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EFFECTS OF RICE HUSK BIOCHAR ON THE GROWTH CHARACTERISTICS, RHIZOSPHERIC MICROFLORA AND YIELD OF TOMATO PLANTS

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ABSTRACT

Human activities have degenerated nearly 40% of the universe soil through excessive chemical inputs, tilling and industrial activities. Biochar can increase soil fecundity, agricultural producti- vity and protects soil-borne diseases. This study aimed to assess the efficacy of rice husk biochar on the growth and yield of tomato plants. Biochar was produced through feedstock (rice-husk) pyrolysis and used as soil amendments at different concentrations (20 t/ha and 40 t/ha) and an unamend soil served as control. Physicochemical properties of the soil, rice husk, biochar and biochar soil before and after planting were determined using standard methods. Rhizospheric microflora of the plants were isolated and identified based on morphological and biochemical characterizations. Agronomic parameters such as plant height, stem girth, leaf area, number of leaves, flowers, fruits and weight of fruits were determined. Results revealed that rice husk biochar (40 t/ha) had the highest physical and chemical parameters while the unamend soil had the lowest constituents. Predominant bacterium and fungus are *Bacillus subtilis* and *Aspergillus niger*. Agronomic parameters: plant heights, stem girths, leaf areas, number of leaves, flowers, fruits and weight of fruits were higher in all the amended soil than the control at the various growth stages. The unamend soil yielded no fruit at 10th week while the amended soil yielded an average fruit weight of 34.95g ± 8.76 and 21.53g ± 5.16 at 20 t/ha and 40 t/ha respectively. This study revealed that biochar produced from rice husk could be used to improve growth and yield of tomato plants.

Keywords: Biochar, pyrolysis, rice-husk, tomato, bacteria, fungi

INTRODUCTION

Waste biomass (about 140 billion metric tonnes) are generated globally from agricultural activities every year (UNEP 2009). One of the most widely available agricultural wastes is rice-husk and it produces about 20–23% by weight of the entire paddy processed [1, 2]. In Nigeria, for example, an estimated amount of 1,032,993.6 metric tonnes of rice husk are generated annually [3, 4]; this has led to the growing interest in the conversion of waste or biomass into resources through pyrolysis to biochar [5].

Biochar is a carbon-rich substance produced through pyrolysis; a thermal decomposition of biomass in the presence of little or no oxygen [6]. It is a stable and recalcitrant form of organic matter that can persist in the soil for several years [7]. Biochar is black, highly porous and finely grained, with light weight and large surface area that has a positive effect when incorporated into the soil [3]. Biochar improve the water holding capacity and availability of nutrients [8], influence mycorrhizal abundance and allows improved uptake of nutrients by plants [9].

Amendment of soil with biochar improves the quality of the soil and crop yield by increasing the soil pH, moisture-holding capacity, cation-exchange capacity and the soil microflora [10]. It also has environmental benefits as it helps to mitigate global warming and a potential of toxic element adsorption [11]. Lashari *et al.* [12] have reported remarkable agronomic values and yield potential in poor degraded soils upon the addition of biochar to soil. Agricultural wastes that are mainly composed of cellulose, hemicellulose and lignin polymers have been widely used as feedstock (biomass) for biochar production [13]. Some of the agricultural wastes widely used are grass, rice husk, wood chips, saw dust, bagasse, cow dung etc. The type of agro waste used as biomass and the pyrolysis condition largely affect the properties of the biochar produced [14].

Rice husk is a hard shell that covers a grain of rice. It is usually removed during milling process thereby becomes an agro waste generated during rice production [15]. In Nigeria, local production of rice has increased in the last few years as the Federal Government banned the importation of rice. This had led to the generation of large quantity of rice husk which need to be managed. Hence this study aimed to evaluate the effect of biochar produced from rice husk feedstock on the growth characteristics, rhizospheric microflora and yield of tomato plants.

