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Soil carbon dynamics in a long-term corn-soybean rotation system with cover crops in succession

Abstract – The objective of this work was to evaluate changes in soil carbon contents and stocks in a ten-year long experiment under no-tillage, with the transition from corn to soybean, with cover crops and spontaneous vegetation in succession. The experimental design was a randomized complete block with three replicates, using ten different off-season covers: pigeon pea (*Cajanus cajan*), Brazilian jackbean (*Canavalia brasiliensis*), sunn hemp (*Crotalaria juncea*), black velvet bean (*Mucuna aterrima*), pearl millet (*Pennisetum glaucum*), radish (*Raphanus sativus*), sorghum (*Sorghum bicolor*), wheat (*Triticum aestivum*), ruzigrass (*Urochloa ruziziensis*), and spontaneous vegetation. Soil samples were collected in 2013, 2018, and 2024 at the depths of 0–10, 10–20, 20–30, 30–40, 40–60, 60–80, and 80–100 cm. Soil carbon stocks increased from 2013 to 2018, but declined between 2018 and 2024 after the introduction of soybean in 2021, except in treatments with sorghum and wheat. Between 2013 and 2024, soil carbon contents varied according to depth and type of cover crop. The corn-soybean rotation under no-tillage, with cover crops in succession, influences soil carbon dynamics. Sorghum shows the highest carbon accumulation between 2013 and 2024.

Index terms: climate change, management, mitigation, no-tillage, sustainability, tropical agriculture.

Dinâmica do carbono do solo em sistema de rotação milho-soja de longa duração com plantas de cobertura em sucessão

Resumo – O objetivo deste trabalho foi avaliar variações no teor e nos estoques de carbono do solo em experimento com duração de dez anos, em plantio direto, com transição de milho à soja, com plantas de cobertura e vegetação espontânea em sucessão. O delineamento experimental foi em blocos ao acaso, com três repetições, tendo-se utilizado dez tipos de cobertura na entressafra: guandu (*Cajanus cajan*), feijão-bravo-do-ceará (*Canavalia brasiliensis*), crotalária (*Crotalaria juncea*), mucuna-preta (*Mucuna aterrima*), milheto (*Pennisetum glaucum*), nabo-forrageiro (*Raphanus sativus*), sorgo (*Sorghum bicolor*), trigo (*Triticum aestivum*), braquiária ruziziensis (*Urochloa ruziziensis*) e vegetação espontânea. Amostragens de solo foram coletadas em 2013, 2018 e 2024, nas profundidades de 0–10, 10–20, 20–30, 30–40, 40–60, 60–80 e 80–100 cm. Os estoques de carbono do solo aumentaram de 2013 a 2018, mas declinaram entre 2018 e 2024 após a introdução da soja em 2021, exceto nos tratamentos com sorgo e trigo. Entre 2013 e 2024, os teores de carbono do solo variaram de acordo com a profundidade e o tipo de cobertura.

A rotação milho-soja sob plantio direto, com culturas de cobertura em sucessão, influencia a dinâmica do carbono do solo. O sorgo apresenta o maior acúmulo de carbono entre 2013 e 2024.

Termos para indexação: mudanças climáticas, manejo, mitigação, plantio direto, sustentabilidade, agricultura tropical.

Introduction

Since the pre-industrial period, atmospheric CO₂ concentration has increased by approximately 47%, from 278.3 ppm in 1750 to 409.9 ppm in 2019, intensifying global warming and climate change (Forster et al., 2021). In this scenario, the food system is a significant source of CO₂, with net CO₂ emissions related to land use and agriculture estimated to represent 14 and 10% of annual anthropogenic contributions, respectively (Le Quéré et al., 2018; Mbow et al., 2019). Therefore, considering the significant contribution of the agriculture sector to global greenhouse gas (GHG) emissions, efforts are necessary to reduce impacts on atmospheric CO₂ levels (Lynch et al., 2021).

Soil carbon plays a critical role in the global C cycle as it is one of the largest terrestrial C reservoirs, storing approximately three times more than the atmosphere (Sanderman et al., 2017). That stock is regulated by the balance between inputs, via plant residues, and outputs, through C mineralization driven by microorganisms (Chowdhury et al., 2021; Bhattacharyya et al., 2022). Agricultural soils can act as C sinks through the adoption of C sequestration practices, which vary depending on climate, soil characteristics, and agricultural management (Paustian et al., 2019; Carvalho et al., 2023). In the Brazilian Cerrado, agroecosystems without soil disturbance, such as the no-tillage system and pasture, show a higher C accumulation, functioning as a soil sink for atmospheric CO₂ (Corazza et al., 1999; Sant-Anna et al., 2017; Ayarza et al., 2022).

