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Original Article

The interaction impact of compost and biostimulants on growth, yield and oil content of black cumin (*Nigella sativa* L.) plants

O impacto da interação de composto e bioestimulantes no crescimento, rendimento e teor de óleo de plantas de cominho-preto (*Nigella sativa* L.)

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Abstract

This study was conducted during the seasons 2020/2021 and 2021/2022 to investigate the effect of the interaction between compost and biostimulants on growth measurements, seed yield, and oil production of black cumin (*Nigella sativa* L.). Four levels of fertilizer (0.6, 12, 18 tons/ha) were used. While the biostimulants were ascorbic acid (AS) at 100 ppm, yeast extract (YE) at 8 g/L, and AS at 100 ppm + 8 g/L YE. The plants were treated with these stimuli as follows: control (without addition), 100 ppm, YE at 8 g/L, and the results showed that fertilization at all levels, as well as foliar spraying with the used stimuli, led to a significant increase. Growth parameters, number of capsules, seed production, as well as fixed and volatile oil production, and plant treatment with organic fertilizers at a high level (18 tons). /ha) recorded the highest values for the trait under study. It was also shown that the foliar treatment at a concentration of 8 g/l YE was more effective in increasing the previously studied variables. All interactions were great. Most of the composite coefficients increased significantly for all the traits studied. Moreover, the application of manure at a high rate (18 t/ha) at a rate of 8 g/l YE was the best treatment. GC-MS analysis of the volatile and persistent oil showed that the main constituents of both species were also affected by the use of organic fertilizers and biotreatments. The combination of high-rate fertilizer (18 t/ha) plus AS at 100 ppm + 8 g/L YE improved main oil components compared to untreated plants.

Keywords: compost, ascorbic acid, yeast extract, black cumin, *Nigella sativa* L.

Resumo

Este estudo foi realizado durante as temporadas 2020/2021 e 2021/2022 para investigar o efeito da interação entre composto e bioestimulantes nas medidas de crescimento, rendimento de sementes e produção de óleo de cominho-preto (*Nigella sativa* L.). Quatro níveis de fertilizantes (0,6, 12, 18 ton/ha) foram usados, enquanto os bioestimulantes foram ácido ascórbico (AS) a 100 ppm, extrato de levedura (YE) a 8 g/L e AS a 100 ppm + 8 g/L YE. As plantas foram tratadas com esses estímulos da seguinte forma: controle (sem adição), 100 ppm e YE a 8 g/L. Os resultados mostraram que a adubação em todos os níveis bem como a pulverização foliar com os estímulos utilizados levaram a um aumento significativo aumentar. Parâmetros de crescimento, número de cápsulas, produção de sementes, bem como produção de óleo fixo e volátil e tratamento de plantas com fertilizantes orgânicos em alto nível (18 ton/ha), registraram os maiores valores para as características em estudo. Também foi demonstrado que o tratamento foliar na concentração de 8 g/l YE foi mais eficaz em aumentar as variáveis estudadas. Todas as interações foram ótimas. A maioria dos coeficientes compostos aumentou significativamente para todas as características estudadas. Além disso, a aplicação de esterco em alto nível (18 ton/ha) na taxa de 8 g/l YE foi o melhor tratamento. A análise GC-MS do óleo volátil e persistente mostrou que os principais constituintes de ambas as espécies também foram afetados pelo uso de fertilizantes orgânicos e biotratamentos. A combinação de fertilizante de alto nível (18 ton/ha) + AS a 100 ppm + 8 g/L YE melhorou os principais componentes do óleo em comparação com plantas não tratadas.

Palavras-chave: composto, ácido ascórbico, extrato de levedura, cominho-preto, *Nigella sativa* L.

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1. Introduction

Medicinal plants have been used since immemorial in foods, spices, and treating diseases. Black cumin (Nigella sativa L.) is a winter annual flowering species in the Ranunculaceae family. This spice seed crop is native to the Mediterranean region and grows widely throughout the Middle East, Europe, and Asia. The plant is cultivated and grown all over the world (Aggarwal et al., 2008; Bayram et al., 2010; Mohamed et al., 2017). The main producers of Nigella sativa are India, Sri Lanka, Bangladesh, Pakistan, Afghanistan, Egypt, Iraq, Iran, Turkey, Syria, and Ethiopia. Ripe black cumin seeds contain about 7% moisture, 4.34% ash, 23% protein, 0.39% fat, 4.99% starch, and 5.44% crude fiber. The seeds are rich in fats, fiber, minerals such as iron, sodium, copper, zinc, phosphorous, calcium, and vitamins such as ascorbic acid, thiamin, niacin, pyridoxine, and folic acid (Takruri and Dameh, 1998; Mozaffari et al., 2000; Sultana et al., 2018). Moreover, Nigella Sativa seeds contain 30-35% fixed oil and 0.5-1.5% volatile oil which has many uses in the pharmaceutical and food industries. Black cumin seeds contain protein, alkaloids (nigericin and nigellone), stable (α-here) saponins, and essential oil (Ashraf et al., 2005; Ozel et al., 2009).

