Effects of *Leucaena* biochar addition on crop productivity in degraded tropical soils

*Dafydd M. O Elias[a], Gin Teng Ooi[b], Mohammad Fadhil Ahmad Razi[b], Samuel Robinson[c], Jeanette Whitaker[a], Niall P. McNamara[a]

[a]UK Centre for Ecology & Hydrology, Lancaster Environment Centre, Library Avenue, Bailrigg, Lancaster, LA1 4AP, United Kingdom

[b]Crops For the Future, Jalan Broga, 43500 Semenyih, Selangor Darul Ehsan, Malaysia (http://www.cffresearch.org/)

[c]Lancaster Environment Centre, Library Avenue, Lancaster University, Lancaster LA1 4YQ, United Kingdom

*Corresponding Author: Dafydd Elias

Email: dafias@ceh.ac.uk

Abstract

Biochar has the potential to increase crop yields on degraded, tropical soils. It can be readily produced in rural community settings using low-cost technology and is most economically feasible if produced from local biomass or waste residues. Biochar was produced from *Leucaena* biomass using low-cost pyrolysis and sequential pot experiments were then conducted in Malaysia on three degraded soils. We first evaluated the effect of *Leucaena* biochar on yields of *Amaranthus*, a leafy vegetable crop and measured changes to soil pH and nutrient availability over two growth cycles. We then tested whether any yield response to biochar was dependent upon the rate of biochar or fertilizer application. We found that biochar application at 30 t ha^{-1} with maximal fertilizer increased yields between 17-53 % on very strongly acidic soil. Biochar added at 15 t ha^{-1} with maximal fertilizer increased yield by 54 % on strongly acidic soil whilst there was no significant yield response on fertilized, slightly acidic soil. Unfertilized biochar treatments showed small yield responses across all soils over 2 growth cycles (9-11 %), but yields were much lower than in fertilized treatments. Biochar also decreased short-term N availability when applied with fertilizers, which may improve nitrogen retention and substantially increased soil pH. This may reduce mobility of Fe, Mn and Al ions, which were negatively associated with yield. Our results suggest that *Leucaena* biochar can elicit a positive crop yield response but only when combined with fertilizer additions on very strongly to strongly acidic tropical soils.

Keywords:

Biochar, Malaysia, Yield, Degraded Tropical Soils, Soil pH, Food Security

1. Introduction:

Global human population is expected to increase to over 9 billion by 2050 with food demand projected to rise by 70 – 100 % as a result [1]. In developing countries, at current levels of productivity, agricultural land area under cultivation will need to increase substantially to meet this demand, with severe implications for natural ecosystems, particularly in tropical regions [2-4]. Closing the current yield gap between potential and realized crop yields could dramatically reduce the requirement for further agricultural expansion, whilst also providing a pathway to poverty alleviation. Biochar has attracted significant interest as an agricultural soil amendment due to its potential for increasing crop productivity, with additional benefits in terms of improved soil fertility and mitigation of climate change [5]. Given the possible benefits from biochar, its potential for supporting a range of the Sustainable Development Goals (SDG) of the United Nations is widely recognised [6].

When applied to soils, biochar interacts with soil physical, chemical and biological properties, potentially conferring improvements to soil quality and crop productivity [7]. It also can provide long term carbon (C) sequestration due to the stable, recalcitrant nature of organic C relative to the original biomass [8]. Biochar addition to soils can improve soil physical properties and water retention by increasing aggregate stability, reducing bulk density and hydraulic conductivity [9, 10]. This may promote nutrient retention by reducing leaching of nutrients in soil solution [11]. Biochar may also ameliorate soil acidity, which in turn can alleviate ion toxicity and increase availability of many soil nutrients [12]. These effects on soil properties have been suggested as a mechanism for the average ~10 % increase in crop yields as a result of biochar addition [13, 14].

However, crop productivity responses to biochar addition are highly variable, with some studies showing positive effects whilst others found neutral to negative effects [13, 15]. The inconsistencies in crop yield responses may be attributable to variation in the properties of biochar applied (due to feedstocks used and specific pyrolysis conditions) and the time since application [16-18]. However, initial soil properties have shown to be stronger predictors of yield response with pH, cation exchange capacity (CEC) and organic carbon negatively correlated and clay content positively associated with crop yield response to biochar addition [15]. This is significant as 30 % of the world's soils are classified as acidic, which includes over 50 % of potential arable land [19]. These soil characteristics are also typically associated with highly weathered and degraded soils across the humid tropics, with a recent meta – analysis showing that biochar addition resulted in a ~25 % increase in crop yields in the tropics whilst there was no significant effect of addition on temperate soils [20].

Despite the potential for biochar application on tropical soils, for its adoption by rural smallholder farmers in developing countries to be economically feasible, small-scale decentralised pyrolysis technology must be utilised with feedstocks obtained locally and a focus on higher-value (non-cereal) crop yield improvement [21, 22]. We therefore tested whether a low-cost, locally produced biochar, using *Leucaena leucocephala* biomass, could elicit a crop yield response in *Amaranthus* across three degraded agricultural soils from Malaysia with low fertility and a range of soil pH. *Leucaena* is a ruderal, fast-growing, tropical leguminous tree, which is drought tolerant, capable of growing across a wide range of soils, pH and is currently utilised for forage and fuelwood production in tropical regions [23, 24]. This adaptability to a wide range of edaphic conditions and ability to grow on degraded lands makes it useful as a tool for land restoration and as a feedstock for biochar production [25]. Using a series of sequential pot experiments, we tested the following hypotheses:

H1: Addition of *Leucaena* biochar to tropical, degraded soils will increase crop yield irrespective of fertilizer addition, with greater yield responses at higher biochar addition rates.

