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BIOCHAR FOR A SUSTAINABLE ENVIRONMENT

Effects of biochar and polyacrylamide on decomposition of soil organic matter and ^{14}C-labeled alfalfa residues

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Abstract
Purpose Various soil conditioners, such as biochar (BC) and anionic polyacrylamide (PAM), improve soil fertility and susceptibility to erosion, and may alter microbial accessibility and decomposition of soil organic matter (SOM) and plant residues. To date, no attempts have been made to study the effects of BC in combination with PAM on the decomposition of soil SOM and plant residues. The objective of this study was to evaluate the effects of BC, PAM, and their combination on the decomposition of SOM and alfalfa residues.
Materials and methods An 80-day incubation experiment was carried out to investigate the effects of oak wood biochar (BC; 10 Mg ha^{-1}), PAM (80 kg ha^{-1}), and their combination (BC + PAM) on decomposition of SOM and ^{14}C-labeled alfalfa (*Medicago sativa* L.) residues by measuring CO_2 efflux, microbial biomass, and specific respiration activity.
Results and discussion No conditioner exerted a significant effect on SOM decomposition over the 80 days of incubation. PAM increased cumulative CO_2 efflux at 55–80 days of incubation on average of 6.7 % compared to the soil with plant residue. This was confirmed by the increased MBN and MB^{14}C at 80 days of incubation in PAM-treated soil with plant residue compared to the control. In contrast, BC and BC + PAM decreased plant residue decomposition compared to that in PAM-treated soil and the respective control soil during the 80 days. BC and BC + PAM decreased MBC in soil at 2 days of incubation indicated that BC suppressed soil microorganisms and, therefore, decreased the decomposition of plant residue.
Conclusions The addition of oak wood BC alone or in combination with PAM to soil decreased the decomposition of plant residue.

Keywords ^{14}C-alfalfa residue decomposition · Biochar · Microbial biomass · Polyacrylamide · Soil organic matter

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1 Introduction

The loss of topsoil with soil organic matter (SOM) because of erosion reduces soil quality and crop production (Nagle 2002; Lee et al. 2015). In particular, SOM improves soil aggregation, water retention, microbial biomass and activity, and plant-available nutrients, contributing to maintenance of soil quality (Mikha and Rice 2004; Atul-Nayyar et al. 2009; Lee et al. 2009; Katsalirou et al. 2010). Therefore, developing a method to maintain SOM and reduce soil erosion is necessary.

From a practical point of view, carbon input is a promising technology to increase SOM and improve soil quality (Awad et al. 2012). As the main source of organic C, adding plant residue is an important strategy to maintain SOM, improve soil quality, and promote crop production (Blagodatskaya et al. 2009; Chen et al. 2009; Guzman and Al-Kaisi 2010).

In addition, plant residue decreased soil erosion and surface runoff, owing to a 12 % reduction of soil loss (Santhi et al. 2006; Panagos et al. 2015). Notably, labile organic C in plant residue stabilizes soil aggregates (Abiven et al. 2008; Langer and Rinklebe 2011). In particular, degraded plant C binds microaggregates (<0.25 mm) to form stable macroaggregates (>0.25 mm) (Abiven et al. 2008). Understanding C turnover, which is associated with plant residue and SOM decomposition, is necessary to predict the effects of agricultural practices on soil C sequestration (Fu et al. 2000; Kuzyakov 2006). Applying ^{14}C-labeled plant residue to soil facilitates the investigation of C flux in plant/soil systems and determines the decomposition rate of plant residue (Chen et al. 2009).

As a good soil conditioner, biochar (BC) improves soil quality and productivity in term of increased SOC content, cation exchange capacity (CEC), and plant-available nutrients (Glaser et al. 2002; Jones et al. 2011; Kuzyakov et al. 2014; Lehmann et al. 2006, 2015; Liu et al. 2016). BC has also been used to remediate organic and inorganic pollutants from contaminated soils (Ahmad et al. 2014; Ok et al. 2015; Rinklebe et al. 2016). Wood BC decreases soil bulk density and improves aggregation, contributing to a pronounced decrease in soil loss (Jien and Wang 2013; Lee et al. 2015). In contrast, the stability of wood biochar against microbial decomposition typically contributes to C storage or sequestration (Ameloot et al. 2013; Bandara et al. 2015). However, little is known about the effects of BC on soil microbial activities (Lehmann et al. 2011). In contrast, anionic polyacrylamide (PAM) improves the physicochemical and biological properties of soil and reduces erosion and runoff as a binding agent by stabilizing the outer surfaces of aggregates, increasing aggregate stability, and therefore, improving water penetration (Levy and Miller 1999; Sojka et al. 2007; Wu et al. 2012; Awad et al. 2013). Moreover, PAM is highly resistant to microbial degradation, with a decomposition rate of ~10 % year^{-1}, and, therefore, can be efficiently used as a soil conditioner for long periods (Lee et al. 2010). The short-term dynamics of C caused by PAM or BC should be assessed to reduce erosion and maintain soil quality (Lu and Zhang 2015). A combination of oak wood BC and PAM improves soil physicochemical properties and reduces runoff and soil loss under simulated and natural rainfall (Lee et al. 2015). Therefore, the interactive effects of BC and PAM on the decomposition of SOM and plant residue should be investigated.

