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Amazonian Dark Earths - potential for replication with the use of biochar

M.Sc. thesis

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2012

Declaration

I declare that I have elaborated my thesis independently and quoted only quotations listed in the references.

Prague, April 15, 2012

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Nikola Teutscherová

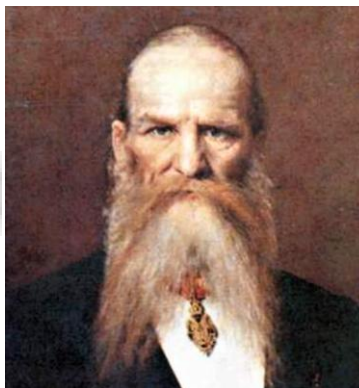
Acknowledgement

I would like to thank my supervisor doc.Ing. Bohdan Lojka, Ph.D. (Head of Department of Crop Sciences and Agroforestry of the Institute of Tropics and Subtropics of the Czech University of Life Sciences Prague) for leading my master thesis, for his help, suggestions, patience and information.

Special thanks belong to Hlavkova Nadace and Universidad Nacional de Ucayali for financial support, without which this project could not take place and the thesis could not be written.

Furthermore, acknowledgement belongs to professors and member of Universidad Nacional de Ucayali for their suggestions and advices, namely to Professor Juan (the head of Laboratory of Soil Sciences) and to all people helping me in the nursery. Also I want to thank to all professors from Czech University of Life Sciences, who helped me to finish this thesis. My thanks belong mainly to Prof. Ingo. Daniela Pavli ova CSU. for arranging nitrogen analysis at the Faculty of Aerobiology, Food and Natural Resources and for helping with evaluation of the results of analysis accomplished in Peru. Furthermore, I would like to thank Ing.Jindrova and doc. Ing. Katerina Berchova Ph.D. for helping me with the statistical analysis.

Last but not least, I want to thank to all my friends and family for their encouragement during whole time of finishing this thesis and to Jesús (Jopo) Morales Ubeda for his help during sample preparation, laboratory analysis and support.



I am very grateful to everyone, who helped me with this thesis.

Abstract

Amazonian Dark Earths (ADE), in Portuguese *Terra Preta de Índio* or, are anthropogenic soils. Generally, soils of Amazonia are mainly acid Oxisols, Ultisols and Inceptisols with low fertility. Characteristic feature of ADE is high C content, which is increased usually by addition of high amount of biochar. According to many previous studies these anthropogenic soils greatly improve the yields of cultivated crops. The aim was to prove the positive influence of biochar application on plant growth and biomass production and further explore the possibility of biochar to improve the current agricultural systems in Peruvian Amazon, and thus, decrease the deforestation. For the experiment we have chosen two locally grown crops and one native tree from three distinct families - rice, (*Oryza sativa* L. – Poaceae); cowpea, (*Vigna unguiculata* [L.] Walp – Fabaceae) and bolaina blanca (*Guazuma crinita* Mart. – Malvaceae). The plants were grown in plastic bags with two kilograms of Amazonian Ultisol and different additions of two types of biochar, partly decomposed chicken manure and inorganic NPK fertilizer. After six weeks of cultivation we measured the stems and roots, weighted above- and belowground biomass and analyzed pH and soil and foliar nutrient contents. Additionally, study of charcoal production and utilization was done with in depth interview using questionnaires among local farmers and charcoal producers. Soil organic matter and pH were increased by all biochar amendments. Generally, biochar improved soil nutrient content and soil properties, but influenced biomass production and foliar content in smaller extent than it was expected. Significant increase of biomass production was observed only in case of cowpea, which was probably caused by the ability of cowpea to balance higher C:N ratio by symbiotic N fixing. Furthermore, we found generally higher influence on root growth which suggests that higher effect on aboveground growth could possibly be observed after longer cultivation time. Residues from charcoal production are potential source low-cost biochar, however, these are not widely used in agriculture. Results indicate that agricultural method using biochar from charcoal production could be a potential improvement of recent agriculture in Peruvian Amazon. However, more especially long-term experiments need to be done.

Key words: biochar, nutrient content, Peruvian Amazon, soil fertility, Ultisol

Abstrakt

Amazonské černé půdy (AČP), v portugalštině *Terra Preta de Índio* jsou půdy vytvořené soustavnou lidskou činností. Obecně jsou půdy Amazonie tvořeny kyselými Oxisoly, Ultisoly a Inceptisoly, které jsou považovány za půdy s velmi nízkou úrodností. Charakteristickým prvkem AČP je zvýšený obsah uhlíku, který je pravděpodobně výsledkem akumulace zbytků biouhlu v půdě. Podle mnoha výzkumů tyto antropogenní půdy výrazně zvyšují výnosy pěstovaných plodin. Cílem práce bylo prokázat pozitivní vliv využití biouhlu na růst rostlin a produkci biomasy a dále analyzovat možnosti využití biouhlu pro zlepšení zemědělských systémů užívaných v Peruánské Amazonii a tím ke snížené míry odlesňování. Pro experiment jsme vybraly dvě tradičně pěstované plodiny a jeden strom ze tří různých čeledí – rýži (*Oryza sativa* L. – Poaceae); vignon (*Vigna unguiculata* [L.] Walp – Fabaceae) and bolainu blancu (*Guazuma crinita* Mart. – Malvaceae). Rostliny byly pěstovány v plastových sáčkách se dvěma kilogramy Ultisolu s přísadou dvou typů biouhlu, organického hnojiva a NPK hnojiva. Po šesti týdnech jsme změřili délky stonků a kořenů, zvážili nadzemní a podzemní biomasu a analyzovali množství živin v rostlinném materiálu i v půdě. Dále jsme analyzovali možnosti využití biouhlu v zemědělství pomocí řízených rozhovorů s místními farmáři a producenty uhlí. Obsah uhlíku v půdě a pH byly výrazně zvýšeny přísadou biouhlu. Obecně lze říct, že přísada biouhlu zvýšil obsah živin v půdě a zlepšil půdní vlastnosti, ale produkci biomasy a růst rostlin ovlivnil v menší míře, než jsme očekávali. Výraznější vliv byl pozorován v případě vignon, což bylo pravděpodobně způsobeno schopností bobovitých rostlin symbioticky poutat vzdušný dusík a tím snižovat C:N poměr. Dále, obecně vyšší vliv biouhlu na produkci podzemní biomasy a růst kořenů naznačuje, že při delším trvání experimentu vliv biouhlu mohl být mnohem vyšší i u nadzemní biomasy. Odpadní produkty z produkce dřevěného uhlí jsou potenciálním zdrojem levného biouhlu, přesto nejsou obecně využívány v zemědělství. Výsledky naznačují, že zemědělské metody spojené s využíváním biouhlu by mohly zlepšit současné zemědělské praktiky v Peruánské Amazonii. Nicméně, další, zejména dlouhodobé, studie jsou nezbytné.

Key words: biochar, biouhel, obsah živin, Peruánská Amazonie, půdní úrodnost, Ultisol

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1 INTRODUCTION

In 1542, after eight months of searching for The City of Gold, the Spanish explorer Francisco de Orellana reported populations living in rain forests on the Amazon basin along the Amazon river. These regions were densely populated by indigenous Indians. Francisco de Orellana is also the first European who transversed the Amazon river. After the discovery of these chiefdoms he reported to Spanish court: "...there could be seen very large cities that glistened in white [...] many roads that entered into the interior [...] and besides this, the land is fertile [...] as our Spain" (O'Grady and Rush, 2007).

In later centuries Orellana's reports were lost or forgotten and the Amazon basin was considered to be an uninhabited virgin rain forest. For many years there was one question which nobody was able to answer: "How were they able to feed such a big population with so poor soils which usually are in rain forests?" This question was finally answered with discovering Amazonian Dark Earths. The origin of these nutrient-rich patches in an otherwise largely infertile soilscape has been the focus of intensive debate in the past (Myers *et al.*, 2003). While their anthropogenic origin is now widely accepted, the human activities which actually led to the high soil organic matter (SOM) and nutrient contents remain unclear (Petersen *et al.*, 2001; Meggers, 2001; Neves *et al.*, 2003). Even today, Anthrosols are intensively cultivated by local population (the "caboclos"), highlighting its importance to the Amazonian social and ecological landscape (Lima *et al.*, 2002).

Amazonian Dark Earths (ADE), in Portuguese *Terra Preta de Índio* or *Terra Preta do Índio*, are anthropogenic soils. Generally soils of Amazonia are mainly Oxisols, Ultisols and Inceptisols with an anthropic A horizon (Lima *et al.*, 2002). Dark color of ADE is caused by the high content of charcoal (biochar). Besides *Terra Preta*, also *Terra Mulata* is classified under ADE, but the organic content is lower than in *Terra Preta* so even the color is lighter. In Ferralsols, soil organic matter is derived from vegetation cover, while in ADE the organic component is principally derived from the debris of human occupation (Kern *et al.*, 2003). It is characterized by presence of this charcoal in high concentrations, animal and fish bones, potshreds and other fragments as residues from the Indian pre-Columbian settlements in this area, manure, turtle carapaces, shells, excrements, urine etc. accumulating for a long period of time in addition to vegetal components. Other characteristic fact about ADE is the high concentration of

nutrients in comparison with adjacent soils. One of the biggest problems of Amazonian earths, nutrient leaching, does not occur at these soils (Steiner *et al.*, 2008; Steiner *et al.*, 2009a; Lehmann *et al.*, 2002b). Total nutrient content in the soil is generally higher than in adjacent soils

Porous structure of biochar is connected with the improvement of soil moisture availability and water retention (Tryon, 1948; Piccolo *et al.*, 1996; Steiner *et al.*, 2009a). Biochar, the most characteristic feature of ADE, is not only promising soil conditioner but can be also used to mitigate climate change by CO₂ and other greenhouse gases sequestration (Ogawa *et al.*, 2006, Amonette *et al.*, 2007; Gaunt and Lehmann, 2008).

Furthermore, the ways of inexpensive biochar production can be found, for example by connection with charcoal production. By recursive optimization model (Labarta *et al.*, 2008) was predicted that after ten years, a representative pioneer farmer producing charcoal would earn 17% higher net income and clear 17% less forested area. Also, biochar amendments can be used for rehabilitating degraded land and bring poor soils into production (Barrow, 2012)

Most of the recent agricultural systems in the humid tropics are facing serious problems connected with population growth and increasing food demand. In many cases this leads to large-scale deforestation and agricultural expansion. The problem arises with the infertility of the soils in most of the developing countries. After clearing of the land, the quality of the soil starts decreasing rapidly. This process is mostly irreversible and the sustainable agriculture is almost impossible without using mineral fertilizers. After exhaustion of the soil the farmers are forced to leave the field, cut down and burn new part of the forest to obtain new agricultural area, leaving the previous one without vegetation which in most cases causes erosion and soil degradation. Recently, erosion is one of the most significant problems in agriculture (Drenge, 1992).

Furthermore, locally produced manure, compost and other organic fertilizers have been substituted by purchased inorganic fertilizers. These are causing pollution and soils degradation.

Integration of biochar into agriculture can be important step to sustainable agriculture. The aim of this thesis is to prove the positive influence on plant growth and biomass production and further explore the possibility of biochar to improve the current agricultural systems in Peruvian Amazon, and thus, decrease the forestation.

2 LITERATURE REVIEW

2.1 History of Amazonian Dark Earths

The origin of these nutrient-rich patches in an otherwise mostly infertile soils has been the focus of intensive debate during the last years (Myers *et al.*, 2003). There are many theories of the creatinon of Amazonian Dark Earths, locally called *Terra Preta*. It was considered to be a result of volcanic activity in the Andes, formed by ashfall. Another theory considers the to be a result of sedimentation in Tertiary lakes (Falesi, 1974).

These theories of *Terra Preta* formation were not proven. Recently, these dark and fertile soils are thought to be formed by indigenous soil management of pre-Columbian indians, which was also confirmed during later research.

ADE are thought to have originated from disposal of organic waste and incomplete burning, creating charcoal (slash-and-char) (Mann, 2002).The original human residents of the river basin used slash-and-char method to improve their soils and to maintain their fertility. It is derived from slash-and-burn method but differentiated by lower temperatures. The effect of this technique on the soil fertility is significantly better than using slash-and-burn method because instead of ashes it brings more charcoal into the soil which helps to keep the soil fertile for a long period of time. The darkest patches correspond to the middens of settlements and are cluttered with crescents of broken pottery (Lehmann, 2006b). There are not very many remains of these indigenous populations, probably because of the unavailability of stone. The wood would rot easily in the humid climate. These soils were most likely created by pre-Columbian Indians from 500 to 2500 years B.C. and abandoned after invasion of Europeans (Smith, 1979). Its thought that these soils were used for agriculture for centuries.

Since discovering of ADE they have been subject of discussion and there are also attempts to recreate ADE (Sombroek *et al.*, 2002).

2.2. Location and extension of ADE

ADE as their name says can be found mostly in South America, especially in Amazonian basin. Amazonia is usually divided into *Várzea*, or floodplain, which occupies perhaps 2% of the basin's 7 million square kilometers, and *Terra Firme*, the never-flooded uplands that comprise everything else (Mann, 2000). *Várzea* is fertile. Upland soils in the Amazon basin are usually rather unfertile with low organic matter content (Sombroek, 2000). However, within these upland soils there are spots of fertile ADE.

The exact area of the ADE is estimated to be between 0.1-0.3% (Sombroek *et al.*, 2005) 10% (Mann, 2002) of the Amazonian basin, from this wide range of estimations the insufficiency of present knowledge is clear. Most commonly, ADE are found near rivers, especially in the middle part of Amazon (Figure 1).



Figure 1: Terra Preta do Índio in Brazil (Bechtold, 2007)

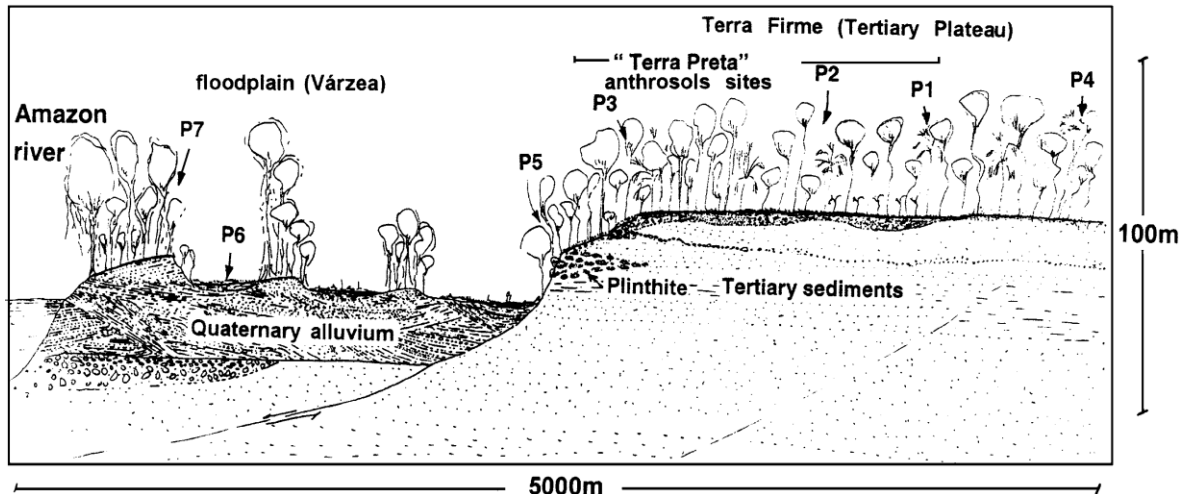


Figure 2: Location of the soils along of the transect Tertiary Plateau (*Terra Firme*) – floodplain (*Várzea*) near Iranduba, Western Amazonia (Lima *et al.*, 2002)

As shown at previous picture (Figure 2) ADE are mostly located near fertile *várzea* but far enough to prevent flooding. This is protecting *Terra Preta* from water erosion. (Lima *et al.*, 2002).

