Response on Uptake of Nutrients and on Grain Yield from Rice Husk Biochar Application on *Oryza sativa* L. Grown in a Low Yielding Granary Area of Tanjung Karang, Selangor, Malaysia

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ABSTRACT

Rice (*Oryza sativa* L.) production plays a major role in enhancing food security in Malaysia. Lower rice yield and improper soil management practices have raised serious concerns about rice cultivation in Malaysia. The objective of this study was to examine the short-term effects of rice husk biochar (RHB) application on rice yields production on low yielding area for two crop cycles. RHB was applied at the rates of 0, 5, 10, and 20 Mg ha⁻¹. Rice husk biochar was applied one week before rice seeds, of variety MR 263, were directly seeded. Results of the study showed that RHB significantly increased grain yield by 44% and 46% in first and second crop cycles, respectively, as compared to the control treatment. Likewise, RHB amended plots showed significant improvement of rice yield components, viz. productive tiller, panicle length, and weight per panicle, than those of the control plots in the first and second crop cycles. Furthermore, RHB significantly increased nitrogen (N), phosphorus (P) and potassium (K) uptake by 17%, 18%, 29%, respectively, in the first crop cycle, and 26%, 23%, 110%, respectively, in the second crop cycle. RHB application also significantly improved soil available P, exchangeable K and exchangeable Mg in the two crop cycles. Another interesting finding was that the use of RHB reduced soil total carbon loss by 4% to 12% compared with 19% by the control treatment. These findings suggest that RHB can potentially be used as a soil amendment to increase rice yield production, enhance soil nutrient availability and nutrient uptake, as well as reduce carbon losses especially during drought period.

Keywords: rice husk biochar, carbonized biomass; grain yield; nutrient uptake and availability, rice (*Oryza sativa* L.); soil amendment

INTRODUCTION

In Malaysia, abundance of rice husk (408,000 Mg per annum) produced after

each harvest period, approximately 20% of crop residue, is burnt as a source of heat energy for drying rice and for rice husk biochar (RHB) production (BERNAS, 2020). The RHB produced has a lower carbon (C) content than pyrolyzed biochar. However, rice husk and RHB are not fully utilized and caused environmental pollution. The application of RHB as soil

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amendment in the rice soils near the mills is deemed feasible, logistically, for the improvement of rice crop and to reduce greenhouse gases (GHGs) emission due to volatilization and denitrification processes.

Rice (*Oryza sativa* L.) is mainly grown under flooded ecosystem mainly in the coastal areas of the total lowland rice cultivation area in Malaysia, and out of which 83% is in Peninsular Malaysia, 11% in Sarawak and 6 % in Sabah, Malaysia (Paddy Statistic of Malaysia, 2015). Total rice production for Malaysia in 2019 was estimated at 1825 Mg within an area of 700 000 ha, with national average yield of 4 Mg ha⁻¹ (Index Mundi, 2020). The total domestic consumption is approximately 2775 Mg and Malaysia imported about 1000 Mg in 2019. Department of Statistics Malaysia (2020) reported that as of 2018, Malaysia had achieved 70% of rice self-sufficiency.

One of the main reasons for low yield of rice in Malaysia is improper soil management practices which lead to high nitrogen (N) loss through volatilization and denitrification processes. Soil amelioration is one of the solutions which has been reported to improve crop performance as well as the yield production. Currently, conversion of crop residues into charcoal, known as biological charcoal (biochar) is gaining attention due to its potential as soil amendment, in addition to abating climate change through sequestering carbon and reducing methane (CH₄) and nitrous oxide (N₂O) emissions (Lehmann et al. 2007; Woolf et al. 2010). Biochar is a byproduct of residual carbonization of biomass after separation from combustible gases under anaerobic condition and high-temperature pyrolysis (Yang et al, 2020). Lehmann and

Joseph (2009) reported that incorporation of biochar helps to conserve and sustain soil productivity as well as reducing environmental impact. As reported in previous studies, application of biochar was found to have dominant effect especially on soil chemical properties rather than crop yield, which was probably due to the application of organic or synthetic fertilizers (Steiner et al., 2007; Asai et al., 2009; Haefele et al., 2011). Thus, biochar application in cultivated land is beneficial in the long-term especially in subtropical and tropical regions due to its roles in improving soil physicochemical properties, organic matter, crop production, and fertilizer use efficiency (Van Zwieten et al., 2010; Deenik et al., 2011). Increase in rice crop yield with biochar application has been reported elsewhere (Steiner et al. 2007; Zhang et al. 2010; de Melo Carvalho et al. 2014).

