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Effects of soil type and biochar rate on sweet pepper productivity and microbial dynamics in regenerative organic soils

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Abstract

Biochar is widely used to improve crop productivity and soil health; however, its adoption remains limited due to insufficient quantification of benefits and unclear mechanisms, particularly within regenerative organic systems. We conducted a pot experiment applying biochar at 0 (control), 11.2, and 22.4 Mg ha⁻¹ to sandy loam and loam soils planted with sweet pepper (*Capsicum annuum* L.) and measured plant performance, microbial biomass, and soil organic carbon (SOC). Yield was higher in sandy loam than loam (282 vs. 146 g plant⁻¹). Biochar increased loam yield from < 100 g plant⁻¹ (control) to ~240 g plant⁻¹ at 22.4 Mg ha⁻¹, while sandy loam yield plateaued beyond 11.2 Mg ha⁻¹. Microbial biomass was greater in sandy loam (6845 ng g⁻¹) than loam (3737 ng g⁻¹) and declined ~17% at the highest rate, from 5628 to 4682 ng g⁻¹. Arbuscular mycorrhizal fungi colonization peaked at the moderate rate and declined at the highest rate. Although biochar increased SOC, SOC related weakly to microbial biomass in sandy loam and a significant negative relationship in loam (declining from ~4200 to 2800 ng g⁻¹ as SOC rose from 1.8 to 5.0%), indicating that biochar-derived carbon was not readily available to soil microorganisms. Fruit nutrient concentrations were unchanged across treatments. These results show biochar effects are soil- and rate-dependent: moderate application (11.2 Mg ha⁻¹) improved productivity without suppressing microbial biomass, whereas the highest rate (22.4 Mg ha⁻¹) further improved loam yield but reduced microbial biomass and decoupled SOC accumulation from microbial activity, without improving fruit nutritional quality.

Keywords Biochar, Microbial biomass, Mycorrhizal fungi, Soil organic carbon, Sweet pepper

1 Introduction

Agricultural soils are increasingly challenged by extreme weather events such as droughts, intense rainfall, flooding, heat waves, and wildfires, which degrade soil health, reduce crop productivity, and threaten global food security. For example, drought can significantly alter soil microbial communities, reduce microbial biomass and enzymatic



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activity and thereby affect soil fertility and plant performance [1, 2]. These changes may also have legacy effects, influencing microbial responses to subsequent disturbances [3]. In response, regenerative agriculture seeks to restore soil health, enhance microbial biodiversity, and reduce reliance on synthetic inputs to improve system resilience [4, 5]. Within this framework, organic amendments such as biochar are increasingly promoted for improving soil fertility and carbon (C) sequestration while supporting sustainable crop production [6, 7].

Biochar is a C-rich material produced through the pyrolysis of organic biomass, typically containing 50 to 80% C. Its porous structure and high surface area contribute to improved soil physical, chemical, and biological properties. Biochar can enhance soil structure, water retention, and nutrient availability by increasing soil pH, organic matter, and cation exchange capacity (CEC) [8–10]. In addition, its pore network provides habitat for soil microorganisms, potentially supporting bacteria, fungi, and arbuscular mycorrhizal fungi (AMF) [11–13]. Consequently, biochar has been associated with improvements in microbial activity, soil fertility, and crop productivity [14, 15].

The influence of biochar on crop yield has been widely reported, although responses vary depending on soil properties, climate, and application rates. Meta-analyses indicate that biochar can increase soil pH, CEC, and soil organic carbon (SOC), contributing to yield improvements, particularly in degraded or low-fertility soils [16–18]. Long-term studies further suggest that biochar effects can persist for several years, due to increased soil C stocks. However, these responses are not universal, and optimal application rates remain uncertain. Excessive biochar inputs may not enhance productivity and can negatively affect microbial communities, indicating the importance of rate-dependent responses [19].

Biochar also interacts with soil microbial communities, including AMF, which play a key role in plant nutrient acquisition. While some studies report increased microbial diversity and AMF colonization following biochar application, these responses are influenced by biochar properties, soil type, and environmental conditions [20, 21]. In some cases, higher biochar rates have been associated with reduced microbial activity or altered community composition, highlighting the complexity of biochar–microbe interactions and the need for site-specific evaluation [22, 23].

Despite the growing body of research, important knowledge gaps remain regarding how biochar application rate and soil type interact to influence plant performance, microbial dynamics, and soil C pools [24, 25]. While low to moderate application rates are generally beneficial, high application rates can introduce unintended negative effects on soil biology. Excessive biochar can suppress soil microbial biomass and significantly reduce arbuscular AMF colonization [22, 26, 27]. This suppression often occurs due to transient increases in soil pH beyond optimal thresholds, the sorption of signaling molecules required for mycorrhizal symbiosis, or the immobilization of essential nutrients within the biochar's porous structure [13, 28]. Understanding these high-rate limitations is critical for defining safe application thresholds.

