderson, 2003; Schmidt et al., 2011). The microbial biomass is related to the processes of organic matter

decomposition, interacting with nutrient dynamics, and the regeneration of soil aggregate stability (Cotrufo et al., 2013; Velasquez et al., 2007).

Other potential indicators are the soil macrofauna and microbiota, which respond rapidly to environmental changes, due to their quick adaptation to new conditions, being able to detect alterations prior to any change of physical and chemical patterns of the soil (Baretta et al., 2007a). Among them, arbuscular mycorrhizal fungi, soil enzymatic activity and the composition and abundance of the soil fauna may indicate changes imposed to the soil environment by management techniques (Andrade et al., 2009; Balota and Chaves, 2010; Chaer et al., 2009; Velasquez et al., 2007).

Nowadays, the use of soil fauna as bio-indicator of soil quality is becoming more common, since they have several crucial functions in soil systems, e.g. soil structure (Brussaard et al., 2007). The faunal groups can be useful for comparing crops and the intensity of human intervention in agroecosystems, considering the fact that their interactions with abiotic factors affect their functions (Coleman, 2008; Karanja et al., 2009; Socarrás, 2013). Thus, a study of faunal groups together with biochemical and other biological features of soil and their correlations can be used as robust indicators of soil quality (Velasquez et al., 2007).

The objective of this study was to evaluate the influence of different coffee management systems on biological parameters. And the hypothesis that TOC, biological attributes and faunal groups in the soil will serve as indicators of soil quality in different coffee management systems was tested.

2. Material and Methods

2.1. Sampling sites description

Soil samples from different coffee management systems were collected from two farms in April, 2006, in the southern part of Minas Gerais State. This region is the largest *C. arabica* producing state in Brazil (69% of the production) (Brasil, 2014). Typical farms that represent popular practices in the region were chosen.

On the first farm (coordinates 21° 42' 30" S e 46° 34' 20" W), conventional and organic coffee fields were sampled. The conventional coffee field was planted in rows spaced $2.0 \text{ m} \times 1.0 \text{ m}$ apart and the coffee cultivar used was Acaiá 417/19 planted in 1976. In 2001, 22.5 ha of this conventional coffee area were converted into organic management. The differences between both fields were that

in the organically managed field organic amendments were added to the soil and no pesticides or chemical fertilizers used. Organic materials added included coffee grain husk (5.0 Mg ha⁻¹ year⁻¹) and castor bean cake (Ricinus communis L.) (6.5 Mg ha⁻¹ year⁻¹). In the conventional area an average of 400 kg ha-1 year-1 of N and K and 100 kg ha⁻¹ year⁻¹ of P were applied as synthetic fertilizers, and coffee grain husk was sporadically applied in very small amounts. In the conventional system, the coffee rust fungus Hemileia vastatrix Berk & Br. was controlled by using thiamethoxam (30% m/m) and cyproconazole (30% m/m), and Hypothenemus hampei Ferrari (the coffee berry borer) was controlled with endosulfan. Soil chemical parameters were analyzed according to van Raij et al. (2001). In the organic fields, the chemical measurements were: pH (CaCl₂), 5.9; P, 93 (mg dm⁻³); K⁺, 8.3, Ca⁺², 72, Mg⁺², 27 (mmol₂ dm⁻³). The chemical measurements in the conventional fields were: pH (CaCl₂), 4.9; P, 38 (mg dm⁻³); K^+ , 5.4, Ca^{+2} , 39, Mg^{+2} , 12 (mmol_c dm⁻³).

In the second farm (coordinates 22° 17' 02" S and 46° 22' 02" W), soil samples from integrated coffee plantation management systems were collected, with two different green manure crops between the coffee rows: Arachis pintoi Krapov & WC Greg. (Fabaceae/Leguminoseae) and Brachiaria decumbens Stapf (Poaceae/Gramineae). In both integrated fields 150 kg ha⁻¹ year⁻¹ of N and 124 kg ha⁻¹ year⁻¹ of K were applied. In the integrated system, pesticides are only used based on need, and this is in contrast to conventional system in which regular applications of the pesticides are made. In the integrated management system, thiamethoxam (30% m/m), cyproconazole (30% m/m) and azoxystrobin (50% m/m) were sometimes used to control the plant pathogens (Hemileia vastatrix Berk & Br. and Cercospora zeae-maydis Tehon & Daniels). Besides, the intercalary plants are pruned periodically, and their clippings remain on the soil in the cultivation area, which allows the accumulation of organic matter, improving nutrient cycling. The coffee cultivar grown in this area is Catuaí IAC 99, planted in 2002, spaced 2.0 m \times 1.0 m apart. The chemical parameters analyzed according to van Raij et al. (2001) for these areas were: pH (CaCl₂) 4.3; P, 19 (mg dm⁻³); K⁺, 3.6, Ca⁺², 20, Mg⁺², 4 (mmol_o dm⁻³), for the A. pintoi area, and, pH (CaCl₂), 4.6; P, 20 (mg dm⁻³); K⁺, 3.5, Ca⁺², 34, Mg⁺², 12 (mmol₂ dm⁻³) for the area with *B. decumbens*.

