Preprints.org

Article

Not peer-reviewed version

Biochar and Organic Fertilizer Co-Application Enhances Soil Carbon Priming Increasing CO2 Fluxes in Two Contrasting Arable Soils

Magdalena Bednik , Agnieszka Medyńska-Juraszek * , Irmina Ćwieląg-Piasecka

Posted Date: 18 September 2023

doi: 10.20944/preprints202309.1124.v1

Keywords: biochar; soil respiration; incubation experiment; CO2 efflux

Preprints.org is a free multidiscipline platform providing preprint service that is dedicated to making early versions of research outputs permanently available and citable. Preprints posted at Preprints.org appear in Web of Science, Crossref, Google Scholar, Scilit, Europe PMC.

Copyright: This is an open access article distributed under the Creative Commons Attribution License which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

Preprints (www.preprints.org) | NOT PEER-REVIEWED | Posted: 18 September 2023 doi:10.20944/preprints202309.1124.v1

Disclaimer/Publisher's Note: The statements, opinions, and data contained in all publications are solely those of the individual author(s) and contributor(s) and not of MDPI and/or the editor(s). MDPI and/or the editor(s) disclaim responsibility for any injury to people or property resulting from any ideas, methods, instructions, or products referred to in the content.

Article

Biochar and Organic Fertilizer Co-Application Enhances Soil Carbon Priming Increasing CO_2 Fluxes in Two Contrasting Arable Soils

Magdalena Bednik, Agnieszka Medyńska-Juraszek * and Irmina Ćwieląg-Piasecka

Wroclaw University of Environmental and Life Sciences, Institute of Soil Sciences, Plant Nutrition and Environmental Protection, Grunwaldzka 53 St., 50-375 Wroclaw, Poland; magdalena.bednik@upwr.edu.pl (M.B.); irmina.cwielag-piasecka@upwr.edu.pl (I.Ć.-P.)

* Correspondence: agnieszka.medynska-juraszek@upwr.edu.pl (A.M.-J.)

Abstract: Biochar soil amendment along with non-tillage agriculture, are often proposed as a strategy for organic carbon sequestration in soil. How the quality of biochar might influence the priming effect on soil organic matter mineralization and whether the addition of fresh organic matter will affect its stability in soil is still questionable. In the study, six biochars of different biomass origin and three exogenous organic matter sources were added to two distinct arable soils. CO_2 emission was monitored for 100 days of incubation and CO_2 flux was estimated. Results showed that biochar application increased soil CO_2 fluxes. The highest peaks were recorded in treatments with food waste biochars, suggesting that this feedstock serves as sources of labile C fractions to soil microbes. Co-application of raw organic materials (manure and fresh clover biomass) enhanced CO_2 emission and estimated carbon losses, especially in sandy soil with low organic carbon content. Biochar properties and content of labile C fractions can stimulate CO_2 emission, however in a long-term period this contribution is negligible. Findings of our study showed that more attention should be paid to priming effects caused by addition of exogenous organic matter e.g. fertilizers or cover crops when applied to biochar amended soils.

Keywords: biochar; soil respiration; incubation experiment; CO_2 efflux

1. Introduction

Some of intensive agriculture strategies contributes to the increase of greenhouse gases (GHG) emission and biochar has been widely recommended as a soil amendment moderating global climate change. Produced by the thermochemical conversion of organic residues in oxygen–limited conditions, biochar (BC) is highly resistant to degradation due to its recalcitrant nature [1,2]. Addition of biochar to soil alters physicochemical properties, e.g. porosity, bulk density, pH, carbon (C) and nitrogen (N) content or water holding capacity, which impact soil CO_2 emissions [3–5]. BCs obtained from various feedstock, under different temperature regimes of pyrolysis, have various properties [6] and their effects after incorporation into the soil may greatly vary with local environmental conditions and cultivation systems [7,8]. In general, biochars produced from plant biomass e.g. straw or wood are rich in recalcitrant C forms and are able to sequester more C in soil in comparison with biochars derived from animal manures [9] or organic residues e.g. food wastes [10]. Biochar application to soil may increase carbon sequestration due to the inputs of recalcitrant organic C [6,11,12], however the effects of biochar application on the soil GHG emission is questionable. Results presented in meta-analysis show that biochar application significantly increased soil CO_2 fluxes by 22.14%, thus contributing to the global warming potential [13]. The mechanisms behind this process are still not well understood. The exogenous inputs of labile C sources such as fresh plant residues or dissolved organic carbon from pyrogenic organic matters (POMs) to soil induce positive priming effect with increasing CO_2 emission [14,15]. On the other hand, some studies reported negative priming and suppression of soil CO_2 emission due to reduced enzymatic activity and the precipitation of CO_2 on the biochar surface [16]. Furthermore, the direction of priming effects may

© 2023 by the author(s). Distributed under a Creative Commons CC BY license.

Preprints (www.preprints.org) | NOT PEER-REVIEWED | Posted: 18 September 2023 doi:10.20944/preprints202309.1124.v1

change over the time from amendment application [17]. To model the possible contribution of biochar in GHG mitigation and its stability in soil it is necessary to include many different factors, that might affect biochar behavior in soil under different climatic conditions and cultivation systems [18–20]. It is also important to answer the question whether all biochars contribute to the GHG mitigation process equally, or maybe more attention should be paid to the final product in terms of finding a proper material for effective CO_2 emission mitigation from cultivated soils. As non-tillage and organic farming strategies to increase the carbon sink in agricultural soils are receiving a lot of attention, biochar co-application with sustainable tillage practices might be a proper approach supporting greenhouse gases emission mitigation from arable soils [21,22]. The knowledge about the effects of co-application of biochar, raw crop residues and organic fertilizers e.g. manure and compost on CO_2 emission from arable soils is limited.

An increasing number of studies have demonstrated that soil organic carbon (SOC) decomposition can be influenced by exogenous organic C (EXOC) input through the priming effect. For example, Sun et al. [23] claimed that the addition of EXOC significantly enhanced native SOC decomposition by 47.5% with the highest value in cropland soils (60.9%) and the lowest value in forest soils (26.2%). SOC decomposition contributes greatly to the CO_2 emission from agricultural activities. This study gives insight into the state of knowledge about biochar CO_2 mitigation potential in soil, answering the question whether the process of carbon sequestration by biochar can be disturbed by application of other exogenous organic matter. We hypothesized that labile organic matter (LOM) from cover crops residues or organic fertilizers e.g. manures or compost may change the C-sequestration potential in biochar-amended soil as both types of C sources will contribute to the SOC priming effect. This may induce changes in native mineralization process of organic matter, which in turn will increase or decrease CO_2 flux from soil. Moreover, the presence of raw organic residues and labile C fractions may influence biochar mineralization rate and this can be indicated by CO_2 emission during respiration processes [24]. Previous studies mainly have examined biochar produced from diverse biomass streams, including forestry and agriculture wastes. As a novel approach, in this study the recalcitrance of conventional straw and wood biochar is compared with biochar produced from kitchen wastes - mainly food scraps, fruit and vegetable peels and all the wastes selectively collected for the composting process. Biochar from food waste can become a sustainable replacement of other black carbons, and food waste conversion to biochar has been widely studied as a method to sequester carbon and mitigate the substantial greenhouse gas emissions associated with wasted food signed in United States 2030 Food Loss and Reduction Goal [25]. Our previous study showed that food waste biochar contains more labile carbon compounds prone to oxidation and thus can contribute to the process of CO_2 emission from BC amended soil or enhance soil organic matter (SOM) mineralization [10], and both processes can be monitored by measuring CO_2 efflux from soil. Biochar recalcitrance is expected to last hundreds of years [1,2], but residence calculation in most of the studies do not take into account the carbon loss due to enhanced mineralization of biochar in the presence of raw organic matter delivered to soil with organic fertilization on non-tillage cultivation strategies.

