



Research Article

DOI: 10.36959/973/446

Biochar Source and Rate Effects on Soil pH and Water-Soluble Nutrients Over Time in Simulated Furrow-irrigated Rice in the Greenhouse

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Abstract

Biochar is the by-product of burning vegetative dry matter and can positively impact soil properties as an organic soil amendment, leading to biochar's increased use in agroecosystems. The objective of this study was to evaluate the effects of biochar source [i.e., an Arkansas (AR) and Colorado (CO) biochar] and rate (i.e., 0, 2.5, and 5 Mg ha⁻¹) on soil pH and water-soluble (WS) soil phosphorus (P), calcium (Ca), magnesium (Mg), iron (Fe), manganese (Mn), and sulfur (S) concentrations in simulated-furrow-irrigated rice (*Oryza sativa*) on a silt-loam soil (Typic Albaqualf) in the greenhouse. Soil pH differed among biochar rates ($P < 0.01$) and over time ($P < 0.01$), while WS-P differed between biochar sources over time ($P < 0.01$), WS-Ca, -Mg, and -Fe differed among biochar source-rate combinations ($P < 0.02$), and WS-Mn and -S differed among rates ($P < 0.02$). Averaged across biochar source and time, mean soil pH was 5.69 from the 5 Mg ha⁻¹ rate, which differed from the no-biochar control (pH = 5.59) but was similar to that in the 2.5 Mg ha⁻¹ rate (pH = 5.64). Water-soluble P was similar between biochar sources and over time for the whole growing season, except for 34 days after planting, where WS-P was eight times greater from the AR than the CO biochar. Results of this study showed that the use of biochar in upland agroecosystems requires considering both biochar source and rate to realize the greatest benefit of biochar on soil nutrient availability over the growing season.

Keywords

Arkansas, Rice production, Biochar, Soil water quality, Dissolved nutrients

Introduction

Increasing global energy production to keep up with the growing global human population has become increasingly important. With ~80% of global energy coming from fossil fuels (i.e., oil, natural gas, and coal), research and utilization of alternative energy sources is imperative to develop sustainable fossil fuel alternatives. Biofuel, which is sourced from renewable resources like vegetative plant matter, is a potential fossil fuel alternative for long-term sustainable energy [1]. Biofuel production requires the burning of plant biomass, which creates a co-product termed biochar [2]. Biochar is the product of burning vegetative dry matter [i.e., Switchgrass (*Panicum virgatum*), corn (*Zea mays*) stover, wood chip waste, and/or rice (*Oryza sativa*) material] in an anoxic environment through a process called pyrolysis, which is commonly used in biofuel production [3]. Due to the large carbon concentration and abundance of biochar

as a byproduct, biochar has been investigated as a soil amendment.

Biochar material has potential positive effects on soil health and plant growth based on biochar's physical and chemical properties, namely biochar's low density and carbon (C) and other nutrient concentrations [2,4]. Biochar application

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Accepted: July 23, 2024

Published online: July 25, 2024

Citation: Brye JB, Gwaltney L, Lunga DD, et al. (2024) Biochar Source and Rate Effects on Soil pH and Water-Soluble Nutrients Over Time in Simulated Furrow-irrigated Rice in the Greenhouse. J Rice Res Dev 6(1):466-476

in agroecosystems has been reported to positively impact soil properties and crop growth through reduced soil bulk density, increased soil water retention in sandy and loamy soils [5], enhanced cation exchange capacity [6], increased soil organic C (SOC) and soil C sequestration [7,8], increased crop growth [9], with minimal impact on yield [10,11]. Khalili, et al. [12] evaluated the effects of biochar treatments (i.e. walnut feedstock biochar) in furrow-irrigated conditions with different water regimes in a sandy loam soil and reported that corn yield was numerically larger from the biochar-treated plots that received 75% of the crop water requirement compared to the full irrigation recommendation. Anyanwu, et al. [13] assessed how rice-husk biochar treatment in soil impacted earthworms, soil microbiota, and early plant growth in potted rice and tomato (*Solanum lycopersicum*) in a tropical agricultural soil and reported that biochar-treated pots resulted in increased aboveground biomass. However, Anyanwu, et al. [13], reported a significant decrease in belowground root biomass during the 90-day soil-biochar contact time, especially for rice, which indicated that biochar may not have a positive effect on plant root growth. The reported benefits of biochar have resulted in increased use and research into biochar as a beneficial soil amendment in agricultural settings [2,4].

Biochar is generally understood to positively impact the soil and plant growth, but further investigations have led to the discovery of potential drawbacks of biochar as a soil amendment [5,13,14]. One major drawback is that biochar sources can chemically and physically vary depending on the original feedstock, combustion temperature and duration [14]. For example, high-temperature pyrolysis causes the release of hydrogen and oxygen groups (i.e. OH⁻), which can increase the C concentration of the biochar and impact resulting biochar pH. The partial detachment of functional groups can increase the presence of negatively charged carboxyl (COOH⁻) and hydroxyl (OH⁻) groups, which affects the cation exchange capacity of the biochar [6,15]. Biochar has also been reported to have liming properties, where increasingly alkaline biochars can positively affect plant growth through improved soil fertility, enhancing nutrient dissolution in acidic or slightly acidic soils [16]. Consequently, the resulting physical and chemical property variability among biochar sources makes predicting the impact of biochar amendment on soil properties and plant growth and productivity difficult when combined with the site-specific soil properties of agricultural fields that can substantially affect the plant response to different biochar sources.

Additionally, biochar's potential large adsorptive capacity [17], specifically in relation to plant-essential nutrients like phosphate, nitrogen (N), and iron (Fe), can negatively impact plant nutrient availability [6,18]. Xu, et al. [18] conducted an incubation study with a saline-sodic sandy soil and five wheat (*Triticum*) straw biochars produced from pyrolysis at 25, 300, 400, 500, and 600 °C for 4 hours, where the objective was to compare biochar effects at different pyrolysis temperatures on P availability. Xu, et al. [18] reported that relatively lower-temperature (< 400 °C) biochar treatments adsorbed less P than biochars created from ≥ 500 °C pyrolysis temperatures,

but, when all biochar sources and fertilizer-P were applied simultaneously, phosphate sorption/precipitation led to lower plant-available P. However, due to the adsorptive capacity of biochar and affinity for cations, there are several studies that reported an increase in P availability due to the ability of biochar to fix Al, Fe, and Ca ions that immobilize P [19]. In addition to variations in original biochar feedstock, the particle size (i.e., degree of fineness) may influence biochar's adsorptive capacity, where the finer the particle size, the greater the expected surface area, reactivity [20], and adsorptive capacity. Consequently, due to the large adsorptive capacity, high application rates of biochar may represent a further complication in the already challenging management of P in rice cropping systems, particularly in the furrow-irrigated rice production system, where the combination of moist and saturated environments can occlude the different forms of P [21].