MATERIALS AND METHODS

Sample Collection

Topsoil was collected at (0-10 cm) from DURFARMS, Federal University of Agriculture, Abeokuta while rice husk was collected from a rice processing factory in Lafenwa, Abeokuta, Nigeria. Tomato (Beske cultivar) seeds were obtained from Institute of Agricultural Research and Training, Ibadan, Nigeria.

Pyrolysis

This was done using the two barrel metal retort method. The retort chamber (114 liters) was filled with rice husk and then suspended in a 208 liters capacity drum perforated at the sides around the bottom and the top. The space between the two drums was then filled with smaller wood chips and then wet with kerosene (starter fluid) and set on fire for 3 hr. The big metallic drum was covered with a lid built with an exhaust pipe which supports chimney process. After the pyrolysis, the drum was allowed to cool and the rice husk char was removed and stored in a cleaned sack [16]

Determination of physicochemical properties of the soil, rice husk and rice husk biochar before and after planting

The physical and chemical parameters: soil texture, acidity, pH, E.C., C: N ratio, P, Na, Ca, K, Mg, H^+, S, Mn, Fe, Cu and B were determined using standard methods [17]

Isolation of rhizospheric microorganisms

Rhizospheric soil samples were serially diluted to 5-fold using sterile distilled water. Using pour plate method, 1 mL inoculum was inoculated on Nutrient Agar (NA) and Potato Dextrose Agar (PDA). The NA plates were incubated at 37 °C for 24 hr while PDA plates were incubated at 28 °C for 48-72 hr. Distinct colonies were physically enumerated on plates and sub-cultured to obtain pure cultures. Bacterial and fungal isolates were identified based on their cultural, morphological and biochemical characteristics with reference to Bergey's Manual [18].

Green house experiment on the growth of tomato plant with different biochar amendment

The experiment was arranged in a completely randomized design with three treatments and four replicates. Five kilogram topsoil was mixed with the rice husk biochar at two different rates (20 t/ha and 40 t/ha) in a 7-litre plastic container. The untreated soil served as the control. The mixtures were allowed to stand for two weeks for homogeneity of the mixture with addition little amount of water. Then nurseries of tomato seeds raised for three weeks were transplanted into each pot at 2 strands per pot and were watered twice daily. Agronomic parameters such as the plant height, number of leaves, leaf area, stem girth, number of flowers, number of fruits and fruit weight were measured at two weeks interval after transplanting [19].

Data Analysis

Experiments were carried out in four replicates. All the data obtained were subjected to Analysis of Variance (ANOVA) and means were separated using Duncan Multiple Range Test (DMRT). Mean values obtained from the DMRT was used to plot bar chart in excel and their error bars and LSD values were indicated.

RESULTS

Physicochemical properties of the soil, rice-husk and rice-husk biochar

The physicochemical properties of the soil, rice husk and rice husk biochar are shown in Table 1. The rice husk biochar had the highest values than the soil without biochar except for the soil acidity. pH (7.80), electrical conductivity (2.04 µS/cm), cation exchangeable capacity (11.70 cmol/kg) and available phosphorus (3.41 g/kg), exchangeable bases (sodium, calcium, potassium and magnesium) were all higher in the rice husk biochar than the soil.

Physicochemical analysis of unamend soil and rice-husk biochar soil after planting

Table 2 shows the results for the analysis of unamend soil (control) and rice-husk biochar soil at different concentrations after planting. The result showed that physical and chemical parameters of the control (soil without biochar) had lower values than the amended soil at the different concentrations except for Na, H^+ and P indicating that unamended soil is also a rich substrate.

