

# Biochar Increases Rice Yield by Improving Root Morphological and Root Physiological Functions in Heavily Saline-sodic Paddy Soil of Northeast China

Xiang Li,<sup>a</sup> Tianxu Yao,<sup>a</sup> Xiaoxuan Huang,<sup>b</sup> Xuebin Li,<sup>a</sup> Pingyue Li,<sup>a</sup> Shuang Du,<sup>a</sup> Wei Wang,<sup>a</sup> Sihao Miao,<sup>a</sup> Dan Wang,<sup>c</sup> Feng Jin,<sup>d,\*</sup> and Xiwen Shao<sup>a,\*</sup>

Saline-sodic soil is one of the major threats to crop growth, production, and quality. Biochar amendment could alleviate the adverse impacts of saline-sodic stress in crops. However, the effect of biochar on root morphological, root physiological functions, and rice yield in saline-sodic paddy soil has not been studied. Here, the underlying mechanisms of positive effects in morphological characteristics and physiological functions of rice roots under heavily saline-sodic paddy soil amended with biochar were evaluated. The soil was amended with biochar at zero-biochar (CK), 15g kg<sup>-1</sup> soil (T1), 30 g kg<sup>-1</sup> soil (T2), or 45 g kg<sup>-1</sup> soil (T3). Biochar addition significantly increased root length, root volume, root dry weight, and root-shoot ratio at all growth stages. The root absorption vigor and root oxidization capacity were enhanced and increased significantly by decreasing Na<sup>+</sup>/K<sup>+</sup> ratio, MDA content, and increasing K<sup>+</sup> concentration in rice root after biochar amendment. The root bleeding rate of biochar treatments at different growth stage were 11.01% to 67.73% greater than these of CK. The yield of rice was increased significantly under saline-sodic paddy soil after biochar amendment. It was concluded that biochar had positive effect on the morphological characteristics and physiological functions in rice roots in saline-sodic paddy soil.

DOI: 10.15376/biores.17.1.1241-1256

*Keywords:* Biochar; *Oryza sativa* L.; Saline-sodic stress; Root function; Yield

*Contact information:* a: Agronomy College, Jilin Agricultural University, Changchun 130118, China; b: Changchun Academy of Agriculture Science, Changchun 130111, China; c: Changchun Agricultural Technology Extension Station; d: Key Laboratory of Straw Biology and Utilization, The Ministry of Education, Jilin Agricultural University, Changchun 130118, China; \*Corresponding author: jinfeng@jlau.edu.cn, 616530670@qq.com; Xiang Li, Tianxu Yao, and Xiaoxuan Huang contributed equally to this work.

## INTRODUCTION

Soil degradation induced by salinization and/or alkalization is a problem that adversely affects agricultural production and sustainable development (Suarez 2001). Crop growth and development are inhibited by poor physicochemical and biological properties in salt-affected soils (Qadir and Oster 2002). The Western Songnen Plain, located in northeast China (42° 30' N, 121° 40' E), is one of the five largest saline-sodic soils in the world (Yang 2015). The growth and development of crops are affected by many direct and indirect ways under saline-sodic soil. Crop growth is restricted due to the Na<sup>+</sup> toxicity and high pH value in soil solution (Al-Karaki 1997; Elgharably 2011). Indirectly, crops growth may be reduced due to (i) decreased availability of water to plants caused by high osmotic

pressure of soil solution (Munns and Tester 2008; Rengasamy 2010), (ii) decreased availability and uptake of crop nutrients due to the high  $\text{Na}^+$  concentration (Qadir and Oster 2002), (iii) poor root growth caused by Na-induced deterioration of soil physical properties (Elgharably 2011), and (iv) low levels of organic carbon and poor of soil microbial activity (Shahram *et al.* 2018).

The physicochemical and biological characteristics of salt-affected soils have been shown to be improved by biochar (Chaganti and Crohn 2015; Ali *et al.* 2017; Wei *et al.* 2020). The role of biochar alleviating the salt-affected stress and promoting crop growth has been attributed directly through the provision of mineral nutrients for crop growth, and indirectly through lower soil bulk density (Akhtar *et al.* 2015; Liu *et al.* 2020b), improved soil stomatal density (Yang *et al.* 2014), and increased osmotic coefficient by enhancement of the K availability for higher  $\text{K}^+ / \text{Na}^+$  ratio (Abrishamkesh *et al.* 2015; Chakraborty *et al.* 2016). The authors' previous research found that biochar could reduce the adverse effect of saline-sodic stress, increase dry biomass and yield, and improve quality of rice largely due to the high adsorption capacity of biochar to  $\text{Na}^+$  and the cell membrane damage improvement (Ran *et al.* 2020; Yao *et al.* 2021). Farhangi-Abriz and Torabian (2017) indicated that biochar application could increase plant growth through degradation of  $\text{O}_2^{2-}$  and  $\text{H}_2\text{O}_2$  concentrations, antioxidant activities, and MDA content in plant cells under salted-soils. Similar findings have been reported in soybean amended with modified biochar (Mehmood *et al.* 2020).

Recent research has shown that biochar applications could optimize morphology and physiological characteristics, facilitate root elongation and root cell viability, enhance root absorption vigor under non-stressed soil by increasing the soil capacity to store water, and improve soil nutrient status and soil structure (Bruun *et al.* 2014; Xiao *et al.* 2016; Shahram *et al.* 2018; Liu *et al.* 2020a). Under salt-stress condition, Mehmood *et al.* (2020) found that the crucial role of biochar in minimizing the adverse effects of high salinity on soybean growth and efficiency of the mechanisms enabling protecting from salinity through a shift of the architecture of the root system and enhancing the antioxidant defense systems and stress-responsive genes. Shahram *et al.* (2018) reported similar results on mung bean.

Although many studies of biochar applications on salted soil have been reported, most studies have focused on dryland crops or on the aboveground part of crops (Chakraborty *et al.* 2016; Farhangi-Abriz and Torabian 2017; Liu *et al.* 2020a; Yao *et al.* 2021). To date, there are few reports on the effect of biochar on root morphological characteristics and physiological functions of rice under saline-sodic paddy soil. And the potential mechanism needs to be clarified. This study examined how the biochar application mediates a response to the saline-sodic stress in rice roots. The aims of this study were to find out whether the biochar application could improve rice yield through root morphological characteristics and physiological functions modifications under saline-sodic stress, and explore the underlying mechanisms of action. It is prognosticated that the research finding will be helpful to health parameters of saline-sodic paddy field and promote the rice production in saline-sodic lands.

## EXPERIMENTAL

### Setup of Experiment

A pot experiment was used to study the morphology and physiological indicators. The experiments were conducted at the Jilin Agriculture University's experimental station located in Sheli Country, Baicheng City, Jilin Province, China (45° 35' N, 123° 50' E). The mean annual temperature in this experimental region is 4.7 °C. The average annual rainfall is 413.7 mm. The average annual evaporation is 1697 mm.

The pot experiment under open-air conditions included one hundred and ninety-two pots (height: 40 cm; internal diameter: 30 cm), which were filled with 8.5 kg soil with varying amounts of biochar. Biochar was added at rates of 0 g kg<sup>-1</sup> soil (CK), 15 g kg<sup>-1</sup> soil (T1), 30 g kg<sup>-1</sup> soil (T2), and 45 g kg<sup>-1</sup> soil (T3). Each treatment was made in three replicated and laid out by completely randomized block design.

### Preparation of Experimental Soil

The experimental soil samples were randomly collected at a depth of 0 to 25 cm for a total amount of approximately 3,000 kg from Jilin Agriculture University's experimental station. The experimental soil sieved through a 2 mm sieve - after air-dried. The soil was the solonchak type according to the World Reference Base for Soil Resources (IUSS Working Group 2014). The salinity, sodicity, pH, sand, silt, clay, bulk density, and organic matter contents of experimental soils are shown in Table 1.