Rice grain yield from the area of study has been reported to achieve up to 10 Mg ha⁻¹ (Personal communication, 2014) with heavy applications of chemical fertilizers by rich farmers. There were several areas that produced yields of 5 Mg ha⁻¹ and below, including the area where the present study was conducted. However, documented reports on the influence of biochar application on rice yield is still greatly lacking, especially in Malaysia. We hypothesized that RHB application helps to increase rice yield through improvement of soil properties and nutrient uptake in areas with low rice productivity. The objective of this study was to examine the short-term effects of RHB application on rice yields in low yielding areas for two crop cycles.

MATERIALS AND METHODS

Field sites

A field experiment was established in a Barat Laut Selangor rice area in Kampung Tengah Pasir Panjang (3°35'27.6"N 101°04'10.5"E), over two rice cropping seasons. Total rainfall was 517 mm in the first crop cycle (September to December 2014) and 321 mm in the second crop cycle (March to June 2015), whereas the average temperature

during this period of study was 27 °C. A long drought during the second crop cycle affected the water level during the rice growth period. Rice cultivation in this area has been carried out for the past 39 years. Soil of the experimental field is of Sabrang series, which is classified as Sulfic Endoaquepts. The soil has a clay texture with pH of 5.50, and the topsoil has total carbon content of 172 g kg⁻¹ with total mineral content of 80% (1.65 % sand, 38.83 % silt, and 60.47 % clay) as shown in Table 1.

Table 1. Selected chemical properties of the soil before treatment and rice husk biochar (RHB) properties

Chemical properties	Initial soil (non-amended soil)	RHB (First crop cycle)	RHB (Second crop cycle)
pH (Flood water)	6.30	~*	-
pH (soil = 1: 2.5; biochar = 1: 50)	5.50	8.50	7.4
Total Carbon (g kg ⁻¹)	172.00	210.00	431.20
Total Nitrogen (g kg ⁻¹)	6.00	1.00	6.80
Available P (mg kg ⁻¹)	64.60	-	-
Exchangeable K (mg kg ⁻¹)	471.9	-	-
Exchangeable Ca (mg kg ⁻¹)	2646	-	-
Exchangeable Mg (mg kg ⁻¹)	559.2	-	-
CEC (cmol ₍₊₎ kg ⁻¹)	15.90	-	-
Total ash (%)	-	88.50	60
Volatile matter (%)	-	9.20	16
Total P (g kg ⁻¹)	-	5.10	1.55
Total K (g kg ⁻¹)	-	2.70	2.92
Total Ca (g kg ⁻¹)	-	0.98	0.85
Total Mg (g kg ⁻¹)	-	0.20	0.87

*: not determined

The RHB used for the field experiment was produced by a carbonization process which was collected from SENDI Enterprise at Sekinchan, Selangor. RHB treatment was applied at the rates of 0, 5, 10 and 20 Mg ha⁻¹ at the beginning of each crop cycle. RHB was spread onto the surface during land preparation after second ploughing and then mixed into soil by ploughing to a depth of 15 cm. The four treatments were arranged in a fully randomized complete block design (RCBD) with four replications and plot size of 4 m x 4 m. Rice variety MR 263 was used and sown through direct broadcasting (200 g plot⁻¹). The plot was irrigated 14 days after broadcast of seeds and then drained at the ripening stage at 100 days after sowing (DAS). Fertilizer application of the rice crop was according to the recommended rates and timing by Sistem Penanaman Padi Lestari (MARDI, 2008).

