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Biochar Affects Essential Nutrients of Carrot Taproots and Lettuce Leaves

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Abstract

Essential nutrient concentrations in crops can affect human health. While biochar has the potential as a soil amendment to improve crop yields, it may also affect the concentrations of nutrients such as Ca, Fe, K, Mg, Mn, and Zn in edible portions of crops. To better characterize effects of biochar on important human nutrients in food crops, we evaluated the effects of biochar on lettuce (*Lactuca sativa* L. cv. Black-Seeded Simpson) leaf and carrot [*Daucus carota* subsp. *sativus* (Hoffm.) Schübl. cv. Tendersweet] developing taproot nutrients. Plants were grown in pots in a

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greenhouse using sandy loam (Coxville, fine, kaolinitic, thermic Typic Paleaquults) and loamy sand (Norfolk, fine-loamy, kaolinitic, thermic Typic Kandiudults,) series soils, amended with biochar produced from four feedstocks: pine chips (PC), poultry litter (PL), swine solids (SS), and switchgrass (SG); and two blends of PC plus PL [Pc/PL, 50%/50% (55) and 80%/20% (82) by weight]. Biochar was produced at 350, 500, and 700 °C from each feedstock. Lettuce leaf and carrot taproot total nutrient concentrations were determined by inductively coupled plasma analysis. Biochar (especially at least in part manure-based, i.e., PL, SS, 55, and 82 at nearly all temperatures) primarily decreased nutrient concentrations in lettuce leaves, with Ca, Mg, and Zn affected most. Carrot taproot nutrient concentrations also deceased, but to a lesser extent. Some biochars increased leaf or taproot nutrient concentrations, especially K. This study indicated that biochar can both decrease and increase leaf and taproot nutrient concentrations important for human health. Thus, potential effects on nutrients in plants should be carefully considered when biochar is used as a soil amendment with vegetable crops.

Keywords

Coxville soil; feedstock; human health; Norfolk soil; plants; pyrolysis temperature

Access to nutritionally adequate and safe food is necessary for human welfare and economic development (Roy et al., 2006). For 2017, the UN FAO (2019) estimated that ≈820 million of the world's people were undernourished. An important component of healthy food is the concentration of trace nutrients (White and Brown, 2010). Inadequacy of trace nutrients in food has been a developing human health issue (Roy et al., 2006), with K, Ca, Mg, Fe, Zn, I, Se, and Cu most commonly inadequate in human diets (Broadley and White, 2010; White and Brown, 2010). Soils supply nutrients, which are essential to plants (Brady, 1974). Thus, soil fertility, including amendments such as fertilizer, which affect the availability of these nutrients, is key to plant nutrition (McGrath et al., 2014), and soil health is i important for human health (Doran et al., 1996).

Biochar is the product from combustion of a wide variety of biomass materials with limited or no oxygen (pyrolysis) (Lehmann and Joseph, 2015). Amending agricultural soils with biochar may improve crop productivity through a variety of mechanisms by increasing soil pH, increasing soil water holding capacity, enhancing biotic interactions, and supplying essential plant macro- and micronutrients (Jeffery et al., 2015). In terms of nutrients essential for human health, : biochar can alter soil fertility by being a source of these nutrients, altering soil properties which affect the absorption and release of these nutrients, and affecting microbes associated with the cycling of these nutrients (Ding et al., 2016). Therefore, the availability of soil nutrients, which are necessary to improve plant mineral nutrition (White et al., 2014), could be increased by biochar.

Biochar can be produced from many different organic "waste materials" from agriculture and forestry, including materials such as poultry litter (Chan et al., 2008) and swine solids (Cantrell et al., 2012), which can cause environmental N pollution problems. As a result, production of biochar can help alleviate animal manure and yard waste disposal problems (Lehmann and Joseph, 2015). Furthermore, technologies are being developed whereby

biochar can be produced at the source of the feedstock materials, potentially providing benefits (while also considering costs) to rural economies (Joseph, 2009).

The source of the feedstock and pyrolysis temperature can greatly affect its nutrient composition. Cellulosic-based feedstocks such as straw or wood produce biochars that are usually low in ash (Novak et al., 2014) and, hence, low in major nutrients such as Ca, Mg, K, and trace nutrients such as Fe, Zn, and Mn (Ippolito et al., 2015; Novak et al., 2013). In contrast, manure-based feedstocks produce biochars that are high in ash and major and trace nutrients (Ippolito et al., 2015; Novak et al., 2013, 2014). When a feedstock comes from a source where nutrients are purposely added at some stage during its production, these can be reflected by high concentrations in the resulting biochar. For example, when swine are fed Zn as a supplement (Sistani and Novak, 2006), the biochar produced from swine waste can have high Zn concentrations (Novak et al., 2015). Increasing the pyrolysis temperature will drive off more volatile compounds and nutrients such as O, H, and N, thus increasing the relative concentration of ash and associated nutrients in the biochar (Ippolito et al., 2015; Novak et al., 2014).

Because of biochar's potential to add nutrients to the soil either directly, or indirectly by affecting soil properties influencing nutrient bioavailability (Chan and Xu, 2009), biochar can affect the concentration of nutrients in plants. A meta-analysis indicated that biochar generally increased plant leaf K concentrations across a variety of crops (Biederman and Harpole, 2013). For example, in maize leaf, K increased with peanut hull biochar (Gaskin et al., 2010) and eucalyptus biochar (Butnan et al., 2015), while willow wood biochar had no effect on maize leaf K (Agegnehu et al., 2016). Butnan et al. (2015) reported decreases in maize leaf Ca, Mg, and Mn with some eucalyptus biochar treatments. Masud et al. (2014) reported increased plant uptake of K and Ca, with biochar application on soybeans. However, while current literature indicates a variety of plant nutrient responses to biochar, additional studies are needed to better understand the effect of biochar applications elements that are important for the nutritional value of food crops (Martin et al., 2011; White and Brown, 2010).

Despite the commercial support for use of biochar as a soil amendment and availability of different products (e.g., KIS Organics, 2019; Pleasant, 2009; Wakefield Biochar, 2019; White, 2018), there has been relatively little research on both the potential positive or negative effects on horticultural crops. For example, biochar has been considered as an amendment to growing media such as peat replacement (Blok et al., 2017) or as a substrate in a soilless hydroponic system for tomato production (Dunlop et al., 2015).

The general focus for biochar and metals has been in terms of either excess heavy metals due to biochar feedstocks and production techniques (Subedi et al., 2017), and the potential use of biochar to reduce potentially high concentrations of toxic heavy metals such as Cd or Pb in plants, which could adversely affect human health (Peng et al., 2018). For example, application of PL or SS biochar, along with compost, reduced Cd and Zn concentrations in a metal-contaminated mine soil and switchgrass roots and shoots, which were associated with increased root and shoot growth (Novak et al., 2019). Addition of cassava stem biochar to a Cd- and Zn-contaminated soil reduced the bioavailability of both nutrients and concentration

of Zn but increased the concentration of Cd in *Vigna radiata* L. roots and shoots (Prapagdee et al., 2014). Based on a meta-analysis, Chen et al. (2018) estimated an average 17% decrease in plant Zn with biochar application across a wide variety of conditions. Because some of these trace nutrients such as Zn and Mn are also essential for humans, any reduction of normally low concentrations in soils due to biochar could also adversely affect human health.

There have been a few reports in the biochar literature specifically relating to its potential beneficial effects on trace nutrients and human health, by increasing the supply of these nutrients to crops. For example, acidified (with S) maize cob biochar increased the concentrations of Fe, K, Mn, and Zn, and to a lesser extent, Mg and Ca (in one case biochar decreased Ca), in quinoa seed for plants growing under different soil stress conditions (Ramzani et al., 2017). While eucalyptus twig biochar by itself had no effect on rice grain Fe concentration, the biochar did enhance the increase in concentrations of these nutrients when Fe fertilizer was also used to biofortify the crop with Fe (Ramzani et al., 2016a). In a study with biosolids and biosolids plus biochar (made from *Pinus radiata* chips) in combination, Gartler et al. (2013) found increased extractable soil Zn concentrations and the concentrations of Zn in the edible portions of several crops, including lettuce leaves and carrot taproots. The biosolids were the source of the Zn; and while biochar by itself had no effect on Zn concentrations in nearly all crops, biochar by itself increased the Zn concentration in radish bulbs (Gartler et al., 2013). When biochar was in combination with biosolids it further increased the concentration of Zn in lettuce leaves but decreased the concentration relative to biosolids alone in radish leaves and bulbs (Gartler et al., 2013). Biochar produced from macroalgae grown in industrial wastewater increased the concentration of Ca, Mg, K, and Mo in radish roots (Roberts et al., 2015), suggesting that high nutrient concentration biochars could be used to supply plants with nutrients essential for human health.

