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# Amending acidic soil with bamboo (*Bambusa blumeana*) biochar: effect on mung bean (*Vigna radiata*) growth rate and yield

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**Abstract**. Biochar is a carbon rich solid residue produced when biomass is heated with little or no available oxygen at relatively low temperature. Its use in agriculture as soil amendment had been found to enhance soil quality and increase crop productivity. Acidic soil amended with biochar produced from waste basal portions of bamboo was tested by growing mung bean in a randomized complete block design experiment. Results showed that plants grown in soil amended with 8.5 and 15.75% biochar started flowering, pod filling and maturing 6 to 7 days earlier than those grown in un-amended soil. After 60 days, plants in the biochar amended soil were significantly taller by about 27% and their pod production was 102% higher than those without biochar. Addition of vermicast to biochar-amended soils did not significantly enhance the flowering and pod filling stages of mungbean and likewise did not significantly increase pod production rate. Root nodules of plants grown in soils amended with 8.5 and 15.75% biochar. The addition of vermicast resulted in fewer nodules but in effect enhanced rootlet formation. There was a continually increasing trend in pH during the vegetative growth of the plant up to maturity both in the control pots and those amended with biochar. The results suggest that bamboo biochar can be an efficient and cost effective way of improving soil quality and aid in sustainable crop production in regions with acidic soils.

Key Words: carbonization, biochar, mung bean, vermicast, root nodulation.

**Introduction**. Biochar is a carbon (C) rich product from the thermal decomposition of biomass under limited supply of oxygen at relatively low temperature (< 700°C). Rather than being burned and oxidized to gaseous carbon dioxide, as would happen in combustion or incineration in an oxygen-rich environment, part of the carbon is retained with the solid residue. Recently, applications have recently gained global interest in agricultural practices and environmental management (Lehmann & Joseph 2009). In addition to bioenergy production and carbon sequestration, Laird (2008) has reported that biochar can be used as a soil amendment material to improve soil quality and increase crop productivity. It was emphasized that while charcoal can be used to generate energy, its application to soil may be a key to sustainability. Day et al (2004) suggested that the use of biochar to sequester carbon in agricultural lands as a means to combat climate change can only be accomplished economically if the sequestered C has beneficial soil amendment and/or fertilizer values.

Although several studies have shown that the use of biochar as soil amendment material has benefited crop growth and yield, its full agronomic value is not yet well understood (Iswaran et al 1980; Glaser et al 2001). Mclaughlin et al (2009) stated that not all biochars are created equal. A variety of biomass feedstock can be used in the production of biochar under different conditions resulting in products of varying properties. Among the feedstocks used in several studies include rice hull, bagasse, cornstalks or straw, coconut shell, peanut and pili hulls, greenwastes, wastewater sludge and animal manure (Chan et al 2007; Gaskin et al 2010; Hossain et al 2010).

Bamboo is one natural resource that has attracted great attention from researchers due to its unique biological structure. It is characterized by high biomass yield, short rotation period and high ecological benefits (Zhang & Flanagan 2007). It is one of the fastest growing plants and is abundantly available in many countries. Its vast distribution, availability and desirable properties provide the local community with many choices of utilization for a wide range of applications.

In the Philippines, a total of 62 bamboo species is found in 39,000 to 53,000 ha of forest lands, government plantation, privately owned plantations, and "natural stands" (Virtucio & Roxas 2003). Iloilo Province has an abundant supply of bamboo pole, mostly *kawayan tinik* (*Bambusa bluemana*) which has an aggregate area of 8,095 ha. The culms of this species can be harvested in three years. However, because of lack of proper management, huge quantities of bamboos are wasted every year. Often times, during harvesting and processing, only the middle portion of the poles are used while the basal and top portions and the branches are abandoned and are left to decay in the growing areas (Virtucio & Roxas 2003).

The conversion of bamboo wastes to charcoal is one of the potential choices for its productive utilization. Combusting charcoal from different biomass in order to process heat is common while a few chars may also undergo further activation to be used in higher value adsorption applications (Fan et al 2004; Boateng 2007; Brewer et al 2009). Another option is to apply the char to soil as a soil amendment and/or a carbon sequestration agent (Laird 2008; Masulili et al 2010).

