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BIOCHAR-BASED FERTILIZATION WITH LIQUID NUTRIENT ENRICHMENT: 21 FIELD TRIALS COVERING 13 CROP SPECIES IN NEPAL

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ABSTRACT

Biochar produced in cost-efficient flame curtain kilns (Kon-Tiki) was nutrient enriched either with cow urine or with dissolved mineral (NPK) fertilizer to produce biochar-based fertilizers containing between 60–100 kg N, 5–60 kg P_2O_5 and 60–100 kg K_2O, respectively, per ton of biochar. In 21 field trials, nutrient-enriched biochars were applied at rates of 0·5–2 t ha^{-1} into the root zone of 13 different crops. Treatments combining biochar, compost and organic or chemical fertilizer were evaluated; control treatments contained same amounts of nutrients but without biochar. All nutrient-enriched biochar substrates improved yields compared with their respective no-biochar controls. Biochar enriched with dissolved NPK produced on average 20% ± 5·1% ($N = 4$ trials) higher yields than standard NPK fertilization without biochar. Cow urine-enriched biochar blended with compost resulted on average in 123% ± 76·7% ($N = 13$ trials) higher yields compared with the organic farmer practice with cow urine-blended compost and outcompeted NPK-enriched biochar (same nutrient dose) by 103% ± 12·4% ($N = 4$ trials) respectively. Thus, the results of 21 field trials robustly revealed that low-dosage root zone application of organic biochar-based fertilizers caused substantial yield increases in rather fertile silt loam soils compared with traditional organic fertilization and to mineral NPK or NPK-biochar fertilization. This can be explained by the nutrient carrier effect of biochar, causing a slow nutrient release behaviour, more balanced nutrient fluxes and reduced nutrient losses, especially when liquid organic nutrients are used for the biochar enrichment. The results open up new pathways for optimizing organic farming and improving on-farm nutrient cycling. Copyright © 2017 John Wiley & Sons, Ltd.

KEY WORDS: biochar-based fertilization, organic fertilizer; urine; nutrient cycling, nutrient flux

INTRODUCTION

Pyrogenic carbon (PyC, biochar) is a natural constituent of soil organic matter (SOM) in many soils worldwide (Kluepfel *et al.*, 2014; Reisser *et al.*, 2016). It can also be found as an amendment in historically modified or modern agricultural soils where it is associated with increased soil fertility (Glaser & Birk, 2012; Solomon *et al.*, 2016). PyC is either the natural product of incomplete combustion during wildfires or it is deliberately produced as biochar in technical pyrolysis systems at low to zero oxygen conditions. PyC persists longer than any other form of organic carbon in soils (Lehmann *et al.*, 2015) and is considered a soil carbon sink (Santín *et al.*, 2015). In the last decade, the challenge to combat accelerating global warming, as well as anthropogenic land degradation, has caused a strong interest in biochar research. Multiple field trials have been initiated, most of them using biochar application rates between 10 and 50 t ha^{-1} (Crane-Droesch *et al.*, 2013). Across a wide range of field and lab studies, yield increases have been only minor, with 10–20% on average (Biederman & Harpole, 2013; Crane-Droesch *et al.*, 2013b; Jeffery *et al.*, 2015; Liu *et al.*, 2013). While some positive examples exist in degraded, mostly tropical acidic sandy soils (Yamato *et al.*, 2006; Steiner *et al.*, 2007; Cornelissen *et al.*, 2013), yield increases in fertile temperate soils were mostly low or absent (Schmidt *et al.*, 2014; Ruysschaert *et al.*, 2016) with some notable exceptions (Genesio *et al.*, 2015). At current industry prices of 600 to 800 Euro per ton of biochar (Schmidt & Shackley, 2016) those minor yield increases at such high application rates are economically not viable for most crops (see also economic evaluation in the Supporting Information).

Contrary to earlier experimental approaches, the formation of highly fertile black anthrosols (Amazon or African Dark Earths) was not due to the presence of pure biochar but rather to the amendment of mixtures of biochar with nutrient-rich organic matter (Glaser & Birk, 2012; Solomon *et al.*, 2016). Some field and pot trials tested therefore the combination of biochar and compost delivering results that indicate a synergy between the highly porous and recalcitrant biochar and nutrient-rich organic substrates (Steiner *et al.*, 2011; Schulz *et al.*, 2013; Ghosh *et al.*, 2015; Kammann *et al.*, 2016; Vandecasteele *et al.*, 2016). Prost *et al.* (2013) showed that biochar was enriched in nitrate,

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phosphate and other potential plant nutrients during the composting process. Biochar nutrients capture increased plant growth considerably when such composted biochar particles were used as soil amendment, compared with the same but untreated, pristine biochar (Kammann *et al.*, 2015). Some studies reported reduced N (nitrate) leaching (Major *et al.*, 2011; Ventura *et al.*, 2013; Haider *et al.*, 2016) when biochar was applied to soil combined with mineral or organic N fertilizer.

Since 2013, a new paradigm has emerged, suggesting that biochar should be considered as the matrix for a new generation of mineral or organic slow-release fertilizers (Joseph *et al.*, 2013; Qian *et al.*, 2014; Schmidt *et al.*, 2015). Schmidt *et al.* (2015) demonstrated that blending biochar with liquid organic manure (cow urine) and compost led to high pumpkin yield increases of more than 300% compared with the treatment with compost and liquid manure only and an increase of 85% compared with the treatment with biochar and compost. The authors hypothesized that enriching biochar with liquid organic manure may provide an organic coating of aromatic biochar surfaces, thus increasing the nutrient capturing ability and nutrient exchange capacity of the blended material. Subsequently, anions like nitrate or phosphate, as well as cations such as ammonium, may reversibly adsorb to the organic coating of the biochar surfaces (Archanjo *et al.*, 2017; Conte & Laudicina, 2017). However, the mechanisms behind the organic or organo-mineral coating and subsequent reversible binding (capture and release) of nutrients is not yet sufficiently understood.

When enriching biochar with liquid nutrients, the biochar may serve as carrier material, holding the nutrients in its highly porous structure, which may slow down the leaching of mobile nutrients, particularly in environments where heavy rainfalls occur. It is supposed that to mine those biochar-captured nutrients, plant roots and their symbionts would have to be in sufficiently close contact to the nutrient-charged biochar particles. It may therefore be more effective to apply the nutrient-enriched biochar into the root zone instead of distributing (and diluting) it homogenously across a field by broadcasting or plowing (Blackwell *et al.*, 2010; Graves, 2013; Schmidt *et al.*, 2015).

Depending on the form of N fertilizer, root zone application can reduce N losses and hence increase fertilizer-N use efficiency even in the absence of biochar. Huijsmans *et al.* (2003) showed that ammonia emissions were reduced by a factor of ten when liquid cow manure was applied to the subsurface zone compared with a broadcasting application. In 115 farmer trials, Mazid Miah *et al.* (2016) achieved yield increases between 11 and 31% at reduced urea fertilizer application rates between 28 and 44% (w/w) when pelletized urea was applied into the root zone of rice, compared with traditional broadcasting of the fertilizer.

Since 1997, the first patents have been issued related to using charcoal or biochar as a slow release fertilizer (Radlein *et al.*, 1997; Kotaka, 2005) for mineral N but not for organic N sources. Several scientific studies tested the patented or slightly modified methods (Magrini-Bair *et al.*, 2009) though mostly with not well-characterized low temperature biochars and with generally neutral or negative effects on plant growth compared with traditional fertilization (Dil *et al.*, 2014; González *et al.*, 2015). However, the biochar was not applied in concentrated form to the root zone but was homogenously distributed in pot trial soils. The latter was also the case in an experiment by Ye *et al.* (2016), where a biochar mineral complex fertilizer was applied at 1·5 t ha^{-1} with and without manure compost in a pak choi pot trial that did not lead to significant yield increase. Blackwell *et al.* (2010) were the first to test banded non-nutrient-enriched biochar application on dryland wheat production, where the substrate was applied to 5–15 cm depth over a width of 10 cm on a row spacing of 30 cm. They concluded that the banding application of biochar at low application rates of 1 t ha^{-1} can provide significant positive effects on yield while reducing fertilizer requirements.

To investigate suitable techniques to use biochar as a fertilizer carrier for on-farm nutrient management, 21 field trials were conducted in 2015 and 2016 with 13 different crops on different soils in different climatic zones of rural Nepal. Biochar was produced by flame curtain pyrolysis (Cornelissen *et al.*, 2016), enhanced either with liquid organic nutrients or with dissolved mineral fertilizer and applied at low biochar doses (0·5–2 t ha^{-1}) into the root zone of mostly annual as well as some perennial crops. The fertilizing effect of root zone applied biochar substrates was compared with that of farmers' traditional compost fertilization, as well as to other organic and conventional fertilization methods without biochar. We hypothesized that (1) liquid blending of biochar with dissolved nutrients would improve the fertilizing effect compared with the individual application of the respective fertilizers or that of the pure non-enhanced biochar and (2) organic nutrient enrichment of biochar will improve plant growth more strongly than mineral NPK enrichment of biochar.

