**AF-944**
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RESEARCH ARTICLE Indian Journal of Agricultural Research

# Effect of Biological Amendment on the Agro-morphology of Cocoa Companion Species (*Irvingia gabonensis, Garcinia kola, Tieghemella heckelii* and *Beilschmiedia mannii*) at the Nursery Stage

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**10.18805/IJARe.AF-944**

## ABSTRACT

**Background:** The use of organic amendments plays a crucial role in improving soil fertility and enhancing plant growth in agroforestry systems. This study evaluates the impact of different organic amendments on the morphophysiological parameters of four companion species of cocoa (*Irvingia gabonensis* (Kplé), *Garcinia kola* (Petit cola), *Tieghemella heckelii* (Makoré) and *Beilschmiedia mannii* (Bitéi)) at the juvenile stage. The aim is to determine the most effective amendment for promoting vegetative growth, biomass accumulation and photosynthetic activity.

**Methods:** The research was carried out on the experimental farm of ICRAF located in the La Mé region. Four treatments were set up: T0, which consisted of soil taken from the top 10 cm of an old cocoa plantation; T1, a blend of two-thirds T0 soil and one-third rice husk biochar; T2, which comprised T0 soil enriched with 0.5 kg of composted poultry manure and T3, a balanced mix of T1 and T2.

**Result:** The results highlight that treatments T0 and T2 stand out due to their significant positive effects on overall vegetative growth, biomass production and the enhancement of photosynthetic activity. In particular, the enriched compost (T2) demonstrated remarkable efficacy in improving plant performance, underscoring its potential as a preferred amendment for crop cultivation. Although biochar (T1) showed lower performance on some global indicators, it exhibited specific and noteworthy effects, particularly on root development and leaf chlorophyll content.

**Key words:** Cocoa companion species, Morphophysiological parameters, Organic amendment, Sustainable agriculture.

## INTRODUCTION

Côte d'Ivoire, with an annual cocoa production exceeding 2 million tons, has experienced severe forest degradation due to agricultural expansion (FAO and SEP-REDD+, 2017). The rapid growth of cash crops such as rubber, oil palm and cashew, coupled with population increase, has exacerbated forest cover decline. Overexploitation of natural resources hinders species regeneration and poses a threat to biodiversity and livelihoods (IPBES, 2019). In response, Côte d'Ivoire has adopted a zero-deforestation policy to restore forest cover and degraded agricultural landscapes, integrating locally valuable species in a sustainable management approach (Gaboriau *et al.,* 2016; Katariya *et al.,* 2024).

However, the adoption of agroforestry as an innovative strategy in the context of climate change raises questions about the domestication, reproduction and integration of species in plantations. Agroforestry plants, diverse and meeting various production criteria, face growth difficulties, particularly during their early stages. This constraint limits their availability at the appropriate time for planting, which hinders the adoption of agroforestry systems by producers. Some slow-growing species, such as *Irvingia gabonensis* (Kplé), *Garcinia kola* (Bitter cola), *Tieghemella heckelii* (Makoré) and *Beilschmiedia mannii* (Bitéi), deserve special attention due to their economic potential for farmers. Their fruits are edible (Louppe *et al.,* 2008) and their barks and seeds are used in traditional pharmacopoeia and as animal forage (Adjanohoun and Aké, 1979). However, none of these species have benefited from systematic selection or improvement programs aimed at optimizing their production in terms of quantity or quality. Intercropping systems, developed empirically by farmers, have rarely

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**How to cite this article:** Alban, M'BO.K.A., Soro, K.I., Kouakou, T.d. and Christophe, K. (2025). Effect of Biological Amendment on the Agro-morphology of Cocoa Companion Species (*Irvingia gabonensis, Garcinia kola, Tieghemella heckelii* and *Beilschmiedia mannii*) at the Nursery Stage. *Indian Journal of Agricultural Research*. 1-9. doi: 10.18805/IJARe.AF-944.

**Submitted:** 04-04-2025 **Accepted:** 16-10-2025 **Online:** 04-11-2025

incorporated scientific advancements that allow for the simultaneous optimization of agronomic, environmental and economic benefits. However, in recent years, stakeholders in the cocoa value chain have been paying increasing attention to the improvement of agroforestry systems, particularly through the optimization of the growth of indigenous species of interest, integrated into cocoa and fruit tree plantations (Wato, *et al.*, 2020).

