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Geoderma Regional 37 (2024) e00807

Contents lists available at ScienceDirect

Geoderma Regional

journal homepage: www.elsevier.com/locate/geodrs

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ARTICLE INFO

Keywords:
Cacao agroforestry systems
Acacia mangium
Albizia lebbeck
Carbon balance
Soil organic carbon
Litter quality
Acrisols

ABSTRACT

Cacao–based agroforestry systems are promoted as adaptation and mitigation solutions for cacao production and carbon sequestration. Based on a 20-year experiment, we assessed the impact of associated shade tree legume (ATL), *Albizia lebbeck* and *Acacia mangium* on the total carbon stock (in soil at 60 cm depth + tree biomass + litter) of cacao stands. This study included cacao systems shaded with either *A. lebbeck* (Cacao-Alb) or *A. mangium* (Cacao-Aca) and full-sun cacao stands (Control). Soil organic carbon (SOC) contents (up to 60 cm deep) were estimated by a calibrated near-infrared spectroscopy model. Total tree biomasses were estimated using allometric equations. Leaf litter was sampled from 1-m^2 quadrats. Compared to Control, Cacao-Aca had a significant negative impact on the carbon stock in the cacao biomass (−47%) as well and in the soil at depths of 10 cm (−23%), 30 cm (−21%) and 60 cm (−12%). In contrast, Cacao-Alb had a nonsignificant effect on carbon storage in the cacao biomass, whereas it generally had a positive influence on the SOC stock regardless of depth, i.e., +6% at the 0–10 cm depth, +7% at 0–30 cm, +20% at 30–60 cm and + 11% at 0–60 cm. Cacao-Aca had a significant positive impact (+71%) on the total carbon stock per hectare. The increase in Cacao-Alb relative to that in the Control reached +38%, but the difference was not significant. These contrasting results between the two tree legume species could be explained by the high-quality litter, reflected by the lower C/N and C/P ratios produced by *A. lebbeck,* and the greater negative impact of *A. mangium* on cacao biomass. The main finding of this study is that the impact of intercropping cacao with shade tree legumes on the stand-level total carbon stock depends on the ATL species.

1. Introduction

In the current context of climate change, the challenge of increasing or at least maintaining carbon (C) stocks in soils and the biomass of agricultural and forestry systems has become crucial. Increasing the organic C content of agricultural soils can reduce or even mitigate anthropogenic greenhouse gas emissions (Lal, 2004). In this context, a policy framework has been established with international guidelines, known as the “4p1000” initiative launched at the “21th conference of the parties (COP21)” in 2015. This initiative encourages an increase in soil organic matter (SOM) content and C sequestration by promoting agricultural practices adapted to local conditions (environmental, social and economic), notably agroecology, agroforestry, conservation agriculture or landscape management. Agroforestry systems are a land management practice that can improve soil health by enhancing SOC storage and nutrient availability and promoting soil organisms activity and diversity (Dollinger and Jose, 2018; Fahad et al., 2022; Kibblewhite et al., 2008; Sauvadet et al., 2020). Increasing SOC enhances soil fertility and productivity, improves soil biological properties and processes, and maintains soil biodiversity (Amelung et al., 2020; Jackson et al., 2017; Suárez et al., 2021). Furthermore, Niether et al. (2020), in their meta-analysis, reported that there was an overall positive but nonsignificant

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https://doi.org/10.1016/j.geodrs.2024.e00807
Received 20 July 2023; Received in revised form 15 April 2024; Accepted 8 May 2024
Available online 9 May 2024

effect of a cacao agroforestry system (cAFS) on SOC. They also reported many differences between various studies on the influence of cAFS on SOC.

Cacao (*Theobroma cacao L.*) cultivation is one of the main drivers of deforestation in the main producing countries like Ivory Coast due to the major strategic challenge of the cacao economy (Wessel and Quist-Wessel, 2015). Cacao stakeholders recommend agroforestry as a way to produce cacao while restoring tree cover, improving soil fertility and diversifying farmers' income (Critchley et al., 2022). The cAFS consists of growing cacao in association with other woody species that provide various ecosystem services (productivity, biodiversity, climate regulation) (Abou Rajab et al., 2016; Somarriba et al., 2013). Trees associated with cacao systems also provide shade for cacao and coproducts for farmers and contribute to soil nutrient replenishment through litterfall and decomposition, recycling leached nutrients beyond the reach of cacao roots (Asigbaase et al., 2019, 2021b; Tondoh et al., 2015). However, opinions concerning the benefits of cAFS in cacao farming are contradictory. Some authors have argued that the association of trees in cacao systems fosters SOM accumulation, C storage, nutrient recycling, biodiversity conservation and climate change mitigation (Abou Rajab et al., 2016; Asase and Tetteh, 2016; Asigbaase et al., 2021a; Batsi et al., 2021; Noumi et al., 2018; Sanial et al., 2022; Sauvadet et al., 2020; Vaast and Somarriba, 2014). According to other authors, shade trees may not enhance soil fertility and SOC sequestration on cacao farms, as one would expect, especially at a rate that could contribute to the long-term sustainability of these systems (Abdulai et al., 2018; Asare et al., 2017; Blaser et al., 2018; Blaser et al., 2017). These authors explained this by the fact that tree association with cacao did not show clear benefits to associated trees at the field level.

Carbon storage in these systems is achieved through the use of different pools, mainly soil, tree biomass, and litter (Norgrove and Hauser, 2013; Saj et al., 2017; Schroth et al., 2016). The ability of an agroforestry system to store significant levels of C is reported to vary according to the associated tree species and tree density and age (Abou Rajab et al., 2016; Blaser-Hart et al., 2021; Saj et al., 2017; Schroth et al., 2016; Silatsa et al., 2017). The SOC stock depends on the soil depth considered, the management strategy, and the quantity and quality of litter inputs (Asase and Tetteh, 2016; Mohammed et al., 2016). Litter decomposition plays a key role in improving the physical, chemical and biological characteristics of soil. Litter is a central nutrient resource and an essential link between plants and soils for the return and recycling of organic matter and nutrients (Hartemink, 2005; Naik et al., 2018; Triadiati et al., 2011; van Vliet and Giller, 2017). Plant litter improves the quantity of SOM and thus SOC. In general, the beneficial effects of legume species on SOM restoration and SOC stocks have been reported in the literature (Dominguez-Núñez, 2022; Koné et al., 2020). Furthermore, legumes are considered to have the ability to produce large amounts of biomass in the short run and fix atmospheric nitrogen, some of which is returned to the soil through litter decomposition. The use of legumes is an interesting alternative, as they can improve or restore soil organic status over short periods (Tian et al., 2000). This topic deserves additional attention because the litter dynamics associated with this function are very poorly known (Asitoakor et al., 2022; Blaser et al., 2017).

