

Effect of Biofertilizer and Spraying Phenylalanine on Mineral Content and Antioxidant Compounds of Dill

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Abstract

The field experiment was carried out in a private field in Babil Governorate, Abi Ghark district, for the winter seasons 2018-2019 and 2019-2020 to study the effect of biological fertilizer and phenylalanine spraying on the content of the shoots and seeds of oil and some active compounds. The treatments were distributed in a global experiment according to a randomized complete block design (RCBD) with three replications. The experiment included two factors, the first is the bacterial vaccine *Azotobacter chroococcum* with two levels (inoculum and without inoculation) and the second is spraying with the amino acid Phenylalanine in three concentrations 0, 75 and 150 ml⁻¹. The averages were compared according to the LSD test at a probability level of 0.05. The results showed that the treatment of biological fertilizer and spraying with the amino acid Phenylalanine was significantly superior in all the studied traits, which included the oil yield (gm.plant⁻¹), the percentage of nitrogen, phosphorus and potassium and the shoots content of total phenols (mg.kg⁻¹) and total flavonoids (mg.kg⁻¹) and total flavonoids (mg. . Kg⁻¹), percentage of free radical suppression and total antioxidant capacity (mg. 100 g⁻¹). The best results were when the two-way treatment (bio-fertilizer×150 ml.l-Phenylalanine), which gave the highest oil yield of 0.677 and 0.907 g. Plant⁻¹, for the two seasons respectively, compared with the comparison treatment that gave the lowest yield for both seasons of the experiment.

Keywords: biological fertilizer, Phenylalanine , Dill(*Anethum graveolens L.*)

Introduction

Anethum graveolens L. is an important vegetable plant that belongs to the Apiaceae family and is native to the eastern and western Mediterranean. The active parts are fresh and dry leaves, seeds, roots and oil. It is consumed fresh or cooked and used as a spice and in the manufacture of food and cosmetics, The plant is highly nutritious and rich in vitamins, and contains antioxidants, polyphenols, and other compounds that are considered anticancer, It is used as a treatment for blood pressure, improves the work of the heart and lungs, calms nerves, appetite suppresses, dyspepsia, anti-spasms, vomiting in children, and a diuretic for mothers' milk ⁽¹⁾.

Azotobacter free-living bacteria It is usually spread in light and neutral alkaline soils, it can stabilize atmospheric nitrogen, in addition to its ability to dissolve

phosphates and produce various types of growth regulators such as indole acetic acid, gibberellins, cytokinins and a number of vitamins that stimulate the growth of roots and the formation of root hairs. Also, *Azotobacter* inhibits plant pathogens and the production of cidrofurates ⁽²⁾.

Amino acids are nitrogenous organic compounds that are the basic unit in building proteins, building proteins, and contribute to maintaining the stability of cell acidity. And spraying plants with amino acids contributes to overcoming the nutritional stresses during the plant life cycle ⁽³⁾ as a result of improving the efficiency of absorption and transport of nutrients. As the amino acid Phenylalanine is one of the cyclic (aromatic) amino acids that are used in the construction of many secondary compounds such as alkaloids and vegetable

pigments. It contributes to plant resistance to abiotic stresses and aids in plant growth and development due to its effect on the biosynthesis of some growth hormones, particularly gibberellins⁽⁴⁾.

Materials and Method

The experiment was conducted during the winter seasons 2018-2019 and 2019-2020 in one of the private fields in Babil governorate, Abi Gharq district, which is 10 km northwest of the city center of Hilla, at longitude 44.333 degrees north and latitude 32.501 degrees south in Babel governorate to study the effect of vaccination of Dill seeds with *Azotobacter chroococcum* at two levels (inoculum, and without inoculation) and spraying with Phenylalanine amino acid in three concentrations 0, 75 and 150 ml.⁻¹ for the two seasons, respectively, on the content of dill from some antioxidant compounds. The soil analysis test was conducted before planting and fertilization for both seasons by taking separate samples of field soil at the depth of 0-30 cm for the purpose of measuring some chemical and physical characteristics of the soil of the experiment site (Table 1).

The land was prepared twice, perpendicularly, then it was flattened and divided into three duplicates, the

distance between them was 2m. Each duplicate includes 18 panels with dimensions of 2 m x 2 m for each plate as the line 9 includes the distance between the plants 20 cm and between a line and another 20 cm, in each slab Nine lines. The experiment included inoculating the seeds at a rate of 1 kg of seeds with 0.5 kg of the bacterial vaccine *Azotobacter chroococcum* loaded on peat moss. As for spraying plants with amino acid, it was done three times, the first after 30 days of germination, and the second after 14 days from the first and third, when plants bloomed by 50%.

As for the amino acid, its concentrations were prepared by taking 75 and 150 mg, and each was added to a liter of distilled water and sprayed all the experimental units, except for the comparison treatment with the amino acid, and the following characteristics were studied: oil yield (gm.plant⁻¹), the percentage of nitrogen, and the percentage of phosphorous by following the method of reduction of Ammonium molybdate-vanadate, the percentage of potassium according to the method used by, and the content of the shoots of total phenols and total flavonoids (mg.kg⁻¹ dry weight), and an estimate of the percentage for free radical suppression and estimation of total antioxidant capacity.