Organic fertilizers have recently gained a lot of popularity as a useful way for sustainable agriculture to provide the nutritional needs of crops. Although organic fertilizers contain trace amounts of nutrients, they enhance soil fertility and production because they contain growth-promoting factors including enzymes and hormones. Applying compost to the soil improved the soil's water-holding capacity, which enhanced crop access to nutrients. Root cover conditions (structure, moisture, etc.) are also greatly improved by compost, and this enhances plant growth by increasing the number of microorganisms (Puma, 2001; Shaheen et al., 2007).

The nutrients in the compost are added gradually and are retained in the soil for a longer period, ensuring residual benefits on subsequent crops. (Ginting et al., 2003). In addition, it can be found in considerable quantities locally and is a less expensive way to increase soil fertility. Increased crop yield was achieved by adding organic manures, especially to sandy soil, which was deficient in organic matter, had undesirable physical and biological characteristics, and had higher N leaching (Awosika et al., 2014).

According to Norman (2004), compost can represent organic manure from both plant and animal sources. Plant sources include green manures, seaweeds, cover plants, crop residues, nitrogen fixed by microorganisms, mulch, and compost. Animal sources also include the dung of sheep, goats, cattle, horses, and poultry. Both major and minor nutrients can be found in compost from both vegetable sources. We used compost and poultry manure in this investigation. Ultimately, environmentally friendly agricultural practices for sustainable food production use organic and bio-fertilizers (Islam et al., 2017).

Ascorbic acid (Vitamin C) is a proven antioxidant and biostimulant that can protect plants from damage caused by aerobic metabolism and a range of pollutants. It acts as an enzyme cofactor. Further, ascorbic acid is highly effective in plant resistance to many plant pathogens such as fungi, bacteria, nematodes, and parasitic plants (Oertli, 1987 Mahdy, 1994). Ascorbic acid also has many other important roles such as antioxidant defense and photoprotection, as well as the regulation of growth and photosynthesis (Blokhina et al., 2003).

Yeast extract (Saccharomyces cerveace) has been used for a long time as a biofertilizer and also a biostimulant that is used in the production of horticultural crops, due to the positive, biological and physiological roles of yeast, which were described by some studies such as Nagodawithana (1991), where he indicated that yeast extract is a good source of many nutrients, Vitamin B, proteins, carbohydrates, enzymes, nucleic acids, and plant hormones which make it suitable for application to the leaves. Yeast extract plays an important role in providing many nutrients to plants (Khalil and Ismael, 2010). This study was planned to study the response of growth, seed yield, and production. Oil of black cumin plants (*Nigella sativa* L.) to compost and some biostimulants (ascorbic acid and yeast extract).

2. Materials and Methods

2.1.Description of the study site

This experiment was conducted at the farm of the Muhammadiyah project, Ma'an, Jordan during the two consecutive experimental seasons 2020/2021 and 2021/2022 to find out the effect of compost control and 6, 8, and 12 t/ha and foliar application of biostimulants represented by a concentration of 100 ppm of ascorbic acid (AS) and yeast extract (YE) at 8 g/L and AS at 100 ppm + 8 g/L YE and the interaction between the two factors on growth characteristics, seed yield, fixed and volatile oil (percentage and yield) of black cumin plants (*Nigella sativa* L.).

2.2. Experimental soil analysis

2.2.1. Experimental design and tested treatments

A split plot design with three replications, compost, 0, 6,12, and 18 tons/ha. was the main plots, Table 1 reviews the physical and chemical analysis of the soil used, while Table 2 indicates the physical and chemical properties of the compost used in this study. Biostimulants were ascorbic acid (AS) at 100ppm, yeast extract (YE) at 8 g/l, and AS at 100 ppm + 8 g /l YE were assigned as the subplots. Black cumin seeds were planted on November 10th of both seasons. The empirical plot was 3.0×2.5 m and contained 4 rows, 60 cm apart. The distances between the hills were 25 cm. and the plants were thinned 35 days later to two plants/hills. Compost was added at its three rates before sowing and during soil preparation, the plants, except the control, were treated with foliar sprayed with the two tested biostimulants, either separately or in combination three times as follows. After 60, 75, and 90 days from sowing respectively, in both seasons. The plants were foliar sprayed till runoff. All usual farming operations were performed. All agricultural practices were performed as usual.

