

Impact of Biochar Application on Carbon Dynamics and Fertility of Soils Over-fertilization with Compost

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Abstract: In Taiwan, farmers often apply excess compost to ensure adequate crop yield in frequent tillage, highly weathered, and lower fertility soils. The potential of biochar (BC) for decreasing soil C mineralization and improving soil nutrient availability in compost over-fertilized soil is promising, but is still under-examined. To test this potential, a 434-day incubation experiment of in vitro C mineralization kinetics was conducted. We added 0%, 0.5%, 1.0%, and 2.0% (w/w) woody BC composed of lead tree (*Leucaena leucocephala* (Lam.) de. Wit) to an Oxisols and two Inceptisols of Taiwan. In each treatment, 5% swine manure compost (two times recommended the amount) was added to serve as the over-fertilized soil. The results indicated that soil type strongly influences the impact of BC addition on soil carbon mineralization potential. Respiration per unit of total organic carbon (total mineralization coefficient) of the three studied soils significantly decreased with BC addition increased. Principal component analysis suggested that for retaining more plant nutrients in addition to the effects of carbon sequestration, farmers could use locally produced biochars and composts in highly weathered and highly frequent tillage soil. Adding 0.5%–1% woody BC in soil should be reasonable and appropriate.

Keywords: biochar; carbon mineralization; over-fertilization soils; compost; Ultisols

1. Introduction

The problem of soil degradation due erosion, salinization, depletion of soil organic matter (SOM), and nutrient imbalance is the most serious bio-physical constraint limiting agricultural productivity in many parts of the world [1]. Maintaining an appropriate level of SOM and ensuring the efficient biological cycling of nutrients are crucial to the success of soil management and agricultural productivity strategies [2,3], including the application of organic and inorganic fertilizers combined with knowledge of how to adapt these practices to local conditions, aiming to maximize the agronomic use efficiency of the applied nutrients and thus crop productivity [3]. In soils with low nutrient retention capacity, strong rains rapidly and easily leach available and mobile nutrients into the subsoil where they are unavailable for most crops [4], rendering conventional fertilization highly inefficient [5]. SOM has declined in the arable lands of Taiwan in the last several decades due to highly frequent tillage in association with high air temperature and rainfall, and farmers often apply excess compost to ensure adequate crop yield.

Depending on the mineralization rate, organic fertilizers, such as compost, mulch, or manure, release nutrients in a gradual manner [6] and may therefore be more appropriate for nutrient retention under high-leaching conditions than inorganic fertilizers. Due to the relatively low levels of nutrients (10–20 g N/kg and less than 10 g P/kg) in compost compared to complete fertilizer, as well as the low plant availability of compost N and P, a large amount of compost is needed to meet the N and P crop requirements [7], and farmers often apply excess compost to ensure adequate crop yield, leading to excessive N and P loading to the environment. In the tropics, however, naturally rapid mineralization of SOM is a limitation of the practical application of organic fertilizers; in addition to repeated application at high doses and the cost of application of organic materials, their rapid decomposition and mineralization may significantly contribute to global warming [8–10]. Excessive manure application often causes heavy metal accumulation (Cu, Pb, Zn, etc.) in the soil, and the soluble fraction of these metals tends to increase due to desorption and remobilization of metals previously bound to the soil matrix, leading to enhanced crop uptake of heavy metals [11]. In acidic and highly-weathered tropical soils, application of organic fertilizers and charcoal increases nutrient stocks in the rooting zone of crops, reduces nutrient leaching, and thus improves crop production [5]. Biochar could be a key input to raise and sustain production and simultaneously to reduce pollution and dependence on fertilizers, and could also improve soil moisture availability and sequester carbon [12]. Biochar (BC) studies have mainly focused on the effects of pure BC addition or artificial fertilizer; however, pure BC does not provide a high amount of nutrients in most cases [13]. Incorporation of BC-compost into poor soil is considered a promising approach to produce a substrate like *terra preta*, and the study result demonstrated a synergistic positive effect of compost and BC mixtures on soil organic

matter content, nutrients levels, and water-storage capacity of a sandy soil under field conditions [13]. BC either helped stabilize manure C, or the presence of manure reduced the effect of BC on the mineralization of soil organic carbon (SOC) [14]. Trupiano et al. [15] showed that both BC amendment (65 g/kg) and compost (50 g/kg) addition to a moderately subalkaline (pH 7.1) and clayey soil poor in nutrients had a positive effect on lettuce plant growth and physiology, and on soil chemical and microbiological characteristics; however, no positive synergic or summative effects exerted by compost and BC in combination were observed compared to the compost alone treatment. BC, compost, and BC-compost blend all fewer environmental impacts than mineral fertilizer from a systems perspective [16].

However, in compost over-fertilized soils, little is known about the impact of BC application rates on the carbon mineralization and soil fertility of mixed-soil (BC, compost, and soil) in highly frequent tillage soil systems. In vitro C mineralization kinetics of various BC addition rates in three selected soils were examined in this study. We hypothesized that BC addition may stabilize compost organic matter, diminish mixed-soil C mineralization, and improve soil nutrient status. The aims of our research were: (1) to quantify the effects of woody BC additions on C mineralization and soil fertility and (2) to evaluate the sustainability of woody BC additions in terms of maintaining high SOM contents and nutrient availability.

2. Materials and Methods

2.1. Soil Characterization

Three representative rural soils derived from different parent material in Taiwan were selected for the incubation experiment. The Pingchen (Pc) soil series is a relict tertiary Oxisol (slightly acidic Oxisol, SAO) in Northern Taiwan [17]. The Erhlin (Eh) soil series is an Inceptisol (mildly alkaline Inceptisol, MAI) developed from calcareous slate old alluvial parent material in Central Taiwan. The Annei (An) soil series is also an Inceptisol (slightly acid Inceptisol, SAI) developed from calcareous sandstone-shale new alluvial parent material in Southern Taiwan. Rice is the commonly grown crop in the sampled fields. The physical and chemical characteristics of the top soils (0–20 cm depth) are presented in [Table 1](#).

Soil pH was determined in a soil-to-deionized water ratio of 1:1 (g/mL) and in soil-to-1 N KCl ratio of 1:1 (g/mL) [18] and electrical conductivity (EC) was determined by saturation extract of the soil sample [19]. Soil particle size was determined using the pipette method [20]. Soil total C (TC) content was determined by dry combustion [21], using an O · I · Analytical Solid Total Organic Carbon (TOC) (O.I. Corporation/Xylem Inc., Texas, USA). Soil TC was assumed to be organic in nature because the low or neutral soil pH precludes carbonates. Soil total nitrogen (TN) content was extracted by digesting 1.0 g dried and powdered sample using concentrated H₂SO₄

in a Kjeldahl flask using K_2SO_4 , $CuSO_4$, and Se powder as catalysts. TN concentration was determined via O · I · Analytical Aurora Model 1030W (O.I. Corporation/Xylem Inc., Texas, USA); content of soil total phosphorus (TP) in the digested solution was determined with inductively coupled plasma optical emission spectrometry (ICP-OES) (PerkinElmer Inc., Optima 2100DV, USA). The exchangeable bases (Ex-K, Na, Ca, and Mg), cation exchangeable capacity (CEC), and base saturation percentage (BS%) were measured using the ammonium acetate method at pH 7 [22]. Mehlich-3 extraction [23] was used for analysis of plant available nutrients. Mehlich-3 extractable (M3-) K, Na, Ca, Mg, Fe, Mn, Cu, Pb, Zn, and P values were measured with ICP-OES.

