



Effect of Organic and Bio-Fertilization on Promoting Vegetative Growth of Pear Seedling Cv. (Basateen MKM) Under Intensive Cultivars Conditions

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ABSTRACT

The experiment was conducted over two successive seasons (2023/2025) on 72 pear seedlings (*Pyrus communis*) grafted on *Pyrus betulaefolia* rootstock its two-year-old at the experimental orchard of the Horticulture Research Institute (ARC), Egypt. The seedlings were planted in clay soil with 1.5×2.0 m spacing and subjected to eight fertilization treatments combining compost, mineral fertilizers (NPK), and microbial inoculants including *Azospirillum brasilense*, *Bacillus circulans*, *Trichoderma harzianum*, and arbuscular mycorrhizal fungi (AMF). The study aimed to optimize fertilization strategies by replacing 50% of chemical fertilizers with sustainable biological alternatives under local conditions. Treatments involved combinations of two levels of NPK (50% and 100% of the recommended dose), two levels of compost (also equivalent to 50% and 100% NPK), and three levels of biofertilizers to assess their effects on vegetative growth. The most effective treatment (T8: 50% compost + 50% NPK + 200 ml/tree of microbial mixture + 200 g/tree of AMF) significantly improved seedling height, stem diameter, shoot number, leaf area, and total chlorophyll content. Leaf nutrient levels of N, P, K, and Ca increased accordingly. T8 also enhanced rhizosphere microbial activity, with bacterial counts reaching 95×10^5 cfu/g, actinomycetes 141×10^3 cfu/g, fungi 97×10^3 cfu/g, and enzyme activities of 106.55 μ g TPF/g/day (dehydrogenase) and 0.57 mg PNP/g/day (phosphatase). Macronutrient concentrations in the soil also rose. In conclusion, integrating organic and biofertilizers with reduced mineral input effectively enhances pear seedling growth and soil health, supporting sustainable orchard management.

Keywords: Pear, bio-fertilization, microbial inoculants, compost.

INTRODUCTION

Pears (*Pyrus* spp.) are among the most renowned deciduous fruits in the *Rosaceae* family, produced in temperate zones. It started in the southwest Chinese mountains and has since spread throughout (Katayama et al., 2016). On recently reclaimed fields in Egypt, the "Le-Conte" pear cultivar (*Pyrus communis*, Red) is grown and propagated using *Pyrus betulaefolia* (*P. Betulifolia*) rootstock (Abd-El-Latif et al., 2017). Techniques for management are essential for lowering the cooling requirements of buds. These include defoliation, fertilization, training, controlling tree vigor, and delaying winter pruning (Westwood, 1978; Lang et al., 1987). Pear flower buds emerge on the tips of stems and short spurs that are at least two years old. The growth of flower buds can be altered by a variety of factors and techniques. According to Isaac (1986), Wei (1987), Edwards and Notodimedjo (1987), and others, shoot bending may increase the production of pear trees,

improve the flowering of juvenile trees, and encourage the growth of flower buds. The cultivated area reached 13870 feddan with productivity of 80993 tons (Food and Agriculture Organization of the United Nations, 2023).

A lack of nitrogen will result in smaller leaves since there will be less normal and functional leaf surface (Khan et al., 2015). Nitrogen is crucial for increasing tree vitality. The chemical fertilizers are expensive, and the small farmers cannot afford to use these fertilizers in a suitable amount and balanced proportion, which results in low production (Ahmad, 2000).

Organic nutrient sources contribute to the preservation of soil health by preserving the balance between organic matter and soil micro-flora, which in turn enhances the soil's physical, chemical, and biological characteristics (Walia and Kler, 2009). Organic biofertilizers enriched with microorganisms play an effective role in



enhancing microbial activity in the soil and increasing the availability of essential nutrients such as nitrogen, phosphorus, and potassium. They also improve the physical and chemical properties of the soil and maintain the productivity of fruit trees such as grapes, oranges, apples, and pears (Kang et al., 2022; Butcaru et al., 2024). Use of beneficial microorganisms is an effective strategy for enhancing nutrient availability in plants. Also, inoculating plants with *Azospirillum brasilense* increases nitrate reductase activity, resulting in reduced nitrate levels in plant tissues and improved nitrogen use efficiency and reducing fertilizer dependency by up to 25%. Furthermore, the fungus *Trichoderma harzianum* is widely used in the biocontrol of plant pathogens and modifies root system architecture by stimulating auxin signaling pathways. This fungus also increases enzyme activity and produces secondary metabolites, enhancing nutrient uptake from the soil and nutrient utilization efficiency in plants. The use of these microorganisms has been shown to improve fertilizer use efficiency and reduce fertilizer consumption in agriculture. In addition, both *Azospirillum brasilense* and *Trichoderma harzianum* act as growth promoters, contributing to a reduction in the need for fertilizer inputs in conventional agricultural systems (Galindo

et al., 2020; Moreira et al., 2022). *Bacillus circulans* bacteria provided potassium to the soil by secreting an organic acid, which facilitates the presence of potassium in the soil and its absorption by the plant (Abd El-Rahman and Al-Sharnouby, 2021). Arbuscular mycorrhizal fungi (AMF) constitute a group of soil microorganisms that establish symbiotic relationships with a wide spectrum of plant species. These fungi actively contribute to the enhancement of plant phosphorus (P) nutrition, in addition to improving the nutritional status of other poorly mobile nutrients in the soil. This is through the secretion of organic acids that function to solubilize less soluble phosphate compounds, thereby rendering them available for uptake by plant roots (Ahmed and Al-Sharnouby, 2021).

This study aimed to evaluate the effectiveness of integrating organic and biofertilizers with reduced mineral NPK application on the vegetative growth, nutrient status, and rhizosphere microbial activity of pear seedlings (*Pyrus communis* L. cv. MKM). The goal was to identify sustainable fertilization strategies that reduce dependence on chemical inputs while enhancing soil biological health and supporting the successful establishment of pear orchards in nutrient-deficient or reclaimed soils.

