



# Response of soil microbial biomass and enzymatic activity to biochar amendment in the organic carbon deficient arid soil: a 2-year field study

Muhammad Irfan<sup>1</sup> · Qaiser Hussain<sup>1</sup> · Khalid Saifullah Khan<sup>1</sup> · Muhammad Akmal<sup>1</sup> · Shahzada Sohail Ijaz<sup>1</sup> · Rifat Hayat<sup>1</sup> · Azeem Khalid<sup>2</sup> · Muhammad Azeem<sup>3</sup> · Muhammad Rashid<sup>1</sup>

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## Abstract

The application of pyrolyzed organic carbon (C) to soils has been assessed worldwide to play a vital role in improving the physical-chemical characteristics of the soil. However, the effects of co-use of biochar and nitrogen (N) fertilizer on soil biological process in an arid region are not well understood. For this, a 2-year field experiment was conducted in an arid region to assess the co-use of biochar and nitrogen (N) fertilizer on soil microbial biomass and enzyme activity in the rhizosphere of the wheat crop. Sugarcane bagasse was used as biochar feedstock and applied with three levels of biochar (0, 0.5, and 1% C ha<sup>-1</sup>) on carbon equivalent basis in the presence and absence of N fertilization (46 kg N ha<sup>-1</sup>). Biochar was incorporated in the soil before sowing of wheat, and the soil samples were taken from each treatment at crop maturity. Findings of the study indicated that biochar amendments enhance the soil organic carbon, DOC, inorganic N, and soil moisture contents, while reducing the bulk density and salinity of soil in both wheat growing season. Microbial biomass carbon and nitrogen increased by 18% and 63% with biochar amended at 1% C ha<sup>-1</sup> with nitrogenous fertilizer and the same trend was observed in the following year. Urease and dehydrogenase activities also significantly increased with biochar applied at 1% C ha<sup>-1</sup> with N fertilization illustrating 15% and 19%, respectively. During the second year of wheat trial, the enzymatic activity also boosted up as the first year. The results revealed that sugarcane bagasse-derived biochar addition can be utilized in improving the soil health, nutrient status, and soil biological functions in the calcareous soil of the arid region.

**Keywords** Biochar · Microbial biomass · Enzyme activity · Wheat · Rainfed

## Introduction

The incorporation of carbon-based organic material to soil assist to sustain the soil organic carbon levels, which in turn typically improve the moisture retention, nutrient status, aeration, nutrient supply, and biological functioning in soil

(Girmay et al. 2008). Addition of various organic residues such as crop residues, green manures, industrial wastes, leaves of different trees, animal wastes, vegetable trashes, and household wastes has been tested to sustain soil quality (Ali et al. 2011; Quilty and Cattle 2011). However, the application of pyrolyzed organic carbon to soils has been gained considerable interest worldwide due to its potential to improve soil nutrient retention capacity (through the sorption/adsorption or stabilization of nutrient ions), maintain pH in acidic as well as alkaline soils, improve water holding capacity, and to sequester carbon from decades to thousands of years (Downie et al. 2009; Spokas et al. 2012).

Biochar amendments to soil play a vital role in improving the physicochemical characteristics (Lehmann et al. 2011) with induced variability in the soil microbial biomass and enzyme activity (O'Neill et al. 2009; Khodadad et al. 2011). Biochar addition has been shown to enhance the microbial biomass and enzyme activity with organic carbon availability as biochar contains a significant portion of dissolved organic

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✉ Qaiser Hussain  
qaiseruaf@gmail.com; qaiser.hussain@uaar.edu.pk

<sup>1</sup> Institute of Soil Science, Pir Mehr Ali Shah Arid Agriculture University, Rawalpindi 46300, Pakistan

<sup>2</sup> Department of Environmental Sciences, Pir Mehr Ali Shah Arid Agriculture University, Rawalpindi 46300, Pakistan

<sup>3</sup> Department of Agriculture, Hazara University, Mansehra, KPK 21300, Pakistan

carbon (Bruun et al. 2011; Zimmerman et al. 2011; Luo et al. 2013). Incorporation of biochar into the soil may have several direct or indirect impacts on soil biota due to the change in several abiotic factors including soil pH or altered quality of substrate as a source of energy (Thies et al. 2015). The alteration in microbial biomass results from the additional supply of nutrients from the labile carbon of biochar, adding the fact that biochar improves microbial living conditions and affords microbes protection from grazers or competitors in biochar pores (Lehmann et al. 2011). Several studies reported that biochar additions may induce detoxification of compounds (allelochemicals) and sorption of various signaling molecules, and alteration in soil physicochemical properties may lead to enhance microbial biomass. However, some research studies revealed no prominent influence of biochar on soil microbial biomass (Kuzyakov et al. 2009; Castaldi et al. 2011). Dempster et al. (2012a) determined that the addition of biochar deprived the soil microbial biomass due to the toxicity effect which depends on the types of feedstock and pyrolysis temperature.