These efforts aim to meet the needs of producers for diversification and resilience in the face of climate change. However, the reproduction and establishment of agroforestry species represent a major challenge for farmers. Indeed, many producers lack knowledge of the reproduction methods and developmental cycles of these species, which limits their adoption and contribution to improving production systems.

Furthermore, the growth of young plants in cocoa plantations is a critical issue, as these forest species take several years to provide sufficient shading to reduce the heat stress experienced by cocoa trees (Sonwa *et al.,* 2014). This shading deficit in the early years compromises the resilience of plantations to drought episodes and high temperatures, thereby reducing orchard productivity. Recent studies have shown that the selection and introduction of agroforestry species adapted to local conditions can improve the sustainability of cocoa plantations while enhancing the ecosystem services provided by these systems (Tscharntke *et al.,* 2011). Moreover, research on crop associations has highlighted that optimal management of shading densities and species can promote the growth of young cocoa trees and mitigate the effects of climate change (Wade *et al.,* 2010). These findings emphasize the importance of an integrated approach to strengthen the productivity and resilience of cocoa plantations in the context of increasing environmental challenges Furthermore, no specific strategy, whether based on organic or inorganic amendments, has been developed to facilitate their growth in nurseries or fields. Yet, these agroforestry species play a key role in the sustainability strategy of agricultural systems, particularly for cocoa production in Côte d'Ivoire. This study thus aims to explore the potential of organic amendments to optimize the domestication of forest fruit species of community interest. To this end, within the scope of this research activity, we hypothesized that organic amendments could improve the juvenile growth of cocoa companion's trees. The objective is to evaluate the effect of these amendments on the agro-morphological parameters of young plants of these species and identify an optimal treatment that promotes their vegetative development and integration into agroforestry systems.

**Study area**

The study was conducted in the Mé region, in Adzopé (Fig 1). The weather conditions during the study period (November 2020 to April 2021) showed an average relative humidity between 45% and 95%. The average rainfall was 79 mm, with an average of 7 rainy days per month. The temperature, on the other hand, ranged on average between 23°C and 35°C.

**Biological material**

- **Plant material**

The plant material used consisted of young plants of *Irvingia gabonensis*, *Garcinia kola*, *Tieghemella heckelii* and *Beilschmiedia mannii* derived from seeds provided by SODEDOR (Fig 2).

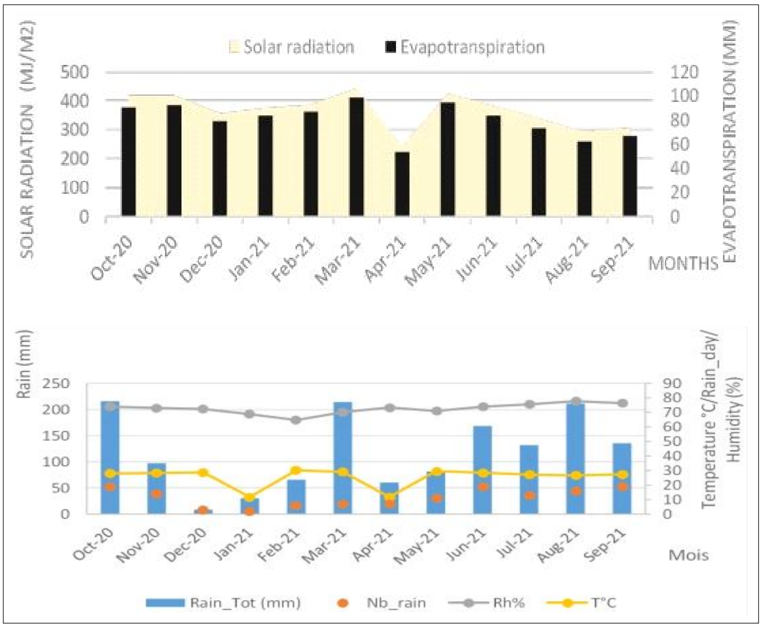


**Fig 1:** Climatic parameters of the study site.

- **Substrates**

Rice husk-based biochar, composted chicken manure (CCM) and soil collected to a depth of 10 cm from an old cocoa plantation (OCP) were used to formulate the different biological amendment treatments (Fig 3).

## MATERIALS AND METHODS

### Experimental design

The experiment followed a split-plot design with two factors and four repetitions. The main factor was the substrate (T0, T1, T2, T3) and the secondary factor was the species (*Beilschmiedia mannii, Tieghemella heckelii, Irvingia gabonensis, Garcinia kola*). It included 64 elementary repetitions (4 substrates × 4 species × 4 blocks), with each treatment containing six plants-96 per species, totaling 384 plants. A schematic representation is shown in Fig 4.

