



# Changing the land use from degraded pasture into integrated farming systems enhance soil carbon stocks in the Cerrado biome

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**ABSTRACT.** Integrated agricultural production systems can increase soil organic carbon stocks over time and contribute to the mitigation of climate change. The present study aimed to evaluate soil carbon stocks, accumulation rates (TOC), total nitrogen (TN), and the quality of soil organic matter (SOM) after the transition of a low-productivity pasture into agrosilvopastoral systems in the Cerrado biome. We evaluated an 11-year-old experiment, and the treatments studied were: Eucalyptus + buffel grass; Eucalyptus + cowpea; Eucalyptus + pigeon pea; eucalyptus + buffel grass + cowpea; Eucalyptus + buffel grass + pigeon pea; Eucalyptus in monoculture (with a 20 × 3 m tree arrangement and no cultivation between rows but with a history of forage and grain crop production); and a low-productivity pasture as additional treatment and reference to the soil condition previously the land-use change. In this study, TOC and TN stocks and accumulation, labile (LC) and non-labile carbon (NLC), and SOM humic fractions were evaluated at 0-10, 10-20, 20-40, and 0-40 cm depth layers. Integrated farming systems have increased TOC and TN, NLC, carbon contents and stocks in SOM chemical fractions in all depths and TOC and TN accumulation of 5.22 Mg ha<sup>-1</sup> year<sup>-1</sup> and 0.23 Mg ha<sup>-1</sup> year<sup>-1</sup>, respectively, at the 0-40 cm depth layer. The integration of Eucalyptus with legumes or buffel grass increased the LC content in the surface layer of the soil. The transition from low-productivity pasture into integrated farming systems can promote the recovery of SOM and soil quality. Hence, our results suggest that agrosilvopastoral systems can be used as sustainable farming systems in the Cerrado biome.

**Keywords:** agrosilvopastoral system; labile carbon; humic substances; soil organic matter; legumes; buffel grass.

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## Introduction

The lack of adequate management practices in pastures contributes to the loss of soil organic matter (SOM), which increases atmospheric greenhouse gas (GHG) emissions (Tadini et al., 2021b). Land-use types that increase the organic carbon content and reduce the GHG emissions must be adopted to control the negative impacts of climate change (Toru & Kibret, 2019). The conversion of low-productivity pastures into integrated farming systems can facilitate the soil carbon accumulation (Frazão et al., 2021).

Integrated farming systems have characteristics similar to others conservationist systems and contribute to CO<sub>2</sub> sequestration (Yadav et al., 2021). They can increase biodiversity (Freitas et al., 2020), biomass supply, and root volume (Cunha et al., 2020), thereby improving soil fertility, SOM storage, soil biological activity (Damian et al., 2021), and soil physical quality (Muchane et al., 2020).

SOM and its fractions (labile carbon and humic substances) have been studied in different land-use and management systems. Organic soil carbon (TOC) is an important indicator of ecosystem recovery (Gomes et al., 2021) and is positively correlated with soil fertility (Tadini et al., 2021b). Labile Carbon (LC) is a sensitive and representative indicator of SOM dynamics and quality. It comprises labile compounds that are used by microorganisms as an energy source (Ndzelu, Dou, & Zhang, 2020), which suggests that these compounds can be easily decomposed and lost (Santos et al., 2019). Gomes et al. (2021) reported an increased LC content in agroforestry systems due to greater species diversity and frequent deposition of plant residues.

Carbon is sequestered by humification into molecules and/or recalcitrant complexes that reduce SOM decomposition and CO<sub>2</sub> emissions, and hence, it is important to study these humic substances (Tadini et al.,

2021a). Chemical fractionation of humic substances helps infer the degree of SOM humification and stability (Ramos et al., 2020). The increase in more recalcitrant fractions indicates higher stabilization of carbon in the soil, which meets the requirement of low-carbon agriculture systems, as verified in integrated farming systems evaluated by Coser et al. (2018).

However, it is essential to highlight that integrated systems may have varied SOM content owing to the differences in properties such as precipitation, litter supply, root biomass, decomposition rate, plant species, edaphoclimatic characteristics, and litter chemical composition (Yadav et al., 2021). Although integrated production systems can increase soil TOC stocks in short term, Almeida et al. (2021) reported that long-term monitoring must be assessed. Ma, Chen, Bork, Carlyle, and Chang (2020a) stated that the implementation time of sustainable production systems is an important factor in the recovery and accumulation of carbon in the soil.

Experiments that incorporate different models of integrated farming systems should be performed to validate SOM accumulation and improvements in soil quality. The present study aimed to evaluate soil carbon and nitrogen stocks, accumulation rates, and SOM quality after the transition of low-productivity pastures into agrosilvopastoral systems in an eleven-year-old field experiment in the Cerrado biome.

## Material and methods

### Location and characterization of the study area

Integrated production systems were adopted in 2009 at the Hamilton de Abreu Navarro Experimental Farm (16°40'03" S, 43°50'41" W, 598 m altitude) located at the Institute of Agrarian Sciences of the Federal University of Minas Gerais, municipality of Montes Claros, Minas Gerais State, Brazil. The study area is located in the Cerrado biome (*stricto sensu*) and features a slightly undulated relief. The climate in the region is tropical savanna (Aw, according to the Köppen classification), with rainy summers and dry winters. The average annual rainfall during the study period was 923 mm, with an average temperature of 24.9°C (Instituto Nacional de Meteorologia, 2022). The soil was classified as Acrisol, and its chemical and physical characteristics are listed in Table 1.