Table (1) Some chemical and physical characteristics of the pre-sowing research soil

pH	N	P	K	O.M.	Sand	Clay	Silt
	mg.kg	mg.kg	mg.kg	gm.kg	gm.kg	gm.kg	gm.kg
7.41	36.5	7.23	108	12.4	305	200	495

Results and discussion

It is evident from Table (2) that the biological fertilizer *Azotobacter* and the Phenylalanine amino acid spraying had a significant effect for all the studied traits, as the plants whose seeds were fertilized with the biological fertilizer significantly exceeded the oil yield and the percentage of nitrogen, phosphorus and

potassium. It achieved the highest averages of 0.580 and 0.730 gm.plant⁻¹, 2.245, 2.575%, 0.338, 0.408%, 1.526 and 1.731%, compared to plants whose seeds were not inoculated with biological fertilizer, which amounted to 0.462 and 0.587 gm. 2.420%, 0.314, 0.385%, 1.391% and 1.598% for the two seasons, respectively.

The reason for this may be that the addition of biological fertilizer provides the plant with nutrients, which leads to the accumulation of carbohydrates inside the plant, which leads to the production of terpenes by entering carbohydrates in the glycolysis pathway reactions and producing Pyruvic acid and then Mevalonic acid, which is the key to the production of Isoprene units that represent the building block Basic for building turbine oils ⁽⁵⁾. These results are in agreement with ⁽⁶⁾ on the fennel plant in which the oil yield was increased after treatment with biological fertilizer. The increase in the percentage of nitrogen, phosphorous and potassium elements in the plant tissue as a result of responding to biological fertilizers may be due to the ability of these bacteria to increase these elements in the soil and provide them in a manner that is easily absorbed by the plant ⁽⁷⁾.

It is noticed from the same table that spraying the plant with the amino acid Phenylalanine at a

concentration of 150 ml⁻¹ achieved a significant superiority also compared to the comparison treatment, as the values reached 0.595 and 0.794 g. Plant⁻¹, 0.275, 2.626%, 0.352, 0.427%, 1.584 and 1.823% compared to the comparison treatment that achieved 0.451 and 0.538 gm. Plant⁻¹, 2.084, 2.363%, 0.299, 0.367%, 1.297 and 1.501% for the study characteristics and for the two seasons, respectively. This increase may be due to the treatment with the amino acid because the chemical composition of the acid contains the amine group NH₂, and this was positively reflected on the plant's nitrogen content ⁽⁸⁾.

On the other hand, spraying amino acids plays a role in changing the osmotic potential in the plant tissue, as it reduces the water effort and this increases the ability of cells to absorb water and elements and this positively affects the increase in vegetative growth and thus increases the absorption of nutrients such as nitrogen, phosphorus and potassium ⁽⁹⁾.

Table (2) The effect of spraying with Phenylalanine biological fertilizer on the oil yield and some chemical properties of dill plant

The Factors		2018-2019 Season				2019-2020 Season			
		Herb oil Yield Gm/plant	N (%)	P (%)	k (%)	Herb oil Yield Gm/plant	N (%)	P (%)	k (%)
Bacteria effect	Without inoculum	0.462	2.115	0.314	1.391	0.587	2.420	0.385	1.598
	inoculum	0.580	2.245	0.338	1.526	0.730	2.575	0.408	1.731
(0.05)LSD		0.0152	0.0231	0.0052	0.03310	0.0337	0.0401	0.0069	0.03712
Phenyl-alanine	0	0.451	2.084	0.299	1.297	0.538	2.363	0.367	1.501
	75	0.518	2.181	0.326	1.495	0.644	2.505	0.396	1.670
	150	0.595	2.275	0.352	1.584	0.794	2.626	0.427	1.823
(0.05)LSD		0.0187	0.0283	0.0064	0.04054	0.0413	0.0491	0.0085	0.04546
Without inoculum	0	0.418	2.047	0.283	1.267	0.512	2.326	0.362	1.468
	75	0.456	2.097	0.314	1.427	0.569	2.402	0.384	1.592
	150	0.513	2.202	0.344	1.478	0.680	2.533	0.409	1.735

Cont... Table (2) The effect of spraying with Phenylalanine biological fertilizer on the oil yield and some chemical properties of dill plant

inoculum	0	0.484	2.121	0.315	1.326	0.565	2.400	0.372	1.533
	75	0.579	2.265	0.338	1.563	0.718	2.608	0.407	1.748
	150	0.677	2.348	0.360	1.690	0.907	2.718	0.446	1.911
(0.05)LSD		0.0264	0.0400	n.s.	0.05733	0.0584	0.0695	0.0120	0.06429

The results in Table (3) indicate that inoculation of dill seeds with the biological fertilizer *Azotobacter* had a significant effect on the shoots content of total phenols (mg.kg^{-1}), total flavonoids (mg.kg^{-1}), percentage of free radical suppression and total antioxidant capacity (mg.100 gm^{-1}). It achieved the highest averages of 0.751 and 0.769 mg.kg^{-1} , 36.453 and 36.592 mg.kg^{-1} , 62.58, 66.03%, 40.33 and 41.86 mg.100 g^{-1} compared to the comparison treatment (without inoculation), which achieved the lowest averages of 0.748 and 0.764. Mg.Kg^{-1} , 36.366 and 36.509 mg.kg^{-1} , 58.60, 61.78%, 39.42 and 40.54 mg.100 gm^{-1} for the two seasons, respectively.

