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The Crop Succession Systems Under No-Tillage Alters the Surface Layer Soil Carbon Stock and Stability

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Abstract: The main challenge of the no-tillage system (NTS) is to reconcile productivity, the maintenance of surface residues, and the stabilization of soil organic matter (SOM). To address this challenge, particularly in tropical regions, various cover crops have been tested. The objective of this study was to test the effects of agricultural crop succession systems on the stock and stability of soil organic carbon in different surface layers of the soils. The research was carried out in the state of Goiás, Brazil, in an experiment set up in 2016, designed in randomized blocks with a split-plot scheme (treatments and soil layers), comprising four repetitions (blocks). The treatments (plots) consisted of crops grown in succession to soybean, which were as follows: T1—soybean/corn (*Zea mays*); T2—soybean/pearl millet (*Pennisetum glaucum*); T3—soybean/*Urochloa ruziziensis* (brachiaria); and T4—corn + *Urochloa ruziziensis*. The subplots represented the following soil layers: 0–5, 5–10, 10–20, and 20–40 cm. We evaluated the biomass dry mass and the soil parameters such as soil density, total porosity, and light organic matter across all layers. The organic carbon, grain size fractionation (mineral-associated organic carbon—MOC; sand-sized carbon—POC), and isotopic composition (δ13C) were determined in the 0–5 and 5–10 cm layers. The highest biomass dry production was observed in the soybean/pearl millet succession, which reduced the soil density and increased the total porosity in the surface layer. The soybean/pearl millet treatment produced high amounts of light organic matter, particularly in the 0–5 cm layer, a result also found for the soybean/brachiaria and soybean/corn + brachiaria systems. The crop successions did not alter the soil carbon stock or stability; however, the surface layer stored the highest amount of carbon, with elevated total organic carbon values and carbon stocks and stability (MOC and POC). Overall, in this study, replacing corn with other crops in succession with soybean did not affect the stock or stability of soil organic carbon. The species grown in succession with soybean contributed to the higher surface carbon stock and stability, promoting the formation of more stable and recalcitrant carbon.

Keywords: crop diversification; pearl millet; soil organic matter; soil density; physical fractionation; soil carbon pool; carbon storage

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1. Introduction

No-tillage system (NTS) is a conservation agriculture approach that fundamentally depends on crop diversification through rotation, the preservation of plant residues or cover crops, and minimal soil disturbance [1], which impacts soil chemical [2], physical [3], and biological properties [4], as well as productivity [5]. Consequently, it enhances water infiltration and storage capacity, reduces surface temperature, increases microbial activity, and promotes the accumulation of organic matter and nutrients in the upper soil layers [3,6].

In Brazil, the NTS is widely adopted, particularly in the local production areas of the Central-West region [7]; however, due to the climate of the Brazilian Savanna biome (i.e., Cerrado), the decomposition of plant biomass occurs at an accelerated rate, affecting the maintenance of crop residues on the soil surface [8]. In this context, employing cover crops, whether in rotation, succession, singly, intercropped, or mixed systems, with species exhibiting distinct functional traits can help maintain soil coverage for extended periods, consequently influencing nutrient availability [9], increasing productivity [10], and improving soil properties [2–4].

Leguminous species, for example, generally possess a low C/N (carbon/nitrogen) ratio and form associations with nitrogen-fixing microorganisms, facilitating nitrogen entry into the system and benefiting subsequent crop productivity [11]. In contrast, grasses, owing to their vigorous and abundant root systems, promote nutrient cycling at depth [12] and maintain soil cover for prolonged periods because of their high straw production and slow, gradual decomposition resulting from a high C/N ratio [13]. Thus, the implementation of NTS associated with different species (i.e., grasses and leguminous species) can contribute to key mechanisms for carbon protection. This process physically shields carbon by minimizing soil disturbance, thereby preserving the aggregate structure, inhibiting access for decomposing microorganisms and leading to enhanced carbon storage and a reduction in CO_2 emissions into the atmosphere [14]. In this sense, the NTS, when integrated with crop succession, can significantly increase organic carbon levels in the soil. These systems generally limit soil disturbance to the seeding rows, thereby reducing carbon loss and enhancing the retention of crop residues on the soil surface, which in turn improves soil properties [2–4]. Additionally, they maintain higher carbon stocks relative to conventional tillage systems. A recent study conducted in the Cerrado region revealed that NTS can accumulate organic carbon values in the surface soil layer (29.33 Mg ha^{-1}) comparable to those found in native forest ecosystems (37.51 Mg ha^{-1}) [15]. Although NTS is acknowledged for its positive impacts on soil properties, a notable knowledge gap persists concerning the effects of different crop cultures on soil organic carbon stocks and their stability.

Soil organic matter (SOM) is a carbon-based composite material that undergoes constant transformation in the soil due to decomposition processes. Owing to the presence of more recalcitrant and more labile fractions [16,17], SOM exhibits varying levels of resistance, which implies its durability and consequently its capacity to store carbon over time, leading to changes in the physical, chemical, and biological properties of the soil [18]. Therefore, investigating not only the total carbon content in the soil but also the distinct fractions in which organic matter is found is highly important. The physical fractionation of organic matter, for example, involves the separation of SOM into two fractions, the particulate organic carbon (POC), which is composed of particles derived from plant residues and hyphae, and mineral-associated organic carbon (MOC), which is the portion of SOM associated with the silt and clay particles in the soil matrix. This fraction interacts with the surfaces of mineral particles, leading to the formation of organomineral complexes, which are protected by colloidal protection mechanisms [19]. These different fractions of SOM play specific roles in nutrient cycling, soil stability, and carbon sequestration, thereby enhancing the understanding of soil quality and dynamics.

The objective of this study was to compare the dry biomass of crops and physical and chemical attributes of the soil across different agricultural crop succession systems under no-tillage in the Cerrado region of Brazil. We sought to answer the following questions: (i) Does agricultural crops succession result in varying levels of biomass dry production? (ii) Do the physical and chemical attributes of the soil differ among crops succession? (iii) Does crops succession lead to differences in the stock and stability of soil organic carbon?

