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Effect of Foliar Spraying with Some Commercial Bio-Stimulants on Physiological Responses of Eggplant (*Solanum melongena* L.)

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تأثير الرش ببعض المحفزات الحيوية التجارية على الاستجابات الفسيولوجية لنبات الباذنجان

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Abstract

This study was undertaken to determine the effects of two bio-stimulants (Humic Power and Amino 24) on the growth and nutrient contents in eggplant (*Solanum melongena* L.) cv. Master F1 was grown under greenhouse conditions using a randomized complete block design with three replications. Different levels of Humic Power were applied (control, 3 mL/L and 5 mL/L) and Different levels of Amino 24 (control, 1 mL/L and 2 mL/L) were sprayed on the foliar every 15 days throughout the study period. The highest vegetative growth was obtained with eggplant supplemented with foliar Humic Power (HP.2) with Amino 24 (AM.2), followed by plants treated with Humic Power (HP.1) with Amino 24 (AM.2) compared to untreated plants. There was a significant interaction between the bio-stimulants (Humic Power and Amino 24) for the content of macro elements in eggplant plants, except for the potassium content, where the difference was non-significant. On the other hand, the highest content of macro elements (NPK) was obtained at Humic Power (HP.2) and Amino 24 (AM.2), respectively as compared to the control.

***Keywords**: Eggplants, Bio-Stimulants, Solanum melongena L., Humic acid, Amino Acid, Physiological responses*.

الملخص

أجريت هذه الدراسة لتحديد تأثير اثنين من المنشطات الحيوية (Humic Power وAmino 24) على النمو ومحتوى المغذيات في نبات الباذنجان (*Solanum melongena. L*) صنف Master F1 المزروع تحت ظروف الصوبة البلاستيكية، استخدم في التجربة تصميم القطاعات كاملة العشوائية بثلاث مكررات. كما تم استخدام مستويات مختلفة من المنشط الحيوي Humic Power هي (الشاهد، 3 مل/لتر، 5 مل/لتر) كما استخدمت مستويات مختلفة من منشط النمو الحيوي Amino 24 هي (الشاهد، 1 مل/لتر، 2 مل/لتر) رشا على المجموع الخضري كل 15 يومًا طوال فترة الدراسة. سجلت أعلى زيادة في صفات النمو الخضري (ارتفاع النبات، عدد الأوراق لكل نبات، مساحة الورقة، محتوى الكلوروفيل الكلي عند المعاملة (HP.2) من Humic Power مع المعاملة (AM.2) من منشط النمو الحيوي Amino 24، تليها النباتات المعاملة بـ (HP.1) مع (AM.2) مقارنة بالنباتات غير المعاملة. من ناحية أخرى، كان هناك تداخل معنوي بين المنشطات الحيوية (Humic Power وAmino 24) لمحتوى العناصر الغذائية في نباتات

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الباذنجان باستثناء محتوى البوتاسيوم حيث كان الفرق غير معنوي. تم الحصول على أعلى محتوى من النيتروجين والفسفور والبوتاسيوم مع المعاملة (HP.2) والمعاملة (2.AM) على التوالي بالمقارنة مع نباتات الشاهد.

الكلمات الدالة: الباذنجان، المحفزات الحيوية، حمض الهيومك، حمض أميني، استجابات فسيولوجية.

1. Introduction

Eggplant (*Solanum melongena* L.) is considered one of the most important cash crops belonging to the Solanaceae family (Hassan *et al*., 2019). The various eggplant cultivars exhibit a wide range of fruit colors and shapes, from white, yellow, and green to nearly black, and from oval or egg-shaped to long club-shaped. It is farmed in some warm temperate regions of the Mediterranean and South America, as well as in Asia, Africa, and the subtropics (India, Central America), where it is an economically significant crop (Azarpour *et al*., 2012). Fruits from eggplants are renowned for having few calories and a mineral makeup that is good for human health. According to Michaelojc and Buczkowska (2008), they are also good sources of potassium, magnesium, calcium, and iron. According to Alicja *et al.* (2019), eggplant is a important source of polyphenols, vitamins, and minerals with anti-inflammatory, anti-microbial, hepatoprotective, and cardioprotective characteristics.

