

Changes in Soil Organic Carbon Fractions in Response to Cover Crops in an Orange Orchard

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ABSTRACT: The cultivation of cover crops intercropped with fruit trees is an alternative to maintain mulch cover between plant rows and increase soil organic carbon (C) stocks. The objective of this study was to evaluate changes in soil total organic C content and labile organic matter fractions in response to cover crop cultivation in an orange orchard. The experiment was performed in the state of Bahia, in a citrus orchard with cultivar 'Pera' orange (*Citrus sinensis*) at a spacing of 6 × 4 m. A randomized complete block design with three replications was used. The following species were used as cover crops: Brachiaria (*Brachiaria decumbes*) – BRAQ, pearl millet (*Pennisetum glaucum*) – MIL, jack bean (*Canavalia ensiformis*) – JB, blend (50 % each) of jack bean + millet (JB/MIL), and spontaneous vegetation (SPV). The cover crops were broadcast-seeded between the rows of orange trees and mechanically mowed after flowering. Soil sampling at depths of 0.00-0.10, 0.10-0.20, and 0.20-0.40 m was performed in small soil trenches. The total soil organic C (SOC) content, light fraction (LF), and the particulate organic C (POC), and oxidizable organic C fractions were estimated. Total soil organic C content was not significantly changed by the cover crops, indicating low sensitivity in reacting to recent changes in soil organic matter due to management practices. Grasses enabled a greater accumulation of SOC stocks in 0.00-0.40 m compared to all other treatments. Jack bean cultivation increased LF and the most labile oxidizable organic C fraction (*F1*) in the soil surface and the deepest layer tested. Cover crop cultivation increased labile C in the 0.00-0.10 m layer, which can enhance soil microbial activity and nutrient absorption by the citrus trees. The fractions LF and *F1* may be suitable indicators for monitoring changes in soil organic matter content due to changes in soil management practices.

Keywords: citriculture, lability, light fraction, oxidizable carbon, soil carbon pools.

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INTRODUCTION

Sustainable management of agricultural soils requires, among other factors, the maintenance and/or a gradual increase in organic matter content, which would enhance soil fertility by nutrient supply, improvements in the soil structure and the maintenance of microbial activity (Johnston et al., 2009; Tobiašová, 2011).

One of the basic prerequisites for sustainable management of an agricultural system is the maintenance of soil cover. Thus, growing crops such as legumes and/or grasses for soil cover is a viable agricultural practice (Caetano et al., 2013). The success of this management strategy depends on the choice of adequate cover crop species that meet the criteria of adaptability to the climatic and agricultural conditions of the region of use. This selection must also consider other characteristics such as ease of control, suitable shoot phytomass production, ease of seed acquisition, high potential for soil cover, and slow residue degradation after harvest (Souza et al., 2013).

Currently, the soil cover between tree rows in commercial orange orchards in the state of Bahia is not prioritized as soil management practice. Contrary to what is considered ideal for soil conservation, disc plowing between the tree rows to eliminate weeds, leaving the soil completely bare, is a normal practice. The rationale for this practice is the empirical observation that this process increases productivity by suppressing weed growth, which in turn decreases competition for water. However, bare soil can enhance the potential for soil and nutrient losses by erosion and oxidation of soil organic matter, increase atmospheric emissions of CO₂-C, and destroy the soil structure (Hernani et al., 1999; Johnston et al., 2009; Tobiašová et al., 2011; Xavier et al., 2013).

In spite of the benefits to the soil, the cultivation of cover crops in-between the rows of orange orchards is still incipient. Studies in orange orchards in the state of Bahia by Carvalho et al. (2003a,b; 2006) showed cover crop cultivation, along with adequate soil tillage, improved both the soil physical and chemical properties and increased productivity. Balota and Auler (2011) also observed improvements in the microbiological soil profile when cover crops were grown in an orange orchard in the state of Paraná, Brazil. However, even though Poeplau and Don (2015) demonstrated that cover crops represent an important management strategy to increase organic C stocks in agricultural soils, this practice has been neglected and its advantages were not adequately quantified.

The rate of SOC accumulation depends mainly on the quantity of dry matter produced by the cover plants (Gonçalves and Ceretta, 1999) and on environmental factors such as humidity and temperature (Kirschbaum, 2006). The change in total SOC content depends on agricultural management practices and is not always detectable in the short term (Xavier et al., 2013). Thus, the partitioning of soil organic matter into the functional compartments with different dynamics represents an important tool to readily detect recent changes in the soil in response to changes in management practices (Sequeira et al., 2011; Blanco-Moure et al., 2013). The labile fractions of soil organic matter such as LF, POC and C fractions extracted using low degrees of oxidation with H₂SO₄ (Chan et al., 2001), may be more sensitive indicators of the changes caused by modifications in soil management practices (Loss et al., 2013; Souza et al., 2014; Marques et al., 2015), and an analysis of these compartments will deepen the understanding of SOC dynamics (Carter, 2001).

The maintenance of soil cover between tree rows in commercial orange orchards by cover crops represents an alternative method for increasing organic C sequestration in the soil. Hence, the identification of one species or a combination of plants that can increase soil organic C stocks when used as cover crops is essential for a successful management of orange orchards. Thus, based on the hypothesis that the introduction of cover crops between the rows of orange trees leads to changes in SOC levels, the objective of this study was to assess the SOC content and C-stocks and measure labile fractions of soil organic matter in response to cover crop cultivation in an orange orchard.