This paper investigates the agronomic benefits of bamboo biochar as soil amendment material using mung bean [*Vigna radiata* (L.) Wilczek] as plant indicator. Mung bean was chosen because it is a short duration legume which fits well to many cropping systems and is often grown in farms after rice/corn harvest. For small-scale farmers and government agencies tasked to provide technical assistance, low-input sustainable agriculture is often found a problem. Soil acidity that may result from continuous and excessive use of inorganic fertilizers could likewise restrict agricultural production. This paper specifically aims to provide information on the effects of amending acidic soil with bamboo biochar on plant growth and yield as well as its effect on soil pH and plant root development. Such information are essential in assessing the potential of using bamboo biochar as soil conditioning material in acidic soils.

#### Material and Method

*Study location*. The pot experiment in this study was conducted in Iloilo City, Philippines during the dry season from April 1, 2011 to May 30, 2011. The location map is shown in Figure 1.

**Biochar preparation**. Biochar was produced at the Science and Technology Bamboobased Farm in Maasin, Iloilo using a fabricated 0.2 m<sup>3</sup> drum carbonizer based on the Central Philippine University-Affiliated Renewable Energy Center (CPU-AREC) kiln design but with additional provisions of a chimney handle and support stand rollers to allow equipment mobility. The feedstock materials were the unutilized basal portions of the *B. blumeana* poles left after harvest. All the waste materials were obtained from a bamboo farm at Brgy. Inabasan, Maasin, Iloilo from mature culms aging more than 3 years. Before the carbonization process, the prepared feedstock materials were cut into small pieces (average length of 10.16 cm and width of 5 cm) and air dried until moisture content was reduced to about 10-15%. Carbonization time was 2.5 hrs at terminal temperature of 628°C. The produced biochar was washed with tap water, dried to 10 to 15% moisture ground and sieved through a 3.175 mm mesh screen. These are used in the preparation of the biochar amended soils for pot experiments. Mungbean was obtained from a local agricultural products store in Iloilo City. **Soil and vermicast preparation**. Four soil samples were obtained from Miag-ao Iloilo and San Jose, Antique and analyzed for pH and nutrient content. The soil with the lowest pH (Brgy Atabay, San Jose Antique) was used as soil material in the experiments. The soil analysis for pH and nutrient content was performed at the Soil's Laboratory of the Department of Agriculture, Region 6, Iloilo. The vermicast used as organic fertilizer in the positive control was obtained from the University of the Philippines Visayas Reforestation and Vermicompost Site. It was air-dried and analyzed at the Soil Laboratory, Department of Agriculture, Region 6, Iloilo for pH and nutrient content.



Figure 1. Map showing the location where the study was conducted. Iloilo City is found in Panay Island, one of the cities in Western Visayas, Philippines (Source: www.googlemaps.com).

**Experimental layout**. The nursery experiment was conducted in a 12 m by 10 m fenced vegetable garden at the Villa Carolina Village, Arevalo, Iloilo City, Philippines from April 1, 2011 to May 30, 2011 when temperature was about  $33\pm2.0^{\circ}$ C and relative humidity about  $49\pm2.0\%$ . A randomized complete block design (RCBD) with four replications (blocks) of six treatments were applied. Since there were shaded areas in the garden, the blocks were arranged as shown in Figure 2 in order to have as much as possible uniform light exposure of the plants to sunlight during the day.



Figure 2. Experimental lay-out showing four blocks, each block containing six soil treatments that are arranged randomly.

Figure 3 shows the pot lay out in block 4 of the experimental set up. Each treatment was represented by three pots planted with five mung bean seeds by direct seeding which were later thinned to three seedlings per pot based on vigor and uniformity.



Figure 3. A block showing six treatments arranged randomly and uses soil, biochar-amended soil, soil with vermicompost and biochar amended soil with vermicompost.

The four blocks accommodated a total of 72 pots and 216 mung bean seedlings grown with the following treatments: A = soil only, B = soil+biochar (8.5%), C = soil + biochar (15.75%), D = soil + vermicompost, E = soil + vermicompost + biochar (7.5%), F = soil + vermicompost + biochar (13.89%). All the soil used in this experiment were taken from the pots where mung bean plants were grown in a previously conducted greenhouse experiment, allowing an additional  $1\frac{1}{2}$  months for biochar to age in the soil before planting. The pots were likewise placed on shallow plates arranged on the plastic mat. Throughout the duration of the experiment, the pots were watered as and when required with the same volume of water to prevent drying out of the top soil surface, while uniformly doing agronomic practices in each block like removal of weeds from each pot and putting potted plants (like green onions) in between mung bean pots for pest control.