H2: Crop yield response will be driven by the effects of *Leucaena* biochar on soil nutrient availability.

2. Methods:

2.1 - Soil Collection and Biochar Production

Agricultural soils were collected from the Crops for the Future field research centre (CFFRC), Malaysia (2.933162 N, 101.878028 W) located east of the town of Semenyih, Malaysia (Figure S1). The research centre agricultural land covers 12.8 ha and forms part of the Balau Estate, which is mainly planted with oil palm. The surrounding estate was first planted with rubber followed by two further rotations of oil palm. The sampling area was subject to one rotation of oil palm prior to being used as an oil palm nursery up until 2014. This was then cleared and used to grow maize, Bambara groundnuts and Napier grass. This area is low lying with high rates of surface runoff and soil erosion, resulting in low soil fertility (Personal Communication - Gin Teng Ooi). The climate is tropical and aseasonal with an average annual temperature of 27.2 °C. Eleven year mean annual rainfall ranged from 1454-2808mm with a mean of 1987mm (unpublished data). Three soils were sampled from this area representing a range of low fertility, degraded tropical soils. Two soils were taken from the Rengam series (Nitisol) which is a coarse, sandy clay derived from acid igneous parent material whilst one soil was a fine sandy clay loam derived from quaternary alluvium parent material. ~200 kg (Exp 1) and ~ 400 kg (Exp 2) of each soil were collected from the top 0 – 10 cm by hand. Five subsamples of each soil type were collected for initial physico-chemical analysis and field bulk density (using a 7.5 cm diameter ring to 5cm depth).

Biochar was produced from *Leucaena leucocephala* woody biomass in a custom made, low cost retort kiln (Model CR-570, Kenaboi Nature Resources, Puchelong Selangor, Malaysia)

(Figure S2 - S3) with a final pyrolysis temperature of 600-700°C held for 4 hours. This was then homogenized to pass through a 2mm sieve prior to application.

2.2 - Experimental Design and Setup

Two sequential experiments were conducted in a shade house located at CFFRC, Malaysia between 03 – 05/2018 (Exp 1) and 02 – 04/2019 (Exp 2). The first experiment explored the crop and soil nutrient response to a single dose of biochar using a standard recommended fertilisation rate. The second experiment expanded on this work to include two levels of biochar applications in combination with standard and reduced fertilisation rates to assess the potential for biochar to improve nutrient management. The three soils collected from CFFRC were firstly homogenised using a trowel/spade and hand mixed with any large stones or coarse woody debris removed. 9 kg of soil was added to 10 L plastic pots and treatments (biochar and fertilizer applications) were applied in a fully factorial design with all combination of treatments being applied to all three soil types. For Exp 1, two rates of biochar (0 and 30 t ha^{-1}) and fertilizer (0 and 0.4 t ha^{-1} NPK (15:10:10), 25 t ha^{-1} poultry manure) addition were applied to each soil (n = 60) (Table 1). For Exp 2, three rates of biochar (0, 15 and 30 t ha^{-1}) and fertilizer (0.4, 0.2 and 0.1 t ha^{-1} NPK (15:10:10) and 25, 12.5, 6.25 t ha^{-1} poultry manure) addition were applied to each soil (n = 135) (Table 1). Fertilizer application rates were based on local growers guidelines [26].

Both biochar and fertilizer were fully incorporated by hand mixing. Water was then added to achieve 75 % of maximum water holding capacity and pots were arranged in the shade house as 5 blocks of 12 (Exp 1: 3 soil types x 4 treatments) and 27 (Exp 2: 3 soil types x 9 treatments) for randomized complete block designs. 1 g of *Amaranthus Tricolor* seeds were dispersed across the soil surface, lightly covered in soil, watered and left to germinate. This plant was selected because it is grown as a leafy vegetable across the region. These were thinned to 5 plants which were then left to grow to maturity over 35 days. Plants experienced ambient photoperiod, but soil moisture was maintained throughout the experiment using an automated drip feed system and weekly water additions back to initial pot weight. Air temperature within the shade house was measured throughout the experiments using Hobo pendant loggers (Onset Computer Corporation, MA, USA) (Table S1). During Exp 1 and Exp 2 the mean air temperature was 30.87 ± 0.05 °C and 30.50 ± 0.01 °C respectively. After 35 days, mature *Amaranthus* plants were harvested by cutting aboveground biomass (AGB) level with the soil surface and drying in an oven at 40 °C until constant weight. For Exp 1, 1g of *Amaranthus Tricolor* seed were re-sown as described above and a repeat application of inorganic fertilizer was applied as a top dressing (0.4 t ha^{-1} NPK). These will subsequently be referred to as Exp 1a (growth cycle 1) and Exp 1b (growth cycle 2).