Among the organic components used as fertilizer additives, bio-stimulants improve nutrient uptake, encourage plant development, and boost plant tolerance to biotic and abiotic challenges. These stimulants are useful for both horticultural and crops (Drobek *et al*., 2019; Elhadi *et al*. 2022). Humic and fulvic acids, protein hydrolysates, nitrogen-containing substances, seaweed extracts, helpful fungi, and bacteria are the key ingredients used in these formulations (Du-Jardin, 2015). In place of pesticides, bio-stimulants are increasingly being used in agricultural production. Many different plants are employed to produce stimulatory effects (Abbas and Hussain, 2020). Bio-stimulant application in crops is intensively investigated as the pro-ecological solution for modern agriculture. The concept of bio-stimulants was formulated at the end of the 20th century and has been continuously developed (Brown and Saa, 2015) Although the composition of a bio-stimulant may be based on single or many components, the synergistic activity of many different components has been documented (Hassan *et al*., 2019). To promote the efficiency of nutrient utilization, tolerance to abiotic stress, and quality features of plants regardless of the nutritional content, bio-stimulants are preparations containing a substance(s) and/or microorganisms designed for use on a plant or root zone. Commercial goods containing combinations of such chemicals and/or microbes are also included in this group (Du Jardin, 2015). Through its impact on numerous physical, chemical, and biological aspects of soil, humic acid has a crucial, indirect function in enhancing plant growth (Usharani *et al*., 2019). Its effects on cell membrane permeability, water and nutrient absorption, respiration, photosynthesis, root cell growth and elongation, seed germination rate, protein synthesis, and enzyme activity are indicative of its direct role (Russo and Berly, 1990). The majority of the elements that raise soil fertility and nutrient availability are found in humic acid, a commercially available organic fertilizer that boosts plant

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development and output while reducing the negative effects of stressors (Doran *et al*., 2003). Humic acid, according to Adani *et al*. (1998), is thought to improve nitrogen utilization efficiency and therefore boost the growth of shoots and roots. The main application of humic compounds is to reduce or eliminate the harmful effects of chemical fertilizers from the soil, which has a significant impact on plant growth (Ghabbour and Davies, 2001). Humic compounds increase nutrient uptake across the cell membrane and into the roots of plants (El-Nemr *et al*., 2015). Improved water holding capacity, pH buffering, and thermal insulation are just a few of the features of soil that are improved by humic acid, a stable portion of carbon (McDonnell *et al*., 2001). Humic acid absorbs both minor and major elements, activates or inhibits enzymes, alters membrane permeability, and activates the formation of biomass, which promotes the growth of plants (El-Ghamry *et al*., 2009). Humic acids have been demonstrated to stimulate plant growth and subsequently yield by acting on mechanisms related to cell respiration, photosynthesis, protein synthesis, water and nutrient uptake, and enzyme activities. The primary fractions of humic substances and the most active parts of organic matter in soil and compost are humic acids (Chen, 2004). Amino acids are bio-stimulants that have favorable effects on plant growth, and yield, and greatly lessen the harm brought on by abiotic stressors (Kowalczyk and Zielony, 2008). It is commonly recognized that consuming enough necessary amino acids would improve the yield and general quality of corps (Shaheen *et al*., 2010). The current study's objective was to examine the impact of foliar spraying eggplant plants with various concentrations of bio-stimulants (Amino 24 and Humic power) on their growth and productivity.

2. Materials and Methods

2.1. Plant material, growth conditions, and treatments

The factorial experiment was carried out in a greenhouse at (lat. 32 52'N, long 14 12'E, altitude 254 m above sea level) in the fall of 2022 using a randomized complete block design with three replications. Eggplant (*Solanum melongena* L.) cv. Master F1 was the vegetable species under the test of the current experiment. The daily air temperature in the greenhouse and natural lighting were used to cultivate eggplant plants. Three different concentrations of bio-stimulant (Humic power) were used (control, 3 and 5 mL/L), as well as three different concentrations of bio-stimulant (Amino 24) were used in this experiment (control, 1 and 2 mL/L), were sprayed on the foliar every 15 days throughout the study period. The list of the chemical compositions employed by the bio-stimulants that were used is shown in (Table 1).

