

## COMPARATIVE PERFORMANCE OF TWO DIFFERENT BIOFERTILIZERS IN ACIDIC AND ALKALINE SOILS ON OKRA GROWTH

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### Abstract

The present study evaluates the effects of two different types of biofertilizers on the growth of okra (*Abelmoschus esculentus*) following completely randomized design with three treatments (control, vermicompost, trichocompost) and three replications using acid soil (pH 4.77) and alkaline soil (pH 7.87). The results showed partial (pH 5.9) and complete (pH 6.4) neutralization of soil acidity by trichocompost and vermicompost whereas pH increased in alkaline soil but EC decreased significantly by both treatments. Besides, total OC, total N, total P, total K, total S all increased significantly ( $P < 0.05$ ) in both soils (except S in alkaline soil) by both biofertilizer application but total Ca, Mg, Fe, Mn decreased in acid soil while results were mixed in alkaline soil. Results of plant height and root length were increased significantly ( $p < 0.01$ ) in both acidic and alkaline soil by vermicompost and trichocompost application. For nutrient uptake, Vermicompost appeared equally useful for acidic and alkaline soil and trichocompost appeared more useful in acidic soil than alkaline soil. The application of the two biofertilizers in both the soils had significant positive effects on the growth of okra.

*Key words:* Biofertilizers, Acidic soil, Alkaline soil, Okra growth.

### Introduction

Biofertilizers are becoming increasingly popular in many countries and for many crops because of the adverse impacts of inorganic fertilizer on environment and plant growth. Biofertilizers are inputs containing microorganisms capable of mobilizing native elements from non-usable form to usable form through biological processes (Bahadur and Manohar, 2001). Vermicompost (vermi-compost, vermiculture) is the product of the decomposition process using various species of worms, usually red wigglers, white worms, and other earthworms, to create a mixture of decomposing vegetable or food waste, bedding materials, and vermicast (also called worm castings, worm humus, worm manure, or worm faeces). Vermicompost contains water-soluble nutrients and is an excellent, nutrient-rich organic fertilizer and soil conditioner (Kelly and Knutzen, 2008).

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Vermicompost stimulates the microbial activity of soil, improves nutrient content and increase growth yield and quality of the plant (Arora *et al.*, 2011). On the other hand, *Trichoderma* spp. is free living fungi that are common in soil and root systems and are well- known to solubilize phosphates and micronutrients. They can produce phosphates and several organic acids. *Trichoderma* spp. recently was suggested as a plant growth promoting fungi due to their ability to produce siderophores, phosphate solubilizing enzymes, and phytohormones (Haidar *et al.*, 2018).

Vegetables are the integral part of the balanced diet of human since time immemorial. Globally, the role of vegetables has been recognized in solving the problem of food and nutritional security. Okra (*Abelmoschus esculentus* L.) is an important vegetable crop of Malvaceae family, which supplies higher nutrition (carbohydrates, fats, protein, minerals and vitamins) in our diet. Okra is tolerant of a wide range of soil pH but prefers neutral pH of 6.5 to 7.0. It is a fast growing annual which has captured a prominent position among the vegetables in Bangladesh. It is a multiple use crop. Dried seeds are nutritious food. It contains upto 20% protein and the fibre from okra canes is a possible paper pulp source, while the dried canes are a fuel source (Lyngdoh *et al.*, 2013).

### **Materials and Methods**

*Soil sampling site:* Acid soil sample was collected from Kalapara, is an Upazila of Patuakhali District in the Division of Barisal, Bangladesh. The sampling area belongs to the agro-ecological zone, AEZ-13. Alkaline soil sample was collected from Matuail Union, Jatrabari Thana of Dhaka metropolitan. The sampling area belongs to the agro-ecological zone, AEZ-19.

