

## Effects of Rice Husk Charcoal Application on Rice Yield, Methane Emission, and Soil Carbon Sequestration in Andosol Paddy Soil

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### Abstract

Biochar application is an effective option for promoting soil carbon sequestration. Rice husk charcoal (RC) produced from pyrolysis of rice husk (RH) is characterized by a higher silicon content as compared to wood-derived biochar. A study was conducted using pots to evaluate the short-term effects of RC application at 0, 0.4, 2.0, 4.0, 20.0, and 40.0 g pot<sup>-1</sup> (CONT, RC0.4, RC2, RC4, RC20, and RC40, respectively), or RH application at 4.0 g pot<sup>-1</sup> (RH4) on rice yield, methane (CH<sub>4</sub>) emission, and soil carbon sequestration in an Andosol paddy soil. The results showed that the significant increase in brown rice yield with RC40 was attributed to increases in the grain number and percentage of ripened grains. Plants treated with RC20 or RC40 absorbed greater amounts of silicon than those treated with lower RC rates. RC application did not significantly increase CH<sub>4</sub> emission. The soil carbon content after rice cultivation increased in proportion to the RC application rate. The balance between cumulative CH<sub>4</sub> emission and soil carbon sequestration, based on the 100-year global warming potential, suggested that RC40 served as CO<sub>2</sub>-equivalent sink. No differences were observed in the measured parameters between RH4 and RC4. These findings suggest that the higher rates of RC application in the RC20 and RC40 treatments function as a silicon fertilizer while promoting soil carbon sequestration.

**Discipline:** Agricultural environment, Crop production

**Additional key words:** biochar, C-N ratio, global warming, greenhouse gas emissions, silicon

### Introduction

Biochar is a solid material processed from the thermochemical conversion of biomass under oxygen-limited conditions (Laufer & Tomlinson 2013). The carbon (C) in biochar persists in the soil for long periods, and biochar application is an effective option for enhancing soil C sequestration (Lehmann et al. 2009). However, the mitigating effect of biochar application on global warming remains controversial as it may affect the production and consumption of greenhouse gases (GHGs) stronger than CO<sub>2</sub>, such as

methane (CH<sub>4</sub>) and nitrous oxide (N<sub>2</sub>O), in soil. Its effect on GHG emissions varies depending on such conditions as soil type, fertility, and moisture content, as well as the feedstock and pyrolysis temperature used for producing the biochar (Lehmann et al. 2009). There is also little consensus on how biochar affects crop production. Several studies have reported that biochar application increased crop yield by improving the physicochemical properties of soil (Glaser et al. 2002, Lehmann & Rondon 2006), whereas other studies have observed a reduction in grain yield due to N limitation caused by the high C-N ratio of biochar (Asai et al. 2009).

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Thus, a mechanistic understanding of how biochar application causes changes in crop yield is essential to guide the promotion of biochar application on agricultural lands.

Rice husk charcoal (RC) processed from pyrolysis of rice husk (RH) is considered to be one of the most cost-effective biochars used in rice-based farming systems (Ogawa & Okimori 2010). RH is generated as a by-product of rice production, equivalent to 22% of the brown rice yield (Ogawa et al. 1988). RC has been used for many years by Japanese farmers as a soil amendment for improving the physical properties of soil. It is also recognized as an economically viable source of several silicon (Si) compounds (Sun & Gong 2001). The traditional use of RC in the Japanese agricultural sector has been on a fairly small scale as a material for the composting, nursery media, mulch, and underdrainage of agricultural land. New perspectives on biochar use suggest that RC application may increase crop productivity and C sequestration in soil. However, information on the effects of RC application on rice yield and global warming is limited and varied (Haefele et al. 2011, Knoblauch et al. 2011, Koyama et al. 2015). Thus, the extent to which the rate of RC application to paddy fields increases the rice yield and promotes global warming remains unclear.

We conducted a study in pots to evaluate the translocation and distribution of RC-derived nutrients and their effects on rice productivity. Andosol paddy soil was used for the experiment, as it is the second largest type of paddy soil in Japan after Gleysols (Takata et al. 2011). This soil is characterized by a relatively high C content (Takata et al. 2013) and low levels of CH<sub>4</sub> emitted under anaerobic conditions (Yagi & Minami 1990), among the Japanese soil types. Our study aimed to address three specific questions: (i) How does RC influence rice growth and yield? (ii) Can RC application with the aim of soil C sequestration have a positive impact on CO<sub>2</sub>-equivalent balance despite its additional CH<sub>4</sub> emission? (iii) Is there a certain rate of RC application that can achieve both higher rice yield and the mitigation of global warming?