From now on the following abbreviations will be used for the different management systems: OC for coffee under organic management, CC for coffee under conventional management, IA for coffee with integrated management and *Arachis pintoi* between rows and IB for coffee with integrated management and *Brachiaria decumbens* between rows.

2.2. Soil sampling

To optimize the soil sampling, three replicate random rows of coffee trees were chosen in each field. In the first two rows three sampling points 20 m apart were chosen and in the third longer row four sampling points were chosen. A total of ten soil samples per management system

were collected, from three replicate rows, considering a random design.

Each sample was composed by five sub-samples, collected at a depth of 20 cm, below the crowns of the coffee trees, at the aforementioned points. The sampling method was standardized for all soil analyzes, to allow the comparison of all variables in the multivariate analysis. Soil samples were kept at 4 °C until the analysis, within approximately a month.

2.3. TOC, C-CO2, MBC and MBN

To determine the TOC a variation of the Walkey-Black method was used, according to van Raij et al. (2001). From the 10 samples collected in each area, five random replications were used. The C-CO $_2$ and q CO $_2$ of the areas was determined by the Anderson and Domsch (1989) methodology, with a soil incubation period of seven days, while the MBC and the MBN were evaluated through the fumigation-extraction method (Vance et al., 1987).

2.4. Enzymatic activity

The acid phosphatase analysis was performed according to the methodology of Tabatabai and Bremner (1969), with the following modification: the extract was diluted 10 times and the absorbency was read at 400 nm. Arylsulphatase activity was determined according to the methodology of Tabatabai and Bremner (1970), dehydrogenase activity according to Casida-Júnior et al. (1964) and urease activity was estimated according to Tabatabai and Bremner (1972). Six replicates were analyzed, randomly selected from the 10 samples collected in each coffee management area.

2.5. Root colonization and AMF spore density

Roots of coffee plants were manually collected at 5 cm depth in the soil below the tree canopies. The sampled roots were rinsed in running water and treated with KOH 10% for 60 minutes at 90 °C. Afterwards, the roots were bleached with 15% $\rm H_2O_2$ for 10 seconds. The fungal structures inside the roots were dyed for 3 min with 5% pen ink, diluted in a 5% acetic acid solution at 90 °C (Vierheilig et al., 1998). For each sample, twenty segments of 1 cm of the dyed roots were mounted on a microscope slides and were evaluated under a microscope.

The spores were extracted of 50 g of soil through wet sieving, according to Gerdemann and Nicolson (1963) and centrifuged in water at 3000 rpm for 3 minutes and in 70% sucrose at 2000 rpm for 2 minutes. The extracted spores were rinsed in running water on a 38 μ m mesh sieve. The spores were counted on a plate with concentric grooves under a stereo microscope. All 10 samples collected in each area were used for the analysis.

2.6. Soil fauna

The soil fauna was analyzed by means of two different methods: pit-fall traps and soil monoliths; soil monoliths are recommended by the "Tropical Soil Biology and Fertility" (TSBF) Program (Baretta et al., 2007a).

The setting of the traps aimed at collecting organisms from the meso and macrofauna with epigean ecologic

behavior (Baretta et al., 2007a). Each trap was filled with 200 mL of a solution composed of 15 mL of detergent per 1 liter of water, and the traps were introduced into the soil at the 10 previously established points for each area.

For the soil monoliths, five locations were randomly selected, and a monolith measuring 25 cm square and 30 cm deep was collected in each area, close to each coffee tree selected for general soil sampling. The soil organisms were manually separated and identified at the level of taxonomic orders, with the aid of a stereo microscope with 100 X magnification.

Shannon's diversity index, Simpson's dominance index, Pielou's evenness index, groups richness, richness estimate by the ACE method modified for more heterogeneous communities and the estimated sample coverage were calculated with the SPADE program (Chao and Shen, 2003).

