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**Nutrient availability and proliferation of spinach in organic amended sandy loam soil**Rahat A. Chaity<sup>1</sup>, Ferdouse Z. Tanu<sup>1\*</sup>, Azizul Hakim<sup>2</sup>, M Hasinur Rahman<sup>1,3</sup> and Hafiz A. Haque<sup>4\*</sup><sup>1</sup>Department of Soil and Environmental Sciences, University of Barishal, Barishal 8254, Bangladesh.<sup>2</sup>Department of Soil Science, University of Chittagong, Chattogram 4331, Bangladesh.<sup>3</sup>Department of Primary Industries and Regional Development (DPIRD), Govt. of Western Australia, Northam, WA 6441, Australia.<sup>4</sup>Department of Coastal Studies and Disaster Management, University of Barishal, Barishal 8254, Bangladesh.

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**Abstract**

A comprehensive in- vitro pot incubation study was conducted to investigate the performance of widely used organic amendments, such as compost, vermicompost, charcoal, and newly used leaf powders of *Sesbania acculeata* and *Typha elephantina* on the proliferation of spinach in sandy loam soil. The results obtained from organic amendments were compared with those obtained from conventional fertilizers (urea, triple superphosphate, and muriate of potash). Leaves of *S. acculeata* and *T. elephantina* were found to be excellent nitrogen (N) providers (3.15% and 1.86%, respectively), with relatively low contents of phosphorous (P) and potassium (K). The spinach growing in vermicompost- amended soil presented the highest weights of both fresh and dry roots. Additionally, spinach presented the greatest degree of fresh shoot development, whereas the second highest degree of dry shoot development in *S. acculeata* leaf- amended soil. The newly used *T. elephantina* leaf amendment significantly improved soil fertility after 30 days of incubation. Tests of postharvest soils revealed that all the organic amendments used in the current study improved soil fertility and plant development far more than the control and traditional fertilizers did by increasing the organic content (OC) content, cation exchange capacity, and nutritional status, which is the ultimate criterion for sustainable agriculture.

**Keywords:** Available nutrients, *Sesbania acculeata* leaf, *Typha elephantina* leaf, Organic amendments, Sandy loam soil, Spinach proliferation

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**1. Introduction**

The sustainable development of agroecosystems is currently of vital importance. The degradation of agricultural soils and reduction in production are important challenges in agroecosystems. Farmers in Bangladesh frequently apply high quantities of agrochemicals in unplanned manners to increase the output of various field crops. Unplanned application methods and excessive use of these agrochemicals reduce soil fertility and pollute ecosystems. As a result of the cumulative nutrient imbalance [1], the farming method is deemed unsustainable. Increasing cropping intensity to accommodate a growing population's food demands has led to the extraction of natural plant nutrients from crop fields and resulted in a considerable decline in agricultural soil quality in Bangladesh over time [2,3]. A high-yielding crop production system cannot be sustained, except that soil nutrients are balanced with nutrient loss. The most reasonable strategy to increase total production from limited resources is to target high yields with high cropping intensity. Because of the detrimental effects of commercial fertilizers, there is a renewed interest in organic additions to ensure global agricultural sustainability [4].

The organic matter (OM) content in most of the soils of Bangladesh is alarmingly low (usually approximately 0.5-2%). This is due to the intensification of agriculture as well as the imbalanced use of chemical fertilizers with little or no use of organic fertilizers. Soil OM is critical and should be maintained at least at the 3% level for nutrient availability and plant productivity. The role of OM is most pronounced in low- fertility soils such as sandy

or sandy loam soils found in ‘char-lands’ in Bangladesh. Bangladesh's charlands are being intensively farmed to meet the country's food demand. However, due to the low fertility of the soil, the gross production of these lands is insufficient. Under these circumstances, to increase fertility and crop productivity, no alternative exists without adding OM to the soils. Organic amendments and inorganic fertilizers, combined with appropriate irrigation procedures, are needed to maintain crop productivity in sandy soils [5]. Organic additions often improve soil properties while preserving the soil moisture retention capacity, resulting in increased crop productivity and crop quality. Although organic amendments offer fewer nutrients for plants than commercial fertilizers do, the addition of growth-promoting hormones and enzymes is crucial for improving soil quality [6]. The application of effective amendments instead of chemical fertilizer to sandy or sandy loam soils may help farmers not only maximize yield but also combat the soil erosion process.

In developing countries such as Bangladesh, labour costs are low, but organic manures cost more than chemical fertilizers do. However, the cost of green manure (GM) may be low because of the extensive growth and availability of desired plants. Farmers in Bangladesh typically use compost, vermicompost, poultry manure, cow dung, vegetable waste, kitchen ash, tree litter, and crop residues as organic fertilizers on the basis of availability. GM is an eco-friendly leguminous or nonleguminous crop with potential for sustainable soil improvement and food grain production. GM is applied in two ways: by in- situ growth of GM crops or by the collection of green leaves and twigs from ex- situ grown plants. The most commonly used leguminous green manuring crops in Bangladesh are *S. aculeata* and *S. rostrata*, whereas the nonleguminous plant is sunflower. The residual nature of organic amendments such as compost (CP), vermicompost (VC), charcoal (CH) and plant leaves is of great interest for crop fields worldwide. However, the use of charcoal and *S. acculeata* leaves in powder form is not popular in Bangladesh. Evidence shows that *S. acculeata* leaf powder (SA) increases nutrient availability, enhances plant growth, and improves the soil nutrient status [7].

A nonleguminous plant, *Typha elephantina*, is a nonfood commercial crop that grows in tidal waterlogged areas in Bangladesh. Its leaves are used for various crafted materials, including mats and baskets, as well as for roof tops and wall partitions. Moreover, *Typha* sp. has been found to be an efficient nitrogen (N) accumulator from sediment [8] and contains more than 10% protein [9] and 1.68% dry wt. N in leaves [10]. Therefore, *T. elephantina* plant leaves are expected to be good organic amendments for Bangladeshi farmers. Nevertheless, no previous research has revealed the use of leaves of *T. elephantina* as an organic remedy for crop growth. In the present study, CP, VC, CH and plant leaf powder from the leguminous plant *Sesbania aculeata* (locally named ‘Prickly Sesban’) and the nonleguminous plant *T. elephantina* (locally named ‘Elephant- grass/ Indian Reed Mace’) were used as organic amendments for the growth of spinach. In this study, the use of *T. elephantina* leaf powder (TE) as an organic amendment represents a new attempt at pot experiments. It is hypothesized that the nutrient contents in TEs and the effects of TEs on soil chemical properties and spinach growth would be comparable to those of the five other studied amendments. Low-cost organic amendments can act as better alternatives to chemical fertilizers for sustainable agricultural production.