The depth of ADE can vary between 0,2 m and more than 1m and often the largest amount of nutrients is found in the deeper horizons (Lehmann *et al.*, 2003a).

2.2 Soil fertility and production potential of ADE

Soil fertility is ability of the soil to provide good conditions for life and growth during whole vegetation period for plants. It is influenced by soil moisture, nutrient content in the soil, presence of toxic substances, the structure of the soil, air content in the soil and biochemical cycles taking place in the soil.

2.2.1 Soil organic matter

Plants obtain nutrients from two natural sources: soil organic matter (SOM) and minerals. SOM includes any plant or animal material that returns to the soil and goes through the decomposition process (Bot and Benites, 2005). SOM is also the source of C and energy for soil microorganisms which improves biological activity of the soil. One of the most important functions of the SOM is nutrient bounding.

Large part of the content of N, P and S is contained in soil organic matter. From this they can be released to the soil solution and used by plants. There are more qualities of SOM and this

quality determines releasing of these nutrients. This is also influenced by the high content of black carbon in the soil. In the absence of direct experimental evidence, we can hypothesises for N that: (i) black C particles have high C:N ratios and do not release any N, but do not influence the N release of non-black C. In this case, the N release from the organic matter that is not associated with black C would need to be determined separately; (ii) black C is finely distributed and has a direct influence on the N release from non-black C (Lehmann *et al.*, 2003a).

2.2.2 Total nutrient content and nutrient availability

While the majority of soils in Amazonia generally have low fertility and high content of exchangeable aluminium (Al), the most striking property of ADE is their high fertility and elevated contents of most nutrients (Sombroek, 1966). In general, ADE have alkaline pH (Yuan and Xu, 2011), higher total phosphorus (P) content, greater exchangeable calcium (Ca) and magnesium (Mg), and increased minor element concentrations. Available manganese (Mn) contents increases to the same extent as total Mn (Lehmann *et al.*, 2003a). Total and available P contents of ADE are associated with microfragments of bone apatite with high P and Ca values. In ADE under cultivation, these values are lower, with increasing Al release. Large amounts of Mn and Zn occur in ADE and in high-fertile floodplain soils (Lima *et al.*, 2002).

The biochar amendments decreased the leaf nitrogen (N), proline- and chlorophyll-concentrations. Despite the larger leaf area compared to control soils, the N accumulation remained the same (Kamman *et al.*, 2010).

Lehmann *et al.* (2003c) compared different P pools of two anthropogenic dark earths with unfertilized and fertilized *Terra Firme* soils from Central Amazonia (Table 1) The highest content of available P was in high fertilized Ferralsol. On the contrary the lowest content was found in low fertilized Ferralsol and in unfertilized Ferralsol. The content of available P can vary but generally in Anthrosols (in this case ADE) is higher than in Ferralsols without high fertilization. Also pH is significantly higher in Anthrosols in comparison with Ferralsols. Also total organic carbon (TOC) is more than twice higher. These findings are explained by history of ADE. Organic material persist in the soil in the high stable form.

Table 1: Characterization of a fimic Anthrosol (ADE) and xanthic Ferralsol from central Amazonia. *n.d.* Not determined (Lehmann *et al.*, 2003c)

Soils	Sampling depth (cm)	pH (H ₂ O)	Sand (%)	Silt (%)	Clay (%)	TOC (g kg ⁻¹)	P _{available} (mgkg ⁻¹)
Anthrosol Marajá	0-10	5.7	58.7	17.0	24.3	n.d.	25.4a
Anthrosol Rio Preto da Eva	0-10	5.7	71.0		29.0	84.7	6.5b
Ferralsol – high fertilization	0-10	4.7	21.4c	19.6	59.0	36.0	142.6
Ferralsol – low fertilization	10- 20	4.2	27.2	14.9	57.9	21.2	4.3b
Ferralsol – unfertilized	0-5	4.1	21.4	19.6	59.0	40.6	4.2b

^a Mehlich-1 extraction

^b Mehlich-3 extraction

^c Particle size distribution determined at a nearby soil pit

However, total nutrient contents may be high but often they are not available to plants (Lehmann *et al.*, 2003a). The N contents of ADE are typically higher than those of adjacent soils (Table 2), however this N does not need to be available to the plants (Lehmann *et al.*, 2003a).

As can be seen in Table 2, the largest proportion of N is usually present in soil in organic forms, and this is also the case for ADE, while more inorganic than organic P is usually present. The examples from cultivated fields indicate that the proportion of inorganic N and P increase in ADE compared to adjacent Ferralsols. The uncultivated fields showed lower proportions on inorganic than organic P in ADE than Ferralsols. No information is currently available for sulphur (S) (Lehmann *et al.*, 2003a).

Table 2: Inorganic and organic nutrient contents of ADE and comparable upland soils (values in brackets are percent of total) (Lehmann *et al.*, 2003a); n.d. - not determined

Location	Nitrogen		Phosphorus		Ref.
	inorganic [mg kg ⁻¹]	organic [mg kg ⁻¹]	inorganic [mg kg ⁻¹]	organic [mg kg ⁻¹]	
ADE	45 (2)	2755 (98)	n.d.	n.d.	[1]
Forest soils	39 (5)	811 (95)	n.d.	n.d.	[1]
ADE Açutuba (TP1)	127 (5)	2773 (95)	1642 (75)	541 (25)	[2]
ADE Caldaráo (TP2)	n.d.	n.d.	837 (79)	222 (21)	[2]
ADE Iranduba (TP3)	n.d.	n.d.	838 (86)	137 (14)	[2]
ADE Rio Preto da					
Eva (TP4)	n.d.	n.d.	662 (63)	389 (37)	[2]
ADE Belterra	407 (7)	5844 (93)	912 (63)	535 (37)	[2]
Adjacent soils (average*)	30 (3)	1100 (97)	287 (85)	52 (15)	[2]
ADE Marajá	n.d.	n.d.	1754 (57)	1342 (43)	[3]
ADE Rio Preto da Eva	n.d.	nd.	118 (62)	74 (38)	[3]
Ferralsol	n.d.	n.d.	23 (38)	36 (62)	[3]

[1] A-horizon of Amazonian Dark Earths (N=2) and clayey forest soils (Ferralsols and Acrisols; N=4) (Klinge, 1962). [2] Topsoil (0-0.1 m) of cultivated and fertilized fields (Glaser, 1999). [3] Topsoil (0-0.1 m) of uncultivated soil (Lehmann *et al.*, 2003c).

* average was chosen for reasons of clarity, although it is recognized that nutrient contents vary significantly between adjacent soils. Conclusions valid for individual comparisons, as well.

The soils of *Terra Firme* (Acrisols, Lixisols, Ferralsols according to WRB Taxonomy; Ultisols, Oxisols according to U.S. Taxonomy) differ from the adjacent soils. Nutrients availability in ADE depends more on nutrient release from exchange sites and soil organic matter through biological processes than weathering of parent material (Lehmann *et al.*, 2003a).

Table 3: Average extractable P (mg kg^{-1}) at 0-0.2m depth measured by 4 different extracting solutions on Amazonian Dark Earths (ADE) soil (Açutuba, $3^{\circ} 30' \text{ S}$ and $60^{\circ} 20' \text{ W}$), non-ADE soil and transition soils (Açutuba and Laranjal $3^{\circ} 30' \text{ S}$ and $60^{\circ} 40' \text{ W}$); means ($n=10$) (Falcão *et al.*, 2003)

Extractors	Dark Earths	Transitions 1	Transitions 2	Non Dark Earths
	$[\text{mg kg}^{-1}]$	$[\text{mg kg}^{-1}]$	$[\text{mg kg}^{-1}]$	$[\text{mg kg}^{-1}]$
Mehlich-1 ^a	131.5	43.5	35.9	3.2
Mehlich-3 ^b	52.3	51.6	47.1	2.5
Bray 1 ^c	98.3	38.5	29.6	5.2
Olsen mod. ^d	75.5	16.8	13.2	7.2

^a Mehlich-1 (0.05N HCl – H₂SO₄ 0.025N) (Mehlich, 1984).

^b Mehlich-3 (CH₃COOH 0.2N – NH₄NO₃ 0.25N – NH₄F 0.015N – HNO₃ 0.013N – 0.001 M EDTA) (Mehlich, 1984)

^c Bray 1 (HCl 0.025N – NH₄F 0.03N) (Bray & Kurtz, 1945).

^d Olsen modified (NaHCO₃ 0.5M + EDTA 0.01M + Superfloc 127. a pH 8.5) (Hunter, 1975).

In Table 3 we can see average extractable P measured by four different extracting solutions. Significant differences can be seen between Dark Earths and transition soils, but also between all four measurements. Despite these differences, the trend of higher amount of extractable P in ADE is obvious.

In Figure 3, the correlation between soil organic C and cation exchange capacity (CEC) is demonstrated. With higher C content in the soils, the CEC is increasing rapidly.

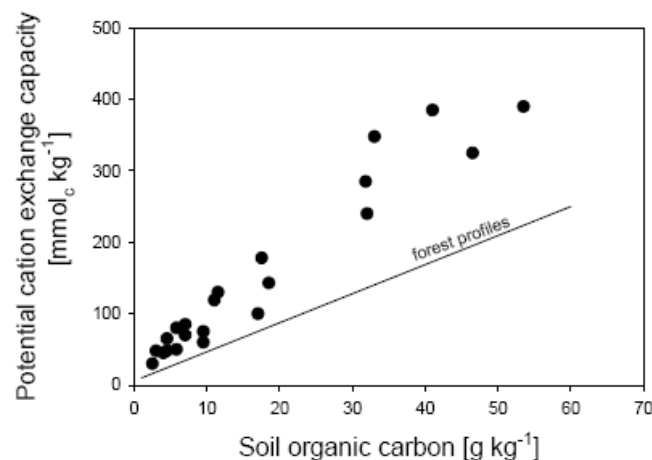


Figure 3: Potential cation exchange capacity of ADE (dots) (CEC determined with ammonium acetate buffered at pH 7) as influenced by organic C contents in comparison to forest soils (line) of similar clay mineralogy redraw after Sombroek *et al.*, 1993 (Lehmann *et al.*, 2003a)

2.2.3 Leaching

In tropical soils, applied nutrients are rapidly leached below the root zone of annual crops (Cahn *et al.*, 1993). In order to increase nutrient use efficiency, techniques must be developed to keep applied nutrients in the topsoil and therefore in the main root zone of the crop. Lehmann *et al.* (2003b) suggested two basic approaches which can be used to reduce nutrient leaching; applying slow-release nutrient forms such as organic fertilizers and increasing adsorption sites thereby retaining applied inorganic nutrients. Leaching of nutrients is minimal in ADE due to the retention capacity which provides an explanation for their sustainable fertility (Figure 4)(Lehmann *et al.*, 2003b). Cumulative leaching of mineral N, K, Ca, and Mg in the ADE was only 24, 45, 79, and 7%, respectively, of the amounts in adjacent soil (Lehmann *et al.*, 2003b). Relatively high leaching of Ca can be explained by high content of Ca in sorption complex.

2.2.4 Production potential

Higher yields on ADE may depend on crop species due to their different nutrient and water requirements. For example, crops with high K requirements, such as bananas, may not grow significantly better on ADE than at other soils, but the growth of crops which have high P requirements, such as legumes will increase significantly (Lehmann *et al.*, 2003a). Legumes usually grow better on ADE and their yields are high. It is caused by biological N fixation which is better in soil with large amount of charcoal.

In an on-farm experiment, maize grain (Figure 5) and growth (Figure 6) were significantly greater on ADE than Ferralsol (German, 2001, in Lehmann *et al.*, 2003a). The variability of grain yield was large and crop yields on ADE with a low production potential were also shown to be lower than those in Ferralsols (Figure 5). The dynamics of maize height (Figure 6) revealed more rapid crop growth on ADE than on adjacent soils during initial stages of development, whereas differences diminished towards maturation (German, 2001 in Lehmann *et al.*, 2003a).

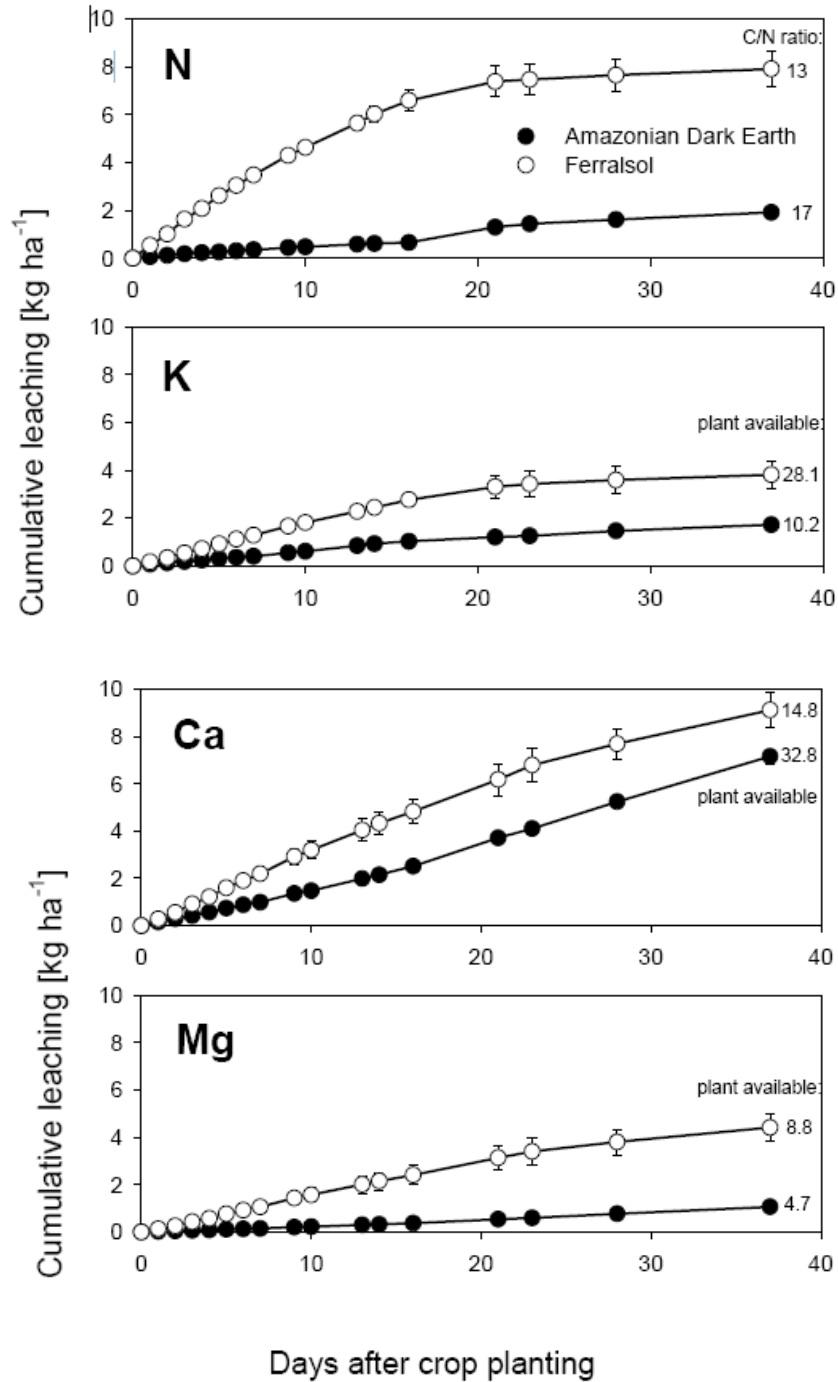


Figure 4: Nutrient leaching from an ADE and a Xanthic Ferralsol determined in lysimeters (0.2 m diameter) cropped to rice (*Oryza Sativa* L.) for 37 days, and nutrient availability (total C and N determined by dry combustion; cations extracted using KCl for Ca and Mg, Mehlich⁻¹ for K); means and standard errors, n=4 (Lehmann *et al.*, 2003a, modified after Lehmann *et al.*, 2003b)

