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Soil fertility in silvopastoral systems integrating tree legumes with signalgrass (*Urochloa decumbens* Stapf. R. Webster)

Ana María Herrera[1] Alexandre Carneiro Leão de Mello[2]
Valéria Xavier de Oliveira Apolinário[3] José Carlos B. Dubeux Jr[4]
Robert Emilio Mora-Luna[5] Erinaldo Viana de Freitas[6]

Agricultural Research Department, Universidad Nacional Experimental del Táchira. Táchira, Venezuela.

Abstract. Silvopastoral Systems (SPS) can increase overall productivity and income in order to stimulate the simultaneous growth and development of trees, forage and livestock. Moreover, the SPS with tree legumes would be important for adding nutrients to the system, mainly N, ensuring soil health and quality. Soil properties were assessed in two SPS, implanted in 2011, using tree legumes (in double rows) intercropped with *Urochloa decumbens* Stapf. R. Webster (Signalgrass). Treatments were Signalgrass + *Mimosa caesalpiniifolia* Benth (Sabiá) and Signalgrass + *Gliricidia sepium* (Jacq.) Kunth ex Walp. (Gliricidia), and they were allocated in a randomized complete block design, with three replications. Soil was sampled in 2013, 2017, and 2018, at 0, 4, and 8 m along transects perpendicular to tree double rows, from 0- to 20- and 20- to 40-cm layers. Soil chemical properties evaluated included pH, P, K^+, Ca^{2+}, Mg^2+, Al^{3+}, $H^+ + Al^{3+}$, cation exchange capacity (CEC), and base saturation. In addition, light fraction of soil organic matter (LF-SOM), soil basal respiration (SBR), and natural abundance of ^{13}C of the respired CO_2 ($\delta^{13}C$-CO_2) were analyzed. Soil pH (5.3, 5.2, 5.1), P (11.3, 7.2, 3.6 mg dm^{-3}), and $CEC_{effective}$ (5.8, 5.1, 5.0 $cmol_c$ dm^{-3}) decreased ($P < 0.05$) along the years 2013, 2017, and 2018, respectively. In 2018, the LF-SOM and $\delta^{13}C$-CO_2 were greater in Sabiá (1.1 g kg^{-1} and -16.4 ‰) compared to Gliricidia (0.7 g kg^{-1} and -18.2 ‰). Silvopastoral systems reduced soil fertility regardless of the tree legume species used as a result of biomass nutrient stock, without maintenance fertilization. Sabiá had greater deposition of LF-SOM, without increasing SBR, providing potential for microbial C use efficiency. Enriched C-CO_2 isotope composition had an efficient SOM oxidization in SPS with Gliricidia or Sabiá. This information can contribute to the assessments related to CO_2 balance and C retention. Both SPS contribute to C sequestration.

Keywords: agroforestry systems; *Gliricidia sepium*; *Mimosa caesalpiniifolia*; soil chemical properties; C sequestration.

Fertilidade do solo em sistemas silvipastoris integrando leguminosas arbóreas com capim-braquiária (*Urochloa decumbens* Stapf. R. Webster)

Resumo. Os Sistemas Silvipastoris (SSP) podem aumentar a produtividade e gerar renda. Também, os SSP com leguminosas arbóreas adicionam nutrientes ao sistema, principalmente N, garantindo a saúde e a qualidade do solo. As propriedades do solo foram avaliadas em dois SSP utilizando leguminosas arbóreas em consorcio com *Urochloa decumbens* Stapf. R. Webster (capim-braquiaria). Os tratamentos foram capim-braquiária + *Mimosa caesalpiniifolia* Benth (Sabiá) e capim-braquiária + *Gliricidia sepium* (Jacq.) Kunth ex Walp. (Gliricídia), sendo distribuídos em delineamento casualizado em blocos (três repetições). As coletas de solo foram realizadas nos anos 2013, 2017 e 2018, a 0, 4 e 8 m ao longo de transectos perpendiculares às fileiras duplas de árvores, nas profundidades de 0-20 e 20-40 cm. As propriedades químicas do solo avaliadas incluíram pH, P, K^+, Ca^{2+}, Mg^{2+}, Al^{3+}, $H^+ + Al^{3+}$, capacidade de troca de cátions (CTC) e saturação por bases. Foram analisadas a fração leve da matéria orgânica (FL-MOS), a respiração basal (RBS) e a abundância natural do ^{13}C do CO_2 respirado ($\delta^{13}C$-CO_2). O pH (5,3; 5,2; 5,1), P (11,3; 7,2; 3,6 mg dm^{-3}) e $CTC_{efetiva}$ (5,8; 5,1; 5,0 $cmol_c$ dm^{-3}) diminuíram ($P < 0,05$) ao longo dos anos 2013, 2017 e 2018, respectivamente. Em 2018, a FL-MOS e $\delta^{13}C$-CO_2 foi maior em Sabiá (1,1 g kg^{-1} e -16,4 ‰) em comparação com Gliricídia (0,7 g kg^{-1} e -18,2 ‰). Os SSP reduziram a fertilidade do solo independentemente das espécies arbóreas utilizadas em decorrência do estoque de nutrientes da biomassa, sem adubação de manutenção. Sabiá teve maior deposição de FL-MOS, sem aumentar a RBS, proporcionando potencial para a eficiência do uso do

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[1] Corresponding author. E-mail address: aherrera@unet.edu.ve

[2] Animal Science Department, Universidade Federal Rural de Pernambuco. CNPq Fellow. Pernambuco, Brazil.

[3] Associated Professor I, Universidade Estadual do Maranhão. Maranhão, Brazil.

[4] North Florida Research & Education Center, University of Florida. Florida, United States.

[5] Animal Science Department. Facultad de Agronomía e Ingeniería Forestal. Pontificia Universidad Católica de Chile. Santiago de Chile. Chile.

[6] Instituto Agronômico de Pernambuco. Pernambuco, Brazil.

C microbiano. A composição enriquecida de isótopos de C-CO_2 mostraram uma eficiente oxidação da MOS em SSP com Gliricídia ou Sabiá. Essas informações podem contribuir para as avaliações relacionadas ao balanço de CO_2 e retenção de C. Ambos SSP contribuem para o sequestro de C.

Palavras-chave: sistemas agroflorestais; *Gliricidia sepium*, *Mimosa caesalpiniifolia*; propriedades químicas do solo; sequestro de C.

