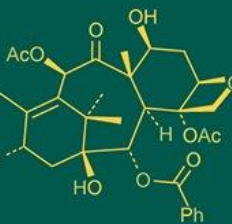
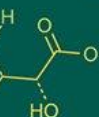
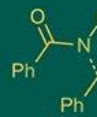


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Marwa Hassan Mayoof
 Department of Horticulture
 and Landscape, Faculty of
 Agriculture and Marshes,
 University of Thi-Qar, Iraq

The effect of organic nutrient spraying and biofertilizer on the nutrient content of sweet pepper (*Capsicum annuum* L.) leaves

Marwa Hassan Mayoof

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Abstract

The field experiment was conducted in one of the protected plastic greenhouses, using a Randomized Complete Block Design (RCBD) with two factors. The first factor was a nutrient solution at three concentrations (0, 2, and 4 g/L), and the second factor was a bio-fertilizer at three concentrations (0, 2, and 4 mL/L). The means of the treatments were compared using the Least Significant Difference (LSD) test at a significance level of 0.05. The results indicated that the treatment with organic nutrient spraying at a concentration of 4 g/L significantly outperformed others in terms of nitrogen percentage (2.643%), phosphorus percentage (0.5900%), and potassium percentage (2.0067%) in the leaves compared to the control treatment. Additionally, the bio-fertilizer spraying treatment at a concentration of 4 mL/L significantly yielded the highest values for nitrogen percentage (2.113%), phosphorus percentage (0.5133%), and potassium percentage (1.8478%) compared to the control treatment.

Keywords: *Capsicum annuum* L, biofertilizer, nutrient content, sweet pepper, control treatment

Introduction

Sweet pepper (*Capsicum annuum* L.) is one of the most important crops in the nightshade family (Solanaceae). It belongs to the genus *Capsicum*, which includes 20 to 30 species of peppers (Hassan, 2001) ^[5]. Sweet pepper requires a moderate climate that leans towards warmth and cannot tolerate cold temperatures significantly. The optimal temperature range for its growth is between 21-27 °C (Zidan *et al.*, 1977) ^[6]. Pepper can successfully grow under unheated protected cultivation conditions in some regions of the world by using early varieties. Each 100 grams of fresh pepper fruit contains (1.2 g) protein, (4.8 g) carbohydrates, (2.6 g) fiber, (0.8 g) fat, (29 mg) calcium, (2.6 mg) iron, and (61 mg) phosphorus (Grubben and Mohamed, 2004) ^[12]. The fruits of sweet pepper are an important source of vitamin C, which is essential for general health as it assists in bone formation and protein metabolism, stimulates appetite, and helps resist inflammation. They also contain vitamins B1, B2, and A, which are known antioxidants. The increase in the world population leads to a higher demand for food, which has resulted in significant attention to raising production rates regardless of quality. This has led to an increase in chemical additives, especially in vegetable crops compared to other crops due to their short growing season and high production and consumption rates. Consequently, this has exacerbated harmful effects on health and the environment through increased levels of nitrates, oxalates, and the toxic effects of pesticides (Othman, 2007) ^[7]. As a result of the growing concern over the contamination of food products with residues of fertilizers and pesticides, many countries worldwide have turned to organic farming systems, which rely on the use of natural resources instead of chemical additives to produce clean food. The harmful effects of fertilizers and pesticides can be mitigated by restoring soil fertility. The use of organic nutrients through foliar spraying is one of the best methods of nutrition, as research has shown that 85% of a plant's nutrient needs can be met through foliar application. Organic matter serves as a source of carbon for soil microorganisms and enhances the availability of nutrients for plants, positively impacting their growth and yield.

One of the key biotechnological techniques involves the addition of bio-fertilizers, which are preparations containing microorganisms capable of providing plants with essential nutrients from natural sources.

