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Assessment of soil organic carbon: The influence of *Ecuadorian cocoa* varieties

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ABSTRACT

This study assessed the soil organic carbon (SOC) stocks in three 15-year-old Ecuadorian cocoa plantations, focusing on the impact of varieties EET 95, EET 103 and EET 116. Twenty sampling points per plantation were established, stratifying by location (under-tree and in-row) and depth intervals (0–5, 5–10, 10–20 and 20–30 cm). Statistical significant differences in SOC concentration (%) and stock accumulation were determined by Analysis of Variance. Results revealed that EET 116 exhibited the highest SOC accumulation, reaching 41 t ha^{-1} under trees and 54 t ha^{-1} in-rows. EET 103 showed intermediate SOC stocks (37 t ha^{-1} under trees, 47 t ha^{-1} in-rows), while EET 95 had the lowest (36 t ha^{-1} under trees and in-rows). Statistical analysis indicated no significant differences in SOC stocks between EET 95 and EET 103, nor between EET 103 and EET 116 under trees. However, EET 116 in-rows demonstrated significantly higher SOC stocks compared to all other treatments. Overall, these results emphasize the complex factors that influence the dynamics of SOC in cocoa plantations, highlighting the importance of considering varietal differences in carbon sequestration strategies for sustainable cocoa production.

Keywords: carbon sequestration, Ecuador, EET varieties, soil health, cocoa crops, sustainable agriculture.

INTRODUCTION

Increased atmospheric concentrations of greenhouse gases (GHGs), including C, N and P compounds, have driven global warming to become a critical environmental concern (Fu and Waltman, 2022). GHG emissions are dominated by carbon dioxide (CO_2), which constitutes approximately 80%, followed by methane (CH_4) at 11% and nitrous oxide (N_2O) at 6% (Benbi, 2013).

One of the dominant contributors of GHG emissions is agriculture, emitting roughly 18.4% of the global output, with CH_4 and N_2O being the primary pollutants (Brown et al., 2021; Kowalska et al., 2017; Laborde et al., 2021; Rehman et al., 2020; Tubiello et al., 2013). When it comes to the strategies for mitigating climate change impacts, soil organic carbon (SOC) sequestration removes atmospheric carbon dioxide through photosynthesis and stabilizes carbon in soil organic matter (OM) (Yang et al., 2022). OM decomposition and consequently SOC storage, are influenced by the conditions present in saline and sodic soils (Guillén et al., 2023; Reyna et al., 2023).

Soil, as the resource that supports life and agriculture, plays a crucial role in carbon storage. Therefore, SOC levels are significantly influenced by soil management practices (Gómez et al., 2022). Conservation tillage and diverse crop rotations, which maintain or enhance SOC, improve soil structure and porosity. Conversely, aggressive tillage and monoculture lead to SOC depletion (Gómez et al., 2022). SOC is essential for maintaining soil health, directly impacting crop productivity and the availability of nutrients, particularly N (Reyna-Bowen et al., 2020).

Regional GHG emissions, expressed as percentages of the global total, are: Asia (44%), North and South America (17%), Africa (15%), Europe (11%), and Australia/Oceania (4%) (Rehman et al., 2020; Tubiello et al., 2013). Multiple features, including climate, soil type and agricultural practices, influence this trend (Mrówczyńska-Kamińska et al., 2021; Shakoor et al., 2021). Within regions like Latin America, emission levels can vary widely due to various agricultural practices. In Ecuador, approximately 70.0% of GHG emissions come from agricultural activities (Cornejo and Wilkie, 2010), further contributing to soil degradation in this country (Padilla and Haro, 2021).

In the agroforestry systems of *Theobroma cacao L.*, high planting densities significantly impact SOC (Monroe et al., 2016). Cocoa is vital for Ecuador, with annual exports growing at approximately 4.5% and a current planting area of 543.6 ha^{-1} (Purcell et al., 2018). Ecuador ranks third in organic cocoa exports, with 10% of production in the Amazon region, mainly in conventional and organic agroforestry systems (Silva et al., 2022).

Previous research suggests that cocoa crops demonstrate a significant capacity for CO_2 assimilation, resulting in carbohydrate synthesis equivalent to approximately 14% of the global cocoa yield (Ortiz-Rodríguez et al., 2016). Consequently, cocoa cultivation presents a promising possibility for SOC sequestration, contributing to both rural socioeconomic development and ecological conservation (Goñas et al., 2022; Reyna-Bowen et al., 2018). Recognizing that cocoa variety influences the SOC sequestration rates through variations in OM production and decomposition, the purpose of this research was to assess the effect of specific cocoa variety plantations on SOC stock.