MATERIALS AND METHODS

Experimental site and plant material:

The field experiment was conducted during two successive growing seasons (2023/2024 and 2024/2025) at the experimental orchard of the Horticulture Research Institute, Agricultural Research Center (ARC), Giza Governorate, Egypt (30°01'N, 31°12'E). The aim of the study was to evaluate the potential of replacing mineral fertilizers with organic and biofertilizer alternatives to enhance the vegetative performance of pear seedlings under open-field conditions. A total of 72 healthy and uniform pear seedlings (*Pyrus communis* L. cv. MKM), grafted onto *Pyrus betulaefolia* rootstock, were planted in clay soil at a spacing of 1.5 × 2.0 meters. Prior to transplanting, composite soil

samples were collected from three depths (0–30, 30–60, and 60–90 cm) and subjected to physicochemical and microbiological analysis following standard procedures (Horneck et al., 2011). The soil was classified as clay with a pH of 8.0, electrical conductivity (EC) of 1.26 dS/m, and moderate nutrient content (Table, 1). Microbiological activity in the baseline soil included total bacterial counts of 25×10^3 cfu/g, actinomycetes 36×10^3 cfu/g, and fungi 19×10^3 cfu/g.

Two rates of chemical fertilizers NPK were employed in this study. The first rate was 100% of recommended dose from NPK (286, 190 and 286 g of N, P and K pure units per tree, respectively (\approx 50–70 N, 30–50 P, and 50–70 K



units/feddan/year)). The second rate was 50% of recommended dose from NPK (143, 95 and 143 g of N, P and K pure units per tree, respectively). Monocalcium phosphate (15.5%) was used as a phosphorus source and added to the soil in a single application around the seedlings at the end of winter. Ammonium nitrate (33.5%) was used as a nitrogen source and potassium sulfate (50%) was used as a potassium source. They were added to the soil around the seedlings in three equal applications during March, May, and June.

Organic fertilization was supplied as compost. The physicochemical and biological characteristics of the compost were determined outlined in APHA (1989). Total nitrogen, phosphorus, and potassium contents were determined according to described by Black (1965). Microbiological analyses, encompassing the enumeration of total and fecal coliform bacteria, *Salmonellae* & *Shigella*, were according to Difco (1985). The weed seeds were assessed by Yu et al. (2010), and nematode populations were examined according to Rice et al. (2017). It showed

high organic matter content (40.23%), with a C/N ratio of 17:1 and no detectable coliforms, pathogens, nematodes, or weed seeds (Table, 2).

Two application rates were used:

- **100% compost** (20.429 kg/tree) \approx equivalent to 286 g N, 194 g P, and 284 g K per tree.

- **50% compost** (10.214 kg/tree) \approx equivalent to 143 g N, 97 g P, and 142 g K per tree.

All compost was applied around the trees once during the winter soil preparation period.

The bio fertilizers used included *Azospirillum brasilense*, *Bacillus circulans*, *Trichoderma harzianum*, and *Arbuscular Mycorrhizal* fungi (AMF). These microbial inoculants were sourced from Agric. Microbiology Dept., SWERI, ARC, Giza, Egypt. AMF was applied to the soil once during the winter service, while the bacterial and fungal inoculants were applied twice: the first during winter soil service (second week of December), and the second 35 days later after the first application.

Table (1). Physical, chemical and biological properties of the experimental soil.

Type of analysis	Unit	Soil
Particle size distribution		
Clay	%	46.20
Silt	%	28.60
Sand	%	25.20
Textural class		Clay
Chemical analysis		
pH (1:2.5) soil – water suspension		8
EC (saturation paste extract)	dS/m	1.26
Organic matter	%	1.25
Organic carbon	%	0.73
N	mg/kg	41.91
P	mg/kg	7.93
K	mg/kg	211.31
Soluble cations and anions		
Ca ⁺⁺	mmol/L ⁻¹	6.06
Mg ⁺⁺	mmol/L ⁻¹	3.03
Na ⁺	mmol/L ⁻¹	2.79
K ⁺	mmol/L ⁻¹	0.68
CO ₃ ⁼	mmol/L ⁻¹	-
HCO ₃ ⁻	mmol/L ⁻¹	3.30
Cl ⁻	mmol/L ⁻¹	6.78
SO ₄ ⁼	mmol/L ⁻¹	2.48
Microbiological analysis		
Total bacterial count	cfu/g $\times 10^5$	25
Total actinomycetes	cfu/g $\times 10^3$	36
Total fungi	cfu/g $\times 10^3$	19

cfu/g: Colony forming unit/gram



‡Table (2). Physicochemical and biological characteristics of compost.

Composition	Units	Compost
Density	kg/m	660.00
Moisture content	%	32.00
pH (1:10)		7.20
EC (1:10)	dSm ⁻¹	1.89
N-NH ₄	mg kg ⁻¹	19.00
N-NO ₃	mg kg ⁻¹	283.00
Total nitrogen	%	1.40
Organic matter	%	40.23
Organic carbon	%	23.33
Ash	%	59.77
C/N ratio		17: 1
Total phosphorus (P ₂ O ₅)	%	0.95
Total potassium (K ₂ O)	%	1.39
Weed seeds		Nil
Nematode	Larava/200g	Nil
Total bacterial count	cfu/g × 10 ⁵	31.10
Total actinomycetes	cfu/g × 10 ⁴	185.00
Total fungi	cfu/g × 10 ⁴	96.00
Total coliform	cfu/g	Not detected
Faecal coliform	cfu/g	Not detected
<i>Salmonella</i> and <i>Shigella</i>	cfu/g	Not detected

Cfu/g: Colony forming unit/gram

Layout of the experimental treatments:

The experiment was conducted using a complete randomized design with three replicates, which carried out on 72 seedlings of pear with 8 treatments as follows:

T₁: 100% NPK Control (286 g of N, 190 g of P and 286 g of K per tree).

T₂: 100% Compost (20.429 Kg/tree).

T₃: 100% Compost + 100 ml/tree of mixture of *Azospirillum brasilense*, *Bacillus circulans* and *Trichoderma harzianum* + 100 g/tree Arbuscular mycorrhizal fungi (AMF).

T₄: 100% Compost + 150 ml/tree of mixture of *Azospirillum brasilense*, *Bacillus circulans* and *Trichoderma harzianum* + 150 g/tree AMF.

T₅: 100% Compost + 200 ml/tree of mixture of *Azospirillum brasilense*, *Bacillus circulans* and *Trichoderma harzianum* + 200 g/tree AMF.

T₆: 50% Compost (10.214 Kg/tree) + 50% NPK (143 g of N, 95 g of P and 143 g of K per tree) + 100 ml/tree of mixture of *Azospirillum brasilense*, *Bacillus circulans* and *Trichoderma harzianum* + 100 g/tree AMF.

T₇: 50% Compost + 50% NPK + 150 ml/tree of mixture of *Azospirillum brasilense*, *Bacillus circulans* and *Trichoderma harzianum* + 150 g/tree AMF.