Table 1: Physicochemical properties of the soil, rice husk and rice husk biochar before planting

Parameters	Soil	Rice husk	Rice husk bio char
pH	6.28	5.60	7.80
Electrical conductivity (µS/cm)	1.26	0.18	2.04
Carbon:Nitrogen (C:N) ratio	9.7:1	92.6:1	65.1:1
Available Phosphorus (g/kg)	1.45	0.87	3.41
Cation Exchangeable Capacity (cmol/kg)	4.11	7.69	11.70
Exchangeable acidity (cmol/kg)	0.10	0.13	0.07
Sodium (cmol/kg)	0.27	0.21	1.30
Calcium (cmol/kg)	2.45	1.17	9.50
Potassium(cmol/kg)	0.47	2.08	12.50
Magnesium(cmol/kg)	5.41	1.20	11.12
Soil texture (cmol/kg)	Sandy-loam	-	-

Table 2: Physicochemical analysis of unamend and rice-husk biochar soil after planting

S/N	SAMPLE CODE	pH	Ca(cmol/Kg)	Mg cmo /Kg	K cmo /g	Na cmo/Kg	H^+	CEC cmol/Kg	P ppm	% org. S	% T.N	Mn ppm	Fe ppm	Cu ppm	B ppm
1	T1(40t/ ha)	7.85	9.94	1.47	0.43	0.21	0.08	12.14	5.67	1.34	0.12	0.12	172.72	2.30	0.22
2	T2(20t/ ha)	7.61	6.27	1.29	0.55	0.23	0.07	8.41	9.47	1.26	0.12	0.12	137.32	1.45	0.14
3	Control	6.19	5.05	1.28	0.43	0.24	0.10	7.09	9.99	0.90	0.07	0.07	153.40	1.40	0.16

Distribution of bacteria at different growth stages of tomato plants

Table 3 shows the counts of the identified bacteria at the vegetative, flowering, fruiting and harvesting stage. Six bacteria were isolated from the rhizosphere of the amended soil. The isolated bacteria and their incidence rate were *Pseudomonas aeruginosa* (24.1 %), *Bacillus subtilis* (41.4 %), *Bacillus megaterium* (10.3 %), *Staphylococcus aureus* (3.5 %), *Staphylococcus saprophyticus* (17.2 %) and *Citrobacter* sp. (3.5 %). *Bacillus subtilis* was seen to have the highest number of occurrence and growth. At the vegetative stage, *B. subtilis* had the highest number (20) followed by *P. aeruginosa* while the lowest populated organism was *S. aureus.*

During the flowering stage, *B. subtilis* was still the most populated having 40 counts, then *B. megaterium* was the least having 15 counts. At the harvesting stage, *P. aeruginosa* was not dectected at all, *B. megaterium* had the lowest value of 3 and *B. subtilis* was still the most dominant organism having 20 counts.

Table 3: Distribution of bacterial isolates at each growth stage of tomato plants

ISOLATE	Vegetative Stage	Flowering Stage	Fruiting Stage	Harvesting Stage
Pseudomonas aeruginosa	20	25	15	-
Staphylococcus saprophyticus	15	20	20	10
Citrobacter sp.	15	20	15	5
Bacillus megaterium	10	15	10	3
Bacillus subtilis	30	40	30	20
Staphylococcus aureus	12	23	14	12