**Table 1.** Soil Chemical and physical characterization<sup>1</sup> in the experimental area at 0-20 cm depth.

Soil attribute	Experimental area	
	AGS	Test
pH in water	6.50	5.00
P Mehlich (mg dm <sup>-3</sup> )	17.52	7.70
Remaining P (mg L <sup>-1</sup> )	39	39
K (mg dm <sup>-3</sup> )	217	232
Ca (cmol <sub>c</sub> dm <sup>-3</sup> )	7.60	8.30
Mg (cmol <sub>c</sub> dm <sup>-3</sup> )	3.28	3.32
Al (cmol <sub>c</sub> dm <sup>-3</sup> )	0.00	0.00
H+Al (cmol <sub>c</sub> dm <sup>-3</sup> )	2.08	2.29
Sum of Bases (cmol <sub>c</sub> dm <sup>-3</sup> )	11.44	12.21
Aluminum Saturation (%)	0.00	0.00
Potential Cation Exchange Capacity (cmol <sub>c</sub> dm <sup>-3</sup> )	13.52	14.50
Base Saturation (%)	84.61	84.21
Organic Carbon (g kg <sup>-1</sup> )	20.37	5.67
Sand (g kg <sup>-1</sup> )	380	340
Silt (g kg <sup>-1</sup> )	320	260
Clay (g kg <sup>-1</sup> )	300	400
Bulk density (g cm <sup>-3</sup> )	1.41	1.78

<sup>1</sup>Sampled in August 2019. AGS: Agrosilvopastoral system; Test: Low-productivity pasture used as a reference area before AGS introduction.

### History of the study area

The AGS system was introduced in 2009 on a low-productivity pasture, which is identified as Test in the present study, in an area of 6,900 m<sup>2</sup>. The low-productivity pasture (which had no defined management, presence of weeds and exposed soil, and a mixture of grass species) lies within an area that previously witnessed cultivation of irrigated vegetables and corn, but the Cerrado vegetation in this region was removed for cultivation in 1971. The tree component of the agrosilvopastoral system was a hybrid of *Eucalyptus urograndis* (*E. grandis* × *E. urophylla*). Over the years, tree thinning and within-row planting was performed to reduce shading between rows, resulting in a tree arrangement of 20 × 3 m (166 trees ha<sup>-1</sup>). After the plantation

of trees, forage species and grain crops were combined in a consortium and/or succession. The management practices and cultures included in this integrated farming system have been described in detail by Rodrigues, Santos, Sampaio, and Fernandes (2017) and Frazão et al. (2021).

In December 2019 (after 11 years of adopting the integrated farming system), pigeon pea, cowpea, and buffel grass were planted between eucalyptus rows. In addition, consortia of pigeon peas with buffel grass and cowpeas with buffel grass within eucalyptus plantation rows were also adopted. Glyphosate Atanor® (4.0 L ha<sup>-1</sup>) was used to desiccate the remaining species of previous crops (predominantly *Brachiaria* sp.) to implement the aforementioned integrated farming systems. The soil was plowed and then fertilizers were applied during plantation, which included 20 kg ha<sup>-1</sup> of P<sub>2</sub>O<sub>5</sub> and, 25 days after sowing (DAS), 20 kg ha<sup>-1</sup> of K<sub>2</sub>O and 20 kg ha<sup>-1</sup> of N.

Pigeon pea (*Cajanus cajan* (L.) Millsp., cv. Iapar 43/Aratã) was sown in a final stand of 10 plants m<sup>-1</sup>, with 1 m row spacing, and cowpea (*Vigna unguiculata* (L.) Walp., cv. BRS Potengi) with 8 plants m<sup>-1</sup> with spacing of 0.5 m between rows. Buffel grass (*Cenchrus ciliaris* L. cv. Aridus) was sowed using 35 kg ha<sup>-1</sup> of seeds (2 kg ha<sup>-1</sup> of viable pure seeds) in rows spaced 0.5 m apart. The sowing of the plots with a consortium between legumes and buffel grass followed the same spacing and population of plants used in the plots with no consortium.

Cowpeas were managed for grain production, whereas pigeon peas and buffel grass were used for forage production. The cowpea beans were harvested approximately 75 DAS. The pigeon pea was cut at 150 DAS, while buffel grass remained in the area after legume production.

### Experimental design and treatments

The experiment used a randomized block design (RBD) with four repetitions and seven treatments, as follows: Eucalyptus and buffel grass (E+B); eucalyptus and cowpea (E+C); eucalyptus and pigeon pea (E+P); eucalyptus, buffel grass, and cowpea (E+B+C); eucalyptus, buffel grass, and pigeon pea (E+B+P); eucalyptus (Euc), also with a 20 × 3 m arrangement, no cultivation between rows, but with a history of forage and grain crops, with weeds being periodically mowed; and low-productivity pasture (Test), the additional treatment described in the previous subsection. The eucalyptus arrangement was 20 × 3 m, and the experimental units were allocated among the rows with a size of 10 × 3 m (30 m<sup>2</sup>), allowing a distance of 1 m from the eucalyptus rows.

### Soil sampling

Soil sampling for density and SOM evaluation was carried out in May 2020, at 165 DAS for legumes and buffel grass. Soil samples were collected from soil layers at a depth of 0-10, 10-20, and 20-40 cm. Sampling was performed along trench lines randomly distributed in the Test treatment (additional treatment), whereas in the integrated farming systems, sampling was performed along the transect between the planting rows of eucalyptus at distances of 2, 4, 6, and 8 m from the trees. These different distances (simple samples) were used to obtain composite samples. Therefore, each composite sample (repetition) was formed by mixing four individual subsamples. After field sampling, the samples were air-dried, passed through sieves of 2 mm, and the remaining plant residue was removed.

