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SOIL FERTILITY & PLANT NUTRITION

Does biochar improve nutrient availability in Ultisols of tree plantations in the Ecuadorian Amazonia?

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Abstract

The application of biochar to strongly weathered soils is thought to supply nutrients and improve nutrient retention. We hypothesized that biochar increases (a) total N, bioavailable macronutrient (NH_4–N, P, K, Ca, Mg), micronutrient (Fe, Mn, Zn, Cu), and plant-beneficial Na concentrations; and (b) nutrient retention in the topsoil. We grew the native leguminous Brazilian firetree [*Schizolobium parahyba* var. *amazonicum* (Ducke) Barneby] and the exotic beechwood (*Gmelina arborea* Roxb.) in a full factorial split-split-plot design at La Victoria and Los Zapotes, Ecuadorian Amazonia. The treatments included amendment of mineral fertilizer plus lime, 3 and 6 t ha^{-1} biochar (locally produced charcoal), and a control. We sampled the 0-to-0.25- and 0.25-to-0.50-m soil depth layers before the start of the experiment in 2009 and six times until 2013. The site at Los Zapotes was more fertile as reflected by a significant site effect on most studied soil properties in both depth layers. Biochar increased modified Olsen ($NaHCO_3$+EDTA)-extractable Ca ($p < .05$) and Zn concentrations ($p < .1$) and total N concentrations ($p < .05$) in topsoil. Mineral fertilizer plus lime increased Olsen-extractable P, K, Ca, Mg, and Zn concentrations (all $p < .05$) but reduced Olsen-extractable Fe concentrations ($p < .05$) in topsoil. Biochar increased Ca ($p < 0.1$) and Zn ($p < .05$) retention in mineral fertilized topsoils but decreased total N retention ($p < .05$) in unfertilized topsoils. The amendment of up to 6 t ha^{-1} biochar did not increase the fertility of the studied degraded Amazonian Ultisols sufficiently to enhance tree growth.

Abbreviations: BS, base saturation; ECEC, effective cation-exchange capacity; GAD, Gobierno Autonomo Decentralizado (Decentralized Autonomous Government); SOC, soil organic carbon; SOM, soil organic matter.

1 | INTRODUCTION

The fertile nutrient- and organic-matter-rich Terra Preta do Indio soils in the Amazon basin, which occur next to infertile, strongly weathered Oxisols and Ultisols, can in part be attributed to the historic long-term amendment of biochar (Cunha et al., 2009; Schulz & Glaser, 2012). This inspired the idea that biochar can generally be used to improve the fertility of degraded, strongly chemically weathered tropical soils (Glaser et al., 2000, 2001, 2002; Lehmann et al., 2003; Lima et al., 2002; Major et al., 2010). Biochar is charcoal produced as an agent for C sequestration from renewable and sustainable biomass by pyrolysis (Lehmann, 2007). The chemical composition of biochar varies widely. Biochar consists of a heterogeneous mixture of elemental C and aromatic structures formed during pyrolysis, metals, chemical compounds inherited from the source material, adsorbed volatiles, and ash (Brewer et al., 2009; Keiluweit et al., 2010; Spokas et al., 2011). Even biochars obtained from the same material under similar pyrolysis conditions but in different batches can have different chemical properties. Furthermore, differences in physical and chemical properties are a function of particle size in the same type of biochar (Francioso et al., 2011; Nocentini et al., 2010). Some biochars can contain high concentrations of toxic metals, which are concentrated from the feedstock biomass during pyrolysis or organic pollutants such as polycyclic aromatic hydrocarbons formed for example, by condensation of volatiles emitted during pyrolysis (Bieser & Thomas, 2019; IBI, 2015; Malev et al., 2016; C. Wang et al., 2017).

The amendment of biochar has been shown to potentially increase pH and the number of cation- and anion-exchange sites (Deluca et al., 2015; El-Naggar et al., 2019; Maia et al., 2011), increase soil organic carbon (SOC) concentrations (Apori & Byalebeka, 2021), soil water retention (Das & Sarmah, 2015), and nutrient availability (Berek, 2019; Berek & Hue, 2016; El-Naggar et al., 2019; Jeffery et al., 2017; Lefebvre et al., 2019). Moreover, biochar absorbed Al^{3+} (Hong & Lu, 2018) and legacy pesticides from previous land use thereby mitigating toxicities (L. Qian & Chen, 2014; Rizwan et al., 2016; Shi et al., 2019). Finally, biochar can provide a long-term effect because of its recalcitrance (Criscuoli et al., 2014; Hernandez-Soriano et al., 2016; J. Wang et al., 2016). Biochar can be produced from locally available feedstocks like fermentation residue (Maroušek, 2014), sewage sludge (Yue et al., 2017), and wood chips (Gonzalez Sarango et al., 2021).

The high deforestation rates in Ecuador, mainly because of conversion to pastures (Singh, 2010; Mosandl et al., 2008) and subsequent unsustainable pasture management (Roos et al., 2013) have resulted in the loss of surface soil organic matter and exchangeable cations, and thus a loss in soil fertility (Mainville et al., 2006). As a consequence, nutrient-poor and degraded Ultisols with low nutrient concentrations and a high risk of Al toxicity occur frequently in the sloping lands of the south Ecuadorian Amazonia region (Province of Zamora-Chinchipe, Machado et al., 2017; Valarezo et al., 1998). In the year 2015, the autonomous province government of Zamora-Chinchipe reported that 2,350 km^2 of their soils showed some degree of degradation and that the area of degraded soils increased at a rate of 86 km^2 yr^{-1} (GAD, 2015; Ministerio del Ambiente, 2017). These soils suffer from a strong risk of erosion (Valarezo et al., 1998). Therefore, adequate remediation approaches to recover the sloping degraded land are urgently needed.

Core Ideas

- Biochar increased total N and available Ca and Zn concentrations in topsoil.
- Biochar increased the retention of available Ca and Zn in mineral fertilized topsoils.
- Biochar decreased total N retention in unfertilized topsoils.
- Biochar amendment of up to 6 t ha^{-1} hardly improved the fertility of an Ultisol.