MATERIALS AND METHODS

The study was carried out on the Fazenda Lagoa do Coco, in the municipality of Rio Real (11° 27' 52" S, 37° 56' 11" W, 186 m asl), on the northern coast of the state of Bahia, Brazil. According to the Köppen classification, the climate is predominantly As, hot (18 to above 35 °C). In the driest month, pluvial precipitation is less than 60 mm. Summers are dry, the mean annual rainfall is 1,000 mm; the wettest months of the year are May through July, whereas the period from October to December is the driest. The mean annual temperature is 24 °C (Santana et al., 2006). The soil of this orchard was classified as cohesive *Latossolo Amarelo Álico* (Haplortox) (Carvalho et al., 2002). At the time of the experiment, the orchard was about eight years old, consisting of trees resulting from grafting orange 'Pera' onto lemon 'Cravo', at a spacing of 6 × 4 m. Previously, this area had been used as orange orchard for 15 years, and the trees were renewed by planting seedlings at their definite places. The experimental plots covered an area of 840 m², with 48 plants per plot. The total experimental area was approximately 12,600 m², and the experiment used a completely randomized block design.

Soil tillage prior to sowing of the cover crops between the tree rows in the orchard consisted of mechanical mowing of the spontaneous vegetation followed by passing a disc harrow in the upper soil layer. The cover crops were sown by hand in the entire area between the orange tree rows and the seeds were surface-incorporated with a disc harrow.

The cover crops evaluated in the experiment were: T1 - Brachiaria (*Brachiaria decumbens* Stapf) (BRAQ), T2 - Pearl millet (*Pennisetum glaucum* R.Br.) (MIL), T3 - Jack bean (*Canavalia ensiformis* (L.) DC.) (JB), T4 - blend of jack bean + millet (JB/MIL) in equal proportions (50% each), and T5 - Spontaneous vegetation (SPV). The cover crops were planted at the beginning of the rainy season (May-June 2013), and were mowed 90 days after sowing, corresponding to the period of full flowering. Mowing was performed in a way that left the shoot phytomass residues below the soil surface after mowing. This study evaluated the effects of only one cultivation cycle of cover crops.

Soil samplings at depths of 0.00-0.10, 0.10-0.20 and 0.20-0.40 m were performed 30 days after mowing of the cover crops. The main soil physical and chemical properties in the orchard are presented in table 1.

The total SOC content was quantified by wet digestion of the soil samples with a mixture of potassium dichromate and sulfuric acid (H₂SO₄) with external heating (Yeomans and Bremner, 1988). The soil bulk density in the different soil layers was measured by the volumetric ring method (Blake and Hartge, 1986). The SOC stock was calculated according to the equation:

$$\text{SOC stock (Mg ha}^{-1}\text{)} = [\text{SOC}\%] \times \text{BD} \times \text{T}$$

where [SOC] is the concentration of total organic C, in dag kg⁻¹; BD is soil bulk density, in Mg m⁻³; and T is the thickness of the layer, in cm.

The soil light fraction (LF) was extracted by density fractionation using NaI according to the method adapted from Sohi et al. (2001). Briefly, 6.5 g of sieved, air-dried soil samples (<2.00 mm mesh) were suspended in 30 mL NaI solution at a density of 1.8 Mg m⁻³, shaken, and subsequently centrifuged at 3,000 rpm for 15 min. The supernatant was then vacuum filtered and the remaining solution collected for reuse. The excess of NaI retained in the filter was removed by rinsing it thoroughly with distilled water. The organic particles trapped in the filter constitute the soil LF, and were oven-dried at 60 °C for 72 h and then weighed. We considered the total LF content as being the sum of the free and occluded LF. Extraction and quantification of LF were performed in duplicate.

The particulate organic carbon (POC) content was determined by physical fractionation, according to the method adapted from Cambardella and Elliott (1992). Approximately 10 g of sieved (<2.00 mm) air-dried soil was mixed with 30 mL sodium hexametaphosphate (5 g L⁻¹) and shaken for 15 h in a vertical shaker. Subsequently, the suspension was passed through

Table 1. Physical and chemical properties of a cohesive *Latossolo Amarelo Álico* (Haplortox) in the 0.00-0.10, 0.10-0.20 and 0.20-0.40 m layers under orange trees

Property	0.00-0.10 m	0.10-0.20 m	0.20-0.40 m
Sand (g kg ⁻¹) ⁽¹⁾	900	900	860
Silt (g kg ⁻¹) ⁽¹⁾	30	3	20
Clay (g kg ⁻¹) ⁽¹⁾	70	97	120
Bulk density (Mg m ⁻³)	1.48	1.66	1.66
pH(H ₂ O)	5.9	5.8	5.7
P (mg dm ⁻³) ⁽²⁾	30.9	24.3	20.3
K (cmol _c dm ⁻³) ⁽²⁾	0.17	0.15	0.13
Ca (cmol _c dm ⁻³) ⁽³⁾	1.63	1.57	1.38
Mg (cmol _c dm ⁻³) ⁽³⁾	0.56	0.54	0.49
Ca+Mg (cmol _c dm ⁻³) ⁽³⁾	2.18	2.11	1.87
Al ³⁺ (cmol _c dm ⁻³) ⁽³⁾	0.03	0.03	0.05
H+Al (cmol _c dm ⁻³)	1.98	1.96	2.17
Sum of bases (cmol _c dm ⁻³)	2.38	2.28	2.02
CEC (cmol _c dm ⁻³) ⁽⁴⁾	4.32	4.24	4.19
Base saturation (%)	54	53	47

⁽¹⁾ Pipette method; ⁽²⁾ Extracted by Mehlich-1; ⁽³⁾ Extracted by 1.0 mol L⁻¹ KCl; ⁽⁴⁾ Cation exchange capacity.

a 0.053-mm sieve using a water jet. The material retained in the sieve, consisting of POC associated to sand fraction, was oven-dried at approximately 50 °C and the relative weight quantified. Differing from the original method, we measured the C content of the material retained in the sieve (POC + sand) by wet digestion with a mixture of potassium dichromate and H₂SO₄ with external heating, as mentioned above (Yeomans and Bremner, 1988). The quantity of organic C associated with the mineral fraction (mAOC) was calculated as the difference between the total SOC and POC content. To determine the proportion of soil POC (g POC kg⁻¹ of soil), we quantified the sand fraction as a proportion of total soil mass.

