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**Biochar quality and its short-term effects on rice yield and soil chemical properties**

**Saowakon Hemwong 1\*, Michael Scott Demyan2 and Georg Cadisch3**

*1 Program of Plant Science, Faculty of Agriculture and Technology, Nakhon Phanom University, Nakhon Phanom, Thailand*

*2 School of Soil Environment and Natural Resources, The Ohio State University, Columbus, United State*

*3 Institute of Plant Production and Agroecology in the Tropics and Subtropics (380a), University of Hohenheim, Stuttgart, Germany*

**\*For correspondence:** saowakon@hotmail.com

**Abstract**

Producing biochar from rice husks instead of rice straw burning might reduce environmental pollution. This study aimed to examine the effects of biochar of different quality and fertilizer levels on rice yield, soil fertility and biochar characterization via thermal and spectroscopic methods. The treatments implemented were: i) control (no biochar and fertilizer amendments), ii) no biochar amendment with received chemical fertilizer at rice transplanting and panicle initiation (PI) phase, while biochar amendment with fertilizer treatments was applied urea only PI phase, e.g., iii) bamboo biochar (BC), iv) BC with fertilizer, v) eucalyptus biochar (EC), vi) EC with fertilizer, vii) rice husk biochar (RHC), viii) RHC with fertilizer, ix) rice straw burned residue (RSB), and x) RSB with fertilizer. The EC amendment with fertilizer resulted in a higher increase of 71% grain yield than the control without biochar. However, BC significantly improved available soil P and exchangeable K to 26 and 0.046 mg kg-1, respectively as compared to non-amended biochar treatment. In contrast, EC had the strongest impact on increasing soil cation exchange capacity (CEC) but leading to initial N immobilization. The carbon characteristics of woody biochar (BC and EC) were high in inorganic OH stretching (3620 cm-1), this is exhibited by certain inorganics and minerals and resulted in the increase in soil CEC and increased plant NUE. Evidence gathered from the experiment indicated that woody biochar additions to lower fertility paddy soils together with fertilizer application could produce yields greater than either only N fertilizer or biochar application alone.

**Keywords:** Soil fertility; DRIFTS-MIRS analysis; Fertilizer; Biochar

**Background**

Northeast Thailand is the most extensive rice production area in Thailand. However, the average yield (2.23 - 2.28 t ha-1 in the year 2017-2019) (Office of Agricultural Economics 2020) is lowest due to the prevailing mostly low fertility sandy soil. Biochar has received a growing interest as a sustainable soil amendment to improve soil fertility and increase plant nutrient use efficiency, thereby increasing crop growth and yield. Biochar is the carbonization product of organic matter under partial or complete exclusion of oxygen. Slow pyrolysis of wood biomass in traditional kilns has been the most widespread application for biochar production (Arni 2018) in Northeast Thailand. Increases in crop growth and yield with biochar application have been previously reported in Indonesia (Masulili et al. 2010) and NE Thailand (Haefele et al. 2011). Charcoal’s enhancement of soil cation-exchange capacity (CEC) suggests that added biochar is not only a soil conditioner but also acts as Fertilizer (Naggar et al. 2019). Although biochar essentially is organic carbon, its very slow decomposition in the soil makes it different from other soil organic carbon pools, but physico-chemically it may provide many of the same soil services as soil organic matter (SOM), such as soil stabilization by aggregation, and retention of nutrients and water (Kerré et al. 2017). Biochar may be highly recalcitrant to microbial decomposition and thus guarantees a long-term benefit for soil fertility (Pluchon et al. 2016), although it is not inert. Bass et al. (2016) has demonstrated that crop yield can be increased upon biochar additions to soils especially in the tropics. Mukherjee and Zimmerman (2013) reported that fresh biochar could act as an N and P source. The nutrient content is affected by charring conditions, e.g., total N and available P in biochar produced at lower temperature was higher than high-temperature biochar (Ding et al. 2016). On the other hand, biochar application might limit soil N availability in N deficient soils due to the high C/N ratio specific to biochar and, therefore, might temporarily reduce crop productivity (Rawat et al. 2019).

The quality of biochar depends not only on thermal conditions used in the pyrolysis process but also on the type of feedstock used (Enders et al. 2012). Soil nutrient and carbon availability have been changed variably due to different feedstock and their quality (Oduor 2012). Thus, soil application of biochar made from different feedstock may result in different impacts on crop yields, soil nutrient dynamics, and biochar stabilities (Bruun et al. 2011). The total carbon content of biochar varies considerably depending on feedstock and treatment and may range from 400 g kg-1 up to 900 g kg-1 (Chan and Xu 2009; Gaskin et al. 2010). In addition, the amount of soil extractable K, Ca, Na and Mg could be increased by 60-670% after biochar application (Wang et al. 2014). Furthermore, Brewer et al. (2011) showed that switchgrass and corn stover biochar had higher ash content and lower aromatic carbon than biochar produced from woody material. The most important measures of biochar quality appear to be high adsorption capacity- as measured by GACS (Gravimetric Adsorption Capacity Scan), and low levels of mobile matter (tars, resins, and other short-lived compounds) (Mclaughlin et al. 2009). Most studies have shown that the characteristics of biochar govern the impact on plant growth over time after biochar incorporation into soil (Major et al. 2010). Cases of reduced plant growth due to biochar application can be attributed to temporary increased levels of pH, volatiles (phenolic compound, aliphatic or aromatic hydrocarbon) and/or nutrient imbalances associated with fresh biochar (McClellan et al. 2007; Deenik et al. 2010). Increased nutrient retention in soils by biochar may be the most important factor for increased crop yield on low fertility sandy soils (Asai et al. 2009). Chen et al. (2007) found that additions of biochar plus fertilizer increased radish yield more than addition of fertilizer alone, indicating reduced N leaching and increased N use efficiency. However, there is need for further investigation on how the application of biochar technology in lowland rice systems on low nutrient status soils could support the replacement of the traditional practice of open burning of rice straw to improve soil fertility and increase yield of rice production system. We hypothesize that biochar produced from rice husk as a soil amendment will improve soil fertility and increase growth and yield of rice compared to the conventional management practice or biochar from woody (bamboo, eucalyptus) feedstock. The objectives of this experiment were: i) to characterize biochar from different feedstocks via thermal and spectroscopic methods, ii) to study the effect of different biochar qualities on soil fertility, and iii) on the potential to enhance growth and yield of lowland rice.

