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### Influence of foliar application with *Moringa oleifera* residue fertilizer on growth, and yield quality of leafy vegetables

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#### KEYWORDS

Biofertilizer

*Moringa oleifera*

Lettuce

Mustard spinach

Organic farming

#### ABSTRACT

Biofertilizers produced from organic materials help to promote the growth, and yield quality of crops and is more environmentally friendly than chemical fertilizers. *Moringa oleifera* is a leafy vegetable whose leaves are also used to make biofertilizers. The use of moringa non-edible parts in biofertilizer preparation remains under-explored. In this study, a procedure to produce moringa foliar biofertilizer (MFB) from non-edible parts was developed. The effect of composting time (3 to 4 months) on the quality of MFB was investigated, and four-month incubation was found suitable for biofertilizers yield with the highest nitrogen content and optimal pH. Furthermore, the influences of MFB doses (20 to 100 mL per Litre) on the growth of lettuce and mustard spinach were studied. The yield of these leafy vegetables was the highest at 100 mL per Litre of MFB spray. Finally, MFB was compared with other commercial foliar sprays, including chitosan fertilizer and seaweed fertilizer. Each foliar treatment was applied every five days until five days before harvest. Plant height, the number of leaves, canopy diameter, leaf area index, actual yield, ascorbic acid content, and Brix were found to be similar in lettuce sprayed with MFB, chitosan, and seaweed fertilizers. In conclusion, the application of MFB promoted the growth and yield of mustard spinach.

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## 1 Introduction

Safe and clean vegetables are important for human health and well-being. Biofertilizers (organic fertilizers) are essential for the production of safe leafy vegetables. Furthermore, the use of biofertilizers helps to protect the environment from soil degradation and groundwater pollution. One of the biofertilizers which are widely investigated for their potential of improving plant yield and growth is moringa leaf extract, produced from *Moringa oleifera* (Zulfiqar et al. 2020; Karthiga et al. 2022). Previous studies demonstrated that moringa leaf extracts increase the growth and yield of various plants such as pepper (Matthew 2016), tomatoes (Culver et al. 2012), and maize (Biswas et al. 2016).

*Moringa oleifera* is a fast-growing softwood species grown in tropical and subtropical regions. Moringa is mainly cultivated for its leaves which are consumed as a vegetable (Price 2007). Recently, aqueous extracts of different parts of moringa (leaves, seeds, and roots) have been used to produce agricultural products. Its aqueous extract reduces the reproduction and galling of root-knot nematodes, and helps to improve plant growth and yield parameters of pea plants (Youssef and El-Nagdi 2021). Moringa leaf and seed extracts are also effective in extending the shelf-life of cut rose flowers (Hassan et al. 2020). Although moringa leaf extract is extensively studied, but the production of moringa biofertilizer and its impact on vegetable growth still remained under-explored. Therefore, this study was carried out to investigate the effect of moringa foliar biofertilizer on the growth and quality of leafy vegetables.

## 2 Materials and methods

### 2.1 Experimental site and planting materials

The study was conducted at the experimental field of the Institute of Biotechnology, Hue University (Hue, Vietnam) from October 2019 to March 2021. This region has a humid tropical climate with average temperatures varying from 26 to 35 °C. In the study, a yellow lettuce (*Lactuca sativa* L.) variety obtained from Phu Nong Seeds company and a mustard spinach (*Brassica juncea*) variety obtained from Ha Noi Xanh company were used.

### 2.2 Moringa foliar biofertilizer (MFB) preparation

Moringa foliar biofertilizer was prepared following the non-aerated process. Briefly, 70 kg of moringa residues (including stems, branches, and leaf petioles) were washed with water to remove dust particles before being chopped into small parts. In a 100-liter container, the chopped moringa residues were spread to form a 20-cm layer. Second, molasses (5 L) and effective microorganism (EM) products (0.2 kg) were subsequently added to the top of the layer. The container was filled with chopped materials and water

was added to 2/3 of the container. The container was then tightly covered. The mixture in the container was stirred once every month until the end of the composting period (three to four months). The extract was collected and filtered. The obtained fertilizer was kept in an airtight container.

### 2.3 Effect of composting time on the quality of MFB

To evaluate the effect of composting time on the quality of MFB, the residue was incubated for either 3, 3.5, or 4 months. Physicochemical properties of the extract including the percentages of nitrogen (N), phosphorus (P), phosphorus pentoxide (P<sub>2</sub>O<sub>5</sub>), potassium (K), potassium oxide (K<sub>2</sub>O), and organic matter (OM) were determined.

### 2.4 Effect of different doses of MFB on growth, yield, and quality of leafy vegetables

Three to four leaf plants in a 10 m<sup>2</sup> plot were sprayed with either 100 mL, 50 mL, 33.3 mL, 25 mL, or 20 mL of MFB diluted in 1 L of water (Nwokeji et al. 2022). MFB has sprayed every five days intervals until five days before harvest. The experiment was designed in a Completely Randomized Design (CRD) with five fertilizer doses and three replicates per treatment.

