# Biochar amendment promotes limited organic carbon increase in saline-sodic soil of the Songnen Plain, Northeast China

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#### Abstract

To what extend that biochar addition promotes organic carbon increase in saline-sodic soils, however, remains poorly understood. Here, we evaluated soil organic carbon (SOC) contents change before and after biochar addition, and deciphered which driving factor or process govern SOC change with biochar application. There was a limited increase in SOC, about by  $+1.16\%^{-}+12.8\%$ , even biochar was applied at the rate of 10% of bulk soil weight. However, soil dissolved organic carbon (DOC) increased significantly by up to 67%. About half SOC was stored in small macroaggregates (250-2000  $\mu$ m, CPOC), and SOC in silt and clay-sized particles (<53  $\mu$ m) decreased obviously with biochar addition. Microorganism biomass, represented by phospholipid fatty acid, increased with biochar amendment, of which actinomycetes, fungi, protozoon, and bacteria with straight-chain saturated fatty acids (OB) increased remarkably. DOC was governed by ACT and soil N:P ratio, while SOC mostly depended on CPOC. Biochar addition aggravated nitrogen limitation in saline-sodic soils, and the roles of microorganisms on regulating SOC greatly depended on nitrogen bioavailability. Biochar amendment had greatly changed interactions between environmental factors and SOC in saline-sodic soils. The effects of nutrients on soil carbon changed to strongly negative from strongly positive after and before biochar addition, meanwhile, aggregation was the only factor with positive effects on soil carbon change. How to mitigate nutrient limitation and improve soil aggregation process should be considered in priority when biochar was used to increase SOC in saline-sodic soils.

## Introduction

Biochar amendment into soils has been suggested as an effective means to abate global warming by storing the carbon rich but decomposition resistant biomass materials in soils, while simultaneously bettering soil properties and increasing plant biomass yields (Christian, 2001; Farji-Brener and Ghermandi, 2000). Biochar technology could potentially reduce about 1.8 Pg CO<sub>2</sub>-C yearly, equal to 12% of current anthropogenic CO<sub>2</sub>-C emissions (Christian, 2001). It was estimated that biochar application would reduce 3–4 folds of current carbon loss from the soil pool (Zhao et al., 2015). Biochar-amended soil could sequester C over a long time by physical stabilization (Novak et al., 2009; Yin et al., 2014). The mean residence time of biochar is estimated to about 2000 years, and the half-life is about 1400 years. Biochar application also has positive effects up to 30% on biomass production with the large-volume application of BC ( $30^{-}60t/ha$ ), and this would yield more plant-derived biomass input into soils (Yin et al., 2014). However, carbon mineralization usually was stimulated or suppressed by biochar through positive or negative priming effects and varied significantly and separately (Prommer et al., 2014; Sui et al., 2016). This might offset the benefit of increasing SOC by biochar addition in short-term time. It was believed that credible data are needed from varied field experiments and soil types while using biochar as a soil amendment may be a potentially useful option to mitigate climate change. Saline-sodic soils cover 3.1% of the global land area, and world soils that are currently saline have lost an average of 3.47 t SOC/ha since they become saline (Yang et al., 2018). Improvement of saline-sodic soil is increasingly undertaken as a means of reclaiming otherwise unproductive agricultural land, and biochar application is a potential choice for fertilizing saline-sodic soils for replantation (Munda et al., 2018). Salinesodic soils are flocculated with high soluble salts and exchangeable Na<sup>+</sup> and would become disperse when pH is higher than 8.5. Organic matter contents in saline-sodic soils are usually low due to poor plant growth, which is restricted by poor aeration, compaction, and lower nutrient bioavailability (Sun et al., 2016). Since biochar materials are charcoal-like, high porous, fine-grained, and have a large surface area, its incorporation application to soils has attracted considerable attentions as an effective and economic soil amendment for improving soil physicochemical properties, enhancing plant biomass and increasing soil organic carbon pool. Biochar has been successfully used to reduce the nutrient deficiency and salt stress (Sun et al., 2016), reclaim degraded soils (Sun et al., 2016), facilitate plant growth (Brodowski et al., 2005). and suppress SOC mineralization (Lin et al., 2015). However, the effects of biochar addition on SOC pool were usually with inconsistent results depending on the nature of soil and biochar, soil types, and incubation time. Besides as a direct carbon source that can increase SOC pool, biochar addition would act as nutrient sources such as nitrogen and phosphorus, which facilitate plant growth and increase the SOC pool indirectly. The large adsorption capacity of biochar could also preserve organic carbon in the pores that prevent them from decomposition by microorganisms. However, most studies about biochar amendment were carried out on nonsalt-affected soils, and knowledge about the effects of biochar application on carbon dynamics in saline-sodic soils is still scant and not well understood, which need further evaluation (Sollins et al., 1996).

