

# Supplementary Materials

# **Title: Effects of Rice Husk Biochar on Carbon Release and Nutrient Availability in Three Cultivation Age of Greenhouse Soils**

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## Supplemental Figure

## Materials and Methods

Scanning electron microscopy (SEM) observations of RHB used powdered samples that were placed on aluminum stubs using carbon tape, and electron micrographs were obtained using FEI Inspect S (Thermo Fisher Scientific Inc., Waltham, Massachusetts, USA). Chemical functional groups were determined by Fourier transform infrared spectra (FTIR), and the biochar FTIR spectra were obtained from pressed pellets of 1:10 biochar/KBr mixtures using a PerkinElmer Pyris Diamond (PerkinElmer, Waltham, Massachusetts, USA). The pellets were then scanned in the reflectance mode in the range 4000~500 cm<sup>-1</sup>. A detailed account of the peak assignments of FTIR spectra refers to the reports of Keiluweit et al. [1] and Shen et al. [2].

## References

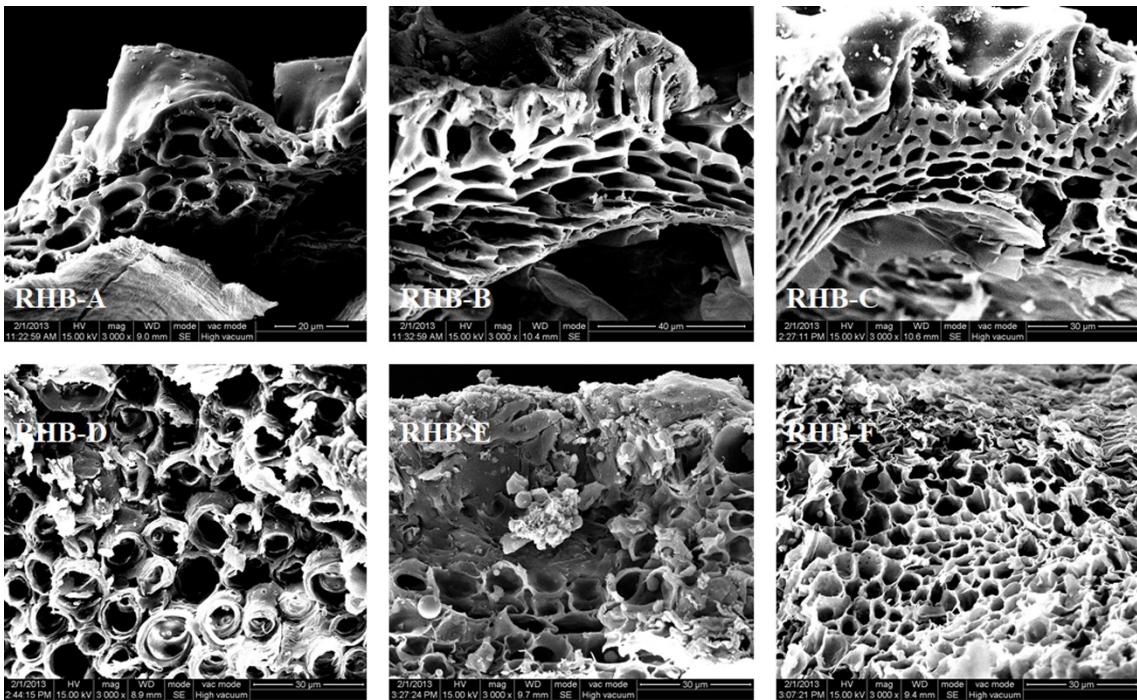
- References**

  - Keiluweit, M.; Nico, P.S.; Johnson, M.G.; Kleber, M., Dynamic molecular structure of plant biomass-derived black carbon (biochar). *Environ. Sci. Technol.* 2010, 44, 1247-1253.
  - Shen, Z.; Zhang, Y.; McMillan, O.; Jin, F.; Al-Tabbaa, A. Characteristics and mechanisms of nickel adsorption on biochars produced from wheat straw pellets and rice husk. *Environ. Sci. Pollut. Res. Int.* 2017, 24, 12809-12819.