## Materials and Methods

### 1. Experimental design

An experiment using pots was conducted under a

plastic-film greenhouse at the Agricultural and Forestry Research Center (AFRC), University of Tsukuba, Japan (36°12'N, 140°09'E, 25 m above sea level) from April to October 2012. Sieved (< 5 mm) wet paddy soil (34.4% moisture content) equivalent to 1.64 kg of dry soil was placed in a Wagner pot (0.02 m<sup>2</sup> of surface area). The soil was collected from the top 15-cm layer of a paddy field at the AFRC, and is classified as a "Haplic Andosol" by the Food and Agriculture Organization of the United Nations (FAO) and the United Nations Educational, Scientific, and Cultural Organization (UNESCO) system (FAO-UNESCO 1990). The soil pH, nitrogen (N) and C contents were 5.81, 4.52 g N kg<sup>-1</sup> dry soil and 58.7 g C kg<sup>-1</sup> dry soil, respectively. The treatments comprised six RC application rates of 0, 0.4, 2.0, 4.0, 20.0, and 40.0 g pot<sup>-1</sup> (CONT, RC0.4, RC2, RC4, RC20, and RC40, respectively), and one RH application rate at 4.0 g pot<sup>-1</sup> (RH4). The RC application rate in RC0.4 is equivalent to 20 g m<sup>-2</sup>, which is the average RC yield from paddy fields in Japan, as 100 kg of RH is converted into about 20 kg of RC (Ogawa et al. 1988, Senoo et al. 2009). We set up higher RC application rates up to RC40 (40.0 g pot<sup>-1</sup>) based on the assumption that increasing the amount of applied RC increases the amount of soil C sequestration. RH4 was included to compare the effects of carbonized and un-carbonized RH. Produced by pyrolysis at 350-400°C for 15 min, the RC was purchased from Pros Co. Ltd., Nagano, Japan. The RH was collected from a country elevator near the experimental site. Table 1 lists the physicochemical characteristics of the RC and RH. The treatments were arranged in a completely randomized design with eight replicates for measuring rice growth and yield parameters. CH<sub>4</sub> fluxes during rice cultivation and soil chemical properties after rice harvest were measured in triplicate using three pots from each treatment, and rice plants at maturity were subjected to chemical analysis using the other three pots of each treatment.

### 2. Crop management

According to a given treatment and six days before transplanting, RC or RH was thoroughly mixed into all layers of the pot soil. The basal compound fertilizer was also applied on the same day at 0.42 g pot<sup>-1</sup> of nitrogen (N), phosphoric acid (P<sub>2</sub>O<sub>5</sub>), and potassium (K<sub>2</sub>O). Two

**Table 1. Physicochemical characteristics of rice husk charcoal (RC) and rice husk (RH) used for the experiment.**

Material	Bulk density (g cm <sup>-3</sup> )	pH (H <sub>2</sub> O)	EC (mS m <sup>-1</sup> )	Total N	Total C	Si			
						K	Ca	Mg	(g kg <sup>-1</sup> )
RC	0.13±0.01	8.40±0.45	35.8±12.7	4.01±0.27	427±23	192.0±15.2	8.71±0.39	2.17±0.15	0.59±0.04
RH	0.11±0.00	6.11±0.20	76.5±16.8	3.36±0.52	377±5	96.7±14.0	4.52±1.04	0.95±0.13	0.43±0.21

EC: electrical conductivity; N: nitrogen; C: carbon; Si: silicon; K: potassium; Ca: calcium; Mg: magnesium.

Values represent mean ± standard deviation (n = 3).

rice seedlings (*Oryza sativa* L. cv. Nipponbare) at the 3-4 leaf stage were transplanted as one hill in each pot on 26 May 2012. During cultivation, irrigation water was added continuously to maintain submerged conditions in the pot soil. Topdressing was applied to all pots at a rate of 0.315 g N pot<sup>-1</sup> at 63 days after transplanting (DAT) using ammonium sulfate. Final drainage was carried out at 120 DAT and the aboveground parts of rice plants were harvested at the maturity stage at 127 DAT. The daily mean air temperature at a height of 1.5 m inside the plastic-film greenhouse ranged from 16.9°C to 33.4°C during the experiment. The mean soil temperatures at a depth of 5 cm in two pots of each treatment did not differ significantly among the treatments, and ranged from 17.8°C to 31.9°C during the experiment.

### 3. Measurements

The CH<sub>4</sub> flux was measured in triplicate using a closed chamber method (Minamikawa et al. 2015). Each pot was covered with a transparent acrylic chamber that was 1.0 m in height with a bottom area of 0.3 m × 0.3 m, and surrounded by water for sealing. One-mL gas samples were collected at intervals of one to two weeks (between 09:00 and 12:00) from 28 to 120 DAT (for a total of 11 times). The CH<sub>4</sub> flux was calculated as a temporal change in the gas concentration at 1, 11, and 21 min after placement in the chamber, using a gas chromatograph equipped with a flame ionization detector (GC-8A, Shimadzu Corp., Kyoto, Japan). The soil redox potential (Eh) at a depth of 5 cm was monitored in the same three pots using platinum-tipped electrodes inserted into the soil and a portable Eh meter (PRN-41; Fujiwara Scientific Co., Ltd, Tokyo, Japan). Cumulative CH<sub>4</sub> emission was estimated using the trapezoidal method by interpolation between the measurement days during the rice growing season. The cumulative CH<sub>4</sub> emission was converted into the CO<sub>2</sub>-equivalent by using the global warming potential (GWP) along with climate-carbon feedback for a 100-year time horizon: 1 g of CH<sub>4</sub> is equivalent to 34 g of CO<sub>2</sub> (Myhre et al. 2013). Yield-scaled CH<sub>4</sub> emission was calculated by dividing the cumulative CH<sub>4</sub> emission by the brown rice yield at a moisture content of 15%.