We hypothesized that oak wood BC may suppress plant residue decomposition in soil. In addition, the combination of BC and PAM may represent a promising technology to sequester C in the soil by decreasing plant residue decomposition. To date, no research evaluated the decomposition of plant residue in response to application of a combination of biochar and polyacrylamide. Therefore, in this study, we investigated the effect of BC, PM, or BC + PAM on the decomposition of SOM and ^{14}C-labeled alfalfa plant residue in soils. Herein, for the first time, we report the interactive effects of BC and PAM on the decomposition of SOM and plant residue in soil by measuring microbial biomass and specific respiration activity.

2 Materials and methods

2.1 Soil collection and analyses

Soil was collected from Ap horizon (0–30 cm) from an agricultural field in Hongcheon, Gangwon Province, Korea. The soil was air-dried, homogenized, and passed through a 2-mm sieve. Soil texture was measured by the pipette method (Sheldrick and Wang 1993), and soil water holding capacity (17.33 %) was determined gravimetrically (Veihmeyer and Hendrickson 1931). Soil pH and electrical conductivity (EC) were measured in 1:5 soil water extract using a pH-EC meter (Orion 3 Star, Thermo, Rockford, IL, USA). Soil total carbon (TC) and nitrogen (TN) contents were determined using a Multi N/C 2100 S analyzer (Analytik, Jena, Germany). The soil exchangeable cations were analyzed in 1 M NH_4OAc extract by inductively coupled plasma spectrometry (Sumner and Miller 1996), as shown in Table 1.

Table 1 Physicochemical properties of the soil and characterization of biochar (BC) and anionic polyacrylamide (PAM)

	Sand	Silt	Clay	Texture	pH	EC[a]	TC[b]	TN[c]	C/N	Exchangeable cations			
	%					dS m^{-1}	%	%		Ca^{2+}	Mg^{2+}	K^+	Na^+
										cmol$_{(+)}$ kg^{-1}			
Soil	45.58	31.13	23.29	Loam	6.15	0.05	1.04	0.09	11.45	7.24	2.17	0.06	0.07
BC	–	–	–		10.17	2.15	83.15	0.39	213.21	17.99	3.24	0.06	12.00
PAM	–	–	–		7.40	-	42.20	16.20	2.60	–	–	–	–

[a] electrical conductivity

[b] total carbon

[c] total nitrogen

2.2 Characteristics of soil conditioners

Oak wood biochar (Sootgage, Hoengseong, Gangwon Province, Korea) produced at 400 °C and anionic polyacrylamide (Magnafloc 336, Ciba Canada Ltd., Mississauga, Ontario, Canada) were applied to the soil in an incubation experiment. Biochar (BC) at 10 Mg ha^{-1} was mixed with soil, and a 500 mg L^{-1} solution of anionic polyacrylamide (PAM) was applied at a rate of 80 kg ha^{-1}. The application rates of BC and PAM were chosen according to Lee et al. (2015). BC comprised 31.4 % mobile matter, 5 % ash, surface area of 270.8 m^2 g^{-1}, a pore volume of 0.12 cm^3 g^{-1}, and an average pore size of 1.1 nm (Ahmad et al. 2012; Table 1).

2.3 Incubation

The first factor was soil conditioners (BC, PAM, and BC + PAM), along with untreated soil as a control, and the second factor was addition of 100-mg ^{14}C-labeled alfalfa residue (21, 400 disintegrations per minute (DPM) mg^{-1}) per 30 g soil. Alfalfa (*Medicago sativa* L.) plants were labeled three times in a $^{14}CO_2$ atmosphere according to the method described by Gocke et al. (2011) to produce the ^{14}C-labeled litter and then ground using a ball mill (MM2, FaRetsch). Plant residues were thoroughly mixed with 30 g of air-dried soil containing one of the four soil conditioners above. The treated soils were placed in closed vessels, maintained at 70 % of water holding capacity with deionized water, and then incubated at 22 °C for 80 days. Four replicates were performed.

Small vials containing 2 mL of 1.0 M NaOH were placed in the vessels to trap CO_2. These vessels were changed at 1, 2, 3, 4, 5, 7, 10, 17, 24, 31, 38, 52, 66, and 80 days of incubation to measure CO_2 and ^{14}C activity. Additionally, four blank vessels containing only NaOH were used as controls.

2.4 CO_2 efflux and ^{14}C analyses

NaOH solution (1.0 M) was prepared to trap CO_2, and trapped CO_2 was precipitated by 0.5 M $BaCl_2$ solution to estimate CO_2 efflux rates. The NaOH solution was titrated with 0.1 M HCl against phenolphthalein indicator (Zibilske 1994). A 0.4-mL aliquot of NaOH solution was mixed with 2 mL of Rothiscint-22x scintillation cocktail (Carl Roth Co., Karlsruhe, Germany) to measure ^{14}C activity after decay of chemiluminescence by a liquid scintillation counter (MicroBeta TriLux, 205 Perkin Elmer Inc., Waltham, MA, USA). The measurement error did not exceed 3 %, and the ^{14}C counting efficiency was ~93 %.