T₈: 50% Compost + 50% NPK + 200 ml/tree of mixture of *Azospirillum brasilense*, *Bacillus circulans* and *Trichoderma harzianum* + 200 g/tree AMF.

The considered treatments were evaluated through the following determinations:

1. Vegetative growth parameters:

a. Stem diameter rate (cm/year): was measured at the end of each season as the increment in stem diameter (above 3 cm from the grafting point, according to Soliman, (2020). Then, it was calculated as a difference between stem diameter at the end season and initial stem diameter at the beginning of the season.

b. Trunk circumference: it was calculated as a difference between the end season and the beginning of the season.

c. Leaves number /shoot: were counted at the end of each season of study.

d. Leaf area (cm²): six mature leaves were taken at the third node from the base of the main stem of the seedlings for estimating leaf area meter (model 1203, CID, Inc., USA).

e. Seedlings highest (cm): tree seedlings were randomly chosen the mean value of each treatment was estimated.



f. Number of shoots / seedling: The new shoot number that formed was counted for each seedling.

g. New shoot length (cm): three shoots were randomly chosen on each seedling for counting the newly formed shoots. The mean value of each treatment was estimated.

h. Shoot diameter: Average shoot diameter measured by using a vernier caliper

i. Main number of roots.

j. Root dry weight.

2. Endogenous leaf chemical contents:

Leaf nutrient contents (macro and micro elements) were determined in the oven-dried leaf samples (5- 7th leaf from the base) collected at the first week of July of both seasons. Leaves were taken as previously described, dried at 70° for three days, and used for the following analysis:

a. Leaf nitrogen content percentages: sample of 5 grams dry weight from each replicate were used to estimate the leaf nitrogen content. Then, it was estimated by Micro-Kjeldahl according to A.O.A.C (2005).

b. Leaf phosphorus content (%): was estimated as described by Chapman and Parker (1961).

c. Leaf potassium content (%): was estimated according to Lilleland and Brown (1946).

d. Chlorophyll analysis:

1. Weight 100 mg of leaf tissue in fractions into the vial containing 7 mL Dimethyl sulphoxide.
2. Chlorophyll will extract into the fluid without grinding at 65 °C by incubating for various times, depending on the degree of cutinization and thickness of the leaf.
3. Transfer the extract liquid to a graduated tube and make up to a total volume of 10 ml with Dimethyl sulphoxide, assay immediately, or transfer to vials and store between 0-4 °C until required for analysis.

4. Take 3 ml of chlorophyll extract and transfer to cuvette, 5. Measure the extract's optical density (OD) at the following wavelengths, 645 and 663 nm, using Dimethyl sulphoxide as a blank after 30 min and 1 hr, incubation.

- Chlorophyll A (mg/g) = $12.7 (OD_{663}) - 2.69 (OD_{645}) \times (V / (1000 \times wt.))$.

- Chlorophyll B (mg/g) = $22.9 (OD_{645}) - 4.68 (OD_{663}) \times (V / (1000 \times wt.))$.

- Total Chlorophyll (mg/g) = $20.2 (OD_{645}) + 8.02 (OD_{663}) \times (V / (1000 \times wt.))$.

OD: optical density at certain wave length (645 or 663 nm).

V: Final volume (10 ml).

Wt.: weight of sample (100 mg).

e. Leaf Calcium content (ppm): It was determined by using Atomic absorption spectrophotometer as described by Lindsay and Norvell (1978).

f. Leaf content of total carbohydrates (mg/100g): It was determined in dry leaf samples collected at the 2nd week of July of each season according to A.O.A.C. (2005) as mg/100 g D.W.

g. C/N ratio: It was calculated according the following equation:

$$\text{C/N ratio} = \frac{\text{Total carbohydrates}}{\text{Total Nitrogen}}$$

3. Soil Microbial Properties:

Total bacterial counts (Allen, 1959), actinomycetes (Williams and Davis, 1965), and fungal counts (Martin, 1950) were determined. The dehydrogenase activity ($\mu\text{g/g}$ dry soil/day) (Skujins, 1976) and total phosphatase (mg/ PNP/g dry soil) (Tabatabai, 1982), were determined.

Statistical Analysis:

Each season was subjected to a completely randomized design using the analysis of variance (ANOVA) as described by Gomez and Gomez (1984). The LSD test, which compares differences between treatment mean values at the 0.05 level of probability, was used to compare the discrepancies. ANOVA was used in the MSTAT-C software program to evaluate the data (Freed *et al.*, 1989).



RESULTS AND DISCUSSION

1. Vegetative growth parameters:

Stem diameter rate (cm/year) and trunk circumference: Data in Table (3) showed that the stem diameter rate (cm/year) and trunk circumference of pear seedlings during the two seasons significantly differ among all treatments under study. Also, the stem diameter rate and the trunk circumference were recorded as the highest results with the combinations of 50 % compost + 50% NPK + 200 ml/tree of mixture of *Azospirillum brasilense*, *Bacillus circulans* and *Trichoderma harzianum* and 200g/tree Arbuscular Mycorrhizal fungi in both seasons compared to the other treatments in the study.

This supports findings by Prasad et al., (2017) found that Bio-fertilization is of

great importance in alleviating the deterioration of natural resources and environmental pollution. Bio-fertilizers combined with organic manure influence plant growth by enhancing root biomass, and an increase in all vegetative growth parameters by reducing consumption of natural sources of energy. Also, Wang et al., (2022) recorded that they recorded that when using the organic or bio-fertilizers on alter soil physical and chemical properties, thus manipulating specific microbial taxa and functions within the rhizosphere microbiome of pear plants to promote all of the vegetative growth characteristics, and they affected to increase the yield, which may help in the design of more efficient biofertilizers to promote the growth of fruit and production.

Table (3). Effect of organic and bio-fertilizers on stem diameter rate (cm/year) and trunk circumference of pear seedlings during two seasons (2023-2024/2024-2025).

