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Research Article

Effects of Cocoa Pod Husk Biochar on Growth of Cocoa Seedlings in Southeast Sulawesi-Indonesia

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Abstract

Background and Objective: High quality cocoa seedlings can be produced by improving soil fertility of the plant growing media through application of biochar during the nursery growing period. This study aimed to evaluate the effect of cocoa pod husk (CPH) biochar on soil temperature, soil moisture and growth attributes of cocoa seedlings. **Materials and Methods:** The experiment was conducted in the glasshouse of Agricultural Faculty, Halu Oleo University, Kendari, Southeast Sulawesi, Indonesia. The experimental design was a randomized block design with seven levels of cocoa pod husk (CPH) biochar (i.e., without biochar (control), 3 g of CPH biochar kg⁻¹ soil, 6 g of CPH biochar kg⁻¹ soil, 9 g of CPH biochar kg⁻¹ soil, 12 g of CPH biochar kg⁻¹ soil, 15 g of CPH biochar kg⁻¹ soil and 18 g of CPH biochar kg⁻¹ soil in 3 replications. Data were analyzed by using two way analysis of variance (ANOVA) followed by Duncan's multiple range test with an error rate of 5% (p<0.05). **Results:** The CPH biochar significantly increased soil temperature, soil moisture, soil fertility and cocoa seedling growth. The application of CPH biochar kg⁻¹ increased soil pH, soil-C, P and CEC (cation exchange capacity). Cocoa seedling growth was significantly improved by CPH biochar and a rate of 9 g CPH biochar kg⁻¹ soil showed the best results in cocoa seedlings in terms of increased seedling height, number of leaves, leaf area and shoot dry weight by 20.99, 26.62, 75.63 and 78.36%, respectively, as compared to control. **Conclusion:** Therefore, CPH biochar has the potential to improve soil temperature, soil moisture, soil pH, soil organic-C, P, CEC and cocoa seedling growth, however, the amount of CPH biochar material applied should be considered.

Key words: Biochar, seedling growth, CEC, CPH biochar, cocoa seedling growth

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Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Improving quality of cocoa seedlings is a major target to increase production, to meet the domestic and export demand since Southeast Sulawesi as a national area center of cocoa development in Indonesia. The key factor of cocoa development is namely the use of high quality of cocoa seedlings. Seedlings used for replanting cocoa projects are produced in nurseries mainly by using polybag as containers. Currently, in most developing countries, polybags are commonly used as containers in the nursery phase. The components of growing media used in containerized nurseries can be an important factor in growth and development of seedlings. A good quality growing medium can improve seedling growth and produce higher quality seedlings. High quality cocoa seedlings can be produced by improving soil fertility of the plant growing media through application of biochar during the nursery growing period. Even, biochar derived from the cocoa pod husk (CPH) can be as a sustainable approach improving the growing media in the nursery phase^{1,2}.

Biochar production has a great potential, mainly as a soil conditioner^{3,4,5}, or as a component of seedling substrates⁶⁻¹⁰. Some studies showed strong positive growth responses of seedlings to the use of biochar in the nursery phase¹⁰⁻¹⁴. CPH biochar has a complex chemical composition which has potential as plant growth media for cocoa seedlings^{1,15}.

Biochar application to soils has been shown to improve soil properties^{16,17}. Biochar application possibly affects soil water relations and rooting patterns¹⁸. Biochar may alter soil physical properties, such as it increases aeration and water holding capacity of certain soils^{19,20}, increases the water holding capacity and water availability to plants²¹. Biochar can increase water holding capacity with additions of only 5% biochar into the mass of top soils²². This occurs because not only the density of biochar is lower than that of some minerals but also biochar contains macro and micropores²³, which can hold air or water, greatly reducing the bulk density of the entire biochar particles¹⁸ and thus a large capacity to hold water at field capacity²⁴. Further, an increase in the soil available water content is possible by their amendment with hydrogels, polymers that can hold water at hundreds of times of their own weight²⁵. Some studies showed that amendment of coarse-texture soils with biochar increases their capacity to absorb water²⁶⁻²⁸. The ability to absorb water is important, where excessive drainage leads to loss of soil nutrients. Simply holding more water, however, does not necessarily result in more water being available to plants, particularly if the water is held so tightly that the plants have no access to it and hence

affects their growth. The other studies found a significant influence of biochar addition on water retention and the water retention capacity differed among soils depending on type of applied biochar^{29,30}. Further, an increasing wood biochar application rate linearly increased with the available soil moisture in a sandy soil³¹. Biochar has been used as soil amendment to improve soil structures and fertility qualities^{32,24}. Biochar amendments enhanced nutrient uptake³³, improved soil fertility^{34,35,36} and improved agronomic performance^{37,38}. Biochar addition increase soil pH^{39,40}, improve soil health and improve plant growth due to higher availability of nutrients⁴¹. Therefore, addition of biochar increases soil field capacity, especially at high application rates and resulting in increased plant growth^{42,43}. Further, the addition of low amounts of biochar had limited or less visible effects on various plants species whereas higher doses of biochar appeared to limit plant growth^{44,45}.