Seedlings were planted directly in the greenhouse soil, and two weeks after planting, the treatments were sprayed on the foliar according to the specified concentrations for 60 days from planting. Various agricultural operations such as irrigation, weed removal, and pest control were carried out as recommended by the Ministry of Agriculture.

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Table 1. Chemical composition of applied bio-stimulants.

Humic power (w/v)	Amino 24 (w/v)
Total Organic Matter 55%	Organic Matter 45%
Humic Acid 31.2%	Free Amino Acid 24%
Water Soluble Potassium Oxide (K_2O) 6.5%	Organic Nitrogen 4%
Organic Nitrogen 2.6%	Organic Carbon 12%
Free Amino Acids 2%	pH 4-6
Total Micronutrients 1%	
pH 4-6	

2.2. Measurements for the vegetative stage

Vegetative measurements and the number of fruits were taken 60 days after seedling cultivation, including those of the eggplant plant height, number of leaves, leaf area, and number of branches per plant. Also, the total chlorophyll, nitrogen (N), phosphorus (P), and potassium (K^+) concentrations of the leaves were measured.

2.3. Experimental design and statistical analysis

The data show the average results of an independent experiment conducted in the 2022 fall season. Three replicates were used for each treatment in the experiment, which used a factorial experimental design (3x3) in a randomized complete block design. The treatments included various concentrations of the bio-stimulants (Humic Power and Amino 24). Using the statistics 10 software program, data were subjected to analysis of variance using a two-way ANOVA test, and means were compared using the Duncan means test (P= 0.05).

3. Results and Discussion

3.1. Changes in vegetative growth and morphological characteristics

Vegetative growth parameters, including plant height, number of leaves, leaf area, and Number of branches/plants, were significantly affected by Humic Power (Table 2), treatment Amino 24 (Table 3), and the interaction of Humic Power X Amino 24 (Fig.1).

The application of Humic Power affected positively the plant height, the number of leaves, leaf area, and the number of branches for each plant compared to the control (Table 2). Similar results were also reported by (Dursun and Turan, 2002). While there was no significant difference between the concentrations of Humic Power (HP.1 and HP.2) in all vegetative measurements that were carried out on eggplant plants, except for the leaf area, where there was a significant difference between the concentrations of Humic Power.

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Table 2. Effect of bio-stimulant Humic Power on plant height, leaves number, leaf area, number of branches/plant of Eggplant plants

Humic Power	Plant height (cm)	Leaves number	Leaf area (cm^2)	Number of branches/plant
Con.	67.3[b]	71.9[b]	162.4[c]	3.21[b]
HP.1	72.1[a]	80.2[a]	186.7[b]	3.84[a]
HP.2	75.4[a]	80.6[a]	191.0[a]	3.94[a]

Means followed by the same letter in each column are not significantly different by Duncan's multiple range test at 5% level. CON= control, HP.1= Humic Power (3 mL/L), HP.2= Humic Power (5 mL/L).

Results in Table (3) show that the average plant height, leaves number, leaf area, and number of branches per plant eggplant were influenced by the bio-stimulant (Amino 24). Where there were significant differences between the concentrations of Amino 24 (AM.1 and AM.2) in all vegetative measurements that were carried out on eggplant plants compared to the control treatment. Table (3) also shows that the third treatment (AM.2) was significantly superior to the second treatment (AM.1) in all vegetative measurements that were conducted. Similar results were also reported by (Haghighi *et al*., 2020).

Table 3. Effect of bio-stimulant Amino 24 on plant height, leaves number, leaf area, number of branches/plant of eggplant plants

Amino 24	Plant height (cm)	Leaves number	Leaf area (cm^2)	Number of branches/plants
Con.	66.5[c]	70.5[c]	157.4[c]	2.91[c]
AM.1	71.8[b]	79.3[b]	179.6[b]	3.57[b]
AM.2	76.5[a]	82.8[a]	203.1[a]	4.51[a]

Means followed by the same letter in each column are not significantly different by Duncan's multiple range test at 5% level. CON= control, AM.1= Amino 24 (1 mL/L), AM.2= Amino 24 (2 mL/L).