The reason for the increase in the shoots content of antioxidant compounds and the amount of their oxidative activity may be due to the result of the effect of biological fertilizer in increasing the vegetative indicators, which led to an increase in the accumulation of dry matter resulting from the increase in the rate of photosynthesis processes mainly producing the primary metabolites, which in turn led to an increased accumulation of intermediate compounds that participate in secondary metabolism to produce phenolic, flavonoid and other compounds⁽⁵⁾. The increase in phenols and flavonoids, the percentage of free radical suppression and the total antioxidant capacity in the leaves may be due to the increase in vegetative growth, which is reflected in the accumulation of carbohydrates in the plant, which enter a direct pathway to produce phenols via Shikmic acid and the interference of amino acids such as Phenylalanine and Tyrosine in building flavonoids⁽¹⁰⁾.

It is noted from the same table the significant superiority of spraying with the amino acid Phenylalanine at a concentration of 150 ml.liter^{-1} , which achieved the highest values in the aforementioned characteristics, which amounted to 0.756 and 0.771 g.kg^{-1} , 36.494, 36.677 g.kg^{-1} , 66.67, 70.15% and 42.01, and 43.62 mg.100 g^{-1} for the two seasons, respectively, compared to the comparison treatment that achieved the lowest values of 0.744 and 0.763 mg.kg^{-1} , 36.331 and 36.415 mg.kg^{-1} , 55.66, 59.17%, 38.23 and 39.33 mg.100 g^{-1} for the two seasons, respectively.

This significant increase when spraying with the amino acid Phenylalanine may be due to the fact that it enters the vital structural steps of phenols and flavonoids that are from the pathway of Shikimic acid after adding a molecule of Phosphoenol pyruvate in several steps to form Prephonic acid, which is the amino acid Phenylalanine by the intermediate compound Phenylpy Phenylalanine on the formation of Cinnamic acid, which is the basic unit for building phenols and flavonoids within this path⁽⁵⁾. In the production of phenolic acids, at a rate of 10% in the production of anthocyanins, accordingly, preparing the plant with the amino acid Phenylalanine and Tyrosine it needs leads to an increase in the phenolic compounds and their derivatives. It is worth noting that phenols and flavonoids are antioxidant compounds that have effectiveness to inhibit free radicals by giving hydrogen electrons to the free radical turning it into a harmless compound⁽⁵⁾.

Table (3) The effect of spraying phenylalanine biological fertilizer on some antioxidant compounds in dill plant

The Factors		2018-2019 Season				2019-2020 Season			
		Total phenolics Gm/kg	Total flavonoid Gm/kg	Free radical suppression %	Total antioxidant mg/100gm	Total phenolics Gm/kg	Total flavonoid Gm/kg	Free radical suppression %	total antioxidant mg/100gm
Bacteria effect	Without inoculum	0.748	36.366	58.60	39.42	0.764	36.509	61.78	40.54
	inoculum	0.751	36.453	62.58	40.33	0.769	36.592	66.03	41.86
(0.05)LSD		0.00091	0.00720	1.581	0.1223	0.00089	0.00796	1.216	0.565
Phenylalanine	0	0.744	36.331	55.66	38.23	0.763	36.415	59.17	39.33
	75	0.750	36.404	59.43	39.38	0.767	36.559	62.39	40.65
	150	0.756	36.494	66.67	42.01	0.771	36.677	70.15	43.62
(0.05)LSD		0.00111	0.00882	1.936	0.1497	0.00109	0.00975	1.489	43.62
Without inoculum	0	0.742	36.284	55.39	37.64	0.759	36.365	58.38	39.19
	75	0.749	36.366	57.34	39.01	0.766	36.519	60.56	39.75
	150	0.754	36.449	63.08	41.60	0.768	36.644	66.39	42.68
inoculum	0	0.746	36.378	55.94	38.81	0.766	36.466	59.96	39.48
	75	0.750	36.443	61.53	39.75	0.768	36.600	64.21	41.54
	150	0.757	36.539	70.27	42.43	0.774	36.710	73.91	44.56
(0.05)LSD		0.00157	n.s.	2.738	0.2118	0.00155	0.01379	2.106	0.979

Conclusions

The addition of bio-fertilizer and spraying with amino acid Phenylalanine at a concentration of 150 ml⁻¹ liter had a positive effect in all the studied traits.

The treatment of the interaction between the biological fertilizer and the amino acid Phenylalanine at

a concentration of 150 ml⁻¹ was superior in most of the studied characteristics.

Conflict of Interest: None

Funding: Self

Ethical Clearance: Not required

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