2.3 – Soil Analysis

Maximum soil water holding capacity was measured for the three soils in triplicate, calculated as the amount of water remaining in the soil after being saturated and left to drain for 24 h in a fully humid airspace [27]. Soil bulk density was determined by drying at 105°C after sieving to 2mm to remove roots and stones [28]. Subsamples of soil and biochar were characterised for pH, total C, N, inorganic P and K. Soil pH was determined on 5 fresh soil samples and biochar using a calibrated pH meter (Hanna Instruments, UK) in a soil - water suspension (1:2.5 ratio of soil to deionised water) after stirring and standing for 30 min. The remaining soil was air dried. Inorganic P was extracted from air dried soils and biochar using 0.5M Sodium Bicarbonate, (Olsen P). K was extracted using 0.1M magnesium acetate. P and K were then determined using a calibrated and blank corrected SoilTest 10 spectrophotometer (Palintest, Gateshead, UK). Subsamples for total C and N were dried at 105°C for 24 hours, sieved to pass a 2mm sieve and ground to a fine powder using a pestle and mortar. These were analysed using dry combustion at 980°C using a LECO Truspec Micro (LECO

Corporation, Michigan, USA). Following biomass harvesting at the end of the experiment, fresh soil samples were collected from each pot and pH measured using the method described above.

2.4 - Soil nutrient availability

At the end of Exp 1a, Exp 1b and Exp 2, soil nutrient availability was assessed using commercial ion exchange membranes (PRS^{TM} Probes, Western AG, Saskatoon, Canada) after biomass harvesting [29]. Pairs of plastic probes (1 anion and 1 cation exchange) housing the membranes were installed within every pot to 0-10cm depth for 24 hours to measure the availability of N, P, K, Mg and micronutrient ions (S, Fe, Mn, Al) in soil solution. Once removed, probes were washed thoroughly with deionised water and shipped to the manufacturer for analysis. Probes were eluted using 0.5M HCl for 1 hour. NO_3^- and NH_4^+ were measured colorimetrically using automated flow injection analysis (FIA). All other elements were analysed using inductively coupled plasma-optical emission spectroscopy (ICP-OES). Results are reported as supply rates per area of membrane available for ion exchange over the burial period (µg/10cm^2/day). In the absence of competing sinks such as plant roots, these membranes mimic plant root uptake dynamics and provide an estimate of potential soil nutrient supply.

2.5 – Statistical Analysis

All statistical analyses were performed using R (Version 3.5.2). Differences between initial soil properties were tested using one-way ANOVA and Tukey's honest significance difference test. To test hypothesis 1 that addition of *Leucaena* biochar would stimulate crop yield, we used ANOVA with AGB as a response variable, soil type, biochar application and fertilizer application rate as explanatory variables and their interactions. Experimental block was included as a covariate to account for potential differences in microclimatic conditions and growth rates within the shade house. Normality was checked by inspecting Q-Q plots and plotting residuals vs fitted values. For Exp 1a and Exp 1b, AGB was log transformed whilst for Exp 2 it was square root transformed to achieve normality of residuals. Where significant treatment effects were detected, post-hoc multiple pairwise comparisons were performed using estimated marginal means (emmeans) [30] implemented in the R package emmeans [31]. P-values were adjusted for multiple comparisons using the Tukey method.

To test hypothesis 2 that crop yield response would be driven by the effects of *Leucaena* biochar on soil pH and nutrient availability we first determined the effect of biochar on soil pH and nutrients using ANOVA with soil pH and each soil nutrient as response variables. Soil type, biochar application and fertilizer application rate were included as explanatory variables with interactions. We then used multiple linear regression with AGB as the response variable, soil pH and nutrient availability (inorganic N, P, K, Mg, S, Fe, Mn, Al) as explanatory variables. Fe and Mn were summed as their availability was highly correlated (r >0.9). Biochar application rate was also specified as a factor within the models to account for the unmeasured, potential physical effects of biochar application to soils and interaction terms between nutrients and biochar were also included. Variables were selected using a forward-backward stepwise procedure using AIC score as a selection criterion [32]. To determine the relative importance of predictors, averaging over order of regressors was performed using the *Relaimpo* R package [33]. To visualise interactions within models, partial regression plots were produced using the *visreg* R package [34].

3. Results:

3.1 – Initial soil and biochar properties

The three soils used in the mesocosm experiments varied predominantly in terms of their pH ($F_{2,12} = 32.58$, $p = <0.001$), being classified as very strongly acidic (pH – 5.08 ± 0.13), strongly acidic (5.45 ± 0.06) and slightly acidic (6.14 ± 0.08) (Table 2) [35]. Total C ($F_{2,12}$ = 40.72, p = <0.001), N ($F_{2,12}$ = 14.45, p = <0.001) and K content ($F_{2,12}$ = 35.06, p = <0.001) also varied between land uses. However, the magnitude of differences were small and all soils had extremely low soil C and N (<1.38 % and N < 0.11 %), indicating low soil quality [36]. Soil P concentrations ($F_{2,12}$ = 0.75, p = 0.49) and field bulk density ($F_{2,12}$ = 2.75, p = 0.10) did not vary between soils (Table 2). *Leucaena* biochar was strongly alkaline with high C, N and K concentrations relative to all soils (Table 2).

3.2 - The effect of Leucaena biochar application rate and fertilization on aboveground biomass