Fertilidad del suelo en sistemas silvopastoriles que integran leguminosas arbóreas con pasto señal (*Urochloa decumbens* Stapf. R. Webster)

Resumen. Los Sistemas Silvopastoriles (SSP) pueden aumentar la productividad general y generar ingresos. Además, los SSP con leguminosas arbóreas adicionan nutrientes al sistema, principalmente N, asegurando la salud y la calidad del suelo. Propiedades del suelo fueron evaluadas en dos SSP utilizando leguminosas arbóreas en asociación con *Urochloa decumbens* Stapf. R. Webster (Barrera). Los tratamientos fueron Barrera + *Mimosa caesalpiniifolia* Benth (Sabiá) y Barrera + *Gliricidia sepium* (Jacq.) Kunth ex Walp. (Gliricidia), distribuidos en un diseño de bloques aleatorizados (tres repeticiones). Se realizaron colectas de suelo en los años 2013, 2017 y 2018, a 0, 4 y 8 m en transectos perpendiculares a las hileras de árboles, en las profundidades de 0-20 y 20-40 cm. Las propiedades químicas del suelo evaluadas incluyeron pH, P, K^+, Ca^{2+}, Mg^{2+}, Al^{3+}, H^++Al^{3+}, capacidad de intercambio catiónico (CIC) y saturación de bases. Se analizaron la fracción activa de la materia orgánica (FA-MOS), respiración basal (RBS) y abundancia natural de ^{13}C del CO_2 respirado ($\delta^{13}C$-CO_2). El pH (5.3, 5.2, 5.1), P (11.3, 7.2, 3.6 mg dm^{-3}) y la $CIC_{efectiva}$ (5.8, 5.1, 5.0 $cmol_c$ dm^{-3}) disminuyeron ($P < 0.05$) a través de los años 2013, 2017 y 2018, respectivamente. En 2018, la FA-MOS y $\delta^{13}C$-CO_2 fueron mayores en Sabiá (1,1 g kg^{-1} y -16,4 ‰) comparada con Gliricidia (0,7 g kg^{-1} y -18,2 ‰). Los SSP redujeron la fertilidad del suelo independientemente de las especies arbóreas utilizadas como resultado de la reserva de nutrientes de la biomasa, sin fertilización de mantenimiento. Sabiá tuvo mayor deposición de FA-MOS, sin aumentar RBS, favoreciendo potencialmente la eficiencia del uso de C microbiano. La composición isotópica de C-CO_2 enriquecida muestra una oxidación eficiente de la MOS en SSP con Gliricidia o Sabiá. Esta información puede contribuir a las evaluaciones relacionadas con el balance de CO_2 y retención de C. Ambos SSP contribuyen al secuestro de C.

Palabras clave: sistemas agroforestales; *Gliricidia sepium*; *Mimosa caesalpiniifolia*; propiedades químicas del suelo; secuestro de C.

Introduction

Silvopastoral systems (SPS), as an agroforestry practice, can increase overall productivity and generate income in order to stimulate the simultaneous growth and development of trees, forage and livestock (Sarabia *et al.*, 2019; Watanabe *et al.*, 2016). Moreover, SPS with tree legumes enhance the delivery of ecosystem services in livestock system (Dubeux Jr. *et al.*, 2019), including biological nitrogen fixation, shade and fodder supply for ruminants, as well as production of marketable timber, increase in producer income, and stability of the soil-plant system (Apolinário *et al.*, 2015; Dubeux Jr. *et al.*, 2019). These combinations of SPS would be important for stabilizing fertilization and ensuring soil health and quality.

This stability of the soil-plant system is possible from beneficial interactions between biological components (soil, water, air, plants, and animals) (Sheoran *et al.*, 2017). Tree legumes, for instance, can increase N input and nutrient availability in the system, via N-fixation, litter deposition and decomposition (Sarabia *et al.*, 2019). However, tree characteristics, such as canopy structure, leaf deposition, and litter quality, may affect soil chemical and biological properties (Alfaro *et al.*, 2018).

The soil is considered a complex and fundamental system for the dynamic of terrestrial ecosystems (Terra *et al.*, 2019); however, some livestock traditional activities may contribute to the depletion of soil productive capacity, leading vast grassland areas to degradation (Santana *et al.*, 2016). Soil chemical and physical properties affect water and nutrient availability for plants and soil microorganisms. The most common soil properties used to assess nutrient availability are pH, mineral content, cation exchange capacity (CEC), soil organic matter (SOM), carbon and mineralizable nitrogen content (Bünemann *et al.*, 2018). Regarding biological properties, microbial processes have fundamental importance for the productivity and sustainability of systems (Santos *et al.*, 2014), being highly related to SOM decomposition derived from plant and animal waste, as well as its recycling. Basal soil respiration, microbial biomass, and C:N ratio are the most valuable variables in the interpretation of SOM dynamics (Alfaro *et al*., 2018; Santos *et al*., 2014), as those are considered indicators of soil organic matter cycling.

Gliricidia sepium (Jacq.) Kunth ex Walp. (Gliricidia) and *Mimosa caesalpiniifolia* Benth. (Sabiá) are tree legumes, introduced and native, respectively, with potential for use in SPS. These species are drought-tolerant and nitrogen fixers (Apolinário *et al*., 2015). Recently, SPS integrating those legumes with the grass *Urochloa decumbens* Stapf. R. Webster (signalgrass) have been evaluated in Northeastern Brazil, in terms of biomass production, chemical composition, litter quality, deposition and decomposition, as well as animal performance (Apolinário *et al*., 2016, 2015; Oliveira *et al*., 2018; Santos *et al*., 2020). However, there are few assessments of changes in soil chemical and biological properties in SPS, including those tree legumes. The hypothesis of this study was that the inclusion of those tree legumes in SPS would improve soil fertility differently with time. The aim of the experiment was to evaluate the changes along a time sequence in soil chemical and biological properties in two SPS, integrating tree legumes, Gliricidia or Sabiá, with signalgrass.

Material and Methods

Location, procedures and experimental design

The research was conducted at the Experimental Station of the Agronomic Institute of Pernambuco (IPA), located in Itambé, Pernambuco, Brazil. The station is located at 7°23′ S and 35°10′ W. Average annual rainfall is 1253 mm (APAC, 2022), and the average annual temperature is 25°C (RIMA, 2014), with Köppen-Geiger climate classified as hot and humid tropical (As). The predominant soil is dystrophic Ultisol (Silva *et al*., 2001).