2.2. Studied BC

BC produced from lead tree (*Leucaena leucocephala* (Lam.) de. Wit) in an earth kiln was constructed by the Forest Utilization Division, Taiwan Forestry Research Institute, Taipei, Taiwan [24,25]. The charring for earth kilns typically requires several days and reaches temperatures about 500 to 700 °C. The highest temperature in the kiln at the end of carbonization was above 750 °C. The BCs were homogenized and ground to <2 mm mesh for analyses. The characterization of the studied BC was described in the previous studies [26,27] (Table 1).

2.3. Incubation Experiment

In amended soils, laboratory incubation is generally used to obtain accurate information about C-mineralization dynamics [28], and the data can then be fitted to or with kinetic models to obtain complementary information such as the C-mineralization rates and the potentially mineralizable C. Therefore, a laboratory aerobic incubation experiment was conducted over 434 days to study and evaluate C-mineralization kinetics in a nonamended (no BC addition) soil (i.e., the control) and in three soils amended with three BC application rates. A total of 12 treatments were used in this study, and each treatment was set up in triplicate. To all soil treatments, we added 5% commercially available swine manure compost as soil fertilizer, which is twice the recommended amount of organic fertilizer in Taiwan. The characteristics of the swine manure compost are listed in Table 1. The application rate of BC, 0%, 0.5%, 1.0%, and 2.0% (w/w), equated to field applications of approximately 0, 12, 24, and 48 tons/ha, respectively, considering 2400 Mg of soil per hectare (soil bulk density equal to 1.2 Mg/m³ and an arable soil layer of 20 cm). Twenty-five grams of mixed soil sample was placed in 30-mL plastic containers, which were subsequently put into 500-mL plastic jars containing a vessel with 10 mL of distilled water to avoid soil desiccation and a vessel with 10 mL of 1 M NaOH solution to trap evolved CO₂. The jars were sealed and incubated at 25 °C. Soil moisture contents were adjusted to 60% of field capacity before the

incubation and was maintained throughout the experiment using repeated weighing. The incubation experiment was run for 434 days with 23 samples taken after 1, 3, 7, 14, 21, 28, 35, 42, 49, 56, 63, 77, 91, 105, 119, 133, 161, 189, 217, 245, 308, 371, and 434 days. After sampling, the vessel with 10 mL of a 1 M NaOH solution was removed, resealed, and stored until analysis for CO₂ and replaced with fresh NaOH. A titrimetric determination method was used to quantify evolved CO₂ [29]. The cumulative CO₂ released and C mineralization kinetics were calculated based on the amount of CO₂-C released during different intervals of time in each treatment. In addition, total mineralization coefficient (TMC) was calculated according to Díez et al. [30] and Méndez et al. [31] as follows:

$$\text{TMC (mg CO}_2\text{-C/g C)} = \text{CO}_2\text{-C evolved/initial TOC} \quad (1)$$

where CO₂-C evolved is expressed as mg CO₂-C/100 g soil and initial total organic carbon (TOC) is expressed as g C/100 g soil.

Samples of the BC-treated soil were collected after incubation 434 for days or analysis of plant available nutrients using Mehlich-3 extraction (M3-) [23]. M3-K, Na, Ca, Mg, Fe, Mn, Cu, Pb, Zn, and P values were measured with ICP-OES. To compare the changes and quantify the impacts of soil BC amendments on nutrients, soil pH, TC, TN, TP, exchangeable bases (Ex-K, Na, Ca, Mg), and CEC of the BC-treated soil on day 434 were also measured.

2.4. Statistical Analysis

The statistical analyses (calculation of means and standard deviations, differences of means) were performed using SAS 9.4 package (SAS Institute Inc., SAS Campus Drive, Cary, NC, USA). Results were analyzed by analysis of variance (one-way ANOVA) to test the effects of each treatment. The statistical significance of the mean differences was determined using the least-significant-difference (LSD) tests based on a *t*-test at a 0.05-probability level. The Pearson correlation coefficient (*r*) calculated and principle component analysis (PCA) was performed using SAS 9.4 software.

3. Results

3.1. Carbon Mineralization

Addition of woody BC showed significantly reduced CO₂ release in SAO soil, no significantly difference in MAI soil, and a significant increase in SAI soil (Figure 1 and Table 2). In SAO soil treatments, the CO₂-C release reduced about 8.8%, 7.0%, and 9.4% for 0.5%, 1.0%, and 2.0% BC addition rates, respectively. No significant difference was observed in the MAI soil treatments; the CO₂-C release reduced about 8.8%, 7.0%, and 9.4% for 0.5%, 1.0% and 2.0% BC addition rates, respectively. In contrast, in SAI soil treatments, the CO₂-C release increase about 6.2%,

15.3%, and 7.9% for 0.5%, 1.0%, and 2.0% BC addition rates, respectively. The results of the total mineralization coefficient (TMC) indicated significantly reduced trend with increasing BC addition in SAO and MAI soil treatments, but in SAI soil, only the 2% addition showed a significantly decrease in comparison with the control. The value of TMC was higher in SAI soil treatments, followed by MAI soil treatments, and much lower in SAO soil treatments. The TMC value decreased by 16.5%, 24.0%, and 37.8% for 0.5%, 1.0%, and 2.0% BC additions to SAO soil, respectively. In MAI soil, TMC reduced by 19.6%, 20.7% and 32.5% for 0.5%, 1.0%, and 2.0% BC additions, respectively. In SAI soil, TMC reduced by 0.7% and 19.8 for 0.5% and 2.0% BC addition, respectively, but increased 2.0% for 1.0% BC addition. We hypothesized that woody BC addition may stabilize compost organic matter and diminish C mineralization in soils over-fertilized with compost, and the results showed that addition of woody BC to SAO soil produced a favorable effect by decreasing the cumulative amount of CO₂-C evolution, but in SAI soil, it produced an unfavorable effect by increasing the cumulative amount of CO₂-C evolution. We observed no effect in MAI soil.

3.2. Changes in Soil Properties and Fertility Characteristics

After 434 days of incubation, all treatments were analyzed to investigate if BC addition could result in increasing (enhancing) or decreasing (reducing) soil properties and fertility characteristics in over-fertilized soils (Table 3). The enhancing effect on soil fertility characteristics suggests that adding BC can retain nutrients in over-fertilized soils, even after one year of incubation. At the end of this year, the higher amount of nutrients that could be retained in soils suggests that the farmer could apply less compost in the following year.

At the end of incubation, TC significantly increased with BC addition increase in the three soils. The significant decreases in CO₂-C evolution and TMC with BC addition increase could explain the soil carbon accumulation (sequence) in soils. That is, in this study, BC addition evidently reduced C-mineralization and TMC and resulted in more soil C sequestered in soils over-fertilized with compost. TN content significantly increased with 1% and 2% BC addition in MAI and SAI soils, but slightly decreased in SAO soil. The application of woody BC with a high C/N ratio in three over-fertilized soils did not obviously result in soil nitrogen fixation, but in contrast, increased the TN contents. The TP content significantly increased with 0.5% and 2.0% BC addition in SAO soil and with 2.0% in MAI soil, but significantly decreased with 1.0% and 2.0% BC addition in SAI soil. The C/N ratio significantly increased with BC addition increased; the values of which were all less than 10:1 (Table 3).

