



## Improvements in wheat productivity and soil quality can accomplish by co-application of biochars and chemical fertilizers



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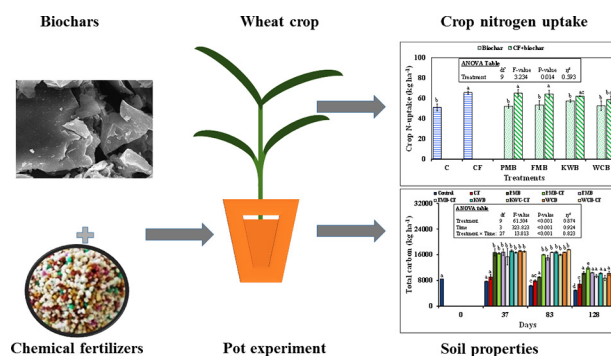
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### HIGHLIGHTS

- Nutrient, structure biochars and chemical fertilizer (CF) tested on wheat growth
- CF increased the wheat yield but decreased soil carbon content.
- Grain yield was higher in *nutrient* (manure) than *structure* biochars.
- *Structure* biochar from wood chips negatively affected grain yield.
- High wheat yield in co-applied *nutrient* biochars and CF than *structure* biochars

### GRAPHICAL ABSTRACT



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### ABSTRACT

The beneficial role of biochar is evident in most of infertile soils, however this is argued that increment in crop yield owing to biochar application does not always achieve in cultivated/fertile soils. The *nutrient* biochar believed to enhance crop yield and soil fertility than *structural biochar* that may offset the positive effect of chemical fertilizer on crop performance but improves soil structural properties. Therefore, we investigated the effect of biochars [produced from nutrient rich feedstocks like poultry manure (PMB) and farmyard manure (FMB) and structural feedstocks such as wood chips (WCB) and kitchen waste (KWB)], and chemical fertilizers (CF) when applied alone or in combination on soil chemical properties, wheat growth, yield and nitrogen uptake in a cultivated clay loam soil. Sole biochar treatments increased the total carbon and mineral nitrogen content that were 21 and 106% higher, respectively compared to control after 128 days ( $P < 0.001$ ). Contrarily, sole biochars application did not increase wheat biological yield and N uptake compared to control ( $P > 0.05$ ) except PMB, the *nutrient* biochar ( $P < 0.05$ ). Compared to control, grain yield was 6 and 12% lower in WCB and FMB, respectively but not differed from KWB, PMB or WCB-CF. Conversely, co-application of biochars and CF treatments increased crop biological yield but the increment was the highest in *nutrient biochars* FMB or PMB (29 or 26%), than *structural biochars* WCB and KWB (15 and 13%), respectively ( $P < 0.05$ ). For N uptake, this increment varies between 16 and 27% and again *nutrient biochar* has significantly higher N uptake than *structural biochars*. Hence, *nutrient biochars* (i.e. PMB) benefited the soil fertility and crop productivity more than *structural biochars*. Therefore, for

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immediate crop benefits, it is recommended to use *nutrient biochar* alone or in combination with chemical fertilizer. Such practice will improve crop performance and the quality of cultivated soil.

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## 1. Introduction

Land degradation will become a global peril to crop production and food security in the coming decades. It is estimated that 30% of the world agricultural soil will be converted into degraded land in the next few decades (Oldeman et al., 1990; Rashid et al., 2016). The situation is even worse in Pakistan where 61% of cultivated soils were under serious threat of degradation in 2006 (Anonymous, 2006). Only in Punjab province, 1.58 million hectares of sandy and loamy soils were strongly deficient in nutrients thus termed as nutrients depleted soils (Alim and Javed, 1993; Hassan and Arshad, 2006). This could be the results of growing exhaustive crops, inappropriate agriculture practices, like tillage (Wu et al., 2017), use of high dosage of chemical fertilizers especially nitrogen enhanced light fraction of soil carbon decomposition (Neff et al., 2002) and low carbon inputs resulted in serious decline of soil organic matter (Lu et al., 2011), which is on average 0.5% at current in Pakistani soils (Hassan and Arshad, 2006). Such causative factors could be among the main roots of decrease in crop yield in the country. For instance, reduction in wheat (*Triticum aestivum* L.) yield, the main staple crop of the region, was approximately 5% from year 2008 to 2013 (FAO, 2013) and further declining trend in yield was observed up to last year (Alam, 2016). The decrement in the yield could be attributed to inefficient use of chemical fertilizers especially nitrogen. Since, this nutrient is applied at the time of sowing mostly in the form of urea therefore, significant amount of applied nitrogen is lost through volatilization, leaching, nitrification and denitrification processes, ultimately less ends up in plant. The problem could be solved by supplying adequate N levels in the soil and its subsequent uptake by the crop (Malhi et al., 2006; Rashid et al., 2013) as well as by increasing soil fertility especially organic matter or carbon content in the soil (Kaneez-e-Batool et al., 2016; Rashid et al., 2014a; Rashid et al., 2014b; Rashid et al., 2017; Rashid et al., 2016; Shah et al., 2012) or co-applied biochar with chemical fertilizers that may synchronize the soil nutrient availability with crop N demand (Agegnehu et al., 2016). According to an estimate, irrigated agricultural soil of Pakistan has a potential to sequester 1.04–2.08 Tg carbon year<sup>-1</sup> (Khan and Lal, 2007). Such huge potential would urge scientists to find suitable approaches or management strategies for carbon sequestration and would be able to increase the organic matter content up to 2% (Lal, 2004). By doing so the soil nutrient deficiency problem might be solved (Khan and Lal, 2007) and would be helpful in improving the crop yields.

Animal manures and poultry litters are among the largest waste streams in Pakistan. It is estimated that annual waste production from animals was approximately 2.5 million tons (Mangalwala, 2014). This huge amount of waste was two times higher than municipal solid waste production in the country which is mainly dominated by food waste (about 60%) (Kamran et al., 2015). Improper management of these wastes, such as open dumping of kitchen waste, poultry litter or animal dung and the later waste also used as an energy source for burning stoves in the houses resulted in an increased greenhouse gaseous emission to the atmosphere that are prone to global warming in the region (Gustafsson et al., 2009; Irwin, 2015). Therefore, sustainable and smart management practices are required to reuse this waste in crop management strategy.

Biochar is a carbonaceous material obtained through pyrolysis of plant biomass or animal wastes (López-Cano et al., 2016) in absence of oxygen. The most recent technology developed to prepare charred material from waste with the intent to mitigate climate change by sequestering carbon when applied to the soil (Lehmann and Joseph, 2015). Likewise, biochar also enhanced other important fertility indicators when applied to the soil including carbon content, reduced nitrate

mineralization (Marks et al., 2016) and therefore decreased the leaching losses of C and N from soil (Bass et al., 2016; Haider et al., 2017). Biochar application to soil proved to be beneficial for improving soil fertility and carbon sequestration of degraded soil (Yeboah et al., 2009). Due to a highly basic product, biochar changes the soil pH through its interaction with H<sup>+</sup> ions (Barrow, 2012; Liu and Zhang, 2012). The high porosity and recalcitrant nature of biochar decreases the mineralization rate thus helps in slow release of plant available nutrients.