The Centre National de Recherche Agronomique (CNRA) launched in 1998, an experiment at the Divo Research Station to evaluate the growth and production of cacao associated with two species of shade tree legume, *Albizia lebbeck* (L.) Benth. (Fabaceae) and *Acacia mangium* Willd. (Fabaceae). After 20 years, the trial was still ongoing, allowing us to assess the total C balance of the soil–plant system and to test the hypothesis of a positive impact of shade tree legumes on the carbon stock of total tree biomass, litterfall, and soil organic carbon stock in cacao-based agroforestry systems. Therefore, this study investigated the impact of intercropping cacao with shade tree legumes on aboveground carbon stocks (trees and litter) and soil carbon stocks, as well as the total carbon balance after 20 years.

2. Materials and methods

2.1. Study area

This study was carried out at the Centre National de Recherche Agronomique (CNRA) in Divo, Ivory Coast (5° 47.528280′ N/−5° 15.041462′ W). The natural vegetation of this region is the Guinean semideciduous forest. The climate is subequatorial, with an annual average air temperature and humidity of 26 °C and 85%, respectively, both of which are subjected to seasonal variations. The rainfall regime is bimodal, with two dry seasons and two wet seasons. The annual average precipitation is over 1200 mm, with more than three consecutive months of the dry season (Ehounou et al., 2019). The soil is dominated by low-activity clays (mainly kaolinite) and a high sesquioxide content. They are classified as Acrisols (WRB, 2015), but they are chemically poor and characterized by low native fertility (Table 1 and ESM 1), resulting from very low nutrient reserves (WRB, 2015). However, these soils are generally considered suitable for cacao cultivation (Ehounou et al., 2019).

2.2. Experimental design

An experimental design was set up in 1998 on a former experimental cacao plantation site, which was left fallow and colonized by *Chromolaena odorata* for >10 years. The tree legumes *A. mangium* and *A. lebbeck* were planted after clearing, and the cacao seedlings were planted two years later. Therefore, the experimental design considered was 20 years old at the date of data collection in 2020 (ESM 2). The plots have been managed similarly to many cacao plantations in Ivory Coast, especially those operated by farmers, despite being on an experimental station. The experiment was thus conducted without fertilization but with regular weeding.

The experiment involved four blocks arranged perpendicular to the slight slope (<1%) over a 2-ha area. Initially, each block was subdivided into five rectangular plots of 726 m^2 (33 m × 22 m): one unshaded cacao monoculture plot (Control), two plots with *A. lebbeck* (Cacao-Alb), and two plots with *A. mangium* (Cacao-Aca). Alleys 4- m wide were used to separate both the blocks and the plots. Cacao trees were planted at a spacing of 2.5 m between trees in a row and 3 m between rows, i.e., at a density of 1333 plants ha^{-1}. Associated tree legumes (ATLs) were planted in a pattern of four trees per plot, i.e., 55 trees ha^{-1}. Some data concerning tree growth and density were collected in the early years, but very few were collected afterwards due to political troubles in the country. However, the experimental design was maintained with minimal management and annual cacao pod harvests, but yields were unfortunately no longer evaluated. In 2020, considering some tree mortality over time, three plots per block were selected for the present study, considering one plot per block for each modality: Cacao-Alb, Cacao-Aca, and the control.

In each Cacao-ATL plot, all the remaining 20-year-old legume trees (Table 2), *A. mangium* and *A. lebbeck*, were identified, and a radius of 10 m around these trees was defined as subplots. In the Control plots, four independent subplots with a 10 m radius were randomly delineated. Therefore, the main factor of this experiment was ATL.

2.3. Methods

In this study, three carbon pools of the soil–plant system were considered: the total tree biomass (per species, cacao and ATL), the litter biomass and the soil organic carbon. The C stock was determined for each pool at the plot level (see below for details), and the three components were summed to estimate the total C stocks for a given cAFS.

2.3.1. Assessment of carbon stock in cacao and associated tree legume biomass

Cacao trees and ATL density per unit area were determined, and their

Table 1
Pedo-horizon characteristics of the soil profiles from each of the studied systems on a 20-year-old experimental cacao plantation in Divo (Côte d'Ivoire), where cacao trees were associated with the tree legume *A. mangium* (Cacao-Aca) or *A. lebbeck* (Cacao-Alb), or not (Control).

	Cacao-Aca			Cacao-Alb			Control		
variables	H1	H2	H3	H1	H2	H3	H1	H2	H3
pH $_{H20}$	5,15	5,39	5,47	5,91	5,44	5,39	5,91	5,53	5,25
pH $_{KCL}$	4,70	4,82	4,65	5	4,68	4,47	4,96	4,59	4,36
Total N (%)	0,78	0,60	0,88	1,04	0,88	0,72	0,89	0,53	0,50
C (%)	6,43	5,59	8,71	9,67	8,62	6,98	8,60	5,07	5,23
P $_{total}$ (mg.kg^{-1})	94,48	102,35	131,38	138,78	108,89	99,41	130,37	84,86	119,83
Available P (mg.kg^{-1})	0,80	0,71	0,94	2,01	0,67	0,54	2,41	0,54	0,63
K^+ (cmol.kg^{-1})	0.09	0.12	0.12	0,19	0,10	0,08	0.15	0.10	0.12
Mg^{2+} (cmol.kg^{-1})	0,40	0,51	0,56	0,64	0,36	0,43	1.10	0.65	0.79
Ca^{2+} (cmol.kg^{-1})	2.07	2.16	2.66	3,35	3,50	3,53	2.39	1.44	1.28
Na^+ (cmol.kg^{-1})	0.04	0.04	0.04	0.03	0.03	0.03	0.02	0.02	0.02
Clay (%)	44.7	42.2	43.6	44,2	20,9	25,5	22.4	36.7	43.6
Fine Silt (%)	5.7	5.9	5.6	5,5	4,2	4,6	6.7	5.6	5.1
Coarse Silt (%)	2.7	4.1	2.7	2,7	2,9	3,7	3.5	3.3	2.9
Fine Sand (%)	12.3	12.6	12.1	11,2	20,7	16,8	21.4	15.8	13.2
Coarse Sand (%)	33.9	33.6	34.9	36,6	51,2	49,6	44.5	37.8	34.8
CEC (cmol.kg^{-1})	12,41	9,78	10,29	8,66	16,68	12,79	5.28	6.33	9.09

Pedo-horizon designation H1: A3(B), H2: B11 and H3: B12.