**Mung bean growth characteristics**. The growth characteristics of the plant were observed per treatment. Data recorded included days to flowering, days to pod formation, days to pod maturation, and plant height (cm). First day of full flowering, pod production and maturing (indicated by mature black pods) were noted for each of the treatment found in the 4 blocks. Five plants were selected randomly from each treatment per block for measuring plant height after the 15<sup>th</sup>, 30<sup>th</sup>, 45<sup>th</sup> and 60<sup>th</sup> day. The number of plants selected was reduced to 3 on the 45<sup>th</sup> and 60<sup>th</sup> day due to some plant deaths and use of some plants for nodule formation during flowering stage. Plant height was measured from the ground to the apex of the plant in each block.

*Mung bean yield determination.* The parameters recorded after 60 days included data on the number of pods/plant and seeds/mature pod. Plants with mature pods were selected from each block for data recording. At maturity after 60 days, the total number of pods/plant and number of seeds per pod were obtained on the net plot basis.

*Mung bean root nodulation*. Growth characteristics below ground of mung bean were analysed using the data collected on the number of root nodules formed. Three plants were randomly selected per treatment for data gathering. The plants were soaked in water to loosen the soil, and the roots carefully cleaned without removing the nodules. The nodules were counted manually after washing the roots to remove the soil.

*Statistical analysis.* The data from pot experiments were statistically analyzed using SPSS 17 (www.brothersoft.com/Windows/Business/Data Management). The data were checked to satisfy the assumptions needed to use the One-Way and Two-Way ANOVA and post hoc tests used were Tukey HSD Statistics, the Games-Howell Procedure and Duncan's Multiple Range Test.

#### **Results and Discussion**

*Growth characteristics of mung bean*. The changes in the mung bean's growth were monitored in terms of number of days to reach flowering, pod formation and maturity. Increases in plant height were likewise monitored. Mung bean plants in biochar-amended soils had earlier flowering, fruiting and maturing stages than those without biochar as shown in Table 1. Means of 33, 34 and 48 days to flowering, pod formation, and maturity respectively were observed to be 6 to 7 days earlier for biochar-amended soils B and C than those planted on soil only.

Table 1

Growth characteristics of mung bean in soil with and without biochar amendment

Treatment	Days to flowering	Days to pod formation	Days to maturity
A (soil only)	$39.75 \pm 1.92$	$41.75 \pm 2.21$	$55.00 \pm 2.16$
B (8.5% biochar in soil)	$33.00 \pm 0.71$	$33.75 \pm 1.26$	$48.50 \pm 0.58$
C (15.7% biochar in soil)	$33.25 \pm 1.09$	$34.25 \pm 1.26$	$48.25 \pm 1.71$
D (vermicast fertilized soil)	$35.50 \pm 1.50$	$38.25 \pm 2.06$	$50.67 \pm 1.73$
E (7.5% biochar in soil-vermicast mixture)	$34.50 \pm 0.50$	$35.50 \pm 0.58$	$49.50 \pm 0.58$
F (13.9% biochar in soil-vermicast mixture)	$33.00 \pm 1.22$	$34.00 \pm 1.41$	$49.00 \pm 1.41$
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\*Significant at 0.05 level.

One way-ANOVA showed significant difference (p < 0.05) in each of the growth stages of mung bean (Table 2) since  $F_{calculated} > F_{critical at 5\% level}$ .

Table 2

One-way ANOVA on the differences of growth stages of mung bean using 6 treatments

		Sum of squares	df	Mean square	F	F(p = 0.05)
	Between groups	135.833	5	27.167	13.04	2.77
Flowering	Within groups	37.5	18	2.083		
	Total	173.33	23			
	Between groups	200.500	5	40.100	16.40	2.77
Pod formation	Within Groups	44.000	18	2.444		
	Total	244.500	23			
	Between groups	126.875	5	25.375	11.49	2.77
Maturation	Within Groups	39.750	18	2.208		
	Total	166.625	23			

Results of post hoc analyses using Tukey HSD test indicated differences in each growth period in terms of number of days from planting to the time of full flowering, pod formation and maturity (Table 3). Generally, the means of these growth stages in treatment A are significantly lower than in the other treatments.