**Supplemental Figure S1.** Results of significant tests of RHB effect on cumulative C release, soil properties and available nutrients within three studies soils.

**Supplemental Figure S2.** Pearson correlation coefficients between measured parameters and PCs (PC1 and PC2) at Day 100 for the three studied soils

**Supplemental Figure S3.** Canonical scores of the first two canonical discriminant functions (Can) of RHB type (A~F) and addition rate (0.5, 1.0, 4.0, 10, and 20%) in 3S, 14S and 24S soil. (C = control, CO = compost, A-0.5 = 0.5% of RHB A, and so on.)



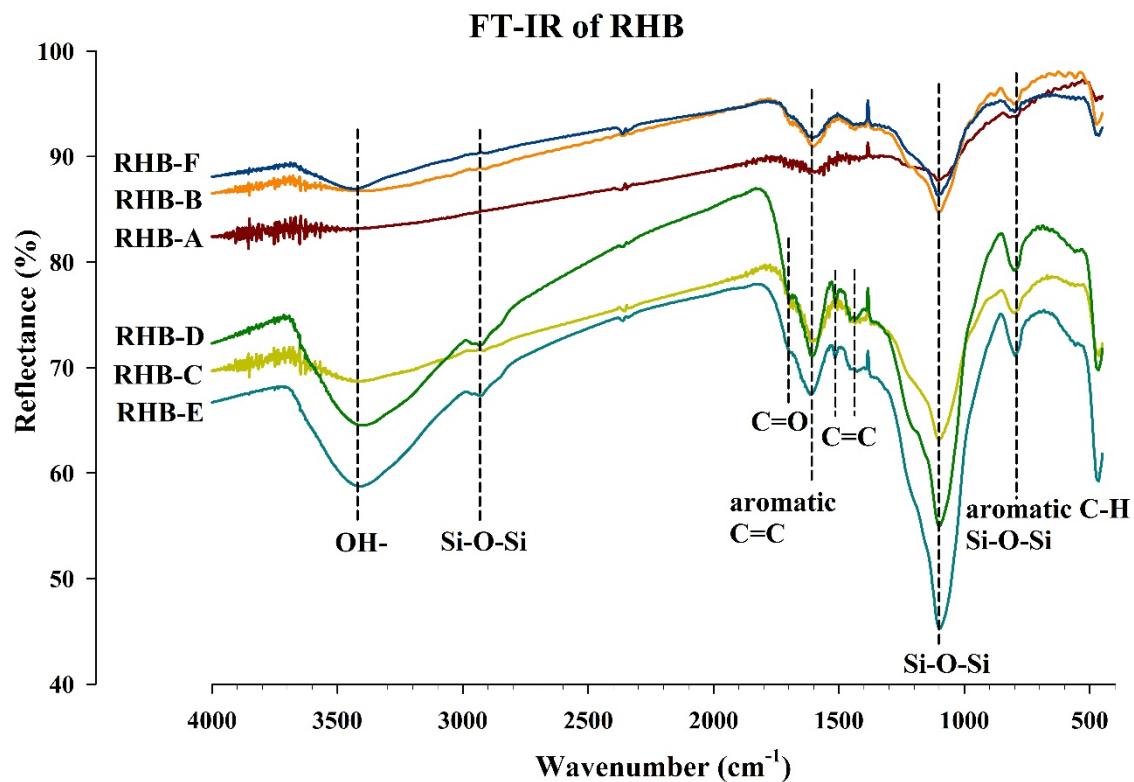
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**Supplemental Figure S1.** SEM images of the studied rice husk biochars.