Table 2
Associated tree legumes characteristics (mean ± standard deviation) of diameter at breath height (DBH), tree height, tree crown diameter) and cacao trees density in the 20-year-old experiment testing cacao with associated tree legumes *A. lebbeck* (Cacao-Alb) or *A. mangium* (Cacao-Aca), or without associated trees (Control) in Divo (Ivory Coast).

	Treatments	
Tree variables	*Acacia mangium*	*Albizia lebbeck*
DBH (cm)	67.5 ± 26.8a*	42.9 ± 13.7b
Heigth (m)	20.8 ± 4.3a	13.9 ± 3.6b
Crown diameter (m)	13.7 ± 3.3a	14.0 ± 4.0a

* For each variable, values followed by the same letter in each line are not significantly different ($p < 0.05$).

biomasses (above- and below-ground) were calculated. The total biomass of cacao trees, including both aboveground (AGB) and belowground (BGB) biomasses, was estimated using allometric equations. AGB was estimated using the modified cacao-specific allometric equation of Somarriba et al. (2013) (Eq. (1), and BGB (Eq. (2)) was estimated using the allometric equations developed by Cairns et al. (1997), considering in the present case the diameter at 50 cm height and cacao total height at the top of the crown. The ATL biomass was estimated using the allometric equations of Chave et al. (2014) for aboveground biomass (AGBi; Eq. (3)) and those of Cairns et al. (1997) for belowground biomass (BGBi; Eq. (4)).

$$AGB = 10^{(-1,684+2,158*log(D50)+0,892*log(H)} \quad (1)$$

$$BGB = e^{[-1,0587+0,8836*ln(AGB)]} \quad (2)$$

$$AGBi = 0.0509*(Wi*(DBHi2)*Hi \quad (3)$$

$$BGBi = e^{[-1,0587+0,8836*ln(AGBi)]} \quad (4)$$

where AGB = aboveground biomass (kg.tree^{-1}), BGB = belowground biomass (kg.tree^{-1}), H = cacao tree height (m), D50 = diameter at 50 cm above the ground (cm), AGBi = ATL aboveground biomass (kg.ha^{-1}), BGBi = belowground biomass (kg.ha^{-1}), Hi = ATL tree height (m), DBHi = diameter at breast height, i.e., by convention measured at 1.3 m above the ground (cm), and Wi = wood density (g.cm^{-3}).

Individual tree biomass estimates were reported to the stand level by multiplying them by the density of trees per hectare. Biomass values were then converted to C stocks using a conversion factor of 0.48 according to the IPCC (2013) and were already used in the area by Sanial et al. (2022). At the plot level, the C stock in total biomass was estimated from the sum of AGB and BGB for both cacao and ATL for the Cacao-ATL treatments and from the sum of cacao biomasses for the control.

2.3.2. Litter sampling and chemical analysis

The litterfall under trees (ATL and cacao trees) was measured between October 2019 and November 2020. On one subplot per plot, three litter traps of 1m^2 each were set up at a height of 50 cm above the ground. The traps were installed at 1.75 m, between 3.25 and 5 m, and between 7 and 9 m from the ATL in both cAFSs, and from randomly chosen points in the control. The trapped litter was collected and weighed every month over this period. The Data were recorded for each trap per treatment. An aliquot was sampled and oven dried at 60 °C for 24 h to assess the dry mass of the litter. All litter materials from the same trap were mixed, crushed to pass through a 0.2- mm mesh sieve, and stored in a dry place before chemical analyses at an ISO-9001 certified laboratory (LAMA, IRD, Dakar). C and N concentrations were determined by dry combustion (Dumas, 1831) using a CHN analyser (Thermo Fischer Scientific CHN NA2000, Waltham, MA, USA). Litter was dissolved in concentrated HNO_3 solution at high temperature (110 °C) for 2 h, supplemented with H_2O_2, and the P content was subsequently analysed by colorimetry using a phosphomolybdic complex reduced with ascorbic acid, and K by MP-AES (Agilent). In November 2020, the ground litter amount was measured in a 1 m^2 quadrat at three randomly selected points within each study plot. The collected litter samples were oven-dried to determine the ground litter biomass (GLB). The GLB was then reported per hectare (Eq. (5)). The C stock of GLB (GLC) was also determined from the C concentration of litterfall under trees in each treatment (Eq. (6)).

$$GLB = HGLW \times 0.01 \quad (5)$$

$$GLC = GLB \times LC \quad (6)$$

where:
GLB: ground litter biomass (Mg.ha^{-1}).
HGLW: harvested ground litter weight in 1m^2 (g.m^2).
GLC: ground litter carbon stock (Mg.ha^{-1}).
LC: litterfall C concentration.
0.01: conversion factor to change reported values in the preferred Mg.ha^{-1}

2.3.3. Soil carbon pool assessment

Soil was sampled in November 2019 in the subplot using a 6-cm diameter cylindrical auger every 10 cm to a depth of 60 cm. In the

ATL subplots, sampling points were located at 1.75 m, 3.25 m, 5 m, 7 m, and 9 m from the tree legume trunk within two diametrically opposed transects (Fig. 1). Soil samples were then pooled 2 by 2 for each distance from the ATL to obtain composite samples per tree distance. In the control plots, the same sampling protocol was used considering a randomly chosen point on each subplot from which the sampling distances were defined. In each subplot, at each distance and for each depth, a composite sample was prepared by mixing two separate samples from two diametrically opposed transects, as was done in the case of ATL. The sampling was performed after the topsoil litter was carefully removed, and 30 samples were collected for each of the 44 subplots for a total of 1320 composite soil samples. The soil samples were air-dried and sieved at 2 mm. The 2-mm-mesh sieved samples were used for laboratory analysis, and the remaining part (Ø > 2 mm) was weighed to calculate the coarse particle content (SCP).

