



Emissions intensity and carbon stocks of a tropical Ultisol after amendment with Tithonia green manure, urea and biochar



Bernard Fungo^{a,b,c,*}, Johannes Lehmann^d, Karsten Kalbitz^{b,e}, Moses Tenywa^f, Margaret Thiongo^c, Henry Neufeldt^c

^a National Agricultural Research Organization (NARO), P.O. Box 1752, Kampala, Uganda

^b Institute for Biodiversity and Ecosystem Dynamics (IBED), Faculty of Science, University of Amsterdam, Science Park 904, 1098 XH Amsterdam, The Netherlands

^c CGIAR Research Program on Climate Change, Agriculture and Food Security (CCAFS), World Agroforestry Center (ICRAF), P.O. Box 30667, UN, Avenue-Gigiri, Nairobi, Kenya

^d Soil and Crop Sciences, Cornell University, Bradfield Hall, Ithaca, NY 14853, USA

^e Soil Resources and Land Use, Institute of Soil Science and Site Ecology, Dresden University of Technology, Piennner Strasse 19, 01737 Tharandt, Germany

^f Collage of Agricultural and Environmental Sciences, Makerere University, P.O. Box 7062, Kampala, Uganda

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ABSTRACT

Biochar has been shown to reduce soil emissions of CO₂, CH₄ and N₂O in short-term incubation and greenhouse experiments. Such controlled experiments failed to represent variable field conditions, and rarely included crop growth feedback. The objective of this study was to assess the effect of biochar, in comparison to green manure and mineral nitrogen, on greenhouse gas Emissions Intensity (EI = emissions in CO₂ equivalents per ton of grain yield) in a low-fertility tropical Ultisol. Using a field trial in western Kenya, biochar (0 and 2.5 t ha⁻¹; made from Eucalyptus wood) was integrated with urea (0 and 120 kg N ha⁻¹) and green manure (*Tithonia diversifolia*; 0, 2.5 and 5 t ha⁻¹) in a factorial design for four consecutive seasons from October 2012 to August 2014. Compared to the control, biochar increased soil CO₂ emissions (9–33%), reduced soil CH₄ uptake (7–59%) and reduced soil N₂O emissions (1–42%) in each season, with no seasonal differences. N₂O emissions increased following amendment with *T. diversifolia* (6%) and urea (13%) compared to the control. Generally, N₂O emissions decreased where only biochar was applied. The greatest decrease in N₂O (42%) occurred where all three amendments were applied compared to when they were added separately. EI in response to any of the amendments was lower than the control, ranging from 9 to 65% (33.0 ± 3.2 = mean ± SE). The amendments increased SOC stocks by 0.1–1.2 t ha⁻¹ year⁻¹ (mean ± SE of 0.8 ± 0.09 t ha⁻¹ year⁻¹). The results suggest decreased net EI with biochar in low fertility soils mainly through greater net primary productivity (89% of the decrease).

1. Introduction

The search for climate-smart agricultural production technologies is directing research to identify innovations that address multiple benefits such as crop productivity, carbon sequestration and mitigation of soil-atmosphere greenhouse gas (GHG) emissions. Addition of biochar (pyrogenic organic matter) to agricultural soils as a management strategy has reportedly increased crop yields in several studies but has shown variable effects on GHG fluxes (Knoblauch et al., 2011; Cayuela et al., 2014). Biochar may affect fluxes of GHGs such as CO₂, CH₄ and N₂O by a variety of mechanisms, including: (i) the turnover rate of soil organic matter (SOM), which in turn determines the availability of C and N, the precursors for GHG production or

consumption, (ii) soil physical properties (e.g. gas diffusivity, aggregation, water retention) (Quin et al., 2014); (iii) soil chemical properties (e.g. pH, Eh, availability of organic and mineral N and dissolved organic C, organo-mineral interactions); and (iv) soil biological properties (e.g. microbial community structure, microbial biomass and activity, macrofauna activity, N cycling enzymes) (Van Zwieten et al., 2010).

Biochar may also change the effects of adding easily mineralizable organic matter as well as mineral N fertilizers on GHG emissions from soil. Additions of legume materials as a fertilizer provide both N and C that typically lead to greater GHG emissions from soils (Gentile et al., 2009). Similarly, the amount of fertilizer N additions is considered proportional to the N₂O emissions (Manzoni and Porporato, 2009; Mori and Hojito, 2011). It is not clear, if simultaneous addition of biochar

* Corresponding author at: National Agricultural Research Organization (NARO), P.O. Box 1752, Kampala, Uganda.
E-mail address: bfungo1@yahoo.com (B. Fungo).

with either fertilizer N or legume mulch or a combination if the two may result in GHG emission reductions. Uncertainty also exists whether any emission reductions would persist over several cropping seasons.

It is not clear what role possible feedback through enhanced crop growth plays to the GHG budget. Greater crop growth and presumably greater C return to soil have been found where the pH is increased by biochar to neutral values (Jeffery et al., 2015) and this feedback would therefore be expected to be greatest in acid tropical soils. Whereas Spokas (2013) suggested that biochar has mainly shorter-term GHG mitigation effect (few days to several weeks) after application, Lentz et al. (2014) indicated that the effects may be long-lived. As such, questions remain concerning the long-term implications in cropping periods particularly for field-based biochar studies. Zimmerman et al. (2011) observed that native SOM mineralization was higher during the early incubation stage (first 90 days) and low during the later incubation stage (250–500 days). Maestrini et al. (2014) also reported pyrogenic OM (PyOM) to have promoted native OM mineralization during the first 18 days and inhibited it afterward (up to 150 days).

The objectives of this study were to determine the effect of biochar on (i) GHG fluxes (CO₂, CH₄ and N₂O), (ii) Emissions Intensity (EI; the net CO₂-equivalent for CH₄ and N₂O per ton of grain yield), and (iii) changes in soil organic carbon (SOC) and ecosystem carbon balance of a low-fertility tropical agricultural soil when integrated with organic and mineral N inputs. The overall hypothesis is that biochar is responsible for controlling the release of labile N and C from high N mineral and organic amendments and an accompanying reduction in CO₂, CH₄ and N₂O emissions. Specifically, we hypothesize that compared to unamended soil, biochar (i) reduces availability of N from both organic and mineral sources such as *T. diversifolia* and urea to thereby reduce N₂O emission resulting from interactions of N₂O with biochar; (ii) increases availability of easily mineralizable C from both soil and organic amendment to reduce the CH₄ soil sink; (iii) affects emissions of CH₄ and N₂O early on, but not in later seasons as active surfaces get saturated with time; and (iv) increases plant growth as a result of biochar additions that are more important than changes in other soil processes affecting GHG emissions.

2. Materials and methods

2.1. Study site

The field experiment was established in September 2012 at Kapsengere on the southern Nandi hills in western Kenya (00° 09' 34"N and 34° 57' 37"E). The site receives ~2000 mm mean annual rainfall in a bimodal distribution, with two growing seasons per year, March–July and September–January. The mean annual temperature is 26 °C. The soils are classified as Typic Kandiodults (USDA, 1999) developed on biotite-gneiss parent material. The experimental field was divided into three blocks. Soil properties before the experiment were determined by taking two samples from each block (six composite samples in total). The composite sample was obtained by mixing soil taken at four random locations. These were assumed to adequately represent the entire field where the experiment was established. The soil samples were analyzed using methods described in Fungo et al. (2014); in addition, particle size distribution was determined by the hydrometer method (Soil Texture Unit 1067; LaMotte Co., Chestertown, MD, USA) (soil properties in Table 1). The natural vegetation is composed of tropical rainforest of the Guineo-Congolian type. The experiment was conducted for four consecutive maize growing seasons: September–December 2012; March–August 2013; September–December 2013; March–August 2014. The seasons are henceforth referred to as Short Rains 2012 (SR2012); Long Rains 2013 (LR2013); Short Rains 2013 (SR2013) and Long Rains 2014 (LR2014), respectively.

Table 1

Physical-chemical properties of the soil (0–0.2 m) and the amendments used in the field trial in western Kenya (n = 6 replicates for soil; triplicate measurements for amendments; means with standard errors in brackets).