Soil samples were collected after the rice harvest and sieved (< 2 mm) for chemical analysis. Soil pH (1:2.5, soil:H<sub>2</sub>O) and electrical conductivity (EC; 1:5, soil:H<sub>2</sub>O) were determined using a pH meter (HM-30R; DKK-TOA Corp., Tokyo, Japan) and an EC meter (CM-30R; DKK-TOA Corp., Tokyo, Japan). To measure pH and EC in the RC and RH, a 1:10 ratio of the material to deionized water was adopted due to the small bulk densities of RC and RH. RC and RH were digested with a mixture of sulfuric acid and hydrogen peroxide, and potassium, calcium, and magnesium concentrations were analyzed using an atomic

absorption photometer (Z-2300; Hitachi High-Technologies Corp., Tokyo, Japan). The total N and C contents of the soil, RC, and RH were analyzed using the dry-combustion method with an NC analyzer (NC-220F; Sumika Chemical Analysis Service, Ltd., Osaka, Japan). Soil C sequestration was calculated using the following equation:

$$\text{Soil C sequestration (g C pot}^{-1}\text{)} = (\text{C}_{\text{tre}} - \text{C}_{\text{bef}}) \times \text{DW}$$

where C<sub>tre</sub> denotes the soil C content of the treatment after rice cultivation (g kg<sup>-1</sup> dry soil), C<sub>bef</sub> is the initial soil C content at the start of the experiment (58.7 g kg<sup>-1</sup> dry soil), and DW is the dry soil weight in the Wagner pot of the treatment (kg pot<sup>-1</sup>). The CO<sub>2</sub>-equivalent balance between the cumulative CH<sub>4</sub> emission and soil C sequestration was calculated to compare the short-term effects of the RC and RH applications.

The harvested aboveground parts of rice plants were air-dried, and then the culm and panicle lengths of the longest culm in each pot were measured. The fully ripened grains were separated from the threshed grains using a saline solution with a specific gravity of 1.06. The yield was defined as the weight of fully ripened brown rice with a water content of 15%. The roots were separated from the soil by rinsing in water. The rice straw and roots were dried in an oven at 80°C for 48 h for measuring the dry weight of each. The dried rice plant was ground to a fine powder (< 2 mm) using a cutting mill (SM-100; Retsch GmbH, Haan, Germany). The total N and C contents of the rice plant were determined using the same method as that used for soil analysis. The Si content in the straw sample, as well as RC and RH, was determined using gravimetric analysis as described by Saito et al. (2005). The N and Si uptakes and C assimilation by the rice plants were calculated by multiplying the nutrient concentrations and the rice panicle or straw weight for each plant.

### 4. Statistical analyses

Statistical analyses were conducted using JMP 8 software (SAS Institute Japan Inc., Tokyo, Japan). To estimate statistical significance, a one-way analysis of variance (ANOVA) and Tukey's honest significant difference (HSD) test at the 5% level were used.

## Results

### 1. Growth, yield, and yield components of rice plants

Among the growth parameters, culm length and straw yield increased with higher RC application rates (Table 2). Among the yield parameters, the grain number per panicle, the percentage of ripened grains, and brown rice yield increased with higher RC application rates. These parameters were significantly higher for RC40 than for CONT. In contrast, the 1000-grain weight decreased with higher RC application rates. RC40 produced significantly

lower 1000-grain weight than the other treatments. Panicle length, panicle number, and root weight did not differ significantly among treatments. No significant differences between RC4 and RH4 were observed in the growth and yield parameters.

## 2. Nutrient concentrations and uptakes in rice plants

The N concentration of mature rice plants did not differ significantly among the treatments (Table 3). The C concentration in rice straw decreased with higher RC application rates. In contrast, the Si concentration in rice straw increased with higher RC application rates. The RC20 and RC40 treatments produced significantly lower C concentrations and higher Si concentrations than the CONT treatment. The N uptake and C assimilation by the rice plants did not differ significantly among treatments (Table 3) because the decreases in C concentration in the rice plants were compensated for by increases in the rice straw weight caused by the RC20 and RC40 treatments (Table 2). Silicon uptake by the rice plants was significantly enhanced by RC20 and RC40. The Si uptake values for the RC20 and RC40 treatments were 196% and 350%, respectively, of that for CONT (Table 3).

## 3. Soil chemical properties and C sequestration

Soil pH, EC, and N content after rice cultivation did not differ significantly among treatments (Table 4). However, soil C content after rice cultivation increased with higher RC application rates. The RC20 and RC40 treatments produced significantly higher soil C contents than did CONT. As a result, soil C-N ratios were significantly higher for RC20 and RC40 than for the other treatments. The increase in the soil C content from the initial value

to that after the rice harvest represents the amount of soil C sequestration. The amounts of soil C sequestration for RC20 and RC40 were 550% and 851%, respectively, of that for CONT (Table 5).