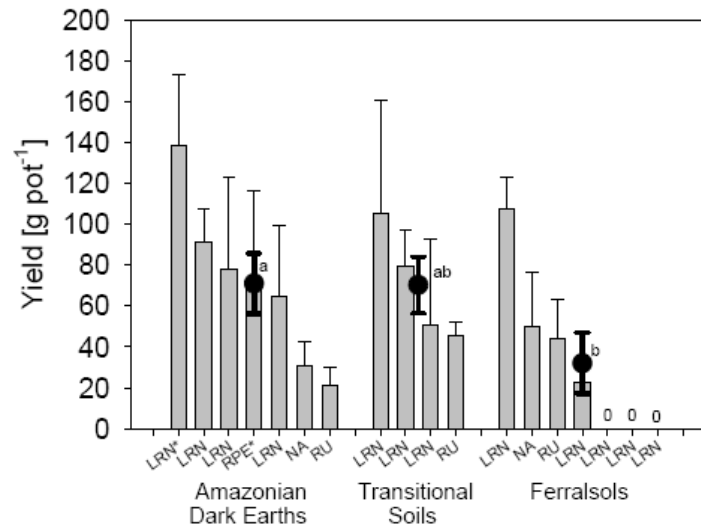


Figure 5: Grain yield of maize (*Zea mays* L.) grown in pots (0.3 m height, 0.2 m diameter) filled with unfertilized topsoil (0-0.3 m) of ADE containing artifacts, transitional soils, and Ferralsols randomly collected from smallholder farms along the Lower Rio Negro (LRN), Rio Preto da Eva (RPA), Novo Airão (NA) and Rio Urubu (RU) areas (bars: N=3 for each location, points: means of all ADE types and Ferralsols, respectively; completely randomized design; * from fertilized soils to commercial farms); three seeds planted per bag (missing plants were replanted in week two), harvest after 12 weeks; rainfed; grain was weighted after drying at 70°C for 24 hours (German, 2001) Adapted from Soils Fertility and Production Potential (Lehmann *et al.*, 2003a).

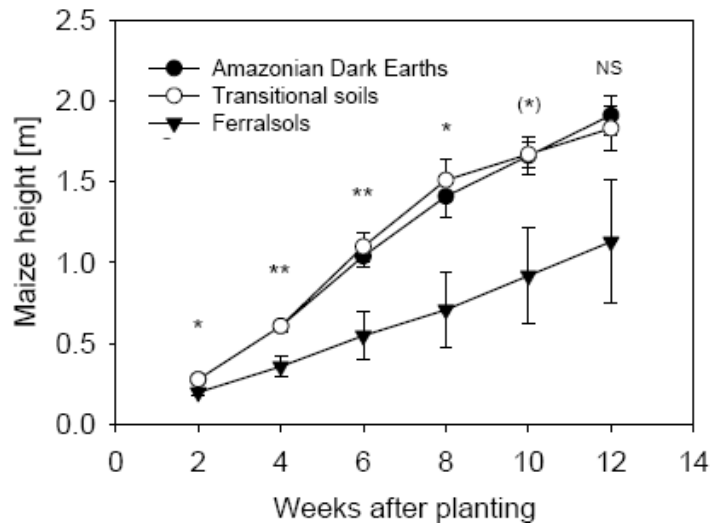


Figure 6: Growth of maize (*Zea mays* L.) grown on topsoil of ADE, transitional soils, and adjacent Ferralsol as described in Figure 5 (N=5, 4, 7, respectively) (German, 2001). Adapted from Soils Fertility and Production Potential (Lehmann *et al.*, 2003a).

Experiments of Lehmann *et al.* (2003b) showed that the mineral P fertilizer and equivalent amounts of manure did not result in higher above- or belowground biomass and nutrition of cowpea (*Vigna unguiculata* (L.) Wapl.) in comparison to the unfertilized control (Table 4). In contrast, a significant growth improvement was found for the Anthrosol and after charcoal amendments in the Ferralsols. P fertilization increased foliar P contents, but not total P uptake. N uptake of cowpea was significantly decreased by charcoal additions and in the Anthrosol, which was the effect of the poor N nutrition (Table 4). P nutrition and uptake were increased when charcoal was added to the Ferralsol and for the Anthrosol. Charcoal amendments improved foliar K nutrition and uptake in contrast to the Anthrosol, whereas K nutrition was even significantly reduced in comparison to the control (Table 4). In contrast, Ca contents and uptake were higher for cowpea grown in the Anthrosol than with charcoal applications, which did not increase compared to the unamended control. Mg contents and uptake of cowpea were reduced in both the charcoal amended soils and Anthrosols. Foliar Zn and Mn contents were improved in the Anthrosol, whereas foliar Zn contents did not change and Mn contents decreased after charcoal application (Table 4). The effect on foliar Cu contents were variable and may indicate a slight increase in charcoal amended soils. Charcoal additions and Anthrosol had no effect on foliar Fe contents of cowpea.

Table 4: Above and below ground biomass production and foliar nutrient contents of cowpea grown with the applications of mineral and organic fertilizers and charcoal in pots without leaching using Xanthic Ferralisol and Fimic Anthrosol in the central Amazon; values in one column followed by the same letter are not significantly different at $p < 0.05$ ($n=5$) (Lehmann *et al.*, 2003b)

Treatment	Shoot biomass (g pot ⁻¹)	Root biomass (g pot ⁻¹)	N (g kg ⁻¹)	P (g kg ⁻¹)	K (g kg ⁻¹)	Ca (g kg ⁻¹)	Mg (mg kg ⁻¹)	Fe (mg kg ⁻¹)	Zn (mg kg ⁻¹)	Mn (mg kg ⁻¹)	Cu (mg kg ⁻¹)
F	2.3 c	2.44 cd	51.9 a	2.00 cd	23.6 b	9.02 cd	7.56 ab	167	41.6 bc	176 c	2.6 d
F + Fert	2.2 d	0.46 cd	55.2 a	2.53 ab	25.3 b	9.04 cd	7.72 a	165	40.6 c	176 c	5.2 abc
F + Manure	1.9 d	0.48 cd	52.9 a	2.03 cd	25.8 b	9.01 cd	7.50 ab	162	43.0 bc	184 c	5.8 abc
F + Manure + Fert	2.6 bcd	0.36 d	52.5 a	2.85 a	24.3 b	9.38 c	7.18 b	337	44.6 b	205 abc	4.2 bcd
F + Charcoal	3.3 ab	0.42 d	23.9 b	1.96 d	33.1 a	8.17 cd	3.67 cd	116	41.2 bc	73.6 d	5.6 abc
F + Charcoal + Fert	2.9 bc	0.48 cd	24.9 b	2.27 bcd	34.8 a	7.88 cd	3.40 d	148	38.4 c	76.4 d	5.6 abc
F + Charcoal + Manure	3.5 ab	0.44 cd	23.4 b	2.32 bcd	35.7 a	7.45 d	3.32 d	77.8	39.8 c	69.6 d	7.0 a
F + Charcoal + Fert + Manure	3.7 a	0.44 cd	24.6 b	2.36 bcd	36.4 a	7.66 d	3.48 d	142	40.8 bc	77.8 d	4.6 ab
A	3.7 a	0.84 a	21.9 b	2.42 abc	17.3 c	13.2 ab	3.96 c	139	51.8 a	205 abc	6.8 ab
A + Fert	3.3 ab	0.62 b	24.1 b	2.65 ab	14.6 c	14.2 ab	3.92 c	168	55.0 a	244 a	3.2 cd
A + Manure	3.4 ab	0.68 b	22.5 b	2.33 bcd	16.9 c	12.7 b	3.72 cd	190	53.2 a	191 bc	2.2 d
A + Fert + Manure	2.9 bc	0.60 bc	23.5 b	2.52 ab	12.7 c	14.5 a	4.00 c	125	54.4 a	226 ab	2.8 d

F – Ferralisol; A – Anthrosol; Fert – fertilized with TSP; Manure – additions of chicken manure; Charcoal – applications at 10% weight.

In the same experiment (Lehmann *et al.*, 2003b), but this time with rice (*Oryza sativa* L.) also soil chemical properties were examined. The Anthrosol contained twice as much of the soil C compared to the Ferralsol (Table 5). Charcoal amendments to the Ferralsol, however, resulted in the highest soil C contents which were doubled compared to the Anthrosol. In contrast, total N contents did not increase in the same order of magnitude, which is reflected by higher C:N ratio in the charcoal-amended Ferralsols than the Anthrosol and than the Ferralsol without charcoal. The Anthrosol had higher pH value than the Ferralsol. Fertilization and liming significantly increased the pH, similar to the amendment with charcoal. However, soil available P contents were not significantly higher after fertilization. The Anthrosol showed a significantly higher P availability by one order of magnitude. Extractable K contents were higher in fertilized than in unfertilized soil and even higher in Ferralsols amended with charcoal. K and Mg availabilities were lower and Ca availability was higher in the Anthrosol than in the Ferralsol. Neither P, Ca, nor Mg contents were higher when charcoal was added to soil. The extractable Al contents were effectively reduced by both liming and charcoal additions. Manuring had no significant effect in soil chemical properties apart from a reduction of exchangeable Al.

Table 5: Soil carbon and nutrient contents, pH, acidity and cation exchange capacity of Xanthic Ferralsol and Fimic Anthrosol amended with inorganic and organic fertilizers and charcoal (only Ferralsol) after rice; (n=4) (Lehmann *et al.*, 2003b)

Treatment	C (g kg ⁻¹)	N (g kg ⁻¹)	C/N	pH (H ₂ O)	P (mg kg ⁻¹)	K (mmol _c kg ⁻¹)	Ca (mmol _c kg ⁻¹)	Mg (mmol _c kg ⁻¹)	Al (mmol _c kg ⁻¹)	CEC (mmol _c kg ⁻¹)
Ferralsol	39.7 d	3.17 c	12.6 c	5.14 e	8.1 c	28.1 e	14.8 e	8.8 de	2.3 a	54.0 e
F + Fert	39.2 d	3.03 c	12.9 c	5.93 b	16.9 c	168.8 d	32.1 c	20.1 b	0.2 de	221.1 d
F + Manure	37.8 d	3.02 c	12.5 c	5.16 e	8.1 c	35.8 e	15.0 e	9.8 d	1.7 c	62.3 e
F+ Manure + Fert	39.5 d	3.09 c	12.8 c	5.80 cd	21.0 c	189.3	36.1 b	22.5 a	0.0 e	247.8bcd
F + Charcoal	159.4 b	3.95 b	40.4 b	5.89 bc	10.5 c	258.3 ab	17.1 e	9.7 d	0.4 d	285.5 bc
F + Charcoal + Fert	156.2 ab	3.92 b	39.8 b	6.29 a	24.1 c	296.7 a	27.5 d	15.3 c	0.0 e	339.4 a
F + Charcoal + Manure	169.0 a	3.88 b	43.6 a	5.80 cd	9.5 c	220.0 bc	13.9	7.4 e	0.5 d	241.7 cd
F+ Charcoal + Fert + Manure	171.1 a	4.00 b	42.7 a	6.22 a	20.0 c	258.3 ab	27.9 d	15.8 c	0.0 e	301.9 ab
Anthrosol	84.7 c	4.96 a	17.1 d	5.71 d	318.4 b	10.2 e	32.8 c	4.7 f	2.0 b	49.7 e
A + Fert	85.0 c	4.93 a	17.2 d	5.93 b	386.1 a	173.9 d	50.6 a	22.2 a	0.0 e	246.7bcd

F – Ferralsol, Fert – fertilized with TSP, KCl and lime; Manure – additions of chicken manure; Charcoal – applications at 20 % weight; A – Anthrosol.

2.3 Amazonian Dark Earths replication

2.3.1 Biochar amendments and the influence on soil nutrient availability

Charcoal added to the soil increases its pH. Additions of biochar are increasing the availability of major cations, P and the total N concentrations (Lehmann *et al.*, 2003b). Availability of nutrients is generally higher than in any other soil in Amazon basin. It is not caused only by adding nutrients in biochar into the soil but also by the high nutrient retention. Charcoal amendments to soil increase cation exchange capacity of the soil which prevent leaching of nutrients. Adding charcoal to soil also increases seed germination (Chidumayo, 1994) (Table 6), plant growth, and crop yields (Glaser *et al.*, 2002b, Kishimoto and Sagiura, 1985) (Table 7).

Table 6: Seed germination rate (%) in seven indigenous woody plants under laboratory conditions in undisturbed and charcoal soils from miombo woodland at sites A and B in Chitemalesa area, central Zambia (Chidumayo, 1994)

Species	Undisturbed soil	Charcoal soil
<i>Acacia polvacantha</i>	5	30
<i>Bauhinia petersiana</i>	30	40
<i>Isoberlinia angolensis</i>	30	50
<i>Pterocarpus angolensis</i>	0	30
<i>Swartzia madagascariensis</i>	0	20
<i>Tamarindus indica</i>	10	70
<i>Ziziphus mauritiana</i>	0	40

Significant improvements in productivity ranging from 20 to 200% can be observed even at very low application rates of 0.4 to 8 t C ha⁻¹ (Lehmann *et al.*, 2006c). Too high amendments can lead to high C:N rates. Optimal rate C:N is 25-30:1. The microorganisms can be immobilized if the ration is too wide. Lehmann *et al.* (2006c) assessed that legumes thrives well even with high biochar amendments. This is probably caused by the biological N fixation of legumes which can compensate the nitrogen missing in the soil and lower the high C:N rate.

As can be seen from the Table 7 (Glaser *et al.*, 2002b) Chidumayo (1994) reported better shoot heights (24%) and biomass production (13%) among seven indigenous woody plants on soils under charcoal kilns compared to the undisturbed Zambian Alfisols and Ultisols. Kishimoto and Sagiura (1985) found that the heights of sugi trees (*Cryptomeria japonica*) increased by a

factor of 1.26-1.35, and biomass production increased by a factor of 2.31-2.36, five years after application of 0.5 Mg of charcoal ha⁻¹. Similar observations were made after additions of humic acids from charcoal deposits, which increased maize growth by up to 1 g kg⁻¹ on Nigerian Alfisols and Inceptisols (Mbagwu and Piccolo, 1997). Also soybean and maize showed diminished yields with addition of charcoal (Kishimoto and Sagiura, 1985).