Depending on the feedstock and temperature used to produce biochar, many studies ended up into conflicting results for crop yield and plant nutrient availability (Bass et al., 2016; Haider et al., 2017; Hussain et al., 2016; Sun et al., 2014). The variations in crop yield and soil properties indexes due to biochar soil application were ranged between –36 to 31% and –21 to 101%, respectively (Hussain et al., 2016). These differences could be linked to the nature of the feedstock used to prepare various biochars, i.e. the *nutrient* or *structure* biochars produced from animal manure or plant waste, respectively (Jeffery et al., 2017). On the other hand, a recent meta-analysis study indicated no significant mean response of biochar application to soil, mineral nitrogen, aboveground plant productivity and crop nitrogen uptake (Biederman and Harpole, 2013). Hussain et al. (2016) concluded from the literature review that biochar application enhanced the crop yield in highly degraded or infertile soil but this parameter was not enhanced when biochar was applied to cultivated, fertile and/or healthy soils. This means that biochar does not always enhance crop yield when applied on cultivated soil. Consequently, integrated application of biochar and chemical fertilizer could be suitable management strategy for improving crop yield in such soils (Agegnehu et al., 2016; Ali et al., 2015; Fageria and Baligar, 2005). However, only few studies have been carried out to evaluate the integrated effect of biochar as soil amendment to improve the efficiency of chemical fertilizers (Agegnehu et al., 2016; Ali et al., 2015; Brantley et al., 2016; Gul and Whalen, 2016; Tammeorg et al., 2014b). Hitherto, Tammeorg et al. (2014b) did not find any increase in wheat yield after combined application of meat-bone meal biochar and chemical fertilizer. Therefore, for integrated application of biochars and/or chemical fertilizer, appropriate fractions of chemical fertilizer with biochar or different biochars in a blend are required (Sigua et al., 2016; Tammeorg et al., 2014b). To elucidate the former blending, a study was conducted in pots under semi-field conditions to investigate the influence of different biochars produced from various organic wastes and chemical fertilizer in enhancing wheat yield and crop nitrogen utilization when applied alone or in combination in a clay loam soil. We hypothesized that i) sole application of biochar will enhance soil carbon content ii) biochar produced from nutrient rich feedstock when applied in cultivated soil will enhance the wheat yield compared to structural biochar that will only improve soil structural properties, iii) co-application of both *structural* and *nutrient* biochar with chemical fertilizer will increase the soil fertility status, wheat yield and nitrogen uptake, iv) the increment in wheat yield would be variable among various biochars (*nutrient* or *structure*) and chemical fertilizer blending since differences in the feedstocks of biochars will influence their end-product quality.

## 2. Materials and methods

### 2.1. Biochar production

Biochar were produced from farmyard manure (FYM), poultry manure (PM), wood chips (WC) and kitchen waste (KW) in a laboratory-scale pyrolysis unit. *Dalbergia sissoo* wood chips were collected from

the furniture shops near the university campus. Organic wastes such as FYM and PM were collected from the poultry and cattle farm of the university, whereas the KW was collected from the university cafeteria. These materials were sun dried for few days and then converted to biochar using laboratory-scale vertical cylinder pyrolysis unit equipped with external electric heating system (Gangil and Wakudkar, 2013) at 500 °C for 5 h under limited oxygen conditions. After production, each biochar was analyzed for pH, EC, total carbon and nitrogen content (Table 1).

## 2.2. Pot experiment

A standard pot experiment was performed with a clay loam soil in the research vicinity of PMAS-Arid Agriculture University, Rawalpindi, Pakistan. Soil was collected from the University research field where sorghum was previously grown, and classified as Rawal series, Udic Haplustalf Alfisols (Govt. of Pakistan, 1974). After collection, soil was passed through 2 mm sieve to remove the root debris and 5 kg of this fresh soil was transferred to each earthen pot with diameter 22 cm and depth 24 cm. All treatments including sole application of biochars or chemical fertilizers (CF) and their combinations were added in these pots. In total, ten treatments, each in triplicates (10 × 3), were allocated in 30 earthen pots. These includes i) control soil (C), ii) CF at the recommended dose, iii) Poultry manure biochar (PMB), iv) farmyard manure biochar (FMB), v) kitchen waste biochar (KWB), vi) wood chip biochar (WCB), vii) PMB and CF (PMB-CF), viii) FMB and CF (FMB-CF), ix) KWB and CF (KWB-CF), and x) WCB and CF (WCB-CF). At wheat sowing time, each biochar was applied/mixed in the soil at top 15 cm soil layer at a rate of 20 tons ha<sup>-1</sup> (Gao et al., 2016). Likewise, in CF treatment, nitrogen, phosphorous and potassium were applied at the wheat crop recommended doses of 87, 87 and 30 kg ha<sup>-1</sup>, respectively corresponding to their source urea (115.1 kg ha<sup>-1</sup>), diammonium phosphate (189.1 kg ha<sup>-1</sup>) and muriate of potash (50 kg ha<sup>-1</sup>). In the blend treatments, same rates of biochar and artificial fertilizer were used as in case of their lone application (Brantley et al., 2016). After treatments application, 17 wheat seeds were hand sown in each pot. To provide the natural environmental conditions to each experimental unit, the pots were arranged in a completely randomized design outside in an open space. Cumulative rainfall and mean temperature during the experimental period is presented in Fig. 1. The pot was irrigated regularly to maintain 60% water holding capacity by adding the water through hand sprinkler with an extra care and increment in this parameter was followed using a low-cost moisture meter (FY-901, Hangzhou FCJ I&E Co., Ltd, China).

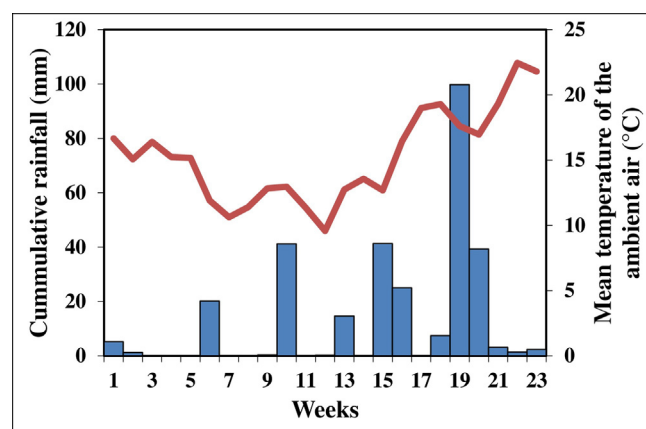
## 2.3. Chemical analysis for soil and biochar

To evaluate the effect of biochar and fertilizer treatments on soil chemical properties, four samplings were done during the course of experiment. Initially, soil was sampled before biochar or fertilizer application in the pots and then at tillering and booting stage while the last sampling was carried out after crop harvest. At each time interval, soil was sampled at three random locations from each pot using hand

**Table 1**

Mean (n = 3, S.E ± 1) of the chemical characteristics such as pH, electrical conductivity (EC), total nitrogen (N<sub>total</sub>) and carbon (TC), and carbon to nitrogen ratio (C:N) of biochars produced from poultry manure (PMB), farmyard manure (FMB), kitchen waste (KWB) and wood chips (WCB).