To ensure the sustainability of a food system, a strategy is the adoption of low-C agricultural techniques, characterized by low direct GHG emissions and a high soil C storage, affecting soil quality and crop yield, in addition to contributing to the mitigation of CO₂ emissions (Sá et al., 2017). An example of a low-C agricultural practice is increasing crop diversification with cover crops under no-tillage, as pointed out by Sá et al. (2017). The same authors highlighted that this system, based on conservation agriculture principles,

has been shown to effectively restore soil organic C stocks in Brazil's biomes, consequently playing an important role in integrating agriculture as a part of the mitigation solutions in climate change.

Soil C stocks are affected by the quality of the crop residue deposited in cropping systems under no-tillage, considered a key factor in C retention and sequestration (Rigon et al., 2021). The chemical composition of crop residues, such as lignin, hemicellulose, and cellulose, as well as their N content, regulates the rate of decomposition, affecting the storage of organic C in the soil (Carvalho et al., 2011, 2013, 2022a). Additionally, residue quality parameters such as the C-to-nitrogen ratio, lignin-to-N ratio, and initial N content, influence residue decomposition and C mineralization. Crop residues with a higher N content and lower C:N ratios tend to decompose more rapidly, contributing to nutrient cycling, whereas residues with lower N content and higher C:N ratios decompose more slowly, providing soil cover and promoting long-term soil C storage (Adhikari et al., 2024).

Low-C agriculture, such as the management strategies employed in the present study, aligns with the objectives of the 30th annual United Nations Climate Change Conference, which aims to prioritize the development of strategic solutions guided by the Agenda for Sustainable Development of the United Nations (United Nations, 2015). In Brazil, numerous studies have been conducted to assess soil C stocks under different management practices and agricultural species, such as cash crops and cover/companion crops (Corazza et al., 1999; Marchão et al., 2009; Ferreira et al., 2016; Ayarza et al., 2022; Carvalho et al., 2023; Oliveira et al., 2023). However, researches are still scarce regarding transitions between the two most representative cash crops in the country – soybean [*Glycine max* (L.) Merr.] and corn (*Zea mays* L.) – grown with a diverse set of cover crop species in succession.

The objective of this work was to evaluate changes in soil C contents and stocks in a ten-year long experiment under no-tillage, with the transition from corn to soybean, with cover crops and spontaneous vegetation in succession.

Materials and Methods

The study, part of a larger multi-year research project, was conducted in an experimental area at

Embrapa Cerrados, located in the municipality of Planaltina, District Federal, Brazil ($15^{\circ}35'30''S$, $47^{\circ}42'30''W$, at 990 m above sea level). The experiment was arranged in a randomized complete block design with three replicates in 12x8 m plots. The ten evaluated treatments consisted of the following nine cover crops plus spontaneous vegetation (Figure 1): pigeon pea [*Cajanus cajan* (L.) Millsp.], Brazilian jackbean (*Canavalia brasiliensis* Mart. ex Benth.), sunn hemp (*Crotalaria juncea* L.), black velvet bean (*Mucuna aterrima* Holland), pearl millet (*Pennisetum glaucum* R.Br.), radish (*Raphanus sativus* L.), sorghum [*Sorghum bicolor* (L.) Moench], and wheat (*Triticum aestivum* L.).

During the experiment, the mean annual rainfall and mean air temperature were 1,310 mm and 21.80°C , respectively (Figure 2). According to the Köppen-Geiger classification, the climate is Aw, a tropical savanna, characterized by dry winters and rainy summers. The soil was classified as an Oxisol, with the following physicochemical characteristics: pH (H_2O) 6.0, 21.7 g kg^{-1} organic matter, 0.9 mg dm^{-3} P (Mehlich-1), $0.1 \text{ cmol}_c \text{ dm}^{-3}$ Al^{3+} , $2.9 \text{ cmol}_c \text{ dm}^{-3}$ $\text{Ca}^{2+} + \text{Mg}^{2+}$, 39 mg dm^{-3} K^+ , 258 g kg^{-1} fine sand, 76.7 g kg^{-1} coarse sand, 101.8 g kg^{-1} silt, and 563.5 g kg^{-1} clay.

Previously to the experiment, soybean and corn were grown in rotation from 1999 to 2004. In each crop season, corn and soybean were sown in November and harvested in March. From the 2004/2005 to 2012/2013