MATERIAL AND METHODS

Experimental Sites

Between January 2015 and March 2016, a total of 21 field experiments were set-up in the eastern half of Nepal between the district of Tanahu (27°56′N, 84°25′E) as the most westerly site, and the district of Ilam (26°54′N, 87°55′E) as the most easterly location. The lowest site was in the Terai region at 90 m above mean sea level (amsl; Parwanipur), and the highest site was in the Kabre district at 2125 m amsl (Dhunkharka). This covered almost all agronomically relevant climatic zones of Nepal, from the tropical lowlands and the sub-tropical foothills to the hills with temperate climate. Ten out of the 21 trials were set-up with at least three, but mostly five replicate plots per treatment and three to seven different factorial treatments using either randomized block or Latin square designs. These trials are called here *primary trials* and are described in detail in the succeeding texts. The remaining trials were set-up as *village trials* with

Table I. Analytical results and quality parameters of the biochars used in five primary farmer trials

		Trial/Location						
		Pumpkin, Dhading	Cabbage, Dhading	Cabbage, Bandipur	Organic tea, Ilam	Maize, Parwanipur		
Kiln		60°, 1·1 m steel	60°, 1·1 m steel	60°, 1·1 m steel	60°, 1·1 m steel	70°, 1·5 m steel	European Biochar Certificate threshold	
Biomass		Eupatorim shrubs	woody feed leftovers	maize straw	tea prunings	rice husks, bamboo, wood	premium	basic
Density	kg m^{-3}	175	188	200	220	194		
Specific surface (BET)	m^{-2} g	215	149	69	82	85		
Water Holding Capacity (WHC)	g L^{-1}	846	720	715	ND	725		
Ash 550 °C	mass-%	21·9	23	48·3	40·3	28·8		
Hydrogen (H)	mass-%	1·33	1·32	0·58	1·3	2·02		
Carbon (C)	mass-%	72	73·3	47·3	53·7	59·6		
Nitrogen (N)	mass-%	0·54	0·59	0·53	0·59	0·74		
Oxygen (O)	mass-%	4·0	1·7	3·1	4·1	8·7		
Carbonate CO_2	mass-%	2·24	0·95	<0·4	<0·4	0·55		
Organic carbon (C_{org})	mass-%	71·4	73	47·3	53·7	59·5	>50	>50
H/C_{org} (molar)		0·22	0·21	0·15	0·29	0·4	<0·7	<0·7
O/C (molar)		0·042	0·017	0·049	0·057	0·11	<0·4	<0·4
pH		9·8	8·1	9·2	9	8·2		
Electric conductivity	µS cm^{-1}	9090	1400	1360	1020	ND		
Salt content	g kg^{-1}	8·25	7·55	7·29	2·7	ND		
Phosphorous (P)	mg kg^{-1}	3,700	7,400	16,000	2,300	2,900		
Magnesium (Mg)	mg kg^{-1}	12,000	14,000	7,600	6,800	6,100		
Calcium (Ca)	mg kg^{-1}	17,000	42,000	18,000	24,000	9,900		
Potassium (K)	mg kg^{-1}	28,000	37,000	45,000	16,000	19,000		
Natrium (Na)	mg kg^{-1}	520	460	610	750	5,800		
Iron (Fe)	mg kg^{-1}	6,000	12,000	15,000	19,000	1,200		
Silica (Si)	mg kg^{-1}	34,000	24,000	140,000	120,000	87,000		
Sulfur (S)	mg kg^{-1}	860	1,700	990	760	1,100		
Lead (Pb)	mg kg^{-1}	<2	<2	5	2	0·9	<120	<150
Cadmium (Cd)	mg kg^{-1}	<0·2	<0·2	<0·2	<0·2	<0·2	<1·5	<1·5
Copper (Cu)	mg kg^{-1}	30	17	19	17	7	<100	<100
Nickel (Ni)	mg kg^{-1}	5	2	10	11	2	<30	<50
Mercury (Hg)	mg kg^{-1}	<0·07	<0·07	<0·07	<0·07	0·18	<1	<1
Zinc (Zn)	mg kg^{-1}	120	70	99	47	65	<400	<400
Chromium (Cr)	mg kg^{-1}	7	2	18	14	1	<80	<90
Bor (B)	mg kg^{-1}	74	20	30	11	16		
Manganese (Mn)	mg kg^{-1}	210	140	310	540	270		
Naphtalene	mg kg^{-1}	2·0	2·5	2·5	5·3	ND		
Fluorene	mg kg^{-1}	0·1	0·2	0·3	0·2	ND		
Phenanthrene	mg kg^{-1}	0·8	0·5	0·7	0·5	ND		
Fluoranthene	mg kg^{-1}	0·6	0·2	0·3	0·2	ND		
Pyrene	mg kg^{-1}	0·5	0·2	0·3	0·3	ND		
Benzo(a)pyrene	mg kg^{-1}	<0·1	<0·1	<0·1	<0·1	ND		
SUM PAH (EPA 16)	mg kg^{-1}	4·9	3·6	4·5	6·7	ND	<4 ± 2	<12 ± 4

All analytical methods followed the European Biochar Certificate (European Biochar Certificate, 2012).

Table II. Soil analyses of all field trial sites including analytical methods

Analyzed parameters	Dhunkharka, primary trial – maize	Bara, primary trial – maize	Jhhapri, primary trial – chili	Jhhapri, primary trial – cabbage	Nalang, primary trial – cabbage	Nalang, primary trial – Japanese melone	Dhunkharka, primary trial – cardamom	Chandragiri, primary trial – cabbage	Rauta, primary trial – cinnamon
pH	4·67	7·05	5·1	4·08	5·91	5·99	3·77	5·92	4·44
Soil organic carbon (SOC)%	5·0	1·7	1·9	4·1	3·2	1·5	2·5	1·8	3·1
Total nitrogen (ppm)	2912	1904	1512	4390	5180	1960	1512	1932	1736
Molar C/N (average)	19·7	10·3	14·3	10·8	7·1	9	18·6	10·6	20·3
Available phosphorus (ppm)	425	344·2	86·6	377·6	292·5	308·4	8	568·7	48
Available potassium (K), (ppm)	718	561	389·4	129·1	148·7	98·98	55	420·2	224
Exchangeable calcium (Ca), (ppm)	680	34·8	853	326·7	1717	40	262·5	2154	245
Exchangeable m agnesium (Mg) (ppm)	186·6	1517	189	89·04	325·2	950	72·65	231·4	80
Exchangeable sodium (Na) (ppm)	13·9	425·2	39·7	15·88	27·38	433·1	6·441	75·56	2·034
CEC (m.e./100 g)	29·2	25·84	32·6	33·1	39	9·42	18·2	39·4	21·8
Clay (%)	20	21	26	22	44	13	17	26	20
Sand (%)	13·18	24·65	12·6	14·86	10·35	35·73	12·5	23·86	63·35
Silt (%)	66·82	54·35	61·5	63·14	45·65	51·27	70·5	50·14	16·65
Texture class	silt loam	silt loam	silt loam	silt loam	silt loam	silt loam	silt loam	silt loam	sandy loam

Village trial parameters (gray background colour) are given as mean values with standard deviation of all individual village sites.

Table II. Soil analyses of all field trial sites including analytical methods

Analyzed parameters	Rauta, primary trial – cinnamon	Dhunkharka, village trial – tomato	Dolakha, village trial – potato	Simara, village trial – onions	Jhhapri, village trials – chili, cabbage and califlower	Nalang, village trial – with cabbage and cauliflower	Barbote, village trial – tea	Test method/instrument
pH	4·44	5·6 ± 0·34	4·7	5·94	4·9 ± 0·32	5·7 ± 1	4·5 ± 0·24	Measured in H_2O suspension
Soil organic carbon (SOC)%	3·1	5·4 ± 0·65	1·4	1·6	1·7 ± 0·2	1·5 ± 0·35	1·9 ± 0·39	Weight loss on ignition at 360 °C
Total nitrogen (ppm)	1736	2678 ± 1509	1010	2184	1305 ± 285	1932 ± 455	1338 ± 293	Kjeldahl method
Molar C/N (average)	20·3	23·2	15·4	8·5	15	9·1	16·7	
Available phosphorus (ppm)	48	397 ± 50·3	31·5	130·8	65·9 ± 31·2	270 ± 179	106 ± 28·2	Olsen P-Method
Available potassium (K), (ppm)	224	621 ± 237	144	160	145 ± 103	227 ± 200	113 ± 92	Ammonium acetate followed by atomic absorbtion spectroscopy
Exchangeable calcium (Ca), (ppm)	245	901 ± 164	ND	52·6	419 ± 195	48·4 ± 12	407 ± 251	Ammonium acetate extraction
Exchangeable m agnesium (Mg) (ppm)	80	189 ± 6·4	600	1133	89 ± 44	889 ± 610	64 ± 21·6	Ammonium acetate extraction
Exchangeable sodium (Na) (ppm)	2·034	30·7 ± 12·8	36	459·1	12·4 ± 12	393 ± 298	23·5 ± 26·2	Ammonium acetate extraction
CEC (m.e./100 g)	21·8	29·3 ± 2·5	ND	44·81	17·3 ± 6	25·6 ± 13·4	30·6 ± 3·5	Ammonium acetate extraction
Clay (%)	20	21·3 ± 4·7	ND	24	31·4 ± 7·5	16·4 ± 4·2	24·1 ± 4·7	Hydrometer method
Sand (%)	63·35	13·1 ± 6	ND	13·29	16·4 ± 12·8	17·2 ± 9·9	44·7 ± 10·4	
Silt (%)	16·65	65·6 ± 2·1	ND	62·71	52·2 ± 11·3	66·4 ± 9·6	31·2 ± 7·6	
Texture class	sandy loam	silty clay loam	silt loam	silt loam	clay loam	silt loam	loam	

4 to 14 participating farmers of the same village. Each farmer managed one control plot and one plot with nutrient-enhanced biochar, respectively, with both plots receiving the same fertilizer amount and having the same size at all village locations. Each participating farmer was hence considered as one paired replicate. The individual village trial set-ups are detailed at the end of the section.