In soil, extracellular enzymes of microorganisms/microbes accomplish organic matter disintegration and nutrient cycling (Burns et al. 2013). Thus, the soil enzyme functions and their responses towards the biochar application have attracted a considerable attention in soil fertility and nutrient status. It has been testified that biochar might generally give a suitable environment for the activities of a number of enzymes related to N and phosphorous (P) utilization (Bailey et al. 2011) and also minimize the activities of enzymes involved in C cycle (Lehmann et al. 2011). The activity of soil enzymes related to C, N, and P cycles which are boosted up by the degradation of the organic matter is strictly correlated with soil physicochemical properties (Kussainova et al. 2013), populations structure, and abundance of soil microorganisms (Nielsen et al. 2014), vegetation (McCormack et al. 2013), or with the occurrence of various anthropogenic factors (Lehmann and Joseph 2015a, b). Moreover, biochar amendment rates and texture of soil also influenced the response of soil microbial biomass activity and decomposition (Lehmann et al. 2011). The variation in microbial biomass response towards biochar additions may include the following: enhanced nutrient availability (DOC, N, P, and K), adsorption of lethal compounds, and a significant effect on soil-water content and pH range; all of these factors affect the soil microbe activity in soil (Lehmann et al. 2011). The key factor of all biochars is its internal porosity which varied with kind of feedstock and temperature of pyrolysis that effects the efficiency of soil microbes (Pietikäinen et al. 2000), C substrates, and other mineral nutrient (N, P, etc.) turnovers in soil (Saito and Marumoto 2002; Warnock et al. 2007).

Enzyme activity is considered a sensitive indicator of soil health. The effect of biochar on soil enzymes is a key to understanding the short and long-term impacts on microbial nutrient turnover. (Khadem and Raiesi 2017; Gul et al. 2015). The key role of microbial biomass and enzyme activity in soil nutrient

cycling with co-use of biochar and N fertilizer on the activity of soil enzymes remains largely unclear. In order to sustain long-term efficiency and crop productivity of the arid land cropping system, an efficient and proper natural resource management needs to be determined. The main objectives of the following study were to examine the biochar impact in the presence and absence of nitrogenous fertilization (N) on soil microbial biomass activity and functions in the wheat crop rhizosphere.

## Materials and methods

### Site description

The present study was performed at the University Research Farm of Pir Mehr Ali Shah-Arid Agriculture University, Rawalpindi, 46300, Pakistan (73°30' E to 73°45' E, 33°1' N to 36°6' N). The average soil organic carbon is less than 1% in arid regions. The texture of the soil is sandy loam; pH of the soil is neutral to alkaline in nature with varying moisture contents that depend on precipitation intensity. The weather is arid to semi-arid, sub-tropical continental to subhumid and has two rain showers occurrence with a maximum in winter-spring days and at the end of summer of the whole year. Rainfall in an arid region is erratic; nearly about 60–70% of the precipitation commonly occurs during the monsoon season (mid-June to mid-September) (Shafiq et al. 2005).

### Biochar production

Sugarcane bagasse biochar was prepared by pyrolysis (partial or no oxygen supply) in the two-barrel conventional pyrolysis chamber at 400 °C (Gunther 2009). Before pyrolysis, sugarcane bagasse was initially air-dried for 2–3 days depending on moisture contents. For the research trial application, the black carbon (biochar) was crushed to pass through a 2-mm sieve (Pan et al. 2011).

### Experimental site

A 2-year field trial was performed with biochar application (on C equivalent basis) with and without N fertilizer (urea) in organic carbon deficient soil under rainfed and arid conditions. The experiment was conducted with the application of different treatments including 0% biochar-C ha<sup>-1</sup> (B0N0); 0.5% biochar-C ha<sup>-1</sup> (B1N0); 1% biochar-C ha<sup>-1</sup> (B2N0); 0% biochar-C ha<sup>-1</sup> plus N (B0N1); 0.5% biochar-C ha<sup>-1</sup> plus N (B1N1); 1% biochar-C ha<sup>-1</sup> plus N (B2N1). The nitrogenous fertilizer was applied @ 46 kg N ha<sup>-1</sup> at sowing of wheat. The recommended rate of phosphorus and potassium fertilizers was applied as basal fertilizers. Biochar doses were applied to plots having dimensions (4.5 m × 1.5 m) following randomized complete block design (RCBD) according to the

experiment plan. Biochar was thoroughly mixed in soil with a hard rake and then plowed to a 12-cm deep 2 weeks before sowing. Each biochar treatment was placed in triplicate plots, and individual plots were separated to each other with a 0.5-m width by border rows.