2.5 Microbial biomass

After 2 and 80 days of incubation, soil microbial biomass C (MBC) and N (MBN) were analyzed by the chloroform fumigation-extraction method (modified after Vance et al. (1987)). Specifically, 5 g of soil were extracted with 20 mL of 0.05 M K_2SO_4. Another portion of 5 g soil was fumigated with alcohol-free chloroform ($CHCl_3$) in a desiccator for 48 h, after which the soil was extracted as described for the unfumigated soil. The extracts were frozen until being analyzed for TC and TN using a multi N/C 2100 S analyzer (Analytik Jena, Germany). To calculate ^{14}C incorporation in microbial biomass derived from plant residue, ^{14}C activity was measured in the K_2SO_4-extractable C in fumigated and unfumigated soils by mixing 2 mL of soil extract with 5 mL of scintillation cocktail (Blagodatskaya et al. 2011).

2.6 Calculations and statistical analysis

$^{14}CO_2$ efflux from labeled plant residue was estimated as the percentage of initial input. ^{14}C activity and CO_2 efflux rates were calculated as described by Kuzyakov and Cheng (2004) and Van Groenigen et al. (2005). MBC and MBN values were estimated as described by Wu et al. (1990) and Brookes et al. (1985), respectively. Microbial biomass ^{14}C (% of ^{14}C input) was calculated as follows:

$$MB^{14}C = \left(\left[MB^{14}C_F - MB^{14}C_{Unf}\right]:{}^{14}C_{input}\right) \times 100$$

where $MB^{14}C_F$ is the activity of ^{14}C in fumigated soil extract (DPM g^{-1}), $MB^{14}C_{Unf}$ is the activity of ^{14}C in unfumigated soil extract (DPM g^{-1}), and $^{14}C_{input}$ is the activity of ^{14}C-labeled plant residue input (DPM g^{-1}). The metabolic quotient (qCO_2) or specific respiration activity was estimated as the CO_2 production per unit MBC and time (mg of C mineralized per g of microbial biomass per unit time; mg CO_2-C h^{-1} g^{-1} microbial biomass C) (Leita et al. 1995).

The standard error of the mean was calculated. A two-way factorial analysis of variance and Tukey's honestly significant difference test at a 0.05 probability level were performed to compare means. Data were analyzed using the SAS/STAT® 9.3 computer software package (SAS 2004).

3 Results

3.1 CO_2 efflux

For soils without plant residue, the highest CO_2 efflux rate was observed after 1 day of incubation and then decreased until 24 days of incubation. CO_2 efflux then remained stable low level until the end of incubation (or 80 days) (Fig. 1a). No significant differences in CO_2 efflux rates were observed in soils with BC, PAM, or BC + PAM compared to the control or no soil conditioner. No significant differences in cumulative CO_2 were found after adding BC, PAM, or BC + PAM compared to the control (Fig. 1b).