The site location, crop, main treatments and more detailed information like latitude, longitude, altitude, plant family, plot size, planting density, number of participating farmers, replicates and biochar feedstock can be found in Table S1.

Biochar and Soil Characterization

All biochars were produced by Kon-Tiki type flame curtain pyrolysis kilns (Cornelissen *et al.*, 2016) installed either in the fields or backyards of participating farmers. While in most primary trials and in all village trials soil pit Kon-Tiki kilns were used, at the following locations, full metal Kon-Tiki kilns were used: Nalang and Jhhapri cabbage trial, Ilam organic tea trial and Parwanipur maize trial. As shown in Cornelissen *et al.* (2016), no significant differences in quality were found between biochars generated from soil versus metal flame curtain pyrolysis kilns. The feedstock for the biochar production was in most cases *Eupatorium adenophorum*, a frequently occurring invasive forest shrub species that local people call 'ban mara' (i.e. forest killer) and woody feed leftover (i.e. animals are frequently fed on leafy tree twigs where the woody parts are leftover). Crop residues like maize straw, rice husks or tree prunings were also used (for details see Table S1), but forest wood was never used. The feedstocks were mostly dry and were pyrolyzed at 650–720 °C (Cornelissen *et al.*, 2016). After a pyrolysis time of approximately 1 h, when the earthen or steel cone was filled with carbonized biomass, the biochar was quenched either with water, with cow urine or with mineral liquid fertilizers (for details see Table S1). As the feedstocks used for the biochar production were thin and the resulting biochar friable, no milling was necessary. The average particle size was estimated to be below 10 mm. The type and general characteristics of the biochars of the different field trials are comparable to those of the Kon-Tiki type biochars listed and characterized in Cornelissen *et al.* (2016) that were produced with the same kilns and from comparable feedstocks within this project.

Only the biochars used for the primary field trial sites were analyzed and fully characterized. As the input materials and the production technique were the same in all trials, we consider the biochar characterization to be representative. The biochars from the cabbage trials in Jhhapri and Nalang, the tea trial in Ilam, the pumpkin trial in Nalang and the maize trial in Parwanipur were fully analyzed following the protocol of the European Biochar Certificate (European Biochar Certificate, 2012). All analytical parameters were well within the thresholds for premium quality of the European Biochar Certificate (European Biochar Certificate, 2012); the results are provided in Table I. As the production technology was the same for all biochars used in the various field trials, the main difference between the biochars was caused by variations in the local feedstock used for biochar production, which concerned mostly the carbon and mineral contents. The feedstocks used for each trial are listed in Table S1.

Soil samples were mixed from 12 randomly distributed soil cores collected from a depth of 2 to 20 cm before each respective trial was established. Soil samples were analyzed at the Aquatic Ecology Centre at Kathmandu University, Nepal, using the analytical methods outlined in Table II.

Cow Urine and Compost Nutrient Analyses

Cow urine was collected fresh usually from the participating farmers' own cows 1 day before the set-up of each respective trial. It was not possible to perform nutrient analyses of each urine sample due to the unavailability of these analytical capacities in Nepal and the difficulties to preserve and transport such organic liquid samples between remote tropical villages and foreign laboratories. One cow urine sample from the tea trial in Barbote (Ilam) was analyzed in Europe and subsequently compared with literature values (Saunders, 1982; Bristow et al., 1992; Haynes & Williams, 1993; Knowlton & Herbein, 2002; Hoogendoorn et al., 2010; Dijkstra et al., 2013; Schmidt et al., 2015). The analyzed sample had 7·1 g L^{-1} total N, 0·21 g L^{-1} phosphate (P_2O_5) and 7·9 g L^{-1} potassium, which is lower than average literature values. Due to the feeding of cows with low protein diets, the total NPK content of cow urine in rural Nepal is very likely lower than average literature values though no scientific data are available. For the field trials of this study, the following average cow urine nutrient values were used: 9·5 g L^{-1} total nitrogen (TN), 0·35 g L^{-1} phosphate (P_2O_5) and 11·0 g L^{-1} potassium oxide (K_2O), corresponding to an agronomic NPK ratio of 100:4:115 at an application rate of 10·5 m^3 cow urine per hectare. The actual NPK content of the cow urine used for the trials was probably lower, though the slightly higher literature values were used for the calculation of corresponding mineral NPK fertilizer rates. Hence, a conservative approach was taken to assure that mineral NPK rates were definitely not lower than the applied organic nutrients in the trials. Urinary P excretion in ruminants is generally considered minimal (Field et al., 2009) and is nearly 100 times lower than in solid cow manure (Knowlton & Herbein, 2002). The average pH of the cow urine samples from 11 trials was 7·6 ± 0·4 measured with a Mettler Toledo® FiveGo F2 directly in the liquid.

Each farmer used her or his own farm-made manure compost, with mostly cow and some goat manure mixed with approximately 30% (vol) rice or maize straw. The composts usually matured for at least 5 months and could be considered mature at the time of use. Due to variations in composition, climate and maintenance, the compost quality could not be considered equal between all farmers and farmer trials. However, because each respective primary or village trial used the same compost and same amount of it for all treatments, we consider the compost variation a contribution to the overall variation, but in a conservative and systematic way (i.e. when treatment effects are found *despite* this inherent variability, they need to be sufficiently large and well developed to be detectable). To represent the more than 100 different composts used in the trials is was decided to use the following average literature values for straw manure compost: 14 g N kg^{-1} (DM), 7 g P_2O_5 kg^{-1} (DM) and 18 g K_2O kg^{-1} (DM) (Eghball *et al.*, 1997; Sommer, 2001; Bernal *et al.*, 2009). Considering that 15% of the compost N and 30% of each of the compost phosphate and potassium are in a plant-available form in the first year (Wen *et al.*, 1997; Eghball, 2000; Bar-Tal *et al.*, 2004), the amount of crop available N, P_2O_5 and K_2O applied with the compost were estimated to be 2·1 kg N t^{-1}, 2·1 kg P_2O_5 t^{-1} and 2·6 kg K_2O t^{-1} respectively.

Root Zone Application of Substrates

Before seeding or planting, 30- to 40-m deep planting pits with a diameter of about 35 cm, or 30-cm deep planting furrows, were dug. According to farmer practice, cow manure compost was applied to each planting/seeding hole or to the bottom of each planting/seeding furrow. In the control treatments, the compost was mixed with either cow urine or NPK fertilizer corresponding to the amounts applied with the biochar treatments. In the biochar treatments, the biochar was first enriched with cow urine or dissolved NPK fertilizer and only afterwards mixed with the compost in the planting/seeding holes or furrows. In the trials with no compost amendment, only the organic or mineral fertilizers with or without biochar were applied to the planting/seeding pit or furrow and mixed with some soil. The planting/seeding pits or rows were then covered with 2 to 5 cm of the set-aside topsoil so that neither seeds nor the young emerging roots of the plantlets were in direct contact with the fertilizing substrates. In the trials with the perennial tea shrubs, 35 cm deep furrows were dug at a distance of 20 to 30 cm from one side of the tea shrub line. The fertilizing substrates were applied to the bottom of these furrows, which were then covered with soil. In both tree crop trials (cinnamon in Udyapur and cardamom in Kabre), new plantations and the planting pits were prepared as described earlier for annual crops, with the difference that planting pits were 40 to 45 cm deep and had a diameter of 50 cm, and direct root contact with the freshly applied substrate was avoided.

Preparation of Organic and Mineral Biochar-Based Fertilizers

Cow urine was thoroughly mixed with water quenched biochar (having a H_2O content of 60%) at a volume ratio of 1:1, which led to an approximate nutrient enrichment of 60 kg N, 2·1 kg P_2O_5 and 66 kg K_2O per ton of biochar. The urine-biochar slurry was soaked for at least 1 h before it was mixed with the compost and some soil inside the planting pits or furrows. In the trials where mineral fertilizer was used, the mineral nitrogen in the form of urea and/or diammonium phosphate, mineral phosphate in the form of diammonium phosphate or 85% phosphoric acid (PA) and mineral potassium in form of muriate of potash was dissolved in as much water as to obtain a saturated solution. The mineral nutrient solution was then mixed with water quenched biochar at a volume ratio of 1:1 and kept soaking for an hour resulting in biochar to NPK concentrations as given in Table S1. The NPK-biochar was applied into the root zone as described earlier for the urine-biochar substrates either mixed with compost or without compost (Table S1). In NPK control treatments, the (non-dissolved) NPK fertilizer was broadcast (standard practice).