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I Flowering	Tractment	Treatmont N (blacks)		Subset for alpha = 0.05		
T. Flowering	meatment	IN (DIUCKS)	1		2	
	В	4	33.00			
	F	4	33.00			
T L LIOD	С	4	33.25			
Tukey HSD	E	4	34.50			
	D	4	35.50			
	А	4			39.75	
	- · ·		Subset	for alpha =	= 0.05	
II. Pod formation	Treatment	N (blocks)	1	2	3	
	В	4	33.75			
	F	4	34.00			
TUKOV HSD	С	4	34.25			
Tukey 115D	E	4	35.50	35.50		
	D	4		38.25		
	А	4			41.75	
III Maturity	Treatment	N(hlocks) -	Subset	Subset for alpha = 0.05		
III. Maturity	neatment	N (DIOCKS)	1		2	
	С	4	48.25			
	В	4	48.50			
Tukey HSD	F	4	49.00			
	E	4	49.50			
	D	4	50.67			
	А	4			55.00	

Tukey HSD post hoc analysis of each stage in mung bean growth

Means for groups in homogeneous subsets are displayed.

According to Shanmugasundaram et al (2009), traditional mung bean cultivars grown in Asia reach maturity in 90 to 110 days. The results of this study indicated earlier growth developmental stages. The results of this study for the three stages are within this range and clearly shows that the addition of biochar to soil could enhance growth development. Agugo et al (2010) studied the growth of four mung bean genotypes and reported significant differences in days from emergence to flowering from 32.00 to 34.1 days a mean of 33.09. The differences, type of soil and varied response of the plant to different environmental conditions to which the plants are exposed. In this study, the early start in the growth development of mung bean in biochar-amended soils than those without biochar could probably be due to greater availability of nutrients resulting from improved soil structure and provision of enhanced conditions for growth.

**Plant height**. The growth performance of mung bean in terms of plant height for the 6 treatment groups at four different periods is shown in Figure 4 and Table 4. Plants grown in treatments B, C and F were observed to be consistently taller than those grown in non-amended soils A and D.



Figure 4. Effect of biochar amendment on the height development of mung bean grown in 6 different soil treatments.

Table 4

Comparison of mean mung bean height after 30 and 60 days of growth

Treatment	Mung bean height (cm)				
meatment	30th day	60th day			
A (soil only)	$12.16 \pm 1.79$	$18.04 \pm 2.62$			
B (8.5% biochar in soil)	15.72±0.91	22.86±3.18			
C (15.7% biochar in soil)	$15.16 \pm 1.32$	$23.37 \pm 4.75$			
D (Soil + vermicast)	$11.39 \pm 2.56$	$20.17 \pm 3.30$			
E (7.5% vermicast fertilized soil	$13.40 \pm 2.39$	$20.39 \pm 3.71$			
F (13.39% vermicast fertilized soil	$15.19 \pm 1.46$	$22.14 \pm 4.82$			

Soil porosity can be enhanced by biochar amendment as reported by previous studies (Troeh & Thompson 2005; Thies & Rillig 2009). The greater capacity of the soil to store moisture and the increased adsorptive capacity to retain nutrients resulting from biochar amendment could have caused more nutrients available for plant growth. In effect, more pronounced and faster growth development was observed for plants grown in biochar amended soils.

Statistical analysis of plant height in biochar amended and un-amended soil are given in Tables 5 and 6 which show significant differences in these two types of soil both after 30 and 60 days.

