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Article

Pine Woodchip Biochar Impact on Soil Nutrient Concentrations and Corn Yield in a Silt Loam in the Mid-Southern U.S.

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Abstract: Biochar has altered plant yields and soil nutrient availability in tropical soils, but less research exists involving biochar additions to temperate cropping systems. Of the existing research, results vary based on soil texture, crop grown, and biochar properties. The objective of this study was to determine the effects of pine (*Pinus* spp.) woodchip biochar at 0, 5, and 10 $Mg \cdot ha^{-1}$ rates combined with urea nitrogen (N) on soil chemical properties and corn (*Zea mays* L.) yield under field conditions in the first growing season after biochar addition in a silt-loam alluvial soil. Biochar combined with fertilizer numerically increased corn yields, while biochar alone numerically decreased corn yields, compared to a non-amended control. Corn nitrogen uptake efficiency (NUE) was greater with 10 $Mg \cdot ha^{-1}$ biochar compared to no biochar. There were limited biochar effects on soil nutrients, but biochar decreased nitrate, total dissolved N, and Mehlich-3 extractable sulfur and manganese concentrations in the top 10 cm. Pine woodchip biochar combined with N fertilizer has the potential to improve corn production when grown in silt-loam soil in the mid-southern U.S. by improving NUE and increasing yield. Further research will be important to determine impacts as biochar ages in the soil.

Keywords: biochar; soil microorganisms; temperate agroecosystem; corn production

1. Introduction

Fertile and carbon-rich soils have been discovered throughout the Amazon River basin, an area where soils are typically nutrient leached and weathered [1,2]. Terra Preta soils, or Amazonian dark earth soils, occur in locations classified as Oxisols and Ultisols with similar mineralogical properties as surrounding soils. However, the Terra Preta soils are differentiated by their darker color due to large amounts of organic matter (reportedly nearly 90 $g \cdot kg^{-1}$ in the surface horizon or 250 $Mg \cdot ha^{-1} \cdot m^{-1}$ compared to around 30 $g \cdot kg^{-1}$ or 100 $Mg \cdot ha^{-1} \cdot m^{-1}$ in surrounding Oxisols), charcoal, and A horizons ranging from 30 to 60 cm opposed to the typical 10- to 15-cm depths in adjacent soils [1,3–5].

In light of the observed fertility of Amazonian dark earth soils, presumably due in part to the presence of charcoal, research is ongoing to determine the effects of charcoal addition in different soils and charcoal's agronomic impact as a soil amendment. Biochar, or charcoal added to soils for the purpose of improving agronomic soil properties, can be produced by pyrolysis. Pyrolysis is the thermal conversion of biomass under no or minimal oxygen conditions with temperatures generally between 300 and 700 °C [6–9]. The wide range of biochar products produced through various combinations of biomass types and production conditions can lead to different results when applied to various soils and at different application rates.

With a focus beyond tropical soils, research has been conducted to investigate the effects of soil application of various biochar products in temperate regions. For example, birch (*Betula* spp.) wood biochar applied at 20 $Mg \cdot ha^{-1}$ to a sandy loam (Typic Hapludalf) in Denmark did not increase oat (*Avena sativa* L.) biomass or yield, but barley (*Hordeum vulgare* L.) grown the following year experienced significant biomass increases with biochar addition [10]. In a sandy clay loam (Eutric Cambisol) in Wales, woodchip biochar applied at 25 and 50 $Mg \cdot ha^{-1}$ had no effect on corn (*Zea mays* L.) growth or nutrient concentration, although hay grass (*Dactylis glomerata* L.) grown the year after corn experienced increased foliar N with 50 $Mg \cdot ha^{-1}$ biochar compared to the control and increased biomass in the third year of the study [11]. Poultry litter biochar addition to an Alfisol in Australia increased radish (*Raphanus sativus* L.) yield with increasing biochar addition from rates of 10, 25, to 50 $Mg \cdot ha^{-1}$ in a pot trial [12]. In a sandy loam in Belgium, there was a general reduction in soil nitrate availability and nitrogen-use efficiency as well as reduced biomass in radish and spring barley with the addition of 10 $g \cdot kg^{-1}$ willow (*Salix* spp.) or pine (*Pinus* spp.) biochar, with greater reduction in soil nitrate with the willow biochar compared to the pine biochar in the pot study [13]. Considering these and other observations, the results regarding biochar addition have been mixed based on the biochar products used, soil textures, and the specific crops grown in these temperate region studies.

Corn is an important commodity crop in the United States, with nearly 35.5 million ha harvested for grain, and grain production at a record high of almost 0.49 billion m^3 across the country in 2013 [14]. Corn requires substantial N inputs, with 0.45 kg N expected to be taken up for every 25 kg corn grain produced (bu corn) [15]. If biochar can enhance soil fertility of corn production systems in temperate agroecosystems, there is potential to improve soil quality characteristics, increase yields, and reduce commercial fertilizer-N inputs, thereby improving the sustainability of production systems. Corn yield but not nutrient uptake increased in field research in Iowa after the addition of mixed hardwood biochar (96 $Mg \cdot ha^{-1}$) to a Typic Hapludoll [16]. Peanut (*Arachis hypogaea* L.) hull biochar increased soil N concentration but did not affect corn tissue N when applied at 11.2 and 22.4 $Mg \cdot ha^{-1}$ to loamy sand in

Georgia, while pine woodchip biochar did not increase soil N or tissue N [17]. Corn yield was reduced with the addition of 11.2 Mg·ha^{-1} of the peanut hull biochar, but the application of 22.4 Mg·ha^{-1} produced yields similar to those of the unamended control. The pine woodchip biochar decreased corn yield with increasing application rate in the first year after biochar addition, but the second corn crop experienced increased grain yields with biochar application compared to the control [17]. Corn biomass increased with giant reed (*Arundo donax* L.) biochar irrespective of biochar concentration (0.1, 0.2, and 0.5 g·kg^{-1}) when added to a silt loam in a pot experiment in China [18]. Plant residue biochars tended to increase corn biomass when applied at rates of 2.6 and 6.5 Mg·ha^{-1} to a Glossoboric Hapludalf in a greenhouse study in New York, with minimal differences at greater applications (*i.e.*, 26 and 91 Mg·ha^{-1}) [19]. Poultry manure biochar application produced a similar pattern, while food waste, paper mill waste, and dairy manure applied at 26 and 91 Mg·ha^{-1} tended to decrease corn biomass [19].