One option to recover the degraded land is reforestation, which at the same time counteracts the high deforestation rate and mitigates climate change through C sequestration (Cunningham et al., 2015; Marin-Spiotta & Sharma, 2013). To facilitate tree growth, soil fertility needs to be improved by the amendment of, for example, fertilizer, lime, or biochar to ensure quick establishment and good growth of tree plantations. Kishimoto and Suguira (1985) found that 5 yr of an annual application of 0.5 t ha^{-1} of charcoal increased the height of Japanese cedar [*Cryptomeria japonica* (L.f.) D. Don] trees by a factor of 1.26–1.35 and the biomass production by a factor of 2.31–2.36. Biochar has been shown to cause strong positive growth responses of 36 woody plant species from the tropical and boreal climate zones (Thomas & Gale, 2015). Pan et al. (2021) reported that the application of biochar to rubber seedlings in pot experiments increased the soil water content and nutrient availability, decreased nutrient leaching, and mitigated soil acidity. However, Gonzalez Sarango et al. (2021) using applications of up to 6 t ha^{-1} of locally produced charcoal as biochar did not detect a biochar effect on the growth of trees at two sites in the same experiment in the south Ecuadorian Amazonia as reported here, while the amendment of a complete mineral fertilizer plus lime had a strong effect on tree growth. At the less fertile of the two study sites, the biochar amendment increased pH, effective cation-exchange capacity (ECEC), and base saturation (BS) significantly in most biochar treatments indicating that Al toxicity was reduced. At the more fertile site with a higher soil pH at the start of the experiment, biochar had weaker effects on pH, ECEC, BS, and the inherently lower Al toxicity. Gonzalez Sarango et al. (2021) therefore suggested that the lack of a biochar effect was attributable to an insufficient nutrient supply by the applied biochar, particularly of N and P.

The tree plantation in Ecuador studied by Gonzalez Sarango et al. (2021) and here included two tree species, Pachaco [Brazilian firetree, *Schizolobium parahyba* var. *amazonicum* (Ducke) Barneby] and Melina (beechwood, *Gmelina arborea* Roxb.). Brazilian firetree, a species in the N_2-fixing family Fabaceae (legumes), is one of the most important planted native tree species in the Amazonia region, mainly used in the plywood industry (Silva et al., 2011). Because Brazilian firetree grows fast and tolerates low soil fertility, it has been frequently planted on degraded soils (Gazel Filho et al., 2007). Beechwood is another frequently planted tree in Amazonia, which is native to India. It grows on little fertile acidic soils and is used as timber wood, firewood, and fodder (Swamy et al., 2004). To explore the effects of the locally available biochar—charcoal mainly produced as fuel—amended at application rates of up to 6 t ha^{-1}, on soil fertility in plantations of both the native Brazilian firetree and exotic beechwood trees at two different sites in the southern Ecuadorian Amazonia region, we tested the hypotheses that biochar (a) increased the availability of macronutrients (N, P, K, Ca, Mg, S), micronutrients (Fe, Mn, Cu, Zn), and beneficial Na concentrations in the topsoil, but less than the amendment of complete fertilizer plus lime and thus insufficiently to promote tree growth; and (b) reduced nutrient leaching from topsoil to subsoil because of improved nutrient retention but not sufficiently to result in a direct benefit for tree growth.

2 | MATERIALS AND METHODS

2.1 | Study sites and selected tree species

The experiment was conducted in the Province of Zamora-Chinchipe, southern Ecuadorian Amazonia (Table 1). The study soils were considered degraded because of the lack of an organic layer, the presence of only a thin A horizon in La Victoria, or even no A horizon in Los Zapotes, which indicated a strong erosion, because of the steep slopes. Seedlings of Brazilian firetree and beechwood derived from a single mother plant were purchased from a local tree nursery. The seedlings were planted at the two study sites when they were 2-mo old and had a height of 35–40 cm.

2.2 | Experimental design and sampling

The experimental design has been described in detail in Gonzalez Sarango et al. (2021) and is summarized in Supplemental Table S1 and Supplemental Figure S1. In brief, we used a full factorial split-split-plot design (Dormann & Kühn, 2011). We split the main plots into two subplots for the two tree species (Brazilian firetree and beechwood). These subplots were then further split into with/or without mineral fertilizer plus lime (200, 150, 200, 118, 183, and 40 kg ha^{-1} of N, P, K, Mg, S, and Zn, respectively, and 5 and 3 t ha^{-1} $CaCO_3$ at La Victoria and Los Zapotes, respectively) × three levels of biochar (0, 3, 6 t ha^{-1}). The split-split-plot design was considered to be particularly well suited to control for small-scale spatial heterogeneity in steep terrain. The main plots had an area of 144 m^2 and included 16 trees, corresponding to 1,111 trees ha^{-1}. The size of our biochar application was based on the only other field study on the use of biochar in tree plantations of Kishimoto and Sugiura (1985), we were aware of before the start of our experiment in 2009, who had applied five times 0.5 t ha^{-1} (i.e., a total of 2.5 t ha^{-1}) of biochar, which resulted in significant effects.

Our biochar was commercially available wood-derived charcoal, which was produced from tabano (*Casearia mariquitensis* Kunth., ∼80%) and a mixture of cashco (*Weinmania fagaroides* Kunth), canelo (*Nectandra laurel* Klotzsch ex Nees), and capulí (*Prunus opaca* Walp., ∼20%). The wood was milled to <0.5 cm and pyrolyzed in a traditional earthen kiln from harvest debris that could not be sold as construction or pulp wood. We bought the charcoal in Jimbilla, around 30 km away from Loja, where it was produced as fuel. The source of the wood is a temperate Andean forest on acidic soils that have mainly developed from schist. The charcoal did not show indications of a high trace metal accumulation (Table 2). Moreover, the low production temperature of <500°C bears a small risk of excessive accumulation of polycyclic aromatic hydrocarbons (C. Wang et al., 2017). The amended element masses with the biochar are summarized in Supplemental Table S1. Nutrients plus lime and biochar were spread on a circular area with diameters of 2.4 and 2 m at La Victoria and Los Zapotes, respectively, and mixed with the topsoil to a depth of 0.25 m before the treelets were planted in the center at the time of planting from 27 July–7 Aug. 2009. The trees were harvested from 4 to 8 Nov. 2013.

