https://doi.org/10.17221/266/2025-PSE

The changes in growth, yield, and biologically active compounds of essential oil in *Trachyspermum ammi* L. upon rhizobacteria and seaweed applications

Aya H. El-Nagar[1], Kholoud Z. Ghanem[2], Fahmy A.S. Hassan[1], Mohammed I. Fetouh[1], Rasha S. El-Serafy[1], Mohamed M. Moussa[3]*

[1]Horticulture Department, Faculty of Agriculture, Tanta University, Tanta, Egypt
[2]Department of Biological Science, College of Science and Humanities, Shaqra University, Shaqra, Riyadh, Saudi Arabia
[3]Horticulture Department, Faculty of Agriculture, Menoufia University, Shebin El Kom, Egypt
**Corresponding author: rasha.elserafi@agr.tanta.edu.eg*

Citation: El-Nagar A.H., Ghanem K.Z., Hassan F.A.S., Fetouh M.I., El-Serafy R.S., Moussa M.M. (2025): The changes in growth, yield, and biologically active compounds of essential oil in *Trachyspermum ammi* L. upon rhizobacteria and seaweed applications. Plant Soil Environ., 71: 565–580.

Abstract: Using biostimulants to enhance plant growth and increase yield and secondary metabolites in medicinal and aromatic plants is an important strategy to achieve sustainable agriculture. The influence of two strains of nitrogen-fixing rhizobacteria (NFB) of *Azotobacter chroococcum* (NFB1) and *Azospirillum lipoferum* (NFB2), three levels of seaweed extract (SWE; 0 (SWE1), 250 (SWE1), and 500 mg/L (SWE2)) and their interactions have been investigated on *Trachyspermum ammi* L. (ajwain) growth, fruit yield, and essential oil constituents for two winter seasons. Growth traits (plant height, number of branches, and fresh and dry weights) and fruit traits (umbel number, 1 000-fruit weight, and fruit yield) were improved following NFB and/or SWE applications. Leaf pigments, total phenols, carbohydrates, free amino acids, and nutrient content were also enhanced. Ajwain plants that received NFB2 soil inoculation and foliarly sprayed with SWE1 observed the highest growth and yield values. Applying this treatment resulted in 27.6% and 32.7% higher fruit yield per plant for the first and second seasons, respectively, compared to the control. The results of GC-MS revealed that γ-terpinene, p-cymene, and thymol are the major components in ajwain essential oil. All applications used changed the percentages of the main components detected in ajwain essential oil. For instance, increasing SWE level caused a reduction in γ-terpinene with an increase in thymol content. The highest conservation rate from γ-terpinene to thymol was detected in NFB2 × SWE1-treated plants, with the highest thymol content and least γ-terpinene. *Azospirillum lipoferum* soil inoculation with SWE1 foliar application is recommended to enhance ajwain production, in terms of fruit yield and oil quality.

Keywords: aromatic herb; nutrition; metabolite; phytochemistry; biofertilisation; Apiaceae

Ajwain (*Trachyspermum ammi* L.) is an aromatic herb that belongs to the Apiaceae family and grows annually in many countries, including Egypt, India, Iran, and Iraq (Lawless 1992). Seeds of ajwain contain carbohydrates (38.6%), fat (18.1%), protein (15.4%), fiber (11.9%), glycosides, tannins, and moisture (8.9%), flavone, saponins, and minerals (7.1%) (Bairwa et al. 2012), in addition to the important vitamins of thiamine and riboflavin (Pruthi 2001). Also, they contain 2% to 4.4% essential oil (Bairwa et al. 2012). Thymol, p-cymene, γ-terpinene, α-pinene and β-pinene are the main ingredients in the ajwain essential oil (Zarshenas et al. 2014). Thymol is the major component in ajwain oil; in some cases, p-cymene and γ-terpinene can increase more than the thymol content (Omer et al. 2014). Ajwain seeds help in the process of healing

https://doi.org/10.17221/266/2025-PSE

and inhibit infections (Pruthi 2001, Bairwa et al. 2012). Ajwain oil has curative and liver protective properties of anti-aggregatory, antiviral, anthelmintic, anti-ulcer, antihypertensive, and anti-fats influences (Bairwa et al. 2012), antimicrobial (Sivropoulou et al. 1996), anti-fungicidal, and antioxidant properties (Singh and Singh 2016). It is also used in the food/flavour industry and therapeutic compositions (Choudhary et al. 2017).

Fertilisation is the essential factor affecting the growth and productivity of medicinal and aromatic plants, including *Trachyspermum ammi* L. Unfortunately, the excessive use of mineral fertilisers causes serious impacts such as deterioration of soil fertility, agro-ecosystem pollution and higher production costs (Kahil et al. 2017, Alshallash et al. 2022, Al-Saif et al. 2024). Importantly, chemical-free production of medicinal and aromatic plants is of great interest to guarantee their quality and safety. Therefore, several strategies such as non-traditional fertilisers and biostimulants have been developed to achieve those goals (Ali et al. 2022a,b, Moussa et al. 2024).