Treatments	Stem diameter rate (cm/year)	Trunk circumference	Stem diameter rate (cm/year)	Trunk circumference
	(2023-2024)		(2024-2025)	
T ₁	73.00 CD	1.550 F	87.00 C	1.930 F
T ₂	70.00 D	1.630 F	81.00 C	2.007 F
T ₃	73.00 CD	2.330 E	87.00 C	2.710 E
T ₄	76.00 C	3.470 D	85.33 C	3.853 D
T ₅	90.33 A	3.700 C	96.00 B	4.080 C
T ₆	82.00 B	3.590 CD	96.00 B	4.000 C
T ₇	90.00 A	3.950 B	104.0 A	4.373 B
T ₈	93.00 A	4.233 A	108.0 A	4.580 A

Means in a column followed by the same letter do not differ significantly according to Duncan's New Multiple Range t-Test at 5 % level.

a. Leaves number/shoot and leaf area (cm²): Leaves number/shoot and leaf areas were significantly different during the two seasons under study in Table (4). The number/shoots were recorded as the highest data with T₈ and T₇ treatments, respectively, in both seasons compared to the other treatments under the study. In the same Table, the leaf area in the first season was recorded as the maximum average with treatments T₈ and T₇. On the other hand, the treatments of T₈, T₇, T₆, and T₅, respectively, gave the highest data of leaf area as followed by T₄, T₃, T₂, and T₁ in the second season. Data harmony

with Kai et al., (2013), they study different organic fertilizers on the growth of the Huangguan pear tree. They found that the applications of organic fertilizer enhance the leaf area and increase the number of leaves per shoot. Also, Yu et al., (2025) studied the effects of organic fertilizers and the rhizosphere microbiome on plant growth. They found that the plant fertilizers maintained the stability of the soil microbial community, beneficial to plant root development, increased carbon cycle pathways, and affected the area of the canopy trees under study.

**Table (4).** Effect of organic and bio-fertilizers on leaves number/shoot, and leaf area (cm²) of pear seedlings during two seasons (2023-2024/2024-2025).

Treatments	(2023-2024)		(2024-2025)	
	Leaves number /shoot	Leaf area (cm ²)	Leaves number/shoot	Leaf area (cm ²)
T ₁	9.000 E	25.40 D	10.32 D	30.12 D
T ₂	9.270 DE	25.70 CD	10.50 D	30.28 CD
T ₃	9.607 D	26.18 BC	11.15 C	31.00 C
T ₄	10.44 C	26.30 BC	11.46 C	32.27 B
T ₅	11.23 B	26.82 B	11.90 B	33.17 A
T ₆	11.52 B	27.63 A	12.16 B	33.30 A
T ₇	12.20 A	27.80 A	13.24 A	33.50 A
T ₈	12.32 A	27.85 A	13.65 A	33.62 A

Means in a column followed by the same letter do not differ significantly according to Duncan's New Multiple Range t-Test at 5 % level.

b. Seedlings highest (cm), number of shoots /seedling, new shoot length (cm) and shoot diameter: The highest seedlings (cm), number of shoots /seedling, new shoot length (cm), and shoot diameter in Table (5) showed that all treatments significantly affected the highest seedlings (cm), number of shoots /seedling, new shoot length (cm), and shoot diameter in both seasons. Also, using treatments T₈ or T₇, respectively, increased all growth parameters in Table 6 compared to the other treatments in the same seasons under study. The results harmony with Haska et al., (2022) found that the vegetative growth parameters speed with the peat moss medium and the biofertilizer applications and biofertilizer application can improve the seedling quality in the production of organic seedlings. Also, Yu et al., (2025) recorded that the Plant fertilizers were enhancing the all growth parameters of plants in the long term, and a guide for organic fertilizer selection from the perspective of soil microecology and promoting sustainable development of organic agriculture.

c. Main number of roots and Root dry weight: Table (6) showed that the main number of roots and root dry weight were significantly affected by all treatments under the two seasons in the study. Data of the main roots and dry weight showed that the treatment of T₈ gave the maximum average of main roots and dry weight as followed by T₇, T₆, T₅, T₄, T₃, T₂, and T₁ in both seasons. Data confirmed that Haska et al., (2022) studied the effect of different organic growing mediums and biofertilizer in organic seedling production. They received that Biofertilizer application improved the root growth rate, such as weight, number, and length, in all seedlings under study. Also, the heaviest roots were obtained from seedlings inoculated with biofertilizer and grown in medium Peat moss for all species. Yu et al., (2025) studied the Effects of organic fertilizers on plant growth and the rhizosphere microbiome they found that animal fertilizers enhanced nitrogen cycle pathways, while plant fertilizers boosted carbon cycle pathways and promoted plant root development.

Table (5). Effect of organic and bio-fertilizers on shoot highest, number of shoots/seedling, new shoot length (cm) and shoot diameter of pear seedlings during two seasons (2023-2024/ 2024-2025).

Treatments	(2023-2024)				(2024-2025)			
	Shoot highest (cm)	Number of shoots/seedling	New shoot length (cm)	Shoot diameter (cm)	Shoot highest (cm)	Number of shoots/ seedling	New shoot length (cm)	Shoot diameter (cm)
T ₁	116.0 F	7.000 E	38.00 D	0.7467 C	134.0 E	8.000 D	43.70 E	0.880 C
T ₂	118.0 EF	7.193 E	38.67 D	0.8533 BC	136.3 E	8.233 D	43.80 E	0.983 C
T ₃	122.0 DE	8.700 D	40.43 C	0.8633 BC	156.7 D	9.747 C	46.13 D	0.950 C
T ₄	125.0 CD	8.950 D	40.72 C	0.9200 B	161.3 CD	9.700 C	46.39 D	0.973 C
T ₅	135.0 B	10.13 B	45.30 B	1.200 A	171.7 BC	10.47 B	51.00 B	1.367 B
T ₆	130.0 BC	9.293 C	45.36 B	0.9567 B	176.7 B	10.42 B	48.81 C	1.217 B
T ₇	143.0 A	12.52 A	48.88 A	1.220 A	194.7 A	13.58 A	54.58 A	1.530 A
T ₈	147.0 A	12.43 A	48.50 A	1.317 A	196.3 A	13.30 A	54.20 A	1.597 A

Means in a column followed by the same letter do not differ significantly according to Duncan's New Multiple Range t-Test at 5 % level. □

**Table (6).** Effect of organic and bio-fertilizers on main number of roots and Root dry weight of pear seedlings during two seasons (2023-2024/2024-2025).