We sampled the mineral soils from the two depth layers 0–0.25, and 0.25–0.5 m at the beginning of the experiment in March 2009 and six times after the start of the experiment in February 2010, February 2011, February 2012, July 2012, January 2013, and November 2013. We

TABLE 1 Location and properties of the study sites and the upper 0.25 m of the mineral soils (ranges or means ± standard deviations) before the start of the experiment in 2009 (Gonzalez Sarango et al., 2021)

Properties	Sites: La Victoria	Sites: Los Zapotes
UTM WGS84 coordinates	17 M 9552550 730470	17 M 95668135741834
Nearest settlement	City of Zamora	Village of Panguintza
Native vegetation	Evergreen tropical forest	
Mean annual precipitation, mm	1,945	
Mean annual temperature, ˚C	22	
Mean annual humidity, %	88	
Elevation, m asl	949–965	875–917
Slope, %	15	60
Texture	sandy loams	loamy silty clays
Parent rock	granodiorite	andesite and tuffite
Soil type (Soil Survey Staff, 2014)	Typic Kandiudults	
Bulk density, g cm^{-3}	1.2 ± 0.05	0.95 ± 0.05
pH (H_2O)	4.5–4.7	4.7–5.2
Soil organic C, g kg^{-1}	30 ± 2.1	28 ± 3.2
C/N ratio	14 ± 0.23	11 ± 0.70
Effective cation-exchange capacity, $mmol_c$ kg^{-1}	49 ± 3.6	81 ± 15
Base saturation, %	33 ± 4.9	65 ± 10

collected four samples from each plot, which were combined to one composite sample, and then air-dried and sieved <2 mm.

2.3 | Measurements

We determined the pH of the biochar with a glass electrode in H_2O (soil/solution ratio: 1:10) and the ash content by combustion at 560˚C and weighing the remains. Total C, N, and S concentrations in the biochar and total N concentrations in the soil were determined in ground aliquots of the biochar and the soil samples from March 2009, February 2010, July 2012, and November 2013 with an Elemental Analyzer (Flash HT Plus, Thermo Fisher Scientific). Total element concentrations in the biochar were determined with a pressurized digestion with concentrated HNO_3 in a Microwave oven (MARS6Xpress, CEM). Plant-available NH_4^+–N, P, K, Ca, Mg, Fe, Mn, Zn, and Cu concentrations were extracted with the modified Olsen procedure (0.5 M $NaHCO_3$ + 0.01 M ethylenediaminetetraacetic acid (EDTA), pH 8.5, soil/solution ratio 1:10) following the suggestion of the Network of Soil Laboratories of Ecuador (RELASE). Olsen-P and NH_4–N concentrations were determined colorimetrically. We used the ascorbic acid method to produce Mo blue, quantified at 880 nm with an ultraviolet-visible (UV-VIS) spectrophotometer (UV-1800, Shimadzu) for phosphate (Crouch & Malmstadt, 1967) and the phenol-hypochlorite method to produce indophenol (Weatherburn, 1967) quantified at 630 nm for ammonium. Plant-available Na was extracted with ammonium acetate (1 M NH_4OAc, pH 7). The concentrations of all elements except for total C, N, and S and NH_4^+–N and PO_4^{3-}–P were measured with inductively coupled plasma-optical emission spectroscopy (ICP-OES) (5100 VDV, Agilent).

2.4 | Calculations and statistical analyses

To assess the effect of biochar on the retention of nutrient (i) in topsoil, we determined the Olsen-extractable stock (S) of all elements except N (total stock) and Na (NH_4OAc-extractable stock) in the 0-to-0.25-m layer as a measure of the bioavailable nutrient pool before the start in 2009 ($t0$) and at the end of the experiment in 2013 ($t1$) and estimated nutrient retention (R) by the biochar in percentage of the initial element stock determined with the same extractants plus the element amendments with mineral fertilizer plus lime and/or biochar (A) (Equation 1).

$$R_i = \frac{S_{i,t1}}{(S_{i,t0} + A_i)100} \quad (1)$$

Because we did not measure nutrient uptake by the trees, Equation 1 was only used to compare plots, which did not show significant differences in the biomass production of the trees. The latter was true for all plots that received

TABLE 2 Properties of the used biochar (commercially available charcoal produced as fuel)

Property	Concentration	Unit
pH (H_2O)	8.55	–
Ash content	2.62	%
C	838	g kg^{-1}
N	3.60	g kg^{-1}
P	332	mg kg^{-1}
K	5.06	g kg^{-1}
Ca	4.94	g kg^{-1}
Mg	1.81	g kg^{-}
S	436	mg kg^{-1}
Fe	392	mg kg^{-1}
Cu	not detected	mg kg^{-1}
Zn	153	mg kg^{-1}
Mn	316	mg kg^{-1}
Na	395	mg kg^{-1}
Al	2.52	g kg^{-1}
Pb	9.54	mg kg^{-1}
B	not detected	mg kg^{-1}
Mo	not detected	mg kg^{-1}
Co	6.06	mg kg^{-1}

mineral fertilizer plus lime with/without biochar and for all plots that did not receive mineral fertilizer plus lime with/without biochar at each site. The amendment of fertilizer plus lime resulted in a significantly higher biomass production than without fertilizer plus lime, while the biochar did not influence biomass production (Gonzalez Sarango et al., 2021). For the fertilized and unfertilized plots, respectively, we roughly assumed that the same tree biomass production of the same tree species resulted in the same nutrient uptake. If this assumption was true, a significant effect of the biochar amendment on R_i could be attributed to changes in nutrient leaching.

Element concentrations and retention were each analyzed for treatment effects using Repeated Measures ANOVA for split-split-plot designs with the software R (R Core Team, 2017) following Dormann and Kühn (2011). In each ANOVA, we accounted for the effects of sampling date (and its interaction terms with all other explanatory variables), site, and site block before the treatment factors tree species, fertilizer plus lime, and biochar amendment and their interaction terms. The Error term was Error (plot/"fertilized plus lime"/"biochar addition"). Residuals were checked graphically for compliance with the requirements of ANOVA. We used the function *lillie.test* from the package *nortest* (Gross & Ligges, 2015) for tests of normality, the function *aov* from the package *stats* (R Core Team, 2017) for the ANOVA, the function *HSD.test* from the package *agricolae* (de Mendiburu, 2019) for Tukey's HSD post-hoc test, and the packages *gplots* (Warnes et al., 2019), and *plotrix* (Lemon, 2006) for plotting data.