### Soil analysis

The soil density was determined using the volumetric soil core method (Blake & Hartge, 1986). TOC was quantified by wet oxidation using potassium dichromate (Yeomans & Bremner, 1988) and total nitrogen (TN) using sulfuric digestion, according to the Kjeldahl method (Bremner & Mulvaney, 1982; Tedesco, Gianello, Bissani, Bohnen, & Volkweiss, 1995). Thereafter, C/N ratios were calculated. TOC and TN stocks (Mg ha<sup>-1</sup>) were obtained by multiplying the density of the soil (g cm<sup>-3</sup>) and the thickness of soil layer (cm) with TOC or TN content (%), respectively. The annual accumulation rates of TOC and TN (Mg ha<sup>-1</sup>) were obtained by subtracting the means of TOC and TN stocks of all integrated systems studied by the stocks of the Test system (additional treatment) divided by 11 years, which was the implantation time.

Labile carbon (LC) was determined by oxidation with potassium permanganate (KMnO<sub>4</sub> 0.033 mol L<sup>-1</sup>) and quantification was performed by colorimetry using a spectrophotometer at a wavelength of 565 nm (Blair, Lefroy, & Lisle, 1995; Shang & Tiessen, 1997). The non-labile carbon (NLC) corresponded to the non-oxidized carbon by KMnO<sub>4</sub> and was therefore obtained from the difference between TOC and LC.

Chemical fractionation of SOM was performed to separate the fractions of fulvic acid (C-FA), humic acid (C-HA), and humin (C-HUM), based on differences in solubility in acid and alkaline media (Mendonça & Matos, 2005). The carbon content of each fraction was quantified by wet oxidation using potassium

dichromate (Yeomans & Bremner, 1988), and the C-HA/C-FA and EA/HUM ratios were calculated. The alkaline extract (AE) matched the sum of the C-FA and C-HA fractions.

### Statistical analysis

The Shapiro-Wilk test was applied to verify the occurrence of normal distributions, and the Bartlett test was used to determine the homogeneity of variances. After validating these assumptions, the analysis of variances was performed and the Scott–Knott test was applied at a 5% significance level for comparisons between the integrated systems. Additionally, each integrated system was individually compared with the additional treatment (Test) using Dunnett's test ( $p \leq 0.05$ ). Statistical analysis was performed using R software version 3.6.2.

## Results

### Total organic carbon (TOC) and total nitrogen (TN) accumulations, and soil C/N ratio

Integrated production systems increased TOC levels at all soil depths ( $p < 0.05$ ). An increase of up to 212% in the 0–40 cm depth was observed between the Test and E+B systems (Figure 1A). There was no variation in the TOC content between the integrated systems for the evaluated soil layers.

TN content was also affected by soil use ( $p < 0.05$ ). Integrated systems contributed to the increase in TN content at all depths, and lower values were obtained in the Test (Figure 1B). The soil profile (0–40 cm) of the E+B and Test systems had TN content of 1.67 and 0.87 g kg<sup>-1</sup>, respectively; this represents an increase of 92%. No differences in the TN content were observed among the integrated systems.

The C/N ratio was lower in the Test system at all depths ( $p < 0.05$ ). The integrated systems had similar C:N ratios (Figure 1C). The C/N ratio in the 0–40 cm soil profile ranged from 6.89 (Test) to 11.90 (E+B+P).

The TOC and TN stocks differed based on the land use type ( $p < 0.05$ ). The integrated systems presented higher stocks than the Test at all depths. Total TOC stocks (0–40 cm) ranged from 44 to 108 Mg ha<sup>-1</sup> in the Test and Euc systems, respectively, indicating an increase in TOC stocks by 146% after 11 years conversion of land use (Figure 1D). No differences were found between TOC stocks among the integrated systems, with values between 98 and 108 Mg ha<sup>-1</sup>.

The Test system presented lower TN stocks than the integrated systems at the 0–10, 10–20, and 0–40 cm depth layers (Figure 1E). Total TN stocks (0–40 cm) of 6.28 Mg ha<sup>-1</sup> under Test up to 9.35 Mg ha<sup>-1</sup> under Euc were observed. There were no variations in TN stocks among the integrated farming systems, with values from 8.38 to 9.35 Mg ha<sup>-1</sup>.

High TOC and TN accumulation potentials were obtained by introducing integrated farming systems in low-productivity pasture areas. The accumulation rates of TOC were 1.97, 1.37, and 1.87 Mg ha<sup>-1</sup> year<sup>-1</sup> at the 0–10, 10–20, and 20–40 cm depths, respectively, adding 5.22 Mg ha<sup>-1</sup> year<sup>-1</sup> in the 0–40 cm layer. TN accumulation rates were 0.10, 0.07, and 0.06 Mg ha<sup>-1</sup> year<sup>-1</sup> at 0–10, 10–20, and 20–40 cm depths, respectively, adding 0.23 Mg ha<sup>-1</sup> year<sup>-1</sup> in the 0–40 cm layer.