Biochar	pH	EC dS m <sup>-1</sup>	N <sub>total</sub> g kg <sup>-1</sup>	TC g kg <sup>-1</sup>	C:N
PMB	6.6 ± 0.19	16.92 ± 1.33	31.5 ± 0.41	530 ± 15.2	17
FMB	7.6 ± 0.21	30.54 ± 2.50	5.8 ± 0.07	542 ± 42.1	93
KWB	6.3 ± 0.15	29.69 ± 3.66	25.1 ± 0.06	495 ± 34.5	20
WCB	6.1 ± 0.55	91.90 ± 7.31	2.2 ± 0.05	509 ± 23.2	231



**Fig. 1.** Climatic data, cumulative rainfall and mean temperature of the ambient air, during the experimental period.

augur and these samples were mixed thoroughly to make a composite sample. Each composite sample was analyzed for EC, TOC and N content.

Electrical conductivity (EC) of the soil/biochar was determined according to Agegnehu et al. (2016) by preparing a biochar water suspension (1:2.5) and the contents were allowed to equilibrate at room temperature for 30 min. A multi-meter (Ino-Lab® Multi 9430 IDS, WTW, GmbH & Co. KG, Germany) was used to measure the EC of the prepared solution from each treatment, after standardizing with 0.01 N KCl solution at 25 °C (Page, 1982). The carbon content of biochar/soil was determined through wet oxidation of the samples by means of chromic acid, hydrogen peroxide, and sulfuric acid (Walkley and Black, 1934) as carried out by Agegnehu et al. (2016) for carbon determination in soil and biochar. Total N content in soil, plant or biochar samples were measured through FOSS Kjeltec™ 8400 Auto Sampler System (Eden Prairie, USA).

## 2.4. Plant growth and yield parameters

Wheat crop growth and yield parameters such as germination percentage, chlorophyll content, leaf area, plant height, panicle length, number of spikelets per panicle, number of grains per panicle, hundred grains weight, biological yield, grain yield, root biomass, and harvest index were determined. Germination percentage was calculated by counting the germinated seeds of wheat crop after one week of sowing. Following Eq. (1) was used to calculate wheat germination.

$$\text{Germination (\%)} = \frac{SE}{SS} \times 100 \quad (1)$$

where SE denotes total number of seedlings emerged after one week and SS represents the total number of seed sown.

Chlorophyll content and leaf area was measured by using Spad chlorophyll and leaf area meters. For this purpose, 10 independent measurements were made from each treatment using various wheat plants.

Wheat crop was harvested at maturity stage, and the biological yield was measured by weighing the above ground parts of all plants in each pot after complete drying in an oven for 48 h at 70 °C until constant weight of each sample was obtained. For other parameters, we randomly selected three plants from each pot at maturity stage and their plant height and panicle length were determined with meter rod (cm). From each panicle of the selected plants, spikelets were counted and then they manually crushed to calculate the number of grains per panicle. Subsequently, panicles of all the plants in each pot was manually crushed then 100 and total grains were weighed on a digital balance to determine the 100 grain weight and total grain yield of each pot.

## 2.5. Root biomass

The soil clump from each pot was removed to separate the roots. Laterally, the clump was placed in cold-water container for few hours. Afterwards, each clump was broken into four pieces and placed on 1 mm mesh sieve. The roots were separated from soil with a jet of tap water. The collected roots were completely oven dried for 48 h at 70 °C until constant weight was obtained and then weighed on digital balance to measure their biomass.

## 2.6. Harvest index (%)

Harvest index was calculated by following equation.

$$HI = \frac{EY}{BY} \times 100 \quad (2)$$

where HI is the harvest index of wheat crop. EY represents the economic yield, which is the total grain yield and BY indicates the biological yield of the wheat crop.

## 2.7. Apparent N recovery

Apparent crop N recovery from all the treatment was calculated by using Eq. (3).

$$ANR (\%) = \frac{(NC_{BF} \times DM_{BF}) - (NC_{control} \times DM_{control})}{TN_{applied}} \quad (3)$$

where  $NC_{BF}$  is nitrogen content ( $gN\ 100\ g^{-1}\ DM$ ) of wheat crop (vegetative and reproductive parts) in fertilized treatment,  $DM_{BF}$  indicates wheat dry matter yield ( $kg\ ha^{-1}$ ) in fertilized treatment.  $NC_{control}$  represents wheat nitrogen content ( $gN\ 100\ g^{-1}\ DM$ ) in control (unfertilized) treatment,  $DM_{control}$  is the wheat dry matter yield ( $kg\ ha^{-1}$ ) in control (unfertilized).  $TN_{applied}$  shows the total N rate ( $kg\ ha^{-1}$ ) applied per treatment.

## 2.8. Statistical analysis

All treatments effect was subjected to univariate analysis using SPSS Statistics 17.0 (IBM, New York, USA). Since effect of biochars or chemical fertilizers on soil parameters was observed with different time intervals, therefore time was also considered a main factor in these analyses. The main effects of treatments at three different time intervals were tested by analysis of variance (ANOVA). The significance among treatment was tested at 5% probability level. Tukey's and LSD tests analyzed the multiple comparison among various treatments. The influence of treatments, such as biochars and chemical fertilizer on soil chemical properties and wheat growth and yield parameters and their relationships among each other were analyzed by principal component analysis (PCA) through multivariate analysis software CANOCO 5.0 for Windows (Microcomputer Power Inc., Ithaca, NY) on correlation matrices.

## 3. Results

### 3.1. Biochar influence on soil properties

Overall, treatment and time effects on soil total carbon was highly significant ( $P = 0.000$ ). All biochar treatments including their combination with chemical fertilizer significantly increased this parameter compared to control or CF treatments after 37 days of their application to soil (Fig. 2A). This increment was 135% (18,161 vs. 7732  $kg\ ha^{-1}$ ) in case of KWB, 122% (17,202 vs. 7732  $kg\ ha^{-1}$ ) for WCB, 117% (16,783 vs. 7732  $kg\ ha^{-1}$ ) for FMB and 116% (16,723 vs. 7732  $kg\ ha^{-1}$ ) for PMB ( $P < 0.001$ ; Fig. 2A). In treatments with combination of biochar and chemical fertilizer, no further increment or decrease in total carbon