Corresponding Author:
Marwa Hassan Mayoof
 Department of Horticulture
 and Landscape, Faculty of
 Agriculture and Marshes,
 University of Thi-Qar, Iraq

This reduces reliance on various chemical fertilizers. These bio-fertilizers have the ability to continuously release nutrients, making them sufficient to meet the needs of the treated plants. The term "bio-fertilizers" broadly refers to products containing organisms and algae, either alone or in combination, which, when applied, help in fixing atmospheric nitrogen or dissolving and mobilizing soil nutrients (International Code of Conduct, 2019). Research indicates that humic acid has significant effects on carbon assimilation efficiency, increases cell membrane permeability, and enhances phosphorus absorption. This promotes the growth of plants and the availability of macronutrients and micronutrients by improving their readiness for uptake, thereby enhancing the absorption of elements such as potassium, calcium, phosphorus, and manganese. Mikkelsen (2005) [14] also mentioned these effects. In a study conducted by Abdel-Mawgoud *et al.* (2010) [9] on improving the growth and yield of sweet pepper (California Wonder variety) through the foliar application of nutrient, organic, and bio-solutions, it was found that the addition of the nutrient solution Stimufo at a level of 3 g/L resulted in the highest increase in potassium percentage in the plant leaves. Al-Marjani (2011) [3] noted that the addition of amino acids (Tyrosine, Proline, and Arginine) at a concentration of 200 mg/L to tomato plants resulted in the highest increase in nitrogen, phosphorus, and potassium content compared to the levels of 0 and 100 mg/L. This research aims to evaluate the effect of organic nutrients at a concentration of 4 g/L and the bio-fertilizer Biophilur at a concentration of 4 mL/L on the nutritional elements in the leaves.

Materials and Methods

Implementation of the experiment

The study was conducted in one of the greenhouses in Dhi Qar Governorate, using clayey loam soil to investigate the effects of organic nutrients and bio-fertilizers on the nutrient content of sweet pepper plants. Soil samples were collected from different areas of the greenhouse at a depth of 0-30 cm. The samples were air-dried, ground, and passed through a sieve with 2 mm openings. Chemical and physical analyses were conducted to determine various soil properties. Seeds were sown in plastic trays, with one seed per cell, and the trays were placed in a shaded area covered with green saran cloth. Agricultural operations were uniformly applied to all seedlings. Once the seedlings were ready for transplanting, they were moved to the permanent field in the greenhouse. All necessary operations were carried out to prepare the field for the cultivation of sweet pepper.

Experimental Treatments

This study was conducted using a Randomized Complete Block Design (R.C.B.D) with three replications, resulting in a total of 27 experimental units. It was designed as a factorial experiment with two factors. The treatments were randomly assigned to the experimental units for each of the three replications. The organic nutrient concentrations were as follows: (0) g/L, (2) g/L, and (4) g/L. The concentrations of the bio-fertilizer were: (0) mL/L, (2) mL/L, and (4) mL/L. The first application of the nutrients and bio-fertilizer

was done six weeks after planting, with a second application occurring 15 days after the first. Each treatment concentration was individually placed in a watering can for application. Data analysis was performed using the Genstat program (2011), and means were compared using the Least Significant Difference (L.S.D) test at a significance level of 0.05, as noted by Al-Rawi and Khalaf Allah (2000) [2].

Studied Traits

Five homogeneous plants were randomly selected from each experimental unit for all replications and marked for the required measurements. The study focused on estimating the percentage of nutrient elements (N, P, K) in the leaves. The percentages of nitrogen, phosphorus, and potassium were determined according to the method of Cresser and Parsons (1979) [10]. A weight of 0.2 g of the dry, ground sample was digested using sulfuric acid and perchloric acid to prepare the samples for measuring the nutrient content.