### 2.5 Effect of different foliar fertilizers on growth, yield, and quality of leafy vegetables

Three to four leaf plants in a 10 m<sup>2</sup> plot were sprayed with MFB (100 mL per Litre), commercial chitosan fertilizer, seaweed fertilizer, and water (control). MFB has sprayed every five days intervals until five days before harvest. Commercial fertilizers were diluted with water at a ratio of 1.25:1 (volume: volume). The experiment was designed in a Completely Randomized Design (CRD) with five fertilizer doses and three replicates per treatment.

### 2.6 Statistical analysis

Growth time (day) was the time taken from sowing to harvest. Growth parameters including plant height (cm), canopy diameter (cm), the number of leaves, and leaf area index (leaf area/ground area) were determined for five plants in each treatment. The plant height (cm) was measured from the ground to the highest point of the leaves. The leaf area index is the multiplication of the number of plants/ground area (m<sup>2</sup>) and the leaf area (m<sup>2</sup>)/plant. The yield components included (i) fresh mass/plant (g/plant) (combined weight of stem, leaves, and roots); (ii) estimated yield (ton/ha) (average fresh mass/plant × plant density); (iii) actual yield (ton/ha). Statistical analysis was performed using one ways analysis of variance (ANOVA) followed by Turkey's test in IBM SPSS Statistics 20.0 (SPSS Inc., Chicago, IL, USA). Data represented significant differences as  $p < 0.05$ .

### 3 Results

#### 3.1 Effect of composting time on the quality of moringa foliar fertilizer (MFB)

Results of the study revealed that the chemical properties of MFB depended on the composting time (Table 1). Results presented in table 1 showed that the nitrogen content and pH increased with composting time. These parameters peaked after composting for four months (nitrogen content of 11.9% and pH of 5.04). On the other hand, the contents of P and P<sub>2</sub>O<sub>5</sub> were similar between 3.5 and 4 months, which were higher than those of 3 months. However, the contents of K and K<sub>2</sub>O at 3 months were higher than those of 3.5 and 4 months. OM varied between 29% and 38% after 4 months of composting.

#### 3.2 MFB doses influence growth, yield and quality of leafy vegetables

Lettuce was grown from 35 days to 37 days in the first planting, and from 32 days to 34 days in the second planting (Table 2). Plant

height, number of leaves, canopy diameter, and leaf area index were found to be the highest when MFB was applied at 100 mL per litre (Table 2). Foliar application of MFB at 100 mL per litre significantly increased the fresh mass and estimated yield compared to the lower doses (Table 3). The actual yields were comparable between 100 and 50 mL per litre treatments and were significantly higher than those of other treatments. Higher ascorbic acid content and Brix were observed in the first planting with 100 and 50 mL per Litre treatments, however, these observations were not reproducible in the second planting.

Mustard spinach also has a similar grown period to lettuce and it was recorded from 33 to 36 days in the first planting, and from 28 to 32 days in the second planting (Table 4). Plant height, number of leaves, canopy diameter, and leaf area index slightly changed and tended to decrease with decreasing amounts of MFB. Similarly, fresh mass, estimated yield, and actual yield of mustard spinach also decreased when fewer MFB was applied (Table 5). The highest dose of MFB (100 mL per Litre) correlated with the freshest weight and highest yield of mustard

Table 1 Effect of composting time on the physicochemical properties of moringa foliar biofertilizer (MFB)

Composting time	N (%)	P (%)	P <sub>2</sub> O <sub>5</sub> (%)	K (%)	K <sub>2</sub> O (%)	OM (%)	pH
Three months	4.20 <sup>c</sup>	2.21 <sup>b</sup>	5.06 <sup>b</sup>	7.20 <sup>a</sup>	8.68 <sup>a</sup>	37.73 <sup>a</sup>	3.37 <sup>b</sup>
Three and a half months	8.52 <sup>b</sup>	3.04 <sup>a</sup>	6.97 <sup>a</sup>	5.39 <sup>b</sup>	6.49 <sup>b</sup>	29.13 <sup>a</sup>	4.82 <sup>a</sup>
Four months	11.90 <sup>a</sup>	2.63 <sup>ab</sup>	5.89 <sup>ab</sup>	5.07 <sup>b</sup>	6.11 <sup>b</sup>	32.77 <sup>a</sup>	5.04 <sup>a</sup>

Values given in table are mean of three replicates; mean value followed by the different letter in same column are significantly different at P<0.05

Table 2 Effect of different doses of moringa foliar biofertilizer (MFB) on the growth of lettuce