To highlight the most important results we combined treatments, which did not differ significantly in the element concentration or retention and re-ran the ANOVA on the resulting "simplified" design (with a reduced number of treatments and/or dates) as indicated in the figure legends.

3 | RESULTS

3.1 | Effect of biochar amendment on element concentrations

3.1.1 | Macronutrients and Na

Our ANOVA revealed significant effects of the site on total N and Olsen-extractable P, Ca, Mg, and NH_4OAc-extractable Na concentrations and marginally significant effects ($p < .1$) on Olsen-K concentrations in the topsoil (Supplemental Table S2) reflecting the fact that the site at Los Zapotes was inherently more fertile than that at La Victoria (Gonzalez Sarango et al., 2021). As expected, the mineral fertilizer plus lime amendment had significant positive effects on Olsen-P, -K, -Ca, and -Mg concentrations but not on total N, Olsen-NH_4^+–N, and NH_4OAc-Na concentrations in topsoil. The biochar amendment increased total N and Olsen-Ca concentrations in topsoil significantly (Figures 1a–c and 2a–c, Supplemental Table S2). The positive effect of the biochar amendment on the total N concentration was, however, restricted to February 2010. Against our expectation, the biochar amendment did not have a significant effect on the Olsen-P concentrations (Figure 1d–f; Supplemental Table S2). Again as expected, date had a significant effect on all bioavailable macronutrient concentrations and Na but not on the total N concentrations mainly reflecting the different nutrient status before the start and during the experiment (Figures 1–3; Supplemental Table S2; Supplemental Figures S2–S7). The fact that several interactions of date with site, site block, tree species, and amendment of mineral fertilizer plus lime had significant effects on most nutrient concentrations indicates that the development of the nutrient concentrations was different at the different study sites and sometimes even site blocks, between the two tree species and in fertilizer plus lime-amended or not mineral fertilized plots (Supplemental Figures S2–S7). There were only two significant interactions involving biochar in the topsoil, that is, the interaction date/biochar on Olsen-NH_4^+–N concentrations and the interaction date/tree species/biochar on Olsen-Mg concentrations suggesting an influence of biochar on NH_4^+–N and Mg availability on some dates, which was different for the two tree species in the case of Mg. In the subsoil layer 0.25–0.5 m, we found the same significant main effects of our treatments on the total N and

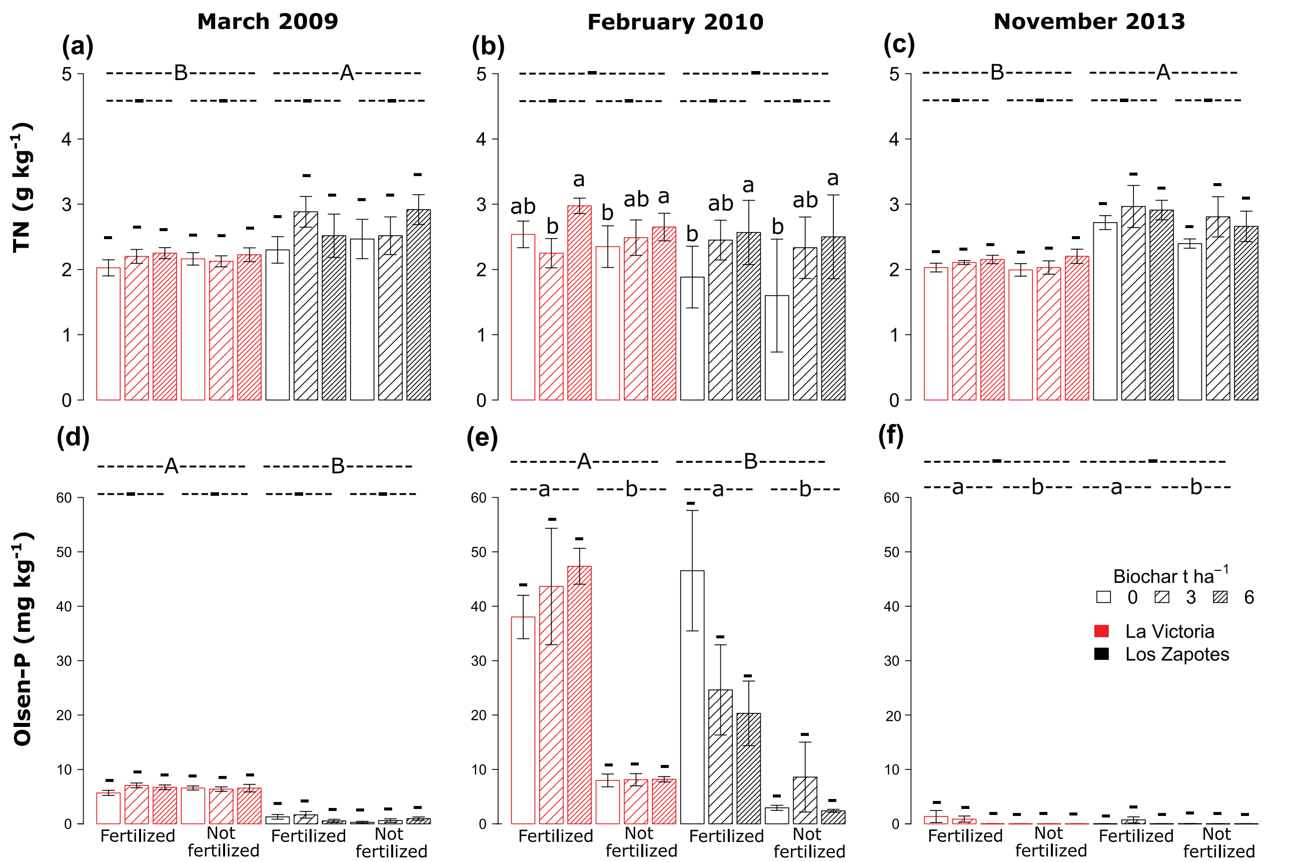

FIGURE 1 (a–c) Mean total nitrogen and (d–e) Olsen-P concentrations in the 0-to-0.25-m depth layer of the mineral soil before the start in (a, d) March 2009, (b, e) shortly after the start in February 2010, and (c, f) at the end of the experiment in November 2013 at Los Zapotes and La Victoria in all plots that received 0, 3, or 6 t ha^{-1} of biochar, respectively. The error bars show standard errors ($n = 8$ for La Victoria and $n = 6$ for Los Zapotes). Different upper-case letters indicate significant differences between the study sites. Different lower-case letters in the middle indicate significant differences between the plots amended with mineral fertilizer plus lime and those without mineral fertilizer plus lime and different lower-case letters at the bottom indicate significant differences among the amendments of 0, 3, and 6 t ha^{-1} of biochar. A minus illustrates no significant differences and the dashed lines span all treatments to which the letters apply

Olsen-extractable nutrient concentrations as in the topsoil, except that biochar additionally increased Olsen-Mg concentrations but did not have an effect on total N concentrations in the subsoil (Figure 2d–f; Supplemental Table S3).