Table 7: Relation between charcoal amendment to soil and crop response (Lehmann *et al.*, 2002b)

Treatment	Amendment (Mg ha ⁻¹)	Biomass production (%)	Plant height (%)	Root biomass (%)	Shoot biomass (%)	Plant type	Soil type	Reference
Control	-	100	100	-	-	Bauhinia wood	Alfisol/Ultisol	Chidumayo (1994)
Charcoal	Unknown	113	124	-	-	Bauhinia wood	Alfisol/Ultisol	
Control	-	100	-	-	-	Soybean	Volcanic ash soil, loam	Kishimoto and Saguira (1985)
Charcoal	0.5	151	-	-	-	Soybean	Volcanic ash soil, loam	Iswaran et al.(1980)
Charcoal	5.0	63	-	-	-	Soybean	Volcanic ash soil, loam	Kishimoto and Sagiura (1985)
Charcoal	15.0	29	-	-	-	Soybean	Volcanic ash soil, loam	
Control	-	100	-	-	-	Pea	Dehli soil	Iswaran et al. (1980)
Charcoal	0.5	160	-	-	-	Pea	Dehli soil	
Control	-	100	-	-	-	Moong	Dehli soil	
Charcoal	0.5	122	-	-	-	Moong	Dehli soil	
Control	-	100	-	100	-	Cowpea	Xanthic Ferralsol	Glaser et al. (2002a;2002b)
Charcoal	33.6	127	-	-	-	Oats	Sand	
Charcoal	67.2	120	-	-	-	Rice	Xanthic Ferralsol	
Charcoal	67.2	150	-	140	-	Cowpea	Xanthic Ferralsol	
Charcoal	135.2	200	-	190	-	Cowpea	Xanthic Ferralsol	
Control	-	100	100	100	100	Maize	Alfisol	Mbagwu and Piccolo (1997)
Coal humic acid	0.2	118	114	122	114	Maize	Alfisol	
Coal humic acid	2.0	176	145	186	166	Maize	Alfisol	
Coal humic acid	20.0	132	125	144	120	Maize	Alfisol	
Control	-	100	100	100	100	Maize	Inceptisol	
Coal humic acid	0.2	125	119	122	127	Maize	Inceptisol	
Coal humic acid	2.0	186	148	198	173	Maize	Inceptisol	
Coal humic acid	20.0	139	131	147	130	Maize	Inceptisol	
Control	-	100	100	100	-	Sugi trees	Clay loam	Kishimoto and Sagiura (1985)
Wood charcoal	0.5	249	126	130	-	Sugi trees	Clay loam	
Bark charcoal	0.5	324	132	115	-	Sugi trees	Clay loNo am	
Activated carbon	0.5	244	135	136	-	Sugi trees	Clay loam	

2.3.2 Microbial response to charcoal amendments

Also the microbial processes in soil are influenced by biochar amendments as it was assessed by Nishio (1996) by the stimulation of the growth of indigenous arbuscular fungi. In the same research, nodule formation, nodule weight (2.3 times), and nitrogen uptake (2.8 - 4 times) were improved by biochar additions. Pietikäinen *et al.*(2000) used biochar as an adsorbent of growth-inhibiting organic substances.

Its generally known and accepted that microorganisms have positive influence on the soil. But it is necessary to think about soil and microorganisms as the complex. Not only adding microbial fertilizers to soil can improve soil properties and fertility but also adding charcoal for improving conditions for microorganisms can help.

When the soil was sterilized by chloropicrin, alfalfa growth was greatly reduced. The stimulatory effect of charcoal on plant growth also diminished. On the other hand, vigorous plant growth and the stimulatory effects of charcoal addition were clearly observed when the sterilized soil was mixed with a large amount of native soil. This clearly indicates that the stimulatory effect of added charcoal may appear only when a certain level of indigenous AMF are present (Nishio, 1996).

Charcoal may stimulate the growth of AMF by the following mechanism. Charcoal particles have a large number of continuous pores with a diameter of more than 100m. They do not contain any organic nutrients, because of the carbonization process. The large pores in the charcoal may offer a new microhabitat to the AMF, which can obtain organic nutrients through mycelia extended from roots. This may enable the AMF to extend their mycelia far out from the roots, thus collecting a larger amount of available phosphate (Nishio, 1996).

Although many legume species live in symbiosis with microorganism of the *Rhizobium* genus even in the soils without biochar, this biological fixation is influenced by biochar amendments. Soils with large concentrations of biochar, such as ADE, have usually low nitrate concentration of available nitrate and high concentration of available Ca, P, and micronutrients. This is ideal for maximum biological N fixation (Lehmann *et al.*, 2003a). Biological N fixation determined by nitrogen difference was found to be 15% higher when biochar was added to the soil at early stages of alfalfa development, and 227% higher when nodule development was greatest (Nishio, 1996).

2.3.3 „Terra Preta Nova“ project

Data mentioned above led to the formulation of the multi-disciplinary and multi-institutional Brazilian research+development project called Terra Preta Nova (TPN), with participation of scientists from other Latin American countries, the USA and Europe (Sombroek *et al.*, 2002). These „new“ soils could be used to sequester carbon dioxide from the atmosphere. This participant countries, institutions and organizations cooperate and continue in reserach and investigation of ADE properties and biochar utilization in their recreation.

In the process of photosynthesis CO₂ is consumed by the plants. Therefore, Kyoto Protocol allows countries with large areasa of forests or other vegetation deduct a certain amount from their emissions (Kyoto Protocol, 1998). There are several techniques of incorporation of C to the soil in form of biochar.

2.4 *New perspectives of biochar*

Lehmann *et al.* (2006a) proposed a new approach to C sequestration in terrestrial ecosystems through the application of biochar to soil, which offers both large and long-term C sink (figure 3). This C sequestration in the soil has been occuring in nature for centuries or thousands of years, typical example are ADE. Even now after hundreds and tousands of years without being cultivated these earths contain large amounts of biochar derived C stocks. The total C storage is as high as 250 Mg C ha⁻¹m⁻¹ compared to typical values of 100 Mg C ha⁻¹m⁻¹ in Amazonian soils derived from similar parent material (Glaser *et al.*,2001).

Climate change and the anthropogenic greenhouse effect, caused by carbon dioxide (CO₂), methane (CH₄) and nitrous oxides (NO_x), are some of the most important challenges facing the modern world. These gases are mostly generated by burning of fossil fuels and from decomposition of above- and belowground biomass (Lehmann *et al.*, 2006a).

There are many strategies trying to prevent global warming. One of them is sequestration of carbon in environment which leads to decreasing C concentration in the athmosphere.

However, carbon sequestration is not the only positive effect of biochar on the environment.

The most important opportunities to incorporate a biochar soil management technique are in (Lehmann *et al.*, 2006a):

- Shifting cultivation
- Charcoal production
- Recycling of agricultural and forestry wastes
- Energy production using renewable fuels (bio-fuels)
- Cropping for biochar using fast-growing trees

In all five systems, biochar can be produced and applied to soil.

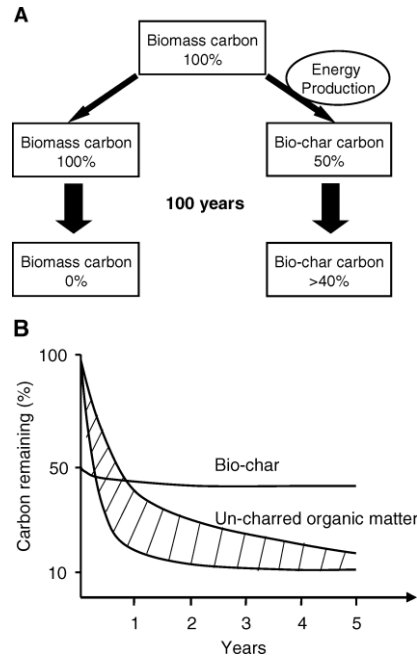


Figure 7: Schematics of differences between C remaining after decomposition in case of biomass and biochar (Lehmann *et al.*, 2006a)

- (A) C remaining from biomass after 100 years from IPPC (1996) ; C remaining after charring or pyrolysis (FAO 1983); biochar C remaining after decomposition
- (B) Range of biomass C remaining after decomposition of crop residues from Jenkinson and Ayanaba (1977), in Lehmann *et al.*, 2006a.

2.4.1 Shifting cultivation slash-and-burn vs. slash-and-char

Shifting cultivation is one of the traditional land use systems which are usually being used in tropics and subtropics, especially in small areas. Farmers start cultivation with cutting down the vegetation, it can either be primary rain forest or vegetation growing there in the period of fallow. After cutting everything down they usually burn new area so they can start to

use soil for crop cultivation. After cropping period which is usually 1-5 years long the fallow period comes for about 5-25 years. Under this system soil fertility declines rapidly and weed pressure increases (Lehman *et al.*, 2006a).

At this time of fast population growth it is necessary to intensify agriculture to secure food supply. Traditional shifting cultivation is being modified. Fallows are getting shorter and sometimes they are even skipped which leads to decreasing soil fertility and irreversible degradation of the soil. In traditional slash-and-burn systems people have to find more and more areas for cultivating crops to feed increasing population. This leads to extensive deforestation. Burning also produce large amount of carbon dioxide which has been recently important ecological problem in the world. However, slash-and-burn technique can improve the soil fertility, but only for short period of time. In highly weathered soils in tropics the leaching is very fast and causes significant nutrient losses. That is the reason why it is necessary to add fertilizer to soils regularly to sustain soil fertility (Glaser *et al.*, 2002b). Same problem is with organic matter which mineralizes quickly into CO₂ (Lehmann *et al.*, 2006c).

Using slash-and-char technique for making biochar and putting it back to the soil would sequester more than 50% of C in highly stable form (Lehmann *et al.*, 2006a). For comparison, only about 3% of the aboveground biomass would be converted into forms similar to biochar under a typical slash-and-burn system (Glaser *et al.*, 2002). Slash-and-char technique does not advocate the destruction of existing primary forests. It should be a carbon- and nutrient-conserving alternative to the existing slash-and-burn technique. In this way, carbon will rather be retained in the system compared to slash-and-burn, since only the biomass from the same cropping area will be used for producing the charcoal (Lehmann *et al.*, 2002b).

If shifting cultivation is to be successful, (i) the quality of applied charcoal must be produced from the same area of land which is to be cropped, and (ii) the periods of charcoal production must at least correspond to those of land clearing practiced so far (Lehmann *et al.*, 2002b). The long-term dynamics of soil fertility with charcoal applications are remarkable. It may be assumed that nutrients bounded to charcoal are more persistent than those in ash but direct evidence needs to be gathered (Lehmann *et al.*, 2002b).

2.4.2 Biochar wastes from charcoal production

In the areas where has always been enough of wood this wood has been used for making charcoal since ancient times. People were collecting wood and sawdust and making big piles of it. At the bottom of these piles there was a hole so the air could get into the middle of the pile. Characteristic feature of these charcoal making piles was that they were empty with a flue in the middle. The burning starts near the hole at the bottom. It was necessary to cover whole pile with some wet material such us clay, turfs, mud or bricks to ensure limited access of the air into the pile which prevents the charcoal from burning. This process has been still used in many developing countries.

Half of the world's population uses biomass fuels for cooking. In 1992, 24 million tones of charcoal were consumed worldwide. Developing countries account for nearly all of this consumption, and Africa alone consumes about half of the world's production. Charcoal production has increased by about a third from 1981 to 1992, and is expected to increase with the rapidly growing population in the developing world (Kammen *et al.*, 2005).

In small-scale production systems, waste charcoal remains within the area of production or is discarded locally. The proportion of charcoal waste varies significantly depending on the production procedure, the wood properties, the charcoal processing, and the market demands (FAO, 1983).

Charcoal producers can be devided into two groups. First of them are small-scale farmers selling small amounts of charcoal to make money for their subsistence. The second group is made of people selling charcoal as a business, making charcoal just for the purpose to make money.

Recent studies point to the promise of rain forest extraction for more sustainable rural development in Amazonia but often overlook important differences within traditional communities in terms of relative economic reliance upon specific forest resources (Coomes, and Burt, 2001). The results of their study indicate that peasant charcoal production can provide significant cash income to for the forest peoples and high returns per hectare, particularly when intergrated into swidden-fallow agroforestry systems, without causing notable forest destruction. Also Labarta *et al.* (2008) confirmed this in their study in Peruvian Amazon.

Although most of the peasants are making charcoal for market to be used as fuel these information can be also used also in biochar production. Charcoal and biochar are both produced by the same process, slow pyrolysis. For charcoal production bigger pieces of wood are being used, such as logs and big blocks, and the purpose of charcoal production is usage as a fuel. On the contrary for biochar production smaller pieces of wood are being used, such as swarf and chips, and purpose of production is application to the soil.

2.4.3 Recycling of agricultural and forestry wastes

In many agricultural and forestry production systems, waste is produced in significant amounts from crop residues (Table 2) such as (Walsh *et al.*, 1999):

- Forest residues (logging residues, dead wood, excess saplings, pole trees)
- Mill residues (lumber, pulp, veneers)
- Field crops residues
- Urban wastes (yard trimmings, site clearing, pallets, wood packaging)

Rice husk are typically regarded as a waste product, but can be used to sequester C by producing biochar. Global rice paddy production is 0.589 Pg yr⁻¹. From this it was calculated the sequestration potential to be 0.038 Pg C yr⁻¹ (calculated estimating 32% husk, 38% C concentration, and 53.5% conversion from husk C to biochar C) (Lehmann *et al.*, 2006a). Using rice husks for biochar production can play very important role in C sequestration in soils due to big world production of rice.

Table 8: Availability, suitability and global production of agricultural waste materials (Lehmann *et al.*, 2006a)

Waste materials	Availability	Suitability	Potential global production of biochar (Pg yr ⁻¹)
Forest residues	Medium	High	0.04
Mill residues	High	High	0.05
Rice husks	High	Medium	0.04
Groundnut shells	High	High	0.002
Urban waste	High	High	0.03
Total			0.162

2.4.4 Energy production using renewable fuels

Besides the most common way of energy production from biomass, pyrolysis and gasification of biomass can be promising. Furthermore, pyrolysis is of interest due to the relatively high biochar production. In the context of biochar sequestration, only pyrolysis is of interest due to the simultaneous biochar production (Lehmann *et al.*, 2006a).

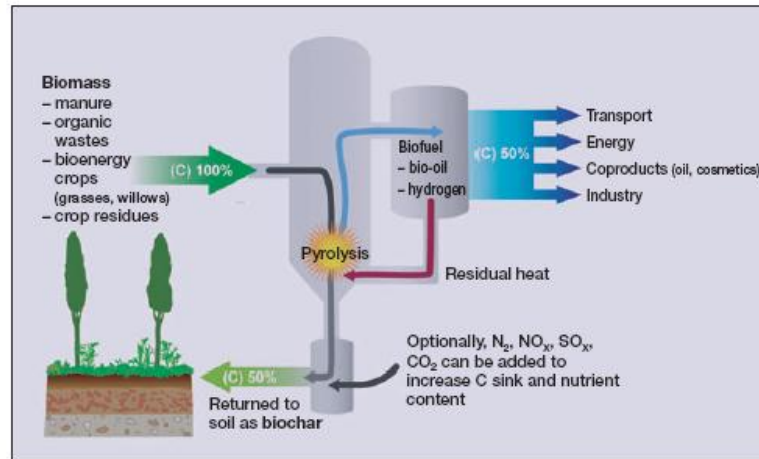


Figure 8: Concept of low-temperature pyrolysis bio-energy with biochar sequestration. Typically, about 50% of the pyrolyzed biomass is converted into biochar and can be returned to soil (Lehmann, 2007)

Pyrolysis is the basic method to produce fuel from biomass (Figure 6). It is chemical decomposition of organic materials by heating without access of the air. The temperatures necessary for different types of organic materials to decompose vary but generally they are between 300 and 600°C. Products of pyrolysis are bio-oil, biochar and syngas. The amounts of these components vary according to the process. Most biochar can be obtained in process of slow pyrolysis, up to 35%. On the other side for bio-oil production the best process is fast pyrolysis in which we can obtain up to 75% of bio-oil (Table 4).