**Materials and methods**

*Production and chemical properties of biochar*

Biochar feedstocks were obtained from bamboo (*Bambusa* sp.) stems, eucalyptus (*Eucalyptus camaldulensis* Dehnh.) branches and rice (*Oryza sativa*) husk and all biochar productions based on farmers practice in Thailand. Bamboo and eucalyptus biochar was produced in a biochar kiln (made from a 200 L oil drum) (Steiner et al. 2018) at maximum temperatures of 500 and 525 °C, respectively, while rice husk biochar was produced by traditional kiln (Ogawa and Okimori 2010) made from an 18 L zine bucket at a maximum temperature of 188 °C. Production efficiencies of bamboo, eucalyptus and rice husk biochar were 16, 21 and 59% of total feedstock weight, respectively. The biochar was ground to pass through a 1 mm sieve. The biochar pH was measured in a 1:5 biochar/water suspension with a compound glass electrode (Gaskin et al. 2008). The CEC of the biochars was measured by the sodium acetate method (Chapman 1965). Total C and N contents of biochar were determined using a Vario Max CN analyser (Elementar, Germany) at 1200 °C.

*FTIR-EGA and DRIFTS-MIRS analysis of biochar*

Fourier transform infrared-evolved gas analysis (FTIR-EGA) and Fourier transform mid-infrared spectroscopy (DRIFTS-MIRS) analysis of soil alone, biochar and rice straw burned residue were performed according to Demyan et al. (2013). Briefly, pure biochar and soil samples were ball milled and dried overnight at 40 ºC for biochar alone and 32 ºC for soil samples before analysis. A high-temperature heating system was used in conjunction with a Bruker Tensor-27 (Bruker optic GmbH, Ettlingen, Germany) infrared spectrometer. The high-temperature reaction chamber (HTC) (Harrick Scientific Products, Pleasantville, NY, USA) had an integrated sample holder which approximately 50-70 mg of biochar or soil samples and was equipped with a cartridge-type heating element and a K-type thermocouple. The HTC included an automatic temperature controller and integrated temperature/process controller (Wat-low Winona, Minnesota, USA) for programmed heating rates and setpoint temperature. The HTC was closed by a gas-tight dome with a high-temperature O-ring. The dome had three windows, two made from KBr to enable diffuse reflectance measurement (DRIFTS) of soil, while the third window was made from quartz glass. For measuring FTIR-EGA in the pure biochar and soil samples, synthesis air was used as the purge and carrier gas to 15 L hr-1. The chamber was purged for 5 min after introducing the sample. An optimized heating rate of 68 ºC min-1 and set-point temperature of 700 ºC were used. Once at 700 °C the temperature was held for an additional 10 minutes. An integration of main CO2 area between 2400-2200 cm-1 was done for every spectrum in order to have a time-temperature resolved thermogram of CO2 evolution during heating. The temperature of the largest integrated peak area was then taken as the point of maximum CO2 evolution (CO2max). The individual scans from an FTIR-EGA analysis were then assembled into a single file or thermogram for further processing using the spectral processing software OPUS v 6.5 (Bruker Optik GmbH). In DRIFTS-MIRS analysis of biochar samples, a Praying Mantis diffuse reflectance (DRIFTS) chamber (Harrick Scientific) was fitted in the Tenson-27 (Bruker Optik GmbH) and the HTC (Harrick Scientific) placed inside the DRIFTS chamber to recorded from 4000-400 cm-1 by combining 16 individual scans at a resolution of 4 cm−1 every 4 seconds during heating recorded. Mid-infrared spectra were recorded on a Tensor-27 Fourier transform spectrometer using a potassium bromide (KBr) beam splitter and a liquid nitrogen cooled mid-band mercury-cadmium-telluride detector. The spectrometer was mounted with a Praying Mantis diffuse reflectance chamber, purged with dry air from a compressor (Jun-Air International, Nørresundby, Denmark) a flow rate of 200 L hr-1. The acquisition mode was double forward-backward and the Blackman-Harris-3 apodization function was used. The spectra were recorded in absorbance units (AU). Individual scans were recorded in triplicate. For each replicate scan, the sample was returned to the sample container, mixed and then transferred to the measuring cup. Spectral pre-processing included atmospheric correction for carbon dioxide (CO2) and water, baseline correction and vector normalization to compensate for slight variations in air humidity, temperature and CO2 concentration at the time of measurement. Peak area integration on the corrected spectra was performed according to Demyan et al. (2012) using the spectral processing software OPUS version 6.5 (Bruker Optik GmbH). Major peaks and assigned functional groups of biochar as measured by DRIFTS-MIRS are illustrated in Figure 1 and further described in Table 1.

*Experimental design*

A pot experiment was conducted in an open greenhouse of the Section of Plant Science, Faculty of Agriculture and Technology, Nakhon Phanom University, Thailand (17°24'N, 104°47'E). The experiment used a sandy loam paddy soil collected from 0-15 cm soil depth of an experimental field, air-dried, crushed and passed through a 2 mm sieve. Six kilograms of sieve soil were then put in a plastic pot (height, 24 cm; diameter, 24 cm). Initial soil characteristics were: pH 5.67, total carbon 4.49 g kg-1, total nitrogen 0.60 g kg-1, and Bray II P 0.185 mg kg-1. Soil exchangeable K and cation exchange capacity (CEC) were 0.024 mg kg-1 and 5.61 cmolc kg-1, respectively.