## 4. Soil redox potential (Eh) and CH<sub>4</sub> emission

Soil Eh values gradually decreased and remained steady at approximately  $-200$  mV from about 50 DAT until rice harvesting (Fig. 1a). No significant differences were observed in soil Eh among the treatments throughout the measurement period. When soil Eh decreased below  $-150$  mV, CH<sub>4</sub> fluxes began to increase in all treatments until the heading stage of the plants (around 70 DAT) (Fig. 1b). CH<sub>4</sub> fluxes did not differ significantly among RC application treatments, except at 77 DAT when the RC40 treatment caused significantly higher CH<sub>4</sub> flux than the other treatments. The cumulative CH<sub>4</sub> emission increased in the RC application treatments by between 9% and 24% of that in the CONT (Table 5). The increases were not significant or proportional to the RC application rates. CH<sub>4</sub> fluxes in response to the RH4 treatment significantly increased and remained at relatively higher levels than those of the other treatments from 91 to 113 DAT. Consequently, the cumulative CH<sub>4</sub> emission for RH4 was 17% higher than that for RC4, although differences between the treatments were not significant.

## 5. Yield-scaled CH<sub>4</sub> emission, and CO<sub>2</sub>-equivalent balance between CH<sub>4</sub> emission and soil C sequestration

The yield-scaled CH<sub>4</sub> emission was the highest for RH4 (50.8 g CH<sub>4</sub> kg<sup>-1</sup> of brown rice yield) and the lowest for RC40 (31.0 g CH<sub>4</sub> kg<sup>-1</sup> of brown rice yield), although

**Table 2. Effects of rice husk charcoal (RC) and rice husk (RH) applications on growth, yield and yield components of rice plants.**

Treatment	Culm length (cm)	Panicle length (cm)	Rice biomass yield (g DW pot <sup>-1</sup> )		Panicle number (pot <sup>-1</sup> )	Grain number		Percentage of ripened grains (%)	1000-grain weight (g)	Brown rice yield (g pot <sup>-1</sup> )
			Straw	Root		(panicle <sup>-1</sup> )	(pot <sup>-1</sup> )			
CONT	61.9 b	20.9 a	51.3 b	16.1 a	40.8 a	70.2 ab	2846 b	69.8 b	24.0 a	47.5 b
RC0.4	61.9 b	20.3 a	53.4 b	16.3 a	42.6 a	66.0 b	2805 b	70.9 ab	23.9 ab	47.4 b
RC2	63.8 ab	20.7 a	53.6 b	17.4 a	43.5 a	68.5 ab	2980 ab	71.8 ab	23.9 ab	51.1 b
RC4	63.8 ab	20.2 a	53.2 b	16.1 a	43.1 a	68.5 ab	2946 ab	71.1 ab	23.7 ab	49.5 b
RC20	63.8 ab	20.8 a	56.9 ab	16.5 a	43.1 a	70.5 ab	3035 ab	76.0 ab	23.6 ab	54.4 ab
RC40	66.2 a	21.4 a	61.7 a	17.0 a	42.5 a	75.7 a	3205 a	82.7 a	23.3 b	61.8 a
RH4	60.2 b	20.6 a	51.8 b	16.0 a	42.4 a	67.5 b	2852 b	74.0 ab	23.8 ab	50.3 b

Rice straw and root weight was indicated on a dry weight basis. 1000-grain weight and brown rice yield were adjusted to have a moisture content of 15%. CONT: control treatment without any application of RC and RH. Means (n = 8) followed by the same letter within each column were not significantly different at the 5% level in Tukey's HSD test.

**Table 3. Effects of rice husk charcoal (RC) and rice husk (RH) applications on nutrient concentration, uptake and assimilation in rice plants at the mature stage.**

Treatment	Panicle		Straw					
	N conc. (%)	N uptake (g pot <sup>-1</sup> )	Conc. (%)			Uptake (g pot <sup>-1</sup> )		C assimilation (g pot <sup>-1</sup> )
			N	Si	C	N	Si	
CONT	1.23 a	0.89 a	0.46 a	1.49 c	45.6 a	0.23 a	0.74 c	23.1 a
RC0.4	1.26 a	0.87 a	0.47 a	1.56 c	45.7 a	0.26 a	0.84 c	24.8 a
RC2	1.24 a	0.94 a	0.45 a	1.48 c	45.6 a	0.24 a	0.78 c	24.3 a
RC4	1.19 a	0.89 a	0.48 a	1.52 c	45.3 a	0.26 a	0.84 c	24.8 a
RC20	1.16 a	0.90 a	0.45 a	2.77 b	44.0 b	0.23 a	1.45 b	22.8 a
RC40	1.12 a	0.97 a	0.37 a	4.20 a	42.4 c	0.23 a	2.59 a	26.1 a
RH4	1.24 a	0.90 a	0.49 a	1.72 c	45.7 a	0.27 a	0.93 c	24.7 a

Conc.: concentration, N: nitrogen, C: carbon, Si: silicon, CONT: control treatment without any application of RC and RH. Means (n = 3) followed by the same letter within each column were not significantly different at the 5% level in Tukey's HSD test.