MATERIALS AND METHODS

This research was developed in the coast region of Ecuador, in Manabí province, Calceta canton specifically in an area located at 0° 51' 46" S, 80° 80' 61" W, spanning an elevation gradient from 19 to 80 meters above sea level (Figure 1). Soil samples were collected from three 15-years-old *Theobroma cacao L.* plantations. With a tropical climate, this region holds two seasons: rainy (from January to May) and dry (from June to December), the main meteorological annual parameters include an average temperature of 25.6 °C, precipitation of 838.7 mm and a potential evapotranspiration of 1365.2 m.

The cocoa crop, consisting of varieties EET 95, EET 103 and EET 116, was planted in March 1990 on 0.84 hectares. The trees are spaced 4 meters apart within rows. Weed control is performed every 8 to 15 days with manual scything, targeting broad-leafed and thin weeds. Phytosanitary measures involve pruning twice a year to remove diseased fruit, sucker shoots and branches. Insect management occurs every 8 days using traps and targeted applications. Harvesting is manual, and fertilization includes direct soil applications and foliar biostimulant sprays. Irrigation is applied

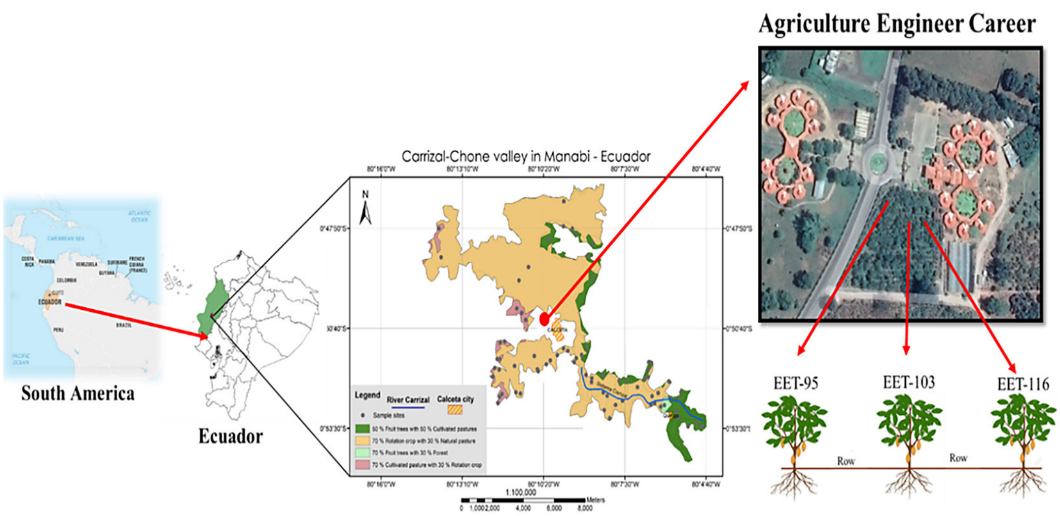

Figure 1. Study area of three species of *Theobroma cacao L* cultivations (varieties EET 95, EET 103 and EET 116) at the ESPAM MFL in Calceta, Manabí, Ecuador

twice a week by sprinklers, except during January, February and March when rain suffices. Crop residues, including husks and prunings, are left in the field.

Although the importance of soil and air temperature in comprehending ecological processes was recognized, the authors' capacity to monitor these factors was constrained by a lack of funding and appropriate equipment. Therefore, by collecting soil samples between July and August 2022, the authores were able to compare soil properties across several cocoa kinds and assess the impact of plants on SOC. Eight sampling locations were chosen (below trees and in-row sections) within each region, and samples were taken down to a depth of 30 cm. This produced 240 soil samples in total (10 at each layer), a method intended to offer a thorough examination of the biological dynamics pertinent to the research aim (Figure 2).

Soil bulk density (BD) was assessed using 45 samples collected from five representative soil pits. For this parameter, the depths were: 0–10, 10–20 and 20–30 cm with three replicates per depth and location (under-tree and in-row). To achieve a stable dry weight, the samples were oven-dried at 105 °C for 72 hours, a 98.2 cm^3 cylinder was used to obtain BD as the dry soil mass divided by the known sampler volume (Hao et al., 2008). From these subsamples, the soil moisture content was determined, while the remaining soil was air-dried in preparation for further analysis.