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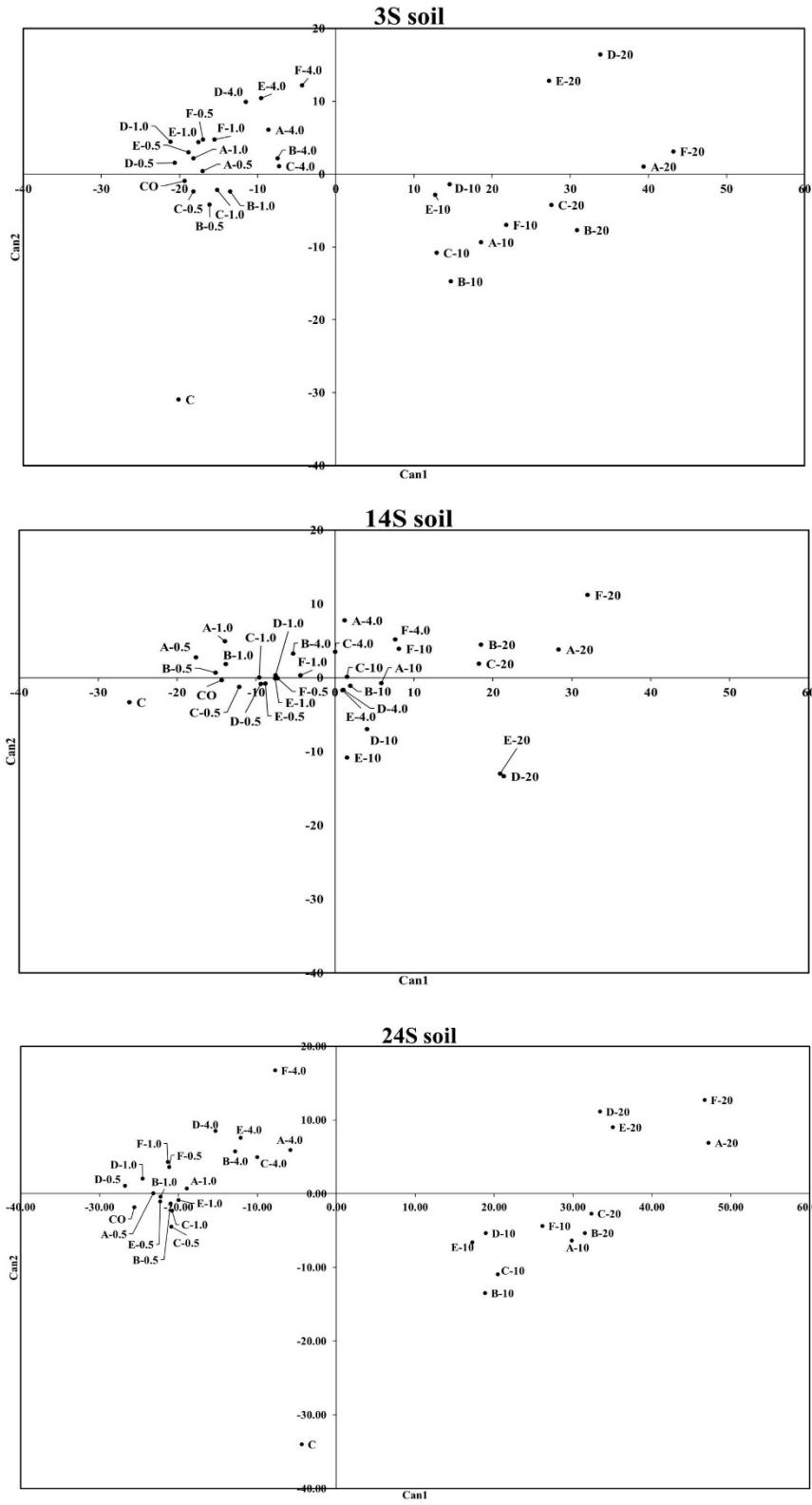
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**Supplemental Figure S2.** The FT-IR spectra of studied rice husk biochars.

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41      **Supplemental Figure S3.** Canonical scores of the first two canonical discriminant functions  
42      (Can) of RHB type (A~F) and addition rate (0.5, 1.0, 4.0, 10, and 20%) in 3S, 14S and 24S soil. (C =  
43      control, CO = compost, A-0.5 = 0.5% of RHB A, and so on.)