Figure (1) shows the effect of the interaction between Humic Power and Amino 24 on plant height, leaves number, leaf area, and number of branches per plant eggplant. There was a significant interaction between the bio-stimulants (Humic Power and Amino 24) for vegetative growth parameters except for plant height (Table 6). Maximum vegetative growth was obtained with eggplant plants supplemented with foliar Humic Power (HP.2) with Amino 24 (AM.2), followed by plants treated with Humic Power (HP.1) with Amino 24 (AM.2) compared to untreated plants (Fig.1 A, B, C, and D).

The plant height of eggplant plants supplemented with Humic Power bio-stimulant (HP.2) with Amino 24 bio-stimulant (AM.2) increased by 26.82%, followed by Humic Power (HP.1) with Amino 24 (AM.2) by 23.3% compared to untreated plants (Fig.1A). Whereas, the number of eggplant leaves with Humic Power (HP.1) bio-stimulant with Amino 24 (AM.2) bio-stimulant increased by 28.4%, followed by Humic Power (HP.2) with Amino 24 bio-stimulant (AM.2) by 27.2%. compared to control plants (Fig.1B). Similar results were obtained in the leaf area, where the highest value was in the third treatment of humic growth bio-stimulant with the

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third treatment of Amino 24 bio-stimulant as it increased by 49.5%, followed by the second treatment Humic Power with the third treatment of amino 24 bio-stimulant by 48.7% compared to untreated plants (Fig. 1C). The results showed in (Fig. 1D) that the number of branches per plant increased by 77.4 % in the plants that were sprayed foliar with Domeic (HP.1) with Amino 24 (AM2). Followed by plants of the third treatment of Amino 24 with the second treatment of Humic Power as it increased by 75.9 % compared to untreated plants (Fig. 1D).

The best results of vegetative growth parameters were noted with Eggplant plants applying foliar with all treatments of Humic Power bio-stimulant and Amino 24 bio-stimulant. This improvement may be correlated to the higher amount of free amino acids, humic acids, and organic matter, essential elements of bio-stimulants (Table 1).