## 2. Materials and methods

### 2.1 Soil sampling site

An agricultural field from a village named ‘Charaicha’ of Barishal Sadar Upazila in the Barishal district in Bangladesh was chosen for soil sampling for the pot experiments. The coordinates of the sampling site are 23°32.460 N and 90°13.288 E. The sampled soil was from the Barishal soil series, a calcareous fluvisol belonging to the Ganges River floodplain, and was grouped as Typic Haplaquepts in the USDA soil subgroup. The field was poorly to moderately well drained and because getting seasonally flooded. The cropping pattern of the agricultural field was rice-vegetable-rice.

### 2.2 Soil collection and preparation

The soil samples were taken from 0 -15 cm depth according to the Soil Survey Staff of the USDA (United States Department of Agriculture). The soils were air dried and separated from visible plant residues. Large aggregates were broken and mixed thoroughly for a composite sampling. Approximately 500 g of soil was sieved through a 2 mm sieve, whereas a small amount of sample was passed through a 0.5 mm sieve and preserved for physical and chemical analysis. The remainder of each sample was saved for the plant growth response experiment.

### 2.3 Collection and preparation of amendments

#### 2.3.1 Compost and vermicompost

Commercially produced compost and vermicompost were collected from the local market of Barishal Sadar Upazila. The main ingredients of compost and vermicompost are cow dung, kitchen waste, and household waste. The collected compost and vermicompost were air dried, ground, and passed through a 0.5 mm sieve for chemical analysis and 2 mm sieve for plant cultivation.

#### 2.3.2 Charcoal

Wood residue is utilized for charcoal manufacture via traditional methods by local farmers. Wood waste was air dried, fired in a mud kiln, and pyrolyzed. Although a few air pockets were initially kept open to allow steam and smoke to escape, the kiln was mostly sealed to produce an oxygen-free environment. The prepared charcoal was powdered and passed through a 0.5 mm sieve for chemical analysis and a 2 mm sieve for plant cultivation.

#### 2.3.3 Plant leaves

Finely ground leaves of two plants, one leguminous (*S. acculeata*, locally named Prickly sesban), and one nonleguminous (*T. elephantina*, elephant grass), were used as amendments. Prickly sesban is a popular green manure crop, whereas elephant grass is a common plant on the coasts of Bangladesh. To the best of the authors' knowledge, the use of ground elephant grass leaves as an organic amendment in a pot experiment is practised for the first time with respect to the country as well as the world. Leaves of prickly sesban and elephant grass plants were collected at the age of 6 months from local farmers. Leaves were air-dried, ground, and sieved with a 0.5 mm sieve for chemical analyses and for use as organic amendments.

#### 2.3.4 Inorganic fertilizer

Commercially used fertilizers such as urea, triple superphosphate (TSP), and muriate of potash (MoP) were collected from the local market.

### 2.4 Laboratory analysis and analytical procedure

The soil particle size was analysed via the hydrometer method [11]. Marshall's triangle method was used to determine the textural groups of the soils [12]. The core method [13] was used to calculate the bulk density of each soil sample. The soil pH and pH of the compost, vermicompost and charcoal were determined with a pH meter (HACH instruments, PHC10101). The ratios of the soil samples, compost, vermicompost, charcoal, and plant leaves to distilled water as follows: soil:water = 1:2.5; compost:water = 1:5; vermicompost:water = 1:5; charcoal:water = 1:10; and plant leaf powder:water = 1:10. The electrical conductivity (EC) of the soil and amendments was determined via an EC meter (HACH HQ30D, port-CDC40101). The ratios of soil and amendments to distilled water were as follows: soil: water = 1:5 and amendments:water = 1:10. The ammonium acetate extraction method was used for the analysis of the cation exchange capacity (CEC) of soil, compost, vermicompost and charcoal at a sample-to-extraction ratio of 1:20 [14]. The Walkley and Black wet oxidation method [15] was used for the analysis of the organic carbon (OC) content in the soil.

For total nitrogen (TN) analysis of the soil, compost, vermicompost, charcoal, and leaves of *S. acculeata* and *T. elephantina*, samples were digested via wet oxidation by using concentrated sulfuric acid ( $\text{H}_2\text{SO}_4$ ) via the micro Kjeldahl method [16]. The nitric acid and perchloric acid digestion methods were followed for total phosphorus (TP) and total potassium (TK) analysis [14].

The samples were extracted with 1 M potassium chloride (KCl), and the steam distillation method with Devarda's alloy was used for the analysis of available N ( $\text{NH}_4^+ + \text{NO}_3^-$ ) [17]. Available P from soil was extracted via the Bray and Kurtz method [18] and the Olson method [19] and was estimated via the colorimetric method via a spectrophotometer (fixed at 880 nm). The available K of the samples was measured by a flame photometer (Jenway 500701 PFP7 Industrial Flame Photometer).

## 2.5 Experimental setup

All amendments were analysed for their physicochemical properties and nutrient contents. A total of 21 plastic pots (5×3 inches in size) were collected and prepared with soils and amendments at a rate of 10 t ha<sup>-1</sup> as recommended by the Soil Resource Development Institute (SRDI) of Bangladesh (Supplementary Table S1). The treatment used in the pots for nutrient availability and plant growth experiments are listed in Table 1. The experimental design was completely randomized design (CRD) with three replications.

**Table 1** Pot experimental set up with their symbol of respective amendments.

Arrangement of amendments	Symbol
Control	C
Fertilizer	F
Compost	CP
Vermicompost Soil	VC
Charcoal	CH
<i>S. acculeata</i> Leaves	SA
<i>T. elephantina</i> Leaves	TE

Each pot received 100 g of soil with 0.5 g of treatment mixture (amendments and fertilizers). The pots were incubated for four months. All the pots were placed in a randomized manner weekly in the incubation experiment. The pots were maintained at 50% saturated moisture content until the incubation period was over. After each incubation period of 30, 60, 90 and 120 days, one set of pots was separated, and the soil pH was determined immediately. The soil was subsequently air dried, ground, weighed, and stored for further analysis of available nutrients. Thus, the pH, OC, and available N, P and K of the amended soils were recorded after four incubation periods.