Plant and Soil Sampling and Analysis

Rice was harvested at 110 days after sowing. Sampling with 1 m² quadrant was carried out to determine rice yield components such grain yield (GY), number of productive tillers (PT), number of panicles (NP), percent filled grains (FG), weight of 1000 grains, panicle length (PL), weight per panicle (WPP), straw dry matter weight (DMWs) and harvest index (HI). Three points of composite soil samples were taken before applying treatment for soil characterization and after harvest to determine the soil pH, total carbon (TC), total nitrogen (TN), available phosphorus (P), exchangeable potassium (K), calcium (Ca), magnesium (Mg) and soil cation exchange capacity (CEC). Harvested plant sample was

oven-dried at 65 °C to constant weight before weighing to determine the dry matter weight and soils were air-dried, ground and sieved with 2 mm mesh. The oven-dried plant samples were ground into a 0.25 mm size and subjected to wet digestion before being analyzed for macronutrients (P, K, Ca, and Mg). Soil pH was determined using a pH meter with 1:2.5 ratio soil solutions with deionized water. Total C (soil) and total N (soil and plant samples) were determined using LECO TruMac CNS analyzer. The soil available P was determined using the method of Bray and Kurtz (1945). Exchangeable cations (K, Ca and Mg) and soil CEC were determined using ammonium acetate of pH 7.0. For RHB analysis, LECO TruMac CNS analyzer was used to measure the RHB total C and total N. The pH of RHB was determined in water at ratio of 1:5 (w/v). Total elements (P, K, Ca, and Mg) were determined using the dry ashing method. Volatile matter and ash content were measured using ASTM D 3172 Standard practice for Proximate Analysis of Coal and Coke.

Statistical Analysis

One-way analysis of variance (ANOVA) was performed to determine the significant differences in yield parameters, nutrient uptake and soil properties between different treatments. The differences in means were separated by least significant difference (LSD) test at $p \leq 0.05$. Pearson correlation coefficients were calculated to determine the relationship between GY and nutrient uptake, soil properties and yield parameters using SAS 9.2 software (SAS Institute Inc., USA).

RESULTS

Effect of RHB Amendment on Soil Characteristics

Data describing the chemical properties at the end of field experiment under different rates of RHB treatments are presented in Table 2.

Table 2. Soil chemical characteristic as influenced by RHB application in first and second crop cycle in highly organic topsoil (0-15 cm depth)

Variables	First crop cycle (Mg ha ⁻¹ RHB)				Second crop cycle (Mg ha ⁻¹ RHB)			
	0	5	10	20	0	5	10	20
pH	5.24c* (±0.07)	5.77a (±0.07)	5.75a (±0.03)	5.46b (±0.08)	5.62a (±0.07)	5.59a (±0.10)	5.57a (±0.04)	5.51a (±0.09)
Total C (g kg ⁻¹)	166.7a (±3.75)	167.4a (±5.62)	166.0a (±3.98)	164.6a (±2.63)	139.7c (±2.66)	155.9a (±1.30)	149.7b (±0.71)	153.3a (±1.53)
Total N (g kg ⁻¹)	6.6a (±0.21)	6.7a (±0.10)	6.5a (±0.30)	6.7a (±0.20)	5.1c (±0.14)	5.5b (±0.05)	4.7d (±0.11)	6.2a (±0.01)
Available P (mg kg ⁻¹)	72.75b (±2.52)	72.82b (±3.95)	76.30b (±1.71)	91.07a (±3.14)	55.13b (±0.79)	60.02a (±2.90)	61.64a (±2.63)	62.04a (±3.23)
Exch. K (mg kg ⁻¹)	150c (±19.57)	174c (±7.69)	222b (±36.87)	256a (±7.84)	3740d (±56.34)	4772b (±50.67)	5192a (±92.73)	3906c (±28.84)
Exch. Mg (mg kg ⁻¹)	342c (±1.35)	373b (±3.86)	405a (±12.28)	353c (±8.14)	742b (±12.86)	821a (±25.23)	829a (±29.37)	829a (±29.37)
Exch. Ca (mg kg ⁻¹)	3514a (±126.92)	3605a (±108.18)	3646a (±139.27)	3655a (±114.02)	108c (±4.80)	109c (±11.96)	154a (±12.26)	135b (±10.16)
CEC (cmol (+) kg ⁻¹)	67.52a (±4.91)	73.04a (±3.35)	73.77a (±2.90)	74.73a (±6.15)	30.08a (±2.84)	30.61a (±3.00)	30.77a (±2.95)	32.09a (±2.42)

* Means with different letters indicates significantly difference between treatment at 0.05 level by LSD test and ± value indicate mean standard deviation.