**Table 1.** Characteristics of the Analyzed Soil

Soil Characteristic	Value
Sand content (%)	22.13 ± 0.97
Silt content (%)	39.14 ± 1.59
Clay content (%)	35.60 ± 1.11
Bulk density (g cm <sup>-3</sup> )	1.38 ± 0.01
EC <sub>ep</sub> (s m <sup>-1</sup> )	23.91 ± 0.59
pH	9.64 ± 0.06
SAR <sub>e</sub> (mmolc L <sup>-1</sup> ) <sup>1/2</sup>	352.11 ± 2.21
ESP (%)	69.38 ± 2.21
Organic matter (%)	0.81 ± 0.03

EC<sub>e</sub>: electrical conductivity of soil saturation extract, SAR: sodium adsorption ratio, ESP: exchangeable sodium percentage

### Biochar Properties

Biochar was made by pyrolysis of peanut shells under anoxic condition and 350 °C to 550 °C for 4 h in a vertical kiln. The biochar mass was ground to small pieces and then sieved through a 2 mm sieve. The biochar was thoroughly mixed to obtain a powdery consistency to ensure a uniform distribution in soil. The mineral contents were determined using an AAS elemental analyzer (GBC-906, GBC Science instruments Co. Ltd., Melbourne, Australia). The total content of organic carbon and nitrogen were measured using the Vario-Max Cube analyzer (Analysensysteme GmbH, Langensfeld, Germany) according to a previous study (Lu 2000). The pH was measured with a 1:5 biochar / water suspension using the Seven Easy pH-meter and electrode comprehensive tester (S30K Mettler Toledo, Shanghai, China). The properties of raw peanut shell and biochar are presented in Table 2.

**Table 2.** Properties of Raw Peanut Shell and Biochar

pH and Elemental Component	Peanut Shell	
	Raw Material	Biochar
pH	5.56 ± 0.11	7.94 ± 0.32
C (mg·g <sup>-1</sup> )	429.19 ± 13.05	540.64 ± 26.58
N (mg·g <sup>-1</sup> )	10.85 ± 0.61	15.93 ± 1.01
S (mg·g <sup>-1</sup> )	2.58 ± 0.05	6.85 ± 0.34
Mg (mg·g <sup>-1</sup> )	0.29 ± 0.00	0.74 ± 0.03
K (mg·g <sup>-1</sup> )	1.46 ± 0.01	0.25 ± 0.00
P (mg·g <sup>-1</sup> )	5.51 ± 0.21	12.53 ± 0.51
Ca (mg·g <sup>-1</sup> )	6.32 ± 0.43	2.01 ± 0.02
Fe (mg·g <sup>-1</sup> )	2.77 ± 0.09	2.07 ± 0.00
Be (mg·g <sup>-1</sup> )	0.14 ± 0.00	13.41 ± 1.22
Mn (mg·g <sup>-1</sup> )	0.09 ± 0.00	0.06 ± 0.00
Ni (mg·g <sup>-1</sup> )	0.00 ± 0.00	0.00 ± 0.00
Cu (mg·g <sup>-1</sup> )	0.01 ± 0.00	0.01 ± 0.00
Zn (mg·g <sup>-1</sup> )	0.02 ± 0.00	0.15 ± 0.00
B (mg·g <sup>-1</sup> )	0.02 ± 0.00	0.01 ± 0.00

C: carbon, N: nitrogen, S: Sulfur, Mg: magnesium, K: potassium, P: phosphorus, Ca: calcium, Fe: ferrum, Be: beryllium, Mn: manganese, Ni: nickel, Cu: copper, Zn: zinc, B: boron

### Planting Rice

The rice variety Changbai 9 was cultivated for this experiment. The seeds were placed in tray with cattle manure compost and germinated in an unheated greenhouse. On May 22, 2018, thirty-nine days old seedlings were transplanted to each pot in the way of two hills per pot and one seedling per hill. A base fertilizer of 30 g (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub> per pot, 15 g diammonium-phosphate per pot, and 7.5 g K<sub>2</sub>SO<sub>4</sub> per pot was applied before transplanting. A total of 15 g per pot of urea was applied at the tillering stage (7 days after transplanting), and 6 g per pot urea and 5 kg K<sub>2</sub>SO<sub>4</sub> were applied at the panicle stage. The total N, P, and K were 5.25 g, 2.25 g, and 1.875 g per pot, respectively. Biochar was broadcast on the soil surface and incorporated into the soil by plowing to depth of about 15 cm 10 days before transplanting. Water, weeds, insects, and diseases were controlled as required to minimize yield loss.

### Root Physiological and Yield Measurements

For each treatment, rice plants from 24 pots were analyzed at the following growth stages: tillering (July 13), jointing (August 1), heading (August 16), and grain-filling (September 5). Rice roots together with soil were first placed in a 40-mesh nylon sieve bag and carefully rinsed with running water. Washed roots were then gently wiped dry with filter paper. The main root length was measured by ruler. The fresh weight of root and the upper part of the plant were measured with an electronic balance (AUY220, Shimadzu, Kyoto, Japan), after which the root-shoot ratio was calculated. Root volume was measured by a drainage method (Zhang *et al.* 1999). The total absorption (RTA) and root active absorption area (RAA) of the root system were measured by colorimetry of methylene blue as described by Zhang *et al.* (1999). The root oxidation capacity (ROC) was measured based on the methods of  $\alpha$ -NA oxidation as described by Zhang (1992). Root bleeding rate during different rice growth was determined according to the Hirasawa *et al.* (1983) method at different growth stages. Rice plants were cut 5 cm above the soil level at 6:00 p.m. during the tillering, jointing, heading, and grain-filling stages. Each decapitated stem was firstly covered with absorbent cotton on the top and then with a polyethylene sheet.

Moreover, paper bag was placed on the stems to prevent the stems from night dews and exposure to direct sunlight. At 8:00 a.m. of the next day, the absorbent cotton containing root sap was collected, and the root sap bleeding rate ( $\text{mg h}^{-1} \text{ plant}^{-1}$ ) was calculated according to the increase of cotton weight. The root malondialdehyde content (MDA) was quantified using the method of Stewart and Bewley (1980). The suspension was mixed and heated at  $95\text{ }^{\circ}\text{C}$  for 25 min; the reaction was stopped by quickly placing tubes on ice. The suspension was filtered and the absorbance of the supernatant was read at 532 and 600 nm and MDA content was calculated using the extinction coefficient of  $155\text{ mM}^{-1}\text{ cm}^{-1}$  and the content was expressed as  $\text{nmol g}^{-1}\text{ FW}$ .

During the physiological maturity stage (September 25), the rice roots collected from three pots per treatment were rinsed with tap water and then wiped dry with filter paper. These samples were dried in an oven at  $80\text{ }^{\circ}\text{C}$  until the constant weight and ground into fine powder. The contents of  $\text{K}^{+}$  and  $\text{Na}^{+}$  were subsequently measured with the flame photometer (M410, Sherwood Ltd, Cambridge, UK) after nitric-perchloric acid (1%) digestion (Bastías *et al.* 2004). At maturity, rice plants of each treatment from five pots were harvested by cutting at the soil level to determine the rice yield. Numbers of panicles per hill (NPP), number of spikelets per panicle (SP), Seed setting rate (SSR), 1,000-grain weight (GW) and grain yield per hill (YP) were recorded.

### Statistical Analysis

Descriptive statistics were used to test the mean and standard deviation of the measured parameters. The effects of biochar on the measured parameters were evaluated using one-way ANOVA (analysis of variance). Significant differences among means were detected using the least significant difference (LSD) test at  $p < 0.05$ . The SPSS 18.0 software (IBM, Inc. Chicago, IL, USA) was used for all statistical analysis.