2. Materials and Methods

2.1. Characterization of the Study Area

The study was conducted in the experimental area of the Federal University of Jataí (UFJ), Jatobá Campus, in the municipality of Jataí, state of Goiás, Brazil, at geographic coordinates 17°52′53″ S and 51°42′52″ W (Figure 1).

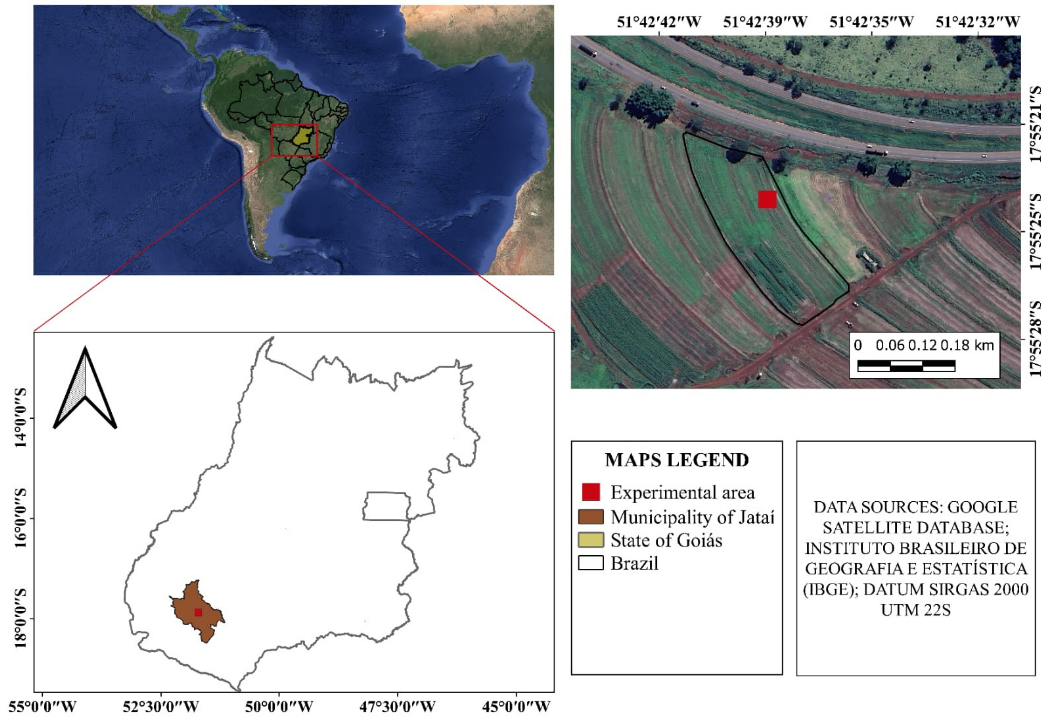

Figure 1. Study area location in Federal University of Jataí, Goiás State, Brazil.

The climate in the region is classified as Aw, a tropical savanna, according to the Köppen classification, featuring two well-defined seasons: a rainy summer and a dry winter, with an average annual temperature and average annual precipitation index of 23.7 °C and 1800 mm, respectively [20,21]. The average humidity recorded throughout the experiment was 67.72%, with a minimum daily value of 7% and a maximum of 83%.

The soil in the area is classified as dystrophic Red Latosol (Oxisol), characterized by a very clayey texture [22], with a pH in $CaCl_2$ of 5.2, cation exchange capacity (CEC) of 91 $mmol_c\ dm^{-3}$, base saturation (BS) of 58%, and organic matter (OM) content of 24 $g\ kg^{-1}$ (Table 1).

Table 1. Soil characteristics of the experimental area before the cultivation of the 2020/2021 soybean crop, in Jataí, Goiás.

Soil Layer	pH	OM	OC	P	S	Al	H + Al	K	Ca	Mg	SB	CEC	BS	Clay	Sand	Silt
0–20 cm	$CaCl_2$	$g\ kg^{-1}$		$mg\ dm^{-3}$	$mmol_c\ dm^{-3}$									%		
	5.2	24	14	25.2	16	0	38	2	35	15	53	91	58	59	24	17

pH: hydrogen ion potential in $CaCl_2$; OM: organic matter; OC: organic carbon; P: phosphorus; S: sulfur; Al: aluminum; H: hydrogen; K: potassium; Ca: calcium; Mg: magnesium; SB: sum of bases; CEC: cation exchange capacity; BS%: base saturation.

Figure 2 shows the climatological conditions during the experiment, collected from the Automatic Surface Observation Meteorological Station located at UFJ, belonging to the National Institute of Meteorology (INMET) (Figure 2).