Although long-term effects of biochar on soil properties and processes may differ from short-term, there are a range of mechanisms by which biochar may impact soils and agronomic production in the first growing season. Biochar has decreased organic N release but increased nitrification [20]. Maize biochar to loamy sand initially sorbed released ammonium, but also showed short-term increases in N mineralization and nitrification [21]. Through effects on soil properties and alteration of N cycling processes, biochar may interact with fertilizer to increase N availability and N uptake efficiency in plants. Giant reed biochar reduced N leaching and increased nitrogen utilization efficiency, or the amount of corn biomass produced per unit N, in pot experiments [18].

In Arkansas, nearly 60% of the state, or 7.5 million ha (18.6 million ac), is commercial timberland [22]. Thus, pine woodchip biochar could be a potential use of regional wood waste from the forestry industry. Additionally, to the authors' knowledge, there are no published studies conducted in Arkansas regarding biochar application to soils. The objective of this study was to determine the effects of pine woodchip biochar in combination with varying amounts of inorganic N fertilizer on soil biological and chemical properties and corn yield under field conditions in the first growing season after biochar addition. Specifically, it was hypothesized that adequate corn yields and nutrient uptake would occur with biochar addition that could reduce inorganic fertilizer inputs because of increased N uptake efficiency. Despite any sorption or immobilization that may be caused by biochar, more N would be taken up by plants in the presence of biochar and fertilizer than in the absence of biochar.

2. Results

2.1. Initial Biochar and Soil Properties

The pine woodchip biochar had an alkaline pH, EC over 5 dS·m^{-1}, and a C:N ratio of 366:1 (Table 1). The surface texture of the soil was confirmed to be silt loam with percentages of sand, silt, and clay of 26, 65, and 9%, respectively (Table 2). The soil possessed a near-neutral pH of 6.4 and EC of 0.16 dS·m^{-1}. Ideal soil pH for corn growth ranges from 5.8–7 [23]. Since the initial soil pH fell within this range, no additional liming was necessary. All initial soil property variables except for Mehlich-3 extractable soil P were statistically similar among all plots (Table 2). Mehlich-3 soil P ranged from 29.3 to 35.2 μg·g^{-1}. Soil bulk density differed among fertilizer-biochar treatment combinations when analyzed during the middle of the experiment (data not shown). However, a clear pattern was lacking between fertilizer rate and biochar rate in terms of how treatments affected soil bulk density, which ranged from 1.18 to 1.33 g·cm^{-3}.

Table 1. Initial mean (± standard error (SE)) pH, electrical conductivity (EC), total carbon (C), total nitrogen (N), C:N ratio, and total recoverable mineral concentrations determined using a nitric acid digest for pine (*Pinus* spp.) woodchip biochar (*n* = 2).

Biochar Property	**Mean (± SE)**
pH [a]	8.7 (0.03)
EC (dS·m^{-1}) [a]	5.3 (0.2)
Total C (mg·g^{-1})	244.5 (21)
Total N (mg·g^{-1})	0.7 (0.2)
C:N ratio	366:1 (64)
Potassium (mg·g^{-1})	2.1 (0.1)
Calcium (mg·g^{-1})	10.1 (0.5)
Magnesium (mg·g^{-1})	2.7 (0.2)
Phosphorus (μg·g^{-1})	770.5 (17)
Sulfur (μg·g^{-1})	128.5 (1.5)
Sodium (μg·g^{-1})	321.5 (13)
Iron (μg·g^{-1})	868.0 (57)
Manganese (μg·g^{-1})	420.5 (30)
Copper (μg·g^{-1})	6.5 (0.04)
Boron (μg·g^{-1})	10.4 (0.7)
Zinc (μg·g^{-1}) [b]	0.01 (0)

[a] pH and EC were determined using a 1:2 (wt:vol) soil:water mixture; [b] Zinc in the woodchip biochar was below the detection limit of the method. Therefore, the detection limit of 0.01 was used for statistical analysis.

Table 2. Mean (± standard error (SE)) soil properties for the Razort silt loam prior to treatment (*n* = 36).

Soil Property	**Mean (± SE)**
Particle-size distribution (g·g^{-1})	
Sand	0.3 (0.3)
Silt	0.6 (0.3)
Clay	0.1 (0.2)
pH [a]	6.4 (0.03)
Electrical conductivity (dS·m^{-1}) [a]	0.2 (0.1)
Organic matter (mg·g^{-1})	27.1 (0.4)
Dissolved organic carbon (C) (μg·g^{-1})	33.8 (1.2)
Microbial biomass C (μg·g^{-1})	46.1 (2.3)
Microbial biomass nitrogen (N) (μg·g^{-1})	8.8 (0.3)
Microbial biomass C:N ratio	5.3:1 (0.2)
Dissolved total N (μg·g^{-1})	9.2 (0.2)
Nitrate-N (μg·g^{-1})	7.6 (0.2)
Ammonium-N (μg g^{-1})	0.02 (0.02)

