Turkish Journal of Agriculture - Food Science and Technology, 6(3): 346-351, 2018

Turkish Journal of Agriculture - Food Science and Technology

Available online, ISSN: 2148-127X
www.agrifoodscience.com,
Turkish Science and Technology

Effect of Biochar and Nitrogen Applications on Growth of Corn (*Zea mays* L.) Plants

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ARTICLE INFO

Research Article

Received 08 December 2017
Accepted 07 March 2018

Keywords:
Biochar
Corn plant
Limestone
Soil amendment
Nitrogen

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ABSTRACT

The objective of the study was to determine the effect of three different biochars (Pin, Poplar, and Oak biochars), four different biochar doses (0, 1, 2, and 4%), and four different nitrogen rates (0, 70, 140, and 210 mg kg^{-1}) on soil fertility, growth, and nutrient uptake of corn plants. The experiment was conducted in a greenhouse, and corn (*Zea mays* L.) was used as the test plant. The biochar types, biochar doses, and nitrogen rates significantly affected many soil and plant parameters. The highest leaf dry matter yield was obtained with a combination of the poplar biochar, 4% biochar dose, and 140 mg kg^{-1} N application.

DOI: https://doi.org/10.24925/turjaf.v6i3.346-351.1746

Introduction

Biochar term refers to the charcoal that is used as an additive in agricultural production or environmental purposes. It is produced from pyrolysis of various organic materials, i.e. wood, bones and crop residues (Anonymous, 2010). Once it is converted into biochar, it can be mixed with soil to improve its fertility.

Pyrolysis is a thermo-chemical decomposition process in which organic material is converted into a carbon-rich solid and volatile matter by heating in the absence of oxygen. The solid, termed variously as char, biochar, charcoal or coke, has generally high carbon content and may contain around half of the carbon of the original organic material (Lehmann et al., 2006).

Biochar can increase plant growth in a variety of soils by improving soil chemical, physical, and biological properties (Glaser et al., 2002; Lehmann and Rondon, 2006; Yamato et al., 2006). The positive biochar responses are associated with a combination of increased soil pH of acidic soils (Chan et al., 2007; Rondon et al., 2007; Yamato et al., 2006), enhanced physical properties such as water holding capacity (Iswaran et al., 1980), retention of soil nutrients, and reduced leaching losses of nutrients (Hoshi, 2001; Lehmann et al., 2003; Lehmann, 2007). Biochar applications result in increased N availability and retention when applied along with N fertilizers (Chan et al., 2007; Steiner et al., 2008; Van Zwieten et al., 2010).

Studies have shown that biochar applications affect crop yields ranging from yield increases as much as 266% in radish when applied with N fertilizer, to modest increases (between 38-140%) and even decreases in some conditions (Chan et al., 2007; Gaskin et al., 2010; Major et al., 2010). Negative yield responses have been reported and explained with micronutrient deficiencies caused by high pH values of biochars (Mikan and Abrams, 1995).

The differences in plant response to biochar applications depend on the properties of the biochar, application rate, soil type, and plant variety studied. The objective of the present study was to determine the effect of different biochar types and doses along with different nitrogen application rates on corn growth and nutrient uptake.

Materials and Methods

The effect of biochar on plant growth was tested using three different biochars (pine (*Pinus nigra*), poplar (*Populus tremula* L.), and oak (*Quercus coccifera*) biochar), four different biochar doses (0, 1, 2, and 4 % biochar w/w), and four nitrogen rates (0, 70,140, and 210 mg kg^{-1} N). A factorial experiment (3×4×4) in randomized block design with three replications was used in this study.

A soil formed on limestone was taken from Çokyaşar Village of Kahramanmaraş, Turkey. The biochars were produced from pine, poplar, and oak woods by a small-scale pyrolysis system (double barrel design). The particle size of biochar was fit to pass a 2 mm sieve. The biochar was mixed manually at each application rate until the biochar - 4kg soil mixture was homogenous for each pot to make up the four application rates of 0, 1, 2, and, 4% biochar. The greenhouse conditions consisted of nearly 12 hr light cycle, 25–35ºC daytime temperature and 18–23ºC nighttime temperatures.

