



Full Length Article

Biochar and Slow Release Urea Effects on Root Morphology, Grain Yield, Nitrogen Uptake and Utilization in *Brassica napus*

Xiaoqin Tian^{1,2}, Zhuo Li², Longchang Wang^{1*}, Yifan Wang¹ and Biao Li¹

¹College of Agronomy and Biotechnology, Southwest University/Key Laboratory of Eco-environments in Three Gorges Reservoir Region, Ministry of Education/Engineering Research Center of South Upland Agriculture, Ministry of Education, Chongqing, 400715, P.R. China

²Crop Research Institute, Sichuan Academy of Agriculture Sciences, Chengdu, Sichuan, 610066, P.R. China

*For correspondence: wanglc@swu.edu.cn

Received 26 July 2019; Accepted 25 November 2019; Published 04 February 2020

Abstract

Low nitrogen (N) utilization efficiency is a serious problem in rapeseed production. Slow-release urea (SRU) or the combined application of slow-release urea and common urea could enhance N utilization, but this effect is limited and long-term N fertilizer application would lead to soil acidification. Biochar has the potential to effectively improve acid soil and increase N utilization efficiency. A pot experiment was conducted to investigate the effects of the combined application of biochar and different urea on the root morphology, grain yield, and nitrogen uptake and utilization of rapeseed. The two-factorial complete randomized block design was adopted with three schemes of urea (100% common urea, 100% slow-release urea, and 60% common urea and 40% slow-release urea, named UR, SRU, and combined (60%+40% SRU), respectively), and three biochar levels (0, 2 and 4% of the soil weight, named C0, C1 and C2, respectively). Biochar was found to significantly affect rapeseed root morphology, grain yield, nitrogen uptake and utilization when compared with no biochar, but these effects varied with different urea and biochar application rate. C1 combined with 60%+40% SRU significantly promoted the diameter of root crown, total root volume, average root diameter, total nitrogen accumulation (TNA), and nitrogen-use efficiency (NUE); C1 combined with UR also significantly increased the total root volume, total root length, and average root diameter of rapeseed. However, C2 combined with UR and 60%+40% SRU, and the combined application of biochar (C1 and C2) and SRU, had an inhibitory effect on root morphology, nitrogen uptake and utilization, and yield due to the slow release effect of biochar on available nitrogen. The diameter of root crown, total root volume, total root surface area, average root diameter, TNA, NUE, effective pod number, number of grains per pod, and yield were highest under the combined application of 60%+40% SRU and C1. NUE and yield of 60%+40% SRU+C1 treatment increased by 12.36–63.68% and 2.82–46.59%, respectively, than with urea application only. This combination may be an effective mean to further improve the yield and NUE of rapeseed in dryland in southwest China. © 2020 Friends Science Publishers

Keywords: Biochar; Nitrogen fertilizer; Rapeseed; Soil amendment; Dryland in southwest China

Introduction

Low nitrogen (N) utilization efficiency of crops is a serious problem throughout the world, especially in China. Good root morphology is important to achieve both high yield and high efficiency of rapeseed (Comas *et al.* 2013). The southwest region of China is the main rapeseed production area and crucial to ensuring the security of edible oil industry. Nitrogen plays an important role in rapeseed growth and development, and increase of N application has been a major management strategy for high-yielding rapeseed cultivation (Cassman *et al.* 2003). As a result, nitrogen fertilizer input has increased year by year in this area. With the development of mechanized light and simple

production of rapeseed, people have increased the investment of nitrogen fertilizer in the early stage to avoid the trouble of late fattening, which have reduced nitrogen fertilizer-use efficiency, have enhanced N loss and volatilization, and have also caused a series of environmental problems (Lassaletta *et al.* 2014; Duran *et al.* 2016; Clark and Tilman 2017; Kostyanovsky *et al.* 2019). Thus, reasonable N fertilization strategies should be developed and utilized to achieve high crop yields and high nitrogen efficiency and to minimize negative effects on the environment.

Without changing the chemical structure of common urea, slow-release urea reduces the urea release rate by physical or chemical treatment so that the rate of ammonia

production is consistent with the speed of crop utilization, thus improving N utilization efficiency. Several studies have demonstrated that the application of slow-release urea has numerous benefits, including achieving one-time simplified fertilization, improving N utilization rate, reducing soil nitrogen surplus, increasing crop yield, and reducing the risk of environmental pollution (Saha *et al.* 2019). At present, the combined application of slow-release urea and ordinary urea in an appropriate ratio has attracted widespread attention from scholars in both China and other countries (He *et al.* 2017; Pirtle *et al.* 2019; Rafael *et al.* 2019). Studies showed that appeared greedy to crop resulting in lower yield and higher costs because of rich nitrogen supply under 100% slow-release urea treatment, while the combined application of slow-release urea and ordinary urea had a good comprehensive effect (Xie *et al.* 2019). No matter it is applied alone or in combination with ordinary urea, the effect of improving N use efficiency is limited, and long-term chemical fertilizer application would cause soil acidification (Cheng *et al.* 2017). It is therefore, desirable to find efficient comprehensive strategies that can improve N utilization efficiency based on ensuring good soil quality.