### Application method of treatments

Four (04) treatments were used for the implementation of the experiment (Fig 5).

- **T0:** Soil collected from the first 10 cm of soil from an old cocoa plantation.
- **T1:** 2/3 of T0 mixed with 1/3 rice husk-based biochar.
- **T2:** T0 mixed with ½ kg of composted chicken manure.
- **T3:** Proportional combination of ½ of T1 and T2.

The two substrates were treated with the insecticide-nematicide CARBALM 5 G (active ingredient: Carbofuran 50 g/kg) four days before their application. The applied dose was equivalent to one teaspoon per nursery bag. A two-week acclimatization period was respected to allow the plants to regain their vigor, after which data collection was carried out over a period of three (03) months.


**Fig 2:** Young plant of *Irvingia gabonensis* (A); *Garcinia kola* (B); *T. heckelii* (C) and *Beilschmiedia mannii* (D).

### Study of the effect of biological amendment on the agro-morphology of species

- **Leaf emission and height and radial growth of species under amendment**

Leaf emission was observed monthly for three months, counting new leaves. Height growth was calculated by the difference in initial and final height measurements, while radial growth was determined by the difference in collar diameter over three months. Measurements were taken using a tape measure and caliper.

- **Vegetative vigor index**

The vegetative vigor index of the plants under amendment was determined using the formula by Alexandre (1977).

$$\text{Vigor index (V)} = \frac{\text{Height}}{\text{Collar diameter}}$$

- **Method for evaluating the impact of substrates on leaf area of species**

After three months, leaf area per species and treatment was estimated by measuring leaf length and width using a 50 cm ruler. A non-destructive method was used, allowing assessment of many leaves. The surface area of 30 leaves was traced on graph paper and the product of length and width was correlated with actual leaf surface area through regression analysis to establish a calculation formula.

The formula obtained was:

$$y = 0.7409 \times (L \times l)^{0.9476} \text{ with a determination coefficient } R^2 = 0.9809$$

- **Dry biomass**

The biomass (vegetative and root) and water content was conducted destructively after uprooting (at the end of the experiment). This involved measuring the fresh weight of different plant organs (leaves, stem, roots) and their dry weight after being oven-dried at 60°C for 48 hours (Fig 6). The plant samples were previously cut before drying.

### Determination of chlorophyll content in species under biological amendment

Chlorophyll content was measured monthly using the SPAD-502_Plus meter, The SPAD value, proportional to chlorophyll content, was recorded for both young and old leaves at three parts (proximal, central, distal) to estimate chlorophyll levels (Fig 6).

### Statistical analyses

Data were organized into Excel files. The Excel files were validated with SAS Insight and outlier data were corrected. The general linear model (GLM) of variance analysis in SAS software (9.4) was used to determine the amendment effect on the species, using the LSD test at a 5% significance level.