The paper assesses the effects of labile carbon content in biochar on CO_2 efflux from soils amended with biochar derived from different waste materials. It also verifies the questionable effect of exogenous raw organic matter on biochar recalcitrance under conditions imitating non-tillage soil cultivation, promoted as a sustainable method of soil conservation and reduction of agriculture impact on GHG emission.

2. Materials and Methods

2.1. Incubation experiment setup

The incubation experiment was carried out with two different soil types, six biochars and three types of additional organic matter mixed with soil. Both soils used in this study are common in Central Europe and the main intended difference between them is the texture – silt loam (SiL), and loamy sand (SA) (Table 1). Samples were collected from the topsoil (0-25 cm) layer of arable land in

Preprints (www.preprints.org) | NOT PEER-REVIEWED | Posted: 18 September 2023 doi:10.20944/preprints202309.1124.v1

two locations close to Trzebnica, Poland (51°15'46.8"N 17°06'13.3"E and 51°24'13.2"N 17°06'31.6"E). Prior to the incubation experiment, moist soil samples were stored in closed containers in the refrigerator, at 4 °C to keep soil biological activity. Six different feedstocks commonly produced in urban areas and farmlands were chosen for biochar production: kitchen wastes (BC1), cut grass (BC2), coffee grounds (BC3), wheat straw (BC4), sunflower husks (BC5) and beech wood chips (BC6). All the feedstock are accepted as a permissible biomass for biochar production in Europe [26]. Before the pyrolysis, feedstock materials were air-dried and stored at ambient air humidity. Production of biochars was performed in September 2020 at Wroclaw University of Technology. Pyrolysis was conducted in the nitrogen atmosphere, at 550 °C and the conditions remained constant for every type of feedstock. The duration of the process was 60 minutes for each biomass. Organic matter applied in this experiment included compost (CO), cattle manure (MA) and fresh legume biomass (LE). Compost was produced in a home composter located in Wroclaw, Poland, from kitchen waste (vegetable and fruit peels) and garden waste (cut grass, leaves, small twigs). Dried cow manure in a form of granules was purchased from Fertigo fertilizer supplier (Suchy Las, Poland). Legume biomass consisting of whole plants of white and red clover (*Trifolium repens* L., *Trifolium pratense* L.) was collected from green areas in Wrocław, Poland.

Biochars, compost and manure were air dried and grounded in a soil mill to obtain particle sizes <2 mm. Fresh legume biomass was carefully washed with distilled water, to avoid the pollution of incubation systems and cut into pieces <2 mm with scissors to obtain materials with uniform fraction sizes. Biochars and organic materials with particle sizes <2 mm were mixed thoroughly with the soil in the following proportions: biochars 2% (v/w) (corresponding to additions of 0.565 – 0.915 t ha^{-1} depending on biochar's bulk density, assuming the thickness of plowing layer 25 cm and soil density 1.50 g cm^{-1}) and organic matter 1% (w/w) (corresponding to the dose of 37.5 t ha^{-1}). Then, 100 g of each mixed sample was placed in a 550-mL glass vessel. All treatments are summarized in Table 2. Vessels were incubated in a place protected from direct sunlight, at a constant air temperature of 22 °C. They were left open most of the time to allow soil respiration, with possibility to close tightly if needed. Moisture of the incubated material was maintained at approx. 20% by weight, by watering with distilled water when necessary.

Table 1. Summary of the treatments in incubation experiment.

Description	Abbreviation	Dose equivalent [t ha^{-1}]
Sandy soil without amendments	SA	-
Sandy soil with 6 types of biochar	SA BC1 - SA BC6 [1]	0.57 – 0.92 (2% v/w)
Sandy soil with 6 types of biochar and three types of organic matter	SA BC1- BC6 CO for compost SA BC1- BC6 MA for manure SA BC1- BC6 LE for legumes	biochar: 0.57 – 0.92 (2% v/w) organics: 37.50 (1% w/w)
Silt loam soil without amendments	SiL	-
Silt loam soil with 6 types of biochar	SiL BC1 - SiL BC6	0.57 – 0.92 (2% v/w)
Silt loam soil with 6 types of biochar and three types of organic matter	SiL BC1- BC6 CO for compost SiL BC1- BC6 MA for manure SiL BC1- BC6 LE for legumes	biochar: 0.57 – 0.92 (2% v/w) organics: 37.50 (1% w/w)

[1] - respectively for all six biochar types

2.2. Analysis of substrates

To determine standard characteristics of the substrates, samples of the soils, biochars, compost and manure were air-dried, sieved with 2 mm mesh and further prepared following the specific methodologies of analyses. The pH was determined in H_2O in 1:5 suspension (v/v) using pH-meter (Mettler-Toledo, Graifensee, Switzerland). For soil samples, particle size distribution was measured using mesh and hydrometer method, and content of calcium carbonates as an equivalent was determined using Scheibler apparatus (according to DIN 18129, ISO 10693 method) – approach frequently applied in Poland to determine $CaCO_3$ content in soil samples [27,28]. Cation exchange

Preprints (www.preprints.org) | NOT PEER-REVIEWED | Posted: 18 September 2023 doi:10.20944/preprints202309.1124.v1

capacity was measured by MP-AES 4200 Spectrometer (Agilent Technologies, Santa Clara, CA, USA) after extraction with 1 M ammonium acetate. For biochars, a modification of the method was used, based on rinsing the samples with isopropanol, as proposed by Munera-Echeverri et al. [29]. Total organic carbon and total nitrogen was measured on enviro TOC/TN analyzer (Elementar, Langenselbold, Germany). Ash content was calculated based on mass loss after sample combustion in a muffle furnace at 550 °C (Czylok, Jastrzębie Zdrój, Poland). Characteristics of the soils, biochar and organic materials used as substrates for the experiment are summarized in Table 2.

Table 2. General properties of the soils, biochars and organic amendments used in the experiment.