The soil pH significantly increased with 2.0% BC addition of three soils, about 0.3 pH unit for SAO soil, 0.1 pH unit for MAI soil, and 0.2 pH unit for SAI soil (Table 3). Within the

exchangeable bases, Ca and Mg both showed insignificant difference from the control in the three soils, but obviously increased in MAI and SAI soils. Addition of 0.5% BC resulted in a significant increase in the K and Na contents in SAO soil but a decrease with 1.0% and 2.0% additions. The 2% BC addition in MAI and 1.0% and 2.0% BC additions in SAI soil significantly increased K content. CEC showed variable changes—significant increases occurred in 1.0% BC addition in MAI soil but significant decreases occurred with 2.0% addition in SAI soil.

In SAO soil in terms of soil fertility characteristics, the contents of M3-P, K, Mg, Fe, and Mn obviously and significantly decreased with increasing BC addition (Table 4). In contrast, Ca, Cu, Pb, and Zn increased with increasing BC addition, especially with the 2.0% addition. The contents of Cu, Pb, and Zn in SAO soil were about 8–9, 10–12, and 26–30 mg·kg⁻¹, respectively. These values not very high and could not result in plant toxicity. However, we should pay more attention in SAO soil to ensure that these metals are not fixed by BC, and the availability may increase after BC addition. In MAI soil, P, K, Ca, Mg, Fe, and Mn increased after BC addition, but only K content significantly increased with 1.0% and 2.0% BC addition. Significant decreases of Cu, Pb, and Zn occurred with 0.5%, 1.0%, and 2.0% BC addition (except for Zn with 2.0%). The application of woody BC in over-fertilized MAI soil could retain some nutrients and significantly reduce heavy metal availability. Similar results for K, Cu and Pb were found for SAI soil. However, the P content with 1.0% BC addition and Zn content with 0.5% and 1.0% addition significantly increased in SAI soil. The contents of Ca, Mg, Fe, and Mn decreased after BC addition. Adding BC to SAI soil could result in some nutrients decreasing and reduce the availability of Cu and Pb, but we should pay attention to the risk of increased Zn availability.

3.3. Principal Component Analysis

The PCA described substantial differences in soil physicochemical characteristics (pH, TC, TN, TP, M3-P, M3-K, M3-Cu, M3-Pb, and M3-Zn), and cumulative CO₂-C among the BCs (Figure 2). The PCA identified two primary components of SAO soil fertility, and PC1 and PC2 accounted for 49.1% and 21.0% of the total variance, respectively. AdditioPC1 and PC2 explained 43.0% and 19.78%, and 52.3% and 23.3% of the total variance in the MAI and SAI soil, respectively.

PCA showed two groupings for each of the three soils. The two grouping of SAO soil were: pH, TC, TP, M3-Pb, M3-Zn, and M3-Cu (Group 1), and TN, M3-P, M3-K, and cumulative CO₂-C (Group 2). The 2% BC addition clustered near Group 1, whereas the 0.5% BC addition clustered closer to Group 2. For the MAI soil, two groupings stood out: pH, TC, TN, TP, M3-P, and M3-K (Group 1), and M3-Cu, M3-Pb, M3-Zn, and cumulative CO₂-C (Group 2). The addition of 1% BC clustered near Group 1. Lastly, the PCA for the SAI soil showed two main groupings: pH,

TC, TN, M3-P, M3-K, M3-Zn, and cumulative CO₂-C (Group 1), and TP, M3-Cu, and M3-Pb (Group 2). Addition of 1% BC clustered closer to Group 1, whereas 0.5% BC addition clustered closely to Group 2.

4. Discussion

4.1. Effect of BC on Carbon Mineralization

Whereas proper use of compost promotes soil productivity and improves soil quality, excess application degrades the soil and water quality and inhibits crop growth [32]. The net decrease in CO₂ emission with BC is clear, both directly through sequestration of BC C and indirectly through altering soil physical, chemical, and microbiological properties [5,33]. The BC used in our study was a high-temperature pyrolysis product of wood with an accumulation of black C. This property makes it inert and resistant to microbial degradation [34]. In this study, we hypothesized that the addition of relatively small amounts of a woody BC to soils with excess swine manure compost application could stabilize compost organic matter and decrease C mineralization. Decreasing C mineralization could contribute to reducing the decomposition of compost organic matter, enhance C sequestration, retain some nutrients, and may reduce the application rate of manure compost in the following year.

Carbon mineralization in each soil type was obviously greater in the initial days of the incubation (Figure 1), especially on the first day of incubation, as reported in other studies [35–37]. Swine manure compost contains a significant amount of easily degradable organic C, and consequently, and intense increases in soil microbial activity should occur after its application to soil, leading to high C mineralization. The BC treatments significantly reduced C mineralization in SAO soil, and showed insignificant difference in MAI soil (Table 2), but has significantly increased C mineralization in SAI soil (1.0% and 2.0% BC treatments). Mukome et al. [38] showed that emissions of CO₂ from the interaction of BC with compost organic matter (COM) are dependent on the BC feedstock and pyrolysis temperature; however, the net CO₂ emissions were less for the BC and compost mixtures compared to compost alone, suggesting that BC may stabilize COM and diminish C mineralization. The presence of easily metabolized organic C or additional labile organic carbon sources has been shown to accelerate BC decomposition (or increased soil CO₂ effluxes) [39–42], suggesting that co-metabolism contributes to BC decomposition in soils. Respiration per unit of TOC (TMC) of the three studied soils significantly decreased with increasing BC addition. The four treatments in SAO soil had significantly lower TMC than in MAI and SAI soils. Méndez et al. [31] suggested that a high TMC results in a more fragile humus and thus in a lower quality soil. In contrast, the lower TMC means that organic matter is conserved more efficiently and maintains the activity of the microorganisms responsible for soil organic matter biodegradation.

BC amendments clearly had effects on soil CO₂ evolution, which varied with soil type. In the coastal saline soil (pH 8.09), the peanut-shell-derived BC addition increased the cumulative CO₂ emissions and the cumulative SOC mineralization due to the labile C released from BC and the enhanced microorganism proliferation [37]; however, the increased mineralized C only accounted for less than 2% in the 0.1%–3% BC treatments, indicating that BC may enhance C sequestration in saline soil. Rogovska et al. [14] indicated that BC additions sometimes increase soil respiration and CO₂ emissions, which could partially offset C credits associated with soil BC applications, and many uncertainties are related to estimation of mineralization rates of BC in soils. In this study, the result of CO₂ evolution and TMC both suggest that when adding excess swine manure compost in Oxisols, a higher BC application rate can stabilize and prevent the rapid mineralization of compost. BC addition in mildly alkaline Inceptisols can stabilize compost organic matter but only slightly decrease the mineralization of compost. In slightly acid Inceptisols, a higher BC application rate can stabilize compost organic matter but may significantly increase the mineralization of compost.