Dose (mL/Litre)	Growth time (day)	Plant height (cm)	Number of leaves/plant	Canopy diameter (cm)	Leaf area index
First planting					
100	36	22.9 <sup>a</sup> ± 1.10	12.1 <sup>a</sup> ± 0.51	30.9 <sup>a</sup> ± 1.68	57.65 <sup>a</sup> ± 2.94
50	37	20.3 <sup>ab</sup> ± 1.22	11.2 <sup>ab</sup> ± 1.40	30.8 <sup>a</sup> ± 1.59	55.36 <sup>ab</sup> ± 3.61
33.3	36	20.9 <sup>bc</sup> ± 0.56	10.8 <sup>ab</sup> ± 0.40	30.7 <sup>a</sup> ± 2.31	44.67 <sup>c</sup> ± 3.42
25	35	19.4 <sup>c</sup> ± 0.57	10.5 <sup>b</sup> ± 0.42	27.3 <sup>b</sup> ± 0.98	45.57 <sup>c</sup> ± 3.12
20	36	22.0 <sup>ab</sup> ± 1.26	11.0 <sup>ab</sup> ± 0.81	29.4 <sup>ab</sup> ± 1.83	49.43 <sup>bc</sup> ± 3.17
LSD <sub>0.05</sub>		1.98	1.42	3.34	5.59
Second planting					
100	32	23.0 <sup>a</sup> ± 1.35	12.2 <sup>a</sup> ± 1.41	29.6 <sup>a</sup> ± 0.87	51.30 <sup>a</sup> ± 2.23
50	33	20.9 <sup>ab</sup> ± 0.75	10.7 <sup>ab</sup> ± 1.05	27.3 <sup>b</sup> ± 1.36	48.42 <sup>ab</sup> ± 2.85
33.3	34	19.7 <sup>b</sup> ± 1.06	10.5 <sup>b</sup> ± 0.62	26.7 <sup>bc</sup> ± 0.45	45.71 <sup>b</sup> ± 1.89
25	33	19.5 <sup>b</sup> ± 1.26	11.5 <sup>ab</sup> ± 0.53	26.5 <sup>bc</sup> ± 0.72	45.40 <sup>b</sup> ± 3.07
20	34	19.7 <sup>b</sup> ± 1.14	10.2 <sup>b</sup> ± 0.91	25.7 <sup>c</sup> ± 1.03	45.92 <sup>b</sup> ± 1.52
LSD <sub>0.05</sub>		2.02	1.54	1.51	3.41

Values given in table are mean of three replicates; mean value followed by the different letter in same column are significantly different at P<0.05

spinach at both times of planting. The ascorbic acid content decreased from 8.07 (100 mL per Litre) to 5.26 (20 mL per Litre) remained relatively constant across a range of MFB doses. On the other hand, the data for Brix were not reproducible and it

decreased from 8.07 (100 mL per Litre) to 5.26 (20 mL per Litre) in the first planting but it did not significantly change in the second planting.

Table 3 Effect of different doses of moringa foliar biofertilizer (MFB) on the yield and quality of lettuce

Dose (mL/Litre)	Fresh weight (g /plant)	Estimated yield (ton/ha)	Actual yield (ton/ha)	Ascorbic acid (%)	Brix (%)
First planting					
100	127.3 <sup>a</sup> ± 9.02	33.7 <sup>a</sup> ± 2.40	21.3 <sup>a</sup> ± 0.60	2.67 <sup>a</sup> ± 0.12	5.53 <sup>a</sup> ± 0.25
50	108.6 <sup>b</sup> ± 6.43	29.0 <sup>b</sup> ± 1.07	19.7 <sup>ab</sup> ± 0.95	2.57 <sup>ab</sup> ± 0.15	5.10 <sup>a</sup> ± 0.15
33.3	106.0 <sup>bc</sup> ± 4.01	28.0 <sup>bc</sup> ± 1.71	18.3 <sup>bc</sup> ± 1.03	2.34 <sup>bc</sup> ± 0.21	4.53 <sup>b</sup> ± 0.11
25	96.0 <sup>c</sup> ± 6.24	26.7 <sup>bc</sup> ± 0.53	18.2 <sup>bc</sup> ± 0.67	2.19 <sup>c</sup> ± 0.07	4.47 <sup>b</sup> ± 0.18
20	100.0 <sup>bc</sup> ± 2.18	25.6 <sup>c</sup> ± 1.66	17.7 <sup>c</sup> ± 0.43	2.16 <sup>c</sup> ± 0.16	4.43 <sup>b</sup> ± 0.24
LSD <sub>0.05</sub>	10.88	2.95	1.68	0.28	0.43
Second planting					
100	140.2 <sup>a</sup> ± 8.26	34.4 <sup>a</sup> ± 1.83	21.7 <sup>a</sup> ± 1.26	3.45 <sup>a</sup> ± 0.38	5.45 <sup>a</sup> ± 0.15
50	117.0 <sup>b</sup> ± 6.15	28.7 <sup>b</sup> ± 1.91	20.0 <sup>ab</sup> ± 0.95	2.94 <sup>a</sup> ± 0.27	4.94 <sup>a</sup> ± 0.26
33.3	107.3 <sup>bc</sup> ± 5.23	27.0 <sup>bc</sup> ± 1.34	19.0 <sup>bc</sup> ± 0.78	3.01 <sup>a</sup> ± 0.41	5.01 <sup>a</sup> ± 0.68
25	101.6 <sup>c</sup> ± 2.55	26.3 <sup>bc</sup> ± 0.95	18.0 <sup>bc</sup> ± 1.14	3.07 <sup>a</sup> ± 0.06	5.07 <sup>a</sup> ± 0.22
20	99.3 <sup>c</sup> ± 4.79	25.8 <sup>c</sup> ± 1.06	17.3 <sup>c</sup> ± 0.87	3.04 <sup>a</sup> ± 0.09	5.04 <sup>a</sup> ± 0.17
LSD <sub>0.05</sub>	10.85	2.54	2.36	0.72	0.71