Five seeds of sweet corn were seeded in each pot at a depth of 2–3 cm. A week after germination, plants were thinned to 1 plant per pot. Nitrogen was added as NH_4NO_3 (33%N) at four rates (0, 70,140, and 210 mg kg^{-1} N) in two split applications, which were 18 and 35 days after germination. In addition to nitrogen, phosphorous was added 8 days after germination to all pots in the form of triple super phosphate (TSP) with a single application rate of 50 mg kg^{-1} P. All pots were watered evenly and on an "as needed" basis. Watering frequency ranged from once per week at the beginning of the experiment when plants were small to three times per week towards the end of the experiment when plants were larger and required more water. Plants were harvested 90 days after planting. The biomass (leaves, stem, and root) was then dried in an oven at 60-80°C.

Soil samples were air dried and crushed to pass a 2-mm sieve. The samples were analyzed for organic carbon by wet oxidation (Nelson and Sommers, 1996), soluble salts (Rhoades, 1996), pH (Thomas, 1996), $CaCO_3$ (Loeppert and Suarez, 1996), Olsen P (Kuo, 1996), ammonium acetate extractable K, Ca, Mg, and Na (Helmke and Sparks, 1996), plant available Cu, Fe, Mn, and Zn by DTPA method (Lindsay and Norvell, 1978). The Kjeldahl method was employed for total N analysis (Bremner, 1996).

The plant samples were digested with $HNO_3/HClO_4$ mixture for P, K, Ca, Mg, Fe, Mn, Zn, and Cu concentrations (Jones and Case, 1990). Total plant phosphorus was determined by vanadomolybdophosphoric acid method (Kuo, 1996), and total K, Ca, Mg, Fe, Mn, Zn, and Cu were measured in the digest using an atomic absorption spectrophotometer (Perkin Elmer, 3110).

Some chemical and physical properties of the soil used in this study and biochars are given in Table 1. The soil was slightly alkaline in reaction, low in plant available P, K, and Zn.

Although it was not statistically tested, there were variations for the measured chemical and physical properties among the biochars (Table 1). Biochars had very high pH values, and the poplar biochar contained the highest amounts of P, K, and Mg.

The differences among treatments were investigated by analysis of variance (ANOVA) using IBM SPSS Advanced Statistics version 20.0.0 software. The Duncans' multiple comparison test was used to evaluate the main effects of treatments that differed when the F-value was significant at $P \leq 0.05$.

Results

Biochar Effect on Post Harvest Soil Properties

Biochar types did not affect pH and electrical conductivity of soils (Table 2). The soils mixed with pine and poplar biochars had higher lime content compared with that of oak biochar. The poplar biochar caused higher soil organic matter content relative to the oak and pine biochars.

The biochars did not increase plant available P, K, and Zn values relative to the critical levels given for each nutrient (Table 3). The poplar and pine biochar amended soils had significantly higher plant available P compared with the oak biochar amended soils (Table 3). There was no significant difference for Ca and Mg concentrations among the different biochar amended soils. The plant available K and extractable Na of oak biochar amended soils were significantly higher compared with the poplar biochar, and the pine biochar caused the lowest values. The soils amended with poplar biochar had the highest plant available Zn, followed by the soils amended with the oak and pine biochars, respectively. The available Cu decreased significantly in the order of pine, poplar, and oak biochar soils. The DTPA-extractable Mn was lower in the soils of pine biochar compared with the soils amended with poplar and oak biochars.

Table 1 Some plant available nutrients, pH, and EC values of the soil and the three biochars

Material	P	K	Ca	Mg	Na	Zn	Mn	Cu	pH	EC
	mg kg^{-1}									dS m^{-1}
Soil	3.52	152.99	5100.94	300.54	49.67	0.60	6.90	2.56	7.77	2.19
Pine	17.96	4931.36	1352.93	280.72	163.84	7.23	21.91	0.32	9.05	-
Poplar	145.78	7739.06	1237.60	613.50	268.78	9.24	5.28	0.70	9.45	-
Oak	27.32	5122.46	1410.59	158.59	81.52	3.49	25.47	0.32	9.25	-

Table 2 The effect of biochar types on some physical properties of soils

Biochar Type	pH	EC (dS m^{-1})	$CaCO_3$ (%)	Organic Matter (%)
Pine	7.79	2.01	13.75^a	2.23^b
Poplar	7.81	1.99	13.91^a	2.69^a
Oak	7.82	2.03	13.45^b	2.19^b

Means in the same column followed by the same symbol are not significantly different at $P \leq 0.05$ level based on Duncan test.