In particular, the relationship between increases in SOC and biologically active microbial biomass is not well understood [29, 30]. To address these gaps, this study evaluated the effects of biochar application rates on sweet pepper production in two contrasting soils (sandy loam and loam). Specifically, we examined plant growth, yield, fruit quality, soil chemical properties, phospholipid fatty acid (PLFA)-derived microbial biomass, and

AMF root colonization. In addition, depth-wise variation in microbial biomass and its relationship with SOC were assessed. By integrating plant, soil, and microbial responses, this study aims to provide a more comprehensive understanding of how biochar functions within soil systems and to inform its effective use in regenerative agricultural practices.

2 Materials and methods

2.1 Experimental design and soil sourcing

The pot experiment was conducted at Rodale Institute, Kutztown, PA, following a two-factor factorial randomized complete block design with four replications in 2024. Treatments consisted of (i) soil type (sandy loam and loam) and (ii) biochar application rate (0, 11.2, and 22.4 Mg ha⁻¹). The corresponding biochar amounts applied per pot were 0, 61, and 122 g, respectively; however, results are presented using field-equivalent application rates for broader interpretation. Field-equivalent rates were calculated from the incorporation depth (top 6 cm) and bulk density (1.4 Mg m⁻³): for 1 ha to 6 cm depth, soil mass = 10,000 m² × 0.06 m × 1.4 Mg m⁻³ = 840 Mg ha⁻¹. Biochar was applied at 1.33% and 2.67% (w/w) of this incorporated soil mass, giving 11.2 and 22.4 Mg ha⁻¹, respectively; per pot amounts (61 and 122 g) were scaled from the same w/w ratio applied to the incorporated soil mass in each pot.

At the beginning of the experiment, two surface soils (0 to 20 cm depth) were collected in spring 2024. The first soil was a Pacolet sandy loam (fine, kaolinitic, thermic Typic Kanhapludults, 6 to 10% slope) with pH 6.2, 5.2% organic matter (OM), and 34.1, 80.1, and 233.1 mg kg⁻¹ of nitrate-N (NO₃-N), P, and K, respectively, and a CEC of 10.1 meq 100 g⁻¹. This soil was collected from the Rodale Institute Southeast Organic Center in Chattahoochee Hills, GA. The second soil was a Clymer loam (coarse-loamy, siliceous, active, mesic Typic Hapludults, 1 to 3% slope), hereafter referred as loam, with pH 6.6, 3.2% OM, and 12.5, 27.2, and 50.1 mg kg⁻¹ of NO₃-N, P, and K, respectively, and a CEC of 5.9 meq 100 g⁻¹. This soil was collected from the Rodale Institute Pocono Organic Center in Long Pond, PA, a regenerative organic certified site. Both soils were sourced from long-term organically managed systems. The sandy loam soil was previously planted with hairy vetch, whereas the loam soil was collected from fields previously used for sweet corn production.

Soils were air-dried, adjusted to 10% gravimetric water content, and repacked into 18.9 L plastic pots (28 cm height, 25 cm diameter). Each pot contained 12.8 kg of soil packed to a height of 20 cm, corresponding to a bulk density of 1.4 Mg m⁻³ (Fig. 1). Pots were equipped with drainage holes and placed on saucers to collect leachate. Biochar (pH: 9.5, moisture: 23.8% wet weight, organic C: 83.8%, total ash: 8.3%, N: 0.7%, CO₃: 5.7%, electrical conductivity: 0.9 dS m⁻¹, and surface area: 456 m² g⁻¹ dry), sourced from High Plains Biochar (Laramie, WY), was incorporated into the top 6 cm of soil at rates equivalent to 61 and 122 g per pot. After a 48-hour stabilization period, 3 to 4-week-old sweet pepper seedlings (cultivar: Sprinter Organic; Johnny's Selected Seeds, Winslow, ME) were transplanted (two plants per pot). Plants were grown under ambient conditions and irrigated as needed.



Fig. 1 (a) Sweet pepper plants during outdoor cultivation in pots; (b) destructive sampling at the end of the experiment, demonstrating the lead author handling an experimental pot during soil and root tissue collection; (c) root system visual differences between sandy loam (top) and loam (bottom) soils under three biochar rates (left to right): control, 11.2 Mg ha⁻¹, and 22.4 Mg ha⁻¹

2.2 Sample collection and measurements

Plant height and stem diameter were measured at multiple growth stages. Fruits were harvested repeatedly throughout the growing season, and total yield per pot was recorded. Subsamples of harvested fruit were analyzed for mineral nutrient composition at the Pennsylvania State University Agricultural Analytical Services Laboratory (University Park, PA). At the end of the experiment, soils were destructively sampled by dividing each pot into three depth intervals (0 to 6, 6 to 12, and 12 to 20 cm). Root samples were collected to determine root length (not included in the quantitative analysis, Fig. 1c) and AMF colonization, following the procedures described in Panday, Heller [31]. Total yield per pot was calculated as the cumulative fresh weight of all harvests summed across the season; per plant yield was obtained by dividing this cumulative value by two plants per pot.