The fractionation of total SOC was determined using an aqueous sulfuric acid solution under a gradient of oxidizing conditions, according to the method adapted from Chan et al. (2001). Organic C fractions were quantified by oxidation using 0.167 mol L⁻¹ K₂Cr₂O₇ in an acidic medium containing the different H₂SO₄ concentrations without external heating (Walkley and Black, 1934). The amount of oxidizable organic C was estimated using 2.5, 5, and 10 mL of concentrated sulfuric acid with final H₂SO₄ concentrations of 3, 6, and 9 mol L⁻¹, while the potassium dichromate concentration was maintained constant. The organic C concentration determined by this three acid-aqueous solutions method allowed separation of SOC into the following four fractions (*F*) of decreasing oxidizability/labability:

F1 (very labile): oxidizable organic C below a H₂SO₄ concentration of 3 mol L⁻¹;

F2 (labile): difference between oxidizable organic C extracted at 6 and 3 mol L⁻¹ H₂SO₄;

F3 (less labile): difference between oxidizable organic C extracted at 9 and 6 mol L⁻¹ H₂SO₄; and

F4 (recalcitrant): difference between total SOC and oxidizable organic C at 9 mol L⁻¹ H₂SO₄.

The organic C recovered in the *F1* fraction was considered as labile C (C_L), while the sum of organic C in the *F3* + *F4* fractions represented non-labile C (C_{NL}) (Xavier et al., 2009). Thus, the labability index of organic C in the soil was calculated as C_L/C_{NL}.

The statistical analyses were performed using analysis of variance for a randomized block design. When the results of the F test were significant, the treatment means were compared by the Scott-Knott test at a significance level of p<0.05. The software package ASSISTAT 7.7 (Silva and Azevedo, 2006) was used for statistical analyses.

RESULTS AND DISCUSSION

Total soil organic carbon storage

The SOC levels (Table 2) were low, ranging between 6.7 and 13.3 g kg⁻¹. Such low levels of SOC may be due to the low physical-colloidal protection of organic matter due to the predominance of sand in the soil (Table 1), and the presence of a large sand fraction also facilitates the processes of organic C oxidation. In general, SOC levels were higher in the deeper than the upper layers, suggesting vertical movement of organic matter, which is facilitated by sandy texture of the soil.

The effect of cultivating different cover crops on SOC content was not readily apparent, as no definite increase was observed in total SOC that could be attributed to a specific cover crop. There was no significant effect of cover crop cultivation on SOC levels in the 0.00-0.10 m layer. However, in the 0.10-0.20 m layer, the JB/MIL combination induced lower SOC levels than BRAQ and JB, while in the 0.20-0.40 m layer, compared to all other cover crops, grasses significantly increased SOC contents. Given the contribution of the root system to maintaining SOC levels, Bressan et al. (2013) evaluated the effect of grasses on the chemical quality of *Latosolo Amarelo* (Oxisol) in the Cerrado biome and, similar to the results reported here, found that millet and Brachiaria species increased organic matter levels to a depth of 0.40 m. The SOC levels in the SPV were similar to those of the other cover crops in the 0.00-0.10 m layer, and did not differ from JB/MIL treatment in the 0.10-0.20 m layer or from that of JB/MIL and JB in the 0.20-0.40 m layer. These results indicate that an adequate management of native spontaneous vegetation is as important as cover crops for maintaining soil organic C levels, and that the removal of this vegetation, which is a common practice in commercial citrus orchards, will not only expose the soil but also result in the loss of soil organic C and nutrients.

A significant increase in SOC stocks due to cultivation of cover plants was observed only in the 0.20-0.40 m layer (Table 2). At this depth, SOC stocks were significantly increased by BRAQ and MIL (mean increase of 9.8 Mg ha⁻¹ C), compared to the other treatments. A soil profile of SOC stocks for all sampled layers (0.00-0.40 m) showed that grass cultivation resulted in a significant increase in SOC levels (20 %), compared to the other treatments (Table 2). This response was probably due to the addition of organic matter from the abundant root system of these grasses.