3.1.2 | Micronutrients

Site had a significant effect on the Olsen-extractable micronutrient concentrations in the topsoil except for Zn (Supplemental Table S4), because of the higher inherent fertility at Los Zapotes. The amendment of mineral fertilizer plus lime increased the availability of Zn and marginally also of Cu and decreased that of Fe (Figure 3). The amendment of biochar did not influence micronutrient availability, except that it increased the Olsen-Zn concentrations marginally significantly. Again, date and several interactions of date with other treatments had significant effects on all four micronutrients. However, only Fe availability was significantly or marginally significantly affected by interactions involving biochar. The experimental treatments had a similar effect on micronutrient availability in the subsoil as in the topsoil (Supplemental Table S5) except that the amendment of fertilizer plus lime did not change Fe availability and site had an additional significant positive effect on Zn availability.

3.2 | Effect of biochar on nutrient retention in the topsoil

In November 2013, at the end of the experiment, there were significant effects of the site on the Olsen-NH_4^+–N, –Ca, –Mg, –Fe, –Mn, and –Cu retention and a marginally significant effect on total N retention in the topsoil derived with Equation 1 on the plots amended with mineral fertilizer plus lime and on Olsen-NH_4^+–N, –Ca, –Mg, –Fe, –Mn, and –Cu retention in the topsoil on the plots that did not receive mineral fertilizer plus lime (Supplemental Tables S6 and S7). There were additionally significant effects of the tree species on the retention of Olsen-K in the topsoil on the mineral

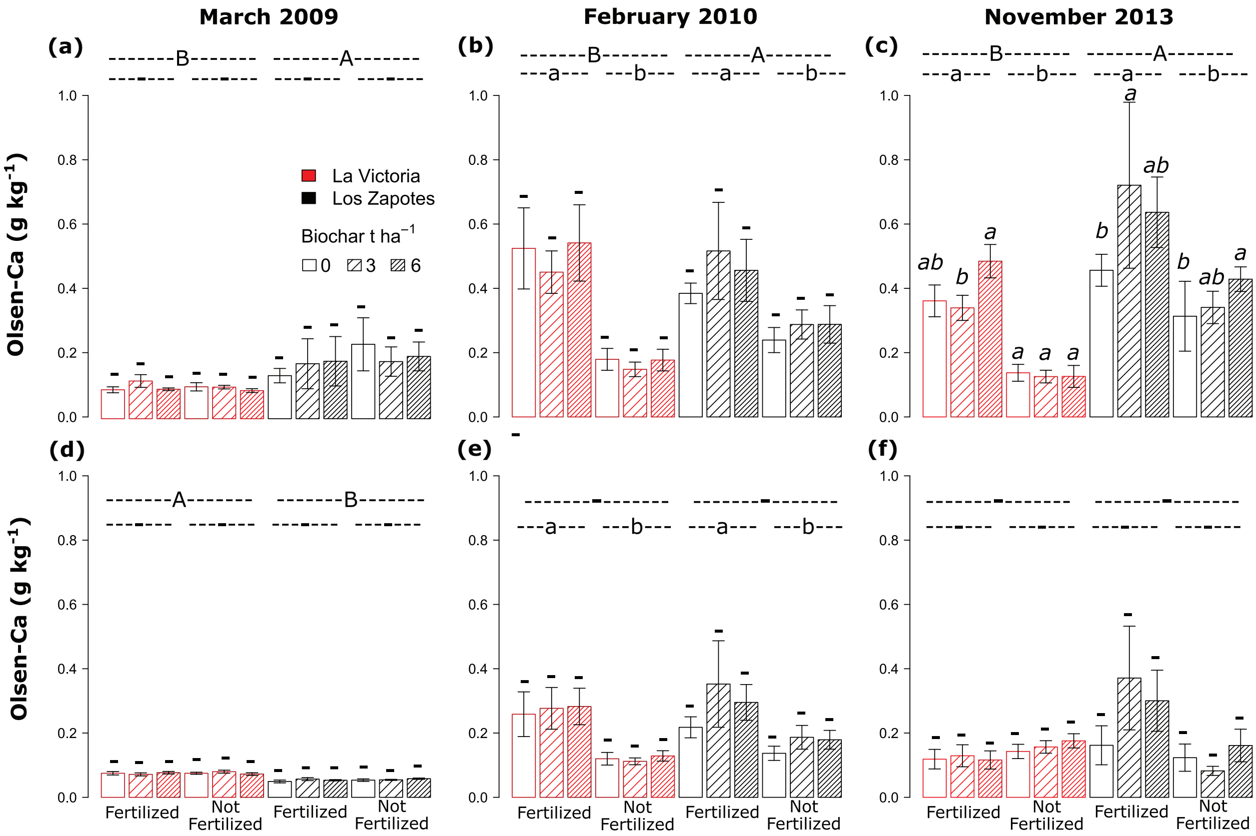

FIGURE 2 Mean Olsen-extractable Ca concentrations in the (a–c) 0–to-0.25 m and (d–f) 0.25–to-0.50 m depth layers of the mineral soil before the start in (a, d) March 2009, shortly after the start in (b, e) February 2010 and (c, f) at the end of the experiment in November 2013 at Los Zapotes and La Victoria. The error bars show standard errors ($n = 8$ for La Victoria and $n = 6$ for Los Zapotes). Different upper-case letters indicate significant differences between the study sites. Different lower-case letters in the middle indicate significant differences between the plots amended with mineral fertilizer plus lime and those without mineral fertilizer plus lime and different lower-case letters at the bottom indicate significant differences among the amendments of 0, 3, and 6 t ha^{-1} of biochar (in italics if marginally significant, $p < .1$). A minus illustrates no significant differences and the dashed lines span all treatments to which the letters apply

fertilized plots and Olsen-Ca and -Mg in the topsoil on the unfertilized plots and a marginally significant effect on the Olsen-NH_4^+–N retention in the topsoil on the mineral fertilized plots. Biochar amendment significantly increased the retention of Olsen-Zn in the topsoil on the mineral fertilized plots and decreased total N retention in the topsoil of the unfertilized plots at the application rate of 6 t ha^{-1} (Figure 4; Supplemental Tables S6 and S7). In addition, there was a marginally significant effect of biochar on the retention of Olson-Ca in the topsoil on the mineral fertilized plots.