Table 9: Fate of initial feedstock mass between products of pyrolysis processes (Sohi *et al.*, 2009)

Process	Liquid (bio-oil)	Solid (bio-char)	Gas (syngas)
FAST PYROLYSIS			
Moderate temperature (~500°C), Short hot vapour residence (<2s)	75% (25% water)	12%	13%
INTERMEDIATE PYROLYSIS			
Low-temperature, Morderate hot vapour residence time	50% (50% water)	25%	25%
SLOW PYROLYSIS			
Low moderate temperature, Long residence time	30% (70% water)	35%	35%
GASIFICATION			
High temperature (>800°C), Long vapour residence time	5% tar 5% water	10%	85%

2.4.5 Cropping for Biochar

Cropping for biochar production for atmospheric CO₂ sequestration can seem to be promising method. Lehmann *et al.*, (2006a) suggests the combination of biochar production with recent agricultural land-use method rather than sole purpose crop production for biochar. This system can be more beneficial due to the possible high inputs.

Also the quality of biochar is important for the following use of biochar as the soils conditioner. Because how was mentioned earlier different biomass types produce biochar of different quality. Especially properties such stability against decomposition, efficiency to improve soil fertility, and ability to provide other ecosystem services should be considered (Lehmann *et al.*, 2006a).

2.4.6 Environmental benefits of biochar

Biochar in the soil influence the environment in many ways and all of them are positive. The most important environmental benefits are:

- Mitigation of climate changes, CO₂ sequestration
- Improvement of the soil
- Reduction of pollution of waterways
- Scrubbing of air pollutants (Lehmann, 2007)

2.4.6.1 Mitigation of climate change, CO₂ sequestration

When CO₂ emissions from fossil fuels become airborne, their full elimination from the atmosphere takes a very long time. One quarter of fossil fuel-derived CO₂ emissions remains in the atmosphere for several centuries and complete removal of CO₂ may take 30 000-35 000 years (Archer, 2005).

The Kyoto Protocol (KP) agreement on reducing the net emission of greenhouse gases in general, and of CO₂ in particular, recognizes the importance of C in the soil as a store, source and potential sink of CO₂, in addition to supporting functions of the aboveground biomass (Sombroek *et al.*, 2003).

Basically there are three ways of CO₂ offset from the atmosphere discussed lately such as forestations projects, offsetting by landfilling purpose-grown biomass and carbon offsetting by biochar.

As regard to climate changes the biggest advantage of biochar in the soil is its capability to absorb CO₂ from atmosphere. CO₂ from atmosphere is assimilated by plants. These plants are pyrolysed as biomass. Pyrolysis produces energy and biochar going back to soil as carbon store and sink. (Figure 5). The amount of biochar produced in pyrolysis depends on the temperature of pyrolysis and also on the material used for pyrolysis. Preliminary results indicate that biochar bio-energy not only leads to a not sequestration of CO₂, but that presence of biochar in soil may decrease emissions of two even more potent greenhouse gases, nitrous oxide (NO_x) and methane (CH₄). It can be caused by a higher C:N ratio (Lehmann, 2007).

2.4.6.2 Improvement of the Soil

The potential of biochar as a soil condition has been mentioned. For bio-energy production biomass removal from the land is necessary, which can lead to erosion and the loss of nutrients in the ecosystems (Lehmann, 2007). However, returning biochar back to the soil leads to the opposite situation, because of the larger amount of charcoal than in biomass. Additionally, this biochar C is more stable.

2.4.6.3 Other Benefits

Charcoal as material preventing leaching is well known. Due to this property of biochar waterways can be protected by retaining nutrient in the soil by biochar. Another way

to reduce waterways pollution is the fact that biochar improves nutrient retention in the soil which leads lowering amount of fertilizers needed to grow a crop (Lehmann, 2007).

Additionally, the ability to scrub CO₂, nitrous oxides, and sulfur dioxide from flue gas is another positive characteristics of biochar (Day *et al.*, 2005). This process produce nitrogen-enriched, slow-release, carbon-sequestering fertilizer and it consists in char-affinity for capturing CO₂ through gas phase reaction with mixed nitrogen-carrying nutrient compounds within the pore structure of carbon char (Day *et al.*, 2005).

Hilber *et al.*, (2009) performed experiment about the influence of activated charcoal amendments to contaminated soils on dieldrin and nutrient uptake by cucumbers. During this experiment it was shown that dieldrin fresh weight concentrations in cucumber fruits were significantly reduced. Although biochar does not help to remove dieldrin from the soil, decrease plant uptake by biochar can help to keep this pollutant out of the food chain. Dieldrin is chlorinated hydrocarbon which is being used as insecticide, especially against ticks and termites. Furthermore, it is extremely persistent organic pollutant toxic for mammals. For this reason it used to be used also as rodenticide.

Yu *et al.* (2009) realized experiment confirming the theory about capability of biochar to decrease plant uptake of contaminants from the soil. Spring onions were grown on soil containing biochar and on control soil samples. The analysis showed that although the pesticides (chlorpyrifos and carbofuran) are more persistent in soils with biochar the plant uptake is much lower.

3 OBJECTIVES

Based on literature review we have settled the hypothesis that addition of the biochar to the unfertile soil can enhance its fertility, which can lead to substantial improvement of the plant growth. This positive influence can probably be used to improve traditional shifting cultivation in Peruvian Amazon.

The first objective of this study is to assess the influence of biochar on soil fertility and biomass production in pot experiment. Biochar addition into the Amazonian Ultisol is expected to improve water retention and the structure of the soil, increase soil nutrient content by preventing leaching, and influence uptake of nutrients, and thus improve the biomass production and the quality of this biomass. This hypothesis, based on previous literature study, is the positive effect of biochar on soil nutrient content, plants growth and foliar nutrient uptake. The plant growth improvement is expected in the case of all soil amendments used during the research. However, the soil fertility improvement potential after biochar addition is expected to be higher than in case of chicken manure or inorganic fertilizer application.

Additionally, the aim is the assessment of the potential of biochar obtained from wood shaving for the soil improvement potential in comparison with biochar obtained from charcoal production. Different effects of two types of biochar on plant growth are expected according to their different nutrient content, C content and CEC.

The second objective is the analysis of the over-all recent utilization of biochar for agricultural purposes and the potential of agricultural biochar production from wood shavings from saw-mills in Pucallpa, one of the most important timber centers in Peruvian Amazon.

4 MATERIALS AND METHODS

4.1. Study area

4.1.1. Climatic conditions and vegetation in study area

Peruvian Amazon is located in the lowlands to the east from the Andes. The climate is tropical with low temperature fluctuation during the year. The relatively drier months with precipitation lower than 100 mm are June, July, August and September (Figure 9). The mean accumulated precipitation in Pucallpa is 1569mm (Rivas Martinez, 2009), in year 2011 1614 mm (Anonym, 2012).

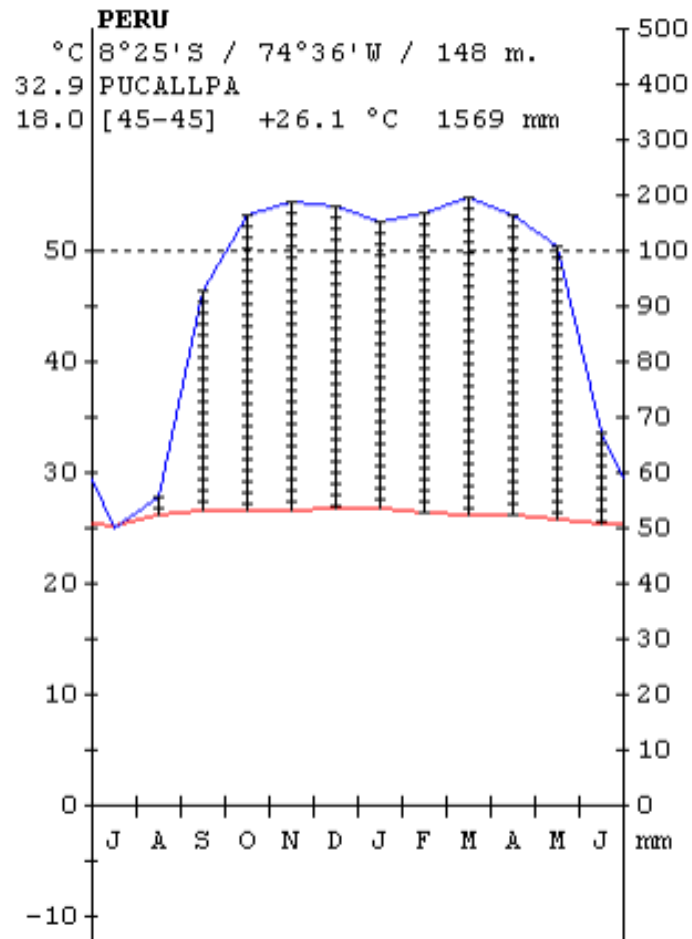


Figure 9: Climate diagram of Pucallpa during 1996-2009 (Rivas Martinez, 2009)

Dominant vegetation of Peruvian Amazon is evergreen forest (Benites, 1982) formed by groups of tree species with an important commercial value. The major soil orders occurring in

Peruvian Amazon are listed in Table 10. The main agricultural system on these soils is slash-and-burn agriculture leading to the expansion of agriculture into the tropical rain forest and causing deforestation. Despite the low fertility and very low pH of these soils, their large extension in comparison with fertile soils suggests that the soils fertility improvement efforts and food production should focus on Ultisols and Inceptisols which are located on the slopes and uplands (Benites, 1982).

4.1.2. Charcoal production

Pucallpa is considered to be one of the three access gates into Peruvian Amazon. Thus, it is one of the largest timber and charcoal producers in Peru. Charcoal production plays significant role in indigenous peoples' lives and is an important cash income without causing significantly deforestation, which was concluded by Coomes and Burt (2001) in the study of peasant charcoal production in the northern part (Iquitos area) of Peruvian Amazon. In the same study peasants mostly integrate charcoal production into swidden-fallow agroforestry systems. The main constrain is low labor return, therefore, charcoal production does not promise prosperity. Low labor return in Iquitos area can be causing significant reduction in shifting from agriculture to charcoal production, compared to Pucallpa, where the returns to labor are much higher (Labarta *et al.*, 2008). Also the fact that charcoal production has a potential to decrease deforestation by 17% and increase net income by 17% within only 10 years should not be neglected (Labarta *et al.*, 2008).

4.1.3. Predominant soils in Peruvian Amazon

The most typical soil in this area is Ultisol (according U.S. Soil Taxonomy, 1999). According to Land Survey unit, CIAT, Ultisols occupy 59.9% of the Peruvian Amazon (Table 10). Ultisols are red clay acid soils with low base status with a vast potential for agricultural production. The production of Ultisols is generally good in first few years, but cutting of native forests often leads to rapid degradation in soil fertility. This is caused by the dependence of these soils on nutrient recycling by deep-rooted plants for maintenance of fertility in the surface soil. The Ultisol order is subdivided into five suborders, using criteria of profile wetness and organic matter content (Buol *et al.*, 1989).

Table 10: Proportion of soil orders occurring in Amazon basin of Peru (Land survey unit, CIAT, as cited in Benites, 1982)

Soil orders	Proportions (%)
Ultisols	59.9
Alfisols	22.4
Inceptisols	11.8
Oxisols	5.4
Mollisols	0.5

Acid, infertile soils (Oxisols and Ultisols) are the most common soils in whole Amazon basin (Cochrane and Sanchez, cited in Sanchez *et al.*, 1982), occupying 361 millions of hectares which is 75% of Amazon basin, respectively.

The cause of the release of bases in most cases is weathering rather than leaching. Vegetation plays a significant role in the distribution of the base cation in the soils horizons. Therefore, with the depth the base saturation decreases as a result of the concentration of the bases in the shallow depth by the vegetation. Therefore, if there are no amendments applied into the soils, the only cultivation system is shifting cultivation (U.S. Soil Taxonomy, 1999).

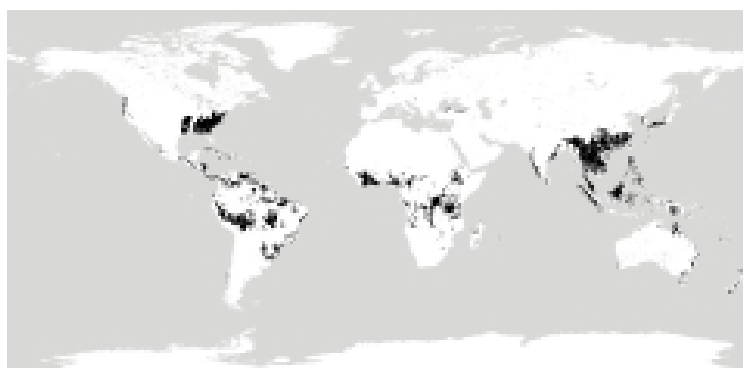


Figure 10: Ultisol distribution in the world (U.S. Soil Taxonomy, 1999)

4.1 Pot experiment

4.1.1 Experimental design

During the pot experiment we wanted to assess the influence of the biochar on the plant growth. The experiment took place in the agroforestry nursery at The National University of Ucayali (UNU - Universidad Nacional de Ucayali) in Pucallpa (8° 24' S; 74° 34' W) from July

25th to September 5th 2011. The mean month precipitation for the three months period was 26°C (Table 11).

Table 11: Climatic conditions during the pot experiment (Anonym, 2012)

Month	Mean temperature (°C)	Maximum temperature (°C)	Minimum temperature (°C)
July	25.3	32	18.4
August	25.9	34	18
September	26.9	33.8	20

For the experiment we have chosen two locally grown crops and one native tree from three distinct families - rice, (*Oryza sativa* L. – Poaceae); cowpea, (*Vigna unguiculata* [L.] Walp – Fabaceae) and bolaina blanca, (*Guazuma crinita* Mart. – Malvaceae). The plants were grown in plastic bags with two kilograms of Amazonian Ultisol and different additions of two types of biochar, partly decomposed chicken manure and inorganic fertilizer, all with six repetitions. Both types of biochar, chicken manure were applied in amount of 350 g per plastic bag as it is listed in Table 12.

Table 12: Experimental design of pot experiment with cowpea and rice

Treatment code	Ultisol (kg)	Biochar I. (g)	Chicken manure (g)	NPK (g)	Biochar II. (g)
U (control)	2000	-	-	-	-
B I.	2000	350	-	-	-
MB	2000	350	350	-	-
MBF	2000	350	350	7	-
M	2000	-	350	-	-
B II.	2000	-	-	-	350

Biochar I. – ground charcoal from local market

Biochar II. – biochar made from wood shavings.