Property	Biochar		Soil		Green manure (<i>T. diversifolia</i>)		
					Property		
pH	6.3	(0.1)	6.0	(0.1)	N (mg kg ⁻¹)	21.5	(0.5)
C (g kg ⁻¹)	868	(11)	23.3	(0.1)	P (mg kg ⁻¹)	2.3	(0.1)
N (g kg ⁻¹)	27.0	(0.9)	21.0	(0.9)	K (mg kg ⁻¹)	43.2	(1.2)
P (mg kg ⁻¹)	135	(3.7)	9.30	(0.2)	Ca (mg kg ⁻¹)	13.6	(0.2)
K (mg kg ⁻¹)	1490	(14)	223	(10)	Na (mg kg ⁻¹)	72.7	(0.9)
Ca (mg kg ⁻¹)	1920	(17)	1950	(10)	Fe (mg kg ⁻¹)	951	(10)
Na (mg kg ⁻¹)	180	(7.3)	15.9	(0.6)	Zn (mg kg ⁻¹)	89.7	(1.6)
Mg (mg kg ⁻¹)	150	(4.5)	312	(9.4)	Mg (mg kg ⁻¹)	2.6	(0.0)
Al (mg kg ⁻¹)	559	(9.8)	939	(16)	S (mg kg ⁻¹)	2.5	(0.0)
S (mg kg ⁻¹)	36.5	(1.4)	14.0	(0.8)	Mn	264	(5)
					(mg kg ⁻¹)		
Fe (mg kg ⁻¹)	164	(5.7)	67.2	(1.6)	Cu (mg kg ⁻¹)	11.0	(0.2)
Zn (mg kg ⁻¹)	108	(2.4)	13.5	(0.4)	B (mg kg ⁻¹)	53.2	(1.6)
Mn (mg kg ⁻¹)	188	(4.9)	782	(14)	Mo	1.3	(0.0)
					(mg kg ⁻¹)		
Cu (mg kg ⁻¹)	0.77	(0.1)	1.97	(0.1)			
B (mg kg ⁻¹)	1.07	(0.0)	0.33	(0.0)			
C.E.C (meq 100 g ⁻¹)	18.2	(0.6)	16.2	(0.5)			
EC (S mm ⁻¹)	196	(6.5)	88.0	(1.2)			
Silt (%)	nd		17.5	(0.3)			
Sand (%)	nd		10.7	(0.4)			
Clay (%)	nd		71.6	(2.0)			

nd = not determined.

2.2. Biochar and green manure

Biochar was produced from eucalyptus wood by chopping and grinding to pass through a 2-mm sieve. The ground material was pyrolyzed to a maximum temperature of 550 °C using a thermostat-regulated kiln with continuous agitation to provide homogeneous charring conditions and retained at this temperature for one hour before cooling to room temperature. Green manure from *T. diversifolia* was prepared by cutting leaves from the field and chopping them into 50-mm lengths, air-drying and grinding to pass through a 2-mm sieve before field application. The physical and chemical characteristics of the soil were analyzed following the same procedures as in Fungo et al. (2014) and are presented in Table 1.

2.3. Experimental design

The experiment was laid out in a randomized complete block design with three replicates. Treatments included the following: two levels of biochar (0 and 2.5 t ha⁻¹); three levels of *T. diversifolia* green manure (0, 2.5 and 5 t ha⁻¹); and two levels of urea application (0 and 120 kg N ha⁻¹) in a full factorial design (Table 2). Treatments were indicative of the range of conventional management practices of many small-holder farmers in integrated soil fertility management systems. Each treatment was established in 2 × 2-m plot separated by a one meter distance within and between rows.

2.4. Management of experiment

Precipitation and air temperature were monitored throughout the experiment with the help of a weather station on site. Application of biochar was done only once at the start of the first season in October 2012. The same amounts of green manure (2.5 or 5.0 t ha⁻¹), were applied to each plot once at the start of each season (four applications in total). Mineral N (Urea; 261 kg ha⁻¹) was applied in two splits at a total of 120 kg N ha⁻¹ per season; 40% at planting and 60% at 30 days after planting. Due to the inherently low fertility of the soil, 30 kg ha⁻¹

Table 2

Experimental treatments for determining the effect of biochar, *T. diversifolia* green manure and urea on fluxes of CO₂, CH₄ and N₂O in a maize field in western Kenya. Biochar was applied only once at the start of the experiment while urea and tithonia were applied every season for four consecutive seasons.

Treatment	Biochar		<i>T. diversifolia</i>		Mineral N (Urea)	
	Rate (t ha ⁻¹)	Code	Rate (t ha ⁻¹)	Code	Rate (kg N ha ⁻¹)	Code
1 (B ₀ T ₀ U ₀)(Control)	0	B0	0.0	T0	0	U0
2 (B ₀ T _{2.5} U ₀)	0	B0	2.5	T2.5	0	U0
3 (B ₀ T ₅ U ₀)	0	B0	5.0	T5	0	U0
4 (B ₀ T ₀ U ₁₂₀)	0	B0	0.0	T0	120	U120
5 (B ₀ T _{2.5} U ₁₂₀)	0	B0	2.5	T2.5	120	U120
6 (B ₀ T ₅ U ₁₂₀)	0	B0	5.0	T5	120	U120
7 (B _{2.5} T ₀ U ₀)	2.5	B2.5	0.0	T0	0	U0
8 (B _{2.5} T _{2.5} U ₀)	2.5	B2.5	2.5	T2.5	0	U0
9 (B _{2.5} T ₅ U ₀)	2.5	B2.5	5.0	T5	0	U0
10 (B _{2.5} T ₀ U ₁₂₀)	2.5	B2.5	0.0	T0	120	U120
11 (B _{2.5} T _{2.5} U ₁₂₀)	2.5	B2.5	2.5	T2.5	120	U120
12 (B _{2.5} T ₅ U ₁₂₀)	2.5	B2.5	5.0	T5	120	U120

of P as Triple Super Phosphate (TSP) (55 kg P₂O₅ ha⁻¹) and 30 kg ha⁻¹ of K as Muriate of Potash (MoP) (45 kg K₂O ha⁻¹) were applied to each plot at the start of each season. The amendments were applied by broadcasting on the soil surface by hand and immediately incorporating into the 0.1 m top soil. In plots where the combinations were applied, biochar was applied first, followed by Tithonia and then urea. Two seeds of a maize cultivar HB 513 were planted at the start of every season at a spacing of 0.25 m within and 0.5 m between rows (40 plants per plot). Weeding was done at 30 and 50 days after planting using a hand hoe. Thinning was done during the first weeding to retain one plant per planting hole.

2.5. Gas measurements

Measurements of CO₂, CH₄ and N₂O fluxes were conducted using a static closed chamber method (Nefel et al., 2006; Morris et al., 2013). The chamber consisted of a PVC tube (diameter = 0.3 m; height = 0.15 m) transversely divided into two parts to make a base (0.05 m) and a cover (0.1 m). The base was driven into the soil to reach ~0.02 m below the soil surface. To ensure air-tight conditions, a rubber ring was placed between the base and the cover. A photo-acoustic infrared field gas monitor (INNOVA 1402, Lumasense Technologies A/S, Ballerup, Denmark) was used to determine the gas concentrations. The accuracy and precision of standard gas concentration measurements with Photo Acoustic Spectroscopy (PAS) and Gas Chromatography (GC) have been shown to be comparable (Jung et al., 2012; Zhao et al., 2012; Iqbal et al., 2013). The gas monitor was connected to the chamber by two 0.7 m-long teflon tubes as gas inlet and outlet. Inside the cuvette, air humidity and temperature were monitored by a digital thermo-hygrometer (PCE-313 A, Paper-Consult Engineering Group, Meschede, Germany) attached to the cover from the outside while only the sensor reached inside the chamber through a rubber screw connector. Two chambers, marked “1” and “2”, were set up in each plot. All chambers marked “1” in all the treatments were sampled on day 1 and those marked “2” were sampled the following day. The values for chamber 1 and 2 for each plot were then averaged. For each gas sampling event INNOVA recorded four measurements at 2-min intervals after closing the chamber. Flux measurements were conducted weekly except during dry periods where bi-monthly measurements were taken. Fluxes are generally constant during dry, low moisture soil conditions. A total of 53 data points were obtained and used in the analysis.

2.6. Harvesting and yield determination

Above ground biomass was considered as the sum of stover, cobs and grain. Corn ears from each plot were removed from their shucks 120 days after planting, bagged, and dried for six days in a shed. Stover in each plot was cut ~0.02 m above the soil surface, weighed and dried in a shed for six days. All above ground residues from the plot were never returned to the plot in order to determine how much of the SOC could be credited to above ground vs. below ground inputs. After drying, ears were mechanically shelled, and cob and kernel biomass was determined for each plot. Moisture content in the samples was determined by taking biomass of five fresh randomly selected plants to the oven for 48 h at 60 °C. Soil samples were taken from a 0–0.2 m depth at the beginning and end of the experimental period. For the soil samples, total organic carbon (TOC) was determined by the dry combustion method (Elementar Vario EL, Hanau, Germany) and assumed that TOC = SOC since these acid soils have negligible amounts of carbonates. The available phosphate content was determined using the Lancaster method (RDA, 1988). Exchangeable cations were estimated using Inductively Coupled Plasma Spectroscopy (OPTIMA 4300DV, Perkin Elmer, USA) after extraction with 1N ammonium acetate (pH 7.0) using a soil:water ratio of 1:20 w/v.