	Abbr. in paper	Substrate	pH (H_2O)	CEC[1] [cmol (+) kg^{-1}]	TOC [g 100 g^{-1}]	TN [g 100 g^{-1}]	C:N	Ash [%]	$CaCO_3$ [%]
Soils	SA	Loamy sand sand / silt / clay [%] 81 / 17 / 2	4.62	1.62	0.72	0.04	16.9	n/a	0.25
	SiL	Silt loam sand / silt / clay [%] 22 / 64 / 15	6.40	11.70	0.99	0.07	13.7	n/a	0.00
Biochars	BC1	Food wastes	9.41 ± 0.05	228	53.0 ± 1.10	0.98 ± 0.02	54.1	10.1 ± 1.00	n/a
	BC2	Cut green grass	10.43 ± 0.04	228	52.0 ± 1.00	2.70 ± 0.05	19.3	31.3 ± 3.10	n/a
	BC3	Coffee grounds	6.91 ± 0.07	35.0	68.0 ± 1.40	3.60 ± 0.07	18.9	3.70 ± 0.40	n/a
	BC4	Wheat straw	7.20 ± 0.13	7.41	76.0 ± 1.50	0.24 ± 0.05	317	1.30 ± 0.1	n/a
	BC5	Sunflower husks	10.29 ± 0.02	35.3	78.0 ± 1.60	0.63 ± 0.01	124	5.60 ± 0.60	n/a
	BC6	Beech wood chips	6.96 ± 0.07	22.7	70.0 ± 1.40	1.40 ± 0.03	50.0	9.80 ± 1.00	n/a
Organics smatter	CO	Compost	5.66	10.8	17.6	2.01	8.77	n/a	n/a
	MA	Manure	7.00	n/a	28.0	4.00	7.00	n/a	n/a
	LE	Legume biomass	n/a	n/a	51.8	n/a	n/a	12.20	n/a

[1] in table: Abbr. = abbreviation, CEC = cation exchange capacity, TOC = total organic carbon, TN = total nitrogen, n/a = not applicable. Values are means ± standard deviation (SD) from three replicates.

2.3. Respiration measurements

Soil respiration was measured during the incubation as an amount of CO_2 released by the unit of soil + treatment in the unit of time. To determine this value, a portable gas detector with infrared (IR) sensor dedicated for CO_2 concentration measurements (GasHunter II, Alter S.A., Tarnowo Podgórne, Poland) was used. Measuring range of the device was 0-5000 ppm with a resolution of 50 ppm. The assumption was that in closed vessels concentration of CO_2 will increase over time only as an effect of the soil respiration. The test began with the sealing of the jars for one hour, to allow CO_2 to accumulate. Then, carbon dioxide concentrations were measured in the air inside the vessel, by inserting the probe with the pump through a dedicated valve in the cap and collecting the sample for 60 seconds (please see the scheme below – Figure 1).

Preprints (www.preprints.org) | NOT PEER-REVIEWED | Posted: 18 September 2023
doi:10.20944/preprints202309.1124.v1

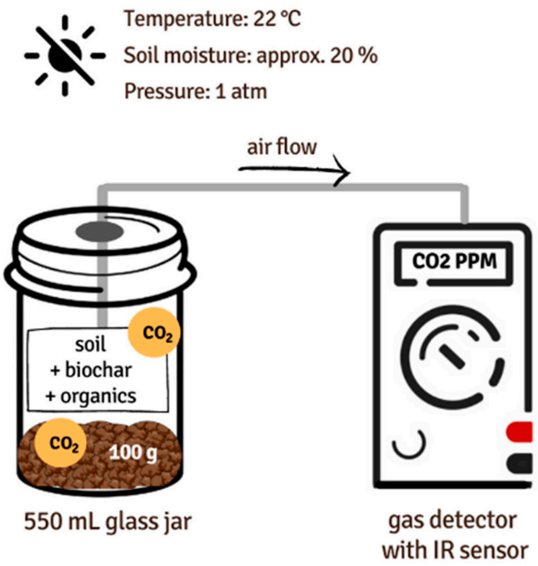

Figure 1. Scheme of the soil respiration measurements.

Measurements were conducted in three replicates and the final value is an average. Zero value as a reference point was CO_2 concentration in the air in the laboratory. Measurements were carried out at 1, 3, 5, 7, 14, 35, 42, 55, 70, 84 and 98 days of an incubation. After this time the CO_2 was constant and at a very low rate, close to the detection limit of the device. The temperature and air humidity in the room were constant during the measurements (22 °C, humidity approx. 50%). To control these conditions, automatic sensors of air parameters were used and conditions in the incubation room were adjusted by air-conditioning if needed.

Values recorded by CO_2 sensor were displayed in ppm, therefore it was necessary to perform some calculations. We adapted protocol proposed by Fierer [30], based on the universal gas law to convert ppm CO_2 to C-CO_2 [μg]. We assumed that the pressure and temperature were constant during all the measurements (1 atm and 22 °C), and volume of free space in the vessel was 490 mL (the remaining volume from 550 mL was taken up by soil). To calculate the number of moles of air in the vessel (n) the modified ideal gas law was applied:

$$n = \frac{pV}{RT}$$

Where V = volume of air in the vessel (490 mL), p = pressure (1 atm), R = const. [82.05 mL atm mol^{-1} K^{-1}], T = temperature in K = 273 + °C [273 + 22 = 293 K]. According to the calculations each vessel contained 20.38 mmol of air. To determine the exact amount of C-CO_2 released by the incubated mixture, the following equation was applied, on the basis of laboratory protocol by Fierer [30].

$$\mu g\ C - CO_2 = mmol\ air\ \times ppm\ CO_2\ \times 10^{-3}\frac{mol}{mmol} \times 12\ \frac{\mu gC}{\mu molC}$$

Calculated values relate to μg of C-CO_2, released by 100 g of soil in one hour.

Graphs and figures were prepared with GraphPad Prism 5 Software for Windows (GraphPad Software Inc., San Diego, CA, USA). Calculations of results were performed using MS Excel Software (Microsoft, Redmond, WA, USA) and GraphPad Prism 5 Software for Windows.

2.4. Carbon loss estimation

Carbon loss was balanced as a percentage of carbon released during the respiration measurements in relation to whole carbon pool present in incubated vessels, originated from native soil organic matter, biochars and organic amendments (compost, manure or legume biomass). Results

Preprints (www.preprints.org) | NOT PEER-REVIEWED | Posted: 18 September 2023 doi:10.20944/preprints202309.1124.v1

of cumulative respiration were calculated to obtain mass of released carbon. Carbon content in soil, biochar and organic amendment was determined before the experiment, in dry substrates. Then, the amount of C introduced with biochar and organic amendment required the following calculations.

For biochars, calculations were based on carbon content in dry substrates and bulk density of biochars, using the formula:

$$BCC = \rho \times V \times \%C\ [g]$$

Where BCC = carbon originating from biochars, ϱ = bulk density of biochars [g cm^{-3}], V = amount of biochar in vessel (2 cm^3), %C = carbon content in biochars.

For compost, manure and legume biomass, calculations included dry mass and carbon content in the substrate:

$$COA = m \times d.m. \times \%C\ [g]$$

Where COA = carbon originating from organic amendment, m = mass of amendment in vessel (1 g), d.m. = content of dry mass [%], %C = carbon content in dry mass of the amendment.

Then, carbon pool introduced from organic amendments, biochars and present in soil was summarized to obtain total C content in incubated vessels (g 100 g^{-1} soil). Total carbon content was compared with carbon losses during the respiration, what allowed to express the loss as a percentage of carbon pool. Results were summarized in the Table S1 – supplementary.