**Table 4. Effects of rice husk charcoal (RC) and rice husk (RH) applications on soil chemical properties after rice cultivation.**

Treatment	pH (H <sub>2</sub> O)	EC (mS m <sup>-1</sup> )	Total N	Total C	C-N ratio
			(g kg <sup>-1</sup> of dry soil)		
CONT	5.72 a	77.9 a	4.56 a	60.0 c	13.2 c
RC0.4	5.69 a	80.8 a	4.52 a	59.8 c	13.2 c
RC2	5.70 a	74.7 a	4.63 a	61.5 bc	13.3 c
RC4	5.70 a	76.8 a	4.53 a	60.4 bc	13.3 c
RC20	5.69 a	78.8 a	4.70 a	65.8 ab	14.0 b
RC40	5.78 a	70.1 a	4.61 a	69.5 a	15.1 a
RH4	5.62 a	87.5 a	4.73 a	62.1 bc	13.1 c

EC: electrical conductivity; N: nitrogen; C: carbon; C-N: carbon to nitrogen ratio. CONT: control treatment without any application of RC and RH. Means (n = 3) followed by the same letter within each column were not significantly different at the 5% level in Tukey's HSD test.

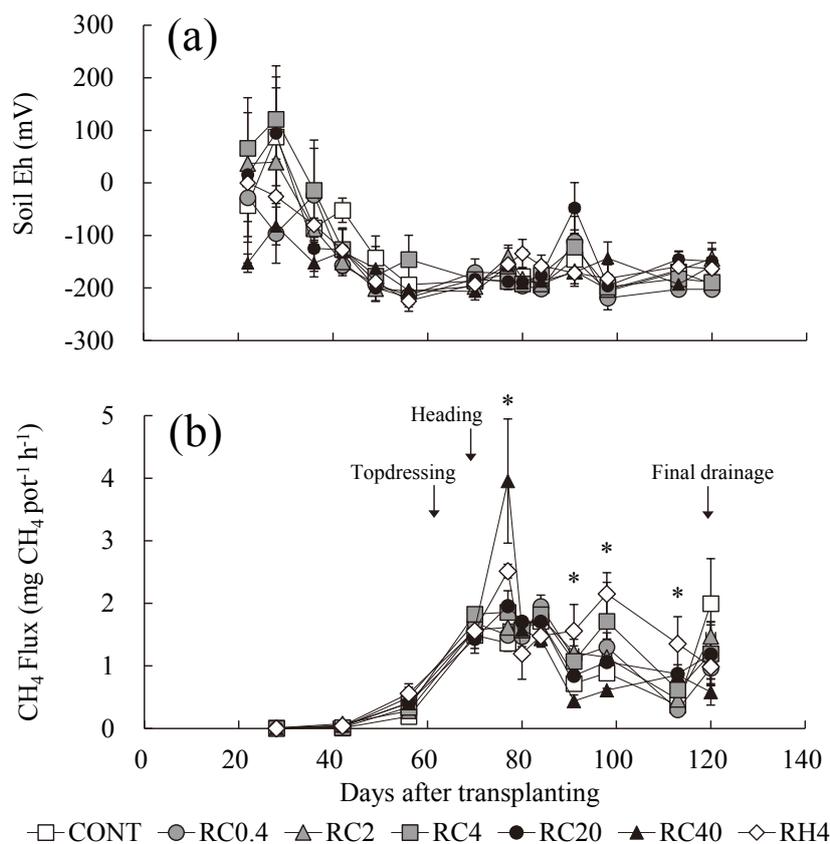
not significantly different from CONT (Table 5). The CO<sub>2</sub>-equivalent balance indicated that RC40 functioned as a CO<sub>2</sub>-equivalent sink, whereas the other treatments served as a CO<sub>2</sub>-equivalent source. The differences from CONT were not significant when the RC application rate was less than or equal to 20.0 g pot<sup>-1</sup> (RC0.4, RC2, RC4 and RC20). The yield-scaled CH<sub>4</sub> emission and CO<sub>2</sub>-equivalent balance between RH4 and RC4 did not differ significantly.

## DISCUSSION

### 1. Effect of RC application rate on rice productivity

The RC40 treatment caused significant increases in

both brown rice and straw yield compared to the CONT treatment (Table 2). The increase in brown rice yield was attributed to significant increases in the grain number and percentage of ripened grains, although a decrease in the 1000-grain weight was observed for RC40. Haefele et al. (2011) showed varying effects of RC application on paddy rice yield, depending on such site characteristics as cation exchange capacity (CEC), water retention capacity and N availability in the soil. The soil in the pots used in this study did not undergo nutrient leaching and water stress, however, because the soil was maintained under submerged conditions until 120 DAT with the recommended amounts of basal and topdressing fertilizers. Although not measured



**Fig. 1. Seasonal variations in soil redox potential (Eh) (a) and methane (CH<sub>4</sub>) flux (b) during rice cultivation as affected by rice husk charcoal (RC) and rice husk (RH) applications.**

Soil Eh was monitored at a pot soil depth of 5 cm. CONT shows the control treatment without any application of RC and RH. Asterisks (\*) in the CH<sub>4</sub> flux denote a significant difference between treatments by ANOVA at the 5% level. Vertical bars represent standard errors of the mean values (n = 3).