As one of the largest saline-sodic soil areas in the world, the salt-affected areas were estimated to  $3.84 \times 10^6$  ha in the Songnen Plain of northeast China. Saline-sodic soils here have high montmorillonite clay and sodium bicarbonate and very low SOC content, and carbon sequestration rates were <60 gC/m<sup>2</sup>/yr in the Momoge wetland site (Sollins et al., 1996). How to improve soil physicochemical properties, fertilizer saline-sodic soil for more biomass yield, and to increase soil carbon pool for mitigating rising atmospheric CO<sub>2</sub> is one the most important environmental spot here. The objectives of the present work are: (1) to compare impacts of biochar addition on soil organic carbon pools based on incubation experiments in the field, and (2) to reveal potential factors that govern SOC contents before and after biochar addition. Based on these two aims, it is hoped to manageably provide carbon farming solutions to the global climate and satisfying food demand using biochar technology.

## 2. Methods and Materials

## 2.1 Soil Sampling for incubation experiments

Saline-sodic soils for incubation experiments were collected from a degraded wetland on 4 April 2018 in the Momoge region, Jilin Province, China. The predominant vegetation cover was *Leymus chinensis (Trin.)* Tzvel. with a total vegetative cover of less than 10%. The climatic averages for the year are 4.4 and a rainfall of 392 mm/y. Surface soils within 40 cm depth were collected and brought back to the laboratory. Soils were well mixed and passed through a 5 mm sieve to remove stones before incubation experiments.

#### 2.2 Incubation experiments design

To investigate the effects of biochar additions on soil organic carbon change, soil microcosms were designed and constructed using 1mx1mx1m polypropylene boxes. Biochar materials were prepared from rice straw at 550 in anaerobic conditions and contained 422.6 g/kg of carbon, 8.4 g/kg of nitrogen, 2.2 g/kg of phosphorus, and pH of the biochar was 8.34.

Incubation experiments were carried out in the field. The application rates of biochar were set to 0.5% (T0.5), 1% (T1), 2% (T2), 5% (T5), and 10% (T10) by biochar weight to soils, and each treatment was performed with three replications. Soil alone was the control treatment (CK). Soil and biochar materials were fully mixed before they were put into the microcosm box. The soil depth was set to 50 cm. T0.5, T1, and T2 treatment were categorized into the lower level of additions (LK), while T5 and T10 were at a higher level (HK) (Yoo and Kang, 2012).

Incubation boxes were placed in the field without extra water addition but natural precipitation, aiming to simulate natural soil water conditions. Experiments started in May 2018, and finished in November 2018, spanning a growing season in Northeast China. Soil samples were collected monthly for chemical analysis.

## 2.3 Phospholipid fatty acid (PLFA) analysis

PLFA extraction and analysis were performed according to the method described by (Zhang, et al., 2012a). In brief, fresh soils were freeze-dried and extracted with a chloroform-methanol-citrate buffer mixture (1:2:0.8). The phospholipids were separated from other lipids on a silicic acid column. Phospholipid phosphate concentration was determined using the spectrometric method. Phospholipids were subjected to a mild-alkali methanolysis, and the resulting fatty acid methyl esters were separated by gas chromatography with a flame ionization detector (Agilent 6890N). The carrier gas was helium, and the temperature increased to 260 from 170 at a rate of 5/min. The inner standard, a mixture of 37 fatty acid methyl ester (FAME), was used to identify and quantify the response of individual fatty acids (Steinbeiss et al., 2009). The PLFA makers used for taxonomic microbial groups were shown in Table 1.