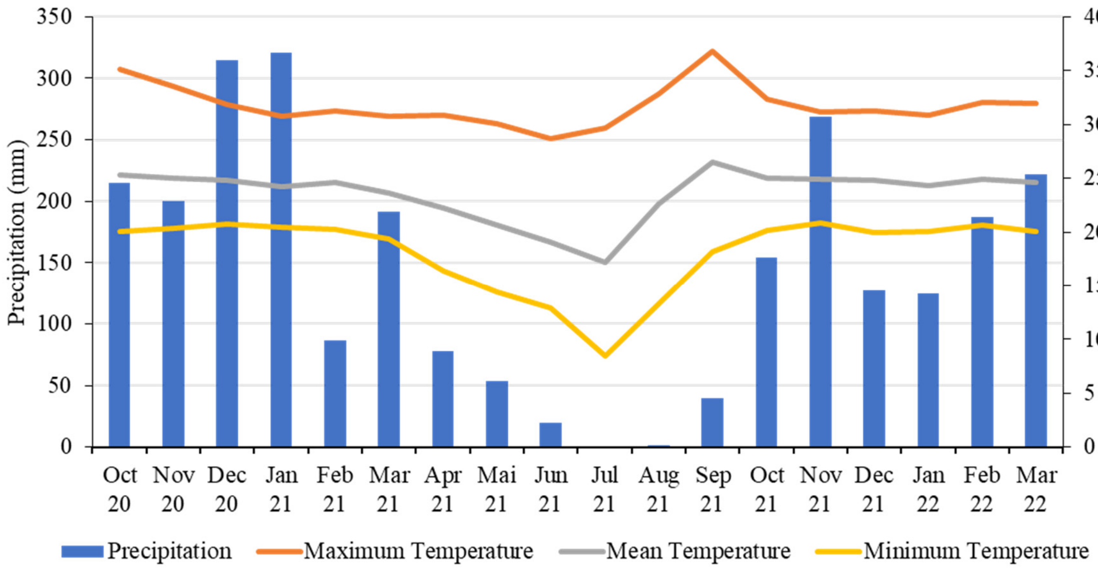

Figure 2. Maximum temperatures (T° max), Average temperatures (T° avg), Minimum temperatures (T° min), Rainfall (Precipitation mm) during the study, Jataí, GO, Brazil.

2.2. Experimental Design and Treatment Description

The experiment was established in a randomized block design with four treatments and four replications, in an experimental area of 1080 m^2 (60 × 18 m), where each experimental unit measured 67.5 m^2 (15 × 4.5 m). Agricultural crop succession treatments were set up in October 2016 with soybean sowing, corresponding to the spring/summer season, while the succeeding species were planted in March 2017 during the autumn/winter season. The treatments consisted of four agricultural succession systems: T1—soybean in the harvest and single corn (*Zea mays*) in the second harvest (Soybean-Corn); T2—soybean in harvest and millet (*Pennisetum glaucum*) in second harvest (Soybean-Pearl millet); T3—soybean in the harvest and brachiaria (*Urochloa ruziziensis*) in the second harvest (Soybean-brachiaria); and T4—soybean in the harvest and in the second harvest corn intercropped with brachiaria (Soybean-Corn + brachiaria) (Figure 3).

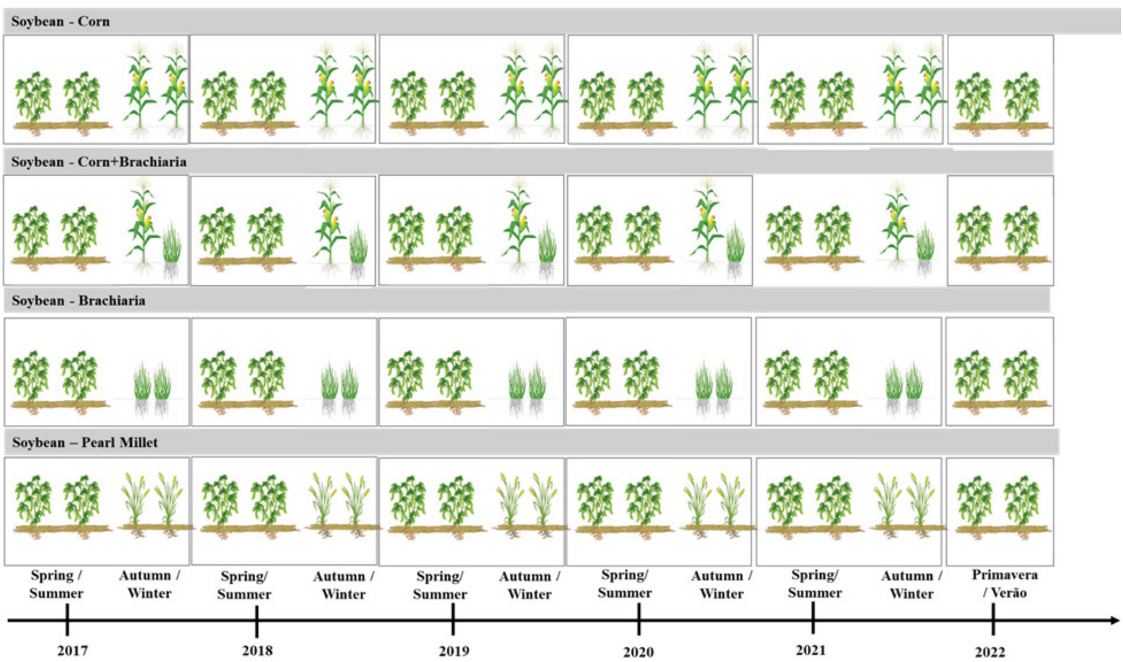

Figure 3. Arrangement of treatments since the installation of the experiment.

Soybean sowing throughout all experimental years (2016–2022) was carried out in October, at a spacing of 0.45 m and using cultivars recommended for the region, and crops sown in autumn/winter were also sown at a spacing of 0.45 m. Crop fertilization was carried out in all agricultural years in soybean crops (spring/summer) and in off-season crops only in corn and corn + brachiaria (autumn/winter) (Table S1, Supplementary Materials). For this study, the production systems characterized the treatments, which were sown on the same day during the autumn/winter season in March 2021. At the end of the cover crop cycle, only corn was harvested from ears on July 2021, while the other crops entered natural senescence and remained in the area.

2.3. Sampling and Evaluations

Sampling of biomass (successive species) and soil was conducted after the harvest of the autumn/winter 2020/21 crops on July 2021, and the biomass of soybean was collected after the harvest of the spring/summer 2021/22 crop on February 2022.

The biomass was sampled using a wooden frame measuring 30 × 30 cm (0.09 m^2). In each plot, three random simple samples were collected [23]. The samples were dried in an oven at 65 °C for 72 h and subsequently weighed to determine the dry biomass of crops, expressed in kg ha^{-1}.

Soil samples were collected 60 days after the harvest of corn from the autumn/winter crop, five years after the establishment of the experiment, with successive species under no-till conditions. A 40 cm deep trench was excavated in each plot, and both disturbed and undisturbed soil samples were collected from the 0–5, 5–10, 10–20, and 20–40 cm layers [22], which were sent to the Soil Laboratory at UFJ. The disturbed samples were air-dried, crushed, and sieved through a 2 mm mesh to obtain air-dried fine soil, whereas the undisturbed samples, which were collected via volumetric rings with diameters of 5.7 cm and heights of 6.3 cm, were standardized by removing excess soil (trimming) and used for mass and volume determination. The soil density was determined using the volumetric ring method, while particle density was evaluated through the volumetric balloon method applied to disturbed samples. Total porosity was determined using an indirect method [22].