4.2. Effect of BC on Soil Properties and Fertility Characteristics

In the tropics, naturally rapid mineralization of soil organic matter is a limitation of the practical application of organic fertilizers, despite the application having a positive effect in enhancing soil fertility [32]. Thus, the repeated application of organic materials at high doses can significantly contribute to global warming, plant toxicity, accumulation in plants of heavy metals, and ground and surface water pollution due to nutrient leaching. Some studies have indicated that the simultaneous application of BC and compost resulted in enhanced soil fertility, water holding capacity, crop yield, and C sequestration [43–46]. Schulz and Glaser [46] found that the overall plant growth and soil fertility decreased in the order of compost > BC + compost > mineral fertilizer + BC > mineral fertilizer > control. The combination of BC with mineral fertilizer further increased plant growth during one vegetation period but also accelerated BC degradation during a second growth period. Combination of BC with compost showed the best plant growth and C sequestration but had no effects on N and P retention. The blending of BC with compost has been suggested to enhance the composting performance by adding more stable C and creating a value-added product (BC-compost blend) that can offset both the potential negative effects of the composting system and the pyrolysis BC system [16].

As well as diminishing C mineralization in soils over-fertilized with swine manure compost, we further examined the positive or negative effects of soil nutrients and heavy metals on mineralization and availability after 434 days of incubation. The results suggested that the effects of adding woody BC vary with soil types and elements (Table 4). In SAO soil, 0.5% BC treatment

significantly increased TC, TP, C/N ratio, Ex-K, Ex-Na, and M3-Ca, but had no negative impact on mixed soil. The 1% BC treatment significantly increased TC, C/N ratio, M3-Ca, and Cu, but significantly decreased M3-P, K, Mg, Fe, and Mn. The 2% BC treatment significantly increased TC, TP, C/N ratio, pH, M3-Ca, Cu, Pb, and Zn, but significantly decreased M3-P and Fe. For MAI soil, the 0.5% BC treatment significantly increased TC and pH, and significantly decreased M3-Cu, Pb, and Zn. The 1% BC treatment significantly increased TC, TN, C/N ratio, CEC, and M3-K, but the M3-Cu, Pb, and Zn contents significantly decreased. The contents of TC, TN, and TP, C/N ratio, pH, Ex-K, and M3-K significantly increased, but the contents of M3-Cu and Pb significantly decreased. For SAI soil, pH, TC, C/N ratio, and M3-Zn significantly increased in the 0.5% BC treatment. The 1% BC treatment significantly increased pH, Ex-K, TC, TN, C/N ratio, M3-P, K, and Zn, but significantly decreased in TP, M3-Cu, and Pb. The 2% BC treatment significantly increased pH, Ex-K, TC, TN, C/N ratio, and M3-K, but TP, M3-Cu, and Zn significantly decreased.

Without amendment with compost, the soils used in this study had low plant available contents of some nutrients as well as low CEC. Soils with low CEC are often not fertile and are vulnerable to soil acidification [45]. The CEC of the studied soils followed the order: SAI soil > MAI soil > SAO soil. After incubation, the soil pH of the four treatments in SAO soil (Table 3) were lower than in bulk soil (Table 1), suggesting low soil buffering capacity and that the soil acidification occurred after adding excess manure compost. In a Dystric Cambisol with a loamy-sand texture, a maize (*Zea mays* L.) field trial with five treatments (control, compost, and three BC-compost mixtures with constant compost amount (32.5 Mg/ha) and increasing BC amount, ranging from 5–20 Mg/ha) was conducted [13], and the results demonstrated that total organic C content could be increased by a factor of 2.5 from 0.8% to 2% ($p < 0.01$) at the highest BC-compost level compared with control. TN content only slightly increased and plant-available Ca, K, P, and Na contents increased by factors of 2.2, 2.5, 1.2, and 2.8, respectively. Trupiano et al. [15] indicated that, compared to the addition of compost alone, the compost and BC combination did not improve soil chemical characteristics except for an increase in total C and available P content. These increases could be related to BC capacity to enhance C accumulation and sequestration, and to retain and exchange phosphate ions by its positively charged surface sites. Oldfield et al. [16] suggested that BC recycles C and P; whereas compost recycles C, N, P, and K; and a blend of both resulted in the recycling of C, N, P, and K. Regional differences were found between BC, compost, and BC-compost blend, and the BC-compost blend offered benefits in relation to available nutrients and sequestered C [16].

4.3. BC Addition Rate Effects on Soil Carbon Mineralization and Soil Fertility

Deteriorating soil fertility and the concomitant decline in agricultural productivity are major concerns in many parts of the world [44], and it is a critical problem in Taiwan. Biochar and biochar-compost applications positively impact soil fertility, for example, through their effect on SOC, CEC, and plant available nutrients [43]. [Naeem et al. \[47\]](#) suggested that application of BC in combination with compost and inorganic fertilizers could be a good management strategy to enhance crop productivity and improve soil properties. [Agegnehu et al. \[44\]](#) indicated that as the plants grew, compost and biochar additions significantly reduced leaching of nutrients; separate or combined application of compost and biochar together with fertilizer increased soil fertility and plant growth. Application of compost and biochar improved the retention of water and nutrients by the soil and thereby the uptake of water and nutrients by the plants [44].

The PCA of soil carbon mineralization and soil fertility from the different BC addition treatments in the three soils over-fertilized with compost supported the results discussed above. In SAO soil, 2% BC addition clustered near Group 1 (pH, TC, TP, M3-Pb, M3-Zn, and M3-Cu) whose values were negatively correlated, indicating that 2% BC addition reduced soil C mineralization and stabilized compost organic matter, but slightly increased the soil pH, the content of TC, TP, M3-Pb, M3-Zn, and M3-Cu (smaller, positive loading scores for PC1). In contrast, the 0.5% BC addition clustered closer to Group 2, whose variables were positively correlated, suggesting that the content of TN, M3-P, and M3-K slightly decreased (smaller, negative scores for PC1) with reducing soil C mineralization. In MAI soil, the addition of 1% BC similarly clustered near the negatively correlated Group 1 (pH, TC, TN, TP, M3-P, and M3-K), indicating its positive contribution to soil fertility. In SAI soil, 1% BC addition clustered closer to Group 1 whose variables were positively correlated, suggesting that pH and the content of TC, TN, M3-P, M3-K, and M3-Zn of slightly promoted (smaller, positive scores for PC1) with increasing soil C mineralization. In contrast, the 0.5% BC addition clustered closely to Group 2 whose values were negatively correlated, indicating that 0.5% BC addition increased soil C mineralization and cannot stabilized compost organic matter, but slightly reduced the content of TP, M3-Pb, and M3-Cu (smaller, negative loading scores for PC1). The application of woody BC has potential for stabilizing compost organic matter, diminishing soil C mineralization, and improving soil nutrient availability in soil over-fertilized with compost, but depending on soil type and application rate. Addition of BC in SAO soil and MAI soil led to substantial improvement in physicochemical properties, as well as to significant and insignificant lower C mineralization, respectively (Figure 1 and Table 2). The 0.5% BC addition reduced the content of available P and K, and 2% addition could result in the risk of Cu, Pb, and Zn in SAO soil. In MAI soil, 1% addition increased pH and the content of TC, TN, TP, M3-P, and M3-K. In contrast, BC addition in SAI soil resulted in significant higher C mineralization. The addition of 1% BC

increased in soil pH and the contents of TC, TN, M3-P, M3-K, and M3-Zn, but 0.5% BC addition would reduce the contents of TP, M3-Cu, and M3-Pb.