## 2.4 Separation of different aggregation fractions

Soil samples were air-dried for separating different aggregation fractions using a nest of 4 sieves having diameters of 2000, 250, and  $53\mu$  m (Zhang et al., 2012b). In brief, about 250g dry soil samples were placed on the uppermost of the nest and had plane-rotary shaken mechanically for 20min. The microsieve size ( $<250 \ \mu$  m) was further sieved by hand. Mechanism sieving was done 3 times. Finally, three aggregation fractions were got, which were small macroaggregates part (250-2000  $\mu$  m, CPOC), macroaggregates fractions (53-250 $\mu$  m, FPOC), and silt and clay-sized particles ( $<53\mu$  m, MOC). Particles larger than 2000  $\mu$  m were not got in the present work. Weight and SOC contents of every aggregation fraction were determined for calculating proportions of carbon storage.

#### 2.5 Chemical analysis

Total soil organic carbon (SOC) and nitrogen (TN) were determined using an elemental analyzer after carbonate was removed by 1 N HCl solution (Elementar Vario Microcude, Hesse, Germany). Total phosphorus contents (TP) in soils were measured by the ammonium molybdate-ascorbic acid method. Soil samples were dried in an aluminum box to a constant weight, and soil moisture contents (SWC) and bulk density (BW) were calculated by weighting mass loss before and after soil oven at 105 for 8 h.

Dissolved organic carbon (DOC) was extracted by mixing 5.00 g soil with 30.0 mL deionized water in Erlenmeyer flasks. After shaking for 30 min, the mixtures were centrifuged and filtered through a  $0.45\mu$  m filter. Filtrates were analyzed for total organic carbon using a TOC-VCPH analyzer (Gangdong, Tianjin, China).

#### 2.6 Statistical analysis

All statistical analyses were performed by R software. Pearson correlation analysis was applied to explore relationships between SOC, DOC, and environmental factors, including pH, SWC, BW, nutrients, and microorganisms. Analysis of variance (ANOVA) is used to compare differences among different treatments. Principle component and multiple linear regression analyses using the stepwise regression method were carried out to decipher potential links and predominant factors that affected SOC and DOC change. Considering environment factors were closely and intercorrelated, a partial least square path model (plspm) was used to explore and visualize effects of which ecological process or components on SOC and DOC before and after biochar amendment (Brown and Human, 1997). The plspm R package can be download at https://cran.r-project.org/src/contrib/Archive/plspm/.

#### 3. Results

## 3.1 Soil physical-chemical properties changes with biochar amendment

Overall, the trend of the results illustrated that BW, SWC, and TN decreased with biochar addition, while

pH, TP, SOC, and DOC contents showed a noticeable or small increas. There was a steady increase, about 46%, for TP from 0.24g/kg in CK to 0.35g/kg in T10. A significant drop of TN contents, about 10%, was observed with biochar addition. A small change was observed when SOC contents increased from 12.06g/kg in CK to 13.60g/kg in T10. Despite the small change of DOC contents between CK and LK, it increased rapidly in HK treatment, and was about 25% and 67% in T5 and T10 more than that in CK, respectively. As expected, an increment in soil pH was observed but within a small range, from 8.22 in CK to 8.44 in T10 treatment (Table 2).

#### 3.2 SOC contents in different aggregate fractions

Over the whole, FPOC contributed about 41.6%<sup>49.7</sup>% of total SOC in soils. It was observed that CPOC rise irregularly but slowly from 27.9% in CK to 36.7% in T10 as biochar addition increased (Fig.1). MOC showed a noticeable decrease trend with biochar addition.

#### 3.3 Soil microorganism community change with biochar amendment

ANOVA analysis indicated that total PLFAs did not vary significantly among CK, LK, and HK treatments. PLFAs contents in CK were nearly equal to that in LK and increased to 320 nmol/g in HK treatment.

Biochar addition had great impacts on microorganism species identified by PLFA markers. It was obvious that biomass of BAC and AMF change little and even slightly reduce under LK, but increased greatly by 21% and 24% compared with that in CK treatment though not at a significant level. The increase trends were more obvious for ACT, FUN, PRO, and OB, which increased by 11.5%, 44.1%, 24%, and 108% in HK treatment than those in CK treatment, respectively. The difference of FUN between in HK and CK was at a statistically significant level (Table 3).

#### 3.4 Correlation analysis

Significant and positive correlations were observed for DOC vs. pH, TP, MOC, PLFA, BAC, ACT, PRO, and OB, while the opposite correlations were found for DOC vs. BW, C:N, C:P, N:P, and COPC. For SOC, positive correlations were found with BW, C:N, C:P, and CPOC, while negative correlations were observed with SWC, FOPC, MOC, PLFA, PRO, and OB to a significant level (Fig.2).