However, Hartley et al. (2016) reported that woody material from green waste composting facilities, and rhododendron and softwood all reduced Cu, Zn, Fe, and Mn uptake into wheat grains. In other studies, Sorrenti et al. (2016) found that kiwifruit had Fe chlorosis symptoms when grown in biochar amended soil, while Moreno-Jimenez et al. (2016) reported no effect of oak biochar on barley grain Cu and Zn concentrations. Thus, the relationship between biochar and trace nutrient concentrations in crops is complex, and there have been few suggestions that changes in crop nutrient concentrations solely due to biochar additions could adversely affect mineral nutrients in food for human consumption.

Thus, we conducted a study which indicated if amending soils with biochar could have beneficial or detrimental effects on the concentrations of key inorganic nutrients for human nutrition in the edible portions of crops. The focus was on the effects of different biochars on concentrations of major (Ca, K, Mg) and minor (Fe, Mn, Zn) plant nutrients contained in lettuce leaves and carrot taproots. Lettuce is a food source for Ca, Fe, and K (Noumedem et al., 2017), while carrot taproots are an important source of K, Mg, and Mn (Bjarnadottir, 2019; da Silva Dias, 2014). In addition, because biochar characteristics should be tailored to particular soil problems (Novak and Busscher, 2012), we evaluated biochar effects using two South Carolina coastal plain soils, the Coxville and Norfolk series.

Materials and Methods

Soils and biochar

Two agricultural soils used in this experiment were collected from a field at the Clemson University Pee Dee Research and Education Center farm in the Coastal Plain region of South Carolina near Florence, SC. The soils were the sandy loam (fine, kaolinitic, thermic Typic Paleaquults) Coxville Series and loamy sand (fine-loamy, kaolinitic, thermic Typic Kandiudults) Norfolk Series. Soil characteristics are summarized in Table 1 and are based on J. Novak, personal communication (2018), Olszyk et al. (2018), and Sigua et al. (2014) for the nutrients B, Ca, Cu, K, Mg, Mn, Na, P, and Zn. Analysis for these nutrients was by Clemson University (2019) and used the Mehlich 1 extraction procedure followed by ICP analysis. Overall, the Coxville soil had a slightly lower pH and higher C and N concentrations than the Norfolk soil (N not detectable for the Norfolk soil at a limit of 1 $g \cdot kg^{-1}$). The *P* value was over twice as high for the Coxville than for the Norfolk soil, with values for the other elements varying between the two soils.

Four biochar feedstocks were used: switchgrass straw (*Panicum virgatum*) (SG), loblolly pine chips (*Pinus taeda*) (PC), swine solids (SS), and poultry litter (PL). There were also two blends of PC and PL: 50% PC and 50% PL (55), and 80% PC and 20% PL (82) by weight. Biochars were produced as indicated in Novak et al. (2013), Novak et al. (2014), and Olszyk et al. (2018). For mixtures, PC and PL feedstocks were combined before being made into pellets. Feedstocks and blends were formed into cylindrical pellets using a PP220 pellet mill (Pellet Pros., Inc., Davenport, IA) equipped with a 6-mm die as described by Novak et al. (2014). Resulting pellets were sieved using a 4-mm sieve to eliminate fine material. Pellets retained on the 4-mm sieve were used in the study. Pellets for each feedstock were then was pyrolyzed at a low (350 °C), medium (500 °C) and high (700 °C) temperature using a furnace-retort system (Lindberg/MPH, Riverside, MI) for 1 to 2 h (Novak et al., 2013), depending on sample size.

Soil and biochar pellets from the appropriate feedstock or blend were combined for each biochar treatment as described by Novak et al. (2014). For a 1% mixture of soil and biochar, a target of 450 g of soil was weighed into a bag and then a target of 4.5 g of biochar was added. Each bag was then thoroughly mixed by hand, spread, and placed in the pots.

Details concerning properties of the biochar used in this study are found in Olszyk et al. (2018), Novak et al. (2013), Novak et al. (2014), and Sigua et al. (2014); and details are summarized in Tables 2 and 3. The biochar pH, electrical conductivity (EC), and extractable phosphorus (EP) values were measured for four samples per biochar as discussed previously in Olszyk et al. (2018). In brief, pH and EC were measured using a 2:1 water to biochar or soil ratio (v/v) with MilliQ water. The EP was measured spectro-photometrically (Olsen and Sommers, 1982). As indicated earlier (Olszyk et al., 2018), biochar pH was highest for PL, 82 and 55, and lower for SS, PC, and SG (Table 2). The EC was highest with PL, followed by 55; with 82 and SS moderate, and PC and SG lowest. The EP was highest with SS, 55 and 82; with PL moderate, and PC and SG low. For a particular feedstock or blend, the pH and EC tended to increase with increasing pyrolysis temperature, while EP deceased with increasing temperature.

Biochar total nutrient (i.e., elemental Al, Ca, Cu, Fe, K, Mg, Mn, P, S, and Zn) concentrations were measured for one sample by a commercial laboratory (Bureau Veritas Minerals, Vancouver, BC, Canada) for one sample for each biochar and temperature combination. In brief, samples were digested using Aqua Regia digestion followed by quantifying nutrients using inductively coupled plasma mass spectrometry (ICP-MS) analysis. Each sample was cold leached with HNO, followed by a hot water digestion and cooling. A modified Aqua Regia solution (HCl, HNO_3, and DI H_2O) then was added to each sample, followed by heating in a hot water bath. Samples were brought to volume with dilute HCl, then filtered, and subsequently analyzed using a Perkin Elmer Inc. (Waltham, MA) NexION 300 ICP-MS. The S and P concentrations were presented elsewhere (Olszyk et al., 2018), while the concentrations for the other elements are discussed for the first time in this article.

Plants

This study focused on two crops used for direct human consumption, carrot and lettuce. Seeds for each crop plant were planted into soil and soil-plus-biochar mixtures in 10-cm diameter green plastic pots with geotextile cloth lining the bottom of the pot. There were about eight lettuce or five carrot seeds per pot.

Lettuce (*Lactuca sativa* L. cv. Black-Seeded Simpson) was planted 8 Nov. 2013 and harvested 15 Jan. 2014, and carrot [*Daucus carota* subsp. *sativus* (Hoffm.) Schübl. cv. Tendersweet] was planted 13 Nov. 2013 and was harvested 22 Jan. 2014. After seeds were planted, deionized H_2O was added to the soil with and without biochar at an amount necessary to obtain a soil moisture content of 10% (w/w). Soon after initial water addition, lettuce received 0.068 g KH_2PO_4 and 0.076 g NH_4NO_3 per pot, equivalent to 67 $kg{\cdot}ha^{-1}$ P, 85 $kg{\cdot}ha^{-1}$ K and 112 $kg{\cdot}ha^{-1}$ N, respectively. Slightly more fertilizer was added to the carrot pots; i.e., ≈0.073–0.076 g KH_2PO_4 and 0.082–0.084 g NH_4NO_3 per pot, equivalent to ≈72–74 $kg{\cdot}ha^{-1}$ P, 92–94 $kg{\cdot}ha^{-1}$ K, and 121–124 $kg{\cdot}ha^{-1}$ N. The fertilizer was dissolved in reverse-osmosis (R/O) water with a pipettor delivering the desired amount of N, P, and K to each pot. Pots subsequently were watered with R/O water whenever the top of the soil was dry to the touch. Plants were thinned to one plant per pot after germination. Plants were grown in a greenhouse under 1000-W high intensity discharge lights with a 16-h light/8-h dark photoperiod. Average daily greenhouse environmental conditions from emergence to harvest were 15.3 °C (both crops), 195 (carrot) or 188 (lettuce) $\mu mol\text{-}m^{-2}{\cdot}s^{-1}$ photosynthetically active radiation from 400 to 700 nm (PAR), and 36% (carrot) or 39% (lettuce) relative humidity (RH).

At harvest, lettuce and carrot leaves were removed from the plants and dried at about ambient air temperature (no extra heat) for 29 and 22 d, respectively, before drying at 60 °C. For carrot root systems, pots were randomly assigned to two groups. For group A carrot pots, root systems were immediately washed with R/O water to remove soil, followed by separation of young, developing taproot and diffuse roots; drying at 60 °C; and weighing. Group B carrot pots were used for a leachate study of soil water chemistry. Thus, the B pots were placed in plastic bags and put into a cool room at ≈4 °C for ≈24 to 42 d until leached with Milli-Q water, and the leachate collected. After leaching was complete, all B pots were

processed using the same procedures as for group A pots. Data for group A and B root systems were combined for analysis.

Lettuce leaf and carrot taproot samples were ground using a Wiley® mill. Samples were digested using automated block digestors using EPA method 3050B (U.S. EPA, 1996). The digestate was analyzed for total nutrient concentrations with a Varian Vista-PRO ICP-OES (Mulgrave, Victoria, Australia) or a PerkinElmer Optima 8300 ICP-OES (Waltham, MA). The lettuce leaf and carrot taproot nutrient concentrations were expressed on a μg00B7g^{-1} basis, except for mg·g^{-1} for K.