Values given in table are mean of three replicates; mean value followed by the different letter in same column are significantly different at P<0.05

Table 4 Effect of different doses of MFB on the growth of mustard spinach

Dose (mL/Litre)	Growth time (day)	Plant height (cm)	Number of leaves/plant	Canopy diameter (cm)	Leaf area index
First planting					
100	34	35.1 <sup>a</sup> ± 2.97	11.4 <sup>a</sup> ± 0.31	31.9 <sup>a</sup> ± 2.07	46.30 <sup>a</sup> ± 3.71
50	33	27.2 <sup>b</sup> ± 3.23	11.3 <sup>a</sup> ± 0.35	30.9 <sup>ab</sup> ± 1.58	43.55 <sup>ab</sup> ± 2.96
33.3	33	31.7 <sup>ab</sup> ± 4.15	10.2 <sup>bc</sup> ± 0.50	28.8 <sup>bc</sup> ± 2.00	40.06 <sup>b</sup> ± 2.28
25	34	30.7 <sup>ab</sup> ± 2.24	9.5 <sup>c</sup> ± 0.45	26.7 <sup>cd</sup> ± 1.68	39.53 <sup>b</sup> ± 4.33
20	36	26.8 <sup>b</sup> ± 3.56	10.3 <sup>b</sup> ± 0.37	25.8 <sup>d</sup> ± 1.45	39.09 <sup>b</sup> ± 2.57
LSD <sub>0.05</sub>		5.76	0.68	2.39	5.22
Second planting					
100	31	29.7 <sup>a</sup> ± 1.15	11.5 <sup>a</sup> ± 1.01	31.2 <sup>a</sup> ± 3.07	44.52 <sup>a</sup> ± 3.12
50	29	27.1 <sup>ab</sup> ± 2.24	10.7 <sup>ab</sup> ± 0.75	29.9 <sup>a</sup> ± 3.21	40.19 <sup>ab</sup> ± 1.14
33.3	29	27.8 <sup>ab</sup> ± 1.63	10.5 <sup>ab</sup> ± 0.31	31.4 <sup>a</sup> ± 2.87	39.43 <sup>ab</sup> ± 2.41
25	28	25.5 <sup>b</sup> ± 2.41	10.3 <sup>b</sup> ± 0.54	28.8 <sup>a</sup> ± 2.12	37.50 <sup>b</sup> ± 3.97
20	32	24.9 <sup>b</sup> ± 3.01	10.0 <sup>b</sup> ± 0.16	29.8 <sup>a</sup> ± 1.93	37.21 <sup>b</sup> ± 2.71
LSD <sub>0.05</sub>		3.99	1.17	3.61	5.31

Values given in table are mean of three replicates; mean value followed by the different letter in same column are significantly different at P<0.05

Table 5 Effect of different doses of moringa foliar biofertilizer (MFB) on the yield and quality of mustard spinach