The treatments were defined by two SPS: Gliricidia + signalgrass (Gliricidia) and Sabiá + signalgrass (Sabiá). The experimental area consisted of six paddocks of 1.0 ha each. Tree legumes were established in double rows spaced by 15.0 m (between double rows) x 1.0 m (between rows) x 0.5 m (within rows) (Fig. 1). Establishment of signalgrass occurred in open pits (approx. 5 cm deep) and seeds were placed manually (10 kg of commercial seed ha^{-1} with 40 % cultural value). Signalgrass was previously established in one of the blocks since 1969. In the other two blocks were established along with the tree legumes, between the double rows. Legume seeds were planted in a greenhouse and inoculated with specific *Bradyrhizobium* strains obtained from the Soil Microbiology Laboratory at Universidade Federal Rural de Pernambuco (UFRPE). All paddocks were fertilized in July 2011 with 44 kg ha^{-1} of P (as single superphosphate) and 100 kg ha^{-1} of K (as potassium chloride) on the entire area. Legume seedlings were transplanted to the field in June 2011 at approximately 30-cm height and planted in 20-cm deep furrows. Paddocks were fully established in October 2011.

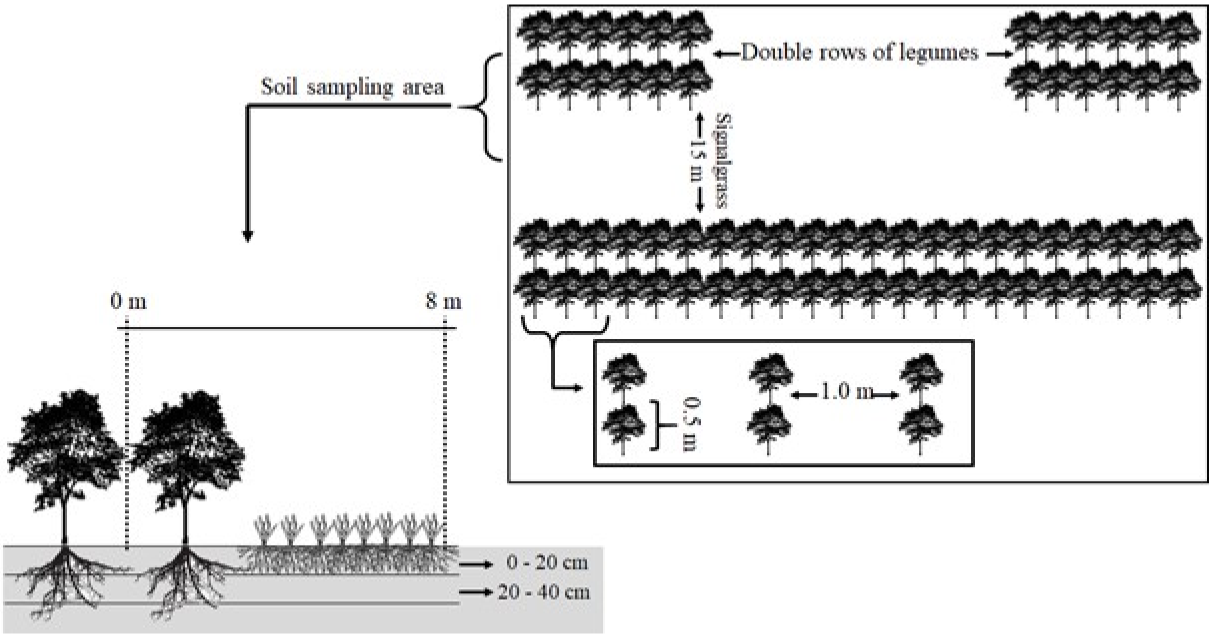

Fig. 1. Tree spacing in the double row silvopastoral system and soil sampling area. Each one-ha plot had 14 double rows.

Initial soil physical characteristics (2011) from 0- to 20-cm depth were loam-clay-sandy texture with proportions of coarse sand (0.2-2 mm), fine sand (0.05-0.2 mm), silt (0.002-0.05 mm), and clay (< 0.002 mm) of 44, 17, 16, and 22 %, respectively, and soil and particle density of 1.33 and 2.52 g cm^{-3}, respectively. Average monthly rainfall for the experimental years is shown as supplementary material.

Each plot had 14 double rows with a tree density of approximately 2500 plants ha^{-1}. Treatments were allocated in a randomized complete block design, with three replications. Yearling crossbred Holstein × Zebu steers, with approximately 200 kg of body weight (BW) were used as experimental animals, under continuous stocking with variable stocking rate (Mott and Lucas, 1952). Stocking rate was adjusted every 28 days following the recommendation of Sollenberger et al. (2005). The target herbage allowance was 3 kg DM d^{-1} of green herbage mass per kilogram of BW. Water and a mineral mixture (Ca: 107 g kg^{-1}, P: 88 g kg^{-1}, S: 12 g kg^{-1}, Na: 126 g kg^{-1}, Co: 55 mg kg^{-1}, Cu: 1530 mg kg^{-1}, Fe: 1800 mg kg^{-1}, I: 75 mg kg^{-1}, Mn: 1300 mg kg^{-1}, Zn: 3630 mg kg^{-1}, F: 880 mg kg^{-1}) were offered *ad libitum* on each paddock.

Soil chemical properties

Soil chemical properties were evaluated in 2013, 2017 and 2018, at 0- to 20-cm depth. Three soil samples (sampling units) were analyzed per paddock (experimental unit), at two distances within each paddock, in between the double rows and at 8 m from them in a perpendicular transect to the tree row (Fig. 1), resulting in a composite sample for each distance per plot. Soil variables included pH, P, K^+, Ca^2+, Mg^2+, Al^3+, and H^++Al^3+, obtained from the Soil Chemical Laboratory at UFRPE. Subsequently, the total exchangeable bases (TEB), effective (TEB + Al^3+) and potential (TEB + H^++Al^3+) CEC and base saturation were estimated following EMBRAPA (2017) procedures.