**Table 5. Effects of rice husk charcoal (RC) and rice husk (RH) applications on cumulative methane (CH<sub>4</sub>) emission and soil carbon (C) sequestration, and their carbon dioxide (CO<sub>2</sub>) equivalent balance.**

Treatment	Cumulative CH <sub>4</sub> emission (g CH <sub>4</sub> pot <sup>-1</sup> )	Soil C sequestration (g C pot <sup>-1</sup> )	Yield-scaled CH <sub>4</sub> emission (g CH <sub>4</sub> kg <sup>-1</sup> of brown rice yield)	CO <sub>2</sub> -equivalent balance (g CO <sub>2</sub> -eq pot <sup>-1</sup> )
CONT	1.76 a	2.13 c	37.1 ab	-52.0 b
RC0.4	1.93 a	1.84 c	40.8 ab	-59.0 b
RC2	1.97 a	4.56 bc	38.6 ab	-50.4 b
RC4	2.18 a	2.79 c	44.0 ab	-63.7 b
RC20	1.97 a	11.71 ab	36.2 ab	-24.0 ab
RC40	1.92 a	18.13 a	31.0 b	1.3 a
RH4	2.55 a	5.59 bc	50.8 a	-66.4 b

Cumulative CH<sub>4</sub> emission was estimated using the trapezoidal method by interpolation between the measurement days during the rice growing period. Yield-scaled CH<sub>4</sub> emission = [Cumulative CH<sub>4</sub> emission] / [Brown rice yield with a moisture content of 15%]. CO<sub>2</sub>-equivalent balance = [CO<sub>2</sub>-equivalent in soil C sequestration] - [CO<sub>2</sub>-equivalent in CH<sub>4</sub> emission]. Negative figures in CO<sub>2</sub>-equivalent balance indicate that the effect of CH<sub>4</sub> emission is greater than that of soil C sequestration at rice harvesting, and positive figures indicate vice versa. CONT: control treatment without any application of RC and RH. Means (n = 3) followed by the same letter within each column were not significantly different at the 5% level in Tukey's HSD test.

in this study, CEC and water retention capacity were probably not limiting factors affecting the yield of brown rice. Our results did not show any marked enhancement in other soil chemical properties such as pH, EC, or total N content; however, the results indicated a significant increase in the C-N ratios for the RC40 treatment (Table 4), which could have caused the immobilization of N in the soil and led to a lower rice yield, as reported by Asai et al. (2009). The culm length, straw yield, grain number per pot, and percentage of ripened grains in RC40 showed higher values than those of the CONT plants (Table 2), suggesting that the rice plants were not subject to N limitation during cultivation. An explanation for the increase in rice yield can be drawn from nutrient analysis of the rice plants that revealed significantly higher Si uptake by the plants for the RC20 and RC40 treatments (Table 3). Imaizumi & Yoshida (1958) found that Si application significantly increased the rice yield when the Si content of the rice plants was less than 5% (equivalent to 11% SiO<sub>2</sub>). Rice plants grown in pots generally have lower Si content than those grown in a paddy field because the plants in pots grow larger without light and space competition with other plants, and the small volume of pot soil results in less Si being supplied to the plant. In a previous field study on the same paddy soil type (Koyama et al. 2015), no significant effect on yield was observed among the treatments. The rice plants of the control plots were presumably supplied with enough Si from the soil and irrigated water; thus, additional Si from RC application did not impact the rice yield in field experiments. In this study, the Si concentration in the CONT treatment plants was as low as 1.49% (Table 3). Thus, we speculate that the RC and RH applications functioned as Si fertilizer for plants with low Si content. Silicon uptake by rice plants improves photosynthesis by suppressing excessive transpiration and enhancing the light-intercepting structure of rice plants growing in a community (Ma et al. 1989, Hossain et al. 1999). Mori & Fujii (2009) reported that Si application increased the rice grain number through enhanced N acquisition and dry matter production at the panicle formation stage. In addition, other studies demonstrated that Si application reduced the root toxicity of certain minerals such as manganese and iron, especially at the ripening stage, and hence increased the percentage of ripened grains (Ma et al. 1989, Uchimura et al. 2000). In our study, RC40 showed increases in Si uptake, grain number and the percentage of ripened grains (Table 2 and 3). The higher Si uptake for RC40 compared to that for the CONT accounted for 24% of the total amount of Si supplied from the RC. This Si recovery percentage from RC is similar to the range of slag Si fertilizer at 21-38% (Ando et al. 1988). Increases in Si concentration up to 4.2% in rice plants were reported to significantly improve the mechanical strength of culms and leaf sheaths (Hossain et al. 1999). These results suggest that RC40 may provide

such additional beneficial effects as resistance to fungus and insect damage, and tolerance to lodging.