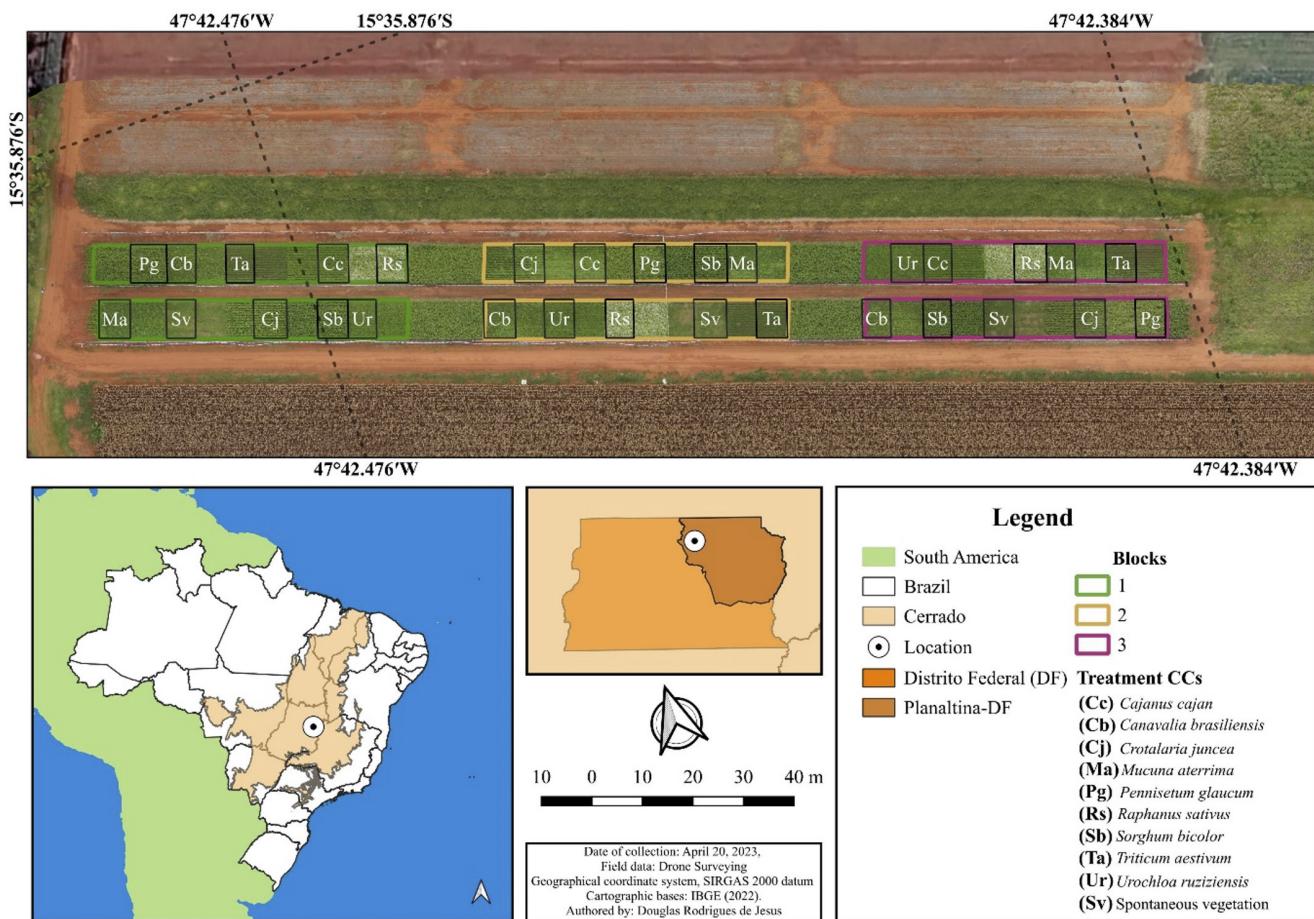


Figure 1. Sketch of the plots and location of the experimental area at Embrapa Cerrados, in the municipality of Planaltina, Distrito Federal (DF), Brazil. CCs, cover crops; Cc, pigeon pea (*Cajanus cajan*); Cb, Brazilian jackbean (*Canavalia brasiliensis*); Cj, sunn hemp (*Crotalaria juncea*); Ma, black velvet bean (*Mucuna aterrima*); Pg, pearl millet (*Pennisetum glaucum*); Rs, radish (*Raphanus sativus*); Sb, sorghum (*Sorghum bicolor*); Ta, wheat (*Triticum aestivum*); Ur, ruzigrass (*Urochloa ruziziensis*); and Sv, spontaneous vegetation.

crop season, corn and cover crops were grown in succession under a no-tillage system. The cover crops were sown in March and cut at flowering (between May and August), depending on the plant species. Off-season occurred from May to November, depending on the cover crop cultivated.

Between the 2013/2014 and 2020/2021 crop seasons, the 30F53VYHR corn hybrid was sown in November under a no-tillage system (directly on top of the cover crop residues), using five seeds per meter in rows spaced 0.75 m apart, resulting in a population of 65,000 plants per hectare. At sowing, all plots and subplots received 500 kg ha⁻¹ N-P-K (4-30-16), 2.0 kg ha⁻¹ Zn (ZnSO₄ 7H₂O), and 10 kg ha⁻¹ of the FTE BR 12 fertilizer (Nutriplant, Barueri, SP, Brazil) used as a micronutrient source (3.2% S, 1.8% B, 0.8% Cu, 2.0% Mn, 0.1% Mo, and 9.0% Zn). Nitrogen was sidedressed as urea at a rate of 130 kg ha⁻¹ N, split into two applications in the subplots at the V4 and V8 growth stages. Harvest was carried out in March.