A leafy vegetable crop, spinach (*Basella rubra* Linn) was selected as an indicator plant and grown on amended sandy loam soil in 21 pots to assess the proliferation induced by the amendments. General information about the indicator crop is provided in supplementary Table S2. For growing spinach, each pot received 3 kg of soil and 15 g of each amendment at a rate of 10 t/ha. Healthy, plump, and large seeds of spinach were collected from a local market, sown in pots, and left in a dark chamber for germination. To evaluate the aftereffects of amendments with plant harvesting on soil chemical properties and nutrient availability, each type of amended soil was prepared as described in section 2.2 for pH, OC, CEC and available N, P and K analyses following the methods described in section 2.4. The chemical and physicochemical properties of the amended soils were analysed and compared with the respective values of nonamended soil.

## 2.6 Harvesting, preparation, and analysis of plant samples

The plants were uprooted manually from the pots after 120 days. The shoots and roots were separated and cleaned. The total number of leaves on each plant were counted. The average surface area (cm<sup>2</sup>) of each plant's leaves was measured. Fresh roots and shoots of each plant were weighed. The plant samples were dried in an oven at 65°C for 24 hours, after which the dried weights of the separated roots and shoots were determined. The concentrations of N, P, and K in the roots and shoots were analysed via the methods described in section 2.4. The protein contents of the roots and shoots were calculated by multiplying the % N by 5.7 [20]. The uptake of N, P and K by spinach was calculated by multiplying the concentrations of N, P and K in the plant (root + shoot) with their corresponding dry matter production. The uptake was expressed as mg pot<sup>-1</sup>.

## 3. Results

### 3.1 Characteristics of the soil and amendments

The physical and chemical properties of the soil are listed in Table 2, and those of the amendments are presented in Table 3. The soil pH was slightly alkaline (7.3).

**Table 2** Physical and chemical properties of the studied soil.

Soil properties	
Physical	
Sand (%)	60
Silt (%)	26
Clay (%)	14
Textural class	Sandy loam
Bulk density (g/cm <sup>3</sup> )	1.32
Particle density (g/cm <sup>3</sup> )	2.42
Chemical	
OC (%)	0.97
Total N (%)	0.182
Total P (%)	0.043
Total K (%)	0.063
Available N (mg/kg)	14.23
Available P (mg/kg)	2.30
Available k (mg/kg)	34.12
pH	7.30
EC (dS/m)	1.09
CEC (meq./100 g)	11.23

The pH of all the organic amendments was slightly alkaline to alkaline in nature (pH 7.3 to 9.3), among which charcoal had the highest pH and SA had the lowest pH. The relatively low electrical conductivity (EC) of all amendments indicates low soluble salt concentrations. VC presented the greatest EC and the highest CEC among all the amendments. The CEC of VC is nearly double than that of the most mineral soils ( $\leq 15$  meq. / 100g; Table 3). The lowest EC and CEC are found in the TE.

**Table 3** pH, EC, CEC, and total NPK contents of the organic amendments.

Amendments	Parameters					
	pH	EC (dS/m)	CEC (meq. / 100g soil)	TN (%)	TP (%)	TK (%)
CP	7.4	23	20.8	1.67 $\pm$ .01	1.21 $\pm$ .03	1.67 $\pm$ .03
VC	9.1	33	30.9	1.88 $\pm$ .03	1.37 $\pm$ .04	1.86 $\pm$ .00
CH	9.3	30	30.0	1.75 $\pm$ .01	1.45 $\pm$ .05	1.47 $\pm$ .01
TE	9.1	18	15.3	1.86 $\pm$ .02	0.41 $\pm$ .02	0.81 $\pm$ .03
SA	7.3	24	15.6	3.15 $\pm$ .03	0.32 $\pm$ .00	0.52 $\pm$ .01

The total contents of N, P and K in the organic amendments ranged from 1.67 to 3.15%, 0.32 to 1.45%, and 0.52 to 1.86%, respectively. The highest contents of N, P and K were detected in SA, CH, and VC, respectively, whereas the lowest content of N was detected in CP, and both P and K were detected in SA. However, the contents of both P and K were considerably higher in TE than in SA.

### 3.2 Effects of amendments and incubation days on soil chemical properties

The effects of amendments and incubation periods on soil pH, OC, and nutrient availability are presented in Figure 1. The initial pH of the soil (7.3) decreased after 30 days and 60 days of incubation for all the treatments, including the control. However, the pH of all amended soils except those in the C and F treatments further increased after 90 days and 120 days of incubation (Figure 1A). The application of all the organic amendments and fertilizer resulted in an increase in the soil OC content after 30 days of incubation and a gradual decrease throughout the next incubation period (Figure 1B). The control soil also presented the lowest OC content, which gradually decreased throughout the entire incubation period. At final stage of incubation after 120 days, the order of relatively high OC content in the treatments was TE>CH>SA>VC>CP>F>C.