RHB amendment significantly increased soil pH in the first crop cycle, whereas no significant effect was detected in the second crop cycle. It was observed that soil pH in the first crop cycle increased up to 10% with 5 Mg ha⁻¹ RHB application compared to control. However, there was no significant effect of RHB on soil TC in the first and second crop cycle. However, there was significant difference of TC in the second crop cycle in comparison to to unamended plots. A small increase of soil TC (7-11%) as compared to unamended plots was observed during the

second crop cycle. As observed for soil total N, there was no significant response in the first crop cycle, but found significantly influenced soil total N in the second crop cycle. Soil total N increased under RHB treated soil with peak at 20 Mg ha⁻¹ (6.28 g kg⁻¹) in the second crop cycle. Application of RHB was statistically significant only for the 20 Mg ha⁻¹ in the first crop cycle, while significantly increased under all RHB treatment in the second crop cycle. In the first crop cycle, soil exchangeable K was significantly affected by RHB at rate of 10 and 20 Mg ha⁻¹, while significantly increased in all RHB treatments compared to control treatment. A

similar result was observed for exchangeable Mg in the second crop cycle, whereas a significant difference only found at application of 5 and 10 Mg ha⁻¹ during the first crop cycle. However, RHB amendment only significantly influenced soil exchangeable Ca during the second crop cycle with application rates of 10 and 20 Mg ha⁻¹. Furthermore, soil CEC was not significantly different between plots with or

without RHB applications for both crop cycles.

Effects of RHB Amendment on Nutrient Uptake

Rice husk biochar addition significantly ($P \leq 0.05$) increased aboveground biomass N uptake as compared to the control in both crop cycles (Table 3).

Table 3. Total nutrient uptake by aboveground biomass after rice husk biochar (RHB) application at different rates in first and second crop cycles

Variables	First crop cycle (Mg ha ⁻¹ RHB)				Second crop cycle (Mg ha ⁻¹ RHB)			
	0	5	10	20	0	5	10	20
N (kg ha ⁻¹)	142.60c* (±2.56)	166.70a (±3.23)	146.91b (±1.67)	145.35bc (±3.19)	119.11d (±1.62)	145.34b (±3.69)	134.93c (±3.00)	150.23a (±2.74)
NHI (%)	47.71c (±2.78)	53.58b (±2.57)	57.86ab (±3.73)	58.89a (±1.18)	-	-	-	-
P (kg ha ⁻¹)	50.79b (±3.31)	59.32a (±2.55)	60.07a (±4.06)	49.09b (±1.39)	22.58c (±1.25)	25.82bc (±2.75)	28.18ab (±0.36)	29.62a (±1.99)
K, (kg ha ⁻¹)	125.83b (±6.51)	161.57a (±3.77)	162.33a (±10.25)	156.08a (±5.27)	116.99d (±2.89)	166.37c (±3.55)	207.57b (±4.31)	245.52a (±4.55)
Ca (kg ha ⁻¹)	10.15a (±0.95)	11.61a (±1.47)	12.90a (±1.83)	12.09a (±0.54)	14.34a (±2.86)	14.68a (±2.73)	16.19a (±1.42)	16.94a (±1.36)
Mg (kg ha ⁻¹)	17.46ab (±1.46)	18.54a (±0.85)	17.21b (±0.23)	15.58c (±0.47)	12.79b (±1.09)	16.68a (±0.22)	16.14a (±0.98)	16.37a (±0.92)
Zn (kg ha ⁻¹)	9.08a (±1.14)	10.01a (±0.10)	8.54a (±0.36)	9.15a (±0.73)	10.86bc (±1.35)	14.85b (±0.85)	14.89b (±1.26)	19.16a (±0.80)

* Means with different letters indicates significantly difference between treatment at 0.05 level by LSD test and ± value indicate mean standard deviation; N=nitrogen; NHI=nitrogen harvest index, P=phosphorus; K=potassium; Ca=Calcium; Mg=Magnesium; Zn=zinc.