Tukey's HSD post analysis (Table 7) on the 30th day indicated that plants in treatments B, C and F are significantly taller (p < 0.01) than those in treatments A, D and E. After 60 days, plants grown in treatments B and C were the tallest and differed significantly with treatment A. However, they did not differ significantly with the fertilized treatments D, E and F (p > 0.01). Treatments A, D, E and F did not differ significantly with each other at the end of 60 days

Table 5

One-Way ANOVA on the differences of plant heights of mung bean when categorized by 6 different treatments (April 30, 2011)

Source	Sum of squares	df	Mean square	F	F(p = 0.01)
Between Groups	325.262	5	65.052	14.123	3.187
Within Groups	525.117	114	4.606		
Total	850.380	119			

Table 6

One-Way ANOVA on the differences of plant heights of mung bean when categorized by 6 different treatments (May 30, 2011)

Source	Sum of squares	df	Mean square	F	F(p = 0.01)
Between groups	309.620	5	61.924	4.206	3.289
Within groups	1148.361	78	14.723		
Total	1457.980	83			

Table 7

Tukey HSD Post Hoc Analysis of plant height after 30 days (April 30, 2011) (left part of the table); Games-Howell Post Hoc Analysis of Plant height after 60 days (May 30, 2011) (right side of the table)

		-			-	-		
Experimental	Ν/	Sub	oset	Experimental	Λ/	Sub	Subset	
groups	/ 1	1	2	groups	/ V	1	2	
D	17	11.39		А	16	18.04		
А	20	12.16		D	10	20.17	20.17	
E	19	13.40		E	10	20.39	20.39	
С	20		15.16	F	16	22.14	22.14	
F	20		15.19	В	16		22.86	
В	20		15.72	С	16		23.37	

*Growth yield of plants.* The data used to determine growth yield were pods/plant and seeds/mature pod. Table 8 shows the yield components of mung bean in un-amended (A, D) and biochar amended soils (B, C, E, F). Plants grown in biochar amended soils showed more positive response than the un-amended ones. The number of pods in treatments B and C were higher than those in A, D, E and F but C and F did not differ significantly. The results have shown that for unfertilized treatments, biochar amendment had enhanced growth yield (pod formation) whereas no significant effect on growth yield of fertilized soils resulted from the addition of biochar.

Since the count data on the number of pods/plant were found to be positively skewed, the square root transformation was used to satisfy the assumption for homogeneity of variance. Two-way ANOVA showed significant differences on the number of pods per plant among the treatments (p < 0.01) (Table 9). The post hoc Tukey HSD test on pods/plant revealed that treatments A, D, E and F do not differ significantly, as well as treatments F and C. It is interesting to note however that the means of the number of pods of treatments B and C do not differ significantly but are significantly higher than the means of the other treatments. This indicates that more pods are developed in unfertilized but biochar-amended acidic soils (Table 10).

Table 8

Treatment	Pods/plant (60 days)	Number of seeds/ mature pod (55 days)
A (soil only)	$2.15 \pm 0.74$	$5.82 \pm 2.02$
B (8.5% biochar in soil)	$4.56 \pm 1.63$	$5.14 \pm 0.81$
C (15.7% biochar in soil)	$4.13 \pm 2.02$	$6.63 \pm 1.96$
D (vermi-cast fertilized soil)	$2.19 \pm 0.75$	$7.81 \pm 1.40$
E (7.5% biochar in soil-vermicast mixture)	$3.06 \pm 1.15$	$6.50 \pm 2.07$
F (13.9% biochar in soil-vermicast mixture)	$3.19 \pm 1.05$	$5.82 \pm 1.78$

Yield components of mung bean in biochar amended and un-amended soils

Table 9

One-Way ANOVA on the differences of number of pods per plant of mung bean when categorized by 6 different treatments

Source	Sum of squares	df	Mean square	F	F (p = 0.01)
Between Groups	81.125	5	16.225	9.513	3.255
Within Groups	153.500	90	1.706		
Total	234.625	95			

Table 10

Tukey HSD Post Hoc Analysis of pods/plant in six different soil treatments

Experimental group	Λ/	Subset			
Experimental group	74	1	2	3	
A	20	2.1500			
D	16	2.1875			
E	16	3.0600			
F	16	3.1875	3.1875		
С	16		4.125	4.125	
В	16			4.5625	

One-way ANOVA on the number of seeds/mature pod was found not to vary significantly among treatments since F calculated was less than  $F_{critical}$  at 1% level (Table 11). Therefore, no post hoc evaluation was performed.