Biochar has long been subjects of scientific interest as amendments for acid soil improvement, crop production, and increase of N use efficiency (Abrishamkesh *et al.* 2015; Kraska *et al.* 2016; Sowiński and Glab 2018; El-Naggar *et al.* 2019). Moreover, agronomic performances of biochar were variable (positive or negative effects) with soil, crop, and biochar types and application modes including fertilizer types and application modes (Hussain *et al.* 2017; Sarma *et al.* 2018). It is generally accepted that biochar can increase the pH of acidic soils due to alkaline substances in biochar which neutralize some soil acidity (Wang *et al.* 2016). However, it is still unclear whether the effects of biochar on the N use efficiency and yield of rapeseed would vary with different urea types in the southwest purple soil region in China. Moreover, it is also unclear whether the combination of biochar and slow-release urea, and the combination of biochar and both common urea and slow-release urea would significantly increase N use efficiency and yield of rapeseed, compared with applying urea alone. Therefore, the effects of combined application of biochar and different urea on root morphology, yield and N uptake and utilization of rapeseed are studied with the purpose to: (1) provide a theoretical basis for the reasonable application of biochar under different types of urea; (2) provide technical support for achieving high yields and high N use efficiency of rapeseed in southwest China.

Materials and Methods

Experimental materials

The tested soil was collected from the cultivation-layer (*i.e.*, the top 0–20 cm) at the experimental farm of Southwest University in Chongqing City, China (29°49'N, 106°25'E).

Pre-processing of soil was done according to the methods of Šas dková *et al.* (2018). The soil was classified as a typical purple soil and its physico-chemical properties were as follows: 7.22 g·kg⁻¹ organic matter, 0.62 g·kg⁻¹ total N, 36.75 mg·kg⁻¹ available N, 9.46 mg·kg⁻¹ available P, 80.00 mg·kg⁻¹ available K, and pH 6.91. Selected properties of the applied biochar, which was pyrolyzed at 500°C for 2 h from straw of rice (*Oryza sativa* L.), were as follows: 0.61 g·kg⁻¹ total N, 1.99 g·kg⁻¹ total P, 27.15 g·kg⁻¹ total K, 537.97 g·kg⁻¹ total C, and pH 8.70. The sustained release period of slow-release urea (polymer-coated urea, 44.5% N) was about 90 d in soil.

Experimental design

A pot experiment was conducted in the greenhouse of Southwest University. The treatments were defined by a two-factorial complete randomized block design using three schemes of urea (100% common urea, 100% slow-release urea, and 60% common urea and 40% slow-release urea, named UR, SRU, and 60%+40% SRU, respectively), and three biochar levels (0, 2, and 4% of the soil weight, named C0, C1, and C2, respectively). A control treatment (CK) which received no biochar and no N fertilizer was also included. The ten treatment combinations were replicated 12 times. The prepared materials were mixed and then filled into pots (25 cm inner diameter and 35 cm height; each pot contained 5 kg of soil).

The sowing rate of rapeseed (Sanxiayou No. 5) was 5–6 seeds·pot⁻¹. The rapeseed experiment was conducted from October 20, 2017, to May 15, 2018. N, P, and K were applied at rates of 0.20 g N, 0.15 g P₂O₅ and 0.15 g K₂O per kg of soil (equal to 1g N·pot⁻¹, 0.75 g P₂O₅·pot⁻¹, and 0.75g K₂O·pot⁻¹) as a basal fertilizer for rapeseed. Other management practices applied were in accordance with those used by local farmers.

Measurement items and methods

Soil samples (0–20 cm) were collected from each plot at 7, 15, 30, 45 and 60 days after urea application. During sampling and transportation, all the samples were kept in an insulated box with ice. Prior to analysis, soil samples were stored at 4°C to determine NO₃⁻ and NH₄⁺ concentrations of soil. The plants were harvested from each pot at maturity in May 2018 and separated into roots, straw, pods, and grains to determine the root morphology, dry weight, grain yield, and total N content of rapeseed. Other yield-related agronomic traits, including the effective number of pods, the number of grains per pod, and thousand-seed weight, were determined simultaneously. NO₃⁻ and NH₄⁺ concentrations of soil were determined using the dual-band UV-spectrometer and indophenol blue colorimetry, respectively (Zhang and Gong 2012). The root morphology was determined using a Root Scanner device (Regent Instrument Inc., Québec, Canada) after the root samples were collected

and cleaned during the bud and maturing stages. The samples of all organs were desiccated at 105°C for 30 min and then dried at 70°C to a constant weight to determine the dry weight during the bud and maturing stages. These were then finely ground into powder to pass through a 0.2 mm sieve, to determine the total N values using the semi-micro Kjeldahl method. N use efficiency was calculated according to Sang *et al.* (2018).

N utilization efficiency (NUE, %) = (total N uptake by plant with added N – total N uptake by plant with no N)/N application amount × 100.

Statistical analysis

A two-way ANOVA was used to determine differences between the treatments. Means were compared using Duncan’s test at a 0.05 probability level. The data were analyzed using the S.P.S.S. 17.0 software for Windows (SPSS Inc., Chicago, USA).