Indicators of soil organic matter cycling

Indicators of soil organic matter cycling were evaluated for two years (2017 and 2018) at 0- to 20 and 20- to 40-cm depth (Fig. 1). The sampling protocol was similar to the one used for soil chemical properties. Indicators of soil organic matter cycling included the light fraction of SOM (LF-SOM), soil basal respiration (SBR), and natural abundance of ^{13}C of the respired CO_2 ($\delta^{13}C$-CO_2).

The light fraction of SOM was estimated using the SOM fractionation methodology proposed by Meijboom *et al.* (1995) and adapted by Dubeux Jr. *et al.* (2006). Soil basal respiration was determined by CO_2 emission from soil samples incubated for 21 d, with soil humidity at around 60 % of field capacity, without light and temperature between 25- to 28- °C, quantified and estimated by titration (Harris *et al.*, 1997; Silva *et al.*, 2007). After titration, the suspension was processed following the method of Ramnarine *et al.* (2012), and subsequently dried in a forced-air oven (65±2 °C) to constant weight for storage. Samples were sent to the University of Florida, to determine the $\delta^{13}C$-CO_2 using a CHNS analyzer through the Dumas dry combustion method (Vario Micro Cube, Elementar) coupled to an isotope ratio mass spectrometer (IsoPrime 100, IsoPrime). The $\delta^{13}C$-CO_2 estimate was performed according to Unkovich *et al.* (2008) and Fry (2006).

Statistical analysis

Variance analyses were performed using the PROC MIXED procedure of the SAS (SAS University Edition software), using the Tukey test, when the F test was significant ($P < 0.05$). For chemical and biological properties, SPS, distance from the double rows of legumes, soil depth, and year (as appropriate), were considered fixed effects, and blocks were the random effect. The year effect was analyzed using the repeated measurement procedure.

Results and Discussion

Soil chemical properties

Interaction between treatment × distances for H^+ +Al^{3+} ($P = 0.04$) and TEB ($P = 0.02$), year × distances for K+ ($P = 0.05$) and year × treatment × distances for Mg^{2+} ($P = 0.04$) and base saturation ($P = 0.004$) were observed. Soil pH, P, $CEC_{effective}$ and $CEC_{potential}$ (Tables 1 and 2) were different ($P < 0.05$) among years, with no treatment differences ($P > 0.05$) in any of the variables considered in soil chemical properties.

Soil pH significantly reduced from 2013 to 2018 (Table 1), showing increased acidity of soil reflecting the activity of nitrifying bacteria as they release NO_3^- and H^+ during oxidizing the ammonium NH_4+-N (Neina, 2019). Further, this is favored by the presence of nitrogen-fixing species (e.g., Gliricidia and Sabiá) and nitrate concentrations (Mathesius, 2022). Moreover, these species deposited N-rich litter that will further add H^+ during nitrification of the decomposing litter. According to Johan *et al.* (2021) soils with elevated acidity, usually have low base concentration, elevated Al^{3+} and high P fixation. The lower soil pH values observed in 2018 compared to 2013 may also be explained by the extractions of cations by trees and the pasture. This result is probably associated with decreased P, Mg^{2+} K^{2}, TEB and $CEC_{effective}$ over the years ($P < 0.05$).

Table 1. Soil chemical properties between 0- to 20-cm depth in silvopastoral systems intercropped with signalgrass in Itambé, Pernambuco state, Brazil.

	pH	P	Ca^{2+}	Mg^{2+}	K^{+}	Al^{3+}
Factor	(water, 1: 2.5)	mg dm^{-3}		cmolc dm^{-3}		
Year (Y)						
2013	5.3^{A}	11.3^{A}	2.5	2.7^{A}	0.23^{A}	0.23^{A}
2017	5.2^{AB}	7.2^{B}	3.0	1.1^{C}	0.17^{AB}	0.17^{AB}
2018	5.1^{B}	3.6^{C}	2.9	1.5^{B}	0.12^{B}	0.12^{B}
Standard error	0.05	0.07	0.29	0.08	0.06	0.06
P-value	0.0061	<0.0001	0.1864	<0.0001	0.0122	0.0122
Treatment (T)						
Gliricidia + Signalgrass	5.3	6.1	3.1	1.8	0.21	0.21
Sabiá + Signalgrass	5.1	7.3	2.5	1.6	0.14	0.14
Standard error	0.05	0.07	0.33	0.07	0.06	0.06
P-value	0.1239	0.4993	0.3070	0.3514	0.1038	0.1038
Distance (D)						
0 m	5.2	6.5	2.7	1.7	0.19	0.19
8 m	5.2	6.8	2.9	1.8	0.15	0.15
Standard error	0.04	0.06	0.26	0.07	0.05	0.05
P-value	0.2074	0.8010	0.2563	0.4255	0.1857	0.1857
Y × D						
Standard error	0.06	0.08	0.35	0.10	0.09	0,11
P-value	0.0974	0.1084	0.8288	0.3055	0.2246	0,0493
Y × T × D						
Standard error	0.09	0.11	0.50	0.15	0.13	0.11
P-value	0.4229	0.6171	0.4893	0.0017	0.2464	0.3893

P: phosphorus (Mehlich-I); Ca^{2+}: calcium; Mg^{2+}: magnesium; K^{+}: potassium; Al^{3+}: exchangeable aluminum.

Different uppercase letters in the column, within each factor, differ significantly ($P < 0.05$). Other interactions were not significant ($P > 0.05$).

Both legume species, Gliricidia and Sabiá, had an average biomass accumulation during the initial 5.5 years of approximately 10 Mg ha^{-1} $year^{-1}$ (Apolinário *et al.*, 2015). Biomass fractions included thin, intermediate, and thick branches and total leaves (19, 22, 54, and 5 %, respectively). During these years, nutrient absorption increased exponentially with growth (mainly due to woody tissue production) until it reached its maximum growth (between 7 to 10 years, depending on the species). The order of nutrient concentrations for all components of the above-ground biomass, in general, follows this sequence: N> Ca^{2+}> K^{+}> Mg^{2+}> P (Moura *et al.*, 2006). This could result in nutrient depletion in the soil over time (Alvarado, 2015), especially when maintenance fertilization is not considered. Nutrients associated with the residues, as litter, represent 50 % of total nutrients associated with aerial biomass, contributing to chemical soil properties with 40–45 % of the N, 54 % of the K^{+}, 56 % of the Mg^{2+}, and 28 % of the P (Montagnini, 2000).