Thus, the amount of CPH biochar should be considered for improving soil water relations, soil fertility and cocoa seedling growth. The aim of the study was to investigate the effects of CPH biochar application on soil temperature, soil moisture, pH, soil-C, P, CEC and cocoa seedling growth attributes.

MATERIALS AND METHODS

The experiment was conducted from May-August, 2016 in the glasshouse of Agricultural Faculty, Halu Oleo University, Kendari, Southeast Sulawesi, Indonesia. The location of the experiment site was at the geographical coordinate 122°31'32.89" E; 04°00'33.90" S and the altitude at 25 m from sea level. The experimental design was a randomized block design with seven levels of cocoa pod husk (CPH) biochar (i.e., without biochar (control), 3 g of CPH biochar kg⁻¹ soil, 6 g of CPH biochar kg⁻¹ soil, 9 g of CPH biochar kg⁻¹ soil, 12 g of CPH biochar kg⁻¹ soil, 15 g of CPH biochar kg⁻¹ soil and 18 g of CPH biochar kg⁻¹ soil in 3 replications. The mean daily temperatures in the glasshouse varied from 22-30°C and the relative humidity ranged from 68-88%.

Biochar was produced from cocoa pod husk (CPH) by using a drum kiln, in which carbonization was done within 4-6 h⁴⁶. The hot biochar produced after pyrolysis was quenched with distilled water, collected, air-dried, crushed and sieved through a 2 mm sieve before being used. The soil for trial was collected from the sandy loam (76% sand, 21% silt and 11% clay) of the experimental farm of Agricultural Faculty, Halu Oleo University.

Cocoa seedlings were raised on germination media for 14 days and each seedling was then transplanted into a

polybag of 25 × 30 cm size which had been filled with seedling media of 5 kg dry soil mixed with a treatment-based rate of biochar from cocoa pod husk at planting space of 20 × 20 cm. One seedling was raised in one polybag. The amount of water applied was 200 mL/plant for 3 months under glasshouse conditions with every 2 days of water frequency. Seedling growth attributes, soil moisture and soil temperature were monitored for 3 consecutive months. The data collected for seedling growth attributed included: Seedling height, number of leaves, leaf area, root dry weight and shoot dry weight. The soil moisture was monitored with a soil moisture meter (model: PMS-714), while soil temperature with soil thermometer at the depth of 12 cm below the surface every 2 days at 17.00 pm (before being irrigated). Seedling height, number of leaves and leaf area were measured 90 days after planting. Thereafter, seedlings were removed from the nursery and sent to the laboratory, in order to obtain their dry weight of root and shoot. Dry weight was obtained after drying the material at 85 °C for 48 h. At 90 days after planting, three soil samples were taken from each polybag at random positions, mixed, carried to the Laboratory to measure pH, soil organic-C, phosphorus and CEC.

Statistical analysis: To detect effect of the treatments on soil media characteristics and seedlings growth, used two way analysis of variance (ANOVA), if the ANOVA indicated significantly at the $p < 0.05$ level than applied Duncan's multiple range test (DMRT) to detect the different among the treatments at the $p < 0.05$ level.

RESULTS

Addition of CPH biochar to soil media of cocoa seedlings significantly increased soil temperature and soil moisture as shown in Table 1. This indicates that CPH biochar has the potential to influence temperature and soil moisture. The biochar treated soil media of cocoa seedlings had higher soil temperature and soil moisture than control. Cocoa seedlings grown on soils treated with 15 g of CPH biochar kg^{-1} soil were significantly higher in soil temperature (28.19 °C) than control (27.80 °C), but insignificantly different from cocoa seedlings grown on soils with 18 g of CPH biochar kg^{-1} soil (28.13 °C). Table 1 also showed that soil media of cocoa seedlings treated with 18 g of CPH biochar kg^{-1} soil were significantly higher in soil moisture (17.44%) than control (12.68%) and the other treatments (12.72-15.70%).