Grey correlation analysis was performed on highly significantly related indicators. Five main traits of rapeseed were selected and the optimal value of each trait was determined. The mean value of each trait was dimensionless, and the greater the range of each trait, the better. The upper limit effect measure was used. The grey relational coefficient $\xi_i(k)$ can be expressed as follows (Lei 1996):

$$\xi_i(k) = \frac{\min \min |x_0(k) - x_i(k)| + \rho \max \max |x_0(k) - x_i(k)|}{|x_0(k) - x_i(k)| + \rho \max \max |x_0(k) - x_i(k)|}$$

$$x_i^k = \frac{x_i^k}{\max x_i^k}$$

$$\gamma_i = \frac{1}{n} \sum_{k=1}^n \xi_i(k)$$

$$\omega_{i(r)} = \frac{1}{n} \sum_{i=1}^n \gamma_i$$

$$G_i^k = \sum_{r=1}^n (\xi_i \times \omega_{i(r)}),$$

$$k=1, 2, 3, \dots, n; i=1, 2, 3, \dots, n$$

Where: x_i^k - The i trait observation value of treatment k ;

$\max_i x_i^k$ - The maximum value of the i trait in all treatments;

$\min_i x_i^k$ - The minimum value of the i trait in all treatments;

$\min_i \min_k |x_0(k) - x_i(k)|$ - Sec level minimum difference;

$\max_i \max_k |x_0(k) - x_i(k)|$ - Sec level maximum difference; ρ - Resolution coefficient (0.5).

Results

Root morphology

Biochar significantly affected rapeseed root morphology (except total root length and total root surface area at bud

stage), but this effects varied with different urea and biochar application (Table 1). Under UR treatment, 2% biochar significantly promoted the average root diameter and total root volume at the bud stage, and total root length at the maturing stage, which were 10.98, 28.66 and 12.87% higher, respectively, than those with 0% biochar. The treatment of 2% biochar also significantly promoted the average root diameter at the bud stage, root crown diameter at the maturing stage, and total root volume at the maturing stage, when combined with 60%+40% SRU treatment, which were 12.20, 9.26 and 15.13% higher, respectively, than with no biochar. However, the combined application of 4% biochar with 60%+40% SRU and UR as well as the combination of biochar (2 or 4%) with SRU had an inhibitory effect on the root morphology of rapeseed. The root crown diameter and total root volume in SRU+C1 were significantly reduced by 11.19 and 38.54%, and in SRU+C2 were significantly reduced by 12.59 and 30.73%, respectively, compared with those of SRU+C0 at the bud stage. The root crown diameter of plant in SRU+C1 and SRU+C2 was also 13.24 and 14.88% lower, respectively, than in SRU+C0 at the maturing stage. Additionally, the root crown diameter and total root volume of 60%+40% SRU+C2 were significantly lower by 7.25 and 21.08% at the bud stage and 11.11 and 22.99% at the maturing stage, respectively than with 60%+40% SRU+C0; the average root diameter and total root length of UR+C2 were 13.81 and 12.83% lower, respectively, than with UR+C0 at the maturing stage. This indicates that the appropriate amount of biochar (2%) was beneficial to promote the growth of rapeseed roots under UR and 60%+40% SRU treatments, while high biochar application (4%) had a certain inhibitory effect on rapeseed roots. Additionally, the combined application of biochar (2 or 4%) and SRU was not conducive to the growth of rapeseed roots.

At the maturing stage, the interaction of urea and biochar on the root morphology of rapeseed reached a significant level except for total root surface area. The root crown diameter, total root surface area and total root volume of rapeseed treated with 60%+40% SRU+C1 were the highest than other treatments, and the average root diameter was only lower than UR+C1. This indicates that the combined application of 2% biochar with 60%+40% SRU was most conducive to promoting root growth in all treatments.

Nitrogen accumulation and utilization

Application of 4% biochar significantly reduced the total N in UR and 60%+40% SRU treatments (Fig. 1). Compared with C0, the total N in C2 under the UR and 60%+40% SRU treatments was 18.32 and 13.43% at bud stage, while 12.85 and 17.60% at maturing stage but lower, respectively. In addition, the total N in SRU+C1 and SRU+C2 was significantly lower by 15.71 and 25.29% at bud stage, and 21.29 and 19.08% at maturing stage, respectively than for

Table 1: Root morphology of rapeseed in different treatments

Treatment	Bud stage					Maturing stage				
	Root crown diameter (mm)	Total root length (cm)	Total root volume (cm ³)	Total root surface area (cm ²)	Average root diameter (mm)	Root crown diameter (mm)	Total root length (cm)	Total root volume (cm ³)	Total root surface area (cm ²)	Average root diameter (mm)
URC0	6.70bcd	261.81a	1.64c	19.18a	0.82b	7.43bcd	200.72b	2.35bc	16.15abc	1.81abc
URC1	6.76bc	269.39a	2.11a	19.52a	0.91a	7.84bc	226.55a	2.51b	16.32abc	1.99a
URC2	6.48cd	262.26a	1.62c	19.41a	0.73c	6.98d	174.97d	2.25cd	15.61bc	1.56d
SRUC0	7.15ab	275.16a	1.92b	19.75a	0.81bc	7.93bc	192.20bc	2.33bc	16.37abc	1.71bcd
SRUC1	6.35cd	272.32a	1.18e	19.62a	0.81bc	6.88d	189.86bc	2.30bcd	15.66bc	1.64bcd
SRUC2	6.25d	268.31a	1.33d	19.24a	0.76bc	6.75d	186.81c	1.97d	15.42bc	1.61cd
60%+40% SRUC0	7.31a	273.45a	1.85b	19.53a	0.82bc	8.10b	183.65cd	2.71b	16.88ab	1.85ab
60%+40% SRUC1	7.47a	275.53a	1.97b	19.55a	0.92a	8.85a	195.23bc	3.12a	17.30abc	1.93a
60%+40% SRUC2	6.78bc	273.39a	1.46d	19.46a	0.73c	7.20cd	173.98d	2.01cd	16.23a	1.78abc
N	**	*	**	ns	ns	**	**	**	**	**
C	**	ns	**	ns	**	**	**	**	*	**
N×C	**	ns	**	ns	*	**	**	**	ns	*

ns: non-significant, *: $P \leq 0.05$, **: $P \leq 0.01$. Values with the same letter within a column are not significantly difference at $P = 0.05$. N: different urea types, C: biochar levels, N×C: interaction effect between biochar and urea