The rice experimental treatments implemented were: i) control with no biochar amendment and without fertilizer, ii) no biochar amendment with received solid compound fertilizer at rice transplanting and urea at panicle initiation (PI) phase (farmer practices), while biochar amendment with fertilizer treatments was applied urea only PI phase, e.g., iii) bamboo biochar (BC), iv) BC with fertilizer, v) eucalyptus biochar (EC), vi) EC with fertilizer, vii) rice husk biochar (RHC), viii) RHC with fertilizer, ix) rice straw burned residue (RSB), and x) RSB with fertilizer. Sixty grams of biochar (equivalent to 1% by weight or 13 t ha-1) were mixed into the top 10 cm. In case of rice straw burned residue treatment, rice straw was placed onto the soil surface in the pots at a rate equivalent to 5 Mt ha-1 (the average amount of rice straw residue in paddy field of Thailand), then open burned for 10 minutes to simulate field burning of rice straw residues. The burnt rice straw was then left to air cool for 2 days prior to rice transplanting. Then, water was added to the pots until the water level was 5 cm above the soil surface. Urea was applied during the panicle initiation (PI) phase of rice (60 days after transplanting). The mineral fertilizer treatment received solid 15-15-15 compound fertilizer containing 0.106 g N pot-1, 0.106 g P2O5 pot-1 and 0.106 g K2O pot-1 at transplanting and 0.130 g N pot-1 as urea at PI phase.

The experiment was arranged in a completely randomized design with 4 replications.

*Rice growth and harvesting*

Thirty-day old rice seedlings of cultivar Chai-nat 1 were transplanted (3 plants pot-1) on February 28, 2011. Weeds, pests and diseases were controlled by pesticides as needed.

Rice was sampled at 30 and 60 days after transplanting (DAT) to assess tiller number, height, shoot dry weight and total N content. At maturity (120 DAT), rice was harvested, i.e. cut at ground level, dried at 70 °C and seed yield determined. The shoot and seed grain were weighed and the % of filled grains and harvest index (HI) to measure the efficiency of plants in producing seed as the ratio of grain yield to total aboveground biomass (Thomas and Prasad 2003), as well as total N content, were determined by flow injection analysis (Tecator 1984). Nitrogen use efficiency (NUE) (Svechjak and Rengel 2006), internal efficiency of N (IEN) (Segda et al. 2004), agronomic yield N use efficiency (ANUE) (Smith et al. 1988) and apparent fertilizer use efficiency (AFUE) (Eagle et al. 2001) were calculated by these equations.

NUE = Total dry weight/Total N uptake

IEN = Gain yield/Total N uptake

ANUE = Gain yield/N applied

AFUE = (N plantfert- N plantunfert)/(Nfert) x 100

*Soil sampling and analysis*

Soils were sampled from 0-10 cm soil depth before rice transplanting and after rice harvesting and air dried to determine soil chemical properties before and after rice transplanting, i.e., pH (1:2.5 soil: H2O), total carbon and total nitrogen by Vario Max CN (Elementar, Germany) at 1200 °C, available P (Bray II), exchangeable K, Ca, Mg and CEC by 1 M sodium acetate extraction at pH 7.

*Statistical analysis*

An one-way ANOVA was performed on the data using the completely randomized design using the Statistix 8 program (Analytical Software 2003). Standard error and Standard error of the different mean (SED) were calculated from the ANOVA.

**Results**

*Chemical properties of biochar*

The lowest pH (6.78) was found in rice husk biochar which was significantly (*P* <0.001) different from that of the bamboo and eucalyptus biochars (Table 2). Biochar prepared from rice husk also had a higher CEC than bamboo and eucalyptus biochar (*P* <0.05). The C content of the eucalyptus biochar was highest (660 g kg-1) compared to bamboo (545 g kg-1) and rice husk (307 g kg-1) biochars. However, the N content of rice husk biochar was higher (10.4 g kg-1) than those of bamboo (8.9 g kg-1) and eucalyptus (5.7 g kg-1) biochars. The biochar C/N ratio of the eucalyptus biochar was higher (177) than those of bamboo (62) and rice husk (30) biochars.

*DRIFTS-MIRS and FTIR-EGA characterization of biochar*

Via both DRIFTS-MIRS and FTIR-EGA several vibrational and thermal differences between the four types of biochar were identified (Table 2). In the DRIFTS-MIRS (Figure 1) characterization, the BC and EC appeared relatively similar and had visible peaks at 3620 (inorganic OH stretching) and 3055 cm-1 (aromatic C-H stretching) which were not apparent in the RHC and RSB. The rice straw burned residue had a very prominent peak at 2930 cm-1 (C-H stretching of aliphatics), while this was a minor component in the other three biochars. Smaller differences were found at 1700 (carboxyl group) and 1616 cm-1 (C-O vibrations) which were more evident in the rice straw burned residue compared to the other three biochars. The peak at 811 cm-1 was relatively constant across the biochar types.

The characterization via FTIR-EGA (Figure 2 and Table 7) showed that rice residue burned had the lowest temperature of maximum CO2 evolution (CO2max), as a measure of thermal stability, compared to the other three biochars and was in a similar range as unamended soil. Bamboo biochar had the highest CO2max at 578 ºC, while those of EC and RHC were similar at 560 ºC. The FTIR-EGA also revealed different carbon contents of the four biochars, as the total peak area is directly related to evolved CO2 with the peak area expressed by sample weight, so evaluated C contents ranged from RSB<RHC<EC<BC.

*Rice growth, yield and nutrient uptake*

During the initial 30 DAT, the eucalyptus biochar treatment resulted in significantly (*P* <0.05) lower height and shoot dry weight than bamboo and rice husk biochars and non-biochar amendments, both with and without fertilizer. However, there were no significant (*P* >0.05) differences in tiller number (Table 3). The highest total N concentration of rice shoot was obtained in the eucalyptus biochar and no-biochar amendments with fertilizer treatments. At 60 days, before the second split N application, the control treatment with no-biochar amended but with fertilizer treatment had significantly (*P* <0.01) highest shoot height, tiller number and dry weight. Nevertheless, rice shoot dry weights in biochar amendment treatments were significantly higher than in no-amended without fertilizer treatment. The bamboo biochar treatment led to the highest (*P* <0.01) total N content (1.041 g pot-1) when compared with EC, RHC, RSB and no-amended biochar treatments (0.320, 0.244, 0.301 and 0.220 g pot-1, respectively).

