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# Large Losses of Pyrogenic Carbon (Biochar) and Native Soil Carbon During a 15-Month Field Study in North Florida, USA

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**Abstract:** Although the application of biochar to soils has been proposed as a method of carbon sequestration for climate mitigation while improving crop yields, losses of biochar carbon (BC) through mineralization may reduce these benefits. However, few field studies have been conducted that control for biochar migration so that the rates and processes of BC remineralization from soils, as well as the effects of biochar on native soil organic carbon, can be accurately determined. Here, biochar made from different biomass types (oak, pine wood, and grass) and temperatures (lightly charred at 250 °C and pyrolyzed at 400 and 650 °C) were added to fine sandy Entisol in an open agricultural field and Spodosol in a shaded forested site in North Central Florida. After 15 months, BC losses, determined by the Kurth–Mackenzie–Deluca chemical–thermal oxidation method, ranged from 17.5 to 93.3% $y^{-1}$ (14.0–51.5% $y^{-1}$ for the 650 °C biochar). These losses were correlated with but much greater than the 0.4–3% $y^{-1}$ BC losses recorded in a one-year laboratory study using the same biochars and those of several previous field studies (1–14% $y^{-1}$). The losses of non-BC native carbon stocks (i.e., positive priming) also varied with biochar and soil type and ranged from 1.5 to 15.8% $y^{-1}$. The high BC losses observed in this study may be attributed to the subtropical and temporally variable climate conditions at the study site. Greater efforts should be made to base BC long-term stability estimates on field studies that monitor or control for biochar migration rather than reliance only upon laboratory incubations.

**Keywords:** black carbon; chemical thermal oxidation; field study; priming; mineralization; stability; Entisol; Spodosol



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## 1. Introduction

Biochar is a carbonaceous material that is produced, optimally from waste biomass, by combustion under oxygen-limited conditions for use as a soil ameliorant or carbon sequestration tool [1]. Large-scale production and the use of biochar as a soil amendment may offset a large part of the carbon released by burning fossil fuels and improve soil fertility and soil C storage [2]. The condensed polyaromatic portion of biochar is inherently resistant to microbial degradation and thus has the potential to remain in the soil for hundreds or even thousands of years [3]. However, the longevity of biochar in soil must be predictable for the accurate assignment of carbon offsets/climate change mitigation credits to producers or users, e.g., farmers and land managers.

The relatively high persistence of biochar carbon (BC) is broadly accepted. For example, a recent meta-analysis found mean BC decomposition rates of 1.8% $y^{-1}$ for studies, mostly laboratory incubations, lasting more than 1 year [4]. Another recent synthesis and

modeling of current data estimated 63 to 82% of BC to persist over a 100-year timeframe [5]. But biochar's resistance to degradation in soil largely depends on its physical and chemical properties, which are mainly controlled by charring duration and temperature and feedstock biomass type [6]. For example, biochars made from grasses have been found to be less stable than those made from woody materials, and biochars produced at higher temperatures are more resistant to mineralization than those produced at lower temperatures [6–10]. The size of biochar particles may also have a significant impact on BC mineralization, as smaller particles have more available surface area, which facilitates surface reactions such as solubilization and enzymatic attack [11].

In addition to biochar type, biochar mineralization rates may be controlled by environmental factors such as climate and soil type [11,12]. For example, as with all organic matter, the rate of BC mineralization has been found to be directly [13,14] but non-linearly related to soil temperature [6]. Soil structure can also influence BC mineralization directly by controlling water and oxygen penetration or indirectly by influencing microbial and fungal community activity and accessibility to the biochar itself [15,16]. Increases in soil water holding capacity with biochar addition may result in greater BC mineralization by improving conditions for microbial activity [17,18], but it may delay abiotic and biotic BC mineralization if saturated conditions limit the supply of oxygen [19]. Studies have shown that BC mineralization is very sensitive to moisture levels [20], with alternating moisture conditions causing the greatest enhancement of BC mineralization [9]. However, we are unaware of any investigations that have examined the effects of varying moisture levels on BC mineralization under field conditions.

Biochar interaction with native soil organic matter (SOM) may alter the mineralization of both, thus affecting the amount of C that is ultimately sequestered following biochar amendment. Biochar addition to soil may either increase or decrease SOM mineralization (positive and negative priming, respectively), and the presence of soil may increase or decrease BC mineralization (reverse positive and reverse negative priming, respectively) [8,21–24]. In the case of positive priming, the introduction of a labile C source, such as root exudates or manure, can stimulate the production of microbial exoenzymes that lead to the co-metabolism of a more refractory C source. Nutrient mining and improved microbial habitat can also lead to enhanced C mineralization [25]. In contrast, the addition of biochar may lead to decreased rates of native SOM mineralization (negative priming) via sorptive protection or substrate switching [8,26,27]. However, variations in SOM priming with different biochar types have rarely been investigated in a field setting.

Thus far, the great majority of studies examining BC stability have been conducted as laboratory incubations. However, several limitations of laboratory studies can lead to inaccuracies in the predicted long-term stability of biochar. Because they are typically closed systems versus the open systems of natural soils, laboratory incubations are known to progressively deviate from natural conditions over longer experimental durations [28]. Some of the natural processes not realistically simulated in laboratory incubations include rainwater infiltration, bioturbation, gas diffusion, and alternating conditions, all of which can increase mineralization rates. Shifts in microbial communities and their activities may also occur over time in closed cultures due to non-ideal conditions such as nutrient and substrate limitation and the accumulation of metabolites [28]. While these can be avoided by shorter-term incubations of less than a month, the extrapolation of long-term decay rates from such short-term incubations is problematic due to the chemical heterogeneity of fresh biochar, which causes mineralization rates to decrease exponentially over time [10]. For example, an 8.5-year incubation experiment [29] calculated mean residence times (MRTs) of BC twice as much as those measured during the first 2.5 y of the experiment, at 400 versus 200 years, respectively [30].

Though field studies are needed to understand the rates and processes that control BC loss from soils and biochar's effects on native SOM, these types of studies are relatively scarce. Only a few studies have examined on-site losses using biochar of known starting composition and amount. The rest relied on natural charcoal produced from fires or slash-and-burn agriculture. In a review of 311 papers providing quantitative estimates of biochar stability from measurements of biochar stocks or soil respiration [31], only seven were field studies and only three of these involved experimental additions of biochar to soils. None of these field studies monitored or controlled for losses due to lateral or vertical BC transport, which can be significant (discussed below). Similarly, only three of 24 studies used in a recent meta-analysis of biochar on SOM priming [4] were field-based, and none of these controlled for the transport losses of biochar. Of the biochar field studies conducted, only a few examined biochar mineralization in tropical or subtropical soils, where conditions may be most conducive to BC mineralization [32–36].

In many field experiments, losses of biochar from soil can be attributed to decomposition, erosion (horizontal transport), or vertical migration downward through the soil profile. A field study showed that 7–55% of the BC added to a soil through a controlled experimental grass fire was lost by erosion and 23–46% had moved downward in the soil profile [37]. Four and a half years after mixing biochar into the upper 7 cm layer of loamy sandy Acrisol in Zambia, Obia et al. [38] estimated that 4–25% of the BC migrated below this layer and 25–60% was transported laterally out of the experimental plots.

To address these research voids, the goal of this study was to examine changes in BC and native soil C over a 15-month emplacement at two North Florida field sites. In addition, we examine the controls on biochar C stability by using multiple biochar types (different biomass types and production temperatures) and irrigated and non-irrigated field plots. Through the containment of the field soils, the experiment was designed to prevent plant growth and physical (erosional or downward migration) loss of biochar. Because previous biochar degradation laboratory studies were conducted using the same biochar materials as this study [10,24], another goal of the study, the comparison of biochar C loss rates determined in the laboratory and the field, could be carried out. We hypothesize that BC mineralization rates measured in the field will be much greater than those measured in the laboratory, but that the factors governing the BC mineralization rate will be the same. While the single time point destructive sampling design used in this study does not allow for longer-term prediction of C residence times, it does provide insight into the effects of relatively extreme field conditions on BC mineralization in soil and its interaction with soil C.

## 2. Materials and Methods

### *2.1. Materials and Field Methods*

Biochar was produced from three dried biomass types, namely the living wood portion of oak (Laurel oak, *Quercus laurifolia*), pine (Loblolly pine, *Pinus taeda*), and the stems and blades of Eastern gamma grass (*Tripsacum dactyloides*). These were pyrolyzed in a tubular oven with continuous flushing of 99% pure gaseous $N_2$ for 3 h at the highest treatment temperatures (HTTs) of 400 and 650 °C (designated as Oak-400, Pine-650, etc.). An additional biochar was produced under full atmosphere in an oven at 250 °C for 3 h (designated as charred Oak, Pine, and Grass). These less fully charred samples, as indicated by their lower C contents, may be representative of the products of prescribed burns, which tend to be cooler than wildfires, and on biomass below the soil surface [39]. All biochars were lightly crushed and sieved into particle sizes of <0.25 (fine) and 0.25–2 mm (coarse). Detailed information on the chemical properties of these biochars is provided in

Supplementary Information Table S1 and has been reported previously by Zimmerman et al. [10] and Mukherjee et al. [40].

Biochars were mixed with two North Florida soils, fine sandy Entisol from an open agricultural field at the University of Florida, Plant Science Research and Education Unit (PSREU), Citra, FL (TOC = 10.1 mg $g^{-1}$, designated as Agricultural Soil, AS) and, using a smaller set of biochars, Spodosol from a shaded forested site in Gainesville, FL (TOC = 24.2 mg $g^{-1}$, designated as Forest Soil, FS). The burn history of these sites is unknown, but neither has seen fire for at least 50 years. Detailed information on the chemical properties of these soils is provided in Supplementary Information Table S1. Soils were collected from the 0–10 cm depth interval, sieved to <2 mm to remove plant roots and debris, air dried for 4 days, and homogenized with selected biochars in a large bowl by hand mixing. Mixture ratios of 3.2 g for oak or 2.4 g for pine or grass biochar $kg^{-1}$ soil at the AS site were used so as to obtain similar BC addition rates. Due to limited biochar availability, 2.3 g $kg^{-1}$ soil biochar of all types was used at the FS site.

The soil collection sites were used as field emplacement sites, with separate irrigated (12 mm every other day) and non-irrigated plots at the AS site. Each treatment type was duplicated and placed randomly within each 3 × 2 m plot. The soil–biochar mixtures (or soil-alone controls) were placed into 2.4 L plastic buckets with eight 1 mm holes and 0.25 mm metal mesh screening in the bottom to deter the downward loss of particles (but allow for moisture flow through and potential BC leaching). These buckets were buried to a depth of 20 cm so that the soil in the buckets was level with the native soil and about 10 cm of the bucket sides extended above the soil surface so as to deter lateral particle loss. Each bucket was covered on top with landscaping cloth (Blue Hawk™ fabric) for shading and to deter weed growth and photodegradative loss.

Both soil-alone and soil–biochar mixtures were emplaced for 15 months (June 2009 to September 2010) at the two sites, after which the whole buckets were retrieved. This time period was chosen so as to include two summer periods, when biochar degradation was assumed most likely to occur. The emplacements were kept free from debris and weeds by periodic cleaning. The climate of North Central Florida is subtropical, with an average temperature at the study site during the experimental period of 21.7 °C and an average summer (June–August) temperature of 29.1 °C. During the experimental period, the site received 203.3 cm of precipitation (about 60% during the two summers) and an additional 560.1 cm was added to the irrigated plots. After the experimental period, soils were retrieved, homogenized by hand mixing, air-dried, and stored until chemical analysis.

### *2.2. Analytical Methods*

Initial soil and soil–biochar mixture samples were stored in a freezer until the end of the experiment and were analyzed together with the 'final' collected samples. So, all the same reagents and methods were used. Prior to analysis, whole-bucket samples were dried (80 °C, 48 h) then subsampled using a sample splitter (W.S. Tyler® SS-50). Total organic carbon (TOC) contents of the samples (pre- and post-emplacement) were analyzed after HCl acidification to remove inorganic C on a Carlo-Erba NA-1500 CHS Elemental Analyzer and were analyzed in duplicate (or additional times until reaching < 5% relative error).