Table 3 The effect of biochar types on some extractable nutrients and Na (mg kg^{-1})

Biochar	P	K	Ca	Mg	Na	Cu	Zn	Mn
Pine	7.67^{a}	90.76^{c}	6443.08	339.26	36.64^{c}	0.64^{a}	0.48^{c}	2.88^{b}
Poplar	7.53^{a}	105.33^{b}	7086.16	333.82	46.07^{b}	0.53^{b}	0.79^{a}	4.37^{a}
Oak	6.96^{b}	128.77^{a}	7379.02	346.99	51.13^{a}	0.47^{c}	0.51^{b}	4.26^{a}

Means in the same column followed by the same symbol are not significantly different at P≤0.05 level based on Duncan test.

Table 4 The effect of biochar doses on some physical properties of soils

Biochar Doses (%)	pH	EC	$CaCO_3$ (%)	Organic Matter (%)
0	7.53^{c}	1.70^{c}	16.06^{a}	1.41^{d}
1	7.86^{b}	2.02^{b}	13.84^{b}	2.10^{c}
2	7.90^{a}	2.12^{a}	12.80^{c}	2.66^{b}
4	7.93^{a}	2.19^{a}	12.12^{d}	3.32^{a}

Means in the same column followed by the same symbol are not significantly different at P≤0.05 level based on Duncan test.

Table 5 The effect of biochar doses on some extractable nutrients and Na (mg kg^{-1})

Biochar Doses (%)	P	K	Ca	Mg	Na	Cu	Zn	Mn
0	5.93^{d}	78.61^{d}	6380.22	303.92^{c}	35.06^{d}	0.47^{b}	0.29^{d}	2.88^{b}
1	6.65^{c}	90.68^{c}	5155.73	365.18^{a}	41.58^{c}	0.57^{a}	0.57^{c}	4.15^{a}
2	7.10^{b}	108.33^{b}	9113.74	348.19^{b}	47.03^{b}	0.57^{a}	0.67^{b}	4.18^{a}
4	8.98^{a}	155.53^{a}	7227.91	342.81^{b}	54.78^{a}	0.57^{a}	0.86^{a}	4.13^{a}

Means in the same column followed by the same symbol are not significantly different at P≤0.05 level based on Duncan test.

Table 6 The effect of biochar types on leaf elemental content

Biochar	N	P	K	Ca	Mg	Na	Cu	Zn	Mn	Fe
	%					mg kg^{-1}				
Pine	2.45^{b}	0.19^{a}	1.98^{b}	0.78^{b}	0.28^{a}	233.98^{a}	24.14	40.40^{a}	101.00^{b}	140.34^{c}
Poplar	2.66^{a}	0.19^{a}	1.91^{c}	0.76^{b}	0.25^{b}	173.78^{c}	24.42	35.36^{b}	95.21^{c}	179.68^{a}
Oak	2.32^{c}	0.17^{b}	2.12^{a}	0.81^{a}	0.26^{b}	189.17^{b}	24.00	34.82^{b}	119.74^{a}	168.31^{b}

Means in the same column followed by the same symbol are not significantly different at P≤0.05 level based on Duncan test.

Table 7 The effect of biochar doses on leaf elemental content

Biochar Doses (%)	N	P	K	Ca	Mg	Na	Cu	Zn	Mn	Fe
	%					mg kg^{-1}				
0	2.19^{d}	0.16^{b}	1.68^{d}	0.65^{c}	0.28^{a}	214.07^{a}	24.00	28.57^{c}	90.15^{c}	157.40^{b}
1	2.52^{c}	0.19^{a}	2.03^{c}	0.80^{b}	0.28^{a}	195.74^{b}	24.68	39.08^{b}	113.74^{a}	162.67^{b}
2	2.58^{b}	0.20^{a}	2.08^{b}	0.81^{b}	0.25^{b}	185.70^{c}	23.88	37.78^{b}	104.48^{b}	159.81^{b}
4	2.62^{a}	0.19^{a}	2.23^{a}	0.86^{a}	0.24^{c}	200.39^{b}	24.19	41.99^{a}	112.91^{a}	171.24^{a}

Means in the same column followed by the same symbol are not significantly different at P≤0.05 level based on Duncan test.

Biochar applications caused a sharp increase in soil pH, and then a steady increase was observed with each application dose up to 2%. There was no significant difference between the highest two application doses (Table 4). The changes in EC values with biochar doses also followed the same trends as in soil pH. Soil organic matter levels increased and lime content decreased significantly as biochar doses increased (Table 4). The decrease of lime content was probably due to addition of low lime containing biochars to the soils.

Biochar applications affected the concentration of plant available nutrients (Table 5). Increasing the biochar doses increased the plant available P, K, Na, and Zn. The availability Cu and Mn increased with biochar applications over the control but there was no significant differences among biochar application rates. Although there were some degree of variability among the treatments, the plant available Ca concentrations did not change significantly with the biochar doses. Plant available Mg increased with biochar doses over the control but the highest Mg levels were measured in soils that received 1% biochar.