2.7. Statistical analysis

Data obtained from the different analysis methods were submitted to analysis of variances (ANOVA) and the means of the microbial variables by the Duncan's post-hoc test and the density of the macrofauna by the LSD post-hoc test.

Canonical correlation analysis (CCA) was used to associate the different attributes and management systems. The biochemical and microbiological soil attributes were used in the CCA as environmental variables to be correlated with the density of the soil fauna groups for each management system. Analyses were performed using the software CANOCO (Leps and Smilauer, 2003).

3. Results

3.1. TOC, C-CO₂, qCO₂, MBC and MBN

The OC system had the highest TOC, followed by IB, CC, and finally the IA (p<0.05). With respect to C-CO₂, the greatest values were found in the IB, CC and OC, while

the smallest value was found in the IA. The IA had the highest MBC as well as OC which did not differ from IB and CC. The CC presented a higher MBN, followed by IA, OC and IB. CC had the lowest MBC/MBN ratio while IA had a MBC/TOC ratio superior to the others. IA had the lowest qCO $_2$ of all management systems (Table 1).

According to the CCA, TOC was the most efficient attribute to discriminate the four management systems. The other indicators (MBC, MBN and C-CO₂) were able to discriminate one or the other system, and only when all parameters were used together it was possible to discriminate all four systems (Figure 1).

3.2. Enzymatic activity

The IA area presented the lowest values and differed from OC for all enzymes evaluated (Table 1). Phosphatase and urease activity were greater in OC than in CC (p<0.05). CC and IB differed only in the phosphatase activity. In general, OC presented a higher enzymatic activity while CC, IB and IA showed lower values, which was similar to the pattern observed for TOC (Figure 2).

3.3. Mycorrhizal colonization and AMF spore density in the soil

In all samples the roots were colonized by AMF with the presence of hyphae, arbuscules and vesicles. The greatest colonization percentage was observed in IB areas (Table 1). No significant difference was found in the number of spores under different management systems using univariate analysis, though the CCA had greater number of spores associated with OC (Figure 1).

3.4. Soil fauna

Structural differences in the faunal communities (p<0.1) were verified with the use of both sampling methods, monoliths and traps (Tables 2 and 3).

Table 1. Soil attributes of coffee plantations under conventional (CC), organic (OC) and integrated management systems intercropped with *A. pintoi* (IA) or *B. decumbens* (IB).

| Analysis | Unit | CC | | OC | 1 | IA | | IB | | CV (%)a |
|-------------------|--|-------|---------|-------|----|-------|----|-------|----|---------|
| TOC | g kg ⁻¹ dry soil | 14.5 | c^{b} | 20.6 | a | 12.8 | d | 16.9 | b | 5.3 |
| MBC | μg g ⁻¹ dry soil | 287.4 | b | 350.0 | ab | 449.6 | a | 273.4 | b | 4.3 |
| MBN | μg g ⁻¹ dry soil | 52.6 | a | 29.3 | b | 30.3 | b | 18.6 | c | 15.6 |
| MBC/MBN | - | 5.6 | b | 12.1 | a | 15.1 | a | 16.4 | a | 14.2 |
| MBC/TOC | - | 11.5 | b | 9.9 | b | 20.5 | a | 9.4 | b | 9.9 |
| C-CO ₂ | ${ m mg~C\text{-}CO}_2~{ m kg}^{-1}~{ m dry}$ soil ${ m h}^{-1}$ | 8.7 | a | 7.4 | ab | 6.7 | b | 8.8 | a | 26.2 |
| qCO ₂ | mg C-CO ₂ g MBC ⁻¹ h ⁻¹ | 30.4 | a | 21.3 | a | 15.0 | b | 32.1 | a | 19.6 |
| Dehydrogenase | μg TTC g ⁻¹ h ⁻¹ | 12.2 | a | 10.4 | a | 6.7 | a | 9.7 | a | 32.8 |
| Urease | $\mu g \text{ N-NH}_4^+ g^{-1} h^{-1}$ | 3.7 | b | 5.7 | a | 2.7 | b | 3.2 | b | 15.5 |
| Phosphatase | mg PNP g ⁻¹ h ⁻¹ | 6.0 | b | 8.3 | a | 4.6 | b | 7.8 | a | 22.0 |
| Arylsulphatase | $\mu g \; PNP \; g^{-1} \; h^{-1}$ | 4.1 | ab | 4.9 | a | 3.9 | b | 4.4 | ab | 11.9 |
| Colonization | 0/0 | 19.7 | b | 18.8 | b | 23.0 | ab | 31.4 | a | 23.1 |
| Spores | spores 100 g ⁻¹ soil | 239.0 | a | 274.4 | a | 209.6 | a | 205.7 | a | 20.8 |

 $^{^{}a}$ CV = Coefficient of Variation (ANOVA). b Data followed by the same small letter in the line do not differ by the Duncan test (p<0.05).