## RESULTS AND DISCUSSION

### Root Morphological Characteristics

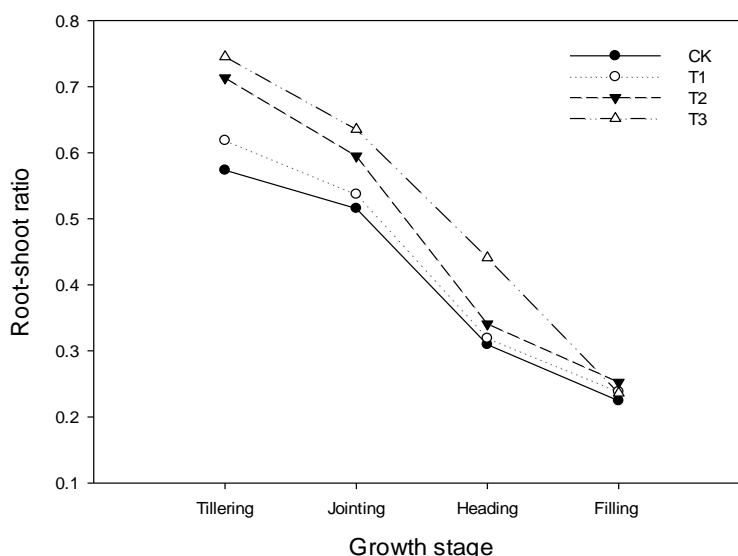
Biochar application positively affected the rice root morphological characteristics under saline-sodic paddy soil (Table 3). The main root length, root volumes and root dry weight were significantly increased by adding biochar at all analyzed growth stages, and the values kept rising with increasing of biochar application. There were significant differences ( $P < 0.05$ ) between biochar treatment and CK during the tillering, jointing, heading, and filling stages. Thus, applying biochar to the saline-sodic paddy soil could significantly improve root morphological index, promote root growth, and increase nutrient uptake of rice.

**Table 3.** Effects of Biochar on Rice Root Morphology in Different Growth Stages

Trait	Treatment	Tillering stage	Jointing stage	Heading stage	Filling stage
Main root length (cm)	CK	10.95 ± 0.17 c	14.50 ± 0.27 b	16.33 ± 0.13 c	16.70 ± 0.27 b
	T1	14.50 ± 0.16 b	16.10 ± 0.19 a	17.85 ± 0.12 b	17.75 ± 0.05 a
	T2	14.75 ± 0.12 b	16.35 ± 0.11 a	20.65 ± 0.31 a	17.25 ± 0.16 a
	T3	16.35 ± 0.24 a	18.50 ± 0.19 a	21.55 ± 0.43 a	18.35 ± 0.39 a
Root volume (cm <sup>3</sup> )	CK	4.10 ± 0.04 c	14.20 ± 0.06 b	35.10 ± 2.22 c	36.50 ± 0.41 c
	T1	7.40 ± 0.09 b	19.60 ± 0.10 a	50.70 ± 4.19 b	41.30 ± 1.03 b
	T2	9.30 ± 0.14 a	21.10 ± 0.27 a	65.50 ± 2.43 b	50.70 ± 2.12 a
	T3	11.50 ± 0.33 a	21.50 ± 0.23 a	95.30 ± 5.17 a	53.50 ± 1.99 a
Root dry weight (g plant <sup>-1</sup> )	CK	0.35 ± 0.01 c	1.25 ± 0.04 c	2.30 ± 0.11 c	2.25 ± 0.13 c
	T1	1.21 ± 0.00 b	2.08 ± 0.03 b	4.37 ± 0.07 b	4.12 ± 0.19 b
	T2	1.31 ± 0.06 a	2.32 ± 0.01 b	5.00 ± 0.18 b	4.27 ± 0.07 b
	T3	1.42 ± 0.05 a	2.70 ± 0.06 a	5.72 ± 0.27 a	4.97 ± 0.13 a

Different letters indicate significantly different values ( $P < 0.05$ ). CK: non-biochar; T1: 15 g biochar per kilogram soil; T2: 30 g biochar per kilogram soil; T3: 45 g biochar per kilogram soil.

The effect of biochar on root-shoot ratio is shown in Fig. 1. The root-shoot ratio of biochar treatments was higher ( $P < 0.05$ ) than that of the non-biochar control during the whole growth stage. The ratio peaked at the tillering stage, with the T3, T2, and T1 treatments increasing the ratio by 7.16%, 13.97%, and 4.5%, respectively. However, the differences between biochar treatments and CK gradually decreased as rice growth progressed, with no significant differences during the later stages.



**Fig. 1.** Effects of biochar on the root-shoot ratio at different growth stage. CK: non-biochar; T1: 15 g biochar per kilogram soil; T2: 30 g biochar per kilogram soil; T3: 45 g biochar per kilogram soil

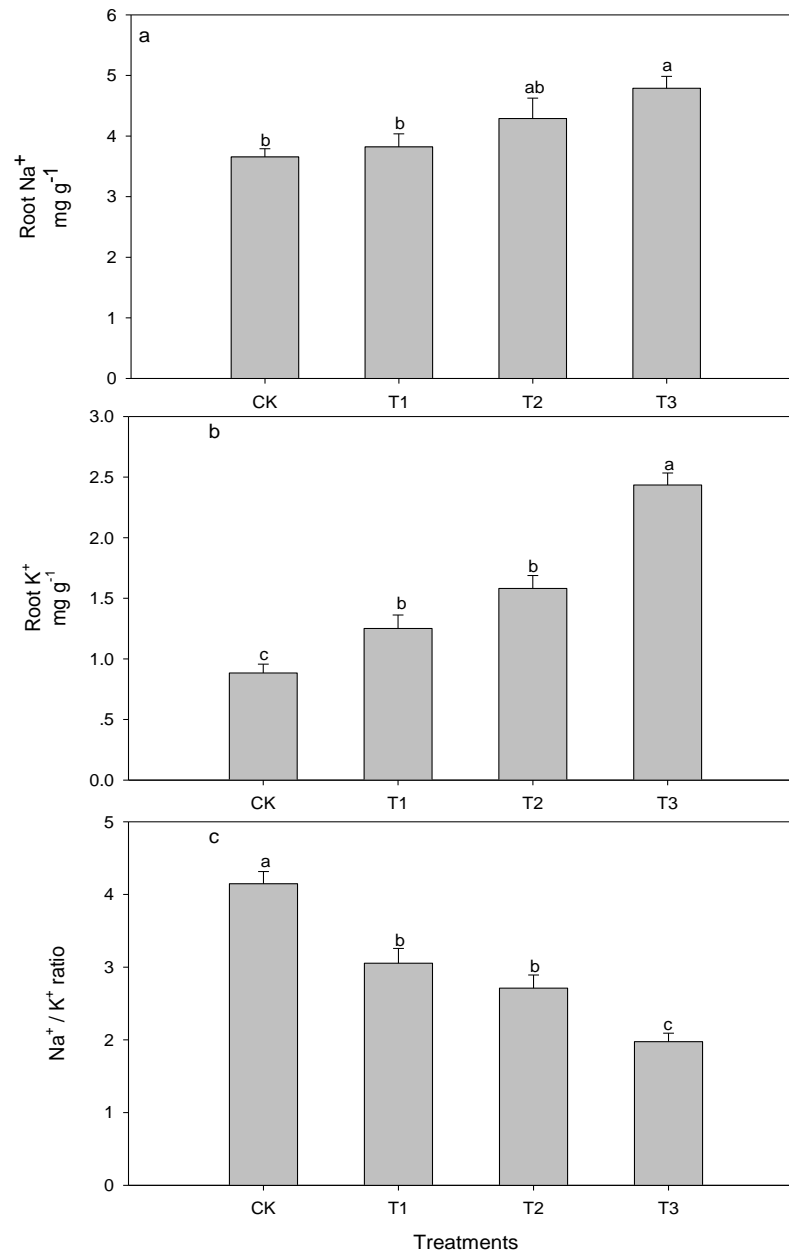
### Root Na<sup>+</sup> and K<sup>+</sup> Accumulation Content and Na<sup>+</sup> / K<sup>+</sup> Ratio

Figure 2 reveals that root Na<sup>+</sup> accumulation content increased compared to CK. However, the significant difference was only observed between T3 and non-biochar treatment. Biochar addition significantly increased K<sup>+</sup> accumulation content ( $p < 0.05$ ) and enhanced with the addition rate of biochar as compared to CK, especially T3 increased by 176%. Compared to non-biochar treatment, biochar application significantly lowered Na<sup>+</sup> / K<sup>+</sup> ratio of root in saline-sodic soil.