At the end of the experiment in May, the following measurements were taken: plant height, number of branches/plants, shoot fresh and dry weight g / plant, number of capsules/plants, seed yield g /plant, and seed yield kg /ha. Was calculated, fixed oil percentage, fixed oil yield ml/plant, and fixed oil yield L / ha. Volatile oil percentage, volatile oil yield ml /plant, volatile oil yield L /ha. and volatile oil and fixed oils components.

2.3. Time and method of treatments

N, P, and K fertilizers (control) were added to the soil in half of the recommended dose as follows; Ammonia nitrate (33.5%) at 357 kg/ha, calcium superphosphate (15.5% P2O5) at 476 kg/ha, potassium sulfate (48% K2O) at 89.25 kg/ha, and phosphorus fertilizer was added during soil preparation. Nitrogen and potassium fertilizers were divided into two equal doses. The first dose was added 30 days after sowing and the second dose was added 30 days after the first dose.

2.4. Volatile and fixed oil percentage

The percentage of volatile oil in the air-dried seeds was determined according to the method of the British Pharmacopoeia (1963). However, the fixed oil percentage was predestined by the Soxhlet apparatus using petroleum ether (BP 40-60 °C) as solvent according to the Association of Official Agricultural Chemists (AOAC, 1980).

2.5. Statistical analysis

All acquired data were recorded in tables and statistically analyzed according to MSTAT-C (1986) using the L.S.D. test at 5% test for differences between all treatments according to Mead et al. (1993).

Table 1. Physical and chemical analysis of the soil used during the seasons 2020/2021 and 2021/2022.

Soil traits		experimental season	
		2020/2021	2021/2022
Physical analysis	Particle size distribution (%)		
	Coarse sand	5.85	6.28
	Fine sand	74.93	73.06
	Silt	14.14	15.74
	Clay	7.88	7.62
	Texture class	Sandy	Sandy
Chemical Analysis	EC. Dsm-1(1:1 ex.)	0.91	0.88
	pH (1:1 w/v)	7.82	7.92
	Organic matter (%)	0.77	0.82
	saturation capacity %	28.49	28.85
	Ca Meq/100g soil	0.60	0.71
	Mg Meq/100g soil	0.40	0.49
	NaMeq/100g soil	0.08	0.09
	KMeq/100g soil	0.03	0.05
	ClMeq/100g soil	0.25	0.32
	HCO_3Meq/100g soil	0.30	0.43

Table 2. The physical and chemical properties of the compost used in this study.

Properties	Value		Properties	Value	
	2020	2021		2021	2022
The dry weight of 1 m^3	450 kg	450	Total N %	1.5	1.6
Moisture (%)	25-30	25-30	Total P (%)	0.9	0.88
pH (1:10)	7.6	7.8	Total K (%)	1.4	1.31
E.C. (m mhose/cm)	2-3.5	2-3.6	Fe (ppm)	290	297
Organic matter %	37	38.6	Mn (ppm)	29.2	30.3
Organic carbon %	19.8	21.5	Cu (ppm)	150	151
C/N ratio	14.2	13.9	Zn (ppm)	152	175

The E.C. (m mhose/cm) represents electrical conductivity, while the C/N ratio represents the ratio of carbon to nitrogen.

3. Results and Discussion

3.1. Growth measurements

The data recorded in (Table 3) indicated a significant increase in plant height, number of main branches/ plants, plant freshness, and dry weight (g) of black cumin (*Nigella sativa* L.). Compost fertilizer at all levels, in both seasons, it can be seen that these parameters gradually increased with increasing fertilizer levels compared to untreated plants in both seasons. Therefore, the use of high-level organic fertilizer (18 t/ha) gave the best vegetative growth of plants ranging from 65.79, 68.15, 44.74, 42.86, 47.38, 56.62, 43.40, and 48.17% of control in both seasons. , respectively.

The efficiency of organic manures in improving growth parameters was appeared by Sanjeeva et al. (2018), Ali and Hassan (2014), Badran et al. (2012), Sayed and Hossein (2011) and Hassan et al. (2009) on Nigella sativa L. plants, Al-Fraihat et al. (2011) on marjoram, Abd El-Azim et al. (2017) on fennel, Pradeepa (2016) on chili pepper, Hady et al. (2016) on cluster bean, Hassan et al. (2015) on dill, Abdou and Badr (2022) on caraway plants and Mohamed et al. (2022) on basil plant.

About foliar spray AS and YE, the datum in (Table 3) evident that foliar application of black cumin plants with the two materials whether alone or in combination, in two experimental seasons, led to a significant improvement in all growth traits, in two seasons, as compared to non-sprayed plants.

Table 3. Effect of compost, ascorbic acid (AS), and yeast extract (YE) on growth measurements of *Nigella sativa* L. plants through the 2020/2021 and 2021/2022 seasons.