Experimental design and statistical analysis

For each crop there were 18 biochar treatments (six feedstocks × three pyrolysis temperatures), plus a no-biochar control; with the treatments and controls repeated for each soil type. There were six replicates per treatment and soil type. The study used a completely randomized design with pots randomly located across a greenhouse bench and rotatedto change position at least once during the experiment.

Nutrient data were $\log_{10}$ transformed before statistical analysis, as treatment effects were assumed to be additive on a log scale. A small value (0.1 μg·g^{-1}, or 0.5 μg·g^{-1} for carrot taproot Fe) was added to “0” values before log transformation. Because of heterogeneity of variance, a weighted analysis of variance (ANOVA) was used with weights proportional to the inverse of the variances for the transformed data for each soil and biochar treatment (Welch, 1951).

The PROC MIXED ANOVA procedures in SAS/STAT® software (SAS Institute Inc., Cary, NC) were used to analyze data separately for each crop. The ANOVAs were carried out to compare treatments. The factors were soil and treatment (18 biochar treatments plus control plants) and soil × treatment interactions. When there was a significant soil × treatment interaction, a Dunnett’s test at $P < 0.05$ was used to compare individual treatments to the control plants separately for each soil. The means and standard errors in the figures were based on back-transformations of the least squares means and standard errors from the statistical analysis to provide results with the same nontransformed units as the original data. The back-transformed standard errors used in figures are the average based on upper and lower least square standard errors.

Results

Biochar nutrients

The manure-based biochars (PL and SS) generally had higher nutrient concentrations (Olszyk et al., 2018, Table 3). The SS biochars had the highest Cu, Fe, Mg, Mn, P, and Zn. The PL biochars had the highest K, as well as high concentrations of Mg, Mn, P, and Zn. Both SS and PL had high Ca and S. The other elements were intermediate for SS and PL, and the 55 and 82 mixtures of PL and PC had intermediate concentrations of all nutrients. The cellulosic biochars PC and SG had low concentrations of all nutrients. For the PL and SS biochars, nutrient concentrations usually increased with increasing pyrolysis

temperatures. In general, nutrient concentrations varied less consistently with temperature for the mixtures and cellulosic-based biochars.

Lettuce leaf nutrients

Compared with the no-biochar control plants, lettuce leaf Ca and Mg concentrations were reduced substantially by nearly all biochars (Fig. 1). For the Coxville soil, the only biochar treatment that did not significantly reduce the nutrient concentration was for Ca with SG at 500° (Fig. 1A), while all biochar treatments caused significant reductions in lettuce leaf Mg (Fig. 1C). For the Norfolk soil, the cellulosic biochars had less effect than the other biochars on Ca and Mg concentrations—with no response to PC for either nutrient—and with SG only a reduction in leaf Ca and Mg at 500° (Fig. 1B and D). The 55 biochar at 700° also had no effect on leaf Mg for the Norfolk soil (Fig. 1D). There were differences in biochar effects on lettuce leaf Ca and Mg among pyrolysis temperatures for different biochars, but no consistent pattern in responses for either soil.

In contrast to the general decreases in leaf Ca and Mg with many biochar treatments, there were increases as well as decreases in lettuce K with biochar. Leaf K decreased for SS with both soils, PC for the Coxville soil, SG at 350 and 700 °C for the Coxville soil, and 82 at 350 °C for the Norfolk soil (Fig. 2A and B). Leaf K concentrations increased for PL at 350 °C for the Coxville soil (Fig. 2A), and for PL at all temperatures as well as 55 and SG at 700 °C for the Norfolk soil (Fig. 2B).

In terms of micronutrients, biochar decreased lettuce leaf Mn concentrations with PC at all temperatures and SG at 700 °C, but increased Mn with PL at 350 and 500 °C for the Coxville soil (Fig. 2C). Biochar decreased leaf Mn for the Norfolk soil with the 55, 82 and PL biochars at all temperatures, and PC at 700 °C (Fig. 2D). Lettuce leaf Fe concentrations decreased for the Coxville soil only with 55 and SS at 500 °C and PL at 700 °C, even though there was a trend toward lower Fe with most other biochar treatments (Fig. 3A). For the Norfolk soil, nearly all biochar feedstocks decreased Fe concentrations, with the exceptions of PC at all temperatures and SG at 350 and 700 °C (Fig. 3B). Leaf Zn concentrations decreased with PL, SS, 55 and 82 for both soils, PC for the Coxville soil, and SG at 700 °C for the Coxville soil; and SG at 500 °C for the Norfolk soil (Fig. 3C and D). The reductions in lettuce leaf Zn followed a pattern like that for Ca and Mg for the Coxville soil, and Ca, Mg, and Fe for the Norfolk soil.

Carrot taproot nutrients

Biochar effects on developing carrot taproot nutrient concentrations followed a different pattern of response compared with lettuce leaves. Biochar had no effect on taproot Ca concentrations for the Coxville soil (Fig. 4A), but decreased taproot Ca with PL at all temperatures, 55 at 500 and 700 °C, and SS at 500 °C for the Norfolk soil (Fig. 4B). Taproot Mg for the Coxville soil, decreased with 55, 82, PC, SG, and SS with biochars produced at 700°; and 55 at 500 °C (Fig. 4C). For the Norfolk soil, only 55 at 700 °C and PL at 500 °C decreased taproot Mg, while SS at 350 °C increased taproot Mg (Fig. 4D).

Taproot K concentrations increased with the all biochars with the Coxville soil, for at least one temperature (Fig. 5A). The greatest increases in taproot K were with PL and 55. In

contrast, biochar had little effect on taproot K with the Norfolk soil, except for increasing K with Pl at 350 and 700 °C and decreasing K with SS at all temperatures (Fig. 5B).

Biochar increased taproot Mn with PL at 350 and 500 °C, but decreased Mn with 82 at 700 °C for the Coxville soil (Fig. 5C). Biochar generally reduced taproot Mn for the Norfolk soil, but there was a large amount of variability among replicate plants, and the decreases were significant only with 55 at 700 °C and PC at 500 °C (Fig. 5D).

Biochar decreased taproot Fe with a few biochars scattered across all feedstocks, except PC with the Coxville soil, with the largest decrease for 82 at 700 °C (Fig. 6A); while biochar had no effect on taproot Fe for the Norfolk soil (Fig. 6B). Taproot Zn for the Coxville soil decreased with 55, 82, SG, and SS at 700 °C and 55 at 500 °C; while Zn increased with PL and SS at 350 °C (Fig. 6C). The only biochar effect on Zn for the Norfolk soil was a decrease with 55 at 700 °C (Fig. 6D).

Discussion

Despite the many potential benefits for crop growth due to amending soil with biochar, there may be unexpected consequences if the effects of the biochar on plant nutrients are not carefully considered. Successful plant growth depends on an adequate supply of plant nutrients, with growth rates increasing with increasing nutrient concentrations (McDonald, 1994). There is considerable literature on biochar and nutrients relating to plants (Chan and Xu, 2009; De Luca et al., 2015). Fewer studies relate biochar and plant nutrients to human health. Some of these emphasized an excess of heavy metals in plants with some biochars (Subedi et al., 2017), or use of biochar to reduce heavy metals in plants (Peng et al., 2018). A few studies considered use of biochar and nutrients in plants relative to human health. For example, nutrient increases were due to use of a biochar feedstock with a high nutrient concentration (e.g., macroalgae from wastewater, in Roberts et al., 2015), or an enhanced increase in essential nutrients when biochar was applied with a concentrated source of nutrients such as Fe fertilizer (Ramzani et al., 2016a, 2016b) or biosolids (Gartler et al., 2013). Ramzani et al. (2017) applied only biochar and found increases in essential nutrients, but the biochar had been acidified with sulfur. When biochar by itself was applied to soil, essential nutrients were reduced in wheat grains (Hartley et al., 2016).

In our study, some biochar treatments enhanced K concentrations, i.e., in lettuce with PL for both soils with at least one temperature (Fig. 2A and B); possibly this result was due to an extra fertilization effect of the biochar (in addition to the added fertilizer), as soil K concentrations were low in this study. This increase in K with biochar was like the results in other studies (Biederman and Harpole, 2013). In a similar study with lettuce and PL biochar, shoot K also increased (Gunes et al., 2014). Using charcoal instead of biochar, Deenik et al. (2010) observed a similar increase in leaf K concentration for lettuce for plants receiving high levels of charcoal (10% and 20% by weight) when no fertilizer was used. Deenik et al. (2010) observed an increase in lettuce leaf K uptake with 10% charcoal, but a decrease in leaf K uptake with 20% charcoal. Deenik et al. (2010) also noted increases in lettuce leaf Ca, Mg, Fe, Mn, and Zn; but decreases in leaf K Ca, Mg and Zn uptake, primarily with the highest charcoal rate. Sorrenti et al. (2016) reported an increase in leaf K when tree fruit

wood biochar was applied to kiwifruit vines. Potassium also increased in carrot taproots for many biochars with the Coxville soil, and PL at 350 and 700 °C with the Norfolk soil (Fig. 5A and B). If these increases persisted in the harvest- able crops, their nutritive value in terms of lettuce leaf or carrot taproot K would further increase beyond the healthy levels already present in the tissues (Rudrappa, 2019).