#### 4. Discussion

#### 4.1 Effects of biochar amendment on change of SOC and DOC

Biochar could reduce soil carbon loss by lowering greenhouse gas emissions, enhancing productivity, and stabilizing organic matter. However, the impacts of biochar on soil carbon dynamics on longevity and magnitude varied widely from weeks to serval years (MacKenzie and Quideau, 2010). Biochar amendment has positive, neutral, or negative effects on SOC and DOC (Liu et al., 2016). SOC contents are usually stimulated by biochar amendment within a great change range, from a few percents to several folds, while DOC contents increased or reduced varied in different researches (Table 4). For instance, Smebye's work (2016) indicated that DOC contents increased by 2775% when biochar was added into arable soils at the rate of 10%. DOC was also reduced by -5.59%~26.67% when biochar was applied into crop field (Yang et al., 2018). In contrast to results seen in arable soils, magnitude of SOC contents changes in the present work were relatively smaller compared with significant SOC increase in sugarcane, paddy field, and other agriculture fields (Table 4). SOC contents only increased within a little range, from 1.16% to 12% even biochar was added at the rate of 10%. A remarkable increase of DOC was observed, from no effect to 66.7%, and higher than that in arable soils. It indicated that driving factors affecting SOC dynamics in saline-sodic soils differed greatly those in crop soils, which might due to differences of nutrient limitation, microorganisms, and unique soil pH conditions (Zimmerman et al., 2011).

Biochar addition had fewer facilitation effects on SOC increase in saline-sodic soils than that in agricultural and coastal saline soils, and the effects varied with biochar addition. SOC contents in CK treatment had no difference with LK treatment but differed significantly with HK treatment, but only 12.8% increase was observed, which was due to serious nutrient limitation to plant growth and microorganism activities in saline-sodic soils. Besides as direct carbon source into soil carbon pool, biochar addition improved soil physicochemical properties, facilitated plant growth, and yielded more litter biomass, which introduced more carbon to SOC pools. Principal components analysis indicated that carbon and nitrogen had positive loadings on the first principal component (PC1, 38.71% of total variance), and PLFA had positive loadings on the second principal component (PC2, 17.03% of total variances) (Fig.3). This meant that nutrient limitation and microorganism activities were the predominant factors controlling SOC and DOC contents. The dependence of CPOC on SOC is presented by the neighboring location of SOC and CPOC in Fig.3. The increasing contribution of CPOC to SOC might be caused by direct biochar addition.

Proportions of MOC to SOC decreased with biochar addition, which implied that SOC decomposed mainly comes from MOC. MOC was closely related to OB in the present work. OB was as indicators of physiological or nutritional stress in bacterial communities and lower proportions meant lower stress (Bossio et al., 1998). Proportions of OB to PLFA increased to 3.64% from 2.95% with biochar addition, and this implied that bacteria face more resource stress and nutrient limitation after biochar addition, which was confirmed by negative correlations between OB and C:N and C:P (Fig.2). Bioavailable nutrient input by biochar addition, specifically total P increment, stimulated microorganism growth like FUN and AFM, this was in good agreement with previous results (Liu et al., 2018). Correlations analysis confirmed that PLFA, BAC, FUN, and AMF all significantly and positively related to TP. FUN and AMF biomass increased obviously with biochar, which would compete with bacteria for space and resources. Biochar is the solid material produced from the thermochemical conversion of biomass under oxygen limitation and is dominantly composed of condensed aromatic C (Liu et al., 2018), which is not bioavailable for bacteria. Particles less than 53  $\mu$  m contain an abundance of polysaccharides, proteins, and lipids, which composed 41.7%, 4.2%, and 11.1% of MOC, respectively (Grandy and Neff, 2008). These compounds could be as alternative carbon and nutrient sources for bacteria and consumed easily.