To determine SOC, the soil samples were sieved in a 2 mm mesh and homogenized. Stoniness, expressed as a mass percentage, was determined gravimetrically. To reach a stable dry weight, the samples were dried at 40 °C for 72 hours in an oven, SOC concentration was quantified using the Walkley wet oxidation method (Walkley, 1947). For SOC stock, the calculus was made according to Equation 1, for each depth interval and the entire soil profile according to the Intergovernmental Panel on Climate Change (IPCC) guidelines.

$$SOC_{stocki} = 10000 SOC_i \times \times BD_i \times d \times (1 - \delta) \quad (1)$$

Equation 2 determines the soil total organic carbon stock by summing the contributions from each layer, while accounting for stoniness. This is achieved by multiplying the organic carbon concentration ($g \cdot g^{-1}$) by the BD of the soil layer ($g \cdot cm^{-3}$) and its thickness (cm), and then subtracting the influence of coarse particles (represented by the fraction δ, ranging from 0 to 1). The summation is performed across all n soil layers, yielding the overall carbon stock Equation 2.

$$SOC_{stock} = \sum_{i=1}^{i=n} SOC_{stocki} \quad (2)$$

To evaluate differences in mean SOC concentration (%) and stock across soil depths and locations within the three cocoa varieties, parametric analysis of variance (ANOVA) was performed using InfoStat 2018. RStudio was operated to conduct correlation analyses and principal component analysis (PCA) to explore the main relationships among the parameters.

RESULTS AND DISCUSSION

Figure 3 shows that the moisture behavior in the soil profile has variability over the position of each of the sampled areas. In the EET 116 cocoa variety, the area with the highest moisture is between 5 and 20 cm, unlike the other two varieties where the highest percentage of moisture is on the surface 0–5 cm and 20–30 cm. From this behavior of moisture on the surface, it can be understood that the effect of crop residues and pruning in

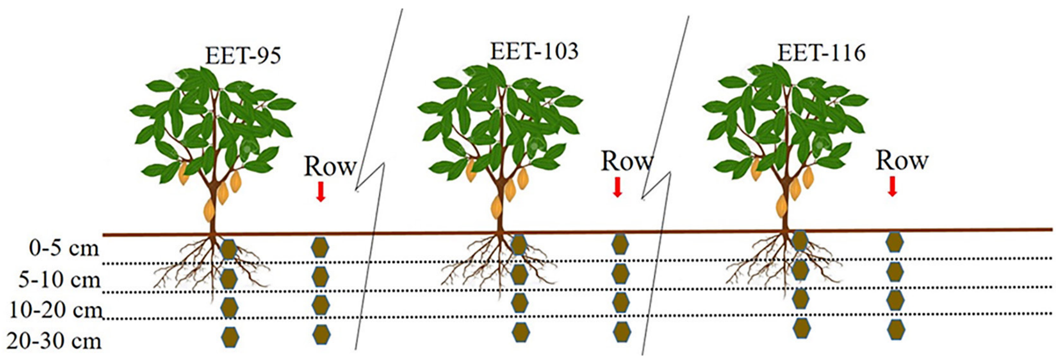

Figure 2. Soil samples were collected at four depths, both under trees and in-rows

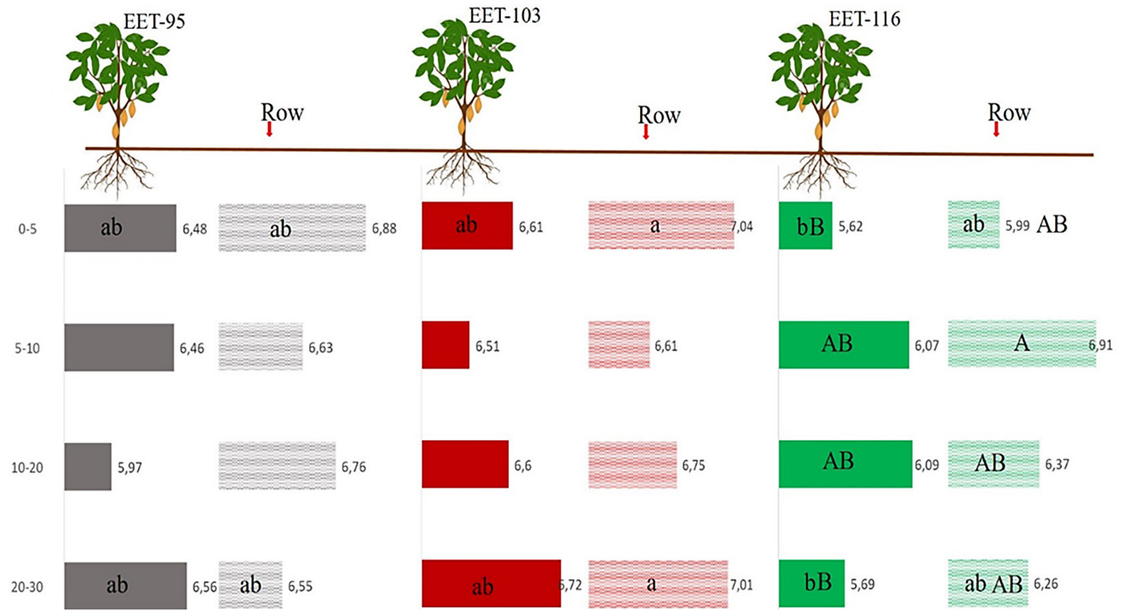

Figure 3. Soil moisture content (%) for EET 95, EET 103 and EET 116. Capital letters (vertical comparisons by location); lowercase letters (horizontal comparisons by depth section. Statistically differences are denoted by different letters ($p > 0.05$)