PCA of the soil properties measured in [Speratti et al. \[48\]](#) found that both BC feedstocks had positive correlations between Ca, Fe, and Mn. Metals such as Fe and Mn, along with lower soil pH, can contribute to the formation of organo-mineral and/or organo-metallic associations that decrease BC mineralization [\[49\]](#). This can increase BC-C stability in the soil, which may improve soil structure [\[50\]](#). In this study, the free Fe oxides (dithionate-citrate-bicarbonate extractable) content was very high (43.1 g/kg) in SAO soil, followed by MAI soil and SAI soil at 8.80 g/kg and 6.96 g/kg, respectively. Along with lower soil pH (< pH 6.0), BC, compost, and soil Fe oxides can contribute to the formation of organo-mineral and/or organo-metallic associations that improve soil structure, stabilize compost organic matter, and decrease mixed-soil C mineralization in SAO soil. The soil pH in MAI soil was highest. The potential of BC for reducing C mineralization in MAI soil was insignificant between the control and BC treatments but showed minor reductions after BC addition treatments. After BC addition, the mixed-soil C mineralization significantly increased, which could contribute to less formation of organo-mineral and/or organo-metallic associations due to the lower amount of Fe oxides and higher soil pH (7.1–7.2). [Berek et al. \[51\]](#), adding two biochars at 2% (w/w) composed of lac tree wood and mixed wood (scrapped wood and tree trimmings) with and without vermicompost or thermocompost at 2% (w/w) in Hawaii in highly weathered soils (Ultisols and Oxisols), indicated that soil acidity, nutrient in the soils, plant growth, and nutrient uptake improved with the amendments compared to the control. [Berek et al. \[51\]](#) also suggested that increases in nutrients and reduced soil acidity by additions of biochar combined with compost were the probable cause, and the use of locally produced biochars and composts was recommended to improve plant nutrient availability in highly weathered soils.

5. Conclusions

In this study, we assessed the capacity of woody BC in soils over-fertilized with compost to stabilize compost organic matter, diminish C mineralization, and improve nutrient availability in three highly weathered and frequent tillage soils in Taiwan (Oxisols, SAO; and Inceptisols, MAI and SAI). The effect of BC addition varied strongly according to the soil type. Soil carbon mineralization significantly decreased with increasing BC addition in SAO soil, and produced insignificant changes in MAI soil, but significant increases in SAI soil. Respiration per unit of TOC (TMC) significantly decreased with increasing BC addition. In this study, a higher BC application rate stabilized and prevented the rapid mineralization of swine manure compost. The soil pH, exchangeable bases, and CEC only showed minor increases with increasing BC addition.

BC addition had a positive effect soil fertility, including TC, TN, TP, M3-P, K, Mg, Fe, Mn, Pb, and Zn, but had slightly positive effect on exchangeable Ca and negative effect on extractable Cu. For improving soil nutrient availability, adding BC generally increased the levels of plant macronutrients and reduced the concentrations of micronutrients. Principal component analysis indicated that adding BC has a positive impact on diminishing soil carbon mineralization (carbon sequestration), sustaining soil fertility, and preventing heavy metals contamination in compost over-fertilized soil, and suggested that adding 0.5% woody BC in SAO soil and adding 1% in MAI and SAI soil should be reasonable and appropriate in Taiwan.

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References

1. Lal, R. Sequestering carbon and increasing productivity by conservation agriculture. *J. Soil Water Conserv.* **2015**, *70*, 55A-62A.
2. Bationo, A.; Kihara, J.; Vanlauwe, B.; Waswa, B.; Kimetu, J. Soil organic carbon dynamics, functions and management in West African agro-ecosystems. *Agric. Syst.* **2007**, *94*, 13-25.
3. Vanlauwe, B.; Bationo, A.; Chianu, J.; Giller, K.E.; Merckx, R.; Mkwunye, U.; Ohiokpehai, O.; Pypers, P.; Tabo, R.; Shepherd, K.D.; Smaling, E.M.A.; Woomer, P.L.; Sanginga, N. Integrated soil fertility management: operational definition and consequences for implementation and dissemination. *Outlook Agric.* **2010**, *39*, 17-

- 24.
4. Renck, A.; Lehmann, J. Rapid water flow and transport of inorganic and organic nitrogen in a highly aggregated tropical soil. *Soil Sci.* **2004**, *169*, 330-341.
 5. Steiner, C.; Teixeira, W.G.; Lehmann, J.; Nehls, T.; Vasconcelos de Macêdo, J.L.; Blum, W.E.H.; Zech, W. Long term effects of manure, charcoal and mineral fertilization on crop production and fertility on a highly weathered Central Amazonian upland soil. *Plant Soil*, **2007**, *291*, 275-290.
 6. Burger, M.; Jackson, L.E. Microbial immobilization of ammonium and nitrate in relation to ammonification and nitrification rates in organic and conventional cropping systems. *Soil Biol. Biochem.* **2003**, *35*, 29-36.
 7. Hornick, S.B.; Sikora, L.J., Sterrett, S.B.; Murray, J.J.; Millner, P.D.; Burge, W.D.; Colacicco, D.; Parr, J.F.; Chaney, R.L.; Willson, G.B. Utilization of sewage sludge compost as a soil conditioner and fertilizer for plant growth. Agriculture Research Service, United State Department of Agriculture, USA. 1984; Agriculture Information Bulletin No. 464, 32 p.
 8. Zech, W.; Senesi, N.; Guggenberger, G.; Kaiser, K.; Lehmann, J.; Miano, T.M.; Miltner, A.; Schroth, G. Factors controlling humification and mineralization of soil organic matter in the tropics. *Geoderma* **1997**, *79*, 117-161.
 9. Kaur, T.; Brar, B.S.; Dhillon, N.S. Soil organic matter dynamics as affected by longterm use of organic and inorganic fertilizers under maize–wheat cropping system. *Nutr. Cycl. Agroecosyst.* **2008**, *81*, 59-69.
 10. Srivastava, A.; Das, S.; Malhotra, S.; Majumdar, K. SSNM-based rationale of fertilizer use in perennial crops: a review. *Indian J. Agric. Sci.* **2014**, *84*, 3-17.
 11. Leita, L.; De Nobili, M. Water-Soluble Fractions of Heavy Metals during

- Composting of Municipal Solid Waste. *J. Environ. Qual.* **1991**, *20*, 73-78.
12. Barrow, C.J. 2012. Biochar: potential for countering land degradation and for improving agriculture. *Appl. Geogr.* **2012**, *34*, 21-28.
 13. Liu, J.; Schulz, H.; Brandl, S.; Miehtke, H.; Huwe B.; Glaser, B. Short-term effect of biochar and compost on soil fertility and water status of a Dystric Cambisol in NE Germany under field conditions *J. Plant Nutr. Soil Sci.* **2012**, *175*, 698-707.
 14. Rogovska, N; Laird, D.; Cruse, R.; Fleming, P.; Parkin, T.; Meek, D. Impact of biochar on manure carbon stabilization and greenhouse gas emissin. *Soil Sci. Soc. Am. J.* **2011**, *75*, 871-879.
 15. Trupiano, D.; Coccozza, C.; Baronti, S.; Amendola, C.; Vaccari, F.P.; Lustrato, G.; Di Lonardo, S.; Fantasma, F.; Tognetti, R.; Scippa, G.S. 2017. The effects of biochar and its combination with compost on lettuce (*Lactuca sativa* L.) growth, soil properties, and soil microbial activity and abundance. *International Journal of Agronomy* vol. 2017, Article ID 3158207, 12 pages.
<https://doi.org/10.1155/2017/3158207>.
 16. Oldfield, T.L.; Sikirica, N.; Mondini, C.; López, G.; Kuikman, P.J.; Holden, N.M. Biochar, compost and biochar-compost blend as options to recover nutrients and sequester carbon. *J. Environ. Manage.* **2018**, *218*, 465-476.
 17. Tsai, C.C.; Chen, Z.S.; Hseu, Z.Y.; Guo, H.Y. Representative soils selected from arable and slope soils in Taiwan and their database establishment. *Soil and Environ.* **1998**, *1*, 73-88. [in Chinese, with English abstract]
 18. Thomas, G.W. Soil pH and soil acidity. In *Methods of Soil Analysis. Part 3. Chemical Methods*; Bigham, J.M. Eds.; Agronomy Society of America and Soil Science Society of America: Madison, WI., USA, 1986; pp. 475-489.