Soil pH, C, P and CEC were significantly increased with biochar addition (Fig. 1-4). The highest soil pH, soil-C, P and

Table 1: Effects of biochar from cocoa pod husk (CPH) on soil temperature and soil moisture

Treatments	Soil temperature (°C)	Soil moisture (%)
Without CPH biochar	27.80 ^c	12.68 ^c
3 g biochar kg^{-1} soil	27.90 ^c	12.72 ^c
6 g biochar kg^{-1} soil	27.82 ^c	12.67 ^c
9 g biochar kg^{-1} soil	27.89 ^c	13.76 ^c
12 g biochar kg^{-1} soil	27.98 ^c	15.38 ^b
15 g biochar kg^{-1} soil	28.19 ^a	15.70 ^b
18 g biochar kg^{-1} soil	28.13 ^{ab}	17.44 ^a

Values in the same column with different superscript letters are significantly different according to the Duncan's multiple range test at $p < 0.05$ for each variable

CEC were found in the experimental unit having 18 g of CPH biochar kg^{-1} soil while the lowest was found in the control. Soil media of cocoa seedlings treated with 18 g of CPH biochar kg^{-1} soil were significantly higher than control, 3 g of CPH biochar kg^{-1} soil and 6 g of CPH biochar kg^{-1} soil in soil pH, but insignificantly different from soil media of cocoa seedlings treated with 15 g of CPH biochar kg^{-1} soil. Soil pH was insignificantly different among soil media of cocoa seedlings treated with 3 g of CPH biochar kg^{-1} soil up to 15 g of CPH biochar kg^{-1} soil. Carbon in soil media of cocoa seedlings treated with 18 g of CPH biochar kg^{-1} soil were significantly different with control, 3 g of CPH biochar kg^{-1} soil and 6 g of CPH biochar kg^{-1} soil, but insignificantly different from soil media of cocoa seedlings treated with 9 g of CPH biochar kg^{-1} soil, 12 g of CPH biochar kg^{-1} soil and 15 g of CPH biochar kg^{-1} soil. Carbon was insignificantly different among soil media of cocoa seedlings treated with 9 g of CPH biochar kg^{-1} soil up to 18 g of CPH biochar kg^{-1} soil. Phosphorus in soil media of cocoa seedlings treated with 18 g of CPH biochar kg^{-1} soil were significantly different from control, but insignificantly different from soil media of cocoa seedlings treated with 3 g up to 15 g of CPH biochar kg^{-1} soil. Figure 4 also showed that soil media of cocoa seedlings treated with 18 g of CPH biochar kg^{-1} soil were significantly different with control, 3 g of CPH biochar kg^{-1} soil, 6 g of CPH biochar kg^{-1} soil, 9 g of CPH biochar kg^{-1} soil and 12 g of CPH biochar kg^{-1} soil in CEC, but insignificantly different from soil media of cocoa seedlings treated with 15 g of CPH biochar kg^{-1} soil. CEC was insignificantly different among soil media of cocoa seedlings treated with 3 g of CPH biochar up to 12 g of CPH biochar kg^{-1} soil.

Table 2 showed that cocoa seedlings treated with 45 g of CPH biochar were significantly higher in seedling height and number of leaves than control and the other treatments except those treated with 6 g of CPH biochar kg^{-1} soil and 12 g of CPH biochar kg^{-1} soil. Leaf area of cocoa seedlings on media treated with 9 g of CPH biochar kg^{-1} soil was

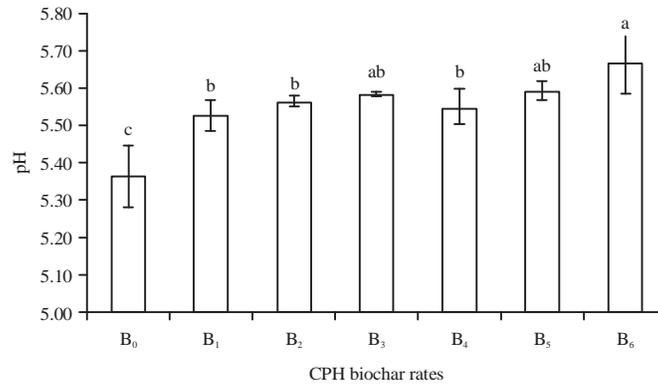


Fig. 1: Variation in soil pH under different CPH biochar application rate

Error bars indicate standard deviations and different letters on bars indicate significant differences due to CPH biochar rates treatment. Without biochar (B₀), 3 g CPH biochar kg⁻¹ soil (B₁), 6 g of CPH biochar kg⁻¹ soil (B₂), 9 g of CPH biochar kg⁻¹ soil (B₃), 12 g of CPH biochar kg⁻¹ soil (B₄), 15 g of CPH biochar kg⁻¹ soil (B₅), 18 g of CPH biochar kg⁻¹ soil (B₆)