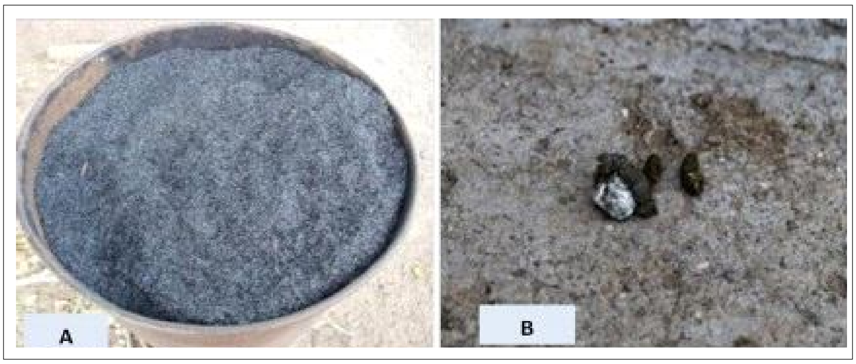


**Fig 3:** Rice husk-based biochar (A), Composted chicken manure (B).

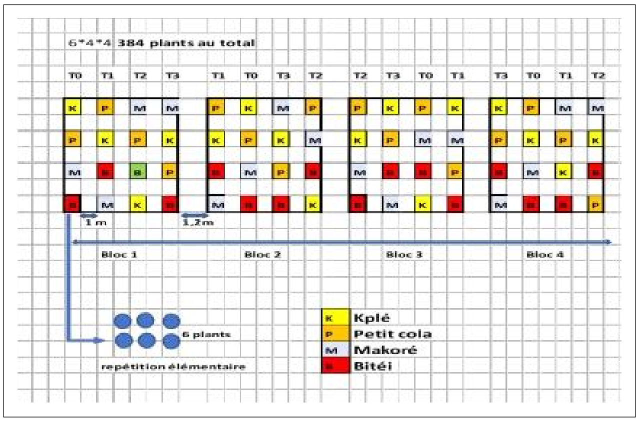


**Fig 4:** schematic design of experimental.

**Fig 6:** Measurement of the width (A) and chlorophyll content of a leaf (B).


**Fig 5:** Treatments of the experiment, Soil collected from old cocoa plantation (A); Rice husk-based biochar + Soil collected from old cocoa plantation (B); Composted chicken manure + Soil collected from old cocoa plantation (C); ½ of B and C (D).

## RESULTS AND DISCUSSION

### Effects of organic amendments on growth and development of species

The effects of organic amendments on the growth and development of the species showed varying results (Table 1). In Bitéi plants, no significant differences were observed for height ($p \geq 0.0505$), vigor index ($p \geq 0.3149$) and root water content ($p \geq 0.7072$). However, significant effects were noted for radial growth ($p \leq 0.036$), number of leaves ($p \leq 0.0136$), water content in aerial parts ($p \leq 0.0345$) and root ($p \leq 0.0259$) and aerial ($p \leq 0.0114$) biomass. For Kplé plants, only the number of leaves produced showed significant differences ($p \leq 0.0416$). In Makoré plants, amendments had no impact on height ($p \geq 0.9726$), vigor index ($p \geq 0.5111$), number of leaves ($p \geq 0.391$), or root water content ($p \geq 0.3391$), but significant differences were found for circumference ($p \leq 0.0489$), water content in aerial parts ($p \leq 0.0288$) and root ($p \leq 0.0029$) and aerial ($p \leq 0.011$) biomass. In Petit Cola, only the number of leaves produced was affected by the amendments ($p \leq 0.0031$). Radial growth, measured by circumference, showed notable variations. For Bitéi, it ranged from 0.11667 cm (T2) to 0.05833 cm (T0), while for Makoré, it ranged from 0.075 cm (T3) to 0.125 cm (T0). The number of leaves produced varied significantly, with maximum values of 6.379 for Bitéi, 4.7917 for Kplé and

4.9167 for Petit Cola under treatment, particularly the control (T0). Water content in aerial parts varied, with Bitéi ranging from 35.239% (T2) to 46.476% (T0) and Makoré from 37.299% (T0) to 48.655% (T2). Root biomass was lowest for Bitéi under T2 (0.3342 g) and highest for T0 (1.4613 g). For Makoré, root biomass ranged from 0.4045 g (T0) to 0.8123 g (T1). Aerial biomass showed significant differences, with Bitéi's maximum value under T2 (3.8559 g) and the minimum under T0 (1.5071 g). Makoré's maximum aerial biomass (2.7299 g) was found with T0.

**Variation in leaf area of species under each treatment**

Statistical analysis revealed significant effects of organic amendments on the foliar parameters of the studied species. In Bitéi, leaf length ($p \leq 0.0044$), width ($p \leq 0.0015$) and area ($p \leq 0.004$) varied significantly under different treatments. Similarly, in Kplé, significant differences were observed in leaf length ($p \leq 0.0025$), width ($p \leq 0.0041$) and area ($p \leq 0.0007$). For Makoré, treatments had no significant effect on leaf width ($p \geq 0.6601$) or area ($p \geq 0.3839$), but leaf length showed significant variation ($p \leq 0.0428$). In Petit Cola, although leaf area was unaffected ($p \geq 0.842$), leaf length ($p \leq 0.0129$) and width ($p \leq 0.0143$) were significantly impacted. Leaf length ranged from 8.594 cm (T2) to 12.754 cm (T0) in Bitéi, with Kplé showing the longest leaves (21.896 cm) under T1. Makoré and Petit Cola had the longest leaves under T1, measuring 13.4917 cm and 17.488 cm, respectively. Leaf width in Bitéi varied from 2.5854 cm (T2) to 4.0833 cm (T0), while the widest leaves in Kplé (7.614 cm) and Petit Cola (6.0458 cm) were observed under T1. The largest leaf area in Bitéi (373.56 $cm^2$) and Kplé (1118 $cm^2$) was seen under T0 and T1, respectively. Treatment T2 had the least impact across species (Table 2).