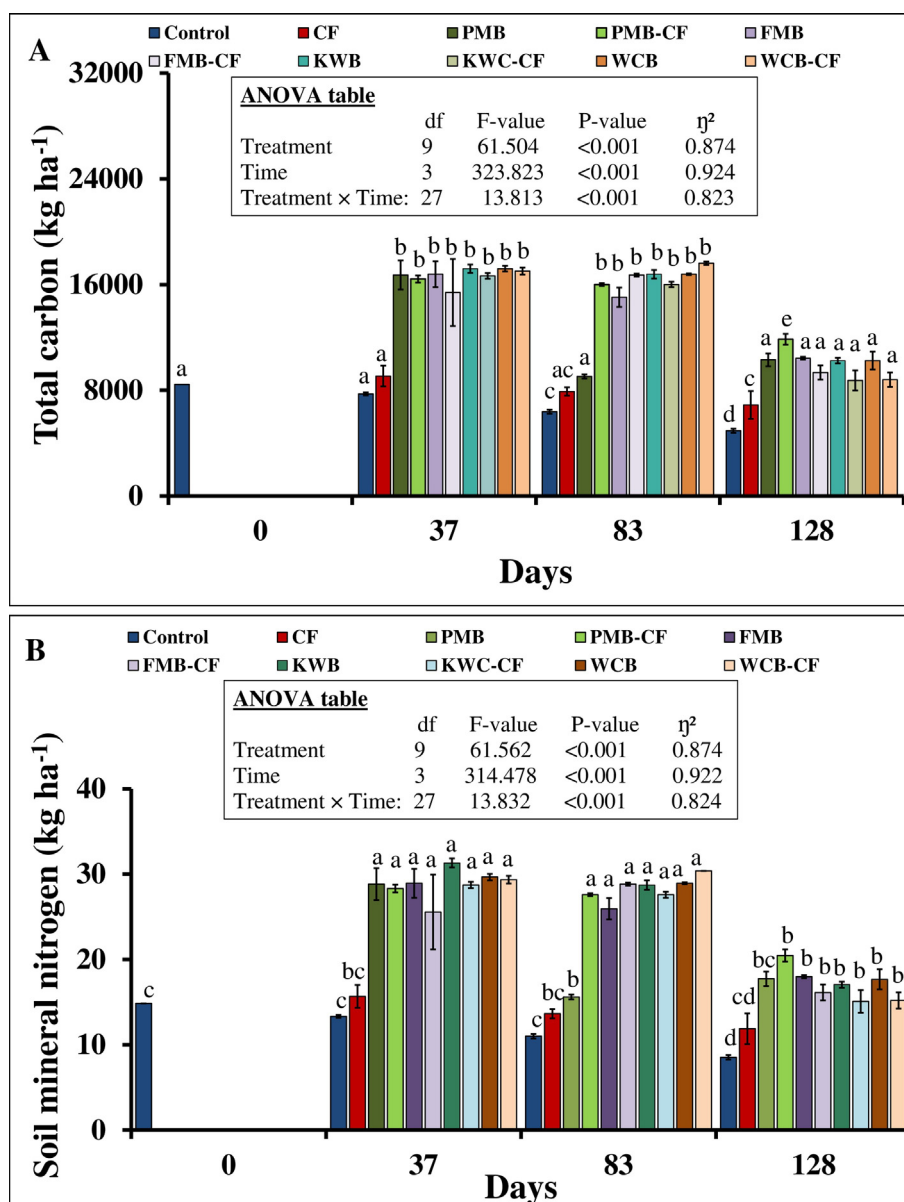
content was observed ( $P > 0.05$ ). Effect of time on soil total carbon was also significant, showing an overall decrease in this parameter ( $P < 0.001$ ). This decrease was observed at each time interval for control treatment. However, for biochar treatments, the decrease in total carbon was only observed after 128 days compared to the carbon content after 37 days. After 128 days of application (at the end of experiment), soil carbon content in control was decreased by 41% (4945 vs. 8450  $kg\ ha^{-1}$ ) whereas all biochar treatments increased carbon content in the soil to about 21% (10,219 vs. 8450  $kg\ ha^{-1}$ ). In case of PMB-CF treatments, the increase in soil carbon content was 40% (11,868 vs. 8450  $kg\ ha^{-1}$ ) at the end of experiment ( $P < 0.05$ ; Fig. 2A). Like total carbon, similar trend in soil mineral nitrogen ( $NH_4^+-N$  and  $NO_3^- -N$ ) was observed (Fig. 2B). In general, biochar treatments significantly increased the mineral N in soil. This increment was observed for all biochar treatments after 37 ( $P < 0.001$ ) and 83 days of treatments application ( $P < 0.001$ ), however at the end of experiment (after 128 days) biochar associated increment in soil mineral N was decreased ( $P < 0.001$ ). Although this decrease was not significantly different from initial mineral N of the soil at 0 day ( $P > 0.05$ ) but it was significantly higher in the biochar treatments compared to control ( $P < 0.05$ ). After 37 days, the highest increase (111%: 31.3 vs. 14.9  $kg\ ha^{-1}$ ) in mineral N content was observed in KWB compared to soil initial mineral N while in other biochar treatments the increment was 100 (29.7 vs. 14.9  $kg\ ha^{-1}$ ), 95 (28.9 vs. 14.9  $kg\ ha^{-1}$ ), 94% (28.8 vs. 14.9  $kg\ ha^{-1}$ ) for WCB, FMB and PMB, respectively (Fig. 2B;  $P < 0.001$ ). In treatment with combination of chemical fertilizer and biochars, similar trend was observed but here the highest increase in mineral N was for WCB (98%: 29.4 vs. 14.9  $kg\ ha^{-1}$ ) followed KWB (93%: 28.7 vs. 14.9  $kg\ ha^{-1}$ ), PMB (91%: 28.3 vs. 14.9  $kg\ ha^{-1}$ ) and FMB (79%: 26.6 vs. 14.9  $kg\ ha^{-1}$ ). However, mineral N in CF treatment was not significantly differed from initial or control treatment ( $P > 0.05$ ). Like total carbon, mineral N content in soil with time decreased and the lowest decline was observed at the end of experiment. At this time, no difference in mineral N content in the soil was observed among all treatments expect control that was 43% (8.5 vs. 14.9  $kg\ ha^{-1}$ ) lower compared to initial soil mineral N (Fig. 2B;  $P < 0.001$ ) and on average 52% (9 vs. 18  $kg\ ha^{-1}$ ) lower than all biochar treatments.

### 3.2. Wheat growth parameters

In general, wheat germination was not significantly affected by any treatment ( $P > 0.05$ ). However, multiple comparison revealed that germination in PMB-CF and FMB-CF was 9 (78.7 vs. 86.0%) and 17% (71 vs. 86%) lower compared to control treatment ( $P < 0.05$ ). Similarly, chemical fertilizer application in KWB treatment significantly decreased (17%: 72.7 vs. 88.0%) the wheat germination compared to this sole biochar application (Table 3). Other treatments were not significantly different from each other or control. Chlorophyll content of crop leaves was significantly higher in the CF treatment compared to control ( $P < 0.001$ ; Table 3). However, any biochar application or biochar in combination with CF did significantly increased chlorophyll content of wheat leaves than control ( $P > 0.05$ ). Similarly, sole biochar treatments and CF, or their combination ( $P > 0.05$ ; Table 3) did not affect the plant height. Root biomass differed among all treatments in following order PMB-CF > CF > FMB-CF = KWB-CF = KWB > Control > PMB > FMB = WCB ( $P < 0.001$ ; Table 3). A 73% (855 vs. 494  $kg\ ha^{-1}$ ) increase in root biomass was observed in case of PMB-CF treatment compared to control and this increase was 58% (779 vs. 494  $kg\ ha^{-1}$ ) for CF. However, WCB/FMB and PMB decreased this parameter by 33% (330 vs. 494  $kg\ ha^{-1}$ ) and 16% (416 vs. 494  $kg\ ha^{-1}$ ), respectively than control ( $P < 0.05$ ; Table 3).