Between the 2021/2022 and 2024/2025 crop seasons, soybean replaced corn as the main crop, being sown at

the beginning of the rainy season in November. At the end of the rainy season in March, the cover crops were sown after soybean harvest, at a row spacing of 0.5 m, resulting in a population of 220,000 plants per hectare. The plant density of each cover crop was as follows: 20 plants per meter for pigeon pea, sunn hump, sorghum, wheat, and ruzigrass [*Urochloa ruziziensis* (R.Germ. & C.M.Evrard) Crins]; 40 plants per meter for pearl millet and radish; and 10 plants per meter for black velvet bean and Brazilian jackbean. The cover crops were cut at flowering between May and August, depending on the plant species.

Soil samples were collected in 2013, 2018, and 2024 in order to determine C contents and stocks at seven different depths: 0–5, 5–10, 10–20, 20–40, 40–60, 60–80, and 80–100 cm. For each plot, samples were obtained from the 0 to 20 cm layers by homogenizing eight subsamples and from the deeper layers, consisting of five subsamples each. Soil sampling for C stocks was conducted for the treatments and the native Cerrado vegetation in 2013, but only for the treatments in 2018 and 2024.

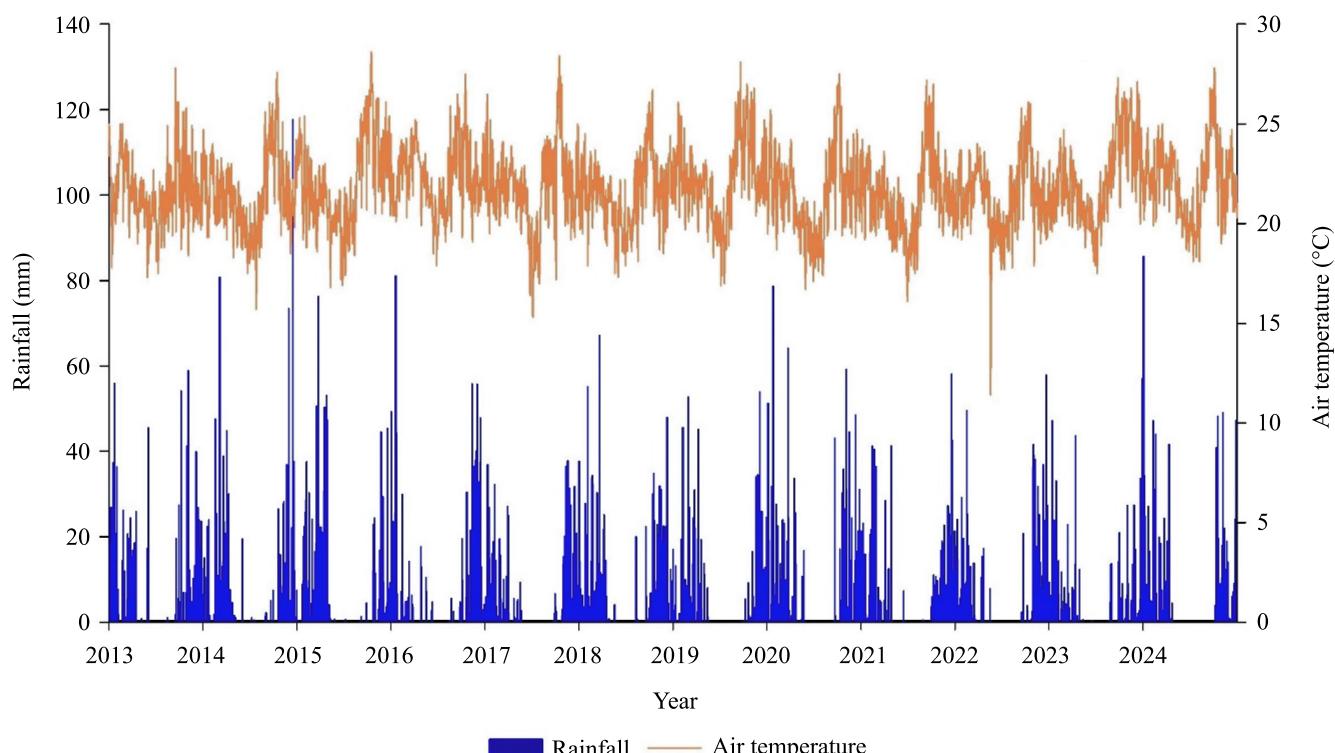


Figure 2. Means for rainfall and air temperature from 2013 to 2024, in the municipality of Planaltina, District Federal, Brazil.