To assess the SOC content, infrared spectrometry and chemical analysis of soil C were used according to the procedure applied by Malou et al. (2021). Visible and near-infrared reflectance spectra (VisNIR) of each sample ($n = 1320$) were acquired at 2-nm intervals between 350 and 2500 nm with a LabSpec 4 spectrophotometer (Analytical Spectral Devices, ASD, Boulder, CO, USA). The numerical processing of spectral data was carried out using Unscrambler® X 10.4 (Camo Software, Oslo, Norway). A sample subset was selected from the spectral information ($n = 136$) as the most representative samples and analysed conventionally in the laboratory to construct a model and estimate the C content of the other soil samples ($n = 1184$). The soil C content of the subset samples was determined via dry combustion of 100 mg aliquots of soil (ground to <0.2 mm) using a CHN elemental analyser (Thermo Finnigan Flash EA1112, Milan, Italy) (NF-ISO 10694, 1995). These 136 samples were then assigned to calibration ($n = 80$) or validation ($n = 56$) subsets. The NIRS prediction models (see Table 3, for details) were built as described by Barthès et al. (2019) using a six segment random validation procedure on the calibration subset of samples, as well as a global partial least squares regression (Booksh and Boysworth, 2007) on (1) the results

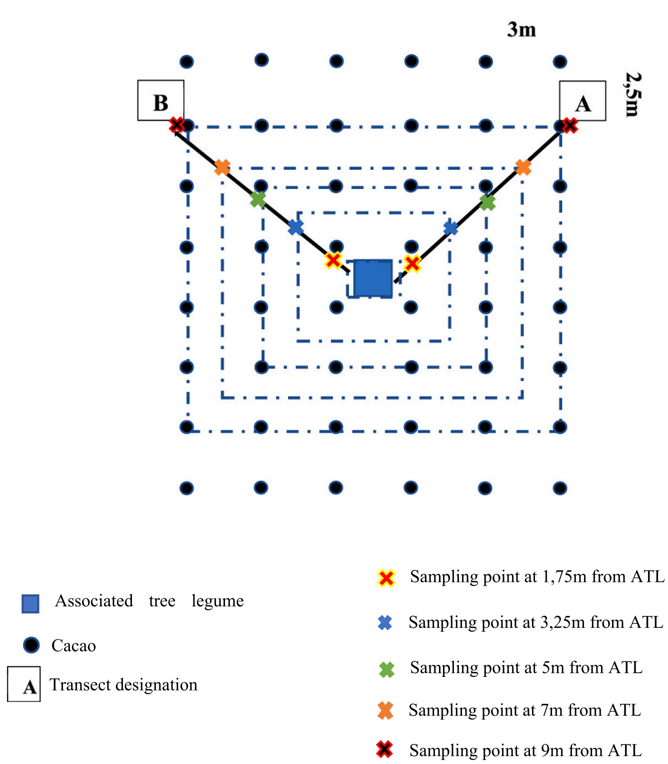

Fig. 1. Schematic representation of the soil sampling plan in a plot planted with a tree legume.
* For each depth, the values of variables are significantly different ($p < 0.05$).

obtained from laboratory measurements and (2) the VisNIR spectra. The validation model was sufficiently accurate for SOC prediction (see Table 3 for model figures of merit). It was applied to 1184 remaining samples that were not analysed by the conventional chemical method. The SOC contents were used to estimate the SOC stocks, as described below.

The soil bulk density (SBD) was measured by the cylinder method in each 10 cm depth layer using a 406,94 cm^3 cylinder on three sides of a 60 cm depth pit excavated at the centre of each study plot. The collected soil samples were then oven dried at 105 °C for 48 h and weighed to calculate the SBD. The SOC stock was then determined according to Eq. (7).

$$\mathrm{SOC}\left(\mathrm{Mg\ C.ha^{-1}}\right) = \mathrm{SOC} \times \mathrm{SBD} \times \left(1 - \left(\mathrm{SCP}/100\right)\right) \times \mathrm{d} \tag{7}$$

where SOC is the soil organic carbon stock ($Mg.ha^{-1}$), SCP is the coarse particle (>2 mm) content ($g.kg^{-1}$), SOC is the soil organic carbon content ($g.kg^{-1}$), SBD is the soil bulk density ($g.cm^{-3}$) and d is the soil thickness (m), all measured at the specified soil depth.

2.3.4. Plot total C stock assessment

The total C stock at the plot level was estimated as the sum of the SOC stock over the 0–60 cm depth, the ground litter C stock, the total biomass C stock of the cacao trees and, where applicable, the total biomass C stock of the ATL in the cAFS plots.

2.3.5. Data analysis

The effect of the tree legumes *A. lebbeck* and *A. mangium* on the studied parameters after 20 years of association with cacao was tested against the treatment without ATL (i.e., the control) using a linear mixed model, with the block effect considered a random effect. The distance effect was not considered for the litterfall and ground litter C stocks. A linear mixed model was also used to test the effect of ATLs on the studied parameters in interaction with soil depth and distance to the ATL. The Post hoc HSD Tukey tests (*lmer* R package, Kuznetsova et al., 2017) were used to compare the means of different factor levels, revealing a significant effect at the 5% level. The data are reported as the mean ± standard deviation.

Random Forest (RF) regression (Breiman, 2001; Breiman et al., 2018) was also used as a modelling tool to assess the relative importance of litter quality indicators (as predictors) to the SOC content in the 0–10 cm soil layer because the SOC stock and the presence or absence of ATL are not independent. RF is a classification method based on machine learning that builds numerous decision trees by randomly splitting the dataset (bagging). Unlike linear regression, RF analysis can handle nonlinearities and allows the definition of both continuous and categorical variables for predictors. The default RF function options in the R package were applied. We defined the following parameters required by the model: the number of trees to build in the forest (*ntree* = 500) and the number of predictors to use in each tree-building process (*mtry* = 2). The performance of the global RF regression models was evaluated through the percentage increase in the root mean square error, which allows us to determine how much the error would increase if the predictor was completely random; in other words, it prefigures the loss of precision of the model if the predictor is not considered to estimate the variable. Thus, high values of error gain (%) indicate greater explanatory factors in the RF model.