### 3.3. Wheat yield parameters

Table 3 presents the wheat yield parameters as influenced by the application of chemical fertilizers, biochars and their combination.



**Fig. 2.** Mean ( $n = 3$ ) soil carbon (A) and mineral nitrogen content (B) as influenced by time, biochars produced from poultry manure (PMB), farmyard manure (FMB), kitchen waste (KWB) and wood chip waste (WCB) and combination of all biochars with chemical fertilizers at various time intervals. Significant difference among treatments was represented by small letter. Multiple comparisons among treatments were analyzed by Tukey test.

Biological yield of the wheat from the treatments PMB, FMB and WCB was not significantly different from control ( $P > 0.05$ ). This parameter was 35% (7679 vs. 5699  $\text{kg ha}^{-1}$ ) and 7% (6100 vs. 5699  $\text{kg ha}^{-1}$ ) higher in CF and KWB treatments, respectively than control ( $P < 0.05$ ; Table 3). Chemical fertilizer application increased the biological yield by 29 (6956 vs. 5378  $\text{kg ha}^{-1}$ ), 26 (7144 vs. 5672  $\text{kg ha}^{-1}$ ), 15 (6448 vs. 5592  $\text{kg ha}^{-1}$ ) and 13% (6903 vs. 6100  $\text{kg ha}^{-1}$ ) when applied in combination with FMB, PMB, WCB and KWB, respectively ( $P < 0.05$ ; Table 3). Like biological yield, grain yield was also significantly affected by all treatment application ( $P < 0.001$ ). A 12% (1980 vs. 2256  $\text{kg ha}^{-1}$ ) and 6% (2116 vs. 2256  $\text{kg ha}^{-1}$ ) decrease in wheat grain yield was observed in FMB and WCB treatments, respectively compared to control. Grain yield obtained from PMB, KWB and WCB-CF was not differed from control. The highest increase in this parameter was observed for PMB-CF treatment, which was 32% (2973 vs. 2256  $\text{kg ha}^{-1}$ ) than control and 29% (2973 vs. 2313  $\text{kg ha}^{-1}$ ) than its lone counterpart (PMB). This was followed by FMB-CF who increased the grain yield by 30% (2924 vs. 2256  $\text{kg ha}^{-1}$ ) from control and 48% (2924 vs.

1980  $\text{kg ha}^{-1}$ ) from FMB. Sole application of CF increased the grain yield by 27% (2866 vs. 2256  $\text{kg ha}^{-1}$ ) compared to control treatment. However, CF when co-applied with KWB increased grain yield by only 6% (2459 vs. 2313  $\text{kg ha}^{-1}$ ) compared to its sole counterpart. Harvest index (%) was not significantly affected by any treatment ( $P = 0.06$ ). In general, the number of grains per panicle ranged between 22 and 31 grains panicle<sup>-1</sup>. Application of biochars decreased this parameter by 4–14% compared to control ( $P < 0.001$ ). However, in the treatments where biochar was applied in combination with chemical fertilizer grains panicle<sup>-1</sup> were significantly increased from control and their sole counterpart. For instance, application of chemical fertilizer in PMB increased this parameter by 26% (31 vs. 24.7 grains panicle<sup>-1</sup>) and the highest increase (29%: 28.3 vs. 22 grains panicle<sup>-1</sup>) was observed in case WCB. However the lowest increase was from KWB treatment where CF increased this parameter by only 15% (28.3 vs. 24.7 grains panicle<sup>-1</sup>) but it was significantly different than control. The CF treatments alone did not increase grains panicle<sup>-1</sup> compared to control. Number of spikelets per panicle was also significantly

lower in FMB, KWB, WCB compared to control however, this parameter was significantly higher in CF treatment compared to control but not differed between control and PMB treatments (Table 3). Chemical fertilizer application in biochars was able to increase this parameter which was significantly higher in PMB (30%: 17.3 vs. 13.3 spikelets panicle<sup>-1</sup>) and FMB (31%: 16.7 vs. 12.7 spikelets panicle<sup>-1</sup>) treatments compared to their lone counterpart as well as from control. Panicle length was also significantly influenced by the application of various treatments. Chemical fertilizer and PMB significantly increased the panicle length compared to control, however this parameter was lower for KWB and WCB than control. Combined application of CF and KWB or WCB significantly enhanced panicle length than control as well as from their sole counterpart. In general, the highest length was observed in PMB-CF (27%: 9.5 vs. 7.5 cm) and FMB-CF (24%: 9.3 vs. 7.5 cm) treatment compared to control (Table 3).

### 3.4. Wheat nitrogen uptake and crop apparent nitrogen recovery

Crop nitrogen uptake was also significantly influenced by the application of biochar or chemical fertilizer ( $P = 0.014$ ; Fig. 3A). Sole application of biochars from all waste sources did not increase the crop N uptake ( $P > 0.05$ ). Chemical fertilizer application increased the crop N uptake by 29% (66 vs. 51 kg ha<sup>-1</sup>) compared to control. Similarly,

combined application of biochar and chemical fertilizer also significantly increased the crop uptake from control (16–27%) as well as from their sole counterparts (9–25%). Apparent N recovery by wheat crop was significantly lower in biochar or biochar-CF treatments compared to CF treatment only ( $P < 0.001$ ; Fig. 3B). This parameter was ranged between 0.2 and 17% among all treatments. Apparent N recovery was observed 0.2–4% in biochar treatments while this parameter was between 2 and 7% when biochar was co-applied with chemical fertilizer. Although they were not significantly differed among each other ( $P > 0.05$ ; Fig. 3B).

PCA analysis of all treatments, soil chemical properties and wheat growth and yield parameters indicates that most of the variation in the data is explained by first two axis (92.3%; Fig. 4). Direction and size of the arrows reveal that a number of plant growth parameters were mainly associated with CF, KWB-CF and FMB-CF treatments. However, germination (%) was negatively correlated with aforementioned treatments. On the other hand, plant growth parameters such as growth yield, nitrogen uptake, biological yield, plant height and chlorophyll content were negatively related to WCB, PMB, Control, KWB and FMB treatments. Germination (%) was closely associated with KWB and FMB treatments (Fig. 4). Among the environmental variables, soil EC was correlated with Spikelet's per panicle, grain yield and N uptake. Soil mineral N was closely linked to chlorophyll content, leaf area