**Effect of organic amendment on the variation of chlorophyll content**

Organic amendments had a variable impact on the chlorophyll content of young and old leaves across species during the three-month trial. In Bitéi, no significant difference in chlorophyll content was observed in young leaves during the first month ($p \geq 0.0511$), but significant variations were noted in the second ($p \leq 0.0066$) and third months ($p \leq 0.0339$). For old leaves, significant differences were observed in all three months ($p \leq 0.0229$; $p \leq 0.0099$; $p \leq 0.0052$), showing a marked response to the amendments. In Kplé, there was no significant change in the chlorophyll content of young leaves throughout the trial ($p \geq 0.1964$; $p \geq 0.7053$; $p \geq 0.2389$). However, old leaves showed significant differences in each month ($p \leq 0.0265$; $p \leq 0.0331$; $p \leq 0.0435$). The young leaves of Makoré exhibited significant changes in chlorophyll content during the first and third months ($p \leq 0.0245$; $p \leq 0.0384$), with no significant variation in the second month ($p \geq 0.0624$). Old leaves of Makoré showed significant variation only in the first month ($p \leq 0.0372$), with no changes in the subsequent months ($p \geq 0.2214$; $p \geq 0.6253$). For Petit Cola, significant differences in chlorophyll content in young leaves were only observed in the third month ($p \leq 0.0454$), while old leaves showed significant variation only in the third month. The greatest variation in young leaves occurred in Bitéi during the second month, where T2 (7.658%) had a marked difference compared to T0 (20.204%). The largest variation in old leaves of Bitéi was seen in the third month, between T1 (40.333%) and T0 (56.025%). In Kplé, the largest variation was seen in the second month with T2 (36.33%) compared to T0 (48.292%). Treatments T1 and T2 had the greatest impact on chlorophyll content, especially in young leaves of Bitéi and Makoré (Table 3).

This study assessed the influence of biological amendments on the agro-morphological characteristics of four companion species of the cacao tree at the juvenile stage: *Irvingia gabonensis* (Kplé), *Garcinia kola* (Petit cola), *Tieghemella heckelii* (Makoré) and *Beilschmiedia mannii* (Bitéi). Four treatments were applied: T0 (soil from an aged cacao plantation), T1 (T0 soil with one-third biochar from rice husks), T2 (T0 soil enriched with composted poultry manure) and T3 (a combination of T1 and T2). The results revealed significant variations in the effects of organic amendments on growth and development, reflecting complex interactions between morpho-physiological traits and soil amendments. In *Beilschmiedia mannii* (Bitéi), *Tieghemella heckelii* (Makoré), *Irvingia gabonensis* (Kplé) and *Garcinia kola* (Petit cola), certain treatments significantly influenced radial growth and leaf production. The control (T0) favored these parameters, possibly due to greater availability of essential nutrients in unamended soils, which may contain natural organic reserves. These findings align with Eghball and Power (1999) and Truong *et al.*, (2024), who demonstrated that organic amendments enhance nutrient availability and promote plant growth. Similarly, Garnier *et al.* (2004) reported that organic nutrients directly influence leaf formation by improving photosynthesis and biomass production. While T0 and T2 positively influenced leaf emission and aerial biomass production, the T1 treatment (biochar and old cacao plantation soil) yielded the best results for root biomass. The aerated structure of rice husk biochar likely enhanced root growth and nutrient uptake, supporting findings from Lehmann and Joseph (2009), who reported biochar improves nutrient retention and soil structure. Water content in aerial and root parts varied among species, with *Tieghemella heckelii* (Makoré) showing high water content under T2, suggesting improved soil water retention. These results align with Rawls *et al.* (2003), who found that organic amendments enhance soil water-holding capacity and plant hydration. Overall, T0 outperformed other treatments in radial growth, leaf production and root biomass. However, T2 contributed to increased water content and aerial biomass, emphasizing species-specific responses to amendments. Ouedraogo *et al.* (2001) highlighted that plant responses depend on the physicochemical properties of inputs and species-specific requirements. The moisture content of composted poultry manure (T2) likely improved

**Table 1:** Vegetative vigor, number of leaves produced, water content and dry biomass produced under the effect of substrates.