3. Results

3.1. Effect of soil and biochar type on respiration

During 100 days of incubation, in each variant, regardless of soil characteristics and type of biochar, the highest respiratory activity was indicated during the first 7 days. After the initial peaks and some fluctuations of respiration, strong decreases of CO_2 release were noted and after about 10 days of the experiment recorded values were definitely lower and stable. Despite similar trends over time (the highest CO2 evolution in the first week of incubation, followed by sharp decrease and stabilization of the values), meas-ured values differed between sandy (SA) and loamy (SiL) soil. Carbon dioxide evolu-tion tended to be higher on SA, compared with SiL amended with analogous doses and types of biochars (Figure 2 and Figure 3). The largest CO2 emission was from sandy soil with BC1, up to 162 µg C-CO2 h−1 100 g−1 of soil (mean value), recorded on the 1st day of in-cubation with biochar made from food waste (Figure 2). The treatment with BC1 was as-sociated with the highest respiration also in loamy soil (Fig 3). The second biochar that led to remarkably higher respiration rate on both soil types was derived from coffee grounds (BC3). The pattern between all the treatments reveals that the non-amended control soils had lower respiration rate than samples incubated with biochars. Moreo-ver, CO2 evolution from SiL tends to be lower than from sand mixed with the same bi-ochar types, regardless of the fact that loamy soil had higher initial carbon content (0.99 g 100 g−1 vs. 0.72 g 100 g−1 on SA) (Table 1).

Carbon losses were calculated on the basis of TOC in soil and amendments, com-pared with the amount of carbon lost as C-CO2 during the respiration. Although in none of the treatments calculated C depletion exceeded 1%, there were some clear dif-ferences between variants of the experiment. During 100 days of incubation, sandy soil (SA) amended only with biochars lost from 0.22% (SA + BC5) to 1.01% (SA + BC1) of the total organic carbon present in incubated mixture, whereas silt loam (SiL) mixed with the same biochar types exhibited lower declines of C content in the range of 0.21% (SiL + BC5) to 0.52% (SiL + BC1) (Table S1 – supplementary).

Preprints (www.preprints.org) | NOT PEER-REVIEWED | Posted: 18 September 2023 doi:10.20944/preprints202309.1124.v1

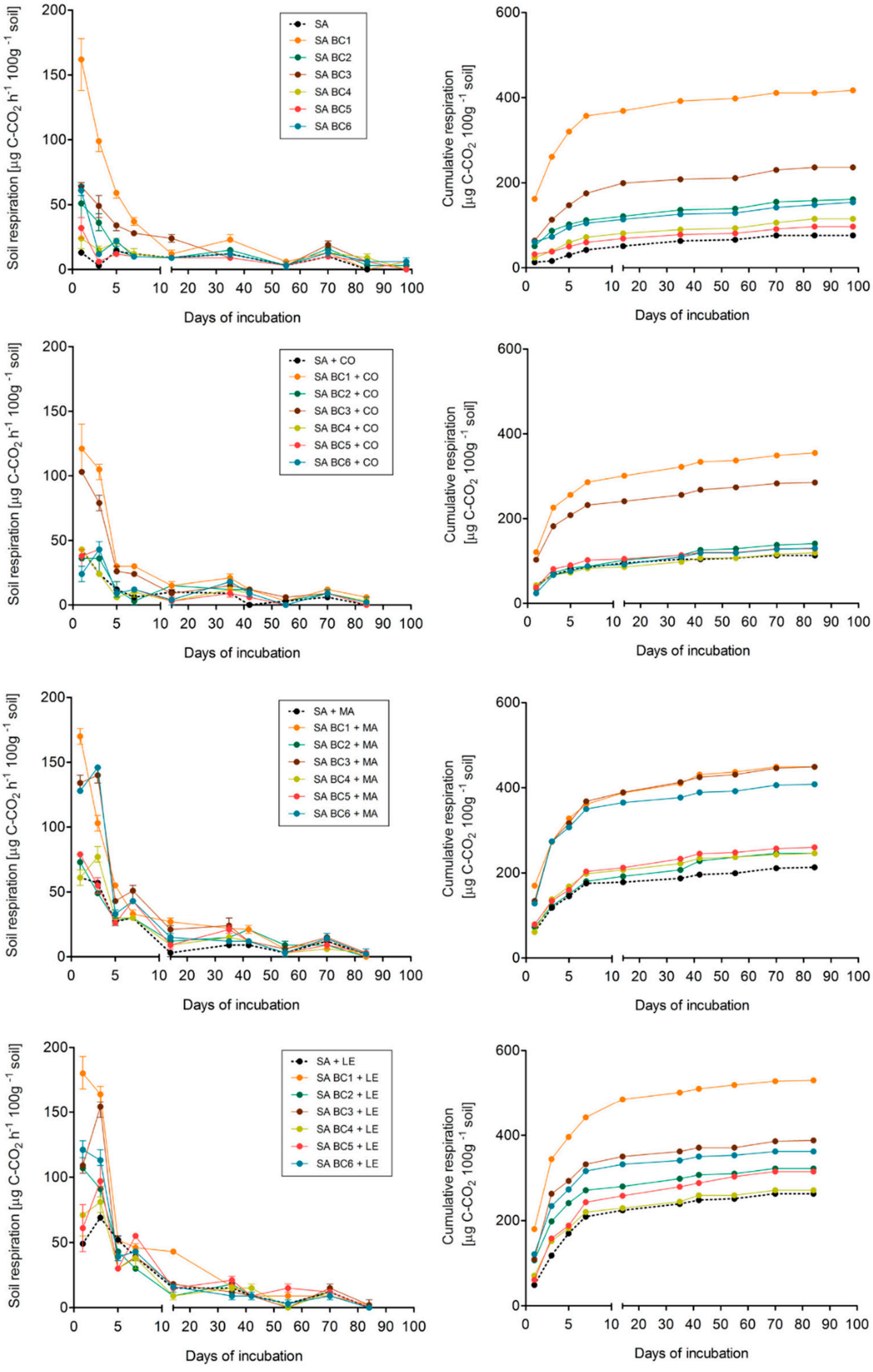

Figure 2. Respiration of sandy soil (SA) amended with biochars (BC1-BC6) and organic materials: compost (CO), cattle manure (MA) and legume biomass (LE). Point = mean, bars = minimum and maximum. BCs origins: BC1 = kitchen waste, BC2 = cut grass, BC3 = coffee grounds, BC4 = wheat straw, BC5 = sunflower husks, BC6 = beech wood chips.