Table 11

One-Way ANOVA on the differences of seeds/mature pod after 55 days of mung bean when categorized by 6 different treatments

Source	Sum of squares	df	Mean square	F	F(p = 0.01)
Between groups	47.03	5	9.4	3.12	3.34
Within groups	181	66	3.02		
Total	228.03	83			

**Root nodulation in mung bean plants**. Root nodulation indicates  $N_2$  fixation and the availability of  $NH_3$  and  $NO_3$  ions needed for plant growth. Table 12 shows that mung bean possesses the inherent property to fix  $N_2$  as indicated by the formation of nodules in its

roots. Among the six treatment groups studied, the rate of nodule formation was highest in non-fertilized biochar amended soils, B and C. It is interesting to note that the combination of soil and vermicast (D) has hindered nodule formation as the number of nodules formed was less in fertilized soils amended with biochar than those treatments without fertilizer. Table 13 shows significant differences in nodules formed in the roots both at the on set of flowering (35 days) and maturity stage (54 days).

#### Table 12

Nodule formation in the roots of mung bean in the different soil treatments					
	No. of nodules/plant				
Treatments	35 days (flowering)	54 days (maturation)			
A (soil only)	$15.67 \pm 6.36$	$25.00 \pm 6.08$			
B (8.5% biochar in soil)	$26.67 \pm 3.06$	$43.33 \pm 13.53$			
C (15.7% biochar in soil)	$53.33 \pm 12.58$	$49.67 \pm 8.50$			
D (vermicast fertilized soil)	9.67±4.51	$17.00 \pm 9.00$			
E (7.5% biochar in soil-vermicast mixture)	$11.33 \pm 3.21$	12.67±8.33			
F (13.9% biochar in soil-vermicast mixture)	$10.33 \pm 4.16$	$14.33 \pm 2.31$			

Nodule formation in the roots of mung bean in the different soil treatments

Table 13

One-Way ANOVA on the differences of root nodule formation in mung bean using 6 treatments

Nodulation after days		Sum of squares	df	Mean square	F	F (p = 0.05)
35 days	Between groups	4324.500	5	864.900	13.80	3.11
	Within groups	752.000	12	62.667		
	Total	5076.500	17			
54 days	Between groups	3536.278	5	707.256	4.673	3.11
	Within groups	1816.000	12	151.333		
	Total	5352.278	17			

Post hoc analysis (Table 14) showed that at flowering stage 35 days after germination, the mean of nodule formation in treatment C is significantly higher than the other 5 treatments (p < 0.05). Treatments A and B did not differ significantly while no significant difference was likewise found in treatments A, D, E and F. The results clearly showed that addition of vermicast has hindered nodule formation in the roots.

Table 14

Post hoc analysis of root nodule formation in mung bean after 35 days at the on-set of flowering

Nodulation	Treatment	N -	Subset for alpha = 0.05		
			1	2	3
Duncan test	D	3	9.67		
	F	3	10.33		
	E	3	11.33		
	А	3	15.50	15.50	
	В	3		26.67	
	С	3			53.33

At plant maturity, after 54 days, the post hoc analysis of nodule formation in the roots (Table 15) indicated that treatments with soil-biochar combination (B and C) differed significantly from the un-amended treatment (A) and those fertilized with vermicast (D, E and F).

Nodulation	Treatment	<u>.</u>	Subset for $alpha = 0.05$		
		74	1	2	3
Duncan test	E	3	12.6667		
	F	3	14.3333		
	D	3	17.0000		
	А	3	25.0000	25.0000	
	В	3		41.0000	41.0000
	С	3			49.6667

Post hoc analysis of root nodule formation in mung bean after 54 days at maturation stage

Table 15

According to Lindemann & Glover (2003), legumes do not respond to nitrogen fertilizer as long as they are capable of fixing nitrogen. When large amount of nitrogen is applied as could have happened when vermicast was added, the plant literally slows or shuts down the nitrogen fixation process since it is easier for the plant to absorb nitrogen from the soil than to fix it from air. Vermicompost are reported to contain 1.5 to 2.2% N (Adhikary 2012). The vermicast added to the soil could have provided the nitrogen needed by the plant thereby slowing down the  $N_2$  fixation process as well as its nodule formation.

The appearance of nodules formed at the roots of the plants during pod formation stage is shown in Figure 5. The nodules formed were pink in color in unfertilized treatments with and without biochar. This agrees with the finding of Lindemann & Glover (2003) that pink or red nodules predominate on legumes in the middle of the growing season.