19. Rhoades, J.D. Salinity: Electrical conductivity and total dissolved solids. In *Methods of Soil Analysis. Part 3. Chemical Methods*; Bigam, J.M. Eds.; Agronomy Society of America and Soil Science Society of America: Madison, WI., USA, 1986; pp. 417–435.
20. Gee, G.W.; Bauder, J.W. 1986. Particle-size analysis. In *Methods of soil analysis, Part 1*, 2nd ed.; Klute, A. Eds.; Agronomy Society of America and Soil Science Society of America: Madison, WI., USA, 1986; Agronomy Monograph 9, pp. 383-411.
21. Tabatabai, M.A.; Bremner, J.M. 1991. Automated instruments for determination of total carbon, nitrogen, and sulfur in soils by combustion techniques. In *Soil analysis: Modern instrumental techniques*; Smith, K.A. Eds.; Marcel Dekker: New York, USA, 1991; pp. 261-289.
22. Rhoades, J.D. Cation exchange capacity. In *Methods of Soil Analysis, Part 2*, 2nd ed.; Page, A.L., Miller, R.H., Keeney, D.R., Eds.; Agronomy Society of America and Soil Science Society of America: Madison, WI, USA, 1982; Agronomy Monograph 9, pp. 149-157.
23. Mehlich, A. Mehlich-3 soil test extractant: a modification of Mehlich-2 extractant. *Commoun. Soi. Sci. Plant Anal.* **1984**, *15*, 1409-1416.
24. Hwang, G.S.; Ho, C.L.; Yu, H.Y.; Su, Y.C. 2006. Bamboo vinegar collected during charcoal making with an earth kiln and its basic properties. *Taiwan Journal of Forest Science* **2006**, *21*, 547-557. [in Chinese with English abstract].
25. Lin, Y.J.; Ho, C.L.; Yu, H.Y.; Hwang, G.S. Study on charcoal production with branches and tops wood of *Cryptomeria japonica* using an earth kiln. *Quarterly Journal of Chinese Forestry* **2008**, *41*, 549-558. [in Chinese with English abstract].

26. Tsai, C.C.; Chang, Y.F.; Hwang, G.S.; Hseu, Z.Y. Impact of wood biochar addition on nutrient leaching and fertility in a rural Ultisols of Taiwan. *Taiwanese Journal of Agricultural Chemistry and Food Science* **2013**, *51*, 80-93.
27. Tsai, C.C.; Chang, Y.F. Viability of biochar on reducing C mineralization and improving nutrients status in a compost-treated Oxisols. *Taiwanese Journal of Agricultural Chemistry and Food Science* **2016**, *54*, 74-89.
28. Ribeiro, H.M.; Fangueiro, D.; Alves, F.; Vasconcelos, E.; Coutinho, J.; Bol, R. Carbon-mineralization kinetics in an organically managed Cambic Arenosol amended with organic fertilizers. *J. Plant Nutr. Soil Sci.* **2010**, *173*, 39-45.
29. Zibilske, L.M. 1994. Carbon Mineralization. In *Methods of Soil Analysis, Part 2, Microbiological and Biochemical Properties*; Weaver, R.W., Angle, J.S., Bottomly, P., Eds.; Soil Science of America Inc.: Madison, Wisconsin, USA, 1994; pp. 835-863.
30. Díez, J.A.; Polo, A.; Guerrero, F. Effect of sewage sludge on nitrogen availability in peat. *Biol. Fertil. Soils* **1992**, *13*, 248-251.
31. Méndez, A.; Gómez, A.; Paz-Ferreiro, J.; Gascó, G. Effects of sewage sludge biochar on plant metal availability after application to a Mediterranean soil. *Chemosphere* **2012**, *89*, 1354-1359.
32. Yun, S.I.; Ro, H.M. Natural ^{15}N abundance of plant and soil inorganic-N as evidence for over-fertilization with compost. *Soil Bio. Biochem.* **2009**, *41*, 1541-1547.
33. Lehmann, J.; Pereira da Silva, J.; Steiner, C.; Nehls, T.; Zech, W.; Glaser, B. Nutrient availability and leaching in an archaeological anthrosol and a ferralsol of the central Amazon basin: Fertilizer, manure and charcoal amendments. *Plant Soil* **2003**, *249*, 343-357.

34. Spokas, K.A. Review of the stability of biochar in soils: Predictability of O:C molar ratios. *Carbon Manage.* **2010**, *1*, 289-303.
35. Liang, B.; Lehmann, J.; Solomon, D.; Sohi, S.; Thies J.E.; Skjemstad, J.O.; Luizão F.J.; Engelhard, M.H.; Neves, E.G.; Wirick, S. Stability of biomass-derived black carbon in soils. *Geochim. Cosmochim. Acta* **2008**, *72*, 1598-1610.
36. Streubel, J.D.; Collins, H.P.; Garcia-Perez, M.; Tarara, J.; Granatstein, D.; Kruger, C.E. Influence of contrasting biochar types on five soils at increasing rates of application. *Soil Sci. Soc. Am. J.* **2011**, *75*, 1402-1413.
37. Luo, X.X.; Wang, L.Y.; Liu, G.C.; Wang, X.; Wang, Z.Y.; Zheng, H. Effects of biochar on carbon mineralization of coastal wetland soils in the Yellow River Delta, China. *Ecol. Eng.* 2016, *94*, 329-336.
38. Mukome, F.N.D.; Six, J.; Parikh, S.J. The effects of walnut shell and wood feedstock biochar amendments on greenhouse gas emissions from a fertile soil. *Geoderma* **2013**, *200-201*, 90-98.
39. Hamer, U.; Marschner, B.; Brodowski, S.; Amelung, W. Interactive priming of black carbon and glucose mineralisation. *Org. Geochem.* 2004, *35*, 823-830.
40. Kuzyakov, Y.; Subbotina, I.; Chen, H.; Bogomolova, I.; Xu, X. Black carbon decomposition and incorporation into soil microbial biomass estimated by ¹⁴C labeling. *Soil Biol. Biochem.* **2009**, *41*, 210-219.
41. Liang, B.; Lehmann, J.; Sohi, S.P.; Thies, J.E.; O'Neill, B.; Trujillo, L.; Gaunt, J.; Solomon, D.; Grossman, J.; Neves, E.G.; Luizão, F.J. Black carbon affects the cycling of non-black carbon in soil. *Org. Geochem.* **2010**, *41*, 206-213.
42. Novak, J.M.; Busscher, W.J.; Watts, D.W.; Laird, D.A.; Ahmedna, M.A.; Niandou, M.A.S. Short-term CO₂ mineralization after additions of biochar and switchgrass to