### Soil sampling

After harvesting the wheat crop, composite soil samples from each treatment plot were collected, preserved in polythene bags, and shipped to the lab within not more than 3 h after the collection. For physicochemical analysis, some portion of the soil samples was air-dried and passed through a sieve (< 2 mm) while the remaining portion of the soil samples was kept in the freezer at 4 °C for microbial biomass and enzyme activity analysis.

### Biochar and soil characteristics

The moisture content of soil and biochar was determined gravimetrically (Gardner et al. 1991). The electrical conductivity (EC) in a saturated paste extract of soil was measured by an electrical conductivity meter (Rhoades 1996), and soil acidity and alkalinity were examined by the ratio of 1:1 soil-water suspension (Thomas 1996). Biochar characteristics like EC and pH were recorded in a 1:10 (*w:v*) biochar-water mixture (Cayuela et al. 2013). Organic carbon content (OC) of sugarcane bagasse biomass (biochar) was determined by burning the biochar samples into ashes in the muffle furnace at high-temperature range (400–500 °C) for 4 h and OC was calculated by applying the formula defined by Brake (1992). Soil organic carbon (SOC) was determined by the using wet digestion procedure involving the use of 1 N potassium dichromate ( $K_2Cr_2O_7$ ) solution and concentrated sulfuric acid ( $H_2SO_4$ ) (Nelson and Sommers 1982). The Kjeldahl method was used for the determination of total nitrogen (TN) involving wet digestion with concentrated sulfuric acid ( $H_2SO_4$ ) and distillation with boric acid and NaOH. During distillation, the titration of the digested mixture was done with 0.01 N  $H_2SO_4$  till pink color was developed (Van Schouwenberg and Walinge 1973).

### Microbial biomass

Microbial biomass carbon (MBC) was estimated by the commonly used technique, i.e., the fumigation-extraction method. Ten grams of soil was fumigated for 24 h at 25 °C with pure ethanol-free chloroform ( $CHCl_3$ ). The samples were then added with 50 mL 0.5 M potassium sulfate ( $K_2SO_4$ ) for 1/2 h on a horizontal shaker at 200 rev per minute. The suspensions were then filtered by a filter paper (Whatman No. 42). Similarly, 10 g of soil was extracted for non-fumigation at the same time (Brookes et al. 1985). SOC in the extracts was measured by the titration technique. Then MBC was determined as microbial biomass  $C = (C \text{ fumigated} - C \text{ non-fumigated}) \times 2.64$ .

Microbial biomass nitrogen (MBN) was also examined by the same method as used for MBC, the fumigation-extraction technique. For determining the total N, fumigated and non-fumigated soil samples were extracted with potassium sulfate ( $K_2SO_4$ ) and the filtered extract was measured for total N by using the Kjeldahl digestion procedure. For digestion, 01 g of soil was digested with a digestion mixture ( $FeSO_4$  10:  $CuSO_4$  1: Se 0.1) and 4.5 mL of concentrated sulfuric acid ( $H_2SO_4$ ) in each digestion tube for 3 h. After digestion, the mixtures were carried out for distillation by pouring the samples into the steam distillation chamber of Kjeldahl with 10 M NaOH and 2%  $H_3BO_3$ . After completion of distillation, 40 mL of samples was taken from the distillation chamber and at the end, titrated against 50 mM  $H_2SO_4$  for the endpoint, i.e., bluish red color (Wu et al. 1990). The soil MBN was calculated by a formula as microbial biomass  $N = (N \text{ fumigated} - N \text{ non-fumigated}) \times 1.46$ .

### Soil enzymes analysis

Soil dehydrogenase activity (DE) was determined by measuring the concentration of triphenyl formazan (TPF). After filtering the samples through a filter paper (Whatmann-42), the optical density of soil filtrate was measured at 546 nm on a spectrophotometer. The activity of the enzyme (TPF  $\mu g g^{-1}$  dwt soil) was noted as  $TPF (\mu g mL^{-1}) \times 45/dwt/5$  (Alef 1995). Urease enzyme activity (UA) was measured by 50 mL potassium chloride (KCl) solution to collect the soil extract. Soil extract was passed through a filter paper. After the filtration of the extract is done, then ammonium content in the filtrate was calculated by 690 nm optical density and at the end, reading was measured to find the urease activity (Kandeler and Gerber 1988).