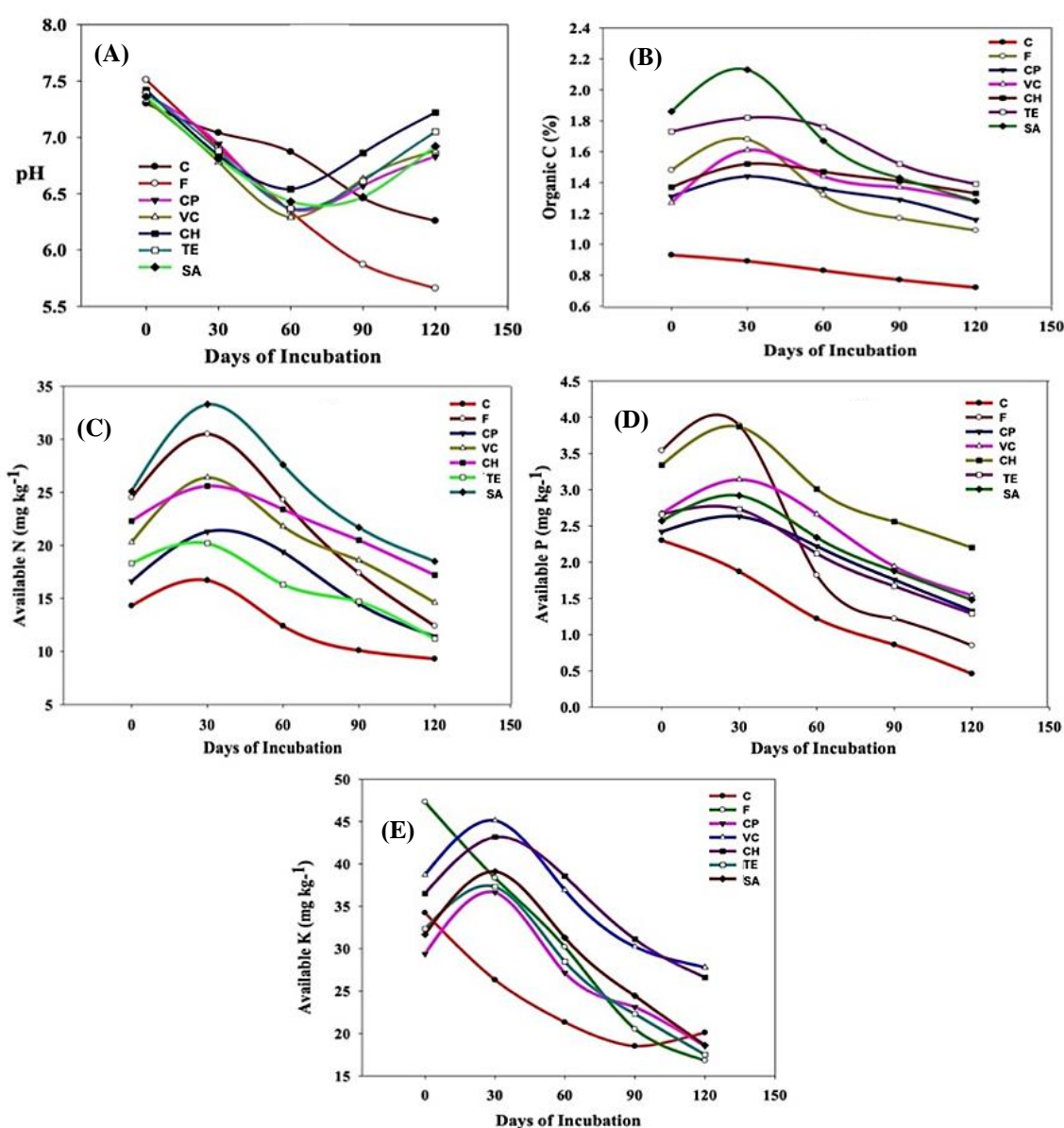
Additionally, all the treatments except the control presented an increase in available N and P after the first 30 days of incubation, and a decreasing trend during the following incubation periods (Figure 1C and 1D). The order of the available N content of the treatments after 30 and 120 days of incubation was SA>F>VC>CH>CP>TE>C and SA>CH>VC>F>CP>TE>C, respectively, whereas the order of the available P content was F>CH>VC>SA>TE>CP>C and CH>VC>SA>CP>TE>F>C, respectively. In the case of available K, the control and F treatments gradually decreased over the four incubation periods, whereas all the other treatments initially increased but then decreased after the final stage of incubation (120 days) (Figure 1E). Additionally, the order of the K availability of the treatments after 30 and 120 days of incubation was VC>CH>SA>F>TE>CP>C and VC>CH>C>SA>CP>TE>F, respectively.

### 3.3 Macrocosm study

A macrocosm study was conducted to ensure the effects of the laboratory incubation performance of the various treatments on plant proliferation and soil properties. The germination rates of the collected spinach seeds are listed in supplementary Table S3. The growth and visual appearance of the indicator plants were observed and are presented in Table 4.

The growth of spinach was equally better in fertilized and organic amended soils than in the control. However, plant performance in terms of root and shoot growth and biomass was better in response to organic amendments in the soil. The order of the treatments causing greater leaf number was SA>F>CH>VC>CP>TE>C, whereas that causing greater leaf surface area was SA>F>VC>CP>TE>CH>C (Table 4). The leaf number and surface area varied significantly ( $p<0.01$ ) among the treatments but were not significantly different among the treatments.

The root and shoot lengths of spinach ranged from 8.14 to 16.06 cm and from 10.11 to 23.65 cm, respectively. The order of treatment for greater root length was VC>SA>CH>CP>F>TE>C, whereas the order of shoot length was SA>CH>VC>CP>F>TE>C (Table 4). All the treatments significantly increased ( $p<0.01$ ) both the fresh and dry weights of the roots and shoots of spinach. Among all the treatments, the VC-treated plants presented the highest weights of both fresh and dry roots. However, the SA-treated plants presented the highest weight of fresh shoots (45.71g) and the CH-treated plants presented the highest weight of dry shoots (4.51 g).



**Figure 1** Changes in pH (A), OC (B), availability of nitrogen (C), phosphorus (D), and potassium (E) in amended soils after incubation.