Biochar carbon (BC) contents of pre- and post-emplacement soil–biochar mixtures and control samples were analyzed using a chemothermal oxidation method (CTO), the Kurth–Mackenzie–Deluca (KMD) method, which quantifies BC by removing presumably non-biochar carbon (non-BC) via oxidative digestion and measuring C in the remaining material. This method has been widely applied to estimate BC quantities in soil [41–45]. The assumption that all non-BC and only non-BC is removed in the KMD digestion process has been questioned based on the observations of strong correlations between soil TOC

and CTO-derived BC across many studies [46] and low CTO-derived BC recoveries from a variety of biochars (e.g., [45,47]). On the other hand, BC quantified by the KMD method has been found to be well correlated with that of the more widely accepted $^{13}C$ isotope approach [48]. In addition, $^{13}C$ cannot be used to quantify BC when there are not significant differences between the $^{13}C/^{12}C$ ratio of native SOC and the biochar added, as is the case in the current study.

Following Kurth et al. [49], about 1 g of sample ground to <0.76 µm, 20 mL of 30% $H_2O_2$, and 10 mL of 1M $HNO_3$ were added into 250 mL glass beakers. The mixtures were then heated to 100 °C on a heating plate for 16 h with periodic stirring. After complete digestion (no observation of further effervescence), samples were filtered through Whatman #2 filter paper, after which the sediment-laden filters were oven-dried at 60 °C. Samples were then carefully scraped from the filters, weighed, and analyzed for C content.

Biochar C content measured via the KMD method was calculated as

$$BC\left(\frac{mg\ C}{g\ soil}\right) = \left(C_{post\ digestion}\right)\left(\frac{mg}{g}\right) \times \left(\frac{m_{post_{digestion}}}{m_{pre_{digestion}}}\right) \quad (1)$$

where $m_{pre\text{-}digestion}$ and $m_{post\text{-}digestion}$ are the soil weights before and after digestion, respectively. The average analytical standard error of the KMD method was found to be 4%.

### *2.3. Data Treatment*

Because the BC content of the pre-treatment soil was non-zero, we concluded there was pre-existing pyrogenic carbon in the soil. This likely occurred because of fire activity of more than 50 years ago or from non-pyrogenic processes that have recently been found to produce condensed aromatic carbon [50] that may be resistant to oxidative digestion. Alternatively, the KMD method may include other non-pyrogenic forms of soil C that are resistant to CTO treatment. Since we are interested here in changes to the quantity of the added BC and we assume that no significant changes to the pre-existing pyrogenic C occurred during the period of the study due to its likely refractory nature, we calculated the BC initially present in each treatment sample ($BC_{init.}$) as

$$BC_{init.} = BC_{meas.init.} - BC_{meas.ctl.init.} \quad (2)$$

where $BC_{meas.init.}$ is the BC content of the initial soil–biochar mixtures and $BC_{meas.ctl.init}$ is the BC content of the initial control soil samples, both as measured by the KMD method. Similarly, the BC content of each treatment sample at the end of the experiment ($BC_{fin.}$) was calculated as

$$BC_{fin.} = BC_{meas.fin.} - BC_{meas.ctl.fin.} \quad (3)$$

where $BC_{meas.fin}$ is the BC content of the final treatment samples and $BC_{meas.ctl.fin}$ is the BC content of the final control samples, both as measured by the KMD method.

The proportion of BC lost during field emplacement (BC %Loss) was calculated as

$$BC\ \%Loss = (BC_{init.} - BC_{fin.})/BC_{init.}\ \% \quad (4)$$

The initial and final native soil C (non-BC) in each sample was calculated as

$$non\text{-}BC_{init.} = TOC_{init.} - BC_{meas.init.} \quad (5)$$

and

$$non\text{-}BC_{fin.} = TOC_{fin.} - BC_{meas.fin.} \quad (6)$$

The proportion of non-BC lost or gained in a soil–biochar mixture due to the biochar addition (positive or negative priming, respectively) during field emplacement (non-BC %Loss) was calculated as

$$\text{non-BC \%Loss} = [(\text{non-BC}_{\text{init.}} - \text{non-BC}_{\text{fin.}}) - (\text{non-BC}_{\text{ctl.init.}} - \text{non-BC}_{\text{ctl.fin.}})]/\text{non-BC}_{\text{init.}} \quad (7)$$

The data were analyzed using open-source R statistical software (version 4.3.3). Paired *t*-test tests were used to test for significant differences in BC and native soil C changes among treatment types. The BC %Loss or non-BC %Loss of soil–biochar (coarse samples only) mixtures were analyzed using a two-way analysis of variance (ANOVA) to test for significant effects of biochar biomass type (grass, oak, and pine) or pyrolysis temperature (charred, 400, and 650 °C). Multiple comparisons among paired treatment types (with duplicated treatments included separately) were performed using Tukey's HSD tests. In all cases, significant differences were accepted at a $p < 0.05$ (unadjusted) level.

## 3. Results

The biochars produced had TOC contents ranging from 83 wgt.% for Pine-650 to 52.7 wgt.% for charred grass (Table S1) and generally increased with production temperature, as has been found previously (e.g., [24]). The $H/C_{org}$ ratios of the biochars ranged from 0.33 to 0.83 for Oak-650 and Pine-400, respectively.

The average initial and final TOC, BC, and non-BC contents of the soils and soil–biochar mixtures are listed in Supplementary Information Tables S2 and S3 (and each individual measurement can be found in Tables S6 and S7). The average relative standard deviation of TOC and BC measured in duplicate treatments was found to be 3.3 and 7.3%, respectively. The average initial and final BC contents of each treatment soil at AS and FS, measured by the KMD method, are plotted in Figures S1 and S2, respectively. The initial TOC content in FS (24.2 mg $g^{-1}$) was much greater than that of the AS (10.1 mg $g^{-1}$), and both slightly decreased after 15 months likely due to microbial stimulation during transfer to buckets and homogenization. Average initial BC measured by KMD was 0.4 and 1.0 mg $g^{-1}$ in AS and FS, respectively, which represented pre-existing 'pyrogenic carbon' in the soil, as determined by the KMD method.

With biochar addition, the soil organic carbon (SOC) of initial soil–biochar mixtures ranged from 12.4 to 15.0 mg $g^{-1}$ in AS and 25.4 to 29.8 mg $g^{-1}$ in FS, as the C content of each biochar differed. Thus, the average initial BC content of the mixtures ranged from 0.3 to 2.4 mg $g^{-1}$ in AS and from 0.5 to 2.1 mg $g^{-1}$ in FS. The final BC content ranged from 0 to 1.7 mg $g^{-1}$ in AS and from 0.1 to 1.2 mg $g^{-1}$ in FS after field emplacement. Initial non-BC content of the mixtures ranged from 11.2 to 13.2 mg $g^{-1}$ in AS and from 23.5 to 27.5 mg $g^{-1}$ in FS. After the field period, final non-BC content had decreases ranging from 8.7 to 12 mg $g^{-1}$ in AS and 21.8 to 25.7 mg $g^{-1}$ in FS.

### *3.1. BC Loss Variation*

After field emplacement, while TOC in all treatment soils was greater than their respective controls soils, the average BC content decreased in all biochar-amended treatment types, and this decrease was statistically significant in about half of the treatment types despite the high degree of variation among replicates. The average BC %Loss from biochar-amended treatments ranged from 19.7 to 72.3% in AS (Figure 1 and Table S2) and 17.5 to 93.3% in FS (Figure 2 and Table S3) and varied with both biochar parent material and production temperature. Among all treatments, mean BC% losses at the AS and FS sites were 53 and 55%, respectively.

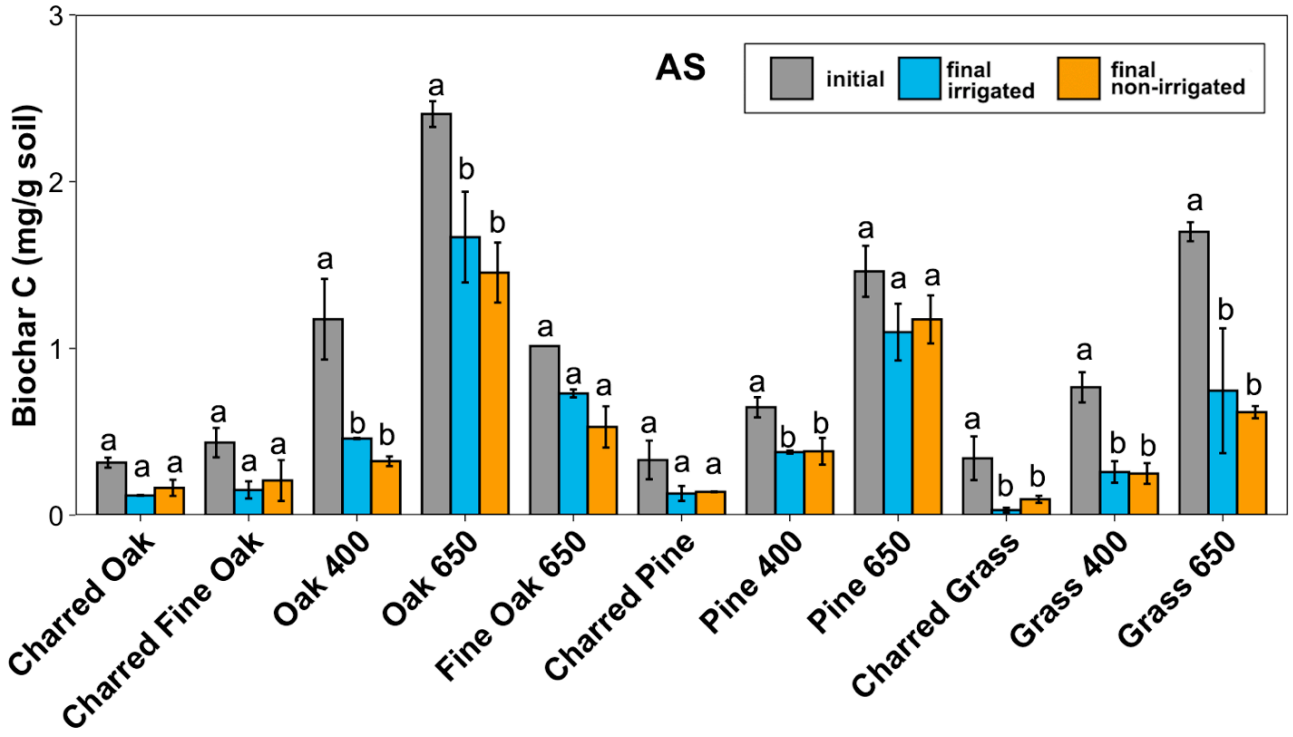


**Figure 1.** Biochar carbon content in agricultural soil (AS) soil–biochar treatments before (initial) and following (final) 15-month field incubation in irrigated and non-irrigated plots (means and standard deviations shown). Different letters above bars indicate treatments with significantly different means ($p < 0.05$) within each treatment type.