Table 2. Soil organic carbon (SOC) contents and stocks in the 0.00-0.10, 0.10-0.20 and 0.20-0.40 m layers in an orange orchard with different cover crops

Cover crop ⁽¹⁾	0.00-0.10 m	0.10-0.20 m	0.20-0.40 m	0.0-0.40 ⁽²⁾ m
SOC (g kg ⁻¹)				
BRAQ	7.97 a	11.22 a	12.60 a	-
MIL	8.58 a	12.05 a	13.34 a	-
JB	6.75 a	7.73 b	10.40 b	-
JB/MIL	7.24 a	11.04 a	8.56 b	-
SPV	8.08 a	9.48 b	10.76 b	-
SOC (Mg ha ⁻¹)				
BRAQ	12.16 a	18.78 a	40.50 a	71.44 a
MIL	12.52 a	19.31 a	45.23 a	77.06 a
JB	9.45 a	18.33 a	28.33 b	57.55 b
JB/MIL	10.89 a	12.88 a	34.81 b	57.15 b
SPV	12.28 a	16.05 a	36.04 b	64.37 b

⁽¹⁾ BRAQ: brachiaria; MIL: pearl millet; JB: jack bean; JB/MIL: blend of JB+MIL in a proportion of 50 %; SPV: spontaneous vegetation. ⁽²⁾ Sum of SOC contents from 0.0 to 0.40-m. (-) not measured. Means followed by the same lower-case letters in a column, within each soil layer, do not differ significantly by the Scott-Knott test ($p < 0.05$).

Soil light fraction

The light fraction (LF) content in the 0.00-0.10 m layer ranged from 1.69 to 15.76 g kg⁻¹ (Figure 1). The cultivation of BRAQ and JB resulted in the highest LF concentration, which was, in the mean, 78 % higher than that observed for the other cover crops. Compared to the control treatment (SPV), the percentage of LF content increased 83 and 89 %, respectively, in response to BRAQ and JB. In BRAQ treatment, a combination of higher shoot phytomass production compared to the other treatments with the slow rate of biomass decomposition (data not shown), probably increased the LF levels. The LF is derived from the decomposition of plant and animal residues by the soil microbiota. It consists of organic debris in different decomposition stages, as well as of C forms derived from pyrogenic coal (Janzen et al., 1992; Murage et al., 2007). This compartment can be considered a sensitive indicator of soil management for responding more quickly to the changes in agricultural practices compared to total SOC (Marin et al., 2006; Maia et al., 2007). An increase in soil LF was attributed to the quantity and quality of shoot biomass left in the soil and to a very high proportion of fine roots by Bu et al. (2012). This observation may explain the results obtained in this study with BRAQ cultivation, since *Brachiaria* has a fasciculate root system with a high amount of fine roots. In the case of JB, the increase of LF may be due to the faster decomposition of the shoot phytomass (data not shown) which would lead to a more rapid accumulation of LF. Contrary to SOC levels (Table 2), LF levels appear to be more responsive to the effects of cover crops in the 0.00-0.10 m layer, confirming that this compartment can be used as a sensitive indicator of changes in soil organic matter in response to modifications in management practices. These results are also in line with other reports (Loss et al., 2011; Matos et al., 2011; Xavier et al., 2013).

Particulate organic carbon

The POC levels were not significantly affected by the cover crops (Table 3), even though the previous reports show that POC is a sensitive indicator of changes in the soil organic matter level due to modifications in management practices (Banger et al., 2010; Covalada et al., 2011; Rossi et al., 2012). One reason for the absence of a significant change in POC levels may be the short time span of the experiment, and it is possible that subsequent cover crop cycles would raise the POC levels. The POC levels represent organic C associated with the sand fraction (>0.053 mm), and therefore, have low colloidal protection. In addition, POC is considered a labile fraction of soil organic matter, as it has a fast cycling rate and consists mainly of partially humidified plant residues (Haynes, 2005). Particulate organic C accounts for about 18 % of the SOC, implying that only a small fraction of SOC consists of more labile organic forms. The low POC/SOC ratio (Table 3) obtained in this study suggests that regular applications of organic matter should be

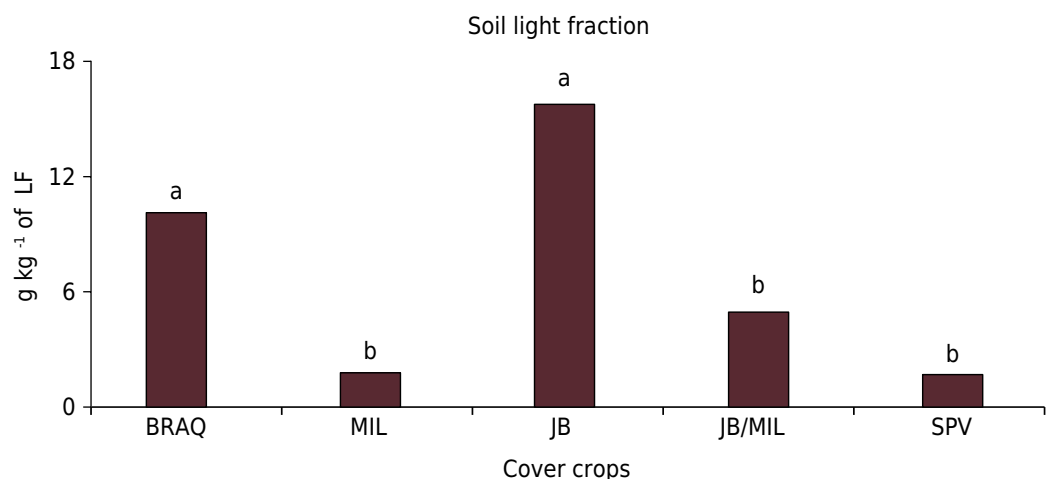


Figure 1. Soil light fraction (LF) contents extracted from the 0.00-0.10 m layer in an orange orchard with different cover crops. Means followed by the same lower-case letters do not differ significantly by the Scott-Knott test ($p < 0.05$).

considered in soil management planning in order to maintain or increase POC, for being a source of energy for soil microbiota, which play an important role in soil nutrient cycling.

