Effect of spraying with tryptophan and seaweed extract on some vegetative and chemical traits and leaf content of the active compounds of papaya seedlings.

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Abstract

The study was conducted in the nursery belonging to the private nursery in the Al-Hafiz area in the holy governorate of Karbala for the period from 1/5/2021 to 1/11/2021. To study the effect of spraying with T-tryptophan at a concentration of (0,50,100 mg/L-1) and seaweed extract (Agazone) at a concentration of(0.5, ,10 ml/L^{-1}) and their interactions on vegetative traits and leaf content of the active compounds of papaya seedlings at the age of 6 months were chosen. 81 seedlings of homogeneous growth as much as possible, A factorial experiment was conducted according to the Randomized Complete Block Design R.C.B.D with three replicates, the results were analyzed statistically and the averages were tested by testing the least significant difference LSD at the 0,05 level. The results can be summarized as follows:

1- The treatment of spraying with T2 tryptophan was excelled in most of the studied traits, such as stem diameter (47.6 mm), leaf area (226 cm^2).As well as the percentage of nitrogen element (4.76 %), phosphorous (0.41 %,) total chlorophyll content of leaves (70.68 mg 100 g $^{-1}$fresh weight), and flavonoid content of leaves (46.56 µg/g).

2- Treatment S2 of seaweed extract was excelled in stem height (47.1 cm), stem diameter (53.7 mm), number of leaves (5.79) leaves.$plant^{-1}$, percentage of nitrogen element 5.54%, leaf area(762 cm^2 ,The percentage of phosphorous element (0.51%), percentage of potassium element (5.31%), the content of the leaves from the total chlorophyll (73.00 mg 100 g 1-fresh weight), the content of the leaves of flavonoids (43.02 µg / g).

3- The T2S2 interaction treatment achieved the highest values in the nitrogen content of the leaves 5.67% (5.67%), phosphorous(0.56%), potassium (5.39%) and flavonoids (48.05 µg/g), and the T1S2 interaction treatment had a significant effect on stem height (54.1 cm). The average number of leaves (6.83 leaf.$plant^{-1}$), the average total leaf area (885 cm^2), and the content of leaves from chlorophyll (76.44 mg 100 gm^{-1} fresh weight).

Introduction

Papaya belongs to the Caricaceae family, (*Carica Papaya L.*) It is a tropical evergreen fruit, and it is believed that it originated on the Caribbean coast of Central America as a result of natural crossbreeding [1] Mexico and the West Indies are its original homeland. Papaya is of great economic and nutritional importance due to its early and year-round fruiting, as well as the palatable taste of the fruits. The global market demand for tropical fruits has increased steadily over the first two decades of the twenty-first century, and global production of papaya in 2017 amounted to about 13,016.281 tons. India is the largest producer of this fruit in the world, with a production volume of about 5,699,000 tons annually, while Brazil comes In the second place, with an annual production of about 1,424,650 tons. Thus, India and Brazil together produce more than half of the world's production [2]. Tryptophan is a heterocyclic acid that has a positive role in stimulating growth, increasing production and improving