Preprints (www.preprints.org) | NOT PEER-REVIEWED | Posted: 18 September 2023 doi:10.20944/preprints202309.1124.v1

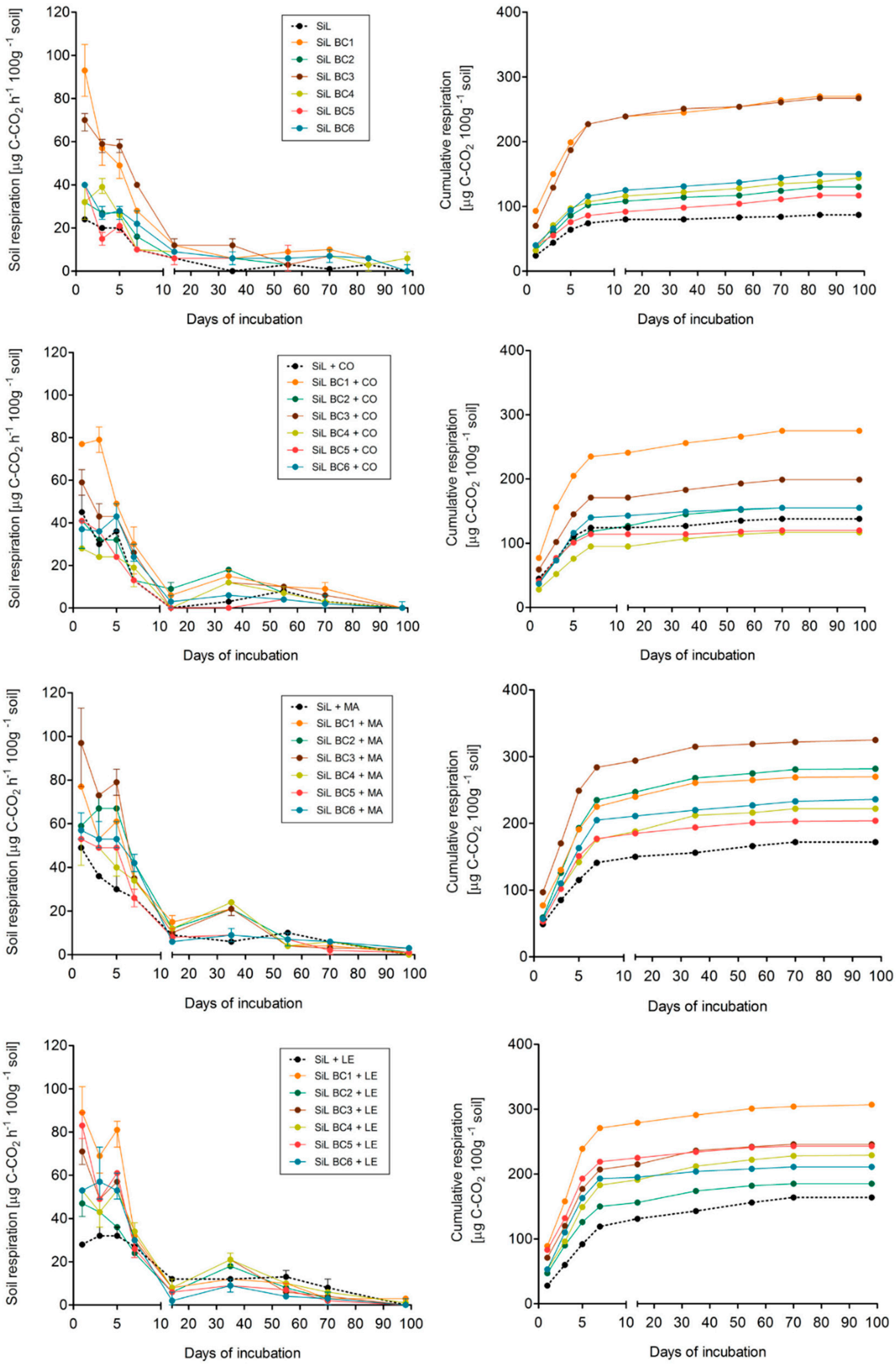

Figure 3. Respiration of silt loam (SiL) amended with biochars (BC1 – BC6) and organic materials: compost (CO), cattle manure (MA) and legume biomass (LE). Point = mean, bars = minimum and maximum. BCs origins: BC1 = kitchen waste, BC2 = cut grass, BC3 = coffee grounds, BC4 = wheat straw, BC5 = sunflower husks, BC6 = beech wood chips.

Preprints (www.preprints.org) | NOT PEER-REVIEWED | Posted: 18 September 2023 doi:10.20944/preprints202309.1124.v1

3.2. Effect of exogenous organic matter on soil respiration

Considering variants with additional organic amendments – compost, manure and legume biomass, higher CO_2 evolution rates were obtained from soils treated with BCs + manure and legume biomass than with compost. For sandy soils, in variants with manure, respiration day-by-day reached up to 145–170 µg C-CO_2 h^{-1} 100 g^{-1} in 3 out of 7 tested combinations, whereas legume biomass addition caused even higher CO_2 release, with the maximum of 180 µg C-CO_2 h^{-1} 100 g^{-1} (SA BC1 + LE). In addition, it was noted that SA + MA and SA + LE treatments showed values of respiration around 30-40 µg C-CO_2 h^{-1} 100 g^{-1} for longer time (approx. 30 days) than soils amended with compost or with biochars only (Figure 2). Treatments with SiL follow the same trend. As within the sandy soils, respiration rate increased in silt loams amended with biochars and legume biomass or manure, whereas compost had lesser effect on CO_2 evolution. Moreover, organic amendments, especially legumes and manure resulted in longer persistence of high soil respiration values – after 14 days of incubation the CO_2 evolution in SiL + MA or SiL + LE treatments rate was still relatively high, around 20-40 µg C-CO_2 h^{-1} 100 g^{-1} (Figure 3). Considering carbon loss percentage in biochar + organic amended soils, addition of easily decomposable organic matter generally increased percentage of carbon loss, especially in manure treated soil with BC (up to 0.85% in SA BC1 + MA) and legume biomass (1.10% in SA BC1 + LE). Effect of compost on the dynamics of C losses during respiration was less evident, as the maximum reached 0.73% (SA BC1 + CO), whereas most values in compost-amended soil were 0.2-0.3%, both in sandy and silty soils (Table S1 – supplementary).

To sum up, initial soil carbon status (native organic carbon pool) had no effect on observed CO_2 efflux – respiration rate was even lower on SiL than on SA soil. Regardless of the soil type, BCs showed similar patterns of respiration among tested treatments. Carbon dioxide emissions were the highest directly after application of the amendment and maximum values were observed for BCs rich in labile organic compounds, such as BC1 or BC3. This strongly suggests that initial peak of CO_2 evolution was a result of labiles decomposition from biochars. Moreover, among tested exogenous organic matter sources, raw materials (legume biomass and manure) had greater impact on C mineralization rates than biologically stable OM from compost, what was reflected in remarkably lower respiration in CO_2 amended treatments, compared with LE or MA. Generally, the trend in CO_2 evolution due to the type of biochar is: BC1 > BC3 > BC6, BC2 > BC5, BC4, and due to the additional exogenous organic matter source is: legume biomass > manure > compost.