Treatments	Main number of roots (2023-2024)	Root dry weight (2023-2024)	Main number of roots (2024-2025)	Root dry weight (2024-2025)
T ₁	7.790 C	19.50 D	8.740 B	20.00 F
T ₂	8.007 BC	19.52 D	8.000 BC	20.85 E
T ₃	8.120 BC	21.17 C	7.233 C	22.50 D
T ₄	8.153 B	21.50 C	8.243 BC	23.29 D
T ₅	8.333 B	21.50 C	8.780 B	25.51 C
T ₆	8.347 B	24.18 B	8.853 AB	25.79 BC
T ₇	8.757 A	24.70 B	9.333 AB	26.66 AB
T ₈	8.890 A	25.75 A	10.17 A	27.15 A

Means in a column followed by the same letter do not differ significantly according to Duncan's New Multiple Range t-Test at 5 % level.

2. Endogenous leaf chemical contents:

a. Chlorophyll A, chlorophyll B, and total chlorophyll (mg/g): Data in Table (7) cleaned that the chlorophyll A, chlorophyll B, and total Chlorophyll (mg/g) of pear seedlings during the two seasons significantly differ among all treatments under study. Also, the data of chlorophyll A, chlorophyll B, and total chlorophyll recorded as the highest results with the combinations of T₈ (50% Compost + 50% NPK + 200 ml/tree of mixture of *Azospirillum brasilense*,

Bacillus circulans and *Trichoderma harzianum* + 200g/tree Arbuscular Mycorrhizal fungi) as followed by T₇, T₆, T₅, T₄, T₃, T₂, and T₁ in both seasons. This supports findings by Ritchie, (2008) stated that the different sources of organic fertilizer in the study affected all leaf chemical content, and changed the physiological interactions in plants over a long period, as followed by the growth characteristics and the crop yield.

Table (7). Effect of organic and bio- fertilizers on chlorophyll (A), chlorophyll (B), and total Chlorophyll of pear seedlings during two seasons (2023-2024/ 2024-2025).

Treatments	Chlorophyll (A) (2023-2024)	Chlorophyll (B) (2023-2024)	Total chlorophyll (2023-2024)	Chlorophyll (A) (2024-2025)	Chlorophyll (B) (2024-2025)	Total chlorophyll (2024-2025)
T ₁	0.761 B	0.277 E	1.037 F	0.917 B	0.327 E	1.244 E
T ₂	0.819 B	0.611 D	1.430 E	1.100 B	0.758 D	1.858 D
T ₃	0.854 B	0.627 CD	1.484 DE	1.108 B	0.843 D	1.951 D
T ₄	0.878 B	0.672 C	1.562 D	1.220 AB	0.802 D	2.022 D
T ₅	0.790 B	0.660 CD	1.545 D	1.233 AB	0.800 D	2.033 D
T ₆	0.862 B	0.750 B	1.777 C	1.427 AB	1.160 C	2.587 C
T ₇	1.155 A	1.015 A	2.170 B	1.882 AB	1.358 B	3.080 B
T ₈	1.265 A	1.030 A	2.262 A	1.919 A	1.600 A	3.519 A

Means in a column followed by the same letter do not differ significantly according to Duncan's New Multiple Range t-Test at 5 % level.

b. Leaf nitrogen content (%), leaf content of total carbohydrates (mg/100g) and C/N ratio: Table (8) showed that the nitrogen content, total carbohydrates, and C/N of pear seedlings during the two seasons significantly differ among all treatments under study. Also, the combination of T₈ (50% Compost + 50% NPK + 200 ml/tree of mixture of *Azospirillum brasilense*, *Bacillus circulans* and *Trichoderma harzianum* + 200g/tree Arbuscular Mycorrhizal fungi) recorded the highest results of N%, carbohydrates, and C/N ratio in both seasons as compared to the other

combination treatments under study. **Haska et al. (2022)** reported that biofertilizer application can improve the seedling quality and increased the leaf mineral content. **Abdel-Rahman et al. (2024)** The maximum N%, total carbohydrates, and C/N ratio were recorded with Cyanobacteria + *B. subtilis* + *B. amylo* compared to the other applications under study.

c. Leaf phosphorus content (%), leaf potassium content (%) and leaf calcium content (ppm): Table (9) showed that the P, K and Ca of pear seedlings during the two seasons significantly differ among all treatments



under study. Also, the combination of T₈ (50% compost + 50% NPK + 200 ml/tree of mixture of *Azospirillum brasilense*, *Bacillus circulans* and *Trichoderma harzianum* + 200g/tree Arbuscular Mycorrhizal fungi) recorded the highest results of P, K, and Ca contents in both seasons as compared to the other combination treatments under study. Ritchie, (2008) found that the

various organic fertilizer sources used in the study altered the physiological interactions in plants and had an impact on all leaf chemical composition. Also, El-Banna and Fouda (2018) found that 30 % of mineral fertilizer combined with farmyard manure at 30 m³/fed., in the presence of bio-fertilizer and sprayed with humic acid increased the leaf mineral on plants compared control.

Table (8). Effect of organic and bio-fertilizers on leaf nitrogen content (%), leaf content of total carbohydrates (mg/100g) and C/N ratio of pear seedlings during two seasons (2023-2024/2024-2025).

Treatments	Leaf nitrogen content (%)	Total carbohydrates (mg/100g)	C/N ratio	Leaf nitrogen content (%)	Total carbohydrates (mg/100g)	C/N ratio
(2023-2024)			(2024-2025)			
T ₁	1.723 E	9.180 E	0.188 B	1.800 D	9.683 D	0.185 B
T ₂	1.800 E	9.520 DE	0.189 B	1.910 D	9.760 D	0.195 B
T ₃	2.130 D	9.540 DE	0.223 AB	2.250 C	9.680 D	0.232 AB
T ₄	2.237 CD	9.803 CD	0.228 AB	2.327 C	9.870 D	0.235 AB
T ₅	2.657 AB	10.60 AB	0.251 A	2.800 AB	10.75 B	0.260 A
T ₆	2.473 BC	10.18 BC	0.243 A	2.760 B	10.53 C	0.262 A
T ₇	2.700 AB	10.62 AB	0.254 A	2.810 AB	11.13 A	0.252 A
T ₈	2.900 A	10.95 A	0.265 A	2.953 A	11.22 A	0.263 A

Means in a column followed by the same letter do not differ significantly according to Duncan's New Multiple Range t-Test at 5 % level.

Table (9). Effect of organic and bio- fertilizers on Leaf phosphorus content (%), leaf potassium content (%), and leaf calcium content (ppm) of pear seedlings during two seasons (2023-2024/ 2024-2025).