Compost(A)	The 2020/2021 season					x				
	Bio stimulant (B)									
	Control	AS	YE	AS+ YE	Mean	Control	AS	YE	AS+ YE	Mean
	Plant height (cm)									
Control	35.0	37.0	39.0	41.0	38.0	37.0	38.3	41.7	45	40.5
COM (1)	40	51	55.3	58	51.1	44.3	53.0	57.0	60.3	53.7
COM (2)	53.0	58.0	60.0	65.0	59.0	54.3	58.3	62.7	69.3	61.2
COM (3)	55.0	57.3	68.00	71.7	63.0	59.3	67.7	71.0	74.3	68.1
Mean	45.8	50.8	55.6	58.9		48.8	54.3	58.1	62.3	
L.S.D at $_{5\%}$	A= 2.9 B= 2.5 AB= 5.0					A= 1.8 B= 2.2 AB= 4.3				
	Number of branches/plants									
Control	6.9	7.4	7.9	8.2	7.6	7.9	8.3	8.6	8.8	8.4
COM (1)	8.5	8.7	9.0	9.2	8.9	9.2	9.4	9.9	10.2	9.7
COM (2)	9.5	9.8	10.1	10.3	9.9	10.3	10.6	10.9	11.4	10.8
COM (3)	9.9	10.4	11.4	12.4	11.0	10.9	11.5	12.5	13.2	12.0
Mean	8.7	9.1	9.6	10.0		9.6	10.0	10.4	10.9	
L.S.D at $_{5\%}$	A= 0.1 B= 0.1 AB= 0.2					A= 0.2 B= 0.1 AB= 0.3				
	Herb fresh weight (g)									
Control	62.3	66.2	68.0	70.3	66.7	63.4	66.7	70.3	71.7	68.0
COM (1)	72.8	75.2	78.0	80	76.5	77.9	80.8	82.2	84.8	81.4
COM (2)	92.8	94.5	96.5	97.8	95.4	98.8	100.3	102.7	104.3	101.5
COM (3)	94.5	97.5	99.8	101.5	98.3	103.1	104.5	108.3	110.2	106.5
Mean	80.6	83.4	85.6	87.4		85.8	88.1	90.9	92.8	
L.S.D at $_{5\%}$	A= 7.0 B= 1.3 AB= 2.6					A= 9.1 B= 1.6 AB= 3.2				
	Herb dry weight (g)									
Control	15.1	15.5	16.1	16.8	15.9	15.3	16.0	16.8	17.5	16.4
COM (1)	16.0	16.9	17.4	17.9	17.0	16.1	17.1	17.7	18.7	17.4
COM (2)	17.4	18.4	19.4	20.5	18.9	18.5	20.2	20.9	21.4	20.2
COM (3)	20.6	22.1	23.5	25.2	22.8	22.0	23.5	25.2	26.7	24.3
Mean	17.3	18.2	19.1	20.1		18.0	19.2	20.1	21.1	
L.S.D at $_{5\%}$	A= 1.5 B= 0.3 AB= 0.6					A= 1.7 B= 0.5 AB= 1.0				

COM (1) = 6, COM (2) = 12, and COM (3) = 18 ton/ha. of compost. AS = ascorbic acid (100 ppm). YE = yeast extract (8 g/l).

Spray application with AS at 100 ppm + 8 g /l YE record to be more efficient in improving plant height, branch number/plants, and fresh & dry weights of plants (g) than those gained by control and other treatments. The previous superior treatment improved these traits with percentages ranging from 28.60, 27.66, 14.94, 13.54, 8.44, 8.16, 16.18, and 17.22 higher than the control for the two experimental seasons, respectively.

The aforementioned results which show the effect of Ascorbic acid and yeast extract on growth parameters was In line with the findings of Ali (2001) on *Calendula officinalis*, Youssef and Talaat (2003) on rosemary, El-Sherbeny et al. (2007) on replants, El-Leithy et al., (2011) on geranium, Abdou et al. (2012) on Salvia officinalis, Abokutta (2016) on fennel, Abdou and Badr (2022) on caraway plants and Mohamed et al. (2022) on basil plant.

According to the nested transactions compost and biostimulant treatments, it is noted that it gave a highly significant on all studied growth traits in two seasons. The data specified that the best results it was received when using the high level of compost plus addition to AS at 100 ppm + 8 g / L YE when compared to other treatments in two experimental seasons, as shown in (Table 3).