2.7. Data analysis

2.7.1. Data management

Cumulative gas fluxes were obtained by calculating the area under the flux-time curve and summing the results while assuming linear changes in measurements between time intervals. Global Warming Potential (GWP, the sum of cumulative gas emissions of CH₄ and N₂O, multiplied by the radiative forcing factor of each gas 25 and 298, respectively, for a time horizon of 100 years) (USEPA, 2007) calculated using the following equation:

$$\text{GWP} = 25 \times (\text{E-CH}_4) + 298 \times (\text{E-N}_2\text{O})$$

Where, GWP is the emission in CO₂-equivalents per hectare, E-CH₄ and E-N₂O are the emissions of CH₄ and N₂O per hectare during a given year, respectively. Emissions Intensity (EI) was then obtained by dividing the cumulative CO₂-equivalent of the gas fluxes by the cumulative grain yield of each treatment over the experimental period.

The cumulative emission for each treatment was derived using a linear trapezoidal rule with sampling dates as the time intervals. For seasonal comparisons, the cumulative flux was restricted to the 120 days between planting and harvesting. Changes in SOC stock were calculated as the difference between values at the beginning and end of the experiment, after subtracting the C addition from biochar and *T. diversifolia*.

2.7.2. Statistical analysis

Differences in the net EI for each treatment were calculated as the difference between the treatment value and that of the control. Data for CO₂ was normally distributed and did not require transformation but CH₄ and N₂O data were natural log-transformed before ANOVA. Treatment effects and their interaction were examined with repeated-measures ANOVA using comparisons with seasons. A fixed-effect model in Stata 12 (StataCorp LP, 4905 Lakeway Drive, College Station, Texas 77845 USA) was used with a nested design with biochar and Tithonia within urea. Post hoc separation of means was done using Least Significant Difference (LSD) at a 5% level of significance using the Stata 12 statistical software.

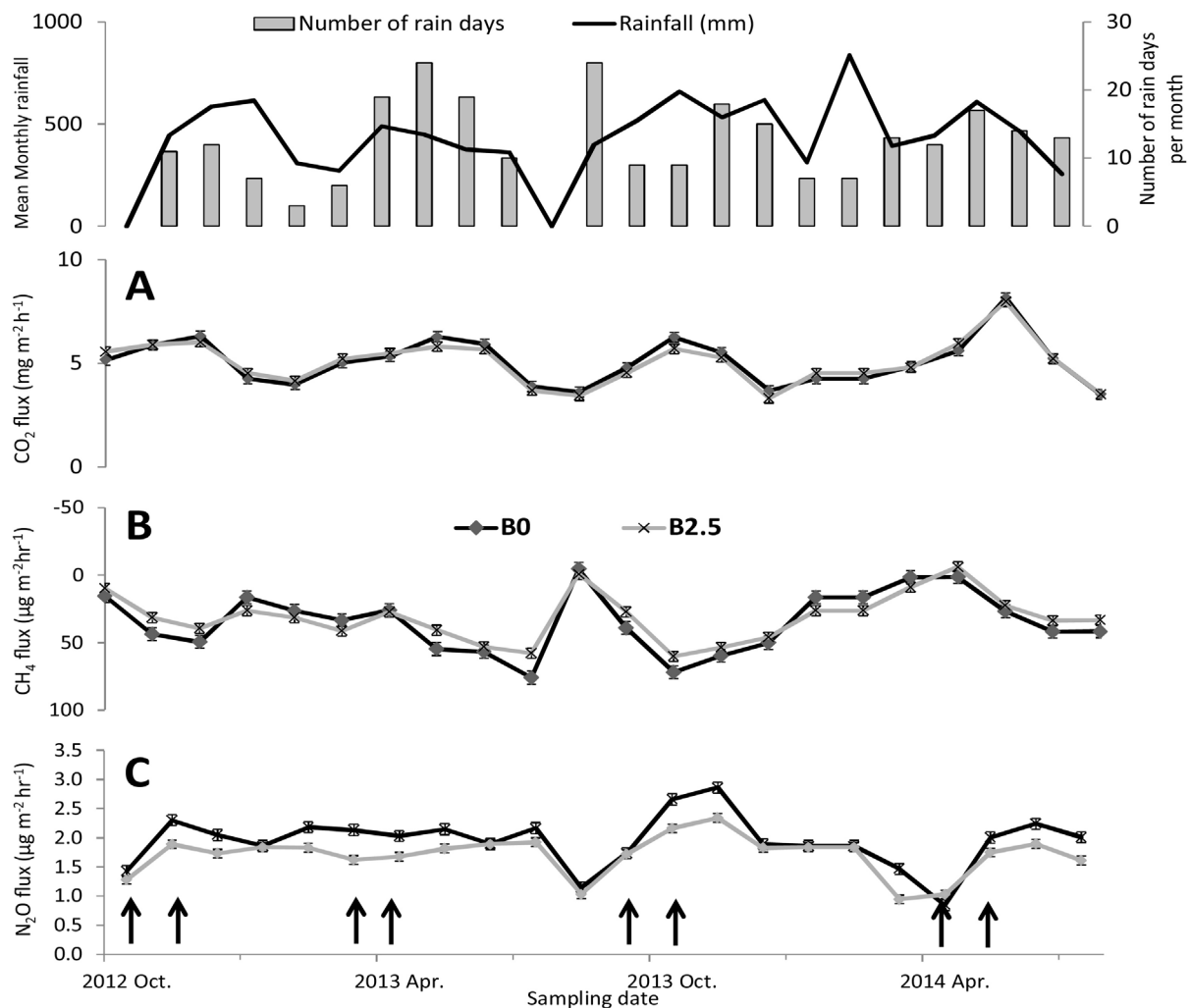


Fig. 1. Weather patterns (top graph), CO₂ (A), CH₄ (B) and N₂O (C) fluxes during four seasons of growing maize in western Kenya after amendment with mineral fertilizer (urea, 120 kg N ha⁻¹) and green manure (*T. diversifolia*, 2.5 and 5.0 t ha⁻¹) and biochar (0 or 2.5 t ha⁻¹). Error bars are standard errors. Dates on the x-axis indicate the planting time. n = 3. Arrows at the x-axis show the dates when urea was applied.

3. Results

3.1. Dynamics of GHG fluxes

3.1.1. Daily dynamics

The mean daily fluxes of CO₂, CH₄ and N₂O were 5.4 mg m⁻² h⁻¹, 39 μg m⁻² h⁻¹ and 1.96 μg m⁻² h⁻¹, respectively (Fig. 1). Emissions closely followed weather patterns with higher CO₂ and N₂O emissions as well as lower CH₄ uptake during the wet seasons. During the dry seasons, fluxes of CH₄ and N₂O were generally below average. Variability in daily fluxes, as expressed by the coefficient of variation (CV), was generally higher for CH₄ (60%) compared to CO₂ and N₂O (21%). Biochar effects were not observable for daily CO₂ measurements, but its effect was observed for CH₄ and N₂O, where it generally reduced the CH₄ sink capacity of the soil and reduced N₂O emission.

3.1.2. Seasonal dynamics

Seasonal cumulative CO₂ emission increased by 17% (Fig. 2A) throughout the four seasons. CH₄ uptake was reduced by 17% (Fig. 2B) in biochar-amended compared to control plots, and this reduction was observed in all the four seasons. The decrease in CH₄ uptake due to biochar was maintained in all the seasons, but there was no significant difference in CH₄ uptake among the seasons. Among all the seasons, LR2013 experienced the highest cumulative uptake of CH₄. Consistent 15% reduction of N₂O emissions were observed due to biochar

additions, irrespective of season (Fig. 2C). Similar to CH₄, LR2013 experienced the highest cumulative emission of N₂O. Emission of N₂O in this season was higher (P = 0.03) than those of all the other seasons likely because the rains were highest in this season.

3.1.3. Annual GHG emissions

All the three amendments, except urea (which had no effect on CH₄), affected each of the GHGs (Table 3). Annual flux of CO₂ was increased by all amendments (Table 3).

The increase in CO₂ emissions ranged from 6 to 33%. The highest increase was observed where both *T. diversifolia* and urea were present (P = 0.02). On average, amendments of biochar, tithonia or urea increased CO₂ emissions by 10% (P = 0.034). No interaction between either biochar and *T. diversifolia* or biochar and urea was observed (P > 0.05). Also, no effect of increasing *T. diversifolia* from 2.5 to 5.0 t ha⁻¹ in terms of CO₂ emissions was observed.

Both biochar and *T. diversifolia* reduced the soil uptake of CH₄, while urea had no effect. Reduction in soil CH₄ uptake ranged from 7 to 59% (Table 4). There was no interaction between amendments, and the level of *T. diversifolia* additions on CH₄ uptake between the 2 and 4 t ha⁻¹ application rates. The largest reduction in CH₄ uptake was observed when biochar, tithonia and urea were added together at the highest quantities. CH₄ uptake decreased progressively as more amendments were added.