*Processing of the sample:* After collection the soil samples were divided into two parts- one was used for required background analysis of various parameters and the larger portion was kept separately for the development of pot experiment. The former part was air-dried for 4 days by spreading in a thin layer and then hammered to make the clods smaller. Visible roots and debris were discarded from the soil sample. For hastening the drying process, the soil sample exposed to sunlight. After that, larger and massive aggregates were broken down into smaller aggregates by gently crushing them using a wooden hammer. The ground samples were screened through a 2mm stainless steel sieve. The sieved sample was then stored into plastic containers and the mouth was well-capped. Then the container was kept in a cool dry place in the laboratory. The other part of the soil sample was dried in the sun and larger clods were crushed by hammer and roots were discarded. This sample was used for pot experiment. The initial soil sample

was analyzed before set up of the pot experiment. This pot experiment was conducted in the premises (net house) of the Department of Soil, Water & Environment, University of Dhaka. A total number of 18 pots were used for the analysis. Irrigation water quality and amount were carefully maintained. For the pot experiment, vegetable crop called okra (*Abelmoschus esculentus*), in the family of Malvaceae was used.

*Pot preparation and treatment:* Five kilogram (5kg) of soil samples were taken in each seven kilogram (7kg) plastic pot for the culture of 4 okra seeds and growth of okra plants. In order to study the effect of biofertilizer, vermicompost and trichocompost were taken as treatments. Vermicompost was collected from Siddique bazar, Dhaka. Trichocompost was collected from online nurseries and gardening store. Collected biofertilizers were air dried, passed through a 2 mm stainless steel sieve and preserved in airtight plastic containers. For treatment each pot contains 27 g of biofertilizers. The experiment was laid out in a Completely Randomized Design (CRD) having three (3) replications. For each soil, three (3) treatments were distributed randomly.

*Growth performance study:* Plant height were measured using a meter scale from the ground level to the apex. From each pot two plants were measured and averaged (centimeter). Leaf number and pod number were also recorded. The leaf area has been calculated using the linear equation  $LA$  (leaf area) =  $11.98 + (0.06 \times L \times W)$  where  $L$  = length of leaf and  $W$  = width of leaf (Bhatt and Chanda, 2003).

*Digestion of biofertilizer and soil:* For digestion, 0.5 g of biofertilizer was taken. It was digested with aqua-regia (HCl: HNO<sub>3</sub> =3:1). (Jackson, 1962). For digestion of soil, 2.5 g of sieved samples were weighed and digested with aqua-regia (HCL: HNO<sub>3</sub> =3:1). (Jackson, 1962).

*Analysis of soil and biofertilizer:* The pH of the samples were measured electrochemically by using a glass electrode pH meter as described by Jackson (1958). The ratio of sample to water was 1:2.5. The electrode was calibrated using standard buffer solutions at pH 4.0 and 7.0. Electrical conductivity was measured by an EC meter at a ratio soil samples to water as 1:5 as described by USSL staff (1954). Cation exchange capacity was measured by 1N ammonium acetate at pH 7.0 (Chapman and Pratt, 1965). Organic carbon was determined by wet oxidation of Walkley and Black (1934). In this method, the soil carbon was oxidized with chromic acid (derived from potassium dichromate). Then the excess chromic acid left after the oxidation of organic carbon was determined volumetrically with standard ferrous sulphate solution and the quantity of substance oxidized was calculated from the amount of chromic acid required. Organic matter was calculated by multiplying the percentage of organic carbon with

conventional Van- Bemmelen's factor of 1.724 (Piper, 1950). Total nitrogen was determined by Kjeldahl's digestion with concentrated sulfuric acid ( $H_2SO_4$ ) as described by Jackson (1958). The distillation of the digest was done with 40% sodium hydro-oxide (NaOH) and the distillates were collected at 2% Boric acid mixed indicator. The distillates were titrated against sulfuric acid ( $H_2SO_4$ ). Total phosphorus of the samples were determined colorimetrically using a spectrophotometer at 490 nm by developing yellow color with vanadomolybdate after digesting the samples ((Jackson, 1973). Total potassium of the samples were determined from the digest by a flame photometer (Jackson, 1962). Total sulfur content of the samples were determined from digest by turbidity of suspended barium sulfate using Tween-80 stabilizer. The turbidity was measured by spectrophotometer at 420 nm as described by Page *et al.* (1989). Calcium, Magnesium, iron, manganese, zinc, chromium and sodium contents were determined by Atomic Absorption Spectrophotometer (AAS) as described in Jackson (1973).