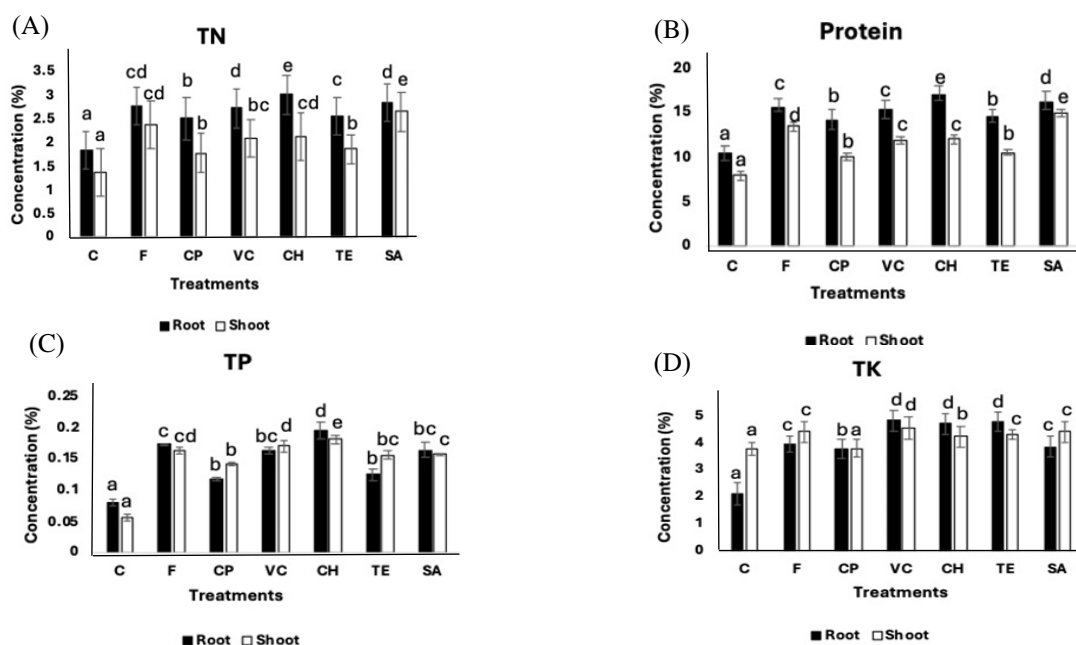
**Table 4** Growth parameters of *Basella rubra* (Indian spinach) under different treatments.

Treatments	Leaf No. (per pot)	Leaf surface area (cm <sup>2</sup> )	Length (cm)		Fresh weight (g pot <sup>-1</sup> )		Dry weight (g pot <sup>-1</sup> )	
			Root	Shoot	Root	Shoot	Root	Shoot
C	6.44 <sup>a</sup>	40.19 <sup>a</sup>	8.14 <sup>a</sup>	10.11 <sup>a</sup>	3.1 <sup>a</sup>	20.12 <sup>a</sup>	0.60 <sup>a</sup>	2.81 <sup>a</sup>
F	11.21 <sup>e</sup>	105.42 <sup>e</sup>	10.01 <sup>b</sup>	17.3 <sup>cd</sup>	4.5 <sup>bc</sup>	32.40 <sup>c</sup>	1.40 <sup>b</sup>	3.39 <sup>c</sup>
CP	9.41 <sup>c</sup>	84.62 <sup>c</sup>	10.03 <sup>b</sup>	16.21 <sup>c</sup>	3.5 <sup>a</sup>	23.20 <sup>b</sup>	0.86 <sup>a</sup>	3.06 <sup>ab</sup>
VC	10.64 <sup>d</sup>	96.45 <sup>d</sup>	16.06 <sup>d</sup>	18.32 <sup>d</sup>	5.1 <sup>c</sup>	31.81 <sup>c</sup>	1.72 <sup>c</sup>	3.33 <sup>bc</sup>
CH	10.71 <sup>d</sup>	74.29 <sup>d</sup>	13.12 <sup>c</sup>	22.05 <sup>e</sup>	4.2 <sup>b</sup>	37.12 <sup>d</sup>	1.55 <sup>bc</sup>	4.51 <sup>d</sup>
TE	8.56 <sup>b</sup>	74.50 <sup>b</sup>	9.05 <sup>ab</sup>	13.24 <sup>b</sup>	3.4 <sup>a</sup>	20.81 <sup>a</sup>	0.81 <sup>a</sup>	2.92 <sup>ab</sup>
SA	13.31 <sup>f</sup>	145.71 <sup>f</sup>	15.67 <sup>d</sup>	23.65 <sup>f</sup>	4.8 <sup>bc</sup>	45.71 <sup>e</sup>	1.58 <sup>bc</sup>	4.25 <sup>d</sup>
Level of significance	0.05	0.05	0.01	0.01	0.01	0.01	0.01	0.01

In addition, all the treatments resulted in better yields than did the control. Both the fresh and dry weights of roots and shoots varied significantly with treatment ( $p < 0.01$ ). The dry weights of roots and shoots increased with treatment. The sequences of treatments for both fresh root weight and dry root weight were VC>SA>F>CH>CP>TE>C. On the other hand, the fresh shoot weight followed the order SA>CH>VC>F>CP>TE>C, and the dry shoot weight followed the order CH>SA>F>VC>CP>TE>C. The increase in the dry weights of both the roots and shoots in the different treatments was also significant ( $p < 0.01$ ).

### 3.4 Total contents of NPK and protein in spinach

The total contents of N, P, K and protein in both the roots and shoots (stem + leaves) are presented in Figure 2. The contents of TN (Figure 2A) and protein (Figure 2B) in the roots and shoots of all the spinach treatments were greater than those in the control. Compared with those in the roots, the contents of TN and protein in the shoots were lower (Figure 2A and 2B). The CH-treated plants presented the greatest TN (3.01%) in the roots followed by the SA-treated plants (2.84%), whereas the lowest TN (1.83%) was detected in the control (C) plants. On the other hand, the SA-treated plants presented the highest TN (Figure 2A) and protein (Figure 2B) contents in the shoots. The TN and protein contents of both the roots and the shoots significantly differed among the treatments ( $p < 0.01$ ). ANOVA indicated significant effects of the treatments on TN ( $p < 0.01$ ).