4 | DISCUSSION

4.1 | Effect of biochar on nutrient and Na concentrations

The significant increase of the N concentrations in the topsoil after amendment of biochar in February 2010 (Figure 1b) is in line with findings of Barrow (2012), Chan et al. (2008), and N. Xu et al. (2016). N. Xu et al. (2016) attributed this increase in part to reduced N leaching losses because of the improved N retention after amendment of biochar. However, the increase in total N concentrations in topsoil at our two study sites did not result in improved growth in the treatments with only biochar but no mineral fertilizer plus lime (Gonzalez Sarango et al., 2021). Most of the additional N must have been stored in nonbioavailable forms, which is in line with the finding that the Olsen-NH_4^+–N concentration did not respond significantly to the biochar amendment (Supplemental Table S2). However, the biochar effect on the total N concentrations had already disappeared at the end of the experiment in November 2013 (Figure 1c). The significant effect of date on the total N concentrations in topsoil was therefore attributable to the increase in total N concentrations after amendment of biochar in February 2010 relative to March 2009, but there was no long-term effect of the biochar amendment on total N concentrations in soil (Figure 1, Supplemental Figure S2a and S2b).

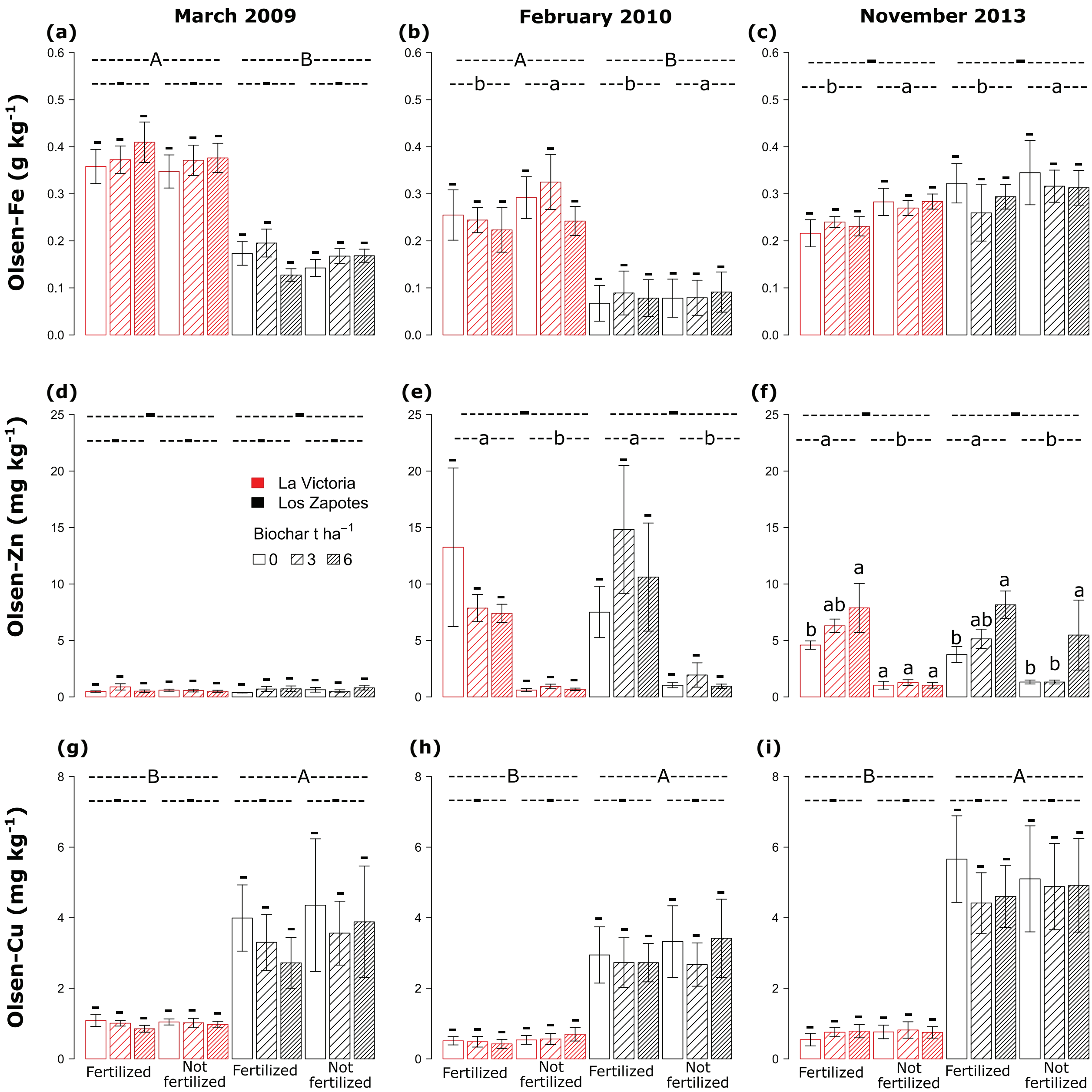

FIGURE 3 (a–c) Mean Olsen-extractable Fe, (d–f) Zn, and (f–h) Cu concentrations in the 0-to-0.25 m depth layers of the mineral soil before the start in (a, d, g) March 2009, (b, e, h) shortly after the start in in February 2010 and (c, f, i) at the end of the experiment in November 2013 at Los Zapotes and La Victoria. The error bars show standard errors ($n = 8$ for La Victoria and $n = 6$ for Los Zapotes). Different upper-case letters indicate significant differences between the study sites. Different lower-case letters in the middle indicate significant differences between the plots amended with mineral fertilizer plus lime and those without mineral fertilizer plus lime and different lower-case letters at the bottom indicate significant differences among the amendments of 0, 3, and 6 t ha^{-1} of biochar. A minus illustrates no significant differences and the dashed lines span all treatments to which the letters apply