In contrast, the decreases in leaf nutrient concentrations we observed with many of the biochar treatments across all elements and both crops could reduce crop quality for direct human consumption. For example, the reported data suggest that reductions with some biochars, for example, Ca, Fe, and Mg in lettuce (Figs. 1A–D and 3A and B), or Ca and Mn in carrot taproots (Figs. 4B and 5C and D) could adversely affect the nutritional values of these crops, as they have healthy amounts of these nutrients (Rudrappa, 2019). In a previous study with lettuce and PL biochar alone, Gunes et al. (2014) also reported decreases in shoot Mn and possibly Fe concentrations, but no changes in shoot Ca or Mg concentrations.

Soil pH would be a key factor in availability of the nutrients, especially for the metals evaluated in this study (Barber, 1984). In a previous seed germination study, we showed that the PL, 55, SS, and to a lesser extent 82, SG, and PC biochars increased pH vs. the controls across plant species depending to some extent on soil type, with the largest increase to about pH 6.7 for PL pyrolyzed at 700 °C vs. 5.6 for the controls across the Coxville and Norfolk soils (Olszyk et al., 2018). A similar increase in soil pH was seen with PL biochar in research with a low pH mine soil (Novak et al., 2019). These increases in soil pH would lower the solubility of Fe, Mn, and Zn, thus decreasing their concentrations in solution (Barber, 1984). This explanation could, at least in part, explain the decreases in those nutrients in our plants; especially with the manure-based biochars for lettuce leaves, and to a lesser extent for carrot taproots.

In general, increases in soil pH, especially with the PL biochar (Olszyk et al., 2018), should have made Ca and Mg (to pH 6.5) more available (Barber, 1984; USDA, 2014) and increased lettuce leaf concentrations, instead of the widespread decreases that we observed. Thus, decreases in Ca and Mg were likely associated with other factors that affect the availability of nutrients from the soil (Barber, 1984). For example, an increase in pH leads to lower concentrations of plant-available P due to binding with Ca in the biochar (Novak et al., 2019), which could lead to lower leaf Ca. Above pH 6.5, as occurred for the Norfolk soil especially with the PL and 55 biochars in a seed germination study (Olszyk et al., 2018), exchangeable Mg could decrease under some conditions (Barber, 1984), and could contribute to decreases in leaf Mg concentrations.

It was possible that the general lower leaf Ca and Mg concentrations we found in lettuce could be due to excess Zn in the biochars. O'Toole et al. (2013) suggested that the Zn in galvanized metal containers used during their studies likely increased biochar Zn, which reduced plant Ca and Mg. However, we used ceramic bowls to make biochar, and, thus we did not have a metal container factor. Instead, we found high Zn concentrations (≈5000 to 6800 ppm) in our SS biochar (Table 3) likely due to the addition of Zn to swine feed as a supplement (Sistani and Novak, 2006). The Zn concentration also was elevated in the PL biochar in our study (Table 3); however, neither the SS nor PL biochar increased Zn in

lettuce leaves (Fig. 3C and D). For carrot taproots, the Zn concentration increased only with PL and SS pyrolyzed at 350 °C with the Coxville soil (Fig. 6C), while SS decreased the taproot Zn concentration at the highest pyrolysis temperature with the Coxville soil.

Instead of increases, we primarily found reductions in Zn concentrations, especially for lettuce leaves and most often with manure-based biochars (Fig. 3C and D). In a previous study with lettuce and PL biochar, Gunes et al. (2014) found a reduction in shoot Zn. Kim et al. (2015) also reported a decrease in lettuce leaf Zn concentration with biochar application, but only tested a cellulosic rice hull biochar. Decreases in Zn concentrations in *Lolium perenne* L. shoots were found with increasing levels of 80% coniferous and 20% hardwood chips biochar and two soils (Rees et al., 2015). Rees et al. (2015) also reported that the Zn concentration decreased in shoots of the Cd- and Zn-hyperaccumulator plant *Noccaea caerulescens* (J. Presl & C. Presl) F.K. Mey. when grown on the lowerpH (5.89) soil, while Zn did not change with biochar on the higher pH (8.07) soil. Both Kim et al. (2015) and Rees et al. (2015) used high heavy metal concentration soils from near smelters, while we used uncontaminated soils.

In both Kim et al. (2015) and Rees et al. (2015), the decrease in leaf Zn was likely related to an increase in soil pH with addition of biochar, especially for a more acidic soil. This suggests that an increase in pH with addition of biochar can be related to reduced plant Zn concentration. As indicated earlier, in a seed germination study we found a general increased soil pH with the same biochars, biochar level, and soils as in this study, with the greatest increase with PL pyrolyzed at 700 °C (Olszyk et al., 2018).

Feedstock type is also a likely factor in the crop response to biochar. In our study, the cellulosic PC biochar had no statistically significant effects comparted to control plants for carrot tap root Zn for both soils (Fig. 6C and D) or lettuce shoot Zn with the Norfolk soil (Fig. 3D). This was similarto the results of Gartler et al. (2013), who found no effect on lettuce Zn with PC biochar; and Chen et al. (2018), who indicated little overall effect of wood (cellulosic) biochar on plant Zn concentrations in their metaanalysis. However, there was a reduction in lettuce Zn concentration with PC for the Coxville soil in our study. We saw a decrease in lettuce leaf Zn primarily with the manure-based biochars, whereas Chen et al. (2018) reported little change in plant Zn with manure biochars.

We saw a reduction in Fe concentrations in lettuce leaves (both soils, Fig. 3A and B) and carrot taproots (Coxville soil, Fig. 6A), primarily with manure-based biochars. Similarly, with fruit tree wood trimming biochar, Sorrenti et al. (2016, p. 16) found that biochar alone reduced kiwifruit plant Fe uptake. The authors hypothesized that "…in potted conditions Fe in soil solution was attracted and retained by biochar...thereby limiting its availability…."

There was no clear difference between the two soil types in terms of response of nutrients to biochar in our study. For lettuce leaf and other carrot taproot nutrients, either Coxville or Norfolk had a larger decrease or increase concentration, or there was essentially no difference in response between soils across biochars. While, in general, more coarse or medium texture soils have been shown a greater increase in crop productivity with biochar addition than fine texture soils (Jeffery et al., 2011), there is no clear-cut picture of the

relationship between soil characteristics, biochar, and concentrations of nutrients in plant tissues. Based on their meta-analysis, Chen et al. (2018) reported a larger decrease in plant Zn concentrations with biochar applications for a fine soil than with coarse or medium texture soils. However, we saw little difference in lettuce leaf and a highly variable difference in carrot taproot Zn between the finer texture Coxville and coarser texture Norfolk soil. In the reported study, the largest differences between soils were for carrot, where there was a large increase in taproot K for many biochars with the finer Coxville soil, but little effect on taproot K for most biochars with the sandier Norfolk soil—except for the small increase in K with PL and decreases with SS (Fig. 5A and B). Similarly, Gaskin et al. (2010) found no effect of pine chip biochar on maize leaf K growing on a sandy loam Ultisol. However, Gaskin et al. (2010) also reported that a peanut hull biochar increased maize leaf K on the sandy soil, but primarily in one of two years. We found a reduction in lettuce leaf Ca and Mg with the sandier Norfolk soil in addition to the finer Coxville soil, and Syuhada et al. (2016) found a reduction in maize leaf Ca and Mg concentrations when biochar was added to a fertilized sandy Pozol.

Finally, changes in nutrients could also occur due to the overall impact of biochar on the crop growth; i.e., if there is an increase in crop growth leaf, concentrations could decrease due to growth dilution, as suggested by Syuhada et al. (2016). However, further discussion of mechanisms by which biochar results in changes in soil quality and, hence, concentrations of leaf or taproot nutrients, are beyond the scope of this article and will be discussed in future work.

Thus, there is a critical need to produce food with the quality and quantity ofnutrients that promote human health (Martin et al., 2011), which may be impacted by biochar amendments to soils. While biochar can have positive effects on the amount of a crop available by increasing yields (Jeffery et al., 2011), especially for tropical soils (Jeffery et al., 2017), the potential role of biochar in affecting the nutrient quality should be further evaluated to optimize its desirable traits and minimize any undesirable characteristics such as absorbing plant nutrients (Kavitha et al., 2018). In addition to evaluating factors such as feedstocks, pyrolysis temperature, and particle size when “designing” a biochar for a specific application (Novak and Busscher, 2012; Novak et al., 2014), researchers should also consider potential impacts on plant nutrients important for human health. The goal should be to produce a biochar that contains inorganic nutrients that can serve as a fertilizer to biofortify crops with essential nutrients (White and Brown, 2010), while avoiding any reductions in these nutrients that would reduce their food value.