## 2. Effect of RC application rate on CO<sub>2</sub>-equivalent balance

Although biochar C has reportedly been recalcitrant in soil for hundreds to thousands of years, some of its labile components are vulnerable to abiotic and microbial oxidation (Zimmerman 2010). We demonstrated that RC application increased soil C sequestration proportional to the RC application rate, while the cumulative CH<sub>4</sub> emissions from the RC application treatments did not differ significantly from that of the CONT (Table 5). Larger amounts of CH<sub>4</sub> emission per unit area were observed in the present experiment when compared to our previous field experiment (Koyama et al. 2015) because draining practices, which reduce CH<sub>4</sub> emission from rice paddy fields (Yagi et al. 1996), were not employed. In addition, rice plants, through which CH<sub>4</sub> is transported from the soil to the atmosphere (Inubushi et al. 1989), produced a greater number of stems in the pot experiments. Similar to our previous field experiment, the present pot experiment demonstrated that RC application proportionally increased the soil C content, but did not directly enhance CH<sub>4</sub> emission from Andosol paddy soil. Zhang et al. (2012) argued that disproportionately lower CH<sub>4</sub> emission at a higher biochar rate could be explained by the effect of N limitation on methanogens due to the high C-N ratio in the biochar-amended soil (14.2 C-N ratio at 40 Mg ha<sup>-1</sup>). In this study, the RC20 and RC40 treatments showed significantly higher C-N ratios (14.0 and 15.1, respectively) than the other treatments (Table 4), although there were no significant effects on CH<sub>4</sub> emission among the treatments (Table 5). The mechanism for relatively lower CH<sub>4</sub> emission at higher RC application rates remains unclear. The CO<sub>2</sub>-equivalent balance gradually improved with higher RC application rates and reached a positive value in RC40 (Table 5). The specific difference in RC application rates between RC20 and RC40 caused the CO<sub>2</sub>-equivalent balance to shift from a source to a sink.

The results also showed that RH4 and RC4 did not differ in CH<sub>4</sub> emission and soil C sequestration in this study. RC contains approximately 13% more C than RH (Table 1), but reportedly contains fewer labile components than RH (Shackley et al. 2012). The amount of C used as a source for CH<sub>4</sub> emission and soil C sequestration showed little difference between RH4 and RC4. The effect of pyrolysis of RH on CH<sub>4</sub> emission and soil C sequestration would be more pronounced at higher application rates of RH and RC, as Knoblauch et al. (2011) concluded that RC application to paddy fields reduces CH<sub>4</sub> emission by as much as 80% compared to RH application at 41 Mg ha<sup>-1</sup>.

## Conclusion

This study demonstrated that RC application in Andosol paddy soil improved the short-term soil carbon budget and rice yield. RC supplied significant amounts of C and Si to the paddy soil. The Si uptake by rice plants increased significantly due to the RC20 and RC40 treatments. RC40 significantly enhanced both the grain number and percentage of ripened grains in the rice plants, thereby resulting in a significant increase in rice yield. The CO<sub>2</sub>-equivalent balance between cumulative CH<sub>4</sub> emission and soil C sequestration indicated that RC40 functioned as a CO<sub>2</sub>-equivalent sink, while the other treatments served as a CO<sub>2</sub>-equivalent source (Table 5). The RC40 application rate is approximately a hundred times greater than the average yield of RC from Japanese paddy fields. In practice, RC40 (equivalent to 20 Mg ha<sup>-1</sup> of RC) should be applied once over a period of several decades, or by using smaller RC amounts in continuous applications. Long-term field studies are needed to determine the optimum RC application rates and frequencies for increasing rice yield and enhancing the CO<sub>2</sub>-equivalent balance under different soil types and environmental conditions.