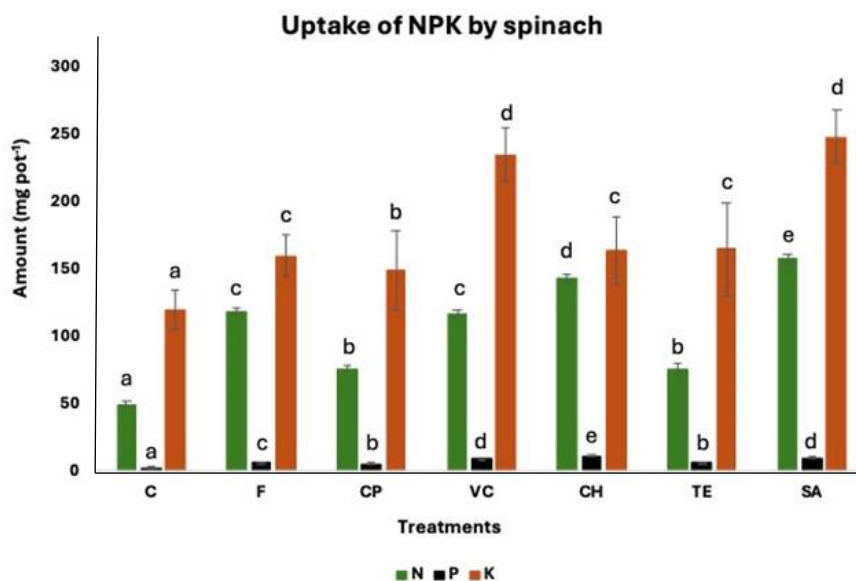


**Figure 2** Total contents of nitrogen, N (A), protein (B), phosphorus, P (C), and potassium, K (D) in spinach treated with fertilizer (F), compost (CP), vermicompost (VC), charcoal (CH), *T. elephantina* leaf powder (TE), *S. acculeata* leaf powder (SA), and the control (C). Different letters in each graph indicate significant differences in each plant part (root or shoot) among treatments, and a similar letter indicates nonsignificant variation in DMRT.

Additionally, among all the treatments the highest accumulation of P in both the roots and the shoots of CH-treated spinach was observed (Figure 2C). The order of treatments for higher TP content in roots was CH>F>SA>VC>TE>CP>C, whereas in shoots, the order was CH>VC>SA>TE>F>CP>C. ANOVA indicated that there were significant effects of the treatments on the TP content ( $p<0.01$ ). The concentration of TP varied significantly across the treatments. The order of the treatments in terms of the TK content of the roots was VC>CH>TE>SA>F>CP>C, and that of the shoots was VC>F>SA>CH>TE>CP>C. The greatest amount of K in both roots and shoots was found in the VC-treated plants.

### 3.5 Uptake of NPK by spinach

Nutrient uptake by spinach was calculated to understand the effects of different amendments on the phytoavailability of nutrients. The uptake of NPK is presented in Figure 3.



**Figure 3** Uptake of nitrogen (N), phosphorus (P), and potassium (K) by spinach subjected to different fertilizer (F), compost (CP), vermicompost (VC), charcoal (CH), *T. elephantina* leaf powder (TE), *S. acculeata* leaf powder (SA), and control (C) treatments. For each colored bar, different letters indicate significant differences among treatments according to DMRT.

The order of N uptake by spinach was SA>CH>F>VC>CP>TE>C. ANOVA revealed significant effects of the various treatments on N uptake ( $p<0.01$ ) by spinach. The order of the treatments causing greater P uptake by the plants was CH>SA>VC>F>TE>CP>C, whereas charcoal (CH) caused the greatest amount of P uptake by the plants. ANOVA indicated that there were significant effects of the various treatments on P uptake by plants ( $p<0.01$ ). Although, the greatest amount of K in both roots and shoots was detected in VC-treated plants, the maximum uptake of K occurred in SA-treated plants with the maximum dry weight of biomass (248.5 mg/pot). The order of the treatments with the greatest amount of K uptake was SA>VC>TE>CH>F>CP>C. The TK content varied significantly with treatment. ANOVA indicated that there were significant effects of the treatments on the TK content ( $p<0.01$ ) and uptake by the plants ( $p<0.01$ ). The lowest uptake was found in the control (119.1 mg /pot).

### 3.6 Effects of treatments and plant growth on soil properties

When the post-harvest soil properties were compared with their initial values, the pH of the amended soils, with the exception of the fertilized soils, decreased, whereas the OC content increased slightly (Table 5). The available N and K contents of all organic matter amended postharvest soils increased from the initial values. In the case of CEC and available P, all amended postharvest soils, including the control, presented higher values than the initial (Table 5) values did. The CH-amended soil presented the greatest increase in OC, available P and K, whereas the SA-amended added soil presented the highest values of available N and CEC. However, the amendments had statistically insignificant effects on the soil properties.



**Table 5.** Comparison between initial values of soil parameters with those after the effects of treatments and crop harvesting.

Treatments	Soil pH		OC (%)		Available N (mg/kg)		Available P (mg/kg)		Available K (mg/kg)		CEC (meq./100g soil)	
	Initial	After	Initial	After	Initial	After	Initial	After	Initial	After	Initial	After
C		7.1 <sup>a</sup>		1.6 <sup>b</sup>		10.3 <sup>a</sup>		2.7 <sup>a</sup>		33.3 <sup>a</sup>		13.67 <sup>a</sup>
F		7.5 <sup>b</sup>		0.9 <sup>a</sup>		13.2 <sup>b</sup>		2.8 <sup>a</sup>		33.5 <sup>a</sup>		13.11 <sup>a</sup>
CP		7.2 <sup>ab</sup>		1.9 <sup>c</sup>		17.3 <sup>c</sup>		3.7 <sup>b</sup>		35.1 <sup>b</sup>		13.56 <sup>a</sup>
VC	7.3	6.9 <sup>a</sup>	0.97	1.8 <sup>c</sup>	14.3	18.7 <sup>d</sup>	2.3	3.9 <sup>b</sup>	34.2	37.9 <sup>c</sup>	12.83	13.87 <sup>a</sup>
CH		7.1 <sup>a</sup>		2.5 <sup>d</sup>		22.4 <sup>e</sup>		4.5 <sup>bc</sup>		39.6 <sup>d</sup>		13.93 <sup>a</sup>
TE		7.1 <sup>a</sup>		2.3 <sup>cd</sup>		16.2 <sup>c</sup>		3.3 <sup>ab</sup>		37.1 <sup>c</sup>		13.45 <sup>a</sup>
SA		6.9 <sup>a</sup>		2.1 <sup>cd</sup>		26.5 <sup>f</sup>		3.9 <sup>b</sup>		35.8 <sup>b</sup>		14.18 <sup>b</sup>
Coefficient of variation (%CV)		2.86		25.98		30.59		18.28		6.42		2.57

Different letters in each column show significant differences among treatments and the similar letter with the respective data indicates nonsignificant variation at 1% level as per DMRT.