Soil P also decreased over the years ($P < 0.05$), being considered as medium level in the years 2013 and 2017 and as low in 2018 (Table 1). This decrease may be associated with the concentration of the element in the above-ground biomass. For Sabiá with 8 years of establishment and diameter at breast height (DBH) between 3-4 cm, P concentrations of 0.88 g kg DM^{-1} were determined in the aerial biomass (Moura *et al.*, 2006). Considering the biomass accumulation of 50 Mg ha^{-1} reported by Apolinário *et al.* (2015), it is estimated that approximately 44 kg ha^{-1} of P were stocked in the arboreal biomass, not to mention the root system.

In Gliricidia, Barreto and Fernandes (2001) reported P concentrations of 1.7 g kg^{-1} for 4-year-old trees, resulting in an estimated P stock of 85 kg ha^{-1} of P, not considering the roots. In addition to the element depletion generated by the absorption of the plant throughout its growth, according to Larsen (2017) the total P content of the soils is relatively low (between 500 and 10,000 kg ha^{-1} P up to 50-cm deep) and a portion is usually in forms unavailable for plants absorption. These assumptions indicate the need to supply this element. The availability and absorption of this element stimulate the root growth of crops, increasing the area of soil explored by vegetation and greater absorption of other nutrients. Consequently, P deficiency could affect plant growth, seed formation, crop maturation, and nitrogen-fixing capacity of legume species (Dubeux Jr *et al*., 2014; Sanz-Saez *et al*., 2017).

In the interaction between treatment × year × distances ($P < 0.05$) for Mg^{+2}, values were greater in 2013 for both SPS and distances (Fig. 2A). This response is possibly associated with the lower requirement of trees because of younger age (2 years), enabling greater Mg^{2+} concentration from the deposited arboreal components.

Table 2. Potential acidity, total exchangeable bases, cation exchange capacity and base saturation between 0- to 20-cm depth, in silvopastoral systems intercropped with signalgrass in Itambé, Pernambuco state, Brazil.

Factor	H^++Al^{3+}	TEB	$CEC_{effective}$	$CEC_{potential}$	Base Saturation
			$cmol_c$ dm^{-3}		%
Year (Y)					
2013	5.0^B	5.3^A	5.8^A	10.3^B	53^A
2017	9.6^A	4.6^B	5.1^B	14.2^A	32^B
2018	9.8^A	4.6^B	5.0^B	14.4^A	32^B
Standard error	0.4	0.3	0.3	0.5	2.1
P-value	<0.0001	0.0446	0.0404	<0.0001	<0.0001
Treatment (T)					
Gliricidia + Signalgrass	7.5	5.3	5.6	12.7	43
Sabiá + Signalgrass	8.7	4.4	5.0	13.2	35
Standard error	0.6	0.4	0.3	0.6	2.60
P-value	0.1842	0.2175	0.2508	0.6464	0.1027
Distance (D)					
0 m	8.4^A	4.7	5.2	13.2	37^B
8 m	7.8^B	5.0	5.4	12.8	41^A
Standard error	0.4	0.3	0.3	0.5	2.0
P-value	0.0216	0.2904	0.5455	0.3013	0.0440
T × D					
Standard error	0.5	0.5	0.4	0.7	2.8
P-value	0.0438	0.0240	0.0666	0.8775	0.0008
Y × T × D					
Standard error	0.7	0.6	0.4	0.9	3.4
P-value	0.0510	0.1975	0.3802	0.5867	0.0039

H^++Al^{3+}: potential acidity; TEB: total exchangeable bases; CEC: cation exchange capacity

Different uppercase letters in the column, within each factor, differ significantly ($P < 0.05$). Other interactions were not significant ($P > 0.05$).

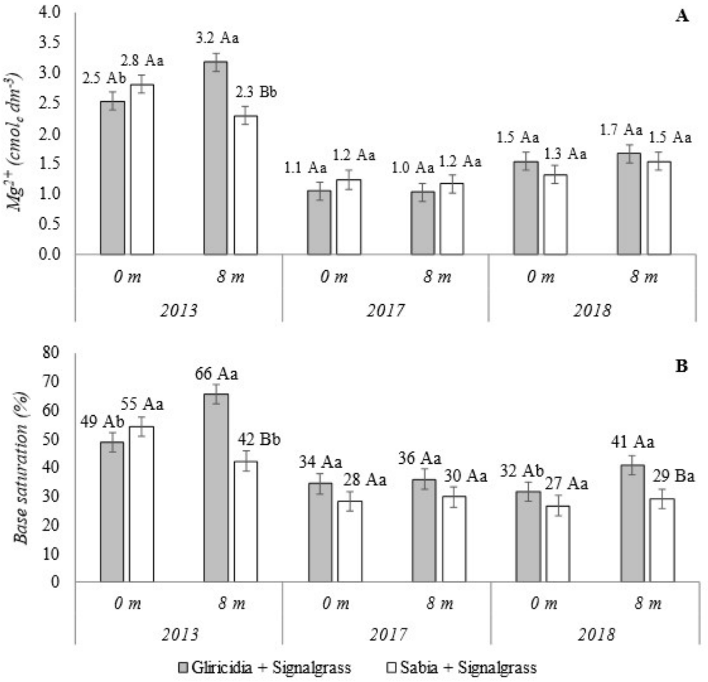

Fig. 2. Interaction between treatment × year × distances for soil Mg^{2}+ (A) and base saturation (B), in silvopastoral systems intercropped with signalgrass in Itambé, Pernambuco state, Brazil.
The same uppercase letters for treatments and lowercase for distance, across years, do not differ significantly ($P > 0.05$).

In 2017 and 2018, the average soil Mg^{2+} was lower than in 2013; however, soil Mg^{2+} concentration was still adequate for plant growth. According to Karley and White (2009), Mg^{2+} concentrations in the soil solution should be between 0.0125 - 0.85 cmolc dm^{-3} to support plant growth. This reduction observed between 2013 and 2017, may be related to the increase in tree nutrient requirement along 7 years of growth, promoting depletion of the element into the soil. Barreto and Fernandes (2001) and Moura *et al*. (2006), respectively, reported above-ground biomass Mg^{2+} concentrations in Gliricidia (6.6 g kg^{-1}) and Sabiá (2.8 g kg^{-1}). Considering these values, and biomass accumulation of 50 Mg ha^{-1} for both species during the initial 5.5 years of tree development (Apolinário et al., 2015), it is estimated that approximately 330 and 140 kg ha^{-1} were stocked in the biomass of Gliricidia and Sabiá, respectively. Magnesium is often stored in root cells and released to the xylem if shoots become Mg^{2+} deficient (Karley and White, 2009). Furthermore, the pH reduction promotes the leaching of some elements, including Mg^{2+} (Neina, 2019).