Effect on Nutrient Content of Corn Leaves

The biochar types affected the nutrient composition of corn leaves (Table 6). The leaves of the corn plants grown in the soils amended with poplar biochar removed more N than those grown in the soils of pine biochar, while the oak biochar amended soil provided the least available N (Table 6). The corn plants grown in oak biochar amended soils removed the highest K, Ca, and Mn but the lowest P among the three biochar types. The plants grown in the pine biochar amended soils removed the highest Mg, Na, and Zn but the lowest Fe concentrations with their leaves. The poplar biochar, on the other hand, was superior for the removal of N and Fe but failed to provide adequate amounts of K and Mn compared with the other two.

Increasing the biochar dose increased the concentrations of N, K, Ca, Zn, and Mn in the corn leaves (Table 7). The biochar applications increased significantly the leaf P compared to the control (0% biochar). The Fe content of the leaves was significantly higher only with the highest biochar application rate, and Cu content of the leaves was not affected from the biochar applications. The

Na content of the leaves decreased with the biochar doses and the leaf Mg also decreased with biochar application doses over 1%.

The nitrogen applications resulted in higher N uptake by the leaves, although with the 70 mg kg^{-1} N application N content of the leaves was not statistically different from the control (0 mg kg^{-1} N) (Table 8). The nitrogen applications significantly increased leaf P, K, Ca, and Mg over the control. The micronutrient uptake of the leaves generally increased with N rates (Table 8).

Effect on Corn Plants Dry Matter Yield

The dry matter of all plant parts (leaf, stem, and root) of the corn plants grown in the soils of the poplar biochar was higher than that grown in the soils of the pine and the oak biochar. The pine biochar amended soil produced the lowest stem weights (Table 9).

Increasing the biochar dose also increased significantly the weights of leaf and root of the corn plants (Table 10). The biochar applications just increased stem weights significantly over the control (0% biochar).

The leaf weights increased up to 140 mg kg^{-1} N applications and then decreased at the highest rate (Table 11). The stem and root weights of corn plants did not respond to N applications and a decrease was observed especially at the highest N application rate.

The best combination among the biochar doses, biochar types, and nitrogen rates for the leaf dry weight was a combination of the 4% biochar, the poplar biochar, and the 140 mg kg^{-1} N application.

Table 8 The effect of nitrogen rates(mg kg^{-1}) on leaf elemental content

Nitrogen Rates	N	P	K	Ca	Mg	Na	Cu	Zn	Mn	Fe
	%					mg kg^{-1}				
0	1.96^c	0.17^b	1.95^b	0.72^c	0.20^c	202.73^a	20.26^c	33.56^d	84.25^c	151.95^c
70	2.38^c	0.18^a	2.02^a	0.77^b	0.27^b	190.18^b	23.39^b	34.92^c	103.02^b	137.81^d
140	2.77^b	0.19^a	2.04^a	0.82^a	0.29^a	195.45^b	26.71^a	40.58^a	119.06^a	169.81^b
210	2.81^a	0.19^a	2.00^a	0.82^a	0.29^a	207.55^a	26.40^a	38.35^b	114.94^a	191.54^a

Means in the same column followed by the same symbol are not significantly different at P≤0.05 level based on Duncan test.

Table 9 The effect of the biochar types on the dry masses (DM) of the leaf, stem, and root of the corn plants

Biochar Types	Leaf D.M. (g)	Stem D.M. (g)	Root D.M. (g)
Pine Biochar	6.35^b	12.53^c	4.21^a
Poplar Biochar	7.33^a	15.01^a	4.56^a
Oak Biochar	6.11^b	13.76^b	3.74^b

Means in the same column followed by the same symbol are not significantly different at P≤0.05 level based on Duncan test.

Table 10 The effect of the biochar doses on the dry masses (DM) of the leaf, stem, and root of the corn plants

Biochar Doses (%)	Leaf D.M. (g)	Stem D.M. (g)	Root D.M. (g)
0	5.45^d	11.41^b	2.74^d
1	6.16^c	13.96^a	3.93^c
2	7.22^b	14.90^a	4.58^b
4	7.56^a	14.80^a	5.43^a

Means in the same column followed by the same symbol are not significantly different at P≤0.05 level based on Duncan test.

Table 11 The effect of the nitrogen rates on the dry masses (DM) of the leaf, stem, and root of the corn plants.