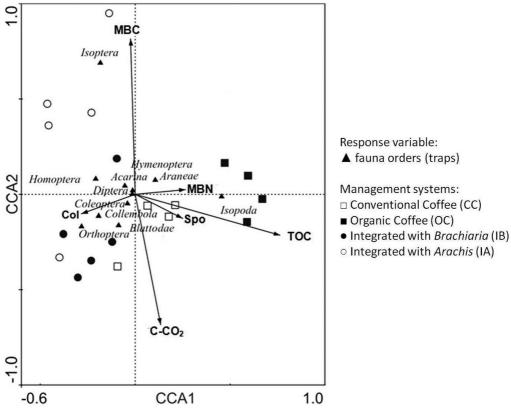


Figure 1. Canonical correlation analysis (CCA) associating biological and biochemical attributes to the management system used. Response variable: fauna groups (traps). Management systems: Conventional Coffee (CC); Organic Coffee (OC); Integrated with *Brachiaria* (IB); Integrated with *Arachis* (IA). Vectors indicate attributes used as explanatory variables: TOC, MBC, MBN, C-CO₂, Col (arbuscular mycorrhizal colonization) and Spo (AMF spore density). *Diptera* is larvae.

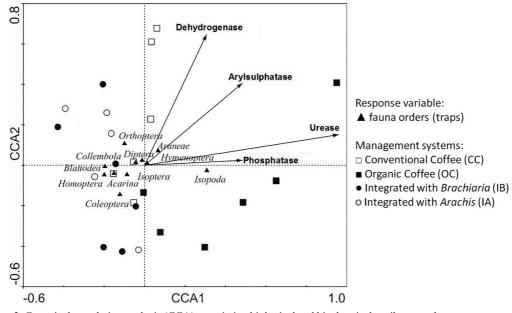


Figure 2. Canonical correlation analysis (CCA) associating biological and biochemical attributes to the management system used. Response variable: fauna orders (traps). Management systems: Conventional Coffee (CC); Organic Coffee (OC); Integrated with *Brachiaria* (IB); Integrated with *Arachis* (IA). Vectors indicate enzymes used as explanatory variables.

As for the soil monoliths, the great number of *Mollusca* and *Hymenoptera* in IB was obtained in only one of five replications. The same occurred with the *Isoptera* in IA and with *Hymenoptera* in CC. The best reproducibility of the data for the different repetitions occurred with *Coleoptera* and *Oligochaeta*, where the former was notably

less numerous in OC while the latter was clearly more numerous in OC (Table 2).

The traps presented highly reproducible data for the different replications, and displayed a greater fauna richness and diversity compared with the monolith method (Table 4). Using the trap method, there was a greater occurrence of *Acarina* in IB and *Araneae* in CC, and a smaller presence

Table 2. Soil macrofauna obtained from soil monoliths sampled under coffee trees under conventional (CC), organic (OC) and integrated management systems intercropped with *A. pintoi* (IA) or *B. decumbens* (IB), compared with values reported from different coffee plantations by other authors (Velasquez et al., 2007) compared coffee intercropped with *Schizolobium amazonicum and Hevea brasiliensis*; Barros et al. (2002) single cropping).