To determine the light organic matter (LOM) content, 50 g of air-dried fine soil was weighed and placed in a 250 mL beaker, to which 100 mL of 0.1 mol L^{-1} NaOH solution was added, and the mixture was allowed to rest overnight [24]. After this period, the suspension was stirred with a glass rod, and all the material was passed through a 0.25 mm sieve, eliminating all the clay and silt fractions. The material retained on the sieve (LOM and sand) was subsequently transferred back to the beaker, the volume was filled with water, and the content was sieved again via a 0.25 mm sieve. Care was taken to separate the LOM from the sand fraction, and this operation was repeated until all the material floated and was removed. Finally, all material retained on the sieve (LOM) was transferred to pre-weighed trays, dried in an oven at 65 °C for 72 h, and then weighed, with the mass expressed in g kg^{-1}.

The stability of organic carbon was assessed using the physical fractionation method of SOM [25], which yields particulate organic carbon (POC) and mineral-associated organic carbon (MOC) from silt and clay. To achieve this goal, 10 g of soil was weighed and transferred to a Falcon tube, and 30 mL of sodium hexametaphosphate (5 g L^{-1}) was added. The tubes were agitated for 16 h on a horizontal shaker, and then all the material was passed through a 53 μm sieve and slowly washed with water. The material retained on the sieve, referred to as POC, was dried in an oven at 50 °C, quantified for its mass, ground in a mortar, and analyzed for total organic carbon (TOC) content [26]. The MOC was obtained from the difference

The total organic carbon (TOC) levels in the soil and their isotopic composition (δ13C) were determined via a Finnigan Delta Plus mass spectrometer at the Stable Isotope Center Prof. Dr. Carlos Ducatti—Institute of Biosciences, UNESP, Botucatu Campus, São Paulo, Brazil. The TOC results were expressed in g kg^{-1}, and the δ13C results were reported in ‰ relative to the international standard PDB (*Belemnitella americana* from the Pee Dee formation). Carbon stocks (CStock) were determined using the equation [27]:

$$\text{CStock} = (\text{C} \times \text{BD} \times \text{t})/10 \quad (1)$$

In which: CStock represents the accumulated carbon (Mg ha^{-1}), C represents the carbon content (g kg^{-1}), BD represents the bulk density (g cm^{-3}), and t, the thickness of the layer under analysis, is measured in meters.

2.4. Statistical Analyses

The data were subjected to analysis of variance using the F test at a 5% probability of error. When statistical significance was observed, the treatment means were grouped using the Scott–Knott clustering method at a 5% probability of error. Data analysis was conducted using the AgroEstat statistical software 1.1.0.712 [28]. Additionally, Pearson correlation analysis among the soil attributes was performed using R software® version 4.3.0 [29]. For this analysis, data from the 0–10 cm soil layer were used, which were derived from the arithmetic means of the 0–5 cm and 5–10 cm layers.

3. Results

3.1. Dry Biomass of Crops and Light Organic Matter

The accumulated dry biomass of crops in the spring/summer and autumn/winter seasons varies from 14,813.89 kg ha^{-1} year^{-1} (soybean/brachiaria) to 18,055.57 kg ha^{-1} year^{-1} (soybean/pearl millet) (Figure 4). The mean dry biomass values differed only for the autumn/winter season and were grouped into three clusters (Figure 4). The first group was the soybean/pearl millet treatment, which produced the highest biomass, followed by the soybean/brachiaria and soybean/corn + brachiaria treatments in the intermediate group, and the soybean/corn treatment had the lowest biomass (Figure 4).