Treatments	Leaf phosphorus content (%)	Leaf potassium content (%)	Leaf calcium content (ppm)	Leaf phosphorus content (%)	Leaf potassium content (%)	Leaf calcium content (ppm)
	(2023-2024)			(2024-2025)		
T ₁	0.110 D	0.460 C	1.300 B	0.264 D	0.615 C	1.320 C
T ₂	0.180 CD	0.581 BC	1.300 B	0.340 CD	0.725 BC	1.320 C
T ₃	0.221 B-D	0.550 BC	1.320 B	0.342 CD	0.863 B	1.340 BC
T ₄	0.232 A-D	0.580 BC	1.357 B	0.362 C	0.727 BC	1.370 BC
T ₅	0.307 AB	0.579 BC	1.400 B	0.640 B	0.889 B	1.420 A-C
T ₆	0.301 A-C	0.731 AB	1.440 A-C	0.631 B	0.920 B	1.450 AB
T ₇	0.330 AB	0.653 A-C	1.470 AB	0.704 B	0.880 B	1.500 AB
T ₈	0.350 A	0.832 A	1.530 A	0.867 A	1.369 A	1.657 A

Means in a column followed by the same letter do not differ significantly according to Duncan's New Multiple Range t-Test at 5 % level.

3. Soil microbial properties:

a. Effect of different treatments on total counts of bacteria, actinomycetes and fungi in soil rhizosphere:

The results in Table (10) showed that the numbers of total bacterial counts, total actinomycetes, and total fungi in the rhizosphere of pear tree roots zone during the two seasons were increased. The results showed that organic and biofertilizers played an effective role in increasing the number of microbes in the pear tree root zone. So, the numbers of total bacterial, actinomycetes, and fungi in the tree root zone were higher in the second season than to the first season in all treatments. Also, the total number of bacteria was higher than the numbers of both actinomycetes and fungi in the tree

root zone for all treatments. The results also showed that T₈ (50% compost + 50% NPK + 200 ml/tree of mixture of *Azospirillum brasilense*, *Bacillus circulans* and *Trichoderma harzianum* + 200g/tree Arbuscular Mycorrhizal fungi) gave the highest counts of total bacteria (59 and 95 × 10⁵ cfu/g soil), total actinomycetes (125 and 141 × 10³ cfu/g soil) and total fungi (84 and 97 × 10³ cfu/g soil) during two seasons, respectively. This is due to the presence of compost and its content of microorganisms and nutrients that increase the reproduction of microorganisms (*Azospirillum brasilense*, *Bacillus circulans*, *Trichoderma harzianum* and Arbuscular mycorrhizal fungi). In contrast, the results showed that using only chemical fertilizers, as in T₁ (control),



resulted in a significant decrease in the numbers of total bacteria, actinomycetes, and fungi, as it was recorded (27 and 36×10^5 cfu/g soil), (41 and 63×10^3 cfu/g soil) and (25 and 31×10^3 cfu/g soil) through the two seasons, respectively. These results are similar to those obtained by Talwar et al., (2017), Mohamed and Massoud (2017) and Khamis et al., (2018), they found that adding organic and biofertilizer to agriculture increases the activity of microbes in the soil and thus increases their numbers in the soil. Also, inoculating the soil with microorganisms increases the biological diversity in the soil and thus increases the concentration of nutrients available to the plant, thus increasing the yield (Ahmed and Ibrahim, 2023). Mohamed and Massoud (2017) found that the increased microbial population in the root zone of trees

inoculated with biofertilizers was due to integrated between *Azotobacter* and Arbuscular mycorrhiza fungi, with the fungi providing tubes for nutrient transport. In addition, *Azotobacter* symbiotically fixes nitrogen within cell plant, leading to increased numbers of total bacteria, actinomycetes, and total mycorrhizae in soil inoculated with organic and biofertilizers. Using of biofertilizers and organic fertilizers increases the total number of bacteria, fungi, and actinomycetes in the soil in the root zone of plants. Where, microbes use carbon from organic matter as an energy source, increasing their reproduction and thus increasing the number of microbes. In addition, mixed biofertilizers integrated with chemical fertilizers increased the number of microbes in the soil rhizosphere (Sinha et al., 2024).

Table (10): Changes of total bacterial, actinomycetes and fungi counts in the rhizosphere of pear seedlings during two seasons (2023-2024/ 2024-2025).

Treatments	Total bacterial counts (10^5 cfu/g soil)			Total actinomycetes (10^3 cfu/g soil)			Total fungi (10^3 cfu/g soil)		
	(2023-2024)	(2024-2025)	Mean	(2023-2024)	(2024-2025)	Mean	(2023-2024)	(2024-2025)	Mean
T1	27.00	36.00	31.50	41.00	63.00	52.00	25.00	31.00	28.00
T2	31.00	40.00	35.50	56.00	79.00	67.50	32.00	42.00	37.00
T3	37.00	51.00	44.00	62.00	88.00	75.00	39.00	49.00	44.00
T4	40.00	62.00	51.00	75.00	97.00	86.00	49.00	55.00	52.00
T5	42.00	66.00	54.00	86.00	105.00	95.50	58.00	69.00	63.50
T6	46.00	72.00	59.00	97.00	120.00	108.50	64.00	77.00	70.50
T7	51.00	84.00	67.50	111.00	129.00	120.00	78.00	85.00	81.50
T8	59.00	95.00	77.00	125.00	141.00	133.00	84.00	97.00	90.50
Mean	41.63	63.25	-	81.63	102.75	-	53.63	63.13	-

Means in a column followed by the same letter do not differ significantly according to Duncan's New Multiple Range t-Test at 5 % level.

b. Effect of organic and biofertilizers treatments on dehydrogenase and phosphatase enzyme activities in tested soil:

The data presented in Fig. (1 and 2) illustrate the biological activity of dehydrogenase and phosphatase enzymes in the rhizosphere of pear root zone during the two study seasons 2023-2024. In both seasons, it was observed that the treatments that were fertilized with organic and biofertilizers had higher enzyme activity than the treatments that were not fertilized. Microorganisms in the soil play an effective role in increasing the activity of dehydrogenase and phosphatase enzymes. Data showed that T₈ (50% Compost + 50% NPK + 200 ml/tree of

mixture of *Azospirillum brasilense*, *Bacillus circulans* and *Trichoderma harzianum* + 200g/tree Arbuscular Mycorrhizal fungi) gave the highest enzyme activity for both enzymes during the two study seasons, recording 79.11 and 106.55 μ g TPF/g dry rhizosphere/day for dehydrogenase, also it recorded 0.54 and 0.57 mg PNP/g dry soil for phosphatase during the 2023 and 2024 study seasons, respectively. On the other hand, the results showed that T₁ (control) gave the lowest enzyme activity (dehydrogenase and phosphatase) in the soil of the tree root zone during both seasons. In addition to, the results showed that the activity of dehydrogenase and phosphatase enzymes in the tree root zone were higher during

the 2024 season compared to the 2023 season. These results are agreement consistent conforming to present by Khamis et al., (2018), Ahmed et al., (2024), Abdel-Rahman et al., (2024) and Okba et al., (2025). The enzyme dehydrogenase is found in living cells and is an indicator of soil health and the number of microbes present. The increased activity of this enzyme when small amounts of mineral fertilizer are added to fruit trees is due to the increased microbial population (Mohamed and Massoud, 2017). Abd El-Rahman and Al-Sharnouby (2021) found that the increase in enzyme activity is due to the increase in soil acidity, which leads to an increase in the mineralization of organic compounds. The increase in the phosphatase enzyme is due to the use of

Arbuscular mycorrhiza fungi and *Bacillus megatherium* in combination with a small amount of mineral fertilizer. This leads to an increase in the concentration of this enzyme in the soil, which increases the conversion of phosphorus from the unavailable form to the available form for absorption by tree roots, thus increasing tree growth (Ahmed and Al-Sharnouby, 2021). Soil enzyme activity was linked to the availability of nutrients in the soil. Adding biofertilizers integrated with organic fertilizers increases enzyme activity in the soil. The presence of *Trichoderma harzianum* also increases the decomposition of organic matter in the soil, which leads to an increase in the organic carbon required for microbial activity (Sinha et al., 2024).

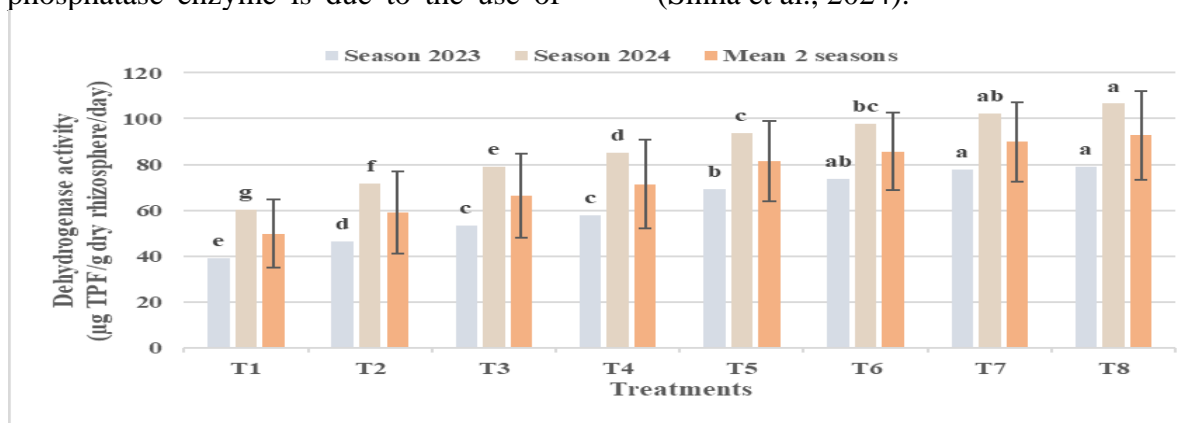


Fig. (1): Effect of mineral, organic and bio fertilizers on dehydrogenase activity in tested soil of pear seedlings in 2023 and 2024 seasons. Values are means of three replicates \pm standard deviation. Histograms sharing the same letter are not statistically different using Duncan's multiple range test ($p \leq 0.05$).

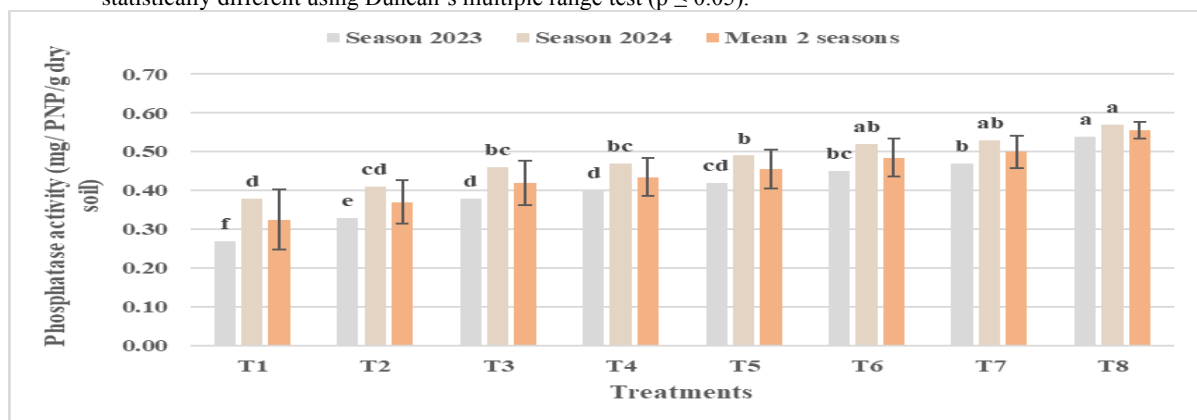


Fig. (2): Effect of mineral, organic and bio fertilizers on phosphatase activity in tested soil of pear seedlings in 2023 and 2024 seasons. Values are means of three replicates \pm standard deviation. Histograms sharing the same letter are not statistically different using Duncan's multiple range test ($p \leq 0.05$).

c. Effect of organic and biofertilizers treatments on macronutrients contents in tested soil:

The results in Table (11) indicated that the use of organic fertilizers (compost)

and biofertilizers (*Azospirillum brasilense*, *Bacillus circulans*, *Trichoderma harzianum* and Arbuscular Mycorrhizal fungi) had a positive impact on increasing the availability of major nutrients in the



soil. The results showed that T₈ (50% compost + 50% NPK + 200 ml/tree of mixture of *Azospirillum brasilense*, *Bacillus circulans* and *Trichoderma harzianum* + 200g/tree Arbuscular Mycorrhizal fungi) recorded a clear increase in N, P, and K, where N was recorded 58.76 and 63.11 mg kg⁻¹, P was recorded 13.07 and 21.91 mg kg⁻¹, and K was recorded 220.13 and 263.18 mg kg⁻¹ in both seasons, respectively, compared to the control treatment (T₁). This is due to the presence of *Azospirillum brasilense*, which works to fix nitrogen symbiotically in the soil, and the presence of *Bacillus circulans*, which works to increase the availability of K in the soil, in addition to the presence of Arbuscular Mycorrhizal fungi, which secretes external enzymes that work to increase the solubility of phosphorus in the soil, and *Trichoderma harzianum*, which works to increase the decomposition of organic matter in the

soil, thus increasing soil acidity and increasing the availability of elements in the soil. All these factors played an effective role in increasing the availability of major elements in the treatments. It was clear that that treatment content compost 100% (T₂) gave results higher than control (T₁) 100% NPK where compost was very mature so, all elements were released and they were available for seedlings in addition to, it contents other factors (humic substances, organic matter, enzymes, vitamins, antibiotics, growth regulators, nitrogen-fixing, phosphorus-solubilizing and potassium facilitating microorganisms). These results are agreement consistent conforming to present by Ahmed et al., (2024) and Sinha et al., (2024). They found that adding organic and biofertilizers to the soil increases the availability of major elements in the soil.

Table (11): Effect of different treatments on available macronutrients contents in the rhizosphere of pear seedlings during two seasons (2023-2024/ 2024-2025).

Treatments	N (mg Kg ⁻¹)		P (mg Kg ⁻¹)		K (mg Kg ⁻¹)	
	(2023-2024)	(2024-2025)	(2023-2024)	(2024-2025)	(2023-2024)	(2024-2025)
T ₁	43.28 E	45.69 D	8.41 B	8.91 E	212.01 E	213.91 F
T ₂	46.03 DE	48.33 CD	9.64 AB	10.21 E	212.65 DE	215.63 F
T ₃	47.66 C-E	50.09 B-D	10.11 AB	11.53 DE	213.17 C-E	219.03 F
T ₄	48.25 B-E	51.03 B-D	10.63 AB	13.03 DE	214.61 B-E	225.63 E
T ₅	51.88 A-D	55.63 A-C	11.81 AB	15.23 CD	216.63 A-D	241.13 D
T ₆	54.63 A-C	57.88 AB	11.98 AB	17.66 BC	217.19 A-C	247.11 C
T ₇	56.26 AB	59.09 AB	12.51 AB	19.71 AB	218.13 AB	252.91 B
T ₈	58.76 A	63.11 A	13.07 A	21.91 A	220.13 A	263.18 A

Means in a column followed by the same letter do not differ significantly according to Duncan's New Multiple Range t-Test at 5 % level.

Conclusion:

The study demonstrated that combining 50% mineral NPK with 50% compost and 200 ml/tree of mixture of *Azospirillum brasilense*, *Bacillus circulans* and *Trichoderma harzianum* beside to 200 g/tree Arbuscular Mycorrhizal fungi (AMF) significantly improved pear seedling growth, nutrient uptake, and rhizosphere microbial activity. This treatment (T₈) resulted in the highest seedling height (196.3 cm), shoot diameter (1.597 cm), leaf chlorophyll content (3.52 mg/g), soil microbial (total bacterial counts: 95×10^5 cfu/g; actinomycetes: 141×10^3 cfu/g; fungi 97×10^3 cfu/g), enzyme activity

(dehydrogenase: 106.55 µg TPF/g/day; phosphatase: 0.57 mg PNP/g/day) and soil macronutrients (N: 63.11 mg kg⁻¹; P: 21.91 mg kg⁻¹; K: 263.18 mg kg⁻¹). These findings highlight the effectiveness of integrated organic and biofertilization in reducing chemical inputs while enhancing seedling vigor and soil health.

Recommendation:

Therefore, applying 50% NPK + 50% compost and 200 ml/tree of mixture of *Azospirillum brasilense*, *Bacillus circulans* and *Trichoderma harzianum* beside to 200 g/tree Arbuscular Mycorrhizal fungi are recommended for optimal best growth of



young pear trees under conditions of this

trail.

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تأثير التسميد العضوي والحيوي في تعزيز النمو الخضري لشتلات الكمثرى صنف بساتين MKM تحت ظروف الزراعة الكثيفة

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أجريت التجربة خلال موسمين متتاليين (2024/2023 و 2025/2024) على 72 شتلة كمثرى (*Pyrus communis*) مطعومة على أصل *Pyrus betulaefolia* بعمر سنتين، مزرعة في تربة طينية بمزرعة المعهد البحثي

للبياتين بمسافه 1.5×2.0 م . هدف التجربة هو تقييم تأثير بدائل التسميد الكيميائي على النمو الخضري والكفاءة الغذائية للشتلات، من خلال دمج السماد العضوي والمخصبات الحيوية مع مستويات مخفضة من الأسمدة المعدنية (NPK). شملت المعاملات ثمانية توليفات تجمع بين مستويين من NPK (50 % و 100 %) من الجرعة الموصى بها، (ومستويين من الكمبوست) مكافئين لنفس النسب، بالإضافة إلى ثلاث مستويات من التسميد الحيوي باستخدام *Azospirillum*، *Trichoderma harzianum*، و *Bacillus circulans*، و *Trichoderma harzianum*، و *Bacillus circulans*، و *Azospirillum brasilense* (AMF). أظهرت النتائج أن المعاملة الثامنة "T8" NPK 50% + 50% كمبوست + 200 مل/شجرة من خليط من *Trichoderma harzianum* و *Bacillus circulans* و *Azospirillum brasilense* (AMF) كانت الأكثر كفاءة، حيث حسنت بشكل معنوي جميع الصفات المدروسة مثل الطول، القطر، عدد الأغصان، مساحة الأوراق، والكلوروفيل الكلي. كما ارتفعت تراكيز العناصر الغذائية نيتروجين، فوسفور، بوتاسيوم وكالسيوم في الأوراق، وزادت الأنشطة الميكروبية في التربة المحيطة بالجذور، حيث وصلت أعداد البكتيريا إلى 95×10^5 CFU/g والأكتينوميسيتات إلى 141×10^3 CFU/g والفطريات إلى 97×10^3 CFU/g، وبلغت أنشطة إنزيمات الديهيدروجيناز والفوسفاتيز 106.55 و 0.57 على التوالي. تُظهر الدراسة أهمية دمج التسميد العضوي والحيوي في تحسين نمو شتلات الكمثرى وتقليل الاعتماد على الأسمدة الكيميائية، مما يدعم ممارسات الزراعة المستدامة تحت الظروف المصرية.