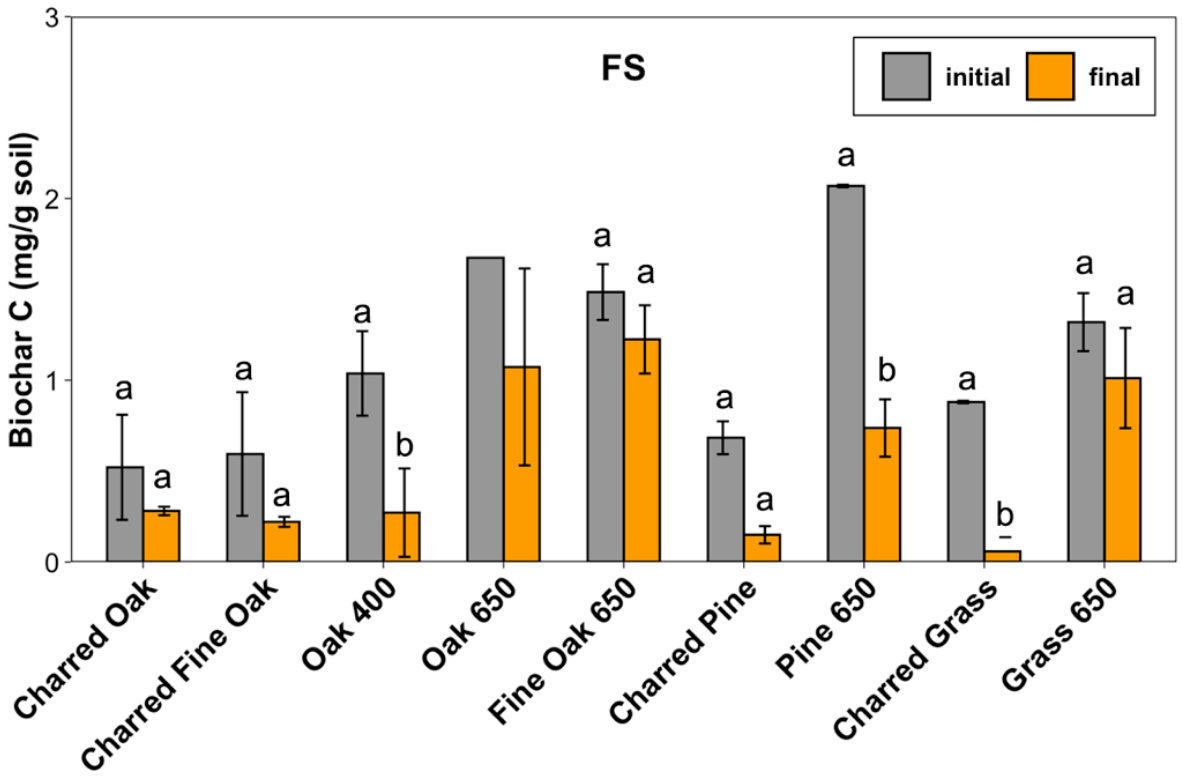


**Figure 2.** Biochar carbon content in forest soil (FS) soil–biochar treatments before (initial) and following (final) 15-month field incubation (means and standard deviations shown). Different letters above bars indicate treatments with significantly different means ($p < 0.05$) within each treatment type.

At the AS site, average BC %Loss for grass, pine, and oak biochar treatments differed significantly at 70, 41, and 51%, respectively (Figure 3). The mean loss of 650 °C biochars in AS was 39%, which was significantly lower than the 64 and 58% BC loss for charred and 400 °C biochar, respectively. Also at the AS site, although the average BC %Loss was slightly greater in fine versus coarse biochar-amended treatments (45.7 versus 43.8%, respectively) and at the irrigated versus non-irrigated plots (Table S4), these differences were not statistically significant due to the high variability among different biochar types, even within duplicate treatment types. The average difference in BC %Loss between duplicate treatments in AS was 8%.

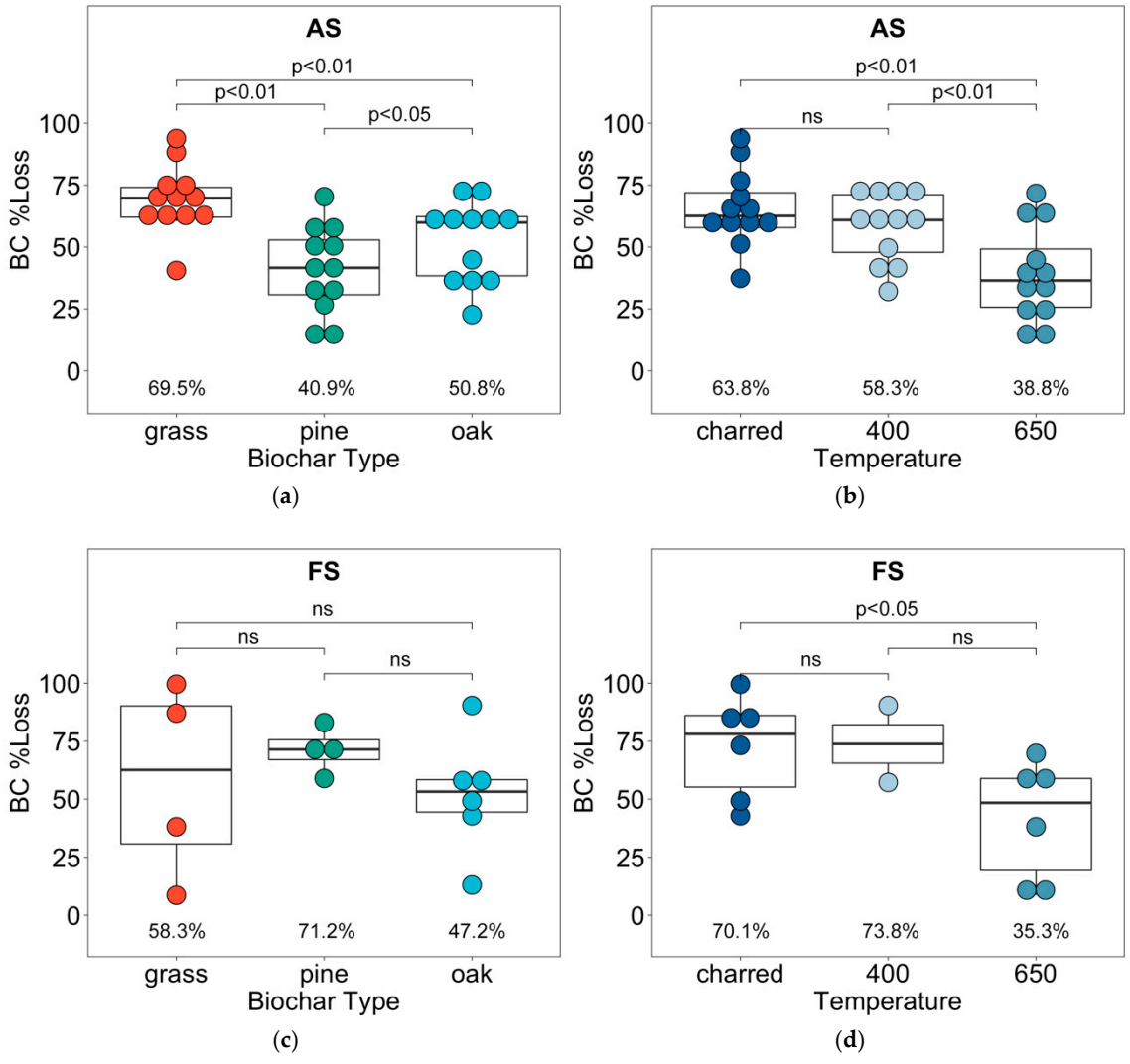


**Figure 3.** Boxplot of BC %Loss in agricultural soil (AS) and forest soil (FS) soil–biochar treatments with two-way ANOVA comparisons of biochars of different biomass and pyrolysis temperature. (**a**) agricultural soil (AS) biochar type and (**b**) temperature, forest soil (FS) by (**c**) biochar type and (**d**) temperature. 'ns' indicates a non-significant difference.

The average BC %Loss in FS for grass, pine, and oak biochar was 58, 71, and 47%, respectively, but in contrast to the AS samples, these differences were not significant due to a larger variance among duplicate treatments (mean of 9% at FS, Figure 3 and Table S4). As with AS treatments, 650 °C biochar C losses were significantly less than that of the charred or 400 °C biochar (35 versus 70 and 74%, on average, respectively). Overall, the mean BC %Loss was slightly greater at the FS versus AS site (55 versus 53%, respectively). But again, this difference was not statistically significant.

### 3.2. Native SOC (Non-BC) Loss Variation

After field emplacement, there were significant losses of non-BC content in more than half of the biochar-amended treatments in the AS and FS plots (Figures 4 and 5), ranging from 1.5 to 15.8% (Tables S2 and S3). Mean non-BC %Loss was significantly greater in the AS compared to the FS treatment plots (9.5 versus 4.7%, respectively, Table S4), but it was not significantly different between irrigated and non-irrigated plots in AS or in fine and coarse-sized oak biochar-amended plots.

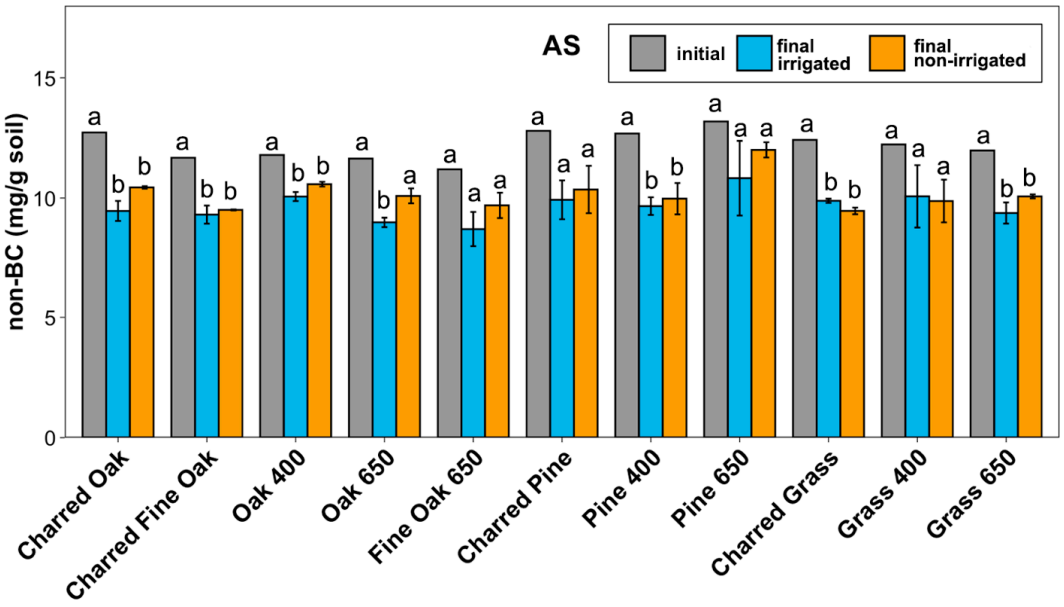


**Figure 4.** Native soil C (non-BC) in agricultural soil (AS) soil–biochar treatments before (initial) and following (final) 15-month field incubation in irrigated and non-irrigated plots (means and standard deviations shown). Different letters above bars indicate treatments with significantly different means ($p < 0.05$) within each treatment type.

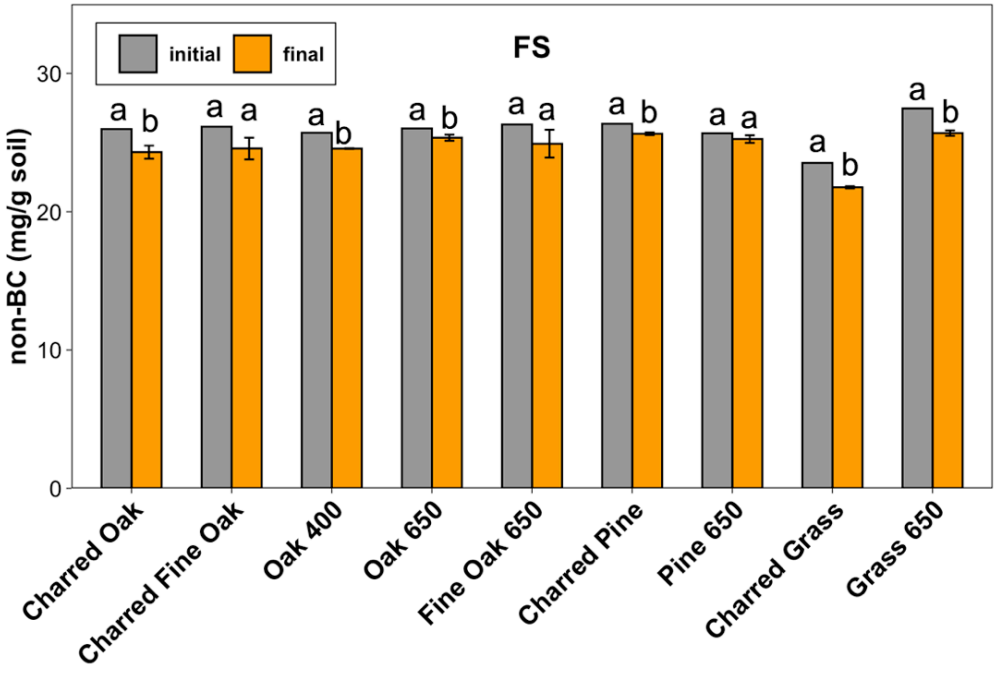


**Figure 5.** Native soil C (non-BC) in forest soil (FS) soil–biochar treatments before (initial) and following (final) 15-month field incubation (means and standard deviations shown). Different letters above bars indicate treatments with significantly different means ($p < 0.05$) within each treatment type.