At final harvest, in most cases, N fertilizer application at panicle initiation stage increased spike number per pot, shoot dry weight and grain yield under biochar amendment treatments (Table 4). The highest grain yield of 15 g pot-1 was observed in bamboo biochar with fertilizer and in no-amended with fertilizer treatments. However, no-amended with fertilizer and eucalyptus biochar without fertilizer treatments were significantly lowest (*P*<0.01) in the percentage of filled grains (85%). The rice harvest index (HI) of bamboo biochar treatment was significantly larger (*P* <0.01) than no-amended, eucalyptus and rice husk biochar with and without fertilizer treatments.

At harvest, the total amount of N accumulated in rice was significantly (*P <*0.01) largest in no-biochar amended with fertilizer treatment. However, total N grain yield was not significantly different between no-amended with fertilizer (0.214 g pot-1) and eucalyptus biochar without fertilizer (0.219 g pot-1) treatments (Table 4). On the other hand, eucalyptus biochar with fertilizer treatment resulted in the lowest grain N yield (0.085 g pot-1). Nitrogen use efficiency (NUE) was highest in rice husk burned residue (RSB) without fertilizer (178 g DWg-1 N) and eucalyptus biochar (EC) with fertilizer treatments (173 g DWg-1 N), respectively (Table 5). However, the lowest NUE, internal efficiency of N (IEN) and agronomic yield N use efficiency (ANUE) were no-amended with fertilizer treatment (61, 9 and 66 g DW g-1 N, respectively).

*Soil chemical characteristics*

At final harvest, there was a significantly lower pH in the no-amended with fertilizer treatment than under all three of the bamboo, eucalyptus and rice husk biochar treatments. Soil chemical properties after rice harvest in the without fertilizer application treatments showed that total C at 0-10 cm depth increased significantly (*P* <0.01) after harvest of rice when applying bamboo, eucalyptus and rice husk biochars compared to no-amended treatments (Table 6). Soil total N was significantly highest (*P* <0.01) in eucalyptus biochar treatment. However, bamboo biochar treatment led to a significantly higher soil C/N ratio in both with and without fertilizer application treatments compared to rice husk and eucalyptus biochar and no-amended treatments. Bamboo biochar significantly improved (*P* <0.001) the available P to 26.9 mg kg-1 and exchangeable K to 0.046 mg kg-1 as compared to the no-amended biochar treatment (Table 6). On the other hand, eucalyptus biochar treatment exhibited the highest soil CEC (9.03 cmolc kg-1).

**Discussion**

*Impact of biochar application on soil chemical properties*

A higher pH in biochar amended soil than in those of no biochar amendments in this study corresponds with the results of other research (Yuan et al. 2011; Bell and Worrall 2011) and was probably due to biochars having high initial pH values of between 6.78-9.90 (Table 6) and high exchangeable Ca2+, Mg2+ and K+ depending on feed stock types and pyrolysis temperature (Chan and Xu 2009; Yuan et al. 2011). In this study, the wood biochar types (bamboo and eucalyptus biochars) produced with high pyrolysis temperature also had higher exchangeable Ca2+ (29.17 - 36.71 mg kg-1), Mg2+ (10.24 - 4.52 mg kg-1) and K+ (38.77 – 41.24 mg kg-1) when compared to rice husk biochar (23.02, 1.23 and 2.27 mg kg-1, respectively). Hence, the pH values of the biochar produced from bamboo and eucalyptus at 500 and 525 °C, respectively, were higher than rice husk biochar at 188 °C (Table 1). Similarly, Yuan et al. (2011) found that biochar pH increased with increased temperature and suggested that at higher pyrolysis temperature, alkalinity and carbonate content of the biochar increased leading to the higher pH of biochars, while the biochar production efficiency was reduced with increased temperature. However, after biochar additions, the pH of soils may increase or decrease, depending on the soil pH and liming value of the biochar (Lehmann et al. 2011). In this study, the short-term experiment observed slightly increased soil pH in biochar additions (5.53 - 5.68) when compared with no biochar amended (5.18) and rice straw burned residue (5.31) but similar soil pH value of initial soil (5.67). These values suggest that biochar application in acidic soil under flooded rice conditions maintained soil pH values probably due to increased buffering capacity of soil and the high presence of carboxylic functional groups (Figure 1) of added biochar when compared to no amended biochar and rice straw burned residue applications.

The amount of biochar application to the soil was 1% by weight (equivalent to 13 t ha-1). The rate used in the current study was a moderate amount compared with other studies, which applied up to 10% by weight (Haefele et al. 2011; Deenik et al. 2010). Some research indicated that much lower application rates yielded positive results (Butnan et al. 2015; Minerzwa-Hersztek et al. 2020; Kaewpradit and Toomsan 2019). The chosen rate did not represent an optimal rate for rice growth, which was unknown and would depend on several factors, including soil properties and concurrent nutrient and organic matter additions (Haefele et al. 2006; Lehmann et al. 2006). However, our rate is a feasible and sustainable option to initiate an application in farming areas of Asia. In addition, the biochar used in this experiment was produced by traditional production via the process of pyrolysis (conventional or slow pyrolysis).

Biochar soil additions can have an ameliorating effect on infertile soils by increasing CEC (Lehmann et al. 2003). This was also observed in our experiment where biochar amendments increased CEC, due to the high exchangeable K, Ca and Mg in biochar. Biochar application also clearly increased soil C and N but the wider C/N ratio might have caused reduced N availability and lower grain yields (Table 4), especially in eucalyptus biochar. The ratio of C to N within biochars can vary depending on feedstock and pyrolysis conditions. This ratio not only influences the recalcitrant properties of the biochar but may also affect the rate of C and N release during mineralization (Krull et al. 2009). However, biochar application has the potential to sequester C in soil due to its high recalcitrant C in aromatic forms which offers a high degree of stability and resistant, although not completely inert to decomposition (Yadav et al. 2017) and its potential to reside in soils over a long period (Sparkes and Stoutjesdijk 2011). We observed that biochar applications improved soil C and N (ranging from 8.15-14.96 g C kg-1 and 0.79-1.36 g N kg-1) when compared to initial soil conditions (4.4 g C kg-1 and 0.66 g N kg-1). Lehmann et al. (2006) suggested that the conservation of biomass C to biochar C leads to sequestration of approximately 50 percent of initial C but this is highly dependent on the feedstock used and the pyrolysis condition. This was also found in our study, i.e. C in bamboo and eucalyptus conversion to biochar lead to products with 32 and 46 % of initial C, while rice husk biochar had a C content below initial C of the raw material.