The data frames were managed with the *plyr* R package (Wickham, 2009), and the graphs were designed with the *ggplot2* R package (Wickham, 2009). All the statistical analyses were performed using R software (version 4.3.0; http://www.r-project.org/), under the R studio interface.

Table 3
Parameters describing the selected NIRS prediction models that gave the best combination of performances on both the calibration and validation sub-sets for soil organic carbon contents (SOC in g kg^{-1}).

Predicted variable	Optimal data processing	LV[a]	Subset	n[b]	Mean[c]	SD[c]	IQ[c]	RMSE[d]	R^2	RPD[e]	RPIQ[f]
SOC	SNV[g]	4	cal[h]	80	1.50	1.20	1.60	5.4	0.79	2.21	2.96
	SNV	4	val[i]	56	1.12	0.69	0.87	3.1	0.79	2.24	2.80

[a] Number of latent variables used in the model.
[b] number of samples in the sub-set.
[c] mean, standard deviation (SD) of variable mean and interquartile range (IQ) in the sub-set, expressed in $g.kg^{-1}$.
[d] root-mean-square error by subset, expressed in $g.kg^{-1}$.
[e] ratio of SD to RMSEP.
[f] ratio of performance to interquartile distance.
[g] standard normal variate (SNV).
[h] calibration.
[i] validation.

3. Results

3.1. Carbon stocks in cacao and associated tree legume biomass

The C stock of the cacao biomass which included both above- and below-ground components, showed significant variation ($p < 0.001$; Table 4) between treatments. These data showed a significant decrease in the C stock of cacao trees in the Cacao-Aca plots compared to that in the control plots. Cacao trees in the Cacao-Alb and control plots had similar biomass C stocks. The C stock in the biomass of cacao and ATL at the stand level revealed a greater increase for the Cacao-Aca association than for the Cacao-Alb association ($p = 0.01$; Table 4).

Table 4
Stocks of C (mean ± standard error; $Mg.ha^{-1}$) in ground litter, cacao or associated tree legumes (ATL) biomasses (below + above-ground) and as soil organic carbon (SOC) at different depths for cacao-based systems associated with legume trees *A. lebbeck (Cacao-Alb) or A. mangium (Cacao-Aca)*, or in monoculture (Control) in a 20-year-old experiment at Divo (Cote d'Ivoire).

Variables	Soil depth (cm)	Cacao-Aca	Cacao-Alb	Control	Probability legumes Tree effect (lmer test) df	F	P
Ground litter C stock ($Mg.ha^{-1}$)	–	2.0 ± 0.6a*	1.5 ± 0.6b	1.3 ± 0.4b	2	7.1	<0.01
Cacao biomass C stock ($Mg.ha^{-1}$)	–	17.0 ± 13.7b	28.3 ± 17.2a	31.9 ± 22.1a	2	18.9	<0.001
Cacao+ATL C stock ($Mg.ha^{-1}$)	–	102.9 ± 91.1a	59.8 ± 27.0ab	31.9 ± 17.9b	2	6.4	0.01
SOC stock (Mg C. ha^{-1})	0–60	50.1 ± 23.8b	63.5 ± 28.5a	57.2 ± 30.8ab	2	6.1	<0.01
SOC stock (Mg C. ha^{-1})	0–10	15.5 ± 6.1b	21.3 ± 10.8a	20.1 ± 9.8a	2	10.9	<0.001
SOC stock (Mg C. ha^{-1})	0–30	32.7 ± 16.2b	44.3 ± 23.9a	41.3 ± 26.5a	2	7.5	<0.001
SOC stock (Mg C. ha^{-1})	30–60	17.4 ± 9.5ab	19.1 ± 8.9a	15.9 ± 5.9b	2	3.2	<0.05
Total C stock ($Mg.ha^{-1}$)**	0–60	154.9 ± 89.2a	124.7 ± 37.8ab	90.5 ± 43.9b	2	5.1	0.01

* For each variable, values followed by the same letter are not significantly different ($p < 0.05$).
** calculated using SOC value at 0–60 cm deep.

3.2. Litter production and quality

Compared with the control, the ATL treatment yielded approximately twice as much litterfall during one year ($p < 0.001$; Table 5). The yield of litter on the ground in the Cacao-Aca plots, unlike that in the Cacao-Alb plots, was significantly greater than that in the control plots ($p < 0.01$; Table 5). The Cacao-Alb and control treatments did not significantly affect the litter biomass C stock.

The litter quality variables showed significant variations between treatments ($p < 0.001$, Table 5). The Cacao-Aca and Cacao-Alb litters had significantly greater total C concentrations than did the control litter. The litter of Cacao-Alb had significantly greater N, P, and K contents than the Cacao-Aca and control litter. The total N, P and K contents were not significantly different between the Cacao-Aca- and control-derived litter. The C/N and C/P ratios of the litter ($p < 0.001$) were lower for the Cacao-Alb treatment than for the control and Cacao-Aca treatments. The Cacao-Aca and control treatments had similar litter C/N and C/P ratios. However, the litter N/P ratios were similar among the three treatments.

3.3. Soil organic carbon

For all treatments, the vertical distribution of the SOC content (Fig. 2) showed significant variations between treatments according to soil depth. The SOC content in the 0–10 cm layer of the Cacao-Alb soil was significantly greater than that in the control, at 17.1 ± 9.4 and 15.2 ± 6.7 $g.kg^{-1}$, respectively. In contrast, the SOC content in the Cacao-Aca treatment was significantly lower than that in both the control and Cacao-Alb. No significant difference in SOC content was detected between the Cacao-Alb and the control in the other soil layers. In the 30–60 cm layer, the SOC content showed very little variation between treatments, notably around the average of 0.60 $g.kg^{-1}$.