During Exp 1a, biochar addition was a significant predictor of AGB (Figure 1A, Table 3) (p = 0.009). Post-hoc pairwise comparisons revealed that the response to biochar addition varied according to soil type. A significant crop yield response was observed when added to the very strongly acidic soil with AGB increased by ~17 % from 15.99 ± 0.90 to 19.24 ± 1.08 g, in combination with fertilizer addition (*emmeans*: p = 0.02) (Figure 1A). However, there was no effect of biochar addition on AGB when added to strongly acidic (No Biochar: 18.65 ± 1.05, Biochar: 18.98 ± 1.07, p = 0.83) or slightly acidic soil (No Biochar: 16.64 ± 0.94, Biochar: 16.53 ± 0.93, p = 0.94) with fertilizer addition (Figure 1A). Averaged across all soils, biochar addition increased AGB without fertilizer by ~11 % (No Biochar: 6.74 ± 0.22, Biochar: 7.56 ± 0.25 g dry weight biomass, *emmeans*: p = 0.02) but yields were less than half of those in fertilized treatments (Figure 1A). During Exp 1b the effect of biochar on AGB was dependent on fertilization (biochar x fertilizer: p = 0.01) (Table 3) as biochar increased AGB yield across all soils without fertilizer addition (~9 %) (No Biochar: 3.98 ± 0.10, Biochar: 4.37 ± 0.12 g dry weight biomass, *emmeans*: p = 0.02) but did not influence yield when combined with fertilizer (Figure 1B). Exp 2 showed that biochar application, fertilizer addition and soil type were all significant predictors of *Amaranthus* AGB (Table 3). However, a three-way interaction between biochar, fertilizer and soil type indicated that the effect of biochar on AGB was dependent upon fertilizer addition and this effect varied between soils ($p = 0.03$) (Table 3). Post-hoc pairwise comparisons showed that biochar increased AGB by ~53 % (relative to no biochar application) when applied to very strongly acidic soil at 30 t ha^{-1} in combination with the highest rate of fertilizer application (No Biochar: 7.81 ± 1.52, 30 t ha^{-1} Biochar: 16.25 ± 2.22 g dry weight biomass, *emmeans*: $p = 0.006$) (Figure 1C). Biochar application also increased AGB by ~54 % (relative to no biochar application) when applied to the strongly acidic soil at 15 t ha^{-1}, in combination with the highest rate of fertilizer addition (No Biochar: 7.95 ± 1.54, 15 t ha^{-1} Biochar: 17.30 ± 2.29 g dry weight biomass, *emmeans*: $p = 0.003$) (Figure 1C). There was no significant effect of biochar application on the slightly acidic soil, irrespective of the rate of biochar or fertilizer addition (Figure 1C).

3.3 – Biochar effects on soil nutrients and pH as drivers of aboveground biomass

Soil pH was significantly increased by both biochar application and fertilizer addition (Figure 2, Table 4) although an interaction between biochar and fertilizer indicated that biochar increased soil pH more strongly in the absence of fertilizer than in combination (Exp 1a: $p = 0.02$, Exp 1b: $p = 0.001$, Exp 2: $p = 0.05$). Addition of *Leucaena* biochar increased soil inorganic N availability without fertilizer addition and decreased N availability with fertilizer (Exp 1a: $p = 0.07$, Exp 1b: $p = 0.009$, Exp 2: $p = 0.001$) (Table S2, Figure 3A, D, Figure 4A). In contrast, soil K availability was significantly increased by biochar addition (Exp 1a: $p = <0.001$, Exp 1b: $p = <0.001$, Exp 2: $p = <0.001$) and accounted for ~15-73 % of explained variation (Table S3, Figures 3C, F, Figure 4C). Soil P availability was not explained by biochar addition and was explained overwhelmingly by the rate of fertilization throughout both experiments (Exp 1a: $p = <0.001$, Exp 1b: $p = <0.001$, Exp 2: $p = <0.001$) (Table S4, Figures 3B, E, Figure 4B). Soil Mg was increased by biochar in Exp 1a and Exp 1b although the effect was greatest without fertilizer (Biochar x Fertilizer: Exp 1a: $p = 0.02$, Exp 1b: $p =$

<0.001) (Table S5, Figure 3D, H). Soil S, Fe, Mn and Al were not influenced by biochar addition.

Variance in AGB was positively related to Mg availability (Exp 1a: $p = <0.001$, Exp 1b: $p = 0.02$, Exp 2: $p = <0.001$) and negatively associated with Fe + Mn availability (Exp 1a: $p = 0.03$, Exp 1b: $p = <0.001$, Exp 2: $p = 0.005$) across all experiments (Table 5). Mg availability explaining between 12.35-37.85 % of explained variation in AGB whilst Fe + Mn explained between 3.39-7.45 % (Table 5). Biochar addition was also a significant predictor of AGB ((Exp 1a, Exp 1b, Exp 2: $p = <0.001$) and explained between 2.35-10.71 % of explained variation in AGB (Table 5) whilst interactions between inorganic N, S (Exp 1b), Mg, Al (Exp 2) and biochar also explained variation in AGB. In Exp 1b, greater inorganic N availability was associated with increased AGB without biochar and reduced AGB with biochar ($p = 0.005$) (Figure S6) whilst the positive effect of S on AGB was stronger with biochar addition ($p = 0.005$) (Figure S7). In Exp 2, higher soil Al was associated with lower AGB without biochar but had little effect on AGB with biochar added at 30 t ha^{-1} ($p = 0.04$) (Figure S8), whilst the positive effect of Mg on AGB was marginally stronger with higher rates of biochar addition ($p =0.09$) (Figure S9). Across Exp 1a and Exp 2, AGB was positively related to soil P whilst, K availability was negatively related to AGB (Table 5).

Discussion

Biochar has been proposed as a potential soil amendment to improve soil quality and increase crop yields, particularly on degraded or highly weathered tropical soils [20, 37, 38]. However, for biochar to be an economically feasible and scalable solution, it must confer crop productivity benefits, be produced from feedstocks available locally and using low-cost, decentralized pyrolysis technologies [21]. We produced biochar from *Leucaena* using a low-cost retort kiln which, although offering limited control over pyrolysis conditions, is particularly suited for use in low and middle income countries and in smallholder farming systems as a shared community resource [39]. We then performed a series of mesocosm experiments to assess the potential of locally produced *Leucaena* biochar to improve soil quality and crop yields across three degraded tropical soils in Malaysia.