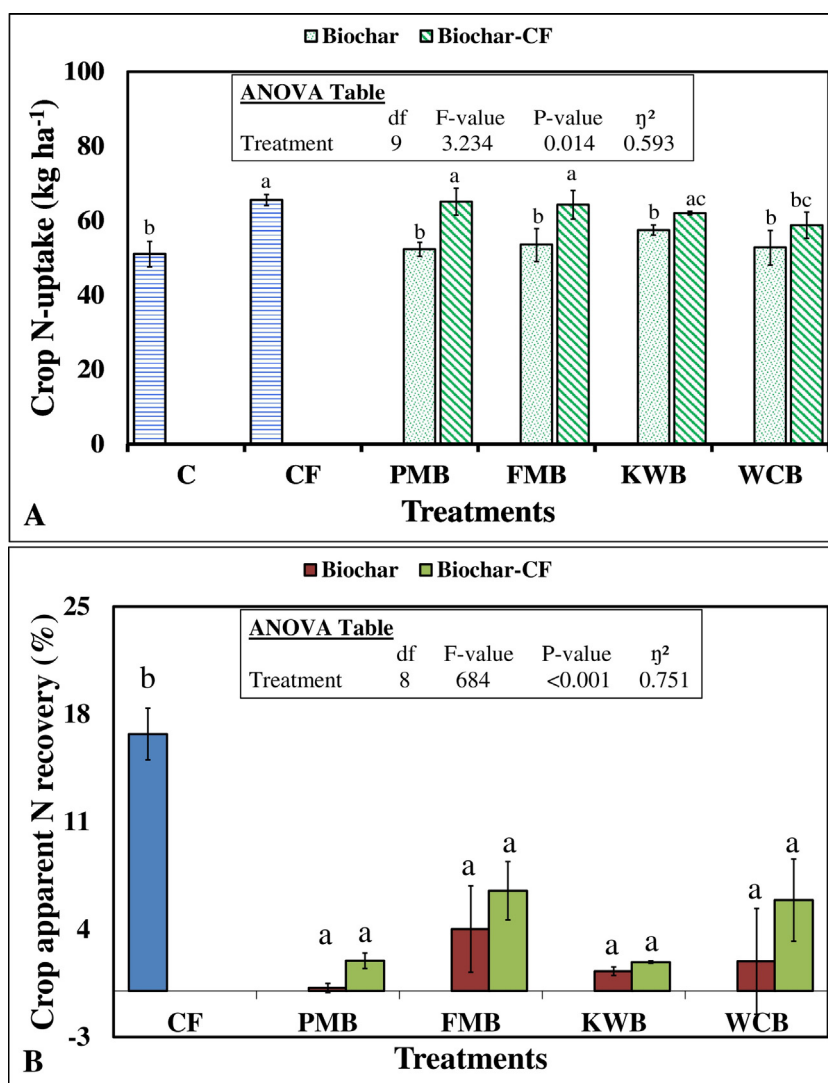
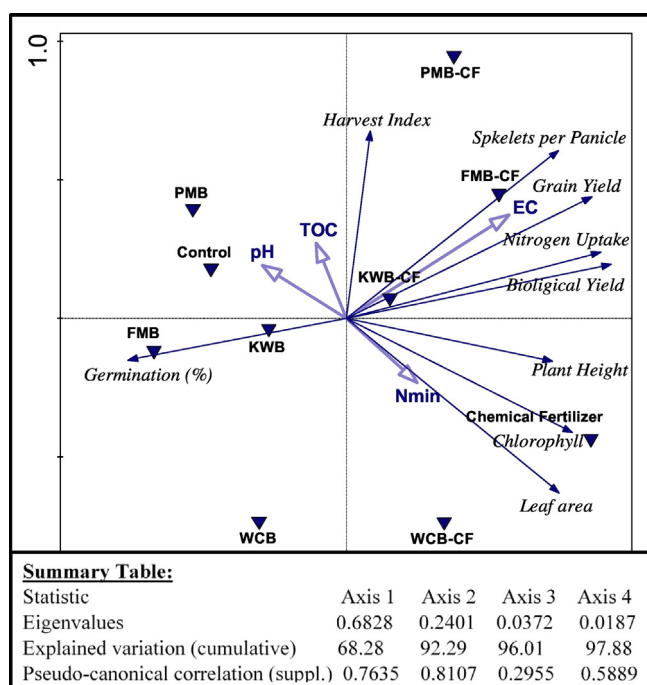


Fig. 3. Crop nitrogen uptake (A) and apparent nitrogen recovery from biochar and chemical fertilizer treatments. Values are mean of three replicates. Treatment abbreviation can be found at caption of Fig. 2. Small letters indicate significant difference among treatments at 5% probability level. Significance was tested by multiple comparison LSD test. Error bars signify the standard error of the mean ( $n = 3$ ).



**Fig. 4.** Principal component analysis of soil pH, electrical conductivity (EC), total carbon (TOC), mineral nitrogen ( $N_{min}$ ) and plant growth and yield parameters. Treatment abbreviation can be found at caption of Fig. 2. Variations in the data are mainly elucidated by first two components, PC1 (68.3%) and PC2 (24.0%) as specified in the inset statistics table, their individual score is unitless.

index and plant height while TOC and pH were negatively related with aforementioned parameters (Fig. 4).

#### 4. Discussion

According to our expectation, we found an increase in soil carbon content due to biochar application in our study that remained the same till day 83 of incubation, however at the end of experiment (after 128 days) soil carbon content was significantly decreased but it was higher than control or treatment with sole application of chemical fertilizer (Fig. 2A). The initial increase in carbon content in the soil are in line with Biederman and Harpole (2013), who found that much of increase in carbon content was mainly associated with recalcitrant nature of carbon in biochar. According to Hussain et al. (2016) carbon storage from biochar in the soil depends on its recalcitrant or labile nature. For instance, biochar produced at low temperatures partly pyrolyzed the feedstock and therefore results in more labile carbon that had less potential to store in the soil (Bruun et al., 2011). According to Yang et al. (2007), increase in pyrolysis temperature upsurges the decomposition of cellulose and hemicellulose feedstock during pyrolysis. Moreover, thermo-chemical reactions during pyrolysis of organic feedstock may be varied between liquefaction and pyrolysis at lower temperatures ranging between 250 and 600 °C (Demirbaş, 2000). That could explain fairly high volatile matter content in the fescue biochar produced at 500 °C (Keiluweit et al., 2010). Likewise, Bruun et al. (2011) found that biochar produced at 525 °C had ~50% lower labile compounds such as cellulose, hemicellulose and cellulose + hemicellulose derived carbon than biochar produced at 500 °C. Also in their study (Bruun et al., 2011), a strong positive correlation of hemicellulose and cellulosic carbon in biochar and  $CO_2$  evolution from biochar amended soil was observed after 115 days of incubation. Similarly El-Naggar et al. (2015) also found a decrease in soil carbon content with time after biochar application. They explained that presence of decomposable compounds in biochar stimulated the microbial activity which may slowly degraded

the labile carbon and resulting in more stable carbon over time (El-Naggar et al., 2015). Therefore, a decrease in soil carbon content in the soil of our study with short period of time (from 83 to 128 days) in all biochars or biochars integrated with chemical fertilizer might be attributed to presence of labile carbon in biochar since we produced biochar at 500 °C.

In our study, we could not find the difference in soil total carbon content after application of biochar produced from various feedstocks. In line with our findings, Streubel et al. (2011) found that biochar feedstock did not impact the increment in soil carbon. They concluded that variation in the quality of feedstock could not influence carbon level in the temperate soil. Rather they found a significant positive correlation between the amount of carbon added to the soil through biochar and increment in carbon content after biochar application to the soil. Accordingly, in our study, the amount of carbon applied through various biochars was also positively correlated with carbon content in the soil after 128 days ( $R^2 = 0.84$ ,  $P < 0.05$ ). Therefore, initial carbon content in biochar would play an important role in increasing carbon level in the soil. Consequently, in our study no significant difference in the initial carbon content of different biochars (Table 1) led to no difference in carbon content in the soil with biochar or integrated biochar and chemical fertilizer treatments at the end of experiment (Fig. 2A).