#### 4. Discussion

This study revealed that five types of organic amendments—compost (CP), vermicompost (VC), charcoal (CH), green leaf powder of *S. acculeata* (SA), and *T. elephantina* (TE) were able to increase nutrient availability in sandy loam soil and promote the proliferation of the vegetable crop Indian spinach. The performance of organic addition was typically superior to that of commercial fertilizer.

##### 4.1 Effects of organic amendments and incubation periods on soil quality

Although all the organic amendments were neutral to alkaline in nature (Table 3), the pH of the organic amended soils slightly decreased from the initial value after 120 days of incubation (Figure 1A). All the pH values (6.8 - 7.3) were within a suitable range for agriculture; however, the control (pH 6.4) and fertilized (pH 5.7) soils were slightly acidic. The pH decreased sharply from 0 to 120 days of incubation in fertilized soil, possibly because of the conversion of ammonium nitrogen to nitrate nitrogen through nitrification, the loss of nitrogen by leaching and the uptake of ammonium nitrogen by plants, leading to soil acidification [21]. The conventional fertilizer contains fast-release nutrients, whereas the organic amendments have slow-release nutrients. Compared with OM, plant litter has a faster decomposition rate because of its low C/N ratio [22]. Increasing the efficacy of nitrogen accelerates the OM decomposition rate and the release of various organic acids in soil. Simultaneously, N input also inhibits the decomposition rate of soil OC by decreasing microbial activity and encouraging the formation of aggregates [23]. The net effect of the mineralization of soil organic N is the depletion of H<sup>+</sup>. This is mainly due to the decomposition of organic acids, emanation of CO<sub>2</sub> and mobilization of base cations [24]. Accordingly, the oxidation of organic matter, and the formation of both inorganic and organic acids during incubation might cause the pH to decrease in organic-amended soils after 30 and 60 days of incubation and again increase after 90 and 120 days of incubation. However, the overall pH of organic-amended soils decreased. The response of the soil to the addition of amendments mainly depends on the type, quality, and quantity of amendments added and the intrinsic soil properties, particularly CEC [25].

With the two highest contents of EC and CEC (Table 3), VC and CH could be two significant organic amendments for sandy loam soil. Different components, such as K, Ca, Mg, Na, and P, in biomass enhance the development of O-containing groups on the surface of VC and CH during pyrolysis and composting, resulting in an increase in the CEC [26]. On the other hand, with the lowest contents of EC and CEC, the newly used organic amendment TA (18 mds) is superior to many other organic amendments [21].

The soil OC content increased in each amended soil after incubation for 30 days but decreased after 120 days (Figure 1B). Nevertheless, the final content of OC in the-amended soils was greater than that in the fertilized soils. The initial increase in OC during incubation can be explained by initial microbial biomass production, whereas a further decrease may occur for the aerobic decomposition of organic matter. However, long-term field observations have been shown that soil OC does not necessarily increase after the incorporation of a large amount of plant litter [27]. The dose [21], chemical composition [28], and surface properties [29] of organic amendments generally affect the soil organic matter content.

The nutrient contents of the organic amendments varied significantly ( $p < 0.05$ ) throughout the incubation period. SA had the greatest N%, with the lowest P% and K%. Another green leaf amendment, TA, also had significantly higher N% than CH and CP. However, all the treated soils, including the control, presented increased N availability after an initial incubation of 30 days but decreased N availability at the final stage of incubation of

120 days (Figure 1C). The sharp decrease in available N in fertilized soil throughout the incubation period indicates rapid mineralization and a low nutrient absorption capacity of conventional fertilizers. Among the organic amendments, SA-amended soil presented the highest content of available N, followed by CH- and VC-treated soils. The leaves of *S. acculeata* may contain materials with high N contents and low lignin contents [30], which can be attributed to easy decomposition and substantial release of mineral N [31]. Despite having a high N content (3.15%), SA might have quick mineralization capacity because of the presence of simple phenolic compounds. This finding also indicates that plant residue in powder form consumes less time to decompose than does that in the litter form. In addition, despite having a TN content of 1.86% in TA, TA-amended soil presented the lowest amount of available N (11.2 mg/kg). The reason might be the presence of more lignin, complex phenolic acid compounds and recalcitrant carbon in *T. elephantina* leaves, which decreases the N mineralization rate [32]. After biochar application, both an increase in N availability due to a decrease in leaching loss of nitrogenous compounds [33] and decrease in N availability due to changes in organic N turnover [34] have been reported in the literature.

Furthermore, organic matter mineralization may cause an initial increase in P availability in organically modified soils (Figure 1D) [35]. An increase in P availability may occur since organic molecules compete with adsorbed P to be fixed on the soil surface and release P in the soil solution [35]. The initial decrease in pH in fertilized soil after 30 days of incubation (Figure 1A) may release aluminium and iron-sorbed P by releasing H<sup>+</sup> ions, resulting in increased P availability. However, all amended soils presented reduced P availability after 120 days of incubation and the sharpest reduction was found in fertilized soil. This may be due to the rapid mineralization and use of fertilizer by the microbial biomass. OM application increases microbial biomass [36], which further uses soil P and decreases P availability over time. The CH-amended soil presented the highest P availability, whereas the CH-amended soil presented the highest TP content (1.45%). On the other hand, the TA-treated soil presented a significantly greater content of available P than did the control and commercial fertilizer-treated soils, while the TA-treated soil presented a TP content of 0.41%. Organic amendments with reduced exchangeable acidity and aluminium and iron contents act as soil pH buffers offering favorable environments for P availability in soil [37]. However, the overall effects of the treatments on P availability were not statistically significant in this study.

Organic amendments have the most prominent influence on K release [21]. The fertilizer treatment sharply decreased throughout the incubation period, although the maximum K content (47.31 mg/kg) initially occurred. The VC-amended soil presented the highest content of available K after 120 days (27.8 mg/kg), followed by the CH-amended soil (26.6 mg/kg). These values were significantly different ( $p < 0.05$ ) from those of the other organic-amended soils. The average K availability of the VC-amended soil was almost double than that of the untreated-soil, which confirms that vermicompost was the best amendment for K needs [38]. Earthworms are capable of uniformly grinding and mixing the minerals in VC, and plants can easily obtain nutrients from VC. The worm mucus present in VC also helps to increase nutrient content. The various organic amendments significantly ( $p < 0.01$ ) affected the phytoavailability of K.