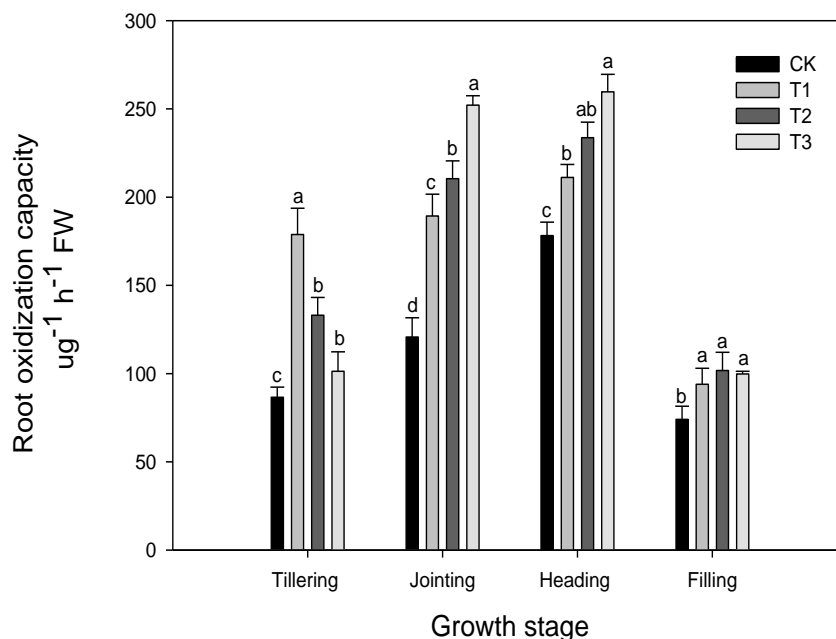
## Root Oxidization Capacity

The effect of biochar on root oxidization capacity of rice is shown in Fig. 3. Compared to CK (non-biochar), all the biochar treatments were more efficient ( $P < 0.05$ ) in enhancing the root oxidization capacity of rice (ROC) at all different growth stages. However, the performance of among biochar treatments was not consistent at different growth stages.

During the tillering stage, T1 was the highest and then decreased with the increase of biochar addition amount. At jointing and heading stages, it was shown as a trend of  $T3 > T2 > T1 > CK$ . But during the grain-filling stage, the highest ROC was observed for the T2 treatment, followed by the T3 and T1.



**Fig. 2.** Accumulation content of Na<sup>+</sup> (a), K<sup>+</sup> (b) and Na<sup>+</sup>/K<sup>+</sup> ratio (c) in rice root as affected by biochar treatment. CK: non-biochar; T1: 15 g biochar per kilogram soil; T2: 30 g biochar per kilogram soil; T3: 45 g biochar per kilogram soil



**Fig. 3.** Effects of biochar on root oxidation capacity (ROC) at different rice growth stages. CK: non-biochar; T1: 15 g biochar per kilogram soil; T2: 30 g biochar per kilogram soil; T3: 45 g biochar per kilogram soil

### Root Total and Active Absorption Areas

Table 4 reveals that biochar had significant effects on root total absorption areas (RTA) and root active absorption areas (RAA) of rice. RTA and RAA at whole growth stages was increased significantly as compared to CK (non-biochar), while no significant difference was found among biochar treatments. RTA was shown as T3 > T2 > T1 > CK at different stages and increasing with the rise of biochar dosage. RAA also had shown a trend of T3 > T2 > T1 > CK at the tillering stage, jointing stage, and heading stage, but showed a trend of T2 > T3 > T1 > CK at the filling stage.

**Table 4.** Effects of Biochar on the Root Total and Active Absorption Areas (M<sup>2</sup>) During Different Growth Stages

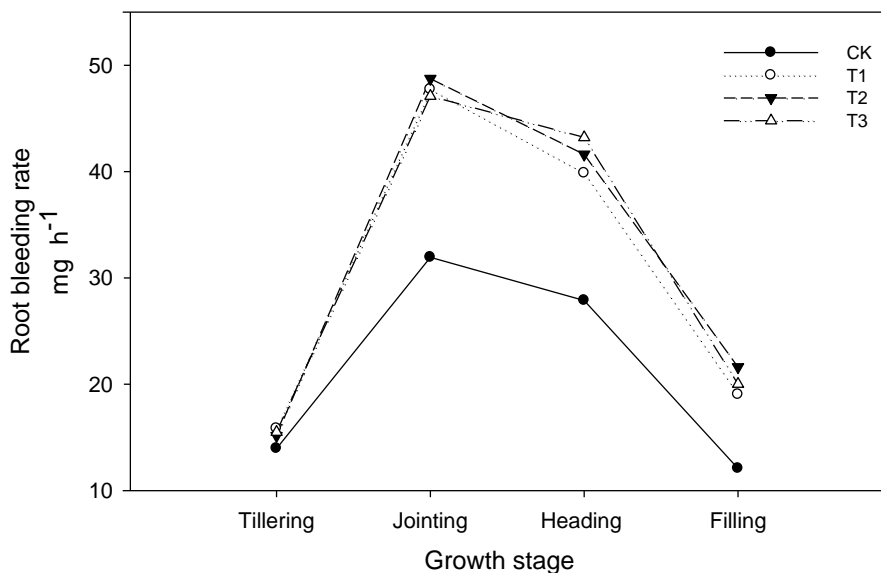
Treatment	RTA			
	Tillering stage	Jointing stage	Heading stage	Filling stage
CK	29.21 ± 1.19 b	56.39 ± 3.32 b	84.66 ± 2.11b	89.17 ± 0.99 b
T1	34.77 ± 2.77 a	72.71 ± 3.89 a	99.27 ± 2.53 a	106.13 ± 3.74 a
T2	37.47 ± 1.89 a	74.81 ± 2.31 a	101.43 ± 1.88 a	110.36 ± 4.10 a
T3	37.94 ± 2.01 a	76.44 ± 3.27 a	103.55 ± 1.49 a	111.01 ± 3.77 a
Treatment	RAA			
	Tillering stage	Jointing stage	Heading stage	Filling stage
CK	16.46 ± 1.39 b	27.84 ± 2.21b	35.42 ± 2.01 b	36.08 ± 0.19 b
T1	22.17 ± 2.17 a	34.35 ± 2.17 a	41.60 ± 2.23 a	43.39 ± 3.27 a
T2	23.57 ± 1.98 a	35.13 ± 2.49 a	43.89 ± 1.18 a	47.89 ± 2.57 a
T3	25.27 ± 2.25 a	37.46 ± 3.76 a	45.34 ± 3.14 a	46.91 ± 3.11 a

Different letters indicate significantly different values ( $P < 0.05$ ). RTA: root total absorption area; RAA: root active absorption area. CK: non-biochar; T1: 15 g biochar per kilogram soil; T2: 30 g biochar per kilogram soil; T3: 45 g biochar per kilogram soil.