soil bacteria, which facilitates the absorption of nutrients and makes the plant more resistant and withstand environmental stresses.This acid is the basic building block for the formation of auxin[3]. It was found [4] that spraying tryptophan on citrus trees at a concentration of 100 mg.L-1 led to an increase in leaf area.It was noted [5] when spraying tryptophan on seedlings of tangerine (*Citrus reticulate* L.) Clementine cultivar grafted on the rootstock of Citrus aurantium every two weeks at concentrations of 100,50.0 mg. L^{-1} Significant increase in most vegetative and root growth traits.It was found that [6] [7] spraying tryptophan with different concentrations on thua and mango seedlings led to a significant increase in the leaves content of the mineral elements NP K seaweed extracts are one of the important organic sources used in agricultural production. They are a complement to, and not a substitute for, fertilizers. These extracts stimulate physiological functions in the plant because they contain macro and micro nutrients and contain many growth-stimulating substances such as cytokinins, auxins, gibberellins, and organic acids [8] . It was shown [9] that spraying the seedlings of (*Eriobotrya japonica* L.)at 1 year old with seaweed extract (Kelpak) at a concentration of 4 ml. L^{-1}It showed a significant increase in seedling height, number of leaves and leaf area compared to the control treatment.The concentration of 10 ml. L^{-1} of seaweed extract recorded the highest rate in stem height and diameter, number of leaves and leaf area of banana plant compared to the control treatment [10].The concentration of 3 ml. L^{-1} of seaweed extract Algo600 achieved a significant increase in the content of strawberry leaves of nutrients NPK [11], and when spraying olive seedlings with seaweed extract, a significant increase in the content of chlorophyll leaves was obtained at a concentration of 2 ml. l^{-1} [12].In view of the lack of studies related to this type of fruit and in order to obtain plants with a strong structure and rapid vegetative growth and to fill their needs of nutrients, this study was conducted to show the effect of treatment with different concentrations of each of the amino acid tryptophan and the extract of seaweed Agazone and their interactions in the vegetative traits and the content of leaves from the active compounds for papaya seedlings.

Materials and Method

The study was coundected in the private nursery located in Al-Hafiz area in the holy governorate of Karbala for the period from 1/5/2021 to 1/11/2021. On papaya seedlings of 6 months old growing in an agricultural medium consisting of 3 soil: 1 with moss inside anvils with a diameter of 17 cmTo study the effect of spraying with Tryptophan and seaweed extract (Agazone) and their interactions on the vegetative traits and leaf content of the active compounds of papaya seedlings.The homogeneous seedlings were selected as much as possible, taking into account the conduct of service operations when needed. A factorial experiment was carried out with two factors (3 × 3) and with three replications. Each experimental unit included three seedlings, thus the total number of seedlings became 81 seedlings using the Randomized Complete Block Design (RCBD) design in the implementation of the experiment. The data were analyzed according to the Excel program, and the arithmetic averages were compared with the LSD test at a probability level of 0.05.The first factor: the amino acid tryptophan, symbolized by the symbol T, and at three levels: T0, T1, T2 (0,50. 100 mg.L^{-1}), the second factor: seaweed extract, symbolized by the symbol S, and at three levels: S0, S1, S2 (0, 5, 10 ml .L^{-1})

studied traits

-stem height (cm)

The stem height was measured with a tape measure from the soil surface to the growing tip of the stem before treatment and at the end of the experiment and the difference between the two readings represents the average increase.

- stem diameter (mm)

The diameter of the stem was measured using the foot (Vernier caliper) at a height of 5 cm from the surface of the soil before treatment and after the end of the experiment and the difference between the two readings represents the increase in diameter.

-number of leaves (leaf.plant^{-1})

The number of leaves in the experimental unit was calculated at the end of the experiment and divided by the number of plants in it to extract the mean.

-leaf area (cm^2)

Four fully-grown leaves were tested at the end of the experiment from each experimental unit and placed in marked plastic bags to preserve them from wilting, then wiped with a wet cloth to get rid of the dust stuck to them, and then weighed with a sensitive scale, and pieces of known area were taken and weighed and calculated.

The leaf area on the basis of the fresh weight and according to the following equation:

leaf area (cm^2)= $\frac{Weight\ of\ fresh\ leaves\ (gm) \times area\ of\ cut\ discs}{Fresh\ weight\ of\ the\ cut\ leaves}$

-Total leaf area (cm^2)

It was calculated by multiplying the area of one leaf by the average number of leaves per plant

-Determination of leaf nitrogen content (%)

The nitrogen ratio was estimated by the method [13] by applying the following equation:

$N=\frac{100\times0.014\times Standard(0.04)\times HCl\ acid\ volume}{Sample\ Weight\ (gm)}\%$

-Determination of phosphorous content of leaves (%)

Phosphorous was estimated using the method of ammonium molybdate and ascorbic acid and measured by a spectrophotometer at a wavelength of 622 nm as stated in the method [14].

p=% (solution volume x concentration from the curve)/(plant sample weight x 10,000)

-Determination of potassium content of leaves (%)

A flame photometer was used to estimate the percentage of potassium according to the method [15].