#### 4.2 Indian spinach growth under the effects of organic amendments on sandy loam soil

The patterns of nutrient distribution among leaves, stems, and roots indicate the ability of plants to store and transport nutrients. The results of this study revealed that among the five organic amendments, SA had the most prominent effect on the proliferation of spinach in sandy loam soil. The greatest number of leaves, leaf surface area, shoot length, and maximum weight of fresh shoot and shoot biomass (including leaves) were observed in the SA-amended plants (Table 4). Furthermore, VC treatment promoted the proliferation of spinach in terms of root length, fresh and dry weights of roots, and fresh and dry weights of root biomass. Additionally, the influence of SA and the newly used TE on growth parameters was explored first in this study. Compared with the control treatment, the treatment improved spinach growth but resulted in a shorter plant height than did the fertilized plants. The reason may be the excess amount of lignin and phenolic acid compounds present in the TE [39], which cause slow decomposition and slow release of nutrients. The value of shoot length per plant varied significantly with respect to the control treatment ( $p < 0.01$ ), but the variations were insignificant among the treatments.

#### 4.3 Phytoavailability of nutrients by spinach under the effects of organic amendments

Compared with the control, all amendments significantly promoted nutrient concentration and nutrient uptake by spinach (Figure 2). The maximum N concentration and N uptake in shoots were detected in SA-treated plants (Figure 1A). These results corroborate those of Sajjad et al. [7], who reported the highest N uptake in wheat crops when SA powder was used. The steady status of nutrients is maintained through biological processes, which significantly reduces the chances of N volatilization or leaching. The highest root N and the second-highest N uptake by spinach were found in the CH-amended plants in this study. Additionally, CH-amended plants presented the highest P contents in roots and shoots and P uptake by spinach. These findings are consistent with recent work

by Varela et al. [40]. The roots and shoots of the control plants presented a minimal P level that could be taken up from the reserve soil P. Furthermore, the VC-treated plants accumulated the greatest amount of K in both the roots and the shoots. Unlike chemical fertilizers, it is difficult for VC to drain from the soil because of the worm mucus it contains. Like fertilizer, TE and SA also resulted in significantly high K contents in roots and shoots. The control group presented little K in the roots and shoots, indicating the capacity of the soil to release reserve K.

#### 4.4 Changes in soil properties resulting from organic amendments and spinach growth

The physicochemical properties of the postharvest sandy loam soils were significantly altered by amendment application. All organic-amended postharvest soils presented a decrease in pH, possibly due to the release of organic acids during decomposition and organic C inputs through root biomass and leaf litter. Generally, the effects of compost, manure, or other organic matter, such as leaf litter, may increase [41] or decrease [42] the soil pH, which depends on organic residues and the initial pH of soils [43]. Amendments have a nonsignificant positive effect on organic C, which may be due to the discharge of root exudates. Organic matter has a large surface area because of its colloidal properties when decomposed. The significant increase in the CEC of the organic-amended postharvest soils was probably due to the high Na, K, Ca, and Mg contents in the amendments. The findings of the present study revealed a significant increase in N and K availability in the postharvest organic-amended soils compared with the control, whereas an increase in P availability was detected in all the amended-soils, including the control.

The charcoal (CH)-amended soil had the greatest amount of OC, available P and K, whereas the SA-amended soil had the greatest amount of available N and CEC among all the organic-amended soils. However, the TE-treated postharvest soils also presented significantly greater CEC and available NPK values than did the fertilized postharvest soils. Furthermore, the OC content of TE-amended postharvest soil was greater than that of F-, CP-, VC- and SA-amended postharvest soils. These findings indicate the potential use of TEs as organic amendments for the growth of vegetable crops such as spinach. The fertilizer releases nutrients quickly and is present in the soil for a relatively short period, whereas the organic amendments provide an adequate environment for essential nutrient availability for long-term plant growth.

## 5. Conclusion

In this study, organic amendments included compost, vermicompost, charcoal, and green leaf powder from *S. acculeata* and *T. elephantina* plants. Compared with the control, all the organic additions increased the nutrient availability of the sandy loam soil and the proliferation of vegetable crop spinach. There is a clear conclusion on the improvement in soil quality with respect to the parameters of pH, EC, CEC, OC, and availability of NPK during the incubation study due to the combined effects of organic amendments and plant growth. In this study, vermicompost, charcoal and leaf powder of *S. acculeata* performed better than *T. elephantina* leaves as organic amendments; however, the use of *T. elephantina* leaf powder as an organic amendment is considered promising with respect to its availability and cost. There is an urgent need to investigate the functional and surface features of TA leaves for future use as organic amendments in soil and address site-specific concern for sustainable agricultural production. Additionally, a field study is needed for cost-benefit analysis and to determine whether effective production of *T. elephantina* is possible. The effectiveness of *T. elephantina* leaf application as an organic amendment may depend on the cropping systems, crop variety, soil type and agroecological region.

## 6. References

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#### Supplementary information

**Table S1** Recommended fertilizer dose for Pui Shak by Soil Research and Development Institute, Bangladesh

Serial No.	Nutrient element	Fertilizer	Dose (per ha)	Fertilizer application time
1	Nitrogen	Urea (If TSP is used)	663 g	-
	or Nitrogen	Urea (If DAP is used)	541 g	-
2	Phosphorus	TSP	314 g	During bed preparation
	or Phosphorus	DAP	314 g	During bed preparation
3	Potassium	MOP	247 g	During bed preparation

**Table S2** Information about the Indicator crop.

Name	Indian Spinach
Local name	Pui Shak
Scientific name	<i>Basella rubra</i> Linn
Purity	98%
Germination rate	80%
Seed rate	5 kgha-1

**Table S3** Result of germination of Pui seed with unsieved and sieved amendments

Amendments	Days of germination	
	Unsieved	Sieved
<b>Compost</b>	15	7
<b>Vermicompost</b>	10	4
<b>Charcoal</b>	30	6
<b>Hogol leaves powder</b>	Not germinated*	7
<b>Dhaincha leaves powder</b>	Not germinated*	5

\*The pots were observed for 3 months to notice the germination of seeds. But hogol and dhaincha leaves powder couldn't germinate at unsieved condition. Hence, it was concluded as not germinated.