Our findings did not support hypothesis 1 as we found that biochar addition at 30 t ha^{-1} only had a substantial yield benefit (+17-53 % in AGB across two sequential experiments) when applied with fertilizer and on very strongly acidic soils (Figure 1). This agrees with many previous studies that have shown the potential for biochar to increase crop yields is greatest on the most acidic, tropical soils [15, 20, 40]. However, a previous study using biochar produced from low-cost pyrolysis on tropical soil found a greater yield response to biochar on acidic soils under nutrient limited conditions [41]. Our results contradict these findings as the level of yield stimulation was highly dependent upon the rate of fertilization, with the greatest yield response observed with a maximal dose of nutrients (Figure 1). When we applied biochar and fertilizer at multiple rates (Exp 2), there was no clear relationship between biochar addition rate and AGB (Figure 1). In very strongly acidic soils, biomass increased by 53 % at a rate of 30 t ha^{-1} whilst crop yields were increased by 54 % on strongly acidic soils at 15 t ha^{-1} with no significant yield response on slightly acidic soils (Figure 1C). Previous studies have demonstrated conflicting relationships between biochar application rate and crop yields with positive, neutral or negative associations with yield being reported, with increased rates of biochar addition [14, 42-45]. The lack of relationship between biochar dose rate and crop productivity may be related to soil-biochar interactions which can regulate soil pH and nutrient availability [46].

We hypothesised that any crop yield response would be driven by the positive effects of biochar on soil nutrient availability (Hypothesis 2). However, in disagreement we found that only soil K availability was consistently increased by biochar application (Figure 3-4) and this was negatively associated with AGB (Table 5). Increased soil K availability likely reflects direct supply from biochar addition as *Leucaena* biochar K concentrations were high (Table 2) relative to all soils and studies of nutrient release dynamics from other biochar's have shown that K is rapidly leached from biochar ash into soil solution [47, 48]. As we found strong positive relationships between Mg availability and AGB across all experiments, the negative relationship between K and AGB may be due to nutrient antagonism as high K concentrations may interfere with root Mg uptake [49].

We did find that when biochar was applied with fertilizer, soil inorganic N was reduced and there appeared to be a negative relationship between soil inorganic N and the rate of biochar application (Figures 3A, Figure 4A, Table S2). As we used ion exchange membranes which depend on equilibrium dynamics [29], this may be explained by a slower N release from biochar amended soils. Our findings agree with previous studies that have observed higher nutrient retention and lower soil available N after biochar additions to soil [50-52] and are consistent with studies showing that biochar can act as a slow-release ion sink at low dose rates [53], whilst at higher application rates it may effectively immobilize N [53-57].

In highly weathered tropical soils, Fe, Mn and Al commonly accumulate at levels which cause toxicity to plants [58, 59]. We found that soil Fe, Mn and Al were negatively related to AGB (Table 5). Although we did not find an effect of *Leucaena* biochar addition on soil Fe, Mn or Al concentrations, the effect of Al on AGB was negated at the highest biochar application rate (Figure S8). As Al solubility decreases with increasing pH and the *Leucaena* biochar used in this study was strongly alkaline (Table 1) (pH 10) (Figure 2, Table 4), its application may therefore be useful to alleviate toxicity in strongly acidic tropical soils.

4. Conclusions

Our results show that biochar produced locally using *Leucaena* biomass and low-cost pyrolysis can elicit a positive crop yield response but only on very strongly to strongly acidic tropical soils, in combination with fertilizer application. Biochar decreased short-term soil N availability when applied with fertilizers, which may improve nitrogen use efficiency by reducing rates of nitrogen leaching. *Leucaena* biochar also substantially increased soil pH. This may benefit crop productivity as increased soil pH reduces the mobility of Fe, Mn and Al ions, which we found were negatively associated with yield.

Acknowledgments

We thank Ms Chong Yuet Tian for assisting with the experimental work. We also thank Peter Gregory and Chin Ong Kooi for their advice and support.

Conflict of Interest

The authors declare no conflict of interest

Funding

The experiments in 2018 were supported by the Supergen Bioenergy Hub Flexibility Funding Programme (Engineering and Physical Sciences Research Council (EPSRC) & Department for Transport (DfT); Grant ref. R121197). The experiment in 2019 was supported by the UK Natural Environment Research Council grant Sustainable Use of Natural Resources to Improve Human Health and Support Economic Development (SUNRISE, Grant No. NE/R000131/1).

References

[1] H.C.J. Godfray, J.R. Beddington, I.R. Crute, L. Haddad, D. Lawrence, J.F. Muir, J. Pretty, S. Robinson, S.M. Thomas, C. Toulmin, Food Security: The Challenge of Feeding 9 Billion People, Science 327(5967) (2010) 812-818.

[2] A. Balmford, R.E. Green, J.P.W. Scharlemann, Sparing land for nature: exploring the potential impact of changes in agricultural yield on the area needed for crop production, Global Change Biology 11(10) (2005) 1594-1605.

[3] K. Klein Goldewijk, A. Beusen, J. Doelman, E. Stehfest, New anthropogenic land use estimates for the Holocene; HYDE 3.2, Earth System Science Data Discussions (2016) 1-40.

[4] F.A.O.o.t.U. Nations, FAOSTAT statistical database -Land Use, 2019.