K=% (Volume x Concentration)/(Plant sample weight x 10,000)

-Determination of the total chlorophyll content of leaves (mg 100gm^{-1} fresh weight)

The chlorophyll was estimated according to the method [16]

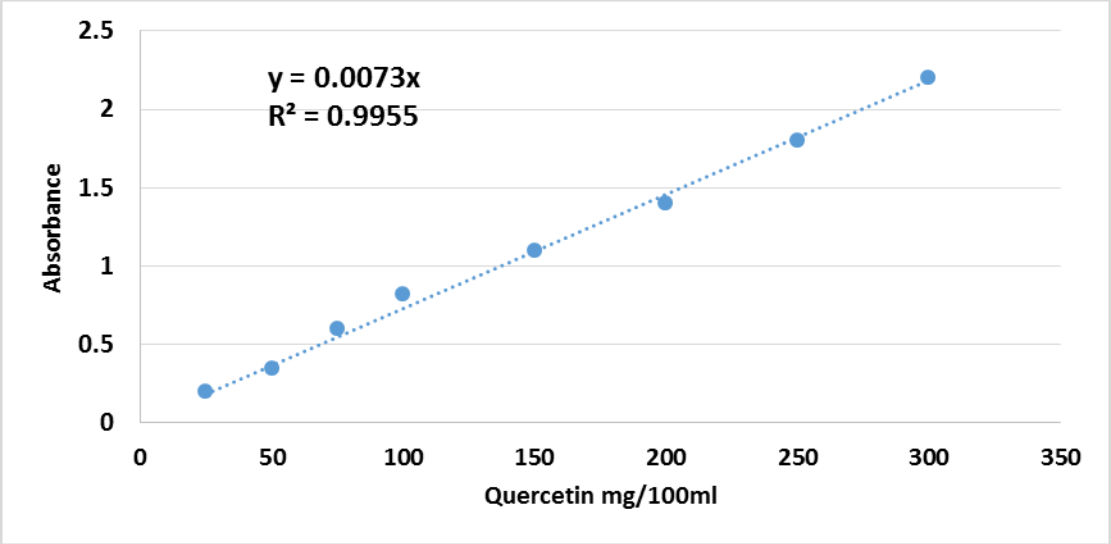
Chlorophyll pigment was estimated according to the following equation:

$Chl.T = \left(\frac{10\times100}{0.5\times1000}\right)\times B\ 8.02\ \ 645+663\ A\ 20.2$

-Flavonoids content (µg/g)

According to the method [17,18]

Calculations: The amount of total flavonoids was extracted from the following standard curve:



Results and Discussion

The results showed a significant effect when spraying with the amino acid tryptophan on the rate of plant height, as treatment T1 was significantly superior to the highest rate of 45.4 cm compared to the comparison treatment T0, which gave the lowest rate of 32.3 cm. Also, treatment with marine algae extract (Agazone) had a positive effect on the average of plant growth .As treatment S2 gave the highest average of 47.1 cm compared to the control treatment S0, which gave the lowest average of 30.4 cm for this trait. The bi-interaction between tryptophan and marine algae extract T.S had a significant effect on stem height average, where the treatment T1S2 was achieved by giving it the highest rate of 54.1 cm compared to the control treatment which gave the lowest average of 25.1 cm.

The results indicate that the spraying of tryptophan led to a significant increase in stem diameter, as the treatment T2 gave the highest rate of 47.6 mm compared with control treatment T0, which gave an average of 41.7 mm.Also, spraying with seaweed extract Agazone caused a significant increase in treatment S2, which recorded the highest rate of 53.7 mm compared to the control treatment S0, which gave 28.7 mm. The bi-interaction between tryptophan and seaweed extract T.S had a significant effect on the average stem diameter. T0S2 treatment gave it the highest average of 58.3 mm compared to the interaction treatment T0S0 which gave the lowest average of 18.8 mm.