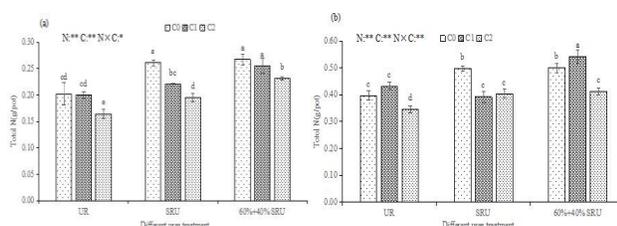


Fig. 1: Total N determined at (a) bud stage and (b) maturing stage of rapeseed in different treatments

N: different urea types, C: biochar levels, N×C: interaction effect between biochar and urea. Bars superscribed by different letters are significantly different at $P < 0.05$

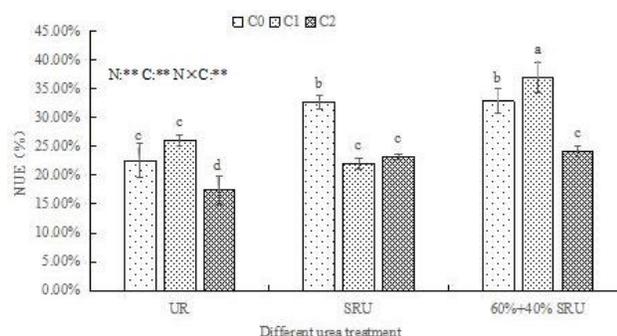


Fig. 2: Nitrogen utilization efficiency in different treatments

N: different urea types, C: biochar levels, N×C: interaction effect between biochar and urea. Bars superscribed by different letters are significantly different at $P < 0.05$

SRUC0. However, compared with C0, the total N in C1 under the 60%+40% SRU treatment was increased by 8.20% at the maturing stage, which was significantly higher than other treatments.

Same as the accumulation of total N, 4% biochar significantly reduced the NUE in UR and 60%+40% SRU treatments (Fig. 2). Compared with C0, the total N in C2 under the UR and 60%+40% SRU treatments was 22.79 and 26.73% lower, respectively. Additionally, the NUE in SRUC1 and SRUC2 was significantly lower by 32.50 and 29.03% than in SRUC0 respectively. However, compared with C0, the NUE in C1 under the 60%+40%

SRU treatment was significantly increased by 12.36%. Moreover, the NUE of 60%+40% SRUC1 treatment was highest in all treatments, and significantly higher by 12.36–63.68% than urea alone. This indicates that the combined application of high biochar application (4%) with different urea types inhibited the accumulation and absorption of nitrogen in rapeseed. Compared with 2% biochar combined with UR or SRU, and compared with applying urea alone, 2% biochar combined with 60%+40% SRU could promote the accumulation and absorption of nitrogen in rapeseed.

Yield and its components

The effects of biochar on effective pods number, thousand-seed weight and grain yield were significant, and the interaction of urea and biochar also reached a significant level (Table 2). Yields in C1 and C2 under SRU treatment were 23.62 and 24.27% lower than in C0 which was due to the number of effective pods in SRUC1 and SRUC2 that were 24.66 and 28.77% lower than in SRUC0 respectively, and yield under 60%+40% SRUC2 was also reduced by 14.93% than under 60%+40% SRUC0, whereas biochar had little effect on yields under UR. With 60%+40% SRUC1 treatment, the rapeseed yield was highest (3.65 g·plant⁻¹) significantly than other treatments except for 60%+40% SRUC0, and was 2.82, 18.12, 46.59 and 223.01% higher than 60%+40% SRUC0, SRUC0, UR+C0 and control, respectively. This indicates that the effect of biochar on rapeseed yield varied with different urea types. Compared with 2% biochar combined with UR or SRU, 2% biochar combined with 60%+40% SUR was more conducive to increase rapeseed yield.

Correlation and grey correlation analysis

Root crown diameter, total root surface area, effective pod number and number of grains per pod showed highly significant positive correlations with rapeseed yield, which was positively correlated with total root volume, and had no correlation with total root length, average root diameter and

Table 2: Yield and its components of rapeseed in different treatments

Treatment	Effective pods number-pod ⁻¹	Number of grains per pod	Thousand-seed weight (g)	Grain yield (g-pot ⁻¹)
CK	52c	14a	3.26c	1.13d
URC0	59bc	16a	3.24c	2.49c
URC1	62bc	16a	3.34bc	2.67c
URC2	55bc	15a	3.76a	2.42c
SRUC0	73ab	16a	3.35bc	3.09b
SRUC1	55bc	14a	3.35bc	2.36c
SRUC2	52c	14a	3.57ab	2.34c
60%+40% SRUC0	80ab	17a	3.57ab	3.55a
60%+40% SRUC1	86a	18a	3.58ab	3.65a
60%+40% SRUC2	70abc	16a	3.65a	3.02b
N	**	ns	**	**
C	*	ns	**	**
N×C	*	ns	*	**

ns: non-significant, *: $P \leq 0.05$, **: $P \leq 0.01$. Values with the same letter within a column are not significantly difference at $P = 0.05$. N: different urea types, C: biochar levels, N×C: interaction effect between biochar and urea