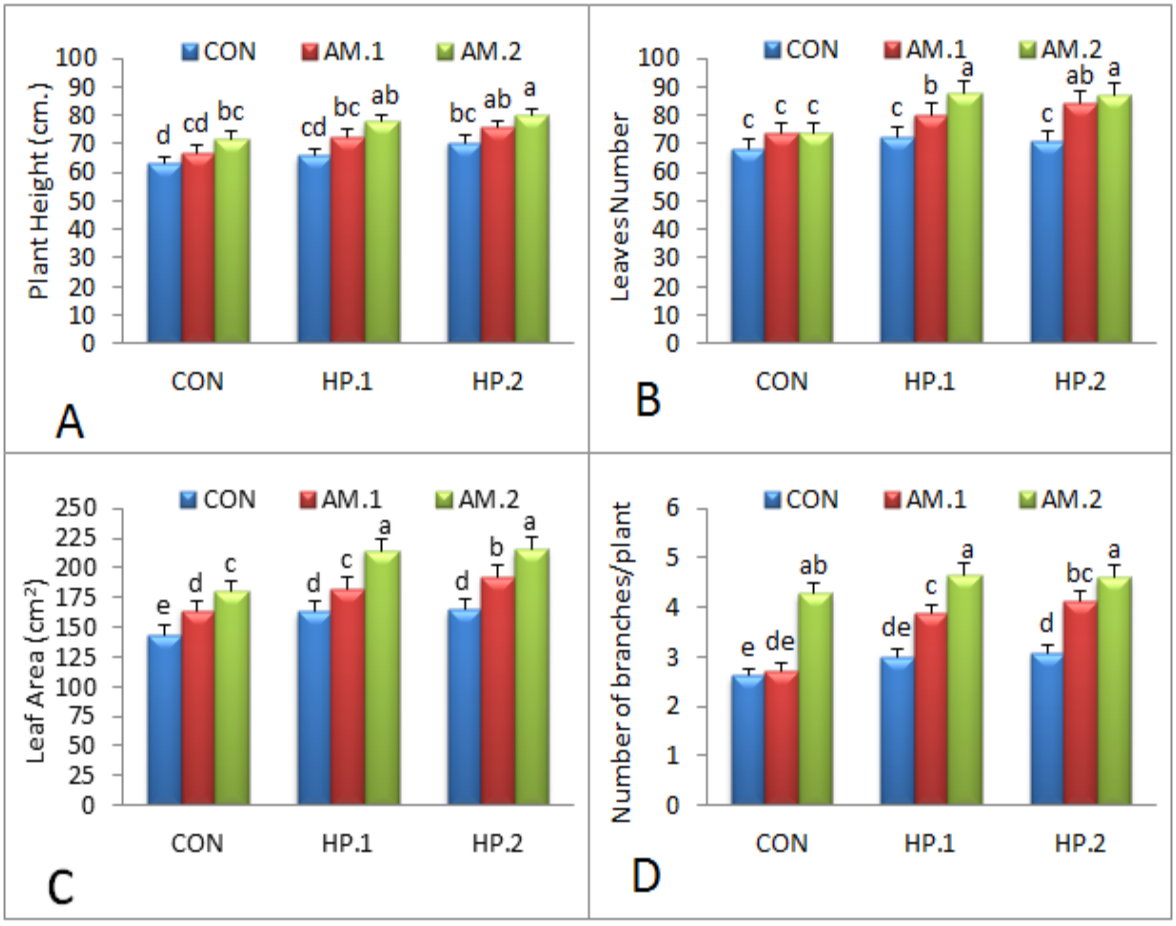

Figure 1. Effect of interaction between bio-stimulants Humic Power and Amino 24 on plant Hight (cm) Leaves number, Leaf Area (cm^2), number of branches per plant of eggplant.
Mean followed by the same letter in each column are not Significantly different by Duncan Multiple range test at 5% level. CON= control, HP.1 (3 mL/L), HP.2 (5 mL/L), Am.1= Amino 24 (1 mL/L), Am.1= Amino 24 (2 mL/L).

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3.2. Effect of bio-stimulators on the total chlorophyll and chemicals contents of eggplant plants

The results in Table (4) indicated an increasingly significant in the total chlorophyll in leaves of eggplant plants in both levels of bio-stimulant (Humic Power) compared with control plants. The effects of different levels of bio-stimulant (Humic Power) on N, P, and K contents in leaves of eggplant plants are shown in Table (4). Results showed that the total chemical of N, P, and K increased with increasing the amount of humic power level (3 and 5 mL/L) respectively compared to untreated plants. The obtained results are supported by Dursun and Turan (2002) and El-Nemr *et al*. (2015).

Table 4. Effect of bio-stimulant Humic Power on the total of chlorophyll, the content of nitrogen (N), phosphorus (P), and potassium (K^+) of Eggplant

Humic Power	Chlorophyll mg/g	Nitrogen %	Phosphorus %	Potassium %
Con.	13.51^c	3.24^c	0.45^c	2.22^c
HP.1	16.13^b	4.61^b	0.51^b	2.57^b
HP.2	20.81^a	5.01^a	0.57^a	2.84^a

Means followed by the same letter in each column are not significantly different by Duncan's multiple range test at 5% level. CON= control, HP.1= Humic Power (3 mL/L), HP.2= Humic Power (5 mL/L).

The results show in Table (5) the effect of the commercial growth stimulant Amino 24 on the leaves' content of total chlorophyll and the elements of nitrogen, phosphorus, and potassium, where the two treatments of Amino 24 had a significant effect on the leaves content of chlorophyll and elements (NPK) compared to the untreated plants, as the third treatment increased (5 mL/L) significantly compared to the second treatment (3 mL/L). Similar results were also reported by El-Nemr *et al*. (2015).