- a Typic Kandudult. *Geoderma* **2010**, *154*, 281-288.
43. Agegnehu, G.; Bass, A.M.; Nelson, P.N.; Muirhead, B.; Wright, G.; Bird, M.I.; Biochar and biochar-compost as soil amendments: effects on peanut yield soil properties and greenhouse gas emissions in tropical North Queensland, Australia. *Agric. Ecosyst. Environ.* **2015**, *213*, 72-85.
44. Agegnehu, G.; Bird, M.; Nelson, P.; Bass, A. The ameliorating effects of biochar and compost on soil quality and plant growth on a Ferralsol. *Soil Res.* **2015**, *53*, 1-12.
45. Agegnehu, G.; Bass, A.M.; Nelson, P.N.; Bird, M.I. Benefits of biochar, compost and biochar–compost for soil quality, maize yield and greenhouse gas emissions in a tropical agricultural soil. *Sci. Total Environ.* **2016**, *543*, 295-306.
46. Schulz, H.; Glaser, B. Effects of biochar compared to organic and inorganic fertilizers on soil quality and plant growth in a greenhouse experiment. *J. Plant Nutrit. Soil Sci.* **2012**, *175*, 410–422.
47. Naeem, M.A.; Khalid, M.; Aon, M.; Abbas, G.; Amjad, M.; Murtaza, B.; Khan, W.D.; Ahmad, N. Combined application of biochar with compost and fertilizer improves soil properties and grain yield of maize, *J. Plant Nutri.* **2017**, *41*, 112-122.
48. Speratti, A.B.; Johnson, M.S.; Martins Sousa, H.; Nunes Torres, G.; Guimarães Couto, E. Impact of different agricultural waste biochars on maize biomass and soil water content in a Brazilian Cerrado Arenosol. *Agronomy* **2017**, *7*, 49.
49. Speratti, A.B.; Johnson, M.S.; Martins Sousa, H.; Dalmagro, H.J.; Guimarães Couto, E. Biochar feedstock and pyrolysis temperature effects on leachate: DOC characteristics and nitrate losses from a Brazilian Cerrado Arenosol mixed with agricultural waste biochars. *J. Environ. Manage.* **2018**, *211*, 256-268.

50. Fang, Y.; Singh, B.; Singh, B.P.; Krull, E. Biochar carbon stability in four contrasting soils. *Eur. J. Soil Sci.* **2014**, *65*, 60-71.
51. Berek, A.K.; Hue, N.V.; Radovich, T.J.K.; Ahmad, A.A. Biochars improve nutrient phyto-availability of Hawai'i's highly weathered soils. *J. Agronomy* **2018**, *8*, 203-220.

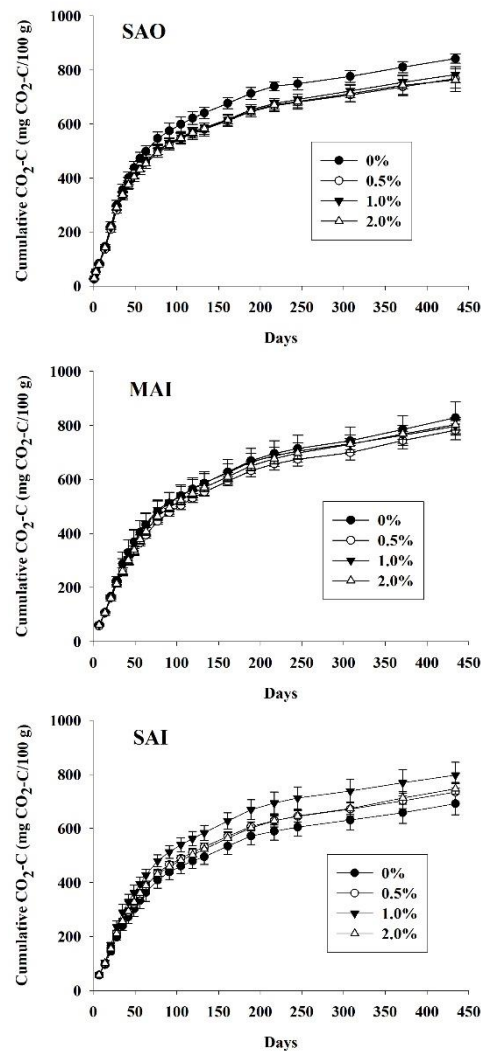


Figure 1. Cumulative CO₂-C (mg CO₂-C/100 g soil) from the three studied soils treated with 0%, 0.5%, 1.0%, and 2.0% woody BC. Error bars indicated the standard deviation of the mean.

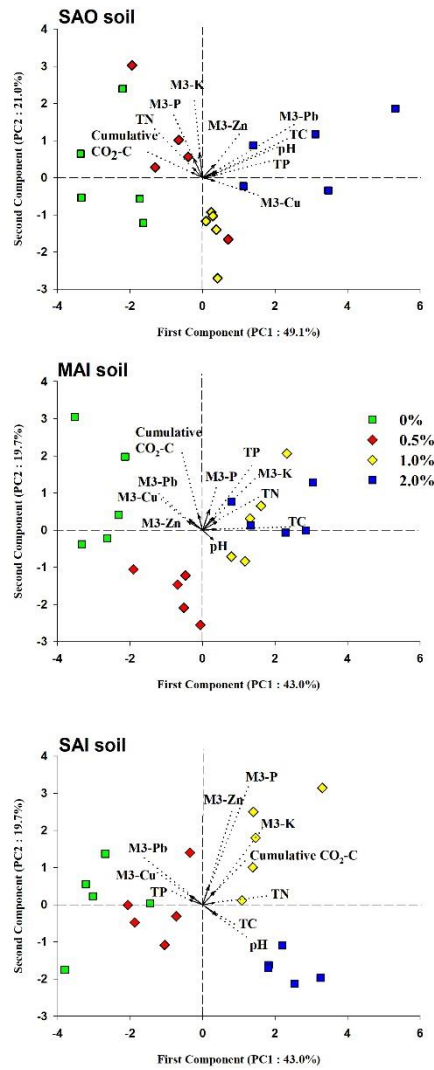


Figure 2. Principal component analysis based on the soil chemical characteristics and cumulative CO₂-C (mg CO₂-C/100g soil) after 434-d incubation period in SAO, MAI, and SAI soils treated with 0%, 0.5%, 1.0%, and 2.0% woody BC.

Table 1. Characteristics of biochar, compost, and three studied soils.

Characteristics	Biochar (BC)	Compost	Pc Soil (SAO)	Eh Soil (MAI)	An Soil (SAI)
pH	9.9 ¹	8.41 ¹	6.1/5.0 ³	7.5/7.2 ³	6.5/6.2 ³
EC (dS/m)	0.77 ¹ /1.36 ²	3.79 ¹	0.45	2.21	0.81
Sand (%)			11	24	33
Silt (%)			30	36	33
Clay (%)			59	39	34
Soil Texture			Clay	Clay loam	Clay loam
Total C (%)	82.5	23.3	2.03	1.11	0.94
Total N (g/kg)	6.99	22.6	2.71	2.32	1.58
Total P (g/kg)	0.55	10.2	1.16	0.98	0.77
Ex. K (cmol(+)/kg soil)	1.91	6.43	0.32	0.29	0.21
Ex. Na (cmol(+)/kg soil)	1.26	1.09	0.31	0.26	0.37
Ex. Ca (cmol(+)/kg soil)	3.62	2.70	4.85	2.94	2.24
Ex. Mg (cmol(+)/kg soil)	0.40	2.72	0.64	0.80	0.36
CEC (cmol(+)/kg soil)	5.20	19.7	8.58	11.5	14.2
BS (%)	100	69	71	37	22
M3-P (mg/kg)	96.6	6874	163	236	94.0
M3-K (mg/kg)	616	8911	68.4	108	94.1
M3-Ca (g/kg)	4.09	14.5	2.03	8.22	2.99
M3-Mg (mg/kg)	278	3972	143	344	401
M3-Fe (mg/kg)	65.5	396	524	589	1199
M3-Mn (mg/kg)	20.9	188	29.0	213	185
M3-Cu (mg/kg)	0.02	6.22	9.77	9.95	3.17
M3-Pb (mg/kg)	ND ⁴	1.23	10.8	11.7	1.54
M3-Zn (mg/kg)	0.35	62.4	20.4	7.98	5.28