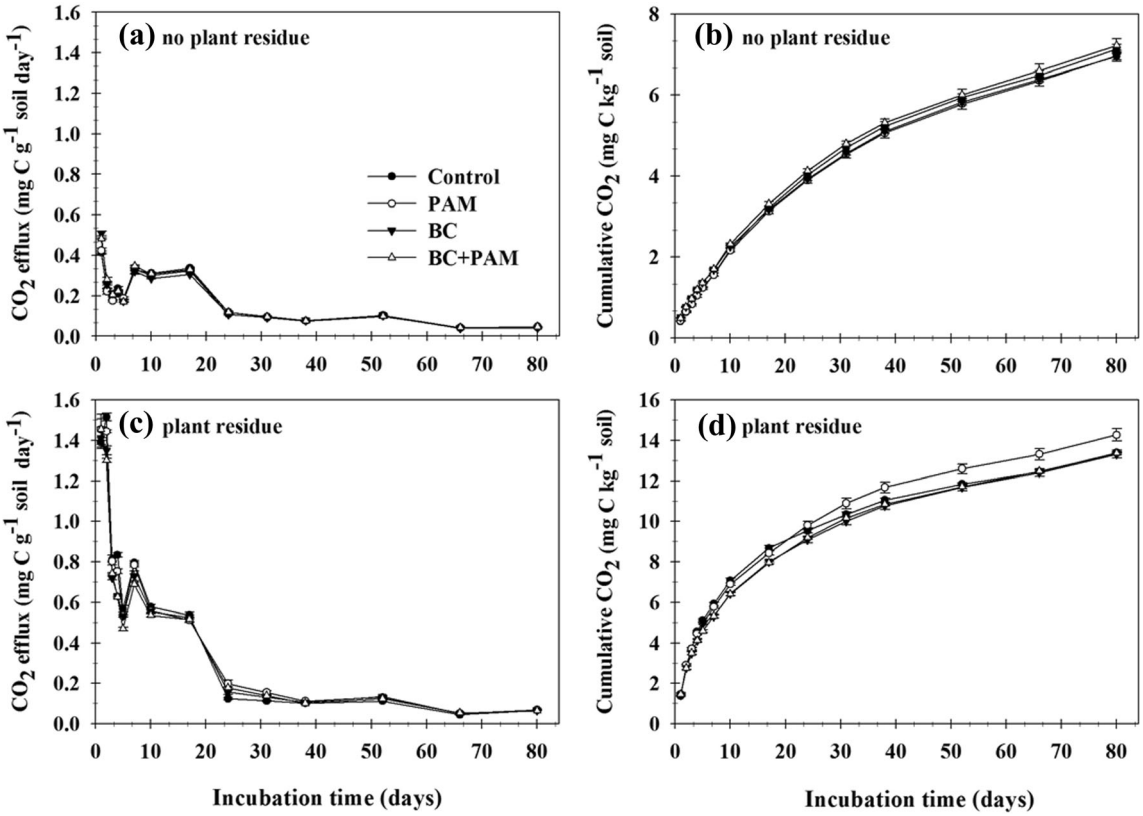

Fig. 1 CO_2 efflux rates (mg C day^{-1} g^{-1} soil) and cumulative CO_2 efflux (mg C kg^{-1} soil) from soils treated with conditioners (control: untreated soil, PAM: polyacrylamide at 80 kg ha^{-1}, BC: biochar at 10 Mg ha^{-1}, and BC + PAM: a combination of biochar and polyacrylamide) without (**a**, **b**)/with (**c**, **d**) addition of plant residue. *Error bars* represent the standard error of the mean ($n = 4$)

For the soils with ^{14}C-labeled plant residue, the CO_2 efflux at 1 day of incubation was 3.4-fold higher than that of soil without plant residue (Fig. 1a, c). BC and BC + PAM addition decreased CO_2 efflux during 2–7 days of incubation by 9.6–25.1 % compared to the control (Fig. 1c). BC and BC + PAM addition decreased cumulative CO_2 efflux by 7.8 and 9.5 % on average at 4–24 days of incubation, respectively, compared to the control (Fig. 1d). In contrast, PAM increased the cumulative CO_2 efflux by an average of 6.6 % at 52–80 days of incubation, compared to the control.

3.2 Decomposition of ^{14}C-labeled plant residue in treated soils

Decomposition of ^{14}C-labeled plant residue occurred in two phases: 1–24 days and 24–80 days of incubation. Based on $^{14}CO_2$ efflux, the maximum plant residue decomposition rates occurred after 1 day of incubation, ranging from 13.1 to 15.2 % of ^{14}C input day^{-1}, and then declined (Fig. 2a). PAM addition induced a slight short-term increase in $^{14}CO_2$ efflux by 7.2 % at 1 day of incubation compared to the control. In contrast, BC and BC + PAM decreased ^{14}C plant residue mineralization rates by 8.2–17.7 % at 1–10 days of incubation compared to the control (Fig. 2a).

During 80 days of incubation, the cumulative $^{14}CO_2$ efflux of the BC and BC + PAM treated soils was significantly lower by 6.1–12.3 and 5.2–11 %, respectively, than that of the control (Fig. 2b, c). Additionally, BC and BC + PAM addition resulted in lower cumulative $^{14}CO_2$ efflux at 80 days of incubation by 6.1 and 5.7 %, respectively. However, PAM induced a slight short-term increase in cumulative $^{14}CO_2$ efflux from the soil after 1 and 2 days of incubation by an average of 5.3 % compared to the control (Fig. 2b, c). Based on $^{14}CO_2$ efflux, ~64–68 % of ^{14}C input from plant residue was mineralized in soils during the 80 days of incubation.

3.3 Specific respiration activity

For soils without plant residue, BC and BC + PAM addition increased qCO_2 by 2.3-fold on average at 2 days of incubation when compared to the control (Fig. 3a). However, no significant difference in qCO_2 among the conditioners with added

