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Response of Black Cumin (*Nigella Sativa* L.*)* Plants to Some Bio-Stimulants Treatments on Growth, Production and Chemical Components

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ABSTRACT

A field study was conducted at the experimental farm of Sids (Horticultural Research Station, Beni-Suef Gov.) with the goal of enhancing the growth, productivity, and seed oil yield of black cumin plants through foliar application of algae extract, kinetin, and bio-phosphate. According to the findings, the ideal vegetative traits of plant height , stem diameter and root length, yield components "number of capsules, seeds yield/ plant (g) and per feddan (Kg)", oil determinations characters (fixed oil %, fixed oil per plant and per fed.), three photosynthetic pigments and percentages of NPK values obtained as a result of foliar spray of the bio-phosphate at 800 g/l in combination with the high dose of Algae extract (8 ml/l) or Kinetin (100 mg/l) and no significant discrepancies found between Algae extract (8 ml/l) and Kinetin (100 mg/l). In order to improve growth, seed yield, and fixed oil components under the same research conditions, it may be advised to spray *Nigella sativa* L. plants with high concentrations of algae extract (8 ml/l) or kinetin (100 mg/l) in combination with bio-phosphate 800 g/l.

KEYWORDS: *Nigella sativa* L., Algae extract, Kinetin, Bio-phosphate, Fixed oil.

1. INTRODUCTION

One of the most significant fragrant and therapeutic herbs is black cumin. Usually found growing in Eastern Europe, the Middle East, and Western Asia, it belongs to the Ranunculaceae family of plants. The tiny black seeds found in its full fruit are called "El-Habba Al-Saudaa" in Arabic, and black seed or black cumin in English Shabana *et al*., (2012). It is mostly planted as a winter annual herb in the Middle Egypt Governorates, particularly Assiut, El-Minia, and Beni-Suef.

Datta *et al.,* (2012), discovered that the essential oil ranged from 0.4% to 0.7% of the black cumin seeds' weight, whereas the fixed oil ranged from 30% to 35%. Lino-leic, oleic, linolenic, arachidic, palmitic, stearic, and myristic acid components are among the variety of oils that make up black seed fixed oil's chemical composition. The efficient use of bio- fertilizers for crops enhances and preserves soil fertility and sustainability in the natural soil ecosystem in addition to offering growers financial gains. Rhizobacteria that promote plant growth have positive effects on growth not only because they fix nitrogen in the rhizosphere but also because they can produce antibiotics and other growth-promoting chemicals like phytohormones and, occasionally, solubilize phosphates, Hassan and Ali (2013). To reduce the amount of chemical P fertilizer that pollutes the environment, phosphate-solubilizing bacteria must be used (Gendy *et al.,* 2012, Hassan and Mahfouz 2012). Commercial products like algae extracts, which contain nutrients like "N, P, K, Ca, Mg, and S" as well as "Zn, Fe, Mn, Cu, Mo, and Co," along with some growth regulators, polyamines, and vitamins, can be applied topically to improve vegetable growth. Bacterial inoculants have been used to boost plant yields in a number of countries (Zhang and Ervin, 2004 and Papenfus *et al*., 2013). cytokinins, auxins, abscisic acid, vitamins and nutrients (Chojnacka and Kim, 2013). Kinetin is the most synthetic known cytokinin according to Barciszewski *et al.* (2000) it was detected in both plant tissue extracts and animal cellular DNA, and it has a furfuryl ring at the N6-position of adenine. Kinetin is known to be essential to plants and to be a growth regulator that these species need.

Even though its function in animals is well established, more research is necessary to fully understand its significance in plants. Higher amounts of kinetin are harmful, whereas lower concentrations have a beneficial effect on plants.

Therefore, the current study was created to find out the most suitable treatment of bio-phosphate and some bio-stimulants (Algae extract and Kinetin) treatments on the growth characters, seed yield, volatile & fixed oil and some chemical components, of *Nigella sativa* L. under environmental conditions in Beni- Suef Governorate, Egypt.

2. MATERIALS AND METHODS

The current study examined how black cumin plants responded to foliar applications of bio-phosphate and certain biostimulants, such as "algae extract and kinetin," over an interval of two seasons in 2021/2022 and 2022/2023, at the research farm of the Sids Horticultural Research Station in the Beni-Suef governorate. Prior to land preparation, several soil samples (0-40 cm depth) were taken for nutrient and trace element analysis (Table, 1) according to, Jackson, 1975.

Table 1. Chemical and physical properties of the experimental soil.

Particle size distribution*			Textural Class	Chemical properties*									
Clay %	Silt %	Sand %	Clay	SP	EC, dSm^{-1} (at 25°C)	mg/kg .soil N	P	K	Fe	Cu	Zn	Mn	pH
82.5	15.0	2.5		82.0	2.26	83.0	9.74	449.5	0.642	0.040	0.152	0.266	8.1

*Soil sample analysis was conducted at the Agricultural Research Center in Giza (ARC).

The black cumin seeds were acquired from the Sids Hort. Res. Station and were planted on November 1st in the two experimental seasons. The unit of experimentation (plot) was 2 x 2.25 = 4.5m^2 and contained 3 rows, 60 cm apart. The growing seedlings were thinned to two plant per hill, 3 week after planting. The other agricultural practices were done as recommends.

2.1.Experimental design:

This experiment was conducted using a randomized complete block design (RCBD) in three repetitions using the split-plot arrangement,

with four bio-phosphate treatments in the main plots (A) i.e. control, 200, 400 and 800 g/l of bio-phosphate (Bacteria, one package has 400 g of *Bacillus magterium* and one (g) has 109 of bacteria and was obtained from Giza ARC). The sub- plots (B) were control, Algae extract at 2, 4 and 8 ml/l & Kinetin at 50, 100 and 200 g/l.