Interaction ($P < 0.05$) occurred between distances × year for K^+ (Fig. 3), with the greatest values in 2013, regardless of the distances and, in 2017, only between the double rows of legumes. The decrease in soil K^+ from 2013 to 2017 can be associated with tree nutrient requirements for tree development over time. Greater removal of K^+ and Mg^{2+} from the soil is a possible consequence of increased accumulation of these elements in biomass components, with younger trees having greater nutrient demand in leaves and branches (Ali *et al.*, 2017; Dick and Schumacher, 2019). For K^+, it is estimated that approximately 0.8 (16 g kg^{-1}) and 0.7 Mg ha^{-1} (14 g kg^{-1}) were stocked in the biomass of Gliricidia and Sabiá, respectively (Apolinário *et al.*, 2015; Barreto and Fernandes, 2001; Moura *et al.*, 2006).

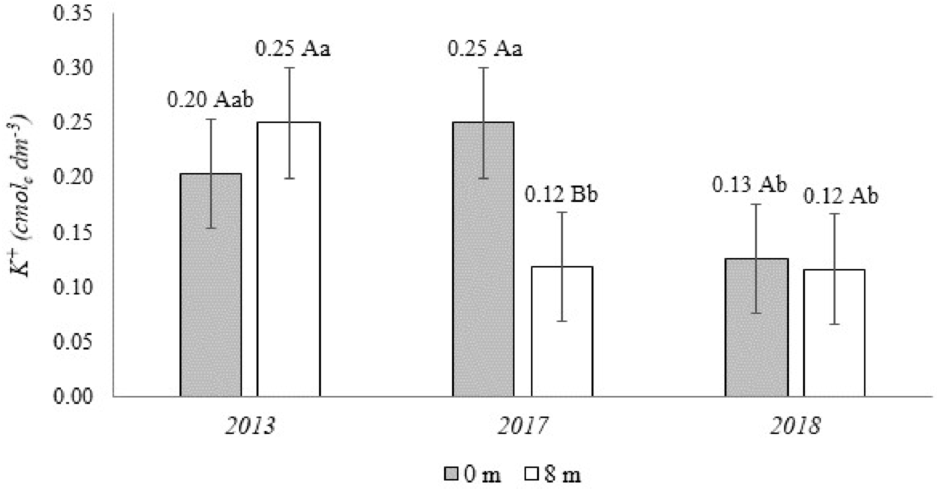

Fig. 3. Interaction between year × distances for soil K^+ in silvopastoral systems intercropped with signalgrass in Itambé, Pernambuco state, Brazil.
Same letters, lowercase for year within each distance and uppercase for the distance within each year, do not differ significantly ($P > 0.05$).

Similar to N, K^+ is the nutrient required in the largest quantity by plants (5 to 10 times more than P), and the requirement for optimal plant growth is between 20–50 g kg^{-1} in vegetative parts and fruits (Hawkesford *et al.*, 2023; Larsen, 2017). Overall reduction in soil K^+ concentration was observed in 2018 compared to 2013 (Fig. 3), with no differences between distances. A gradient with the distance from the trees is not observed, possibly, because these legumes have a length and distribution of homogeneous roots up to 8 m from the crown. Soil K^+ declined almost 50 % from the initial sampling in 2013 until the last sampling in 2018.

Greater soil K^+ between legume double rows in 2017 compared to the signalgrass strip (8 m away from double rows) in the same year is probably associated with the presence of animals in 2017 (Fig. 3). Before soil sampling, grazing animals were in continuous stocking for six months for the year 2017, and for a month prior to the 2018 sampling (before this, the continuous stocking was interrupted for four months). K^+ is eliminated in greater proportion by urine (Battisti *et al.*, 2018) and fecal K^+ from grazing livestock may present a faster release rate into the soil solution during the decomposition process compared with other elements (Lima *et al.*, 2018). Furthermore, greater fecal deposition might occur under the tree canopy because cattle seek shade during the warmer hours of the day (Dubeux Jr *et al.*, 2014). The litterfall is another factor that can affect the concentration of nutrients at different distances from the double row of legumes, however in both years, the evaluations were at the end of the rainy season.

In the interaction between treatment × sampling site ($P < 0.05$) for H^{+}+Al^{3+} and TEB, Gliricidia showed greater and lower values, respectively, between double rows, while Sabiá did not show differences between sampling sites (Fig. 4). This increase is probably associated with the H^{+} ions released by the roots, to balance loads, when the plant absorbs cations products of litter decomposition (Battisti *et al.*, 2018), since Al^{3+} remained stable through assessments (Table 2). Likewise, an increase of ion H^{+} release is expected to occur in highly nitrifying systems (Barth *et al.*, 2019), mainly from the Gliricidia litter, which contains more N than Sabiá litter (Apolinário *et al.*, 2016). In the signalgrass strips, 8 m away from the Gliricidia double row, the release of H^{+} ions must have occurred at a lower rate, mainly due to the lower litter mineralization, especially nitrification, related to the litter quantity and quality (Barth *et al.*, 2019; Battisti *et al.*, 2018; Dubeux Jr *et al.*, 2014). Under the Sabiá canopy, the overall greater litter deposition for Sabiá (Apolinário *et al.*, 2016) and probably greater tree development could lead to more root exudation and litter mineralization. Moreover, the greater nutrient uptake (mainly K^{+} and Mg) with immobilization in the biomass of trees that results in more ion H^{+} occupying the base cations sites in the CEC.