Primary Cabbage Trials in Nalang and Jhhapri

In mid-September 2015, two white cabbage (*Brassica oleracea* L. convar. *Capitata* local var. *green hero*) trials were set-up in Nalang in the Dhading district of Nepal (27°50′N, 84°50′E) and 1 day later in Jhhapri close to the city of Bandipur in the Tanhu district of Nepal (27°55′N, 84°25′E). While the climate is subtropical at both locations with a single rainy season between mid-June to mid-September, the annual mean temperature and precipitation vary slightly (22·8 °C and 2330 mm in Nalang; 20·8 °C and 2100 mm in Jhhapri) mainly due to the difference in altitude amsl that is 450 m for the site in Nalang and 900 m for the site in Jhhapri. During the period of the experiment from mid-September 2015 to mid-January 2016, precipitation was 92 and 82 mm with an average temperature of 20·6 and 18·2 °C for Nalang and Jhhapri, respectively, both corresponding to the normal climate averages of this period for the respective regions. Due to the difference in altitude and geophysical exposition, the Nalang site had more fog and morning dew from mid-November through mid-January than the site in Jhhapri. The Jhhapri site is situated in a forest clearing. In the winter months, it receives less direct sunshine than the Nalang site that is surrounded only by some smaller individual trees. The trees surrounding the field were located far enough away to ensure that tree roots did not influence the crop growth.

Both soils are classified as silt loam though the soil characteristics were quite different. The Jhhapri site was only recently re-cultivated from a sal tree (*Shorea robusta*) overgrown terrace explaining its high SOM content of 7% and low pH of 4·1, while the long-term cultivated house garden field in Nalang also had a rather high SOM content of 5·4% but a less acidic pH (5·9). Soil nutrients and cation exchange capacities (CEC) were rather similar (Table II). Both farms participating in this trial usually practice organic farming methods. No mineral fertilizer, herbicides or chemical pesticides were used either during the trial or in the decade before the trial, except for the mineral fertilizer of both NPK treatments of the trial itself.

The experiment was laid out with an identical set-up at both field sites using a fully randomized block design with five replicates. The plot size was 1·9 m^2 containing eight cabbage plants at an interplant distance of 40 by 60 cm. Cabbage plantlets were bought for both sites at the same plant nursery on the same day. Mineral fertilizer (urea, PA and muriate of potash) for both sites was used from the same bags. Pure cow urine was collected fresh from the respective farmer's cows. No compost was used in both field trials. The biochar was produced in a 60°, 110 cm diameter full metal octagonal Kon-Tiki type kiln on the respective day of the field trial set-up. Feedstock for the biochar was woody feed leftovers and *Eupatorium* shrubs in Nalang and maize straw in Jhhapri (analyses see Table I).

The objective of the trial was to compare the fertilizing effect of NPK enriched biochar with cow urine-enriched biochar, where the nutrient enrichment was either done with glowing hot biochar or with cold water quenched biochar. For both NPK treatments, the mineral fertilizer (NPK) was dissolved in water (10 m^3 ha^{-1}) at per hectare rates of 100 kg N, 50 kg P_2O_5 and 115 kg K_2O. For both organic treatments 46 kg ha^{-1} P_2O_5 (in the form of H_3PO_4) was dissolved in 10·5 m^3 ha^{-1} fresh cow urine (assuming the NPK content detailed earlier), which led to the same or slightly lower nutrient contents in both organic treatments compared with both NPK treatments.

For the *hot quench* treatments, glowing hot biochar was taken directly from the kiln and immersed in the organic, respective mineral nutrient solution, and stirred as described by Pandit *et al.* (2017). For both *cold quench* treatments, biochar from the same kiln was first quenched with water vapor letting water flow from the bottom of the kiln up to the top. The cold, water quenched biochar was then shoveled into the buckets containing the same organic, respective mineral nutrient solution as in the *hot quench* treatments, and was also stirred. The blending of biochar and the nutrient solutions was done at volume ratios of 1:1 in all treatments although the water content of the *cold quench* treatment was thus higher. The nutrient solution- biochar mixes were left soaking for at least 1 h. The root zone application of the four different biochar fertilizer substrates and the planting of the cabbage plantlets were then carried out as described earlier.

During the dry season, the trial plots were irrigated with watering cans providing the same amount of water for each plot. Weeding was done manually. No major pests or pathogenic insects were observed. Plant height and the leaf area were measured after 7 and 9 weeks in Nalang and Jhhapri respectively. The leaf area was calculated as the sum of (length of leaf × mean width at ¼, ½ and ¾ of leaf length) for all leaves per plant. The plots were harvested on 8 and 12 January in Jhhapri and Nalang, respectively, using the same scales for each trial, weighing the fresh aboveground biomass and the fresh weight of the cabbage heads of each plot.

Primary Organic Tea Trial in Barbote (Ilam)

In the organic tea trial in Barbote, Ilam, the traditional farmer fertilization practice with 15 t ha^{-1} broadcast compost only (control) was compared with (2) root zone applied cow urine, (3) biochar only, (4) hot-quenched urine-biochar and (5) cold-blended urine-biochar, with all experimental plots having received 15 t ha^{-1} of broadcasted compost. The experiment was laid out using a fully randomized block design with three replicates. The cow-urine-biochar treatment was prepared as described earlier mixing 6 m^3 ha^{-1} fresh cow urine with 6 m^3 ha^{-1} [corresponding to 1 t (DM)] water quenched biochar. For the hot-quenched urine-biochar treatment, the same amount of biochar as in the cold urine-biochar treatment was taken glowing hot from the kiln and soaked in an identical amount of fresh cow urine. Both the hot and the cold mixed substrates were left impregnating for between 1 and 2 h to allow the liquid solution to penetrate into the porous structure of the biochar. For the cow urine only treatment, the urine was diluted 1:5 with water to avoid plant toxicity.

At the lower side of tea-tree rows planted on a northeastern-exposed slope, 35-cm deep furrows were dug by hand. The substrates were applied at the bottom of the furrows that were subsequently covered again with soil. In the control treatment where no substrate was root zone applied, the same type of furrow was dug and subsequently closed with soil again to assure same conditions for all treatments. Harvesting took place eight times between the beginning of June and end of November 2015 according to local harvesting practices.

Primary Cinnamon Trial in Rauta (Udaypur)

In the cinnamon trial in Udaypur, a total of 16 cinnamon trees with heights of 20 to 23 cm were newly planted in a randomized block design with plant-to-plant distances of 4·50 m. Tree planting pits were dug to a depth of 40 to 45 cm with a diameter of 50 cm. At the bottom of the planting pit, the following respective substrates were applied and subsequently mixed with soil: (A) 5 kg compost; (B) 5 l biochar + 5 kg compost, (C) 5 l cow urine + 5 kg compost and (D) 5 l cow urine mixed with 5 l biochar + 5 kg compost; application rates corresponded to 400 kg ha^{-1} biochar, 2·5 m^3 ha^{-1} (0·25 mm) cow urine and 2·5 t ha^{-1} compost. The substrates were covered with 3 to 5 cm soil, and the trees were planted into the pits avoiding direct contact of the roots with the substrates. After planting, the soil of the pit surface was mulched with grasses.

Primary Japanese Melon Trial in Nalang (Dhading)

The trial with Japanese melons (*Solanum muricatum*) was set-up in September 2015 immediately after uprooting the pumpkin plants from the earlier pumpkin biochar trial sites (Schmidt *et al.*, 2015). The same set-up as for the pumpkin trial was repeated, and the same types and amounts of substrates that were used 7 months before in the pumpkin trial were applied again to planting pits dug at the exact locations of the former pumpkin plants; the new pits were 40 cm deep, with a diameter of 35 cm. Four kilogram of cow manure compost (10·5 t ha^{-1}) were applied to each new planting hole of each treatment. In treatment (A), the compost was mixed with 2·4 l of fresh cow urine (6·3 m^3 ha^{-1}) and some soil. For each plot of treatment (B), 290 g (DM) of biochar (0·75 t ha^{-1}) was mixed with the compost and some soil. For each plot of treatment (C), 290 g (DM) of biochar (0·75 t ha^{-1}) was mixed with 2·4 L of fresh cow urine

($6{\cdot}3\ m^3\ ha^{-1}$) and compost. The substrates were again covered with a few centimetre of soil to avoid direct root contact with the plantlet roots that were planted above the root-zone applied materials. Japanese melons were harvested five times between March and May 2016.

Further Primary Trials

Four further primary trials: chili in Jhhapri (Tanahu), cabbage in Chandragiri (Kathmandu), cardamom in Dhunkarka (Kavre) and maize in Dhunkharka and in Parwanipur (Bara), were set-up with similar treatments and experimental guidelines as the previously presented trials and are not described in detail here. All relevant information about treatments, set-up, field sites, replicates etc. are given in Table S1. The set-up of the primary pumpkin trial in Nalang (Dhading) was reported earlier by Schmidt *et al.* (2015).

Village Trials

Beside the ten primary trials described above, 11 village trials on a total of 88 field sites with as many participating farmers were set-up. In the village trials, common farmer's fertilization practice was compared with an optimized biochar-based fertilization method, using the same amounts of the major plant nutrients (NPK). For each village trial at least four, but mostly eight to ten farmers of the same village participated, using exactly the same general set-up, so that each farmer within a respective trial is considered to be one replicate, using paired *t*-tests for the statistical analyses. Village trials were set-up on relatively large ($3{\cdot}7$ to $14\ m^2$) plots per farmer site with one replicate per treatment (as in this type of village trials, the individual farmer sites within the village were considered replicates).