Nitrogen Rate mg kg^{-1}	Leaf D.M. (g)	Stem D.M. (g)	Root D.M. (g)
0	5.41^c	14.26^a	4.75^a
70	7.01^a	14.03^{ab}	4.15^b
140	7.40^a	13.71^{ab}	3.66^c
210	6.47^b	13.08^b	4.13^b

Means in the same column followed by the same symbol are not significantly different at P≤0.05 level based on Duncan test.

Discussion

The addition of biochars influenced nutrient availability and growth of corn plants; however, the biochars showed different responses. The most significant effect of biochar applications associated with a steady increase in soil organic matter levels. Addition of each 1% biochar dose resulted in an avarage of 0.64% increment in soil organic matter levels. Considering biochar is a quite stable organic carbon form in soils, its application can be an effective mechanism to increase organic matter levels of soils and to reduce carbon emissions.

Biochars improved soil fertility by increasing availability of some plant nutrients that resulted in increased uptake of these nutrients by corn plants. The availability of Ca, K, P, Cu, and Zn increased with the biochar applications especially at 4% dose. The results are in line with Major et al. (2010) who reported that the availability of K, Ca, and Mg increased with the biochar applications. An increased nitrogen availability measured in the study with biochar applications was also reported by Liang et al. (2014) who observed that biochar applications increased total nitrogen and nitrate concentrations but caused a decrease in ammonium levels of soils.

Addition of base cations from the pine, poplar and oak biochars increased soil pH, which is consistent with the report of increased pH due to base cations in hardwood charcoal by Tryon (1948). The other reports also emphasized that biochar can indirectly affect nutrient availability by altering soil pH. Since biochar typically has a higher pH than a soil it can act as a liming agent resulting in a change in soil pH (Glaser et al., 2002; Lehmann and Rondon, 2006).

The most important drawback of biochar applications found in this study was the effects of high pH biochars on soil pH. Slightly alkaline soil pH values (7.53) increased to moderately alkaline values (7.90-7.93) with biochar applications. These pH increases will affect availability of plant nutrients. Therefore, pH of both selected biochar and the soil that will receive the application are the two important parameters for biochar amendments. Naeem et al. (2016) reported that as pyrolysis temperature increased from 300 to 500°C, pH, soluble salt, and total C contents of biochars increased significantly.

Cao et al. (2018) also measured higher pH, total C, and available N and K contents in biochars in the expense of biochar yields when pyrolysis temperature and time increased. Manolikaki and Diamadopoulos (2017) found that biochar applications increased crop yields and phosphorus in slightly acidic sandy loam soil but similar results were not observed in alkaline loam soil. They also observed no yield gains if biochar applications were not supplemented with inorganic fertilization with the exception of P.

Biochar type was found to be a significant factor in the study. It has been shown that the biochar type can have positive, neutral, and negative impacts on plant yields (Chan et al., 2007; Gaskin et al., 2010; Major et al., 2010; Van Zwieten et al., 2010). As shown previously, compared to the pine-derived biochars, switchgrass-derived biochars had higher ash contents resulting in higher inorganic elements, many of which are plant nutrients (Edmunds, 2012).

Nitrogen was added as an experimental factor in this study because previous research suggested that the application of N together with the biochar positively affected plant growth (Van Zwieten et al., 2010). Similar synergetic effects have also been reported in other field (Yamato et al., 2006) and greenhouse experiments (Chan et al., 2007). The results further confirm that biochar as a soil amendment can efficiently increase availability of the nutrients by holding ammonium ions in soils and inhibiting nitrification of nitrogen (Spokas et al., 2009). The neutral or negative plant growth responses have been observed usually where biochar applied to soils without any plant nutrients especially N (Asai et al., 2009; Gaskin et al., 2010). However, when biochar amendments were combined with fertilizers, crop yields increased to a much greater extent than with fertilizer additions in the absence of biochar (Asai et al., 2009; Blackwell et al., 2009). Van Zwieten et al. (2009) reported no significant effects of biochar in the absence of fertilizer for certain plant and soil types, while the greatest biomass increase was observed with the application of biochar plus N fertilizers. Biochar applications along with N fertilizer caused increases in biomass yield in this study.

Conclusion

Biochar types and doses, and nitrogen fertilizer doses were all effective factors on soil fertility and corn growths. In this respect, biochar applications can be used as an effective mechanism to increase stable soil organic matter levels. Soil and biochar pH values were found to be the two important parameters, which may restrict use of biochars in soils. Further studies are needed to understand crop and soil responses to biochar and to develop recommendations for particular biochars in different soils.

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