The SOC stocks calculated for the 0–10, 0–30, 30–60 and 0–60 cm soil layers showed significant differences between treatments ($p < 0.05$, Table 4). Indeed, there was a significant decrease in the SOC stock in the 0–10 cm and 0–30 cm layers (−23% and − 21%, respectively) under Cacao-Aca compared to that in the control. In contrast, Cacao-Alb and the control exhibited similar values at these depths. At the 30–60 cm depth, the SOC content was significantly greater in the Cacao-Alb treatment (+20%) than in the control. Moreover, there was no significant difference in the SOC stock at this depth between the Cacao-Aca treatment (+9%) and the control. At the 0–60 cm depth, the SOC stock was significantly lower in the Cacao-Aca treatment than in the Cacao-Alb treatment and the control. The difference in SOC stocks at the 0–60 cm depth was not significantly different between the Cacao-ATL treatment and the control, despite a trend towards an increase in one case (Cacao-Alb; +11%) and a decrease in the other case (Cacao-Aca; −12%) (Table 4).

Fig. 3 illustrates the order of importance of the selected litter quality parameters as predictors of the SOC content in topsoil (i.e., the 0–10 cm

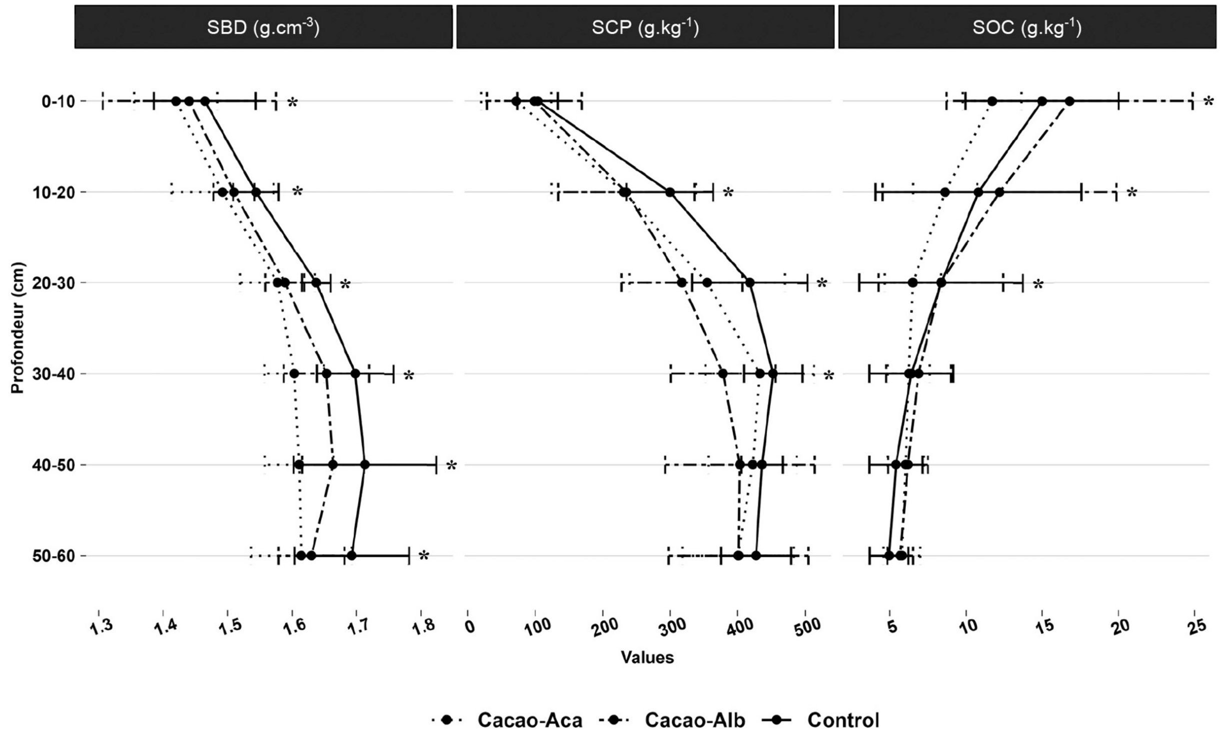

Fig. 2. Soil profile of bulk density (SBD), coarse particles (SCP) and carbon content (SOC) over 0–60 cm depth.

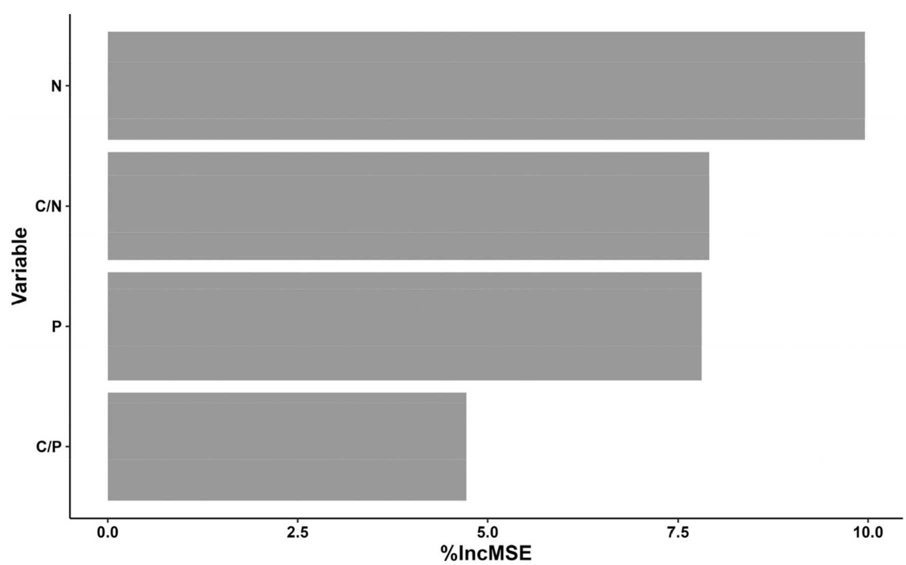

Fig. 3. Relative importance of litter quality parameters as predictors of soil organic carbon content in the 0–10 cm layer, as expressed by the increase in root mean square error (%IncMSE) in the Random Forest regression models.

layer). The N content of the litter explained the most of the SOC content in the 0–10 cm soil layer (Fig. 3). The C/N ratio of the litter and its P content may also explain the SOC content in the topsoil of the different treatments.