The results indicate that there are significant differences in the number of leaves as a result of treatment with tryptophan The treatment T2 outperformed, as it gave the highest average of 5.72 leaves . plant^{-1}, while the number of leaves decreased in the control treatment T0 of 3.48 leaves . plant^{-1}Also, spraying with seaweed extract Agazone had a significant effect in increasing the average number of leaves within treatment S2, which gave 5.79 leaves . plant^{-1}, while the number of leaves decreased in the control treatment S0 to 2.98 leaves . plant^{-1}The bi-interaction between tryptophan and seaweed extract T.S had a significant effect on the average number of leaves, where the T1S2 treatment achieved the highest average of 6.83 leaves . plant^{-1}, compared to the control treatment, which gave the lowest average of 1.92 leaves . plant^{-1}

It was shown in the table that spraying with tryptophan had a significant effect on the leaf area through treatment T2, which gave the highest average of 226 cm ^{2} compared to control treatment T0, which gave the lowest average of 104 cm ^{2},The spraying with seaweed extract Agazone S1 also had a positive effect on the leaf area which gave the highest average of 227 cm^{2} compared to control treatment S0, which gave the lowest average of 83 cm^{2} The bi-interaction between

tryptophan and seaweed extract T.S had a significant effect on the average leaf area, where the treatment T2S1 excelled by giving it the highest average of 454 cm^2 compared to the comparison treatment which gave the lowest rate of 78 cm^2.

The results indicate the superiority of the spraying treatment with tryptophan T1 by giving the highest average of leaf area 650 cm ^{2} compared with control treatment T0, which gave the lowest average of 393 cm ^{2} Also, spraying with marine algae extract had a significant effect on this trait through treatment S2, which recorded the highest average of 762 cm ^{2} compared to the control treatment, which gave the lowest rate of 249 cm ^{2}, The bi-interaction between tryptophan and seaweed extract T.S Agazone had a significant effect on the average total leaf area, where the T1S2 treatment was achieved by giving it the highest average of 885 cm^2 compared to the control treatment which gave the lowest average of 149 cm^2.

Table (1) effect of spraying with tryptophan and seaweed extract on the vegetative traits of papaya seedlings

leaf area	Total leaf area	number of leaves	Stem diameter	Stem height	treatments
					1-tryptophan T
104	393	3.48	41.7	32.3	concentration 0
110	650	5.72	39.1	45.4	concentration 50
226	529	4.40	47.6	39.1	concentration 100 .
T:192.5	**T:79.4**	**T: 0.73**	**T:5.86**	**T:6.28**	**LSDT**
					2- seaweed extract S
83	249	2.98	28.7	30.4	0 . concentration
227	560	4.83	46.1	39.4	5 . concentration
130	762	5.79	53.7	47.1	10 . concentration
S :192.5	**S:79.4**	**S:0.73**	**S:5.86**	**S:6. 28**	**LSD S**
					T.S
78	149	1.92	18.8	25.1	**T0.S0**
109	420	3.83	48.1	33.2	**T0.S5**
126	608	4.69	58.3	38.7	**T0.S10**
83	348	4.17	26.1	37.3	**T50.S0**
116	716	6.17	42.0	44.8	**T50.S5**
130	885	6.83	49.1	54.1	**T50.S10**
88	251	2.87	41.3	28.7	**T100.S0**
454	544	4.50	48.0	40.2	**T100.S5**
136	792	5.83	53.6	48.5	**T0.S10**
ST: 333.4	**ST:137.5**	**ST:1.27**	**ST:10.15**	**ST:10.88**	**LSD T.S**

It is noted from the results received that the nitrogen content of the leaves was significantly affected by the increase of the tryptophan spraying treatments, where the T2 treatment excelled and gave an average of 4.76% compared to the comparison treatment T0 which gave the lowest average of 3.75% Also, spraying with seaweed extract had a significant effect, where treatment S2 excelled on the rest of the treatments as it gave a rate of 5.54% compared to the control treatment S0, which gave the lowest average of 2.55%.The bi-interaction between tryptophan and seaweed extract T.S had a significant effect on the nitrogen content of the leaves, where the T2S2 treatment achieved the highest value of 5.67% compared to the control treatment which gave the lowest rate of 1.28%.