Dose (mL/Litre)	Fresh weight (g /plant)	Estimated yield (ton/ha)	Actual yield (ton/ha)	Ascorbic acid (%)	Brix (%)
First planting					
100	133.0 <sup>a</sup> ± 8.47	35.3 <sup>a</sup> ± 1.47	28.0 <sup>a</sup> ± 1.17	5.76 <sup>a</sup> ± 0.12	8.07 <sup>a</sup> ± 0.09
50	115.7 <sup>b</sup> ± 5.32	30.7 <sup>b</sup> ± 2.21	24.3 <sup>b</sup> ± 1.35	5.54 <sup>a</sup> ± 0.07	7.13 <sup>b</sup> ± 0.11
33.3	113.0 <sup>bc</sup> ± 2.19	30.3 <sup>bc</sup> ± 1.05	24.6 <sup>b</sup> ± 0.98	5.69 <sup>a</sup> ± 0.05	7.01 <sup>b</sup> ± 0.10
25	112.0 <sup>bc</sup> ± 6.20	29.6 <sup>bc</sup> ± 2.14	23.7 <sup>b</sup> ± 1.61	5.68 <sup>a</sup> ± 0.10	6.77 <sup>b</sup> ± 0.07
20	101.7 <sup>c</sup> ± 7.56	27.0 <sup>c</sup> ± 3.02	22.3 <sup>b</sup> ± 2.21	5.62 <sup>a</sup> ± 0.09	5.26 <sup>c</sup> ± 0.13
LSD <sub>0.05</sub>	11.67	3.41	3.14	0.23	0.48
Second planting					
100	137.7 <sup>a</sup> ± 4.41	37.0 <sup>a</sup> ± 1.92	29.7 <sup>a</sup> ± 0.66	5.52 <sup>a</sup> ± 0.21	4.80 <sup>a</sup> ± 0.24
50	126.0 <sup>b</sup> ± 6.92	33.7 <sup>b</sup> ± 2.04	27.3 <sup>b</sup> ± 1.05	5.02 <sup>a</sup> ± 0.34	4.20 <sup>a</sup> ± 0.19
33.3	119.3 <sup>bc</sup> ± 4.65	31.6 <sup>bc</sup> ± 1.99	25.3 <sup>c</sup> ± 1.24	4.73 <sup>a</sup> ± 0.08	4.53 <sup>a</sup> ± 0.20
25	114.7 <sup>c</sup> ± 8.07	30.7 <sup>c</sup> ± 2.31	24.0 <sup>c</sup> ± 0.68	5.28 <sup>a</sup> ± 0.17	4.43 <sup>a</sup> ± 0.16
20	102.3 <sup>d</sup> ± 5.42	27.3 <sup>c</sup> ± 2.11	21.7 <sup>d</sup> ± 0.41	5.20 <sup>a</sup> ± 0.09	4.40 <sup>a</sup> ± 0.32
LSD <sub>0.05</sub>	9.53	2.50	1.91	0.86	0.62

Values given in table are mean of three replicates; mean value followed by the different letter in same column are significantly different at P<0.05

### 3.3 Effect of various foliar fertilizers on growth, yield, and quality of leafy vegetables

The results suggested that the application of MFB promoted the growth of lettuce (Table 6). Furthermore, the growth time, the number of leaves, canopy diameter, and leaf area index of lettuce plants applied with MFB was comparable to those sprayed with commercial biofertilizers. The plant height of lettuce slightly changed among foliar treatments in the second planting and peaked

at 24.3 cm in plants treated with MFB. The yield of lettuce was enhanced by spraying foliar fertilizers at both plantings. The treatment of MFB increased the fresh weight of lettuce. Estimated yields ranged from 33.8 tons per ha to 37.5 tons per ha and actual yields ranged from 21.3 tons per ha to 23.9 tons per ha across foliar treatments. On the other hand, the ascorbic acid content was not influenced by foliar treatments. Lettuce treated with MFB and chitosan fertilizer had higher Brix in the first planting but these results were not reproducible in the second planting seasons.

Table 6 Effect of various foliar fertilizers on the growth of lettuce

Treatment	Growth time (day)	Plant height (cm)	Number of leaves/Plant	Canopy diameter (cm)	Leaf area index
First planting					
MFB	34	25.4 <sup>a</sup> ± 1.21	12.8 <sup>a</sup> ± 1.02	23.6 <sup>ab</sup> ± 1.33	41.9 <sup>a</sup> ± 2.57
Chitosan fertilizer	33	23.8 <sup>a</sup> ± 1.83	11.5 <sup>ab</sup> ± 1.00	24.9 <sup>a</sup> ± 1.65	38.6 <sup>ab</sup> ± 4.98
Seaweed fertilizer	35	24.6 <sup>a</sup> ± 0.92	11.6 <sup>ab</sup> ± 0.25	24.4 <sup>a</sup> ± 0.61	38.8 <sup>ab</sup> ± 2.81
Control	35	18.4 <sup>b</sup> ± 2.97	10.2 <sup>b</sup> ± 0.82	21.1 <sup>b</sup> ± 1.51	34.0 <sup>b</sup> ± 3.24
LSD <sub>0.05</sub>		3.18	1.48	2.96	5.68
Second planting					
MFB	35	24.3 <sup>a</sup> ± 0.69	12.1 <sup>a</sup> ± 0.52	23.9 <sup>a</sup> ± 1.76	42.2 <sup>a</sup> ± 3.04
Chitosan fertilizer	36	21.5 <sup>bc</sup> ± 1.14	11.2 <sup>ab</sup> ± 0.31	24.9 <sup>a</sup> ± 0.55	39.0 <sup>a</sup> ± 2.56
Seaweed fertilizer	35	22.9 <sup>ab</sup> ± 0.76	11.8 <sup>a</sup> ± 0.67	25.4 <sup>a</sup> ± 1.15	40.1 <sup>a</sup> ± 2.18
Control	35	20.5 <sup>c</sup> ± 1.41	10.3 <sup>b</sup> ± 0.71	21.8 <sup>b</sup> ± 1.37	34.8 <sup>b</sup> ± 1.19
LSD <sub>0.05</sub>		1.74	0.96	1.84	3.61