Table 2. *Cont.*

Soil Property	**Mean (± SE)**
Inorganic N ($\mu g \cdot g^{-1}$)	7.6 (0.2)
Dissolved organic N ($\mu g \cdot g^{-1}$)	1.6 (0.1)
Acid phosphatase activities ($\mu g \cdot g^{-1} \cdot h^{-1}$)	271.9 (14)
Alkaline phosphatase activities ($\mu g \cdot g^{-1} \cdot h^{-1}$)	102.8 (7.2)
Water soluble phosphorus ($\mu g \cdot g^{-1}$)	5.3 (0.3)
Mehlich-3 extractable phosphorus ($\mu g \cdot g^{-1}$)	31.7 (1.1)
Mehlich-3 extractable potassium ($\mu g \cdot g^{-1}$)	104.4 (4.7)
Mehlich-3 extractable calcium ($\mu g \cdot g^{-1}$)	923.4 (13)
Mehlich-3 extractable magnesium ($\mu g \cdot g^{-1}$)	46.4 (0.8)
Mehlich-3 extractable sulfur ($\mu g \cdot g^{-1}$)	5.5 (0.2)
Mehlich-3 extractable iron ($\mu g \cdot g^{-1}$)	51.1 (1.7)
Mehlich-3 extractable manganese ($\mu g \cdot g^{-1}$)	161.7 (3.7)
Mehlich-3 extractable copper ($\mu g \cdot g^{-1}$)	1.9 (0.1)

[a] pH and EC were determined using a 1:2 (wt:vol) soil:water mixture.

2.2. Post-Harvest Soil Characteristics

In terms of treatment effects on various post-harvest soil characteristics, biochar had minimal influence. Neither fertilizer nor biochar as an interaction or main effect had an impact on microbial biomass C, N, or C:N (Table 3). However, alkaline phosphatase enzyme activities differed among fertilizer-biochar treatment combinations. Alkaline phosphatase enzyme activities were greater in the no fertilizer with 5 $Mg \cdot ha^{-1}$ biochar treatment combination and the 112 $kg \cdot ha^{-1}$ fertilizer with 0 $Mg \cdot ha^{-1}$ biochar treatment combination than those in the 10 $Mg \cdot ha^{-1}$ biochar with 224 $kg \cdot ha^{-1}$ fertilizer combination and the no fertilizer with 0 or 10 $Mg \cdot ha^{-1}$ biochar treatment combinations (Figure 1). Water-soluble P also differed among fertilizer-biochar treatment combinations (Table 3), with similar concentrations across fertilizer rates with 10 $Mg \cdot ha^{-1}$ biochar, but with greater WSP in the 224 $kg \cdot ha^{-1}$ fertilizer and no biochar treatment combination compared to other treatments without biochar (Figure 2). Among the 5 $Mg \cdot ha^{-1}$ biochar treatments, WSP concentrations were lowest with the 112 $kg \cdot ha^{-1}$ fertilizer rate.

Table 3. Analysis of variance summary of the effects of fertilizer, biochar, and their interaction (Fert × BC) in post-harvest soil and on corn plant variables.

Soil Variable	***p*-Value**		
	Fertilizer	**Biochar**	**Fert × BC**
Soil moisture	0.754	0.065	0.335
pH	0.248	0.105	0.417
Electrical conductivity	0.639	0.308	0.865
Soil organic matter	0.587	0.104	0.671
Dissolved organic carbon (C)	0.041 *	0.356	0.241
Dissolved total nitrogen (N)	0.093	0.022 *	0.727
Microbial biomass C	0.257	0.778	0.435
Microbial biomass N	0.255	0.768	0.477
Microbial biomass C:N	0.385	0.577	0.419

Table 3. *Cont.*

Soil Variable	***p*-Value**		
	Fertilizer	**Biochar**	**Fert × BC**
Nitrate-N	0.031 *	0.044 *	0.570
Ammonium-N	1.000	1.000	1.000
Inorganic N	0.031 *	0.044 *	0.570
Dissolved organic N	0.002 *	0.644	0.203
Acid phosphatase activities	0.513	0.603	0.882
Alkaline phosphatase activities	0.143	0.235	0.046 *
Water soluble phosphorus	0.015 *	0.767	0.045 *
Mehlich-3 extractable phosphorus	0.103	0.403	0.661
Mehlich-3 extractable potassium	0.708	0.697	0.785
Mehlich-3 extractable calcium	0.094	0.146	0.064
Mehlich-3 extractable magnesium	0.641	0.580	0.903
Mehlich-3 extractable sulfur	0.582	0.008 *	0.519
Mehlich-3 extractable iron	0.035 *	0.453	0.520
Mehlich-3 extractable manganese	0.150	0.045 *	0.105
Mehlich-3 extractable copper	0.952	0.226	0.942
Yield	<0.001 *	0.439	0.011 *
Grain total N	0.014 *	0.491	0.477
NUE	0.170	0.003 *	0.300
Ear-leaf weight	0.005 *	0.548	0.333
Ear-leaf N	<0.001 *	0.555	0.710

* $p < 0.05$.