3.4. Total carbon stock of the cacao system

The total C stocks of the cacao systems, i.e., all the C pools, were 154.9 ± 89.2, 124.7 ± 37.8 and 90.5 ± 43.9 Mg C.ha^{-1} for the Cacao-Aca, Cacao-Alb, and control systems, respectively, with a significant effect of the treatment (Table 4). The total C stocks of the cacao systems increased by approximately 71% in the presence of *A. mangium* and 38% in the presence of *A. lebbeck* (Table 4). However, the total C stock value for the cAFS with *A. lebbeck* was not significantly different from that of the cacao monoculture (Table 4). The contributions of the ATL and cacao tree biomasses to the total C of the Cacao-Aca association were found to be large. In fact, *A. mangium* biomass contributed almost 50% of the total C stock in the Cacao-Aca plots, while cacao trees contributed 12% of the total C stock in the cacao plots (Fig. 4). Both *A. lebbeck* and cacao trees represented approximately 23% of the C stocks; therefore, cacao trees are an equivalent contributor to the total C stock in the Cacao-Alb treatment. In contrast, SOC was the largest relative contributor to Cacao-Alb, similar to that in the monoculture control, with >50% of the total C stock of the system (Fig. 4).

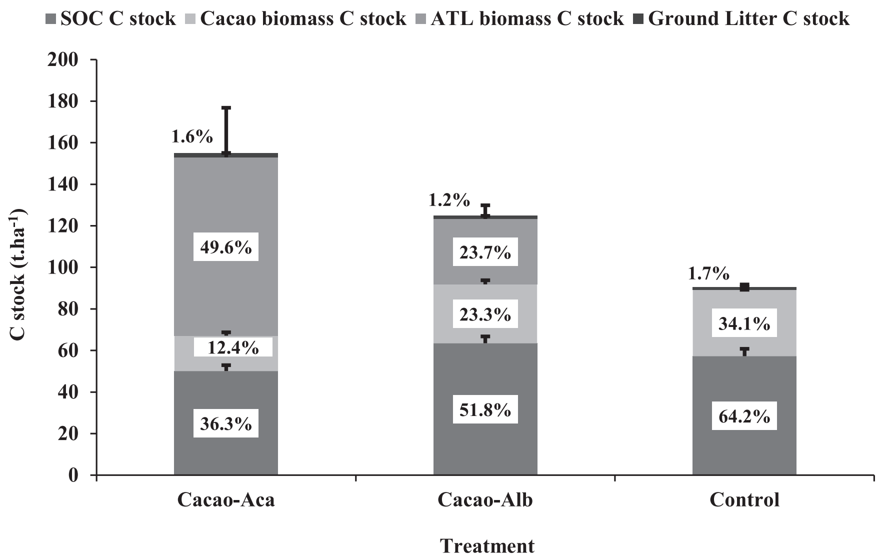

Fig. 4. Different carbon pool proportion in the total carbon stock balance at the plot level for each treatment.

4. Discussion

The greater amount of ground litter in Cacao-Aca probably reflects its recalcitrance. In fact, litter decomposition and disappearance rates are known to correlate with litter quality variables that shape the activity of the involved microbial communities (Bahram et al., 2020; Dawoe et al., 2010; Prieto-Rubio et al., 2023). Specifically, litter recalcitrance, as reported by (Yao et al., 2021), is known to interfere with decomposition, similar to biochemical compounds such as lignin and tannins. *A. mangium* leaves (phyllodes) were deemed to be rich in tannins, lignin and polyphenols, resulting in a 40–60% lower annual decomposition rate than that of *A. lebbeck* leaves, for which a 60–70% annual decomposition rate was reported (Bernhard-Reversat and Schwartz, 1997; Ngoran et al., 2006). Indeed, litters that are rich in tannins, phenols or lignin decompose more slowly than litters that are rich in cellulose. High-lignin litter usually increases the plant residue contribution to the intermediate stability SOM pool (Cotrufo et al., 2015). The Cacao-Alb litter materials had the lowest C/N and C/P ratios but the highest N/P ratio. These factors are conducive to a faster decomposition rate (Chae et al., 2019; Konan et al., 2021; Koné and Yao, 2021). Cacao-Alb litter with an average C/N ratio of 24.6 could therefore be considered more easily decomposed than the other two litter treatments (Kuo and Jellum, 2000; Yao et al., 2021). For a better effect of cAFS on SOC storage, Yao et al. (2021) emphasized the importance of growing cacao in association with trees that produce good litter quality for better availability of cacao nutrients and faster C storage in the soil. Despite the amount of C litter that reached the soil, the present study revealed through the RF approach that simple litter quality parameters such as the N content, C/N ratio or P content could be used to help explain the SOC content in the topsoil.

The vertical distribution of SOC indicated a decrease with depth in terms of SOC content and thus in stock. The SOC stock in the 0–30 cm soil layer was almost two-thirds that in the 0–60 cm layer. In turn, the 0–10 cm layer represented two-thirds of the SOC stock at the 0–30 cm depth. Changes in SOC stocks are controlled by the balance between C inputs from plant residues and C losses mainly through decomposition (i.e., heterotrophic soil respiration). The positive effect of *A. lebbeck* compared to that of *A. mangium* on the SOC stock up to 30 cm depth was probably related to the quality and faster decomposition of its litter. The presence of ATL also positively impacted the SOC stocks at the 30–60 cm depth, with values of +9.4% and + 20.1% for *A. mangium* and *A. lebbeck* respectively, although these values were only significantly greater for *A. lebbeck,* than for to control. It has been reported that *A. mangium* and *A. lebbeck* have extensive lateral root systems, with approximately 80% of the length concentrated in the top 60 cm of soil (Orwa et al., 2009; Saifuddin et al., 2022). Root systems, through both root turnover and exudates, are among of the main pathways by which plants release organic C in the soil, particularly at this depth (Rajab et al., 2018). The presence of companion trees in a cacao plantation could also concomitantly modify the soil bulk density, as was observed in the ATL plots at this soil depth, where the SBD tended to be lower than that of to the control.

Globally, the 20-year intercropping of cacao trees with *A. mangium* and *A. lebbeck* did not result in more soil C in the 0–60 cm layer than in the monoculture. However, it induced a positive trend with *A. lebbeck*. Thus, the impact of the associated shade trees on cAFS is likely to vary depending on the tree species; particularly with regard to SOC stock but also to cacao productivity (biomass and bean yields) as highlighted by Sauvadet et al. (2020). Monroe et al. (2016) showed that in cAFS, cacao trees in pure stands were more efficient at accumulating C than were those intercropped with rubber trees. Blaser et al. (2017) suggested that this might also be the case for many shade tree species. For these authors, the lack of a clear beneficial effect of shade trees on cAFS SOC storage could be explained by the fact that cacao trees are perennial small trees (5 to 8 m high) that also produce nonnegligible amounts of litter annually, contributing substantially to SOC and thereby hiding the contribution of ATLs to the SOC stock.