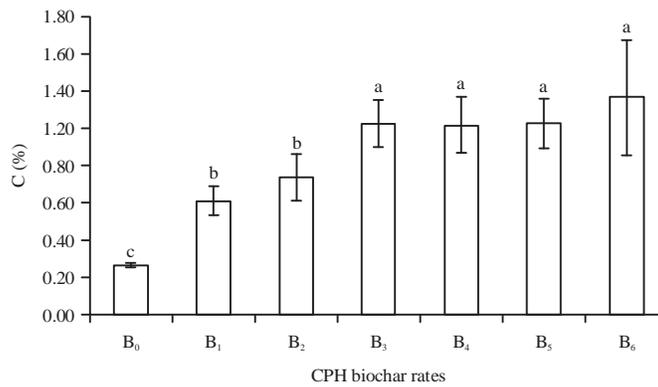


Fig. 2: Variation in soil organic-C under different CPH biochar application rate

Error bars indicate standard deviations and different letters on bars indicate significant differences due to CPH biochar rates treatment. Without biochar (B₀), 3 g CPH biochar kg⁻¹ soil (B₁), 6 g of CPH biochar kg⁻¹ soil (B₂), 9 g of CPH biochar kg⁻¹ soil (B₃), 12 g of CPH biochar kg⁻¹ soil (B₄), 15 g of CPH biochar kg⁻¹ soil (B₅), 18 g of CPH biochar kg⁻¹ soil (B₆)

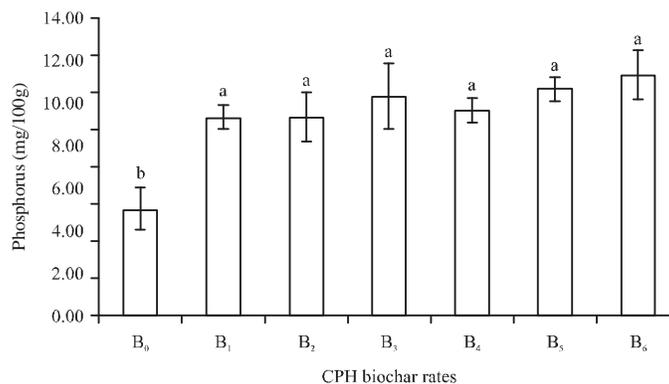


Fig. 3: Variation in soil phosphorus under different CPH biochar application rate

Error bars indicate standard deviations and different letters on bars indicate significant differences due to CPH biochar rates treatment. Without biochar (B₀), 3 g CPH biochar kg⁻¹ soil (B₁), 6 g of CPH biochar kg⁻¹ soil (B₂), 9 g of CPH biochar kg⁻¹ soil (B₃), 12 g of CPH biochar kg⁻¹ soil (B₄), 15 g of CPH biochar kg⁻¹ soil (B₅) and 18 g of CPH biochar kg⁻¹ soil (B₆)

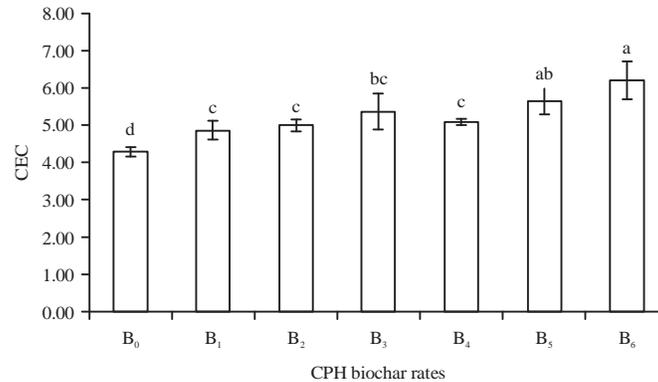


Fig. 4: Variation in cation exchange capacity (CEC) under different CPH biochar application rate