As expected, we observed a significance difference in biological or grain yield and reproductive organs (spikelets per panicle or panicle length) among *nutrient* or *structural* biochar treatments. Biological and grain yield was higher in kitchen waste and poultry manure biochars than other biochar treatments or control (Table 3). Jeffery et al. (2017) found in a very recent meta-analysis study that crop yield increased in tropical soil is mainly related to the nutrient status of the biochar applied. They divided the biochar into two sub-category called *nutrient biochar* produced from manure and *structure biochar* produced from plant-derived material such as wood. According to them *nutrient biochar* increased the crop yield by three time compared to structural biochar (Jeffery et al., 2017). These differences could be explained by the fact that structural biochar improves soil physio-chemical and biological properties such as structure, bulk density or biota than *nutrient biochar* which also provides nutrients for plant growth (Warnock et al., 2007). However, in our study, the wheat yield was lower in farmyard manure biochar than poultry manure or kitchen waste (Table 3). This could be ascribed to very low nutrient content in farmyard manure feedstock and biochar than kitchen waste and poultry manure biochar treatments (Table 1). Moreover, the difference in yield parameters might also be attributed to initial C:N ratio of biochars (Rajkovich et al., 2012; Tammgeorg et al., 2014a), that associates negatively with plants available nitrogen (Rajkovich et al., 2012). In our study the C:N ratio of poultry manure (17) and kitchen waste (20) biochars was very low compared to C:N ratio of farmyard manure or wood chips (93 and 231, respectively) biochars (Table 1). According to Sigua et al. (2014) such high C:N ratio biochar was subjected to low microbial degradation than the biochar with low C:N ratio therefore had lower nutrient mineralization rate (Sigua et al., 2014). However, the soil mineral N content in our study was not significantly differed over time among biochar treatments with contrasting C:N ratio showing that nitrogen availability might not be the factor influencing wheat yield (Fig. 2B, Table 1). Biochar also influenced soil EC or pH that could be the other factors influencing wheat yield (Sigua et al., 2016). For instance, biochar increased the nutrient ions (EC) in soil that might benefited the crop yield/production due to superfluous offtake of nutrients by crop from biochar (Sigua et al., 2016). This could also be one of the plausible reasons for the improvement of wheat yield of PMB and KWB treatments in our study since, soil EC was tended to be higher in these treatments after 37 days, whereas in case of PMB it remained higher throughout the experimental period (data not shown). This was also confirmed by the significant correlation between EC and biological or grain yield as indicated by PCA analysis in our study (Fig. 4).

**Table 2**  
Mean (n = 3, S.E ± 1) of the initial physico-chemical characteristics of the soil used in experiment.

Characteristics	Unit	Values
Bulk density	g cm <sup>-3</sup>	1.55 ± 0.15
Electrical conductivity	dSm <sup>-1</sup>	1.57 ± 0.24
pH-H <sub>2</sub> O extract	–	7.8 ± 0.06
Organic matter	%	0.47 ± 0.01
Total organic carbon	%	0.27 ± 0.001
Total nitrogen	kg ha <sup>-1</sup>	782.5 ± 10.25
Mineral nitrogen	kg ha <sup>-1</sup>	15.6 ± 1.21

According to our third hypothesis, we found a higher biological or grain yield (also reproductive organs), and nitrogen uptake in integrated application of biochars and chemical fertilizer treatments than sole biochars or control (Fig. 3A, Table 3). Albuquerque et al. (2013) found that biochars appeared to have good nutrient retention capacity. Therefore, they retained the mineral nutrients in the soil (Bass et al., 2016). In addition to this biochars also stimulated microbial activity and their combined application with chemical fertilizer would result in persistent nutrients release (Bass et al., 2016) that would result in reduction of nutrient losses through leaching or greenhouse gaseous emission (Bass et al., 2016; Gul and Whalen, 2016; Haider et al., 2017), thus resulting in improved crop yield and nitrogen uptake or efficiency of the applied chemical fertilizer (Gul and Whalen, 2016; Li et al., 2015). In our standard short-term pot experiment, chances of nutrient losses through leaching was not possible due to controlled irrigation as was also the case for Albuquerque et al. (2013), so the nutrient adsorption or retention capacity of biochar might limit the nitrogen losses especially in the treatments in combination with chemical fertilizer. Asai et al. (2009) and Rajkovich et al. (2012) found that biochar with low inherent nutrient counterbalanced the influence of chemical fertilizer in the soil through limiting the nitrogen availability to plants. In such scenario under field condition, these biochars can retain the nutrients in the soil and reduce their losses through leaching or N<sub>2</sub>O emission (Bass et al., 2016) from chemical fertilizer and thus enhance nutrient availability to crop for longer period of time.

Similar to wheat yield, crop nitrogen uptake in our study was significantly higher in treatment with combination of biochar and chemical fertilizer compared to control or lone biochar application (Fig. 3A).

This was also confirmed by PCA analysis where chemical fertilizers, or treatment with co-application of chemical fertilizers and biochars were closely associated with soil mineral N that was linked to biological, or grain yield (Fig. 4). Therefore, wheat biological yield and nitrogen uptake in combined treatments (biochar with chemical fertilizer) was not differed than sole chemical fertilizer treatment (Fig. 3A, Table 3). In contrast to our findings, Tammeorg et al. (2014b) did not find any effect of chemical fertilizer or biochar on spring wheat grain yield, they explained that less plant available nutrients from biochar and high soil inherent organic matter content would be the possible cause of non-increase in wheat productivity. In our study the inherent soil organic matter was very low and the biochar was produced at low temperature (Tables 1, 2) than in the study of Tammeorg et al. (2014b) that could be most plausible explanation of higher biological or grain yield in our study. Our findings are also in agreement with Li et al. (2015) who found that crop yield response in treatment with combined application of chemical fertilizer and biochar was similar to sole application of former fertilizer but higher than biochar alone. In our study, the crop apparent nitrogen recovery was very low in all treatments with sole biochars or co-application of biochar and chemical fertilizer compared to the treatment with chemical fertilizer alone (Fig. 3B). In accordance with our findings, Gul and Whalen (2016) calculated the crop nitrogen use efficiency from co-application of biochar and chemical fertilizer in many studies in the literature and found that this parameter was negative or close to zero in all studies despite of high increment in crop from this treatment. They did not explain the reason for such low nitrogen use efficiency nor we were able to elucidate this, therefore remained the necessary domain for future research.