Organic biomass derived from manures and compost contains large amounts of C and macro- and micro-nutrients (Chan and Xu 2009). It appears that utilization of these sources of organic matter, as biochar feed stocks, will alter the availability of key macro-nutrients such as N and P, and some metal ions (e.g. Ca and Mg), when incorporated into soil (Gundale and Deluca 2006; Major et al. 2010). Results in Table 6 indicate that the biochar type had different positive effects on soil chemical properties. The soil organic C content increased with the addition of wood (bamboo and eucalyptus) and rice husk biochar, with the highest content shown in wood biochar giving similar result to Varela et al. (2013). Thus, soil application of biochar made from bamboo and eucalyptus resulted in largest impacts on soil C sequestration, available soil nutrients (N, P and K) and CEC (Table 6), and although effects rice husk biochar were smaller, it proved still to be a valuable alternative to burning.

*The effects of biochar application on grain yield*

The eucalyptus biochar showed highest C/N ratio resulting in the poorest initial rice growth development (30 DAT, Table 3) possibly due to associated immobilization of N by labile C from biochar addition that can be readily utilized by soil microorganisms (Smith et al. 2010), and finally resulting in reduced grain yield and HI without N fertilizer addition. Given the high C/N ratios of biochar there is an expectation that N immobilization occurs, inducing plant N deficiency (Atkinson et al. 2010), while at the same time, the recalcitrant nature of the C restricts N immobilization (Chan and Xu 2009). In our study, potential N immobilization induced by eucalyptus biochar amendment could be overcome by adding N fertilizer resulting in a higher increase (71%) in grain yield and NUE as compared to eucalyptus biochar amendment without fertilizer. In contrast, with bamboo or rice husk biochar and rice residue burnt only 25, 18 and 0% yield improvement were observed with N fertilizer, respectively, pointing to less N immobilization or other limiting factors. Rice burnt residue and non-amended biochar treatment had very low NUE after N fertilizer application (39% and 58% reduction, respectively) when compared with no fertilizer application. Moreover, the improved ANUE with biochar application indicated a promotion of better nutrient preservation in the soil system. On the other hand, fertilizer addition without biochar amendment reduced internal N use efficiency (IEN) but led to a higher AFUE, indicating that the decline in N grain yield relative to the constant increase in shoot dry weight with the increase in N supply (low %filled grains and HI) were due to increased vegetative growth competing for assimilates available for grain formation and grain filling (Yesuf and Balcha 2014) and N loss. On the other hand, biochar application in combination with fertilizer led to a higher ANUE and lower AFUE at PI stage, although with higher %filled grains yield. This indicated a high quality of rice yield but low fertilizer use efficiency, as biochar application may have decreased root N uptake (Clough et al. 2013). Thus, biochar amendments can substitute only the basal fertilizer addition, but rice needs an additional N supplement at later stages especially when associated with low quality wood biochar application. These findings are in line with previous research which recommended combining biochar with inorganic or organic fertilizer for crop production (Steiner et al., 2007; Yamato et al., 2006). Moreover, the variation in physico-chemical nature of biochar causes variability in the availability of nutrients within each biochar to plant (Sparkes and Stoutjesdijk 2011).

*Soil organic matter composition quality under different biochar types*

The different biochar types used in this study demonstrated their potential as soil amendments to improve soil fertility and alter soil organic matter composition. A strong band at 3500 – 3400 cm-1 in the regions of OH phenolic groups were found in RSB (Figure 1). These peaks indicate the presence of water, carboxylic acid, phenol and alcohol groups (Aghoghovwia 2018). On the other hand, these functional groups decreased at high temperature of pyrolysis used in the production of BC and EC. These results support observations by Yuan et al. (2011) who showed that a larger number of bonds representing functional groups are present in biochars obtained at lower temperature (300 and 500 ºC) and are absent or reduced in those derived at 700 ºC. The C characteristics of woody biochar (BC and EC) showed presence of high non-hydrogen-bonded hydroxy groups (3620 cm-1), often an alcohol or phenol. This is also exhibited by certain inorganics and minerals and is indicative of free OH groups either on the surface or embedded within a crystal lattice and free from interactions with other ions or groups (Coates 2000). This might be the cause of the increase in soil cation exchange capacity from oxidation of aromatic C on the EC surface to form carboxylic groups (Mikutta et al. 2005) and hence increase in grain yield and nitrogen use efficiency of rice (Table 4 and 5). At the same time, the representative peaks for aromatic C observed as strong C-H stretching (3055 cm-1) in wood biochar might be associated with charring at higher temperatures than used in RHC and RHB. The presence of an intense peak at 2930 cm-1 assigned to C-H stretching vibrations in RHC and RSB (Figure 1) was possibly due to a higher proportion of labile compounds (cellulose and hemicellulose) (Demyan et al. 2012; Kunlanit et al. 2014) and associated with higher total N uptake and N use efficiency under without fertilizer application than BC and EC. This indicates that incorporation of burned rice straw residues during soil preparation of rice production has a high potential for C loss via microbial decomposition especially under paddy cultivation due to possible contribution as a substrate and source of C and energy for soil microorganisms (Khodadad et al. 2011) and possibly it has negative C priming effect. As the pyrolysis temperature increased, DRIFT-MIR spectra of biochar revealed a decrease in C-H stretching (2930 cm-1), this was attributed to the acceleration of pyrolysis reaction in biomass, which suggested a decrease in the polar functional groups with an increase in pyrolysis temperature (Zhao et al. 2018). However, the feedstocks from rice (rice husk and rice straw) showed the presence of a band at 811 cm-1, which was assigned to SiO2. SiO2 is a major chemical component in the structure of rice material (Jindo et al. 2014). The strong bands in the 1637-1558 cm-1 region were found in all biochar types and represent aromatic C=C stretching being an indication of stability of biochar in soil as discussed above.