Aeration is also essential to  $N_2$  fixing bacteria activity. Bhattarai et al (2005) stated that good growth occurs in deeper layers of soil if the medium is aerated. It should be noted that most of the nodules that formed in A (soil only) are found near the surface of the soil (Figure 5A).

Treatments B and C had nodules formed on the roots extending deeper on the soil layer indicating a probable  $N_2$  fixing activity of microorganisms deeper down the soil and expectedly holding more N in the soil than those in un-amended ones. Again, this could be attributed to the increased soil porosity made possible by biochar addition. The void spaces in the pores allowed passage of air necessary to support microbial activity that aids in the mineralization of nutrients.

Treatments with vermicast and biochar likewise exhibited root nodule formation (Figure 5D, 2E, 2F). However, the number of nodules are less in these fertilized treatments than those without fertilizer. According to Lindemann & Glover (2003), legumes do not respond to nitrogen fertilizers as long as they are capable of fixing nitrogen from air. It appears that the addition of vermicast slowed down or could have shut down the nitrogen fixing process and prevented nodule formation. Instead of fixing nitrogen, mung bean seemed to have chosen the easier and less energy consuming process of absorbing nutrient from the soil. It is interesting to note that the absence of nodules is compensated by the development of more rootlets in the plants. The presence of these rootlets/roothairs could aid in the absorption of nutrients needed by the plants for growth.



Figure 5. Root nodule formation in the six soil treatments. Treatments A and D are without biochar while B and C are unfertilized soil with biochar. Treatments E and F are fertilized with biochar amendment.

The quantity of nitrogen fixed for plant use is controlled by the plant itself (Mia et al 2014; Rondon 2007. When there is limited soil moisture, nitrogen fixation slows down, dictated by the plant's diminishing nitrogen requirement. This is remedied by the addition of biochar to soil to improve soil porosity and the ability of the soil to retain water. Water held in the pores can dissolve nutrients and render them useful for plant growth and reproduction.

*Effect of biochar amendment on soil pH*. There was a continually increasing trend in pH during the vegetative growth of the plant up to maturity both in the control pots and those amended with biochar (Figure 6).



Figure 6. pH changes in the different soil treatment from an initial soil pH of 4.99.

Clearly, the greater the amount of biochar added, the more basic the soil becomes. This is expected since bamboo biochar has an alkaline pH. The pH in B and C treatments increased by 5.8% and 8.1% respectively compared to the negative control A. In the case of the fertilized soils, E and F treatments increased by 3.8% and 5.3% respectively compared to the positive control D.

Even without biochar addition, soil pH increased throughout the duration of the experiment in treatments A and D. This could probably be due to interactions brought about by the N<sub>2</sub>-fixing bacteria that thrive in leguminous crops like mung bean. Several researches have reported increase in pH upon incorporation of biochar in the soil (Yuan & Xu 2011; Gaskin et al 2010; Chan et al 2007). These researches also reported increase in exchangeable base cations, effective cation exchange capacity and base saturation upon biochar addition. Yuan & Xu (2011) further suggested that the liming effects of biochar samples produced from crop residues on soil acidity closely correlated with alkalinity. This implies that biochar alkalinity is a key factor in controlling the liming effect on acid soils.

Conclusions. The results of this study revealed that the growth development of mung bean in acidic soil could be enhanced by biochar amendment. Evidently, earlier stages of flowering, pod formation and maturity of plants occurred in treatments where soils were amended with biochar than those without biochar. The presence of biochar in the soil could also enhance plant height. Plants were significantly taller in biochar amended soil both in unfertilized soil and in vermicast-fertilized soil. The results also show that significant increase in pod formation per plant can be made possible by biochar amendment. However, it appears that increasing the amount of biochar amendment (8.5 to 15.7% in unfertilized soil and 7.5 to 13.4% in fertilized soil) can not cause significant difference in the number of pods formed at maturation stage. Soil quality could likewise be improved by biochar amendment as indicated by reduced acidity (higher pH) and development of nodules in the roots. These changes in pH and nodule formation are essential in the growth of legumes like mung bean. They provide the necessary conditions for the release and mineralization of nutrients and fixed nitrogen needed in plant growth. The study has shown that biochar amendment is a promising technology in improving mung bean production in acidic soils.

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