3.2. Yield measurements

The data in Table 4 shows the number of capsules per plant, seed yield per plant (g), and hectares (tons). The effect was significant on *Nigella sativa* L. plants at all levels of organic fertilizer during the two study seasons. It was clear that all of them led significantly to an increase in yield measurements. Moreover, the highest values were the capsules number per plant and the total seed yield per plant (gm) and hectares (ton). It was obtained when black cumin plants were supplied with fertilizer at a high rate (18 tons/ha), which led to an increase that ranged between 29.98, 25.08, 45.51, 45.40, 45.36, and 45.23 over the control in the two seasons, respectively. The effectiveness of compost on increasing yield measurements was revealed by Sanjeeva et al. (2018), Ali and Hassan (2014), Badran et al. (2012), Sayed and Hossein (2011), and Hassan et al. (2009) on Nigella sativa L. plants, Hassan et al. (2015) on dill, Pradeepa (2016) on chili pepper, Abd El-Azim et al. (2017) on fennel, Ahmad H. Al-Fraihat et al. (2011) on marjoram, Abdou and Badr (2022) on caraway plants and Mohamed et al. (2022) on basil plant.

Table 4. Impact of compost, ascorbic acid (AS), and yeast extract (YE) on yield measurements of *Nigella sativa* L. plants during the 2020/2021 and 2021/2022 seasons.

Compost (A)	**2020/2021 season**					**The 2021/2022 season**				
	Bio stimulant (B)									
	Control	**AS**	**YE**	**AS+ YE**	**Mean**	**Control**	**AS**	**YE**	**AS+ YE**	**Mean**
	Capsules number/plant									
Control	51.7	59.3	60.7	63	58.7	55.7	62.0	63.3	64.7	61.4
COM (1)	63.7	64.7	66.0	68.0	65.6	65.7	66.7	68.0	69.7	67.5
COM (2)	69.0	70.7	72.0	74.3	71.5	72.0	74.0	76.0	78.7	75.2
COM (3)	72.3	74.0	78.3	80.7	76.3	73.0	75.0	78.0	81.0	76.8
Mean	64.2	67.2	69.3	71.5		66.6	69.4	71.3	73.5	
L.S.D at $_{5\%}$	A= 6.1 B= 1.1 AB= 2.2					A= 5.7 B= 1.3 AB= 2.6				
	Seed yield/plant (g)									
Control	13.8	15.2	16.1	17.3	15.6	14.3	16.2	17.0	17.7	16.3
COM (1)	16.6	18.2	19.2	20.2	18.5	16.7	18.7	20.2	21.0	19.1
COM (2)	19.2	20.2	21.2	22.2	20.7	19.7	20.5	22.0	23.2	21.3
COM (3)	21.2	22.2	23.2	24.2	22.7	22.2	23.2	24.2	25.2	23.7
Mean	17.7	18.9	19.9	21.0		18.2	19.6	20.8	21.8	
L.S.D at $_{5\%}$	A= 1.2 B= 0.3 AB= 0.6					A= 1.1 B= 0.4 AB= 0.8				
	Seed yield/ha. (ton)									
Control	1.328	1.457	1.542	1.661	1.497	1.364	1.552	1.632	1.696	1.561
COM (1)	1.59	1.744	1.840	1.936	1.778	1.606	1.792	1.936	2.016	1.838
COM (2)	1.84	1.936	2.032	2.128	1.984	1.888	1.968	2.112	2.224	2.048
COM (3)	2.032	2.128	2.224	2.320	2.176	2.128	2.224	2.320	2.396	2.267
Mean	1.698	1.816	1.910	2.011		1.747	1.884	2.000	2.083	
L.S.D at $_{5\%}$	A= 0.026 B= 0.021 AB= 0.041					A= 0.031 B= 0.032 AB= 0.064				

COM (1) = 6, COM (2) = 12, and COM (3) = 18 ton/ha. of compost. AS = ascorbic acid (100 ppm). YE = yeast extract (8 g/l).

Concerning Spraying with stimulants, data in (Table 4) show that the influence of them the growth parameters of black cumin was significant in both seasons. The recorded data show that the highest number of capsules/plant, seed yield is g/plant, and seed yield is ton/ha. Recorded data show that the highest number of capsules/plant, seed yield is gram/plant and seed yield is tons/ha. The result of foliar spraying with ascorbic acid and yeast extract together (100 ppm AS + 10 g/L YE) was better in increasing these measurements than those other treatments in both seasons. Numerically, this pre-treatment increased this fraction by 11.37, 10.36, 18.64, 19.78, 17.84, and 19.23 higher than the unsprayed plants for the two experimental seasons, respectively. The improvement of yield parameters due to treating ascorbic acid was explored by, Khalil et al. (2010), on sweet basil, and Ahmad Al-Fraihat et al. (2023) on rosemary, Concerning the use of yeast extract, there were similar results by Salman (2006), El-Keasy et al. (2011), Abd El-Salam Nora (2014), Abo kutta (2016) on fennel, Abdou and Badr (2022) on caraway plants and Mohamed et al. (2022) on basil plant., Yield measurements increases due to the interaction effect between compost and biological treatments on all black cumin growth treatments were significant in two seasons. The maximum effective treatments were obtained from treating the plants with a high rate of compost (18 tons/ha) with ascorbic acid (AS) at a rate of 100 ppm and 8 g/L of yeast extract (YE) as listed in Table 4.