### Statistical analysis

Effects of several treatments (biochar, fertilization, and their interaction) on different physicochemical properties, microbial biomasses, and enzymatic activities in soil were studied by analysis of variance (two-way) by using statistical software Statistics 8.1. The significance of the main differences was verified by using LSD (least significance difference) test at  $p < 0.05$  level (Steel and Torrie 1997).

## Results

### Physicochemical properties

Biochar application significantly affects the soil physical and chemical properties with and without fertilizer application (Table 1). In treatments without N fertilization (B2N0), SOC, TN, and soil moisture contents improved by 23%, 27%, and 24% under 1% biochar-C  $ha^{-1}$  (B2N0) amendment and by 9%, 13%, and 10% under 0.5% biochar-C (B1N0) as

**Table 1** The physical-chemical features of soil and biochar (influenced by different treatments of biochar with and without N fertilizer incorporation)

Treatments	Without N fertilizer				With N fertilizer		
	Biochar	B0N0	B1N0	B2N0	B0N1	B1N1	B2N1
pH	7.95	8.15a	8.09a	8.22a	8.18a	8.23a	8.24a
EC (dS m <sup>-1</sup> )	0.31	0.53a	0.52a	0.55a	0.56a	0.54a	0.55a
SOC (g kg <sup>-1</sup> )	497	6.07b	6.59ab	7.47a	6.20b	6.54ab	7.27a
DOC (g kg <sup>-1</sup> )	0.44	0.41e	0.47c	0.45d	0.48ab	0.49ab	0.52a
TN (%)	1.51	3.03c	3.44bc	3.86a	3.70ab	3.79ab	3.77ab
Bulk density (g cm <sup>-3</sup> )	–	1.44a	1.41ab	1.39bc	1.38bc	1.38c	1.37c
Soil moisture contents (%)	–	9.66b	10.67ab	12.00ab	10.55b	12.33ab	14.00a

Biochar amendment with the rate of 0, 0.5% C, and 1% C ha<sup>-1</sup> (B0, B1, and B2, respectively) with (N1) and without N Fertilizer addition (N0). Lettering in a column shows a statistical difference between the treatments at  $P < 0.05$ . EC, electrical conductivity; DOC, dissolved organic carbon; SOC, soil organic carbon; TN, total nitrogen

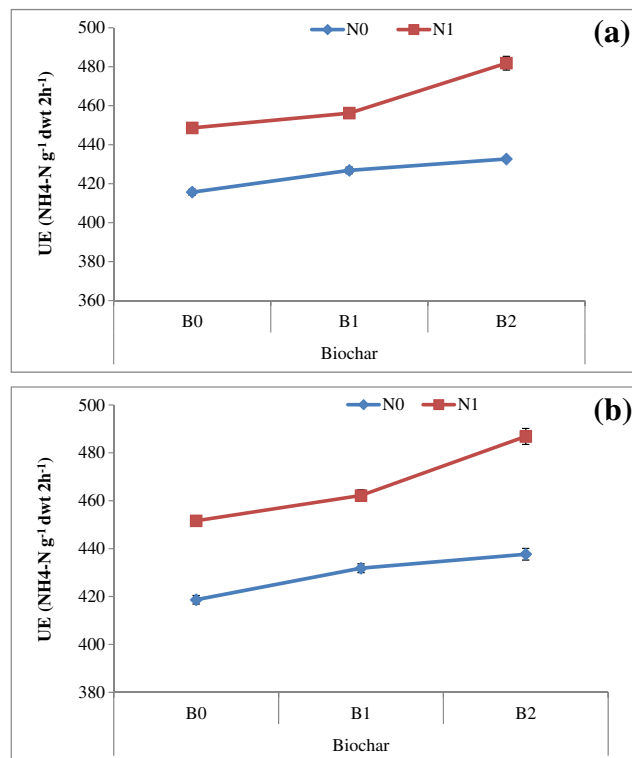
compared to B0N0 (no biochar and fertilizer), respectively. However, treatments with nitrogen fertilizer (B2N1) significantly enhanced the SOC, TN, and soil moisture by 19%, 24%, and 44% and promoted 8%, 25%, and 15% with (B1N1) as compared to B0N1, respectively. In treatments without N, DOC was significantly enhanced by 9% under biochar amendment at 0.5% C (B2N0) as compared to control (B0N0). While in treatment with N, DOC was boosted up to 26% under biochar addition at 1% C (B2N1). However, biochar addition had no prominent result on soil pH and only a minimal effect on EC was observed.