Average non-BC %Loss in the oak biochar treatments in AS was 8.7%, which was lower than the 10.4 and 11.1% average losses in grass and pine biochar treatments, respectively (Figure 6). In FS, the average non-BC %Loss in the grass biochar treatment was 6.9%, significantly greater than the 4.9 and 2.1% losses in oak and pine biochar treatments, respectively (Figure 6).

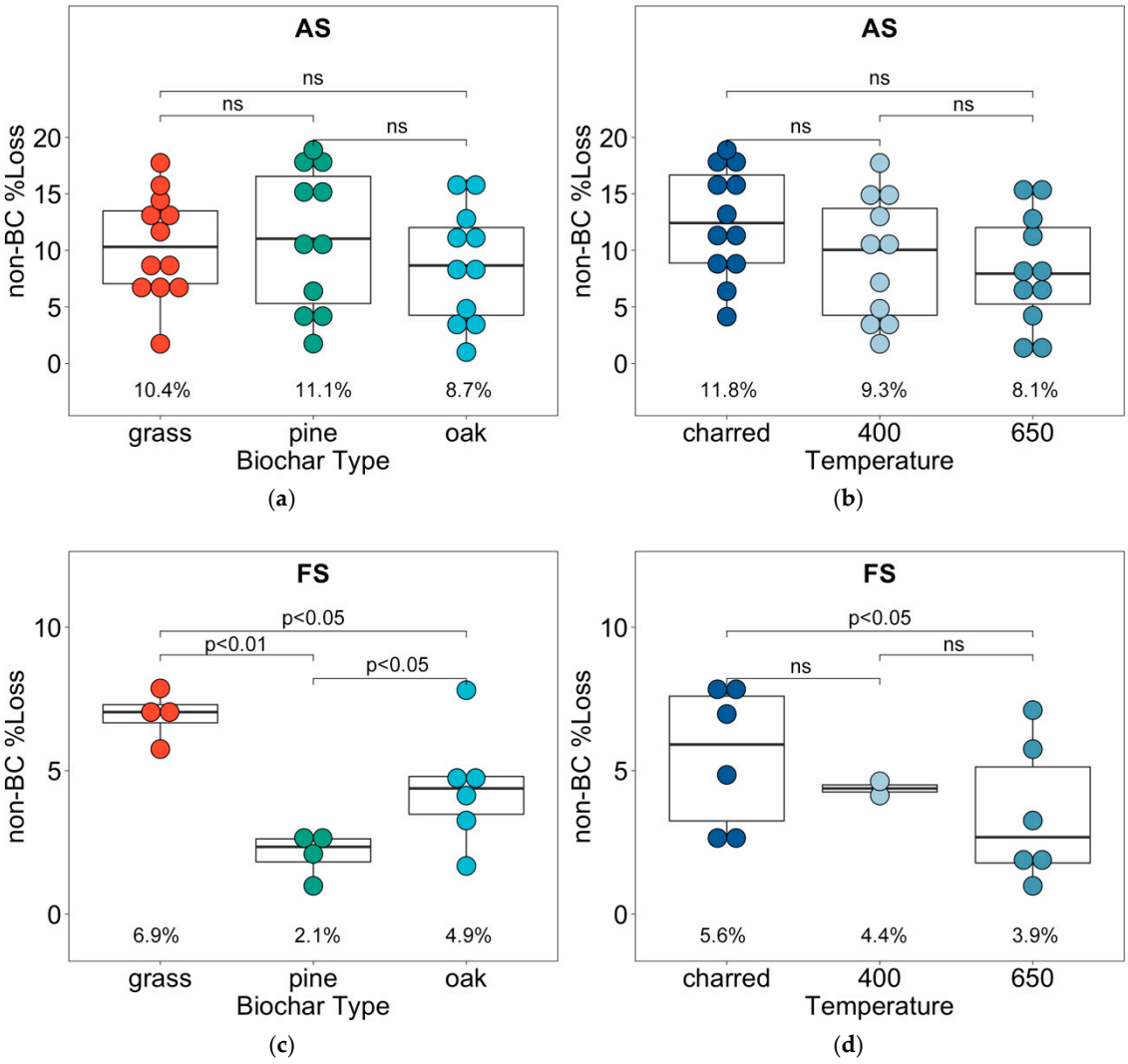


**Figure 6.** Boxplots of native soil C (non-BC) %Loss in agricultural soil (AS) by (**a**) biochar type and (**b**) temperature, and forest soil (FS) by (**c**) biochar type and (**d**) temperature. Treatments are compared via two-way ANOVA. 'ns' indicates a non-significant difference.

Average non-BC %Loss also varied with the biochar pyrolysis temperature in the following order: charred biochar > 400 °C > 650 °C biochar, both in AS and FS plots (Figure 6). Of these, only the difference between the charred and 650 °C biochar non-BC %Loss in FS was significant (Figure 6).

## 4. Discussion

The very high BC loss rates observed in this study will be discussed after first examining the factors and processes that may have controlled biochar and native soil C loss during the field emplacement. Potential BC loss mechanisms include microbial mineralization, solubilization, and physical loss via biochar migration.

### *4.1. Controls on Biochar C Loss*

The factors potentially influencing BC loss in this field study include biochar biomass type, biochar pyrolysis temperature, biochar particle size, soil moisture levels, and soil type. The results suggest that the loss of BC was mainly controlled by biochar biomass type and production temperature, as biochar particle size, soil type, and moisture conditions were not observed to have a significant influence on BC loss (Table S4).

#### 4.1.1. Effects of Biochar Parent Material

At the AS site, BC %Loss in grass biochar-amended plots (70%) was significantly greater than that of oak (51%), and both were significantly greater than that of pine biochar (41%) (Figure 3). Many laboratory studies have found that biochar parent material influences its chemical composition and thus its stability [51]. Grass biochar has often been found to be of greater lability than biochar made from woody materials in laboratory studies [6,10,52,53]. This study is the first to observe this in a field setting. This has been attributed to the lower degrees of aromatic condensation of grass-derived biochars, probably due to the high cellulose relative to the lignin content of grass [54,55]. In contrast, wood-derived biochar decomposes more slowly [4] likely because of its higher aromatic C content [52,56]. In line with findings in AS, laboratory incubations of biochars from hard woods such as oak have usually been found to be of greater stability than those from soft woods such as pine [10,57].

In contrast to the results from AS, in FS, pine biochar-amended treatments had the highest average BC %Loss (71%), higher than the BC %Loss for grass (58%) and oak (47%) biochar, although this difference was not significant. However, this result may be related to the high variability of BC %Loss data in FS due to the lower number of treatments (only four treatments each for grass and pine biochar).

#### 4.1.2. Effects of Biochar Production Temperature

It is clear from many laboratory studies that biochar produced at higher temperatures is more resistant to mineralization than biochar produced at lower temperatures [6–10,53,58]. In a meta-analysis of mainly lab studies, Wang et al. [4] found biochar produced at 200–375 °C decomposed, on average, three times faster than biochar pyrolyzed above 375 °C. In laboratory incubations of the same biochars used in this study, Zimmerman [10] found that charred and 400 °C BCs mineralized at rates 1.5 to 2.7 times faster than BCs produced at 525 and 650 °C. Similarly, in the present study, BC %Loss of charred and 400 °C biochar at AS was 1.7, 2.2, and 1.2 times greater than that of 650 °C BC from oak, pine, and grass, respectively (Figure 3).

The greater stability of higher temperature biochars is closely related to chemical changes that occur during pyrolysis. Elemental analyses, along with solid-state nuclear magnetic resonance (NMR) and Fourier transform infrared (FTIR) spectroscopy, reveal that when beginning at a temperature of 250 °C, the primary components of plant dry matter, such as cellulose and lignin, predominantly dehydrate into phenols, furans, and small aromatic units with high levels of O, H, and S substitutions [7,55]. At temperatures ranging from 400 to 500 °C and higher, stepwise depolymerization, the loss of functional groups, aromatization, dehydrogenation reactions, and the removal of substituents result in the formation of larger fused aromatic ring systems. These condensed structures are likely the most resistant to both biological and abiotic degradation [10].

#### 4.1.3. Effects of Biochar Particle Size

Previous laboratory studies have found that the rates of mineralization of larger sized biochar particles are lower than those of smaller sized particles [10,59,60]. For example,

during a 1 y incubation of the same materials used in the current study, the rate of biochar C mineralization was 12% greater for fine (particle size < 0.25 mm) compared to coarse biochar fractions (0.25–2 mm) in incubations with microbes and 41% greater in abiotic incubations, on average [10]. This is likely because of the greater amount of surface exposure of fine particles to dissolution, oxidation, and microbial exoenzyme attack. However, in the current field study, BC %Loss from coarse and fine-sized biochar-amended treatments was not significantly different (43.8 versus 45.7%, respectively) (Table S4). This may be because over longer time periods and with the very high degrees of BC mineralization that occurred, kinetic limitations dictated by diffusion into biochar inner surfaces were minimized. In addition, over time, biochar may be physically broken down into smaller particles [61]. This is more likely to occur under field conditions, where temperature and moisture conditions vary in contrast to the constant conditions of laboratory incubations. This may also partly explain the greater BC mineralization rates observed in this field study compared to those from laboratory incubations (discussed further below).

#### 4.1.4. Effects of Soil Type

The FS had higher initial organic matter content than AS (mean TOC of 24.2 versus 10.1 mg $g^{-1}$, respectively) as well as higher clay content (10% versus 2%, respectively, Table S1). Correspondingly, the average BC %Loss of each biochar type (except for pine biochar) was lower in FS (Figure 3), though not significantly, given the lower level of replication. In addition, fewer treatments showed significant BC content decreases in FS compared to in AS (Figures 1 and 2).

Previous studies have found that biochar stability varies with soil type. For example, in a laboratory study of biochar applied to five different soils, Yang et al. [62] found that biochar stability was greater in clay-rich acidic soils compared to those of lower clay content. Similarly, BC mineralization was found to be the lowest in clay soils (Oxisol < Vertisol), followed by sandy clay loam soils (Entisol) and highest in sandy soils (Inceptisol) [63]. The mineralization of BC may also be influenced by the amount and type of native organic matter present. In laboratory incubations of rice straw biochar in various rice paddy soils, biochar mineralization increased with native SOC content [64]. On the other hand, BC mineralization in a sandy clay loam was unaffected by repeated amendments of plant residues over seven years [65]. These contrasting results are likely due to the dependence of BC mineralization on SOM lability, where a labile C source may stimulate microbial activity, resulting in BC loss via co-metabolism versus the protective or diluting effect of a more refractory C source. For example, Zimmerman and Ouyang [27] found that the addition of sucrose enhanced BC mineralization, whereas dissolved humic acid suppressed it. A recent meta-analysis [66] found that biochar additions increased soil aggregation in neutral and acidic soil but not in alkaline soil. Thus, biochar may also have been better protected from degradation in FS due to its lower pH (pH 5.8 and 6.3 in FS and AS, respectively, Table S1) in addition to its greater clay and SOC content.

#### 4.1.5. Effects of Soil Moisture

Increased soil moisture, achieved in this study through irrigation applied to one of the plots, could have had several effects on BC loss. First, higher soil moisture may enhance microbial activity and thus BC mineralization [67]. On the other hand, saturated conditions may limit oxygen availability. For example, the mineralization of 350 °C oak BC under saturated conditions was half that of under unsaturated conditions [9]. Second, moisture addition may increase biochar solubilization, making it more available to microbes as a substrate (30–40% of dissolved biochar was found to be microbially mineralizable, [68]). Previous laboratory studies of the biochars used in the present study found the trend in BC

leaching was pine ≈ oak < grass biochar and low > high temperature biochar [40,69,70]. These trends were similar to the trends in BC %Loss observed in this study.