Conclusions

Biochar amendments to soil could increase or decrease concentrations ofessential elements in edible parts of plants, potentially affecting human health. In this study, some biochars increased K concentrations in both lettuce leaves and carrot taproots, especially the high-K concentration PL biochar. In contrast, a number of biochars primarily decreased Ca, Fe, Mg, Mn, and Zn concentrations in lettuce leaves; and to a lesser extent, carrot taproots. These decreases occurred despite higher concentrations of these nutrients in the biochar, such as poultry litter, compared with the soil; and these may be related to effects of biochar on soil

properties such as pH. Thus, while biochar is a potent component in the array of tools scientists possess to enhance crop productivity, unintended consequences (especially in terms of reducing nutrient concentrations in crops) should be carefully considered when designing biochar field amendments.

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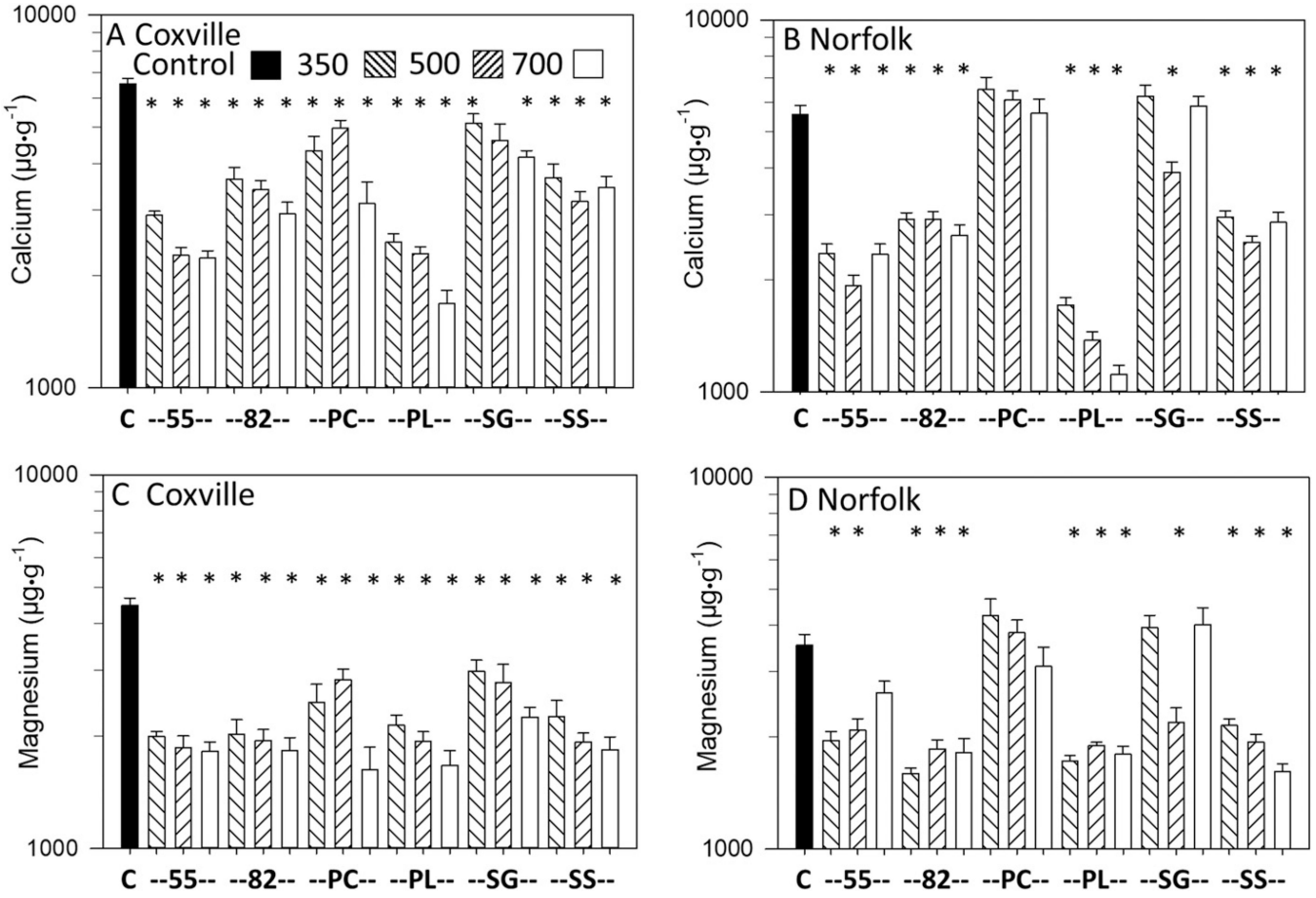

Fig. 1.

Effects of biochar on lettuce leaf Ca and Mg in $\mu g \cdot g^{-1}$. Data are for Ca for the Coxville (**A**) or Norfolk (**B**) soil, and Mg for the Coxville (**C**) or Norfolk (**D**) soil. C = no biochar control, PL = poultry litter, PC = pine chips, 55 = 50% PC and 50% PL, 82 = 80% PC and 20% PL, SS = swine solids, SG = switchgrass. Temperatures in °C are indicated above the graph. Each bar is based on back-transformed least square mean and upper standard error (see methods) for six pots except for 5 for Coxville PL 500, Norfolk PC 350, Norfolk 82 350 and 500, Norfolk PL 500 and 700. An "*" above a bar indicates a significant difference vs. control plants according to Dunnett's test.

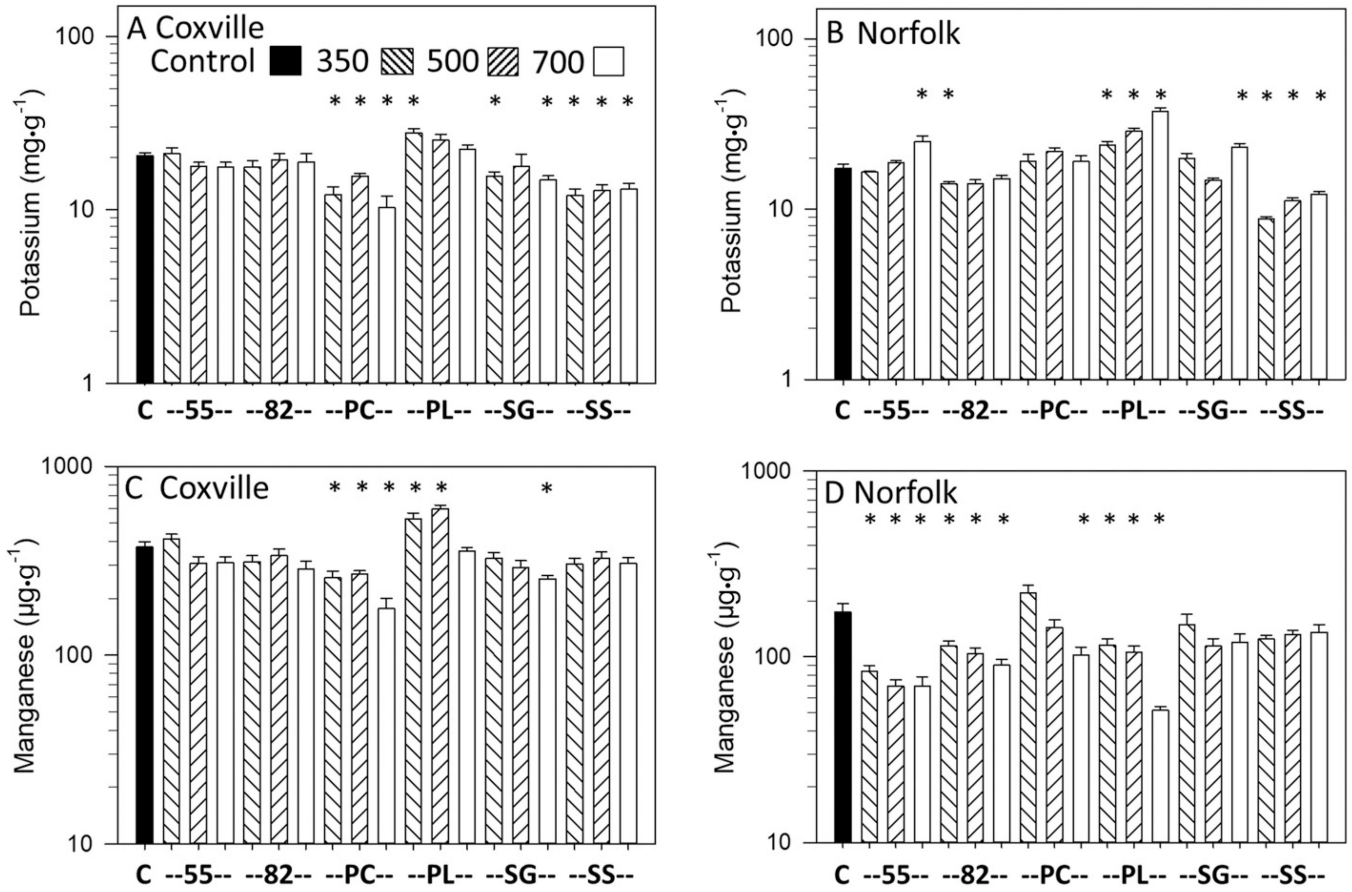

Fig. 2.