RHB treatment indicated that N uptake by aboveground biomass significantly increased up to 17% and 26% in the first and second crop cycle, respectively. It should be noted that Nitrogen Harvest Index (NHI) was only measured in the first crop cycle. With the addition of RHB (20 Mg ha⁻¹), significant

increases in NHI was observed as compared to control. In the first crop cycle, P uptake was significantly influenced by application of 5 and 10 Mg ha⁻¹ RHB as compared to control treatment. The highest value was recorded at application of 10 Mg ha⁻¹ RHB (60.07 kg ha⁻¹), whereas the lowest value was recorded at control

treatment (50.79 kg ha⁻¹). However, in the second crop cycle RHB treatment was statistically significant with application of 10 and 20 Mg ha⁻¹ compared with the control. The highest reading was recorded at 20 Mg ha⁻¹ (29.62 kg ha⁻¹). The K uptake also showed significant differences in all RHB treated plots compared with control in both first and second crop cycle. In contrast, the application of RHB had no effect on Ca uptake. Similar results were observed for Mg and Zn uptake in the first

crop cycle. However, there was a significant difference in Mg and Zn uptake for the second crop cycle with an increment of 28% and 76%, respectively.

Effects of RHB Amendment on Yield Parameters

The effect of RHB application on rice yield parameters shown in Table 4.

Table 4. Effect of rice husk biochar (RHB) on rice yield parameters at first and second crop cycles

Variables	First crop cycle (Mg ha ⁻¹ RHB)				Second crop cycle (Mg ha ⁻¹ RHB)			
	0	5	10	20	0	5	10	20
GY (Mg ha ⁻¹)	5.47b* (±0.44)	7.48a (±0.13)	7.21a (±0.36)	7.89a (±0.59)	3.23d (±0.17)	3.53c (±0.11)	4.70a (±0.23)	4.16b (±0.18)
PT (%)	80.95c (±1.88)	83.38b (±1.28)	86.25a (±0.20)	86.00a (±1.97)	84.70b (±1.64)	90.59b (±1.30)	90.76a (±1.87)	91.45a (±2.25)
NP (m ⁻²)	416b (±10.81)	536a (±5.12)	413b (±8.85)	411b (±10.25)	306d (±4.66)	332c (±6.18)	371b (±7.45)	413a (±4.11)
FG (%)	82.22a (±3.06)	78.46a (±3.20)	76.41a (±2.80)	78.86a (±0.56)	66.63c (±2.25)	88.20a (±0.19)	77.86b (±3.01)	78.04b (±2.95)
TG (g)	10.08b (±0.13)	10.05b (±0.04)	9.80c (±0.17)	10.83a (±0.18)	11.79b (±0.48)	12.60a (±0.50)	13.09a (±0.42)	11.92b (±0.41)
PL (cm)	20.32c (±0.31)	21.78ab (±0.69)	21.05bc (±0.63)	22.01a (±0.35)	21.79c (±0.38)	22.64b (±0.43)	23.5a (±0.34)	23.33a (±0.17)
WPP (g)	1.22bc (±0.14)	1.43b (±0.16)	1.16c (±0.21)	1.81a (±0.10)	0.46bc (±0.10)	0.56bc (±0.02)	1.04a (±0.07)	0.63b (±0.14)
DWS (Mg ha ⁻¹)	8.45a (±0.32)	8.63a (±0.20)	7.34b (±0.09)	8.77a (±0.60)	7.25d (±0.23)	9.41b (±0.37)	8.38c (±0.44)	10.29a (±0.06)
HI	0.39c (±0.017)	0.46b (±0.005)	0.50a (±0.014)	0.47ab (±0.019)	0.30b (±0.008)	0.27d (±0.010)	0.36a (±0.012)	0.29c (±0.008)

* Means with different letters indicates significantly difference between treatment at 0.05 level by LSD test and ± value indicate mean standard deviation; GY=grain yield; PT=productive tiller; NP=no. of panicle; FG=filled grain; TG=1000-grains; PL=panicle length; WPP=weight per panicle; DWS=dry matter weight straw; HI=Harvest index.