| Species | Morphologgical parameters | Treatments | | | | Average | LSD ($p \leq 0.05$) |
|---|---|---|---|---|---|---|---|
| | | T0 | T1 | T2 | T3 | | |
| Bitéi | Height | 3.8542a | 3.625a | 2.9583a | 3.00a | 3.35 | 0.0505 |
| | Circumference | 0.05833b | 0.07417ab | 0.11667a | 0.05833b | 0.07 | 0.036 |
| | Vigor index | 0.02958a | 0.03583a | 0.02708a | 0.0125a | 0.02 | 0.3149 |
| | Number of leaves | 6.379a | 2.375b | 1.375b | 2.042b | 3.04 | 0.0136 |
| | Root water content | 40.171a | 45.652a | 47.429a | 48.638a | 45.47 | 0.7072 |
| | Aboveground water content | 46.476a | 40.332ab | 35.239b | 45.591ab | 41.9 | 0.0345 |
| | Root biomass | 1.4613a | 0.4839ab | 0.3342b | 0.6054ab | 0.72 | 0.0259 |
| | Aboveground biomass | 1.5071b | 2.7319ab | 3.8559a | 2.0595b | 2.53 | 0.0114 |
| Kplé | Height | 11.583a | 8.688a | 9.417a | 10.146a | 9.95 | 0.57 |
| | Circumference | 0.06667a | 0.06667a | 0.06250a | 0,05833a | 0.06 | 0.0881 |
| | Vigor index | 0.21000a | 0.07958a | 0.05708a | 0.10458a | 0.11 | 0.0581 |
| | Number of leaves | 4.7917a | 2.1250b | 2.2083b | 1.9583b | 2.77 | 0.0416 |
| | Root water content | 38.983a | 37.783a | 44.237a | 36.709a | 39.42 | 0.4173 |
| | Aboveground water content | 41.288a | 38.568a | 37.733a | 37.633a | 38.8 | 0.055 |
| | Root biomass | 0.42618a | 0.47608a | 0.38242a | 0.49400a | 0.44 | 0.881 |
| | Aboveground biomass | 2.8566a | 2.3426a | 3.4523a | 2.5231a | 2.79 | 0.6113 |
| Makoré | Height | 4.646a | 4.958a | 4.813a | 6.646a | 5.26 | 0.9726 |
| | Circumference | 0.12500a | 0.08750ab | 0.08750ab | 0.07500b | 0.09 | 0.0489 |
| | Vigor index | 0.11833a | 0.065a | 0.05708a | 0.07125a | 0.07 | 0.5111 |
| | Number of leaves | 9.958a | 10.167a | 9.500a | 8.542a | 9.54 | 0.391 |
| | Root water content | 42.702a | 38.491a | 44.065a | 41.00a | 41.56 | 0.3391 |
| | Aboveground water content | 37.299b | 43.973ab | 48.655a | 46.978a | 44.22 | 0.0288 |
| | Root biomass | 0.4045b | 0.8123a | 0.7001ab | 0.7983a | 0.67 | 0.0029 |
| | Aboveground biomass | 2.7299a | 1.7231b | 1.9596b | 1.9659b | 2.09 | 0.011 |
| Petit cola | Height | 6.896a | 7.354a | 7.50a | 5.521a | 6.81 | 0.088 |
| | Circumference | 0.06667a | 0.07083a | 0.05833a | 0.0625a | 0.06 | 0.8465 |
| | Vigor index | 0.0525a | 0.06875a | 0.10292a | 0.03958a | 0.06 | 0.0715 |
| | Number of leaves | 4.9167a | 4.5417ab | 3.4583ab | 2.9167b | 3.95 | 0.0031 |
| | Root water content | 40.601a | 35.564a | 41.117a | 42.650a | 39.98 | 0.0539 |
| | Aboveground water content | 42.220a | 35.945a | 39.087a | 39.311a | 39.14 | 0.252 |
| | Root biomass | 0.5251a | 0.5777a | 0.4873a | 0.4194a | 0,5 | 0.0718 |
| | Aboveground biomass | 2.4288a | 3.2767a | 3.6988a | 3.1738a | 3.14 | 0.1631 |

On the rows, means followed by the same letter are statistically identical at the 5% threshold (LSD). LSD ($p \leq 0.05$): Probability LSD.