Error bars indicate standard deviations and different letters on bars indicate significant differences due to CPH biochar rates treatment. Without biochar (B₀), 3 g CPH biochar kg⁻¹ soil (B₁), 6 g of CPH biochar kg⁻¹ soil (B₂), 9 g of CPH biochar kg⁻¹ soil (B₃), 12 g of CPH biochar kg⁻¹ soil (B₄), 15 g of CPH biochar kg⁻¹ soil (B₅) and 18 g of CPH biochar kg⁻¹ soil (B₆)

Table 2: Effects of biochar from cocoa pod husk (CPH) on cocoa seedling growth

Treatments	Seedling height (cm)	Number of leaf	Leaf area (cm ²)	Root dry weight (g)	Shoot dry Weight (g)
Without biochar	23.72 ^b	12.96 ^{cd}	24.75 ^d	1.39 ^c	4.53 ^c
3 g biochar kg ⁻¹ soil	23.79 ^b	13.07 ^{cd}	31.16 ^c	1.77 ^{abc}	5.22 ^{bc}
6 g biochar kg ⁻¹ soil	26.95 ^{ab}	14.74 ^{abc}	37.00 ^b	2.057 ^a	6.42 ^{abc}
9 g biochar kg ⁻¹ soil	28.70 ^a	16.41 ^a	43.47 ^a	1.90 ^{ab}	8.08 ^a
12 g biochar kg ⁻¹ soil	25.90 ^{ab}	15.07 ^{ab}	38.73 ^b	1.74 ^{abc}	7.39 ^{ab}
15 g biochar kg ⁻¹ soil	25.24 ^b	13.29 ^{bcd}	36.83 ^b	1.47 ^{bc}	5.87 ^{abc}
18 g biochar kg ⁻¹ soil	24.58 ^b	11.52 ^d	29.77 ^c	1.43 ^c	4.47 ^c

Values in the same column with different superscript letters are significantly different according to the Duncan's multiple range test at p<0.05 for each variable

significantly higher than control and the other treatments. Root dry weight of cocoa seedlings on media treated with 6 g of CPH biochar/polybag was significantly higher than control, 15 and 18 g of CPH biochar kg⁻¹ soil but insignificantly different from cocoa seedlings grown on soils with 3, 9 and 12 g of CPH biochar kg⁻¹ soil. Table 2 also showed that shoot dry weight of cocoa seedlings on media treated with 9 g of CPH biochar kg⁻¹ soil was significantly higher than the control, 3 and 18 g of CPH biochar kg⁻¹ soil, but not significantly different from those treated with 6, 12 and 15 g of CPH biochar kg⁻¹ soil.

DISCUSSION

Biochar from cocoa pod husk (CPH) significantly influenced soil moisture and soil temperature ss shown in Table 1. CPH biochar addition leads to an increase in soil temperature. Moreover, the soils receiving higher biochar rates retained more moisture and the differences were statistically significant. The application of biochar increases soil organic carbon levels^{47,48}, improves soil structure²⁴ and improves the soil's ability to retain moisture^{4,26,49}. The significant increase in soil moisture under treatment of CPH

biochar was in conformity with the findings that biochar from CPH increases pore aeration and water availability². As also shown in Table 1, the soil temperature under different treatments is statistically similar. An increase in biochar rates were followed by an increase in soil temperature. Because of its color, CPH biochar is expected to change the albedo of the soil surface and increase the capture of radiation, thereby increasing its temperature⁴⁴.

Biochar was significantly influenced soil pH, soil-C, P and CEC. Soil pH, soil-C, P and CEC were significantly increased with biochar addition ss shown in Fig. 1-4. Soil media of cocoa seedlings treated with higher rate of biochar led to relatively higher increase in soil pH, soil-C, P and CEC. The application 3 g of biochar kg⁻¹ soil up to 18 g of biochar kg⁻¹ soil increased soil pH, soil-C, P and CEC by 5.7, 284.4, 126.7 and 45%, respectively, as compared to the control. Previous studies also suggest that the application of cocoa's shell biochar that improved soil pH, organic-C, N, P, K and CEC. This is presumably a consequence of cocoa's shell biochar has a high soil pH value, containing 35.14% C-organic, 0.87% P, 2.24% K, CEC 21.25 meq and C/N 32⁵⁰. Many researchers also had reported that biochar has the potential to improve soil pH, CEC and nutrient holding capacity^{23,24,32,16}. These findings

showed that biochar addition to soil is important for the soil C-sequestration and soil fertility in soil⁵¹, as it leads to an increase in soil pH due to biochar application^{52,53,54} and a slight increase in CEC due to biochar application^{5,52}.