Soil samples were analyzed for chemical and biological properties at Ward Laboratories Inc., Kearney, NE. Organic matter was determined by loss-on-ignition, total C and N by combustion, and NO₃-N and NH₄-N by 2 M KCl extraction. Macronutrients and micronutrients were measured using inductively coupled plasma optical emission spectrometry following microwave digestion. Cation exchange capacity and base saturation were calculated from exchangeable cations. Microbial community composition and abundance were determined using PLFA analysis following Buyer and Sasser [32].

2.3 Data analysis

All statistical analyses were conducted in R [33]. A two-way analysis of variance (ANOVA) was performed to evaluate the effects of soil type, biochar application rate, and their interaction on plant, soil, and microbial variables. When main or interaction effects were significant, means were separated by the least significant difference tests. Statistical significance was determined at $P < .05$ unless otherwise stated. Depth-wise microbial biomass data were analyzed separately using ANOVA with soil type, biochar rate, and depth as factors. Relationships between SOC and microbial biomass were assessed using linear regression analysis. Data visualization was performed using ggplot2 package, and summary statistics were calculated using dplyr.

3 Results

3.1 Plant growth and yield

Plant performance was significantly influenced by soil type, with consistently higher growth (27.4 vs. 22.2 cm), stem diameter (10.7 vs. 9.3 mm) observed in sandy loam compared to loam (Table 1). Biochar affected plant height and root colonization as a main effect. Plant height also varied over time; although measurements were taken at four different stages (Supplementary Fig. S1), only mean values are presented in Table 1. Biochar treatments showed lower plant height than the control at early growth stages, but these differences diminished as growth progressed, particularly in sandy loam. In contrast, in loam, biochar treatments consistently resulted in greater plant height than the control throughout the growing period.

A significant soil $\times$ biochar interaction was observed for yield and root colonization (Table 1; Fig. 2). In sandy loam, yield increased to > 300 g plant⁻¹ at 11.2 Mg ha⁻¹, whereas in loam yield increased from < 100 g in the control to ~ 240 g plant⁻¹ at 22.4 Mg ha⁻¹. Root colonization followed a similar pattern, with maximum values observed at 11.2 Mg ha⁻¹ (~ 45 to 48%) and substantial reductions at the highest rate. Fruit nutrient concentrations remained stable across treatments (N: 1.4 to 1.6%, P: $\sim 0.4\%$, K: 2.6 to 2.7%), except for Ca, which was significantly influenced by soil type (Table 1), indicating minimal influence of biochar on fruit nutritional composition. Residual soil nutrient concentrations (0 to 6 cm) varied among treatments but showed no consistent patterns that aligned with changes in plant performance or microbial responses (Supplementary Table S1).

3.2 PLFA microbial biomass response

Living microbial biomass and community components were strongly influenced by soil type (Table 2). Sandy loam comprised 6845 ng g⁻¹ microbial biomass compared to 3737 ng g⁻¹ in loam. Similarly, total bacterial biomass was 3346 vs. 1707 ng g⁻¹, fungal biomass was 1137 vs. 577 ng g⁻¹, and AMF biomass was 223 vs. 130 ng g⁻¹ in sandy loam and loam, indicating consistently greater microbial abundance in sandy loam.

Biochar had no significant effect on PLFA parameters; however, numerical declines were observed at the highest rate. Microbial biomass decreased from 5628 ng g⁻¹ in

Table 1 Effects of soil type and biochar application rate on phenological traits, root colonization, and nutrient concentration in harvested fruits of sweet pepper

Source of variation	Plant height cm	Stem diameter mm	Yield g	Root colonization %	N	P	K	Ca	Mg	S
	nutrient concentration, %									
Soil (S)										
Sandy loam	27.4a	10.7a	282.1	36.9	1.5	0.4	2.7	0.08b	0.2	0.2
Loam	22.2b	9.3b	146.4	29.5	1.5	0.4	2.7	0.12a	0.2	0.3
Significance	***	**	***	**	NS	NS	NS	*	NS	NS
Biochar (B)										
Control	24.0b	9.9	206.4	22.9	1.6	0.4	2.7	0.1	0.2	0.2
11.2 Mg ha ⁻¹	26.5a	10.4	224.7	42.1	1.4	0.4	2.6	0.1	0.2	0.2
22.4 Mg ha ⁻¹	23.9b	9.7	211.6	35.2	1.5	0.4	2.7	0.1	0.2	0.2
Significance	*	NS	NS	***	NS	NS	NS	NS	NS	NS
Interaction										
S $\times$ B	NS	NS	***	*	NS	NS	NS	NS	NS	NS