Table 3: Correlation coefficients of root morphology and yield factors with yield of rapeseed

Root morphology	Yield	Yield factors	Yield
Root crown diameter	0.871**	Effective pod number	0.958**
Total root length	-0.072	Number of grains per pod	0.892**
Total root volume	0.723*	Thousand-seed weight	0.212
Total root surface area	0.936**		
Average root diameter	0.592		

* and ** denote significant correlation at the 0.05 and 0.01 probability levels, respectively

Table 4: Grey judgement analysis of root traits and yield factors

Treatment	Correlation coefficient (ξ)					G
	Root crown diameter	Total root volume	Total root surface area	Effective pods number	Number of grains per pod	
URC0	0.451	0.645	0.340	0.971	0.385	0.600
URC1	0.456	0.639	0.379	0.924	0.451	0.605
URC2	0.507	0.690	0.351	0.846	0.432	0.596
SRUC0	0.723	0.564	0.565	0.983	0.753	0.742
SRUC1	0.497	0.588	0.333	0.948	0.496	0.610
SRUC2	0.515	0.930	0.341	0.780	0.486	0.636
60%+40% SRUC0	0.693	0.554	0.976	0.753	0.821	0.750
60%+40% SRUC1	1.000	1.000	1.000	1.000	1.000	1.000
60%+40% SRUC2	0.903	0.414	0.539	0.906	0.678	0.707
CD (γ)	0.638	0.669	0.536	0.901	0.611	
WC (ω)	0.190	0.199	0.160	0.269	0.182	

G: Grey comprehensive evaluation value, CD: Correlation degree, WC: Weight coefficient

thousand-seed weight (Table 3).

The grey comprehensive evaluation values for the different treatments followed the order: 60%+40% SRUC1 > 60%+40% SRUC0 > SRUC0 > 60%+40% SRUC2 > SRUC2 > SRUC1 > URC1 > URC0 > URC2 (Table 4). The overall pattern under the UR and 60%+40% SRU treatments was C1 > C0 > C2 and C0 showed better results than C1 and C2 under the SRU treatment. Besides, the combined application of 60%+40% SRU and C1 could make the root morphology and yield traits of rapeseed close to the optimal level.

Discussion

Crop root growth is directly affected by the soil environment, including soil temperature, moisture, aeration, porosity and nutrients. Biochar has a certain dilution effect after being applied to the soil due to its developed pore

structure and low density, which can change the aeration and porosity of the soil, thus indirectly affecting the crop root growth. Several studies have also shown that the application of biochar to soil decreased soil bulk density, and improved soil porosity and pore size (Laird *et al.* 2010; Devereux *et al.* 2012). These results may explain the observations of this study, which indicated that 2% biochar improved the root morphology of rapeseed under the UR and 60%+40% SRU treatment (Table 1). However, the high biochar application (4%) inhibited the root growth. Soil porosity may be too high under high levels of biochar application, which accelerated water and nutrient loss, leading to their insufficient absorption and utilization, thus affecting root growth (Xu *et al.* 2016). Previous study has also pointed out that the root epidermal cells were almost completely shed and cortical hair breeding was inhibited by higher biochar application, which directly inhibited root elongation and root thickening (Zhou *et al.* 2017). In this