¹ The pH and electrical conductivity (EC) of biochar and compost were measured using 1:5 solid: solution ratio after shaking for 30 min in deionized water; ² Biochar EC was measured after shaking biochar-water mixtures (1:5 solid: solution ratio) for 24 hr; ³ Soil pH was determined in soil-to-deionized water ratio of 1:1 (g mL⁻¹) and in soil-to-1N KCl ratio of 1:1 (g mL⁻¹); ⁴ ND = not detected.

Table 2. CO₂-C evolved (mg C/100 g dry weight) and total mineralization coefficient (TMC) for control and amended soils after incubation experiment¹.

Rate	CO ₂ evolved (mg C/100 g dry weight)	TMC (mg CO ₂ -C/g C)
<i>SAO Soil</i>		
0%	842 ± 8.7 A	333 ± 3.4 A
0.5%	768 ± 18 B	278 ± 6.4 B
1.0%	783 ± 15 B	253 ± 4.7 C
2.0%	763 ± 21 B	207 ± 5.7 D
<i>MAI Soil</i>		
0%	829 ± 30 A	526 ± 19 A
0.5%	782 ± 18 A	423 ± 9.6 B
1.0%	797 ± 17 A	417 ± 8.7 B
2.0%	803 ± 10 A	355 ± 4.5 C
<i>SAI Soil</i>		
0%	692 ± 20 C	455 ± 14 A
0.5%	735 ± 18 BC	452 ± 11 A
1.0%	798 ± 24 A	464 ± 14 A
2.0%	747 ± 10 B	365 ± 4.9 B

¹ Each value is the average ± standard deviation from three independent experiments.

Means compared within a column followed by a different uppercase letter are significantly different at $p < 0.05$ using a one-way ANOVA (multiple comparisons vs. studied soil + 0% biochar as a control).

Table 3. Mean values of total soil carbon (TC), nitrogen (TN), and phosphorus (TP), soil pH, exchangeable bases (K, Na, Ca, and Mg), and cation exchangeable capacity (CEC) of four treatments of three soils after 434-day incubations¹.

Rate	pH	Ex-K	Ex-Na	Ex-Ca	Ex-Mg	CEC	TSC	TSN	TSP	C/N	
		-----coml(+)/kg soil-----					-----g/kg-----				
<i>SAO Soil</i>											
0%	5.66 b	2.55 b	0.72 b	14.9 a	3.58 a	16.4 a	23.9 c	4.37 ab	1.55 c	5.5 c	
0.5%	5.75 b	2.87 a	0.91 a	15.0 a	3.73 a	16.4 a	28.0 b	4.43 a	1.77 b	6.3 b	
1.0%	5.76 b	2.40 b	0.73 b	14.4 a	3.36 a	16.0 a	31.8 a	4.28 b	1.69 bc	7.4 a	
2.0%	5.93 a	2.55 b	0.63 b	15.5 a	3.41 a	16.3 a	34.5 a	4.27 b	2.21 a	8.1 a	
<i>MAI Soil</i>											
0%	7.53 c	2.64 b	0.66 a	22.9 a	3.37 a	9.7 b	18.2 c	3.64 b	0.88 bc	5.0 c	
0.5%	7.58 b	2.92 b	0.68 a	25.5 a	3.78 a	10.1 b	21.9 b	3.62 b	0.75 c	6.0 bc	
1.0%	7.58 bc	2.92 ab	0.68 a	25.5 a	3.78 a	10.7 a	22.2 b	4.06 a	1.05 ab	5.5 b	
2.0%	7.65 a	3.24 a	0.76 a	25.9 a	3.75 a	10.0 b	32.4 a	4.15 a	1.18 a	7.8 a	
<i>SAI Soil</i>											
0%	7.04 c	2.14 c	0.59 a	13.9 a	4.24 a	13.4 a	13.7 c	2.86 b	1.26 a	4.8 c	
0.5%	7.11 b	2.30 bc	0.54 a	15.3 a	4.52 a	13.3 a	18.3 b	2.89 b	1.11 a	6.3 b	
1.0%	7.14 b	2.61 a	0.62 a	15.6 a	4.56 a	13.6 a	21.4 b	3.06 a	0.88 b	7.0 b	
2.0%	7.24 a	2.45 ab	0.54 a	14.9 a	4.23 a	12.8 b	26.6 a	3.07 a	0.64 c	8.7 a	

¹ Each value is the average from three independent experiments. Means compared within a column followed by a different lowercase letter are significantly different at $p < 0.05$ using a one-way ANOVA (multiple comparisons vs. studied soil + 0% biochar as a control).

Table 4. Mean values of soil fertility characteristics (Mehlich 3 extraction) (mg/kg) of four treatments of three soils after 434-day incubations¹.

Rate	P	K	Ca	Mg	Fe	Mn	Cu	Pb	Zn
<i>SAO Soil</i>									
0%	645 a	461 a	2701 b	533 a	953 a	5.7 ab	8.64 c	10.3 b	26.8 b
0.5%	653 a	467 a	3216 a	556 a	948 a	37.2 a	9.02 bc	10.2 b	28.6 b
1.0%	486 b	408 b	3118 a	444 b	739 b	31.6 c	9.36 ab	11.0 b	27.8 b
2.0%	537 b	457 a	3188 a	474 ab	777 b	3.5 bc	9.83 a	12.3 a	30.6 a
<i>MAI Soil</i>									
0%	769 ab	474 c	7594 a	636 ab	694 ab	286 a	8.73 a	12.9 a	12.2 a
0.5%	671 b	481 bc	7799 a	611 b	621 b	271 a	7.79 b	11.4 b	10.6 b
1.0%	832 a	545 a	7142 a	712 a	739 a	310 a	7.60 b	10.7 b	10.3 b
2.0%	795 a	534 ab	7697 a	660 ab	707 ab	301 a	7.66 b	11.3 b	11.1 ab
<i>SAI Soil</i>									
0%	476 b	384 c	3569 a	750 a	1257 a	197 a	1.70 a	0.84 a	9.48 c
0.5%	462 b	392 bc	3292 a	712 a	1147 a	186 a	1.66 a	0.67 ab	10.1 b
1.0%	564 a	474 a	3313 a	759 a	1200 a	196 a	1.54 b	0.57 bc	11.1 a
2.0%	470 b	437 ab	3648 a	726 a	1183 a	194 a	1.29 c	0.48 c	9.53 bc

¹ Each value is the average from three independent experiments. Means compared within a column followed by a different lowercase letter are significantly different at $p < 0.05$ using a one-way ANOVA (multiple comparisons vs. studied soil + 0% biochar as a control).