Means in a column followed by same lowercase letter are not significantly different. When interaction effect was significant, main effect was not reported. * $P < .05$, ** $P < .01$, *** $P < .001$, and NS = not significant

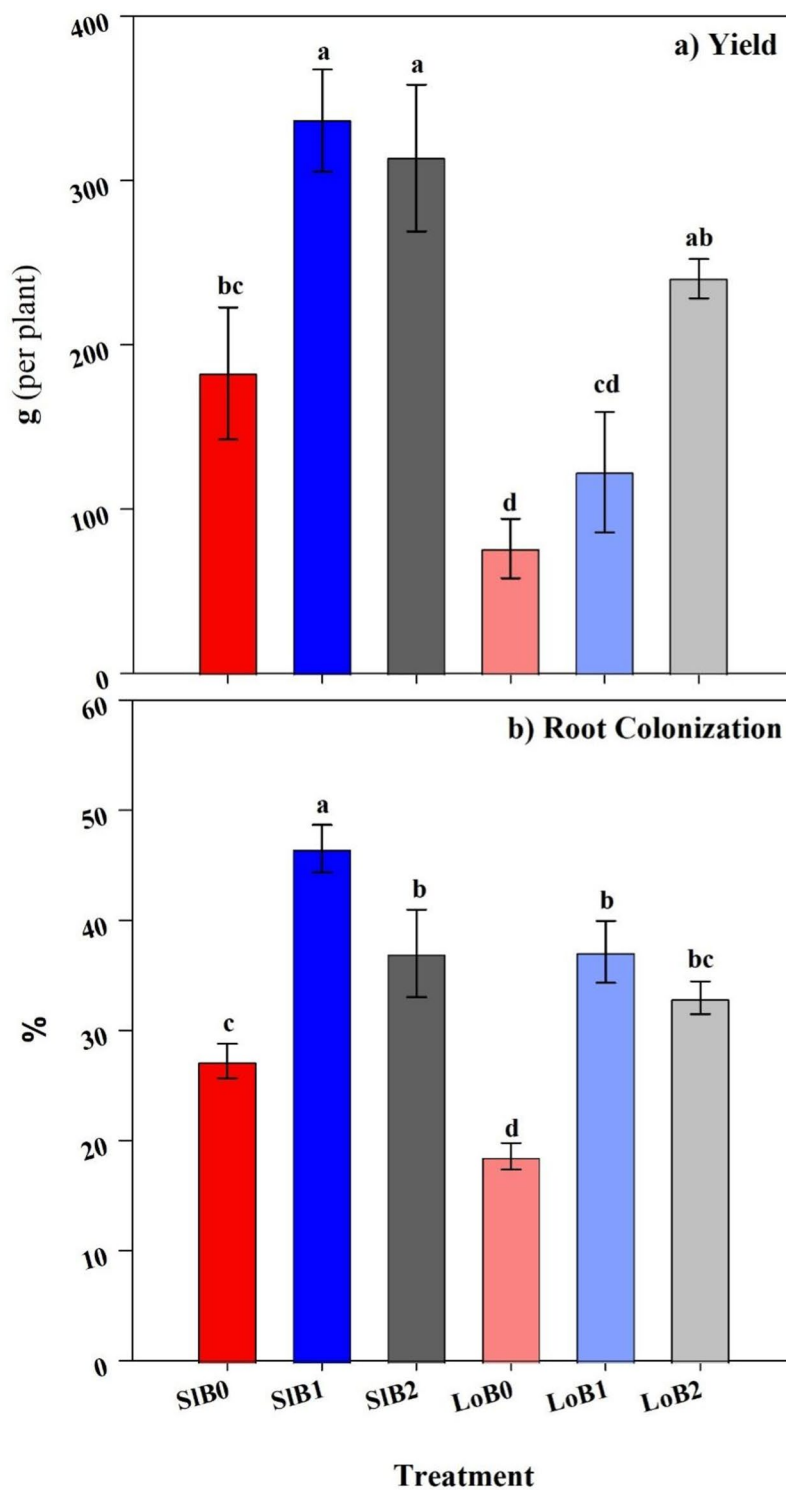
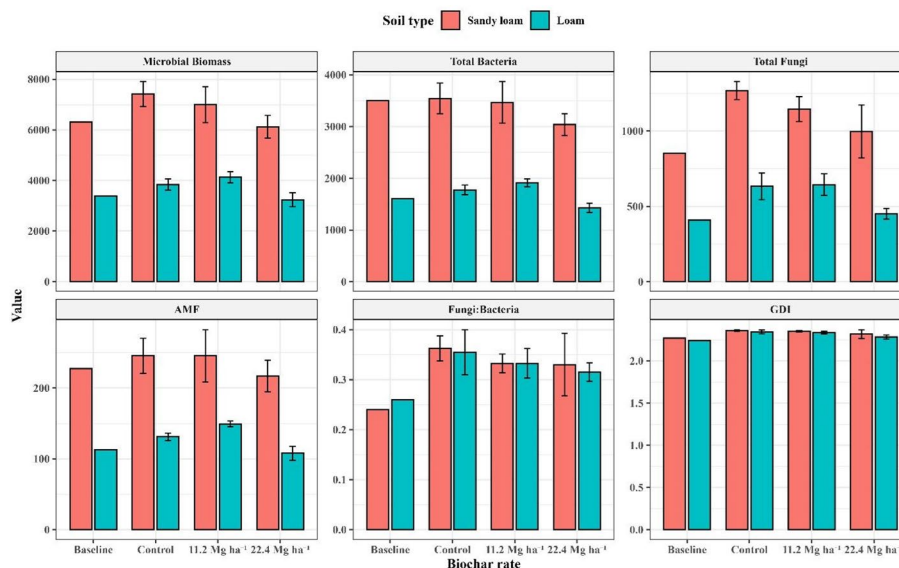