The soil for the pot experiment was taken from devastated and deforested area from about 40 km from Pucallpa, district Campo Verde. This soil had been without vegetation and was supposed to be used only for road construction purposes. Analysis (Table 13) showed high clay content (63%) and low pH (4.9) (measured in 1:1 soil and water suspension).

Table 13: Macronutrient contents of the Ultisol (U) and used amendments analyzed before planting

Amendment	C ¹⁾ (g kg ⁻¹)	P ²⁾ (g kg ⁻¹)	K ³⁾ (g kg ⁻¹)	Ca ³⁾ (g kg ⁻¹)	Mg ³⁾ (g kg ⁻¹)
U	32	0.004	1.2	1.6	0.21
B I.	460	2	1.5	15.5	1.7
B II.	380	4.3	4.6	26	1.7
M	93	51	34	65	67

¹⁾ oxidizable organic carbon determined according to Walkley (Walkley, 1947);

²⁾ determined by spectrophotometry, TURNER system, Model 39-Olsen modified method (NaHCO₃ 0.5M + EDTA 0.01M + Superfloc 127) with ammonium molybdenate, ascorbic acid and antimony (Olsen and Sommers, 1982)

³⁾ Mehlich III extraction and atomic absorption spectrophotometry (Mehlich, 1984)

The charcoal was bought on local market and originally prepared from a tree species *Calycophyllum spruceanum* (Benth.) Hook.f. ex K.Schum., that is locally widely used hardwood species. The high quality wood is mainly used for construction purposes and the rest for charcoal production. Charcoal was manually ground and sieved through a 6-mm aperture sieve. This charcoal (biochar I.) had relatively low content of carbon, 460 g kg⁻¹ respectively. pH of biochar I. was 8.01 measured with the same method like the soil.

Pucallpa is relatively large producers of sawdust and wood shaving. This material has potential to be used for biochar production and later application to the soil as soil conditioner. In this experiment, biochar (biochar II.) was prepared from the wood shavings from *Calycophyllum spruceanum* (Benth.) Hook.f. ex K.Schum. which were obtained from the local wood-processing factory. This charcoal was created in a barrel (appendix 4) with limited access of the air and had relatively low carbon content (380 g kg⁻¹). The pH of this biochar was 6.68 determined with the method described above.

For analysis of both types of biochar and chicken manure were used the same methods as in case of soil analysis.

Cowpea was sown in three planting holes per pot, rice in eight planting holes per pot. Not germinating seeds were replaced by germinating seeds within the first four days after the establishment of the experiment. Therefore, the total amount of plants in case of rice was eight plants per pot and in case of cowpea three plants per pot. Then 40 days old seedlings of *G. crinita* were bought from the local nursery and planted one plant per pot. In case of *G. crinita* the stem and root lengths were measure before planting to be compared with the stem and root length after six weeks of the duration of experiment.

The experiment was established as completely randomized block design using three plant species, six soil treatments, each with six repetitions. The treatment number six with biochar II. (made from wood shavings) was used only in case of cowpea and rice. Altogether there were 102 plastic bags (36 with rice, 36 with cowpea and 30 with *G. crinita*). The plants were regularly watered, controlled and shaded to prevent water stress.

4.1.2 Data collection and evaluation

Biomass production, stem and root length were measured six weeks after the establishment of the experiment. The plants were cut at the soil surface level, the roots were dug out by hands and washed out. Both parts (above- and belowground biomass) were dried in 70°C until the constant weight and weighted. In case of *G. crinita* the stem and root lengths were compared with the stem and root length before the experiment.

The dried aboveground biomass of plants was analyzed in laboratory for macro- and micronutrients. The K, Ca, Mg, Zn, Cu and Fe contents were measured by wet digestion with Atomic Absorption Spectrometry with flame atomizer (Lin and Coleman, 1960). The P content was determined by spectrophotometry (TURNER system, Model 390) from the same extract (molybdenum-blue method) (Olsen and Sommers, 1982). Total Kjeldahl nitrogen (Bremner and Mulvaney, 1982) was measured using GERHART system, Vapodest.

The substrates from the pots were dried, homogenized and ground and samples were obtained from each pot. pH was determined in 1:1 (soil:water) suspension with pH meter (McKeague, 1978). Oxidizable organic carbon was determined according to Walkley (Walkley, 1947). The P content was determined with spectrophotometer (Spectrophotometry, TURNER system, Model 390) using Olsen modified method (NaHCO_3 0.5M + EDTA 0.01M + Superfloc 127) with ammonium molybdenate, ascorbic acid and antimony (Olsen and Sommers, 1982). The Ca and Mg were measured by instrumental analysis with atomic absorption spectroscopy with flame atomizer. Atomic absorption spectroscopy was used also in case of measurement of K content in the extract (Lin and Coleman, 1960). The Al content was measured with atomic absorption spectroscopy with flame atomizer after extraction with KCl (Lin and Coleman, 1960) using titrimetric method with back-titration.

All obtained data were compared and statistically analyzed using StatSoft CR STATISTICA 10. Firstly the presumptions for parametric testing were tested by Levene's test (homogeneity of variances) and Shapiro-Wilk test (normality). If homogeneity and normality

were confirmed, parametric testing was used, in this case analysis of variance, one-way ANOVA ($p < 0.05$) with later post-hoc testing with Scheffe's method. In cases without homogeneity of variances or normality, nonparametric testing was used with Kruskal-Wallis one-way analysis of variance.

4.2 Charcoal production and use in study area

As additional information about the possibilities of the biochar production and use in Pucallpa area, charcoal producers, local farmers and saw-mills were surveyed by in-depth interviews using semi-structured questionnaires. This experimental part took place in Pucallpa and two villages in the Pucallpa area, San Alejandro and Campo Verde districts between July 20th and September 18th 2012.

In Pucallpa, 9 charcoal producers, 17 farmers and 15 saw mills was found and questioned using the questionnaire (appendices 1- 3). In San Alejandro, one charcoal producer and five farmers responded the questionnaires. In Campoverde, we found one charcoal producer and four farmers (Table 12). All charcoal producers, farmers and saw-mills were selected randomly.

Table 14: Numbers of questionnaires respondents

	Charcoal producers	Farmers	Saw-mills
Pucallpa	9	17	15
San Alejandro	1	5	-
Campoverde	1	4	-
Total	11	26	15

Charcoal producers were interviewed about the amount of residues from charcoal production and their possible use, about integration of biochar into the soil and generally about the present state of knowledge of the possibility of biochar utilization in agriculture. Farmers were asked whether or not they are familiar with the potential of biochar in the agriculture and if they use it. The importance of saw-mills arises with the possible biochar production from wood shavings and other wood residues, as it is described in the pot experiment of this research.

5. RESULTS

5.1. Pot experiment

5.1.1. Soil nutrient contents

In the experiment with *G. crinita* pH was increased in all treatments compared to control (Table 15). However, manure containing treatments had slightly more alkaline reaction. Also C was significantly increased by all biochar applications. The K soil contents was increased by both manure and biochar treatments. In case of manure+biochar I.+NPK application the K content was increased almost 10 times. However, the increases in K contents were higher after manure additions in comparison with biochar applications. Ca content was increased by manure applications, biochar did not have any significant effect on Ca content. Also Mg was not affected by biochar amendments. All manure amendments significantly increased the Mg content, the highest effects had only manure application. P content was not affected by biochar addition. Manure increased the P soils content significantly in all manure containing treatments. Al was significantly reduced by all soil condition or fertilizer applications.

Table 15: Soil nutrient contents after six weeks of cultivation of *G. crinita* grown in peruvian Ultisol with amendments of biochar, chicken manure and inorganic fertilizer (NPK); values in one column followed by the same letter are not significantly different at $p < 0.05$; (n=6)

Treatment	pH (H ₂ O)	C (g kg ⁻¹)	K (g kg ⁻¹)	Ca (g kg ⁻¹)	Mg (g kg ⁻¹)	P (g kg ⁻¹)	Al (mg kg ⁻¹)	CEC (mmol kg ⁻¹)
U	4.9c	35.4c	1.1d	0.6c	0.1c	0.4d	190a	51.3d
B I.	5.9bc	107.8ab	3.3c	0.7c	0.1c	0.5d	30b	106.6c
M	6.6ab	57.0bc	6.2b	1.5a	0.7a	17.6b	30b	226.5b
MB	7.2a	109.1a	6.2b	0.9b	0.5ab	13.4c	30b	204.4b
MBF	6.4abc	108.7a	10.7a	1.9a	0.6ab	21.2a	20b	345.3a

While biochar I. slightly increased soil pH, biochar II. decreased it significantly (Table 16). Manure had the most evidential. effect and increased the soil pH to 7.2. C content was significantly increased by biochar II. addition (almost by 100%). However, the pH of all biochar I. containing treatments were significantly higher in comparison with biochar II. treatment. All manure applications increased the K content significantly. The K increases after biochar I. and biochar II. applications were significantly smaller. Ca was increased by both biochar types, by

manure+biochar I. application and by manure only and manure+biochar I.+NPK additions. Mg and P were significantly increased only by manure applications. Al reduced in all treatment. However, the effect of biochar II. was significantly smaller than effect of the rest of the treatments.

In case of rice pH was significantly increased by application of all fertilizers with the exception of biochar II, which did not affect the pH (Table 17). Both types of biochar increased the C content in all biochar-amended treatments. K and Ca were increased by both biochar types and even more by manure treatments. Mg and P contents were not affected applications of biochars, but were significantly increased by manure application.

Table 16: Soil nutrient contents after six weeks of cultivation of cowpea. grown in peruvian Ultisol with amendments of biochar, chicken manure and inorganic fertilizer (NPK); values in one column followed by the same letter are not significantly different at $p < 0.05$ (n=6)

Treatment	pH (H ₂ O)	C (g kg ⁻¹)	K (g kg ⁻¹)	Ca (g kg ⁻¹)	Mg (g kg ⁻¹)	P (mg kg ⁻¹)	Al (mg kg ⁻¹)	CEC (mmol kg ⁻¹)
U	5.8bc	46.2c	0.7d	1.2d	0.1d	0.7d	270a	60.7d
B I.	6.7b	109.1a	3.2c	3.5b	0.2d	0.9d	30c	176.8c
B II.	5.5b	88.0b	4.0c	3.8b	0.2d	1.5d	130b	210.1c
M	6.8b	46.2c	6.4b	4a	1.4a	18.9b	30c	323.4b
MB	7.2a	101.4a	5.7b	3.2c	0.9c	13.2c	30c	261.7b
MBF	6c	102.4a	12.8a	4.2a	1.2b	22.3a	30c	482.6a

Table 17: Soil nutrient contents after six weeks of cultivation of rice grown in peruvian Ultisol with amendments of biochar, chicken manure and inorganic fertilizer (NPK); values in one column followed by the same letter are not significantly different at $p < 0.05$ (n=6)

Treatment	pH (H ₂ O)	C (g kg ⁻¹)	K (g kg ⁻¹)	Ca (g kg ⁻¹)	Mg (g kg ⁻¹)	P (mg kg ⁻¹)	Al (mg kg ⁻¹)	CEC (mmol kg ⁻¹)
U	5.1b	48.0b	0.4d	0.9c	0.2c	0.8c	280a	50.4e
B I.	6.5a	99.0a	3.4c	3b	0.2c	1.0c	50c	173.1d
B II.	5.4b	95.0a	4.0c	2.9b	0.3c	1.4c	130b	189.7d
M	6.6a	55.0b	6.5b	4.1a	2.3a	18.2a	40c	364.5b
MB	7.3a	98.0a	6.8b	3.2b	1.3b	13.1b	30c	308.3c
MBF	6.0ab	101.0a	10.8a	4.4a	1.5b	18.8a	30c	448.7a

All treatments in all three plants cultivation significantly increased the CEC. However, manure applications increased it more than the biochar amendments. The highest CEC has all treatments amended with manure+biochar I.+NPK fertilizer.

5.1.2. Biomass production and plant growth

Above- and belowground biomass, shoot and root lengths of all three species were significantly influenced by the addition of biochar, manure and mineral fertilizer to the Ultisol. In the case of tree species *G.crinita* aboveground biomass production was not influenced by any treatments. Belowground biomass production was significantly improved by biochar I. amendment. The growth of shoots was significantly improved by all amendments. Root length was also significantly (by 500%) improved only by biochar I. additions (Figure 11).

Table 18: Aboveground and belowground biomass, shoot length differences (shoot length Δ) and root length differences (root length Δ) of *G.crinita* grown in substrate with biochar, manure and fertilizer; values followed by same letter are not statistically different at $p<0.05$; $n=6$

Treatment	Aboveground biomass (g pot ⁻¹)*	Belowground biomass (g pot ⁻¹)*	Shoot length Δ (cm)*	Root length Δ (cm)*
U	0.179 \pm 0.049a	0.058 \pm 0.031b	1.667 \pm 0.946b	1.833 \pm 0.753b
B I.	0.198 \pm 0.051a	0.128 \pm 0.052a	2.750 \pm 0.989ab	11.117 \pm 3.307a
M	0.318 \pm 0.162a	0.075 \pm 0.029ab	6.233 \pm 1.675a	2.250 \pm 1.578b
MB	0.193 \pm 0.071a	0.039 \pm 0.030b	6.433 \pm 3.227a	4.650 \pm 2.016b
MBF	0.150 \pm 0.073a	0.021 \pm 0.011b	8.033 \pm 4.817a	1.567 \pm 1.721b

* – mean value \pm standard deviation; Δ – the differences between the lengths (heights) after six weeks of experiment and the lengths (heights) before planting (at the 40 days age of seedlings)

The addition of only biochar I. did not influence the aboveground biomass production, but shoot lengths were increased by 135%. Surprisingly, in case of biochar I.+manure+NPK fertilizer, the increase of shoot heights was 588% (Figure 11).

Aboveground biomass production of cowpea was significantly ($p<0.05$) improved by all amendments (Table 19). However, biochar addition had better influence than manure amendments. No treatment had significant effect on belowground biomass production of cowpea. Shoot heights were increased by all amendments. However, the best results were obtained after biochar I.+manure amendment, the second highest increments after biochar I. and biochar II.

addition. Root lengths were increased by both types of biochar, to a lesser extent also by manure application (Figure 12).

Table 19: Above- and belowground biomass, shoot and root length of cowpea grown in substrate with biochar, manure and fertilizer; values followed by same letter are not significantly different at $p < 0.05$; $n=6$

Treatment	Aboveground biomass (g pot ⁻¹)*		Belowground biomass (g pot ⁻¹)*		Shoot length (cm)*		Root length (cm)*	
U	0.519	±0.126b	0.134	±0.034a	17.361	±2.750d	16.500	±2.126b
B I.	1.449	±0.343a	0.186	±0.066a	33.556	±5.336b	19.778	±2.177a
B II.	1.396	±0.158a	0.182	±0.055a	33.278	±1.831b	21.445	±0.861a
M	1.026	±0.316ab	0.159	±0.069a	20.889	±3.538c	13.000	±1.211c
MB	1.632	±0.274a	0.145	±0.064a	42.833	±5.940a	18.389	±0.491b
MBF	0.942	±0.401ab	0.127	±0.050a	28.389	±3.941bc	10.667	±0.843c

* – mean value ± standard deviation

In case of rice, neither aboveground nor belowground biomass production was influenced by any treatment (Table 20). All amendments decreased the shoot heights significantly. The largest decrease was in case of biochar I.+manure+NPK application, the second largest in case of both biochar types. On the other hand, root lengths were increased by biochar I. application (by 39%) and by biochar I. manure application (by 24%) (Figure 13).