| | | | 7 | his | worl | ζ. | - | | | Velasquez et a | ıl. (2007) | Barros et al. |
|---------------------|----|---------|-------|------|--------|------|-----|----|-----|----------------------------|------------------------------|---------------|
| Group | C | C | О | C | L | A | IJ | В | CV | S. amazonicum ^a | H. brasiliensis ^a | (2002) |
| | | (| indiv | idua | als pe | er m | 2) | | (%) | (in | dividuals per m²) | |
| Araneae | 3 | a^{b} | 13 | a | 10 | a | 6 | a | 173 | 10 | 45 | 29 |
| Blattodea | 0 | a | 0 | a | 3 | a | 3 | a | 316 | - | - | - |
| Chilopoda | 0 | a | 9 | a | 3 | a | 13 | a | 240 | 9 | 38 | 13 |
| Coleoptera | 26 | ab | 0 | b | 22 | ab | 32 | a | 117 | 56 | 61 | 90 |
| Diptera | 0 | a | 0 | a | 3 | a | 0 | a | 447 | 13 | - | - |
| (larvae) | | | | | | | | | | | | |
| Diplopoda | - | | - | | - | | - | | - | 4 | 86 | 58 |
| Homoptera | - | | - | | - | | - | | - | 1 | 99 | 32 |
| Hymenoptera | 61 | a | 3 | a | 16 | a | 268 | a | 329 | 563 | 1322 | 237 |
| Heteroptera | 10 | a | 3 | a | 3 | a | 3 | a | 197 | 4 | - | - |
| Isoptera | 0 | a | 0 | a | 16 | a | 0 | a | 447 | 4 | 262 | 1491 |
| Lepidoptera | 3 | ab | 0 | b | 10 | a | 0 | b | 250 | 1 | - | - |
| (larvae) | | | | | | | | | | | | |
| Mollusca | 10 | a | 42 | a | 22 | a | 38 | a | 204 | 2 | 6 | 4 |
| Oligochaeta | 48 | ab | 96 | a | 3 | b | 42 | ab | 169 | 249 | 90 | 109 |
| Opiliones | 0 | a | 0 | a | 0 | a | 3 | a | 447 | - | - | - |
| Orthoptera | - | | - | | - | | - | | | 1 | 29 | 6 |
| Others ^c | - | | - | | - | | - | | | 60 | 16 | 10 |
| n ^d | 5 | | 5 | | 5 | | 5 | | | 8 | 5 | 5 |

^aCoffee trees intercropped with *S. amazonicum* or *H. brasiliensis*. ^bNumbers with the same letter in a row do not differ significantly (p<0.1). ^cOthers: Sum of organisms of groups appearing less frequently. ^dn = number of replicates.

Table 3. Soil fauna sampled with traps under coffee trees under conventional (CC), organic (OC) and integrated management systems intercropped with *A. pintoi* (IA) or *B. decumbens* (IB).

| Cwarm | C | C | 0 | C | II | В | IA | 4 | CV |
|----------------------|------|-----|------|----------|-------------|----|------|----|-----|
| Group | | | (i | ndividua | ls per trap |) | | | (%) |
| Acarina | 2.9 | abª | 1.2 | b | 3.1 | ab | 6.1 | a | 117 |
| Araneae | 2.0 | a | 1.3 | ab | 0.9 | b | 0.3 | b | 112 |
| Blattodea | 0.1 | b | 0.1 | b | 0.9 | b | 0.3 | ab | 211 |
| Coleoptera | 0.4 | ab | 0.2 | b | 0.4 | ab | 0.9 | a | 136 |
| Collembola | 25.3 | a | 6.2 | b | 16.1 | ab | 24.3 | a | 70 |
| Diptera (larvae) | 26.3 | a | 13.4 | b | 12.7 | b | 26.7 | a | 54 |
| Homoptera | 0.1 | b | 0.2 | b | 4.0 | a | 1.6 | b | 161 |
| Hymenoptera | 8.2 | a | 9.2 | a | 13.8 | a | 12.0 | a | 74 |
| Isopoda | 3.2 | b | 62.1 | a | 0.0 | b | 0.0 | b | 150 |
| Isoptera | 0.0 | b | 0.0 | b | 0.7 | a | 0.0 | b | 300 |
| Lepidoptera (larvae) | 0.2 | a | 0.2 | a | 0.2 | a | 0.3 | a | 182 |
| Opiliones | 0.0 | a | 0.2 | a | 0.1 | a | 0.2 | a | 311 |
| Orthoptera | 0.4 | ab | 0.0 | b | 1.0 | a | 0.4 | ab | 169 |

^aNumbers with the same letter in a row are not significantly different ($p \le 0.1$).