Cumulative N₂O emissions progressively decreased by up to 42%

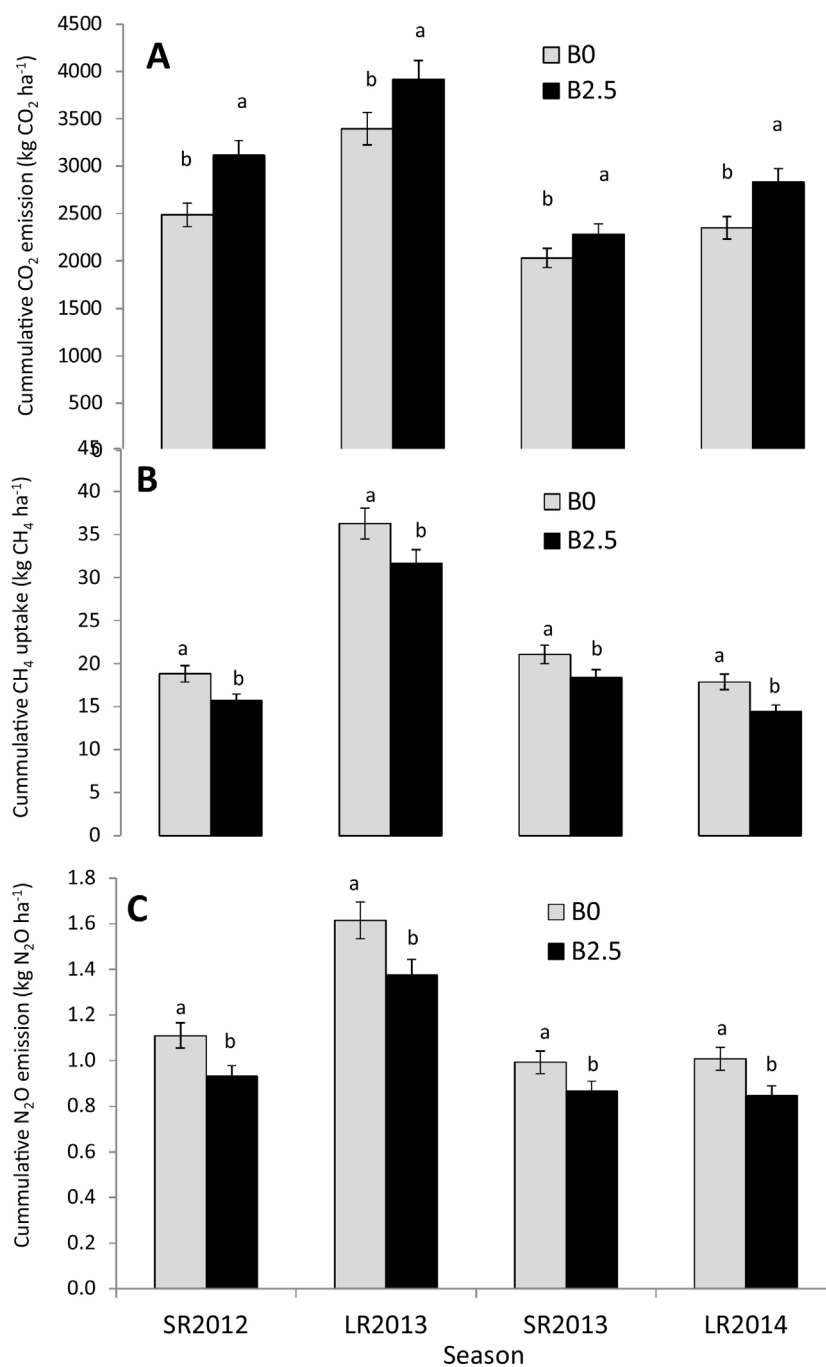


Fig. 2. Seasonal cumulative CO₂, (A) emission, CH₄ uptake (B), and N₂O emission (C) fluxes during four-season maize trial in western Kenya after amendment with mineral fertilizer (urea) and green manure (*T. diversifolia*) and biochar. Within each cluster, bars with different letters are significantly different. Error bars are standard errors, n = 3.

(mean \pm SE of $19 \pm 4.1\%$) where biochar was amended but did not change where *T. diversifolia* or urea were added on their own compared to an un-amended control (Table 4). Biochar-induced decreases in N₂O emission were greatest (42%) where *T. diversifolia* and urea were both present compared to when they were separately applied. N₂O emissions with biochar + urea additions were significantly lower than those with urea alone (Table 4). The interaction between biochar and *T. diversifolia* was significant only at the higher level of *T. diversifolia*.

3.2. Grain yield

Maize grain yields ranged from 12.6 to 19.8 t ha⁻¹ yr⁻¹ (mean = 16.4 t ha⁻¹ yr⁻¹), corresponding to a decrease of 15% and increase of 34%, respectively, compared to the control. Decreases in

maize grain yield were observed where biochar was applied either alone or in combination with low amounts of *T. diversifolia* (B_{2.5}T₀U₀, -15%, and B_{2.5}T_{2.5}U₀, -14%, respectively). High amounts of *T. diversifolia* (T₅) significantly increased yields by 9%. The greatest increase in maize grain yield was observed where biochar was combined with urea (B_{2.5}T₀U₁₂₀, 34%). This was followed by yields where high amounts of *T. diversifolia* (5 t ha⁻¹) were jointly applied with urea (treatment B₀T₅U₁₂₀ at 29%) (Table 4). Grain yield of the combination of the three amendments was higher than either biochar or *T. diversifolia* applied alone (Table 4). Grain yield comprised of 35% (SD = 0.03) of total above ground biomass and this proportion was comparable for all treatments.

Table 3

Three-way ANOVA for the effects of biochar, *T. diversifolia* green manure and urea on fluxes of CO₂, CH₄ N₂O, GWP, maize grain yield and EI in maize field in western Kenya. P-values in bold show means that were significantly different $p = 0.05$, $n = 3$.

Source of variation	CO ₂		CH ₄		N ₂ O		GWP		Grain Yield		EI	
	F	P	F	P	F	P	F	P	F	P	F	P
Biochar	6.11	0.021	6.29	0.026	5.65	0.011	12.5	0.007	10.5	0.031	10.6	0.010
<i>T. diversifolia</i>	12.25	0.000	3.46	0.047	5.02	0.015	10.4	0.025	1.2	0.96	12.0	0.017
Urea	8.62	0.007	3.01	0.095	8.43	0.006	8.0	0.045	11.4	0.034	18.0	0.000
Biochar x <i>T. diversifolia</i>	1.27	0.271	1.13	0.299	0.88	0.358	16.7	0.000	10.8	0.031	9.3	0.021
Biochar x Urea	1.98	0.160	1.13	0.339	4.96	0.037	11.2	0.016	10.1	0.038	15.2	0.000
<i>T. diversifolia</i> x Urea	1.67	0.208	1.04	0.370	1.27	0.098	0.0	0.095	0.0	0.095	11.2	0.018
Biochar x <i>T. diversifolia</i> x Urea	1.36	0.276	1.61	0.220	1.17	0.325	10.8	0.031	4.9	0.042	4.3	0.047

Bold numbers show significant effect at 95% level of confidence.

3.3. Global warming potential (GWP)

Biochar when applied alone caused a significantly lower GWP (8% or 6.3 ± 0.01 t CO₂-eq ha⁻¹ yr⁻¹) compared to the control (Table 4). GWP caused by additions of *T. diversifolia* and urea compared to no additions (control) significantly increased by 32% and 12% corresponding to an increase by 7.7 ± 0.8 and 6.5 ± 0.6 t CO₂-e ha⁻¹ yr⁻¹, respectively. *T. diversifolia* or urea additions resulted in significantly higher yield compared to the control, but were not significantly different from each other. Grain yield was significantly higher with the sole biochar addition. The interactive effect between biochar and urea increased yield significantly but no difference in yield was observed where tithonia and urea were applied together or where biochar was applied with tithonia. In a combined application of biochar, urea and *T. diversifolia*, doubling the amounts of *T. diversifolia* halved the GWP. The difference between GWP as a result of biochar + *T. diversifolia* and biochar + urea additions was significantly lower than the difference in the respective sole applications of *T. diversifolia* versus urea.

3.4. Emissions intensity (EI)

Biochar and tithonia applied alone caused significantly higher EI compared to the control, but EI as a result of urea additions was not significantly different from that of the control. There was no interactive effect between tithonia and urea application with respect to EI. Except for the interaction of 2.5 t biochar ha⁻¹ and urea (B_{2.5}T₀U₁₂₀ in Table 4), all the other treatments showed significantly higher EI than the control ($p = 0.035$), with increases ranging from 8 to 39%. Apart from the control, the highest EI corresponded with additions of only *T. diversifolia* green manure (B₀T_{2.5}U₀ and B₀T₅U₀) or urea additions (B₀T₀U₁₂₀). The lowest EI was observed where biochar and urea were added without or with high *T. diversifolia*. As *T. diversifolia* green

manure additions increased, the EI correspondingly decreased but only in the presence of biochar. The decrease in EI with greater additions of *T. diversifolia* corresponded to increases in CO₂ emission by 14% and decreases in N₂O emission by 16% (Table 4).