2.2.Data recorded and statistical analysis:

The data listed below were noted: the characteristics of vegetative growth, such as plant height, stem diameter, and root length; the components of yield, such as the number of capsules per plant, seed yield per plant, and feddan yield; and the fixed oil percentage, fixed oil per plant, and fixed oil per feddan, which are the oil determination traits Gad *et al.,* (1963) also a three photosynthetic pigments (Chlorophyll a, b and carotenoids), Fadl and Seri- Eldeen (1978) and herb percentages of NPK values Wilde *et al.* (1985), Chapman and Pratt (1975) and Cottenie *et al.,* (1982). The analysis of variance approach was used for all statistical analyses using the MSTATC computer software package, and the least significant differences (L.S.D.) test at 0.05 was used to test for mean differences (Freed *et al.,* 1991).

3. RESULTS AND DISCUSSION

3.1.Vegetative growth criteria:

Data presented in Table 2, declared that bio-phosphate treatments at 200,400 and 800 g/l were significantly increased on plant height, stem diameter and Root length over those of untreated plants in both seasons, Using bio-phosphate 800 g/l gave the best values of growth parameters considerably followed by bio-phosphate 400 g/l and bio-phosphate 200 g/l ,except for Stem diameter parameter the treatment bio-phosphate 200 g/l for first season and Root length parameter the treatment bio-phosphate 200 g/l in both seasons no significant differences were existed in comparison with the control ones in both seasons. The beneficial effect of bio-phosphate on vegetative growth characters was pointed out on *Nigella sativa* (Badran *et al.,* 2012 and Gamal *et al.,* 2012); *Pimpinella anisum* (Mohammad *et al.,* 2012 and Hassan *et al.,* 2013) ; coriander (Rekaby 2013 and Nasr Alla *et al.,* 2016); fennel (Abdou 2020); dill (Walid 2021); basil (Mohamed *et al.,* 2015); chamomile (El-Naggar *et al.* 2020); *Hibiscus sabdariffa* (El-Naggar *et al.,* 2011).

The results obtained in Table 2 for vegetative growth characters, including plant height, stem diameter, and root length at harvest of *Nigella sativa* plants, demonstrated that all six tested treatments—algae extract (2, 4, and 8 ml/l Algae extract) and kinetin (50, 100, and 200 mg/l Kinetin)—caused a significant and substantial increase in the three tested vegetative characters in the two seasons when compared to the untreated treatment. However, in the first season, there were no significant differences. Also, 50 (mg/l) Kinetin was used in the second season, and for the root length parameter at 2 (ml/l) Algae extraction, 50 (mg/l) Kinetin and/or 200 (mg/l) Kinetin were used in both experimental seasons. The highest overall values were given due to 8 (ml/l) Algae extract and/or 100 (mg/l) Kinetin It was interesting to note that no significant differences had been existed between the best treatments 8ml/l Algae ext. and 100 mg/l Kinetin, in both seasons in comparison with control treatment as shown in Table 2. The role of Algae extract and Kinetin in enhancing vegetative growth characters was demonstrated by Shoukat *et al.,* (2011) and Khalid *et al.,*(2014) on black cumin; El Laban *et al.,* (2017) on Dutch fennel; Reem *et al.,*(2022) and Hashim *et al.,*(2023) on Fennel; Radhiyah *et al.,*(2022) on chamomile.

For growth characteristics of plant height, stem diameter, and root length, statistical analysis revealed significant differences in the interaction between bio-phosphate and treatments with algae extract or kinetin in both seasons. The most effective interaction treatments, as indicated in Table 2, were bio-phosphate 800 g/l in combination with algae extract (8 ml/l) and/or kinetin (100 mg/l).

3.2.Yield characteristics:

Table 3 showed that the number of capsules per plant, seed yield per plant (g), and seed yield per feddan (kg) were all significantly higher in the bio-phosphate treatments (200, 400, and 800 g/l) than in the control treatment, which was statistically different in both consecutive

Table 2. Effect of bio-phosphate and Algae extract & Kinetin treatments on vegetative growth of black cumin plants during 2021/2022 and 2022/2023 seasons.