Fig. 2 Interaction effects of soil type and biochar application rate on (a) harvested fruit yield (g plant^{-1}) and (b) AMF root colonization (%). Treatments are denoted as follows: SIB0 = sandy loam without biochar (control), SIB1 = sandy loam with biochar at 11.2 Mg ha^{-1} , SIB2 = sandy loam with biochar at 22.4 Mg ha^{-1} , LoB0 = loam without biochar (control), LoB1 = loam with biochar at 11.2 Mg ha^{-1} , and LoB2 = loam with biochar at 22.4 Mg ha^{-1} . Error bars represent $\pm$ SE. Different letters above bars indicate significant differences among treatments at $P < .05$

Table 2 Effects of soil type and biochar application rate on phospholipid fatty acid (PLFA)-derived microbial parameters, including living microbial biomass, total bacteria, total fungi, and arbuscular mycorrhizal fungi (AMF), in soil samples (0 to 6 cm depth)

Source of variation	Microbial Biomass ng g ⁻¹	Total Bacteria	Total Fungi	AMF
Soil (S)				
Sandy loam	6845.0a	3345.9a	1137.3a	223.1a
Loam	3736.6b	1707.2b	576.5b	129.5b
Significance	***	**	***	**
Biochar (B)				
Control	5627.8	2658.5	950.7	188.4
11.2 Mg ha ⁻¹	5562.9	2690.5	895.5	197.4
22.4 Mg ha ⁻¹	4681.8	2235.2	724.4	162.9
Significance	NS	NS	NS	NS
Interaction				
S x B	NS	NS	NS	NS

Means in a column followed by same lowercase letter are not significantly different. When interaction effect was significant, main effect was not reported. * $P < .05$, ** $P < .01$, *** $P < .001$, and NS = not significant

**Fig. 3** Effects of soil type and biochar rate on PLFA-derived microbial properties (microbial biomass, total bacteria, total fungi, AMF, fungi: bacteria ratio, and general diversity index (GDI)). Bars represent sandy loam and loam soils; error bars indicate $\pm$ SE. Baseline values (initial soil samples before treatment application) are shown for reference but were not included in statistical analysis. Statistical analysis indicated no significant differences among the treatments ($P > .05$)

the control to 4682 ng g⁻¹ at 22.4 Mg ha⁻¹ (~17% reduction; not statistically significant, Table 2), with similar trends for bacteria, fungi, and AMF. The F: B ratio (~0.12 to 0.14) and GDI (~2.2 to 2.3) remained stable across treatments (Fig. 3). Baseline values exceeded treatment means in sandy loam but were comparable in loam.