In the United States, Arkansas is responsible for ~50% of total rice production [22]. Traditional rice production occurs under flooded-soil conditions, where the reducing conditions that develop from an extended period of ponded water tends to increase soil-P availability as Fe-bound P is released [23]. Thus, if biochar is used as a soil amendment in flood-irrigated rice, the potential exists for plant-available P to become even more limited, thus potentially requiring greater fertilizer-P additions to overcome reduced P availability.

As an alternative to flood-irrigated rice, furrow-irrigation has increased in use to about 20% of the planted-rice area in Arkansas due to being a water-conserving and labor-saving practice without a substantial yield penalty [22,24]. Furrow-irrigated rice production consists of planting rice on raised beds along a gently sloping landscape and allowing irrigation water to gravity-flow from the up- to the down-slope position in the field [25]. The frequent wet-and-dry cycles associated with furrow-irrigated rice [26] would not be expected to increase soil-P availability, as would be expected in flood-irrigated rice, because the soil does not generally achieve anaerobic conditions long enough to reduce iron-bound P. Furthermore, if the soil is not reduced, any soluble soil P could subsequently be tightly adsorbed if biochar was applied. The increasing use of furrow-irrigation in rice requires further evaluation into optimal nutrient management and use of soil amendments like biochar [24].

The variable and alternating soil moisture conditions associated with furrow-irrigation may result in increased biochar degradation, thus rendering the potential effects of biochar addition on soil pH, dissolved P, and other water-soluble nutrients over time less predictable than under flood-irrigated conditions. The objective of this greenhouse study was to evaluate the effects of biochar-source/particle-size combination [i.e., large particle size from Douglas Fir (*Pseudotsuga menziesii*) in Colorado (CO) and small particle size from Southern Yellow Pine (*Pinus echinate*) in Arkansas (AR)] and rate (i.e., 0, 2.5, and 5 Mg ha⁻¹) on soil pH and water-soluble (WS) soil nutrients (i.e., P, Ca, Mg, Fe, Mn, and S) in simulated furrow-irrigated rice on a silt-loam soil over the course of one growing season. It was hypothesized that, by the end of the growing season, soil pH would increase

from all biochar treatments, but that soil pH would increase more from the small-particle-size biochar material from AR due to greater reactivity from the smaller particles. It was also hypothesized that soil pH would increase more from the largest biochar rate for both biochar sources due to the greater influence of more biochar with an initially alkaline pH itself. Furthermore, it was hypothesized that soil P, Ca, Mg, Fe and Mn would decrease as biochar rate increased due to biochar’s adsorptive properties regardless of biochar source, and that the AR biochar source would result in a decrease in WS soil nutrients due to the finer particle size. It was finally hypothesized that WS-S would be unaffected by biochar rate because the predominantly negatively charged biochar would have minimal binding affinity for dissolved-S anions.

Materials and Methods

Soil collection, processing, and initial characterization

Research was conducted in the greenhouse at the Milo J. Shult Agricultural Research and Extension Center (SAREC) between 16 April and 29 September 2023. A Dewitt silt loam soil (fine, smectitic, thermic Typic Albaqualf) was collected from the top 10 to 15 cm in a field that had been cropped to furrow-irrigated rice for at least the past 6 years at the Rice Research and Extension Center near Stuttgart, AR [27]. Soil was moist-sieved through a 6-mm screen to remove roots and air-dried for 7 days at 31 °C.

Six sub-samples of air-dried soil were collected, oven-dried for 48 hours at 70 °C, and sieved through a 2-mm mesh screen for initial soil property determinations. Sand, silt, and clay were determined with a modified, 12-hour hydrometer method [28]. Water-soluble (WS) and Mehlich-3 extractable soil nutrients (i.e., P, Ca, Mg, Fe, Mn and S) were determined by a 1:10 soil mass:water volume ratio, and soil was extracted and analyzed for plant available nutrients using inductively coupled, argon-plasma spectrophotometry [29,30]. While WS are typically smaller in magnitude than weak-acid-extractable nutrient concentrations, WS nutrient concentrations were the focus in this study instead of other weak-acid extractions (i.e., Mehlich-1, Mehlich-3, or Bray) because other extractants have regional importance and are usually better indicators of plant-available nutrients, not nutrient concentrations that may directly interact with soil or biochar particles. In addition, under furrow-irrigation, WS nutrients are not as rapidly released, such as under flood-irrigation, thus it is assumed that WS nutrients best represent dynamic, environmentally relevant nutrient availability based on varying soil moisture conditions compared with other weak-acid extractions. Soil pH and electrical conductivity were determined potentiometrically using a 1:2 soil mass:water suspension volume. Soil organic matter concentration was determined by weight-loss-on-ignition at 360 °C for 2 hours of combustion in a muffle furnace [31]. Total carbon (C) and N were determined by high-temperature combustion (Elementar Americas Inc., Mt. Laurel, NJ) [32]. Soil C:N ratio was calculated from the measured TC and TN concentrations.

The soil used in this study had 15% sand, 74% silt, and 11%

clay, which constituted a soil texture of silt loam (Table 1). The initial soil pH was 6.46, which is optimal for rice production in Arkansas on a silt-loam soil [33]. In addition, initial WS-soil P was 2.7 mg kg⁻¹, which would be categorized as very low for rice production in Arkansas [33]. All initial soil properties are summarized in Table 1.

Treatments and experimental design

Two biochar sources were used in this study: i) A biochar material that was sourced from Douglas Fir feedstock in Colorado, and ii) A biochar material sourced from Southern Yellow Pine feedstock in southeastern Arkansas. The CO biochar had 8-25 mm particles size (i.e., coarse biochar), while the AR biochar had a powder (1-3 mm) constitution. In addition, biochar was applied at three rates equivalent to 0, 2.5, and 5 Mg ha⁻¹. The experimental design was a complete full factorial arranged in a randomized complete block design

Table 1: Summary of initial soil physical and chemical property means (n = 6) and standard errors (SE) for the soil used in this greenhouse experiment.