Values given in table are mean of three replicates; mean value followed by the different letter in same column are significantly different at P<0.05

Table 7 Effect of various foliar fertilizers on the yield and quality of lettuce

Treatment	Fresh weight (g /plant)	Estimated yield (ton/ha)	Actual yield (ton/ha)	Ascorbic acid (%)	Brix (%)
First planting					
MFB	146.7 <sup>a</sup> ± 12.12	37.5 <sup>a</sup> ± 3.23	23.9 <sup>a</sup> ± 1.07	4.59 <sup>a</sup> ± 0.37	5.13 <sup>a</sup> ± 0.27
Chitosan fertilizer	132.3 <sup>ab</sup> ± 11.46	35.3 <sup>a</sup> ± 2.39	21.9 <sup>ab</sup> ± 1.92	4.77 <sup>a</sup> ± 0.29	5.10 <sup>a</sup> ± 0.13
Seaweed fertilizer	127.3 <sup>b</sup> ± 4.16	33.9 <sup>a</sup> ± 2.67	21.4 <sup>b</sup> ± 1.06	4.87 <sup>a</sup> ± 0.55	4.53 <sup>b</sup> ± 0.15
Control	105.3 <sup>c</sup> ± 5.04	28.0 <sup>b</sup> ± 1.81	17.7 <sup>c</sup> ± 0.84	3.96 <sup>a</sup> ± 0.77	4.27 <sup>b</sup> ± 0.19
LSD <sub>0.05</sub>	15.17	3.66	2.10	1.92	0.33
Second planting					
MFB	137.7 <sup>a</sup> ± 3.05	34.7 <sup>a</sup> ± 1.55	23.5 <sup>a</sup> ± 1.42	4.77 <sup>a</sup> ± 0.27	5.34 <sup>a</sup> ± 0.34
Chitosan fertilizer	129.6 <sup>b</sup> ± 4.14	34.6 <sup>a</sup> ± 2.01	21.8 <sup>ab</sup> ± 1.15	4.68 <sup>a</sup> ± 0.13	4.93 <sup>a</sup> ± 0.15
Seaweed fertilizer	123.0 <sup>c</sup> ± 2.39	33.8 <sup>a</sup> ± 1.79	21.3 <sup>b</sup> ± 1.08	4.72 <sup>a</sup> ± 0.56	5.00 <sup>a</sup> ± 0.09
Control	101.7 <sup>d</sup> ± 1.81	27.1 <sup>b</sup> ± 1.43	17.8 <sup>c</sup> ± 1.41	3.63 <sup>b</sup> ± 0.48	4.96 <sup>a</sup> ± 0.47
LSD <sub>0.05</sub>	4.92	2.29	1.87	0.88	0.72

Values given in table are mean of three replicates; mean value followed by the different letter in same column are significantly different at P<0.05

Table 8 Effect of various foliar fertilizers on the growth of mustard spinach

Treatment	Growth time (day)	Plant height (cm)	Number of leaves/Plant	Canopy diameter (cm)	Leaf area index
First planting					
MFB	30	26.0 <sup>a</sup> ± 1.21	12.3 <sup>a</sup> ± 0.31	32.9 <sup>a</sup> ± 2.52	41.3 <sup>a</sup> ± 4.44
Chitosan fertilizer	30	24.4 <sup>a</sup> ± 2.00	11.1 <sup>b</sup> ± 0.36	27.7 <sup>b</sup> ± 2.30	37.6 <sup>a</sup> ± 3.08
Seaweed fertilizer	32	25.3 <sup>a</sup> ± 2.24	12.1 <sup>a</sup> ± 0.62	33.5 <sup>a</sup> ± 1.88	38.2 <sup>a</sup> ± 5.42
Control	33	21.2 <sup>b</sup> ± 2.08	10.3 <sup>b</sup> ± 0.57	26.5 <sup>b</sup> ± 0.97	28.9 <sup>b</sup> ± 1.90
LSD <sub>0.05</sub>		3.19	0.84	3.36	6.74
Second planting					
MFB	29	27.0 <sup>a</sup> ± 1.17	12.3 <sup>a</sup> ± 0.41	29.9 <sup>b</sup> ± 1.51	41.2 <sup>a</sup> ± 5.05
Chitosan fertilizer	28	25.9 <sup>a</sup> ± 1.54	11.8 <sup>a</sup> ± 0.61	27.2 <sup>c</sup> ± 1.69	36.9 <sup>ab</sup> ± 4.87
Seaweed fertilizer	28	26.0 <sup>a</sup> ± 1.13	12.6 <sup>a</sup> ± 0.52	31.7 <sup>a</sup> ± 0.94	42.7 <sup>a</sup> ± 4.51
Control	30	21.9 <sup>b</sup> ± 1.36	10.7 <sup>b</sup> ± 0.66	25.4 <sup>d</sup> ± 1.07	31.2 <sup>b</sup> ± 3.01
LSD <sub>0.05</sub>		1.81	0.87	1.77	6.02