### Soil microbial activity

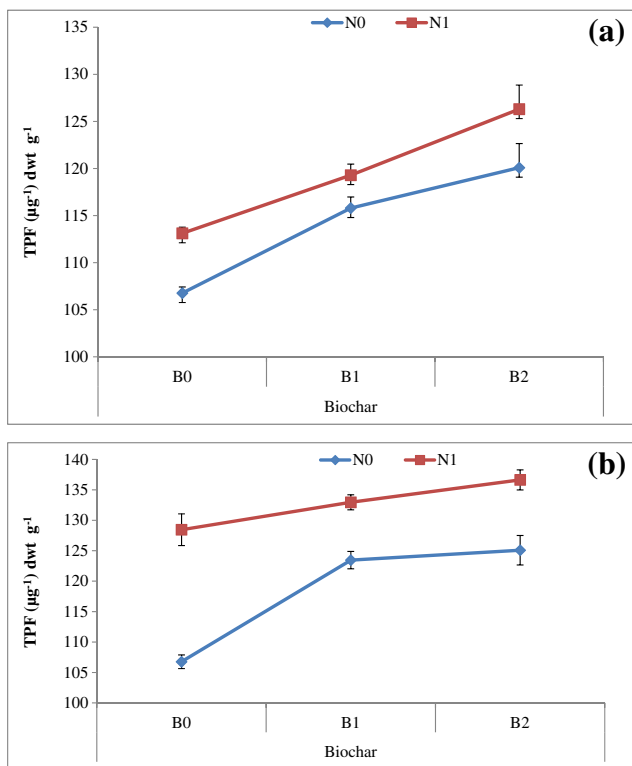
Biochar plays a key role in soil microbial activities to improve the fertility status of soil for long-term agricultural productivity. As given results showed, the interaction of nitrogenous fertilization and biochar had significantly promoted the urease activity in the wheat field during the first year (Fig. 1a). The highest urease enzyme activity (431  $\mu\text{g NH}_4\text{-N g}^{-1} \text{dwt } 2 \text{ h}^{-1}$ ) was detected in B2N0 showing 5% elevate, followed by B1N0 (409  $\mu\text{g NH}_4\text{-N g}^{-1} \text{dwt } 2 \text{ h}^{-1}$ ), showing a 3% increase in the absence of N fertilization (B0N0). In the case of biochar combination with N fertilization, the highest urease activity (490  $\mu\text{g NH}_4\text{-N g}^{-1} \text{dwt } 2 \text{ h}^{-1}$ ) was noticed in B2N1 showing a 15% boosted up afterward and in B1N1 (454  $\mu\text{g NH}_4\text{-N g}^{-1} \text{dwt } 2 \text{ h}^{-1}$ ) showing 9% raise with the combination of nitrogenous fertilizer application respectively, as compared to B0N1. During the second-year field trial, urease enzyme activity showed the same trend as the first year (Fig. 1b). Urease enzymes activity showed significant result during the second-year trial. Biochar-amended soil with chemical fertilizer (urea) has maximum urease enzyme activity as the result noted during 2013.

Dehydrogenase enzyme activity was studied through biochar amendment in the soil in the presence and absence of nitrogen-based fertilizer. Results showed that biochar addition

positively increased the DE activity in the wheat crop field. Biochar and nitrogen fertilizer alone gave a positive response towards the DE activity but the interaction of both N fertilization and biochar-amended soil had a significant impact on DE activity (Fig. 2a). Nevertheless, the greatest DE activity (126 mg TPF kg<sup>-1</sup> 24 h<sup>-1</sup>) was indicated in B2N1 proving an 18% enhancement, followed by B1N1 (119 mg TPF kg<sup>-1</sup> 24 h<sup>-1</sup>) designating a 12% increase by N fertilizer amendment,



**Fig. 1** Effect of biochar on urease (UE) ( $\mu\text{g NH}_4\text{-N g}^{-1} \text{dwt } 2 \text{ h}^{-1}$ ) activity in wheat soil during the 2-year field trial; **a** 2013, **b** 2014: Biochar applied at 0, 0.5% biochar C, and 1% biochar C ha<sup>-1</sup> (B0, B1, and B2) respectively, in the presence (N1) and absence of N fertilization (N0)