each of the species directly affects moisture retention on a different scale compared to the samples under the tree. Traditional management in cocoa plantations leads to a lack of residues or vegetation cover under the tree (Quintana et al., 2019; Quizhpe et al., 2017). This management has the opposite effect to what is seen in rows where better moisture retention is noted (Bronick and Lal, 2005; Quintana et al., 2019).

No statistical differences in SOC were observed between cocoa varieties at depths of 5–10 cm and 10–20 cm, either under-trees or in-rows. The absence of significant spatial variation in soil moisture content at these depths suggests a standardized behavior, with textural properties likely accounting for the observed differences, corresponding to a largely sandy loam soil at this depth and the response of moisture to dry season. However, plantations have sprinkler irrigation, maintaining soils with their respective moisture. Nevertheless, in section 20–30 cm a similar behavior was shown as in the first section, EET103 in-row showed the highest percentage of moisture (7.01%), being significantly different only from EET116 below the tree with 5.69%. There were no significant differences at the other sampling locations. The increase in moisture in the last section can be assumed to be due to the slight change in texture that exists in the area (Bronick and Lal, 2005). A small percentage increase in clay content has had a notable effect on moisture in the 20–30 cm section.

Moisture was also analyzed vertically to see the behavior in each of the varieties of cocoa plants. The results showed that only the EET116 variety showed significant differences between the in-row and below the cocoa plant. The 5–10 cm soil section exhibited the highest moisture percentage (6.91%), while the 0–5 cm and 20–30 cm sections showed lower percentages of 5.62% and 5.69%, respectively (Figure 3).

There was no significant variation across soil depths and sampling locations for BD. In average, EET 95 BD reached 1.12 g cm^{-3} beneath trees and 1.15 g cm^{-3} in rows. The corresponding values for EET 116 and EET 103 were 1.22 g cm^{-3}, 1.17 g cm^{-3}, 1.14 g cm^{-3} and 1.18 g cm^{-3}. The standardization of BD values, observed after 15 years of cocoa plantation management, suggests that BD does not significantly influence the comparison of SOC concentrations.

Figure 4 presents a correlation matrix revealing key relationships between soil properties and cocoa trees. As expected, SOC and SOC stock are perfectly correlated (1.00), as they both quantify soil carbon content. Similarly, OM and SOC show a strong positive correlation (0.72), since SOC is a major component of OM. These findings highlight the central role of soil organic carbon in this system.

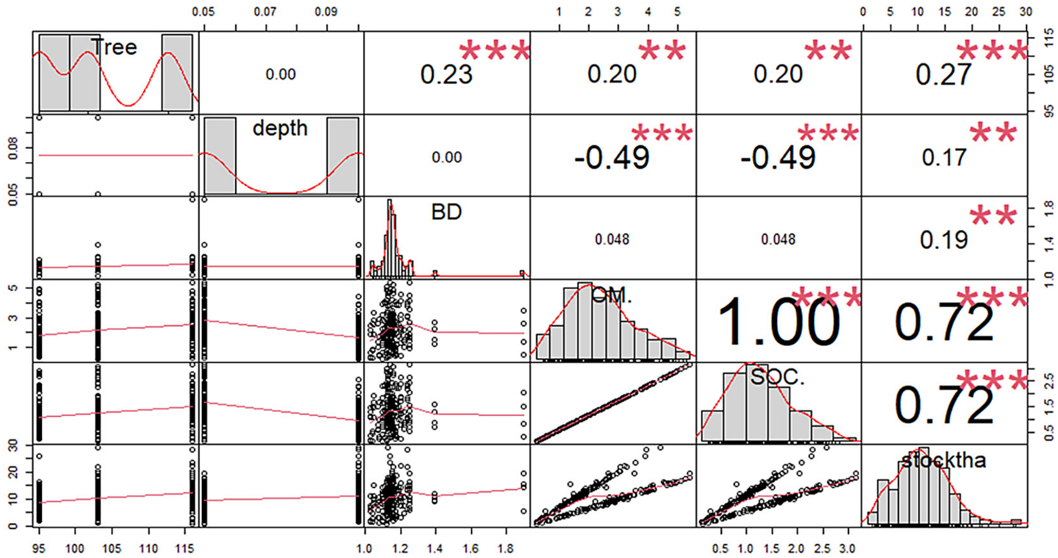

Figure 4. Pearson correlation matrix of soil dynamics: cocoa trees; depth; BD; OM; SOC and SOC stock