In general, in each trial village, the individual farmer field trials were set-up using the same biochar that was jointly produced by all participating farmers at the start of the respective experiment. The compost and the cow urine were provided by each participating farmer from her/his own cowshed. When mineral fertilizer was used, it was provided by the same source in each village. As far as irrigation was necessary, the respective farmer irrigated her/his plots using equal water amounts for all plots. The plots were hand weeded; no supplemental fertilizers or pesticides were applied. In each trial village, two designated lead farmers visited each individual trial at least once a week to ensure equal treatment (weeding, irrigation, mulching etc.). For the trial set-up and the harvest, the local project leader accompanied the farmer leaders and farmers to guarantee the correct experimental procedures, sampling and the correct measuring and recording of the data for plant growth, crop and biomass yield. The application amounts of biochar, cow urine, compost and NPK fertilizer are given for all village trial treatments in Table S1.

Statistical Analysis

For the statistical analyses of the primary field trial results, normality was tested with the Shapiro–Wilk test and homogeneity of variances with Levene's test. Data not following a normal distribution were log-transformed. Treatment effects were analyzed by using one-way analysis of variance (ANOVA) with *post hoc* Tukey tests to detect significant differences among treatment means. Plant growth results of the cabbage trial after 7 and 9 weeks were analyzed with a two-way ANOVA including site location (Nalang or Jhhapri) as second independent variable. The organic tea trial was also analyzed by two-way ANOVA including blocks as second independent variable to evaluate block effects. Both two-way ANOVAs were followed by post hoc Tukey tests to detect differences between treatment means. All data are presented as means ± standard deviation. A *p*-value of $<0{\cdot}05$ was considered significant. Statistical analysis of the village trials with two treatments (with and without biochar) was done by paired *t*-test, using individual farmer sites as replicates and the control versus biochar treatment of each farmer as pair.

RESULTS

In all 21 trials, the nutrient-enriched biochar treatments provided significantly higher yields compared with treatments with the same nutrient concentrations but without biochar. An average yield increase of 20% ± 5·1% was obtained with NPK-charged biochar compared with NPK only (one primary trial, three village trials and four different crops). When organic biochar-based fertilizers were compared with the same organic fertilizers without biochar (seven primary, six village trials and eight crops), an average yield increase of 123% ± 76·7% was obtained. Organic-enriched biochar outcompeted NPK-charged biochar in three primary and one village trial with three different crops showing an average yield increase of 103% ± 12·4%. In the nutrient-enhanced biochar treatments, the average crop yields per hectare were in the range of commercial crop production for all tested crops (see the succeeding texts for references for each crop). The comparatively low but highly significant yield increases in the NPK-biochar treatments compared with NPK only ranged from 15% ± 8·7% in the potato village trial and 16% ± 7·8% in the onion village trial to 22% ± 17·1% in the primary maize trial and 28% ± 14·6% in the village tea trial (Figure 1 and Table S1).

Organic Biochar-Based Fertilizers

Most biochar field trials were undertaken with cow urine-charged biochar that was subsequently blended with manure compost and compared with the control treatment with cow urine-blended compost. The yield increases varied strongly depending on the crop species and location, but increases in all trials were substantial and statistically significant in the range of +15 to +298% (Figure 1). The average yield per hectare for tomato (University of Georgia, 2016), tea (Simple Leaf, 2006), pumpkin (Penn State, 2016) and cauliflower (North Carolina State, 2016) in the organic biochar based treatments were all at the higher end or higher than the respective reference values cited earlier for optimized industrial farming. The yield values for white cabbage

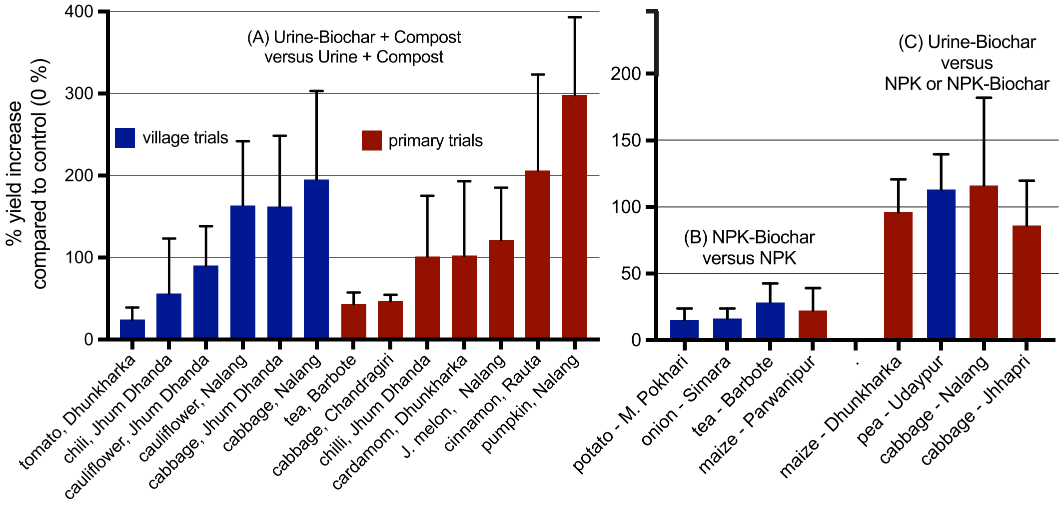

Figure 1. Average yield increases of (A) urine-biochar + compost treatments compared with urine + compost, (B) NPK-biochar compared with NPK only and (C) urine-biochar compared with NPK only or with NPK-biochar. Yield increases are given as the absolute percentage increase above the control yield value. The bars show means and standard deviation. All percentage yield increases were significant ($p < 0{\cdot}05$). The red coloured columns indicate the primary trials, and the blue columns indicate the village trials. [Colour figure can be viewed at wileyonlinelibrary.com]

(University of California, 2013) were slightly lower than the reference value, and only maize yields (Langemeier, 2015) were clearly lower (for all reference values see Table S1). The latter was likely due to the local cultivar, which is growing on average more than 3·50 m high but with only a few leaves and smaller cobs.

In the primary trials with organic tea, cinnamon and Japanese melon (Figure 2), the root zone application of urine-biochar always caused significantly higher yields compared with the application of biochar (+34%, +100% and +71% respectively) or compared with the application of water diluted urine (+43%, +166% and +176% respectively); compost was provided in equal amounts to all treatments (Figure 2). In these three trials, the pure-biochar treatment did not show significant differences compared with the water diluted urine treatment (only in the first 3 months of the tea trial, a significant difference was seen (Figure 2a) but not over the entire tea harvest season (Figure 2b), see discussion in the succeeding texts).

In the Japanese melon trial, set-up directly after uprooting the pumpkin plants of the earlier trial with the same substrates, the melon plantlets had been planted into the same pits as the pumpkins before. The melon yield results again showed the same effect of urine-biochar + compost application for the second crop in a row. The yield increase of Japanese melon was 102% compared with urine + compost and 68% to biochar + compost, which is a considerable but lower increase than in the pumpkin trial (+298% and +85% compared with the same respective treatments) performed at the same site.

Comparison of Organic and Mineral Biochar-Based Fertilization

In Jhhapri and Nalang, the organic biochar-based fertilization increased the cabbage head yield significantly ($p < 0{\cdot}001$) by on average + 86% and +116%, respectively, compared with the cold charged NPK-biochar-based fertilization. Cold charging of biochar led in both cases to higher yields although the differences were not significant ($p > 0{\cdot}05$) when compared with hot charging (Figure 3).

The total yield of all treatments in Nalang was 21% higher than in Jhhapri, and the cold urine treatment was 41% higher compared with the same treatment in Jhhapri. Although the SOM in Jhhapri was higher (7% versus 5·4%), the pH in Nalang was much closer to the optimal range for cabbage (5·9 in Nalang versus 4·1 in Jhhapri) and the CEC was also 15% higher. The soil in Nalang could thus be considered to be more fertile than in Jhhapri; the organic biochar-based fertilizer produced a higher yield increase in the more fertile soil. Temperatures during the winter growing period were milder in Nalang, which is the lower altitude village. Moreover, yield differences between both sites might also be due to differences in biochar feedstock (woody feed leftover in Nalang and maize straw in Jhhapri), which resulted in a biochar with lower carbon content, lower specific surface area and higher mineral content in Jhhapri compared with Nalang although the water holding capacity was equal for both biochars (table I).

The cabbage village trials, done in parallel, compared a modified farmer practice (compost + urine root zone application) with the same cold-urine-biochar charging treatment than in the primary trial, except that no PA was added as P source, which was provided by the compost in the village trial. The yield increase of the urine-biochar + compost treatment, when compared with the standard village organic farmer practice, was higher (+162% in Jhhapri and +195% in Nalang) than the yield increase compared with the NPK-biochar fertilization in the corresponding primary trials (+86% in Jhhapri and +116% in Nalang) (Figure 3). The urine-biochar treatment led in both the village and the primary trials to comparable high yields per hectare.