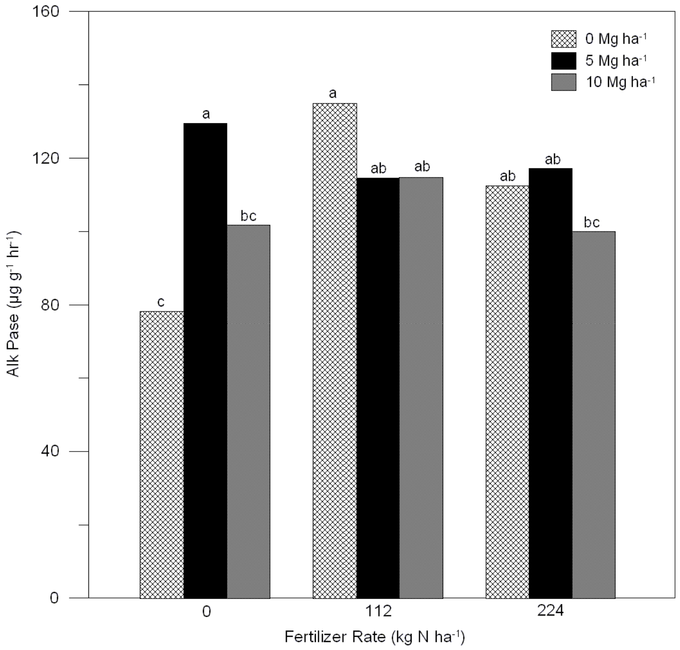

Figure 1. Alkaline phosphatase (Alk Pase) enzyme activity as influenced by pine woodchip biochar and fertilizer rates. The fertilizer rates are 0, 112, and 224 kg·nitrogen (N)·ha^{-1} rates. The biochar treatments are displayed in the shaded boxes at rates of 0, 5, and 10 Mg·ha^{-1}. Bars with different letters are statistically different from each other ($p < 0.05$).

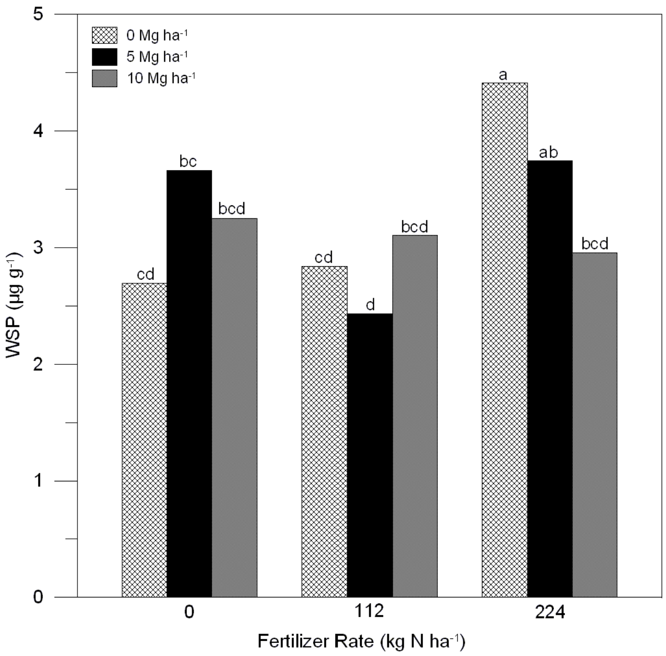

Figure 2. Water soluble phosphorus (WSP) concentrations as influenced by pine woodchip biochar and fertilizer rates. The fertilizer rates are 0, 112, and 224 kg·nitrogen (N)·ha^{-1} rates. The biochar treatments are displayed in the shaded boxes at rates of 0, 5, and 10 Mg·ha^{-1}. Bars with different letters are statistically different from each other ($p < 0.05$).

Nitrate-N and dissolved organic N concentrations differed among fertilizer rates (Table 3). Nitrate increased with the 224 kg·ha^{-1} fertilizer treatment compared to the 0 kg·ha^{-1} fertilizer treatment, while DON was lower with the 224 kg·ha^{-1} fertilizer treatment compared to the 0 and 112 kg·ha^{-1} fertilizer treatments (Table 4). Nitrate and DTN concentrations differed among biochar rates (Table 3). The addition of biochar, irrespective of rate, decreased dissolved total N concentrations compared to no biochar addition, while nitrate concentrations decreased with the 10 Mg·ha^{-1} biochar treatment compared to no biochar addition (Table 5).

Table 4. Mean corn grain total nitrogen (N), ear-leaf weight, and ear-leaf N concentration, and soil dissolved organic N (DON), nitrate (NO_3^--N), inorganic N, dissolved organic C (DOC), and Mehlich-3 extractable soil iron concentration as affected by fertilizer rate.

Plant/Soil	Variable	Fertilizer Rate (kg·N·ha^{-1})		
		0	112	224
Plant	Grain total N (mg·g^{-1})	11.1 b	11.4 b	12.4 a
	Ear-leaf weight (g)	14.0 b	15.7 a	16.7 a
	Ear-leaf N (mg·g^{-1})	23.1 b	26.7 a	28.0 a
Soil	DON (µg·g^{-1})	2.0 a	2.1 a	1.6 b
	NO_3^--N (µg·g^{-1})	2.4 b	2.8 ab	3.9 a
	Inorganic N (µg·g^{-1})	2.4 b	2.8 ab	3.9 a
	DOC (µg·g^{-1})	8.6 b	12.4 a	11.0 ab
	Iron (µg·g^{-1})	25.3 b	28.5 ab	30.6 a

Means followed by different letters in the same row are statistically different ($p < 0.05$).

Table 5. Mean nitrogen uptake efficiency (NUE) in corn and soil dissolved total N (DTN), nitrate (NO_3^--N), inorganic N, and Mehlich-3 extractable soil sulfur and manganese concentrations as affected by biochar rate.

Plant/Soil	**Variable**	**Biochar Rate ($Mg \cdot ha^{-1}$)**		
		0	**5**	**10**
Plant	NUE (%)	12.2 b	21.8 b	44.4 a
Soil	DTN ($\mu g \cdot g^{-1}$)	5.7 a	4.8 b	4.4 b
	NO_3^--N ($\mu g \cdot g^{-1}$)	3.8 a	2.8 ab	2.5 b
	Inorganic N ($\mu g \cdot g^{-1}$)	3.8 a	2.8 ab	2.5 b
	Sulfur ($\mu g \cdot g^{-1}$)	12.3 a	11.4 a	9.4 b
	Manganese ($\mu g \cdot g^{-1}$)	80.6 a	78.2 ab	72.3 b

Means followed by different letters in the same row are statistically different ($p < 0.05$).