Effects of biochar on lettuce leaf K in $mg \cdot g^{-1}$ and Mn in $\mu g \cdot g^{-1}$. Data are for K for the Coxville (**A**) or Norfolk (**B**) soil, and Mn for the Coxville (**C**) or Norfolk (**D**) soil. C = no biochar control, PL = poultry litter, PC = pine chips, 55 = 50% PC and 50% PL, 82 = 80% PC and 20% PL, SS = swine solids, SG = switchgrass. Temperatures in °C are indicated above the graph. Each bar is based on back-transformed least square mean and upper standard error (see methods) for six pots except for 5 for Coxville PL 500, Norfolk PC 350, Norfolk 82 350 and 500, Norfolk PL 500 and 700. An "*" above a bar indicates a significant difference vs. control plants according to Dunnett's test.

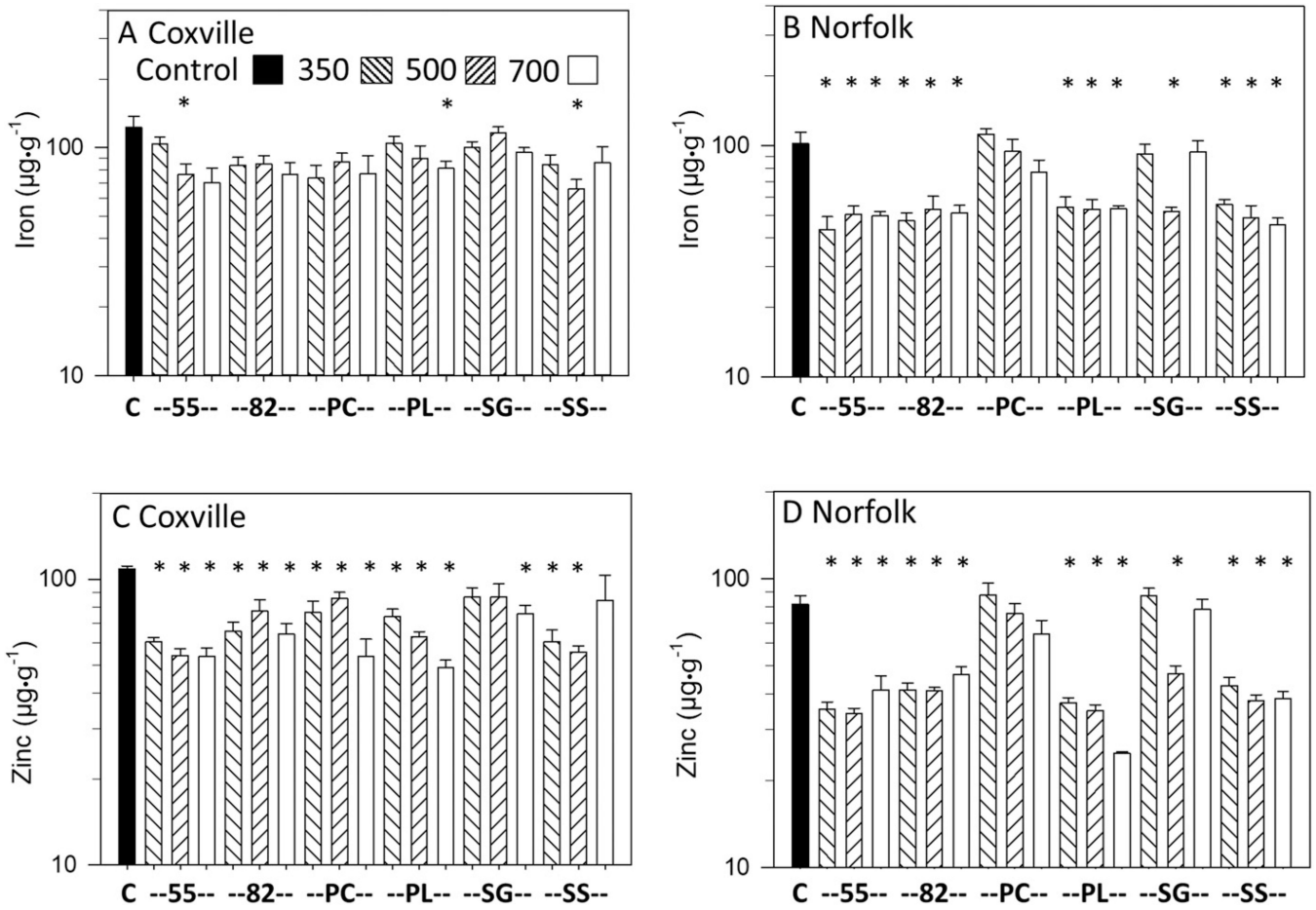

Fig. 3.

Effects of biochar on lettuce leaf Fe and Zn in $\mu g \cdot g^{-1}$. Data are for Fe for the Coxville (**A**) or Norfolk (**B**) soil, and Zn for the Coxville (**C**) or Norfolk (**D**) soil. C = no biochar control, PL = poultry litter, PC = pine chips, 55 = 50% PC and 50% PL, 82 = 80% PC and 20% PL, SS = swine solids, SG = switchgrass. Temperatures in °C are indicated above the graph. Each bar is based on back-transformed least square mean and upper standard error (see methods) for six pots except for 5 for Coxville PL 500, Norfolk PC 350, Norfolk 82 350 and 500, Norfolk PL 500 and 700 for both Fe and Mn, and 5 for Coxville SS 700 for Fe. An "*" above a bar indicates a significant difference vs. control plants according to Dunnett's test.