In order to determine bulk density, undisturbed soil samples were collected from the walls of a 120 cm deep trench in a native Cerrado area located 50 m from the experimental site, specifically from the 0–10, 10–20, 20–30, 30–40, 40–60, 60–80, and 80–100 cm soil layers. The samples were collected in duplicate, at each depth, from opposite sides of the trench, using stainless-steel rings. Then, the samples were oven-dried at 105°C, and soil bulk density was calculated based on the internal volume of the cylinders.

To measure total organic C, the soil samples were air-dried, sieved (< 2.00 mm), ground using a pestle and mortar, and sieved again (< 150 µm). Total C was analyzed by dry combustion in the 2400 Series II CHNS elemental analyzer (PerkinElmer, Waltham, MA, USA) through high-temperature oxidation at 1,000°C. To calculate C stock in Mg ha⁻¹, C concentrations were converted into C stock for each sampled layer, according to Ferreira et al. (2016), as follows:

$$\text{Soil C stock} = \frac{\text{TC} \times \text{BD} \times d}{10}$$

where TC is soil total organic C (g kg⁻¹), BD is bulk density (g cm⁻³), and d is the soil layer (cm).

Total soil organic C data were analyzed using linear mixed models to assess the fixed effects of treatments (cover crops and spontaneous vegetation), years (2013, 2018, and 2024), and of their interaction on the response variables. The model structure included treatments and years as fixed factors, while blocks were considered random effects, ensuring robustness in parameter estimation even in cases of unbalanced data.

Carbon stock and content data were obtained through the application of linear mixed models, which allow considering fixed effects (treatments, years, and their interaction) and random effects (blocks or repetitions), providing a greater flexibility to deal with data from experiments with a hierarchical structure or repeated measures.

Adjusted means (marginal means or estimated marginal means) for treatments and years were estimated using the method of Kenward & Roger (1997) to calculate degrees of freedom, allowing a greater precision in inferences, especially in small samples. Multiple comparisons between treatments and years were performed with p-value adjustment

using Tukey's method (Tukey, 1953), maintaining the control of the family-wise error rate at $\alpha = 0.05$.

Interactions between treatments and years were also evaluated, enabling the identification of potential variations in treatment performance over time. Means were grouped based on statistical similarity, using lettered clusters, according to the 5% significance level adjusted using the method of Šidák (1967).

For the variations in soil C stocks (ΔC), statistical analyses were performed to evaluate differences in values among treatments within each time period (year). Initially, descriptive statistics were calculated, including means, standard deviations, and coefficients of variation for each treatment group.

The adequacy of the models was verified by a graphical inspection of the residuals (residuals vs. fitted values and quantile-quantile plots) and by formal tests to assess the assumptions of normality of the residuals, homogeneity of variances (homoscedasticity), linearity, and independence of errors. Normality of residuals was evaluated using Shapiro-Wilk's test and the visual inspection of quantile-quantile plots, whereas homoscedasticity was assessed using Breusch-Pagan's test and the graphical analysis of residuals vs. fitted values. Linearity and the presence of influential observations were examined through standard diagnostic plots.

When model assumptions were not met, the non-parametric Kruskal-Wallis' test ($\alpha = 0.05$) was applied to compare treatment distributions, as for the ΔC variable. When significant differences were detected, post-hoc multiple comparisons using Dunn's test or Kruskal-Wallis' multiple comparison procedure (from the *agricultural* package) were conducted to identify distinct groups. All analyses were performed in the R environment (R Core Team, 2025), version 4.3.3, using the following packages: *lme4* (Bates et al., 2015), for fitting linear mixed models; *emmeans* (Lenth et al., 2025), for calculating adjusted means and multiple comparisons; and *multcomp* (Hothorn et al., 2008), for simultaneous testing and grouping of means.

Results and Discussion

Soil C stocks down to a depth of 100 cm ranged from 110 to 119 Mg ha⁻¹ in 2013, from 131 to 142 Mg ha⁻¹ in 2018, and from 112 to 134 Mg ha⁻¹ in 2024 across the crop rotation systems (soybean and corn), with cover crop and spontaneous vegetation in succession

(Table 1). Overall, the greatest changes in C stocks were driven by switching the main crop rather than by using cover crops. Replacing corn with soybean reduced soil C stocks ($p < 0.05$) in most of the used cover crop systems (Table 1).

Soil C stocks were not affected by the cover crops and spontaneous vegetation in 2013 and 2018 during the corn phase ($p > 0.05$). However, in 2024, during the soybean phase, sorghum promoted higher soil C stocks than pigeon pea, black velvet bean, ruzigrass, and spontaneous vegetation ($p < 0.05$). This higher C stock value can be attributed to the input of residues with a high lignin:N ratio, which leads to a slower decomposition (Carvalho et al., 2022a). Slowly decomposing residues, in addition to providing soil cover, can enhance long-term soil organic C storage (Adhikari et al., 2024). In 2024, only sorghum and wheat showed higher C stocks than spontaneous vegetation ($p < 0.05$), which can be partly explained by the absence of mowing in these plots, contributing to the incorporation of biomass and C. Carvalho et al. (2013) confirmed this hypothesis when studying the same species, concluding that the aboveground biomass in plots with spontaneous vegetation was statistically equivalent to the biomass of plots with other cover crops.