Total Nitrogen Content in Leaves (%)

The total nitrogen content in the leaves was estimated using the Micro Kjeldahl distillation method, as described by Page *et al.* (1982). The percentage of nitrogen was calculated using the following equation:

$$N = (1000 \times \text{Weight of digested sample} \times \text{Volume of the taken sample})$$

$$(\text{Final dilution volume} \times 14 \times \text{Titration acid normality} \times \text{Volume of consumed HCl})$$

Total Phosphorus Content in Leaves (%)

The phosphorus content in the leaves was estimated colorimetrically using the method described by Murphy and Riley (1962), known as the Ascorbic Acid method. This procedure involves the use of ammonium molybdate and ascorbic acid. After the blue color developed, the intensity of the color in the solution was measured using a UV-700 spectrophotometer at a wavelength of 700 nanometers. This measurement allows for the quantification of the phosphorus content in the leaf samples based on the color intensity.

Total Potassium Content in Leaves (%)

The potassium content in the leaves was estimated using a flame photometer, following the method described by Page *et al.* (1982). The results were expressed based on a standard curve created using potassium chloride. The data were analyzed according to the Randomized Complete Block Design (R.C.B.D.). Statistical analysis of the results was performed using the GenStat statistical software to conduct analysis of variance (ANOVA). The Least Significant Difference (L.S.D) test was employed to compare the means of the treatments at a significance level of 0.05, as noted by Al-Rawi and Khalaf Allah (2000) [2].

Results and Discussion

Percentage of nitrogen in leaves (%)

The results in Table 1 show the effect of adding organic nutrients and biofertilizer on the percentage of nitrogen in the leaves. The organic nutrient treatment significantly exceeded with a concentration of (4 g/L^{⁻¹}), providing the highest percentage of nitrogen.

Table 1: The effect of adding organic nutrients and biofertilizer, and their interaction, on the percentage of nitrogen in leaves (%)

Average Organic Nutrient	Biofertilizer			Organic Nutrient
	Concentration		Concentration (0ml/L)	
(4 mL/L)	Concentration (2ml/L)	Concentration	0.213	gm/L ⁻¹ 0
(0ml/L)			1.287	gm/L ⁻¹ 2
0.599	1.273	0.310	2.390	gm/L ⁻¹ . 4
1.727	2.157	1.737	1.263	Average Biofertilizer
2.643	2.910		2.730	L.S.D
	2.113		1.592	

In the leaves, the nitrogen percentage reached 2.643%, compared to the lowest nitrogen percentage of 0.599% in the control treatment. The same table indicates that the biofertilizer had a significant effect on this trait, as the treatment at a concentration of (4 mL·L⁻¹) significantly exceeded and provided the highest nitrogen percentage in the leaves at 2.113%, compared to the lowest nitrogen percentage of 1.263%. Moreover, the interaction between the studied factors had a significant effect on this trait, as the treatment with organic nutrient (4 g·L⁻¹) and biofertilizer at a concentration of (4 mL·L⁻¹) yielded the highest nitrogen percentage in the leaves at 2.910%, while the control treatment with organic nutrient and biofertilizer gave the lowest nitrogen percentage of 0.213%.

Percentage of Phosphorus in Leaves (%)

The results in Table (2) show the significant effect of adding organic nutrients on the percentage of phosphorus in the

leaves. The organic nutrient treatment at a concentration of (4 g·L⁻¹) yielded the highest phosphorus percentage in the leaves at 0.5900%, compared to the lowest phosphorus percentage of 0.2900% in the control treatment. The same table also indicates that the biofertilizer had a significant effect on this trait, as the concentration of (4 mL·L⁻¹) significantly exceeded and provided the highest phosphorus percentage in the leaves at 0.5133%, compared to the lowest phosphorus percentage of 0.3700%. The interaction between the studied factors had a significant effect on this trait, as the treatment with organic nutrient (4 g·L⁻¹) and biofertilizer at a concentration of (4 mL·L⁻¹) yielded the highest phosphorus percentage in the leaves at 0.6833%. In contrast, the control treatment with organic nutrient and biofertilizer produced the lowest phosphorus percentage of 0.2400%.