Soil Property	Mean (± SE)
Sand (g g ⁻¹)	0.15 (0.6)
Silt (g g ⁻¹)	0.74 (0.5)
Clay (g g ⁻¹)	0.11 (0.7)
pH	6.46 (0.1)
Electrical conductivity (dS m ⁻¹)	0.090 (< 0.01)
Water-soluble soil nutrients (mg kg ⁻¹)	
P	2.7 (0.06)
K	27.7 (3.1)
Ca	20.3 (0.4)
Mg	3.1 (0.25)
S	11.6 (0.15)
Na	60.1 (0.45)
Fe	11.2 (2.0)
Zn	0.6 (0.04)
Mn	1.1 (0.07)
Mehlich-3 extractable soil nutrients (mg kg ⁻¹)	
P	25.5 (0.2)
K	97.2 (0.8)
Ca	679 (6.8)
Mg	125 (0.9)
S	15.1 (0.1)
Na	74.6 (0.6)
Fe	365 (3.9)
Zn	15.4 (0.1)
Mn	284 (2.8)
Soil organic matter (%)	2.53 (0.01)
Total C (%)	1.25 (< 0.01)
Total N (%)	0.12 (< 0.01)
C:N ratio	10:1 (< 0.01)

with three blocks. The six biochar source-rate treatment combinations were randomized within each block for a total of 18 experimental units, arranged on a single bench. Each treatment combination therefore had 3 replicates.

Soil tub preparation and management

On 22 April 2023, approximately 26.4 kg of air-dried, sieved soil were placed into 18 plastic tubs (51 cm wide by 67 cm long by 15 cm deep). On 30 April 2023, each tub received N, P, and potassium (K) fertilizer based on initial soil-test recommendations for furrow-irrigated rice production on a silt-loam soil [34]. Each tub received 3.34 g (14.7 kg ha⁻¹) of P in the form of chemically precipitated struvite (i.e., trade name Crystal Green, Ostara, Inc., Vancouver, Canada; fertilizer grade: 6-27-0) [34] and 2.46 g (55.8 kg ha⁻¹) of K as muriate of potash (fertilizer grade: 0-0-60). Nitrogen was added to the tubs in three different applications in the form of N-(n-butyl) thiophosphoric triamide (NBPT)-coated urea (46% N) to reach the recommended 202 kg N ha⁻¹. The N applications were on 21 and 28 May 2023 and 4 June 2023, and the soil was kept relatively dry right before and during applications to prevent N volatilization. All recommended fertilizer application amounts were increased by 20% to account for the limited space for root growth because of the shallow depth of the tubs [35]. Weeds were manually removed as needed over the course of the study. The 18 tubs were arranged on the same greenhouse bench with stiff, wooden planks under the tubs to ensure uniform soil settling and water distribution upon irrigation.

Each tub was manually seeded on 29 April 2023 with a hybrid rice variety 'RT 7302' (RiceTec, Inc., Alvin, TX) based on furrow-irrigation management recommendations from the University of Arkansas [34]. Tub s were manually seeded at a depth of 1.9 cm among 3 rows with 9 seeds per row to account for a recommended seeding rate of 124 seeds m⁻² [33]. Seeds were planted 8-cm from the short edge of the tub, 4-cm from the long edge of the tub, 5-cm between seeds within a row, and 15-cm between rows. To simulate dry seeding, only the soil surface was moistened prior to seeding, and the seeds were manually covered with remaining air-dried soil.

The volumetric water content of the soil was measured in the top 6 cm of each tub with a Theta probe (model SM150, Dynamax, Inc., Houston, TX) daily. Tub s were irrigated 2-3 times per week, with volumes of water dictated by the soil moisture status to reach a volumetric water content of 0.56 cm³ cm⁻³ that represented a value slightly below saturation. The irrigation method is referred to hereafter as simulated furrow-irrigation because the watering regime attempted to mimic the alternating wet-and-dry cycles reported and described in furrow-irrigated rice fields [26].

Soil sample collection over time and analyses

Soil samples were collected from each tub on a weekly basis from 20 May until 16 September 2023 for a total of 19 samples. Soil from each tub was collected by vertically inserting a copper tube with a diameter of 1.4-cm from the soil surface to the bottom of each tub. Samples randomly rotated around the four corners of each tub from week to

week. Soil sampling always occurred in between rice rows. Soil samples were oven-dried for at least 48 hours at 70 °C, ground manually using mortar and pestle, and sieved through a 2-mm screen to remove any coarse fragments in preparation for soil chemical analyses. Soil pH and WS soil nutrients (i.e., P, Mg, Ca, S, Fe, and Mn) were determined using procedures previously described for initial soil characterization.

Statistical Analyses

Based on a repeated-measure design arranged in randomized complete block (RCB), a linear mixed model was used to evaluate the effects of biochar source, biochar rate, time (i.e., weekly sample dates), and their interactions on soil pH and WS soil nutrient (i.e., P, Mg, Ca, S, Fe, and Mn) concentrations using the ASReml package (version 4.1.0.90; Butler, 2018) in R (version 4.3.2, R Foundation for Statistical Computing, Vienna, Austria). Biochar source, biochar rate, time, and blocks were considered fixed effects. The interaction between blocks and other fixed effects were not included in the model. The between-subject factors (i.e., biochar source and biochar rate) were randomized within three blocks in a RCB design at the beginning of the growing season. The repeated sampling of the within-subject factor (i.e., time) were equally spaced in time. Normality was checked using the residual with qqplots. As a result, a normal distribution was used for soil pH, soil WS P, Fe, and S while a gamma distribution was used for soil WS Ca, Mg, and Mn concentrations. Restricted maximum likelihood (REML) was used as the convergence method for the model, where different variance-covariance structures were evaluated, including compound-symmetry, diagonal, autoregressive first order, autoregressive second order, and unstructured. The best fit was determined by the evaluation of the Akaike information criteria (AIC) and the likelihood ratio test (LRT). Heteroskedasticity was included for each variance-covariance structure and statistically compared to the equivalent model defined by homoskedasticity. As a result, all the response variables were characterized by autoregressive first order variance-covariance structure; soil pH, soil WS P, Ca, Fe, and S were characterized by homogenous variance, while soil WS Mg and Mn were characterized by heterogenous variance. Each response variable dataset was complete and balanced. No outliers were observed for any of the response variables. The Wald test was performed on the best-fit model to extract the analysis of variance (ANOVA, type III sum of squares) results for the fixed effects. Pairwise, multiple comparisons were performed using Tukey's method. Significance for all tests was judged at the 0.05 level.