Table 20: Above- and belowground biomass, shoot and root length of rice grown in substrate with biochar, manure and fertilizer; values followed by same letter are not significantly different at $p < 0.05$; $n=6$

Treatment	Aboveground biomass (g pot ⁻¹)*		Belowground biomass (g pot ⁻¹)*		Shoot length (cm)*		Root length (cm)*	
U	0.299	±0.071a	0.061	±0.028a	16.886	±0.332a	1.450	±0.105cd
B I.	0.177	±0.033a	0.055	±0.019a	14.198	±0.455c	2.017	±0.117a
B II.	0.218	±0.073a	0.037	±0.027a	14.209	±0.431c	1.600	±0.063c
M	0.258	±0.128a	0.045	±0.028a	15.938	±0.158b	1.367	±0.052d
MB	0.256	±0.175a	0.044	±0.024a	16.052	±0.174b	1.800	±0.000b
MBF	0.125	±0.060a	0.020	±0.009a	11.896	±0.495d	1.083	±0.117e

* – mean value ± standard deviation

Generally, root lengths were significantly improved by biochar amendments in all three cases. However, the effect of biochar on plant growth and biomass production was not as high as it was expected.

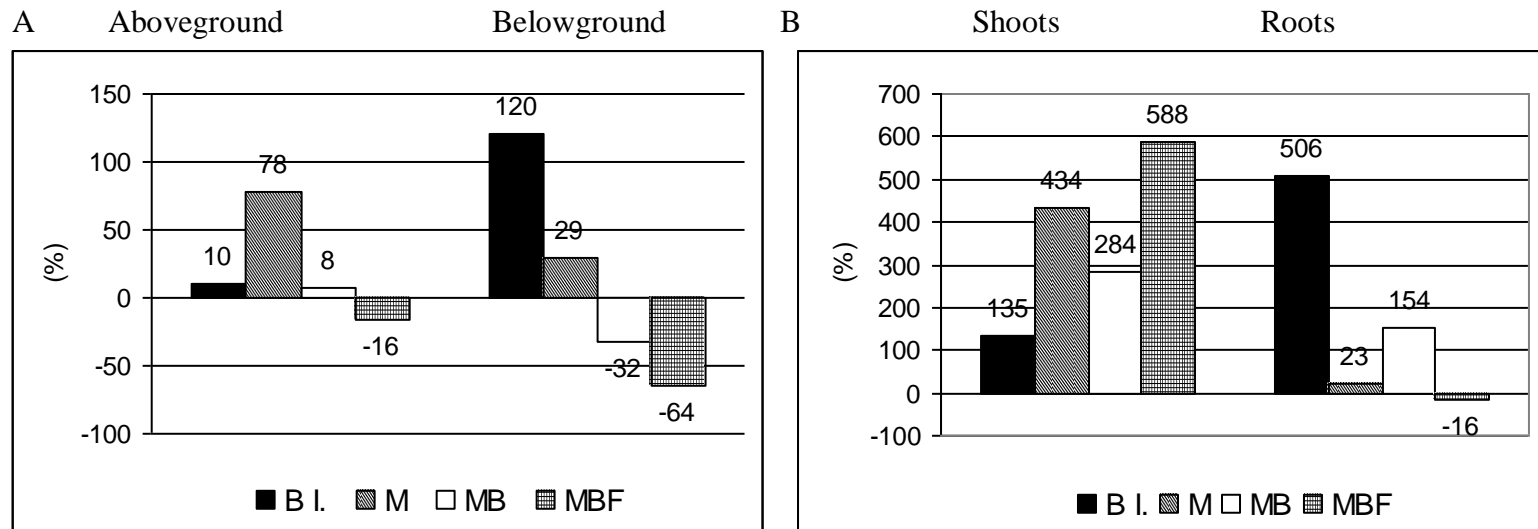


Figure 11: Relative above- and belowground biomass (A) and shoot and root lengths (B) of *G. crinita* compared with control.

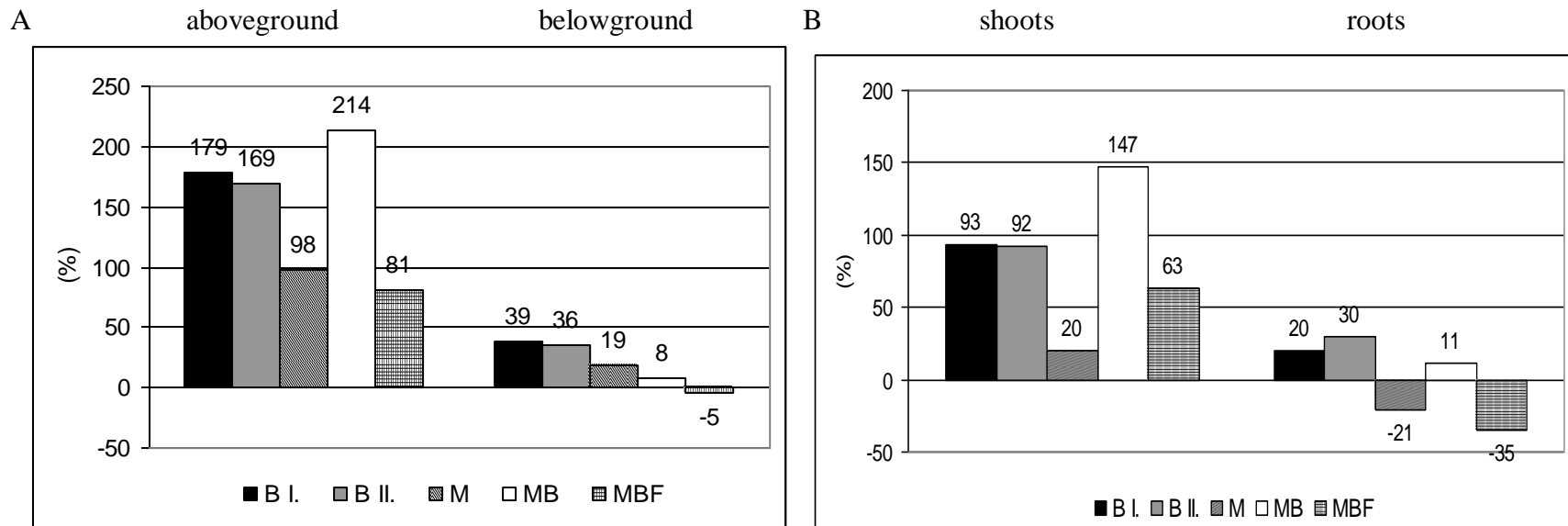


Figure 12: Relative above- and belowground biomass (A) and shoot and root lengths (B) of cowpea compared with control

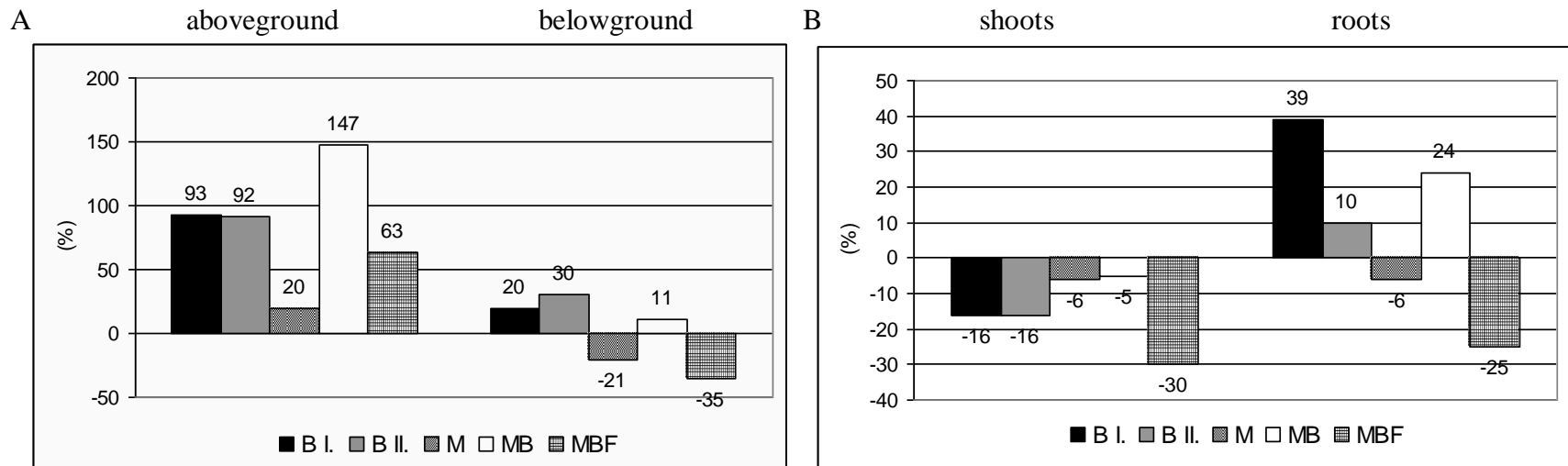


Figure 13: Relative above- and belowground biomass (A) and shoot and root lengths (B) of rice compared with control

5.1.3. Foliar nutrient contents

N content of *G.crinita* was significantly increased by manure application (Table 21). The foliar content of P was decreased by biochar I. amendment. Biochar did not influence the K content. However, it was significantly increased by manure applications. Ca was not affected by any treatment. Mg, similarly to K, was also increased only by manure applications. Zn, Fe, Cu and Mn did not show any significantly change after application of any amendment.

Table 21: Foliar nutrient contents of *G. crinita* grown in plastic bags with two kg of peruvian Ultisol with amendment of biochar, chicken manure and inorganic fertilizer (NPK); values in one column followed by the same letter are not significantly different at $p < 0.05$ ($n=6$); n.d. – not determined.

Treatment	N (g kg ⁻¹)	P (g kg ⁻¹)	K (g kg ⁻¹)	Ca (g kg ⁻¹)	Mg (g kg ⁻¹)	Zn (mg kg ⁻¹)	Fe (mg kg ⁻¹)	Cu (mg kg ⁻¹)	Mn (mg kg ⁻¹)
U	18.7bc	1.1ab	12.0a	9.0a	4.8a	13.5a	311.5a	2.5a	53.0a
B I.	13.5c	0.8b	14.5a	7.8a	4.9a	17.5a	245.5a	2.5a	56.5a
M	29.6a	1.5ab	14.7b	11.5a	9.0b	16.5a	264.5a	2.5a	58.5a
MB	25.4b	1.7a	18.6b	9.8a	10.5b	20.5a	256.0a	2.6a	55.0a
MBF	n.d.	1.4ab	20.1b	12.5a	9.1b	17.0a	268.0a	2.5a	57.0a

Foliar N contents of cowpea were significantly decreased by biochar I. addition and increased by manure application (Table 22). P was significantly increased by manure application and decreased by biochar II. amendment. K content was increased by biochar II. application, biochar I+manure and biochar I.+manure+NPK application. All manure treatments increased Ca and Mg foliar contents. Zn content was significantly reduced by all manure treatments and even more by both biochar types. Only biochar II. and manure had effect on Fe uptake and decreased the Fe foliar content. Cu content was reduced by all amendment, except manure application. However, the largest decrease was obtained after biochar II. addition. Mn foliar content was reduced mainly by biochar II. and biochar I.+manure additions.

Table 22: Foliar nutrient contents of cowpea grown in Peruvian Ultisol with amendments of biochar, chicken manure and inorganic fertilizer (NPK); values in one column followed by the same letter are not significantly different at $p < 0.05$ ($n=6$); n.d. – not determined.

Treatment	N (g kg ⁻¹)	P (g kg ⁻¹)	K (g kg ⁻¹)	Ca (g kg ⁻¹)	Mg (g kg ⁻¹)	Zn (mg kg ⁻¹)	Fe (mg kg ⁻¹)	Cu (mg kg ⁻¹)	Mn (mg kg ⁻¹)
U	33ab	1.3b	16.6b	8.5b	4.9ab	45.2a	294.5a	2.6a	82.5a
B I.	27b	1.2b	25.1ab	7.4b	3.4b	20c	265.3a	2.3b	65.8ab
B II	28ab	0.5c	29.9a	8.8b	2.1c	21.8c	117.2b	2.0c	62.5b
M	36a	1.8a	24.6ab	12a	13.1a	26.5b	214.3ab	2.5ab	65.0ab
MB	28ab	1.6ab	28.7a	10.7a	6.4ab	25.5b	273a	2.4b	56.2b
MBF	n.d.	2.0a	38.2a	11.6a	9a	28.3b	268.6a	2.3b	72.2a

In foliar N contents of rice, there were differences only between biochar I. amendment and manure application (Table 22). P foliar content was not significantly affected by any treatment used in this research. Ca content was significantly increased by all amendments with manure, similarly to Mg with the exception of biochar I.+manure treatment, which did not influence Mg foliar content of rice. Zn was reduced by both biochar types, mainly by biochar I. applications and Fe by all amendments. No treatments significantly influenced Cu foliar content of rice. Mn was increased by biochar addition and decreased by manure and biochar I.+manure application.

Table 23: Foliar nutrient contents of rice grown in Peruvian Ultisol with amendments of biochar, chicken manure and inorganic fertilizer (NPK); values in one column followed by the same letter are not significantly different at $p < 0.05$ ($n=6$); n.d. – not determined.

Treatment	N (g kg ⁻¹)	P (g kg ⁻¹)	K (g kg ⁻¹)	Ca (g kg ⁻¹)	Mg (g kg ⁻¹)	Zn (mg kg ⁻¹)	Fe (mg kg ⁻¹)	Cu (mg kg ⁻¹)	Mn (mg kg ⁻¹)
U	22.2ab	0.7a	0.4d	8.3b	3.5b	39.5a	287.5a	2.3a	111.5bc
B I.	17.6b	0.6a	3.4c	7.5b	2.2b	18.5b	220.0b	2.2a	186.5a
B II.	21.4ab	0.6a	3.4c	9.0b	2.4b	28.0ab	117.5c	2.0a	168.5ab
M	28.0a	1.1a	6.5b	12.3a	7.9a	45.0a	222.0b	2.7a	99.5c
MB	24.3ab	1.0a	6.8b	11.5a	4.2b	32.0ab	122.5c	2.1a	64.5c
MBF	n.d.	1.5a	10.8a	11.8a	6.1a	40.0a	117.5c	2.3a	142.5ab

5.2. Charcoal production and use in study area

All 11 charcoal producers in Pucallpa area use *Dipteryx micrantha* Harms or *Dipteryx alata* Vogel for charcoal production. Both species of *Dipteryx* genera, in Peru commonly known as shihuahuaco and internationally traded as “cumarú” or Brazilian teak, have for the past years been a target of an extractive boom. Shihuahuaco is valued for its high wood density and resistance to rot, making it ideal for outdoor applications such as decking and patio furniture for which it is used in North America and Europe (Putzel *et al.*, 2011). Local woodsmen recognized and distinguish two types of shihuahuaco – *shihuhuaco rojo* and *shihuahuaco amarillo*, which roughly correspond with the two species. Second most common species is *Calycophyllum spruceanum* (Benth.) Hook.f. ex K.Schum. (in 6 cases), one of the indigenous trees suitable for agroforestry in Peruvian Amazon (Weber *et al.*, 2001) which is valued by farmers mainly for its rapid growth, high wood quality for construction and high calorific value for firewood and charcoal (Sotelo Montes *et al.*, 2003, as cited in Boivin-Chabot *et al.*, 2004). One producer uses also *Copaifera* ssp.. All of them buy the wood and are not timber producers themselves.