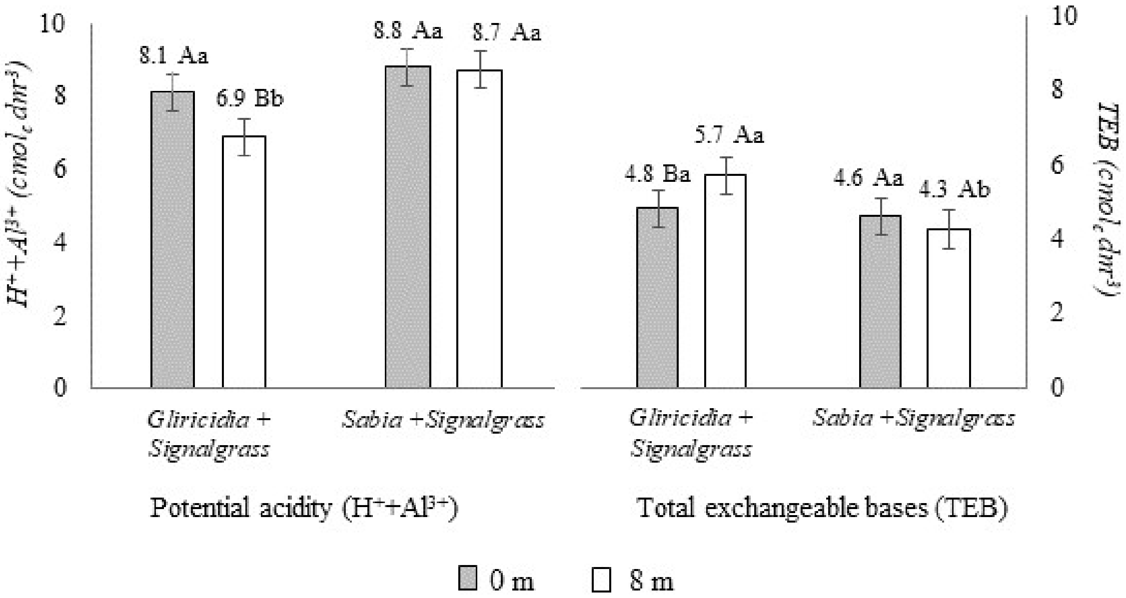

Fig. 4. Interaction between treatment × distances for soil potential acidity and total exchangeable bases in silvopastoral systems intercropped with signalgrass in Itambé, Pernambuco state, Brazil.
Same letters, lowercase for treatment and uppercase for distance, within each response variable, do not differ significantly ($P > 0.05$).

The $CEC_{effective}$ decreased ($P < 0.05$) and the $CTC_{potential}$ values (TEB + H^{+}+Al^{+3}) increased ($P < 0.05$) between years 2013 and 2017 (Table 2). Charges on organic colloids are pH-dependent, limiting the benefits in acid soils since $CEC_{effetive}$ decreases as pH decreases (Gruba and Mulder, 2015). In this case, the exchangeable base cations (Ca^{2}+, Mg^{2}+, K^{+}, Na^{+}) occupied less than 37 % of $CEC_{potential}$, with elevated H^{+} ions occupying the base-cations sites, associated probably with an increase of soil organic matter, as observed by Gruba and Mulder (2015).

Treatment × year × sampling sites interaction ($P < 0.05$) occurred for base saturation (Fig. 2B), with greater values in 2013, at 8 m away from double rows of Gliricidia. However, in this site and between the double rows of Sabiá, the average base saturation was greater than 50 %. Greater base saturation in 2013, with significant reductions in 2017 and 2018, must be a consequence of a negative balance between input and output nutrients 7 years after the SPS were established. This may result from the extraction of large amounts of nutrients from the soil (mainly P, Mg^{2}+ and K^{+}), stored in the tree biomass, and the absence of nutrient replacement through soil correction and fertilization. In 2013, as the trees were still small or medium in size, the extraction of nutrients from the soil was probably less, increasing with growth in later years.

Indicator of soil organic matter cycling

Soil basal respiration (SBR) was similar ($P > 0.05$) across years and between silvopastoral systems (Table 3), with lower SBR at the deeper soil layer (20-40 cm), likely related to reduced plant residues and less microbial activity for SOM decomposition (Correia *et al.*, 2015). Soil basal respiration decreases with soil depth and correlates significantly with the SOM content, concentrating biological activity at the top 15-cm soil layer (Rasouli-Sadaghiani *et al.*, 2018).

Table 3. Soil biological properties in silvopastoral systems intercropped with signalgrass in Itambé, Pernambuco state, Brazil.

Factor	LF-SOM	SBR	$\delta^{13}C\text{-}CO_2$
	g kg^{-1}	mg CO_2 kg solo^{-1} h^{-1}	‰
Year (Y)			
2017	1.6 A	0.19	-17.0
2018	0.9 B	0.16	-17.3
Standard error	0.1	0.01	0.3
P-value	<0.0001	0.0987	0.6185
Treatment (T)			
Gliricidia + Signalgrass	1.1	0.17	-17.4
Sabiá + Signalgrass	1.3	0.18	-16.9
Standard error	0.08	0.02	0.4
P-value	0.2556	0.7310	0.4754
Soil depth (cm)			
0-20	1.5 A	0.21 A	-16.9
20-40	1.0 B	0.14 B	-17.4
Standard error	0.08	0.01	0.4
P-value	0.0002	0.0007	0.4005
Distance (m)			
0	1.3	0.17	-17.4
4	1.1	0.17	-16.9
8	1.3	0.19	17.2
Standard error	0.1	0.01	0.5
P-value	0.4228	0.7094	0.8274
Y × T			
Standard error	0.1	0.01	0.6
P-value	0.0128	0.7607	0.0307

LF-SOM: light fraction of SOM; SBR: soil basal respiration; $\delta^{13}C\text{-}CO_2$: natural abundance of ^{13}C of the respired CO_2.
Different uppercase letters in the column, within each factor, differ significantly ($P < 0.05$). Other interactions were not significant ($P > 0.05$).

There was a treatment × year interaction for the light fraction of SOM and the δ13C-CO2 (Table 3). For the treatment × year interaction affecting the LF-SOM (Fig. 5), greater values occurred in 2017, regardless of treatment; however, in 2018 the consortium with Sabiá presented 63% more of this fraction in the soil compared with Gliricidia.