Table 5. Effect of bio-stimulant Amino 24 on the total of chlorophyll, the content of nitrogen (N), phosphorus (P), and potassium (K^+) of eggplant

Amino 24	Chlorophyll mg/g	Nitrogen %	Phosphorus %	Potassium %
Con.	12.55^c	3.24^c	0.34^c	2.28^c
AM.1	16.02^b	4.26^b	0.55^b	2.57^b
AM.2	21.87^a	5.37^a	0.65^a	2.78^a

Means followed by the same letter in each column are not significantly different by Duncan's multiple range test at 5% level. CON= control, AM.1= Amino 24 (1 mL/L), AM.2= Amino 24 (2 mL/L).

Figure (2) shows the effect of the interaction between Humic Power and Amino 24 on the content of total chlorophyll, nitrogen (N), phosphorus (P), and potassium (K^+) in eggplant. There was a significant interaction between the bio-stimulants (Humic Power and Amino 24) for the total chlorophyll content and chemicals in eggplant plants, except for the potassium content, where the difference was non-significant at (P= 0.05) as in Table (6).

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Table 6. Analysis of variation for the variables; plant height (PH), Leaves number (LN), Leaf Area (LA), number of branches (BN), total chlorophyll (CH), content of nitrogen (N), Phosphorus (P), potassium (K) of Eggplant as function of Humic Power and Amino24

Source of Variation	DF	Mean Square							
		PH (cm)	LN (No)	LA (cm^2)	BN (No/Plant)	CH (mg/g)	N (%)	P (%)	K (%)
Blocks	2	2.386	15	12.40	0.08	0.12	0.016	0.0014	0.014
Humic Power	2	152.031*	217*	2135.60*	1.42*	122.80	7.740*	0.0324*	0.884*
Amino 24	2	224.300	359*	4688.80*	8.85*	199.72	10.204*	0.2287*	0.579*
Humic Power + Amino24	4	2.544ns	32*	75.86*	0.27*	11.87*	0.252*	0.0131*	0.0147ns
Error	16	16.051	9	12.95	0.05	3.87	0.036	0.0011	0.00568
CV%	-	5.59	4	2.00	5.86	11.69	4.41	6.40	2.96

CV= Coefficient of Variation; *=Significant at 0.05 probability; ns =Not Significant

Figure (2A) shows an increase in the total chlorophyll content significantly in the third treatment of Humic Power with the third treatment of Amino24, where its value was (28.05 mg/g), followed by the second treatment of Humic Power with the third treatment of Amino24, where its value was (21.04 mg/g) compared to untreated plants that recorded (9.38 mg/g). While the nitrogen content of eggplant plants with Humic Power (HP.1) with Amino 24 (AM.2) increased significantly, recording (6.02%), followed by Humic Power (HP.2) with Amino 24 (AM.2) as it reached its value (5.91%) compared to control plants (2.12%) (Fig. 2B). The phosphorous content of Humic Power (HP.1) with Amino 24 bio-stimulation (AM.2) increased significantly, recording a value of (3.01%), followed by Humic Power (HP.2) with Amino 24 bio-stimulation (AM.2), its value was (2.94%) compared to the control plants, which recorded the lowest value (1.95%), as in (Fig. 2C). This agrees with both Kowalczyk and Zielony (2008) and El-Nemr *et al*. (2015) where Bio-stimulants are thought to boost the effectiveness of how quickly nitrogen is absorbed, which in turn encourages the growth of roots and shoots. Additionally, they discovered that bio-stimulant substances have a significant impact on plant growth and are primarily used to reduce or eliminate the negative effects of chemical fertilizers from the soil. These substances also promote greater uptake of nutrients into the plant root and through the cell membrane.