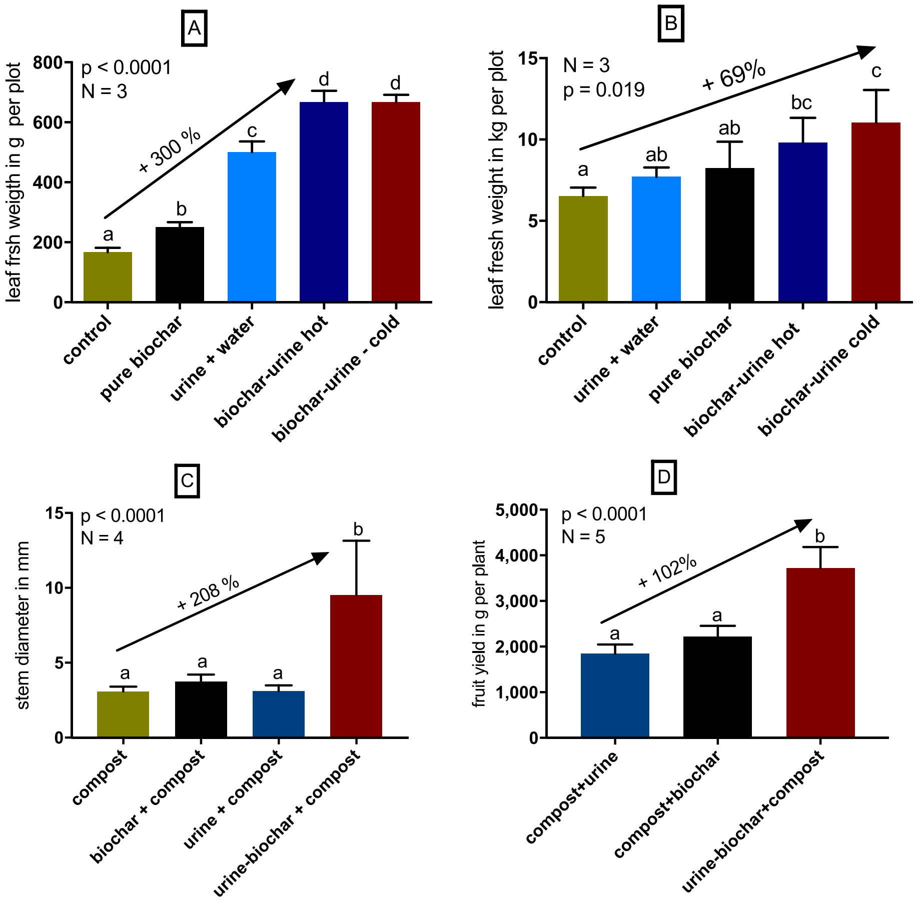

Figure 2. (A) tea yield after two harvests over the first 2 months, or (B) after eight consecutive harvests over the whole tea season. (C) Stem diameter of cinnamon trees 10 months after planting with root substrate applications. (D) Total fruit yield per plant of Japanese melon after the first growing season (three harvests). The bars show the means + standard deviation; the numbers of replicates per treatment are given in the sub-figures. The different letters indicate significant differences between treatments. 'Control' refers in the tea trial to the standard compost broadcasting fertilization, which had also been applied to all other treatments. [Colour figure can be viewed at wileyonlinelibrary.com]

Seven to 9 weeks after cabbage planting, plant growth differences between the treatments were still minimal and mostly not significant (Figure 4). The nutrient availability of cold-charged NPK biochar and both cow urine biochar treatments therefore seem to have been comparable during the first 2 months; only later did it start to differ significantly.

In all four trials at 29 farmer sites where conventional NPK fertilization was compared with cold-charged NPK-

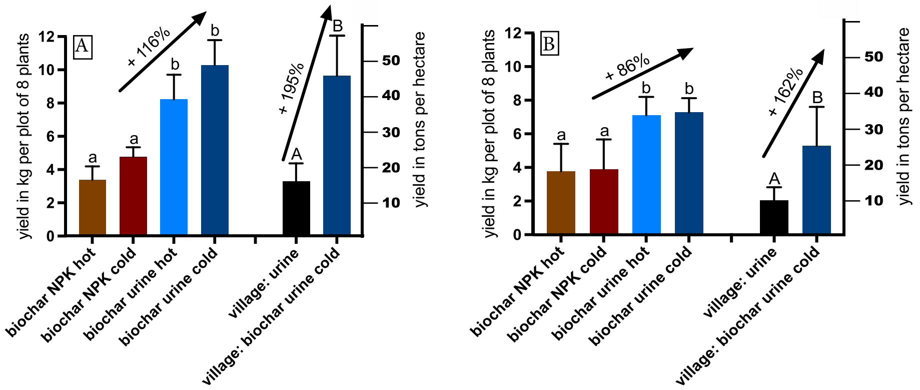

Figure 3. Cabbage head yield of two parallel cabbage field trials in Nalang (left) and Jhhapri (right) including average yields of farmer trials in the corresponding villages, means + standard deviation. $N = 5$ for both primary trials; $N = 11$ and $N = 9$ for Nalang and Jhhapri village trials respectively. The different lowercase (primary trial) and uppercase (village trial) letters indicate significant differences between treatments at $p < 0{\cdot}05$. [Colour figure can be viewed at wileyonlinelibrary.com]

biochar root zone fertilization, the latter caused significantly higher yields, so that NPK enriched biochar can be considered an optimized NPK fertilization method. That the application of biochar in conjunction with NPK-fertilizer never decreased but rather increased yields compared with conventional NPK fertilization is further confirmed by literature (Sinclaire *et al.*, 2008; Schulz & Glaser, 2012; Joseph *et al.*, 2013; Martinsen *et al.*, 2014; Abiven *et al.*, 2015; Tian *et al.*, 2016). The use of cold NPK-enriched biochar in the cabbage trials was therefore considered to be at least equal to conventional fertilization and hence used as the benchmark that has to be matched by the organic biochar based fertilization.

The primary maize trial in Dhunkharka and the village pea trials in Hadiya and Jogidaha (Udaypri) where yield increases with the cow urine biochar treatments were +96% and +113%, respectively, when compared with the pure NPK fertilization (Figure 1), further underpin the potential of organic biochar-based fertilizers compared with mineral NPK fertilization.

DISCUSSION

The results of the 21 trials reported here clearly reveal the potential of biochar to increase crop yield when used as nutrient carrier matrix for fertilizer applications. Depending on crop species, location (soil) and type of nutrients charged into the biochar, the magnitude of the plant growth promoting effects varied. However, compared with either the treatment with the respective nutrients alone or with biochar alone, the plant growth enhancement was always highest when liquid nutrients were charged into the biochar before application to the root zone.

Biochar Enriched with Mineral NPK

The least strong though consistent and always significant yield increase (on average 20%) was obtained with NPK-enriched biochar compared with NPK only. This corresponds well to the 18% average yield increase calculated by Jeffery *et al.* (2015) in their meta-analyses of 60 biochar field trials. However, the experiments included by Jeffery and colleagues did not use nutrient-enriched biochars and they only observed positive yield responses at application rates larger than 5 t ha^{-1}. Our results thus demonstrate that smaller amounts of biochar (0·5 to 2 t ha^{-1}) might be sufficient to achieve yield increases in this range if they are enriched beforehand with liquid mineral nutrients and are applied to the root zone.

Biochar Enriched with Organic Fertilizer

The highest increase in yields, which on average more than doubled compared with the equally fertilized farmer practice, was obtained when the biochar was charged with organic, nutrient-rich liquids such as cow urine. The high yields achieved with this practice not only exceeded by far the yields in the organic control without biochar (+123% ± 76·7%) using the same amounts of the main plant nutrients, but they also exceeded the yields in the NPK fertilizer treatments either with or without biochar (+103% ± 12·4%). Similar high yield increases due to organic nutrient enriched biochar were obtained by Kammann *et al.* (2015) when biochar was removed from a compost pile 8 weeks after co-composting with animal manure.

Cold Versus Hot Nutrient Enrichment

Impregnation of glowing-hot biochar with liquid nutrients was not observed to provide any apparent advantage compared with impregnation of cold, water-quenched biochar with the same amount of nutrients (Figure 3). Pandit *et al.* (2017) equally demonstrated that the enrichment of biochar with liquid NPK nutrients was plant growth enhancing but found, at very high nutrient concentrations, the hot nutrient enrichment more beneficial. During the process of hot enrichment, the biochar pores are wider and more flexible (Xiao & Pignatello, 2016), which was assumed to increase the water and nutrient penetration into the porous system, allowing more nutrients to be loaded into the biochar. On the other hand, when the glowing, 400–700 °C hot biochar is immersed into the liquid nutrient solution, water and some volatile nutrients may be volatilized (e.g. as NH_3) or may be released as micro particles with the rising vapor. Also, due to the increased nutrient and water mobility during the hot

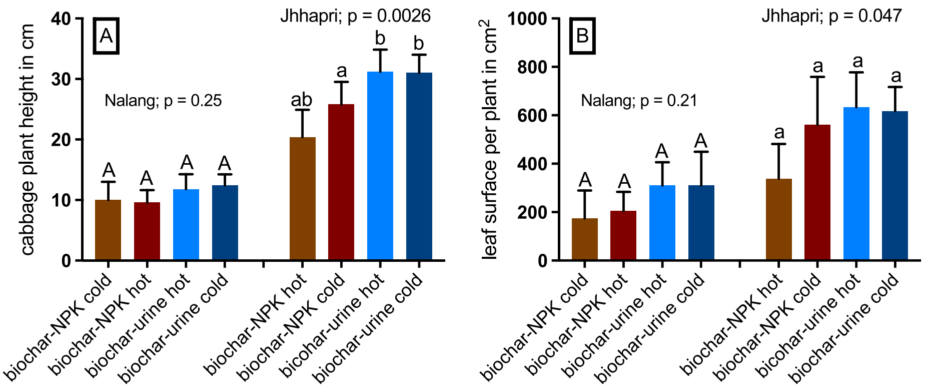

Figure 4. Plant height (left) and leaf surface (right) of cabbage plants 7 weeks after plantation in Nalang (left columns respectively) and 9 weeks after plantation in Jhhapri (right columns respectively); means + standard deviation, $n = 5$ replicates. The different uppercase (Nalang) and lowercase (Jhhapri) letters indicate significant differences in the respective trial due to treatment at $p < 0{\cdot}05$. [Colour figure can be viewed at wileyonlinelibrary.com]

enrichment, these nutrients may penetrate deeper into the pore system, may thus eventually become more strongly bound and subsequently be less bioavailable after cooling down. In the latter case, nutrient losses during charging might be less important and hot-charged biochars would contain similar nutrient amounts, but the release may be restrained. To clarify these mechanisms, nutrient balances before and after hot and cold charging, or leaching and nutrient-release trials using for example stable isotopes, are needed.