During charcoal production, the access of air needs to be limited to prevent burning in favor of pyrolysis. This is generally done by covering the charcoal kiln. There were differences between the charcoal production in Pucallpa, San Alejandro and Campoverde. While in Pucallpa, as one of the most important timber producers in Peru, all charcoal producers use wood shaving, in Campoverde and San Alejandro almost exclusively soil is used (Appendix 4).

Nine of 11 charcoal producers have heard about the possibility of use of biochar in agriculture, only two of them donate or sell the carbon-rich charcoal residues (Table 24). However, all of them would donate or sell them for cheap if the farmers came. In Pucallpa, one producer donates it as a disinfectant and one as a medicine in poultry production. In Campoverde all charcoal residues are used for vegetable production. However, the amounts of the used charcoal residues are so small, that it can be negligible. In San Alejandro there was found no utilization of the residues of charcoal.

Table 24: Farmers' responses about the use of biochar in agriculture

	Pucallpa	San Alejandro	Campoverde	Total
Know potential of biochar* in agriculture	15 (88%)	4 (80%)	4 (100%)	19 (88%)
Know the advantages	8 (47%)	3(60%)	3 (75%)	14 (54%)
Use biochar in agriculture	2 (12%)	0 (0%)	1 (25%)	3 (12%)

* term biochar as an alternative name for charcoal residues was explained prior to questionnaires

It is obvious that the problem of biochar not being used in agriculture is not the lack of knowledge, but mostly the insufficient access to biochar (Table 25). These responses do not correlate with the responses of charcoal producers, who would all provide biochar to farmer if there was a demand from the side of farmers.

Table 25: The reasons why the farmers do not use biochar in agriculture

Reason	number of positive answers
No biochar source	13 (50%)
Use of other organic fertilizers	7 (27%)
Other reason	1 (4%)

The utilization of charcoal residues would cause additional costs for farmer which would need to be paid for the transport and subsequent incorporation of biochar into the soil. Although the potential of biochar on plant growth is generally at least partly known, the real benefits, regarding the increased quality and quantity of produce, are not certain to date. Thus, the final cash benefits of biochar utilization are not widely accepted.

5. DISCUSSION

5.1. Soil and nutrient contents

Soil carbon content was, how it was expected, increased by all biochar amendments. Same results were obtained in most experiments for example by Morales *et al.* (1995), Glaser (1992) or Glaser *et al.* (1992), all done in Brazil. However, the final C content in the soils depends strongly on the biochar type, as well as the content of all nutrients and nutrient uptake by the plants. Nevertheless, only C addition usually does not improve the nutrient uptake of the plants due to the wide C:N ratio which should be compensated by N addition. Similarly, in research of Chan *et al.*(2007), the yields of radish (*Raphanus sativus* var. Long Scarlet) were not increased even by the higher biochar applications (100 t/ha) in absence of N fertilizer.

CEC was significantly increased by all amendments. Nevertheless, the addition of biochar had significantly lower effect than the combination of biochar with manure. The treatment with biochar I.+manure+NPK showed the largest increase of all used amendments. However, biochar used on Norfolk soil by Novak *et al.*(2009) did not significantly improve the CEC. Therefore, biochar nutrient contents, CEC and pH analysis are essential prior the application of biochar in to the soil, because also negative impacts of biochar have been observed (Gundale and DeLuca, 2007).

Biochar application to the soil also significantly increases the soil pH and, thus, decreases the Al content. Also Lehmann *et al.* (2003b), Chan *et al.* (2007) and Novak *et al.* (2009) assessed decreased Al content by biochar application. However, there was significant difference between two biochars. The Al content reduction was significantly higher after biochar I. application in comparison with biochar II.. In the first case, the total Al content was reduced by 89%, in case of biochar II. by only 50% in both cowpea and rice cultivation. These differences are probably, as it has been mentioned above, caused by different composition of biochars. However, Al contents still remained relatively high even after biochar applications. P contents were rather low in all cases suggesting that the limiting factor of plant growth could be P unavailability caused by low pH and high Al content leading to immobilization of P. Interestingly, the growth was not improved even by P-rich chicken manure application with biochar.

5.2. *Plant growth and biomass production*

Despite the positive effect of biochar on soil organic matter, soil nutrients and pH, this did not influence the growth of plants or biomass production of plants in an expected extent. Possibly better results could be obtained in long-term experiments. Better root growth and higher belowground biomass production indicate, that after longer cultivation time this could be reflected in aboveground biomass production, shoot growth and foliar nutrient contents as result of larger and rapidly developed root system. We concluded that cultivation time was probably the most limiting factor in our experiment.

In this experiment, cowpea showed increased aboveground biomass production after amendments of both biochar types and biochar I.+manure application. This is most probably caused by the tolerance of legumes to higher C:N ratio as a result of symbiotic N fixation ability and the potential of balancing C:N ratio.

Aboveground biomass production of *G.crinita* and rice was not influenced by any soil amendment. However, biochar influenced and improved the growth of cowpea. In the similar experiment of Lehmann *et al.* (2003b), the aboveground biomass production was significantly increased by charcoal addition in comparison with control and all other amendment when planted on Ferralsol for the same time of six weeks. However, the belowground biomass was decreased. Oguntunde *et al.* (2004) observed increased biomass production by 44% on biochar amended soil compared to adjacent soil without biochar. Also in Indonesia the yields of maize and peanuts were significantly increased by bark charcoal application (Yamamoto *et al.*, 2006). However, different effects of biochar amendments can be caused by different plant requirements and different biochar production procedure resulting in distinct biochar types. As it is suggested by the experiment of Gundale and DeLuca (2007) with *Koeleria macrantha* (Ledeb.)Schult whose growth was diminished by two charcoal types created from ponderosa pine and douglasfir bark.

Shoot heights were increased by biochar application in case of *G.crinita* and cowpea. The shoot heights of rice were decreased by all types biochar and similarly by all fertilizers applications. Nevertheless, biochar I. application significantly increased the lengths of roots of *G.crinita*, cowpea and rice by 500, 20 and 39%. While in case of the experiment of Lehmann *et al.* (2003b) the highest influence in comparison with control had the treatment with biochar+manure+fertilizer application, in our experiment the treatment with the amendment of biochar I.+manure+fertilizer showed in most cases no effect or even diminishing effect. Rather

negative effect of all treatments on rice growth compared to control could possibly be explained wrong variety selection. Seeds of traditional rice variety were bought on local market. It is possible that this variety was not suitable for this experimental design and was not thriving well when planted in plastic bag in nursery.

Despite of relatively close relationship of *G.crinita* with some of the important cultivated cash crops, such as cacao (*Theobroma cacao* L.) or cotton (*Gossypium* ssp.) there was found no investigation of biochar used in cultivation of plants from this family. Most experiments focus of herbaceous plants, mostly maize (Mbagwu and Picolo, 1997; Yamamoto *et al.*, 2006), rice (Lehmann *et al.*, 2003b) and legumes (Rondon *et al.*, 2004). Chidumayo (1994) reported increased germination, shoot heights and biomass production of seven Zambian indigenous woody plants.

There is no doubt that the origin of biochar and its production procedure has enormous effect on the final results, as it was assessed by many investigations (Deal *et al.*, 2012; Yuan and Xu, 2012; Yuan *et al.*, 2012). The gasifier produced biochar showed better effect on the yield and growth of maize than biochar produced in traditional way using kilns (Deal *et al.*, 2012). During this research, two types of biochar were used, one produced traditionally during the production of commercial charcoal and one using simple kiln (appendix 4). Thus, none of the types was created by modern specialized gasifiers. Nevertheless, the final results on soil and plants were distinct. While in case of cowpea, there was no difference between both biochar types, in case of rice, the root lengths were significantly higher after biochar I. application (biochar from charcoal production) in comparison with biochar II. (biochar created from wood shavings in a specialized simple kiln).

5.3. Foliar nutrient contents

Biochar application significantly decreased the foliar N content of all investigated plants. Similar results were obtained by Lehmann *et al.* (2003b), where total N content as well as N uptake of rice and cowpea were significantly decreased on Brazilian Ferralsol in comparison with ADE. The diminishing effect of biochar additions on N foliar content is, similarly as in the soil (Lehmann *et al.*, 2003b), probably the result of N immobilization by microorganisms. Dempster *et al.* (2011) decreased soil microbial biomass and N mineralization with Eucalyptus (*Eucalyptus marginata* Donn. Ex Sm.) biochar addition to a coarse soil. During the short-term experiments biochar applications can cause N immobilization which can temporarily reduce the plant

available NO₃-N concentrations (Novak *et al.*, 2010). This probably caused the differences between N foliar contents of biochar amended soils and soils without biochar addition.

K, Ca and Mg and micronutrients of *G.crinita* were not affected by biochar amendments, but were significantly increased by manure applications, as we expected. In experiment with cowpea K foliar content was significantly increased by both biochar and manure additions. In the experiment of Lehmann *et al.* (2003b) with cowpea grown in pots with Ferralsol K content was increased only by biochar amendment and addition of only manure did not have any effect on K foliar content. The difference could be caused by different K content of used biochars. Biochar II. in this experiment content relatively high K content, 4.3 g kg⁻¹, respectively. Increase K content in treatments in biochar in both cases can be results of improved CEC preventing leachage of K. Ca and Mg contents were relatively high in applied chicken manure, which explains the increased Ca and Mg content in plastic bags with manure amendments. It also indicates that these nutrients could be limiting in only biochar amended soils. Trakal *et al.*(2011) evaluated metal sorption behavior after biochar application into a metal-contaminated soil, where Zn desorption was reduced after biochar application. The applied amount of biochar in the experiment was not sufficient for metal immobilization in such contaminated soil. Nevertheless, the decreased Zn foliar contents of cowpea and rice confirm the hypothesis of the capability of biochar to influence the sorption/desorption of heavy metals.

We concluded that improved root systems of plants by biochar amendments could lead to increased foliar nutrient contents in long-term experiment, as result of improved nutrient uptake capacity.

6. CONCLUSION

The hypothesis about the soil fertility improvement by biochar amendments was confirmed as soil nutrient contents, organic matter and pH were increased. However, the effect of improved soil properties was generally not related to improved biomass production and plant growth. Although the hypothesis was not disproved, effect of biochar on biomass production, plant growth and foliar nutrient contents was lower than was expected. This could be caused by insufficient cultivation time and most probably by large C:N ratio in the soil after biochar addition, that makes N a limiting nutrient. This limitation was overcome only by growth of legume crops as cowpea that can fix atmospheric N. Other crops like rice and *G.crinita* do not have this advantage and N-immobilization could limit their growth in the soil after biochar addition. From the questionnaires we concluded that carbon rich residues from charcoal production are generally not used in agriculture in Peruvian Amazon despite of being widely available for the farmers. Further research needs to be done especially on distinct biochar types and production procedures. Long-term study fields can recently give more information about the possibilities of biochar utilization. If research confirms positive effect of this biochar type on plant growth, this material could be of high potential in improving local agricultural systems. Great prospects of biochar are becoming more clear and obvious, especially in context of unfertile soils of humid tropics or soils of arid regions. There is no doubt that the tropical areas have a great production potential and, therefore, their melioration and improved management could not only improve the livelihood of the local communities, but also significantly contribute to food security in the future. Moreover, the improvement of the soils of arid region with the increased water retention capacity could undoubtedly diminish desertification and substantially help in fight against hunger in the poorest regions of the world. Nevertheless, the wide acceptance of biochar utilization does not depend only on its effect on the soil and environment, but also on the economic feasibility. Furthermore, the stability of C of particular biochar types in the soils and long-term influences on the soil and plant relationship also need to be evaluated. Even though most experiments show the positive effect of biochar, in this study this was not confirmed. It can be concluded that insufficiency of knowledge about biochar properties or incomplete information could not only have no effect on soils and plants, but actually have even negative effect.

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5 APPENDICES

8.1. Appendix 1

Questionnaire for charcoal producers

Nombre:	Edad:
Pueblo:	Fecha:

1. ¿De cuáles especies produce Ud. el carbón?
2. ¿Ud. compra la madera para la producción del carbón o usa material de su producción?
3. ¿Que material usa Ud. para cubrir las carboneras?
 - a. Suelo
 - b. Aserrín
 - c. Otro :
4. ¿Ha oído Ud. de las posibilidades del aprovechamiento del carbón vegetal en agricultura o forestería?
5. ¿Ud. usa los residuos de la producción del carbón (polvo o trozos pequeños) o los regala a alguien que lo hace?
 - a. No
 - b. Sí, para la producción de verduras
 - c. Sí, para la propagación vegetal
 - d. Sí, para(complete la frase)

8.2. Appendix 2

Questionnaire for farmers

Nombre:	Edad:
Pueblo:	Fecha:

1. ¿Ha oído Ud. de las posibilidades del aprovechamiento del carbón vegetal en agricultura o forestería?
2. ¿Conoce Ud. las ventajas de la utilización del carbon en agricultura?
 - a. Sí, conozco las ventajas
 - b. Sé que hay ventajas, pero no las conozco
 - c. No, no las conozco
3. ¿Ha probado Ud. la utilización del carbon vegetal en agricultura o forestería?
 - a. Sí, frecuentemente
 - b. Sí, una vez sin resultados
 - c. No
 - d. Otra respuesta
4. Si Ud. ha respondido a. a la pregunta numero 3: ¿De dónde tiene Ud. el carbón?
 - a. De mi producción
 - b. Comprado de carbonera
 - c. Regalado
5. Si Ud. ha respondido c. a la pregunta numero 3: ¿Cuáles son los razones?
 - a. No conozco las ventajas
 - b. No tengo ingreso del carbón para la agricultura
 - c. No tengo tiempo
 - d. Otro:

8.3. *Appendix 3*

Questionnaire for saw-mills

Nombre:	Edad:
Pueblo:	Fecha:

1. ¿Que hacen Uds. con los residuos de la madera (aserrín, trozos pequeños etc.)?
 - a. Se vende o regala a los carboneros
 - b. Se vende o regala para la utilización en agrikultura
 - c. No se usa para nada

2. ¿Ha oído Ud. de las posibilidades del aprovechamiento del carbón vegetal en agricultura o forestería?

8.4. Appendix 4

Photos



Photo 1: Biochar production system used at IIAP (Instituto de Investigaciones de la Amazonia Peruana) in Pucallpa for biochar production from rice husks. In this investigation it was used for biochar production from wood shaving. At IIAP charred rice husk is used mostly for vegetative propagation



Photo 2: Biochar made from wood shaving at IIAP, Pucallpa



Photo 3: Charcoal kiln covered with soil, San Alejandro, Peru



Photo 4: Charcoal kiln covered with new wood shaving and/or wood shaving recycled from the previous caron production, Pucallpa, Peru



Photo 5: Air-dried, homogenized and sieved soil samples prepared for the analysis



Photo 6: Different biomass production of cowpea plants with different amendments

(1- Control; 2- biochar; 3- chicken manure; 4- biochar+chicken manure; 5- biochar+chicken manure+NPK fertilizer).