*Collection, digestion and analysis of plant samples:* Collected plant samples were washed first with tap water and then with distilled water. The heights of the collected plants were noted down. The fresh weights of plants and pods were taken separately with an electric balance. The samples were kept one day for air dry. Then the samples were oven dried at  $80 \pm 5^\circ C$  for 48 hours. The oven dry weights of the samples were also taken. Then shoot and root were separated of each plant and grounded with mortar and pestle. The grounded samples were kept in envelopes with proper labeling and sorted in dry place.

Grounded shoot and pod were weighted 0.5 g separately and taken into 100 ml Pyrex beaker. Then all the weighted samples were mixed with 15 ml of conc. Nitric acid ( $HNO_3$ ) and left overnight for predigesting. The predigested samples were heated in a sand bath for about 15 minutes. Then the samples were cooled and 5 ml perchloric acid ( $HClO_4$ ) was added. Again samples were digested until the contents become colorless. The digests were cooled, filtered, diluted to 50 ml and transferred into dry plastic bottles (Jackson, 1973). This extract was used for further analysis.

Total nitrogen of the plant samples (shoot and pod) was determined by Kjeldahl's digestion with concentrated sulfuric acid ( $H_2SO_4$ ) as described by Jackson (1958). The distillation of the digest was done with 40% sodium hydro-oxide (NaOH) and the distillates were collected at 2% Boric acid mixed indicator. The distillates were titrated against sulfuric acid ( $H_2SO_4$ ). Total phosphorus of plant samples were determined colorimetrically using a spectrophotometer at 490 nm by developing yellow color with vanadomolybdate after digesting the plant samples (Jackson, 1973). Total potassium of plant samples was determined from the digest by a flame photometer (Jackson,

1962). Total sulfur content of plant samples was determined from digest by turbidity of suspended barium sulfate using Tween-80 stabilizer. The turbidity was measured by spectrophotometer at 420 nm as described by Page *et al.* (1989). Total calcium, magnesium, iron, manganese, zinc, chromium and sodium contents of plant samples were determined following standard method. From the value of nutrient content, nutrient uptake was calculated as: Nutrient uptake ( $\text{kg ha}^{-1}$ ) = Nutrient content (%)  $\times$  Dry weight ( $\text{kg ha}^{-1}$ )/100 (Huq and Alam, 2005).

The data collected in the experiment were calculated and the calculated results were graphically evaluated by using Microsoft excel (version 2013). Calculated results were statistically analyzed in the form one way ANOVA, using Minitab 17.

### Results and Discussion

The initial characteristics of the two types of soil samples and the two types of biofertilizer samples showed that, Acidic soil had a OC of 0.5 %, OM 0.95%, CEC 3.64 ( $\text{cmol}(+)/\text{kg}$ ), EC 3.17 mS/m, total N 0.22 %, total P 0.04 %, total K 0.07%, total S 0.33%, total Ca 0.01%, total Mg 1.33 %, total Na 0.27 %, total Zn 0.004%, total Fe 2.52%, total Mn 0.02% and alkaline soil had a OC of 0.41 %, OM 0.71%, CEC 2.22 ( $\text{cmol}(+)/\text{kg}$ ), EC 0.27 mS/m, total N 0.16 %, total P 0.07 %, total K 0.02%, total S 0.18 %, total Ca 0.02%, total Mg 0.13 %, total Na 0.05 %, total Zn 0.006%, total Fe 1.98 %, total Mn 0.1% (Table 1). Initial pH value revealed that vermicompost was acidic and trichocompost was alkaline in nature. Vermicompost had a OC of 3.86%, OM 6.64%, CEC 4.73 ( $\text{cmol}(+)/\text{kg}$ ), EC 2.76 mS/m, total N 1.03 %, total P 0.14%, total K 0.93%, total S 0.78 %, total Ca 0.87%, total Mg 0.55%, total Na 0.02%, total Zn 0.03%, total Fe 1.04%, total Mn 0.04%. Trichocompost had a OC of 4.08%, OM 7.01%, CEC 5.97 ( $\text{cmol}(+)/\text{kg}$ ), EC 2.96 mS/m, total N 0.76%, total P 0.12%, total K 0.72%, total S 0.62%, total Ca 2.69%, total Mg 0.46%, total Na 0.07%, total Zn 0.02%, total Fe 1.21%, total Mn 0.07%.