**Conclusion**

Evidence gathered from the experiment indicated that biochar addition to infertile paddy soil, in combination with fertilizer application, can produce yields greater than either only fertilizer or only biochar application alone. Biochar application reduced fertilizer-N input requirements and increased nitrogen use efficiency due to the increased soil cation exchange capacity via biochar. However, the cation exchange capacity properties, soil improvement, and plant production efficiency of the biochar depended on pyrolysis temperature and feedstock type. Woody biochars, e.g., those of eucalyptus and bamboo, were of most suitable quality (high C/N ratio, CEC, pH and increasing nitrogen use efficiency) for use as soil amendments in paddy cultivation if combined with N fertilizer. However, rice husk biochar showed potential as a sustainable (high C, nutrient retention and CEC) alternative to raw material or rice residue burning, thereby reducing potential negative environmental impacts of paddy rice production.

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**Table 1** Major peaks and assigned functional groups of biochars and rice straw burned residue as measured by DRIFTS-MIRS.

|  |  |  |
| --- | --- | --- |
| Peak wavenumber  /cm-1 | Functional group assignments | Source |
| 3620 | Hydroxyl stretching of various inorganics | Ngyuen et al. (1991) |
| 3400 | OH of phenolic groups and/or bound and unbound OH groups | Haberhauer et al. (1998); Baes and Bloom (1989) |
| 3055 | Aromatic CH stretching | Baes and Bloom (1989) |
| 2930 | Asymmetric CH stretch of -CH2 | Baes and Bloom (1989) |
| 1700 | Carboxyl vibrations of carboxyl groups, aldehydes, and ketones | Senesi et al. (2003) |
| 1616 | Aromatic C=C stretch and/or asymmetric -COO- stretch | Baes and Bloom (1989); Demyan et al. (2012) |
| 1510 | Aromatic C=C vibrations | Niemeyer et al. (1992) |
| 1429 | C=C stretching of aromatic rings | Socrates (1980) |
| 1159 | C-O bonds in both polyalcoholic and ether groups | Spaccini and Piccolo (2007) |
| 811 | Fundamental O-Si-O stretching | Nguoyen et al. (1991) |

**Table 2** Chemical properties of biochar used in this experiment.

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
| Organic material type | Moisture  (%) | pH | CEC  cmolc kg-1 | Total C | Total N | | C/N |
| g kg-1 | | |
| BC | 4.59 | 9.90 | 23.95 | 545 | | 8.9 | 61.5 |
| EC | 3.81 | 9.00 | 45.71 | 661 | | 5.7 | 117.3 |
| RHC | 3.75 | 6.78 | 66.52 | 307 | | 10.4 | 29.9 |
| RSB | nd | 10.10 | nd | 424 | | 8.1 | 52.7 |
| SED  C.V. (%) | 0.79ns  27 | 0.04\*\*\*  1 | 7.66\*  16 | 17\*\*  3 | | 1.4ns  17 | 7.6\*  10 |

\*\*\* = Significant different at *P*<0.001, \*\* = significant different at *P*<0.05, \* = significantly different at *P*<0.05 and ns = not significant at *P*>0.05.

nd = not determined; BC - bamboo biochar; EC - eucalyptus biochar; RHC - rice husk biochar; RSB - rice straw burned residue

**Table 3** Effect of biochar application on rice height and dry weight (DW) at 30 and 60 days after transplanting (DAT).

|  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Treatment | | 30 DAT | | | |  | 60 DAT | | | |
| Height  (cm) | Tiller pot-1 | Shoot DW  (g pot-1) | N  content  (g stem-1) |  | Height  (cm) | Tiller pot-1 | Shoot DW  (g pot-1) | N  content  (g pot-1) |
| - Fertilizer | No-amended | 39 | 4 | 1.31 | 0.010 |  | 65 | 10 | 18.75 | 0.220 |
| + fertilizer | No-amended | 36 | 3 | 0.80 | 0.016 |  | 89 | 24 | 40.50 | 0.314 |
|  | BC | 37 | 3 | 1.54 | 0.010 |  | 66 | 13 | 27.00 | 1.041 |
|  | EC | 31 | 3 | 0.46 | 0.015 |  | 71 | 11 | 24.50 | 0.320 |
|  | RHC | 37 | 3 | 1.92 | 0.010 |  | 65 | 11 | 29.50 | 0.244 |
|  | RSB | 34 | 3 | 0.56 | 0.016 |  | 52 | 11 | 27.25 | 0.301 |
| SED  C.V. (%) | | 2\*\*  7 | 0.3ns  14 | 0.13\*\*  15 | 0.002\*\*  24 |  | 7\*  14 | 1\*\*  13 | 2.40\*\*  12 | 0.030\*\*\*  10 |

\*\* = significant different at *P*<0.05, \* = significantly different at *P*<0.05 and ns = not significant at *P*>0.05. BC - bamboo biochar; EC - eucalyptus biochar; RHC - rice husk biochar; RSB - rice straw burned residue

**Table 4** Effect of biochar and N fertilizer application on rice spike numbers, shoot dry weight (DW), seed yield, N content, filled grains percentage and harvest index (HI) at rice harvest.