In the present study, there was no significant difference between BC %Loss in the irrigated and non-irrigated plots at the AS site (Figure 1 and Table S4). There are several potential explanations. First, it may be that because precipitation rates at the study site were relatively high, the effect of additional moisture through irrigation may have been minimized. We know of no BC loss field studies conducted in dryer climates that examined the effects of moisture addition. Second, laboratory leaching studies generally show BC losses in the range of 0.04–2% [40,71–74]. A study using the same biochars and soils used here and extrapolating laboratory leaching results to an equivalent water flow-through of a full year of rain in North Florida, estimated annual BC losses of 1 to 3.7% [40]. Biochar leaching rates have been shown to roughly double following four years of aging [75]. However, these leaching BC losses are still small compared to the 17.5–93.3% BC losses recorded in the present study; thus, they were unlikely to be detected as a significant effect.

#### 4.1.6. Potential for BC Physical Loss via Migration

There is also the potential that some biochar particles may have been lost to lateral erosion or downward migration through the soil [20,32,33]. For example, Pulcher et al. [76] found that 81% of the BC initially added to an Italian tree plantation soil remained in the top 20 cm of soil during the first year, which later decreased to 63.3% after three years. A nine-month field experiment in Switzerland [77] found that approximately 35% of the biochar added to topsoil (0–5 cm) was lost via migration. Obia et al. [36] mixed biochar into the surface (0–7 cm) layer of loamy sandy Acrisol in Zambia. After one year, they found that only 45–66% of the total applied biochar was recoverable from the applied layer. After 3 years, 4–25% of the biochar was detected in the soil layers (7–30 cm) below which it was applied [38].

Unlike any previous field incubation study we know of, here, soils were enclosed in partially buried buckets to inhibit the downward and horizontal migration of biochar. Though the bottom of the bucket had 1 mm holes and was lined with a 0.25 mm metal mesh, biochar particles < 0.25 mm still could have been lost. The biochar added was less than 2 mm in size and has the potential to break into smaller particles in the soil, while the fine biochar particles added were smaller than 0.25 mm. Thus, a significant portion of biochar could have moved downward and evaded quantification in this study. However, several observations suggest that migration was not the major mechanism for the BC losses of the present study. First, visually, there was no apparent accumulation of biochar particles in the bottom portion of the buckets after retrieval from the field. Second, there was no greater loss of BC in irrigated plots, as would be expected if the downward movement of water played a role facilitating downward migration. Third, there were no greater losses from the AS that had greater sand content which therefore would have facilitated downward BC migration. Fourth, coarse and fine biochar treatments showed no significant differences in BC losses. A previous study found that fine biochar particles displayed the greatest downward migration, suggesting its movement through soil inter-particle spaces [38]. Finally, the observed trends in BC losses with biomass type and pyrolysis temperature corresponded to those which would be expected based on laboratory incubations. The BC mineralization rates of the field (at AS) and lab experiments, both carried out over about 1 year, were significantly linearly correlated (Figure S3, $R^2 = 0.229$, $p = 0.024$). These trends would have been unlikely to be maintained had losses been strongly influenced by BC erosion or downward migration.

### *4.2. Biochar Interaction with Native Soil C (Priming)*

Most biochar-amended treatments in AS and FS showed significant losses of native soil C (non-BC) during the study period, ranging from 1.5 to 15.8% (and none showed increases), indicating that biochar had a positive priming effect on the SOC mineralization. While most studies of biochar priming effects have been conducted in the laboratory, available field studies have found the biochar priming effect to be positive [78], negative [79–84], or non-existent [85]. For example, the addition of rice husk biochar to a monsoonal alluvial plain soil reduced native SOC content by 15–42% after five years [78]. In contrast, after six years, carbon stocks in Exira silty clay loam in southwestern Iowa planted with switchgrass were found to increase by twice the amount of the carbon added from biochar [80]. Clearly, there are complex interactions and multiple factors that influence the mechanisms associated with SOC mineralization following biochar amendment. The possible factors that may have influenced native soil C priming in the current experiment include biochar biomass type, biochar pyrolysis temperature, and soil type.

#### 4.2.1. Effects of Biochar Parent Material on Non-BC Priming

An effect of biochar biomass type on priming was observed in FS but not in AS (Figure 6). In FS, grass biochar-amended treatments had significantly greater non-BC %Loss than pine and oak biochar-amended treatments. Similar results were found in a previous laboratory study using same biochar materials in which greater increases in SOC mineralization occurred in grass compared to oak biochar treatments [24]. A similar trend has been reported in other laboratory studies [23,86]. This can be attributed to the higher lignin content of wood, resulting in biochar with lower proportions of polar groups (i.e., high C/N and C/O ratios) and thus a lower value substrate for microbes compared to grass biochar [24,87–89]. Why the effect of biochar biomass type on SOC priming was not observed in AS is unclear, but it may be related to the lower SOC content and presumably lower lability of AS organic matter.

#### 4.2.2. Effects of Biochar Pyrolysis Temperature on Non-BC Priming

A meta-analysis study by Wang et al. [4] reported that the biochar-induced priming effect was not strongly dependent on biochar pyrolysis temperature. However, these results may have been influenced by the range of biochar biomass types and incubation periods included in this study. In a laboratory study, DeCiucies et al. [90] reported a strong temperature effect, with SOC loss generally decreasing (i.e., negative priming) with willow stem biochar pyrolysis temperature. A similar result was found in a previous laboratory study using same biochar materials as the current field study [24]. In line with these findings, in both AS and FS, charred biochar caused the greatest average non-BC %Loss, while 650 °C biochar treatments had the lowest average non-BC %Loss, although the difference in non-BC %Loss between charred and 650 °C biochar was only significant in FS (Figure 6). These findings can be attributed to the greater lability of low-temperature biochar due to its greater abundance of O-containing functional groups, which would result in the co-metabolization of non-BC. At the same time, the greater surface area of high-temperature biochar would offer native SOC greater sorptive protection.

#### 4.2.3. Effects of Soil Type on Non-BC Priming

The magnitude and direction of the biochar-induced priming effect may also be influenced by soil properties, including SOC content, pH, texture, and moisture content [63,91,92]. In this field study, we did not observe significant differences in non-BC %Loss between irrigated and non-irrigated plots in AS (Table S4), so here, moisture did not play a clear role in biochar's priming effect. Similarly, biochar particle size

had no apparent effect. However, average non-BC %Loss in AS (9.5%) was significantly greater than in FS (4.7%, Table S4). Previous laboratory studies have found a general trend of positive priming by biochar in low-carbon soils and negative priming in high-carbon soils [24,87,93]. A likely reason for this is that the microbial population in low-C soils is more readily stimulated by the addition of a labile C source and nutrients provided by biochar in comparison to those in high-C soils that may already have their substrate needs met.

Soil texture may also be an important influence on the priming of native SOC by biochar in this field study, since AS was more sandy and less clayey than FS (Table S1). In line with our findings, previous laboratory studies [4,93,94], including a meta-analysis of these studies [4], have concluded that biochar application to sandy soils (clay content < 10%) produces a stronger positive priming effect than in clay-rich soils. Biochar may stimulate microbial activity in low-clay soils, whereas large amounts of clay can promote the formation of stable soil aggregates (<250 μm) through organic–mineral interactions between natural or added organic matter, biochar, and soil minerals [95,96], which protects native SOC against further decomposition. The finding of only positive priming observed in this study may be related to the relatively low clay content of both AS and FS (<10%), which may have deterred aggregate formation.

The biochar amendment rate also has an important effect on the SOC priming effect [97–99]. The application of low doses of biochar generally enhanced SOM decomposition in low-carbon soils, whereas higher rates of biochar application resulted in a stronger negative priming effect. For example, Liu et al. [98] reported that only the highest of three different biochar doses (1%, 2%, and 5%) reduced SOC mineralization. Thus, the finding of only positive priming observed in this study may be related to the relatively low doses (0.2–0.3% by weight) of biochar added.

Finally, the incubation time period may have played a role in the finding of biochar-stimulated SOC loss. It is generally thought that positive priming dominates early in an incubation period but switches to negative priming over the long term [12,25,87]. The increase in SOC mineralization in soil immediately after biochar addition can be explained by the introduction of a labile substrate and nutrients and perhaps an increased microbial habitat causing microbial stimulation, whereas negative priming during later stages has been attributed to the protection of SOC by adsorption to biochar or encapsulation within biochar-facilitated aggregates [24,25]. One long-term laboratory study found that biochar addition produced a positive priming effect on SOC mineralization for 2.3 years after amendment, with a subsequent decreasing trend after 5 years [56]. Since we only collected samples after 15 months of field incubation, it may be that the soils had yet to switch from positive to negative priming.

### *4.3. Comparison of BC Loss Rates with Other Laboratory and Field Studies*

A comparison of the results of this field experiment with those of previous laboratory experiments using the same biochar material [10] shows that the factors controlling biochar loss were similar (Figure S3). In both lab and field experiments, the biomass pyrolysis temperature was the primary controlling factor with biomass type, a secondary factor. However, the BC mineralization rates observed in this field experiment (16 to 59% BC $y^{-1}$ for 400 and 650 °C biochars) were much higher, by factors of 20 to 30, than those of the previous laboratory incubations of the same biochars (0.6 to 1.9% BC loss $y^{-1}$).

Using the average BC %Loss measured in this study, we can calculate the mean residence time (MRT) of each added biochar based on a linear degradation model (since we only have the initial and final time point). The MRT of BC in AS and FS ranged from 1.3 to 5.6 years (Table S1) versus an MRT of 56 to 161 years using lab incubation data. However,

it should be recognized that linear degradation models yield MRT estimates much greater than those from more widely recommended two-component exponential or power function decay models [100].

In order to compare the BC degradation rates of this study with those of a wider range of studies, one must account for the use of different biochars of varying inherent chemical stability. Biochar is mainly composed of both condensed and non-condensed polycyclic aromatic compounds (poor in H), as well as functional groups (rich in H). Because the former is much more resistant to microbial enzymatic attack, the relative mixture of these forms is a determinative factor in determining the stability of biochar. Thus, the $H/C_{org}$ molar ratio has been suggested as an index of biochar stability, supported by correlations between biochar $H/C_{org}$ and laboratory mineralization data [5,101]. An upper $H/C_{org}$ limit of 0.7 has been proposed by the International Biochar Initiative (IBI) and the European Biochar Certificate (EBC) as the maximum $H/C_{org}$ ratio for a biochar to be considered for use in climate mitigation projects [102]. This is a conservative value to ensure that at least 50% (at a 95% confidence level) of the BC remains in soil after 100 years [101]. Most of the charred and 400 °C biochars used in the current study had $H/C_{org}$ near or greater than 0.7 (Table S1), so they would not qualify for climate mitigation yet may still represent some of the charred materials produced by natural fires. However, the 650 °C biochars had $H/C_{org}$ ranging from 0.33 to 0.45, yet still had MRT estimates of 2.1 to 5.6 years, much lower than observed in previous studies, most of which were conducted in the laboratory. It may be that laboratory studies have vastly overestimated the field stability of biochars.

We know of only two other studies that directly compare the mineralization of the same biochar type in the lab and field. Using *Miscanthus* grass biochar, Rasse et al. [103] calculated a BC mineralization rate of 0.41% $y^{-1}$ after 90 days of laboratory incubation compared to 0.8% $y^{-1}$ in the field, measured over a growing season. These rates are much lower than ours, perhaps due to the biochar's higher production temperature (600–700 °C, $H/C_{org}$ of 0.18), and the lower factor of difference (2x) may be attributed to the much lower temperatures of the Norway field site (annual temperature of 6 °C). Another study using maize silage biochar prepared by gasification (40 min at 1200 °C) found 2.3% $y^{-1}$ BC mineralization loss after 222 days of laboratory incubation [104] compared to 5.1 to 14.2% $y^{-1}$ BC mineralization in the 164- and 245-day field incubations the United Kingdom and Italy, respectively [79,83]. Decreasing mineralization rates are usually found as experimental time lengthens [4,28,29]. Again, these factors of difference are not as great as in the current study but had cooler and dryer climates, respectively.