4. Discussion

Results of study showed that under controlled environmental conditions biochar amendments affected GHG emission increasing CO_2 release from soils. The stimulating effects of biochar application on soil CO_2 fluxes can be ascribed to higher labile C mineralization and inorganic C release from biochar [13]. Furthermore, biochar application supports labile soil organic carbon pools enhancing microbial activity [31]. Microbial available C and nutrients in biochar are strongly correlated with temperature of pyrolysis [1,32], however findings of our study support the thesis that also biomass origin and properties of biochar, especially the content of labile C fractions, will contribute to the process of SOC mineralization, stimulating CO_2 emission from soil. Addition of biochar with more labile C fractions e.g. derived from kitchen wastes contributed to the process of CO_2 emission from soil more prominently than biochars derived from high lignocellulose biomass e.g. wood chips or straw. This observation is in agreement with our previous findings described by Bednik et al. [10]. Higher content of water-soluble carbohydrates (WSC) or dissolved organic carbon (DOC), and also less aliphatic structure of biochars derived from kitchen wastes such as coffee grounds (BC3) or vegetable and fruit peels (BC1) serve as labile C sources for microbes when applied to soil. Similar patterns in BCs mineralization were observed by Farrell et al. [33], showing that soil microbes rapidly utilize easily-available carbon pools delivered with biochar in forms of carbohydrates, dissolved organic carbon or volatile solids, but also a wide range of other organic compounds. Our results indicated that CO_2 fluxes varied over time after biochar application, which is in an agreement with previous findings [34–36]. However, mechanisms involved in soil CO_2 stimulation after biochar application may differ in the short term compared to long term study. The effect of breakdown of organic C and release of DOC from biochar is stimulating CO_2 emission from soil in a very short term of time after biochar application (up to 7 days). After sources of readily available carbon are utilized, CO_2 flux in biochar amended soils was stable, however higher compared to un-amended soils. This confirms that biochar can cause priming effect to native soil organic matter, but in the long time the process of CO_2 emission directly from biochar transformations becomes negligible thus not contributing to the GHG emission in global scale [17,36]. Application of biochar to tested soils also affected carbon pools, causing carbon losses, probably due to disturbance of soil environment (input of nutrients and labile carbon source) [34]. Usually, SOC content increases in the short term are due to the application and incorporation of fresh and C-rich biochar into the soil. This initial exposition of fresh biochar leads to a high microbial response and the turnover of the labile C fractions, often referred to as a positive priming effect [37].

Carbon losses can be also correlated with soil texture and this phenomenon was observed in the study. According to Gross et al. [38] in meta-analysis, biochar applications to clay soils resulted in the highest SOC stock increase, followed by silty soils and loamy soils. The lowest increases were observed for sandy soils. In general, higher clay mineral content in finer textured soils not only provides physical protection of SOC to enzymatic activity and thus turnover, but also increases SOC stability in the form of aggregates [39]. The effects of biochar application on soil CO_2 fluxes can be different depending on experimental design and conditions. Usually, very simple experiments with only biochar and unfertilized soils are preferred, however distinct effects can be observed when inorganic or organic fertilization is performed on biochar – amended soil.

In our study, we hypothesized that labile organic matter (LOM) from cover crops residues or organic fertilizers e.g. manures or compost may change the C-sequestration potential in biochar-amended soil. Both types of C sources will contribute to the SOC priming effect and this may induce changes in native mineralization process of organic matter, which in turn will increase or decrease CO_2 flux from soil [40]. The results of the experiment showed that introduction of EXOC to biochar amended soil enhances CO_2 fluxes from soil, however not equally, and raw materials e.g. cover crop residues will contribute to the process more actively than stable forms of organics like compost. The effect will vary also depending on soil type and properties. More prominent stimulating effect of EXOC on CO_2 emission was observed in sandy soil with biochar amendment. Faster BC-C mineralization on soil with low organic matter content is associated with good adaptation of microbes for the limited nutrients, and more effective utilization of available labile compounds, in comparison with soils rich in native organic matter [1,2,14,41–43]. In terms of loamy soil, lower CO_2 emission can be explained by organo-mineral interactions and protection of organic matter against mineralization process, which is claimed as a main factor of reduced GHG emission from soils with high clay minerals content [44,45].

Food waste is one of the society's highest volumes and most environmentally impactful waste streams. Upcycling of food waste into usable materials can be integral to mitigate the substantial greenhouse gas emissions associated with wasted food [25]. Inference on the high stability of biochar in the soil environment is limited to a very narrow group of biochars produced from basic and generally available types of biomasses, more attention should be paid to 'new biochars' obtained by utilizing household and food waste. As a very valuable source of nutrients and active compounds its utilization as a soil amendment seems to be a natural way of waste upcycling. This work highlights the problem of future implications related to incorporation to soil new types of black carbon. Variability of soil CO_2 fluxes in biochar amended soils can be attributed to biochar and soil properties, but also inputs of exogenous organic matter from soil fertilization and other agronomic practices.

5. Conclusions

Performed study confirms that biochars, when applied into the soil, are the subject of slow mineralization process with CO_2 release. The key factor that affects CO_2 efflux from amended soil is feedstock type used for biochar production, that determines further properties of biochar. CO_2 efflux was the highest for food waste biochars containing more labile C fraction and consequently more susceptible for decomposition processes, compared to high-cellulose biochar. Application of exogenous organic matter, especially raw organic plant residues and cow manure to biochar amended soils enhanced CO_2 release and carbon losses, however in the long – term contribution of the process might be negligible. To predict biochar behavior in soil under different farming practices it is necessary to develop field trials and provide data from long-term observation under natural conditions.

Supplementary Materials: The following supporting information can be downloaded at the website of this paper posted on Preprints.org, Table S1: Carbon balance in incubated treatments.

Author Contributions: For research articles with several authors, a short paragraph specifying their individual contributions must be provided. The following statements should be used "Conceptualization, (M.B., A.M.-J.); methodology, (M.B.); software, (M.B.); validation, (M.B.); formal analysis, (M.B.); investigation, (M.B., A.M.-J.); resources, (A.M.-J.); data curation, (M.B.); writing—original draft preparation, (M.B.); writing—review and editing, (A.M.-J., I.Ć.-P.); visualization, (M.B.); supervision, (A.M.-J., I.Ć.-P.); project administration, (M.B., A.M.-J. and I.Ć.-P.); funding acquisition, (M.B.). All authors have read and agreed to the published version of the manuscript.

Funding: This research was funded by research program "Innowacyjny Doktorat" supported by Wroclaw University of Environmental and Life Sciences (Poland), grant number N070/0009/20" The APC is co-financed by Wroclaw University of Environmental and Life Sciences.

Institutional Review Board Statement: Not applicable.

Data Availability Statement: Data sharing is not applicable to this article.

Acknowledgments: Many thanks to Michał Dudek for support in conducting the experiment and for useful discussions.

Conflicts of Interest: The authors declare no conflict of interest.