Interestingly, cocoa trees show significant positive correlations with SOC (0.20), OM (0.20), SOC stock (0.27), and soil depth (0.23). This suggests that cocoa trees thrive in deeper soils with higher organic matter content, likely due to improved nutrient and water availability. Supporting this idea, BD shows a moderate negative correlation with soil depth (-0.49), indicating that deeper soils tend to be less compacted, promoting root growth and nutrient uptake.

While BD exhibits weak positive correlations with OM and SOC (0.048) and a slightly stronger correlation with SOC stock (0.19), these relationships warrant further investigation. Understanding these complex interactions could help implement informed management practices aimed at optimizing soil conditions for cocoa cultivation. However, it is crucial to remember that correlation does not equal causation, and other factors may be influencing these relationships.

The results presented in Figure 5 indicate that for the first soil layer (0–5 cm depth) SOC concentration average for all cocoa varieties was 2%. However, significant differences in SOC concentration were observed, with the highest concentration (2.51%) in rows of EET 116 and the lowest (1.57%) under EET 103 trees. In contrast, no significant differences were detected in SOC concentration at other locations or depths.

Nevertheless, SOC concentration declined with increasing depth, with average values of 1.3%, 1.0% and 0.89% at each depth, respectively. While (Yang et al., 2022) reported a fourfold difference in SOC concentration between the upper and lower soil layers (0–100 cm), the findings of wthis research (0–30 cm) revealed a nearly threefold difference between the upper and second lower layers, suggesting a potentially steeper decline in SOC concentration at shallower depths.

An important behavior on surface carbon dynamics is that waste management has played an important role in these systems (Goñas et al., 2022). Pruning and the shells of the cocoa pod are accumulated in the rows between the plants. This residue has been incorporated and decomposed during the 15 years of plantation operation. Residue accumulation significantly contributes to the superficial soil layer, which exhibits the highest microbial activity (Yang et al., 2022). The observed SOC concentration patterns are linked to soil moisture dynamics. The greater surface cover in rows, comprising both living and dead vegetation (pruning and harvesting residues), promoted SOC accumulation in the 0–5 cm layer through enhanced decomposition, a process further influenced by environmental factors, including temperature (Solly et al., 2014).

These factors in a tropical climate cause microbial activity to be more active than in the samples located under the tree where no pruning or harvesting residues are found (Jadan et al., 2015; Jobbagy and Jackson, 2000). In contrast, as seen in Figure 5, below the cacao tree the organic carbon concentrations were lower than in the rows. This explains the importance of the incorporation of the organic residues in the rows for their