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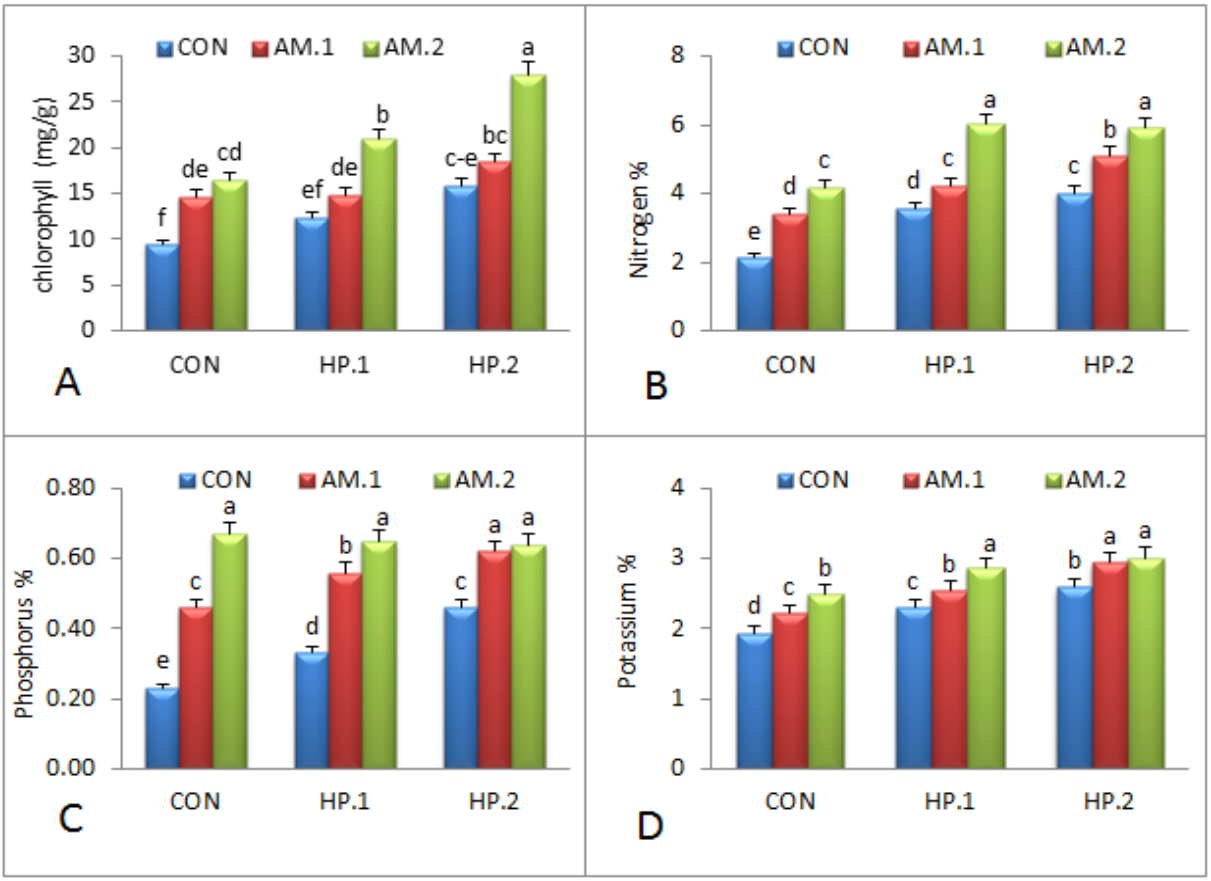

Figure 2. Effect of interaction between bio-stimulants Humic Power and Amino 24 on total chlorophyll (mg/g), content of nitrogen (%), Phosphorus (%), potassium (%) of eggplant.
Mean followed by the same letter in each column are not Significantly different by Duncan Multiple range test at 5% level. CON= Control, HP.1 (3 mL/L), HP.2 (5 mL/L), Am.1=Amino 24 (1 mL/L), Am.1=Amino 24 (2 mL/L).

4. Conclusion

The main conclusion from this research that using commercial bio-stimulants sprayed on shoots has a positive effect on vegetative growth characteristics) plant height, leaves number, leaf area, number of branches per plant, and a total of chlorophyll) and leaf nutrient content from (NPK). This may be due to the role of humic acid in increasing the uptake of nutrients from the soil very effectively, and the availability of amino acids helps in the formation of proteins that plants need for growth and development. The results of this research showed a significant effect of the interaction between the bio-stimulants Humic Power and Amino 24 in most of the growth characteristics studied, which enhanced the plant's ability to utilize the nutrients in the soil solution. Therefore, it is recommended to use them together at the specified rates to obtain the best characteristics of vegetative growth, which positively affects the overall productivity of the eggplant.

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