Table 4. Mean ± standard deviation of diversity and richness indices of the macrofauna sampled with monoliths or traps under coffee trees under conventional (CC), organic (OC) and integrated management systems intercropped with A. pintoi (IA) or B. decumbens (IB).

| Fauna survey | | | | | Diversity indices | ses | | | | | Richness indices | ndices | 70 | |
|----------------|-------------|------------------|-------------|----|-------------------|-----|-------------------|----|----------------------------|---|------------------|--------|--------------|----|
| methods/ Areas | ESC | | Shannon | | Simpson | | Pielou's evenness | | \mathbf{SR}^{b} | | ACE ° | | Chao-1bc ° | |
| MONOLITHS | | | | | | | | | | | | | | |
| CC | 0.82 + 0.19 | a^{d} | 0.96 + 0.15 | В | 0.35 + 0.12 | þ | 0.79 + 0.19 | ap | ab $3.5 + 0.5$ | а | 5.85 + 1.75 | а | 4.13 + 0.89 | B |
| OC | 0.87 + 0.17 | а | 0.41 + 0.37 | þ | 0.74 + 0.22 | а | 0.49 + 0.40 | þ | 1.8 + 0.7 | þ | 1.74 + 2.60 | þ | 2.00 + 1.09 | þ |
| IA | 0.59 + 0.36 | а | 1.21 + 0.30 | В | 0.33 + 0.09 | þ | 0.95 + 0.03 | В | 3.8 + 1.2 | а | 5.44 + 3.66 | ap | 4.60 + 1.39 | В |
| IB | 0.56 + 0.46 | а | 0.82 + 0.44 | ap | 0.54 + 0.23 | ap | 0.73 + 0.30 | ap | 3.6 + 1.6 | а | 3.28 + 2.98 | ap | 4.50 + 1.67 | В |
| TRAPS | | | | | | | | | | | | | | |
| CC | 0.98 + 0.02 | а | 1.30 + 0.17 | ap | 0.33 + 0.06 | þ | 0.74 + 0.07 | В | 6.0 + 1.3 | а | 7.93 + 5.45 | þ | 6.67 + 2.15 | þ |
| OC | 0.97 + 0.02 | а | 1.12 + 0.31 | þ | 0.45 + 0.17 | а | 0.60 + 0.17 | þ | 6.4 + 1.0 | а | 12.19 + 10.14 | ap | 8.50 + 3.21 | ab |
| IA | 0.90 + 0.11 | а | 1.47 + 0.22 | В | 0.30 + 0.08 | þ | 0.76 + 0.07 | В | 7.1 + 1.7 | а | 20.13 + 15.88 | а | 10.22 + 5.52 | В |
| IB | 0.97 + 0.02 | а | 1.30 + 0.25 | ap | 0.35 + 0.12 | þ | 0.70 + 0.10 | а | 6.4 + 1.2 | а | 11.10 + 5.98 | ap | 7.50 + 1.84 | ab |

***ESC – Estimated Sample Coverage. ***PSR – Species richness of soil fauna groups. **ACE – Method according to Chao and Shen (2003). ***Immbers with the same letter in the column to each method are not different by the Duncan post-hoc test ($p \le 0.1$).

of *Collembola* in OC. The presence of *Diptera* was greater in CC and IB, while *Homoptera* were present in larger numbers in IA and IB. However, the most evident finding was the high density of *Isopoda* in OC (Table 3).

In general, soil monoliths data in OC had the lowest richness and diversity values, while IA had the highest diversity. The fauna sampled by traps was apparently more reliable to evaluate richness, diversity and equitability values in the different areas, since it presented lower standard deviation (Table 4).

3.5. Canonical correlation analysis (CCA)

The result of the CCA correlating edaphic fauna obtained with the trap method (response variable) and biological attributes (explanatory variables) was shown to be useful in distinguishing between management systems and to visualize the relationships among variables (Figure 1). In this analysis the separation of the soil management systems according to the TOC content became clear. The OC was characterized by a greater TOC and higher *Isopoda* values. The CC showed the greatest MBN values and a different faunal composition from the OC. The IA was positively correlated with greater MBC values and greater density of organisms belonging to *Isoptera*.

The evaluated enzymes did not show a good correlation with all the soil attributes data together, and, therefore, an independent analysis was performed (Figure 2). The OC was related with the enzymes urease, phosphatase and *Isopoda* and all the other samples and variables were not clearly discriminated.

The CCAs that were calculated with the data from the monolith method were unable to discriminate among the management systems studied and are not shown.

4. Discussion

4.1. TOC, C-CO, MBC and MBN

The main difference among the studied management systems is the TOC content. The highest TOC level was found in OC, and this certainly is a consequence of the greater addition of organic materials, grain husks and castor bean cake, which have a C/N ratio around 31/1 and 10/1, respectively (Conceição et al., 2005; Monaco et al., 2008). There was addition of organic matter in the CC as well, but only in the form of a much smaller amount of coffee husk, while in the IA and IB there was the addition of leaf residues from *A. pintoi* and *B. decumbens* plants with a C/N ratio of about 30/1 and 60/1 respectively.