The LF-SOM in the system indicates recent changes in SOM, related to the amount of litter deposited in the soil (Alfaro *et al*., 2018; Lima *et al*., 2018) and according to Yang *et al*. (2019). The LF-SOM is an important C source for soil microorganisms. Lira Junior *et al*. (2020) after five years of the silvopastoral systems establishment, in the same area observed changes in microbiological attributes and quality of the soil organic matter on the first 20 cm of soil up, at the distance of up to 4 m away from the legume rows. This was observed through average soil C and N contents increase of 34 and 77 %, respectively. Apolinário *et al*. (2016), in an experimental area similar to the present study, observed greater litter deposition of Sabiá (5395 kg DM ha^{-1}) compared with Gliricidia (5204 kg DM ha^{-1}). However, differences between species in leaf production along the period under evaluation could have influenced the leaf litter fall and, consequently, LF-SOM. In 2018, leaf production of Sabiá was 133 % (5.6 Mg DM ha^{-1}) greater in relation to Gliricidia (2.4 Mg DM ha^{-1}), and 61% greater in 2017. Both soil assessments were carried out at the end of the rainy season when the greatest litter fall for these species occurs (Apolinário *et al*., 2015). Increasing the amount of LF-SOM input could result in a linear increase of C-CO_2 emission (Rui *et al*., 2016). However, the SBR did not show differences between SPS, probably reflecting the capacity of soil microorganisms to form new biomass (cell growth) rather than microbial respiration (Rui *et al*., 2016) in SPS with Sabiá.

Interaction between treatment × year occurred for $\delta^{13}C\text{-}CO_2$ (Fig. 5), because similar values occurred between treatments in 2017, but lower values were obtained in 2018 for the SPS with Gliricidia, indicating depletion of $\delta^{13}C$ in this system, likely due to greater legume-C contribution from Gliricidia. Greater labile C content can induce greater initial microbial activity and, over time, its composition could concentrate more recalcitrant C (Grover *et al*., 2015). Thus, in the SPS with Sabiá, recalcitrant materials likely limited the decomposition process in 2018, because of greater non-labile C concentration. The isotopic measurement of respired CO_2 is a useful method to quantify the abiotic production of CO_2 from carbonates due to the signature of $\delta^{13}C$ exclusive of carbonates and SOM (Santos *et al.*, 2014). According to Mamilov and Dilly (2011), never tilled soil produced $\delta^{13}C$-CO_2 during basal metabolism, which is typical for C_3-dominated plant community (−26‰), and organic matter in all tilled soils is oxidized more efficiently with long-term production of $\delta^{13}C$-CO_2 enriched (-19‰).

Thus, the $\delta^{13}C$-CO_2 respired in the soil can be used for the quantification of labile C (Ramnarine et al., 2012), identifying the system with the higher potential to increase the biological C drainage capacity of atmospheric CO_2 (Santos et al., 2014). Silvopastoral systems soils, implanted with Gliricidia or Sabiá, could represent a source of C-CO_2 with heavier isotope composition (Table 3), information that can be used for developing strategies to improve soil quality, reduce CO_2 emissions, and verify C retention efficiency, as observed by Mamilov and Dilly (2011).

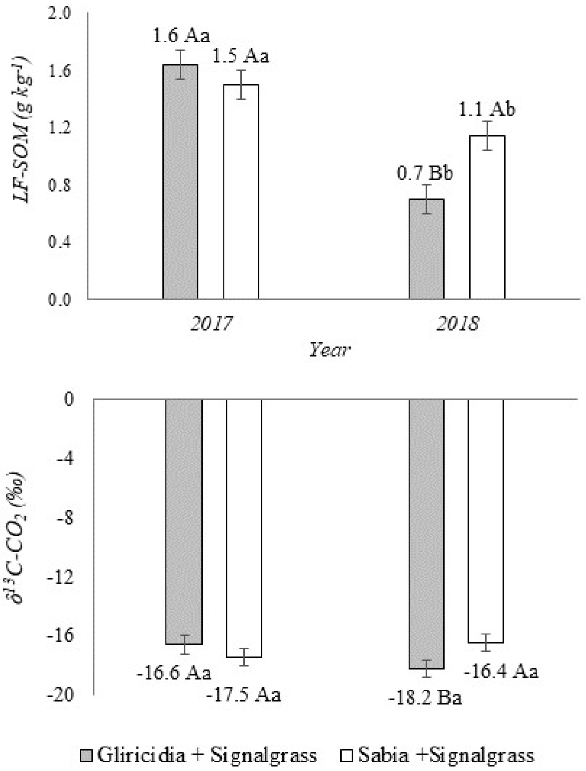

Fig. 5. Interaction between treatment × years for light fraction of SOM (LF-SOM) and δ13C-CO2 in silvopastoral systems intercropped with signalgrass in Itambé, Pernambuco state, Brazil.
Same letters, lowercase for year within each treatment and uppercase for treatment within each year, do not differ significantly (P > 0.05).

Conclusions

Silvopastoral systems integrating tree legumes in double rows with signalgrass caused a reduction in soil fertility regardless of the tree legume species used, evidenced mainly by soil pH, P, $Mg^{2}+$ and K^{+}. This deficiency resulted from the tree growth stage after 7 years of establishment and biomass nutrient stock, without maintenance fertilization. Since this study did not consider the complete development of the trees, it is necessary to evaluate the input and output of nutrients from the soil and nutrient stocks in plants with time, starting from the implementation of the integrated system. Such evaluation could increase knowledge on the dynamics of nutrients and the impact on soil health of when of legumes such gliricidia and sabia are incorporated in silvopastoral systems.

Greater litter fall in silvopastoral systems with sabia, might explain the higher light fraction of SOM obtained, without increases in soil respiration, providing potential for enhanced microbial C use efficiency. Enriched C-CO_2 isotope composition shows an efficient SOM oxidize in silvopastoral systems with Gliricidia or Sabiá. This information can contribute to the assessments related to CO_2 balance and C retention. Both silvopastoral systems contribute to C sequestration.

Conflict of interest. The authors declare that they have not any financial and personal relationships with other people or organizations that could inappropriately influence their work.

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Ethics statement: No animals were used.

Author contributions

Ana M. Herrera, Alexandre C. L. de Mello, Valéria X. O. Apolinário, José C. B. Dubeux Jr. and Erinaldo V. de Freitas, contributed to the study conception and design. Material preparation, data collection and analysis were performed by **Ana M. Herrera, Valéria X. O. Apolinário and Robert E. Mora-Luna.** The first draft of the manuscript was written by **Ana M. Herrera** and **Alexandre C. L. de Mello** and all authors commented on previous versions of the manuscript. All authors read and approved the final manuscript.

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