Algae extract and Kinetin treatments (B)	Bio-phosphate (mg/l) treatment (A)									
	control	Bio-Ph. 200	Bio-Ph. 400	Bio-Ph. 800	Mean (B)	control	Bio-Ph. 200	Bio-Ph. 400	Bio-Ph. 800	Mean (B)
	First season(2021/2022)					Second season(2022/2023)				
Plant height (cm)										
Control (tab water)	65.1	67.1	68.2	70.2	67.7	64.7	66.7	67.3	67.3	66.5
2 ml/l Algae extract	78.3	84.3	88.4	89.7	85.2	77.2	88.3	89.4	92.2	86.8
4 ml/l Algae extract	86.3	94.2	94.4	96.1	90.5	85.4	89.4	98.7	93.4	91.7
8 ml/l Algae extract	96.1	97.1	98.2	99.3	97.7	94.7	96.7	93.2	97.4	95.5
50 mg/l Kinetin	88.1	90.2	91.3	95.4	91.3	88.3	89.7	91.2	95.1	91.1
100 mg/l Kinetin	91.4	94.3	97.1	99.1	95.5	97.3	92.3	90.3	99.7	94.9
200 mg/l Kinetin	83.3	91.1	93.2	94.4	93.0	90.3	91.2	92.1	96.3	92.5
Mean (A)	84.1	88.3	90.1	92.0		85.4	87.8	88.9	91.6	
L.S.D. at 5%	A: 1.7		B: 3.1	AB: 6.2		A: 1.4		B: 2.1	AB: 4.2	
Stem diameter (cm)										
Control (tab water)	0.67	0.70	0.73	0.77	0.72	0.43	0.47	0.50	0.57	0.52
2 ml/l Algae extract	0.70	0.77	0.80	0.90	0.79	0.47	0.50	0.67	0.80	0.61
4 ml/l Algae extract	0.78	0.83	0.87	0.99	0.87	0.50	0.60	0.70	0.97	0.69
8 ml/l Algae extract	0.80	0.85	0.97	0.99	0.90	0.76	0.78	0.83	0.99	0.84
50 mg/l Kinetin	0.80	0.83	0.87	0.90	0.85	0.50	0.53	0.53	0.63	0.55
100 mg/l Kinetin	0.81	0.84	0.92	0.96	0.88	0.73	0.83	0.83	0.86	0.81
200 mg/l Kinetin	0.73	0.87	0.88	0.93	0.85	0.53	0.60	0.63	0.67	0.61
Mean (A)	0.76	0.81	0.86	0.92		0.56	0.62	0.67	0.78	
L.S.D. at 5%	A: 0.06		B: 0.09	AB: 0.18		A: 0.05		B: 0.06	AB: 0.12	
Root length (cm)										
Control (tab water)	19.0	19.3	19.6	20.0	19.5	15.1	15.2	15.5	16.1	15.5
2 ml/l Algae extract	19.1	20.2	21.1	22.2	20.7	15.3	15.7	18.0	18.3	16.8
4 ml/l Algae extract	21.3	21.7	23.0	23.3	22.3	17.1	16.1	21.2	21.6	19.0
8 ml/l Algae extract	27.0	28.7	29.0	30.0	28.7	19.5	22.6	26.1	29.1	24.3
50 mg/l Kinetin	18.2	20.1	21.7	22.0	20.5	13.0	15.7	16.3	16.7	15.4
100 mg/l Kinetin	26.3	27.3	27.7	28.3	27.4	18.2	20.1	27.3	29.7	23.8
200 mg/l Kinetin	19.3	21.3	22.3	23.7	21.7	16.1	16.1	16.6	19.6	17.1
Mean (A)	21.5	22.7	23.5	24.2		16.3	17.4	20.1	21.6	
L.S.D. at 5%	A: 1.7		B: 2.3	AB: 4.6		A: 1.2		B: 1.8	AB: 3.6	

Table 3. Effect of bio-phosphate and Algae extract & Kinetin treatments on yield components of black cumin plants during 2021/2022 and 2022/2023 seasons.

Algae extract and Kinetin treatments (B)	Bio-phosphate (mg/l) treatment (A)									
	control	Bio-Ph. 200	Bio-Ph. 400	Bio-Ph. 800	Mean (B)	control	Bio-Ph. 200	Bio-Ph. 400	Bio-Ph. 800	Mean (B)
	First season(2021/2022)					Second season(2022/2023)				
Number of capsules/ plant										
Control (tab water)	101.4	103.2	102.2	106.2	103.3	100.2	102.6	105.3	107.7	104.0
2 ml/l Algae extract	110.4	102.3	107.3	109.3	107.3	105.3	106.1	108.4	113.4	108.3
4 ml/l Algae extract	117.3	117.3	134.4	138.3	126.8	109.1	111.1	119.2	132.2	117.9
8 ml/l Algae extract	150.7	156.4	167.6	177.3	163.0	126.2	138.1	150.7	161.4	144.1
50 mg/l Kinetin	107.2	119.2	124.2	128.4	119.8	105.7	115.3	123.7	135.7	120.1
100 mg/l Kinetin	146.4	156.2	163.2	175.2	160.3	121.3	135.3	147.9	158.3	140.7
200 mg/l Kinetin	128.4	131.2	139.2	145.2	136.0	111.7	129.1	127.2	142.3	127.6
Mean (A)	123.1	126.6	134.0	140.0		111.4	119.7	126.1	135.9	
L.S.D. at 5%	A: 2.8		B: 6.4	AB: 12.8		A: 2.3		B: 5.2	AB: 10.4	
Seed yield / plant (g)										
Control (tab water)	22.4	26.1	27.3	29.2	26.3	22.3	23.3	24.4	26.1	24.0
2 ml/l Algae extract	25.3	28.4	29.5	31.2	28.6	23.7	24.1	25.1	27.1	25.0
4 ml/l Algae extract	28.1	30.1	33.4	35.4	31.8	24.4	26.1	26.43	33.4	27.6
8 ml/l Algae extract	35.2	37.4	47.1	50.2	42.5	35.4	36.3	38.3	43.3	38.3
50 mg/l Kinetin	26.2	29.4	31.2	32.2	29.8	23.4	24.9	26.3	28.7	25.8
100 mg/l Kinetin	34.1	36.2	44.4	46.3	40.3	31.3	35.3	37.3	40.4	36.1
200 mg/l Kinetin	28.3	32.3	33.2	35.4	32.3	27.3	29.4	31.3	35.3	30.8
Mean (A)	28.5	31.4	35.2	37.1		26.8	28.5	29.9	33.5	
L.S.D. at 5%	A: 1.7		B: 2.4	AB: 4.8		A: 1.4		B: 2.1	AB: 4.2	
Seed yield / fed. (kg)										
Control (tab water)	403.2	419.3	435.5	451.6	427.4	364.6	390.7	416.8	433.2	401.3
2 ml/l Algae extract	422.3	441.6	463.5	499.7	456.8	385.4	397.1	423.2	485.5	422.8
4 ml/l Algae extract	465.6	483.5	538.4	554.5	510.5	403.2	429.3	434.7	552.2	454.9
8 ml/l Algae extract	561.5	612.6	760.3	775.3	677.4	560.6	601.8	642.6	693.5	624.6
50 mg/l Kinetin	419.3	483.5	479.7	515.8	474.6	370.7	424.7	445.5	518.4	439.8
100 mg/l Kinetin	552.2	596.7	739.6	755.7	661.1	509.7	612.2	652.6	674.8	612.3
200 mg/l Kinetin	451.6	535.8	548.4	584.5	530.1	456.8	483.6	499.7	569.9	502.5
Mean (A)	468.0	510.4	566.5	591.0		435.9	477.1	502.2	561.1	
L.S.D. at 5%	A: 13.9		B: 18.5	AB: 37		A: 11.3		B: 16.7	AB: 33.4	