Distribution of fungi at different growth stages of tomato plants

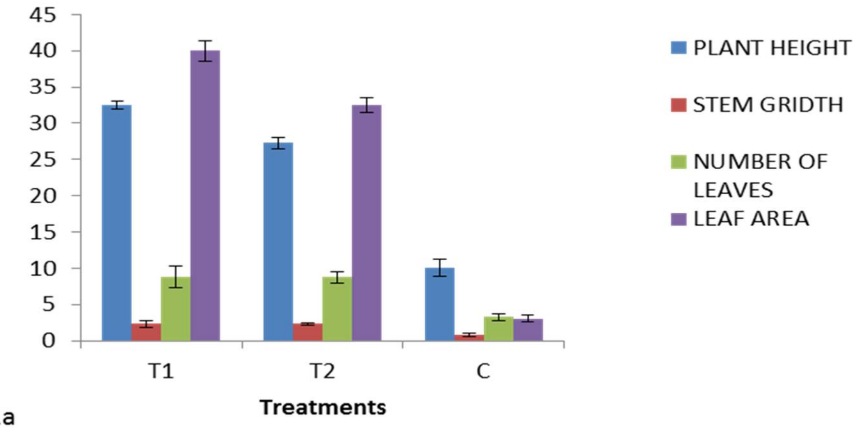
Table 4 shows the fungi species that was isolated at the growth stages of the tomato plant. *A. niger* and *Mucor* were identified in the unamend soil while *Fusarium* sp., *A. niger*, *A. fumigatus, A. flavus*, *Penicillium* sp. and *Mucor* sp were obtained from the rhizosphere of the biochar soil at different stages of the tomato plant. However, only *A. fumigatus* and *A. niger* were obtained at the harvesting stage.

Table 4: Identified fungi from the amended and unamend rice-husk biochar soil

Growth stages of tomato plant	Identified fungi isolates
Vegetative stage	*A.niger, A. fumigatus* and *Penicillium* sp.
Flowering stage	*Fusarium sp.*, *A.niger* and *Rhizopus* sp.
Fruiting stage	*A.flavus, Penicillium* sp. and *Mucor* sp.
Harvesting stage	*A.niger* and *A. fumigatus*
Unamend (control)	*Mucor* and *A.niger*

Planting of tomato with different biochar amendment: T_1 (40/ha), T_2 (20t/ha) and Control (0t/ha biochar)

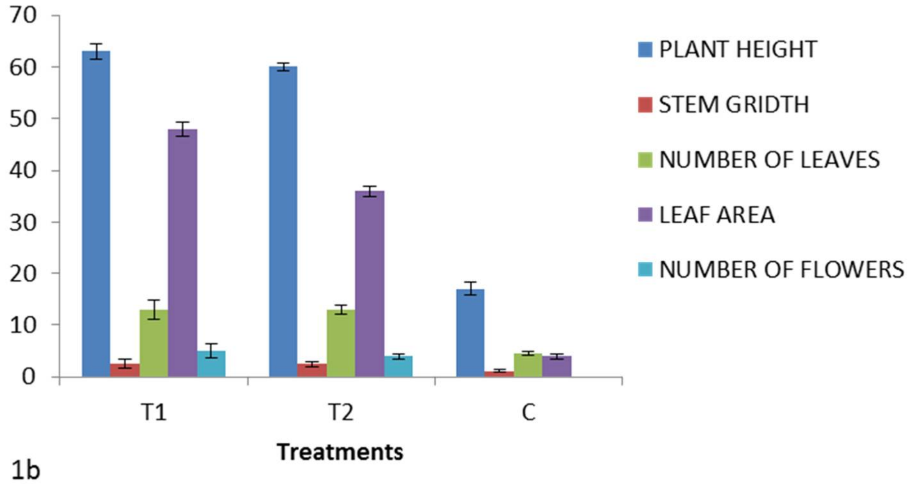
Figure 1(a-e) show the effect of rice husk biochar on agronomic parameters of the tomato plants. Plant height and stem girth were measured in centimetre (cm), leaves area was measured in centimetre square (cm^2) and fruit weight was measured in gram (g). The agronomic parameters were significantly higher in the biochar amended soil than the control at various growth stages under different biochar doses. The values were higher in the plants amended with 40 t/h biochar than those treated with 20 t/h at the vegetative and flowering stage. At the fruiting stage, the plants treated with 20 t/h biochar had higher values than those treated with 40 t/ha except for number of fruits and weight of fruits. The control had delayed flowering up to the 8th week and yielded no fruit at 10th week. Plate II and Plate III depict the flowering and fruiting stages. Flowering of the tomato plant commenced after the vegetative stage ended and the flowering process started after the fourth week of transplanting. The fruiting process started from about 9th week of transplanting but was well pronounced after the 10th week.