Results and Discussion

Soil pH

Soil pH fluctuated over time throughout the study period and was affected by biochar rate ($P < 0.01$) and time ($P < 0.01$; Table 2). Averaged across biochar source and rate, soil pH ranged from a minimum of 4.9 at 49 days after planting (DAP) to a maximum of 6.2 at 118 DAP (Figure 1). Averaged across biochar source and time, soil pH was 5.69 from the 5 Mg ha⁻¹ rate, which differed from a soil pH of 5.59 from the no-biochar

Table 2: Analysis of variance summary of the effects of biochar source, biochar rate, time, and their interaction on soil pH and water-soluble (WS) phosphorus (P), calcium (Ca), magnesium (Mg), iron (Fe), sulfur (S), manganese (Mn), and sulfur (S) over the 2023 growing season under simulated-furrow-irrigated rice on a silt-loam soil in the greenhouse.

Soil Property	Source of Variation						
	Source	Rate	Time	Source × Rate	Source × Time	Rate × Time	Source × Rate × Time
	<i>P</i>						
pH	0.13	< 0.01	< 0.01	0.25	0.99	0.81	0.99
WS-P	0.04	0.87	< 0.01	0.60	< 0.01	0.98	0.99
WS-Ca	0.68	0.04	< 0.01	< 0.01	0.45	0.03	0.41
WS-Mg	0.07	< 0.01	< 0.01	0.01	0.45	0.08	0.50
WS-Fe	< 0.01	< 0.01	< 0.01	0.02	0.92	0.11	0.97
WS-Mn	0.77	< 0.01	< 0.01	0.29	0.94	0.15	0.65
WS-S	0.16	0.02	< 0.01	0.15	0.88	0.31	0.95

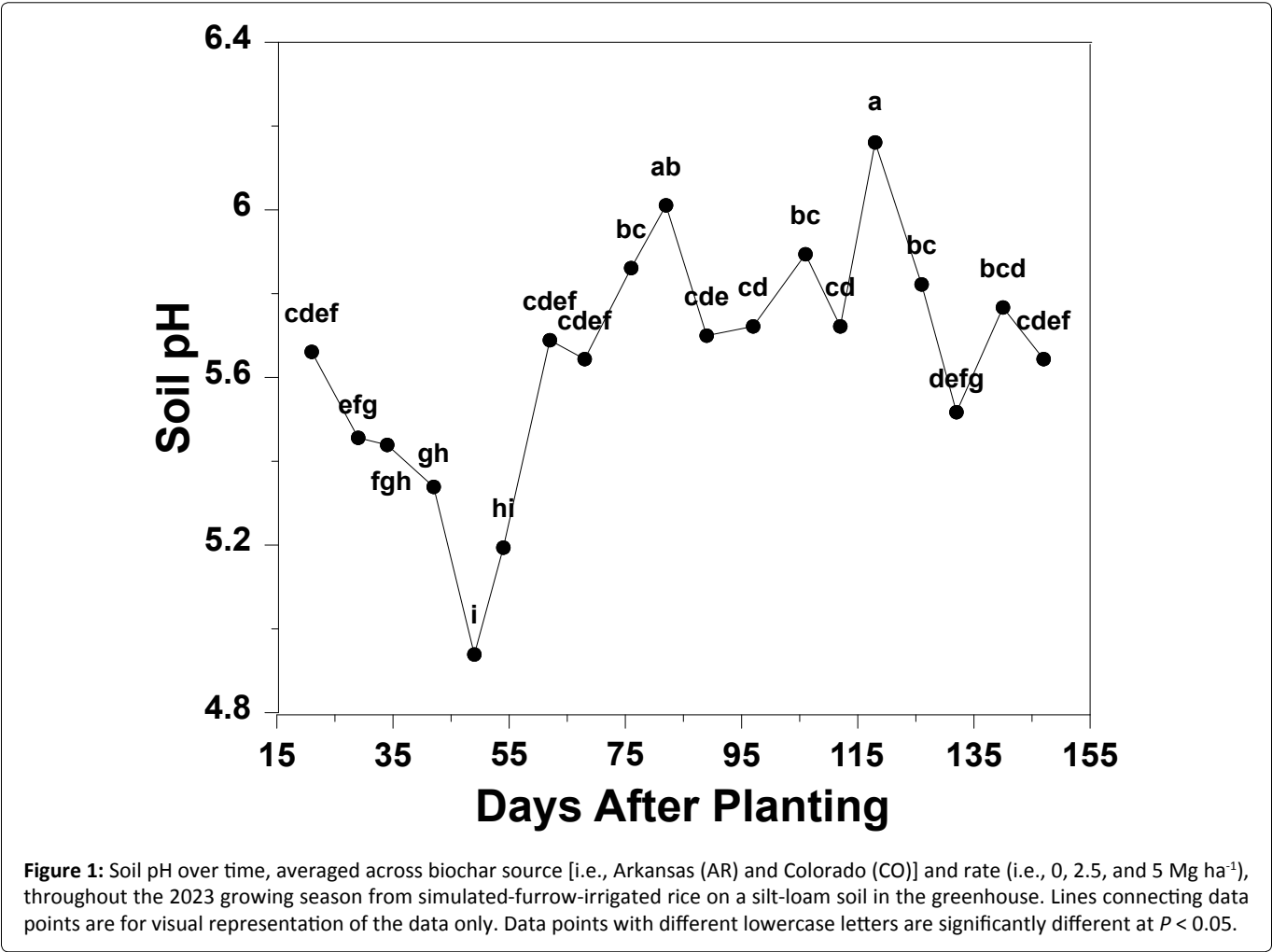


Figure 1: Soil pH over time, averaged across biochar source [i.e., Arkansas (AR) and Colorado (CO)] and rate (i.e., 0, 2.5, and 5 Mg ha⁻¹), throughout the 2023 growing season from simulated-furrow-irrigated rice on a silt-loam soil in the greenhouse. Lines connecting data points are for visual representation of the data only. Data points with different lowercase letters are significantly different at *P* < 0.05.

control. Soil pH was 5.64 from the 2.5 Mg ha⁻¹ rate, which did not differ from either the 5 Mg ha⁻¹ rate or the no-biochar control. Similar to the results of the current study, biochar has been reported to impact soil pH [36–38]. However, the overall decrease in soil pH measured in this study was in contrast to results of other studies, but the other studies did not include a fertilizer-N source [36,37]. The nitrification of ammonium, as the decomposition product of urea, likely contributed

to the decrease in soil pH in the current study, where the largest decrease in soil pH was from the unamended control and without biochar’s ability buffer a change in soil pH. In an incubation study that evaluated the effect of corn (*Zea mays*) stover and switchgrass (*Panicum virgatum*) biochar and rate (i.e., 52-, 104-, and 156 Mg ha⁻¹) on the chemical properties of an acidic (pH < 4.8) clay loam, Chintala, et al. [36] reported that all application rates increased soil pH. Specifically, the

corn stover biochar treatment increased soil pH by 0.73, 0.99, and 1.36 units over the 165-day study period, in which the changes were numerically greater in magnitude than the values reported in the current study. The numerically greater differences in soil pH reported by Chintala, et al. [36], were likely due to the magnitude of the biochar rates that were used being at least 10 times greater than in the current study.