## 4.2 The predominant factor affecting SOC and DOC

SOC and DOC contents were governed by multiple environmental factors, and the multiple linear regression analyses could identify the most important one. ACT biomass and N:P ratios were the common factors that govern DOC contents and the proportions of DOC to SOC (RC) (Table 5). ACT species, as pioneers on nitrogen-poor sites, have a predilection for barren soils and can tolerate environmental stress such as drought, high salinity, and extreme pH. Biochar addition introduced soil pores to increase (low BW), drought (low SWC), and high pH, which were suitable for ACT growth but meanwhile restricted other microorganisms. Actinomycetes are effective in decomposing C compounds with poor nutrient and they are usually booming when N is limiting in soil (MacKenzie and Quideau, 2010). DOC is expected to be derived from ACT excretion or products of refractory SOC degraded by ACT and other microorganisms, and this was supported by the positive Beta coefficients of ACT in regression models, which was 0.635.

N:P ratios were the common negative factor predominant DOC and RC in soils, which suggested the high nitrogen limitation presence in saline-sodic soils. Soil C:P and N:P ratios decreased while N:P ratios increased with biochar addition, and this confirmed rising N limitation to microorganisms after biochar addition. In the regression models, the Beta coefficients of N:P ratios to DOC and RC were both negative values, which matched well with the diagonal location of DOC, PLFA, and TN and N:P (Fig.3). Globally, there is a Redfield-like atomic C:N:P ratio, 60:7:1, for the soil microbial community (Lehmann et al., 2011). Nitrogen limitation to microorganisms in saline-sodic soils was aggravated after biochar addition, which the well-constrained N:P ratios reduced to 5.3 in HK, 7.1 in LK from 8.3 in CK treatment on average.

Only the CPOC was remained in the regression model for predicting SOC contents, and this matched well with positive loading of CPOC on SOC in Fig.3. CPOC could explain 73% of SOC change as the Beta coefficient showed (Table 5). Considering there was no aggregate that larger than 2000  $\mu$  m was separated and biochar was applied as fine powders into soils, it was guessed that CPOC increase was due to biochar powder addition directly. Biochar interacts with minerals in soils and forms an organic-inorganic complex, which resulted in the protection of the enclosed biochar carbon against further decomposition in soils (Brodowski et al., 2005). Besides, SWC and BW decrease implied soil pores increase, which accelerates

 $Fe^{3+}$  and  $Al^{3+}$  deposition in biochar surface. This reduces the microbial accessibility to biochar and protects biochar-derived C from decomposition (Sollins et al., 1996). However, this hypothesis needs more evidence or parameters, such as black carbon biomarker, to confirm links between carbon in CPOC aggregates and biochar carbon.

#### 4.3 How biochar amendment changes the soil carbon pool?

Change of soil carbon pool are usually results of multiple combined factor blocks including soil physical condition, nutrient availability, microorganism activity, and aggregation processes. Attempting to apportion the impacts of their interactive effects on SOC is vital to evaluate the benefit of biochar application on soil properties and fertility. The partial least squares path model (plspm) could provide visual structural equation modeling that studying complex multivariate relationships among observed and latent variables. In the present work, five blocks were established aiming to reveal which blocks had great impacts on carbon change in saline-sodic soils before and after biochar addition.

The Phy block contained SWC, BW and pH variables, the Nut block contained TN, TP, C:N, C:P and N:P variables, the agg block contained CPOC, FPOC, and MOC variables, the carbon block contained DOC and SOC variables, and the mic block contained PLFA, BAC, ACT, FUN, AMF, PRO and OB variables. The Phy, nut, agg, carbon, and mic blocks represent information of soil basic physical-chemical properties, nutrient availability, aggregation process, carbon dynamics, and microorganism communities, respectively.

The plspm models indicated that the biochar amendment had greatly changed interactions among these five blocks. In CK treatment, Phy, nut, mic, and agg all had varied but positive effects on carbon, of which nut had the largest effects on carbon while mic had the smallest. After biochar was added, agg was the only block that had positive effects while other blocks had negative effects on carbon, of which nut still had the largest values in absolute (Fig.4).