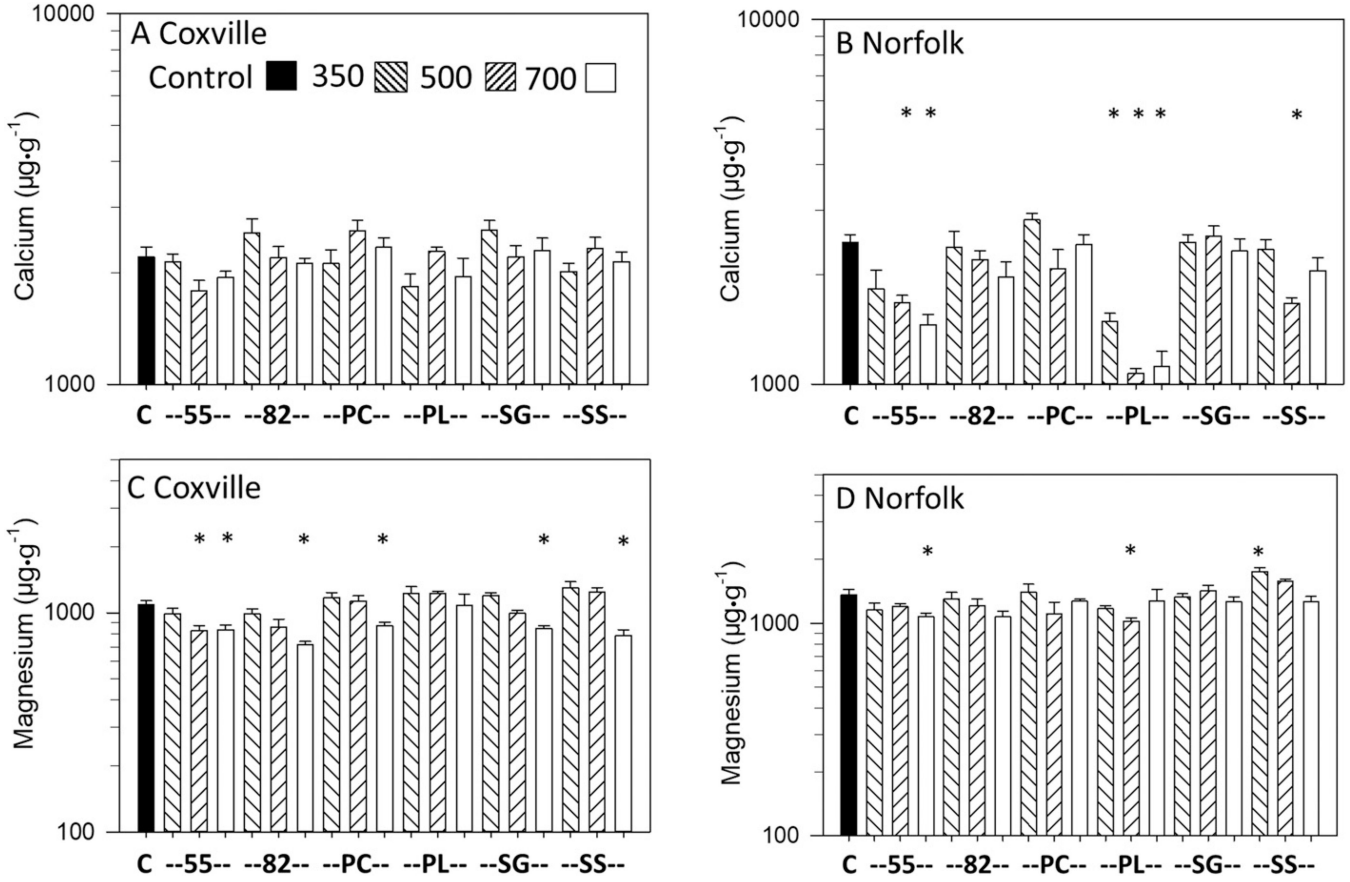

Fig. 4.

Effects of biochar on carrot taproot CaandMg in µg·g^{-1}. Data are for Ca for the Coxville (**A**) or Norfolk (**B**) soil, and Mg for the Coxville (**C**) or Norfolk (**D**) soil. C = no biochar control, PL = poultry litter, PC = pine chips, 55 = 50% PC and 50% PL, 82 = 80% PC and 20% PL, SS = swine solids, SG = switchgrass. Temperatures in °C are indicated above the graph. Each bar is based on back-transformed least square mean and upper standard error (see methods) for six pots except for 3 for Coxville PL 500,4 for Norfolk PL 700, and 5 for Coxville PL 700, Norfolk PC 350, Norfolk 82 350 and 500, Norfolk PL 350 and 500 and 55 700. An "*" above a bar indicates a significant difference vs. control plants according to Dunnett's test.

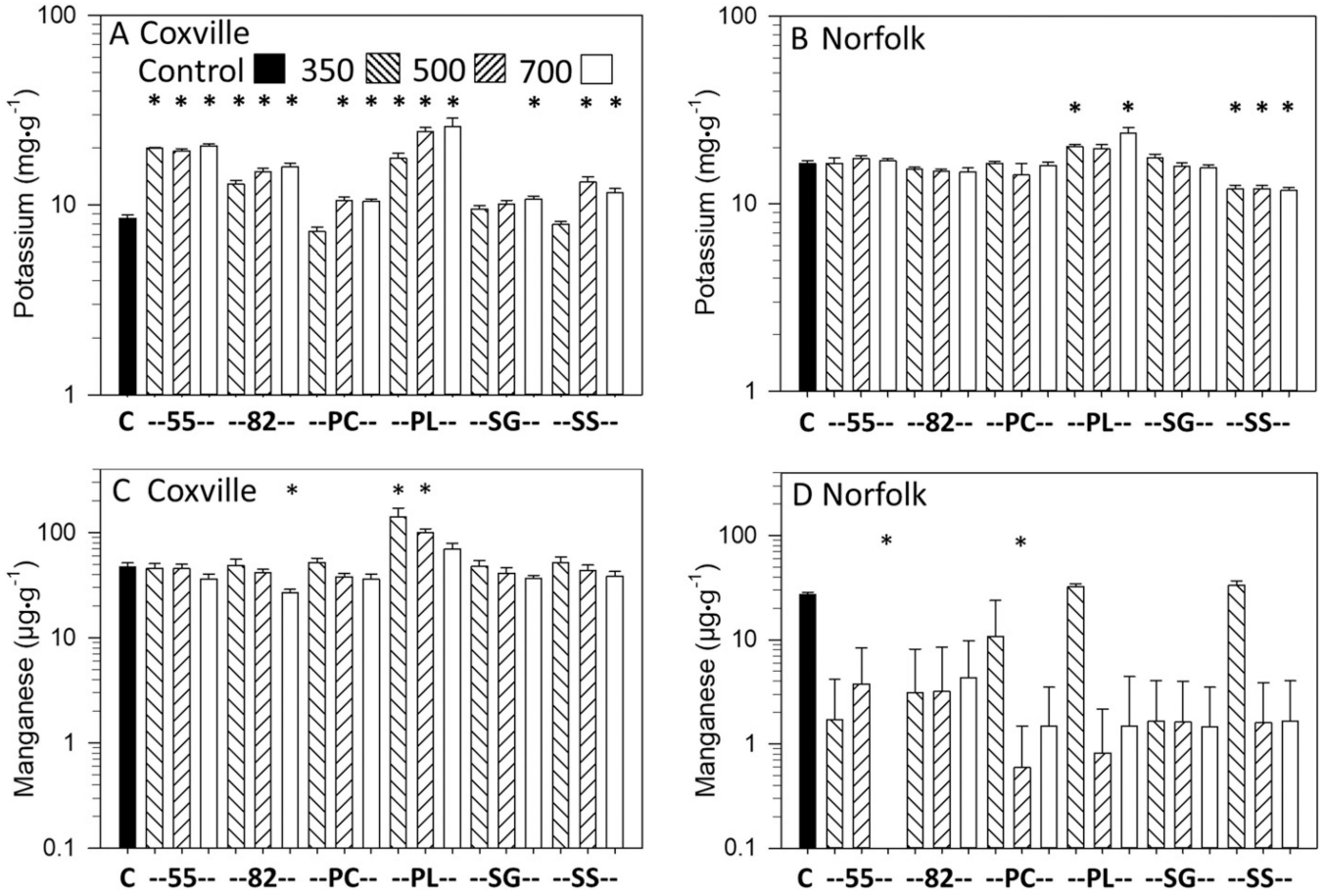

Fig. 5.

Effects of biochar on carrot taproot K in $mg \cdot g^{-1}$ and Mn in $\mu g \cdot g^{-1}$. Data are for Ca for the Coxville (**A**) or Norfolk (**B**) soil, and Mn for the Coxville (**C**) or Norfolk (**D**) soil. C = no biochar control, PL = poultry litter, PC = pine chips, 55 = 50% PC and 50% PL, 82 = 80% PC and 20% PL, SS = swine solids, SG = switchgrass. Temperatures in °C are indicated above the graph. Each bar is based on back-transformed least square mean and upper standard error (see methods) for six pots except for 3 for Coxville PL 500,4 for Norfolk PL 700, and 5 for Coxville PL 700, Norfolk PC 350, Norfolk 82 350 and 500, Norfolk PL 350 and 500 and 55 700. An "*" above a bar indicates a significant difference vs. control plants according to Dunnett's test