Although a meta-analysis found no statistical difference between BC stability in lab- and field-derived experiments [4], no effort was made in the study to standardize for differences in the types of biochar used. Thus, to compare BC mineralization rates derived from lab incubations and field studies more widely, BC mineralization rates of studies that published the $H/C_{org}$ ratio of the biochar used (listed in Table S5) were plotted against the biochar $H/C_{org}$ ratio (Figure 7). The BC mineralization rates of all the laboratory-incubated biochars were < 10% $y^{-1}$, though most had mineralization rates in the range of 0.1–2% $y^{-1}$ and showed a linear increase with $H/C_{org}$ ($p = 0.06$). Field-derived BC mineralization rates, excluding the current study, ranged from 0.6 to 15% $y^{-1}$ and generally increased with the $H/C_{org}$ ratio ($p = 0.20$). The slope of the field-derived BC mineralization in the $H/C_{org}$ plot was also nine times greater than that for lab-derived BC losses, suggesting a greater dependance of BC stability on biochar type in the field compared to the lab. It should be noted, however, that unlike the present study, only one of these field studies [105] controlled for BC migration losses or lateral translocation.

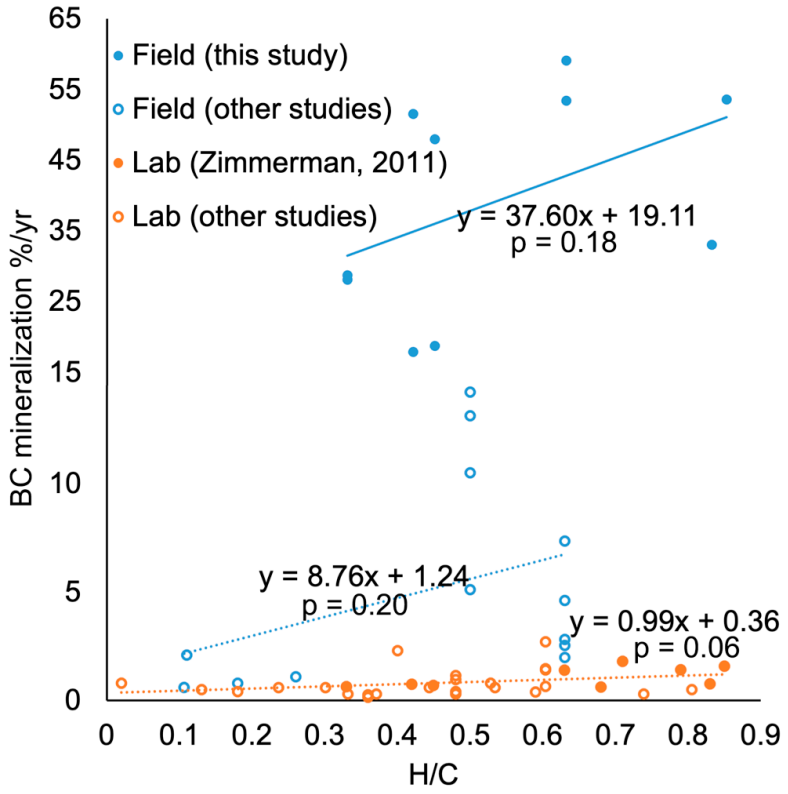


**Figure 7.** Comparison of laboratory versus field-based biochar carbon (BC) mineralization rates from this and previous studies including one [24] using the same biochars as this study. Studies included are those that published $H/C_{org}$ biochar values. Raw data and sources used.

Several factors may play a role in enhancing BC mineralization in the field compared to laboratory studies. The variability of conditions such as moisture and temperature, particularly freeze/thaw and drying/rewetting cycles, are known to stimulate microbial activity and thus organic matter degradation [106,107]. In contrast, the closed and constant conditions of a laboratory system inhibits microbial activity [28] and cannot simulate additional environmental processes such as water and air flow-through, exposure to ozone and UV radiation, and soil fauna activity including bioturbation.

The BC degradation rates in the current field experiment (MRT of 1.3 to 5.3 y) were higher than those reported in the limited number of previous field trials. However, several previous field biochar mineralization studies reported a BC MRT on the same order as those of this study, including 12.6 years at a coppice plantation in northern Italy [79,82], 11 years in Ferralsol in a dairy pasture in Tasmania, Australia [79,82], and 3.8 to 9.9 years in continuous permafrost-affected forest soils in northern Canada [108]. Factors that may have played a role include climate and biochar and soil type. The field site was located in the subtropics, with a strong rainy season and where the average temperature could reach 27 °C for half of the incubation time, which may have accelerated the physical degradation and chemical oxidation of biochar particles. In contrast, Ventura et al. [79] and Singh et al.'s [82] studies were carried out in cooler temperate regions. Clay particles have also been found to play a crucial role in stabilizing BC [11]. In this study, the relatively low clay content of the field site may have provided poor protection for biochar from physical and biological degradation and may have deterred aggregate formation. Finally, the high $H/C_{org}$ of the added biochar implies that the biochar was of lower chemical stability than that used in previous field studies.

### *4.4. Consideration of KMD Method of BC Quantification*

The KMD method used in this study to quantify BC is considered a conservative approach that tends to underestimate the BC content [45,47,109]. Soucémarianadin et al. [45] found that the recovery of BC in soil by the KMD method was only 20% for feathermoss BC produced at 250 °C and reaches 70 and 100% for black spruce needle biochar produced at 425 and 600 °C, respectively. Thus, while the losses recorded in this study would still be accurate, they may only apply to the most stabile portion of the biochar added and BC mineralization rates would likely have been even greater had the KMD method detected all the added BC.

Another issue is the detection of BC in the control soil by the KMD method that we assume to be pre-existing pyrogenic carbon in the soil. However, an alternative explanation is a false positive detection due to the existence of a highly refractive portion of the native SOM that was not completely digested during the KMD chemical thermal oxidation process. This was addressed by quantifying only added BC, calculated as the difference between BC in treatment samples and the control soil at the start and end of the incubation. However, it would have resulted in the inclusion of this pre-existing possibly pyrogenic carbon in the pool identified as non-BC. While there are issues with the KMD method, we find the method to be effective for use in biochar mineralization studies such as this one that is not amenable to the use of the stable isotopic differentiation of C sources, either because the amount of added biochar is low compared to native soil C or because it seeks to compare different biochar types with stable C isotope signatures that cannot all be different from that of native soil C.

## 5. Conclusions

The most important finding of this field study was BC mineralization rates (19.7 to 72.3% in the agricultural soil and 17.5 to 93.3% in the forest soil) that were much greater than those of previous laboratory and field studies. We realize that these rates are much higher than previously measured, but they are supported by the observation that the factors controlling BC loss, including biochar biomass and soil type and biomass pyrolysis temperature, were similar to those found previously. In addition, positive priming reduced native soil carbon stocks by 1.5 to 15.8% and also varied with the characteristics of the biochar and the soils, as found previously in laboratory incubations.

While the BC and biochar-induced native soil C mineralization rates measured here may have been unusually high due to the local climate and the biochar and soil types used, it further supports previous findings that laboratory incubation studies are likely to overestimate the stability of biochar in soil. This and the long period of positive priming of SOC are also of concern from the perspective of the use of biochar to sequester C and offset climate change. We therefore recommend much greater efforts to determine BC stability under varying field conditions (climates and soil types) through the establishment of a research network of long-term field trials. These field trials should either control and monitor biochar migration loss or be of large enough spatial extent to eliminate biochar migration as a concern. They also should utilize a set of standard biochar materials.

A further outcome of this study was the demonstration of the ability of the simple and low-cost KMD method to conveniently distinguish changes in BC from those of native SOC across a range of biochar types. This may have important applications to the certification of negative emissions for the purpose of C offset assignment and imbursement and mitigation pledge validation. Furthermore, having the ability to simultaneously measure changes in BC and SOC allows for the employment of biochar amendment along with other soil C enhancement methods while avoiding the risk of double counting.

**Supplementary Materials:** The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/agriculture15030300/s1.

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## References

1. Lehmann, J. A handful of carbon. *Nature* **2007**, *447*, 143–144. [CrossRef] [PubMed]
2. Woolf, D.; Lehmann, J. Modelling the long-term response to positive and negative priming of soil organic carbon by black carbon. *Biogeochemistry* **2012**, *111*, 83–95. [CrossRef]
3. 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. [CrossRef]
4. Wang, J.Y.; Xiong, Z.Q.; Kuzyakov, Y. Biochar stability in soil: Meta-analysis of decomposition and priming effects. *Glob. Change Biol. Bioenergy* **2016**, *8*, 512–523. [CrossRef]
5. Woolf, D.; Lehmann, J.; Ogle, S.; Kishimoto-Mo, A.W.; McConkey, B.; Baldock, J. Greenhouse gas inventory model for biochar additions to soil. *Environ. Sci. Technol.* **2021**, *55*, 14795–14805. [CrossRef] [PubMed]
6. Nguyen, B.T.; Lehmann, J.; Hockaday, W.C.; Joseph, S.; Masiello, C.A. Temperature Sensitivity of Black Carbon Decomposition and Oxidation. *Environ. Sci. Technol.* **2010**, *44*, 3324–3331. [CrossRef] [PubMed]
7. Baldock, J.A.; Smernik, R.J. Chemical composition and bioavailability of thermally, altered *Pinus resinosa* (Red Pine) wood. *Org. Geochem.* **2002**, *33*, 1093–1109. [CrossRef]
8. Hamer, U.; Marschner, B.; Brodowski, S.; Amelung, W. Interactive priming of black carbon and glucose mineralisation. *Org. Geochem.* **2004**, *35*, 823–830. [CrossRef]
9. Nguyen, B.T.; Lehmann, J. Black carbon decomposition under varying water regimes. *Org. Geochem.* **2009**, *40*, 846–853. [CrossRef]
10. Zimmerman, A.R. Abiotic and Microbial Oxidation of Laboratory-Produced Black Carbon (Biochar). *Environ. Sci. Technol.* **2010**, *44*, 1295–1301. [CrossRef]
11. Lehmann, J.; Czimczik, C.; Laird, D.; Sohi, S. Stability of biochar in soil. In *Biochar for Environmental Management*; Routledge: London, UK, 2012; pp. 215–238.
12. Zimmerman, A.R.; Gao, B. The stability of biochar in the environment. *Biochar Soil Biota* **2013**, *1*, 240.
13. Cheng, C.H.; Lehmann, J.; Thies, J.E.; Burton, S.D.; Engelhard, M.H. Oxidation of black carbon by biotic and abiotic processes. *Org. Geochem.* **2006**, *37*, 1477–1488. [CrossRef]
14. Zimmermann, M.; Bird, M.I.; Wurster, C.; Saiz, G.; Goodrick, I.; Barta, J.; Capek, P.; Santruckova, H.; Smernik, R. Rapid degradation of pyrogenic carbon. *Glob. Change Biol.* **2012**, *18*, 3306–3316. [CrossRef]
15. Pietikainen, J.; Kiikkila, O.; Fritze, H. Charcoal as a habitat for microbes and its effect on the microbial community of the underlying humus. *Oikos* **2000**, *89*, 231–242. [CrossRef]
16. Hockaday, W.C.; Grannas, A.M.; Kim, S.; Hatcher, P.G. The transformation and mobility of charcoal in a fire-impacted watershed. *Geochim. Cosmochim. Acta* **2007**, *71*, 3432–3445. [CrossRef]
17. Yerli, C.; Cakmakci, T.; Sahin, U. $CO_2$ emissions and their changes with $H_2O$ emissions, soil moisture, and temperature during the wetting–drying process of the soil mixed with different biochar materials. *J. Water Clim. Change* **2022**, *13*, 4273–4282. [CrossRef]
18. Yu, O.-Y.; Raichle, B.; Sink, S. Impact of biochar on the water holding capacity of loamy sand soil. *Int. J. Energy Environ. Eng.* **2013**, *4*, 44. [CrossRef]
19. Knicker, H. Pyrogenic organic matter in soil: Its origin and occurrence, its chemistry and survival in soil environments. *Quat. Int.* **2011**, *243*, 251–263. [CrossRef]
20. Foereid, B.; Lehmann, J.; Major, J. Modeling black carbon degradation and movement in soil. *Plant Soil* **2011**, *345*, 223–236. [CrossRef]