Tropical grasses, such as sorghum, contribute to subsoil C accumulation due to their deeper root systems (Silva et al., 2017). However, this is not the case for ruzigrass even though it is a grass species with a deep rooting system because of the high N content in its tissues and its low lignin:N ratio. These factors

favor the formation of labile pools of soil organic matter, which are more readily decomposable, as well as a low production of dry matter, resulting in a lower long-term accumulation of soil organic C (Carvalho et al., 2022b).

Soil C stocks also underwent changes over the years, with a significant increase from 14 to 21% observed from 2013 to 2018 for all cover crops, including spontaneous vegetation ($p < 0.05$). From 2018 to 2024, there was a depletion in soil C stocks, except in plots with sorghum and wheat, which maintained their soil C stock levels. According to Yang et al. (2018), the depletion of soil C stocks after the introduction of a legume (in this case, soybean) as the main crop can be explained by the lower residue input and lower C:N ratio compared with those of the previous main crop (corn). In this line, Dou et al. (2018) found that continuous corn cropping would be better for a stable C accumulation than the corn-soybean rotation. Similarly, Luo et al. (2024) concluded that reducing the frequency of soybean cultivation within the corn-soybean rotation system is essential to mitigate the depletion of soil C stocks, as the decrease in C input into the system promotes the consumption of C and N from soil organic matter by microorganisms for their growth and activity. However, after nine years of data collection on crop residues, soil C stocks, and soil organic matter mineralization, Sattolo et al. (2022) were still not sure about the contribution of legumes and grasses in rotation in increasing soil C stocks, which suggests a new steady-state level in a clayey

Table 1. Mean (standard deviation) of soil carbon stocks at the 0–100 cm depth of soil cultivated with soybean-corn (*Glycine max-Zea mays*) in rotation, with ten different cover crops in succession, in 2013, 2018, and 2024.

Cover crop	Soil carbon stock ⁽¹⁾ (Mg ha ⁻¹)		
	2013	2018	2024
Pigeon pea (<i>Cajanus cajan</i>)	114 (± 6.3) aB	133 (± 2.8) aA	115 (± 15.2) cdB
Brazilian jackbean (<i>Canavalia brasiliensis</i>)	119 (± 4.8) aB	142 (± 1.5) aA	128 (± 9.6) abcB
Sunn hemp (<i>Crotalaria juncea</i>)	117 (± 1.2) aB	136 (± 4.1) aA	126 (± 3.2) abcB
Black velvet bean (<i>Mucuna aterrima</i>)	111 (± 3.2) aB	133 (± 2.0) aA	112 (± 7.2) dB
Pearl millet (<i>Pennisetum glaucum</i>)	116 (± 5.5) aC	140 (± 3.3) aA	127 (± 1.6) abcB
Radish (<i>Raphanus sativus</i>)	110 (± 5.7) aC	133 (± 3.1) aA	122 (± 4.6) abcdB
Sorghum (<i>Sorghum bicolor</i>)	112 (± 4.5) aB	136 (± 4.1) aA	134 (± 2.6) aA
Wheat (<i>Triticum aestivum</i>)	115 (± 4.1) aB	131 (± 6.6) aA	130 (± 6.7) abA
Ruzigrass (<i>Urochloa ruziziensis</i>)	119 (± 6.1) aB	141 (± 2.6) aA	121 (± 2.3) bcdB
Spontaneous vegetation	111 (± 1.0) aB	132 (± 3.1) aA	116 (± 8.4) cdB

⁽¹⁾Means followed by equal letters, lowercase in the columns (cover crop) and uppercase in the lines (year), do not differ by Tukey's test ($\alpha = 0.05$).

Oxisol under a soybean-corn production system in the Cerrado.

The treatments with sorghum and wheat maintained the levels of soil C stock from 2018 to 2024. According to Ngidi et al. (2024), both of these cover crops have the potential to enhance soil C sequestration through C inputs from aboveground biomass and root systems.

Regarding total soil C contents at different depths and in different years (Table 2), no significant differences were observed between cover crops at any depth in 2013. In 2018, only at the 0–5 cm depth,

plots with ruzigrass in succession showed a higher C content than those with sunn hemp and spontaneous vegetation ($p < 0.05$). According to the literature, the intercropping of ruzigrass with corn influences above- and belowground biomass, enhancing soil organic C levels (Colombi & Keller, 2019; Bassegio et al., 2025). For this reason and due to its abundant and deep root system, ruzigrass has been incorporated into intercropping systems (Rosolem et al., 2019). Root growth belowground and the addition of organic residues are active sources of organic exudates, which serve as

Table 2. Means of soil carbon content at seven different layers of soil cultivated with soybean-corn (*Glycine max-Zea mays*) in rotation, with ten different cover crops in succession, in 2013, 2018, and 2024⁽¹⁾.