It was found that the tryptophan treatment had a positive effect on the phosphorous content of the leaves, where the T2 treatment gave the highest average of 0.41% compared to the control treatment, which gave an average of 0.27% The effect of spraying seaweed extract on the average leaf content of phosphorous element, treatment S2 gave the highest rate of 0.51% compared to the control treatment S0, which gave the lowest rate of 0.16%. The bi-interaction between tryptophan and T.S seaweed extract had a significant effect on the percentage of phosphorous in leaves. Whereas, treatment T2S2 excelled by giving it the highest value, which amounted to 0.56%, compared with the control treatment, which gave a value of 0.13%.

The table indicates that there was no significant effect of treatment with tryptophan on the potassium content of leaves, where treatment T2 gave the highest average of 3.92% compared to the control treatment T0 which gave an average of 3.22% Spraying with seaweed extract also affected the average potassium content of leaves, where treatment S2 gave the highest average of 5.31% compared to the control treatment S0, which gave. 1.49 .%The bi-interaction between tryptophan and seaweed extract T.S had a significant effect on the percentage of potassium in leaves, where the T2S2 treatment excelled by giving it the highest value, which reached 5.39% compared with the control treatment, which gave a value of 1.19%.

It is noticed from the results presented in the table that there was a significant increase in the content of chlorophyll in leaves when treated with tryptophan. Compared to control treatment T0, which gave 61.32 (100 g^{-1} fresh weight), spraying with seaweed extract also affected that trait.Where treatment S2 excelled on the rest of the treatments by giving it the highest value (73.00 mg 100 g $^{1-}$ fresh weight) compared to the control treatment S0, which gave a value of 61.54 (mg 100 g $^{-1}$ fresh weight).The effect of a bi-interaction between tryptophan and T.S seaweed extract significant effect on the chlorophyll content of leaves.Whereas, the treatment T1S2 excelled with the highest value of 76.44 (mg 100 g^{-1} fresh weight) compared to the control treatment, which gave a value of 57.13 (mg g^{-1} fresh weight).

Table results indicate a significant increase in the leaves content of flavonoids when sprayed with tryptophan Treatment T2 gave the highest average of 46.56 (µg / g) compared to the control treatment T0, which gave an average of 33.65 (µg / g).Spraying with seaweed extract also had a significant increase in this trait, where the treatment with a concentration of S2, which gave the highest value of 43.02 (µg /g), was excelled on the control treatment, which gave a value of 37.89 (µg /g).The effect of the bi- interaction between tryptophan and seaweed extract T.S significant effect on the leaves content of flavonoids.Whereas, treatment T2S2 excelled with the highest value of 48.05 (µg/g) compared to the control treatment, which gave a value of 29.11 (µg/g).

Table (2) effect of spraying with tryptophan and seaweed extract on the leaves content of NP K, total chlorophyll and flavonoids in papaya seedlings.

Flavonoids µg/g	Chlorophyll mg 100gm^{-1} fresh weight	% K	% P	% N	traits treatments
					1-tryptophan T
33.65	**61.32**	**3.22**	**0.27**	**3.75**	concentration 0
42.19	**70.11**	**3.29**	**0.33**	**4.26**	concentration 50
46.56	**70.68**	**3.92**	**0.41**	**4.76**	concentration 100 .
T: 2.22	**T: 4.95**	**T:0.18**	**T:0.04**	**T: 0.17**	**LSDT**
					2- seaweed extract S
37.89	**61.54**	**1.49**	**0.16**	**2.55**	0 . concentration S
41.48	**67.57**	**3.63**	**0.34**	**4.67**	5 . concentration S
43.02	**73.00**	**5.31**	**0.51**	**5.54**	10 . concentration S
S : 2.22	**S : 4.95**	**S :0.18**	**S :0.04**	**S : 0.17**	**LSD S**
					T.S
29.11	**57.13**	**1.19**	**0.13**	**1.28**	**T0.S0**
35.18	**60.36**	**3.23**	**0.23**	**4.62**	**T0.S5**
36.64	**66.46**	**5.25**	**0.46**	**5.34**	**T0.S10**
39.60	**62.56**	**1.27**	**0.13**	**2.58**	**T50.S0**
42.58	**71.33**	**3.29**	**0.36**	**4.59**	**T50.S5**
44.38	**76.44**	**5.31**	**0.50**	**5.61**	**T50.S10**
44.96	**64.93**	**2.01**	**0.23**	**3.81**	**T100.S0**
46.68	**71.01**	**4.37**	**0.43**	**4.81**	**T100.S5**
48.05	**76.11**	**5.39**	**0.56**	**5.67**	**T0.S10**
ST: 3.85	**ST: 8.58**	**ST:0.32**	**ST:0.07**	**ST:0.30**	**LSD T.S**