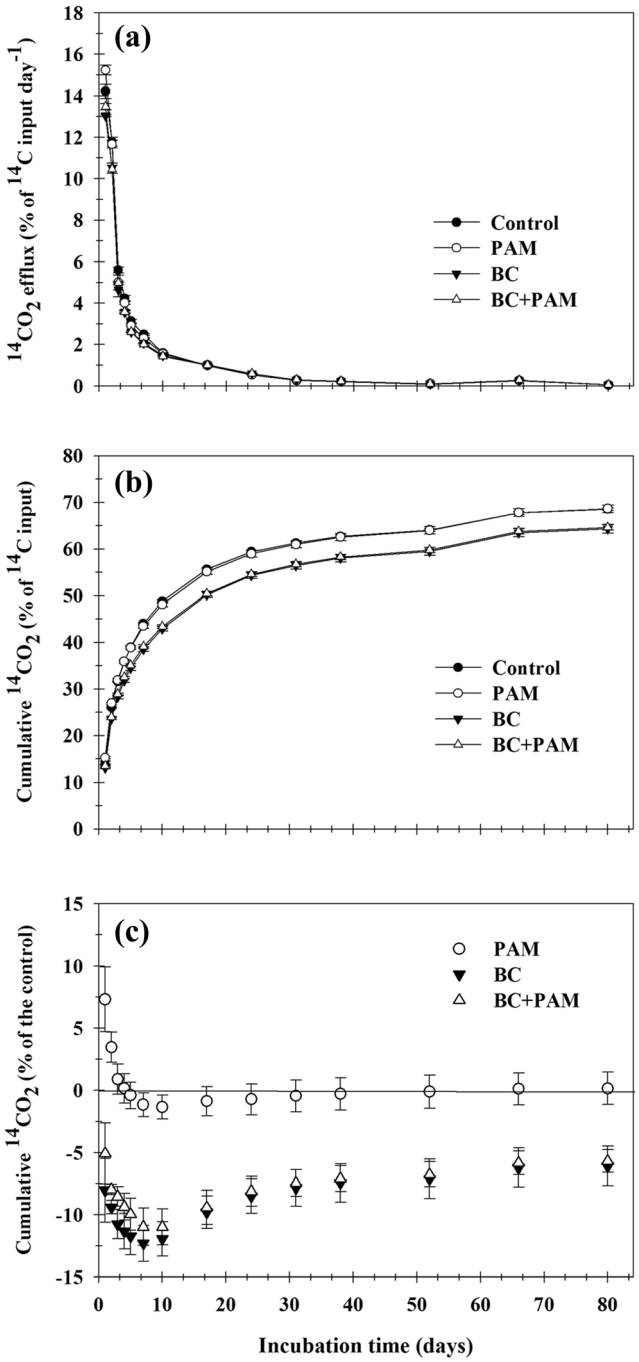

Fig. 2 $^{14}CO_2$ efflux rates (**a**, % of ^{14}C input day^{-1}), cumulative $^{14}CO_2$ efflux (**b**, % of ^{14}C input) and changes in cumulative $^{14}CO_2$ relative to control (**c**) from decomposition of ^{14}C-labeled alfalfa residue in soils treated with conditioners (control: untreated soil, PAM: polyacrylamide at 80 kg ha^{-1}, BC: biochar at 10 Mg ha^{-1}, and BC + PAM: a combination of biochar and polyacrylamide). *Error bars* represent the standard error of the mean ($n = 4$)

plant residue including the control were observed at 2 and 80 days of incubation (Fig. 3b). For soils with plant residue, the qCO_2 at 2 days of incubation was 6.5-fold higher than that of soil without plant residue.

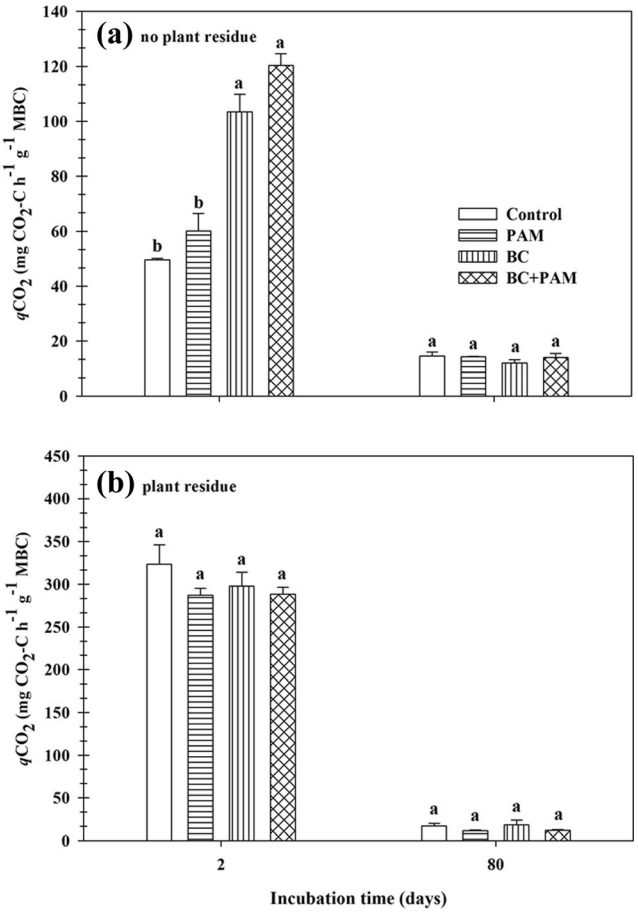

Fig. 3 The metabolic quotient (qCO_2) at 2 and 80 days in soils treated with conditioners (control: untreated soil, PAM: polyacrylamide at 80 kg ha^{-1}, BC: biochar at 10 Mg ha^{-1}, and BC + PAM: a combination of biochar and polyacrylamide) without (**a**)/with (**b**) addition of plant residue. *Different letters* above each bar indicate a significant difference at $P \leq 0.05$. *Error bars* represent the standard error of the mean ($n = 4$)

3.4 Microbial biomass C, N, and ^{14}C

For soils without plant residue, BC and BC + PAM decreased soil MBC by 46.0 and 49.7 %, respectively, at 2 days of incubation (Fig. 4a). BC and BC + PAM addition to the soil suppressed MBC only at 2 days of incubation. No significant difference in MBN at 2 and 80 days of incubation was observed among conditioners and the control (Fig. 4b).