Year	Cover crop ⁽²⁾	Soil carbon content (g kg ⁻¹) at different depths (cm)						
		0–5	5–10	10–20	20–40	40–60	60–80	80–100
2013	Cc	24aA	20aA	17aB	15aA	11aB	9aB	9aB
	Cb	25aA	20aA	18aB	15aB	11aB	9aB	9aB
	Cj	24aA	19aA	18aB	15aB	11B	9aC	9aB
	Ma	24aA	18aB	17aA	14aA	11aB	9aB	8aB
	Pg	24aB	19aB	19aB	15aA	11aC	9aB	8aB
	Rs	22aB	18aB	17aB	14aB	11aB	9aAB	9aB
	Sb	23aB	19aA	17aB	14aB	11aB	9aB	8aC
	Ta	23aB	19aA	18aA	14aA	11aB	9aB	9aB
	Ur	26aB	21aAB	19aB	16aB	11aB	9aB	9aB
	Sv	23aA	19aAB	17aA	14aA	11aB	9aB	8aB
2018	Cc	28abA	21aA	20aA	15aA	14aA	12aA	10aA
	Cb	28abA	23aA	21aA	18aA	14aA	12aA	10aA
	Cj	26bA	22aA	20aA	17aA	13aA	12aA	11aA
	Ma	27abA	22aA	19aA	16aA	13aA	11aA	10aA
	Pg	28abA	24aA	21aA	17aA	14aA	12aA	10aA
	Rs	29abA	22aA	20aA	16aA	14aA	11aA	10aA
	Sb	29abA	22aA	20aA	17aA	14aA	11aA	10aB
	Ta	27abA	21aA	20aA	16aA	13aA	11aA	9aAB
	Ur	31aA	22aA	22aA	18aA	14aA	11aA	10aA
	Sv	25bA	21aA	19aA	16aA	13aA	12aA	10aB
2024	Cc	28aA	18abA	17abB	15aA	12aB	11abA	9bcAB
	Cb	27abA	16bB	15bcC	15aB	13aA	11abA	10abA
	Cj	23abA	19abA	17abB	14aB	12aB	11bcB	9bcB
	Ma	25abA	21aAB	18abA	16aA	12aA	11abA	10bcA
	Pg	26abAB	21aAB	19aAB	15aA	12aB	11abA	10abcA
	Rs	23abB	19abAB	18abAB	16aA	12aAB	9cB	8cB
	Sb	24abB	20abA	18aAB	16aAB	13aA	12aA	11aA
	Ta	23abB	19aba	19aA	16aA	13aA	11abA	10abA
	Ur	25abB	17abB	18abB	15aB	11aB	10bcAB	10abcA
	Sv	22bA	17abB	13cB	15aA	12aA	10bcB	8bcAB

⁽¹⁾Means followed by equal letters, lowercase in the columns (cover crop) and uppercase in the lines (year), do not differ by Tukey's test ($\alpha = 0.05$).

⁽²⁾Cc, pigeon pea (*Cajanus cajan*); Cb, Brazilian jackbean (*Canavalia brasiliensis*); Cj, sunn hemp (*Crotalaria juncea*); Ma, black velvet bean (*Mucuna aterriformis*); Pg, pearl millet (*Pennisetum glaucum*); Rs, radish (*Raphanus sativus*); Sb, sorghum (*Sorghum bicolor*); Ta, wheat (*Triticum aestivum*); Ur, ruzigrass (*Urochloa ruziziensis*); and Sv, spontaneous vegetation.

effective stabilizing agents in soil aggregation (Silva et al., 2016; Shen et al., 2020; Ma et al., 2022). Colombi & Keller (2019) highlighted that roots can influence soil aggregation through various mechanisms, including the direct formation or modification of soil pores and the consequent enhancement of soil organic C.

In 2024, at the 0–5 cm depth, plots with soybean and pigeon pea exhibited a higher C content than those with soybean and spontaneous vegetation ($p < 0.05$). At the 5–10 cm depth, plots with soybean and pearl millet, soybean and sorghum, and soybean and wheat showed a higher C content than plots with soybean and Brazilian jackbean and soybean and spontaneous vegetation ($p < 0.05$). At the 10–20 cm layer, plots with soybean and pearl millet in succession also presented higher C levels than those with soybean and Brazilian jackbean and with soybean and spontaneous vegetation ($p < 0.05$). The net C balance in the system with soybean and wheat was 4.33 ± 0.6 Mg ha⁻¹ C over one year, indicating a contribution to the maintenance of soil C storage (Veeck et al., 2022). At the 60–80 cm soil layer, plots with soybean and sorghum showed a higher C content than those with soybean and sunn hemp, soybean and radish, soybean and ruzigrass, and soybean and spontaneous vegetation ($p < 0.05$). At the 80–100 cm depth, the plots with soybean and sorghum presented a higher C content than those with soybean and pigeon pea, soybean and sunn hemp, soybean and radish, and soybean and spontaneous vegetation ($p < 0.05$).