seasons. The optimal results for the number of capsules per plant, seed yield per plant (g), and seed yield per feddan (kg) were obtained with 800 g/l bio-phosphate. Bio-phosphate 800 g/l, followed by 400 g/l and 200 g/l in both seasons, enhanced the number of capsules per plant of black cumin plants compared to the control treatment. In terms of numbers, the increase in seed output per plant in the first and second seasons was 30.2% and 25.0%, respectively. In contrast, the 200 g/l and 400 g/l bio-phosphate treatments yielded intermediate values, respectively, above that check treatment. Additionally, the two bio-phosphate treatments both increased seed yield/fed, with 400 g/l bio-phosphate producing a slightly higher yield/fed than 200 g/l bio-phosphate treatments. The increase in seed yield/fed reached 26% in the first season and 29% in the second season when compared to untreated plants. The obtained results of yield components in this concern are in harmony with the findings of Gamal *et al.*,(2012) on *Nigella sativa*; Badran *et al.*, (2017) and Abdou (2020) on fennel; Mohammad *et al*., (2012), Hassan *et al.*, (2013) and Mahmoud *et al*., (2021) on anise; Rekaby (2013), Nasr Alla *et al.*, (2016) and Ghatas (2020) on coriander; Walid (2021) on dill El-Naggar *et al.*,(2011) on *Hibiscus sabdariffa.*

The results in Table 3 show that all treatments of kinetin (50, 100, and 200 mg/l kinetin) and algae extract (2, 4, and 8 ml/l kinetin) were substantially significant in generating more capsules per plant, seeds per plant, and feddan than unfertilized plants over the two seasons. Out of the six therapies that were tested, 8 ml/l of algae extract was recommended. Followed by 100 mg/l Kinetin, 200 mg/l Kinetin, 4 ml/l Algae extract, 50 mg/l Kinetin. Such two superior treatments increased seed yield/ plant, over that of the control treatment by 61.6 & 53.2 % respectively in the first season and 59.6 & 50.4 %, respectively in the second one .for seed yield per feddan had followed the same trend shown previously for seed yield per plant and recorded, in descending order 677.4, 661.1, 530.1, 510.5, 474.6, 456.8 and 427.4 Kg for the treatments of 8 ml/l Algae extract, 100 mg/l Kinetin, 200 mg/l Kinetin, 4 ml/l Algae extract, 50 mg/l Kinetin, 2 ml/l Algae extract and control, respectively, in the first season. The related yield values for the same individual treatments were 624.6, 612.3, 502.5, 454.9, 439.8, 422.8 and 401.3 Kg in the second season, (table 3). As an increasing percent for the six respective treatments over the control one, it came to 59, 55, 24, 19, 11 and 7 % in the first season, and 56, 53, 25, 13, 10 and 5 % in second season. In harmony with these results in respect with Algae extract or /and Kinetin treatments on number of capsules / plant, seeds yield / plant and / feddan reported by Shoukat *et al.,* (2011), Khalid *et al*.,(2014) and Khalid (2017) on black cumin ; Toaima *et al.*,(2017)on dill; Reem *et al*.,(2022) on fennel; El Laban *et al*., (2017)on Dutch fennel; Faizan *et al*.,(2011) and Adeleh *et al.*,(2015) on Safflower; Yasser *et al*.,(2022) on *Hibiscus sabdariffa* .

The effect of the interaction between bio-phosphate as a main factor and the Algae extract & Kinetin treatments as a secondary factor was significant in both seasons. except 2 ml/l Algae extraction treatment, had no significant in both seasons ,for number of capsules and except 2 ml/l Algae extraction treatment in both seasons and 50 mg/l Kinetin in the second season for seeds yield / plant ,for seeds yield / fed. Such treatment gave a significant, yield (775.3 or 755.7 kg/fed. in the first season and 693.5 or 674.8 kg/fed. in the second season) to that given by the traditional treatment (control which gave 403.2 kg/ fed. in the first season and 364.6 Kg/ fed. in the second season) as obviously indicated in Table 3.The best and economical results concerning for number of capsules / plant, seeds yield / plant and / feddan were obtained due to bio-phosphate 800 g/l in the combination with Algae extract (8 ml/l) or Kinetin (100 mg/l), as illustrated in Table 3.

3.3.Seed oil productivity:

The oil characteristics of *Nigella sativa* plants were dramatically and significantly increased owing to the use of tested bio-phosphate treatments (200,400 and 800 g/l), resulting in a notable and considerable rise in black cumin, fixed oil percent in seeds, and oil output per plant and per feddan in the two seasons. For oil percent in seeds and oil production per plant, the application of 800 g/l bio-phosphate generated the

Table 3. Effect of bio-phosphate and Algae extract & Kinetin treatments on fixed oil parameters of black cumin plants during 2021/2022 and 2022/2023 seasons.