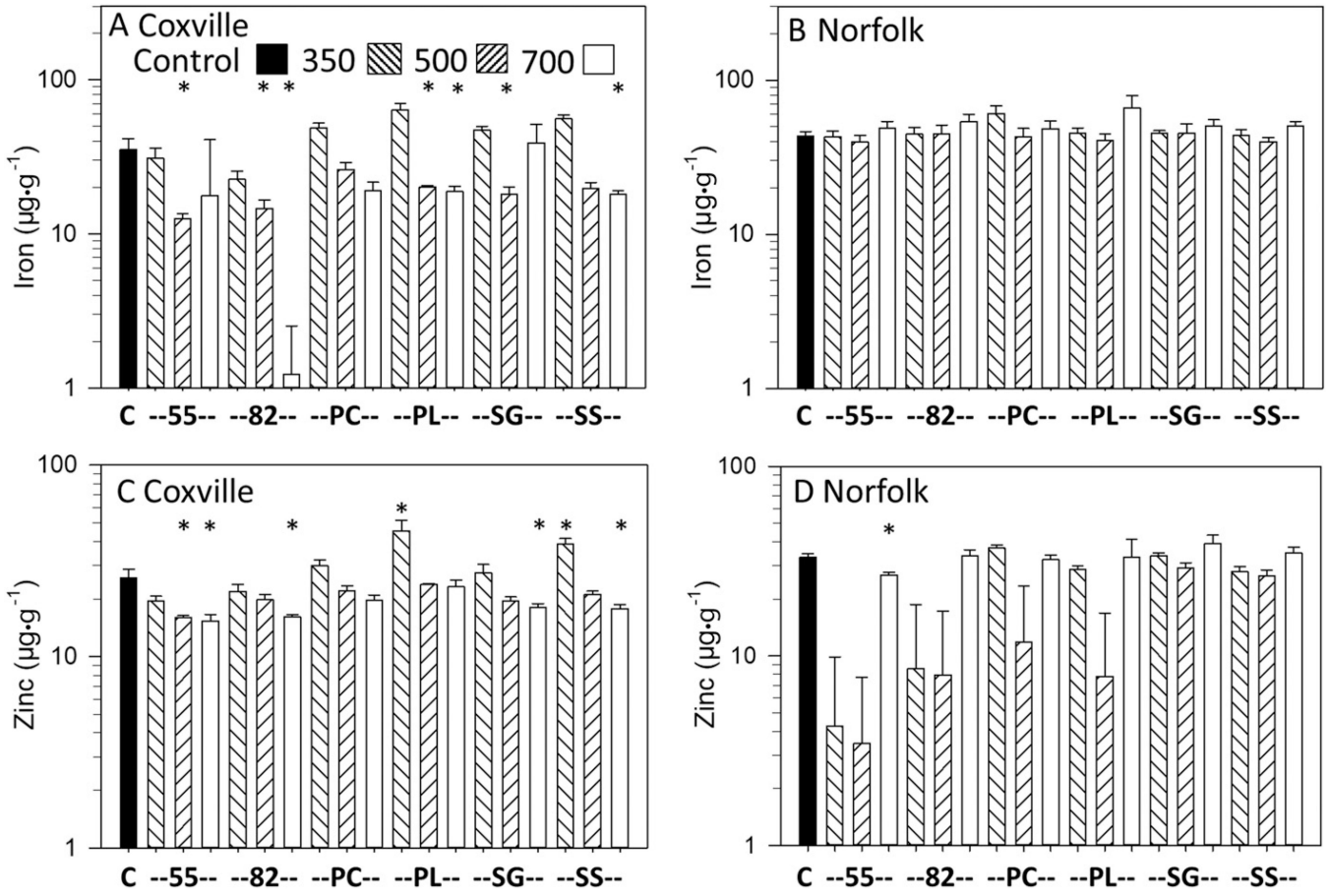

Fig. 6.

Effects of biochar on carrot taproot Fe and Zn in µg·g^{-1}. Data are for Fe for the Coxville (**A**) or Norfolk (**B**) soil, and Zn for the Coxville (**C**) or Norfolk (**D**) soil. C = no biochar control, PL = poultry litter, PC = pine chips, 55 = 50% PC and 50% PL, 82 = 80% PC and 20% PL, SS = swine solids, SG = switchgrass. Temperatures in °C are indicated above the graph. Each bar is based on back-transformed least square mean and upper standard error (see methods) for six pots except for 3 for Coxville PL 500,4 for Norfolk PL 700, and 5 for Coxville PL 700, Norfolk PC 350, Norfolk 82 350 and 500, Norfolk PL 350 and 500 and 55 700. An "*" above a bar indicates a significant difference vs. control plants according to Dunnett's test.

Table 1.

Characteristics of the soils used in this study.[z]

Soil	pH[y]	Sand	Silt	Clay	C[x]	N[w]	K	P	Ca	Mg	Zn	Cu	Mn	B	Na
		---------- g·kg^{-1} ----------					---------- mg·kg^{-1} ----------								
Coxville	5.1	421	434	145	26.3	1.8	40 (1)	44 (1)	321 (2)	53 (0.5)	3.0 (0.1)	0.8 (0.03)	10 (0.4)	0.2 (0)	8 (0.4)
Norfolk	5.9	807	167	26	3.9	ND[v]	53 (2)	17 (0.2)	257 (2)	35 (1)	3.7 (0.1)	0.5 (0)	6 (0.2)	0.1 (0)	4 (0)

[z]Adapted from Sigua et al. (2014) and Olszyk et al. (2018). C = carbon, N = nitrogen, K = potassium, P = phosphorus, Ca = calcium, Mg = magnesium, Zn = zinc, Cu = copper, Mn = manganese, B = boron, Na = sodium. The K, P, Ca, Mg, Zn, Cu, Mn, B, and Na data are based on a Mehlich 1 extraction (Clemson University, 2019; Novak, J. personal communication, 2018) on a dry weight basis, with values averages with standard error in parentheses for three samples.

[y]Soil/water ratio of 1:2.

[x]Organic.

[w]Total.

[v]Not detected, i.e., less than detection limit of 1 g·kg^{-1}.

Table 2.

The pH, electrical conductivity (EC), and extractable phosphorus concentrations (EP) of biochars used in this study.[z]

Feedstock	°C	pH	EC (mS·cm^{-1})	EP (mg·L^{-1})	Feedstock	°C	pH	EC (mS·cm^{-1})	EP (mg·L^{-1})
Poultry litter	350	8.73 (0.01)	16.45 (0.07)	81.3 (2.3)	Swine solids	350	6.94 (0.01)	3.14 (0.01)	181.4 (8.2)
Poultry litter	500	9.76 (0.004)	18.94 (0.07)	61.0 (0.9)	Swine solids	500	7.80 (0.03)	2.98 (0.01)	195.2 (0.6)
Poultry litter	700	10.30 (0.01)	20.39 (0.05)	16.4 (0.2)	Swine solids	700	8.74 (0.09)	1.64 (0.04)	136.8 (3.1)
PC:PL 55	350	7.68 (0.01)	8.59 (0.04)	201.4 (1.9)	Pine chips	350	5.74 (0.03)	0.37 (0.003)	7.2 (0.2)
PC:PL 55	500	9.99 (0.01)	8.99 (0.04)	100.1 (2.3)	Pine chips	500	7.57 (0.01)	0.42 (0.004)	3.6 (0.2)
PC:PL 55	700	10.44 (0.005)	9.92 (0.12)	67.4 (1.6)	Pine chips	700	8.92 (0.01)	0.51 (0.01)	0.04 (0.01)
PC:PL 82	350	7.69 (0.01)	2.54 (0.01)	195.0 (1.6)	Switchgrass	350	5.76 (0.05)	0.33 (0.003)	13.6 (0.5)
PC:PL 82	500	9.66 (0.003)	3.04 (0.07)	104.2 (1.8)	Switchgrass	500	8.38 (0.02)	0.79 (0.01)	44.7 (0.2)
PC:PL 82	700	10.08 (0.002)	3.78 (0.02)	60.3 (1.6)	Switchgrass	700	9.56 (0.01)	0.80 (0.003)	30.2 (0.1)

[z]PC = pine chips, PL = poultry litter, 55 = 50% PC and 50% PL, 82 = 80% PC and 20% PL. The pH, EC, and EP data are adapted from Olszyk et al. (2018). Values are averages with standard error in parentheses for four samples.

Table 3.

Total elemental concentrations of biochars used in this study.[z]

		Al	Ca	Fe	K	Mg	P	S	Cu	Mn	Zn
Feedstock	**°C**	**g·kg^{-1}**							**mg·kg^{-1}**		
Poultry litter	350	0.4	35	2.8	56	12	25	9.8	225	868	911
Poultry litter	500	1.1	47	5.4	70	17	34	11.6	261	1125	1233
Poultry litter	700	1.4	50	7.0	77	18	35	9.8	361	1171	1241
PC:PL 55	350	0.4	21	1,8	34	8	13	5.0	140	501	552
PC:PL 55	500	0.7	26	4.8	36	9	18	4.8	221	627	617
PC:PL 55	700	0.8	27	3.3	43	9	18	3.0	177	638	490
PC:PL 82	350	0.3	9	2.6	12	3	5	1.5	52	220	202
PC:PL 82	500	0.4	12	3.3	16	4	6	1.4	64	284	272
PC:PL 82	700	0.3	10	8.0	14	3	5	<0.1	237	238	142
Swine solids	350	1.0	37	5.8	13	31	50	10.8	1920	1855	5010
Swine solids	500	1.6	53	9.1	21	42	>50	10.1	2384	2545	6836
Swine solids	700	2.2	56	11.6	21	44	>50	7.2	2628	2717	6790
Pine chips	350	0.3	3	0.5	2	0.9	0.3	<0.1	9	82	36
Pine chips	500	0.4	5	1.6	3	1.2	0.6	<0.1	36	125	78
Pine chips	700	0.2	3	2.7	2	0.4	0.2	<0.1	64	65	26
Switchgrass	350	<0.1	3	0.5	2	1.9	0.8	0.7	11	49	28
Switchgrass	500	<0.1	4	1.4	4	2.5	1.5	0.7	44	93	63
Switchgrass	700	<0.1	3	2.9	4	1.0	0.6	<0.1	93	55	24

[z]Values are on a dry weight basis. Al = aluminum, Ca = calcium, Fe = iron, K = potassium, Mg = magnesium, P = phosphorus, S = sulfur, Cu = copper, Mn = manganese, Zn = zinc, PC = pine chips, PL = poultry litter, 55 = 50% PC and 50% PL, 82 = 80% PC and 20% PL. The nutrient data are for one sample with analysis details given in the methods section of this article.