Averaged across biochar source and rate, soil pH was 5.7 at 21 DAP, after having started at a pH of 6.46 prior to any biochar addition (0 DAP, Table 1). Soil pH tended to decrease from 21 to 49 DAP, when the minimum value of 4.9 was reached that however did not differ from the soil pH at 54 DAP (5.2, Figure 1). Soil pH then generally tended to increase until 82 DAP (pH = 6.0), which was followed by 30 days of generally lower soil pH until the season-long peak at 118 DAP (pH = 6.2), which did not differ from the soil pH at 82 DAP (6.0, Figure 1). From 118 DAP to the end of the study, soil pH tended to decrease again, with similar values as at 21 DAP for the remainder of the study period (Figure 1). In contrast to the current study, Chintala, et al. [36] reported a spike in soil pH among all biochar rates (i.e., 52-, 104-, and 156 Mg ha⁻¹), with an increase of at least 0.5 units in the first 15 days of incubation from the corn stover and switchgrass biochars.

The decrease in soil pH during the first 30 days after measurements began may be explained by the applied urea fertilizer being converted into ammonium and then nitrate, which tends to reduce soil pH as hydrogen ions are released in the nitrification process [39]. The relatively large increase in soil pH that occurred after the season-long minimum was achieved at 49 DAP was likely a result of the neutralization of the released hydrogen ions from nitrification of the applied urea. Though the soil pH experienced increases and decreases throughout the growing season under simulated furrow-irrigation, by the end of the growing season, soil pH reverted back to values similar to the soil pH at 21 DAP (Figure 1), most likely due to the soil's buffering capacity and tendency to resist change [40]. The difference, although not formally compared, between initial soil pH (Table 1), prior to any biochar addition, management practice (i.e., irrigation applications, fertilization) implementation, and plant/soil interaction, and the soil pH values during the growing season, along with the differences reported during the season, suggests that soil pH was impacted by some combination of biochar addition, rice cropping, and/or furrow-irrigation.

Water-soluble nutrients

Biochar source and/or biochar rate significantly affected WS-P, -Ca, -Mg, -Fe, -Mn, and -S concentrations over time (Table 2). In contrast to soil pH, WS-P concentrations differed between biochar sources over time ($P < 0.01$; Table 2). Averaged across biochar rates, WS-P concentrations were similar between biochar sources and among measurement dates and generally followed the same temporal pattern at each of the sampling dates, except at 34 DAP. At 34 DAP, the WS-P concentration was eight times greater from the AR biochar source (16.9 mg kg⁻¹) than from the CO biochar source (2.0 mg kg⁻¹; Figure 2A). The spike in WS-P for the AR biochar source was likely due to sampling a random, undissolved

fertilizer pellet. Aside from 34 DAP, WS-P concentrations ranged from 1.4 mg kg⁻¹ at 49 DAP from the CO biochar to 6.2 mg kg⁻¹ at 106 DAP from the AR biochar (Figure 2A). Water-soluble P at 34 DAP was at least 2.6 times greater than the next lowest measured WS-P concentration at 106 DAP (Figure 2A). The numerical increase of WS-P following the sampling at 97 DAP, considering the temporal distance from P-fertilization, might be related to the dissolution of biochar material that released P (Figure 2A).

Studies have reported mixed results when evaluating biochar effects on soil-P availability [41]. A meta-analysis of biochar effects on P availability in agricultural soils reported that biochar application typically increased plant-available P, but biochar rates above 10 Mg ha⁻¹ were generally needed to affect soil-P availability [41]. Thus, in the current study, WS-P concentration was likely unaffected by biochar rate because not enough biochar was added to impact WS-P concentration. However, the chosen biochar rates used in this study represented more realistic and practical biochar application rates than those exceeding 10 Mg ha⁻¹.

Water-soluble Ca differed ($P < 0.01$) among biochar source-rate combinations (Table 2). Averaged across time, WS-Ca concentrations were numerically largest from the 0 Mg ha⁻¹ rate from the AR biochar (23.6 mg kg⁻¹), which only differed from the 5 Mg ha⁻¹ rate from the CO biochar (18.9 mg kg⁻¹; Table 3). An incubation study that evaluated the effects of rape- (*Brassica napus*), paddy-rice, wheat- (*Triticum*), and corn- (*Zea mays*) straw biochars at a rate of approximately 4.2 Mg ha⁻¹ on WS -Na, -Ca, and -Mg in a soil grown with peach (*Prunus persica*) seedlings reported that WS -Ca concentrations from the rape- and wheat-straw biochar treatments were reduced from the control, but paddy-rice straw led to an increase in WS-Ca [42]. Thus, WS-Ca response was likely dependent on biochar source properties, including particle size, feedstock, and adsorption capacity. Furthermore, the WS-Ca levels reported in Wu, et al. [42] were at least 3.2 times greater than from the current study, which could be due to added Ca through irrigation, as experimental pots were watered daily.

In addition, WS-Ca differed ($P = 0.03$) among biochar rates over time (Table 2 and Figure 2B). Averaged across biochar source, WS-Ca concentrations were numerically largest at 49 DAP from the 2.5 Mg ha⁻¹ rate (111 mg kg⁻¹), and only differed at 97 DAP from the 0 Mg ha⁻¹ rate (10 mg kg⁻¹; Figure 2B). All other WS-Ca concentrations were similar to 111 mg kg⁻¹ at 49 DAP from the 2.5 Mg ha⁻¹ rate (Figure 2B). The numerical largest values of WS-Ca measured at 49 DAP corresponded with the lowest value of soil pH reinforcing the strong correlation between pH and nutrient concentrations [43].