Algae extract Algae extract and Kinetin treatments (B)	Bio-phosphate (mg/l) treatment (A)									
	control	Bio-Ph. 200	Bio-Ph. 400	Bio-Ph. 800	Mean (B)	control	Bio-Ph. 200	Bio-Ph. 400	Bio-Ph. 800	Mean (B)
	First season(2021/2022)					Second season(2022/2023)				
	fixed oil %									
Control (tab water)	23.1	23.6	24.3	25.0	24.0	23.3	25.1	25.4	27.2	25.3
2 ml/l Algae extract	25.4	27.2	28.2	31.2	28.0	25.1	26.6	27.1	27.2	26.5
4 ml/l Algae extract	26.2	28.4	29.4	31.3	28.8	27.2	29.1	30.1	30.2	29.2
8 ml/l Algae extract	29.7	33.3	34.3	34.7	33.0	32.4	33.4	34.2	35.3	33.8
50 mg/l Kinetin	25.3	25.4	26.3	32.3	27.3	23.3	26.2	28.4	30.4	27.1
100 mg/l Kinetin	28.4	31.3	34.3	34.3	32.1	29.1	32.1	33.6	34.6	32.3
200 mg/l Kinetin	25.4	27.9	29.2	30.2	28.2	25.3	27.3	29.3	31.4	28.3
Mean (A)	26.2	28.2	29.4	31.3		26.5	28.5	29.7	30.9	
L.S.D. at 5%	A: 1.8		B: 2.6	AB: 5.2		A: 1.4		B: 1.7	AB: 3.4	
	Oil yield/ plant (ml)									
Control (tab water)	5.75	6.16	6.57	6.99	6.37	5.05	5.44	5.84	6.25	5.65
2 ml/l Algae extract	6.76	7.57	8.40	9.61	8.09	5.97	7.11	8.76	10.54	8.10
4 ml/l Algae extract	7.29	9.64	10.52	11.36	9.70	6.49	8.55	8.99	12.67	9.18
8 ml/l Algae extract	10.5	12.55	16.82	17.33	14.30	10.8	12.31	13.04	14.91	12.77
50 mg/l Kinetin	6.50	7.79	8.97	11.25	8.63	5.92	6.84	7.59	9.63	7.50
100 mg/l Kinetin	9.57	11.46	14.96	15.44	12.86	8.99	10.58	12.92	13.74	11.56
200 mg/l Kinetin	7.56	8.80	10.86	13.67	10.22	6.51	8.02	8.88	10.57	8.50
Mean (A)	7.70	9.14	11.01	12.24		7.10	8.41	9.43	11.19	
L.S.D. at 5%	A: 1.39		B: 1.24	AB: 2.48		A: 1.26		B: 0.89	AB: 1.78	
	Oil yield/ fed. (kg)									
Control (tab water)	119.9	135.4	156.9	179.6	148.0	100.6	116.3	139.2	154.2	127.6
2 ml/l Algae extract	132.5	165.4	184.9	197.2	170.0	111.5	135.4	155.3	178.5	145.2
4 ml/l Algae extract	159.3	173.6	192.8	205.2	182.7	123.3	139.1	154.3	193.9	152.6
8 ml/l Algae extract	202.3	251.3	279.6	293.8	256.8	198.5	210.3	235.4	251.4	223.9
50 mg/l Kinetin	125.6	141.7	175.2	197.9	160.1	120.7	134.5	140.7	177.3	143.3
100 mg/l Kinetin	211.3	252.9	269.1	284.9	254.6	186.7	218.3	231.5	249.6	221.5
200 mg/l Kinetin	135.9	142.8	206.8	273.4	189.7	144.7	152.7	162.7	189.9	162.5
Mean (A)	155.3	180.4	209.3	233.1		140.9	158.1	174.2	199.3	
L.S.D. at 5%	A: 10.3		B: 13.9	AB: 27.8		A: 8.7		B: 11.3	AB: 22.6	

greatest values of oil%, followed by 400 g/l bio-phosphate and 200 g/l bio-phosphate as shown in Table 4. for Oil seed yield/ fed. was also increased due to the high dose of bio-phosphate treatments and reached (50% in the first season and 41% in the second season) the two bio-phosphate treatments, among these two treatments, 400 g/ l bio-phosphate proved to give slightly heavier yield/ fed. than 200 g/ l bio-phosphate treatments. Our results are, in harmony with those of Badran *et al.,* and Gamal *et al.,* 2012) on *Nigella sativa;* Ghatas *et al*., (2020) on *Oenothera biennis.*

The presented data in Table 4, showed that spraying all six treatments (2,4 and 8 ml/l Algae extract & 50,100 and 200 mg/l Kinetin) was significant for fixed oil percent in seeds and oil yield per plant and per feddan in the two seasons, compared to control treatment (tab water) in both seasons. Among the six sprayed treatments the highest fixed oil percent in seeds was given by sparying 8 ml/l Algae extract or /and100 mg/l Kinetin as shown in Table (4) followed by 4 ml/l Algae extract, 200 mg/l Kinetin, 2 ml/l Algae extract in the first season, while the lowest values were produced by 50 mg/l Kinetin except 2 ml/l Algae extract in the second season no significant differences existed. And for Fixed oil yield per plant the the highest Fixed oil yield per plant was given by sparying 8 ml/l Algae extract or /and100 mg/l Kinetin as shown in Table (3), also for fixed oil percent per feddan except 50 mg/l Kinetin in the first season. In descending order, the highest values were provided by 8 ml/l Algae extract, 100 mg/l Kinetin, 200 mg/l Kinetin, 4 ml/l Algae extract, 2ml/l Algae extract while the least oil yield was produced due to the 50 mg/l Kinetin. The numerically increased oil yield for such six treatments, in comparison with that of unfertilized treatment, reached 73.5, 72.1, 28.2, 23.4, 14.9 and 8.2% respectively in the first season. The comparable rise, resulting from the same treatments, came to 75.5, 73.6, 27.4, 19.6, 13.8 and 12.3% in the second season. These results are consistent with those of (Khalid *et al*., 2014 and Khalid 2017) on *Nigella sativa*.