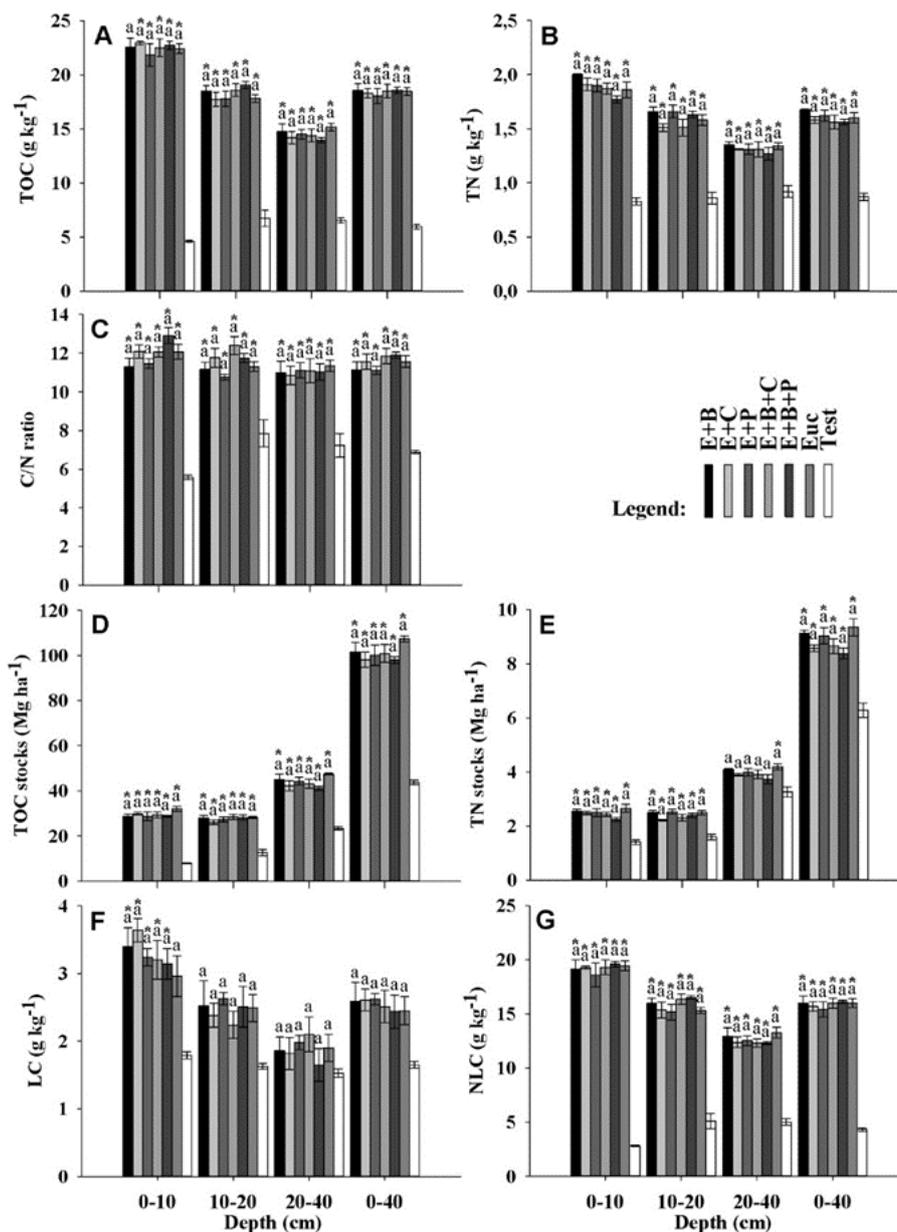
### Labile Carbon (LC) and Non-labile Carbon (NLC)

Differences in LC and NLC contents were observed between land-use systems ( $p < 0.05$ ). The LC value of the test system in the surface layer (0–10 cm) similar to Euc, which was lower than those of the other systems (Figure 1F). The LC content was similar between the Test and integrated systems at other depths (10–20, 20–40, and 0–40 cm), ranging from 1.65 (Test) to 2.62 g kg<sup>-1</sup> (E+P) in the profile (0–40 cm). Similar LC content was obtained for the integrated systems at all depths.

The NLC content in the Test system was lower than the integrated farming systems at all depths (Figure 1G). The NLC values ranged from 4.32 (Test) to 16.14 g kg<sup>-1</sup> (E+B+P) in the 0–40 cm layer, a variation that represented a 273.61% increase by implementing integrated production systems. A similar NLC content was observed in the integrated farming systems.

### Carbon in the fulvic acid (C-FA), humic acid (C-HA), and humin (C-HUM) fractions

The organic carbon content of the soil in the C-FA, C-HA, and C-HUM fractions was higher in the integrated systems when compared to the Test system ( $p < 0.05$ ). Values from 0.23 to 1.52 g kg<sup>-1</sup> of C-FA were obtained, respectively, in the Test and E+P systems (0–40 cm) (Table 2). For the same layer, the C-HA content in the Test and E+B+C systems was between 1.16 and 4.38 g kg<sup>-1</sup>. The C-HUM content was 5.38 and 13.49 g kg<sup>-1</sup> in the Test and E+B+P systems, respectively.



**Figure 1.** Soil total organic carbon (TOC) (A), total nitrogen (TN) (B), C/N ratio (C), TOC stocks (D), TN stocks (E), labile carbon (LC) (F) and non-labile carbon (NLC) (G) under different land use systems in the Cerrado biome. E+B: Eucalyptus and buffel grass; E+C: eucalyptus and cowpea; E+P: eucalyptus and pigeon pea; E+B+C: eucalyptus, buffel grass, and cowpea; E+B+P: eucalyptus, buffel grass, and pigeon pea; Euc: eucalyptus with no cultivation between rows, but with a history of forage and grain crop production; Test: low-productivity pasture. Means followed by the same letter in the row between the integrated systems and at the same soil depth did not differ by the Scott-Knott test ( $p \leq 0.05$ ). Bars with \*, at the same soil depth, differed in relation to additional treatment (Test) by Dunnett’s test ( $p \leq 0.05$ ).