**Table 2:** Leaf area under the effect of biological amendment.

| Species | Paramters | Traitements | | | | Average | LSD ($p \leq 0.05$) |
|---|---|---|---|---|---|---|---|
| | | T0 | T1 | T2 | T3 | | |
| Bitéi | Length | 12.754a | 11.492ab | 8.594b | 11.040ab | 10.97 | 0.0044 |
| | Width | 4.0833a | 3.5875a | 2.5854b | 3.4646ab | 3,43 | 0.0015 |
| | Leaf area | 373.56a | 340.27ab | 223.16b | 325.79ab | 315.69 | 0.004 |
| Kplé | Length | 20.269ab | 21.896a | 18.188b | 21.333a | 20.42 | 0.0025 |
| | Width | 7.0354ab | 7.6146a | 6.5125b | 7.5583a | 7.18 | 0.0041 |
| | Leaf area | 975.1ab | 1118.0a | 886.2b | 1101.3ab | 1020.15 | 0.0007 |
| Makoré | Length | 12.6396ab | 13.4917a | 12.4688b | 12.6458ab | 12.81 | 0.0428 |
| | Width | 5.2896a | 5.5417a | 5.4333a | 5.3104a | 5.39 | 0.6601 |
| | Leaf area | 440.73a | 499.46a | 451.98a | 448.21a | 460.09 | 0.3839 |
| Petit cola | Length | 16.885ab | 17.488a | 13.981b | 16.442ab | 16.19 | 0.0129 |
| | Width | 5.9521ab | 6.0458a | 4.7958b | 5.7188ab | 7.18 | 0.0143 |
| | Leaf area | 705.76a | 729.32a | 565.91a | 691.33a | 673.07 | 0.842 |

In the rows, means followed by the same letter are statistically identical at the 5% threshold (LSD). LSD ($p \leq 0.05$): LSD probability.

**Table 3:** Variation in chlorophyll content under the effect of organic amendments.

| Species | Parameters | Months | Treatments | | | | Average | LSD |
|---|---|---|---|---|---|---|---|---|
| | | | T0 | T1 | T2 | T3 | | ($p \leq 0.05$) |
| Bitéi | JFM | 1 | 26.913a | 21.800a | 18.300a | 22.764a | 22.44 | 0.0511 |
| | | 2 | 20.204a | 12.971ab | 7.658b | 15.400a | 14.07 | 0.0066 |
| | | 3 | 11.758ab | 15.179a | 9.692ab | 6.364b | 10.7 | 0.0339 |
| | VFM | 1 | 52.854b | 52.754b | 58.425a | 53.392b | 54.34 | 0.0229 |
| | | 2 | 57.017a | 47.850ab | 45.625b | 52.652ab | 50.8 | 0.0099 |
| | | 3 | 56.025a | 40.333b | 41.296b | 40.448b | 44.48 | 0.0052 |
| Kplé | JFM | 1 | 23.538a | 16.158a | 16.213a | 18.113a | 18.5 | 0.1964 |
| | | 2 | 17.963a | 24.292a | 23.100a | 19.835a | 21.31 | 0.7053 |
| | | 3 | 10.088a | 8.146a | 11.558a | 9.513a | 9.82 | 0.2389 |
| | VFM | 1 | 47.371a | 42.500b | 45.242ab | 45.474ab | 45.14 | 0.0265 |
| | | 2 | 48.292a | 43.058ab | 36.338b | 41.613ab | 42.33 | 0.0331 |
| | | 3 | 53.979a | 52.004a | 54.254a | 44.909b | 51.35 | 0.0435 |
| Makoré | JFM | 1 | 39.050ab | 34.542b | 41.642a | 35.313ab | 37.63 | 0.0245 |
| | | 2 | 41.496a | 40.904a | 41.504a | 39.404a | 40.82 | 0.0624 |
| | | 3 | 30.504b | 29.562b | 38.221a | 30.329b | 32.14 | 0.0384 |
| | VFM | 1 | 66.929ab | 63.400b | 68.700a | 65.658ab | 66.17 | 0.0372 |
| | | 2 | 69.733a | 66.192a | 68.950a | 67.283a | 68.03 | 0.2214 |
| | | 3 | 70.046a | 66.821a | 69.858a | 68.500a | 68.8 | 0.6253 |
| Petit cola | JFM | 1 | 42.600a | 35.367ab | 31.721ab | 29.804b | 34.87 | 0.0176 |
| | | 2 | 27.033a | 21.763a | 24.204a | 24.129a | 24.28 | 0.7474 |
| | | 3 | 18.225a | 21.908a | 14.317a | 17.654a | 18.02 | 0.7438 |
| | VFM | 1 | 65.404a | 64.933a | 64.563a | 65.663a | 65.14 | 0.3233 |
| | | 2 | 69.425a | 71.171a | 64.467a | 67.208a | 68.06 | 0.1219 |
| | | 3 | 68.125ab | 69.350a | 62.625ab | 57.679b | 64.44 | 0.0454 |