The results showed that GY increased significantly ($P \leq 0.05$) by 44% with RHB amendment in the first cycle, while 46% in the second crop cycle. Similarly, the PT was greater under RHB application with peak at 10 Mg ha⁻¹ (86.25%) than observed in the control for the first crop cycle. However, in the second crop cycle PT was only significantly affected at 10 and 20 Mg ha⁻¹ of RHB application with peak value of 91.45%. In contrast, NP for the first crop cycle was only statistically significant under application of 5 Mg ha⁻¹ RHB. Meanwhile, NP on RHB amended soil for the second crop cycle was significantly ($P \leq 0.05$) higher in all RHB applications compared to the control. Furthermore, NP was not significantly different between plots with or without RHB in the first crop cycle. Unlike in the second crop cycle, NP was significantly increased with the increasing rate of RHB application compared to control. No significant difference was found between treatments for FG in the first crop cycle. However, in the second crop cycle RHB found to have significant effect on FG at application rate of 5 and 10 Mg ha⁻¹ compared with the control. Thousand grain weight significantly increased only in RHB rate of 20 Mg ha⁻¹ for first crop cycle, while statistically significant at 5 and 10 Mg ha⁻¹ RHB application for second crop cycle. Compared to no RHB amendment, PL was significantly increased by 8% in the first crop cycle and 7% in the second crop cycle. Weight per panicle significantly increased when 20 Mg ha⁻¹ RHB was applied in the first crop cycle. In contrast, for the second crop cycle WPP significantly influences at application rate of 10 Mg ha⁻¹ compared to control. In the first crop cycle, there was no significant difference for the DWS and only significantly increased with

application of 20 Mg ha⁻¹ RHB with an increment of 42%. Rice husk biochar application increased HI in the first crop cycle at all application rates, while HI significantly responded to RHB when 10 Mg ha⁻¹ were applied.

DISCUSSION

The RHB application rate used in this study was chosen to study the response on high organic topsoil and did not represent an optimal rate for rice growth. Studies have shown that application of biochar can enhance soil carbon pool, resulting in beneficial effects on soil active organic carbon components, including increase in crop biomass and input of fresh organic carbon, improvement in soil structure, promotion of soil aggregate formation, and provision of an ideal habitat for soil microorganisms (Piccolo et al., 1996; Glaser et al., 2000; Whalley et al., 2006; Steinbeiss, et al., 2009; Yang et al., 2020). Biochar is suggested to improve soil quality, and there are reports of stimulated microbial response and loss of native SOC (Steinbeiss et al., 2009). As reported by Lehmann and Rondon (2006), good agronomic effects in other crops have been achieved with rates of biochar from wood between 0.4 and 8.0 t C ha⁻¹, but no negative effects have been reported below that of 20 t C ha⁻¹. The applications of RHB in this study (Table 2) indicated positive effects on soil chemical properties. Results indicated that the RHB amendment showed increment in soil pH at harvest in high organic rice topsoil for the first crop cycle, but did not exhibit a positive effect in the second crop cycle. The high pH of RHB (8.5) in the first crop cycle could be attributed to an increase in soil pH in the first crop cycle. Apart from that, the high ash content of RHB (89%) and liming potential of the biochar used could be

the reason in increment of soil pH in the first crop cycle. This result is in accordance with studies done by Masulili et al. (2010), who reported an increase in soil pH under application of RHB with pH of 8.5.

The application of RHB showed higher soil TC compared to unamended plot at harvest in the second crop cycle ranging from 136.2 to 157.6 g kg⁻¹. This can be explained by the relatively higher C content (413.23 g kg⁻¹) of the used RHB in the second crop cycle and the cumulative effect of the applied RHB from the first crop cycle. A similar result has been reported by Masulili et al. (2010), who reported high levels of SOM under RHB treated plot although low C organic content compared to raw rice straw or rice husk. However, lower level of soil TC was observed in the second crop cycle compared with the first crop cycle could be attributed by C losses through soil decomposition and mineralization which directly link with reducing water table during drought period in the second crop cycle (Figure 1).