3.3 Depth-wise variation in microbial biomass

Microbial biomass varied with soil depth (Fig. 4), increasing from the surface (0 to 6 cm) to the intermediate layer (6 to 12 cm) and declining at 12 to 20 cm. This pattern was consistent across other key PLFA variables, with treatment effects becoming progressively weaker with increasing depth (data not shown), indicates that microbial activity was not

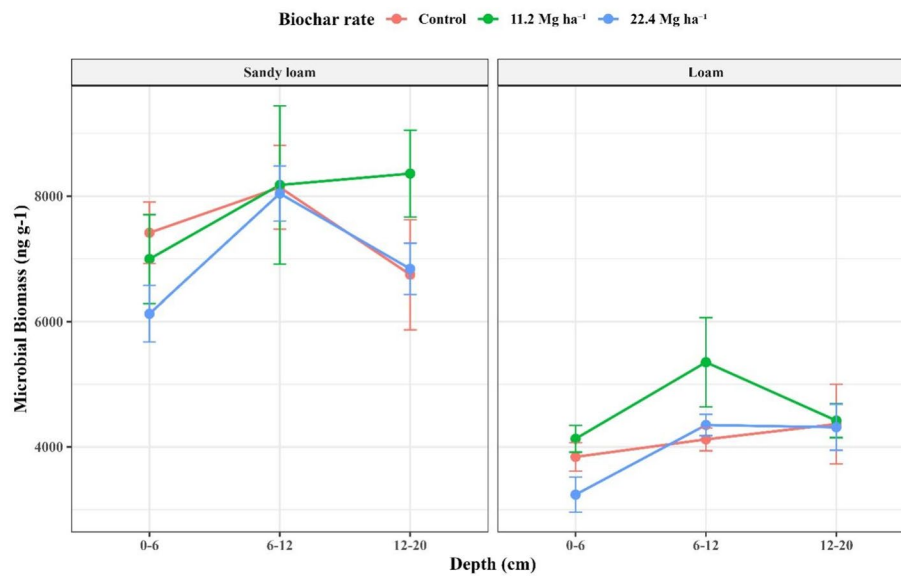


Fig. 4 Effects of biochar application rate and soil depth on microbial biomass (mean $\pm$ SE) in sandy loam and loam soils. Living microbial biomass was evaluated at depths of 0 to 6, 6 to 12, and 12 to 20 cm under three biochar treatments: control, 11.2 Mg ha⁻¹, and 22.4 Mg ha⁻¹ at the end of the experiment. Statistical analysis indicated no significant differences among the treatments or soil depths ($P > .05$)

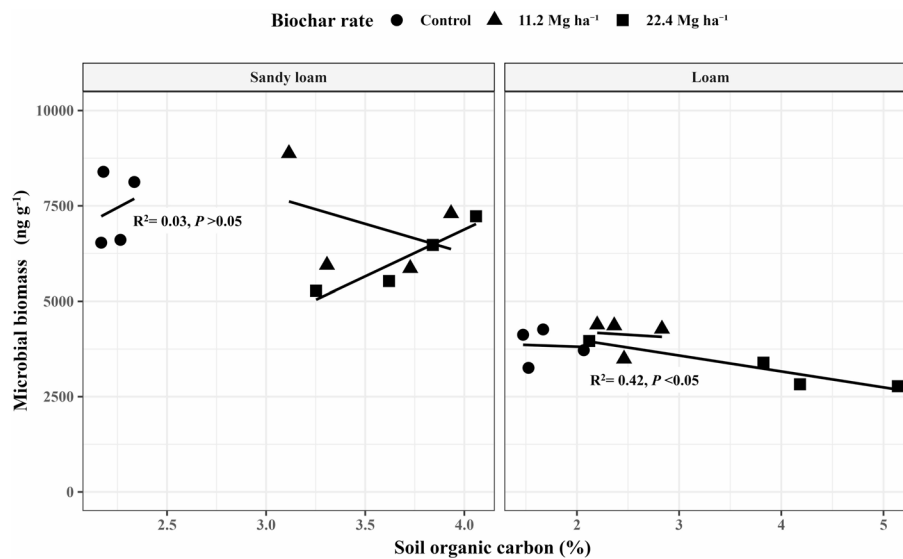


Fig. 5 Relationship between soil organic carbon and living microbial biomass in sandy loam and loam soils under three biochar application rates (control, 11.2 Mg ha⁻¹, and 22.4 Mg ha⁻¹). Symbols represent individual observations, lines denote fitted regressions, and R^2 and P values are shown for each soil type

strictly confined to the immediate surface but extended into the subsurface layer. Biochar effects were most pronounced in the upper soil layers and diminished with depth (Fig. 4). Across all depths, microbial biomass remained approximately two-fold greater in sandy loam than loam soils.

3.4 Soil organic carbon and microbial biomass relationship

The relationship between SOC and microbial biomass differed by soil type (Fig. 5). In sandy loam, SOC (2.5 to 4.0%) showed a weak or no clear relationship with microbial

biomass (~ 5500 to 8500 ng g^{-1}). In loam, increasing SOC (1.8 to 5.0%) was associated with a decline in microbial biomass (~ 4200 to 2800 ng g^{-1}), indicating a significant negative relationship. Across both soils, increases in SOC were not consistently associated with increases in microbial biomass. This decoupling suggests that a substantial portion of accumulated C was not readily available for microbial utilization, highlighting the distinction between total SOC and biologically active C pools.