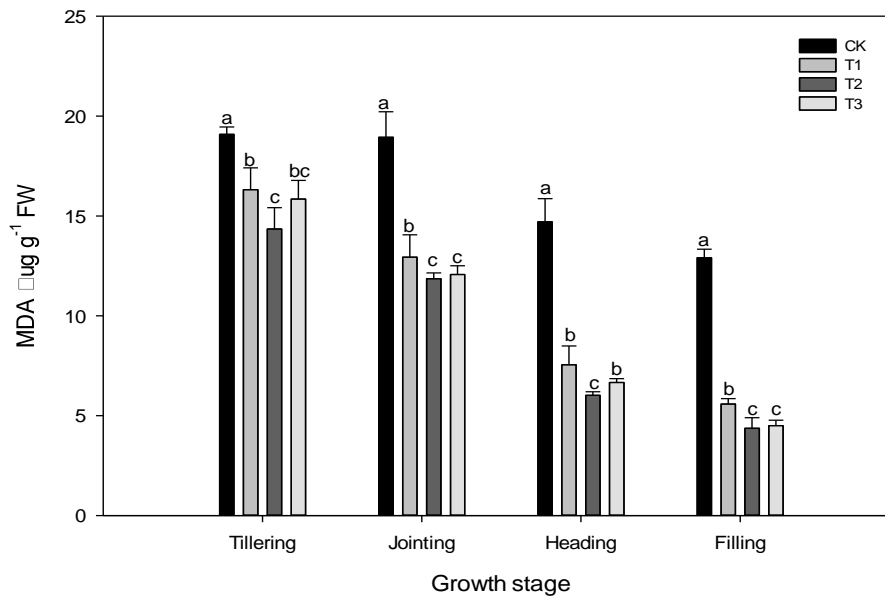


### Root Bleeding Rate

Figure 4 reveals that root bleeding rate was significantly affected by biochar. The root bleeding rate with biochar treatments remarkably increased ( $P < 0.05$ ) compared with CK (non-biochar treatment) at all growth stages; however, no significantly difference was observed among biochar treatments. Root bleeding rate of biochar treatments at tillering, jointing, heading, and filling stage increased by 11.01%, 49.07%, 49.81% and 67.37%, respectively, as compared to non-biochar treatment. The result showed that with the rice growth stage elongation, the difference between biochar treatments and non-biochar treatment increased gradually.



**Fig. 4.** Effects of biochar on root bleeding rate at different growth stages. CK: non-biochar; T1: 15 g biochar per kilogram soil; T2: 30 g biochar per kilogram soil; T3: 45 g biochar per kilogram soil



**Fig. 5.** Effects of biochar on root malondialdehyde content (MDA) at different rice growth stages. CK: non-biochar; T1: 15 g biochar per kilogram soil; T2: 30 g biochar per kilogram soil; T3: 45 g biochar per kilogram soil

### Root Malondialdehyde (MDA) Content

Saline-sodic stress can cause membrane lipid peroxidation of crop cell, and the malondialdehyde is an indicator of membrane damage by reactive oxygen species. The effect of biochar on root MDA content is shown in Fig. 5. Compared to the CK (non-biochar), biochar addition significantly decreased MDA content ( $p < 0.05$ ) during the whole growth period. MDA was shown as CK > T1 > T3 > T2 at different stages, and the significant difference was observed between T2 and CK, T1. MDA content appeared as tillering stage (TS) > jointing stage (JS) > heading stage (HS) > filling stage (FS) among different growth stages.

**Table 5.** Effects of Biochar on the Yield and Yield Components of Rice Plants in Saline-Sodic Paddy Soil

Treatment	NPP	SP	SSR (%)	GW(g)	YP (g•hill <sup>-1</sup> )
CK	9.21 ± 0.11 c	49.08 ± 1.21 a	46.07 ± 1.03 c	21.82 ± 0.12 a	6.11 ± 0.19 c
T1	14.33 ± 0.19 b	50.66 ± 1.04 a	67.09 ± 0.22 b	22.10 ± 0.30 a	11.98 ± 0.20 b
T2	15.83 ± 0.23 b	52.25 ± 2.33 a	72.83 ± 0.79 a	23.33 ± 0.23 a	13.27 ± 0.17 b
T3	17.20 ± 0.17 a	54.04 ± 3.17 a	71.26 ± 0.64 a	23.48 ± 0.31 a	16.60 ± 0.32 a

Different letters indicate significantly different values ( $P < 0.05$ ). CK: non-biochar; T1: 15 g biochar per kilogram soil; T2: 30 g biochar per kilogram soil; T3: 45 g biochar per kilogram soil. NPP: No. of panicles per hill, SP: spikelets per panicle, SSR: seed setting rate, GW: 1000-grain weight, YP: yield per hill

### Yield and Yield Components

The differences of yield and yield components of rice are shown in Table 5. All investigated yield components (NPP, SP, SSR, GW, and YP) were promoted after biochar applied. Compared to CK, the yield and no. of panicle of biochar treatments increased significantly and showed as T3>T2>T1>CK. The difference between biochar treatments and non-biochar treatment (CK) reached a significant level ( $P < 0.05$ ) and T3 was also significantly higher than T2 and T1. Compared to CK, seed setting rate was shown as T2>T3>T1>CK, and the difference between biochar treatment and non-biochar treatment was significant. The spikelets per panicle and 1,000-grain weight were also shown as a trend of T3>T2>T1>CK, but no significant difference between biochar treatments and CK (non-biochar) was observed.

### Effect of Biochar on Root Morphological Characteristics in Heavily Saline-Sodic Paddy Soil

The restrictions of saline-alkali soil stress on crops growth are largely because of ion toxicity, osmotic imbalances, and high pH (Al-Karaki 1997). The root system is a part of the crops that directly suffered from salinity stress (Sun *et al.* 2019). Many studies have shown that under salinity stress, the length, root volume, root superficial area and root weight were significantly decreased and the proportion of roots with different diameters was changed, thus changing the root composition and morphological configuration (Hakim *et al.* 2010; Zhang *et al.* 2011; Van Zelm *et al.* 2020). These effects can severely restrict the uptake of essential nutrients and water by roots, inhibit root respiration, thus limiting the growth of aboveground plant parts and eventually leading to yield losses (Shahram *et al.* 2018). In this study, it was found that under saline-sodic paddy soil, root morphological characteristics such as main root length, root volumes, root dry weight (Tab. 3) and root-shoot ratio (Fig. 1) were significantly improved at all growth stages by biochar application compared with no-biochar. The underlying reason might be the biochar addition to saline-

sodic paddy soils to a large extent counteracted saline-alkali stress by the release of essential macro and micro nutrients such as Ca, K, N, and P in the soil, as indicated in the authors' prior study (Yao *et al.* 2021); simultaneously, as a highly porous material, biochar raised the total porosity, bulk density, and water holding capacity of salt-affected soils and increased the osmotic coefficient (Abrishamkesh *et al.* 2015; Yang 2015; Ran *et al.* 2019; Mehmood *et al.* 2020). Sun *et al.* (2017) found similar results on maize. The development of an enhanced root system was conducive to soil exploration, nutrient transfer, and absorption efficiency, thus contributing to yield increase (Prendergast-Miller *et al.* 2014; Albuquerque *et al.* 2015; Kamau *et al.* 2019; Chew *et al.* 2020).