3.5. Changes in SOC stocks

Amendments increased SOC stocks in the range of 0.7–7.1%, corresponding to 0.1–1.2 t C ha⁻¹ year⁻¹ (mean \pm SE of 0.8 ± 0.09 t C ha⁻¹ year⁻¹). Except for the control (where no amendment was applied), which showed no significant difference in SOC over time, all the other treatments showed gains in SOC (Fig. 3). The smallest increment (0.2 ± 0.01 t C ha⁻¹ yr⁻¹) was observed without any additions while the highest increase (1.2 ± 0.04 t C ha⁻¹ yr⁻¹) was recorded where urea was applied with *T. diversifolia* irrespective of biochar additions. The increase in SOC stocks due to biochar or *T. diversifolia* additions was comparable (0.7 ± 0.02 t C ha⁻¹ yr⁻¹, or 2.8%) to the control. Soil OC stocks in response to additions of 2.5 t ha⁻¹ of tithonia increased by 27% compared to the control, and was comparable to additions of 5 t ha⁻¹ of tithonia. Additions of 2.5 t ha⁻¹ of tithonia together with urea increased SOC by 39% compared to the control.

4. Discussion

4.1. Dynamics of GHGs

Stimulation of CO₂ emissions following biochar addition observed in this study (Figs. 1A and 2A) concurs with what has been observed in previous studies and has been attributed to the supply of easily mineralizable C and improvement in soil physical properties for microbial activity (Lehmann et al., 2011; Sagrilo et al., 2014) and increased root respiration (Major et al., 2010). However, reduction in

Table 4

Annual grain yield (two seasons), CH₄ and N₂O emissions, GWP (calculated only from CH₄ and N₂O) and EI of maize production over four consecutive seasons under biochar, *T. diversifolia* and urea amendment. Means (\pm S.E.) in the same column followed by different letters are significantly different at $p < 0.05$, $n = 3$.

Treatment	CO ₂ (t ha ⁻¹ yr ⁻¹)	CH ₄ uptake (t ha ⁻¹ yr ⁻¹)	N ₂ O emission (t ha ⁻¹ yr ⁻¹)	GWP (t CO ₂ -eq ha ⁻¹ yr ⁻¹)	Maize Grain yield (t ha ⁻¹ yr ⁻¹)	EI (t CO ₂ -eq t ⁻¹ grain yr ⁻¹)
1 (B ₀ T ₀ U ₀)	3.7 (\pm 0.6)d	0.061 (\pm 0.017)a	0.003 (\pm 0.009)a	0.67 (\pm 0.5)a	14.8 (\pm 1.0)f	0.05 (\pm 0.01)a
2 (B ₀ T _{2.5} U ₀)	4.9 (\pm 0.1)a	0.057 (\pm 0.023)b	0.003 (\pm 0.002)a	0.57 (\pm 0.34)b	15.1 (\pm 0.6)f	0.04 (\pm 0.03)b
3 (B ₀ T ₅ U ₀)	4.8 (\pm 0.3)a	0.058 (\pm 0.006)b	0.003 (\pm 0.006)a	0.59 (\pm 0.18)b	14.9 (\pm 0.1)f	0.04 (\pm 0.02)bc
4 (B ₀ T ₀ U ₁₂₀)	4.1 (\pm 0.6)ca	0.057 (\pm 0.007)b	0.003 (\pm 0.004)a	0.60 (\pm 0.11)b	15.9 (\pm 0.8)e	0.04 (\pm 0.02)c
5 (B ₀ T _{2.5} U ₁₂₀)	5.0 (\pm 0.1)a	0.052 (\pm 0.009)bc	0.003 (\pm 0.007)a	0.47 (\pm 0.04)c	18.4 (\pm 1.4)c	0.03 (\pm 0.03)c
6 (B ₀ T ₅ U ₁₂₀)	4.9 (\pm 0.1)c	0.053 (\pm 0.010)bc	0.002 (\pm 0.009)b	0.59 (\pm 0.22)b	19.1 (\pm 0.1)b	0.03 (\pm 0.05)d
7 (B _{2.5} T ₀ U ₀)	4.0 (\pm 0.6)b	0.052 (\pm 0.018)bc	0.002 (\pm 0.005)b	0.59 (\pm 0.32)b	12.8 (\pm 1.8)g	0.05 (\pm 0.01)a
8 (B _{2.5} T _{2.5} U ₀)	4.4 (\pm 0.3)a	0.050 (\pm 0.006)c	0.003 (\pm 0.005)a	0.42 (\pm 0.06)d	12.6 (\pm 1.8)g	0.03 (\pm 0.06)d
9 (B _{2.5} T ₅ U ₀)	5.0 (\pm 0.1)b	0.046 (\pm 0.029)d	0.002 (\pm 0.001)b	0.47 (\pm 0.18)d	17.4 (\pm 1.6)d	0.03 (\pm 0.01)e
10 (B _{2.5} T ₀ U ₁₂₀)	4.6 (\pm 0.6)b	0.047 (\pm 0.014)d	0.002 (\pm 0.007)b	0.44 (\pm 0.36)cd	19.8 (\pm 1.0)a	0.02 (\pm 0.03)f
11 (B _{2.5} T _{2.5} U ₁₂₀)	4.6 (\pm 0.9)b	0.055 (\pm 0.025)b	0.003 (\pm 0.001)ab	0.58 (\pm 0.34)b	17.0 (\pm 3.2)d	0.03 (\pm 0.01)cd
12 (B _{2.5} T ₅ U ₁₂₀)	4.4 (\pm 0.1)b	0.034 (\pm 0.020)e	0.002 (\pm 0.007)c	0.26 (\pm 0.31)e	16.9 (\pm 0.8)d	0.02 (\pm 0.03)g

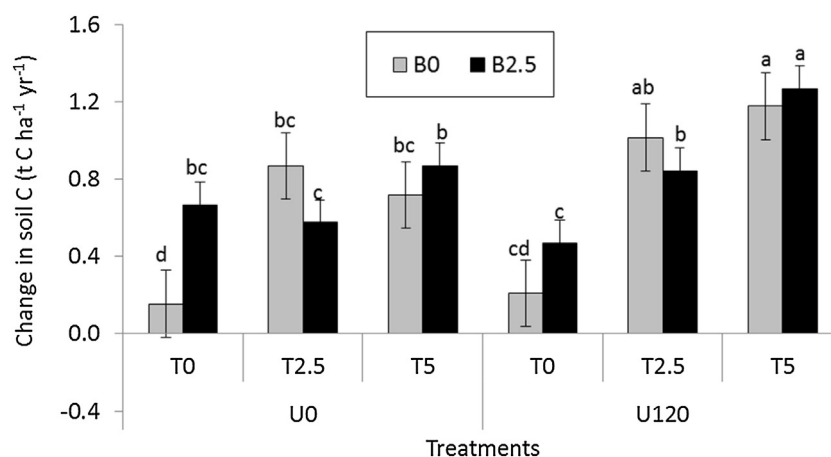


Fig. 3. Annual changes in soil organic C stock (top 0.15 m) under biochar (B in t ha⁻¹), *T. diversifolia* (T in t ha⁻¹) and urea (U in kg N ha⁻¹) amendments compared to the control. Means (\pm S.E.) followed by different letters are significantly different at $p < 0.05$, $n = 3$.

CO₂ emission after biochar addition has also been reported in incubations without plants, and is usually associated with limited N supply due to N immobilization by amended biochar (Laird et al., 2010; Wang et al., 2015). In our case, the amount of biochar was relatively low (0.2% compared to the average 2% w/w in most studies) and could not immobilize significant amounts of soil N while crop growth significantly increased, hence the consistent increase in CO₂ (that included root respiration) with all biochar plots. The lack of an interactive effect between biochar and tithonia as well as urea and tithonia with respect to CO₂ emission (Table 3) was possibly due to the fact that sufficient C was supplied by biochar or *T. diversifolia* making further inputs temporarily unnecessary for microbial use. Contrary to our findings, Rogovska et al. (2011) found reduced CO₂ emission after 500 days of incubation for a green manure-biochar mixture. The authors attributed this observation to the fact that biochar stabilized green manure C by influencing biochemical recalcitrance or physical protection of green manure C (Krull et al., 2003), or that green manure additions reduced the ability of biochar to enhance mineralization of soil organic matter. Our results did not show interactions between biochar and green manure possibly because the mixing of the two amendments was not as uniform as it was in the incubation of Rogovska et al. (2011). Whereas biochar largely remains unchanged in space, tithonia and urea may, after incorporation into the soil, leach to lower soil layers. The subsequent reactions of these amendments may thus remain independent. Some studies on placement of amendments may elucidate this notion. It is important to note that CO₂ can be produced by both enhance mineralization of soil organic matter or biochar itself (Wardle et al., 2008). Our study measured only soil-atmosphere fluxes of CO₂, and could not attribute the increase to a particular CO₂ production process. Nonetheless, the fact that there was an increase in SOC with biochar-tithonia additions (Fig. 3) suggest an overall net positive balance towards soil C sequestration. Future studies should estimate the net effect of biochar application on soil respiration and biochar mineralization. Life cycle assessment to determine C sequestration including CO₂ emitted during biochar production will also be relevant.