Table 2: The effect of adding organic nutrients and biofertilizer, and their interaction, on the percentage of phosphorus in leaves (%)

Average Organic Nutrient	Biofertilizer			Organic Nutrient
	Concentration		Concentration (0ml/L)	
(4 mL/L)	Concentration		0.2400	gm/L ⁻¹ 0
(0ml/L)	Concentration		0.3867	gm/L ⁻¹ 2
(0ml/L)			0.4833	gm/L ⁻¹ . 4
0.2900	0.3467	0.2833	0.3700	Average Biofertilizer
0.4456	0.5100		0.4400	L.S.D
0.5900	0.6833		0.6033	

Percentage of Potassium in Leaves (%)

The results in Table (3) show the significant effect of adding organic nutrients and biofertilizer on the percentage of potassium in the leaves. The organic nutrient treatment at a concentration of (4 g·L⁻¹) significantly exceeded and provided the highest potassium percentage in the leaves at 2.0067%, compared to the lowest potassium percentage of 1.4249% in the control treatment. The same table indicates that the biofertilizer also had a significant effect, as the biofertilizer treatment at a concentration of (4 mL·L⁻¹) significantly exceeded and yielded

the highest potassium percentage in the leaves at 1.8478%, compared to the lowest potassium percentage of 1.5797% in the control treatment. Moreover, the interaction between the studied factors had a significant effect on this trait, as the treatment with organic nutrient (4 g·L⁻¹) and biofertilizer at a concentration of (4 mL·L⁻¹) yielded the highest potassium percentage in the leaves at 2.7967%. In contrast, the control treatment with organic nutrient and biofertilizer produced the lowest potassium percentage of 1.2657%.

Table 3: The effect of adding organic nutrients and biofertilizer, and their interaction, on the percentage of potassium in leaves

Average Organic Nutrient	Biofertilizer			Organic Nutrient
	Concentration		Concentration (0ml/L)	
(4 mL/L)	Concentration		1.2657	gm/L ⁻¹ 0
(0ml/L)	Concentration		1.5833	gm/L ⁻¹ 2
(0ml/L)			1.8900	gm/L ⁻¹ . 4
1.4249	1.6400	1.3690	1.5797	Average Biofertilizer
1.7022	1.7967		1.7267	L.S.D
2.0067	2.1067		2.0233	

It is evident from Tables (1, 2, and 3) that the addition of organic nutrients has a significant effect on the content of

leaves in nitrogen, phosphorus, and potassium. Organic nutrients play an effective role in providing essential

elements for plants (N, P, K) by preventing the appearance of deficiency symptoms of these elements in the plants. Additionally, organic nutrients have an impact on the percentage of macronutrients and the content of micronutrients in the leaves. Singer *et al.* (1998) noted that humic acid makes nutrients more available to plants and encourages the absorption of elements such as potassium, phosphorus, and calcium. Conversely, fulvic and humic acids have the ability to retain mineral elements in complexes and in chelated forms. The improvement in vegetative growth indicators is attributed to the role of these organic fertilizers in supplying the plant with both macronutrients and micronutrients, as well as enhancing the biological properties that increase the availability of these elements (Weil & Magdoff, 2005) ^[13]. It is evident from the tables that the application of biofertilizer has an impact on nitrogen, phosphorus, and potassium. The superiority of biofertilization here is attributed to its positive role in nutrient absorption, particularly nitrogen. This is due to the vital role of nitrogen in the formation of amino acids, cellular membranes, and vitamins, including the B vitamin group, which collectively contribute to increased plant height and the number of leaves. The superiority of plants treated with biofertilizer, nitrogen, phosphorus, and potassium is evident. This response to biofertilization is due to its content of various types of microorganisms that fix nitrogen and solubilize phosphorus and potassium, which play a significant and impactful role in the availability of nutrients to plants. This is achieved through the secretion of growth regulators, organic acids, and chelating compounds that lead to increased concentrations of various elements.

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