[5] D. Woolf, J.E. Amonette, F.A. Street-Perrott, J. Lehmann, S. Joseph, Sustainable biochar to mitigate global climate change, Nat. Commun. 1 (2010).

[6] K. Mehmood, E. Chávez Garcia, M. Schirrmann, B. Ladd, C. Kammann, N. Wrage-Mönnig, C. Siebe, J.M. Estavillo, T. Fuertes-Mendizabal, M. Cayuela, G. Sigua, K. Spokas, A.L. Cowie, J. Novak, J.A. Ippolito, N. Borchard, Biochar research activities and their relation to development and environmental quality. A meta-analysis, Agron. Sustain. Dev. 37(3) (2017) 22.

[7] B. Glaser, J. Lehmann, W. Zech, Ameliorating physical and chemical properties of highly weathered soils in the tropics with charcoal – a review, Biology and Fertility of Soils 35(4) (2002) 219-230.

[8] J. Lehmann, J. Gaunt, M. Rondon, Bio-char Sequestration in Terrestrial Ecosystems – A Review, Mitigation and Adaptation Strategies for Global Change 11(2) (2006) 403-427.

[9] M.O. Omondi, X. Xia, A. Nahayo, X. Liu, P.K. Korai, G. Pan, Quantification of biochar effects on soil hydrological properties using meta-analysis of literature data, Geoderma 274 (2016) 28-34.

[10] Z. Liu, B. Dugan, C.A. Masiello, R.T. Barnes, M.E. Gallagher, H. Gonnermann, Impacts of biochar concentration and particle size on hydraulic conductivity and DOC leaching of biochar–sand mixtures, Journal of Hydrology 533 (2016) 461-472.

[11] Y. Zhou, F. Berruti, C. Greenhalf, X. Tian, H.A.L. Henry, Increased retention of soil nitrogen over winter by biochar application: Implications of biochar pyrolysis temperature for plant nitrogen availability, Agriculture, Ecosystems & Environment 236 (2017) 61-68.

[12] J.H. Yuan, R.K. Xu, The amelioration effects of low temperature biochar generated from nine crop residues on an acidic Ultisol, Soil Use Manage. 27(1) (2011) 110-115.

[13] S. Jeffery, F.G.A. Verheijen, M. van der Velde, A.C. Bastos, A quantitative review of the effects of biochar application to soils on crop productivity using meta-analysis, Agriculture, Ecosystems & Environment 144(1) (2011) 175-187.

[14] L.A. Biederman, W.S. Harpole, Biochar and its effects on plant productivity and nutrient cycling: a meta-analysis, Global Change Biology Bioenergy 5(2) (2013) 202-214.

[15] A. Crane-Droesch, S. Abiven, S. Jeffery, M.S. Torn, Heterogeneous global crop yield response to biochar: a meta-regression analysis, Environ. Res. Lett. 8(4) (2013).

[16] E. Sagrilo, S. Jeffery, E. Hoffland, T.W. Kuyper, Emission of CO2 from biochar-amended soils and implications for soil organic carbon, GCB Bioenergy 7(6) (2015) 1294-1304.

[17] M.L. Cayuela, S. Jeffery, L. van Zwieten, The molar H:Corg ratio of biochar is a key factor in mitigating N2O emissions from soil, Agriculture, Ecosystems & Environment 202 (2015) 135-138.

[18] G. Cornelissen, Jubaedah, N.L. Nurida, S.E. Hale, V. Martinsen, L. Silvani, J. Mulder, Fading positive effect of biochar on crop yield and soil acidity during five growth seasons in an Indonesian Ultisol, Sci. Total Environ. 634 (2018) 561-568.

[19] H.R. von Uexküll, E. Mutert, Global extent, development and economic impact of acid soils, Plant Soil 171(1) (1995) 1-15.

[20] S. Jeffery, D. Abalos, M. Prodana, A.C. Bastos, J.W. van Groenigen, B.A. Hungate, F. Verheijen, Biochar boosts tropical but not temperate crop yields, Environ. Res. Lett. 12(5) (2017) 053001.

[21] S. Robb, S. Joseph, A. Abdul Aziz, P. Dargusch, C. Tisdell, Biochar's cost constraints are overcome in small-scale farming on tropical soils in lower-income countries, Land Degradation & Development.

[22] K. Pratt, D. Moran, Evaluating the cost-effectiveness of global biochar mitigation potential, Biomass and Bioenergy 34(8) (2010) 1149-1158.

[23] R.S. Zeigler, S. Pandey, J. Miles, L.M. Gourley, S. Sarkarung, Advances in the selection and breeding of acid-tolerant plants: Rice, maize, sorghum and tropical forages, in: R.A. Date, N.J.

Grundon, G.E. Rayment, M.E. Probert (Eds.), Plant-Soil Interactions at Low pH: Principles and Management: Proceedings of the Third International Symposium on Plant-Soil Interactions at Low pH, Brisbane, Queensland, Australia, 12–16 September 1993, Springer Netherlands, Dordrecht, 1995, pp. 391-406.

[24] E. Hutton, Selection and breeding of leucaena for very acid soil, Leucaena Research in the Asian Pacific Region: proceedings of a workshop held in Singapore, 23-26 Nov. 1982, IDRC, Ottawa, ON, CA, 1983.

[25] P. Jha, S. Neenu, I. Rashmi, B.P. Meena, R.C. Jatav, B.L. Lakaria, A.K. Biswas, M. Singh, A.K. Patra, Ameliorating Effects of Leucaena Biochar on Soil Acidity and Exchangeable Ions, Communications in Soil Science and Plant Analysis 47(10) (2016) 1252-1262.