1a

Figure 1a: Agronomic parameters for T_1 (40/ha), T_2 (20t/ha) and Control (0t/ha biochar) after 2 weeks of transplanting

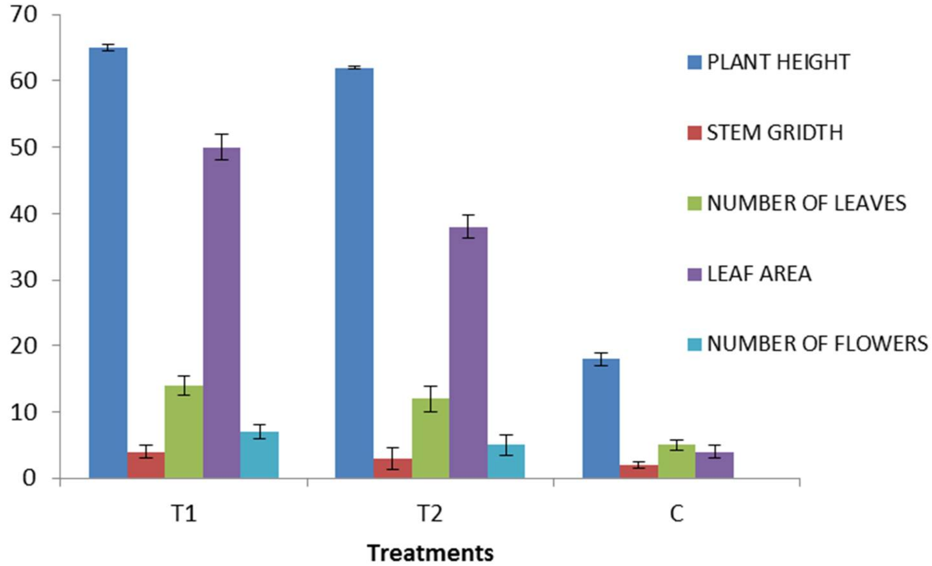
LSD: Plant height = 5.20**, Stem Gridth = 1.50, Number of leaves = 5.5, Leaf Area = 7.53**
Note: Values with Asterisks connotes significant Difference



1b

Figure 1b: Agronomic parameters for T_1 (40 t/ha), T_2 (20t/ha) and Control (0t/ha biochar) after 4weeks of planting

LSD: Plant height=3.00*, Stem Gridth = 1.21, Number of leaves = 3.42, Leaf Area = 12.00**, Number of flowers = 1.31, Note: Values with Asterisks connotes significant Difference



1c

Figure 1c: Agronomic parameters for T_1 (40 t/ha), T_2 (20t/ha) and Control (0t/ha biochar) after 6 weeks of planting

LSD: Plant height = 3.00**, Stem Gridth = 1.00**,Number of leaves = 2.00**, Leaf Area = 12.00**, Number of flowers = 1.50*. Note: Values with Asterisks connotes significant Difference