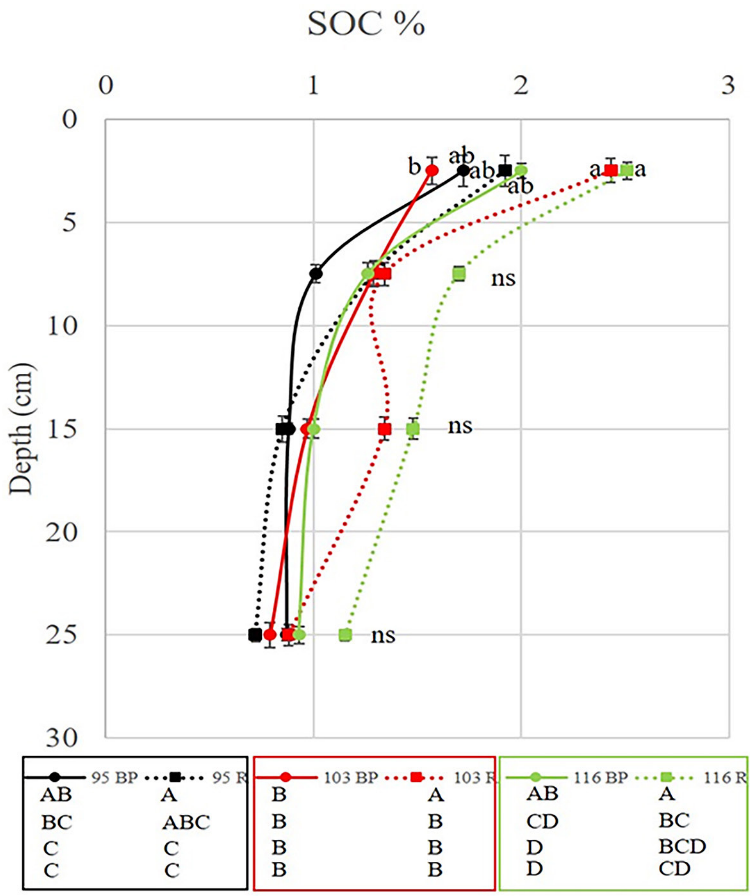

Figure 5. Comparative SOC concentration (%) for EET 95 (black), EET 103 (red) and EET 116 (green). Lowercase letters (horizontal comparisons across soil depth sections); uppercase (vertical comparisons across locations). Statistically differences are denoted by different letters ($p > 0.05$)

incorporation through decomposition in time (Jobbágy and Jackson, 2000). Likewise, the microorganisms at depth do not have this access to nutrients, so the difference in carbon concentration is notable (Fontaine et al., 2007).

In rows, at the 0–5 cm depth, SOC stocks reached 14.44 t ha^{-1} for EET 116 and 14 t ha^{-1} for EET 103. The lowest SOC stock (9.04 t ha^{-1}) was found beneath EET 103 trees. While no significant differences were observed among other sampling locations at subsequent depths, a fluctuating trend in SOC stock was evident: 7.56 t ha^{-1} at 5–10 cm, 12.5 t ha^{-1} at 10–20 cm and 10.2 t ha^{-1} at 20–30 cm. This pattern reflects the influence of root dynamics on SOC distribution across the soil profile.

The elevated SOC stock at the 10–20 cm depth (Figure 6) suggests a significant contribution from root biomass and exudates associated with cocoa tree growth. In long summers with sprinkler irrigation, moisture is retained for much longer under pruning and harvest residues. This has a direct effect on the development of the roots, causing them to drift towards the direction of the rows in search of water. This leads to a greater presence of roots in that section. According to studies, the highest amounts of roots are between 10 to 20 cm deep (Hayes and Seastedt, 1987; Solly et al., 2014).

With the analysis of the entire profile analyzed (0–30 cm), it was observed that the species EET 116 stored 41 t ha^{-1} and 54 t ha^{-1} below tree and row, respectively. For the samples taken from EET 103 in-row reached 47 t ha^{-1}. On the other hand, EET95 and EET 103, with 36 t ha^{-1}, 37 t ha^{-1} and 47 t ha^{-1}, below tree and in-row, respectively, got the lowest values, without significant differences (Figure 7). The fundamental patterns identified in this study could be partially applicable to the areas with similar soil and climate conditions, although more precise understanding would need more research covering a greater range of variables. While this research advances authors' knowledge of cocoa production in tropical regions, it is improbable that particular quantitative results will be directly

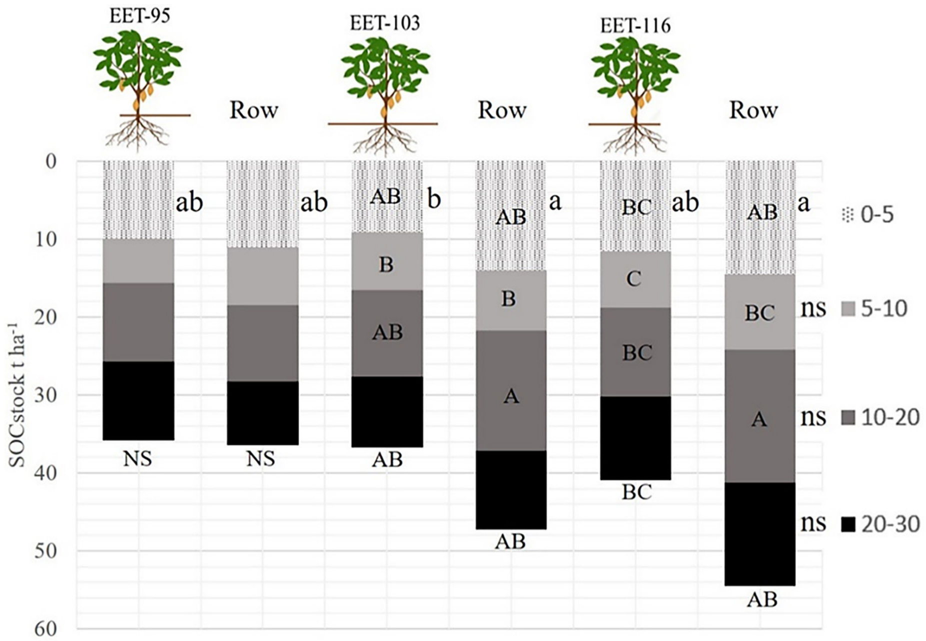

Figure 6. Distribution of SOC stock (t ha⁻¹) across soil depth profiles, comparing EET 95, EET 103 and EET 116 cocoa tree at both beneath-tree and row locations. Letters denote statistically significant differences