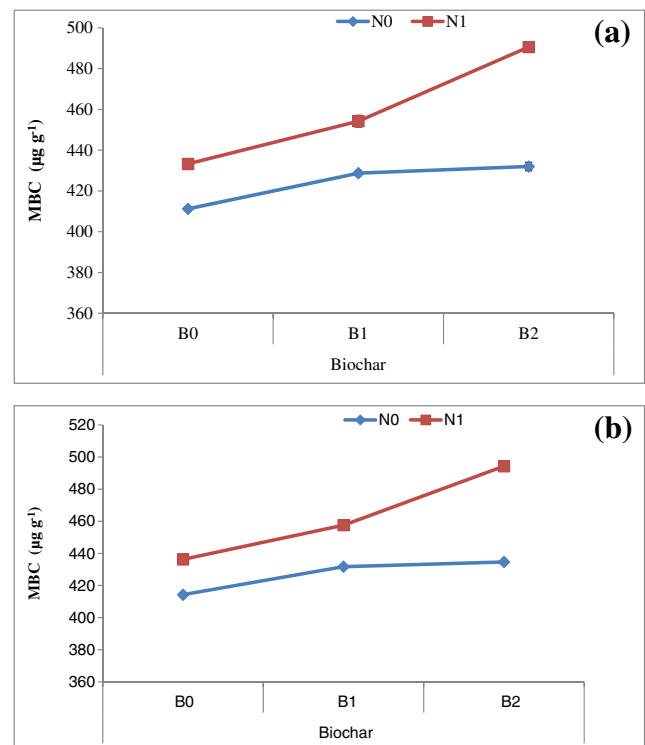


**Fig. 2** Effect of biochar on dehydrogenase ( $\text{mg TPF kg}^{-1} 24 \text{ h}^{-1}$ ) activity in the 2-year wheat field trial; **a** 2013, **b** 2014: Biochar amendment at 0, 0.5% biochar-C, and 1% biochar-C  $\text{ha}^{-1}$  (B0, B1, and B2) respectively, with (N1) and without N fertilization (N0)

as compared to B0N0 (no fertilizer and biochar). According to the first wheat trial, the same trend was observed in DE activity in Fig. 2b. Dehydrogenase enzyme activity significantly increases with the time period but not significantly increased as the first-year result. The maximum DE activity was obtained in B2N1 where a high dose of biochar with chemical fertilizer was applied like the first trial in an arid climate region.

### Soil microbial biomass

The interaction of biochar and nitrogen fertilizer amendment revealed the significant impact on soil microbial biomass carbon (MBC) (Fig. 3a). The highest MBC ( $490 \text{ mg kg}^{-1} \text{ soil}$ ) was obtained in B2N1 representing a 19% enhancement, followed by B1N1 ( $454 \text{ mg kg}^{-1} \text{ soil}$ ) showing a 10% increase with N fertilization, as related to B0N0. Likewise, the highest MBC ( $431 \text{ mg kg}^{-1} \text{ soil}$ ) was noted in B2N0 indicating a 5% increase followed by B1N0 ( $428 \text{ mg kg}^{-1} \text{ soil}$ ) with a 4% increase in the MBC without N fertilizer-amended soil as compared to B0N0 (no biochar and fertilizer). Figure 3b represents the microbial biomass carbon activity during the second-year field trial that evaluates the positive impact of biochar addition with nitrogenous fertilizer. The same trend was seen in Fig. 3b as the 2013 wheat trial significantly enhanced the MBC in biochar-



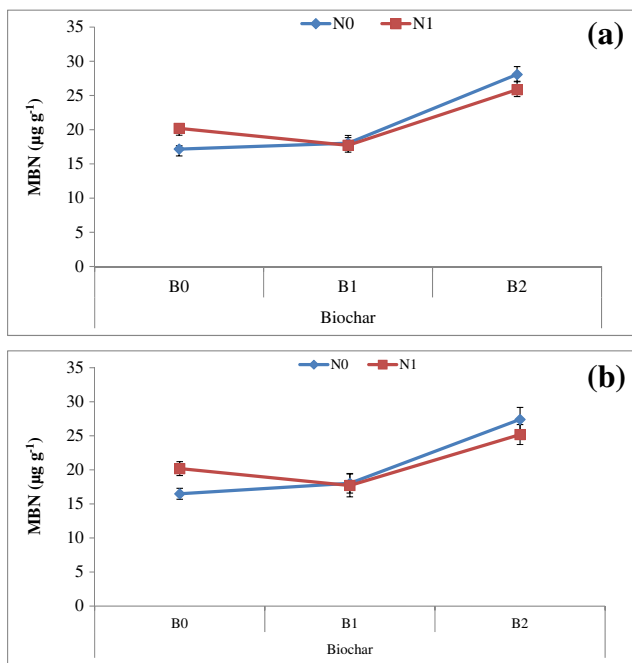
**Fig. 3** Effect of biochar on MBC ( $\mu\text{g g}^{-1}$ ) in the 2-year field trial of wheat; **a** 2013, **b** 2014: Biochar amendment at 0%, 0.5% C, and 1% C  $\text{ha}^{-1}$  (B0, B1, and B2) respectively, with (N1) and without N fertilization (N0)

amended soil. Interactive effects of biochar with N fertilization have a beneficial effect on MBC during both field trials.