Biochar significantly influenced seedling height, number of leaves, leaf area, root dry weight and shoot dry weight as shown in Table 2. This indicates that adding CPH biochar could significantly affect cocoa seedling growth. The above result was supported by another study that adding biochar affects the growth of plants⁴³. The positive effects of biochar on seedling growth are strongly related to water holding capacity^{9,10,45}, since CO₂ assimilation is regulated by water availability and it has a great influence on seedling development for height and stem diameter⁵⁵. In general, the addition of 9 g of biochar kg⁻¹ soil resulted in higher seedling height, number of leaves, leaf area and shoot dry weight than no biochar addition, but insignificantly different from the addition of 6 g of biochar kg⁻¹ soil. Interestingly, the addition of 6 g biochar kg⁻¹ soil significantly increased seedling root dry weight by 47.48% compared to no biochar addition, but the application rates exceeding 6 g of biochar kg⁻¹ soil significantly decreased root dry weight. This indicates that CPH biochar strongly influenced root growth. The presence of biochar may significantly change growth behavior of roots¹⁸. Root development is largely influenced by soil moisture⁵⁶. The application of 9 g of biochar kg⁻¹ soil increased seedling height, number of leaves, leaf area and shoot dry weight by 20.99, 26.62, 75.63 and 78.36%, respectively, as compared to control. Similar results had been reported by a study that biochar increased plant biomass by 189%⁵⁷. This is a consequence of the changes in soil C content (Fig. 2) and soil moisture (Table 1). Soil organic carbon levels increase with biochar addition^{47,48} and improves the soil's ability to retain moisture^{39,58}. Soil water content was a crucial component that influenced root growth, with possible effects on leaf growth and cocoa seedling growth as a whole. Further, these conditions also led to relatively increase in soil pH, P and CEC (Fig. 1, 3, 4). The effect of biochar on pH which is directly associated with availability of P⁵⁹ and CEC (are likely to explain increased cocoa seedlings growth, therefore, cocoa seedlings growth increased with increase in biochar rate. The other study confirms that biochar with higher ash content led to relatively higher increase in plant growth due to increased plant availability of nutrient⁶⁰. Further, other researcher observed that application of biochar increases soil pH^{52,53} and supplies essential plant nutrients^{61,24}.

The results also showed that the treatments exceeding 9 g of biochar kg⁻¹ soil significantly decreased seedling growth. The application of 18 g of biochar kg⁻¹ soil greatly decreased seedling height, number of leaves, leaf area, root

dry weight and shoot dry weight compared to control. The addition of higher doses of biochar appeared to limit plant growth^{44,45}. Further, the decrease in seedling growth may be due to restricted aeration or higher soil moisture at a rate of 18 g biochar kg⁻¹ soil as compared to control and other treatments (Table 1). This indicates that roots are exposed to limited oxygen concentrations and high water content conditions. This has been specified that root development may be damaged by lack of oxygen in compacted soils⁶².

CONCLUSION

Results showed that CPH biochar significantly influenced soil temperature and soil moisture. The application of 3 g CPH biochar kg⁻¹ soil up to 18 g of CPH biochar kg⁻¹ soil increased soil pH, soil-C, P and CEC by 5.7, 284.4, 126.7 and 45%, respectively, as compared to control. Cocoa seedling growth was significantly improved by CPH biochar and a rate of 9 g CPH biochar kg⁻¹ soil showed the best results in cocoa seedlings in terms of increased seedling height, number of leaves, leaf area and shoot dry weight by 20.99, 26.62, 75.63 and 78.36%, respectively, as compared to control, while the higher root dry weight showed at a rate of 6 g CPH biochar kg⁻¹ soil and increased by 47.48% as compared to control. CPH biochar exceeding a rate of 6 g CPH biochar kg⁻¹ soil decreased root dry weight, while seedling height, number of leaves, leaf area and shoot dry weight started to decrease at a rate of biochar exceeding 9 g CPH biochar kg⁻¹ soil. The results indicate that CPH biochar has the potential to improve soil temperature, soil moisture, soil pH, soil organic-C, P, CEC and cocoa seedling growth, however, the amount of CPH biochar material applied should be considered.

SIGNIFICANCE STATEMENT

This study discovers the possible cocoa pod husk (CPH) biochar to be developed as soil amendments that can be beneficial for improving soil fertility and plant growth and due to higher availability of nutrients. This study will help the researcher to uncover the appropriate amount of CPH applied in the nursery growing period. This study finding revealed that biochar derived from the cocoa pod husk (CPH) can be a sustainable approach improving the growing media in the nursery phase and field grown cocoa as well.

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