3.3. Fixed oil production

The data shown in (Table 5) shows the fixed oil production (percentage, yield per plant (ml), and hectare (L) of Nigella sativa L. plants was greatly affected by compost treatment in both experimental seasons. Fertilizing the plants with a high rate of compost 18 tons/ha. it gave the best values as ranged 18.39& to 19.27,71.91 &73.23 and 71.67& 73.21 over the Untreated plants in both seasons, respectively. The role of organic fertilizer is increasing with the results obtained by Ali and Hassan (2014) on black cumin, Hassan et al. (2015) on Rosemary.

Regarding ascorbic acid (AS) and yeast extract (YE) treatments, the results gave out in Table 5 showed that foliar spraying with them either alone or together, in two experimental seasons, gave a high significance in constant fixed oil yield ml/plant and l/ha, for two seasons, comparing to unsprayed plants. spray the plants with the 100 ppm

Table 5. Effect compost, ascorbic acid (AS), and yeast extract (YE) on fixed oil percentage and yield of black cumin plants during the 2020/2021 and 2021/2022 seasons.

Compost (A)	2020/2021 season					The 2021/2022 season				
	Bio stimulant (B)									
	Control	AS	YE	AS+ YE	Mean	Control	AS	YE	AS+ YE	Mean
	Fixed oil %									
Control	21.87	22.27	23.12	23.67	22.73	22.50	23.05	23.67	24.00	23.30
COM (1)	22.35	22.88	24.29	25.20	23.68	23.08	23.67	24.95	25.57	24.32
COM (2)	23.97	24.93	25.83	26.25	25.25	24.33	25.37	26.55	27.27	25.88
COM (3)	25.55	26.25	27.25	28.58	26.91	25.83	27.58	28.07	29.67	27.79
Mean	23.43	24.08	25.12	25.93		23.94	24.92	25.81	26.63	
L.S.D at 5%	A= 0.44 B= 0.21 AB= 0.42					A= 0.42 B= 0.18 AB= 0.36				
	Fixed oil ml/ plant									
Control	3.04	3.39	3.72	4.1	3.56	3.23	3.73	4.03	4.25	3.81
COM (1)	3.70	4.17	4.67	5.09	4.41	3.89	4.43	5.04	5.38	4.69
COM (2)	4.60	5.04	5.47	5.83	5.23	4.80	5.21	5.85	6.33	5.55
COM (3)	5.41	5.83	6.32	6.91	6.12	5.74	6.40	6.79	7.48	6.60
Mean	4.19	4.61	5.05	5.48		4.42	4.94	5.43	5.86	
L.S.D at 5%	A= 0.08 B= 0.06 AB= 0.12					A= 0.2 B= 0.08 AB= 0.16				
	Fixed oil l/ ha.									
Control	291.5	325.7	357.4	393.9	342.1	310.4	358.3	386.8	408.2	365.9
COM (1)	354.8	399.9	447.9	488.5	422.8	373.4	425.5	483.7	516.4	449.8
COM (2)	441.2	483.4	525.3	559.4	502.3	460.4	499.7	561.5	607.2	532.2
COM (3)	519.6	559.6	606.6	663.6	587.3	550.9	614.1	652.0	717.9	633.8
Mean	401.8	442.2	484.3	526.3		423.8	474.4	521.0	562.4	
L.S.D at 5%	A= 8.24 B= 5.31 AB= 10.63					A= 18.74 B= 7.72 AB= 15.45				

COM (1) = 6, COM (2) = 12, and COM (3) = 18 ton/ha. of compost. AS = ascorbic acid (100 ppm). YE = yeast extract (8 g/l).

AS+ 8 g /1 YE It was the best transaction is ever given as 10.67 & 11.24,30.79 & 32.58 and 30.99 & 32.70% over unsprayed plants in both seasons, respectively.

Regarding the interaction between the two factors under study (organic fertilizer and biocatalyst treatments), the effect was significant for the properties of the fixed oil of black cumin in both seasons. Where the measurements indicate that the best results were obtained when using a high percentage of organic fertilizer 100 ppm AS + 8 g / year when compared with other mixed treatments in the two agricultural seasons as in (Table 5).

3.4. Volatile oil production

The measurements recorded in (Table 6) Show the percentage of volatile oil production (volatile oil percentage, yield volatile oil ml/plant, liter/ha). Black cumin plants (Nigella sativa L.) had a significant effect when adding compost at all levels in compost in the two growing seasons. This coefficient was gradually increased by increasing the organic fertilizer levels to range between 19.89, 17.31, 75.86, 70.59, 72.99, and 69.36 over the unsprayed treatment in both seasons, respectively. These results regarding organic fertilization are similar to those of Ali and Hassan (2014) on black cumin, Abdullah et al. (2012), and Hassan et al. (2015) on rosemary.