The increase in vegetative growth indicators and leaf content of chlorophyll and nutrients as a result of treatment with tryptophan is due to its role in stimulating cell growth. As it

serves as a source of carbon and energy and the manufacture of other organic compounds such as proteins, amines, purines, alkaloids, vitamins, enzymes, and terpenes [19]] [20].Spraying with marine algae extract has contributed to improving vegetative growth indicators because it contains auxins, gibberellins and cytokines and their effective role in stimulating cell division and elongation [21].The increase in these qualities may be due to the extract containing amino acids and nutrients and its contribution to increasing the vital activities of the plant [22].The increase in the stem diameter when spraying with seaweed extract is due to the fact that this extract contains nutrients that increase the metabolic activities of the plant, which positively affects the increase in its growth and thus increases the stem diameter [23].The increase in the average number of leaves and leaf area, when treated with seaweed extract, is due to the cytokines that these extracts contain, which have a role in improving the physiological activities inside the plant and increasing the content of chlorophyll in its leaves, which positively affects the photosynthesis process [24].In addition, cytokines work to transport nutrients from the roots to the top and then to vegetative growth and leaves [25].The effect of the amino acid tryptophan in increasing the leaves' content of nutrients (NP) and chlorophyll may be due to the fact that it contains nitrogen, which is directly absorbed when this acid is sprayed on the leaves. This, in turn, leads to the activation of the root system and an increase in the absorption and representation of nitrogen, which is reflected in the absorption of other elements such as phosphorous, and this explains the increase in its concentration in the leaves, as well as the effect of the elements in increasing the efficiency of carbon representation, which results in an increase in the efficiency of absorption of nutrients and this, explains the increase in their concentration in papers [26].[27] mentioned that the elements nitrogen and potassium have an important role in the formation of amino acids and that the treatments of spraying with tryptophan excelled in increasing the nitrogen content of the leaves, especially in higher concentrations may be due to increased levels of endogenous growth stimulants, which act as a source of Sink withdrawal of minerals and nutrients [28] on the other hand, amino acids play an important role in the osmotic effort in the plant tissue, where it reduces the water effort and this increases the ability of cells to absorb water and elements, and this positively affects the increase in the vegetative growth of the plant and thus increases the absorption of nutrients such as nitrogen, phosphorous and potassium [29].Increased elements (NPK) when spraying seaweed extract, where nitrogen directly enters the composition of the chlorophyll molecule, and the seaweed extract contains gibberellin, which increases the effectiveness of ribosomes and the formation of proteins, nucleic acids and amino acids that enter into the construction of the chlorophyll molecule [30]Also, the seaweed extract and the amino acid tryptophan had a role in increasing the content of flavonoids in leaves, which may be due to the increase in vegetative growth, which was reflected in the accumulation of carbohydrates in the plant [31] and the combined effect between tryptophan and seaweed extract, which led to enhancing the nutritional status of the plant and increasing the absorption of elements As the increase in the plant's content of flavonoids depends on the level of elements available to the plant, whether from their source in the soil or through the addition of plant extracts containing the elements [32] and this increase is linked to the rapid growth represented by the vegetative growth criteria [33].

Conclusions

The results of the study showed a clear response of papaya seedlings to spraying with the amino acid tryptophan and extract of seaweed Agazone, especially in high concentrations, which was positively reflected in improving the characteristics of vegetative growth and increasing the leaf content of minerals, chlorophyll, carbohydrates, flavonoids and protein.

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