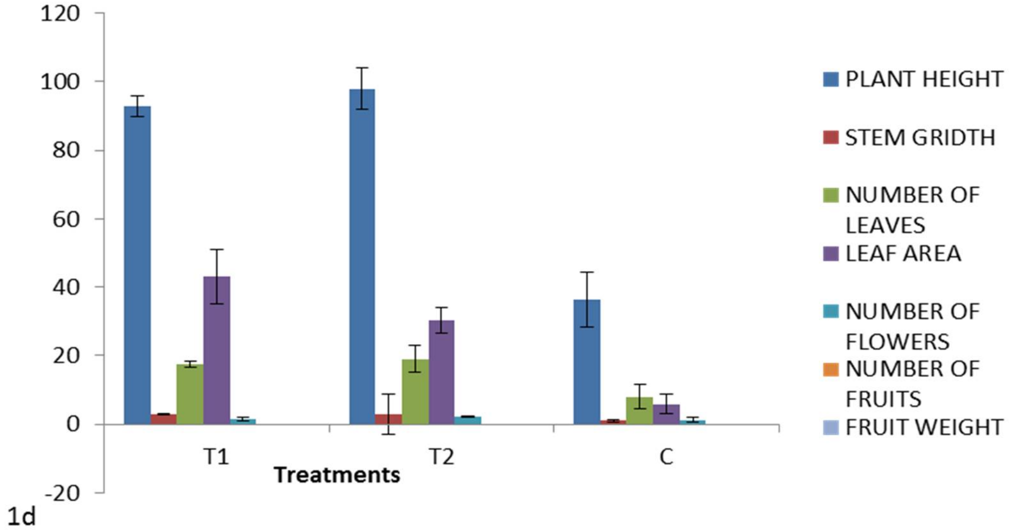


Figure 1d: Agronomic parameters for T_1 (40 t/ha), T_2 (20t/ha) and Control (0t/ha biochar) after 8 weeks of planting

LSD: Plant height = 5.07**, Stem Gridth = 0.25**, Number of leaves = 1.50**, Leaf Area = 12.00**, Number of flowers = 0.12, Number of fruits = 0.10, Fruit weight = 0.01
Note: Values with Asterisks connotes significant Difference

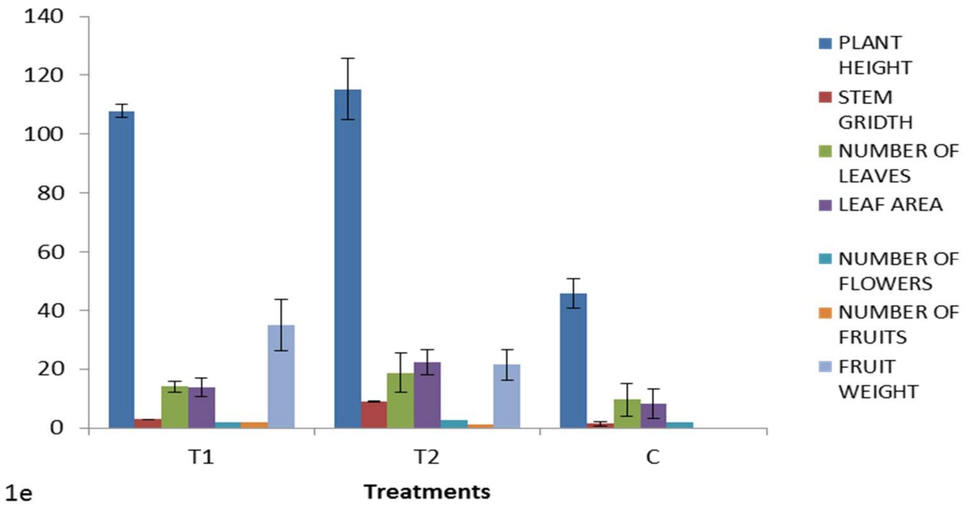


Figure 1e: Agronomic parameters for T_1 (40 t/ha), T_2 (20t/ha) and Control (0t/ha biochar) after 10 weeks of planting

LSD: Plant height = 7.47**, Stem Gridth = 5.90**, Number of leaves = 4.75**, Leaf Area = 8.65**, Numberof flowers = 0.02,Number of fruits = 0.01, Fruit weight = 2.50**
Note: Values with Asterisks connotes significant Difference

Plate 2: Flowering stage of tomato plants

Plate 3: Fruiting stage of tomato plant

DISCUSSION

Biochar science has been regarded as a reliable source of energy. Demerits involved with the use of chemical fertilizers could be replaced with biochar application due to biochar's ability to bio-fortifify, increase soil microbial population, microbial activities and reduce loss of soil nutrients from leaching.