For soils with plant residue, no significant differences in MBC or MBN among conditioners including the control were observed at 2 and 80 days of incubation, with the exception of 35.5 % increase in MBN in PAM-treated soil at 80 days compared to the control (Fig. 4c, d).

Only PAM addition increased MB^{14}C significantly by 33.8 % at 80 days of incubation compared to the control (Fig. 5).

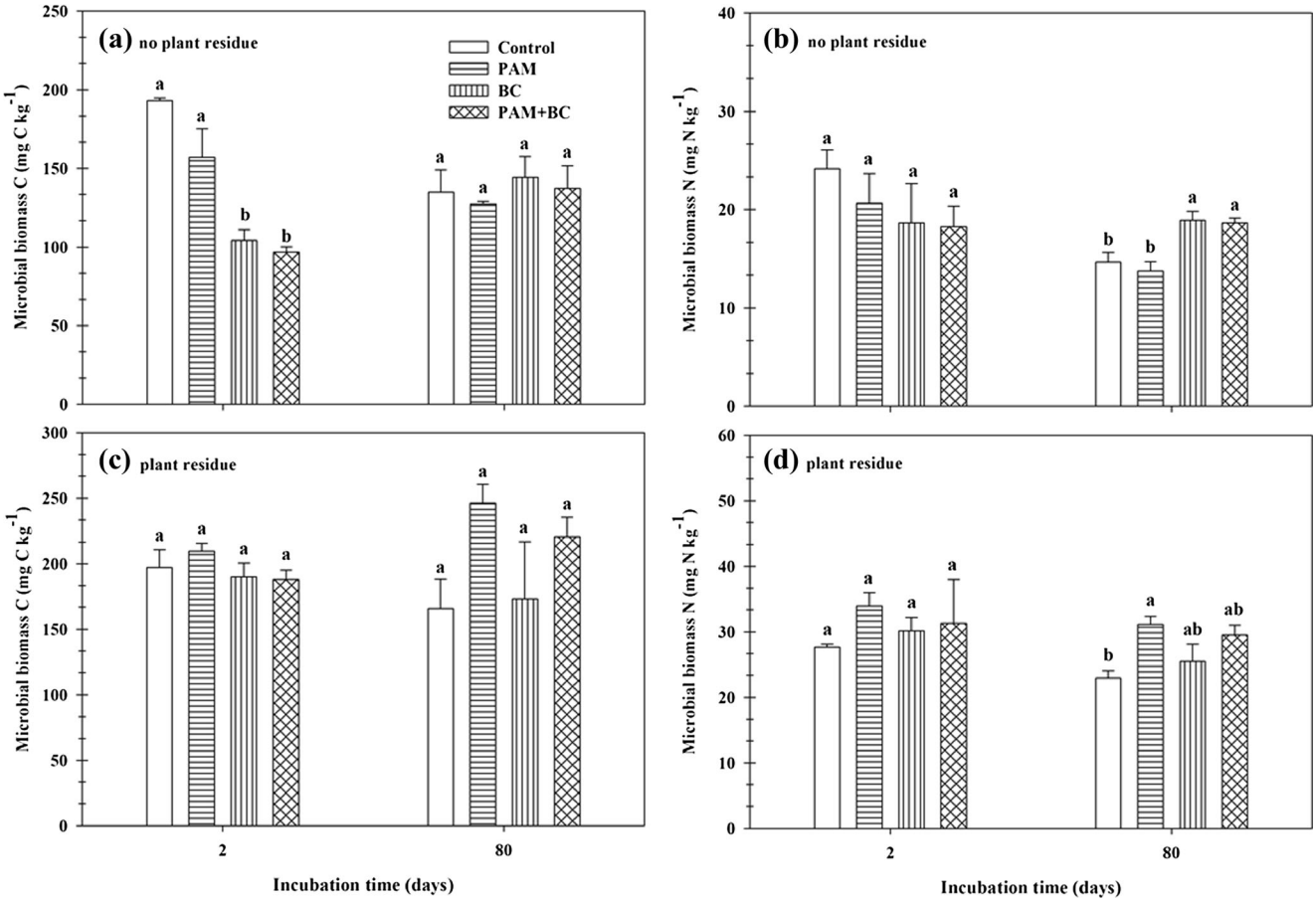

Fig. 4 Microbial biomass carbon (MBC) and nitrogen (MBN) at 2 and 80 days of incubation in soils treated with conditioners (control: untreated soil, PAM: polyacrylamide at 80 kg ha^{-1}, BC: biochar at 10 Mg ha^{-1}, and BC + PAM: a combination of biochar and polyacrylamide) without (**a**, **b**)/ with (**c**, **d**) addition of plant residue. *Different letters* above each bar indicate a significant difference at $P \leq 0.05$. *Error bars* represent the standard error of the mean ($n = 4$)