4 Discussion

The present study suggests that the effects of biochar on plant performance and soil biological properties are strongly dependent on soil type and application rate. Sandy loam (initial pH: 6.2) consistently supported greater plant growth and microbial biomass than loam (initial pH: 6.6), with yield nearly doubling (282 vs. 146 g plant^{-1}) and microbial biomass increasing by $\sim 83\%$. Biochar (pH: 9.5) improved yield in loam, particularly at 22.4 Mg ha^{-1} , whereas in sandy loam yield plateaued or declined at higher rates. These results support previous findings that biochar benefits are often greater in lower-performing soils, while excessive application may reduce performance in more fertile systems [34–36].

Plant growth responses were dynamic over time, highlighting that biochar effects may not be immediate [37, 38]. Early growth stages showed reduced plant height under biochar treatments compared to control, whereas differences diminished as growth progressed, particularly in sandy loam. These contrasting temporal patterns suggest that biochar-induced changes in soil conditions initially altered nutrient availability but became more favorable as the system stabilized. This is consistent with the buffering effect of alkaline biochar, which can modify soil pH and nutrient dynamics through the release of basic cations (Ca, K, Mg, Na), especially in slightly acidic soils [39, 40]. Such transient responses emphasize that early growth measurements may not reflect final plant performance. In contrast, plant height in loam was consistently higher under biochar treatments, suggesting that these buffering and structural effects may have been immediately beneficial in that soil.

Fruit nutrient concentrations remained relatively stable (Table 1), indicating that yield increases were not accompanied by improvements in nutritional quality and may reflect dilution effects [34]. To better understand the biological mechanisms underlying these crop responses, we evaluated AMF. Arbuscular mycorrhizal fungi were particularly responsive to biochar application rate. Root colonization increased at the moderate biochar rate (11.2 Mg ha^{-1}) but declined at the highest rate (22.4 Mg ha^{-1}), although remaining higher than the control (Table 1). A similar pattern was observed for AMF biomass (Table 2), suggesting that moderate biochar inputs may stimulate, whereas excessive application may partially suppress symbiotic activity. Such responses have been linked to changes in soil pH, nutrient availability, and microbial habitat conditions under high biochar inputs [13, 41].

Although biochar altered residual soil nutrient concentrations in the surface layer, these changes were not consistently associated with variations in microbial biomass, root colonization, or yield (Supplementary Table S1), potentially indicating that nutrient availability was not the primary driver of treatment effects. This pattern persisted despite differences in baseline soil fertility, as residual nutrient concentrations did not consistently explain variations in plant performance or microbial responses. However, reduced

AMF did not consistently correspond to reduced yield, particularly in loam (Fig. 2), where soil N and P availability were maintained or increased under biochar application [42]. This suggests that plant nutrient acquisition may have become less dependent on AMF under these conditions.

Biochar also affected entire soil microbial biomass. Overall, microbial biomass declined non-significantly at the highest application rate (from 5628 to 4682 ng g⁻¹; ~17% reduction), despite the concurrent increase in SOC [41, 42]. Despite this decline, overall yield remained higher in sandy loam than in loam (282 vs. 146 g plant⁻¹), while loam exhibited a stronger relative response to biochar application, with yield increasing from <100 g plant⁻¹ in the control to ~240 g plant⁻¹ at 22.4 Mg ha⁻¹ (Fig. 2). This pattern indicates that plant productivity and microbial activity can respond independently to biochar amendment [43]. Such variability is consistent with recent meta-analytical evidence showing that biochar effects on crop growth, biomass accumulation, and soil quality are highly variable across studies and depend on soil conditions, management, and application rates [44]. Microbial responses were strongly depth dependent. Biomass increased from the surface to the 6 to 12 cm layer, where maximum values were observed, and declined at greater depths, with treatment effects diminishing downward (Fig. 4). This pattern is consistent with previous syntheses indicating that biochar effects are concentrated in the surface soil but may extend into subsurface layers, with progressively weaker responses due to limited vertical movement and redistribution processes [45].

A key outcome of this study is the apparent decoupling between SOC, microbial biomass, and plant productivity. Organic carbon exhibited weak or negative relationships with microbial biomass (Fig. 5), indicating that biochar-derived C is largely recalcitrant and not readily available for microbial utilization [41, 42]. It is important to distinguish total SOC, measured by combustion (including both biochar C and native soil C), from labile C, which supports short-term microbial activity. Because biochar C is largely aromatic and recalcitrant, increases in total SOC do not necessarily increase labile C, providing a plausible explanation for why SOC increased while microbial biomass did not. In loam, the significant negative relationship between SOC and microbial biomass (Fig. 5) may reflect this mechanism more strongly because of its lower baseline SOC and bacterial biomass. It may also partly result from the simplified, repacked pot environment, although the influence of a few high SOC observations cannot be ruled out and should be confirmed under field conditions.