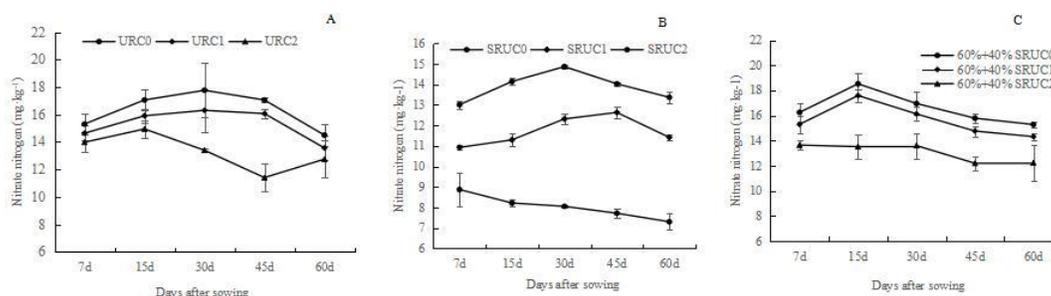


Fig. 3: Nitrate nitrogen content in different treatments

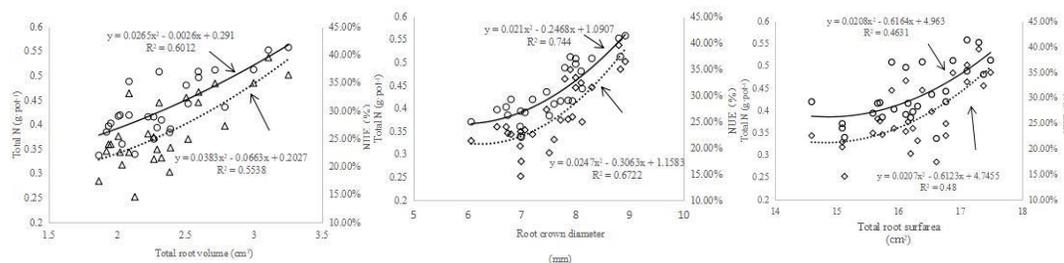


Fig. 4: The effect of root morphology on total N and NUE of rapeseed

study, biochar was alkaline (pH 8.70) and carbon content was high (537.97 g·kg⁻¹ total C). Therefore, soil pH can be largely changed very much by the addition of excess biochar which may cause competitive adsorption of nutrients with roots, thus inhibiting root growth and nutrient uptake (Kishimoto and Sugiura 1985). In addition, changes in soil pH and nutrient conditions, especially C/N, may have a negative effect on the community structure and function of some soil microbes, affecting the physiological functions of roots. Some exploratory studies suggested that certain volatile components of biochar might also inhibit crop growth (Deenik *et al.* 2009), but this viewpoint still required the large-scale accurate and confirmatory tests.

Different from the UR and 60%+40% SRU treatment, root crown diameter and total root volume of the SRU treatment significantly decreased under medium (2%) or high (4%) biochar application. To understand this phenomenon, the soil nitrate nitrogen and ammonium nitrogen content of different treatments within 60 days were tested. Since the ammonium content of each treatment was very low and their differences were not basically significant, only the nitrate nitrogen was analyzed. The nitrate nitrogen content in the soil of the SRU treatment was significantly reduced under 2% biochar application during the 60 days of crop growth, and there were less nutrients for direct absorption by roots (Fig. 3). However, the demand for nitrogen at the seedling stage of rapeseed accounted for nearly 50% of the whole growth period. Thus, the roots of rapeseed could not grow well in the later stage due to the early nitrogen deficiency, resulting in a significant decrease in the root crown diameter (Table 1). It could also be seen that 4% biochar significantly reduced the nitrate nitrogen

content of the soil under various urea treatments, which may be another major reason for the inhibition of crop root development under the addition of high biochar combined with UR and 60%+40% SRU treatment. These results further confirmed the nutrient release effect of biochar (Widowati *et al.* 2011; Li *et al.* 2013).

The nutrient absorption of crops from the soil mainly relies on the root system. The development of roots directly affects the accumulation and absorption of nutrients by the crops. The polynomial regression analysis (Fig. 4) showed that total N and NUE significantly ($P \leq 0.05$) increased with the increase of total root volume ($y=0.0265x^2-0.0026x+0.291$; $R^2=0.60$ and $y=0.0383x^2-0.0663x+0.2027$; $R^2=0.55$), the root crown diameter ($y=0.021x^2-0.2468x+1.0907$; $R^2=0.74$ and $y=0.0247x^2-0.3063x+1.1583$; $R^2=0.67$) and total root surface area ($y=0.0208x^2-0.6164x+4.963$; $R^2=0.46$ and $y=0.0207x^2-0.6123x+4.7455$; $R^2=0.48$). The results of this study showed that 4% biochar significantly reduced total N and NUE under the various urea treatments, but the combined application of 2% biochar and 60%+40% SRU significantly promoted total N and NUE, which were related to the effect of biochar on the root morphology of rapeseed. Abiven *et al.* (2015) showed that biochar amendment resulted in more extensive root systems and improved N uptake in deep soil. In addition, the nutrient content in the soil was also the main influence factor for the nitrogen accumulation of crops (Rasool *et al.* 2018; Zhang *et al.* 2018). Application of 2% biochar was beneficial to the root volume under UR, but the nitrogen accumulation was not significantly increased due to the reduction of available nitrogen content in soil (Fig. 3; Table 1; Fig. 4),

which limited the absorption and utilization of nitrogen by roots to a certain extent.

There are still debates about the impact of biochar on crop yields. Major *et al.* (2010) showed that maize grain yield did not significantly increase under biochar application during first year. Zhang *et al.* (2010) reported that biochar amendments of 10 t·hm⁻² and 40 t·hm⁻² increased rice yields by 12 and 14% in unfertilized soils, and by 8.8 and 12.1% in soils with N fertilization, respectively. However, Albuquerque *et al.* (2013) pointed out that biochar addition to a nutrient-poor, slightly acidic loamy sand soil had little effect on wheat yield in the absence of mineral fertilization, but with the highest mineral fertilizer rate, addition of biochar led to about 20–30% increase in grain yield compared with the use of the mineral fertilizer alone. The reason for these differences is that the biochar will change due to soil nutrient status, biochar application period and crop type, etc. It was found that the effect of biochar on rapeseed yield varied with different urea types in this study. High amounts of biochar significantly reduced the yield of rapeseed under SRU and 60%+40% SRU treatment, but had no effect on the yield under UR treatment. This reduction of yield was mainly due to reduction in the number of effective pods under SRU and 60%+40% SRU treatment. The number of effective pods under C2 was 28.77 and 12.50% lower than that under C0 combined with the SRU and 60%+40% SRU treatment, respectively. Similarly, 2% biochar reduced the yield of SRU treatment due to a reduction in the number of effective pods (Table 2). Previous studies have found that soil organic carbon content increased and soil color deepened under high amount of biochar application, and the surface soil absorbed heat and light leading to the lack of oxygen and low temperature in the deep soil, which caused the root system can not grow well to the deep, thus affecting the nutrient absorption of the crop and reduce the number of effective pods resulting in a reduction in crop yield (Agegnehu *et al.* 2015; Wei *et al.* 2018). This is consistent with the results of this study. Nevertheless, the application of 2% biochar had a certain promoting effect on the yield of rapeseed under 60%+40% SRU and UR treatment, which was related to the increase of root growth and the number of effective pods (Table 3). Although the improvement effect was not significant, on the basis of stable production, the nitrogen utilization efficiency of rapeseed was significantly improved under 60%+40% SRU treatment. Hence, compared with UR and SRU, the combined application of 60%+40% SRU and 2% biochar is an efficient cultivation method in production.