References

1. Budai, A.; Rasse, D.P.; Lagomarsino, A.; Lerch, T.Z.; Paruch, L. Biochar Persistence, Priming and Microbial Responses to Pyrolysis Temperature Series. *Biol Fertil Soils* **2016**, *52*, 749–761, doi:10.1007/s00374-016-1116-6.
2. Shen, Y.; Zhu, L.; Cheng, H.; Yue, S.; Li, S. Effects of Biochar Application on CO2 Emissions from a Cultivated Soil under Semiarid Climate Conditions in Northwest China. *Sustainability* **2017**, *9*, 1482, doi:10.3390/su9081482.
3. Agegnehu, G.; Srivastava, A.K.; Bird, M.I. The Role of Biochar and Biochar-Compost in Improving Soil Quality and Crop Performance: A Review. *Applied Soil Ecology* **2017**, *119*, 156–170, doi:10.1016/j.apsoil.2017.06.008.
4. Medyńska-Juraszek, A.; Latawiec, A.; Królczyk, J.; Bogacz, A.; Kawałko, D.; Bednik, M.; Dudek, M. Biochar Improves Maize Growth but Has a Limited Effect on Soil Properties: Evidence from a Three-Year Field Experiment. *Sustainability* **2021**, *13*, 3617, doi:10.3390/su13073617.
5. Sohi, S.P.; Krull, E.; Lopez-Capel, E.; Bol, R. A Review of Biochar and Its Use and Function in Soil. In *Advances in Agronomy*; Elsevier, 2010; Vol. 105, pp. 47–82 ISBN 978-0-12-381023-6.
6. Li, S.; Chan, C.Y. Will Biochar Suppress or Stimulate Greenhouse Gas Emissions in Agricultural Fields? Unveiling the Dice Game through Data Syntheses. *Soil Systems* **2022**, *6*, 73, doi:10.3390/soilsystems6040073.
7. Saarnio, S.; Heimonen, K.; Kettunen, R. Biochar Addition Indirectly Affects N2O Emissions via Soil Moisture and Plant N Uptake. *Soil Biology and Biochemistry* **2013**, *58*, 99–106, doi:10.1016/j.soilbio.2012.10.035.
8. Tang, Y.; Gao, W.; Cai, K.; Chen, Y.; Li, C.; Lee, X.; Cheng, H.; Zhang, Q.; Cheng, J. Effects of Biochar Amendment on Soil Carbon Dioxide Emission and Carbon Budget in the Karst Region of Southwest China. *Geoderma* **2021**, *385*, 114895, doi:10.1016/j.geoderma.2020.114895.
9. Sarfaraz, Q.; Silva, L.S. da; Drescher, G.L.; Zafar, M.; Severo, F.F.; Kokkonen, A.; Dal Molin, G.; Shafi, M.I.; Shafique, Q.; Solaiman, Z.M. Characterization and Carbon Mineralization of Biochars Produced from Different Animal Manures and Plant Residues. *Sci Rep* **2020**, *10*, 955, doi:10.1038/s41598-020-57987-8.
10. Bednik, M.; Medyńska-Juraszek, A.; Ćwieląg-Piasecka, I. Effect of Six Different Feedstocks on Biochar's Properties and Expected Stability. *Agronomy* **2022**, *12*, 1525, doi:10.3390/agronomy12071525.
11. Mandal, S.; Sarkar, B.; Bolan, N.; Novak, J.; Ok, Y.S.; Van Zwieten, L.; Singh, B.P.; Kirkham, M.B.; Choppala, G.; Spokas, K.; et al. Designing Advanced Biochar Products for Maximizing Greenhouse Gas Mitigation Potential. *Critical Reviews in Environmental Science and Technology* **2016**, *46*, 1367–1401, doi:10.1080/10643389.2016.1239975.

Preprints (www.preprints.org) | NOT PEER-REVIEWED | Posted: 18 September 2023 doi:10.20944/preprints202309.1124.v1

12. Zheng, N.; Yu, Y.; Li, Y.; Ge, C.; Chapman, S.J.; Yao, H. Can Aged Biochar Offset Soil Greenhouse Gas Emissions from Crop Residue Amendments in Saline and Non-Saline Soils under Laboratory Conditions? *Science of The Total Environment* **2022**, *806*, 151256, doi:10.1016/j.scitotenv.2021.151256.
13. He, Y.; Zhou, X.; Jiang, L.; Li, M.; Du, Z.; Zhou, G.; Shao, J.; Wang, X.; Xu, Z.; Hosseini Bai, S.; et al. Effects of Biochar Application on Soil Greenhouse Gas Fluxes: A Meta-Analysis. *GCB Bioenergy* **2017**, *9*, 743–755, doi:10.1111/gcbb.12376.
14. Cross, A.; Sohi, S.P. The Priming Potential of Biochar Products in Relation to Labile Carbon Contents and Soil Organic Matter Status. *Soil Biology and Biochemistry* **2011**, *43*, 2127–2134, doi:10.1016/j.soilbio.2011.06.016.
15. Lu, W.; Zha, Q.; Zhang, H.; Chen, H.Y.H.; Yu, J.; Tu, F.; Ruan, H. Changes in Soil Microbial Communities and Priming Effects Induced by Rice Straw Pyrogenic Organic Matter Produced at Two Temperatures. *Geoderma* **2021**, *400*, 115217, doi:10.1016/j.geoderma.2021.115217.
16. Case, S.D.C.; McNamara, N.P.; Reay, D.S.; Whitaker, J. Can Biochar Reduce Soil Greenhouse Gas Emissions from a Miscanthus Bioenergy Crop? *GCB Bioenergy* **2014**, *6*, 76–89, doi:10.1111/gcbb.12052.
17. Rasul, M.; Cho, J.; Shin, H.-S.; Hur, J. Biochar-Induced Priming Effects in Soil via Modifying the Status of Soil Organic Matter and Microflora: A Review. *Science of The Total Environment* **2022**, *805*, 150304, doi:10.1016/j.scitotenv.2021.150304.
18. Bruun, S.; Clauson-Kaas, S.; Bobuľská, L.; Thomsen, I.K. Carbon Dioxide Emissions from Biochar in Soil: Role of Clay, Microorganisms and Carbonates: CO_2 Emissions from Biochar in Soil. *Eur J Soil Sci* **2014**, *65*, 52–59, doi:10.1111/ejss.12073.
19. Fang, Y.; Singh, B.; Singh, B.P. Effect of Temperature on Biochar Priming Effects and Its Stability in Soils. *Soil Biology and Biochemistry* **2015**, *80*, 136–145, doi:10.1016/j.soilbio.2014.10.006.
20. Liu, X.; Zheng, J.; Zhang, D.; Cheng, K.; Zhou, H.; Zhang, A.; Li, L.; Joseph, S.; Smith, P.; Crowley, D.; et al. Biochar Has No Effect on Soil Respiration across Chinese Agricultural Soils. *Science of The Total Environment* **2016**, *554–555*, 259–265, doi:10.1016/j.scitotenv.2016.02.179.
21. Ding, X.; Li, G.; Zhao, X.; Lin, Q.; Wang, X. Biochar Application Significantly Increases Soil Organic Carbon under Conservation Tillage: An 11-Year Field Experiment. *Biochar* **2023**, *5*, 28, doi:10.1007/s42773-023-00226-w.
22. Shakoor, A.; Arif, M.S.; Shahzad, S.M.; Farooq, T.H.; Ashraf, F.; Altaf, M.M.; Ahmed, W.; Tufail, M.A.; Ashraf, M. Does Biochar Accelerate the Mitigation of Greenhouse Gaseous Emissions from Agricultural Soil? - A Global Meta-Analysis. *Environmental Research* **2021**, *202*, 111789, doi:10.1016/j.envres.2021.111789.
23. Sun, Z.; Liu, S.; Zhang, T.; Zhao, X.; Chen, S.; Wang, Q. Priming of Soil Organic Carbon Decomposition Induced by Exogenous Organic Carbon Input: A Meta-Analysis. *Plant Soil* **2019**, *443*, 463–471, doi:10.1007/s11104-019-04240-5.
24. Stegenta-Dąbrowska, S.; Sobieraj, K.; Koziel, J.A.; Bieniek, J.; Białowiec, A. Kinetics of Biotic and Abiotic CO Production during the Initial Phase of Biowaste Composting. *Energies* **2020**, *13*, 5451, doi:10.3390/en13205451.
25. Kane, S.; Ryan, C. Biochar from Food Waste as a Sustainable Replacement for Carbon Black in Upcycled or Compostable Composites. *Composites Part C: Open Access* **2022**, *8*, 100274, doi:10.1016/j.jcomc.2022.100274.
26. EBC, **2012-2022**. European Biochar Certificate - Guidelines for a Sustainable Production of Biochar.' Carbon Standards International (CSI), Frick, Switzerland. (http://european-biochar.org). Version 10.2 from 8th Dec 2022 (acessed 10 March 2023).
27. Dudek, M.; Łabaz, B.; Bednik, M.; Medyńska-Juraszek, A. Humic Substances as Indicator of Degradation Rate of Chernozems in South-Eastern Poland. *Agronomy* **2022**, *12*, 733, doi:10.3390/agronomy12030733.
28. Łabaz, B.; Kabała, C.; Dudek, M.; Waroszewski, J. Morphological Diversity of Chernozemic Soils in South-Western Poland. *Soil Science Annual* **2019**, *70*, 211–224, doi:10.2478/ssa-2019-0019.
29. Munera-Echeverri, J.L.; Martinsen, V.; Strand, L.T.; Zivanovic, V.; Cornelissen, G.; Mulder, J. Cation Exchange Capacity of Biochar: An Urgent Method Modification. *Science of The Total Environment* **2018**, *642*, 190–197, doi:10.1016/j.scitotenv.2018.06.017.
30. Fierer, N., **2013**. Measuring Soil C Mineralization Rates. Laboratory protocol. https://labs.eemb.ucsb.edu/schimel/josh/Protocols/soil%20respiration.pdf (accessed 10 March 2023).
31. Ameloot, N.; Graber, E.R.; Verheijen, F.G.A.; De Neve, S. Interactions between Biochar Stability and Soil Organisms: Review and Research Needs: Biochar Stability and Soil Organisms. *Eur J Soil Sci* **2013**, *64*, 379–390, doi:10.1111/ejss.12064.
32. Pariyar, P.; Kumari, K.; Jain, M.K.; Jadhao, P.S. Evaluation of Change in Biochar Properties Derived from Different Feedstock and Pyrolysis Temperature for Environmental and Agricultural Application. *Science of The Total Environment* **2020**, *713*, 136433, doi:10.1016/j.scitotenv.2019.136433.
33. Farrell, M.; Kuhn, T.K.; Macdonald, L.M.; Maddern, T.M.; Murphy, D.V.; Hall, P.A.; Singh, B.P.; Baumann, K.; Krull, E.S.; Baldock, J.A. Microbial Utilisation of Biochar-Derived Carbon. *Science of The Total Environment* **2013**, *465*, 288–297, doi:10.1016/j.scitotenv.2013.03.090.