There were no significant effects of cover crop cultivation on of mineral associated organic C (mAOC) levels at soil depths of 0.0-0.10 m (Table 3). In the 0.10-0.20 m layer, JB/MIL and SPV cultivation resulted in the lowest mAOC levels, whereas in the 0.20-0.40 m layer, similar to the trend observed with SOC levels, BRAQ and MIL significantly increased mAOC levels compared to the other cover crops (Table 2). Comparable results were reported by Schiavo et al. (2011), who used *Brachiaria* as cover crop and found that this species induced the greatest increase in POC in the 0.10-0.20 m layer. On average, mAOC accounted for 81 % of total SOC (Table 3), indicating that most SOC in this soil is found in recalcitrant or stabilized forms. As there was a predominant sand fraction (up to 85 %, Table 1) in the soil as determined by soil granulometry, it is possible to assume that only a small fraction of this organic C is associated with clay minerals. Thus, most of what is considered mAOC may, in fact, be related to C in the humidified fraction of soil organic matter, a behavior commonly observed in the tropical soils.

Oxidizable organic C fractions

The lability (bioavailability) of the organic C decreases from fractions *F1* to *F4* (Table 4). In general, the cover crops did not significantly affect organic C levels in the various fractions, but in the 0.00-0.10 m layer, the C levels in the *F1* fraction after MIL and JB were greater than in the other treatments. These differences were not observed for the total SOC levels (Table 2). As the *F1* fraction comprises the C forms that require the lowest degree of oxidation for extraction (Chan et al., 2001; Souza et al., 2014), our results suggest that the cultivation of these two species may enhance the concentrations of the more labile organic C in the soil. This significant difference in organic C also indicates that the *F1* fraction can be used as a sensitive indicator of the changes in soil organic matter due to alterations

Table 3. Particulate organic carbon (POC) and mineral associated organic carbon (mAOC) contents and their proportion in relation to total soil organic carbon (SOC) in the 0.00-0.10, 0.10-0.20 and 0.20-0.40 m layers in an orange orchard with different cover crops

Cover crop ⁽¹⁾	POC	mAOC ⁽²⁾	POC/SOC	mAOC/SOC	POC/mAOC
g kg ⁻¹					
0-0.10 m					
BRAQ	2.03 a	5.94 a	0.25	0.75	0.34
MIL	1.76 a	6.81 a	0.21	0.79	0.26
JB	1.75 a	5.48 a	0.24	0.76	0.32
JB/MIL	2.05 a	4.70 a	0.30	0.70	0.44
SPV	1.73 a	6.35 a	0.21	0.79	0.27
0.10-0.20 m					
BRAQ	1.74 a	9.48 a	0.16	0.84	0.18
MIL	1.63 a	10.42 a	0.14	0.86	0.16
JB	1.56 a	5.99 b	0.14	0.86	0.16
JB/MIL	1.74 a	9.48 a	0.23	0.77	0.29
SPV	1.44 a	8.04 b	0.15	0.85	0.18
0.20-0.40 m					
BRAQ	1.69 a	10.92 a	0.13	0.87	0.15
MIL	1.44 a	11.90 a	0.11	0.89	0.12
JB	1.65 a	6.90 b	0.19	0.81	0.24
JB/MIL	1.72 a	8.68 b	0.17	0.83	0.20
SPV	1.64 a	9.12 b	0.15	0.85	0.18

⁽¹⁾ BRAQ: brachiaria; MIL: pearl millet; JB: jack bean; JB/MIL: blend of JB+MIL in a proportion of 50 %; SPV: spontaneous vegetation. ⁽²⁾ mAOC was calculated as the difference between SOC and POC contents. Means followed by the same lower-case letters in a column, within each soil layer, do not differ significantly by the Scott-Knott test ($p < 0.05$).

in the management practices. These observations are in line with those of other studies (Maia et al., 2007; Loss et al., 2010).

We considered the C recovered in the *F1* fraction as labile C (C_L), while the sum of the C content recovered from the *F3* and *F4* fractions was labeled non-labile C (C_{NL}). In the surface soil layer, the C_L/C_{NL} ratio (lability index) was greater than 1.0 (Table 4), indicating that the cover crops, regardless of the species, increased the proportion of labile C in the soil. Contrarily, in the deeper soil layers, C_{NL} were proportionally higher than the C_L levels. In the mean, C_L represented 42, 35, and 38 % of the total SOC in the 0.00-0.10, 0.10-0.20 and 0.20-0.40 m layers, respectively, while C_{NL} accounted for 34, 52 and 54 % of SOC in these layers. Compared to the other cover crops, the increase in C_L/SOC and C_L/C_{NL} values was greater in the 0.00-0.10 and 0.20-0.40 m layers after JB (Table 4), suggesting that JB as cover crop favors an increase in C_L in both the surface and deeper soil layers. The increase in the C_L in surface soil can be related to the quality of JB shoot biomass, which has a rapid decomposition rate (data not shown), thus facilitating the accumulation of highly labile organic C forms. The increase in C_L in the deeper soil layers may be attributed to the release of low-molecular-weight organic compounds through the JB root system which, due to its structure, is capable of reaching the deepest soil layers (Carvalho et al., 2006). Only a few significant differences were observed in C_{NL} , implying that the more stable organic C forms were not affected by the cover crops. However, an exception was observed in the 0.10-0.20 m layer, where a significant reduction in C_{NL} was recorded after JB/MIL and SPV cultivation (Table 4). It is important to note here that this decrease coincides with a reduction in the mAOC fraction at the same soil depth (Table 3). These two C pools have organic C forms that are similar in nature, which explains the