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## Supplemental Table

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11 **Supplemental Table S1.** Pearson correlation coefficients between measured parameters  
12 and PCs (PC1 and PC2) at Day 100 for the three studied soils.

**13      Supplemental Table S2.** Results of significant tests of RHB effect on cumulative C  
14                  release, soil properties and available nutrients within three studies soils.

**15      Supplemental Table S3.** Results of significant test of RHB effect on cumulative C  
16                  release, soil properties and available nutrients within five application rates.

1      **Supplemental Table S1.** Pearson correlation coefficients between measured parameters  
 2      and PCs (PC1 and PC2) at Day 100 for the three studied soils.

Parameter	3S soil		14S soil		24S soil	
	PC1	PC2	PC1	PC2	PC1	PC2
Total variance	58.6%	16.0%	55.1%	12.2%	55.8%	12.9%
Cumulative C release	-0.431* <sup>2</sup>	0.609*	-0.565*	0.394*	-0.162	0.819*
pH	-0.734*	0.542*	-0.646*	0.618*	-0.868*	0.142
EC <sup>1</sup>	0.614*	0.437*	-0.691*	-0.314	-0.172	-0.527*
TC	-0.839*	0.281	-0.878*	0.350	-0.884*	0.061
TN	0.961*	0.095	0.944*	0.191	0.951*	0.074
TP	0.735*	0.217	0.777*	0.122	0.745*	0.209
CEC	-0.443*	0.565*	-0.522*	-0.046	-0.270	0.639*
P	0.894*	0.335	0.938*	0.209	0.954*	0.122
K	-0.755*	0.543*	-0.569*	0.761*	-0.585*	0.502*
Ca	0.120	0.519*	-0.748*	-0.268	-0.278	-0.278
Mg	0.811*	0.465*	0.598*	0.047	0.893*	0.170
Cu	0.953*	0.070	0.892*	0.223	0.951*	0.016
Pb	0.957*	0.142	0.394*	0.380	0.848*	0.055
Zn	0.933*	0.103	0.945*	0.078	0.969*	0.053

3      <sup>1</sup> EC = electrical conductivity; TC = total carbon; TN = total nitrogen; TP = total  
 4      phosphorus; CEC = cation exchange capacity; <sup>2</sup> The asterisks after the data indicate  
 5      the significant correlations analyzed by SAS ( $P < 0.0001$ ).

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**Supplemental Table S2.** Results of significant tests of RHB effect on cumulative C release, soil properties and available nutrients within three studies soils.