The upland soils are generally identified as sinks for CH₄ (Chan et al., 2001) but additions of biochar and *T. diversifolia* in this experiment reduced the sink capacity (Figs. 1B and 2B). This can likely be attributed to the supply of easily mineralizable C that substitutes for the C oxidized by methanotrophic bacteria (Knoblauch et al., 2008; Zhang et al., 2010). The greatest reduction in CH₄ uptake, coinciding with urea and high *T. diversifolia* input (Table 4), could be attributed to the availability of NH₄⁺-N from urea, which is known to inhibit CH₄ oxidation (Chan and Parkin, 2001; Suwanwaree and Robertson, 2005). Whereas some studies have shown increased uptake of CH₄ with biochar (Liu et al., 2011; Schimmelpfennig et al., 2014), several have

also shown decreases (Spokas et al., 2013; Singla and Inubushi, 2014). This suggests the complexity and variety of factors affecting CH₄ fluxes in biochar-amended soils. The persistence of biochar effects on fluxes over the two years of observation (Fig. 2B) could be related to improvements in physical properties leading to better drainage and adsorption of dissolved organic carbon (Singh et al., 2012; Zhang et al., 2015) rather than N immobilization discussed above that is likely to be a transient phenomenon.

The consistent reduction in N₂O with biochar amendments (Figs. 1C and 2C) is similar to previous studies (Rondon et al., 2005; Xiang et al., 2015). The reduction in N₂O emissions in the present study is lower compared to other studies (such as Zhang et al., 2012a,b; Zhang et al., 2015) partly because the amounts of added biochar were relatively low (2.5 t ha⁻¹ compared with 20–40 t ha⁻¹ in all the above studies). N₂O reduction could have been due to stimulation of microorganisms that can degrade more persistent SOM (Nelissen et al., 2012), resulting in more reactive surfaces. It is also plausible that adsorption of NH₄⁺ (Taghizadeh-Toosi et al., 2012) and other potential allelochemical inhibitors of microbial metabolic pathways, such as monoterpenes and various polyphenolic compounds that are inhibitory to nitrification, could have played a role (Ball et al., 2011). That the interaction between biochar and urea (Table 3) resulted in reduced N₂O emission could probably be due to a direct reaction between the biochar surfaces and N₂O. Thus, the N₂O emission produced from urea is countered by the biochar. Cayuela et al. (2014) have suggested the catalytic activity of biochar as capable of enhancing the reduction of N₂O to N₂. Direct molecular-level interaction between N₂O and reducing agents has been reported (Hitoshi et al., 2002) and Fungo et al. (2014) observed that steam activation of biochar explained 56% of reduction in N₂O emission from the same soil. It is plausible that such a reaction could be taking place in biochar amended soils.

The notion that a stimulatory effect of biochar on soil and/or plant respiration levels off during the first years after application (Spokas, 2013) was not seen within the two years of the experiment, as we observed consistent differences in emissions and above ground biomass production during all four cropping seasons. Several studies (e.g. Kimetu et al., 2008; Major et al., 2010; Koide et al., 2014) have shown that biochar contents remain in soil and decreased only between 0.3 and 6% of the amount applied over a period of three years. Whereas some studies (Nelissen et al., 2014) found short-lived effects of biochar (Spokas, 2013), the two-year effects observed in our study support Lentz et al. (2014), who observed a persistent effect over a 3-year period. Lentz et al. (2014) related this multi-year effect to biochar's physical porosity and chemical binding capacity. The contrasting results in the literature indicate a need for longer-term (> 5 years) studies to test this hypothesis.

Results in Fig. 2 show that biochar effects on CH₄ uptake and N₂O emissions can persist over two years under field conditions. This could be attributed to the fact that non-aromatic organic materials such as sugars and fats in the pores of biochar (Mukherjee et al., 2015; Koide et al., 2014) are susceptible to mineralization over time (Kim et al., 2011). According to Lentz et al. (2014), the disappearance of these organics with soil aging likely (i) increased porosity and surface area akin to the effect that activation has on charcoal (Fungo et al., 2014); and (ii) increased negatively charged sites on biochar (Cheng et al., 2008). The charged sites can bind NH₄, making inorganic N only temporarily unavailable for microorganisms and leaching (Dempster et al., 2012), which can later be readily released (Wang et al., 2015). These mechanisms could have been responsible for observed persistence of biochar effect observed in this study. A change in pore size distribution in the biochar-amended soil could have influenced bacterial community composition (Lentz et al., 2014) that could conceivably also affect GHG emissions. Feng et al. (2012) reported decreased ratios of methanogenic to methanotrophic microorganisms that accounted for the decrease in CH₄ emission from paddy soils. Banerjee et al. (2016) found that moisture affected microbial activity, transcription, composition and ultimately, N₂O emissions. The reduced N₂O mitigation effect observed by Spokas (2013) after 3 years of biochar aging could be related to the relatively large particle size (8–40 mm) of the biochar used.

4.2. Changes in soil C content

As observed in our study, a previous study carried out in the same area (Kimetu et al., 2008) found that application of biochar increased TOC contents by 6.8 times after adding biochar compared to *T. diversifolia* green manure. In fact, the authors observed that biochar not only remained to a greater extent unchanged, but could be protected by aggregation to a greater degree in soil in the long term. The relatively fine texture of the tested soils means that the soil may allow for more SOC protection by a combination of physical occlusion and organo-mineral interactions.

Chivenge et al. (2011a) found that in clayey soils, the addition of low quality organic resources (such as maize stover) resulted in greater stabilization of SOC and SON in the macro-aggregates, while addition of N-fertilizers enhances their decomposition and faster aggregate turnover leading to less accumulation of SOC and SON. Similar to our results, Chivenge et al. (2011b) also found no interaction between *T. diversifolia* with N fertilizers on similar soils with respect to changes in SOC. This suggests that microbial choice of the N source is independent of the source or that the utilization of freshly applied organic matter (C source) is consumed at the same rate as mineral N (source of N). Verchot et al. (2011) observed that most of the newly added SOC gains from green manures ended up in the coarser aggregates, and is therefore subject to turnover and loss in the event that OM inputs decline. However, using soils from the same experiment, Fungo et al. (2017) found that biochar is stored predominantly as free particulate OC in the silt and clay fraction and promoted a movement of native SOC from larger-size aggregates to the smaller-sized fraction. The increase in C stocks when *T. diversifolia* was added but no additive effect of *T. diversifolia* + biochar may be attributed to the fact that there is a balance between increased mineralization of SOM due to biochar addition on the one hand and an increase in SOC due to greater net primary productivity of root biomass on the other.

Our results (Fig. 3) show that even without returning harvested above ground biomass to the soil, any of the additions were still able to demonstrate SOC gains. It follows that returning maize stover and integrated soil fertility management with biochar can achieve even greater SOC sequestration as well as GHG emission objectives. This implies that soil amendment with organic a mineral resources increases net primary productivity and hence below ground biomass. This biomass can later build up soil organic matter stocks. In terms of

climate change mitigation, this is important as it demonstrates the potential for C sequestration associated with soil management.

4.3. System C balance

In relatively low fertility soils similar to the one used in this experiment, increases in biomass production in response to biochar application have been widely reported (Major et al., 2010; Li et al., 2015). The grain and biomass yield increase may most likely be attributed in the studied acid soils to increases in direct mineral additions from biochar, improvement in soil physical properties (e.g., bulk density), greater pH and reduced Al toxicity, improved cation exchange capacity, and possibly effects on soil biota that largely remain speculative. The proportional increase in biomass is comparable to other studies on highly weathered tropical soils (Major et al., 2010; Van Zwieten et al., 2010; Zhang et al., 2011).

The enhanced above ground biomass production that we observed (Table 4) could be due to improved soil physical and chemical properties resulting from biochar amendments. Olmo et al. (2014) found that grain production was correlated significantly and positively with soil moisture, EC, total N, Olsen P and available K, Cu and Zn, and negatively with soil compaction, consistent with the favorable changes in soil physico-chemical properties brought about by biochar additions. According to Vanlauwe et al. (2002) and Lentz et al. (2014), combining biochar with green manure more effectively utilized the two soil amendments, as it eliminates potential plant growth reductions caused by N immobilization after biochar additions and maximized green manure net N mineralization potential.