The presence of ATLs also had a negative influence on the cacao biomass, which was particularly significant for *A. mangium* unlike *A. lebbeck*, compared to cacao the control. Some studies have previously reported a limited influence of specific shade tree species on improving biomass and soil C sequestration in cacao agroforestry stands (Asitoakor et al., 2022; Blaser et al., 2017; Mohammed et al., 2016). However, Cacao-ATL intercropping had an overall positive influence on the total C stock of cacao plantations as reported by N'Zi et al. (2023) and Sauvadet et al. (2020). The increase in the total C stock of these associations was mainly due to the increase in the ATL biomass C stock. This positive influence of ATLs on the total C stock of the cacao stand was significant for *A. mangium* but not for *A. lebbeck*. *A. mangium*, a large tree (with a height of 21 m and canopy diameter of 13.6 m), contributed significantly to the total C sequestration of the Cacao-Aca plantations. Norgrove and Hauser (2013), in Cameroon, also found that shade trees, due to their biomass, were the largest contributors to the carbon stock of cacao agroforestry systems (cAFS), with an average stock of 121.1 Mg.

ha^{-1}, compared to 14.4 Mg.ha^{-1} for cacao trees. Therefore, the Cacao-Aca tree biomass C stock and its litter C possibly compensated for the SOC and cacao C stock decline observed in these plots. The association of cacao with trees producing high wood and litter biomass and with better-quality litter decomposing faster could increase the soil and tree biomass C stock, as reported by Sauvadet et al. (2020), and consequently the total C stock.

The mean C stock of cacao biomass, shade trees and soil and their respective contributions to the total C stock at the stand level were much lower than those reported by Norgrove and Hauser (2013) for Cameroonian cacao systems and Afele et al. (2021) for Ghanaian cacao systems. However, these results are comparable and are within the range reported by Asigbaase et al. (2021a) and Mohammed et al. (2016) for cacao farming systems in Ghana. These values are also close to the mean values reported for similar cacao systems in Central America by Somarriba et al. (2013) but are higher than the values for total C stock and the fractions represented by the different C pools in the total C stock reported by Schneidewind et al. (2019) for Bolivian cacao systems. Moreover, as noted by Asigbaase et al. (2021a), the age, variety and density of cacao and shade trees as well as their diversity influence the capture, storage and sequestration of C in these systems and may account for the differences in the mean C stock of the cacao and shade tree biomass as well as in the total C stock. In addition, direct comparison is limited by the large variation in climatic conditions and soil properties between this work and these studies. However, our study highlighted the potential of shade trees, particularly tree legumes such as *A. mangium* and *A. lebbeck* intercropped with cacao trees on C sequestration in these systems. Norgrove and Hauser (2013) in Cameroon reported the same C sequestration potential for shade trees in cacao stands, with an average stock of 121.1 Mg.ha^{-1}, compared to 14.4 Mg.ha^{-1} for cacao trees. Nadège et al. (2019) in Cameroon also reported an average stock of 94.2 Mg.ha^{-1} for shade trees compared with 30.1 Mg.ha^{-1} for cacao trees. These authors also maintained that cutting shade trees would significantly reduce the C stocks of these cacao farming systems, thus confirming the carbon sequestration potential of shaded cacao farming systems compared with unshaded systems. However, the choice of companion trees for cacao trees must address both the issue of increasing the productivity (biomass and bean yield) of cacao trees and improving the carbon sequestration of these systems, as highlighted by Monroe et al. (2016).

5. Conclusions

A. mangium significantly increased the total C stock of the studied cAFS. However, this ATL had a negative impact on cacao biomass and soil C pools. Due to its high biomass, intercropping *A. mangium* with cacao seems more beneficial for C sequestration at the stand level than at the cacao tree level. In turn, *A. lebbeck* had no significant effect but rather had a positive effect on the SOC stock and total carbon stock at the stand level due to the better quality of the litter it produced.

These findings suggest that intercropping cacao with shade tree species with high biomass and high-quality litter production can maximize C storage in cAFS. This study highlighted that, depending on the specific C pool to be improved, a specific shade tree legume species can be used. The presence of ATL provides additional aboveground biomass that can act as an effective carbon sink, removing carbon from the atmosphere. Given this concern, the selection of *Acacia mangium* as an ATL species in local cacao-based agroforestry systems could be an interesting approach to climate change mitigation but probably not from the cacao farmer's perspective, as it negatively affects both the biomass of the cacao tree and the bean yield.

Although further research into a possible increase in cacao bean yield with less obvious C gain under its influence is needed, *Albizia lebbeck* sounds more promising for locally creating a multifunctional system that benefits both the environment and the farmers.

Funding

This research was supported by the SoCa project granted by the BNP Paribas Foundation through its 2017 Climate initiative.

CRediT authorship contribution statement

Brahima K. Silue: Writing – review & editing, Writing – original draft, Visualization, Methodology, Investigation, Formal analysis, Conceptualization. **Armand W. Koné:** Writing – review & editing, Visualization, Supervision, Methodology, Conceptualization. **Dominique Masse:** Writing – review & editing, Visualization, Supervision, Methodology, Conceptualization. **Patricia Moulin-Esmard:** Writing – review & editing, Formal analysis. **Alain J.A. Kotaix:** Writing – review & editing. **Lydie Chapuis-Lardy:** Writing – review & editing, Supervision, Project administration, Funding acquisition, Conceptualization.

Declaration of competing interest

The authors declare that they have no conflict of interest.

Data availability

The datasets generated during and/or analysed during the current study are available in open access with controlled access and DOI

Acknowledgements

We are grateful to Lucette Adet and Joachim Kouamé and to the field staff of CNRA center in Divo for their contribution to the sample collection. The authors deeply thank the technical staff of the IRD LAMA's laboratory in Dakar for their assistance.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.geodrs.2024.e00807.

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