Soil	n	Cumulative C release	pH	EC <sup>1</sup>	TC	TN	TP	CEC	P	K	Ca	Mg	Cu	Pb	Zn
		mg CO <sub>2</sub> -C kg <sup>-1</sup> soil <sup>-1</sup>		dS m <sup>-1</sup>	%	g kg <sup>-1</sup>	g kg <sup>-1</sup>	coml (+) kg <sup>-1</sup> soil <sup>-1</sup>	g kg <sup>-1</sup>	g kg <sup>-1</sup>	g kg <sup>-1</sup>	g kg <sup>-1</sup>	mg kg <sup>-1</sup>	mg kg <sup>-1</sup>	mg kg <sup>-1</sup>
RHB-A															
3S	15 <sup>2</sup>	2919 B <sup>3</sup>	5.11 C	1.17 B	5.37 A	2.17 A	1.97 B	16.5 C	1.01 B	1.00 B	2.29 C	0.42 C	5.64 B	9.25 A	50.3 C
14S	15	2574 C	5.24 B	2.41 A	5.26 A	1.95 AB	3.76 A	17.5 B	2.39 A	1.24 A	4.23 B	0.81 A	4.72 C	0.97 C	79.5 A
24S	15	3280 A	6.10 A	1.17 B	5.15 A	1.85 B	3.80 A	18.3 A	2.44 A	1.23 A	5.40 A	0.63 B	7.51 A	3.02 B	68.3 B
RHB-B															
3S	15	2834 B	5.10 C	1.13 B	5.75 A	2.07 A	2.15 C	17.0 B	1.03 C	0.90 B	2.49 C	0.43 C	4.63 B	7.56 A	39.7 B
14S	15	2394 C	5.19 B	2.29 A	5.29 B	1.77 B	3.90 B	17.8 A	2.33 B	1.08 A	4.93 B	0.81 A	4.15 C	0.38 C	67.8 A
24S	15	3443 A	5.99 A	1.13 B	5.12 B	1.77 B	4.06 A	18.0 A	2.48 A	1.05 A	5.15 A	0.61 B	7.08 A	2.21 B	69.2 A
RHB-C															
3S	15	2863 B	5.11 C	1.13 C	5.55 A	2.03 B	2.46 B	16.5 B	1.09 C	0.89 B	2.70 C	0.43 C	5.15 B	7.92 A	40.7 C
14S	15	2530 C	5.22 B	2.05 A	5.19 B	1.79 C	3.91 A	18.0 A	2.23 B	1.07 A	5.37 B	0.71 A	4.50 C	0.71 C	70.0 B
24S	15	3124 A	5.97 A	1.23 B	5.48 A	2.31 A	3.80 A	18.2 A	2.35 A	1.05 A	6.29 A	0.62 B	7.90 A	3.01 B	72.7 A
RHB-D															
3S	15	4239 B	5.04 C	1.09 C	4.46 A	2.16 A	2.11 B	16.9 B	1.01 C	0.92 B	2.92 B	0.43 C	6.13 B	9.00 A	47.1 B
14S	15	3505 C	5.17 B	1.87 A	4.27 A	2.28 A	3.74 A	17.2 B	2.44 B	1.17 A	5.04 A	0.69 A	5.05 C	0.77 C	75.1 A
24S	15	5180 A	5.89 A	1.12 B	4.20 A	2.18 A	3.70 A	18.3 A	2.57 A	1.18 A	5.18 A	0.63 B	6.60 A	1.93 B	74.3 A
RHB-E															
3S	15	4190 B	5.02 C	1.18 B	4.51 A	2.14 A	2.20 C	16.9 B	1.13 B	0.96 B	2.60 C	0.49 C	5.53 B	8.71 A	47.3 C
14S	15	3558 C	5.19 B	1.95 A	4.35 A	2.06 A	3.61 B	17.9 A	2.49 A	1.20 A	4.66 B	0.77 A	4.31 C	0.53 C	75.9 A
24S	15	4632 A	5.95 A	1.17 B	4.30 A	2.08 A	4.54 A	18.1 A	2.52 A	1.18 A	5.78 A	0.67 B	7.12 A	2.36 B	69.3 B
RHB-F															
3S	15	3180 B	5.18 C	1.22 B	5.78 A	2.09 A	2.19 C	17.1 A	1.12 C	1.12 C	2.72 C	0.46 B	5.64 B	9.10 A	49.3 C
14S	15	2674 C	5.30 B	2.01 A	5.14 B	1.76 B	4.25 B	17.3 A	2.49 B	1.40 A	4.80 B	0.74 A	4.47 C	0.66 C	76.9 A
24S	15	3586 A	6.07 A	1.27 B	5.65 A	2.17 A	4.90 A	17.4 A	2.67 A	1.36 B	5.89 A	0.69 A	7.05 A	2.48 B	70.1 B

<sup>1</sup> EC = electrical conductivity; TC = total carbon; TN = total nitrogen; TP = total phosphorus; CEC = cation exchange capacity; available nutrients (P, K, Ca, Mg, Cu, Pb and Zn) = Mehlich 3 extraction method; <sup>2</sup> The value is means of five application rate for each RHB (in 3 replicates); <sup>3</sup> Means followed by 18 different capital letter are significantly different for each parameter between three soils ( $P < 0.01$ ).

**Supplemental Table S3.** Results of significant test of RHB effect on cumulative C release, soil properties and available nutrients within five application rates.