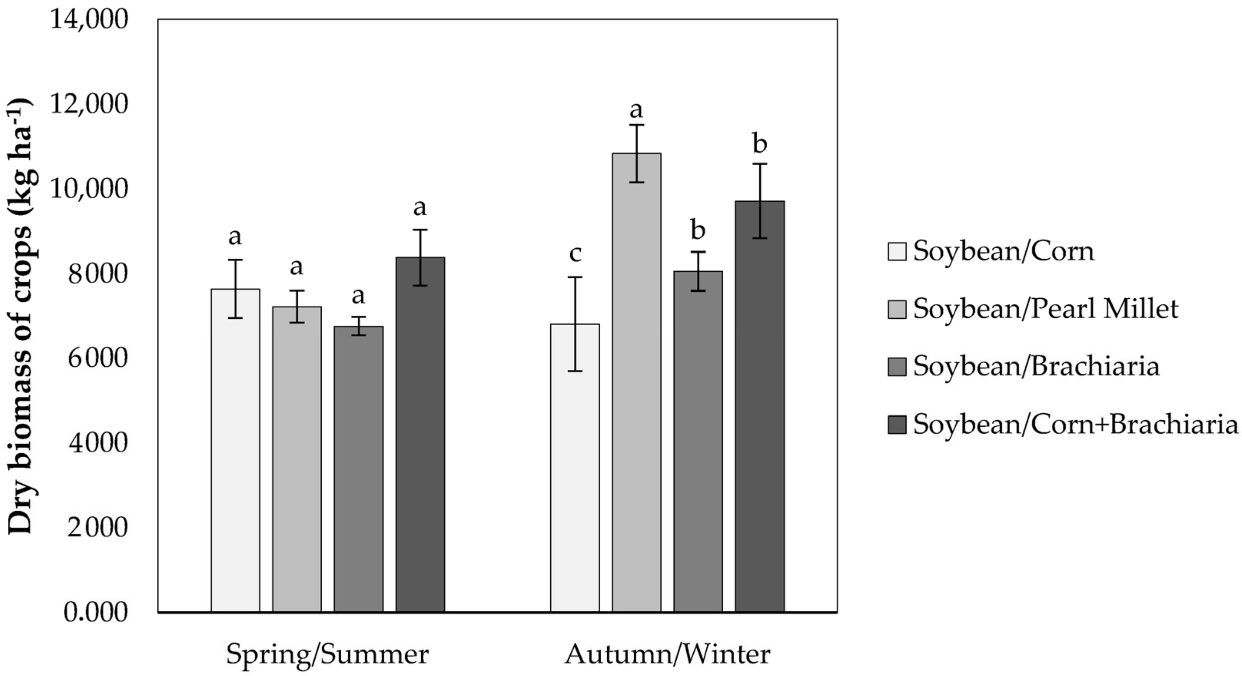

Figure 4. Biomass dry mass of straw (kg ha^{-1}) in the spring/summer and autumn/winter harvests. Jataí—GO, Brazil. Bars indicate mean values ± standard error. Means followed by the same letter for each harvest are grouped together according to the Scott–Knott test at 5% probability (CV%—Coefficient of variation = 7.48 spring/summer; 11.40 autumn/winter).

With respect to the levels of light organic matter (LOM), no differences were detected among the soil layers in the soybean/corn treatment (Table 2). In the soybean/pearl millet and soybean/brachiaria treatments, two groups formed, with the highest LOM concentrations found in the most superficial soil layer. Although the soybean/corn + brachiaria treatment also separated the depths into two groups, the highest LOM values were present in the two upper layers, 0–5 cm and 5–10 cm.

Table 2. Light organic matter (g kg^{-1}) among treatments with crop succession and soil layers, in Jataí, GO, Brazil.

Soil Layer (cm)	Light Organic Matter (g kg^{-1})				CV (%)
	Crop Succession				
	Soybean/ Corn	Soybean/ Pearl Millet	Soybean/ Brachiaria	Soybean/ Corn + Brachiaria	
0–5	0.88 ± 0.15 Ab	2.24 ± 0.41 Aa	1.68 ± 0.26 Aa	1.86 ± 0.39 Aa	52.26
5–10	0.36 ± 0.08 Ab	0.35 ± 0.10 Bb	0.43 ± 0.10 Bb	0.97 ± 0.29 Aa	
10–20	0.25 ± 0.05 Aa	0.23 ± 0.05 Ba	0.23 ± 0.07 Ba	0.30 ± 0.09 Ba	
20–40	0.16 ± 0.02 Aa	0.09 ± 0.02 Ba	0.21 ± 0.05 Ba	0.12 ± 0.05 Ba	
CV (%)	55.92				

Means ± standard error followed by the same uppercase letters in the columns and lowercase letters in the rows belong to the same group according to the Scott–Knott test at 5% probability. CV%—Coefficient of variation.

The treatments were categorized into groups based on light organic matter (LOM) content in the 0–5 cm and 5–10 cm soil layers (Table 2). In the 0–5 cm layer, the group exhibiting the lowest LOM content was exclusively represented by the soybean/corn succession. Conversely, in the 5–10 cm layer, the group with the highest LOM content was comprised of the soybean/brachiaria treatment (Table 2).

3.2. Soil Density and Total Porosity

The density values ranged from 1.16 to 1.45 g cm^{-3}, corresponding to the soybean/pearl millet treatment in the 0–5 cm and 5–10 cm soil layers, respectively (Table 3). Differences in density values among the treatments were observed in the 5–10 cm and 10–20 cm soil layers (Table 3). In the 5–10 cm layer, two groups were formed, one with a lower density for the soybean/pearl millet treatment (1.16 g cm^{-3}) and another with higher density values for the remaining treatments (1.30 to 1.39 g cm^{-3}). In the 10–20 cm layer, two groups also emerged, with lower density values for the soybean/corn and soybean/brachiaria treatments (1.26 and 1.28 g cm^{-3}, respectively) and higher values for the soybean/ pearl millet and soybean/corn + brachiaria treatments, with values of 1.38 and 1.43 g cm^{-3}, respectively (Table 3).

Table 3. Bulk density (g cm^{-3}) and total porosity (%) among crop succession and soil layers, in Jataí, GO, Brazil.

Soil Layer (cm)	Bulk Density (g cm^{-3})				CV (%)
	Crop Succession				
	Soybean/ Corn	Soybean/ Pearl Millet	Soybean/ Brachiaria	Soybean/ Corn + Brachiaria	
0–5	1.41 ± 0.03 Aa	1.45 ± 0.07 Aa	1.36 ± 0.06 Aa	1.38 ± 0.04 Aa	6.25
5–10	1.39 ± 0.03 Aa	1.16 ± 0.02 Bb	1.32 ± 0.05 Aa	1.30 ± 0.06 Aa	
10–20	1.26 ± 0.03 Bb	1.38 ± 0.05 Aa	1.28 ± 0.03 Ab	1.43 ± 0.03 Aa	
20–40	1.39 ± 0.04 Aa	1.36 ± 0.02 Aa	1.33 ± 0.04 Aa	1.33 ± 0.05 Aa	
CV (%)	6.71				

Table 3. *Cont.*

Soil Layer (cm)	Total Porosity (%)				CV (%)
	Crop Succession				
	Soybean/ Corn	Soybean/ Pearl Millet	Soybean/ Brachiaria	Soybean/ Corn + Brachiaria	
0–5	42.0 ± 1.7 Aa	43.3 ± 1.3 Ba	43.3 ± 4.3 Aa	45.0 ± 1.6 Aa	10.46
5–10	44.5 ± 7.4 Ab	53.8 ± 0.8 Aa	45.0 ± 3.4 Ab	45.0 ± 5.0 Ab	
10–20	49.5 ± 4.3 Aa	41.0 ± 3.5 Bb	49.8 ± 1.5 Aa	43.0 ± 1.4 Ab	
20–40	44.3 ± 1.6 Aa	49.5 ± 1.7 Aa	44.8 ± 3.6 Aa	45.5 ± 2.1 Aa	
CV (%)	12.81				

Means ± standard error followed by the same uppercase letters in the columns and lowercase letters in the rows belong to the same group according to the Scott–Knott test at 5% probability. CV%—Coefficient of variation.