The same trend was observed in MBN like MBC in wheat crop, the interactive impact of nitrogen-based fertilizer with biochar also had a prominent effect on the MBN (Fig. 4a). The maximum MBN ( $28.08 \text{ mg kg}^{-1} \text{ soil}$ ) was noticed in B2N0 demonstrating a 63% increase, followed by B1N0 ( $18.03 \text{ mg kg}^{-1} \text{ soil}$ ) elucidating a 6% increase without N fertilizer as compared to B0N0. Correspondingly, the high value of MBN ( $25.85 \text{ mg kg}^{-1} \text{ soil}$ ) was noted in B2N1 showing a 50% increase whereas, B1N1 ( $17.71 \text{ mg kg}^{-1} \text{ soil}$ ) caused a 4% reduction in MBN with N fertilization, as compared to B0N1. Microbial biomass nitrogen showed a positive response in both wheat trials. Figure 4b indicated that MBN showed significant effect with biochar and urea fertilization in the following year. Both figures (a) and (b) showed the similar behavior of MBN by the incorporation of black carbon (Biochar) with the combination of N fertilizer in the soil.

### Discussion

Addition of black carbon alone and with chemical fertilizer has been observed to change soil physical and biochemical properties that indirectly improves the nutrient status (Asai



**Fig. 4** Biochar impact on MBN ( $\mu\text{g g}^{-1}$ ) in the 2-year wheat field trial; **a** 2013, **b** 2014: Biochar amendment at 0%, 0.5% C, and 1% C  $\text{ha}^{-1}$  (B0, B1, and B2) respectively, with (N1) and without N fertilization (N0)

et al. 2009; Major et al. 2010). These variations affect the soil structures (Rillig and Mummey 2006) and nutrient cycling (Steiner et al. 2008) that indirectly affects the plant development and productivity (Warnock et al. 2007). Biochar comes from a variety of feedstocks that have variable and progressive effects on types of soils and climates (Gaskin et al. 2010; Zwieter et al. 2010; Haefele et al. 2011). The results of our study are in line with various studies, which revealed that biochar addition significantly improves the soil physicochemical properties (Table 1) (Asai et al. 2009; Major et al. 2010). All literature studied have the described results alike to our trial findings; highlighting the high recalcitrance of biochar carbon, very little quantity of solubilizing or labile organic compounds is incorporated in soils of different soil textures, but effectively increased total organic C and N in soil with minimum degradation; thus, most authors suggested the use of biochar in soils as an efficient tool for analyzing the soil C sequestration to mitigate the climatic effect (Glaser et al. 2002; El-Mahrouky et al. 2015; Lehmann et al. 2006; Mackie et al. 2015; Marchetti et al. 2012; Zavalloni et al. 2011).

Biochar amendment in soils improved the porosity, nutrient holding capacity, and water holding capacity as well as reduced the hardness of soil (Ogawa and Okimori 2010), which endorsed root development and increased the capacity of soil that was exploited by plant roots indirectly giving significant effect on the nutrient status of the soil. Findings of the study are similar to Genesisio et al. (2012) results, According to Genesisio et al. (2012), biochar addition significantly improves the physical properties and nutrient status of the soil. Other

experiments (pot, incubation, and field) have also informed that charcoal has partial or no effect on soil microbial biomass and activities (Castaldi et al. 2011) because of highly stable carbon of biochar (Kuzyakov et al. 2009). But a major factor to control the microbial activity depends on biochar incorporation quantity and texture of soil that may influence the response of soil microbial biomasses (Lehmann et al. 2011). Explanations for soil microbe activities alter their response by the biochar application that includes improving the soil nutrient availability (DOC, P, N, and K) and also showed positive response to soil characteristics of water contents, Ec, and pH status of soil; all of these factors indirectly affect the activity of soil microorganisms indirectly involved microbial biomass for soil health (Lehmann et al. 2011).

Soil microbial activities are affected by a variety of organic waste to show prominent effect in soil nutrient cycling like microbial biomass carbon and nitrogen (MBC and MBN) and soil enzyme activities (carbon and nitrogen-based) that are related to soil fertility and agricultural productivity (Wang et al. 2009 and Dempster et al. 2012a, b). The meta-analysis of Zhou et al.'s (2017) experiment showed that BC mixing to soil enhanced the activity of MBC (26%) and MBN (21%), respectively; same results were recorded by Zhou et al. (2017) that biochar addition showed a significant result with chemical fertilizer. Remarkably, in different climatic conditions like a field, pot, and the laboratory, incubation experiments showed that biochar addition could significantly improve the MBC contents in soil. Similar to MBC, MBN in the soil also show same behavior that significantly showed a positive response and in case of pot or field studies as MBC, improves by the addition of biochar but did not differ significantly from controls. Whereas, it was concluded that the divergent variation in microbial biomass nitrogen among the pot and field trials could be attributed to N competition by crop requirement (Lehmann et al. 2003).