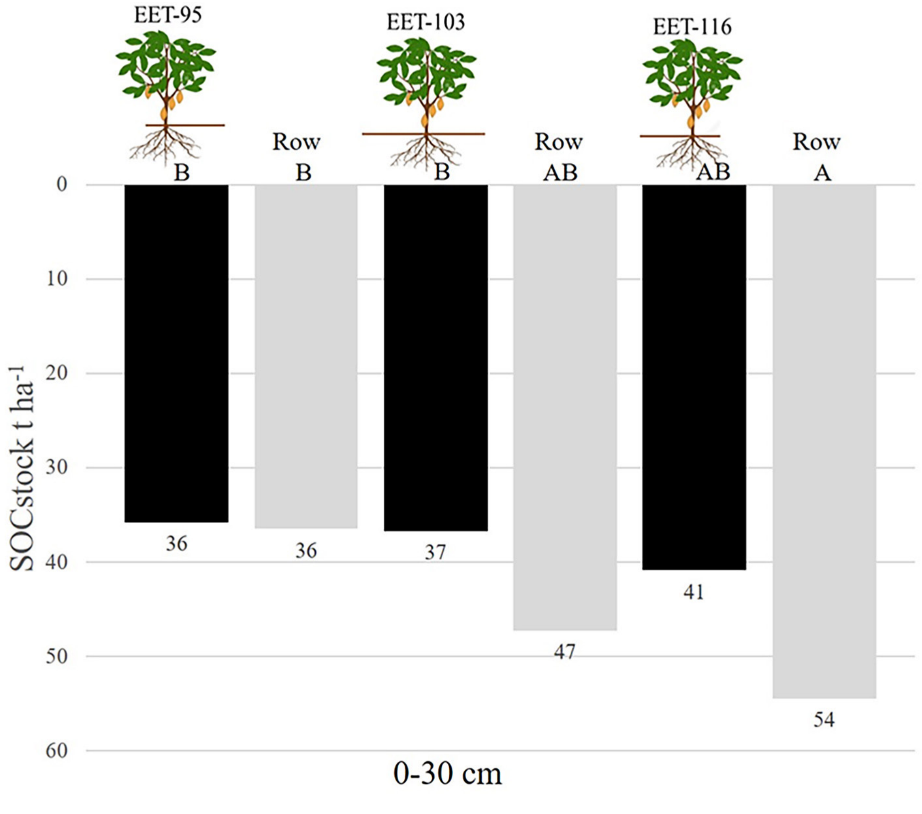

Figure 7. Total accumulated SOC stock (t ha⁻¹) for EET 95, EET 103 and EET 116 cocoa trees at 0–30 cm depths, comparing beneath-tree and row locations. Different letters denote significant differences

interconnected. It is important, because it offers a useful case study that highlights important ecological processes and management factors. Any effort to generalize these findings to other areas or farming systems must thus be carefully considered and backed by specific research.

To justify the selection of principal components in principal component analysis (PCA), the scree plot (Figure 8) was analyzed. The plot reveals a sharp decrease in explained variance after the first component, and an elbow at the second, supporting the retention of only two components to effectively represent the data variability.

The PCA biplot, viewed in the context of the Ecuadorian cocoa study, reveals key insights into the factors driving SOC dynamics (Figure 9). The high variance explained by principal component 1 (PC1, 91.4%) suggests it primarily captures the influence of cocoa variety and location (under tree vs. row), aligning with the significant SOC findings from the study. Principal component 2 (PC2) likely represents secondary variables, such as soil depth, which were not the main focus of the study.

The clear separation between below tree and row samples along PC1 reinforces the observation of location-based differences in SOC made in the study, particularly for the EET103 variety at shallow depths. The influence of SOC stocking (ha) on PC1 might be indirectly linked to location, as row samples likely experience higher stocking due to open conditions. Meanwhile, OM is positively associated with both PC1 and PC2, suggesting its dependence on both location