21. Nocentini, C.; Guenet, B.; Di Mattia, E.; Certini, G.; Bardoux, G.; Rumpel, C. Charcoal mineralisation potential of microbial inocula from burned and unburned forest soil with and without substrate addition. *Soil Biol. Biochem.* **2010**, *42*, 1472–1478. [CrossRef]
22. Keith, A.; Singh, B.; Singh, B.P. Interactive Priming of Biochar and Labile Organic Matter Mineralization in a Smectite-Rich Soil. *Environ. Sci. Technol.* **2011**, *45*, 9611–9618. [CrossRef] [PubMed]
23. Luo, Y.; Durenkamp, M.; De Nobili, M.; Lin, Q.; Brookes, P.C. Short term soil priming effects and the mineralisation of biochar following its incorporation to soils of different pH. *Soil Biol. Biochem.* **2011**, *43*, 2304–2314. [CrossRef]
24. Zimmerman, A.R.; Gao, B.; Ahn, M.-Y. Positive and negative carbon mineralization priming effects among a variety of biochar-amended soils. *Soil Biol. Biochem.* **2011**, *43*, 1169–1179. [CrossRef]
25. Whitman, T.; Singh, B.P.; Zimmerman, A.R.; Lehmann, J.; Joseph, S. Priming effects in biochar-amended soils: Implications of biochar-soil organic matter interactions for carbon storage. *Biochar Environ. Manag. Sci. Technol. Implement.* **2015**, *2*, 455–488.
26. Cui, J.; Ge, T.; Kuzyakov, Y.; Nie, M.; Fang, C.; Tang, B.; Zhou, C. Interactions between biochar and litter priming: A three-source $^{14}C$ and $\delta^{13}C$ partitioning study. *Soil Biol. Biochem.* **2017**, *104*, 49–58. [CrossRef]
27. Zimmerman, A.R.; Ouyang, L. Priming of pyrogenic C (biochar) mineralization by dissolved organic matter and vice versa. *Soil Biol. Biochem.* **2019**, *130*, 105–112. [CrossRef]
28. Spokas, K.A. Review of the stability of biochar in soils: Predictability of O:C molar ratios. *Carbon Manag.* **2010**, *1*, 289–303. [CrossRef]
29. Kuzyakov, Y.; Bogomolova, I.; Glaser, B. Biochar stability in soil: Decomposition during eight years and transformation as assessed by compound-specific $^{14}C$ analysis. *Soil Biol. Biochem.* **2014**, *70*, 229–236. [CrossRef]
30. Kuzyakov, Y.; Subbotina, I.; Chen, H.; Bogomolova, I.; Xu, X. Black carbon decomposition and incorporation into soil microbial biomass estimated by $^{14}C$ labeling. *Soil Biol. Biochem.* **2009**, *41*, 210–219. [CrossRef]
31. Gurwick, N.P.; Moore, L.A.; Kelly, C.; Elias, P. A Systematic Review of Biochar Research, with a Focus on Its Stability In Situ and Its Promise as a Climate Mitigation Strategy. *PLoS ONE* **2013**, *8*, e75932. [CrossRef]
32. Major, J.; Lehmann, J.; Rondon, M.; Goodale, C. Fate of soil-applied black carbon: Downward migration, leaching and soil respiration. *Glob. Change Biol.* **2010**, *16*, 1366–1379. [CrossRef]
33. Haefele, S.; Konboon, Y.; Wongboon, W.; Amarante, S.; Maarifat, A.; Pfeiffer, E.; Knoblauch, C. Effects and fate of biochar from rice residues in rice-based systems. *Field Crops Res.* **2011**, *121*, 430–440. [CrossRef]
34. Knoblauch, C.; Maarifat, A.-A.; Pfeiffer, E.-M.; Haefele, S.M. Degradability of black carbon and its impact on trace gas fluxes and carbon turnover in paddy soils. *Soil Biol. Biochem.* **2011**, *43*, 1768–1778. [CrossRef]
35. Van Zwieten, L.; Singh, B.P.; Tavakkoli, E.; Joseph, S.; Macdonald, L.M.; Rose, T.J.; Rose, M.T.; Kimber, S.W.; Morris, S.; Cozzolino, D. Biochar built soil carbon over a decade by stabilizing rhizodeposits. *Nat. Clim. Change* **2017**, *7*, 371–376.
36. Obia, A.; Børresen, T.; Martinsen, V.; Cornelissen, G.; Mulder, J. Vertical and lateral transport of biochar in light-textured tropical soils. *Soil Tillage Res.* **2017**, *165*, 34–40. [CrossRef]
37. Rumpel, C.; Ba, A.; Darboux, F.; Chaplot, V.; Planchon, O. Erosion budget and process selectivity of black carbon at meter scale. *Geoderma* **2009**, *154*, 131–137. [CrossRef]
38. Obia, A.; Lyu, J.; Mulder, J.; Martinsen, V.; Cornelissen, G.; Smebye, A.B.; Zimmerman, A.R. Biochar dispersion in a tropical soil and its effects on native soil organic carbon. *PLoS ONE* **2024**, *19*, e0300387. [CrossRef] [PubMed]
39. Santín, C.; Doerr, S.H.; Merino, A.; Bryant, R.; Loader, N.J. Forest floor chemical transformations in a boreal forest fire and their correlations with temperature and heating duration. *Geoderma* **2016**, *264*, 71–80. [CrossRef]
40. Mukherjee, A.; Zimmerman, A.R. Organic carbon and nutrient release from a range of laboratory-produced biochars and biochar-soil mixtures. *Geoderma* **2013**, *193*, 122–130. [CrossRef]
41. Ball, P.; MacKenzie, M.; DeLuca, T.; Montana, W.H. Wildfire and charcoal enhance nitrification and ammonium-oxidizing bacterial abundance in dry montane forest soils. *J. Environ. Qual.* **2010**, *39*, 1243–1253. [CrossRef]
42. Bélanger, N.; Pinno, B.D. Carbon sequestration, vegetation dynamics and soil development in the Boreal Transition ecoregion of Saskatchewan during the Holocene. *Catena* **2008**, *74*, 65–72. [CrossRef]
43. Licata, C.; Sanford, R. Charcoal and total carbon in soils from foothills shrublands to subalpine forests in the Colorado Front Range. *Forests* **2012**, *3*, 944–958. [CrossRef]
44. MacKenzie, M.D.; Graham, R.; Quideau, S. *Charcoal in Sierra Nevada Soils: Quantification and Spatial Variability*; Report; Kearney Foundation of Soil Science: Davis, CA, USA, 2006.
45. Soucémarianadin, L.N.; Quideau, S.A.; MacKenzie, M.D. Pyrogenic carbon stocks and storage mechanisms in podzolic soils of fire-affected Quebec black spruce forests. *Geoderma* **2014**, *217*, 118–128. [CrossRef]
46. Zimmerman, A.R.; Mitra, S. Trial by fire: On the terminology and methods used in pyrogenic organic carbon research. *Front. Earth Sci.* **2017**, *5*, 95. [CrossRef]
47. Maestrini, B.; Miesel, J.R. Modification of the weak nitric acid digestion method for the quantification of black carbon in organic matrices. *Org. Geochem.* **2017**, *103*, 136–139. [CrossRef]

48. Lyu, J.; Obia, A.; Cornelissen, G.; Mulder, J.; Smebye, A.B.; Zimmerman, A.R. Comparison of three quantification methods used to detect biochar carbon migration in a tropical soil: A 4.5-year field experiment in Zambia. *Geoderma* **2025**, *453*, 117153. [CrossRef]
49. Kurth, V.J.; MacKenzie, M.D.; DeLuca, T.H. Estimating charcoal content in forest mineral soils. *Geoderma* **2006**, *137*, 135–139. [CrossRef]
50. Goranov, A.I.; Chen, H.; Duan, J.; Myneni, S.C.; Hatcher, P.G. Potentially Massive and Global Non-Pyrogenic Production of Condensed "Black" Carbon Through Biomass Oxidation. *Environ. Sci. Technol.* **2024**, *58*, 2750–2761. [CrossRef] [PubMed]
51. Lehmann, J.; Rillig, M.C.; Thies, J.; Masiello, C.A.; Hockaday, W.C.; Crowley, D. Biochar effects on soil biota—A review. *Soil Biol. Biochem.* **2011**, *43*, 1812–1836. [CrossRef]
52. Hilscher, A.; Heister, K.; Siewert, C.; Knicker, H. Mineralisation and structural changes during the initial phase of microbial degradation of pyrogenic plant residues in soil. *Org. Geochem.* **2009**, *40*, 332–342. [CrossRef]
53. Jindo, K.; Sonoki, T. Comparative assessment of biochar stability using multiple indicators. *Agronomy* **2019**, *9*, 254. [CrossRef]
54. Hammes, K.; Smernik, R.J.; Skjemstad, J.O.; Herzog, A.; Vogt, U.F.; Schmidt, M.W. Synthesis and characterisation of laboratory-charred grass straw (*Oryza sativa*) and chestnut wood (*Castanea sativa*) as reference materials for black carbon quantification. *Org. Geochem.* **2006**, *37*, 1629–1633. [CrossRef]
55. Keiluweit, M.; Nico, P.S.; Johnson, M.G.; Kleber, M. Dynamic molecular structure of plant biomass-derived black carbon (biochar). *Environ. Sci. Technol.* **2010**, *44*, 1247–1253. [CrossRef]
56. Singh, B.P.; Cowie, A.L. Long-term influence of biochar on native organic carbon mineralisation in a low-carbon clayey soil. *Sci. Rep.* **2014**, *4*, 3687. [CrossRef]
57. Jiang, S.; Nguyen, T.A.; Rudolph, V.; Yang, H.; Zhang, D.; Ok, Y.S.; Huang, L. Characterization of hard-and softwood biochars pyrolyzed at high temperature. *Environ. Geochem. Health* **2017**, *39*, 403–415. [CrossRef] [PubMed]
58. Bruun, E.W.; Ambus, P.; Egsgaard, H.; Hauggaard-Nielsen, H. Effects of slow and fast pyrolysis biochar on soil C and N turnover dynamics. *Soil Biol. Biochem.* **2012**, *46*, 73–79. [CrossRef]
59. Sigua, G.C.; Novak, J.; Watts, D.; Cantrell, K.; Shumaker, P.; Szögi, A.; Johnson, M. Carbon mineralization in two ultisols amended with different sources and particle sizes of pyrolyzed biochar. *Chemosphere* **2014**, *103*, 313–321. [CrossRef] [PubMed]
60. Chen, J.; Li, S.; Liang, C.; Xu, Q.; Li, Y.; Qin, H.; Fuhrmann, J.J. Response of microbial community structure and function to short-term biochar amendment in an intensively managed bamboo (*Phyllostachys praecox*) plantation soil: Effect of particle size and addition rate. *Sci. Total Environ.* **2017**, *574*, 24–33. [CrossRef] [PubMed]
61. Spokas, K.A.; Novak, J.M.; Masiello, C.A.; Johnson, M.G.; Colosky, E.C.; Ippolito, J.A.; Trigo, C. Physical Disintegration of Biochar: An Overlooked Process. *Environ. Sci. Technol. Lett.* **2014**, *1*, 326–332. [CrossRef]
62. Yang, F.; Zuo, X.; Zhou, Y.; Wu, S.; Wang, M. Stability of biochar in five soils: Effects from soil property. *Environ. Prog. Sustain. Energy* **2022**, *41*, e13775. [CrossRef]
63. Fang, Y.; Singh, B.; Singh, B.; Krull, E. Biochar carbon stability in four contrasting soils. *Eur. J. Soil Sci.* **2014**, *65*, 60–71. [CrossRef]
64. Wu, M.; Han, X.; Zhong, T.; Yuan, M.; Wu, W. Soil organic carbon content affects the stability of biochar in paddy soil. *Agric. Ecosyst. Environ.* **2016**, *223*, 59–66. [CrossRef]
65. Dharmakeerthi, R.S.; Hanley, K.; Whitman, T.; Woolf, D.; Lehmann, J. Organic carbon dynamics in soils with pyrogenic organic matter that received plant residue additions over seven years. *Soil Biol. Biochem.* **2015**, *88*, 268–274. [CrossRef]
66. Islam, M.U.; Jiang, F.; Guo, Z.; Peng, X. Does biochar application improve soil aggregation? A meta-analysis. *Soil Tillage Res.* **2021**, *209*, 104926. [CrossRef]
67. Chow, A.T.; Tanji, K.K.; Gao, S.; Dahlgren, R.A. Temperature, water content and wet–dry cycle effects on DOC production and carbon mineralization in agricultural peat soils. *Soil Biol. Biochem.* **2006**, *38*, 477–488. [CrossRef]
68. Bostick, K.W.; Zimmerman, A.R.; Goranov, A.; Mitra, S.; Hatcher, P.G.; Wozniak, A.S. Biolability of Fresh and Photodegraded Pyrogenic Dissolved Organic Matter From Laboratory-Prepared Chars. *J. Geophys. Res. Biogeosci.* **2021**, *126*, e2020JG005981. [CrossRef]
69. Bostick, K.W.; Zimmerman, A.R.; Wozniak, A.S.; Mitra, S.; Hatcher, P.G. Production and Composition of Pyrogenic Dissolved Organic Matter From a Logical Series of Laboratory-Generated Chars. *Front. Earth Sci.* **2018**, *6*, 43. [CrossRef]
70. Gibson, C.D.; Hatton, P.-J.; Bird, J.A.; Nadelhoffer, K.; Ward, C.P.; Stark, R.E.; Filley, T.R. Interacting controls of pyrolysis temperature and plant taxa on the degradability of PyOM in fire-prone northern temperate forest soil. *Soil Syst.* **2018**, *2*, 48. [CrossRef]
71. Hockaday, W.C.; Grannas, A.M.; Kim, S.; Hatcher, P.G. Direct molecular evidence for the degradation and mobility of black carbon in soils from ultrahigh-resolution mass spectral analysis of dissolved organic matter from a fire-impacted forest soil. *Org. Geochem.* **2006**, *37*, 501–510. [CrossRef]
72. Abiven, S.; Hengartner, P.; Schneider, M.P.; Singh, N.; Schmidt, M.W. Pyrogenic carbon soluble fraction is larger and more aromatic in aged charcoal than in fresh charcoal. *Soil Biol. Biochem.* **2011**, *43*, 1615–1617. [CrossRef]
73. Jones, D.; Murphy, D.; Khalid, M.; Ahmad, W.; Edwards-Jones, G.; DeLuca, T. Short-term biochar-induced increase in soil $CO_2$ release is both biotically and abiotically mediated. *Soil Biol. Biochem.* **2011**, *43*, 1723–1731. [CrossRef]