The increase in net primary productivity due to soil amendments contributes to atmospheric C capture into terrestrial C (above ground and soil organic C). However, it will be necessary for future studies to investigate other types of green manures to better understand limitations over longer periods of time. Application rates used in several other trials are unrealistic for most farming systems. Despite the relatively low application rates of both biochar and *T. diversifolia* used in this study compared to those of previous studies, we demonstrate that significant ecosystem C gains are practically possible in low fertility soils even with relatively moderate amounts of biochar.

The identical grain yields with additions of 2.5 t ha⁻¹ biochar and 5 t ha⁻¹ tithonia irrespective of urea additions (Table 4) may imply that biochar has the potential to reduce mineral N fertilizer requirements if green manure is added. In terms of GHG emissions, reduced application of mineral fertilizer is one way to reduce the impact of agriculture on climate change. This includes the avoided emissions due to fertilizer production. What remains to be determined is the trade-off between biochar production and fertilizer manufacture in terms of both cost and emissions.

Based on a yield-normalized comparison of our results (EI), biochar can potentially reduce overall GHG emission while improving crop yields and SOC. Nonetheless, the observed reduction in EI due to biochar additions needs to be further analyzed using a full Life Cycle Assessment (LCA) to account for the energy-related emissions needed to produce or transport the biochar among other emissions and emission reductions.

5. Conclusions and recommendations

We have shown that the inclusion of biochar in integrated management of low-fertility tropical agricultural soils can reduce GHG emissions and increase ecosystem C gains. The resultant C gains and GHG emission reduction benefits of biochar can be sustained for at least two consecutive years (four seasons) from the time of application under the environmental conditions studied here. This result points to the importance of plant responses for the GHG balance in biochar systems. Our results contribute to mid-term field studies but longer trials might be necessary to better understand the C balance, including different soil

types and cropping systems especially where stover biomass is returned to the plot. Longer field-based studies and field studies on soil that show different plant response to organic matter additions are needed to improve understanding of linkages between nutrient use efficiency and GHG mitigation.

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References

- Ball, P.N., MacKenzie, M.D., DeLuca, T.H., 2011. Wildfire and charcoal enhance nitrification and ammonium-oxidizing bacterial abundance in dry montane forest soils. *J. Environ. Qual.* 39, 1243–1253.
- Banerjee, S., Helgason, B., Wang, L., Winsley, T., Ferrari, B.C., Siciliano, S.D., 2016. Legacy effects of soil moisture on microbial community structure and N₂O emissions. *Soil Biol. Biochem.* 95, 40–50.
- Cayuela, M.L., van Zwieten, L., Singh, B.P., Jeffery, S., Roig, A., Sánchez-Monedero, M.A., 2014. Biochar's role in mitigating soil nitrous oxide emissions: a review and meta-analysis. *Agric. Ecosyst. Environ.* 191, 5–16.
- Chan, A.S.K., Parkin, T.B., 2001. Methane oxidation and production activity in soils from natural and agricultural ecosystems. *J. Environ. Qual.* 30, 1896–1903.
- Chan, A.S.K., Parkin, T.B., Forest, M., 2001. Methane oxidation and production activity in soils from natural and agricultural ecosystems. *J. Environ. Qual.* 1896–1903.
- Cheng, C., Lehmann, J., Engelhard, M.H., 2008. Natural oxidation of black carbon in soils: changes in molecular form and surface charge along a climosequence. *Geochim. Cosmochim. Acta* 72, 1598–1610. <http://dx.doi.org/10.1016/j.gca.2008.01.010>.
- Chivenge, P., Vanlauwe, B., Gentile, R., Six, J., 2011a. Comparison of organic versus mineral resource effects on short-term aggregate carbon and nitrogen dynamics in a sandy soil versus a fine textured soil. *Agr. Ecosyst. Environ.* 140, 361–371. <http://dx.doi.org/10.1016/j.agee.2010.12.004>.
- Chivenge, P., Vanlauwe, B., Gentile, R., Six, J., 2011b. Organic resource quality influences short-term aggregate dynamics and soil organic carbon and nitrogen accumulation. *Soil Biol. Biochem.* 43, 657–666. <http://dx.doi.org/10.1016/j.soilbio.2010.12.002>.
- Dempster, D.N., Gleeson, D.B., Solaiman, Z.M., Jones, D.L., Murphy, D.V., 2012. Decreased soil microbial biomass and nitrogen mineralization with eucalyptus biochar addition to a coarse textured soil. *Plant Soil* 354, 311–324. <http://dx.doi.org/10.1007/s11104-011-1067-5>.
- Feng, Y., Xu, Y., Yu, Y., Xie, Z., Lin, X., 2012. Soil Biology & Biochemistry Mechanisms of biochar decreasing methane emission from Chinese paddy soils. *Soil Biol. Biochem.* 46, 80–88.
- Fungo, B., Guereña, D., Thiongo, M., Lehmann, J., Neufeldt, H., Kalbitz, K., 2014. N₂O and CH₄ emission from soil amended with steam-activated biochar. *J. Plant Nutr. Soil Sci.* 177, 34–38. <http://dx.doi.org/10.1002/jpln.201300495>.
- Fungo, B., Lehmann, J., Kalbitz, K., Thiongo, M., Okeyo, L., Tenywa, M., Neufeldt, H., 2017. Aggregate size distribution in a biochar-amended tropical Ultisol under conventional hand-hoe tillage. *Soil Till. Res.* 165, 190–197. <http://dx.doi.org/10.1016/j.still.2016.08.012>.
- Gentile, R., Vanlauwe, B., van Kessel, C., Six, J., 2009. Managing N availability and losses by combining fertilizer-N with different quality residues in Kenya. *Agr. Ecosyst. Environ.* 131, 308–314.
- Iqbal, J., Castellano, M.J., Parkin, T.B., 2013. Evaluation of photoacoustic infrared spectroscopy for simultaneous measurement of N₂O and CO₂ gas concentrations and fluxes at the soil surface. *Global Change Biol.* 19, 327–336. <http://dx.doi.org/10.1111/gcb.12021>.
- Jeffery, S., Bezemer, T.M., Cornelissen, G., Kuypers, T.W., Lehmann, J., Mommer, L., Sohi, S.P., van de Voorde, T.F.J., Wardle, D.A., van Groenigen, J.W., 2015. The way forward in biochar research: targeting trade-offs between the potential wins. *Global Change Biol.* 7, 1–13. <http://dx.doi.org/10.1111/gcb.12132>.
- Jung, Y., Han, B., Mostafid, M.E., Chiu, P., Yazdani, R., Imhoff, P.T., 2012. Photoacoustic infrared spectroscopy for conducting gas tracer tests and measuring water saturations in landfills. *Manure Manage* 32, 297–304. <http://dx.doi.org/10.1016/j.wasman.2011.09.016>.
- Kim, E.J., Choi, S.D., Chang, Y.S., 2011. Levels and patterns of polycyclic aromatic hydrocarbons (PAHs) in soils after forest fires in South Korea. *Environ. Sci. Pollut. Res.* 18, 1508–1517. <http://dx.doi.org/10.1007/s11356-011-0515-3>.
- Kimetu, J., Lehmann, J., Ngoze, S., Mugendi, D., Kinyangi, J., Riha, S., Verchot, L., Recha, J., Pell, A., 2008. Reversibility of soil productivity decline with organic matter of differing quality along a degradation gradient. *Ecosystems* 11, 726–739. <http://dx.doi.org/10.1007/s10021-008-9154-z>.
- Knoblauch, C., Maarifat, A., Pfeiffer, E., Haefele, S.M., 2011. Degradability of black carbon and its impact on trace gas fluxes and carbon turnover in paddy soils. *Soil Biol. Biochem.* 43, 1768–1778.
- Koide, R.T., Dell, C., Drohan, P., Skinner, H., Adler, P.R., 2014. Turnover of soil carbon following addition of switchgrass-derived biochar to four soils. *Soil Sci. Soc. Am. J.* 78, 531–537. <http://dx.doi.org/10.2136/sssaj>.
- Laird, D., Fleming, P., Wang, B., Horton, R., Karlen, D., 2010. Biochar impact on nutrient leaching from a Midwestern agricultural soil. *Geoderma* 158, 436–442.
- Lentz, R.D., Ippolito, J.A., Spokas, K.A., 2014. Biochar and manure effects on net nitrogen mineralization and greenhouse gas emissions from calcareous soil under corn. *Soil Sci. Soc. Am. J.* 78, 1641–1655. <http://dx.doi.org/10.2136/sssaj2014.05.0198>.
- Li, B., Fan, C.H., Zhang, H., Chen, Z.Z., Sun, L.Y., Xiong, Z.Q., 2015. Combined effects of nitrogen fertilization and biochar on the net global warming potential, greenhouse gas intensity and net ecosystem economic budget in intensive vegetable agriculture in southeastern China. *Atmos. Environ.* 100, 10–19. <http://dx.doi.org/10.1016/j.atmosenv.2014.10.034>.
- Liu, Y., Yang, M., Wu, Y., Wang, H., Chen, Y., Wu, W., 2011. Reducing CH₄ and CO₂ emissions from waterlogged paddy soil with biochar. *J. Soils Sediments* 11, 930–939. <http://dx.doi.org/10.1007/s11368-011-0376-x>.
- Maestrini, B., Herrmann, A.M., Nannipieri, P., Schmidt, M.W.I., Abiven, S., 2014. Ryegrass-derived pyrogenic organic matter changes organic carbon and nitrogen mineralization in a temperate forest soil. *Soil Biol. Biochem.* 69, 291–301. <http://dx.doi.org/10.1016/j.soilbio.2013.11.013>.
- Major, J., Rondon, M., Molina, D., Riha, S.J., Lehmann, J., 2010. Maize yield and nutrition during 4 years after biochar application to a Colombian savanna oxisol. *Plant Soil* 333, 117–128. <http://dx.doi.org/10.1007/s11104-010-0327-0>.
- Manzoni, S., Porporato, A., 2009. Soil C and nitrogen mineralization: theory and models across scales. *Soil Biol. Biochem.* 41, 1355–1379.
- Mori, A., Hojito, M., 2011. Effect of combined application of manure and fertilizer on N₂O fluxes from a grassland soil in Nasu, Japan. *Agr. Ecosyst. Environ.* <http://dx.doi.org/10.1016/j.agee.2011.07.018>.
- Mukherjee, A., Lal, R., Zimmerman, A.R., 2015. Effects of biochar and other amendments on the physical properties and greenhouse gas emissions of an artificially degraded soil. *Sci. Total Environ.* 487, 26–36. <http://dx.doi.org/10.1016/j.scitotenv.2014.03.141>.
- Nelissen, V., Rütting, T., Huygens, D., Staelens, J., Ruysschaert, G., Boeckx, P., 2012. Maize biochars accelerate short-term soil nitrogen dynamics in a loamy sand soil. *Soil Biol. Biochem.* 55, 20–27. <http://dx.doi.org/10.1016/j.soilbio.2012.05.019>.
- Olmo, M., Albuquerque, J.A., Barrón, V., del Campillo, M.C., Gallardo, A., Fuentes, M., Villar, R., 2014. Wheat growth and yield responses to biochar addition under Mediterranean climate conditions. *Biol. Fert. Soils* 50, 1177–1187. <http://dx.doi.org/10.1007/s00374-014-0959-y>.
- RDA (Rural Development Administration, Korea), 1988. *Methods of Soil Chemical Analysis*. National Institute of Agricultural Science and Technology, RDA, Suwon (in Korean).
- Rogovska, N., Laird, D., Cruse, R., Fleming, P., Parkin, T., Meek, D., 2011. Impact of biochar on manure carbon stabilization and greenhouse gas emissions. *Soil Sci. Soc. Am. J.* 75, 871. <http://dx.doi.org/10.2136/sssaj2010.0270>.
- Rondon, M.A., Ramirez, J.A., Lehmann, J., 2005. Greenhouse gas emissions decrease with charcoal additions to tropical soils. *Proceedings of the 3rd USDA Symposium on Greenhouse Gases and Carbon Sequestration in Agriculture and Forestry* 208.
- Sagrilo, E., Jeffery, S., Hoffland, E., Kuypers, T.W., 2014. Emission of CO₂ from biochar-amended soils and implications for soil organic carbon. *Global Change Biol. Bioenergy* 7, 1294–1304. <http://dx.doi.org/10.1111/gcb.12234>.
- Schimmelpfennig, S., Müller, C., Grünhage, L., Koch, C., Kammann, C., 2014. Biochar, hydrochar and uncarbonized feedstock application to permanent grassland – effects on greenhouse gas emissions and plant growth. *Agr. Ecosyst. Environ.* 191, 39–52.
- Singla, A., Inubushi, K., 2014. Effect of biochar on CH₄ and N₂O emission from soils vegetated with paddy. *Paddy Water Environ.* 12, 239–243. <http://dx.doi.org/10.1007/s10333-013-0357-3>.
- Spokas, K.A., 2013. Impact of biochar field aging on laboratory greenhouse gas production potentials. *Global Change Biol. Bioenergy* 5, 165–176. <http://dx.doi.org/10.1111/gcb.12005>.
- Suwanwaree, P., Robertson, G.P., 2005. Methane oxidation in forest, successional, and No-till agricultural ecosystems. *Eff. Nitrogen Soil Disturb.* 1722–1729. <http://dx.doi.org/10.2136/sssaj2004.0223>.
- Taghizadeh-Toosi, A., Clough, T.J., Sherlock, R.R., Condon, L.M., 2012. A wood based low-temperature biochar captures NH₃-N generated from ruminant urine-N, retaining its bioavailability. *353*, 73–84.
- USEPA, 2007. *Inventory of USA Greenhouse Gas Emissions and Sinks: 1990–2005*. USEPA, Washington, DC.
- Van Zwieten, L., Kimber, S., Downie, A., Morris, S., Petty, S., Rust, J., Chan, K.Y., 2010. A glasshouse study on the interaction of low mineral ash biochar with nitrogen in a sandy soil. *Austr. J. Soil Res.* 48, 569. <http://dx.doi.org/10.1071/SR10003>.
- Vanlauwe, B., Diels, J., Aihou, K., Iwuafor, E.N.O., Lyasse, O., Sanginga, N., Merckx, R., 2002. Direct interactions between N fertilizer and organic matter: evidence from trials with ¹⁵N-labelled fertilizer. In: Vanlauwe, B., Diels, J., Sanginga, N., Merckx, R. (Eds.), *Integrated Plant Nutrient Management in Sub-Saharan Africa*. CAB International, Wallingford, UK, pp. 173–184.
- Verchot, L.V., Dutaur, L., Shepherd, K.D., Albrecht, A., 2011. Organic matter stabilization in soil aggregates: understanding the biogeochemical mechanisms that determine the fate of carbon inputs in soils. *Geoderma* 161, 182–193. <http://dx.doi.org/10.1016/j.geoderma.2010.12.017>.
- Wang, Z., Guo, H., Shen, F., Yang, G., Zhang, Y., Zeng, Y., Wang, L., Xiao, H., Deng, S., 2015. Biochar produced from oak sawdust by Lanthanum (La)-involved pyrolysis for adsorption of ammonium (NH₄⁺), nitrate (NO₃⁻), and phosphate (PO₄³⁻). *Chemosphere* 119, 646–653. <http://dx.doi.org/10.1016/j.chemosphere.2014.07.084>.