Figure 1: Soil total C loss in first and second crop cycle (110 DAS) as compared to initial (before treatment), as affected by RHB application and $\pm$ value indicate mean standard deviation.

Studies have shown that the status of soil water content is the key driving factor in the carbon cycle process. Soil water content manifested a significant correlation with the transformation of organic carbon within a certain range of changes. The changes in soil moisture content and its distribution, for example during a period of drought, will have a significant impact on SOC and its active components. This present study was in accordance to the recent findings of Yang et al. (2020), which was also reported previously that soil moisture losses promoted SOC decomposition (Davidson et al, 1998; Flanagan and Johnson, 2005). Amongst all treatments, the least C loss was recorded at RHB 5 Mg ha⁻¹ (9%) and was the highest in control treatment (19%) as in Table 5.

RHB addition significantly reduced soil C losses from 9 to 12% in biochar treated plot in the second cycle. In particular, the increase in soil total N in this study with RHB application is in agreement with a study by Chan et al. (2008), who reported an increase in soil total N after application on biochar. Similarly, Dong et al. (2015) reported that soil total N increases with addition of biochar application by 14.9% and 11.7% with and without urea, respectively. Application of RHB amendment exerted a significant effect on enhancing soil available P by 25% in the first crop cycle and by 12% in the subsequent crop cycle. A similar finding by Masulili et al. (2010), who reported that soil available P was recorded to be higher in RHB treated plots which could be due to high soil pH under RHB treatment. A similar increase in soil available P also was reported in previous studies (Lehmann et al., 2002; Yamato et al., 2006).

Table 5. Soil total C availability after harvest at first and second crop cycle as influenced by RHB application rates.

RHB treatment (Mg ha ⁻¹)	Soil total C, (g kg ⁻¹)			Reduction, (%)		
	Initial	First crop cycle	Second crop cycle	First crop cycle	Second crop cycle	crop
0	172	167 (±1.88)	140 (±1.33)	2.9	18.6	
5	-	167 (±2.81)	156 (±0.65)	2.9	9.3	
10	-	166 (±1.99)	150 (±0.36)	3.5	12.8	
20	-	165 (±1.31)	153 (±0.77)	4.1	11.1	

± value indicate standard error.

Biochar amendment has previously been shown to increase soil exchangeable cations such K, Ca and Mg. In this study, addition of RHB resulted in a 71% and 4% increased soil exchangeable K in the first and second crop cycles, respectively, and this can be associated with the high K content in the RHB applied (Table 4). Likewise, an increase in soil exchangeable Mg seems to respond up to 10 Mg ha⁻¹ RHB in the first crop cycle, whereas in the second crop cycle only response at 10 and 20 Mg ha⁻¹ RHB application rate. In addition, positive effect of biochar application on soil exchangeable Ca was only observed in the second crop cycle at all RHB application rates. A similar finding has also been reported in previous studies by Belyaeva et al. (2012), who reported an increment in soil exchangeable K, Mg and Ca in pot study using poultry manure biochar.

In both crop cycles, RHB application had a positive effect on plant N uptake which in accordance with Huang et al. (2014), who reported an increased fertilizer N uptake by rice by approximately 23-27%. This might contribute to the increased availability of soil N from decomposition of indigenous organic matter (apart from the fertilizer N) for N uptake

with application of biochar. Similarly, P and K uptake in both crop cycles were slightly higher in biochar application with an increment of 18% and 31%, respectively as compared to control treatment. This might be due to great availability of nutrients content in the soil, which is in accordance with studies done by Major et al. (2010). In addition, an increase in plant K uptake also was reported with biochar application in cowpea crop in a pot study by Lehmann et al. (2003). However, an unusual long drought event that occurred during the second crop cycle led to a significantly lower rice plant uptake of P and K, which might be due to the abiotic stress conditions.