Conclusion

Biochar had significant effects on the nitrogen uptake and utilization, yield and yield related traits of rapeseed, but these effects could be changed by different urea types. Under the UR and 60%+40% SRU treatments, 2% biochar could promote root growth, N-use efficiency and yield of

rapeseed, while it significantly reduced those under the SRU treatment, when compared with no biochar. Besides, 4% biochar inhibited root growth, and reduced nitrogen accumulation, nitrogen use efficiency, and yield of rapeseed under the different urea treatments due to the slow release effect of biochar on available nitrogen. The interaction of urea and biochar on the root morphology, nitrogen use efficiency and yield of rapeseed was also significant. The combined application of 60%+40% SRU and 2% biochar was the best as it provided the highest NUE, grain yield and partial yield related traits. It may be an effective mean for this combination to further improve the yield and NUE of rapeseed in dryland in southwest China.

Acknowledgements

This research was supported by the Public Welfare Industry (Agriculture) Research Project (201503127), and National Natural Science Foundation (31271673, 31700364, 31871583).

References

- Abiven S, A Hund, V Martinsen, G Cornelissen (2015). Biochar amendment increases maize root surface areas and branching: a shovelomics study in Zambia. *Plant Soil* 395:45–55
- Abrishamkesh S, M Gorji, H Asadi, GH Bagheri-Marandi, AA Pourbabae (2015). Effects of rice husk biochar application on the properties of alkaline soil and lentil growth. *Plant Soil Environ* 61:475–482
- Agegnehu G, AM Bass, PN Nelson, B Muirhead, G Wright, MI Bird (2015). Biochar and biochar-compost as soil amendments: Effects on peanut yield, soil properties and greenhouse gas emissions in tropical North Queensland, Australia. *Agric Ecosyst Environ* 213:72–85
- Albuquerque JA, P Salazar, V Barrón, J Torrent, MC Campillo, A Gallardo, R Villar (2013). Enhanced wheat yield by biochar addition under different mineral fertilization levels. *Agron Sustain Dev* 33:475–484
- Cassman KG, A Dobermann, TD Walters, H Yang (2003). Meeting cereal demand while protecting natural resources and improving environmental quality. *Annu Rev Environ Resour* 28:315–358
- Cheng WG, AT Padre, H Shiono, C Sato, T Nguyen-Sy, K Tawarayama, K Kumagai (2017). Changes in the pH, EC, available P, SOC and TN stocks in a single rice paddy after long-term application of inorganic fertilizers and organic matters in a cold temperate region of Japan. *J Soils Sed* 17:1834–1842
- Clark M, D Tilman (2017). Comparative analysis of environmental impacts of agricultural production systems, agricultural input efficiency, and food choice. *Environ Res Lett* 12:1–11
- Comas LH, SR Becker, VMV Cruz, PF Byrne, DA Dierig (2013). Root traits contributing to plant productivity under drought. *Front Plant Sci* 4:1–16
- Deenik JL, AT McClellan, G Uehara, JL Deenik, AT McClellan, G Uehara (2009). Biochar volatile matter content effects on plant growth and nitrogen and nitrogen transformations in a tropical soil. In: *Western Nutrient Management Conference*, pp:26–31. Salt Lake City, Utah, USA
- Devereux RC, CJ Sturrock, SJ Mooney (2012). The effects of biochar on soil physical properties and winter wheat growth. *Earth Environ Sci Trans Royal Soc Edinburgh* 103:13–18
- Duran BEL, DS Duncan, LG Oates, CJ Kucharik, RD Jackson (2016). Nitrogen fertilization effects on productivity and nitrogen loss in three grass-based perennial bioenergy cropping systems. *PLoS One* 11:1–13
- El-Naggar A, SS Lee, J Rinklebed, M Farooq, AK Sarmah, AR Zimmerman, M Ahmad, SM Shaheen, YS Ok (2019). Biochar application to low fertility soils: A review of current status, and future prospects. *Geoderma* 337:536–554