Similar to WS-Ca, WS-Mg differed ($P = 0.01$) among biochar source-rate combinations (Table 2). Averaged across time, WS-Mg concentrations were numerically largest from the 0 Mg ha⁻¹ rate from the AR biochar (3.2 mg kg⁻¹), which only differed from the 5 Mg ha⁻¹ rate from the CO biochar (2.1 mg kg⁻¹; Table 3). The decreased WS-Ca and WS-Mg from the CO biochar at the 5 Mg ha⁻¹ rate could have been due to differential biochar adsorption properties and a greater inner porosity from the larger particle size of the CO biochar

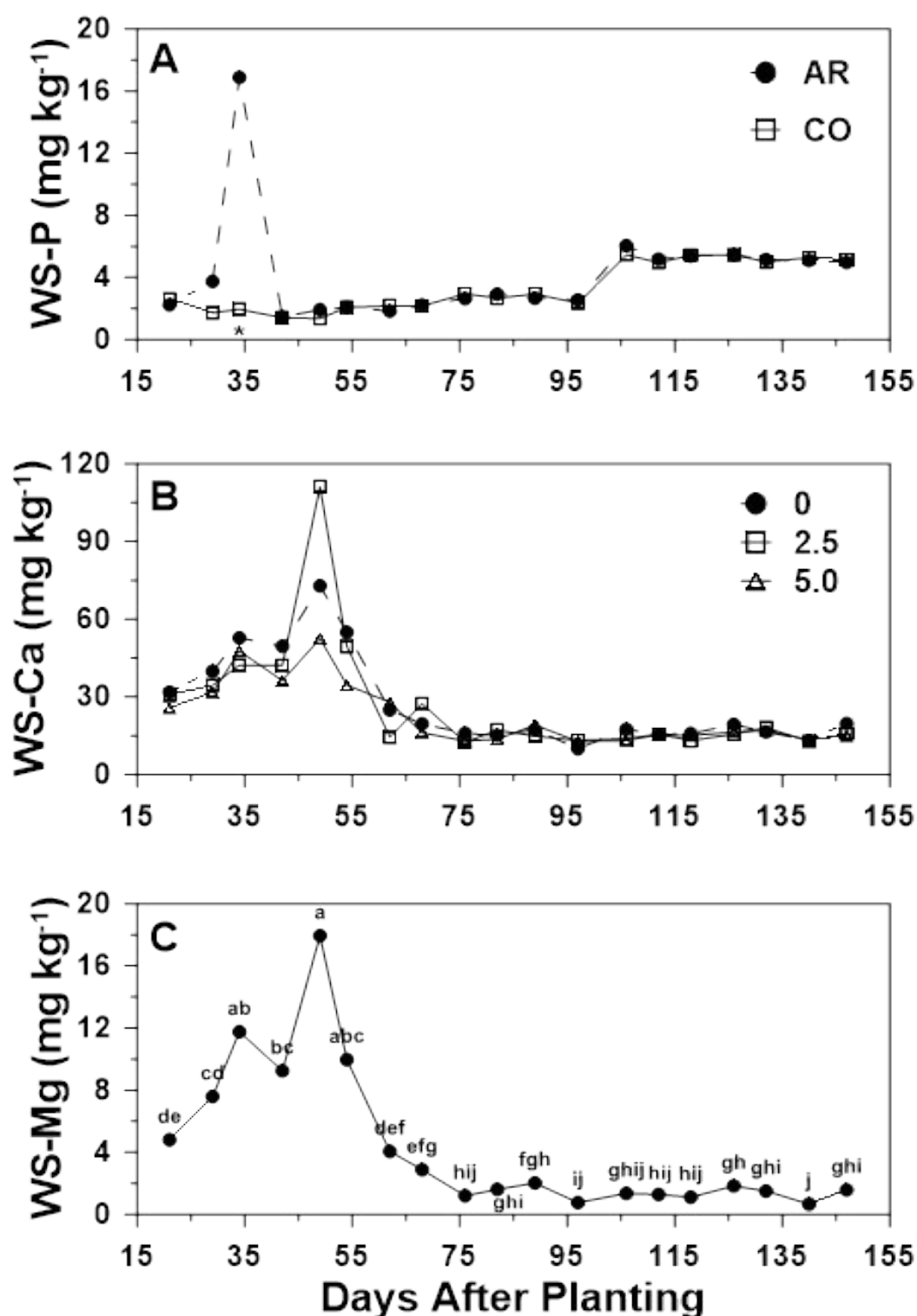


Figure 2: Water-soluble phosphorus (WS-P) from each biochar source [i.e., Arkansas (AR) and Colorado (CO)] over time (panel A), WS-calcium (Ca) from each biochar rate (i.e., 0, 2.5, and 5 Mg ha⁻¹) over time (panel B), and WS-magnesium (Mg) averaged between biochar rate and biochar source over time (panel C) during the 2023 growing season from simulated-furrow-irrigated rice on a silt-loam soil in the greenhouse. Data points within a panel with different lowercase letters are significantly different at $P < 0.05$. Asterisk (*) indicates measurement dates when a significant treatment effect occurred. Lines connecting data points are for visual representation of the data only.

[17]. The larger particle size of the CO biochar and the greater rate at the 5 Mg ha⁻¹ could have allowed the CO biochar to bind more WS-Ca and WS-Mg with exchange sites within

the particles. Munera-Echeverri, et al. [44] described that isopropanol, which is commonly used to clear exchange sites and quantify cation exchange capacity (CEC), could not fully

penetrate the inner micropores of some biochars, leading to potentially lower values in the quantification of biochar CEC.

Similar to soil pH, WS-Mg differed ($P < 0.01$) over time (Table 2 and Figure 2C). Averaged across biochar source and rate, WS-Mg concentration was 4.8 mg kg^{-1} at 21 DAP and generally increased until the season-long peak of 17.9 mg kg^{-1} at 49 DAP, which did not differ from the concentrations at 34 and 54 DAP. After the season-long peak, WS-Mg generally decreased until 76 DAP, where WS-Mg was 1.2 mg kg^{-1} and remained about the same for the remainder of the study (Figure 2C).