Soil microbes are responsible for decomposition of carbon compounds in soil by various enzymes (C and N base) that also control the degree of SOM breakdown and recycling of nutrients in the poor soil (Nannipieri et al. 2012). To understand the black carbon material's impact on the soil enzyme activities is a research priority because soil enzyme activities involved a lot of physicochemical and biological processes in soil. Furthermore, some other studies also testified that BC application to soil usually increases the soil enzyme activities related to N and P cycling and decreases the soil enzyme activities involved in C sequestration and cycling (Bailey et al. 2011 and Ameloot et al. 2013). Our results are also similar with Bailey et al. (2011) and Ameloot et al.'s (2013) findings that showed how to enhance the activity of enzymes (N and C base enzymes) by biochar amendment. On the other hand, various studies have reported inconsistent findings with biochar application rates and pyrolysis temperature. Lammirato et al. (2011)

and Paz-Ferreiro et al. (2014) proposed that BC has variable effects on different soils and enzymes. Our research results indicated that enzyme activity improved with the range of biochar amendment. Our outcomes were similar to the agreement of Bailey et al. (2011) and Ameloot et al. (2013), suggesting that biochar volatile compounds at low pyrolysis temperatures ranging from 350 to 500 °C speed up enzyme activity in a sandy loam soil, including carbon and dehydrogenase based enzymes. The maximum activities of enzymes observed in the study may be due to physicochemical characteristic interactions of the BC with extracellular soil enzymes, which thereby could enhance their activity (Lehmann et al. 2011 and Elzobair et al. 2015).

Biochar addition increases the contributions of enzymes which involved N cycling; it was considered that a variety of microbes promoted N mineralization from the soil to compensate the high C/N ratios in soil (Bailey et al. 2011 and Tian et al. 2016). Generally, these results of the above study proposed that biochar effects on soil enzymatic activities mainly depend on pyrolysis temperature, soil texture, types as well as feedstock of biochar, and interactions of substrates and soil enzymes with BC (Lammirato et al. 2011). Our results are similar to Zhou et al. (2017); biochar application could improve the soil health and quality, considered as a major factor to show significant effect on various soil parameters like MBN, MBC, TN, SOC, and also available K and P (Zhou et al. 2017, and Mastro et al. 2013) which is also similar to following experiment results. These positive changes that could be accredited to biochar have available C, N, and P and gradually release these important nutrients into the soil to improve the soil status (Ouyang et al. 2014). Results of this study clearly indicated that biochar application enhances the soil nutrient availability.

In relation to microbial activity, the addition of biochar is significantly affected in arid climate that depends on biochar physicochemical characteristics. The key factor of microbial activity is porosity of biochar material. The porosity of biochars of various feedstocks may encourage microbe activity to support the nutrient cycling and soil quality; more porous material provide the suitable environment for microorganisms (Pietikäinen et al. 2000) and accumulate C substrates as well as inorganic nutrients to improve the soil health (Saito and Marumoto 2002; Warnock et al. 2007). In the same way, the activity of the enzyme increased when biochar was added to soil then microbial activity increased (Fig. 3). The dehydrogenase enzyme activity is the indication of the positive priming effect of biochar for microbial activity and nutrient cycling. The results also proposed that biochar contains more labile substrates which enhance the activity of soil microbes (Guenet et al. 2010). Our research indicates that sugarcane bagasse biochar had a prominent impact on soil microbial biomass and activity in the wheat crop field in arid regions of Pakistan.

## Conclusion

Biochar application to the cereal crop in the arid area significantly elevates the soil microbial biomasses (MBC and MBN) and enzyme activity. Soil enzyme (urease and dehydrogenase) activities were significantly improved with the co-use of biochar addition at 1% C ha<sup>-1</sup> and N fertilizer amendment to the soil. It was clearly indicated that biochar addition with N fertilizer has positive effects on microbial biomasses and enzymatic activity in the dryland region as compared to biochar addition without N fertilization. The results of the study demonstrated that sugarcane bagasse-biochar incorporation to low organic matter (organic carbon) arid soils has the potential to improve the soil function and crop productivity by revitalizing the microbial biomass activity.

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