According to our fourth hypothesis, the difference in chemical composition of different biochars resulted in much high variability in grain yield that was 32, 30, 9 and 8% higher in treatments with combined application of biochar and chemical fertilizer, PMB-CF, FMB-CF, KWB-CF and WCB-CF, respectively than control (Tables 1, 3). This difference in wheat yield could be attributed to nutrient biochar produced from animal waste with high nutritional value and cation exchange capacity compared to structural biochars produced from plant materials or their by product (Singh et al., 2010). Besides, higher carbon content in biochars also influenced their nitrate adsorption capacity (Kameyama et al., 2016), that resulted in lower N losses through leaching or gaseous emission (Gul and Whalen, 2016; Haider et al., 2017) and may improve the crop nutrient availability with time (Fageria and Baligar, 2005). Such

**Table 3**  
Mean (n = 3, S.E ± 1) growth and yield parameters of wheat crop as influenced by chemical fertilizer (CF) and biochars produced from poultry manure (PMB), farmyard manure (FMB), kitchen waste (KWB) and wood chip waste (WCB). Small letters indicates the significant difference among treatments at 5% probability level after LSD test.

Parameters	Unit	Control	CF	PMB	FMB	KWB	WCB	PMB-CF	FMB-CF	KWB-CF	WCB-CF	P-value
Germination	%	86.0 ± 4.0	75.0 ± 2.0	84.0 ± 2.0	84.3 ± 9.6	88.0 ± 3.5	82.7 ± 5.7	78.7 ± 1.7	71.0 ± 3.5	72.7 ± 7.7	80.3 ± 1.7	0.247
Chlorophyll index	SPAD	32.1 ± 0.6af	37.4 ± 1.1cd	28.9 ± 1.7a	29.5 ± 0.4a	29.8 ± 0.8af	33.8 ± 0.8acdf	33.2 ± 1.5ace	33.3 ± 1.0ac	34.5 ± 0.9cf	36.6 ± 1.0bdef	<0.001
Leaf area	cm <sup>2</sup>	17.1 ± 6.3a	28.7 ± 1.1b	16.3 ± 1.7a	17.3 ± 1.9a	19.4 ± 2.5a	22.1 ± 1.3ab	18.3 ± 2.8a	22.0 ± 1.9ab	19.8 ± 2.5a	26.9 ± 2.0b	0.058
Plant height	cm	67.7 ± 4.8	72.5 ± 4.9	69.9 ± 5.1	68.6 ± 3.1	68.0 ± 3.0	70.3 ± 4.0	70.2 ± 2.2	71.1 ± 3.4	72.9 ± 2.7	70.8 ± 2.8	0.989
Root biomass	kg ha <sup>-1</sup>	493.6 ± 38.4c	779.4 ± 10.3g	415.7 ± 4.9b	329.9 ± 38.6a	568.3 ± 9.7d	329.9 ± 38.6a	854.9 ± 5.6e	646.2 ± 9.1d	601.2 ± 10.5d	590.8 ± 17.3d	<0.001
Biological yield	kg ha <sup>-1</sup>	5699 ± 123ab	7679 ± 141f	5672 ± 117b	5378 ± 241ab	6100 ± 325c	5592 ± 176ab	7144 ± 167d	6956 ± 71d	6903 ± 0d	6448 ± 97c	<0.001
Grain yield	kg ha <sup>-1</sup>	2258 ± 63c	2866 ± 52e	2313 ± 26c	1980 ± 24a	2313 ± 106c	2116 ± 25b	2973 ± 19f	2924 ± 31e	2459 ± 14d	2445 ± 30cd	<0.001
Harvest index	%	39.6 ± 1.0bg	37.3 ± 0.7bh	40.8 ± 1.3cb	36.9 ± 1.3bh	38.3 ± 3.7bdh	37.9 ± 0.8beh	41.7 ± 0.9cdef	42.0 ± 0.1cdg	35.6 ± 0.2ah	37.9 ± 0.5bfh	0.060
100 grain weight	g	4.8 ± 0.1b	5.4 ± 0.04d	5.2 ± 0.02b	4.4 ± 0.01a	5.1 ± 0.03bc	4.7 ± 0.03b	5.3 ± 0.01b	5.6 ± 0.05e	5.2 ± 0.04bc	5.3 ± 0.03bc	<0.001
Grains	No. panicle <sup>-1</sup>	25.7 ± 1.7c	29.0 ± 1.7c	24.7 ± 0.3b	24.0 ± 0.6b	24.7 ± 0.3b	22.0 ± 0.6a	31.0 ± 0.6d	29.7 ± 1.5d	28.3 ± 0.3b	28.3 ± 2.9b	<0.001
Spikelets	No. panicle <sup>-1</sup>	13.7 ± 0.3bf	15.3 ± 0.3cg	13.3 ± 0.3be	12.7 ± 0.3aeh	13.0 ± 0.6aef	12.0 ± 0.6ae	17.3 ± 0.9dg	16.7 ± 0.3dg	14.0 ± 0.6bcfh	14.0 ± 0.9bcf	<0.001
Panicle length	cm	7.5 ± 0.02b	9.1 ± 0.06d	7.7 ± 0.02c	7.4 ± 0.04bf	7.3 ± 0.05af	7.3 ± 0.06af	9.5 ± 0.09e	9.3 ± 0.03g	8.8 ± 0.0h	7.9 ± 0.04i	<0.001



processes may occur in our study that retained the nutrients from the chemical fertilizer in case of its combined application with PMB or FMB, since total carbon content in these biochars was tended to be higher than KWB or WCB (Table 1) that may increase the crop yield in the former treatments. Similarly *structural* biochars such as KWB or WCB in our study can offset the response of chemical fertilizer on crop yield and may mend the soil physio-chemical properties (Albuquerque et al., 2013; Jeffery et al., 2017). Therefore, we observed lower grain yield in KWB or WCB but enhanced soil carbon content. Moreover, PCA analysis also revealed that FMB-CF and PMB-CF are closely associated with crop reproductive parameters and grain yield whereas WCB-CF are more linked to vegetative part (Fig. 4). Above all, despite of the differences in chemical composition of biochar feedstock (Singh et al., 2010), the wheat biological and grain yield was increased in all biochar treatments when applied in combination with chemical fertilizer. Therefore, we can propose that for the immediate benefits on crop yield and soil quality, co-application of biochar and chemical fertilizer would be the better option to avoid land degradation and food security issue.

## 5. Conclusion

Our results from the short-term standard pot experiment concluded that biochars from different organic waste sources resulted in the variation of wheat biological or grain yield. The application of *nutrient* biochars enhanced wheat biological yield than control but this parameter was less than chemical fertilizer. Likewise, co-application of biochars and chemical fertilizer resulted in higher grain yield, nitrogen uptake and soil carbon content than control or from the sole application of biochar. Moreover, in co-application treatments, the *nutrient* biochars have more positive effect on crop productivity than *structural* biochar but the increment in soil carbon content was not differed between these biochars. Therefore, to explore whether the differences in crop productivity would remained among these biochars for longer period, it would be fascinating to study the influence of both *nutrient* and *structure* biochars on crop productivity and soil properties in more than one growing season. Although sole application of chemical fertilizer increased the wheat yield or nitrogen uptake but it significantly reduced soil carbon content and increased the electrical conductivity. Therefore, we propose that co-application of *nutrient* biochars with chemical fertilizer in intensively managed soil would be the most appropriate strategy to achieve short-term benefits in terms of higher wheat grain yield with improving soil quality in infertile cultivated soils.

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