Dissolved organic C concentrations increased (Table 3) with the 112 $kg \cdot ha^{-1}$ compared to the 0 $kg \cdot ha^{-1}$ fertilizer treatment (Table 4). Mehlich-3 extractable soil S and Mn differed among biochar rates (Table 3). Soil S decreased with the 10 $Mg \cdot ha^{-1}$ biochar treatment compared to the 0 and 5 $Mg \cdot ha^{-1}$ treatments (Table 5). Manganese decreased with the 10 $Mg \cdot ha^{-1}$ biochar treatment compared to the no biochar treatment. Conversely, Fe increased (Table 3) with the 224 $kg \cdot ha^{-1}$ fertilizer treatment compared to the no fertilizer treatment (Table 4).

2.3. Corn Characteristics

Corn yield differed among fertilizer-biochar treatment combinations (Table 3), with the 224 $kg \cdot N \cdot ha^{-1}$ fertilizer and 10 $Mg \cdot ha^{-1}$ biochar treatment combination resulting in a greater yield than those produced by treatments with no fertilizer or no biochar (Figure 3). Yields were similar between the 10 $Mg \cdot ha^{-1}$ biochar treatment combined with either the 112 or 224 $kg \cdot ha^{-1}$ fertilizer N. Yields produced with the 5 $Mg \cdot ha^{-1}$ biochar treatment combined with the 112 or 224 $kg \cdot ha^{-1}$ fertilizer treatment were similar to yields produced with all treatment combinations involving fertilizer, except the 112 $kg \cdot ha^{-1}$ fertilizer and 5 $Mg \cdot ha^{-1}$ biochar treatment combination produced lower yields than the 224 $kg \cdot ha^{-1}$ fertilizer and 10 $Mg \cdot ha^{-1}$ biochar combination. When no fertilizer was applied, the 10 $Mg \cdot ha^{-1}$ biochar treatment reduced yield compared to the no fertilizer and no biochar treatment combination.

In contrast to the differences in corn yield among fertilizer-biochar treatment combinations, grain N concentrations differed among fertilizer rates (Table 3). The 224 $kg \cdot ha^{-1}$ fertilizer treatment produced greater grain N than the 112 $kg \cdot ha^{-1}$ fertilizer treatment and the no fertilizer treatment (Table 4). The addition of fertilizer at either rate increased ear-leaf weight and ear-leaf N concentrations compared to the no fertilizer treatment. While biochar did not significantly affect (Table 3) grain N concentrations, ear-leaf weight, or ear-leaf N, NUE was greatest (Table 3) with the 10 $Mg \cdot ha^{-1}$ biochar treatment compared to the 5 or 0 $Mg \cdot ha^{-1}$ biochar treatments (Table 5).

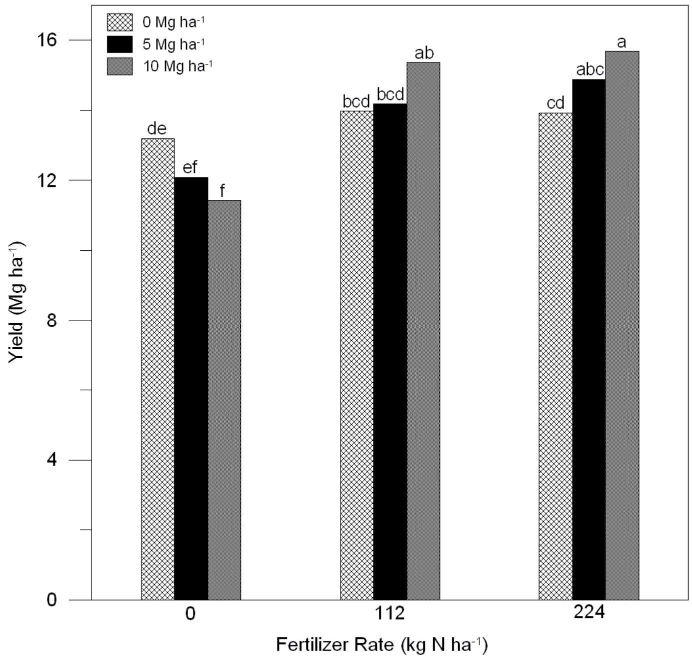

Figure 3. Corn yield as influenced by pine woodchip biochar and fertilizer rates. The fertilizer rates are 0, 112, and 224 kg·nitrogen (N)·ha^{-1} rates. The biochar treatments are displayed in the shaded boxes at rates of 0, 5, and 10 Mg·ha^{-1}. Bars with different letters are statistically different from each other ($p < 0.05$).

3. Discussion

Average corn yields in Arkansas for 2012 and 2013 were close to 11.3 Mg·ha^{-1} and consistent with the lowest yields in this study [14]. The above-state-average yields with the no fertilizer and no biochar treatment were potentially due to residual N in the soil. Furthermore, variety testing in 2013 of irrigated corn in silt loam soil using the specific corn variety used in this study, DEKALB DKC64-69, yielded 15.8 Mg·ha^{-1} when grown at three other University of Arkansas Experiment stations (Stuttgart, Rohwer, and Bell Farm) [24]. The greatest yields from this study were consistent with those reported from the variety testing in the same growing year. In this field experiment, the addition of biochar combined with fertilizer improved crop production to reach the yield potential achieved in the variety testing trials.