Comparisons among the soil layers indicated significant differences for the soybean/corn and soybean/pearl millet treatments. Specifically, the soybean/corn treatment exhibited the lowest density in the 10–20 cm layer, while the soybean/pearl millet treatment demonstrated the lowest density in the 5–10 cm layer (Table 3).

The total soil porosity also differed among the treatments in the 5–10 cm and 10–20 cm layers (Table 3), with differences between the layers observed for the soybean/pearl millet treatment. In the 5–10 cm layer, two groups were formed, one represented by the soybean/pearl millet treatment, which had the highest porosity value (53.8%), and the other comprising the remaining treatments (44.5 to 45.0%), which had lower values. In the 10–20 cm layer, the first group with the highest averages included soybean/corn (49.5%) and soybean/brachiaria (49.8%) treatments, while the soybean/pearl millet and soybean/corn + brachiaria presented values of 41% and 43%, respectively. For the soybean/pearl millet treatment, two distinct groups were identified, with the 5–10 cm and 20–40 cm soil layers exhibiting the highest average values, while the 0–5 cm and 10–20 cm layers displayed the lowest averages (Table 3).

3.3. Organic Carbon in Soil

The values for total organic carbon (TOC), fractionated organic carbon (particulate organic carbon—POC and mineral-associated organic carbon—MOC), and carbon stocks (CStock) did not show significant differences among treatments ($p > 0.05$). However, differences were observed between the 0–5 cm and 5–10 cm layers (Table 4), with the uppermost layer (0–5 cm) exhibiting higher values (Table 4). The distribution of δ13C (‰) ranged from −17.43 to −16.89 ‰ for the soybean/corn and soybean/corn + brachiaria treatments, respectively, with no significant differences observed among the treatments ($p > 0.05$). There were also no significant differences between layers, with values varying from −17.43 ‰ for the 0–5 cm to −16.89 ‰ for the 5–10 cm layer (Table 4).

Table 4. Total organic carbon (TOC), particulate organic carbon (POC), mineral-associated organic carbon (MOC), isotopes (δ13C (‰), and carbon stock in the 0–5 and 5–10 cm layers of the soil. Jataí, GO, Brazil.

Soil Layer (cm)	TOC (g kg^{-1})	POC (g kg^{-1})	MOC (g kg^{-1})	δ13C (‰)	CStock (Mg ha^{-1})
0–5	64.64 ± 3.7 a	1.17 ± 0.70 a	63.46 ± 3.6 a	−17.43 ± 0.2 a	45.08 ± 2.6 a
5–10	44.41 ± 2.5 b	0.53 ± 0.04 b	43.88 ± 2.4 b	−16.89 ± 0.2 a	28.44 ± 1.4 b
CV (%)	17.56	60.73	17.44	−4.46	19.12

Means ± standard error followed by same letters in the columns did not differ in groups by the Scott–Knott test at 5% probability. CV%—Coefficient of variation.

3.4. Correlations Between Soil Attributes

Total organic carbon exhibited correlations with the carbon fractions (COP and MOC), with values of 0.68 and 1.00, respectively (Figure 5). Similarly, the TOC content is strongly correlated with the soil carbon stock (0.96). TOC and soil bulk density (BD) were negatively correlated (−0.66), whereas the correlation of TOC with total soil porosity was positive (0.74). MOC is also strongly correlated with carbon stock (0.96), particulate organic carbon (0.67) and total porosity (0.75), whereas it is negatively correlated with bulk density (−0.66) (Figure 5).

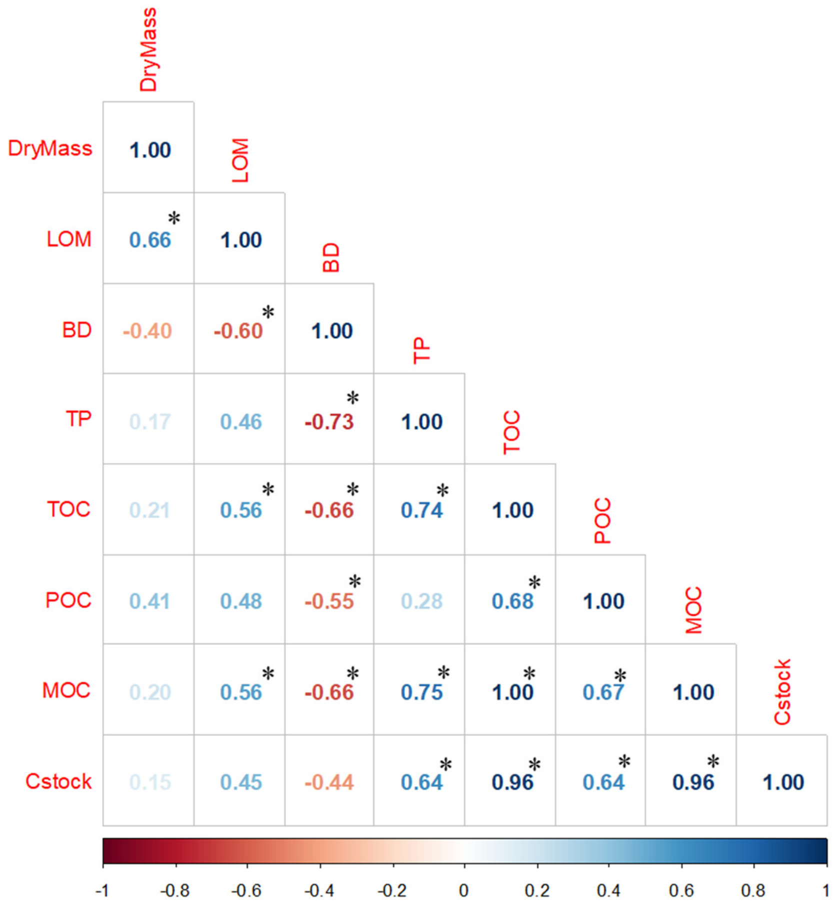

Figure 5. Pearson correlations between soil attributes (0–10 cm) in Jataí, GO, Brazil. * Represents significant correlations ($p < 0.05$). Legend: light organic matter (LOM); bulk density (BD); total porosity (TP); total organic carbon (TOC); particulate organic carbon (POC); mineral-associated organic carbon (MOC); carbon stock (Cstock).