The result showed that, treatment with both biofertilizers in both acidic and alkaline soil took higher time duration to reach the maturity stage over the control (Table 2). This might be due to the initial pH value of vermicompost (6.58) and trichocompost (7.70) optimized the pH of both soils and made soils more suitable to grow with proper time duration. Similar results were reported by Hamidi *et al.*, 2009. Prolonged phenological stages due to biofertilizers application in present study was an indication of suitable condition and time available for plant growth and development. Comparing the results of both soil it can be said that, for vegetative growth stages both the biofertilizers showed almost similar results.

**Table 1. Initial properties of two types of soils and two types of biofertilizers.**

Properties	Acidic soil	Alkaline soil	Vermi-compost	Tricho-compost
pH	4.77	7.87	6.58	7.70
CEC (cmol(+)/kg)	3.68	2.22	4.37	5.97
EC (mS/m)	3.17	0.27	2.76	2.96
Organic carbon (%)	0.55	0.41	3.86	4.08
Organic matter (%)	0.95	0.71	6.64	7.01
Total Nitrogen (%)	0.22	0.16	1.03	0.76
Total Phosphorus (%)	0.04	0.07	0.14	0.12
Total Potassium (%)	0.07	0.02	0.93	0.72
Total Sulfur (%)	0.33	0.18	0.78	0.62
Total Calcium (%)	0.01	0.02	0.87	2.69
Total Magnesium (%)	1.33	0.13	0.55	0.46
Total Sodium (%)	0.27	0.05	0.02	0.07
Total Zinc (%)	0.004	0.006	0.03	0.02
Total Iron (%)	2.52	1.98	1.04	1.21
Total Manganese (%)	0.02	0.10	0.04	0.07

**Table 2. The effect of vermicompost and tricho-compost on phenology of okra.**

Growth stages	Code	Days after sowing (DAS)					
		<u>Acidic soil</u>			<u>Alkaline soil</u>		
		Control	Vermi	Tricho	Control	Vermi	Tricho
Germination	V0	0-6	0-10	0-10	0-5	0-9	0-8
Emergence	V1	6	10	10	5	9	8
Primary leaves	V2	9	14	13	8	13	11
First trifoliolate leaf	V3	17	31	29	17	25	24
Fifth trifoliolate leaf	V4	28	42	38	26	36	34
Pre-flowering	R5	44	52	49	41	54	49
Flowering	R6	48	58	55	47	61	56
Pod formation	R7	No pod	64	61	56	69	63
Pod filling	R8	No pod	73	67	64	76	71
Maturity	R9	No pod	85	83	82	88	86

Post-harvest soil samples were analyzed to evaluate the changes occurred in the physicochemical properties in different treatments (Table 3). In acidic soil, highest pH value was 6.40, total N 0.28%, total P 0.06%, total 0.28%, total Na 1375.20 ppm recorded in vermicompost applied soil, highest EC value was 1.70 mS/m, OC 0.67%, OM 1.15%, total Mg 1.44%, total Mn 294.49 ppm, total Zn 75.21 ppm recorded in trichocompost applied soil, highest total K was 2.01%, total Ca 0.03%, total Fe 2.57% recorded in control. In acid soil pH, EC, OC, OM total S, total Fe, total Mn, total Zn, total Na were differed statistically ( $p < 0.01$  and  $p < 0.05$ ). In alkaline soil, highest pH value was 8.12, CEC 5.17 was cmol (+)/kg, OC 0.65%, OM 1.22%, total K 1.78%, total S 0.26%, total Mg 1.35%, total Fe 2.19%, total Na 1269.68 ppm recorded in vermicompost applied soil, highest EC value was 0.24 mS/m, total N 0.29 %, total P 0.08 %, total Ca 0.06 %, total Zn 76.86 ppm recorded in trichocompost applied soil. Highest total Mn 228.50 ppm was recorded in control. In alkaline soil, OC, OM, total N, total P, total S, total Mg, total Fe, total Mn, total Zn, total Na differed statistically ( $p < 0.01$  and  $p < 0.05$ ). The present study indicates that in both acidic and alkaline soil, vermicompost and trichocompost applied soil showed increased values of pH, OC, OM, total N and almost all nutrients over the control. This might be due to the initial properties of both vermicompost and trichocompost added with soil. Similar results found by Azarmi *et al.*, 2008; Tharmaraj *et al.*, 2011.