Similar to WS-Ca and -Mg, WS-Fe differed ($P = 0.02$) among biochar source-rate combinations (Table 2). Averaged across time, WS-Fe concentrations were numerically largest from the 5 Mg ha^{-1} rate from the CO biochar source (4.8 mg kg^{-1}), which was only similar to the 0 Mg ha^{-1} rate in the CO biochar source (4.0 mg kg^{-1} ; Table 3). In contrast to WS-Ca and WS-Mg, WS-Fe concentration was numerically largest from the CO biochar with the 5 Mg ha^{-1} rate (Table 3). The magnitude of WS-Fe concentration in soil is the result of many potential complex interactions among various elements, soil and biochar particle sizes and surface areas, and biochar adsorptive capacity, thus it was not possible to isolate a single reason why there would be a significant difference in WS-Fe from the CO biochar at the 5 Mg ha^{-1} rate relative to other biochar source-rate combinations. However, one plausible explanation is that, as a reduction-oxidation (redox)-active element, initially oxidized Fe may have been reduced in saturated soil areas within the tubs, which could have increased WS-Fe concentrations [40].

Similar to soil pH, WS-Fe also differed ($P < 0.01$) over time (Figure 3A). Averaged across biochar source and rate, WS-Fe concentration was 2.3 mg kg^{-1} at 21 DAP and increased to 6.3 mg kg^{-1} at 34 DAP, and then decreased to 2.1 mg kg^{-1} at 49 DAP, which was followed by a 33-day increase to the season-long peak of 9.4 mg kg^{-1} (Figure 3A). The season-long peak decreased to 3.9 mg kg^{-1} at 97 DAP, where WS-Fe concentrations were similar over time for the remainder of the study (Figure 3A). Similar to previously, a plausible explanation is that the fluctuating soil moisture conditions from the furrow-irrigation management within the soil tubs created pockets of anaerobic soil that continuously reduced Fe^{+3} to Fe^{+2} , leading to the temporal fluctuations in WS-Fe concentrations [40].

Water-soluble Mn differed ($P < 0.01$) among biochar rates (Table 2). Averaged across biochar source and time, WS-Mn concentrations were numerically greatest from the 0 Mg ha^{-1} rate (4.3 mg kg^{-1}), which was at least 1.2 times greater than from the 2.5 and 5.0 Mg ha^{-1} biochar rates (Table 4). Similar to WS-Ca and -Mg, the decreased WS-Mn concentrations in the 2.5 and 5.0 Mg ha^{-1} biochar rates was likely due to Mn adsorption to negatively charged biochar exchange sites [17].

Similar to soil pH and WS-Fe, WS-Mn differed ($P < 0.01$) over time (Table 2). Averaged across biochar source and rate, WS-Mn concentration was 1.6 mg kg^{-1} at 21 DAP and generally increased to the season-long peak of 17.4 mg kg^{-1} at 49 DAP, which did not differ from the WS-Mn concentration at 54

DAP (Figure 3B). Water-soluble Mn then quickly decreased to 3.7 mg kg^{-1} by 62 DAP and remained about the same for the remainder of the study.

Water-soluble Ca, -Mg, and -Mn all followed a similar temporal trend, where WS-Ca, -Mg, and -Mn all peaked around 49 DAP, then decreased and stabilized for the remainder of the growing season. The similar peaks around 49 DAP aligned closely with the peaks from soil pH, as the season-long minimum of soil pH also occurred at 49 DAP (Figure 1). Normally, lower soil pH tends to decrease available Ca and Mg, but the dissolution of certain minerals, that were either present or formed in the soil, at the low pH could have increased WS-Ca and -Mg concentrations. Furthermore, WS-Mn typically becomes more available as soil pH decreases, thus it is plausible that the increased WS-Mn concentration was due to a decrease in soil pH [40].

Similar to WS-Mn, WS-S differed ($P = 0.02$) among biochar rates (Table 2). However, in contrast to WS-Mn, averaged across biochar source and time, WS-S concentrations were numerically largest from the 2.5 Mg ha^{-1} rate (10.5 mg kg^{-1}), which did not differ from the WS-S concentration at the 0 Mg ha^{-1} rate (10.5 mg kg^{-1} ; Table 4). The decreased WS-S concentration from the 5 Mg ha^{-1} rate could have been due to biochar's partial anion exchange capacity, where carbon functional groups of the organic-derived biochar, that can have both associated negative and positive charges, could have reacted with dissolved-S anions, decreasing WS-S concentrations [45].

Water-soluble S also differed ($P < 0.01$) over time (Table 2). Averaged across biochar source and rate, at 21 DAP, WS-S concentration was 11.4 mg kg^{-1} and WS-S reached the season-long peak of 14.5 mg kg^{-1} by 34 DAP (Figure 3C). Furthermore, WS-S concentration decreased for four weeks to reach a season-long minimum of 7.3 mg kg^{-1} by 62 DAP, then spiked to 10.0 mg kg^{-1} by 68 DAP (Figure 3C). Water-soluble-S concentrations remained similar for 30 days thereafter, until spiking to 13.0 mg kg^{-1} at 118 DAP, which did not differ from the WS-S concentration of 12.4 mg kg^{-1} at 126 DAP (Figure 3C). Water-soluble-S concentrations were similar to 10.8 mg kg^{-1} at 112 DAP for the remainder of the study (Figure 3C). The general increase in WS-S concentrations towards the end of the growing season could have been due to the increase in soil pH, which may have increased WS-S concentrations [40].

Implications

Utilizing biochar as a soil amendment could facilitate soil carbon sequestration and improve soil quality and fertility through enhanced carbon storage, CEC, and water-holding capacity. Based on the results of this study, biochar source and rate can significantly impact soil pH and WS-P, -Ca, -Mg, -Fe, -Mn, and -S over the course of a growing season in furrow-irrigated rice production, which highlights biochar's potential to positively impact the soil environment and agriculture. However, biochar's adsorptive properties appear to bind plant-available nutrients and could cause significant pH fluctuations within a growing season. Excessive pH fluctuation and WS nutrient binding could be problematic in an agronomic setting, while the adsorptive properties of