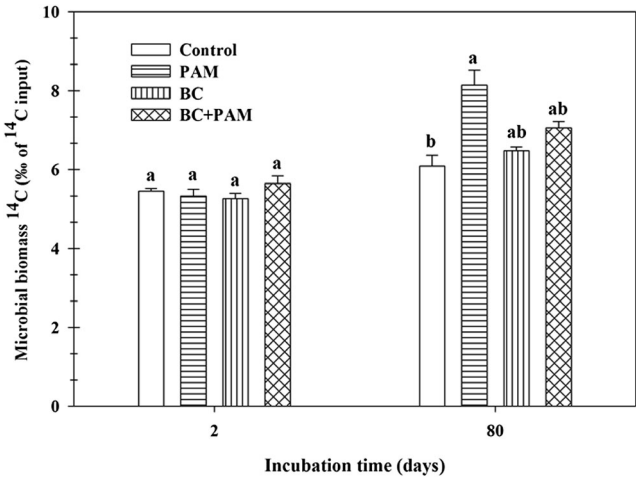

Fig. 5 Microbial biomass ^{14}C (‰ of ^{14}C input) at 2 and 80 days of incubation in soils treated with conditioners (control: untreated soil, PAM: polyacrylamide at 80 kg ha^{-1}, BC: biochar at 10 Mg ha^{-1}, and BC + PAM: a combination of biochar and polyacrylamide) with addition of plant residue. *Different letters* above each bar indicate a significant difference at $P \leq 0.05$. *Error bars* represent the standard error of the mean ($n = 4$)

4 Discussion

4.1 Soil conditioners on SOM decomposition

Labile organic C in soil can be used by microorganisms, resulting in CO_2 emission (Awad et al. 2012, 2013). The sharp increase in CO_2 efflux at the first day of incubation occurred because of the accessibility of labile C in SOM or plant residue to soil microbes (Awad et al. 2012, 2013). Specifically, readily biodegradable organic C in soil and plant residue increases microbial respiration, thereby increasing CO_2 emissions (Usman et al. 2004; Awad et al. 2012; Vourlitis and Fernandez 2015). The mineralization rate of SOM was highest during 24 days of incubation and decreased thereafter (Fig. 1). This is due to the higher decomposition rate of readily biodegradable organic C by microorganisms. Thereafter, a decrease in labile organic C resulted in reduced CO_2 efflux rates at 24–80 days of incubation (Awad et al. 2012, 2013; Usman et al. 2004).

Based on CO_2 efflux, the BC, PAM, and BC + PAM treatments had no significant effects on SOM mineralization (Fig. 1a, b). Because the decomposition rate of biochar is extremely slow, its contribution to CO_2 efflux is negligible (Kuzyakov et al. 2009, 2014). For instance, Hilscher et al. (2009) observed no increase in CO_2 efflux rates after adding pine wood biochar to a loam soil. In addition, the decomposition rate of PAM (~10 % $year^{-1}$) is faster than BC, and PAM can stimulate SOM mineralization by increasing available O_2 in soil (Entry et al. 2008); however, its effect may be negligible due to labile C availability and time limitation. BC and PAM exerted a minor effect on SOM decomposition based on CO_2 evolution because of the stability of these conditioners (Awad et al. 2012, 2013).

4.2 Plant residue decomposition relative to microbial biomass

Application of fresh organic materials to soil increased CO_2 efflux rates over the first few days of incubation (Neale et al. 1997; Usman et al. 2004). Similar to the findings of Aerts (1997), high CO_2 and cumulative CO_2 effluxes evolved to the atmosphere from the decomposition of ^{14}C plant residue during 80 days of incubation compared to soils without plant residue (Fig. 2). Decomposition of plant residue occurred in two phases: 1–24 and 24–80 days of incubation, and indicated rapid decomposition of easily degradable components of plant residue (sugars, starch, and cellulose) and slow decomposition of recalcitrant compounds (lignin and phenolic components) (Dilly and Munch 2004; Chen et al. 2009; Awad et al. 2012).

The addition of PAM increased cumulative CO_2 emission at 52–80 days of incubation (Fig. 1d) and the decomposition of ^{14}C-labeled plant residue after 1 and 2 days of incubation was increased in PAM-treated soil compared to the control soil (Fig. 2). Therefore, PAM increased the decomposition of plant residue due to the improved macroaggregates. Compared to sandy or sandy loam soil in the study by Awad et al. (2012), the higher exchangeable Ca^{2+} (7.2 $cmol_{(+)}\ kg^{-1}$) as electrolyte and clay (23.3 %) in loam soil increased the number of charged sites for bonding with PAM molecules (Levy and Miller 1999; Mamedov et al. 2007; Lee et al. 2010).