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## References

- Ando, J. et al. (1988) Studies on structure, solubility, and agronomic response of industrial slags (Part 5): Effects of alumina on solubility and agronomic response of slags. *Nihon dojo hiryo gaku zashi (Jpn. J. Soil Sci. Plant Nutr.)*, **59**, 27-32 [In Japanese with English summary].
- Asai, H. et al. (2009) Biochar amendment techniques for upland rice production in Northern Laos: 1. Soil physical properties, leaf SPAD and grain yield. *Field Crops Res.*, **111**, 81-84.
- FAO-UNESCO (1990) *Soil map of the world: revised legend. World soil resources report 60*. FAO. Rome, pp140.
- Glaser, B. et al. (2002) Ameliorating physical and chemical properties of highly weathered soils in the tropics with charcoal - a review. *Biol. Fert. Soils*, **35**, 219-230.
- Haefele, S.M. et al. (2011) Effects and fate of biochar from rice residues in rice-based systems. *Field Crops Res.*, **121**, 430-440.
- Hossain, K.A. et al. (1999) Effects of powdered rice chaff application on Si and N absorption, lodging resistance and yield in rice plants (*Oryza sativa* L.). *Plant Prod. Sci.*, **2**, 159-164.
- Imaizumi, K. & Yoshida, S. (1958) Edaphological studies on silicon supplying power of paddy fields. *Nougyou gijyutu kenkyujo houkoku B (Bull. Nat. Inst. Agric. Sci., Ser. B.)*, **8**, 261-304 [In Japanese with English summary].
- Inubushi, K. et al. (1989) Methane emission from the flooded paddy soil to the atmosphere through rice plant. *Nihon dojo hiryo gaku zashi (Jpn. J. Soil Sci. Plant Nutr.)*, **60**, 318-324 [In Japanese with English summary].
- Knoblauch, C. et al. (2011) Degradability of black carbon and its impact on trace gas fluxes and carbon turnover in paddy soils. *Soil Biol. Biochem.*, **43**, 1768-1778.
- Koyama, S. et al. (2015) Increase in soil carbon sequestration using rice husk charcoal without stimulating CH<sub>4</sub> and N<sub>2</sub>O emissions in an Andosol paddy field in Japan. *Soil Sci. Plant Nutr.*, **61**, 873-884.
- Laufer, J. & Tomlinson, T. (2013) Biochar field studies: an IBI research summary. [http://www.biochar-international.org/sites/default/files/IBI\\_Field\\_Studies\\_Final\\_May\\_2013.pdf](http://www.biochar-international.org/sites/default/files/IBI_Field_Studies_Final_May_2013.pdf).
- Lehmann, J. & Rondon, M. (2006) Biochar soil management on highly weathered soil in the humid tropics. In *Biological Approaches to Sustainable Soil Systems*, eds. Uphoff, N. et al., CRC press, Boca Raton, FL, 517-530.
- Lehmann, J. et al. (2009) Stability of biochar in soil. In *Biochar for Environmental Management: Science and Technology*, eds. Lehmann J. & Joseph, J., Earthscan, London, 183-206.
- Ma, J.F. et al. (1989) Effect of silicon on the growth of rice plant at different growth stages. *Soil Sci. Plant Nutr.*, **35**, 347-356.
- Minamikawa, K. et al. (2015) Guidelines for measuring CH<sub>4</sub> and N<sub>2</sub>O emissions from rice paddies by a manually operated closed chamber method. National Institute for Agro-Environmental Sciences, Tsukuba, Japan, pp.76.
- Mori, S. & Fujii, H. (2009) Effectiveness of silica application at the panicle formation stage on the rice production. *Nihon dojo hiryo gaku zashi (Jpn. J. Soil Sci. Plant Nutr.)*, **80**, 136-142 [In Japanese with English summary].
- Myhre, G. et al. (2013) Anthropogenic and natural radiative forcing. In *Climate Change 2013: The Physical Science Basis. Contribution of Working Group I to the Fifth Assessment Report of the Intergovernmental Panel on Climate Change*, eds. Stocker, T.F. et al., Cambridge University Press, Cambridge, United Kingdom and New York, NY, USA. 659-740.
- Ogawa, K. et al. (1988) Biomass production and the amounts of absorbed inorganic elements by crops in arable lands in Hokkaido, and its evaluation. *Hokkaido nougyou sikenjo houkoku (Res. Bull. Hokkaido Natl. Agric. Exp. Stn.)*, **149**, 57-91 [In Japanese with English abstract].
- Ogawa, M. & Okimori, Y. (2010) Pioneering works in biochar research, Japan. *Aust. J. Soil Res.*, **48**, 489-500.
- Saito, K. et al. (2005) Rapid, micro-methods to estimate plant silicon content by dilute hydrofluoric acid extraction and spectrometric molybdenum method: I. Silicon in rice plants and molybdenum yellow method. *Soil Sci. Plant Nutr.*, **51**, 29-36.

- Senoo, K. et al. (2009) Calculation of CO<sub>2</sub> discharge in the rice husk carbonization plant at construction and operation. *Japan Society of Civil Engineers*, <http://www.library.jsce.or.jp/jsce/open/00064/2009/51-07-0005.pdf> [In Japanese].
- Shackley, S. et al. (2012) Sustainable gasification biochar systems? A case-study of rice-husk gasification in Cambodia, Part I: Context, chemical properties, environmental and health and safety issues. *Energy Policy*, **42**, 49-58.
- Sun, L. & Gong, K. (2001) Silicon-based materials from rice husks and their applications. *Ind. Eng. Chem. Res.*, **40**, 5861-5877.
- Takata, Y. et al. (2011) Process of the decline in the cultivated soil area with land use changes in Japan. *Nihon dojo hiryo gaku zashi (Jpn. J. Soil Sci. Plant Nutr.)*, **82**, 15-24 [In Japanese with English summary].
- Takata, Y. et al. (2013) Estimation of carbon and nitrogen content in surface horizon using "Soil Information Web Viewer". *Dojyono butsurisei (J. Jpn. Soc. Soil Phys.)*, **123**, 117-124 [In Japanese with English summary].
- Uchimura, Y. et al. (2000) Effects of silicate application on lodging, yield and palatability of rice grown by direct sowing culture. *Jpn. J. Crop Sci.*, **69**, 487-492.
- Yagi, K. & Minami, K. (1990) Effect of organic matter application on methane emission from some Japanese paddy field. *Soil Sci. Plant Nutr.*, **36**, 599-610.
- Yagi, K. et al. (1996) Effect of water management on methane emission from a Japanese rice paddy field: Automated methane monitoring. *Global Biogeochem. Cycles*, **10**, 255-267.
- Zhang, A. et al. (2012) Effects of biochar amendment on soil quality, crop yield and greenhouse gas emission in a Chinese rice paddy: A field study of 2 consecutive rice growing cycles. *Field Crops Res.*, **127**, 153-160.
- Zimmerman, A. R. (2010) Abiotic and microbial oxidation of laboratory-produced black carbon (biochar). *Environ. Sci. Technol.*, **44**, 1295-1301.