In general, the soil organic carbon content in the C-FA, C-HA, and C-HUM fractions was similar among the integrated farming systems. Differences in C-FA content were observed at 0-40 cm depth, and the E+B and E+P systems had higher contents. Regarding the C-HUM content, the E+B and E+B+P systems showed higher values in the layer of 10-20 cm.

The C-HA/C-FA and EA/C-HUM ratios were also affected by the soil use systems ( $p < 0.05$ ). The E+B and E+P systems presented a lower C-HA/C-FA ratio than the Test at the 10-20 and 0-40 cm depth layers (Table 2). Among the integrated systems, the C-HA/C-FA ratio in the surface layer (0-10 cm) was higher in the treatments E+C, E+B+C, and Euc and lower in E+B, E+P, E, and E+B+P.

The EA/C-HUM ratio was lower at 0-10 cm depth in the Test. In the 10-20 cm layer, the Test presented a lower ratio for the E+C, E+B+C, and Euc systems (Table 2). The EA/C-HUM ratios were similar among the integrated systems, with values ranging from 0.40 to 0.48 (0-40 cm).

**Table 2.** Carbon in fulvic acid (C-AF), humic acid (C-HA) and humin (C-HUM) fractions and C-HA/C-FA e <sup>1</sup>AE/C-HUM fractions under the different land use systems in the Cerrado biome.

Depth (cm)	System						Test	CV %
	E+B	E+C	E+P	E+B+C	E+B+P	Euc		
	C-FA (g kg <sup>-1</sup> )							
0-10	1.98a*	1.72a*	1.97a*	1.65a*	1.83a*	1.55a*	0.28	14.70
10-20	1.30a*	1.25a*	1.48a*	1.23a*	1.17a*	1.13a*	0.20	16.18
20-40	1.07a*	0.95a*	1.12a*	0.92a*	0.75a*	0.92a*	0.23	19.34
0-40	1.45a*	1.31b*	1.52a*	1.27b*	1.25b*	1.20b*	0.23	12.24
	C-HA (g kg <sup>-1</sup> )							
0-10	3.78a*	4.10a*	3.82a*	4.35a*	4.03a*	4.16a*	0.75	16.06
10-20	3.97a*	4.66a*	4.11a*	4.61a*	4.09a*	4.53a*	1.15	19.48
20-40	3.84a*	3.86a*	4.18a*	4.18a*	3.73a*	4.19a*	1.58	20.07
0-40	3.87a*	4.21a*	4.03a*	4.38a*	3.95a*	4.29a*	1.16	16.00
	C-HUM (g kg <sup>-1</sup> )							
0-10	17.12a*	15.48a*	15.71a*	15.36a*	17.20a*	16.11a*	5.74	10.02
10-20	13.14a*	12.22b*	11.76b*	11.81b*	13.77a*	11.13b*	5.79	10.34
20-40	9.24a*	8.74a*	8.99a*	9.07a*	9.50a*	9.05a*	4.62	14.77
0-40	13.16a*	12.15a*	12.16a*	12.08a*	13.49a*	12.10a*	5.38	8.62
	C-HA/C-FA							
0-10	1.94b	2.45a	1.95b	2.67a	2.22b	2.71a	2.79	17.85
10-20	3.03a*	3.78a	2.77a*	3.87a	3.68a	4.16a	5.75	20.19
20-40	3.67a	4.14a	3.93a	4.56a	5.12a	4.72a	7.08	25.51
0-40	2.88a*	3.46a	2.88a*	3.70a	3.67a	3.86a	5.21	16.16
	AE/C-HUM							
0-10	0.34a*	0.38a*	0.37a*	0.39a*	0.34a*	0.36a*	0.19	14.91
10-20	0.40a	0.49a*	0.47a	0.50a*	0.39a	0.51a*	0.25	19.45
20-40	0.55a	0.57a	0.60a	0.57a	0.48a	0.57a	0.41	25.39
0-40	0.43a	0.48a	0.48a	0.49a	0.40a	0.48a	0.28	17.01

E+B: Eucalyptus and buffel grass; E+C: eucalyptus and cowpea; E+P: eucalyptus and pigeon pea; E+B+C: eucalyptus, buffel grass, and cowpea; E+B+P: eucalyptus, buffel grass, and pigeon pea; Euc: eucalyptus no cultivation between rows, but with a history of forage and grain crops production; Test: low-productivity pasture. <sup>1</sup>Alkaline extract (AE) = C-FA + C-HA. Means followed by the same letter in the row, between the integrated systems, did not differ by the Scott-Knott test ( $p < 0.05$ ). Means followed by \* in the row differ in relation to additional treatment (Test) by the Dunnet test ( $p \leq 0.05$ ).

## Discussion

The results of the present study confirmed our hypothesis that the transition of low-productivity pastures into integrated agricultural production systems can significantly increase the SOM content. The contents, stocks, and accumulation rates of TOC and TN, LC, NLC, and TOC contents of the C-FA, C-HA, and C-HUM fractions were generally high in the integrated farming systems.