The high N content in PAM plays an important role in increasing the populations of microorganisms that decompose SOM (Caesar-TonThat et al. 2008). Available inorganic N may also be produced from hydrolysis of acrylamide in PAM under the aerobic conditions in soil macroaggregates (Abdelmagid and Tabatabai 1982; Kay-Shoemake et al. 1998). The addition of PAM together with plant residue increased chitinase activity by 1.3-fold compared to that of untreated soil with plant residue (Awad et al. 2012). This mechanism was confirmed by the higher MBN and MB^{14}C at 80 days of incubation in the PAM-treated soil compared to the control soil (Figs. 4d and 5).

Biochar alone and in combination with PAM suppressed added or native C mineralization based on CO_2 and $^{14}CO_2$ efflux (Figs. 1 and 2). A high portion of ^{14}C-maize residue was stabilized in the soil after addition of BC and PAM as binders due to occlusion of labile residue-C into aggregates (Awad et al. 2013). These negative priming effects of biochar could also be due to its high sorption affinity for OM or labile plant residue substrates to its surface or within its pores, which decreases plant residue decomposition (Lehmann et al. 2011; Zimmerman et al. 2011). Liang et al. (2010) found a 2–3-fold lowered C respiration in BC-treated soil. Also, Bandara et al. (2015) reported reduced activities of extracellular enzymes, such as polyphenol oxidase and catalase, in BC-treated soil. In our study, BC may have acted as a binding agent for labile organic C during aggregate formation and physically inhibited the decomposition of SOM and plant residue (Brodowski et al. 2006). It is noteworthy that biochar sorbs microbial-produced enzymes onto its external surface or suppresses their activity (Zimmerman et al. 2011). This hypothesized mechanism was supported by the high surface area of 270 $m^2\ g^{-1}$ of biochar in this study. A high pyrolytic temperature (400 °C) destroys aliphatic alkyl and ester groups, thereby increasing BC surface area (Chen et al. 2009). Additionally, biochars sorb SOM, which reduces their dissipation in the soil, owing to inaccessibility to microorganisms (Yang and Sheng 2003; Spokas et al. 2009; Kasozi et al. 2010). The effect of BC on the decomposition of SOM and plant residue is driven by pyrolysis conditions and feedstock type (Wang et al. 2012, 2013, 2015; Ahmad et al. 2014). In our study, BC produced from wood at 400 °C sorbed SOM, similar to the findings of Kasozi et al. (2010), in which BC produced from 400 to 650 °C had a larger nanopore surface area than that produced at a low pyrolytic temperature (<400 °C). Furthermore, the dissolved organic C content of BC-treated soil was decreased due to adsorption (Kasozi et al. 2010; Novak et al. 2016).

There was no significant difference in MBC and MBN values in soils treated with BC and BC + PAM because labile plant residue provides C or N to soil microorganisms, increasing their decomposition activity (Awad et al. 2012). Majumder and Kuzyakov (2010) reported that addition of plant residue resulted in a 1.7–5.4-fold increase in MBC compared to soil without added plant residue. The availability of labile C from plant residue is due to the higher microbial activity compared to that in soil without plant residue (Fu et al. 2000; Majumder and Kuzyakov 2010).

4.3 Microbial biomass ^{14}C and metabolic quotient

The metabolic quotient was associated with optimal substrate utilization and environmental stress on soil microorganisms in the current study, as reported previously by Usman et al. (2004). No significant differences in qCO_2, or MB-^{14}C at

2 days of incubation in soil with plant residue were observed (Figs. 3b and 5). This may be due to the large amount of respired C during the 1 day of incubation as well as mineralization of the available labile organic C in soil. These findings are in agreement with previous reports (Fontaine et al. 2003; Hamer and Marschner 2005). Addition of plant residue increased the qCO_2 significantly compared to control soil without plant residue or soil treated with BC and BC + PAM (Fig. 3), due to the large amount of dissolved organic C. Similar to our findings, Usman et al. (2004) reported a higher qCO_2 after application of sewage sludge to soil. The lower qCO_2 values at the end of incubation indicate lower microbial respiration due to reduced available C levels (Griffiths et al. 1998; Usman et al. 2004). Thus, addition of plant residue led to an increase in dissolved organic C, thereby increasing the microbial respired CO_2-C compared to soil without plant residue (Awad et al. 2012).

5 Conclusions

Based on cumulative CO_2 emission, there was no significant difference in SOM mineralization after addition of BC, PAM, or their combination compared to control soil without plant residue. In contrast to PAM, BC and BC + PAM increased specific respiration activity (qCO_2) at 2 days of incubation in soil without plant residue. For soils with plant residue, the addition of PAM alone increased the cumulative CO_2 efflux compared to other treatments and the control. This was due to the increased MBN and MB^{14}C at 80 days of incubation in soil with plant residue compared to the control. BC alone or in combination with PAM suppressed the MBC and decreased the decomposition of plant residue compared to that in the control soil. This was confirmed by the suppression of microorganisms by BC because of the dissolved organic C content of BC-treated soil was decreased due to adsorption.

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