74. Schiedung, M.; Bellè, S.-L.; Sigmund, G.; Kalbitz, K.; Abiven, S. Vertical mobility of pyrogenic organic matter in soils: A column experiment. *Biogeosciences* **2020**, *17*, 6457–6474. [CrossRef]
75. Wozniak, A.S.; Mitra, S.; Goranov, A.I.; Zimmerman, A.R.; Bostick, K.W.; Hatcher, P.G. Effects of Environmental Aging on Wildfire Particulate and Dissolved Pyrogenic Organic Matter Characteristics. *ACS Earth Space Chem.* **2023**, *8*, 104–118. [CrossRef]
76. Pulcher, R.; Balugani, E.; Ventura, M.; Greggio, N.; Marazza, D. Inclusion of biochar in a C dynamics model based on observations from an 8-year field experiment. *Soil* **2022**, *8*, 199–211. [CrossRef]
77. Felber, R.; Leifeld, J.; Horák, J.; Neftel, A. Nitrous oxide emission reduction with greenwaste biochar: Comparison of laboratory and field experiments. *Eur. J. Soil Sci.* **2014**, *65*, 128–138. [CrossRef]
78. Dong, X.; Singh, B.P.; Li, G.; Lin, Q.; Zhao, X. Biochar application constrained native soil organic carbon accumulation from wheat residue inputs in a long-term wheat-maize cropping system. *Agric. Ecosyst. Environ.* **2018**, *252*, 200–207. [CrossRef]
79. 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. [CrossRef]
80. Blanco-Canqui, H.; Laird, D.A.; Heaton, E.A.; Rathke, S.; Acharya, B.S. Soil carbon increased by twice the amount of biochar carbon applied after 6 years: Field evidence of negative priming. *GCB Bioenergy* **2020**, *12*, 240–251. [CrossRef]
81. Shi, S.; Zhang, Q.; Lou, Y.; Du, Z.; Wang, Q.; Hu, N.; Wang, Y.; Gunina, A.; Song, J. Soil organic and inorganic carbon sequestration by consecutive biochar application: Results from a decade field experiment. *Soil Use Manag.* **2021**, *37*, 95–103. [CrossRef]
82. Singh, B.P.; Fang, Y.; Boersma, M.; Collins, D.; Van Zwieten, L.; Macdonald, L.M. In situ persistence and migration of biochar carbon and its impact on native carbon emission in contrasting soils under managed temperate pastures. *PLoS ONE* **2015**, *10*, e0141560. [CrossRef] [PubMed]
83. Ventura, M.; Alberti, G.; Viger, M.; Jenkins, J.R.; Girardin, C.; Baronti, S.; Zaldei, A.; Taylor, G.; Rumpel, C.; Miglietta, F. Biochar mineralization and priming effect on SOM decomposition in two European short rotation coppices. *GCB Bioenergy* **2015**, *7*, 1150–1160. [CrossRef]
84. Weng, Z.H.; Van Zwieten, L.; Singh, B.P.; Kimber, S.; Morris, S.; Cowie, A.; Macdonald, L.M. Plant-biochar interactions drive the negative priming of soil organic carbon in an annual ryegrass field system. *Soil Biol. Biochem.* **2015**, *90*, 111–121. [CrossRef]
85. Reed, E.Y.; Chadwick, D.R.; Hill, P.W.; Jones, D.L. Critical comparison of the impact of biochar and wood ash on soil organic matter cycling and grassland productivity. *Soil Biol. Biochem.* **2017**, *110*, 134–142. [CrossRef]
86. Cross, A.; Sohi, S.P. The priming potential of biochar products in relation to labile carbon contents and soil organic matter status. *Soil Biol. Biochem.* **2011**, *43*, 2127–2134. [CrossRef]
87. Maestrini, B.; Nannipieri, P.; Abiven, S. A meta-analysis on pyrogenic organic matter induced priming effect. *GCB Bioenergy* **2015**, *7*, 577–590. [CrossRef]
88. Yu, Z.; Chen, L.; Pan, S.; Li, Y.; Kuzyakov, Y.; Xu, J.; Brookes, P.; Luo, Y. Feedstock determines biochar-induced soil priming effects by stimulating the activity of specific microorganisms. *Eur. J. Soil Sci.* **2018**, *69*, 521–534. [CrossRef]
89. Raya-Moreno, I.; Cañizares, R.; Domene, X.; Carabassa, V.; Alcañiz, J.M. Biochar Addition to a Mediterranean Agroecosystem: Short-Term Divergent Priming Effects. *Agriculture* **2024**, *14*, 242. [CrossRef]
90. DeCiucies, S.; Whitman, T.; Woolf, D.; Enders, A.; Lehmann, J. Priming mechanisms with additions of pyrogenic organic matter to soil. *Geochim. Cosmochim. Acta* **2018**, *238*, 329–342. [CrossRef]
91. Sun, F.; Lu, S. Biochars improve aggregate stability, water retention, and pore-space properties of clayey soil. *J. Plant Nutr. Soil Sci.* **2014**, *177*, 26–33. [CrossRef]
92. Sun, J.; He, F.; Zhang, Z.; Shao, H.; Xu, G. Temperature and moisture responses to carbon mineralization in the biochar-amended saline soil. *Sci. Total Environ.* **2016**, *569*, 390–394. [CrossRef]
93. Ding, F.; Van Zwieten, L.; Zhang, W.; Weng, Z.; Shi, S.; Wang, J.; Meng, J. A meta-analysis and critical evaluation of influencing factors on soil carbon priming following biochar amendment. *J. Soils Sediments* **2018**, *18*, 1507–1517. [CrossRef]
94. Fang, Y.; Singh, B.; Singh, B.P. Effect of temperature on biochar priming effects and its stability in soils. *Soil Biol. Biochem.* **2015**, *80*, 136–145. [CrossRef]
95. Soinne, H.; Hovi, J.; Tammeorg, P.; Turtola, E. Effect of biochar on phosphorus sorption and clay soil aggregate stability. *Geoderma* **2014**, *219*, 162–167. [CrossRef]
96. Wang, D.; Fonte, S.J.; Parikh, S.J.; Six, J.; Scow, K.M. Biochar additions can enhance soil structure and the physical stabilization of C in aggregates. *Geoderma* **2017**, *303*, 110–117. [CrossRef]
97. Abbruzzini, T.F.; Moreira, M.Z.; de Camargo, P.B.; Conz, R.F.; Cerri, C.E.P. Increasing rates of biochar application to soil induce stronger negative priming effect on soil organic carbon decomposition. *Agric. Res.* **2017**, *6*, 389–398. [CrossRef]
98. Liu, Y.; Chen, Y.; Wang, Y.; Lu, H.; He, L.; Yang, S. Negative priming effect of three kinds of biochar on the mineralization of native soil organic carbon. *Land Degrad. Dev.* **2018**, *29*, 3985–3994. [CrossRef]
99. 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. *Sci. Total Environ.* **2022**, *805*, 150304. [CrossRef] [PubMed]

100. Leng, L.; Xu, X.; Wei, L.; Fan, L.; Huang, H.; Li, J.; Lu, Q.; Li, J.; Zhou, W. Biochar stability assessment by incubation and modelling: Methods, drawbacks and recommendations. *Sci. Total Environ.* **2019**, *664*, 11–23. [CrossRef] [PubMed]
101. Budai, A.; Zimmerman, A.; Cowie, A.; Webber, J.; Singh, B.; Glaser, B.; Masiello, C.; Andersson, D.; Shields, F.; Lehmann, J. Biochar Carbon Stability Test Method: An assessment of methods to determine biochar carbon stability. *Int. Biochar Initiat.* **2013**, *20*, 1–20.
102. *IBI-STD-2.0*; Product Testing Guidelines for Biochar That Is Used in Soil. IBI Biochar Standards, International Biochar Initiative: Norfolk, VA, USA, 2016.
103. Rasse, D.P.; Budai, A.; O'Toole, A.; Ma, X.; Rumpel, C.; Abiven, S. Persistence in soil of Miscanthus biochar in laboratory and field conditions. *PLoS ONE* **2017**, *12*, e0184383. [CrossRef] [PubMed]
104. Naisse, C.; Girardin, C.; Lefevre, R.; Pozzi, A.; Maas, R.; Stark, A.; Rumpel, C. Effect of physical weathering on the carbon sequestration potential of biochars and hydrochars in soil. *GCB Bioenergy* **2015**, *7*, 488–496. [CrossRef]
105. Maestrini, B.; Abiven, S.; Singh, N.; Bird, J.; Torn, M.S.; Schmidt, M.W. Carbon losses from pyrolysed and original wood in a forest soil under natural and increased N deposition. *Biogeosciences* **2014**, *11*, 5199–5213. [CrossRef]
106. Sun, M.-Y.; Aller, R.C.; Lee, C.; Wakeham, S.G. Effects of oxygen and redox oscillation on degradation of cell-associated lipids in surficial marine sediments. *Geochim. Cosmochim. Acta* **2002**, *66*, 2003–2012. [CrossRef]
107. Cravo-Laureau, C.; Hernandez-Raquet, G.; Vitte, I.; Jézéquel, R.; Bellet, V.; Godon, J.-J.; Caumette, P.; Balaguer, P.; Duran, R. Role of environmental fluctuations and microbial diversity in degradation of hydrocarbons in contaminated sludge. *Res. Microbiol.* **2011**, *162*, 888–895. [CrossRef] [PubMed]
108. Schiedung, M.; Bellè, S.-L.; Hoeschen, C.; Schweizer, S.A.; Abiven, S. Enhanced loss but limited mobility of pyrogenic and organic matter in continuous permafrost-affected forest soils. *Soil Biol. Biochem.* **2023**, *178*, 108959. [CrossRef]
109. Maestrini, B.; Alvey, E.C.; Hurteau, M.D.; Safford, H.; Miesel, J.R. Fire severity alters the distribution of pyrogenic carbon stocks across ecosystem pools in a Californian mixed-conifer forest. *J. Geophys. Res. Biogeosci.* **2017**, *122*, 2338–2355. [CrossRef]