Treatment	Cumulative C release	pH	EC <sup>1</sup>	TC	TN	TP	CEC	P	K	Ca	Mg	Cu	Pb	Zn	
RHB	Rate mg CO <sub>2</sub> -C kg <sup>-1</sup> soil <sup>-1</sup>		dS m <sup>-1</sup>	%	g kg <sup>-1</sup>	g kg <sup>-1</sup>	coml (+) kg <sup>-1</sup> soil <sup>-1</sup>	g kg <sup>-1</sup>	g kg <sup>-1</sup>	g kg <sup>-1</sup>	g kg <sup>-1</sup>	mg kg <sup>-1</sup>	mg kg <sup>-1</sup>	mg kg <sup>-1</sup>	
	0.5%	2632 D <sup>2</sup>	5.24 E	1.61 A	2.75 D	3.09 A	3.80 A	17.4 A	2.36 A	0.79 C	3.88 B	0.70 A	7.85 A	5.58 A	83.1 A
	1.0%	2541 D	5.28 D	1.58 AB	2.93 D	3.23 A	3.75 A	17.4 A	2.32 A	0.84 C	3.82 B	0.70 A	7.60 A	5.98 A	85.8 A
<b>RHB-A</b>	4.0%	2825 C	5.36 C	1.54 B	4.03 C	2.95 A	3.68 A	17.3 A	2.25 A	1.17 B	3.86 B	0.67 A	7.96 A	6.80 A	81.3 A
	10%	3049 B	5.65 B	1.58 AB	6.45 B	0.33 B	2.37 B	17.6 A	1.43 B	1.22 B	4.29 A	0.53 B	3.27 B	1.77 B	42.0 B
	20%	3574 A	5.88 A	1.60 A	10.1 A	0.34 B	2.26 B	17.6 A	1.37 B	1.77 A	4.02 AB	0.50 B	3.10 B	1.93 B	37.8 B
	0.5%	2662 C	5.23 D	1.50 A	2.87 D	2.91 A	4.01 A	17.1 CD	2.32 A	0.79 D	4.10 B	0.71 A	6.61 A	4.40 A	75.1 A
	1.0%	2816 B	5.24 D	1.53 A	2.97 D	3.06 A	3.86 A	16.8 D	2.31 A	0.83 D	4.12 B	0.70 A	6.50 A	4.32 A	72.6 A
<b>RHB-B</b>	4.0%	3112 A	5.29 C	1.54 A	4.21 C	2.76 A	3.97 A	17.6 BC	2.29 A	1.07 B	3.78 C	0.70 A	6.78 A	4.81 A	71.9 A
	10%	2853 B	5.59 B	1.54 A	6.56 B	0.32 B	2.71 B	18.1 AB	1.42 B	1.00 C	4.68 A	0.51 B	3.39 B	1.63 B	38.4 B
	20%	3010 A	5.77 A	1.48 A	10.3 A	0.31 B	2.30 C	18.2 A	1.40 B	1.37 A	4.27 B	0.47 C	3.16 B	1.76 B	36.7 B
	0.5%	2692 B	5.23 D	1.52 B	2.82 D	3.38 A	3.76 A	17.0 B	2.23 AB	0.76 C	4.51 B	0.67 A	7.28 A	5.42 A	71.4 B
	1.0%	2800 B	5.32 C	1.41 C	3.00 D	3.03 B	3.81 A	17.3 B	2.26 A	0.78 C	4.54 B	0.64 B	7.43 A	4.94 B	74.0 A
<b>RHB-C</b>	4.0%	2758 B	5.34 C	1.30 D	4.33 C	3.16 B	3.84 A	17.1 B	2.20 B	1.04 B	4.20 B	0.63 B	7.13 A	4.93 B	71.7 B
	10%	2879 B	5.57 B	1.61 A	6.83 B	0.33 C	3.06 B	17.6 B	1.41 C	1.03 B	5.56 A	0.52 C	3.83 B	2.00 C	45.0 C
	20%	3066 A	5.71 A	1.50 B	10.0 A	0.30 C	2.49 C	18.9 A	1.37 C	1.40 A	5.13 A	0.47 D	3.57 B	2.09 C	43.6 C