|  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Treatment | | Spike number  pot-1 | Shoot | |  | Grain yield | | Total  DW | Total  N | %Filled grains | HI |
| DW | N |  | DW | N |
| (g pot-1) | | | | | | |  |  |
| - Fertilizer | No-amended  BC  EC  RHC  RSB | 11  10  9  10  11 | 21  15  29  21  27 | 0.106  0.074  0.148  0.116  0.111 |  | 10  12  7  11  12 | 0.109  0.119  0.219  0.140  0.152 | 31  27  36  32  40 | 0.215  0.193  0.233  0.255  0.263 | 93  94  85  89  90 | 0.32  0.45  0.20  0.35  0.30 |
| + fertilizer | No-amended  BC  EC  RHC  RSB | 25  12  10  11  10 | 89  24  32  21  28 | 1.492  0.153  0.148  0.103  0.085 |  | 15  15  12  13  12 | 0.214  0.168  0.085  0.107  0.151 | 105  39  44  34  40 | 1.706  0.320  0.233  0.290  0.371 | 85  91  93  93  91 | 0.15  0.40  0.32  0.40  0.32 |
| SED | | 3\*\*\* | 6\*\* | 0.015\*\* |  | 2\* | 0.026\*\* | 6.54\*\* | 0.029\*\* | 3\* | 0.05\*\* |
| C.V. (%) | | 36 | 26 | 7 |  | 23 | 27 | 22 | 10 | 4 | 22 |

\*\* = Significantly different at *P*<0.001, and \* = significantly different at *P*<0.05. BC - bamboo biochar; EC - eucalyptus biochar; RHC - rice husk biochar; RSB - rice straw burned residue

**Table 5** Nitrogen use efficiency (NUE), internal efficiency of N (IEN), agronomic yield nitrogen use efficiency (ANUE) and apparent fertilizer use efficiency (AFUE) of rice under different soil management treatments.

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| Treatment | | NUE | IEN | ANUE | AFUE  (%) |
| (g DW g-1 N) | | |
| - Fertilizer | No-amended  BC  EC  RHC  RSB | 145  139  155  153  178 | 47  62  31  53  51 | nd  nd  nd  nd  nd | nd  nd  nd  nd  nd |
| + fertilizer | No-amended  BC  EC  RHC  RSB | 61  122  173  130  108 | 9  47  49  51  33 | 66  115  95  103  93 | 87  40  14  20  37 |
| SED | | 25.41\*\* | 4.72\*\* | 11.80\* | 5.52\*\* |
| C.V. (%) | | 26 | 15 | 18 | 20 |

\*\* = Significantly different at *P*<0.001, and \* = significantly different at *P*<0.05.

nd = not determined. BC - bamboo biochar; EC - eucalyptus biochar; RHC - rice husk biochar; RSB - rice straw burned residue

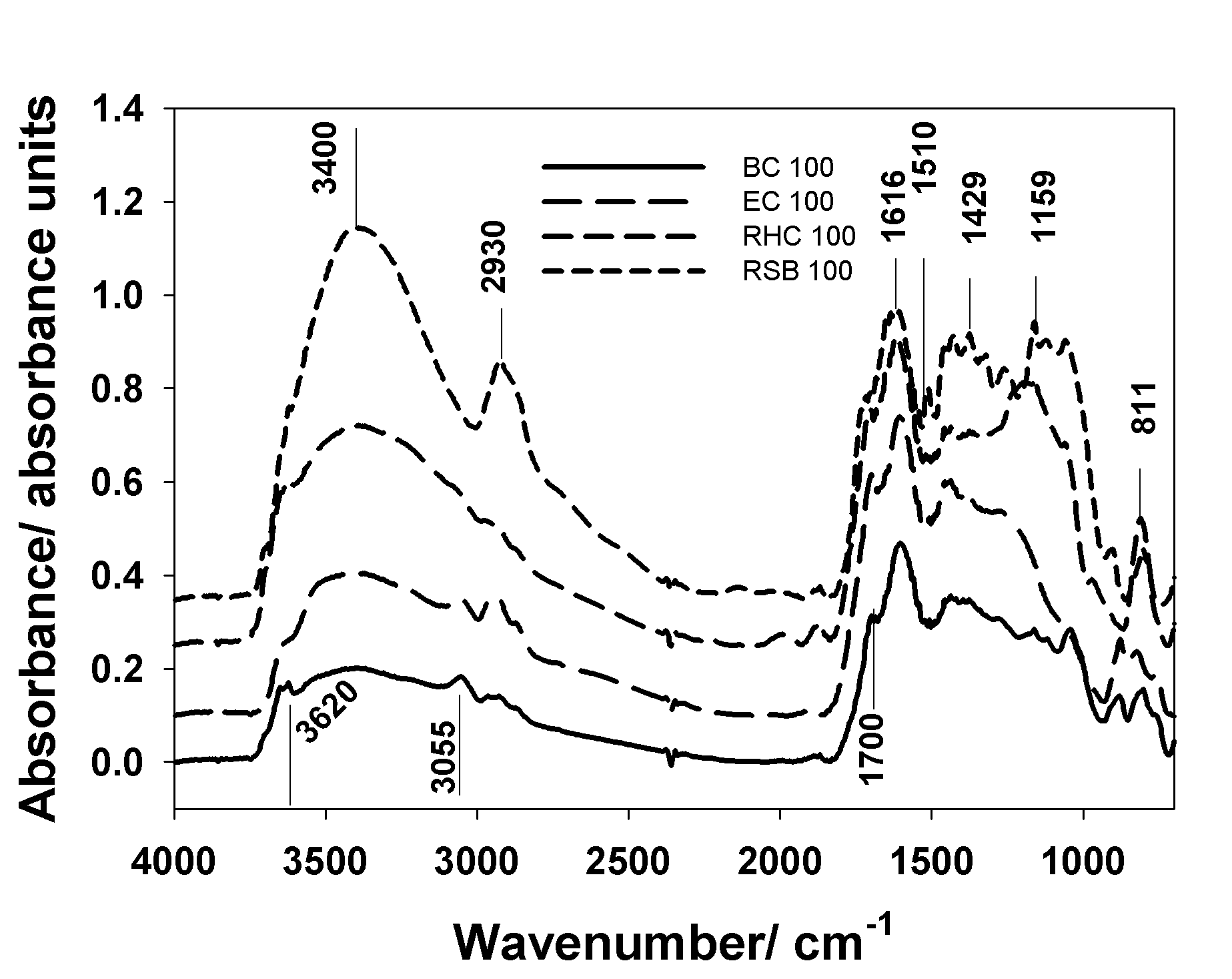
|  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Treatment | | pH | | Total C | | Total N | C/N | Avail. P | Exch. K | CEC |
| (g kg-1) | | |  | (mg kg-1) | | (cmolc kg-1) |
| No amended  BC  EC  RHC  RSB | | 5.18  5.68  5.53  5.60  5.31 | | 5.32  14.34  14.96  8.15  3.26 | 0.70  0.79  1.36  0.88  0.84 | | 7.60  18.31  11.00  9.21  4.34 | 7.80  26.87  8.26  13.20  9.10 | 0.017  0.046  0.024  0.019  0.019 | 1.72  4.12  9.03  4.91  4.69 |
| SED |  | | 0.31ns | 0.20\*\* | 0.01\*\* | | 0.32\*\* | 2.63\*\* | 0.004\*  17 | 0.44\*\*  9 |
| C.V. (%) |  | | 6 | 2 | 6 | | 3 | 18 |