- Wardle, D.A., Marie-Charlotte, N., Olle, Z., 2008. Fire-derived charcoal causes loss of forest humus. *Science* 320, 2.
- Xiang, J., Liu, D., Ding, W., Yuan, J.J., Lin, Y.X., 2015. Effects of biochar on nitrous oxide and nitric oxide emissions from paddy field during the wheat growth season. *J. Cleaner Prod.* 104, 52–58.
- Zhang, D., Genxing, P., Wu, G.K., Wanjiru, G., Li, L., Zhang, X., Zheng, J., Zheng, J., Cheng, K., Joseph, S., Liu, X., 2015. Biochar helps enhance maize productivity and reduce greenhouse gas emissions under balanced fertilization in a rain-fed low fertility inceptisol. *Chemosphere* 142, 106–113. <http://dx.doi.org/10.1016/j.chemosphere.2015.04.088>.
- Zhang, A., Bian, R., Pan, G., Cui, L., Hussain, Q., Li, L., Zheng, J., Zheng, J., Zhang, X., Han, X., Yu, X., 2012a. Effects of biochar amendment on soil quality, crop yield and greenhouse gas emission in a Chinese rice paddy: a field study of 2 consecutive rice growing cycles. *Field Crops Res.* 127, 153–160. <http://dx.doi.org/10.1016/j.fcr.2011.11.020>.
- Zhang, A., Liu, Y., Pan, G., Hussain, Q., Li, L., Zheng, J., Zhang, X., 2012b. Effect of biochar amendment on maize yield and greenhouse gas emissions from a soil organic carbon poor calcareous loamy soil from Central China Plain. *Plant Soil* 351, 263–275. <http://dx.doi.org/10.1007/s11104-011-0957-x>.
- Zhao, Y., Pan, Y., Rutherford, J., Mitloehner, F.M., 2012. Estimation of the interference in multi-gas measurements using infrared photoacoustic analyzers. *Atmosphere* 3, 246–265. <http://dx.doi.org/10.3390/atmos3020246>.
- Zimmerman, A.R., Gao, B., Ahn, M.Y., 2011. Positive and negative carbon mineralization priming effects among a variety of biochar-amended soils. *Soil Biol. Biochem.* 43, 1169–1179. <http://dx.doi.org/10.1016/j.soilbio.2011.02.005>.