### Organic carbon accumulation (TOC), total nitrogen (TN), and soil C/N ratio

The changes in land use when integrated farming systems were adopted likely enabled substantial increases in soil TOC and TN content (Figure 1A, B, D, and E), indicating that the agrosilvopastoral systems can contribute to climate change mitigation (Muchane et al., 2020) and soil quality improvement. Pastures that are not properly managed can decrease soil TOC stocks (Tadini et al., 2021b). However, implementing integrated production systems can facilitate the increase in TOC and TN contents in soil (Frazão et al., 2021; Yadav et al., 2021). According to Ma et al. (2020), agroforestry is an effective strategy to increase TOC storage in tropical regions.

Araujo et al. (2020) verified an increase in the SOM content in the AGS system in the first year of the Cerrado biome. Corroborating the results presented by Araujo et al. (2020), Freitas et al. (2020) reported the increase in TOC and TN stocks after three years of handling adopting agrosilvipastoral systems in the same experimental area. Similarly, increase in soil TOC and TN stocks were observed in degraded pastures of Brazilian Cerrado after four years of silvopastoral system implementation and eight years of agrosilvicultural systems implementation by Almeida et al. (2021) and Frazão et al. (2021), respectively. The present study has also shown that agrosilvopastoral systems have a high recovery potential for soil TOC and TN stocks, indicating the sustainability of production and improvement of soil quality.

Siqueira, Chiba, Moreira, and Abdo (2019) reported an accumulation of 2 Mg ha<sup>-1</sup> year<sup>-1</sup> TOC in the 0-20 cm layer of after five years of adopting an agroforestry system in degraded Argisol, which was attributed to the plant biomass input to the soil surface. Coser et al. (2018) observed an accumulation of 6.14 Mg ha<sup>-1</sup> year<sup>-1</sup> TOC at a depth of 0-40 cm after 28 months of conversion of low-productivity pasture into an agrosilvicultural system in the

Cerrado biome. In the present study, an accumulation of  $5.22 \text{ mg ha}^{-1} \text{ year}^{-1}$  was observed after 11 years of conversion from a degraded pasture into integrated production systems in the 0-40 cm layer.

In integrated farming systems, practices such as no-tillage, annual forage crop cultivation and proper grass and tree management in the first years of implementation facilitate the increase in soil TOC and TN contents (Almeida et al., 2021). The authors attributed this increase to low soil mobilization in sorghum crops in the early years of the integrated production system, along with presence of appropriate soil cover, high root renewal rate of the grass used, and generation of litter input by eucalyptus (which was further intensified by pruning). Similar procedures were also performed in the present study.

Eucalyptus can contribute to the increase in soil TOC content, and higher TOC intensity is observed close to the tree rows due to higher deposition of residues on the soil and below the surface, such as leaf and root extracts, leaves, flowers, seeds, stems, and roots (Abreu et al., 2020). In addition, eucalyptus produces residue continuously during its cycle, which further contributes to an increase in TOC (Cunha et al., 2020). Toru and Kibret (2019) recommended the cultivation of woody perennial plants in conservation systems with the aim of increasing carbon sequestration. Hence, the production of eucalyptus litter over the years contributed to the recovery of TOC and TN content lost in the Test system used in this study.

Associating more species in AGS systems and the presence of grass pastures, which develop a dense root system and cover a large volume of soil, improves SOM levels (Araujo et al., 2020). The high TOC contents obtained in integrated systems can be the result of high inputs of organic eucalyptus and grass residue (Cunha et al., 2020). The authors reported that, in addition to its density, the grass root system consists mainly of fine roots that present fast decomposition, increasing the organic carbon content of the soil. Thus, the grass cultivation history (in all treatments) and buffel grass present in the integrated systems studied (E+B, E+B+C, and E+B+P) contributed to TOC content and stock recovery, and to a significant accumulation of TOC.

Integrated systems that were not cultivated with nitrogen-fixing species (E+B and Euc) at the time of sampling for the evaluation of SOM dynamics showed soil TOC and TN contents and stocks similar to those containing these species (E+C, E+P, E+B+C, E+B+P). According to Wu et al. (2020), plant biomass production can promote increased soil TOC and TN contents, even in integrated systems that do not have nitrogen-fixing plants. In addition, fertilization practices performed over the years may have favored similar results to the soil TN.

Abreu et al. (2020) and Almeida et al. (2021) reported C/N ratios that varied from 10.98 to 14.94 and from 10.36 to 14.85, respectively, in studies involving integrated systems in the Cerrado biome. Corroborating with the authors, the values obtained in the present study ranged from 10.76 to 12.91 in AGS systems, while the Test treatment showed C/N ratios between 5.57 and 7.85 (Figure 1C). According to Wang, Adhikari, Wang, Jin, and Li (2018), different land use types alter the C/N ratio. C/N ratios close to 10 and 12 are considered normal in mineral soils (Toru & Kibret, 2019). The decrease in C/N ratio indicates progress in SOM mineralization and humification processes and reduction of particulate SOM content and primary plant residues that have a wide C/N ratio (Chen, Liu, Jiang, & Wu, 2017), which is in accordance with the lowest LC levels found in the Test system (Figure 1F) due to the low input of fresh organic material at this site.

### Labile Carbon (LC) and Non-labile Carbon (NLC)

The integrated farming systems E+B, E+C, E+P, E+B+C, and E+B+P showed higher levels of LC in the surface layer of the soil in the Test (Figure 1F). Despite the similar LC values ( $p > 0.05$ ) between the Euc and Test systems at a depth of 0-10 cm, an increase of 65.36% was observed in the Euc system. For other soil layers evaluated, gains in LC content of up to 61.34% (E+P at 10-20 cm depth), 37.25% (E+B+C at 10-20 cm depth), and 58.79% (E+P at the 0-40 cm layer) were identified with the conversion of the Test into integrated systems. Variations in LC content occur due to changes in land use and management practices that alter the input of organic matter (Gomes et al., 2021), confirming that LC is sensitive to changes in the soil SOM content and its dynamics can be used to infer SOM quality (Ndzelu et al., 2020), since it consists of a fraction that can be easily lost by decomposition (Santos et al., 2019).