In regard to the effect of the interaction treatments on fixed oil percentage in seeds and oil yield per plant and per feddan in duel seasons, data showed in Table 4 showed that all treatments effectd and the highest values were obtained from the treatment of applaying bio-phosphate 800 g/l in combination with 8 ml/l Algae extract or /and100 mg/l Kinetin in the two seasons as clearly illustrated in Table 4, except 2 ml/l Algae extract in the second season for fixed oil % in seeds and for fixed oil percent per feddan except 50 mg/l Kinetin in the 1st season, no significant differences were lived, in comparison with control treatment which did not receive any treatments.

3.4.Chemical properties:

3.4.1. Photosynthetic pigments

Table 5 presents data on the response of carotenoids, chlorophyll (a), and chlorophyll (b) to bio-phosphate treatments in fresh leaves of black cumin plants. Significant differences were found between the three treatments for Photosynthetic pigments in the two seasons, indicating that all bio-phosphate treatments produced significantly higher values than the control treatment (Table,5). In comparison to the control ones in both seasons, the bio-phosphate treatment produced the highest values for the three photosynthetic pigments (800 g/l), followed by 400 g/l and 200 g/l, respectively. The role of phosphate dissolving bacteria in stimulating the photosynthetic pigments was observed on black cumin (Badran *et al.,* 2012); Anise(Hassan *et al*., 2013) ; fennel (Badran *et al*., 2017) ; coriander (Ghatas 2020) ; dill (Walid 2021) ; basil (Mohamed *et al.,* 2015) and *Hibiscus sabdariffa* (El-Naggar *et al.,* 2011).

The data obtained in Table 5 showed that the amounts of "chlorophyll a, b, and carotenoids" in the leaves of *Nigella sativa* plants throughout both experimental seasons varied significantly and favorably across the different algae extract and kinetin treatments. All six Algae extract and Kinetin treatments resulted in higher values of the three pigments in both seasons than given by untreated control treatment, except 50 mg/l of Kinetin in the 2nd season for the contents of chlorophyll a, no significant differences were existed. However, the same trend for the photosynthetic pigments, in duel seasons, was observed as the highest values, in descending order, were obtained due to the treatments of 8 ml/l Algae extract, 100 mg/l Kinetin, 4 ml/l Algae

Table 5. Effect of bio-phosphate and Algae extract & Kinetin treatments on photosynthetic pigments content (mg/g f.w.) of black cumin plants during 2021/2022 and 2022/2023 seasons.

Algae extract Algae extract and Kinetin treatments (B)	Bio-phosphate (mg/l) treatment (A)									
	control	Bio-Ph. 200	Bio-Ph. 400	Bio-Ph. 800	Mean (B)	control	Bio-Ph. 200	Bio-Ph. 400	Bio-Ph. 800	Mean (B)
	First season(2021/2022)					Second season(2022/2023)				
Chlorophyll (A) content (mg/g f.w.)										
Control (tab water)	1.162	1.238	1.268	1.411	1.270	1.215	1.297	1.336	1.514	1.340
2 ml/l Algae extract	1.257	1.349	1.351	1.472	1.357	1.255	1.349	1.451	1.514	1.392
4 ml/l Algae extract	1.337	1.409	1.419	1.544	1.427	1.355	1.479	1.575	1.678	1.522
8 ml/l Algae extract	1.373	١,٤٤٥	1.669	1.925	1.628	1.464	1.641	1.669	1.825	1.650
50 mg/l Kinetin	1.226	1.281	1.351	1.472	1.332	1.255	1.310	1.322	1.472	1.340
100 mg/l Kinetin	1.448	1.546	1.641	1.781	1.604	1.457	1.595	1.650	1.734	1.609
200 mg/l Kinetin	1.283	1.349	1.419	1.544	1.399	1.323	1.409	1.486	1.575	1.448
Mean (A)	1.298	1.388	1.445	1.593		1.332	1.440	1.498	1.616	
L.S.D. at 5%	A: 0.081	B: 0.052		AB: 0.104		A: 0.062	B: 0.033		AB: 0.066	
Chlorophyll (B) content (mg/g f.w.)										
Control (tab water)	0.237	0.263	0.284	0.361	0.286	0.259	0.285	0.331	0.352	0.307
2 ml/l Algae extract	0.309	0.364	0.382	0.483	0.384	0.381	0.418	0.435	0.454	0.422
4 ml/l Algae extract	0.381	0.463	0.483	0.550	0.469	0.378	0.395	0.417	0.511	0.425
8 ml/l Algae extract	0.618	0.608	0.613	0.676	0.629	0.625	0.648	0.651	0.676	0.650
50 mg/l Kinetin	0.278	0.325	0.350	0.386	0.335	0.338	0.364	0.386	0.415	0.376
100 mg/l Kinetin	0.515	0.561	0.608	0.647	0.583	0.569	0.591	0.608	0.638	0.601
200 mg/l Kinetin	0.381	0.394	0.415	0.454	0.411	0.340	0.427	0.448	0.483	0.424
Mean (A)	0.388	0.426	0.448	0.508		0.413	0.447	0.468	0.504	0.307
L.S.D. at 5%	A: 0.027	B: 0.042		AB: 0.08٤		A: 0.031	B: 0.053		AB: 0.106	
Carotenoids content (mg/g f.w.)										
Control (tab water)	0.542	0.567	0.592	0.616	0.579	0.491	0.513	0.535	0.558	0.524
2 ml/l Algae extract	0.595	0.619	0.669	0.773	0.664	0.538	0.604	0.650	0.673	0.617
4 ml/l Algae extract	0.641	0.665	0.740	0.764	0.703	0.580	0.602	0.669	0.692	0.636
8 ml/l Algae extract	0.756	0.793	0.809	0.838	0.799	0.688	0.724	0.733	0.775	0.730
50 mg/l Kinetin	0.558	0.592	0.619	0.748	0.629	0.505	0.535	0.561	0.740	0.585
100 mg/l Kinetin	0.687	0.785	0.814	0.826	0.778	0.622	0.711	0.737	0.766	0.709
200 mg/l Kinetin	0.595	0.669	0.719	0.743	0.681	0.539	0.561	0.607	0.762	0.619
Mean (A)	0.625	0.670	0.709	0.758		0.566	0.607	0.641	0.709	
L.S.D. at 5%	A: 0.041	B: 0.033		AB: 0.066		A: 0.035	B: 0.027		AB: 0.054	