**Table 6** Soil chemical properties taken after rice harvest as affected by chemical fertilizer and biochar amendment without fertilizer.

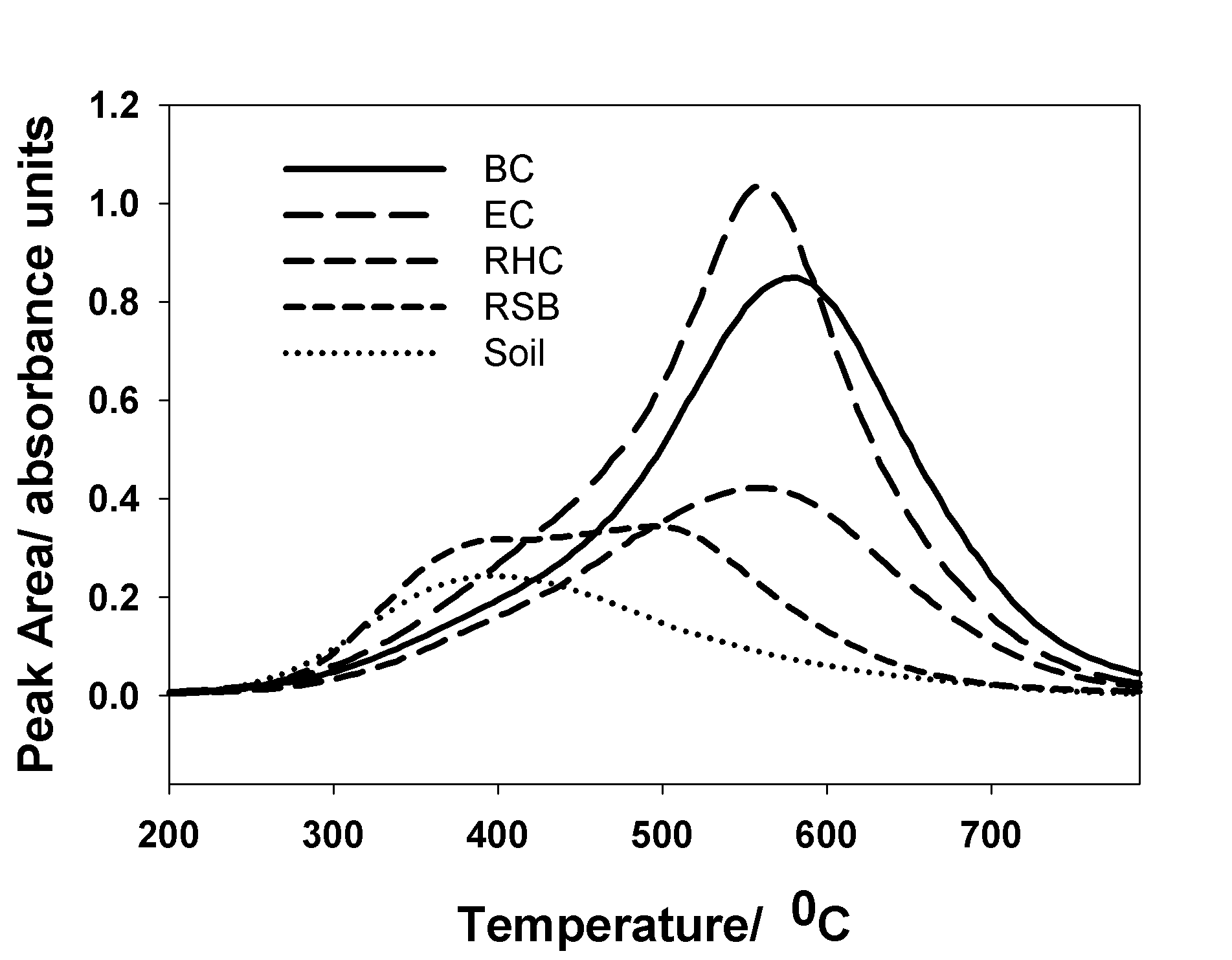
\*\* = significantly different at *P*<0.01, \* = significantly different at *P*<0.05 and ns = not significant at *P*>0.05. BC - bamboo biochar; EC - eucalyptus biochar; RHC - rice husk biochar; RSB - rice straw burned residue

**Table 7** Temperature of maximum CO2 evolution as measured by FTIR-EGA of different biochar types and an unamended soil. Average of two replicates.

|  |  |  |
| --- | --- | --- |
| biochar type | peak temperature/°C | standard error/°C |
| bamboo | 578 | 5 |
| eucalyptus | 560 | 5 |
| rice husk | 560 | 0 |
| rice straw burned | 494 | 2 |
| unamended soil | 401 | 3 |



**Figure 1.** DRIFTS-MIRS spectra in four type of biochars. BC - bamboo biochar; EC - eucalyptus biochar; RHC - rice husk biochar; RSB - rice straw burned residue



**Figure 2.** Evolved CO2 as measured via FTIR-EGA of four types of biochar and an unamended soil. The samples were heated from room 25 to 700°C at a ramping rate of 68°C min-1 and then held at 700°C for an additional 10 minutes. BC - bamboo biochar; EC - eucalyptus biochar; RHC - rice husk biochar; RSB - rice straw burned residue