4. Discussion

4.1. Dry Biomass and Light Organic Matter

The accumulated dry biomass found in this study exceeds the recommended amount to meet the annual straw demand in the Cerrado (6000 to 12,000 kg ha^{-1} year^{-1}) [30,31]. In the Cerrado regions, the high amount of dry biomass required for effective soil protection is attributed to the rapid decomposition rates that occur in tropical regions due to high rainfall levels combined with elevated temperatures (Figure 2). Differences in dry biomass production among preceding crops and soybean have also been reported by other authors [32], who indicate that variations in dry biomass production can arise from species differences, intrinsic management factors, climatic conditions, soil attributes, phytosanitary conditions, and depth of root development [33].

The higher dry biomass production in the soybean/pearl millet treatment during the autumn/winter season (10,833 kg ha^{-1}) supports findings from other studies in tropical climates in Brazil, where the authors reported a biomass production of 9993 kg ha^{-1} in the municipality of Ilha Solteira, São Paulo [34], and between 8600 and 9800 kg ha^{-1} in the Triângulo Mineiro region of Minas Gerais [35]. The greater biomass production of soybean/pearl millet, particularly millet, may be attributed to its adaptation to the climatic conditions of the region, as the species exhibits rapid growth even under water stress. Under favorable conditions, its root system explores a relatively large lateral area and depth [36], contributing to increased water uptake and nutrient cycling. Therefore, the high rainfall levels experienced in the month of crop establishment may have facilitated the growth of aerial biomass, resulting in elevated biomass production [36].

Both treatments with intermediate production of biomass utilized brachiaria grass as a cover crop during the autumn/winter season. The average biomass production between the intercropping system (corn + brachiaria) and corn alone was 2908 kg ha^{-1}, a result similar to that reported by other authors [37], who reported an increase of 2520 kg ha^{-1} in biomass production for corn intercropped with brachiaria compared with corn alone. Moreover, brachiaria grown alone produced 8056 kg ha^{-1}, which is comparable to the results of another study [38], where brachiaria alone yielded 9600 kg ha^{-1}. These results indicate that brachiaria, whether grown in combination or alone, has the capacity to produce biomass within the ideal range for the Cerrado region (6000 to 12,000 kg ha^{-1} year^{-1}). Brachiaria (*Urochloa ruziziensis*) is a grass with a relatively long vegetative cycle, exhibiting high photosynthetic efficiency and the ability to accumulate a significant amount of aerial biomass. A deeper and denser root system contributes to the addition of organic matter to the soil and enhances the soil structure, facilitating the uptake of water and nutrients [39]. By creating a favorable environment for the growth of other crops, such as corn, brachiaria can promote an increase in biomass production, which may ensure sustainability for the following crop. Given its high C/N ratio, the decomposition rate of the biomass is reduced [40]. These characteristics provide greater soil protection against erosion and radiation [41], reduce weed infestations [42], and maintain soil moisture and water retention capacity [41].

Light organic matter (LOM) is an important attribute for determining the quality of an adopted management system. In conservationist systems such as no-till farming, which promotes the accumulation of residues in the surface layer of the soil, there is an increase in SOM levels, as residues are the primary source of SOM formation [43]. This finding confirms the results obtained in this study, where higher levels of SOM were detected in the surface layers for all crop treatments. Similar results were found in a chronosequence study in no-till systems in an area with 20 years of implementation, where the authors also reported higher LOM contents in the surface layer, with values comparable to those in areas of the native Cerrado [43]. Thus, the incorporation of cover crops in conservation systems can enhance light organic matter levels [44], particularly when utilizing grass species. These plants contribute to increased organic matter accumulation at the soil surface due to their continuously renewing fibrous root systems, which facilitate nutrient cycling through enhanced microbial activity [45]. Furthermore, grasses exhibit a significant quantity of roots in the arable layer of the soil, with a high C/N ratio, explaining the prolonged presence of this material on the soil for extended periods, thereby favoring an increase in LOM levels [46], which supports the results obtained in this study.

4.2. Soil Density and Total Porosity

The average soil density values ranged from 1.16 to 1.45 g cm^{-3} (Table 3), which are within the expected range for soils under natural conditions without signs of compaction [47]. Overall, higher average soil density values were observed in the 0–5 cm layer, which may be related to the pressure exerted on the soil by the traffic of agricultural machinery and implements [48], particularly during the planting and harvesting of crops. The density values obtained in the present study, regardless of the layer, were lower than those reported in a study of no-tillage systems conducted on Latosol soils under Cerrado conditions [49]. Conversely, the soybean/corn and soybean/pearl millet treatments resulted in a lower density in deeper layers, which was associated with greater root volume in those layers [50,51].

Total porosity values above 50% are considered ideal for crops [52]. In this study, only the soybean/pearl millet treatment at the 5–10 cm depth presented values above the ideal threshold, whereas the soybean/corn and soybean/brachiaria treatments showed values close to this threshold in the 10–20 cm layer. Some studies have demonstrated the effects of millet on total porosity, with a value of 49.75% in the 0–20 cm layer [53], which can be attributed to the increased organic matter contributed by the crop to the upper soil layer [54]. For corn and brachiaria crops, improvements in density and porosity in subsurface layers have been associated with the root systems of the grasses, which, upon decomposition after death, form biopores, thereby contributing to increased total porosity [55].

The variations observed in soil density and total porosity across layers can be explained by the stabilization process associated with no-till systems. Regardless of the crop used, no-till systems begin to stabilize total porosity and soil density starting in the sixth year of adoption, whereas variances may occur along the soil profile during the initial years [56]. These authors explain that it is common for soils to present higher density values and lower total porosities in subsurface layers because of the natural rearrangement of soil particles. However, other studies have indicated that the benefits associated with no-till systems become noticeably evident during the second phase, referred to as the transitional phase, which typically occurs approximately 10 years following the implementation of the system [57] and may persist for up to 12 years [58].