Values given in table are mean of three replicates; mean value followed by the different letter in same column are significantly different at P<0.05

Like lettuce, mustard spinach growth was also affected by foliar treatments. In the first planting, plant height and leaf area index did not vary between different treatments, however, the number of leaves and canopy diameter was found to be higher in plants treated with MFB and seaweed fertilizer. In the second planting, plant height, the number of leaves, and leaf area index were similar among foliar treatments and higher than those of the control. Canopy diameter ranged from 27.2 cm (chitosan fertilizer) to 31.7 cm (seaweed fertilizer), compared to 25.4 cm of the control. The highest fresh weight and estimated yield of

mustard spinach grown in the first planting were found in those treated with MFB but these results were not reproducible in the second planting. Actual yields of plants treated with MFB were comparable to those treated with seaweed fertilizer and higher than those treated with chitosan fertilizer and the control plants. The ascorbic acid of plants grown in the first planting varied from 3.31% (control) to 5.21% (seaweed fertilizer treated), however, the changes were not significant in the second planting. The Brix of mustard spinach across treatments remained constant (larger than 6.0).

Table 9 Effect of various foliar fertilizers on the yield and quality of mustard spinach

Treatment	Fresh weight (g /plant)	Estimated yield (ton/ha)	Actual yield (ton/ha)	Ascorbic acid (%)	Brix (%)
First planting					
MFB	158.0 <sup>a</sup> ± 5.55	37.1 <sup>a</sup> ± 1.06	26.7 <sup>a</sup> ± 1.29	3.92 <sup>b</sup> ± 0.61	6.47 <sup>a</sup> ± 0.49
Chitosan fertilizer	140.2 <sup>b</sup> ± 3.60	32.9 <sup>b</sup> ± 1.60	24.4 <sup>b</sup> ± 0.76	4.06 <sup>b</sup> ± 0.78	6.60 <sup>a</sup> ± 0.08
Seaweed fertilizer	136.7 <sup>b</sup> ± 6.01	32.1 <sup>b</sup> ± 1.42	25.6 <sup>ab</sup> ± 1.22	5.21 <sup>a</sup> ± 0.30	6.67 <sup>a</sup> ± 0.34
Control	116.0 <sup>c</sup> ± 5.78	27.3 <sup>c</sup> ± 0.95	19.2 <sup>c</sup> ± 0.87	3.31 <sup>b</sup> ± 0.54	6.33 <sup>a</sup> ± 0.44
LSD <sub>0.05</sub>	7.89	1.85	1.75	0.88	1.73
Second planting					
MFB	157.3 <sup>a</sup> ± 10.78	37.1 <sup>a</sup> ± 2.05	25.4 <sup>a</sup> ± 1.75	5.22 <sup>a</sup> ± 0.06	6.73 <sup>a</sup> ± 0.49
Chitosan fertilizer	146.7 <sup>a</sup> ± 12.24	32.9 <sup>b</sup> ± 3.32	23.0 <sup>b</sup> ± 0.99	5.12 <sup>a</sup> ± 0.14	6.82 <sup>a</sup> ± 0.35
Seaweed fertilizer	155.6 <sup>a</sup> ± 13.42	36.6 <sup>a</sup> ± 2.69	25.2 <sup>ab</sup> ± 1.42	5.73 <sup>a</sup> ± 0.45	6.98 <sup>a</sup> ± 0.10
Control	117.3 <sup>b</sup> ± 9.97	27.5 <sup>c</sup> ± 3.02	18.6 <sup>c</sup> ± 1.86	5.08 <sup>a</sup> ± 0.58	6.07 <sup>a</sup> ± 0.38
LSD <sub>0.05</sub>	17.07	3.61	2.33	0.87	1.05

Values given in table are mean of three replicates; mean value followed by the different letter in same column are significantly different at  $P < 0.05$

#### 4 Discussion

In this study, the effects of moringa foliar biofertilizer (MFB), prepared from non-edible parts, on the growth and yield of leafy vegetables was investigated. Results of the study revealed that the composting time impacted the quality of MFB (Table 1) and a four-month composting time yielded biofertilizer with the highest nitrogen content. Further, phosphorus content also slightly increased when the composting time was longer than three months, while the organic matter remained unchanged. Furthermore, the pH of the composite biofertilizer increased from 3.37 to 5.04 with increasing composting time.