Corn yields actually decreased with the addition of 10 Mg·ha^{-1} biochar in the no fertilizer treatment compared to the no fertilizer and no biochar treatment combination. There was the possibility of biochar increasing microbial immobilization or sorption of soil N in the treatments lacking fertilizer. Three Wisconsin cropped soils, a Typic Hapludalf (clay loam), Typic Udipamment (loamy sand), and Typic Argiudoll (silt loam), all experienced increased microbial biomass and activity with mixed manure and pine biochar application in a soil microcosm experiment; microbial biomass and activity increased with increasing biochar concentration (0, 10, 25, 50, and 100 g·kg^{-1}) and extractable-N decreased [25]. Streubel *et al.* [26] reported decreased N mineralization in a silt loam with wood biochar in Washington. Immobilization of soil or biochar N from microbial decomposition of the pine woodchip biochar may have occurred in this study based on the wide C:N ratio of the biochar, but there was no significant biochar effect on microbial biomass C, N, or C:N ratio, thus failing to provide support that changes in the microbial community size explain lower yield in the absence of fertilizer, or explain differences in soil N concentrations or NUE of corn with biochar treatments.

With the addition of fertilizer, corn yield increased numerically with biochar additions, so any reduction of N availability by biochar was presumably not negatively impacting yield when inorganic N fertilizer was applied. Biochar has increased ammonium-N sorption and decreased inorganic N leaching by increasing water holding capacity of soil, N immobilization, and ammonium sorption [18]. Various studies have suggested mechanisms by which biochar may retain N in soil. Zheng *et al.* [18] reported reduced nitrate leaching after nitrate or ammonium fertilizer addition as well as increased microbial activity in giant reed biochar-amended soil. Increased N immobilization in the presence of biochar could have temporarily retained the N in the organic form and reduced the potential for inorganic N leaching [18]. In a study by Güereña *et al.* [27], there was less N leaching with biochar addition combined with the recommended N fertilizer rate than with the fertilizer addition alone. The increases in soil N retention were thought to be due to increases in microbial biomass and thus N immobilization, lower gaseous and erosion losses, and increased retention of organic N on biochar surfaces. Biochar may increase N cycling and availability by providing habitat for microbes and altering food web dynamics [20]. A hardwood biochar decreased N leached from a manure-amended Typic Hapludoll [28], while increasing total soil N [29].

Cycling of N between organic and inorganic pools may be decoupled in the presence of biochar and fertilizer, promoting retention of organic C and N in soil, while increasing plant availability and uptake utilization of fertilizer N [20]. Gajić and Koch [30] found hydrochar reduced early beet growth in a pot and field trial, but that N fertilizer could alleviate yield reductions. They suggested that early plant growth was inhibited by increased microbial immobilization of N especially with use of higher C:N char. Nitrogen may be mineralized later in the growing season, but fertilizer was necessary to compensate for low nutrient availability during early plant growth stages [30].

Biochar decreased soil N concentrations which may have resulted in part from N uptake, or could have resulted from loss mechanisms such as volatilization or erosion which were not directly investigated in this study. With the application of urea fertilizer, there was the potential for N loss, such as by ammonia volatilization [31,32]. However, since biochar did not affect soil water content or pH, and ammonia volatilization was not quantified, there was no evidence to suggest that ammonia volatilization differed among biochar treatments and thus does not explain differences between soil N concentrations based on biochar treatments. Further investigation is warranted to understand the mechanisms responsible for increased yield in the combined presence of fertilizer N and high C:N biochar.

Despite increases in yield in the combined presence of fertilizer and biochar, wood biochar did not affect plant N concentrations, with only fertilizer influencing grain and ear-leaf N concentrations. In another wood biochar field study, increases in soybean (*Glycine max* L.) yields and doubling of forage biomass with 3.9 $\mathrm{Mg \cdot ha^{-1}}$ hardwood biochar were observed in a high-P clay loam in Quebec [33]. However, hardwood biochar applied at rates of 0, 25, and 50 $\mathrm{Mg \cdot ha^{-1}}$ did not affect growth performance (corn height or biomass) and did not affect grain N concentrations in a sandy clay loam (Eutric Cambisol) in a field trial in Wales [11].

Although changes in N concentrations in the grain and ear leaf were not influenced by biochar treatments, there was an increase in NUE with the 10 $\mathrm{Mg \cdot ha^{-1}}$ biochar application compared to the 0 or 5 $\mathrm{Mg \cdot ha^{-1}}$ biochar treatments. Increased radish NUE with greenwaste biochar occurred in an acidic, Australian Alfisol, attributed in part to increases in pH, extractable soil P and K, and field capacity water content in a pot trial [34]. However, increases in pH and extractable soil P and K do not seem to be reasons for improvement in this study. Soil K was not significantly different across treatments in this experiment. Biochar did not alter soil pH despite the biochar's alkaline pH. In this experiment, the soil was initially similar, except for soil P concentrations, despite attempts at homogenizing the soil and uniform field preparation. However, by the end of the experiment, there were no differences based on treatment effects with Mehlich-3 soil P. The changes in WSP based on fertilizer and biochar addition did not seem to have a clear pattern, perhaps impacted by the differences in initial Mehlich-3 soil P and increases in alkaline phosphatase activities by the end of the experiment. The alkaline phosphatase activities were greatest with 5 Mg·ha^{-1} biochar application without fertilizer, possibly related to the greatest residual P in the initial soil later amended with the 5 Mg·ha^{-1} biochar treatment. With fertilizer addition, the activity seemed to level out across biochar treatments without significant differences among fertilizer treatments. Field capacity water content was not analyzed.

In a silt loam in an Italian field study, durum wheat (*Triticum durum* L.) grain production increased with biochar addition (30 and 60 Mg·ha^{-1}) compared to no biochar addition, while there was no biochar effect on grain N concentration [35]. Since grain N concentrations were unaffected by biochar addition, there was no N-dilution effect corresponding with the increase in yield, suggesting greater fertilizer NUE with biochar treatments than without [35]. The lack of a N-dilution effect also resulted in a field study conducted in a Typic Hapludoll in Iowa with hardwood biochar applied at five rates between 0 and 95.8 Mg·ha^{-1}, in which corn yield increased with biochar addition, but biochar did not affect plant tissue N concentrations [16]. Zheng *et al.* [18] reported lower concentrations of N absorbed by corn seedlings in a three-month pot experiment but also greater biomass production with the amount of N that was absorbed compared to the no biochar treatment. Therefore, it was suggested that, based on the lower N accumulation efficiency but greater NUE with compared to without biochar, biochar could have enhanced N availability to the corn plants and therefore decreased N fertilizer demand [18].