Gomes et al. (2021) reported an increase of 26% in labile carbon content in agroforestry systems compared to monocultures, a result of species diversity and frequent deposition of plant residue. In the present study, increases of up to 103.35, 61.34, and 35.25% of LC were observed in layers 0-10, 10-20, and 20-40 cm, respectively, comparing the integrated systems with the Test treatment.

The significant reduction in LC levels in the Test system, mainly in the surface layer, showed a scarcity in biomass input and advance in the soil degradation stage. In the integrated systems, the more significant

deposition of litter and root volume promoted significant gains in LC over the years, mainly at 0-10 cm depth. Li et al. (2019) observed an increase in the carbon levels oxidized by potassium permanganate in management with a return of crop residues to the soil, and Ma et al. (2021) reported that residual crop roots and their exudates increase the soil quality indicator content.

LC is represented by organic compounds with low recalcitrance that can be used by microorganisms as an energy source (Ndzelu et al., 2020). Huang, Rinnan, Bruun, Engedal, and Bruun (2021) reported the presence of amides, polysaccharides, and carbohydrates among the labile constituents of carbon oxidized by potassium permanganate. According to Coser et al. (2018), diversification and an increase in plant residue input resulting from the implementation of integrated systems in the Cerrado may provide SOM accumulation in labile and recalcitrant fractions over a short period. According to the authors, increases in both fractions intensify the availability of nutrients because of the higher availability of labile fractions for mineralization and enable higher TOC storage through higher levels of SOM and more recalcitrant and stable fractions, allowing the principles of sustainable agriculture to be met. In the present study, increases in LC and NLC contents were achieved with the introduction and implementation of integrated systems, concluding that they met the requirements of sustainable agriculture.

According to Tadini et al. (2021b), SOM with more recalcitrant groups generally has a longer remaining in the soil, preventing fast CO<sub>2</sub> decomposition and release into the atmosphere, thus enabling a long-term contribution to climate change mitigation. Therefore, the increases in the NLC content in the integrated systems in the present study indicated an increase in SOM content with higher stability and lifetime (Figure 1G), suggesting that these systems are effective strategies for mitigating climate change. According to Tadini et al. (2021a), carbon is sequestered through humification of molecules and/or recalcitrant complexes. According to Bieluczyk et al. (2020), intercropping species with different root architectures and volumes contributes to improving the soil structure and favoring greater carbon stabilization in the soil.

### **Carbon in the fulvic acid (C-FA), humic acid (C-HA), and humin (C-HUM) fractions**

SOM chemical fractionation showed that most of the TOC was protected in the more stable and recalcitrant fraction (C-HUM), regardless of the system and soil depth (Table 2), corroborating other studies (Freitas et al., 2020; Almeida et al., 2021; Frazão et al., 2021). The C-HUM fraction comprises compounds of higher molecular weight, lower number of carboxylic groups, and higher number of nitrogenous groups, providing higher stability and remaining time in the soil (Pfleger, Cassol, & Mafra, 2017), allowing most TOC to be stored in this fraction. Coser et al. (2018) also related this storage to the formation of humus-clay colloidal complexes.

The organic carbon content of the soil in the C-FA, C-HA, and C-HUM fractions was higher in the integrated systems than in the Test at all depths evaluated (Table 2). Similarly, Coser et al. (2018) observed an increase in all humic fractions of the soil by implementing integrated production systems on low-productivity pastures in the Cerrado region. According to Bai, Guo, Huang, and An (2020), changes in soil plant cover significantly alter the balance between carbon input and output from the soil, affecting the total SOM content and fractions. Diversification and high intake of plant residues can promote the accumulation of labile SOM and stable fractions (Coser et al., 2018; Damian et al., 2021; Frazão et al., 2021).

Changes among integrated systems have also been observed, indicating that SOM fractions are sensitive to changes in soil management and use (Ramos et al., 2020). Higher levels of C-FA were found in E+B and E+P, at a 0-40 cm depth. The increase in C-FA levels in the E+B and E+P systems may have led to a reduction in the C-HA/C-FA ratio in the E+B and E+P in the 0-10 cm layer (Table 2). A lower C-HA/C-FA ratio among the integrated systems was also observed in E+B+P at the same depth, allowing us to infer that the integrated systems mentioned above contributed to increasing C-FA concentrations. It was observed that treatments with pigeon peas allowed good soil cover and biomass production, resulting in positive inputs of plant residues.

Corroborating the results obtained, Ramos et al. (2020) also identified that pigeon peas contribute to increasing the C-FA content of the soil in the Cerrado. According to Segnini et al. (2013), there may be an increase in the compound content of less recalcitrant structures when the plant residue input exceeds the capacity for microorganism metabolism. Otherwise, only the more recalcitrant SOM structures tended to remain. Bordonal et al. (2017) reported a decrease in the humification index (an increase in less recalcitrant structures) for larger carbon inlets.

Buffel grass in the E+B and E+B+P treatments may have contributed to the increase in C-FA content because of the higher production of fine roots. The grass root system is dense and consists mainly of fine roots

with easy decomposition, contributing to improved SOM storage (Cunha et al., 2020). Bai et al. (2020) also verified that pasture restoration increased soil fulvic acid content and observed a correlation with a higher production of root biomass. However, buffel grass did not show good development when intercropped with cowpea (E+B+C), since the cultivar used had indeterminate growth and a semi-erect stance and, consequently, higher competition for light.