### **Effect of Biochar on the Ionic Accumulation and Root Physiological Functions in Heavily Saline-Sodic Paddy Soil**

Farhangi-Abriz and Torabian (2018) found that high  $\text{Na}^+$  can decrease  $\text{K}^+$  content in crops under saline condition because of the exchange of  $\text{K}^+$  with  $\text{Na}^+$ . Na-induced K deduction is a proven competitive process in plant roots (Botella *et al.* 1997; Hafsi *et al.* 2007). Excessive  $\text{Na}^+$  not only interferes with K uptake by roots but it also disrupts the metabolic processes of the root cell, causes nutritional disorders, and hinders the absorption of key nutrients, thus finally resulting in yield losses of the crops (Oster *et al.* 1999; Munns 2002; Ran *et al.* 2020). Increasing the  $\text{K}^+ / \text{Na}^+$  ratio by enhancing  $\text{K}^+$  availability can effectively improve the crop growth and yield in salt-affected soils (Chakraborty *et al.* 2016; Almeida *et al.* 2017; Yao *et al.* 2021). The authors' experiments also consistently found that under saline-sodic paddy soil,  $\text{K}^+$  and  $\text{K}^+/\text{Na}^+$  ratio in rice root system was increased significantly (Fig.2), thus improving cell membrane integrity, increasing water potential, and enhancing salinity tolerance and growth of rice (Jin *et al.* 2018; Ran *et al.* 2020). Yue *et al.* (2016) also found that biochar application could accelerate salt leaching, lower soil EC, and provide a good environment for growth and development of plant roots.

Because plants accumulate excessive  $\text{Na}^+$  and other harmful ions, the stress of saline-sodic soil can significantly increase  $\text{O}_2^{\cdot-}$  and  $\text{H}_2\text{O}_2$  concentration and antioxidant activities in plant cells in salted-soils (Shahram *et al.* 2018). It is known that the stronger root oxidation capacity, the greater the ability of rice roots to secrete oxygen. This not only promotes root respiration and nutrients absorption, but it also helps protect the roots from toxic substances (Prendergast-Miller *et al.* 2014; Farhangi-Abriz and Torabian 2018; Liu *et al.* 2020a). In the present experiment, biochar application significantly increased the root oxidation capacity (Fig. 3) of rice at all growth stages under saline-sodic paddy soil was found. This may be because biochar is beneficial to improving rhizosphere environment (Bruun *et al.* 2014; Chew *et al.* 2020) and facilitating root growth (Tab. 3) and root cell viability (Tab. 4).

Zhang *et al.* (2013) demonstrated that biochar application increases rice root physiological activity, bleeding rate, and root activity in the whole growing period under sandy soils used pot experiment. Consistent with these findings, biochar application significantly increased root total absorption areas and root active absorption areas (Tab. 4), root bleeding rate (Fig. 4), and root MDA content at whole growth stages compared to non-biochar treatment under saline-sodic paddy soil. These results indicated that biochar application can enhance the root continuous absorption ability of water and nutrients, increase the transportation efficiency of matter through the roots and retard root senescence under saline-sodic soil. The potential reasons may include the following three aspects. Firstly, biochar application offsets the negative impacts of  $\text{Na}^+$  and cell osmotic stress, since it could decrease the  $\text{Na}^+/\text{K}^+$  ratio (Fig. 2) and increase  $\text{K}^+$  content (Fig. 2) in rice roots

(Zhang *et al.* 2017; Shahram *et al.* 2018; Mehmood *et al.* 2020). Secondly, biochar could improve root zone processes and activity of soil microorganisms through improving the structure of salt-affected soils (Van Zwieten *et al.* 2010; Kolton *et al.* 2016; Zhang *et al.* 2017). Thirdly, biochar application significantly increased root length, root volumes, root dry weight (Tab. 3), root-shoot ratio (Fig. 1), and root oxidation capacity (Fig. 3) that ultimately increased root absorption areas, root bleeding rate, and root vitality.

### Effect of Biochar on Grain Yield in Heavily Saline-Sodic Paddy Soil

Biochar modifies soil physicochemical and biological parameters of salt-affected soils (Lehmann *et al.* 2011; Fletcher *et al.* 2014; Burrell *et al.* 2016; Futa *et al.* 2020), releasing essential nutrients and increasing crop yield (Thomas *et al.* 2013; Drake *et al.* 2016; Wei *et al.* 2020). In addition, it was found in this work that biochar could increase K concentration and decrease  $\text{Na}^+ / \text{K}^+$  ratio of rice root system (Fig. 2) to help offset the negative impacts of saline-sodic stress. Therefore, these factors could enhance root biomass, optimize root morphology (Tab. 3 and Fig. 1) and physiological traits (Fig. 4 and Tab. 4), and increase rice yield (Zhang *et al.* 2013). The results of this study indicated that biochar application significantly increased grain yield (Tab. 5) by improving root morphological and root physiological functions in heavily saline-sodic paddy soil.

## CONCLUSIONS

1. Biochar application had beneficial effects on root length, root volume, root dry weight, root-shoot ratio, root absorption vigor, root oxidization capacity, and root bleeding rate at all growth stages, which is of great benefit to low fertility soils such as saline-sodic paddy soils.
2. Biochar significantly decreased the root  $\text{Na}^+/\text{K}^+$  ratio and MDA content, and it increased  $\text{K}^+$  concentration in rice root after biochar amendment by transient  $\text{Na}^+$  binding due to its high adsorption capacity and by releasing mineral nutrients into the soil solution.
3. The grain yield increase can be attributed to the improvement of root growth, which helped rice to take up more nutrients and water.

## ACKNOWLEDGEMENTS

This study was funded by the National Natural Science Foundation of China (No. 32071951) and Jilin Province Education Department Planning Project (No. JJKH20200340KJ).

## REFERENCES CITED

- Abrishamkesh, S., Gorji, M., Asadi, H., Bagheri-Marand, G. H., and Pourbabae, A. A. (2015). "Effects of rice husk biochar application on the properties of alkaline soil and lentil growth," *Plant Soil Environment* 11, 475-482. DOI: 10.17221/117/2015-PSE
- Akhtar, S. S., Andersen, M. N., and Liu, F. (2015). "Residual effects of biochar on