A variety of mechanisms may retain N in soil and decrease N fertilizer demand. Laird *et al.* [28] suggested that biochar adsorbed ammonium onto CEC sites and reduced nitrification and in turn the potential for nitrate leaching. Prendergast-Miller *et al.* [36,37] observed nitrate retained by biochar, presumably in solution in biochar pores rather than on CEC sites, and proposed that nitrate could potentially be released from biochar pores by diffusion gradients [36]. Prendergast-Miller *et al.* [36,37] suggested that biochar localized nitrate in the rhizosphere soil and that roots grew preferentially towards biochar particles. Deciduous wood biochar applied at 20 and 60 Mg·ha^{-1} produced longer wheat roots that increased the root-soil contact compared to non-biochar amended soil and could have increased root N uptake in biochar treatments [36]. A more extensive root system could increase plant ability to access nutrients in the soil [18], thus providing an explanation for the decrease in dissolved total N, nitrate, and Mehlich-3 extractable soil S and Mn concentrations with the 10 Mg·ha^{-1} biochar treatment compared to the no biochar treatment in this experiment. Additional belowground sampling would be necessary in future studies to better understand the effects of pine woodchip biochar on rhizosphere soil and corn roots.

4. Experimental Section

4.1. Biochar Characteristics

Pine woodchip biochar (Waste to Energy Solutions Inc., Destin, FL, USA), which was produced through pyrolysis at 500 °C, was selected for this field study. Biochar was dried for 48 h at 70 °C then ground to pass a 40-mesh screen before pH and EC were determined potentiometrically on a 1:2 (wt:vol) sample:water mixture. Total nitrogen and total carbon concentrations were determined by combustion with Elementar Variomax (Elementar Americas, Inc., Mt. Laurel, NJ, USA). The C:N ratio was calculated from the total N and C concentrations. Total recoverable minerals (*i.e.*, P, K, Ca, Mg, sulfur (S), sodium (Na), Fe, Mn, Zn, Cu, and boron (B)) were determined from acid digestion [38] using an ARCOS inductively coupled plasma (ICP) spectrophotometer (SPECTRO Analytical Instruments Inc., Mahwah, NJ, USA).

4.2. Site Description and Experimental Design

The field experiment was conducted at the University of Arkansas Agricultural Research and Extension Center in Fayetteville, Arkansas in summer 2013. The Global Positioning System data for the four corners of the field were collected using World Geodetic System 1984 in latitude, longitude format (36.09780719° N, 94.16717458° W; 36.09780275° N, 94.16708997° W; 36.09846935° N, 94.16705646° W; 36.09846342° N, 94.16713982° W). Annual precipitation from 30-year normal data [39] was 126.5 cm, average annual maximum air temperature was 20.2 °C, and average annual minimum air temperature was 8.7 °C. The soil within the field was classified as a Razort silt loam, occasionally flooded (fine-loamy, mixed, active, mesic Mollic Hapludalf) [40]. There was a slight positive slope from north to south. The 0.26-ha field was planted the previous two years in cotton (*Gossypium hirsutum* L.). The old cotton stalks had been mowed in fall 2012.

The experimental design was a full factorial randomized complete block. There were 36 plots, each 6 m long and 3.6 m wide and consisting of 4 rows. The total field was 73 m by 11 m. A random number generator was used to place the treatments in each block. Pine woodchip biochar was added at rates of 0, 5, and 10 $\text{Mg}\cdot\text{ha}^{-1}$. For the 5 $\text{Mg}\cdot\text{ha}^{-1}$ biochar application rate, 11.2 kg of biochar were used per plot, and 22.5 kg were used per plot for the 10 $\text{Mg}\cdot\text{ha}^{-1}$ biochar application rate. Biochar was manually applied on 28 May and was incorporated with mechanical tillage into approximately the top 5 cm before rows were bedded and knocked down for planting.

Corn, DEKALB hybrid DKC64-69 with the Genuity VT Triple PRO value-added trait, was planted 74,100 $\text{seeds}\cdot\text{ha}^{-1}$, which equated to 7 to 10 $\text{seeds}\cdot\text{m}^{-1}$, on 29 May with a four-row planter. Full emergence occurred after one week, and no thinning of seedlings was required. The rows were watered by furrow irrigation as needed with the use of the Arkansas online irrigation scheduler [41].

Nitrogen fertilizer was applied at 0, half, and full rates in a split application. The full rate was chosen to achieve a theoretical corn yield of 12.5 $\text{Mg}\cdot\text{ha}^{-1}$ [23]. Therefore, 224 $\text{kg}\cdot\text{N}\cdot\text{ha}^{-1}$ were added as the full rate and 112 $\text{kg}\cdot\text{N}\cdot\text{ha}^{-1}$ were added as the half rate of fertilizer, both in the form of urea (46-0-0). Based on soil analyses conducted prior to experiment initiation (data not shown), no other fertilizer amendments were applied. The first urea application was manually applied 20 June, and the split application was applied 9 July. Herbicide application consisted of 1.3 L·ha^{-1} of broadcasted Cornerstone herbicide on 20 June and a directed spray of 1.7 L·ha^{-1} Atrazine plus 1.7 L·ha^{-1} Cornerstone on 10 July.