The effects of microorganisms on carbon dynamic changed to weakly negative in LK+HK (-0.0768) from weakly positive in CK treatment (0.0043). It was concluded that biochar addition triggered the negative priming effects of microorganisms on the soil carbon pool, which coincided with the previous report (Zimmerman et al., 2011). However, no significant effects on carbon from mic were observed as expected in other work (Prayogo et al., 2014). It confirmed that the roles of microorganisms in regulating carbon cycles in saline-sodic soils greatly depended on nutrient limitation, especially nitrogen bioavailability. Biochar initially promoted microorganism biomass via nutrient input by biochar, which was proved by rising  $CO_2$  production greatly over the short term in arable soils (Prommer et al., 2014). However, microorganisms would utilize SOC previously associated with clay minerals as nitrogen or carbon sources when extra nitrogen from biochar was exhausted. This was confirmed by the great change of effects of nut on carbon, which changed to -0.6284 in LK+HK from 0.9684 in CK treatment. Nutrients were the primary driver affect organic carbon pool in saline-sodic soils,

Meanwhile, the effects of agg on carbon changed from 0.0820 in CK, the weakly positive, to 0.3478 in LK+HK, which implied the only positive effects. Soil organic carbon pool would benefit from aggregation change caused by biochar addition, and this was in good agreement with the results of PCA and the regression analysis. However, as mentioned above, SOC increase after biochar addition might come from biochar materials but not the native soil organic matter, and effects of biochar on native SOC preserve should be studied over the long-term timescale (Liu et al., 2018).

#### 5. Conclusions

Our study demonstrated that the addition of a rice straw-derived biochar had limited improvement effects on organic carbon in saline-sodic soils, of which SOC only increased about by +1.16%<sup>~</sup>+12.8% while DOC increased significantly by up to 67%. Biochar amendment facilitated FUN and ACT biomass, and aggravated nitrogen limitation on microorganisms. Bacterial might be forced to utilize SOC associated with minerals. ACT and N:P were the predominant factors governing soil DOC contents, while CPOC accounted for most SOC changes before and after biochar addition. The plspm models implied that mitigating nutrient limitation and improving soil aggregation process should be considered in priority when biochar was used to remediation saline-sodic soils.

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 Table 1 Characteristic of fatty acids of microbial functional groups

Taxonomic group	Specific PLFA markers
Bacteria (BAC)	i15: 0 \ a15: 0 \ i15: 1 ω9c \ i16: 0 \ i17: 0 \ a17:
Actinomycetes (ACT)	16: 0 10-methyl $\sim$ 17: 1 $\omega$ 7c 10-methyl $\sim$ 17: 0 10-
Fungi (Fun)	$18:1\omega9c < 18:2\omega6,9 < 18:3\omega6c$
Arbuscularmycorrhizal fungi (AMF)	$16:1\omega 5c$
Protozoon (Pro)	$20:2\omega6 \le 20:3\omega6 \le 20:4\omega6$
Other bacteria identified with straight-chain saturated fatty acids (OB)	14:0 \ 16:0 \ 17:0 \ 18:0
Gram-positive bacteria	i15: 0 < a15:0 < i16:0 < i17:0 < a17: 0
Gram-negative bacteria	i16:1 $\omega$ 7c $\sim$ 16:1 $\omega$ 9c $\sim$ 18:1 $\omega$ 5c $\sim$ 18:1 $\omega$ 7c $\sim$ cy17:0 $\sim$

Table 2 Soil properties change with different biochar addition treatments.

Treatment	Treatment	pН	$\mathrm{SWC}(\%)$	$\rm BW(g/cm^3)$	$\mathrm{DOC}(\mathrm{g/kg})$	$\mathrm{SOC}(\mathrm{g/kg})$	${\rm TN}({\rm g/kg})$	$\mathrm{TP}(\mathrm{g/kg})$
CK	CK	$8.22{\pm}0.33$	$21{\pm}10$	$0.67{\pm}0.10$	$0.09{\pm}0.02$	$12.06 {\pm} 3.08$	$0.90{\pm}0.07$	$0.24{\pm}0.02$