The significant yield increases with cow-urine-biochar-based fertilization in both primary cabbage trials, compared with NPK fertilizer, demonstrated that organic biochar-based, farmer made fertilization can outcompete chemical NPK fertilization. This is further confirmed by results from maize and pea trials where in both cases, cow urine–biochar treatments more than doubled the yields compared with conventional NPK fertilization with the same level of available nutrients. Understanding the key mechanisms behind the improved plant productivity with organic-enriched versus NPK-enriched biochars will need more advanced experiments and analyses. However, the impressive consistency of the observed effects sheds some first light on the following principles of underlying mechanisms and functions of biochar.

Basic Biochar-Related Mechanisms

While until now positive yield responses to biochar applications have almost exclusively been obtained on poor and degraded soils (Biederman & Harpole, 2013; Cornelissen *et al.*, 2013), all soils of the present biochar field trials can be considered as relatively fertile with their SOM contents ranging from 2·3 to 9·2% and soil pH values ranging from 4·5 to 6·1 with two exceptions at 4·1 and 3·8. Several authors showed that biochar may have a liming effect which depends on biochar and soil type (Silber *et al.*, 2010; Butnan *et al.*, 2015; Camps-Arbestain *et al.*, 2015; Jeffery *et al.*, 2015; Martinsen *et al.*, 2015). Moreover, Jeffery *et al.* (2015) showed in his meta-analysis a clear dependency of biochar-caused yield increases on the initial soil pH. However, in all these trials, relatively large amounts of biochar (>10 t ha^{-1}) were applied. In our trials the biochar was applied concentrated into the root zone; hence, the biochar-containing substrates may have had a considerable local liming effect in parts of the root zone, even at more than 10 times lower per hectare application rates (Cornelissen *et al.*, 2013). If liming would be the dominant effect, results of treatments with biochar only or biochar + compost but without liquid nutrient enhancement should have caused a similar pronounced increase in plant growth as both treatments likely had the same liming or acid neutralizing capacity. However, no such ‘biochar-only’ effect was observed in the pumpkin, Japanese melon, primary tea, cinnamon and the chili trial (Figures 2 and 3). For the same reason, it can be excluded that biochar-contained micronutrients triggered the high yield increases though an additional positive effect due to these supplementary micronutrients is likely (Oram *et al.*, 2014). Liquid nutrient loading of biochar pores might have increased the mobility of ash compounds and thus increased the plant availability of at least some of the micronutrients.

As argued earlier and by Schmidt *et al.* (2015), the strong yield increases obtained with organic nutrient-enhanced biochars cannot simply be explained by a ‘biochar effect’ alone. The yield responses of the presented trials confirm that the mechanisms fostering improved plant growth lie in the combination of liquid organic nutrients and biochar rather than in the effects of each component alone.

Wachendorf *et al.* (2005) showed in a lysimeter trial with ^{15}N-labelled cow urine under an intact pasture grass sod that an average of 60% of the urine-N leached into the subsoil within 1 year in a temperate climate. The tea and cinnamon trial data presented here were conducted over a period that included a complete rainy season with precipitation of >2000 mm. Hence, it is likely that considerable amounts of the urine-applied N in the urine-only plots had leached away before the shrub or tree roots could take them up. Interestingly, the effects of both the ‘urine-biochar’ and the ‘urine-only’ treatments were more pronounced up to the second tea harvest 6 weeks after the application where the leaf yield increase was 300% for urine-biochar, 200% for urine only and 50% for biochar only compared with the control. However, these yield increases levelled out and after 7 months, only the urine-biochar treatment continued to show significant yield increases of +69%, compared with the control, while urine-only and biochar-only treatments showed no-significant increases anymore (Figure 1). These yield-increase dynamics over time are in line with the perception that urine-contained nutrients applied without biochar may be leached to greater depths and out of reach for plant roots rather quickly or become microbially immobilized (Burger & Jackson, 2003) while biochar may capture and release organic nutrients at least partly in a plant available form (Sarkhot *et al.*, 2008; Clough *et al.*, 2013). However, future research including lysimeter trials, plant tissue analyses and SOM and biochar particle analyses are needed to investigate if the hypothesized slow release effects do occur.

The experimental data show that the nutrient release effect of the urine-biochar blends follows a time-dependent dynamic as the growth promoting effect of urine-biochar treatments clearly increases over time, compared with urine–water or to NPK-only applications (e.g. primary cabbage and tea trials). In these trials, significant differences between both treatments were only seen more than 8 weeks after the application (Figures 2 and 4). This indicated that mineral nutrients may have initially been sufficiently available in all treatments. However, later, the organic-enriched biochar substrates in the root zones likely continued to release nutrients over a longer time period. While it could be that the urine-biochar effect is not only a nutrient effect and that hormonal and other growth stimulating substances of the cow urine may play a major role (Gadelha *et al.*, 2002; Oliveira et al., 2009) (because urine in general contains a variety of up to 3000 low-molecular compounds, many of them containing N; Bristow *et al.*, 1992; Bouatra *et al.*, 2013), the increasing growth differences over time indicate a longer-lasting release of nutrients and/or plant growth promoting substances.

Although fertilization with water diluted urine is a standard fertilization practice (Gadelha *et al.*, 2002; Oliveira et al., 2009) showing largely positive plant growth promoting effects, urine may well contain substances provoking also adverse effect on crop growth, which may have been avoided through their adsorption by co-applied biochar. This could be an additional but minor reason for improved yields in the urine-biochar treatments compared with the treatments with urine only (1:10 water diluted).

Schmidt *et al.* (2015) discussed several further hypotheses to explain the observed effect of organic biochar-based fertilization methods, such as (i) improved CEC, (ii) redox potential (Eh) and (iii) water holding capacity. However, since this earlier publication, no new evidence for such effects can be cited. Several authors (Lehmann *et al.*, 2011; Joseph *et al.*, 2015; Rex *et al.*, 2015; Kolton *et al.*, 2016; Ye *et al.*, 2016) recently showed that biochar application (iv) influences the rhizosphere microbiome, may increase microbial diversity or metabolic efficiency and that it can cause changes in the metabolic potential of the rhizosphere. Enriching biochar with microbial nutrients such as cow urine may further modify and probably increase the influence on the rhizosphere microbiome and microbial-root symbioses. However, shifts in microbial function and diversity due to the provision of microbial nutrients via biochar used as carrier substances need to be investigated yet.

As the most likely mechanism behind the high yield increases in the organic biochar-based fertilization, we consider the reduction of leaching of organic nutrients and an increased or more continuous 'on-demand' delivery of the nutrients to microbes and plants over a longer time period, acting like a slow release fertilizer.

Kammann *et al.* (2015) have shown that biochar can capture and release nitrate, phosphate and dissolved organic carbon. The overall amount of captured nitrate revealed in this study ($<1\%$ of the dry biochar weight) was certainly not sufficient to feed plants at application amounts of 1 t biochar per hectare (i.e. 10 kg N ha^{-1} at maximum). However, the mechanisms of nitrate capture (Haider *et al.*, 2016) may be involved in the retention of mineralized N from the soil organic carbon pool or of nitrate released via mineralization–nitrification from compost and urine. Dissolved organic carbon is more strongly sorbed to biochar than anions such as nitrate (Hale *et al.*, 2013; Eykelbosh *et al.*, 2015; Smebye *et al.*, 2015), which is also true for dissolved organic nitrogen, despite carrying a net negative charge (Dempster *et al.*, 2011; Clough *et al.*, 2013). Van Kessel *et al.* (2002) showed that up to 127 kg dissolved organic nitrogen-ha^{-1} a^{-1} can leach out of prairie soil after cow urine application which, when applied with biochar, may be preserved at least to a greater extent than without biochar. However, for conclusive evidence, studies using isotopically labelled ^{15}N for organic and/or mineral nutrient charging of biochar are needed to identify the various gross transformation rates and flows of N applied to the crop root zone.