Comparing the two cover crops, the greater TOC level in the IB than IA is most probably related to a greater primary production and a greater C/N ratio of the residues produced, thus promoting the buildup of carbon in the soil when grasses are used as cover. Legumes, on the other hand, experience a faster decomposition and a smaller accumulation in the soil (Oliveira et al., 2008; Thomas and Asakawa, 1993). In this study, N from *A. pintoi* was not quantified, but legume crop residues, depending on type and quality, usually contain from 80-150 kg ha⁻¹ of

N (Shah et al., 2003), which favors the mineralization of organic residues and consequently results in a smaller accumulation of carbon in the soil.

The addition of residues with a low C/N ratio tends to stimulate microbial biomass growth, as observed for MBC in the IA and OC. Moreover, organic management usually shows higher level of MBC when compared to conventional management system (Ge et al., 2011, 2013; Lagomarsino et al., 2009; Mäder et al., 2002). The MBC could be used as an indicator for the most sustainable management system on coffee production. This is strengthened by the fact that among some biological soil attributes, Partelli et al. (2012) found the MBC as the variable with higher relative contribution to the discrimination between conventional and organic production system of *Coffea canephora*.

Different effects have, however, been observed depending on the type of residue added to the soil. As for MBN, the highest value was observed in IA and that may be due to the immobilization of nitrogen in the microbial biomass as a result of the addition of coffee husk with a high C/N ratio (31/1).

The basal respiratory rate, as well as the specific biomass respiration, $q\text{CO}_2$, is also influenced by the quality of the residue added to the soil. Thus, residues with a greater C/N ratio cause a higher respiration rate during the decomposition of these residues (Nogueira et al., 2006), as observed in CC and IB.

4.2. Enzymatic activity

Soil enzymes are influenced by a number of natural and anthropogenic factors which considerably modify their properties and performance (Chaer et al., 2009; Rao et al., 2014). Practices which favor microbial activity, such as organic fertilization, crop rotation, conservationist management and between-row cover also favor a greater enzymatic activity (Balota and Chaves, 2010; Bowles et al., 2014; Gajda and Martyniuk, 2005).

In general terms, a greater enzymatic activity was found in the organic management area (Table 1, Figure 2). The use of organic residues as coffee husk and castor bean cake for fertilization stimulated the activity of enzymes which act on the decomposition of organic residues, such as, arylsulphatase, urease and phosphatase (Liang et al., 2014; Tu et al., 2006). Dehydrogenase activity is related to microbial activity in general and is stimulated by the addition or prevalence of organic matter in the soil (Liang et al., 2014; Nogueira et al., 2006). In fact, the higher values were associated with higher TOC, and the lowest value was detected in IA that also presented the lowest TOC.

4.3. Mycorrhizal colonization and AMF spore density in the soil

AMF colonization depends on factors such as the AMF species present, the edapho-climatic conditions, the plant cultivar and the land management in use (Andrade et al., 2009). However, most of the studies reporting colonization on coffee were done in inoculation treatments or under greenhouse

conditions (Andrade et al., 2010; Vaast et al., 1997; Siqueira et al., 1998), and there is scarcity of data on field evaluations. From this study, the colonization level of AMF ranged from 18 to 31% in coffee trees growing in intercropped, conventional or organic management.

The low levels of mycorrhizal colonization (most treatment around 20%) could be explained by a satisfactory P nutrition of the plants. *Brachiaria* is a good host for the multiplication of AMF, and this may explain why there was a higher percent colonization in coffee plantations intercropped with this grass, although no differences in spore numbers were shown.

Differences in crop management can influence the spore number and colonization levels (Bainard et al., 2011). Besides, depending on the host plant and AMF species, there could be different strategies for colonization and sporulation of AMF and, sometimes, a change in one of these factors could alter the colonization behavior of the AMF. Coffee plant shaded by leguminous trees stimulates the spore number in soil compared with non-leguminous ones (Muleta et al., 2007). Intercropping also could increase the colonization level and the benefits from the arbuscular mycorrhiza (Bainard et al., 2011).

However, our results indicate that most of the variables did not affect the mycorrhizal status in coffee plants; only the higher colonization level in coffee plantations integrated with *Brachiaria* sp. was clearly correlated. The dense root system of *Brachiaria* sp. increases the chances for association with AM fungi, thereby producing more diverse propagules in soil and enhancing the possibility to colonize the coffee tree roots (Bainard et al., 2011).