extract, 200 mg/l Kinetin, 2 ml/l Algae extract and 50 mg/l Kinetin, (Table,4). Therefore, the 8 ml/l Algae extract or 100 Kinetin proved much more effective than the lower dose of Algae extract or Kinetin. In contrast, the Algae extract at 4 ml/l and 200 mg/l of Kinetin treatments gave intermediate values. The role of Algae extract or /and Kinetin in promoting the three photosynthetic pigments was demonstrated by Hanan *et al*.,(2011) on sunflower; Mohammed *et al*.,(2018)on stevia plants; Shehata *et al*., (2011) on Celeriac Plant.

Concerning the interaction between bio–phosphate and Algae extract & Kinetin treatments, it was significant, in both seasons, as illustrated in Table 5.The best overall results for the three pigments in both seasons were due to some interacting treatments between the two tested factors, a high dose of bio–phosphate at 800 g/l in combination with Algae extract (8 ml/l) or Kinetin (100 mg/l), as clearly illustrated in Table 5.

3.4.2. NPK percentages:

Referring to the effect of supplying treatments on nitrogen, phosphorus and potassium percentage, obtained data in Table 6 indicated that all used bio–phosphate treatments caused significant promotion in the herb contents of NPK % over the check plants (unsupplied), in both experimental seasons. Bio-phosphate at 800 g/l produced the highest percentages of nitrogen, phosphorus, and potassium, followed by 400 g/l and 200 g/l, respectively. In comparison to the control treatments in both seasons, bio-phosphate at 200 g/l produced the lowest percentages. Regarding bio-phosphate, these results agreed with those of *Nigella sativa* (Badran *et al.,* and Gamal *et al.,* 2012); Anise(Hassan *et al*., 2013) ; fennel (Badran *et al*., 2017) ; coriander (Ghatas 2020) ; basil (Mohamed *et al.,* 2015) ; *Hibiscus sabdariffa* (El-Naggar *et al.,* 2011) ; *Camellia sinensis* (Chakrabort *et al.,* 2012).

The results listed in Table 6 show the influence of spraying Algae extract or Kinetin treatments on N, P and K percentages in the dry herb of *Nigella sativa* L. plants in the two experimental seasons. The herb percent of nitrogen, phosphorus and potassium of *Nigella sativa* plants was significantly promoted, in both seasons, due to supplying the plants with each one of the six tested Algae extract or Kinetin treatments comparing to those of untreated treatment as declared in Table (5) The highest values for the N, P and K% among the six tested treatments were given by Algae extract (8 ml/l) or Kinetin (100 mg/l) followed by4 ml/l Algae extract, 200 mg/l Kinetin, 2 ml/l Algae extract and 50 mg/l Kinetin as shown in (Table, 5). The role of Algae extract or Kinetin treatments in stimulating the herb % of such three nutrient minerals was emphasized by Khalid *et al*.,(2014)on *Nigella sativa* Reem *et al*.,(2022) on fennel Shehata *et al*., (2011) on Celeriac Plant.

The interaction between bio-phosphate and Algae extract & Kinetin treatments was significant in the two seasons, Table 6, The highest value of nitrogen, phosphorus and potassium %, was obtained with the treatment of applying *Nigella sativa* plants with a high dose of bio-phosphate 800 g/l in combination with Algae extract (8 ml/l) or Kinetin (100 mg/l) as clearly illustrated in Table (5) in the two experimental seasons.

4. CONCLUSION

Under the same study circumstances, it can be recommended *Nigella sativa* L. plants be applied with high concentrations of algae extract (8 ml/l) or kinetin (100 mg/l) in conjunction with bio-phosphate 800 g/l to improve growth, seed production, and fixed oil components.

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Table 6. Effect of bio-phosphate and Algae extract & Kinetin treatments on NPK percentages of black cumin plants during 2021/2022 and 2022/2023 seasons.