An increase in the most recalcitrant fraction (C-HUM) was also observed in E+B and E+B+P in the 10-20 cm layer among the integrated farming systems (Table 2). Therefore, among the integrated systems, E+B and E+B+P had a higher contribution to the increase in different SOM fractions in lower and higher recalcitrance (C-FA and C-HUM). According to Nath, Brahma, Sileshi, and Das (2018), the chemical composition of cultivated species influences SOM lability, and increases in the recalcitrant and labile fraction concentrations indicate a balance in land use. Frazão et al. (2021) also observed a C-HUM increase in an integrated system with eucalyptus and pigeon pea in the Cerrado, indicating that increases in SOM's more humified fraction of SOM can lead to carbon sequestration over time.

The increase in C-HUM observed in the E+B and E+B+P systems may be related to the chemical composition and forage crop root biomass production, mainly buffel grass, because E+P did not follow the increases observed in C-HUM content (10-20 cm) among the mentioned integrated systems. Bai et al. (2020) reported that stable SOM fractions such as lignin and cellulose are mainly resistant to decomposition. Jalota et al. (2006) showed a lignin content of 14% in buffel grass roots, whereas other species studied, such as *Medicago sativa* and *Triticum aestivum*, presented 2.77 and 8.7%, respectively. Bai et al. (2020) reported that stable SOM fractions such as lignin and cellulose are mainly resistant to decomposition. In addition, Bieluczyk et al. (2020) observed that intercropping species with different root architectures and volumes improved the soil structure and favored greater carbon stabilization in the soil.

The C-HA/C-FA ratio indicates the SOM's humification and carbon mobility in the soil (Ramos et al., 2020) (Table 2). Regardless of the system and depth, the C-HA/C-FA ratio was higher than 1 (1.94 to 7.08), indicating C-HA prevalence. Of the two fractions (C-HA and C-FA), C-HA has higher stability, which usually leads to a higher concentration because C-FA is more labile and susceptible to changes (Gmach et al., 2018). C-FA is a humic fraction of lower molecular weight, higher number of carboxylic groups, higher oxygen content, and lower carbon content, resulting in higher solubility and easier dispersion in the soil profile (Pfleger et al., 2017).

The E+B and E+P systems presented a lower C-HA/C-FA ratio than the Test treatment in the 10-20 and 0-40 cm layers. High plant residue input, a high renewal rate of fine roots, and subsequent decomposition may explain the increase in more labile fractions of SOM (Pfleger et al., 2017). Among the integrated systems, a lower C-HA/C-FA ratio was observed in the surface layer in the E+B, E+P, and E+B+P systems, which can be explained by the increase in C-FA content throughout the soil profile (0-40 cm) (Table 2), as previously discussed.

The EA/C-HUM ratio showed high stability in SOM due to the C-HUM prevalence (Santos et al., 2019; Frazão et al., 2021) ranges from 0.19 to 0.60 (Table 2). However, the lowest EA/C-HUM ratio observed in the Test to the integrated systems in the 0-10 cm layer was a result of the plant residue's low input over the years, leading to oxidation of higher-liability compounds by microorganisms, remaining primarily in more recalcitrant structures of SOM (Segnini et al., 2013). This is supported by the carbon losses found in the total content and different SOM fractions from this system. Thus, the lowest EA/C-HUM ratio in the Test treatment was not correlated with a possible improvement in SOM quality associated with increased stability.

The EA/C-HUM ratios in the E+C (0.49), E+B+C (0.50), and Euc (0.51) systems differed from the Test (0.25) at 10-20 cm depth, possibly due to lower C-HUM values at this layer in these integrated systems, as previously discussed. In the subsequent layer (20-40 cm), the integrated systems had a similar EA/C-HUM ratio to the Test treatment, probably because of the lower influence of plant cover with the increase in depth (Pfleger et al., 2017).

No difference was observed in the EA/C-HUM ratio among the integrated farming systems and, based on the values obtained (<1), it was possible to infer that the integration of different production components promoted better SOM protection (Almeida et al., 2021). An increase in SOM stability is essential for sustainable agricultural intensification, as it reduces CO<sub>2</sub> emissions and enables more significant mitigation of climate change (Tadini et al., 2021a and b).

In summary, the results showed that integrated production systems can be used as a viable strategy for accumulating soil TOC and NT and improving SOM quality in low-productivity pastures. From a practical point of view, the results of the present study indicate how producers and field technicians/extensionists can improve land use in tropical conditions, particularly in the Cerrado biome. However, more studies about these

production models are required. These studies can encourage policies that promote the use of agrosilvopastoral systems by farmers as a strategy to recover degraded areas and/or mitigate greenhouse gas emissions.

## Conclusion

Integrated systems increased the total organic carbon and total nitrogen contents and stocks, non-labile carbon, and carbon content in the chemical fractions of soil organic matter at all evaluated depths, and presented total organic carbon and total nitrogen accumulation of 5.22 and 0.23 Mg ha<sup>-1</sup> year<sup>-1</sup>, respectively, in soil (0-40 cm layer). The integration of Eucalyptus with legumes or buffel grass increased the labile carbon content in the surface layer of the soil. The transition from low-productivity pasture into integrated farming systems can promote the recovery of soil organic matter and soil quality. Hence, our results suggest that agrosilvopastoral systems can be used as sustainable farming systems in the Cerrado biome.

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