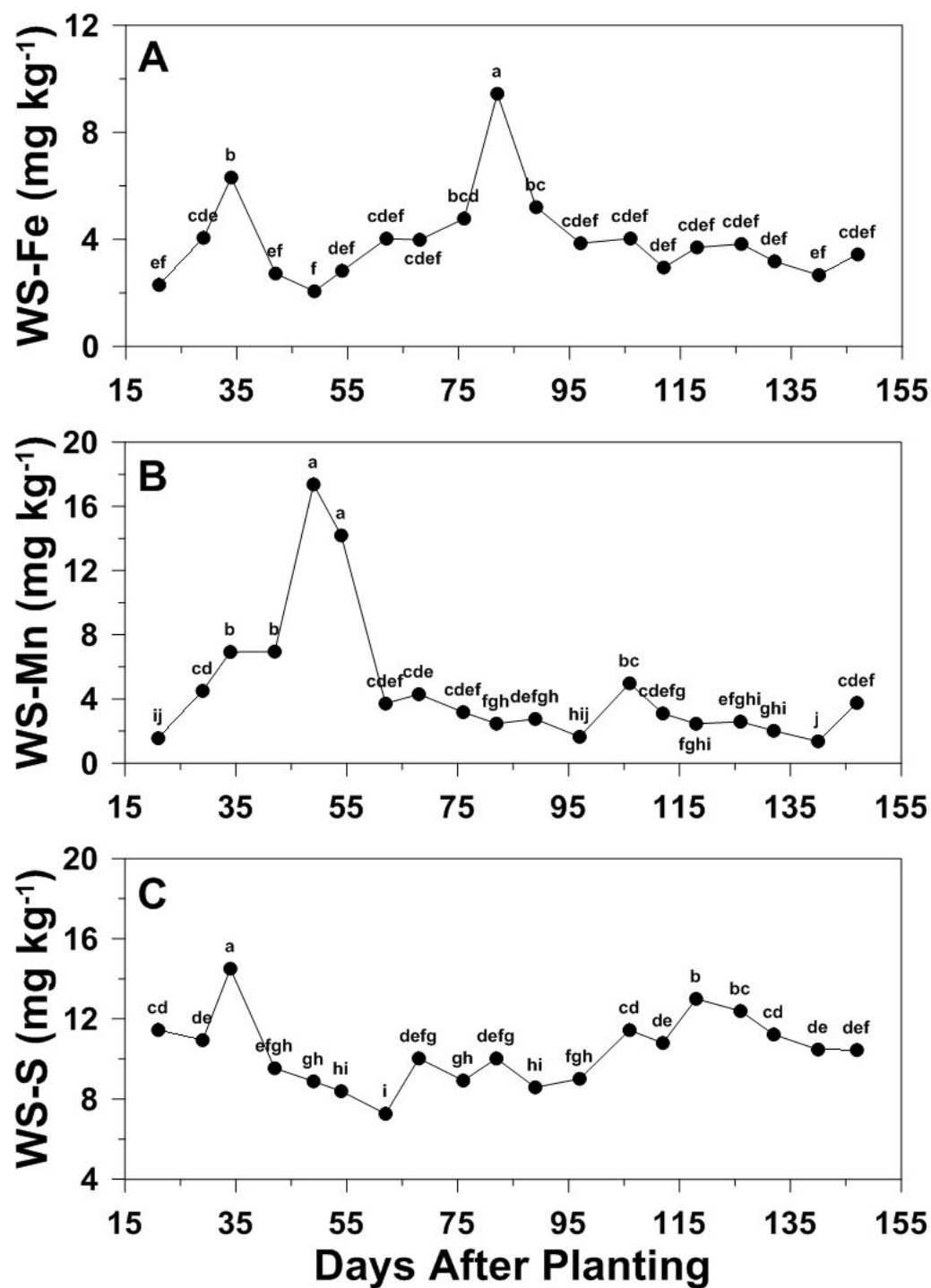


Figure 3: Water-soluble (WS) iron (Fe) (panel A), manganese (Mn) (panel B), and sulfur (S) (panel C) over time during the 2023 growing season from simulated-furrow-irrigated rice on a silt-loam soil in the greenhouse. Data points within a panel with different lowercase letters are significantly different at $P < 0.05$. Lines connecting data points are for visual representation of the data only.

biochar could reduce nutrient leaching and bind soil nutrients close to the soil surface, making essential nutrients potentially more accessible for plants and microbes.

Conclusions

As use of biochar in agroecosystems has gained recent interest, this study evaluated the effect of biochar-source/particle-size combination and rate (i.e., 0, 2.5, and 5 Mg ha⁻¹)

on soil pH and WS soil nutrients (i.e., P, Ca, Mg, Fe, Mn, and S) in simulated furrow-irrigated rice on a silt-loam soil. Contrary to that hypothesized, soil pH was unaffected by biochar source and fluctuated over time, but, similar to that hypothesized, soil pH differed among biochar rates. As hypothesized, WS soil Ca, Mg, and Mn concentrations numerically decreased with biochar treatment from both sources, but, contrary to that hypothesized, the measured decrease was greater from

Table 3: Summary of the combined effects of biochar source [i.e., Arkansas (AR) and Colorado (CO)] and rate on water-soluble (WS) calcium (Ca), magnesium (Mg), and iron (Fe) over the 2023 growing season under simulated-furrow-irrigated rice on a silt-loam soil in the greenhouse.

Biochar Source	Rate (Mg ha ⁻¹)	WS-Ca (mg kg ⁻¹)	WS-Mg (mg kg ⁻¹)	WS-Fe (mg kg ⁻¹)
AR	0	23.6 a [†]	3.2 a	3.7 b
	2.5	20.6 ab	2.5 ab	3.8 b
	5	22.2 ab	2.6 ab	3.8 b
CO	0	23.0 a	3.0 ab	4.0 ab
	2.5	23.2 a	2.9 ab	3.7 b
	5	18.9 b	2.1 b	4.8 a

[†]Means in a column with different letters are different at *P* < 0.05

Table 4: Summary of the effect of biochar rate on water-soluble (WS) manganese (Mn) and sulfur (S) over the 2023 growing season under simulated-furrow-irrigated rice on a silt-loam soil in the greenhouse.

Rate (Mg ha ⁻¹)	WS-Mn (mg kg ⁻¹)	WS-S (mg kg ⁻¹)
0	4.3 a [†]	10.1 ab
2.5	3.5 b	10.5 a
5	3.3 b	10.1 b

[†]Means in a column with different letters are different at *P* < 0.05

the CO biochar with the larger particle size instead of the AR biochar with the smaller particle size. Contrary to what hypothesized, WS-P was relatively unaffected by biochar source and rate, and WS-Fe increased from the CO biochar at the 5 Mg ha⁻¹ rate. Contrary to what hypothesized, WS-S decreased from the 2.5 and 5 Mg ha⁻¹ biochar rates. Overall, the results of this study supported biochar as a generally positive soil amendment, but further quantification of biochar’s adsorptive capacity, especially in a production-scale field, should be further investigated.

Acknowledgments

This study was supported by the Arkansas Rice Research and Promotion Board and the University of Arkansas System Division of Agriculture.

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DOI: 10.36959/973/446