Table 4. Soil organic carbon fractions (*F*) of varying oxidizability and their relationship with total soil organic C (SOC) contents in the 0.00-0.10, 0.10-0.20 and 0.20-0.40 m layers in an orange orchard with different cover crops

Cover crop ⁽¹⁾	Oxidizable C fraction ⁽²⁾				C_L ⁽³⁾	C_{NL} ⁽³⁾	C_L/SOC	C_{NL}/SOC	C_L/C_{NL}
	<i>F1</i>	<i>F2</i>	<i>F3</i>	<i>F4</i>					
g kg ⁻¹									
0.00-0.10 m									
BRAQ	3.03 b	2.14 a	1.58 a	1.22 a	3.03 b	2.80 a	0.38	0.35	1.08
MIL	4.14 a	1.50 a	1.52 a	1.42 a	4.14 a	2.93 a	0.48	0.34	1.41
JB	3.90 a	0.98 a	0.94 a	1.41 a	3.90 a	2.35 a	0.54	0.32	1.66
JB/MIL	2.47 b	2.33 a	0.56 a	1.39 a	2.47 b	1.95 a	0.37	0.29	1.27
SPV	2.77 b	2.22 a	0.70 a	2.40 a	2.77 b	3.10 a	0.34	0.38	0.89
0.10-0.20 m									
BRAQ	3.90 a	1.18 a	1.14 a	5.01 a	3.90 a	6.15 a	0.35	0.55	0.63
MIL	4.51 a	1.38 a	0.79 a	5.38 a	4.51 a	6.16 a	0.37	0.51	0.73
JB	3.67 a	0.54 a	2.45 a	4.38 a	3.67 a	6.83 a	0.33	0.62	0.54
JB/MIL	2.63 a	1.72 a	1.38 a	1.99 b	2.63 a	3.37 b	0.34	0.44	0.78
SPV	3.40 a	1.39 a	1.75 a	2.94 b	3.40 a	4.68 b	0.36	0.49	0.73
0.20-0.40 m									
BRAQ	3.87 a	1.40 a	0.73 a	6.60 a	3.87 a	7.34 a	0.31	0.58	0.53
MIL	4.60 a	1.20 a	0.40 a	7.14 a	4.60 a	7.54 a	0.34	0.57	0.61
JB	4.40 a	1.27 a	0.25 a	2.92 b	4.40 a	3.17 b	0.51	0.37	1.39
JB/MIL	4.03 a	0.50 a	0.86 a	5.33 a	4.03 a	6.19 a	0.39	0.60	0.65
SPV	3.53 a	1.13 a	0.53 a	5.56 a	3.53 a	6.10 a	0.33	0.57	0.58

⁽¹⁾ BRAQ: brachiaria; MIL: pearl millet; JB: jack bean; JB/MIL: blend of JB+MIL in a proportion of 50 %; SPV: spontaneous vegetation. ⁽²⁾ *F1* (very labile): oxidizable organic C under 3 mol L⁻¹ H₂SO₄; *F2* (labile): difference in oxidizable organic C extracted between 6 and 3 mol L⁻¹ H₂SO₄; *F3* (less labile): difference in oxidizable organic C extracted between 9 and 6 mol L⁻¹ H₂SO₄; *F4* (recalcitrant): difference between total SOC and oxidizable organic C under 9 mol L⁻¹ H₂SO₄. ⁽³⁾ C_L (Labile C): comprise C contents extracted in *F1*; C_{NL} (non-labile C): comprise C contents extracted in *F3*+*F4*. Means followed by the same lower-case letters in a column, within each soil layer, do not differ significantly by the Scott-Knott test ($p < 0.05$).

concomitant reduction in SOC levels under these treatments (Table 2), which are both associated with a reduction in the more stable organic C forms.

Determining various fractions of soil organic C at different oxidation degrees proved useful in evaluating the degree of SOC lability. It also permits the separation of the labile from the non-labile organic C forms, providing valuable information on the soil organic matter quality (Chan et al., 2001; Loss et al., 2013).

CONCLUSIONS

The cultivation of Brachiaria and pearl millet as cover crops increased SOC stocks in the 0.00-0.40 m depth profile and represented the most adequate management option of soil cover for soil C sequestration under the given soil and climatic conditions.

Leguminous jack bean as a cover crop promotes greater accumulation of the light fraction in the soil and increases the proportion of the most labile C fractions, both in the surface and the deeper soil layers.

Soil cover by spontaneous vegetation were similar to those observed with the other cover crops, and removal of this vegetation through intensive soil tilling will not only leave the soil bare but will also result in a substantial loss of the soil labile organic C with negative consequences for the nutrient cycling.

Both the soil light fraction and the most oxidizable organic C fraction (*F1*) were sensitive indicators of changes in SOC levels caused by modifications in the management of soil cover and can be useful tools for monitoring soil organic matter levels in the short term.