	0.5%	3124 D	5.20 D	1.27 C	2.56 D	3.26 B	3.76 AB	16.6 C	2.34 A	0.78 D	4.26 BC	0.61 B	7.74 A	5.33 A	79.6 A
	1.0%	3233 D	5.19 D	1.26 C	2.80 D	3.69 A	3.94 A	16.6 C	2.37 A	0.83 C	4.09 BC	0.61 AB	7.29 B	4.87 B	78.6 A
<b>RHB-D</b>	4.0%	3763 C	5.28 C	1.24 C	3.73 C	3.32 B	3.66 B	16.9 BC	2.36 A	1.10 B	3.89 C	0.63 A	7.24 B	4.92 B	75.8 B
	10%	4663 B	5.48 B	1.55 A	5.41 B	0.37 C	2.32 C	17.7 B	1.48 B	1.12 B	5.05 A	0.53 C	3.82 C	2.15 C	47.2 C
	20%	6756 A	5.67 A	1.48 B	7.03 A	0.38 C	2.25 C	19.6 A	1.48 B	1.64 A	4.62 AB	0.53 C	3.54 D	2.23 C	46.2 C
	0.5%	3137 D	5.29 D	1.31 B	2.70 D	3.36 A	3.68 A	16.9 C	2.43 A	0.84 D	4.05 C	0.68 A	7.15 A	5.12 A	75.8 A
	1.0%	3302 D	5.30 D	1.30 B	2.80 D	3.22 A	3.49 A	17.2 BC	2.44 A	0.88 C	4.27 BC	0.68 A	6.98 AB	4.93 A	75.8 A
<b>RHB-E</b>	4.0%	3892 C	5.35 C	1.27 B	3.94 C	3.19 A	3.56 A	17.3 BC	2.43 A	1.12 B	4.16 BC	0.68 A	6.88 B	4.98 A	75.1 A
	10%	4492 B	5.45 B	1.66 A	5.10 B	0.35 B	3.14 A	18.1 AB	1.47 B	1.10 B	4.87 A	0.57 C	3.67 C	2.03 C	48.6 B
	20%	5811 A	5.56 A	1.63 A	7.39 A	0.36 B	3.39 A	18.7 A	1.46 B	1.62 A	4.39 B	0.59 B	3.59 C	2.28 B	45.8 C
	0.5%	3143 B	5.33 CD	1.31 C	2.75 D	3.31 A	4.76 A	17.3 A	2.48 A	0.90 E	4.43 BC	0.70 A	7.04 A	5.15 B	78.2 A
	1.0%	3173 B	5.31 D	1.32 C	2.89 D	2.95 B	4.18 A	17.4 A	2.53 A	0.98 D	3.98 C	0.71 A	7.20 A	5.40 A	78.2 A
<b>RHB-F</b>	4.0%	3374 A	5.37 C	1.45 B	4.09 C	3.02 B	4.12 A	17.4 A	2.51 A	1.36 B	3.99 C	0.68 A	7.09 A	5.54 A	77.7 A
	10%	2944 C	5.66 B	1.72 A	7.02 B	0.36 C	2.83 B	16.6 B	1.47 B	1.30 C	5.16 A	0.53 B	3.64 B	1.97 D	47.7 B
	20%	3100 B	5.92 A	1.71 A	10.9 A	0.39 C	3.01 B	17.6 A	1.47 B	1.93 A	4.80 AB	0.54 B	3.62 B	2.34 C	45.4 C

<sup>1</sup> EC = electrical conductivity; TC = total carbon; TN = total nitrogen; TP = total phosphorus; CEC = cation exchange capacity; available nutrients (P, K, Ca, Mg, Cu, Pb and Zn) = Mehlich 3 extraction method; <sup>2</sup> The value is means (n=9) of three soils for each addition rate (in 3 replicates). Means followed by different capital letter are significantly different for each parameter between treatments (P < 0.01).