The finding that the amendment of biochar did not have an effect on the Olsen-P concentrations in the topsoil (Figure 1d–f) is in contrast to reports of positive effects of biochar on plant-available P concentrations in corn (*Zea mays* L.) (Ch'ng et al., 2017) and cabbage (*Brassica oleracea* L.) cropping (Apori & Byalebka, 2021) and tea [*Camellia sinensis* (L.) Kuntze] plantations (Karim et al., 2020). However, in the latter studies the biochar amendments were 7.5–20 t ha^{-1}, higher than in our experiment. Our finding is also unexpected, because Gonzalez Sarango et al. (2021) had shown that the biochar amendment decreased the acidity of the soil, which should have positive effects on the bioavailability of P because of a reduced precipitation of Al phosphates as a consequence of the reduced exchangeable Al concentrations. An improved P availability in response to a biochar amendment of about 30–60 t ha^{-1} in a laboratory experiment was

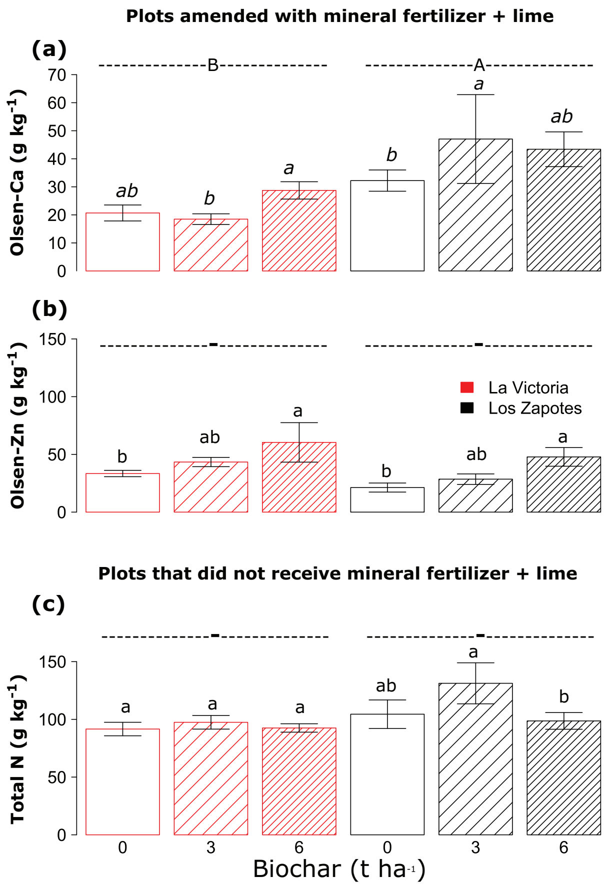

FIGURE 4 Effects of biochar amendment on the Olsen-Ca and -Zn retention in the (a, b) fertilized and (c) total N retention in the unfertilized plots in the 0-to-0.25-m depth layers of the mineral soil at the end of the experiment in November 2013 at Los Zapotes and La Victoria (error bars are standard errors, $n = 8$ for La Victoria and $n = 6$ for Los Zapotes). Different upper-case letters indicate significant differences between the two study sites. Different lower-case letters indicate significant differences among the three biochar treatments (in italics if marginally significant, $p < .1$). A minus illustrates no significant differences

reported by Hong and Lu (2018). We suggest that our tree plantations are strongly P-limited, which is supported by the fact that the plant-available P concentrations were near to or below the detection limit of P at the end of our experiment in 2013 (Figure 1f). Apparently, the addition of P with biochar was not sufficient to decrease the P deficiency substantially, perhaps because we applied less biochar than required. The Olsen-P concentrations before the start of the experiment and in all plots without amendment of mineral fertilizer plus lime were consistently below the lower end of the range of Olsen-P concentrations deemed sufficient for optimal plant growth by Bai et al. (2013) of 10.9–21.4 mg kg^{-1}. Figure 1 also illustrates some temporal variation in Olsen-P concentrations of the plots without amendment of mineral fertilizer plus lime, which is likely attributable to the temporal variation in processes contributing to the Olsen-extracted labile P pool such as the dissolution of minerals, organic matter mineralization, and dieback of microorganisms. Thus, a more complete picture of the P supply would have required a higher temporal resolution of the measurement of the P availability and a more comprehensive characterization of P forms in soil than we could realize.

The fact that the only studied macronutrient concentration in the topsoil and subsoil that was affected by the amendment of mineral fertilizer plus lime and biochar in the same positive direction was Olsen-extractable Ca (Figure 2; Supplemental Table S2) is in line with findings from Y. Wang et al. (2014) who reported an increase of base cation concentrations by up to a factor of 6.7 after incubation of biochar-amended soil in the laboratory. Similarly, Jien and Wang (2013) found significantly increased exchangeable Ca concentrations by a factor of up to 5 after the incubation of soil samples with biochar. There are several more reports showing that biochar amendment increased exchangeable Ca and Mg concentrations in soil (Puga et al., 2015; G. Xu et al., 2014; Yeboah et al., 2009; Zhao et al., 2014). However, Gonzalez Sarango et al. (2021) had previously shown that the amendment of biochar only increased the base saturation at the less fertile site La Victoria. Moreover, the effect of biochar amendment on the Olsen-Ca concentrations was much weaker than that of the mineral fertilizer plus lime amendment (Figure 2). The significant effect of date on the Olsen-Ca concentrations consisted of a strong increase after the start of the experiment. However, the Olsen-Ca concentrations decreased considerably until the end of the experiment because of plant uptake and leaching (Figure 2; Supplemental Figure S3c,d). At the end of the experiment in November 2013, there was only a significant effect of the mineral fertilizer plus lime amendment on the Olsen-Ca concentration in the topsoil while that of the biochar amendment had disappeared (Figure 2c).