The result revealed that, for both soils, significantly ( $p < 0.01$ ) higher plant height and root length were recorded with biofertilizers treated soils over the control soils (Table 4). The increased in plant height and root length in response to applied vermicompost and trichocompost may be due to release of nutrients, biological fixed nitrogen and also by excreting auxin, kinetins, vitamins. Similar results were found by Malik *et al.* (2005); Nuruzzaman *et al.* (2003); Prabhu *et al.* (2003).

The data presented in Table 5 revealed that, In alkaline soil, the values of total N, P, K, S, Ca, Mg uptake in shoot and pod were significantly ( $p < 0.05$  and  $p < 0.01$ ) higher in vermicompost and trichocompost treated soils than control. This might be due to the application of different biofertilizers which enhanced the CEC of soil and established better root system. In acidic soil, similar results found except for total N uptake in shoot and total Mg in pod, they showed higher values in trichocompost treated soil. Similar results were reported by Tanwar *et al.* (2003); Thenua and Sharma (2011).

The data presented in Table 6 revealed that, for both soils, the values of total Na and Zn uptake in shoot and pod were significantly ( $p < 0.01$  and  $p < 0.05$ ) higher in biofertilizers treated soils. This might be due to favorable soil condition and pH condition

Table 3. Chemical properties of post-harvest soil under different treatments.

Soil used	Treatment	pH	CEC (cmol (+)/kg)	EC (mS/m)	OC (%)	OM (%)	Total N (%)	Total P (%)	Total K (%)	Total S (%)	Total Ca (%)	Total Mg (%)	Total Fe (%)	Total Mn (ppm)	Total Zn (ppm)	Total Na (ppm)
Acidic soil	Control	5.57	5.44	0.53	0.51	0.87	0.25	0.05	2.01	0.25	0.03	1.40	2.57	204.50	66.80	1046.40
	Vermi-compost	6.40	4.86	0.89	0.66	1.13	0.28	0.06	1.69	0.28	0.02	1.15	2.28	178.43	56.47	1375.20
	Tricho-compost	5.90	5.44	1.70	0.67	1.15	0.27	0.05	1.98	0.27	0.02	1.44	2.41	294.49	75.21	1175.20
	P-value	<0.05	>0.05	<0.01	<0.01	<0.05	>0.05	>0.05	>0.05	>0.05	<0.01	>0.05	>0.05	<0.01	<0.01	<0.01
Alkaline soil	Control	7.82	3.29	0.20	0.51	0.90	0.24	0.07	1.14	0.16	0.05	0.10	1.73	228.50	74.81	359.60
	Vermi-compost	8.12	5.17	0.19	0.65	1.22	0.21	0.05	1.78	0.26	0.04	1.35	2.19	212.20	74.12	1269.68
	Tricho-compost	8.10	3.18	0.24	0.59	1.02	0.29	0.08	1.01	0.14	0.06	0.11	1.52	223.34	76.86	407.00
	P-value	>0.05	<0.05	>0.05	<0.01	<0.05	<0.05	<0.05	>0.05	>0.05	<0.01	>0.05	<0.01	<0.01	<0.01	<0.01

\*\*\*<0.01 = significant at 1 %, \*\*<0.05 = significant at 5 %, \*>0.05= not significant



**Table 4. Growth performance of *Abelmoschus esculentus* in response to different treatments.**

Soil used for growing plant	Treatment	Plant height (cm)	Root length (cm)	Leaf number per plant	Leaf area (cm <sup>2</sup> )	Pod number per plant
Alkaline soil	Control	28.30	8.50	4.00	18.02	3.00
	Vermicompost	35.70	12.20	6.00	18.92	4.00
	Trichocompost	30.60	11.80	5.00	18.12	4.00
<b>P-value</b>		<0.01	<0.01	>0.05	>0.05	>0.05
Acidic soil	Control	19.30	8.70	4.00	17.02	No pod
	Vermicompost	23.60	10.20	5.00	17.98	3.00
	Trichocompost	23.10	9.20	5.00	17.12	1.00
<b>P-value</b>		<0.01	<0.01	>0.05	>0.05	>0.05

\*\*\* <0.01 = significant at 1%, \*\*<0.05 =significant at 5%, \* >0.05 =not significant.