Treatment	Treatment	pН	$\mathrm{SWC}(\%)$	$\mathrm{BW}(\mathrm{g/cm^3})$	$\mathrm{DOC}(\mathrm{g/kg})$	$\mathrm{SOC}(\mathrm{g/kg})$	${\rm TN}({\rm g/kg})$	TP(g/kg)
LK	T0.5	$8.43{\pm}0.34$	$17 \pm 12$	$0.64{\pm}0.07$	$0.13 {\pm} 0.04$	$12.20 \pm 3.13$	$0.88{\pm}0.10$	$0.27 {\pm} 0.02$
	T1	$8.18{\pm}0.28$	$15\pm10$	$0.62{\pm}0.09$	$0.09 {\pm} 0.02$	$13.55 {\pm} 3.01$	$0.84{\pm}0.12$	$0.28{\pm}0.06$
	T2	$8.13{\pm}0.21$	$14{\pm}10$	$0.59{\pm}0.07$	$0.09{\pm}0.03$	$11.62 {\pm} 4.45$	$0.87{\pm}0.10$	$0.26{\pm}0.03$
HK	T5	$8.31{\pm}0.19$	$16\pm12$	$0.61{\pm}0.06$	$0.12{\pm}0.05$	$12.33 {\pm} 2.75$	$0.81{\pm}0.08$	$0.33{\pm}0.03$
	T10	$8.44 {\pm} 0.19$	$15\pm9$	$0.59{\pm}0.09$	$0.15{\pm}0.05$	$13.60 {\pm} 3.68$	$0.81{\pm}0.09$	$0.35{\pm}0.05$

Table 3 Change of soil microbe's biomass under biochar addition

Treatment	PLFAs(nmol/g)	$Microorganism\ species (nmol/g)$	Microorganism species(nmol/g)	Microorganism species(1
		BAC	ACT	FUN
CK	$265.37 \pm 21.50^{\rm a}$	$171.07 \pm 12.99^{a}$	$36.98 \pm 4.04^{a}$	$25.56 \pm 1.77^{\rm a}$
LK	$264.83 \pm 11.10^{a}$	$163.90{\pm}6.38^{\rm a}$	$37.23 \pm 2.10^{a}$	$30.32 \pm 1.17^{\rm ab}$
НК	$320.52 \pm 20.12^{a}$	$198.76 \pm 11.54^{\rm a}$	$41.23 \pm 2.81^{a}$	$36.84{\pm}2.43^{\rm b}$

Different letters meant a significant difference at 0.05 level

**Table 4** Comparison of SOC and DOC change with biochar addition in the present work and previous studies.

Soils/land	Biochar amendment	SOC change (%)	DOC (%)	Reference
Acidic acrisol	10% (wt/wt)		+2775%	(Smebye et al., 2016)
Arable soil	30%(wt/wt)	+100%~ $+127%$		(Steinbeiss et al., 2009)
Paddy field	10  t/ha - 40(t/ha)	$+10.8\%^{-}+55.6\%$		(Zhang, A. et al., 2012)
Sugarcane field	0.68%-1.04% (wt/wt)	+54%	-11.8%	(Yin et al., 2014)
Arable field	24-72(t/ha)	+153%	-9.75%	(Prommer et al., $2014$ )
Rice field	1.78-29.6 (t/ha)	+63%~ $+65%$		(Sui et al., 2016)
Soil without types	2%-5% (wt/wt)		-1.31%~+31.1%	(Zhao et al., 2015)
Coastal saline soil	0.5%- $2.0%$ (wt/wt)	+5.2%~ $+68%$		(Novak et al., 2009)
Agriculture field	15.75~47.25(t/ha)	+27.08%~ $+92.61%$	-5.59%~-26.67%	(Yang et al., 2018)
Rice field	1~10(t/ha)	+43.8%~ $+169%$		(Munda et al., 2018)
Coastal saline soil	$0.5\%$ $^{\sim}2\%$	-3.83%~ $+187%$		(Sun et al., 2016)
Costal saline soil	$3.2^{\sim}32(t/ha)$	+31%~ $+298%$		(Lin et al., 2015)
Saline soil	0.5%~10% (wt/wt)	+1.16%~ $+12.8%$	$+0\%^{-}+66.7\%$	The present work

Table 5 Summary of multiple linear regression models of DOC, SOC, RC, and environment factors.

Independent	Factor	Coefficient	Beta	$R^2{}_{\rm adj}$	F	p
DOC	ACT	0.002	0.635	0.571	27.648	< 0.000
	NP	-0.015	-0.233			
	$_{\rm pH}$	0.026	0.181			
$\mathrm{RC}$	ACT	0.018	0.516	0.704	72.393	< 0.000
	NP	-0.020	-0.568			
SOC	CPOC	0.156	0.730	0.525	67.314	< 0.000

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Figures.pdf available at https://authorea.com/users/337997/articles/463970-biochar-amendment-promotes-limited-organic-carbon-increase-in-saline-sodic-soil-of-the-songnen-plain-northeast-china