Physical and chemical analyses showed that rice-husk has a rich constituents that led to the enrichment of the rice-husk biochar soil. Higher pH of rice husk biochar implies that the biochar increased the pH of the soil and increased the availability of nutrients to the plants and thereby improving the plant growth. In a similar study, Mishra *et al*. also reported a higher pH (10.50), electrical conductivity (1.67 µS/cm), cation exchangeable capacity (37.40 cmol/kg) and total phosphorus (2.01 g/kg); this also implies that the addition of the rice-husk biochar to the soil helped in the introduction of nutrients to the soil that in turn helped in the various enhancement of the tomato plant. It was observed that most of the mineral contents of the feedstock were available in the biochar after pyrolysis which was similar to the previous research by Amonette and Joseph .

The higher physicochemical values of rice-husk biochar at T2 (40t/h) in soil could also be attributed to its higher tomato yield. Biochar can significantly affect soil moisture, nutrient dynamics and yield of crop and activities of biochar in soil can remain for a longer period of time.

The positive effects of biochar was observed on plant physiology, biomass production and crop yield. Warnock *et al.* indicated that biochar could stimulate microbial communities, increase microbial biomass, provide shelter for microorganisms and increase influx of nutrients while Huang, *et al*. showed that the presence of biochar soil could provide a microhabitat for arbuscular mycorrhizal fungi. Increased in number of bacterial and fungal isolates obtained in this study depicts that the soil ameliorate with biochar is a potential source for the growth of microorganisms. The most dominant bacterial species (*Bacillus subtilis*) obtained in the rice-husk biochar soil have been reported to be used in agriculture to combat a variety of root- pathogens [27]. The isolated bacteria in this study have also been reported for the ability to stimulate the soil microflora in previous study by Thies and Rillig Similar result were obtained in the studies of Akintokun *et al*. where plant growth-promoting bacteria was obtained from the rhizosphere of tomato plants and Kazerooni *et al.* where (*Aspergillus, Fusarium,Penicillium*, *Cladosporium* and *Pythium* were recorded as rhizospheric fungi associated with tomato plants).

Moreover, Molohun *et al*. observed that application of biochar to soil enhances asymbiotic association between microorganisms and plants, promotes growth of soil beneficial microbes involved in plant protection, increased number of root-associated microorganisms and thus, increases crop production. Agbna *et al.* indicated that addition of biochar to soil improved agronomic properties such as plant height, number of leaves as well as fresh and dry of plant parts of tomato. In addition, Lorn *et al*. also reported that amendment of soil with rice husk and Siam weed biochar improved the yield of tomato plants without fertilizers. The result obtained from this study was also in accordance with the work of Elad *et al*. who documented that biochar is a charcoal product that helps to enhance agricultural yields. Khan *et al.* in his work, revealed an increase in plant height to be due to biochar application and attributed it to positive effect of biochar in supplying essential nutrients for vegetative growth of plants.

At the fruiting stage, soil amended with 20 t/ha biochar had highest values than those treated with 40 t/ha, but soil amended with 40 t/ha had highest number of fruits and weight of fruits. However, this study disagrees with the work of Shashi *et al*. [36] who documented that the amendments with 20 t/ha of rice- husk biochar had highest agronomic parameters including the maize yield.

Generally, application of biochar influenced the performance of agronomic parameters and the yield of tomato plant. Many studies have shown yield increases with rice husk biochar, particularly on low fertility soils under both greenhouse and field conditions [8]. Thomas *et al*. [37] and Crane *et al*.[38] in their own contribution stated that biochar improves crop productivity and mitigates drought conditions.

CONCLUSION

This study revealed that biochar produced from rice husk can improve growth and yield of tomato plants. The biochar could be applied at a dose of 40 t/ha as it yielded higher number and weight of tomato fruits. Amendment of soil with rice husk biochar also enhanced the rhizospheric microflora of the tomato plants.

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