- improving growth, physiology and yield of wheat under salt stress,” *Agricultural Water Management* 158, 61-68. DOI: 10.1016/j.agwat.2015.04.010
- Albuquerque, J. A., Cabello, M., Avelino, R., Barrón, V., Campillo, M. C. D., and Torrent, J. (2015). “Plant growth responses to biochar amendment of mediterranean soils deficient in iron and phosphorus,” *Journal of Plant Nutrition and Soil Science* 178, 567-575. DOI: 10.1002/jpln.201400653
- Ali, S., Rizwan, M., Qayyum, M. F., Ok, Y. S., Ibrahim, M., Riaz, M., Arif, M. S., Hafeez, F., Al-Wabel, M. I., and Shahzad, A. N. (2017). “Biochar soil amendment on alleviation of drought and salt stress in plants: A critical review,” *Environment Science and Pollution Research* 3, 1-13. DOI: 10.1007/s 11356– 017-8904-x
- Al-Karaki, G. N. (1997). “Barley response to salt stress at varied levels of phosphorus,” *Journal of Plant Nutrition* 20, 1635-1643. DOI: 10.1080/01904169709365362
- Almeida, D. M., Oliveira, M. M., and Saibo, N. J. M. (2017). “Regulation of Na<sup>+</sup> and K<sup>+</sup> homeostasis in plants: Towards improved salt stress tolerance in crop plants,” *Genetics and Molecular Biology* 40(1), 326-345. DOI: 10.1590/1678-4685-gmb-2016-0106
- Bastías, E. I., González-Moro. M. B., and González-Murua, C. (2004). “*Zea mays* L. amylacea from the Lluta Valley (Arica-Chile) tolerates salinity stress when high levels of boron are available,” *Plant Soil* 267, 73-84. DOI: 10.1007/s 11104–005 -4292-y
- Botella, M. A., Martinez, V., Pardines, J., and Cerda, A. (1997). “Salinity induced potassium deficiency in maize plants,” *Journal of Plant Physiology* 150, 200-205. DOI: 10.1016/s0176-1617(97)80203-9
- Bruun, E. W., Petersen, C. T., Hansen, E., Holm, J. K., and Hauggaard-Nielsen, H. (2014). “Biochar amendment to coarse sandy subsoil improves root growth and increases water retention,” *Soil Use and Management* 30, 109-118. DOI: 10.1111/sum.12102
- Burrell, L. D., Zehetner, F., Rampazzo, N., Wimmer, B., and Soja, G. (2016). “Long-term effects of biochar on soil physical properties,” *Geoderma* 282, 96-102. DOI: 10.1016/j.geoderma.2016.07.019
- Chaganti, V. N., and Crohn, D. M. (2015). “Evaluating the relative contribution of physiochemical and biological factors in ameliorating a saline-sodic soil amended with composts and biochar and leached with reclaimed water,” *Geoderma* 259, 45-55. DOI: 10.1016/j.geoderma.2015.05.005
- Chakraborty, K., Bhaduri, D., Meena, H. N., and Kalariya, K. (2016). “External potassium (K<sup>+</sup>) application improves salinity tolerance by promoting Na<sup>+</sup>-exclusion, K<sup>+</sup>-accumulation and osmotic adjustment in contrasting peanut cultivars,” *Plant Physiology and Biochemistry* 103, 143-153. DOI: 10.1016/j.plaphy.2016. 02.039
- Chew, J. K., Zhu, L. L., Nielsen, S., Graber, E., Mitchell, D. R. G., Horvat, J., Mohammed, M., Liu, M. L., Zwieter, L. V., Donne, S., Munroe, P., Taherymoosavi, S., Pace, B., Rawal, A., Hook, J., Marjo, C., Thomas, D. S., Pan, G. X., Li, L. Q., Bian, R. J., McBeath, A. M., Bird, M., Thomas, T., Husson, O., Solaiman, Z., Joseph, S., and Fan, X. R. (2020). “Biochar-based fertilizer: Supercharging root membrane potential and biomass yield of rice,” *Science of the Total Environment* 713, 136431. DOI: 10.1016/j.scitotenv. 2019.13643
- Drake, J. A., Cavagnaro, T. R., Cunningham, S. C., Jackson, W. R., and Patti, A. F. (2016). “Does biochar improve establishment of tree seedlings in saline sodic soils?,” *Land Degradation & Development* 27, 52-59. DOI: 10.1002/ldr.2374

- Elgharably, A. (2011). "Wheat response to combined application of nitrogen and phosphorus in a saline sandy loam soil," *Soil Science and Plant Nutrition* 57, 396-402. DOI: 10.1080/00380768.2011.582588
- Farhangi-Abriz, S., and Torabian, S. (2017). "Antioxidant enzyme and osmotic adjustment changes in bean seedlings as affected by biochar under salt stress," *Ecotoxicology and Environmental Safety* 137, 64-70. DOI: 10.1016/j.ecoenv.2016.11.029
- Farhangi-Abriz, S., and Torabian, S. (2018). "Biochar increased plant growth-promoting hormones and helped to alleviate salt stress in common bean seedlings," *Journal of Plant Growth Regulation* 37, 591-601. DOI: 10.1007/s 00344-017-9756-9
- Fletcher, A. J., Smith, M. A., Heinemeyer, A., Lord, R., Ennis, C. J., Hodgson, E. M., and Farrar, K. (2014). "Production factors controlling the physical characteristics of biochar derived from phytoremediation willow for agricultural applications," *Bioenergy Research*. 7, 371-380. DOI: 10.1007/s12155-013-9380-x
- Futa, B., Oleszczuk, P., Andruszczak, S., Kwiecińska-Poppe, E., and Kraska, P. (2020). "Effect of natural aging of biochar on soil enzymatic activity and physicochemical properties in long-term field experiment," *Agronomy* 10, 449. DOI: 10.3390/agronomy10030449
- Hafsi, C., Lakhdhar, A., Rabhi, M., Debez, A., Abdelly, C., and Ouerghi, Z. (2007). "Interactive effects of salinity and potassium availability on growth, water status, and ionic composition of *Hordeum maritimum*," *Journal of Plant Nutrition and Soil Science* 170, 469-473. DOI: 10.1002/jpln.200625203
- Hakim, M. A., Juraimi, A. S., Begum, M., Hanafi, M. M., Ismail, M. R., and Selamat A. (2010). "Effect of salt stress on germination and early seedling growth of rice (*Oryza sativa* L.)," *African J. Biotechnology* 9, 1911-1918. DOI: 10.5897/AJB09.1526
- Hirasawa, T., Araki, T., Matsuda, E., and Ishihara, K. (1983). "On exudation rate from the base of the leaf blade in rice plants," *Japanese Journal of Crop Science* 52, 574-581. (in Japanese) DOI: 10.1626/jcs.52.574
- IUSS Working Group WRB. (2014). "World Reference Base for soil resources 2014: international soil classification system for naming soils and creating legends for soil maps," *World Soil Resources Reports* 106, FAO: Roma, Italy.
- Jin, F., Ran, C., Anwari, Q., Geng, Y. Q., Guo, L. Y., Li, J. B., Han, D., Zhang, X. Q., Liu, X., and Shao, X. W. (2018). "Effects of biochar on sodium ion accumulation, yield and quality of rice in saline-sodic soil of the west of Songnen plain, northeast China," *Plant, Soil and Environment* 64, 612-618. DOI: 10.17 221/359/2018-PSE
- Kamau, S., Karanja, N. K., Ayuke, F. O., and Lehmann, J.. (2019). "Short-term influence of biochar and fertilizer-biochar blends on soil nutrients, fauna and maize growth," *Biology and Fertility of Soils* 7(55), 661. DOI: 10.1007/s00374-019-0 1381-8
- Kolton, M., Graber, E. R., Tsehansky, L., Elad, Y., and Cytryn, E. (2016). "Biochar-stimulated plant performance is strongly linked to microbial diversity and metabolic potential in the rhizosphere," *New Phytologist* 213, 1393-1404. DOI: 10.1111/nph.14253
- Lehmann, J., Rillig, M. C., Thies, J., Masiello, C. A., Hockaday, W. C., and Crowley, D. (2011). "Biochar effects on soil biota: A review," *Soil Biology & Biochemistry* 43, 1812-1836. DOI: 10.1016/j.soilbio.2011.04.022
- Liu, X. Y., Wang, H. D., Liu, C., Sun, B. B., Zheng, J. F., Bian, R. J., Drosos, M., Zhang, X. H., Li, L. Q., and Pan, G. X. (2020a). "Biochar increases maize yield by promoting root growth in the rainfed region," *Archives of Agronomy and Soil Science*, DOI:

- 10.1080/03650340.2020.1796981
- Liu, D. F., Feng, Z. Y., Zhu, H. D., Yu, L. H., Yang, K. J., Yu, S., Zhang, Y. F., and Guo, W. (2020b). "Effects of corn straw biochar application on soybean growth and alkaline soil properties," *BioResources* 15(1), 1463-1481. DOI: 10.15376/biores.5.1.1463-1481
- Lu, R. K. (2000). *Methods of Soil and Agro-chemical Analysis*, China Agricultural Science and Technology Press, Beijing (in Chinese).
- Mehmood, S., Ahmed, W., Ikram, M., Imtiaz, M., Mahmood, S., Tu, S. X., Chen, D. Y. (2020). "Chitosan modified biochar increases soybean (*Glycine max L.*) resistance to salt-stress by augmenting root morphology, antioxidant defense mechanisms and the expression of stress-responsive genes," *Plants* 9, 1173. DOI: 10.3390/plants9091173
- Munns, R. (2002). "Comparative physiology of salt and water stress," *Plant, Cell and Environment* 25, 239-250. DOI: 10.1046/j.0016-8025.2001.00808.x
- Munns, R., and Tester, M. (2008). "Mechanisms of salinity tolerance," *Annual Review of Plant Biology* 59, 651-681. DOI: 10.1146/annurev.arplant.59.032607.092911
- Oster, J. D., Shainberg, I., and Abrol, I. P. (1999). "Reclamation of salt affected soils," In: *Agricultural Drainage*, R.W. Skaggs and J. van Schilfhaarde (Eds.), ASA-CSSA-SSSA, Madison, WI, USA, pp. 659-691.
- Prendergast-Miller, M. T., Duvall, M., and Sohi, S. P. (2014). "Biochar-root interactions are mediated by biochar nutrient content and impacts on soil nutrient availability," *European Journal of Soil Science* 65, 173-185. DOI: 10.1111/ejss.12079
- Qadir, M., and Oster, J. D. (2002). "Vegetative bioremediation of calcareous sodic soils: history, mechanisms, and evaluation," *Irrigation Science* 21, 91-101. DOI: 10.1007/s00271-001-0055-6
- Ran, C., Gulaqa, A., Zhu, J., Wang, X. W., Zhang, S. Q., Geng, Y. Q., Guo, L. Y., Jin, F., and Shao, X. W. (2020). "Benefits of biochar for improving ion contents, cell membrane permeability, leaf water status and yield of rice under saline-sodic paddy field condition," *J. Plant Growth Reg.* 39, 370-377. DOI: 10.1007/s 00344-019 -09988-9
- Rengasamy, P. (2010). "Soil processes affecting crop production in salt-affected soils," *Functional Plant Biology* 37, 613-620. DOI: 10.1021/ac970256w
- Shahram, T., Salar, F. A., and Judith, R. (2018). "Biochar and lignite affect H<sup>+</sup> - ATPase and H<sup>+</sup> -PPase activities in root tonoplast and nutrient contents of mung bean under salt stress," *Plant Physiology and Biochemistry* 129, 141-149. DOI: 10.1016/j.plaphy.2018.05.030
- Stewart, R. C., and Bewley, J. D. (1980). "Lipid peroxidation associated with accelerated aging of soybean axes," *Plant Physiol.* 65(2), 245-248. DOI: 10.2307/4266117
- Suarez, D. L. (2001). "Sodic soil reclamation: Modeling and field study," *Australian Journal of Soil Research* 39, 1225-1246. DOI: 10.1071/SR00094
- Sun, C. X., Chen, X., Cao, M. M., Li, M. Q., and Zhang, Y. L. (2017). "Growth and metabolic responses of maize roots to straw biochar application at different rates," *Plant Soil* 416, 487-502. DOI: 10.1007/s11104-017-3229-6
- Sun, G. F., Zhu, Y., Ye, M., Yang, J. Z., Qu, Z. Y., Mao, W., and Wu, J. W. (2019). "Development and application of long-term root zone salt balance model for predicting soil salinity in arid shallow water table area," *Agricultural Water Management* 213, 486-498. DOI: 10.1016/j.agwat.2018.10.043
- Thomas, S. C., Frye, S., Gale, N., Garmon, M., Launchbury, R., Machado, N., Melamed, S., Murray, J., Petroff, A., and Winsborough, C. (2013). "Biochar mitigates negative effects of salt additions on two herbaceous plant species," *Journal of Environmental*

- Management* 129, 62-68. DOI: 10.1016/j.jenvman.2013.05.057
- van Zelm, E., Zhang, Y., Testerink, C. (2020). "Salt tolerance mechanisms of plants," *Annual Review of Plant Biology* 71, 24.1-24.31. DOI: 10.1146/annurev-arplant-050718-100005
- Van Zwieten, L., Kimber, S., Morris, S., Chan, K. Y., Downie, A., Rust, J., Joseph, S., and Cowie, A. (2010). "Effects of biochar from slow pyrolysis of papermill waste on agronomic performance and soil fertility," *Plant Soil* 327, 235-246. DOI: 10.1007/s11104-009-0050-x
- Wei, W. L., Zhang, S. R., Wu, L. P., Cui, D. J., and Ding, X. D. (2020). "Biochar and phosphorus fertilization improved soil quality and inorganic phosphorus fractions in saline-alkaline soils," *Archives of Agronomy and Soil Science* DOI: 10.1080/03650340.2020.1784879
- Xiao, Q., Zhu, L. X., Zhang, H. P., Li, X. Y., Shen, Y. F., and Li, S. Q. (2016). "Soil amendment with biochar increases maize yields in a semi-arid region by improving soil quality and root growth," *Crop & Pasture Science* 6(67), 495-507. DOI: 10.1071/cp15351
- Yang, F., Li, X. Q., Xing, Y., Cheng, H. G., Zhang, L. K., He, Y. Y., and Wang, B. (2014). "Effect of biochar amendment on nitrogen leaching in saline soil," *Journal of Agro-Environment Science* 33, 972-977 (in Chinese).
- Yang, Y. L. (2015). "Effects of biochar on physical and chemical properties of the saline-sodic Soil," *University of Chinese Academy of Sciences, Beijing. (In Chinese)*
- Yao, T. X., Zhang, W. T., Gulaqa, A., Cui, Y. F., Zhou, Y. M., Weng, W. A., Wang, X., and Jin, F. (2021). "Effects of peanut shell biochar on soil nutrients, soil enzyme activity, and rice yield in heavily saline-sodic paddy field," *Journal of Soil Science and Plant Nutrition* 21, 655-664. DOI: 10.1007/s42729-020-00390-z
- Yue, Y., Guo, W. N., Lin, Q. M., Li, G. T., and Zhao, X. R. (2016). "Improving salt leaching in a simulated saline soil column by three biochars derived from rice straw (*Oryza sativa* L.), sunflower straw (*Helianthus annuus*), and cow manure," *Journal of Soil and Water Conservation* 71, 467-475. DOI: 10.2489/jswc.71.6.467
- Zhang, C. S., Lin, Y., Tian, X. Y., Xu, Q., Chen, Z. H., and Lin, W. (2017). "Tobacco bacterial wilt suppression with biochar soil addition associates to improved soil physiochemical properties and increased rhizosphere bacteria abundance," *Applied Soil Ecology* 112, 90-96. DOI: 10.1016/j.apsoil.2016.12.005
- Zhang, J. Q., Pan, F. Y., Dou, J. H., Chen, P., Zhou, Q., and Zhou, R. Y. (2011). "Superiority of resistance in the root growth of kenaf hybrid under salt stress," *Journal of China Agricultural University* 16, 6-12 (in Chinese).
- Zhang, W. M., Meng, J., Wang, J. Y., Fan, S. X., and Chen, W. F. (2013). "Effect of biochar on root morphological and physiological characteristics and yield in rice," *Acta Agronomica Sinica* 39, 1445-1451 (in Chinese).
- Zhang, X. Z. (1992). *Research Methods of Crop Physiology*, Agriculture Press, Beijing. (in Chinese)
- Zhang, X. Z., Chen, F. Y., and Wang, R. F. (1999). *Plant Physiology Experimental Techniques*, Liaoning Science and Technology Publishing House, Shenyang, China. (in Chinese)

Article submitted: October 21, 2021; Peer review completed: December 26, 2021;  
Revised version received and accepted: December 28, 2021; Published: January 5, 2022.  
DOI: 10.15376/biores.17.1.1241-1256