In this study, high nitrogen content in moringa foliar biofertilizer was prioritized as nitrogen is one of the most essential elements to enable fast growth and optimal production of vegetables (Tam and Cong 2018; Hoa and Thanh 2020). Hence, these results suggested that a four-month composting period was suitable to produce biofertilizer from non-edible moringa plant parts. Apart from macronutrients, moringa plant extracts also contain various antioxidant compounds like zeatin, ascorbic acid, phenolic, flavonoids, vitamin E, minerals, and many other growth hormones such as indole-3-acetic acid (IAA), and gibberellins (GAs) (Isman 1997; Rady and Mohamed 2015; Latif and Mohamed 2016). The previous study also indicated that the stem of moringa was found to enrich nutrients such as vanillin,  $\beta$ -sitostanol, 4-hydroxymellin,  $\beta$ -sitosterol, and octacosanoic acid (Faizi et al. 1994).

During the application of MFB to the leafy plants, the higher the dose of MFB enhanced fresh mass and yields (Tables 3 and 5). In both planting seasons, the dose of 100 mL per Litre MFB produced

the highest fresh mass and yields in both lettuce and mustard spinach. It had been reported that the concentration of moringa leaf extract at 200 mg per Litre was sufficient to enhance the quality of baby leaves (Toscano et al. 2021). In this study, the spray of MFB at 25 mL per Litre and 20 mL per Litre did not improve the yields of these vegetables compared to the Control. Similarly, leaf area indices in both lettuce and mustard spinach decreased in these treatments which could be justified by the poor nutrient supply in these treatments (Tables 2 and 4). Previously, it was demonstrated that the extracts derived from moringa stem bark enhanced the leaf area and fruit yield of sweet bell pepper fruit (Nwokeji et al. 2022). Taken together, the application of 100 mL per Litre MFB produced the highest yield and quality vegetables in this study.

Different types of foliar fertilizer used in this study had comparable effects on the growth of lettuce. However, the actual yield was higher when treated with MFB compared to the seaweed fertilizer treatment. Since leaf areas and plant sizes were similar in plants treated with different foliar fertilizers, it is suggested that MFB stimulated root formation in lettuce which resulted in the differences in yield. Consistent with this, previous studies (Culver et al. 2012; Yasmeen et al. 2013) had shown that the application of moringa plant extract increased root dry weight and root length of tomatoes and wheat. Mustard spinach plants grown on the first planting achieved the highest yield when treated with MFB but these results were not reproducible in the second planting. The effects of seaweed fertilizer on the growth and yield of vegetables in this study were similar to those reported by Hoang et al. (2022).

In lettuce, the ascorbic acid content was not significantly influenced by spraying different foliar fertilizers. Yaseen and

Hajos (2022) found no significant difference in the ascorbic acid content between moringa plant extract treated and non-treated lettuce in 2019, but moringa plant extracts were found to improve the ascorbic acid content of lettuce in 2020. This can be explained by the temperature fluctuation in 2020, which caused physical stresses to plants. In this study, lettuce that was grown in the second planting showed a higher percentage of ascorbic acid when treated with foliar fertilizers despite the effect of higher temperatures from February to March (average temperatures at 18.2°C in January, 21.1°C in February and 25.7°C in March, data not shown). Meanwhile, the ascorbic acid content in mustard spinach varied when treated with different foliar fertilizers on the first planting, although no difference was observed in the second planting. Furthermore, MFB did not affect the percentage of ascorbic acid when various doses (20 mL per Litre to 100 mL per Litre) were sprayed on mustard spinach. These results were contradictory to the findings of Cintya et al. (2018) who found an increase in the content of vitamin C with increasing doses of organic fertilizers in spinach (*Amaranthus tricolor* L.), mustard (*Brassica rapa chinensis*). Brix of lettuce tended to decrease when the doses of MFB decreased in the first planting; however, there was no significant difference across doses in the second planting. Similarly, the application of MFB and chitosan fertilizer improved the Brix in lettuce, compared to seaweed fertilizer and control treatments only in the first planting. Meanwhile, in mustard spinach, Brix varied greatly (5.26%–8.07%) across the different doses of MFB in the first planting but remained relatively constant in the second planting. The effects of MFB on the quality of lettuce and mustard spinach were consistent with previous studies on kale and broccoli baby leaves (Toscano et al. 2021).

## Conclusion

In this work, moringa residues including stems, branches, and leaf petioles, were fermented using EM product and molasses to produce moringa foliar biofertilizer (MFB). To obtain optimal MFB, the composting should be allowed to continue for four months. MFB application enhanced the growth and yield of both lettuce and mustard spinach grown in January and February but did not affect the ascorbic acid content and Brix consistently. The application of MFB produced similar effects compared to the chitosan and seaweed fertilizers. To the authors' knowledge, this was the first study to investigate the effects of MFB on the growth, yield, and quality of leafy vegetables grown in the tropical.

## Author contribution

HD designed the experiment. HD, CQN, TDTN, and BQLN carried out the experiments and performed data analyses. HD, CQN, NHTH and LTD prepared all of the tables, and all authors contributed to data interpretation. TTP and HD wrote the first draft

of the manuscript, and HTHT edited the draft. All authors reviewed the manuscript.

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## Declaration of Competing Interest

The authors declare no competing interests.

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