4.3. Soil Analyses

Soil was sampled at the 0- to 10-cm depth prior to treatment application to document initial properties and assess initial plot variability. Soil sampling was also conducted at the end of the growing season to assess potential treatment effects. After moist soil was sieved through a 2-mm mesh screen, soil was analyzed for dissolved organic C (DOC), dissolved total N (DTN), ammonium (NH_4^+-N), and nitrate (NO_3^--N) using a single extraction approach [42]. A Skalar segmented-flow autoanalyzer (Skalar Inc., Norcross, GA, USA) colorimetrically determined NH_4^+ following the salicylate hypochlorite procedure and NO_3^- following a modification of Griess-Ilosvay cadmium-copper reduction of NO_3^- to NO_2^- procedure [43]. Using the chloroform-fumigation method, microbial biomass C and N were quantified by calculating the difference between fumigated and unfumigated samples for both C and N [44]. Fumigated and unfumigated soils were extracted and analyzed for DOC and DTN on a Shimadzu TOC-V PC-controlled total organic carbon with attached total nitrogen analyzer (Shimadzu, Columbia, MD, USA). Inorganic N (N_i) was calculated by summing concentrations of NO_3^- and NH_4^+. Dissolved organic nitrogen (DON) concentration was calculated by subtracting N_i from DTN [42].

Water-soluble phosphorus (WSP) was obtained from extractions from 2-g moist soil samples using a 1:10 (wt:vol) soil:water ratio [45] and analyzed by a Skalar Sans-plus segmented-flow autoanalyzer (Skalar Inc., Norcross, GA, USA) using the ascorbic acid method [46]. Acid and alkaline phosphatase activities were measured using the colorimetric estimation of *p*-nitrophenol produced by phosphatase enzyme activity after soil incubation in buffered sodium *p*-nitrophenyl phosphate solution [47].

After drying soil at 70 °C for at least 48 h, soil pH and EC were measured using a 1:2 (wt:vol) soil:water mixture. Organic matter was determined by loss-on-ignition using a muffle furnace. Mehlich-3 extractable soil nutrients (*i.e.*, P, K, Ca, Mg, S, Fe, Mn, Cu) [48] were determined by ICP spectrometry.

One soil core per plot, 4.8 cm in diameter, was collected at the 0- to 10-cm depth on 29 July. After drying at 70 °C for at least 48 h, soil cores were weighed for bulk density determinations. Oven-dried soil was sieved through a 2-mm mesh screen and particle-size analysis was conducted using an adaptation of the 12-h hydrometer method [49].

4.4. Corn Analyses

Ear leaves were harvested at tasseling from the outer two rows of each plot for leaf tissue-N analysis. Ear leaves were dried at 65 °C, ground to pass a 40-mesh screen, and weighed before ear-leaf total N was determined by combustion [50] using a Model Rapid N III (Elementar Americas, Inc., Mt. Laurel, NJ, USA). The harvested yield area was the center 1.5 m of the center two rows in each plot. Grain was harvested on 28 September once physiological maturity had been reached, and yield was calculated based on grain dry weight. Grain samples were ground prior to analysis for total N by combustion. Nitrogen uptake efficiency (NUE) was calculated using the difference method, where NUE in grain was equal to the difference between N removed in grain and the N removed in the unamended control grain divided by the fertilizer-N applied [51]. The N removed in grain was calculated by multiplying the N concentration in the grain by the mass of grain (yield), assuming 720 kg grain per m^3.

4.5. Data Analyses

A two-way analysis of variance (ANOVA) was performed using SAS (version 9.2, SAS Institute, Inc., Cary, NC, USA) to determine any initial differences in the plots for soil pH, EC, OM, DOC, DTN, microbial C, N, and C:N ratio, NO_3^--N, NH_4^+-N, N_i, DON, WSP, acid and alkaline phosphatase activities, and Mehlich-3 extractable soil nutrient concentrations (*i.e.*, P, K, Ca, Mg, S, Fe, Mn, and Cu). A two-way ANOVA was also performed using the data from the soil harvested at the end of the growing season to determine the effects of biochar, fertilizer, and their interaction on soil characteristics as were analyzed for the initial soil. An additional ANOVA was performed to determine the effect of biochar, fertilizer, and their interaction on soil bulk density and particle-size fractions sand, silt, and clay. A two-way ANOVA was performed to determine the effects of biochar, fertilizer, and their interaction on ear-leaf weight and N, corn yield, grain total N, and NUE. Least significant differences were used to separate treatment means at $\alpha = 0.05$.

5. Summary and Conclusions

Pine woodchip biochar applied at rates of 5 and 10 $Mg \cdot ha^{-1}$ in combination with N fertilizer to a fertile silt loam in northwest Arkansas numerically increased corn yields compared to fertilizer application without biochar. However, biochar addition numerically decreased yields when applied without fertilizer, leading to statistically lower yields with the 10 $Mg \cdot ha^{-1}$ biochar treatment combined with no fertilizer compared to the no fertilizer treatment alone. Nitrogen uptake efficiency was greatest with 10 $Mg \cdot ha^{-1}$ biochar application, while soil nitrate concentrations at the end of the growing season were lowest at the same rate of biochar application. Microbial biomass C and N were statistically similar among all treatments. Biochar potentially altered N availability in soil, although the exact mechanisms regarding biochar-soil-plant nutrient relations need further study to elucidate. Pine woodchip biochar can improve corn NUE even in a fertile, temperate, alluvial soil and can increase corn yields in combination with N fertilizer. Given that results will vary based on soil texture, crop management system, biochar properties, as well as through time as a result of dynamic processes, caution must be exercised generalizing results to other systems. Subsequent research is also important to determine long-term changes in soil resulting from pine woodchip biochar addition.

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Author Contributions

Katy Brantley was a graduate student at the time of this research, and she was responsible for managing the study, collecting and analyzing data, and writing the manuscript. She was aided in the implementation of the experimental design, writing, and editing by Mary Savin. David Longer assisted with the experimental design and field work, while Kris Brye contributed to the writing and editing of the manuscript.

Conflicts of Interest

The authors declare no conflict of interest.

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