[26] W.A. Ebert, T.H. Wu, S.T. Wang, International Cooperators Guide - Vegetable amaranth (Amaranthus L.), International Coopators Guide, AVRDC - The World Vegetable Center.

[27] R. Öhlinger, E. Kandeler, Methods in Soil Physics, in: F. Schinner, R. Öhlinger, E. Kandeler, R. Margesin (Eds.), Methods in Soil Biology, Springer Berlin Heidelberg, Berlin, Heidelberg, 1996, pp. 385-395.

[28] B. Emmett, Z. Frogbrook., P. Chamberlain., R. Griffiths., R. Pickup., J. Poskitt., B. Reynolds., E. Rowe., P. Rowland., D. Spurgeon., J. Wilson., C. Wood., Countryside Survey Technical Report No. 03/07, 2008.

[29] P. Qian, J.J. Schoenau, Practical applications of ion exchange resins in agricultural and environmental soil research, Canadian Journal of Soil Science 82(1) (2002) 9-21.

[30] S.R. Searle, F.M. Speed, G.A. Milliken, Population Marginal Means in the Linear Model: An Alternative to Least Squares Means, The American Statistician 34(4) (1980) 216-221.

[31] R. Lenth, emmeans: Estimated Marginal Means, aka Least-Squares Means. R package version 1.4.1, 2019.

[32] T. Yamashita, K. Yamashita, R. Kamimura, A Stepwise AIC Method for Variable Selection in Linear Regression, Communications in Statistics - Theory and Methods 36(13) (2007) 2395-2403.

[33] U. Groemping, Relative Importance for Linear Regression in R: The Package relaimpo, Journal of Statistical Software; Vol 1, Issue 1 (2007) (2006).

[34] P.J. Breheny, W. Burchett, Visualization of Regression Models Using visreg, R J. 9 (2017) 56.

[35] N.R.C.S. USDA, Soil Quality Information Sheet - Soil Quality Indicators: pH, in: N.R.C.S. USDA (Ed.) 1998.

[36] P. Musinguzi, J. Tenywa, P. Ebanyat, M. Tenywa, D. Mubiru, T. Basamba, A. Leip, Soil Organic Carbon Thresholds and Nitrogen Management in Tropical Agroecosystems: Concepts and Prospects, Journal of sustainable development 6 (2013) 32-43.

[37] M. Laghari, M.S. Mirjat, Z. Hu, S. Fazal, B. Xiao, M. Hu, Z. Chen, D. Guo, Effects of biochar application rate on sandy desert soil properties and sorghum growth, CATENA 135 (2015) 313-320.

[38] J.M. Kimetu, J. Lehmann, S.O. Ngoze, D.N. Mugendi, J.M. Kinyangi, S. Riha, L. Verchot, J.W. Recha, A.N. Pell, Reversibility of Soil Productivity Decline with Organic Matter of Differing Quality Along a Degradation Gradient, Ecosystems 11(5) (2008) 726.

[39] C.R. Lohri, H.M. Rajabu, D.J. Sweeney, C. Zurbrügg, Char fuel production in developing countries – A review of urban biowaste carbonization, Renewable and Sustainable Energy Reviews 59 (2016) 1514-1530.

[40] X. Liu, A. Zhang, C. Ji, S. Joseph, R. Bian, L. Li, G. Pan, J. Paz-Ferreiro, Biochar's effect on crop productivity and the dependence on experimental conditions—a meta-analysis of literature data, Plant Soil 373(1) (2013) 583-594.

[41] N.R. Pandit, J. Mulder, S.E. Hale, V. Martinsen, H.P. Schmidt, G. Cornelissen, Biochar improves maize growth by alleviation of nutrient stress in a moderately acidic low-input Nepalese soil, Sci. Total Environ. 625 (2018) 1380-1389.

[42] C.X. Sun, X. Chen, M.M. Cao, M.Q. Li, Y.L. Zhang, Growth and metabolic responses of maize roots to straw biochar application at different rates, Plant Soil 416(1) (2017) 487-502.

[43] N.V. Gale, T.E. Sackett, S.C. Thomas, Thermal treatment and leaching of biochar alleviates plant growth inhibition from mobile organic compounds, PeerJ 4 (2016) e2385.

[44] K.Y. Chan, L. Van Zwieten, I. Meszaros, A. Downie, S. Joseph, Using poultry litter biochars as soil amendments, Aust. J. Soil Res. 46 (2008).

[45] J. Major, M. Rondon, D. Molina, S.J. Riha, J. Lehmann, Maize yield and nutrition during 4 years after biochar application to a Colombian savanna oxisol, Plant Soil 333(1) (2010) 117-128.

[46] N.V. Gale, S.C. Thomas, Dose-dependence of growth and ecophysiological responses of plants to biochar, Sci. Total Environ. 658 (2019) 1344-1354.

[47] T.E. Angst, S.P. Sohi, Establishing release dynamics for plant nutrients from biochar, GCB Bioenergy 5(2) (2013) 221-226.

[48] L. Wang, C. Xue, X. Nie, Y. Liu, F. Chen, Effects of biochar application on soil potassium dynamics and crop uptake, Journal of Plant Nutrition and Soil Science 181(5) (2018) 635-643.

[49] M. Senbayram, A. Gransee, V. Wahle, H. Thiel, Role of magnesium fertilisers in agriculture: Plant-soil continuum, Crop and Pasture Science 66 (2015) 1219-1229.