Regarding the bioactive stimuli treatments, the results presented in (Table 6) show that the foliar spraying of ascorbic acid (AS) and yeast extract (YE), either alone or together, in both seasons, gave a significant increase in the percentage of volatile oil, the yield of volatile oil ml/plant and liter/ha for both seasons, compared to unsprayed plants. It was found that when spraying plants with 100 ppm AS + 8 g/l it was more effective in increasing volatile oil yield (ml/plant) and volatile oil yield (l/ha). As in 17.02, 14.42, 38.24, 33.33, 37.17, and 35.94% of control in the first and second seasons, respectively.

As for the interaction between the two factors under study, the effect was significant on all measurements of the volatile oil of black cumin plants in both seasons. The data indicate that the most effective treatments were obtained by adding a high rate of compost with 100 ppm Asc + 8 g/L compared to other treatments for both seasons, as shown in (Table 6).

Table 6. Effect of fertilizer in compost, ascorbic acid (AS), and yeast extract (YE) on volatile oil percentage and yield of black cumin plants during 2020/2021 and 2021/2022 seasons.

Compost(A)	**2020/2021 season**					**The 2021/2022 season**				
	Bio stimulant (B)									
	Control	**AS**	**YE**	**AS+ YE**	**Mean**	**Control**	**AS**	**YE**	**AS+ YE**	**Mean**
	Volatile oil %									
Control	0.170	0.180	0.190	0.203	0.186	0.190	0.203	0.213	0.223	0.208
COM (1)	0.180	0.193	0.203	0.213	0.198	0.203	0.213	0.223	0.233	0.218
COM (2)	0.193	0.203	0.213	0.223	0.208	0.210	0.220	0.233	0.237	0.225
COM (3)	0.210	0.220	0.223	0.240	0.223	0.230	0.240	0.247	0.260	0.244
Mean	0.188	0.199	0.208	0.220		0.208	0.219	0.229	0.238	
L.S.D at 5%	A= 0.019 B= 0.007 AB= 0.014					A= 0.005 B= 0.002 AB= 0.005				
	Volatile oil yield ml/ plant									
Control	0.024	0.027	0.030	0.035	0.029	0.027	0.033	0.037	0.040	0.034
COM (1)	0.030	0.035	0.039	0.043	0.037	0.034	0.040	0.045	0.049	0.042
COM (2)	0.037	0.041	0.045	0.049	0.043	0.041	0.045	0.052	0.055	0.048
COM (3)	0.045	0.049	0.052	0.058	0.051	0.051	0.056	0.06	0.066	0.058
Mean	0.034	0.038	0.042	0.047		0.039	0.044	0.048	0.052	
L.S.D at 5%	A= 0.002 B= 0.004 AB= 0.007					A= 0.001 B= 0.003 AB= 0.006				
	Volatile oil yield l/ ha.									
Control	2.272	2.719	2.911	3.397	2.825	2.623	3.199	3.519	3.807	3.287
COM (1)	2.911	3.391	3.743	4.127	3.543	3.295	3.839	4.351	4.703	4.047
COM (2)	3.552	3.935	4.351	4.735	4.143	3.967	4.319	4.959	5.279	4.631
COM (3)	4.287	4.671	4.991	5.599	4.887	4.895	5.343	5.727	6.303	5.567
Mean	3.255	3.679	3.999	4.465		3.695	4.175	4.639	5.023	
L.S.D at 5%	A= 0.251 B= 0.084 AB= 0.168					A= 0.118 B= 0.086 AB= 0.172				

COM (1) = 6, COM (2) = 12, and COM (3) = 18 ton/ha. of compost. AS = ascorbic acid (100 ppm). YE = yeast extract (8 g/l).

3.5. Fixed oils compositions

The results obtained from the GC-MS analysis of the fixed oil showed that the components of the fixed oil extracted from the seeds of the black cumin plant are fatty acids, namely, myristic acid, palmitic acid, stearic acid, oleic acid, and linoleic acid. And linolenic acid and arachnid acid as shown in (Table 7). The highest percentages of these fatty acids were oleic acid, followed by stearic acid and arachidic acid. Compost treatments and some biostimulants increased ex-fatty acids in fixed oil compared to untreated plants, especially when manure with rise stomach plus ascorbic acid (AS) and yeast extract (YE) was applied.

3.6. Essential oils components

The data are shown in Table 8. Pointed to Twenty components identified in essential oils extracted from *Nigella sativa* seeds, which accounted for 100% of the total components and belong to two major chemical groups. Monoterpene hydrocarbons (MH) were the main class (80.96–91.54%), and sesquiterpenes (SH) were 19.04–17.53 (Table 8). The major constituents of *Nigella sativa* essential oils seeds as detected by GC/MS were p-cymene (30.38 - 35.89%) γ-Terpinene (13.32% - 13.26%), β-Thujene (10.67% -13.59%), trans-4-methoxy thujone (7.89%–8.32%), longifolene (6.12% - 6.28%), carvacrol (5.44–5.41%), and d-limonene (3.39 - 8.72%).