In comparison to study by Zhang et al. (2012), rice yield increased 10% in first crop cycle and by 9.5 to 29% in subsequent cycle using wheat straw biochar on clay soil, whereas in this study, grain yield increased significantly by 63% and 51% with a threshold value of 16 Mg ha⁻¹ RHB (8.02 Mg ha⁻¹) and 14 Mg ha⁻¹ of RHB (4.55 Mg ha⁻¹) in first and second crop cycle, respectively (Figure 4). Although grain yield in the second crop cycle seemed to decrease as compared to the first crop cycle, which might be a consequence of an unusual long drought period during this growing season.

Table 6. Correlation between soil properties, nutrient uptake and rice yield parameters to grain yield of two crop cycles.

Variables	First crop cycle (Grain yield) Mg ha ⁻¹	Second crop cycle (Grain yield) Mg ha ⁻¹
Soil pH	0.64**	ns
Available P (mg kg ⁻¹)	ns	0.63**
Exchangeable K (mg kg ⁻¹)	0.72***	0.57*
Exchangeable Mg (mg kg ⁻¹)	ns	0.85***
Exchangeable Ca (mg kg ⁻¹)	ns	0.61*
P uptake (kg ha ⁻¹)	ns	0.72**
K uptake (kg ha ⁻¹)	0.82***	0.75***
Mg uptake (kg ha ⁻¹)	0.51*	0.51*
Cu uptake (kg ha ⁻¹)	0.70**	ns
Zn uptake (kg ha ⁻¹)	ns	0.52*
Productive tiller (%)	0.69**	0.56*
No. of panicle	ns	0.94***
Panicle length (cm)	0.72**	0.86***
Weight per panicle (g)	ns	0.84***

Nevertheless, grain yield under RHB treatments were observed to be higher as compared to control treatment despite growing under stress conditions. This can be attributed to the increase in soil water retention with biochar application (Karhu et al., 2011) which is incredibly important during drought. The optimum soil pH for good rice growth was reported within range of 5.5 to 7.0 (Pedro et al., 1990). A similar increase in grain yield were also reported in previous studies, such in upland rice using wood biochar in Northern Laos (Asai et al., 2009) and on acid sulfate soil using RHB in West Kalimantan, Indonesia (Masulili et al., 2010) and on gley paddy soil using bamboo biochar and RHB in Zhejiang, China (Zhang et al., 2010). The increase in grain yield for both crop cycles as compared to control treatment seemed to be positively correlated with a slight increase in percent of PT, PL and WPP (Table 6).

The improvement in soil chemical properties, viz. soil pH, available P, exchangeable K and exchangeable Mg, and plant macronutrients (N, P and K) uptake by rice plants seems to be a consequence of an increase in grain yield.

Additionally, PT showed an increase of 7 to 8% in RHB amended plots for both crop cycles in comparison to control plots. Such sustainable increasing effect could be supported by other field studies, such as Masulili et al., (2010), who reported increase in PT under RHB treatment on acid sulfate soil in West Kalimantan, Indonesia. The higher PT in biochar amended plots compared to unamended plots may contribute to an increase in plant nutrient uptake, especially of N, and the availability of nutrients in soil. Therefore, RHB application exerted significant positive effects on NP, FG, PL and WPP in both crop cycles. The positive effect of RHB in NP was in accord with studies by Singla et al., (2014), who reported an increase in NP using biochar combined with ammonium sulfate using

pot study in Chiba, Japan. In comparison to the first crop cycle, lower NP, PL and WPP produced in the second crop cycle were clearly affected by the long drought period during this growing season. The DMWs in the first crop cycle had no positive effect toward RHB application, while in the second crop cycle DMWs found to increase under all RHB application rates. Other than the increase in yield parameters by RHB addition, the difference in the HI for first crop cycle was significant compared to control; however, the in second cropping season the HI showed response up to 5 Mg ha⁻¹ as compared to control treatment, which is contrary to the studied by Tammeorg et al. (2014) on faba bean.

CONCLUSION

The present study showed high grain yield production with RHB application in the first and second crop cycles, with an increment of 44% and 46%, respectively. RHB application correspondingly increased yield variables including productive tillers, panicle length and weight per panicle. Application of RHB significantly improved N, P, and K uptake. Further studies are proposed, especially to determine the long term effects of RHB soil amendment on rice cultivated in soils with high organic matter.

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