On the lines, the means followed by the same letter are statistically identical at the 5% threshold (LSD). LSD ($p \leq 0.05$): LSD Probability. JFM: Young leaf mean; VFM: Old leaf mean.

substrate water retention, contributing to optimal plant development (Dróżdż, 2023). Water availability is crucial for photosynthesis, nutrient transport and cell division. Blum (1989) also demonstrated that high water content enhances osmotic adjustment, improving nutrient uptake and root development. Leaf length varied by treatment. In Bitéi and Kplé, the longest leaves were observed under T0 and T1, while T2 resulted in shorter leaves, possibly due to suboptimal nutrient balance. Agegnehu *et al.* (2016) reported similar results in maize, where nitrogen-rich organic compost increased leaf length. Leaf width varied significantly in Bitéi, Kplé and Petit Cola, reaching maximum values under T1, highlighting biochar's positive impact. Leaf area was significantly larger in Bitéi and Kplé under T0 and T1, improving photosynthetic capacity. As Ayoola and Makinde (2012) demonstrated, a larger leaf area enhances carbon assimilation and promotes growth. High leaf area values were observed in T1, T3 and T0, correlating with increased dry biomass. Ramamoorthy *et al.* (2024) and Rutigliano *et al.* (2014) suggested that biochar improves soil properties and microbial diversity, enhancing nutrient availability and photosynthesis. Organic amendments, particularly enriched compost (T2), strengthened photosynthetic capacity and productivity in woody species like Bitéi and Makoré. However, Ahmed *et al.* (2019) recommended further research to refine dosage and formulations based on species-specific needs. Regarding chlorophyll content, biochar significantly improved this parameter in Bitéi, Kplé and Petit Cola, likely due to its high carbon content promoting photosynthesis. These findings align with El Kadiri Boutchich *et al.* (2016) who showed that organic fertilization and biochar application enhance chlorophyll content. Nitrogen, a key chlorophyll component, plays a crucial role in regulating photosynthesis and plant growth. This study highlights the positive impact of organic amendments, particularly biochar, on the growth and biomass production of cacao companion species. The results emphasize the importance of nitrogen and carbon in improving vegetative vigor, photosynthesis and biomass accumulation. T0 and T2 exhibited the most favorable effects on vegetative development, while biochar (T1) promoted root growth and increased chlorophyll content. These findings suggest that strategic combinations of organic amendments can optimize growth in cacao agroforestry systems, enhancing sustainability and productivity.

## CONCLUSION

This study demonstrated that organic amendments positively influence the agro-morphological parameters of four cocoa companion species (Irvingia gabonensis, Garcinia kola, Tieghemella heckelii, Beilschmiedia mannii) at the juvenile stage. Biochar (Bc) and composted poultry manure (FPC) improved dry biomass, leaf count, leaf surface area and chlorophyll content, though their effects on vegetative vigor varied among species. FPC notably enhanced root water content and plant biomass, particularly in Tieghemella heckelii and Beilschmiedia mannii, while biochar increased chlorophyll content and leaf surface area, supporting photosynthesis. The control treatment (SVC) showed competitive results, especially in radial growth and leaf number, whereas the combined treatment (T3: Bc + FPC) was less effective, indicating potential interactions that limit benefits. These findings underscore the need for species-specific amendment selection to optimize growth. Biochar and composted poultry manure emerged as valuable substrates for promoting agroforestry and mitigating forest degradation linked to cocoa farming. However, long-term studies are necessary to refine amendment strategies and better understand environmental interactions affecting companion tree development.

## ACKNOWLEDGEMENT

The authors are grateful to the staff for their technical support. The authors acknowledge the support of the German Government with funding through the Green Innovation Project and ICRAF-The World Agroforestry, Côte d'Ivoire; Deutsche Gesellschaft für International Zusammenarbeit (GIZ).

### Disclaimers

The views and conclusions expressed in this article are solely those of the authors and do not necessarily represent the views of their affiliated institutions. The authors are responsible for the accuracy and completeness of the information provided, but do not accept any liability for any direct or indirect losses resulting from the use of this content.

### Informed consent

All animal procedures for experiments were approved by the Committee of Experimental Animal care and handling techniques were approved by the University of Animal Care Committee.

### Conflict of interest

The authors declare that there are no conflicts of interest regarding the publication of this article. No funding or sponsorship influenced the design of the study, data collection, analysis, decision to publish or preparation of the manuscript.

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