4.3. Organic Carbon in Soil

The higher values for total organic carbon found at the surface are attributed primarily to the maintenance of plant residues on the soil surface. Generally, organic matter levels are obtained through the recurring balance between the addition and decomposition of organic material and microbial activity in the soil. Therefore, the greater the addition and decomposition of this material by soil microorganisms, the higher the total organic carbon content will be [59]. Even in no-till systems, a decrease in total organic carbon with increasing depth is common, following the same pattern observed in other studies [49,54,60]. This stratification is attributed to the surface deposition of aerial residues and the enhanced root growth in the upper layers, especially from species such as grasses [49,61].

The no-till systems may have influenced the enhanced superficial accumulation of particulate organic carbon (POC) due to its practice of maintaining plant residues on the soil surface and the specific type of cover employed. The presence of these residues contributes to increased carbon input into the soil system, thereby promoting higher levels of carbon sequestration [62]. Since a significant portion of particulate organic carbon is formed from residual plant particles directly related to recently added material, the use of crops such as grasses promotes greater residue contributions to the soil surface, leading to increased particulate organic carbon levels [43]. Even greater increases than those observed in NTS can occur in integrated systems, such as integrated crop–livestock systems [63]. In the present study, approximately 2% of the TOC in the 0–5 cm layer was represented by particulate organic carbon, which may be linked to the climatic conditions of the region, as high temperatures and humidity favor microbial activity. This fraction is considered the most labile of the organic matter, forming a fragile carbon reserve that can be rapidly decomposed and lost when the soil is exposed to cultivation [64].

Despite showing a similar response to particulate organic carbon, specifically a decrease in content with increasing depth (this study, [65]), there is a negative correlation between POC and mineral-associated organic carbon (MOC) levels in the soil [66], indicating that the processes forming these fractions are opposed. In other words, for an increase in MOC levels to occur, there must be greater decomposition of POC, which is subsequently associated with silt and clay minerals. This trend was observed in the study area, regardless of the successive species used. Notably, the response of the MOC fraction is observed over the medium to long term [67], as it is more stable than POC is, a characteristic associated with the advancement of humification stages, interactions with the mineral fraction of the soil, and the physical protection provided by its location within microaggregates [68].

In this study, approximately 98% of the TOC content in the 0–5 cm layer was associated with minerals (MOC), differing from the results of another study that noted that 69% of the TOC was represented by MOC in the 0–5 cm layer [64]. Nonetheless, the higher carbon stocks found in the more recalcitrant fractions (MOC) may be attributed to the interactions of organomineral complexes formed in Latosol soils, leading to strong associations of humified organic matter with the kaolinitic and oxidic clays characteristic of these soils. Additionally, the higher clay content in these soils increases the number of micropores, enhancing the stabilization potential of organic matter against biological attacks [69].

The distribution of δ13C (‰) was similar to those obtained for no-till cultivation under Cerrado conditions [70]. The results obtained for δ13C (‰) indicate a greater contribution of carbon from C4 plants (grasses) [71], which may be generally associated with the greater input of crop residues from corn, millet, and brachiaria than from soybean. Additionally, the biomass of fine roots from grasses may have contributed to the isotopic signal of C4 plants, as observed in other studies [70].

A higher carbon stock was found in the surface layer (0–5 cm), which may be related to the amount of crop residue deposited on the soil surface, with a decrease observed with increasing depth (Table 4). A study also noted this decline as the depth increased in Latosol, attributing this result to the greater input of organic material in the surface layer [54]. Moreover, the accumulation of residues on the soil surface significantly contributes to the carbon stock [72], corroborating the results obtained in this research.

Soil texture is directly associated with carbon stocks. In Latosols, which are soils with medium or fine textures, i.e., clayey or very clayey, soil aggregation is favored, playing an important role in preventing the decomposition of organic matter. On average, Latosols have the capacity to store approximately 53.2 Mg ha^{-1} of carbon [73]. The carbon stock obtained in this study (45.08 kg ha^{-1}) is below the average for Latosols, on the basis of the aforementioned study. This result may be explained by the duration of the no-till implementation (6 years), as the accumulation of carbon in these systems occurs very slowly, taking from 10 to 15 years for the stock to become significant [74]. However, other factors may condition carbon stocks in no-till systems beyond the duration of implementation, such as climatic conditions and management practices, as studies in areas with less than 10 years of implementation have reported higher carbon stocks than other management systems [68].

The stronger correlation between total organic carbon (TOC) and mineral-associated organic carbon (MOC) is due to the predominance of this fraction in the studied soil; as previously explained, strong associations of humified organic matter with kaolinitic and oxidic clays occur [64,69]. A similar result was obtained in another study, where the authors explained that the accumulation of TOC depends on the increase in LOM levels, suggesting that the stabilization of organic matter is fundamental for the increase in TOC levels [75]. The soil organic carbon content is negatively correlated with soil density [76], which is explained by the low particle density and the cementing action of organic carbon in macroaggregates, increasing the macroporosity of the soil [77].

5. Conclusions

The soybean/pearl millet succession enhances dry biomass at the soil surface, while contributing to increased soil total porosity and a reduction in soil density.

The replacement of corn with alternative species in succession with soybean did not significantly influence soil carbon levels. However, both crops grown in succession with soybean enhanced carbon stock and stability at the soil surface, promoting the formation of more stable and recalcitrant carbon compounds.

Based on our findings, it is recommended to integrate pearl millet as an alternative species into cropping systems in succession with soybean. This practice enhances dry biomass at the soil surface, improves physical soil properties, and ultimately can contributes to increased carbon stock and stability over time, suggesting a gradual enhancement of soil quality.

Supplementary Materials: The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/agriculture14112085/s1, Table S1: Cultivar, sowing density and fertilizer management carried out in the 2016-2022 harvests in each crop.

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