[50] H.J. Xu, X.H. Wang, H. Li, H.Y. Yao, J.Q. Su, Y.G. Zhu, Biochar impacts soil microbial community composition and nitrogen cycling in an acidic soil planted with rape, Environ Sci Technol 48(16) (2014) 9391-9.

[51] L. Qian, L. Chen, S. Joseph, G. Pan, L. Li, J. Zheng, X. Zhang, J. Zheng, X. Yu, J. Wang, Biochar compound fertilizer as an option to reach high productivity but low carbon intensity in rice agriculture of China, Carbon Management 5(2) (2014) 145-154.

[52] A. El-Naggar, S.S. Lee, J. Rinklebe, M. Farooq, H. Song, A.K. Sarmah, A.R. Zimmerman, M. Ahmad, S.M. Shaheen, Y.S. Ok, Biochar application to low fertility soils: A review of current status, and future prospects, Geoderma 337 (2019) 536-554.

[53] H. Zheng, Z. Wang, X. Deng, S. Herbert, B. Xing, Impacts of adding biochar on nitrogen retention and bioavailability in agricultural soil, Geoderma 206 (2013) 32-39.

[54] N.V. Gale, M.A. Halim, M. Horsburgh, S.C. Thomas, Comparative responses of early-successional plants to charcoal soil amendments, Ecosphere 8(10) (2017) e01933.

[55] S. Rajkovich, A. Enders, K. Hanley, C. Hyland, A.R. Zimmerman, J. Lehmann, Corn growth and nitrogen nutrition after additions of biochars with varying properties to a temperate soil, Biology and Fertility of Soils 48(3) (2012) 271-284.

[56] P. Tammeorg, A. Simojoki, P. Mäkelä, F.L. Stoddard, L. Alakukku, J. Helenius, Biochar application to a fertile sandy clay loam in boreal conditions: effects on soil properties and yield formation of wheat, turnip rape and faba bean, Plant Soil 374(1) (2014) 89-107.

[57] G. Haider, D. Steffens, G. Moser, C. Muller, C.I. Kammann, Biochar reduced nitrate leaching and improved soil moisture content without yield improvements in a four-year field study, Agric. Ecosyst. Environ. 237 (2017) 80-94.

[58] E. Bojórquez-Quintal, C. Escalante-Magaña, I. Echevarría-Machado, M. Martínez-Estévez, Aluminum, a Friend or Foe of Higher Plants in Acid Soils, Front Plant Sci 8 (2017) 1767-1767.

[59] L.V. Kochian, O.A. Hoekenga, M.A. Pineros, How do crop plants tolerate acid soils? Mechanisms of aluminum tolerance and phosphorous efficiency, Annu Rev Plant Biol 55 (2004) 459-93.

Table Captions:

Table 1 – Summary of the factors and number of replicates for each experiment. N = total number of pots within each experiment.

Table 2 – Physical and chemical properties of three soils and chemical properties of biochar produced from *Leucaena* biomass in a low-cost, retort kiln. Soils data represent means (soil: n = 5) ± 1 standard error. Biochar pH, Total C and N represent means (n = 3) ± 1 standard error. Biochar extractable P and extractable K are n = 1.

Table 3 – Three-way ANOVA results for the effects of biochar addition, fertilizer addition and soil type on above ground biomass of 5 *Amaranthus Tricolor* plants. The response variable (above ground biomass) was log-transformed for results of Exp 1a and Exp 1b and square root transformed for results of Exp 2 to achieve normality of residuals. Two and three-way interaction terms were included, and experimental block was included as a covariate. R^2 = adjusted R^2.

Table 4 – Three-way ANOVA results for the effects of biochar addition, fertilizer addition and soil type on soil pH. The response variable (soil pH) was log-transformed to achieve normality of residuals. Two-way interaction terms were included, and experimental block was included as a covariate. Relative importance was calculated by averaging over orders of regressors and is presented as percentages of explained variation and sums to 100%. R^2 = adjusted R^2.

Table 5 – Multiple linear regression results for the effects of biochar addition and measured soil variables on above ground biomass. The response variable (above ground biomass) was log-transformed (Exp 1a and Exp 1b) and square root transformed (Exp 2) to achieve normality of residuals. Two-way interaction terms between soil pH and soil nutrients were included and experimental block was included as a covariate. Forward-backward stepwise

model selection was performed using AIC as a selection criterion. Relative importance was calculated by averaging over orders of regressors and is presented as percentages of explained variance which sum to 100%. R^2 = adjusted R^2.

Figure Captions:

Figure 1 - Dried above ground biomass of *Amaranthus Tricolor* after 35 days of growth during Exp 1a (A.), Exp 1b (B.) and Exp 2 (C.). Bars represent means (n = 5). Error bars represent ± 1 standard error. Points represent overlaid raw data.

Figure 2 - Soil pH measured after harvesting of biomass during Exp 1a (A.), Exp 1b (B.) and Exp 2 (C.).

Figure 3 - The availability of soil inorganic N, P, K and Mg measured using PRS ion exchange membranes over a 1 day burial period. Measurements were made after harvesting of biomass during Exp 1a (A-D) and Exp 1b (E-H). Bars represent means (n = 5). Error bars represent ± 1 standard error.

Figure 4 - Availability of soil inorganic N, P, K and Mg measured using PRS ion exchange membranes over a 24 hour burial period. Measurements were made after harvesting of biomass during Exp 2. Bars represent means (n = 5). Error bars represent ± 1 standard error.