Plots with soybean and sorghum were more efficient in increasing soil C stocks and contents from soil surface to deeper layers. This is explained by the nutrient cycling time of sorghum of 172–222 days (Carvalho et al., 2015) plus its high C:N ratio of 66:1, which slow down the decomposition of organic matter, allowing for a longer residence time in the soil and the gradual release of C and nutrients (Silva et al., 2017). In 2024, the lower C content observed in plots with spontaneous vegetation underscored the importance of including cover crops in low-C agricultural systems. In this sense, Veeck et al. (2022) found that, even during short intervals between cropping seasons, up to 27% of stored C can be lost during periods of spontaneous vegetation.

The ΔC along different periods took into account the use of various cover crop species and spontaneous vegetation from 2013 to 2024 (Table 3). During the first evaluation period from 2013–2018, all species led to positive increases in soil C stocks and treatments did not differ significantly from each other ($p > 0.05$). These results show the importance of cover crops during the second cropping season to promote C accumulation in the early years. This effect was also attributed to the corn crop, which contributed with a higher amount of biomass input compared with that of soybean (Mazzilli et al., 2014).

The positive C accumulation trend did not persist during the second evaluated period from 2018–2024, when most treatments presented losses in soil C stocks. Despite this, when considering the entire period

Table 3. Means of gains and losses in soil carbon stock in three periods with soil cultivated with corn-soybean (*Zea mays-Glycine max*) in rotation, with ten different cover crops in succession⁽¹⁾.

Cover crop	Soil carbon stock in three periods (Mg ha ⁻¹)		
	2013–2018	2018–2024	2013–2024
Pigeon pea (<i>Cajanus cajan</i>)	19.0a	-9.0abcd	10.0abc
Brazilian jackbean (<i>Canavalia brasiliensis</i>)	23.0a	-21.0cd	2.0bc
Sunn hemp (<i>Crotalaria juncea</i>)	18.0a	-9.0abc	9.0abc
Black velvet bean (<i>Mucuna aterrima</i>)	22.0a	-21.0cd	1.0c
Pearl millet (<i>Pennisetum glaucum</i>)	24.0a	-13.0abcd	11.0abc
Radish (<i>Raphanus sativus</i>)	23.0a	-11.0abcd	12.0abc
Sorghum (<i>Sorghum bicolor</i>)	23.0a	-2.0ab	21.0a
Wheat (<i>Triticum aestivum</i>)	17.0a	-0.5a	16.0ab
Ruzigrass (<i>Urochloa ruziziensis</i>)	22.0a	-20.0d	2.0c
Spontaneous vegetation	21.0a	-16.0bcd	6.0bc

⁽¹⁾Means followed by equal letters, lowercase in the columns (cover crop), do not differ by Kruskal-Wallis' test ($p < 0.05$).

from 2013 to 2024, the overall balance was positive, and the treatments exhibited the same behavior as in 2018–2024, i.e., sorghum and wheat showed lower ΔC losses than Brazilian jackbean, black velvet bean, ruzigrass, and spontaneous vegetation ($p < 0.05$).

The evaluated crop succession system, involving wheat in the winter and soybean in the summer in a subtropical region of Brazil, was efficient in C mitigation as it increased soil C stocks, with a gain of 34.7 Mg ha⁻¹ C from the atmosphere (Veeck et al., 2022). Among the studied cover crops, sorghum presented the ability to promote more labile organic fractions on soil surface, contributing to an increase in soil C stocks over time (Ferreira et al., 2020). Furthermore, corn residues may enhance the turnover of soil organic C compared with those of soybean due to their higher C:N ratio (Mazzilli et al., 2014).

Conclusions

1. The evaluated corn-soybean (*Zea mays*-*Glycine max*) rotation under no-tillage and with cover crops in succession can either increase or decrease soil carbon stocks and soil C sequestration, depending on the combination of main crop and cover crops.

2. Systems including sorghum (*Sorghum bicolor*) and wheat (*Triticum aestivum*) in succession as a cover crop exhibit a greater potential for soil C sequestration.

3. Sorghum presents higher soil C contents on soil surface and subsurface layers, which contributes to greater accumulations and lower losses in C stocks after the transition from corn to soybean cultivation.

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Declaration of use of AI technologies

No generative artificial intelligence (AI) was used in this study.

Conflict of interest statement

The authors declare no conflicts of interest.

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