We attribute the increased Zn and Cu availability in the soils of the mineral fertilizer plus lime treatments to the purposeful amendment of Zn (Supplemental Table S3) and an undetected contamination with Cu either in the Zn fertilizer or in the lime (Figure 3; Supplemental Table S3). The marginally significant effect of the amendment of biochar on the Zn availability in the topsoil was similar to findings of Abedin and Unc (2020) that the amendment of biochar to boreal soils had a lasting effect on the bioavailability of Mn, Cu, and Zn beyond the end of their experiment. The Zn concentration in biochar depends on the feedstock of the biochar production (Altland & Locke, 2013; Namgay et al., 2010; Prasad et al., 2019) and the pyrolysis temperature (T. Qian et al., 2016). The loss is smaller at higher pyrolysis temperature, because of increased Zn adsorption to the char (T. Qian et al., 2016).

On the other hand, the significant decrease in Fe availability in the topsoil in response to the amendment of mineral fertilizer plus lime (Figure 3a–c; Supplemental Table S3) was likely attributable to the significant increase of the pH value of the topsoils at La Victoria (Gonzalez Sarango et al., 2021) favoring the precipitation of $Fe(OH)_3$. At Los Zapotes, however, there was only a small and nonsignificant increase in soil pH, which was at the start of the experiment already higher than at La Victoria. It can nevertheless be assumed that at Los Zapotes the liming resulted in increased pH values in the soil solution before the protons were buffered. This short-term increase might have been sufficient to precipitate $Fe(OH)_3$, which then is not easily redissolved. Although the biochar amendment did increase the pH values, particularly at La Victoria (Gonzalez Sarango et al., 2021), the effect was less pronounced, and we suspect that the soil solution pH values did not change similarly strongly as in response to liming. As a consequence, our biochar amendment did not have a significant effect on the Fe availability in the topsoil, which is similar to findings of Abedin and Unc (2020) who reported that an amendment of 15 t ha^{-1} biochar did not significantly change Fe availability in the topsoil. The pH effects after liming and biochar addition in our study are in line with findings that the soil pH values increased after application of lime and gypsum (Murphy & Stevens, 2010) or biochar (Yuan et al., 2011a, 2011b).

4.2 | Effect of biochar on nutrient retention in the topsoil

At the end of the experiment, the significant effects of the site on the Olsen-NH_4^+–N, –Ca, –Mg, –Fe, –Mn, and –Cu retention (Supplemental Tables S6–S7) reflected the higher capacity of the soil at Los Zapotes to retain nutrients, mainly because of a higher ECEC (Gonzalez Sarango et al., 2021). There was additionally a significant effect of the tree species on the retention of Olsen-K and a marginally significant effect of tree species on the retention of Olsen-NH_4^+–N in the topsoil (Supplemental Table S6). We suggest that these effects are attributable to the higher uptake of N and K by beechwood, which showed a higher basal diameter and diameter at breast height than Brazilian firetree (Gonzalez Sarango et al., 2021). In the absence of biochar effects on tree growth (Gonzalez Sarango et al., 2021), the significant biochar effects on the retention of Olsen-Zn and the marginally significant biochar effect on the retention of Olsen-Ca in the mineral fertilized plots (Figure 4a,b) can be attributed to additional sorption sites provided by the biochar. The small but significant negative effect of biochar on total N retention at the application rate of 6 t ha^{-1} in the unfertilized soils (Figure 4c) might be related with a small decrease in bulk density. Unfortunately, we did not determine bulk density at the end of the experiment. The weak effect of biochar on the retention of Olsen-Ca and the absence of any biochar effect on the retention of Olsen-K, and -Mg, and NH_4OAc-extractable Na in spite of the positive effects of the amendment of biochar on pH, ECEC, and BS contrasts findings of Gaskin et al. (2010), Hailegnaw et al. (2019), Lehmann et al. (2003), and Widowati and Asnah (2014) who observed a significant increase in the retention of base cations in response to the amendment of biochar in acidic soils with low ECEC and low BS. However, all cited authors used higher application rates of biochar ranging from 11 to 600 t ha^{-1}. Very high application rates were used in laboratory incubations (Hailegnaw et al, 2019) and pot experiments (Lehmann et al., 2003), while in field or greenhouse experiments 11–30 t ha^{-1} have been used (Gaskin et al., 2010 and Widowati & Asnah, 2014).

5 | CONCLUSIONS

The amendment of up to 6 t ha^{-1} of biochar increased total N and Olsen-extractable Ca and Zn concentrations in the topsoil. However, this increase was not sufficient to enhance tree growth as reported by Gonzalez Sarango et al. (2021). This partly supports our first hypothesis that the amendment of 3–6 t ha^{-1} of biochar increases the nutrient concentrations but only for three of the considered 10 nutrients and not sufficiently to enhance tree growth. This might imply that higher amendment rates would have been necessary as was recently recommended by Gale and Thomas (2019) to achieve a biochar effect. However, amendment rates of 20–30 t ha^{-1} as suggested by Gale and Thomas (2019) would require a sufficient local biochar availability and imply a considerably higher logistic, cost, and labor effort to transport and apply the biochar at the frequently remote degraded Ultisols of the Ecuadorian Amazonia.

The amendment of biochar increased the retention of Ca and Zn in the topsoil of the plots that received mineral fertilizer plus lime and thus indeed reduced leaching losses of these two nutrients included in the fertilizer partly supporting our second hypothesis. The slightly decreased total N retention in the unfertilized plots in response to the amendment of 6 t ha^{-1} biochar likely was related with a small decrease in bulk density.

Overall, there were only weak effects of our comparatively low biochar application rates on soil fertility, which partly had already disappeared at the end of the experiment after 51 mo. We conclude that the amendment of 3–6 t ha^{-1} biochar does not sufficiently improve the fertility of degraded, strongly weathered tropical soils in the South Ecuadorian Amazonia. Further research is necessary to determine, under which circumstances, with which quality, and at which application rates the use of biochar is sufficiently beneficial for the remediation of tropical soils via tree plantations to justify the enormous financial and labor efforts of its application, particularly if considerably higher application rates were necessary to achieve positive effects.

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AUTHOR CONTRIBUTIONS

Esthela Margarita Gonzalez Sarango: Data curation; Formal analysis; Investigation; Methodology; Project administration; Visualization; Writing – original draft. Sophia Leimer: Formal analysis; Methodology; Writing – review & editing. Carlos Valarezo Manosalvas: Conceptualization; Funding acquisition; Investigation; Methodology; Project administration; Supervision; Writing – review & editing. Wolfgang Wilcke: Investigation; Methodology; Resources; Supervision; Writing – review & editing.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

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SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.

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