Algae extract and Kinetin treatments (B)	Bio-phosphate (mg/l) treatment (A)									
	control	Bio-Ph. 200	Bio-Ph. 400	Bio-Ph. 800	Mean (B)	control	Bio-Ph. 200	Bio-Ph. 400	Bio-Ph. 800	Mean (B)
	First season(2021/2022)					Second season(2022/2023)				
Nitrogen percentage										
Control (tab water)	1.272	1.352	1.511	1.670	1.451	1.272	1.375	1.566	1.590	1.451
2 ml/l Algae extract	1.375	1.431	1.590	1.693	1.522	1.431	1.534	1.646	1.693	1.576
4 ml/l Algae extract	1.431	1.566	1.725	1.773	1.624	1.511	1.566	1.773	1.908	1.689
8 ml/l Algae extract	1.725	1.805	1.988	2.091	1.902	1.649	1.742	1.845	2.026	1.815
50 mg/l Kinetin	1.296	1.431	1.455	1.773	1.489	1.352	1.487	1.587	1.705	1.532
100 mg/l Kinetin	1.614	1.749	1.932	2.067	1.840	1.634	1.729	1.864	1.923	1.787
200 mg/l Kinetin	1.352	1.455	1.511	1.805	1.530	1.431	1.534	1.662	1.752	1.595
Mean (A)	1.438	1.541	1.673	1.839		1.468	1.567	1.706	1.800	
L.S.D. at 5%	A: 0.051		B: 0.037	AB: 0.074		A: 0.066		B: 0.044	AB: 0.088	
Phosphorus percentage										
Control (tab water)	0.205	0.246	0.287	0.328	0.267	0.192	0.231	0.269	0.308	0.250
2 ml/l Algae extract	0.287	0.287	0.411	0.411	0.349	0.269	0.269	0.386	0.386	0.327
4 ml/l Algae extract	0.246	0.395	0.452	0.534	0.407	0.231	0.371	0.424	0.501	0.382
8 ml/l Algae extract	0.369	0.452	0.575	0.598	0.498	0.346	0.424	0.539	0.561	0.468
50 mg/l Kinetin	0.246	0.287	0.328	0.369	0.308	0.231	0.269	0.308	0.346	0.288
100 mg/l Kinetin	0.328	0.352	0.493	0.573	0.436	0.327	0.338	0.482	0.538	0.421
200 mg/l Kinetin	0.209	0.328	0.411	0.493	0.360	0.194	0.308	0.386	0.462	0.337
Mean (A)	0.271	0.335	0.422	0.472		0.256	0.316	0.399	0.443	
L.S.D. at 5%	A: 0.043		B: 0.03٥	AB: 0.07٠		A: 0.034		B: 0.029	AB: 0.058	
Potassium percentage										
Control (tab water)	1.642	1.929	2.053	2.586	2.053	1.540	1.810	1.926	2.426	1.926
2 ml/l Algae extract	1.929	2.053	2.586	2.750	2.330	1.810	1.926	2.426	2.580	2.185
4 ml/l Algae extract	2.586	2.750	2.874	3.161	2.843	2.426	2.580	2.696	2.965	2.667
8 ml/l Algae extract	2.791	3.202	3.284	3.530	3.202	2.618	3.004	3.081	3.312	3.004
50 mg/l Kinetin	1.642	1.765	2.340	2.586	2.083	1.540	1.656	2.195	2.426	1.954
100 mg/l Kinetin	2.685	2.997	3.243	3.489	3.103	2.519	2.812	3.042	3.273	2.911
200 mg/l Kinetin	2.176	2.340	2.463	2.997	2.494	2.041	2.195	2.311	2.812	2.340
Mean (A)	2.207	2.434	2.692	3.014		2.071	2.283	2.525	2.828	
L.S.D. at 5%	A: 0.074		B: 0.025	AB: 0.05٠		A: 0.068		B: 0.024	AB: 0.048	

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الملخص العربي

استجابة نباتات حبة البركة لبعض معاملات المنشطات الحيوية في النمو والإنتاجية والمكونات الكيميائية

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أجريت تجربة حقلية خلال الموسمين المتتاليين ٢٠٢٢/٢٠٢١ و ٢٠٢٣/٢٠٢٢ بمزرعة محطة بحوث البساتين بسدس – محافظة بنى سويف – مركز البحوث الزراعية بهدف تحسين نمو وإنتاجية ومحصول الزيت من بذور نباتات حبة البركة بواسطة الرش الورقى بالفوسفات الحيوي وكذلك مستخلص الطحالب والكينتين . أظهرت النتائج أن أفضل صفات النمو الخضري (طول النبات، سمك الساق وطول الجذر)، صفات المحصول (عدد الكبسولات لكل نبات، محصول البذور للنبات الواحد والمحصول للفدان)، صفات الزيت (النسبة المئوية للزيت الثابت ومحصول الزيت الثابت للنبات وللفدان)، صبغات الكلوروفيل ونسبة NPK في العشب الجاف. وذلك من خلال الرش الورقي للفوسفات الحيوي بمعدل ٨٠٠ جم / لتر مع الجرعة الاعلى من مستخلص الطحالب (٨ مل / لتر) أو الكينتين بمعدل ١٠٠ ملجم/لتر ولا توجد فروق معنوية بين مستخلص الطحالب (٨ مل/ لتر) والكينيتين (١٠٠ ملجم/ لتر). من هذه الدراسة يمكن التوصية بمعاملة نباتات حبة البركة بالتركيز العالي من مستخلص الطحالب (٨ مل / لتر) أو الكينتين (١٠٠ ملجم / لتر) مع الفوسفات الحيوي ٨٠٠ جم/لتر بهدف تعزيز النمو ومحصول البذور وكذلك محصول الزيت الثابت، تحت نفس الظروف البيئية لمحافظة بني سويف.