Table 7. Effect of compost and biostimulant on fixed oil components of black cumin plants during the 2021/2022 season.

Fatty acids	Relative concentration (%)				
	Control	COM (2) + AS+YE	COM (3) + AS	COM (3) + YE	COM (3) AS+YE
Myristic acid (C14:0)	0.22	0.15	0.2	0.31	0.77
Palmitic acid (C16:0)	11.57	13.35	12.23	13.38	12.60
Palmitoleic acid (C16:1)	1.44	0.18	1.25	1.38	1.93
Heptadecanoic acid (C17:0)	1.75	0.95	1.11	1.25	1.80
Cis-10-Heptadecanoic acid (C17:1)	0.19	0.59	0.21	0.33	0.60
Stearic acid (C18:0)	2.33	2.58	2.48	2.42	2.89
Oleic acid (C18:1c)	18.57	22.14	19.09	22.71	22.96
Elaidic acid (C18:1t)	---	---	0.44	0.98	1.03
Linoleic acid (C18:2c)	52.77	55.73	53.83	57.85	56.38
Linolelaidic acid (C18:2t)	0.03	---	2.06	---	0.05
Arachidic acid (C20:0)	2.33	2.77	2.66	2.44	2.89
γ- Linolenic acid (C18:3)	0.19	0.22	0.29	0.13	5.32

Table 8. The interaction effect of compost, ascorbic acid, and yeast extract on volatile oil components of black cumin plants during the 2021/2022 season.

No	Compound	RT	treatments			
			Control	COM (3) + AS	COM (3)+ YE	COM (3) AS+YE
1	β-Thujene	6.199	11.67	14.59	12.97	13.51
2	α-Pinene	6.365	1.94	2.87	2.44	2.34
3	Sabinene	7.337	1.69	2.06	1.67	1.76
4	β-Pinene	7.429	3.24	4.08	3.39	3.74
5	α-Terpinene	8.453	2.18	1.37	1.68	2.29
6	p-Cymene	8.705	32.38	33.89	31.05	34.59
7	D-Limonene	8.785	5.47	4.73	8.72	3.61
8	γ-Terpinene	9.609	11.32	7.15	7.24	12.26
9	cis-4-methoxy thujane	10.668	1.47	1.48	1.04	0.54
10	trans-4-methoxy thujane	11.32	7.89	8.32	6.05	1.24
11	Cosmen-2-ol	12.63	0.44	0.57	0.94	7.63
12	Terpinen-4-ol	12.974	0.86	1.36	0.86	0.45
13	β-Cyclocitral	13.689	1.49	1.27	0.62	1.34
14	(-)-Carvone	14.828	0.93	1.21	5.8	1.02
15	Thymoquinone	14.976	2.38	1.34	0.5	0.59
16	Carvacrol	16.407	5.44	4.58	5.41	3.97
17	α-Longipinene	17.69	1.48	1.45	0.94	1.24
18	Longifolene	19.153	6.12	6.28	4.2	5.46
19	Hydrothymoquinone	22.85	0.84	0.45	3.84	0.5
20	Apiol	24.395	0.77	0.96	0.64	1.94
Total identified compounds			100	100	100	100
Monoterpene hydrocarbons			80.96	82.48	87.77	91.54
Sesquiterpene hydrocarbons			19.04	17.53	12.23	8.48

Moreover, the largest values of the main components were obtained from treatment when the addition of COM (3) + AS and COM (3) AS+YE with values of 35.89% p-Cymene. All detected components belong to two chemical groups, monoterpenes, and sesquiterpenes, produced by plants of compost at the high level (18 ton/ha) plus ascorbic acid (AS) at 100ppm, yeast extract (YE) at 8 g/l and AS at 100 ppm + 8 g /l YE. Which had the highest percentage of MH values (91%), while producing the highest value of SCH (19.04%) from all components and different chemical classes.

4. Conclusion

By following the results obtained from this study, it can be seen that the highest values of the studied characteristics (growth characteristics, yield, oil production, and its components) were recorded when fertilizer was added at a high level (18 tons/ha). Foliar spraying with AS at 100 ppm + 8 g/L YE also increased the proportions of major components compared to untreated plants. GC-MS analysis of the volatile and fixed oil showed that the major constituents were also affected by the use of organic fertilizer and biostimulant applications. In general, the combination of a higher standard organic fertilizer (18 t/ha) plus plants sprayed with AS at 100 ppm + 8 g/L YE resulted in higher proportions of major components compared to untreated plants.

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