Multispecies Nutrient Binding Mechanisms and Time-Dependent Release Rates

The capturing and release of cations and anions in biochar are most likely because of the following:

1. Water bridging – i.e. water bridges between the hydration shells surrounding the dissolved ions and the hydrophilic systems, such as inorganic ashes and oxy-genated organic functions, on biochar surfaces (Conte, 2014).
2. Ionic bonding – i.e. direct ion bonding to positive, respectively, negative charged micro sites, functional surface groups or precipitated metal oxides or pyrolytic tars.
3. Covalent bonding – i.e. chemical bonding that involves the sharing of electron pairs between the ion and a valent partner in/on the biochar).
4. Physical entrapment (i.e. dissolved minerals entering narrow or subsequently clogged pores (Pignatello *et al.*, 2006; Jassal *et al.*, 2015).

Organic compounds can further be bound via the following:

5. Charge-assisted intramolecular hydrogen bonds – i.e. H bonds between the electron-rich proton acceptors in the urine-contained organic groups and the proton donors on biochar surface sites, or, vice versa, between the electron-rich biochar aromatic systems and the proton donors in the organic systems from the added urine (Xiao & Pignatello, 2016).
6. Van der Waals interactions – i.e. interactions among the hydrophobic moieties of the urine-added organic molecules and the polyaromatic organic parts of the biochar) (Xiao & Pignatello, 2015).

The main N species in cow urine and their respective total content are as follows: urea (16·2%), hippuric acid (7·2%), allantoine (2·0%), creatinine (1·0%), creatine (0·8%), uric acid (0·4%) and various amino acids (0·78%) with glycine, taurine and alanine being the most abundant (Bristow *et al.*, 1992). All of these compounds are highly soluble and of comparably low-molecular weight and can thus easily infiltrate the biochars' porous system; they should adsorb well to the aromatic heterocyclic biochar ring systems mostly by water bridging, charge-assisted intramolecular hydrogen bonds and Van der Waals interactions. The adsorption capacity of biochar is probably lower for urea than for the other, more complex organic N species. However, urea sorbed in the biochar pore system is probably protected from urease enzyme degradation (González *et al.*, 2015), because the size of biochar pores (much in the order of 1·5 nm as shown by CO_2 adsorption measurements; Kupryianchyk *et al.*, 2016) is smaller than the molecular diameter of enzymes such as urease (Dixon *et al.*, 1980). Urease decomposes urea to ammonia, which is more easily leached or volatilized (depending on the soil pH and water content) than urea, and usually quickly nitrified into the mobile anion nitrate. A slow-down of urea hydrolysis within the biochar pore system, if it occurs, may contribute to a more gradual N release.

Compared with the addition of synthetic urea (Neilen *et al.*, 2016), cow or human urine has more diverse N species (Bristow *et al.*, 1992; Bouatra *et al.*, 2013), which likely modifies the adsorption behaviour within the biochar system, as those different N species are sorbed with varying strengths by different mechanisms. This sorption continuum for various N species has to be considered as major factor for the observed plant nutrition as the r*elease (delivery) rate* of nutrients to plants is more important for plant growth than *the total amounts* (stocks) of the respective nutrients (Ingestad, 1997). The essential property of a fertilizing substrate such as compost, nutrient-enriched biochar or any NPK granulate is not the total analytical amount and concentration but the nutrient amount supplied per unit of time in relation to the growth rate of the plant (Ingestad & Ågren, 1992; Ingestad & Ågren, 1995). The relative nutrient uptake rate thus strongly controls plant nutrition, almost independently of the concentration in the culture solution (Rastetter & Shaver, 1992; Ingestad, 1997).

It has been repeatedly shown that the required concentrations for adequate nutrient uptake and plant growth are much lower than the typical application rates in agriculture (Olsen, 1953; Ingestad, 1982; Pettersson, 1986; Ingestad & Ågren, 1992). It is, however, agronomically very challenging to distribute such low concentrations to the roots in a timely manner; this usually needs advanced technology like computer-aided fertigation. On the basis of the present field trial results, we hypothesize that the nutrient *flux* (i.e. delivery 'at the right moment' to plant growth needs) is improved when biochar is enriched with high-diversity dissolved organic nutrients and applied into the root zone, compared with nutrient enrichment with simple ionic NPK species, even if total amounts are lower than in conventional NPK fertilization. It is most likely that this nutrient flux from organic enriched biochar is aided by a surrounding soil environment, which has large and active gross N transformation rates from other organic pools such as SOC and/or compost.

Limitations, Implications and Research Needs Based on This Study

Climate, soil fertility, cultivation practices and field history between the sites varied largely, providing a multitude of various field-relevant conditions. However, all trials took place in silt loam or loamy-sand soils with relatively high SOC (1·5–5·4%) and acidic to neutral pH values. The proposed methods should therefore be tested in other soil types before general assumptions can be made. It cannot be excluded that the high and concentrated application rates of organic nitrogen in the urine and urine-biochar treatments, which have to be transformed microbially into plant available forms of nitrogen, is limited by the microbial availability of soil organic carbon and that therefore the organic biochar-based fertilization method would be less plant growth promoting in poor soils with low SOM. Moreover, most biochar field trials that have shown highest yield responses in the scientific literature were undertaken on acidic soils (Biederman & Harpole, 2013; Crane-Droesch *et al.*, 2013; Jeffery *et al.*, 2015). The effects of biochar amendments on alkaline soils have generally been lower and were in many cases not significant (Artiola *et al.*, 2012; Schmidt *et al.*, 2014; Ruysschaert *et al.*, 2016). However, based on the conclusion that the effect of low-dosage root-zone applied, nutrient-enriched biochar is mostly due to nutrient delivery effects, it is likely that the suggested method is also efficient in alkaline soils, although experimental confirmation is needed. The nutrient release dynamics of various mineral and organic nutrients in combination with various types of biochar have to be investigated systematically to optimize plant nutrition over the whole growing cycle.

In the trials presented here, the quantity of applied organic and mineral nutrients was defined according to general fertilizer recommendation for the respective crops. A next step would be to test optimal nutrient-to-biochar ratios depending on the organic and mineral nutrient form and the type of biochar. Moreover, the suitability and nutrient efficiency of other organic nutrient-rich liquids such as processing water from tofu or cheese production, yeast slurry from wine making, human urine or fermentation liquids of waste biomass should be investigated in combination with biochar for improving agronomic nutrient cycling and nutrient use efficiencies.

Currently, we suggest to repeat the low-dosage root zone application of nutrient-enriched biochars before planting or seeding of annual crops and to reapply it every 1 to 3 years for perennial crops. The repeated application of biochar will accumulate, and biochar concentrations in soils will likely reach about 40–50 t ha^{-1} over the next 50 years, which would correspond to nutrient-enriched biochar concentrations in Terra preta soils (Glaser *et al.*, 2001; Glaser & Birk, 2012). The long-term effect of accumulating nutrient-enriched biochar needs to be investigated systematically in the upcoming years; however, it can be expected that soil organic carbon, water and nutrient retention will increase from year to year enabling the reduction of fertilizer and of repeated biochar applications as for example seen in novel African Dark Earths (Solomon *et al.*, 2016).

While all 13 crops tested in this study responded positively to the root-zone application of biochar-based fertilizer, the yield increases were highly variable depending on the crop type, biochar, soil system and climate. More systematic trials with a larger spectrum of crop families are necessary to generalize the reaction patterns of different botanical plant families or species traits (e.g. mycorrhizal versus non-mycorrhizal) and adapt the method to the nutritional needs of particular crops.

CONCLUSION

It was demonstrated with a large number of field trials that organic and mineral nutrient enrichment of biochar and its subsequent low-dosage application (0·5 and 2 t ha^{-1}) to the root zone of various crops considerably increased crop yields compared with the same fertilization type and amount without biochar. Organic nutrient enrichment of biochar was consistently more effective than mineral nutrient enrichment opening up new perspectives for the future of organic farming.

The yields obtained in the cow urine-biochar treatments were consistently at the higher end or exceeding those of optimized conventional farming (Table S1), demonstrating that organic biochar-based fertilization methods are able to optimize plant nutrition and hence crop productivity. The methods described and demonstrated here were effective on relatively fertile soils in 'productive' climates and may thus have the potential to develop into a new, long-term approach for sustainable, CO_2-negative (i.e. C-sequestering) and nutrient-efficient agriculture instead of being only a solution for highly degraded soils in suboptimal agronomic systems. Organic biochar-based fertilizers can easily and at high quality be produced on-farm, recycling organic (waste) nutrients and waste biomass carbon, reducing the ecological footprint of agriculture and improving on-farm economics and in particular small-scale farmer income and independence.

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SUPPORTING INFORMATION

Additional Supporting Information may be found online in the supporting information tab for this article.

Table S1. Summary of all presented field trials listing crop species, field size, number of replications per treatment, biochar feedstock, geographical location and altitude above sea level. Yield or growth of the best biochar containing treatment is compared with the equally fertilized control, the level of significance is given with the employed statistical test and the yield is compared with reference values per hectare; literature references for the later value are give in the results section. The first gray section (trials 1 to 13) shows experiments where organic treatments are compared, the second section (trials 14 to 17) compares mineral fertilizer treatments and the third section contains trials where organic treatments are compared with mineral fertilizer treatments. Primary trials are designated with a 'P' and village trials with a 'V' in the second column.