- He J, B Li, CQ Wang, YH Li, BF Yang, JS Zhang, H Xiang, B Yin, HH Li (2017). Effects of different nitrogen fertilization treatments on soil nitrogen supply and yield in rice-rape rotation. *Sci Agric Sin* 50:2957–2968
- Hussain M, M Farooq, A Nawaz, AM Al-Sadi, ZM Solaiman, SS Alghamdi, U Ammara, YS Ok, KHM Siddique (2017). Biochar for crop production: potential benefits and risks. *J Soils Sed* 17:685–716
- Kishimoto S, G Sugiura (1985). Charcoal as a soil conditioner. *Intl Achieve Future* 5:12–23
- Kostyanovsky KI, DR Huggins, CO Stockle, JG Morrow, IJ Madsen (2019). Emissions of N₂O and CO₂ following short-term water and N fertilization events in wheat-based cropping systems. *Front Ecol Evol* 7:1–10
- Kraska P, P Oleszczuk, S Andruszczak, E Kwiecińska-Poppe, K Różyło, E Pałys, P Gierasimiuk, Z Michałojć (2016). Effect of various biochar rates on winter rye yield and the concentration of available nutrients in the soil. *Plant Soil Environ* 62:483–489
- Laird DA, P Fleming, DD Davis, R Horton, B Wang, DL Karlen (2010). Impact of biochar amendments on the quality of a typical Midwestern agricultural soil. *Geoderma* 158:443–449
- Lassaletta L, G Billen, B Grizzetti, J Anglade, J Garnier (2014). 50 year trends in nitrogen use efficiency of world cropping systems: the relationship between yield and nitrogen input to cropland. *Environ Res Lett* 9:1–9
- Lei TS (1996). *Application of Grey System Theory in Agriculture*, pp: 31–61. Henan Science and Technology Press, Zhengzhou, China
- Li YM, MJ Wu, ZY Zhang, JH Lü, JF Li (2013). Effectiveness of low-temperature biochar in controlling the release and leaching of herbicides in soil. *Plant Soil* 370:333–344
- Major J, M Rondon, D Molina, SJ Riha, J Lehmann (2010). Maize yield and nutrition during 4 years after biochar application to a Colombian savanna oxisol. *Plant Soil* 333:117–128
- Pirtle T, L Rumble, M Klug, F Walker, S Cui, N Phillips (2019). Impact of biochar and different nitrogen sources on forage radish production in middle tennessee. *J Adv Agric* 10:1594–1610
- Rafael RBA, ML Fernández-Marcos, S Cocco, ML Ruello, F Fornasier, G Corti (2019). Benefits of biochars and NPK fertilizers for soil quality and growth of cowpea (*Vigna unguiculata* L. Walp.) in an acid arenosol. *Pedosphere* 29:311–333
- Rasool T, R Ahmad, M Farooq, R Ahmad (2018). Nitrogen use efficiency and productivity of barley as affected by nitrogen and sorghum mulching in different cropping systems. *Intl J Agric Biol* 20:1937–1944
- Saha BK, MT Rose, VNL Wong, TR Cavagnaro, AF Patti (2019). A slow release brown coal-urea fertiliser reduced gaseous N loss from soil and increased silver beet yield and N uptake. *Sci Total Environ* 649:793–800
- Sang HH, XY Jiao, SF Wang, WH Guo, MK Salahou, KH Liu (2018). Effects of micro-nano bubble aerated irrigation and nitrogen fertilizer level on tillering, nitrogen uptake and utilization of early rice. *Plant Soil Environ* 64:297–302
- Sarma B, M Farooq, N Gogoi, B Borkotoki, R Kataki, A Garg (2018). Soil organic carbon dynamics in wheat - green gram crop rotation amended with Vermicompost and Biochar in combination with inorganic fertilizers: A comparative study. *J Cleaner Prod* 201:471–480
- Šlapáková B, J Jeřábek, V Voříšek, V Tejneck, O Drábek (2018). The biochar effect on soil respiration and nitrification. *Plant Soil Environ* 64:114–119
- Sowiński J, L Głęb (2018). The effect of nitrogen fertilization management on yield and nitrate contents in sorghum biomass and bagasse. *Field Crops Res* 227:132–143
- Wang GJ, ZW Xu, Y Li (2016). Effects of biochar and compost on mung bean growth and soil properties in a semi-arid area of Northeast China. *Intl J Agric Biol* 18:1056–1060
- Wei BM, YQ Wang, ZW Li (2018). Influence of tobacco biochar on soil physic-chemical properties and maize growth in the soil composited with feldspathic sandstone and sand. *J Soil Water Conserv* 32:217–220
- Widowati UW, LA Soehono, B Guritno (2011). Effect of biochar on the release and loss of nitrogen from urea fertilization. *J Agric Food Technol* 1:127–132
- Xie Y, L Tang, YL Han, L Yang, GX Xie, JW Peng, C Tian, X Zhou, Q Liu, XM Rong, YP Zhang (2019). Reduction in nitrogen fertilizer applications by the use of polymer-coated urea: effect on maize yields and environmental impacts of nitrogen losses. *J Sci Food Agric* 99:2259–2266
- Xu J, WQ Niu, MZ Zhang, Y Li, W Lv, KY Li, XY Zhou, BH Niang (2016). Effect of biochar addition on soil evaporation. *Chin J Appl Ecol* 27:3505–3513
- Zhang AF, LQ Cui, GX Pan, LQ Lia, Q Hussain, XH Zhang, D Crowley (2010). Effect of biochar amendment on yield and methane and nitrous oxide emissions from a rice paddy from Tai Lake plain, China. *Agric. Ecosyst Environ* 139:469–475
- Zhang GL, ZT Gong (2012). *Soil Survey Laboratory Methods*. Science Press, Beijing, China
- Zhang Y, H Chen, G Ji, Y Zhang, J Xiang, S Anwar, D Zhu (2018). Effect of rice-straw biochar application on rice (*Oryza sativa*) root growth and nitrogen utilization in acidified paddy soil. *Intl J Agric Biol* 20:2529–2536
- Zhou JS, P Yan, WM Zhang, FY Zheng, XY Cheng, WF Chen (2017). Effect of biochar on root morphogenesis and anatomical structure of rice cultivated in cold region of Northeast China. *Acta Agron Sin* 43:72–81