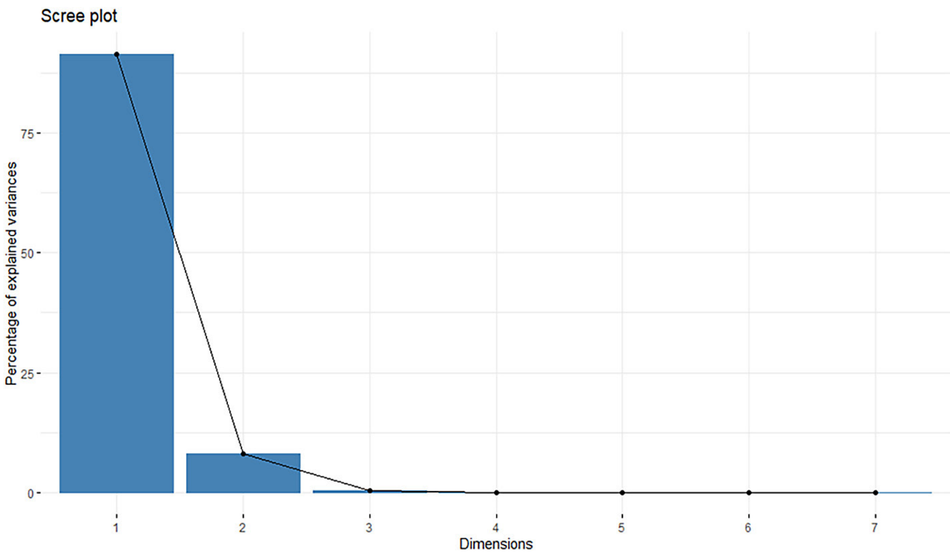

Figure 8. Percentage of explained variances

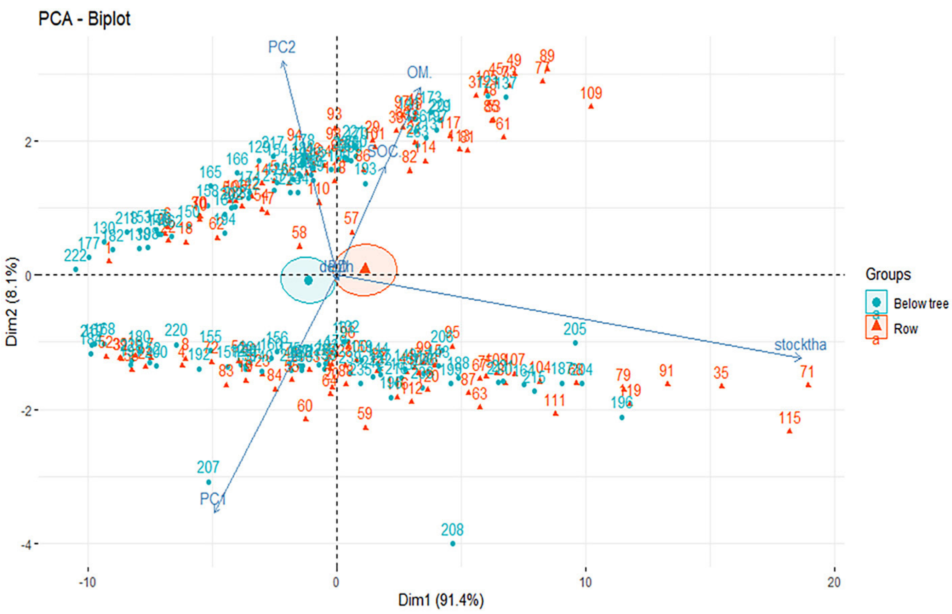

Figure 9. PCA group in two categories below tree and row

and other factors, such as soil depth, which aligns with the study's findings of varying SOC concentrations across depths.

Furthermore, the PCA biplot can be connected to specific study outcomes. The high SOC concentrations in the EET116 variety likely contribute to the separation of some row samples along PC1. The absence of significant SOC differences between beneath-tree and row locations for EET 95 and EET 103 may explain the observed sample overlap.

However, it is important to acknowledge that the PCA biplot does not explicitly show varietal differences. Further analysis focusing on varieties could uncover additional patterns. Moreover, incorporating soil depth and the time factor into the PCA could provide a more comprehensive understanding of SOC dynamics. By integrating the study findings and considering these additional factors, the PCA biplot becomes a powerful tool for visualizing and interpreting the complex interplay of factors influencing SOC in cocoa plantations.

CONCLUSIONS

Under constant climatic circumstances, the EET 116 cocoa variety has greater SOC content and accumulation, which is visibly associated with increased aerial and root biomass growth.

Mature cocoa plantations, especially those over 15 years old, show a substantial ability for carbon sequestration, regardless of the individual cocoa variety produced, with carbon content in the plantation rows equaling or exceeding that of the surrounding soil.

Unlike random sampling techniques, the customized experimental design used in this study highlights the importance of precise management practices in SOC within cocoa agroforestry systems, showing that even traditional methods can have significant positive environmental effects.

Acknowledgements

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