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REFERENCES

- Balota EL, Auler PAM. Soil microbial biomass under different management and tillage systems of permanent intercropped cover species in an orange orchard. *Rev Bras Cienc Solo*. 2011;35:1873-83. doi:10.1590/S0100-06832011000600004
- Banger K, Toor GS, Biswas A, Sidhu SS, Sudhir K. Soil organic carbon fractions after 16-years of applications of fertilizers and organic manure in a Typic Rhodalfs in semi-arid tropics. *Nutr Cycl Agroecosyst*. 2010;86:391-9. doi:10.1007/s10705-009-9301-8
- Blake GR, Hartge KH. Bulk density. In: Klute A, editor. *Methods of soil analysis: physical and mineralogical methods*. Madison: America Society of America; 1986. p.363-75.
- Blanco-Moure N, Gracia R, Bielsa C, López MV. Long-term no-tillage effects on particulate and mineral-associated soil organic matter under rainfed Mediterranean conditions. *Soil Use Manage*. 2013;29:250-9. doi:10.1111/sum.12039
- Bressan SB, Nóbrega JCA, Nóbrega RSA, Barbosa RS, Sousa LB. Plantas de cobertura e qualidade química de Latossolo Amarelo sob plantio direto no cerrado maranhense. *Rev Bras Eng Agríc Amb*. 2013;17:371-8. doi:10.1590/S1415-43662013000400003

- Bu X, Ruan H, Wang L, Ma W, Ding J, Yu X. Soil organic matter in density fractions as related to vegetation changes along an altitude gradient in the Wuyi Mountains, southeastern China. *Appl Soil Ecol.* 2012;52:42-7. doi:10.1016/j.apsoil.2011.10.005
- Caetano JO, Benites VM, Silva GP, Silva IR, Assis RL, Cargnelutti Filho A. Dinâmica da matéria orgânica de um Neossolo Quartzarênico de cerrado convertido para cultivo em sucessão de soja e milho. *Rev Bras Cienc Solo.* 2013;37:1245-55. doi:10.1590/S0100-06832013000500014
- Cambardella CA, Elliott ET. Particulate soil organic matter changes across a grassland cultivation sequence. *Soil Sci Soc Am J.* 1992;56:777-83. doi:10.2136/sssaj1992.03615995005600030017x
- Carter MR. Organic matter and sustainability. In: Rees RM, Ball BC, Campbell CD, Watson CA, editors. *Sustainable management of soil organic matter.* New York: CAB International; 2001. p.9-22.
- Carvalho JEB, Dias RCS, Melo Filho, JF. Produção integrada de citros x convencional - impacto sobre a qualidade do solo. Cruz das Almas: Embrapa Mandioca e Fruticultura; 2006. (Comunicado técnico, 118).
- Carvalho JEB, Santos RC, Araújo AMA. Produção sustentável de citros. Cruz das Almas: Embrapa Mandioca e Fruticultura; 2003a. (Comunicado técnico, 84).
- Carvalho JEB, Santos RC, Souza ALV. Novo preparo e manejo do solo no controle do mato - contribuição ao desenvolvimento do sistema radicular dos citros. Cruz das Almas: Embrapa Mandioca e Fruticultura; 2003b. (Comunicado técnico, 85).
- Carvalho JEB, Souza LS, Caldas RC, Antas PEUT, Araújo LC, Lopes AMA, Santos RC, Lopes NCM, Souza ALV. Leguminosa no controle integrado de plantas daninhas para aumentar a produtividade da Laranja-‘Pêra’. *Rev Bras Frutic.* 2002;24:82-5. doi:10.1590/S0100-29452002000100018
- Chan KY, Bowman A, Oates A. Oxidizable organic carbon fractions and soil quality changes in an oxic Paleustalf under different pasture ley. *Soil Sci.* 2001;166:61-7. doi:10.1097/00010694-200101000-00009
- Covalada S, Gallardo Lancho JF, García-Oliva F, Kirchmann H, Prat C, Bravo M, Etchevers JD. Land-use effects on the distribution of soil organic carbon within particle-size fractions of volcanic soils in the Transmexican Volcanic Belt (Mexico). *Soil Use Manage.* 2011;27:186-94. doi:10.1111/j.1475-2743.2011.00341.x
- Gonçalves CN, Ceretta CA. Plantas de cobertura de solo antecedendo o milho e seu efeito sobre o carbono orgânico do solo, sob plantio direto. *Rev Bras Cienc Solo.* 1999;23:307-13. doi: 10.1590/S0100-06831999000200015
- Haynes RJ. Labile organic matter fractions as central components of the quality of agricultural soils: an overview. *Adv Agron.* 2005;85:221-68. doi:10.1016/S0065-2113(04)85005-3
- Hernani LC, Kurihara CH, Silva WM. Sistemas de manejo de solo e perdas de nutrientes e matéria orgânica por erosão. *Rev Bras Cienc Solo.* 1999;23:145-54. doi:10.1590/S0100-06831999000100018
- Janzen HH, Campbell CA, Brandt SA, Lafond GP, Townley-Smith L. Light fraction organic matter in soils from long-term crop rotations. *Soil Sci Soc Am J.* 1992;56:1799-806. doi:10.2136/sssaj1992.03615995005600060025x
- Johnston AE, Poulton PR, Coleman K. Soil organic matter: its importance in sustainable agriculture and carbon dioxide fluxes. *Adv Agron.* 2009;101:1-57. doi:10.1016/S0065-2113(08)00801-8
- Kirschbaum MUF. The temperature dependence of organic-matter decomposition-still a topic of debate. *Soil Biol Biochem.* 2006;38:2510-8. doi:10.1016/j.soilbio.2006.01.030
- Loss A, Coutinho FS, Pereira MG, Silva RAC, Torres JLR, Ravelli Neto A. Fertilidade e carbono total e oxidável de Latossolo de cerrado sob pastagem irrigada e de sequeiro. *Cienc Rural.* 2013;43:426-32. doi:10.1590/S0103-84782013000300008
- Loss A, Moraes AGL, Pereira MG, Silva EMR, Anjos LHC. Carbono, matéria orgânica leve e frações oxidáveis do carbono orgânico sob diferentes sistemas de produção orgânica. *Comunicata Sci.* 2010;1:57-64.