4.4. Soil fauna

Faunal community structure was more important to differentiate the management systems than diversity. While diversity and richness indexes were not good to separate the management systems, since the values were quite similar (Table 4), the community's structures were clearly different (Tables 2 and 3, Figure 1).

Analyzing the differences of faunal community structure by the monolith method, Oligochaeta, Coleoptera and Lepidoptera (larvae) showed most evident differences among the sampled systems, comparing with the trap technique. Oligochaeta presented higher occurrence in OC. It can be explained by the absence of pesticides and a greater amount of soil organic matter. In fact, a greater occurrence of Oligochaeta in more sustainable management systems, such as the OC, would be expected, since this group is considered a good indicator of soil quality (Baretta et al., 2007b; Bartz et al., 2013). Coleoptera abundance was lower in OC, and its faunal group can be influenced by shadow level, which may be an explanation for the higher abundance in IB areas, where shadow level was higher due the green manure presence (Henderson and Roitberg, 2006; Nestel et al., 1993).

When sampling was performed with the monolith technique, different authors observed great variations in

the structures of the soil faunal communities (Table 2), restricting the possibility of identification of typical patterns (Barros et al., 2002; Velasquez et al., 2007). Thus, the use of monoliths in studies in which the macrofauna will be used as an indicator of soil quality under coffee trees may sometimes not be the best option. In spite of the low reproducibility and the difficulty in finding patterns, the monolith is one of the most frequently used methods to study soil fauna, because it looks at all kinds, not only the more mobile epigeous fauna (Baretta et al., 2007a, b; Barros et al., 2002; Bignell, 2009; Velasquez et al., 2007). But, since it is a technique that is laborious and highly variable, sampling a greater number of replicates becomes necessary and this constitutes a major limitation to this method (Baretta et al., 2007a).

The trap method, although limited to certain groups, is operationally easier; the data obtained had greater reproducibility and provided clearer patterns of the edaphic fauna. This methodology is more reliable in comparative studies, especially when considering organisms with greater mobility in the surface (Baretta et al., 2007a).

By analyzing the faunal structures with the trap method, the high density of *Isopoda* in the OC was the most evident aspect of the soil fauna evaluation, indicating areas with low pesticide use and with high organic matter content (Karanja et al., 2009; Paoletti, 1999). In the IA area a greater occurrence of Isoptera was correlated with a higher MBC, which may be related to higher presence of fungus in these areas (Jouquet et al., 2005; Riggins et al., 2014). The reduced presence of Collembola and Coleoptera in the organic system was a surprise, since they are usually correlated with a high organic matter content and absence of pesticides. Another unexpected result was the larger populations of Araneae, Collembola and Diptera (larvae) in areas where pesticides were applied, since these groups are usually considered as sensitive indicators (Maluche-Baretta et al., 2006).

4.5. Canonical correlation analysis (CCA)

Although the CCA proved to be a good tool to visualize the community structure and the interactions of the soil fauna, it produced a more reliable graph, when only the data of the trap method were used (Figure 1). TOC was the soil attribute most influenced by the different management systems. TOC content was higher for the OC, and lower in IA, and also influenced the other soil attributes. Of all orders identified, *Isopoda* and *Isoptera* stand out. *Isopoda* was an indicator of OC, being associated with high TOC values, while *Isoptera* was an indicator of IA, being associated with high MBC values (Jouquet et al., 2005; Karanja et al., 2009; Paoletti, 1999). For the enzymes, it was possible to visualize a higher activity in the OC, and again the relationship between *Isopoda* and OC was detected (Figure 2).

Since the biological attributes were mainly driven by TOC, and in general were higher in OC, it is very likely that increasing the input of organic matter in the other

management systems will result in a higher biological activity.

5. Conclusions

TOC content was the soil attribute most influenced by the different coffee management systems, being the greatest content found in OC and the lowest in IA. TOC also influenced the other attributes evaluated. Faunal structure was more sensitive to distinguish the management systems than the other biological parameters. Among the faunal organisms, *Isopoda* was the best indicator of OC, being associated with high TOC values, while *Isoptera* was the group linked to IA, being associated with high MBC values. The CC and IB were more similar to each other, and discrimination of all four systems was only possible when the attributes were individually analyzed.

Little is reported about these attributes and its relations to coffee plantations, and an increasing data base will be important to establish parameters of biological sustainability in soils.

The management systems had distinct values for the different attributes, however, OC had higher values for most of the biological attributes and higher number of *Oligochaeta* and *Isopoda*, important soil quality indicators, corroborating the concept that OC is a more sustainable system.

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