Preprints (www.preprints.org) | NOT PEER-REVIEWED | Posted: 18 September 2023 doi:10.20944/preprints202309.1124.v1

34. Liu, C.-H.; Chu, W.; Li, H.; Boyd, S.A.; Teppen, B.J.; Mao, J.; Lehmann, J.; Zhang, W. Quantification and Characterization of Dissolved Organic Carbon from Biochars. *Geoderma* **2019**, *335*, 161–169, doi:10.1016/j.geoderma.2018.08.019.
35. Ouyang, L.; Yu, L.; Zhang, R. Effects of Amendment of Different Biochars on Soil Carbon Mineralisation and Sequestration. *Soil Res.* **2014**, *52*, 46, doi:10.1071/SR13186.
36. Ventura, M.; Alberti, G.; Panzacchi, P.; Vedove, G.D.; Miglietta, F.; Tonon, G. Biochar Mineralization and Priming Effect in a Poplar Short Rotation Coppice from a 3-Year Field Experiment. *Biol Fertil Soils* **2019**, *55*, 67–78, doi:10.1007/s00374-018-1329-y.
37. Wang, S.; Gao, B.; Zimmerman, A.R.; Li, Y.; Ma, L.; Harris, W.G.; Migliaccio, K.W. Physicochemical and Sorptive Properties of Biochars Derived from Woody and Herbaceous Biomass. *Chemosphere* **2015**, *134*, 257–262, doi:10.1016/j.chemosphere.2015.04.062.
38. Gross, A.; Bromm, T.; Glaser, B. Soil Organic Carbon Sequestration after Biochar Application: A Global Meta-Analysis. *Agronomy* **2021**, *11*, 2474, doi:10.3390/agronomy11122474.
39. Zong, Y.; Wang, Y.; Sheng, Y.; Wu, C.; Lu, S. Ameliorating Soil Acidity and Physical Properties of Two Contrasting Texture Ultisols with Wastewater Sludge Biochar. *Environ Sci Pollut Res* **2018**, *25*, 25726–25733, doi:10.1007/s11356-017-9509-0.
40. Dey, D.; Mavi, M.S. Co-Application of Biochar with Non-Pyrolyzed Organic Material Accelerates Carbon Accrual and Nutrient Availability in Soil. *Environmental Technology & Innovation* **2022**, *25*, 102128, doi:10.1016/j.eti.2021.102128.
41. Amoakwah, E.; Arthur, E.; Frimpong, K.A.; Lorenz, N.; Rahman, M.A.; Nziguheba, G.; Islam, K.R. Biochar Amendment Impacts on Microbial Community Structures and Biological and Enzyme Activities in a Weathered Tropical Sandy Loam. *Applied Soil Ecology* **2022**, *172*, 104364, doi:10.1016/j.apsoil.2021.104364.
42. Kolb, S.E.; Fermanich, K.J.; Dornbush, M.E. Effect of Charcoal Quantity on Microbial Biomass and Activity in Temperate Soils. *Soil Science Soc of Amer J* **2009**, *73*, 1173–1181, doi:10.2136/sssaj2008.0232.
43. Steinbeiss, S.; Gleixner, G.; Antonietti, M. Effect of Biochar Amendment on Soil Carbon Balance and Soil Microbial Activity. *Soil Biology and Biochemistry* **2009**, *41*, 1301–1310, doi:10.1016/j.soilbio.2009.03.016.
44. Kalu, S.; Simojoki, A.; Karhu, K.; Tammeorg, P. Long-Term Effects of Softwood Biochar on Soil Physical Properties, Greenhouse Gas Emissions and Crop Nutrient Uptake in Two Contrasting Boreal Soils. *Agriculture, Ecosystems & Environment* **2021**, *316*, 107454, doi:10.1016/j.agee.2021.107454.
45. Yang, Y.; Sun, K.; Han, L.; Chen, Y.; Liu, J.; Xing, B. Biochar Stability and Impact on Soil Organic Carbon Mineralization Depend on Biochar Processing, Aging and Soil Clay Content. *Soil Biology and Biochemistry* **2022**, *169*, 108657, doi:10.1016/j.soilbio.2022.108657.

Disclaimer/Publisher's Note: The statements, opinions and data contained in all publications are solely those of the individual author(s) and contributor(s) and not of MDPI and/or the editor(s). MDPI and/or the editor(s) disclaim responsibility for any injury to people or property resulting from any ideas, methods, instructions or products referred to in the content.