- Loss A, Pereira MG, Giacomo SG, Perin A, Anjos LHC. Agregação, carbono e nitrogênio em agregados do solo sob plantio direto com integração lavoura-pecuária. *Pesq Agropec Bras*. 2011;46:1269-76. doi:10.1590/S0100-204X2011001000022
- Maia SMF, Xavier FAS, Oliveira TS, Mendonça ES, Araújo Filho JA. Organic carbon pools in a Luvisol under agroforestry and conventional farming systems in the semi-arid region of Ceará, Brazil. *Agrofor Syst*. 2007;71:127-38. doi:10.1007/s10457-007-9063-8
- Marin AMP, Menezes RSC, Silva ED, Sampaio EVSB. Efeito da *Gliricidia sepium* sobre nutrientes do solo, microclima e produtividade do milho em sistema agroflorestal no Agreste Paraibano. *Rev Bras Cienc Solo*. 2006;30:555-64. doi:10.1590/S0100-06832006000300015
- Marques JDO, Luizão FJ, Teixeira WG, Sarrazin M, Ferreira SJF, Beldini, TP, Marques EMA. Distribution of organic carbon in different soil fractions in ecosystems of central Amazonia. *Rev Bras Cienc Solo*. 2015;39:232-42. doi:10.1590/01000683rbcs20150142
- Matos ES, Freese D, Mendonça ES, Slazak A, Hütthl RF. Carbon, nitrogen and organic C fractions in topsoil affected by conversion from silvopastoral to different land use systems. *Agrofor Syst*. 2011;81:203-11. doi:10.1007/s10457-010-9314-y
- Murage EW, Voroney P, Beyaert RP. Turnover of carbon in the free light fraction with and without charcoal as determined using the ^{13}C natural abundance method. *Geoderma*. 2007;138:133-43. doi:10.1016/j.geoderma.2006.11.002
- Poeplau C, Don A. Carbon sequestration in agricultural soils via cultivation of cover crops - A meta-analysis. *Agric Ecosyst Environ*. 2015;200:33-41. doi:10.1016/j.agee.2014.10.024
- Rossi CQ, Pereira MG, Giacomo SG, Betta M, Polidoro JC. Frações lábeis da matéria orgânica em sistema de cultivo com palha de braquiária e sorgo. *Rev Cienc Agron*. 2012;43:38-46. doi:10.1590/S1806-66902012000100005
- Santana MB, Souza LS, Souza LD, Fontes LEF. Atributos físicos do solo e distribuição do sistema radicular de citros como indicadores de horizontes coesos em dois solos de Tabuleiros Costeiros do estado da Bahia. *Rev Bras Cienc Solo*. 2006;31:1-12. doi:10.1590/S0100-06832006000100001
- Schiavo JA, Rosset JS, Pereira MG, Salton JC. Índice de manejo de carbono e atributos químicos de Latossolo Vermelho sob diferentes sistemas de manejo. *Pesq Agropec Bras*. 2011;46:1332-8. doi:10.1590/S0100-204X2011001000029
- Sequeira CH, Alley MM, Jones BP. Evaluation of potentially labile soil organic carbon and nitrogen fractionation procedures. *Soil Biol Biochem*. 2011;43:438-44. doi:10.1016/j.soilbio.2010.11.014
- Silva FAS, Azevedo CAVA. New version of the Assistat - statistical assistance software. In: *Proceedings World Congress on Computers in Agriculture*; 2006; Orlando. Orlando: American Society of Agricultural and Biological Engineers; 2006. p.393-6.
- Sohi SP, Mahieu N, Arah JRM, Powlson DS, Madari BE, Gaunt J. A procedure for isolating soil organic matter fractions suitable for modeling. *Soil Sci Soc Am J*. 2001;65:1121-8. doi:10.2136/sssaj2001.6541121x
- Souza LD, Santos CV, Souza LS, Pereira BLS. Resistência à penetração em Latossolo Amarelo dos Tabuleiros Costeiros, sob cobertura vegetal com leguminosas. *Cruz das Almas: Embrapa Mandioca e Fruticultura*; 2013. (Boletim de pesquisa, 58).
- Souza RF, Figueiredo CC, Madeira NR, Alcântara FP. Effect of management systems and cover crops on organic matter dynamics of soil under vegetables. *Rev Bras Cienc Solo*. 2014;38:923-33. doi:10.1590/S0100-06832014000300024
- Tobiašová E. The effect of organic matter on the structure of soils of different land uses. *Soil Till Res*. 2011;114:183-92. doi:10.1515/agri-2015-0010
- Walkley A, Black IA. An examination of the Degtjareff method for determining soil organic matter and a proposed modification of the chromic acid titration method. *Soil Sci*. 1934;37:29-38.
- Xavier FAS, Maia SMF, Oliveira TS, Mendonça ES. Soil organic carbon and nitrogen stocks under tropical organic and conventional cropping systems in Northeastern Brazil. *Commun Soil Sci Plant Anal*. 2009;40:2975-94. doi:10.1080/00103620903261304

Xavier FAS, Maia SMF, Ribeiro KA, Mendonça ES, Oliveira TS. Effect of cover plants on soil C and N dynamics in different soil management systems in dwarf cashew culture. *Agric Ecosyst Environ.* 2013;165:173-83. doi:10.1016/j.agee.2012.12.003

Yeomans JC, Bremner JM. A rapid and precise method for routine determination of organic carbon in soil. *Commun Soil Sci Plant Anal.* 1988;19:1467-76. doi:10.1080/00103628809368027