**Table 5. Macronutrients uptake by *Abelmoschus esculentus* under various treatments.**

Soil used	Treatment	Total N (kg/ha)		Total P (kg/ha)		Total K (kg/ha)		Total S (kg/ha)		Total Ca (kg/ha)		Total Mg (kg/ha)	
		Shoot	Pod	Shoot	Pod	Shoot	Pod	Shoot	Pod	Shoot	Pod	Shoot	Pod
Alkaline soil	Control	17.32	9.75	1.11	0.39	10.28	2.86	1.94	0.80	8.42	3.49	4.07	1.27
	Vermi	40.55	16.5	2.40	0.78	18.92	3.87	5.11	1.34	20.57	5.06	7.06	1.70
	Tricho	21.15	12.4	1.42	0.61	12.27	3.24	1.75	0.67	10.63	6.45	5.25	1.57
<b>P-value</b>		<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.05
Acidic soil	Control	9.88	No pod	0.42	No pod	4.52	No pod	0.53	No pod	2.62	No pod	9.63	No pod
	Vermi	15.60	9.82	0.90	0.43	7.23	3.07	1.39	2.62	4.84	2.43	4.84	1.11
	Tricho	28.34	3.14	0.76	0.23	7.31	1.59	0.60	1.59	4.31	0.93	4.31	1.29
<b>P-value</b>		<0.01	<0.01	<0.05	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01

\*\*\* <0.01 = significant at 1%, \*\*<0.05 =significant at 5%, \* >0.05 =not significant.

after treated with biofertilizers which enhanced nutrient availability and nutrient uptake as well as a better growth and activity of roots. In alkaline soil, there is no significant difference in Fe uptake in pods and Mn uptake in both shoot and pod. In acidic soil, there is no significant difference in Fe and Mn uptake in shoot. These might be due to lower Fe, Mn content of both biofertilizers and soils. Similar findings were observed by Idries and Snadhu (1979); Jagdale *et al.* (1980); Bera *et al.* (2013).

**Table 6. Micronutrient uptake by *Abelmoschus esculentus* under various treatments.**

Soil used	Treatment	Total Na (kg/ha)		Total Fe (kg/ha)		Total Mn (kg/ha)		Total Zn (kg/ha)	
		Shoot	Pod	Shoot	Pod	Shoot	Pod	Shoot	Pod
Alkaline soil	Control	0.25	0.07	0.48	0.05	0.02	0.006	0.26	0.08
	Vermi	0.41	0.11	0.34	0.05	0.04	0.009	0.35	0.10
	Tricho	0.19	0.08	0.07	0.01	0.02	0.02	0.15	0.07
<b>P-value</b>		<0.01	>0.05	<0.01	>0.05	>0.05	>0.05	<0.01	>0.05
Acidic soil	Control	0.09	No pod	0.17	No pod	0.02	No pod	0.09	No pod
	Vermi	0.25	0.06	0.17	0.05	0.03	0.007	0.16	0.06
	Tricho	0.23	0.03	0.14	0.03	0.02	0.003	0.11	0.02
<b>P-value</b>		<0.01	<0.05	>0.05	<0.05	>0.05	<0.05	<0.05	<0.05

\*\*\* <0.01 = significant at 1%, \*\*<0.05 =significant at 5%, \* >0.05 =not significant.

From the study, it can be stated that, the application of vermicompost appeared equally effective in both acidic and alkaline soil over the control on the growth of *Abelmoschus esculentus*. On the other hand, Application of trichocompost appeared more effective in acidic soil than alkaline soil.

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