These contrasting responses of higher SOC but lower microbial biomass and greater loam yield despite reduced microbial biomass suggest that biochar improves plant growth mainly through direct chemical and physical effects rather than microbial processes. Its alkaline pH (9.5), cation content, and porous structure improve soil pH, nutrient retention, water holding capacity, and cation exchange capacity, particularly in the lower fertility loam soil. At the same time, biochar can adsorb labile organic compounds, reducing their availability to microorganisms (negative priming) [46]. Together, these mechanisms provide a possible explanation for the different responses of crop yield and microbial biomass across biochar rates and soil types.

Several limitations should be considered when interpreting these results. Soils were repacked at a uniform bulk density, and changes in soil physical structure following biochar addition were not monitored. In addition, soil physicochemical changes after

biochar application were not measured over time, which may limit interpretation of the mechanisms driving these depth-dependent responses. Because this study was conducted over a single growing season under controlled conditions, longer-term field studies are needed to better understand how biochar effects on soil C dynamics, microbial communities, and crop nutritional quality evolve over time.

Overall, these findings indicate that biochar is a context-dependent amendment. While it can enhance productivity in lower-performing soils such as loam, excessive application may risk suppressing microbial biomass and symbiotic associations without improving crop nutritional quality. This variability aligns with recent syntheses indicating that biochar effects depend strongly on soil properties, application rate, and environmental conditions, and that excessive inputs can sometimes suppress microbial activity despite increasing soil C pools [47]. Effective use of biochar in regenerative systems therefore appears to require moderate application rates and soil-specific management strategies that support existing soil biological processes rather than relying solely on C addition.

5 Conclusions

Biochar effects on sweet pepper productivity and soil microbial properties were soil type- and rate-dependent. Sandy loam produced higher overall yields, whereas loam responded more strongly to biochar. Although the highest biochar rate increased yield in loam, it also reduced microbial biomass and AMF colonization compared with the moderate rate, without improving fruit nutritional quality. In addition, increases in SOC did not correspond to greater microbial biomass, indicating that biochar-derived carbon was largely unavailable for microbial use. Overall, 11.2 Mg ha⁻¹ provided the best balance between crop productivity and microbial health across both soils, while 22.4 Mg ha⁻¹ may be appropriate only when maximizing yield in lower fertility soils is the primary goal. For regenerative organic vegetable systems, biochar should be matched to soil type and applied at moderate rates as part of an integrated soil health strategy. Because this was a single-season pot study using two soils with similar pH, field-based, multi-season studies across a wider range of soil types are needed to validate these recommendations.

Supplementary Information

The online version contains supplementary material available at <https://doi.org/10.1007/s44378-026-00297-8>.

Supplementary Material 1

Acknowledgements

The authors would like to thank Phillip Hinson, Cole Connolly, and Blake Morris for their support in conducting this study.

Author contributions

D.P.: Conceptualization, Data Curation, Formal Analysis, Funding Acquisition, Manuscript Preparation-First Draft, Methodology, Study Design, and Validation, S.D.: Manuscript Preparation-Review Draft, Methodology, and Study Design, and B.S.A.: Manuscript Preparation-Review Draft and Study Design, W.P.H.: Data Curation, Manuscript Preparation-Review Draft, and Methodology, L.M.: Data Curation and Manuscript Preparation-Review Draft, and A.G.: Funding Acquisition, Manuscript Preparation-Review Draft, Project Administration, and Study Design. All authors reviewed the manuscript.

Funding

This research project was funded by Arabella Advisors.

Data availability

The datasets generated during and/or analysed during the current study are available from the corresponding author on reasonable request.

Declarations

Ethics approval and consent to participate

Sweet pepper plants used in this study were commercially cultivated materials and their collection and use complied with relevant local and institutional guidelines. No wild plant species were collected, and no special permits or licenses were required for this study. The biochar used was a commercially produced material derived from wood biomass, and its source, manufacturer, and location have been provided in the manuscript.

Consent of publication

Informed consent for publication was obtained from the individual participant visible in Fig. 1. The individual pictured is an author of this study and has explicitly consented to the publication of their identifiable image in this manuscript.

Disclaimer

Mention of trade names or commercial products in this publication is solely for the purpose of providing specific information and does not imply recommendation or endorsement by the United States Department of Agriculture (USDA). USDA is an equal opportunity employer.

Clinical trial number

Not applicable.

Competing interests

The authors declare no competing interests.

Received: 28 April 2026 / Accepted: 30 July 2026

Published online: 09 August 2026

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