One type of bio-fertiliser is plant growth-stimulating bacteria such as *Azotobacter,* which is important in the biological fixation of nitrogen and can produce and secrete biologically active substances such as vitamin B, nicotinic acid, biotin, auxin, gibberellin, etc., in the rhizosphere area of a plant, which enhances root growth (Bahgat et al. 2023). Concerning the effect of free-living nitrogen-fixing bacteria on the performance of medicinal plants, Ghilavizadeh et al. (2013) examined the effect of biofertilisers and plant density on essential oil content and yield of *Trachyspermum ammi* L., and they found that the yield and essential oil content increased when *Azotobacter* and *Azospirillum* bacteria were applied. *Azotobacter* and *Azospirillum* soil supplementations increased growth, productivity and secondary metabolites in fennel and roselle plants (El-Serafy and El-Sheshtawy 2020, Bahgat et al. 2023).

Seaweed extracts as biostimulants offer a natural substitute for synthetic fertilisers in promoting plant growth and yield (Ali et al. 2021). They enhance fruit set, nutritional absorption, biotic and abiotic stress tolerance, and seedling growth and development (Mukherjee and Patel 2020). Important growth-promoting compounds are found in seaweed extracts, such as auxins, cytokinins, gibberellins, and several nutrients essential for plant development (Mughunth et al. 2024). Plant growth, yield, and essential oil content improved when coriander plants were foliar sprayed with seaweed extract (Tursun 2022). Ali et al. (2023) state that seaweed extract improved the fennel plants' oil components, fruit output, and plant development.

The effect of nitrogen-fixing bacteria (NFB) interacting with seaweed extract (SWE) on *Trachyspermum ammi* L. productivity has not been studied yet. This study hypothesises that NFB soil supplementation and SWE foliar application may enhance ajwain productivity and quality. Therefore, this experiment aimed to evaluate the role of NFB and SWE and their combination on plant growth, fruit yield, secondary metabolites, and active compound alterations in the essential oil of *Trachyspermum ammi* L. plant.

MATERIAL AND METHODS

Study description and experimental site

A field experiment was conducted at the Experimental Farm of Agriculture Faculty, Tanta University, Tanta, Egypt (30°47'18"N; 31°00'06"E), at 8 m a.s.l., during the winter seasons of 2022–2023 and 2023–2424.

Ajwain fruits were planted on 15th and 20th October for the 2022 and 2023 seasons, respectively, at a 30 × 60 cm spacing. After 21 days of cultivation, ajwain plants were thinned to two plants per hill. All weed and pest control farming practices were performed as the Egyptian Ministry of Agriculture recommended. Mineral fertilisation was done according to Sathyanarayana et al. (2017). Before cultivation, samples of the experimental soil were analysed, and its physical and chemical analyses were as follows: sand 21.5%, silt 38.9%, and clay 39.6%. The soil chemical features were electrical conductivity (EC), 1.84 dS/m; pH 8.32; Mg 11.5 mg/L; Ca 14.5 mg/L; HCO_3 14 mg/L; $CaCO_3$ 1.33 mg/L; total N 0.26%; P 0.041%; and K 0.06%.

Treatments and experimental design

The strains of aerobic nitrogen-fixing bacteria (NFB) of *Azotobacter chroococcum* (ATCC 9043) and *Azospirillum lipoferum* (ATCC 29707) were provided from the Soil, Water, and Environment Research Institute, Agricultural Research Centre, Giza, Egypt, and maintained in the refrigerator at 4 °C until soil supplementation.

The current investigation was planned in a split-plot design with nine treatments in two factors: (1) NFB

https://doi.org/10.17221/266/2025-PSE

strains (the NFB0; un-inoculated, NFB1; *Azotobacter chroococcum*, and NFB2; *Azospirillum lipoferum*), and (2) seaweed extract at different doses of 0 (SWE0), 250 mg/L (SWE1), and 500 mg/L (SWE2), each treatment repeated three times. NFB strains were randomly applied in the main plots, while foliar extracts were supplemented in the subplots, which were 9.0 m^2 (3.0 × 3.0 m) to include 100 plants each.

Bacteria suspension (10 mL) of each strain was individually applied to the experimental main plots as a soil drench 21 days post-germination at a 10^9 CFU/mL density. Ajwain plants in each subplot were foliarly sprayed with seaweed extract three times: 30, 60, and 90 days after cultivation. Untreated control plants received foliar supplementation three times with tap water at the same time as extract application for both seasons.

Harvesting

Ten ajwain plants were manually collected at the harvesting stage (when the fruits turn brown) during the second week of May for both seasons to determine the plant height (cm), branch number/plant, plant fresh weight (g), plant dry weight (g), number of umbels/plant, weight of 1 000 fruits, fruit yield/plant (g), and fruit yield/ha (kg).

Essential oil extraction and composition

Ajwan fruits (100 g) were hydro-distilled using a Clevenger-type apparatus for 3 h according to Viuda-Martos et al. (2011). Ajwan fruits from each treatment were distilled in triplicate, and oil contents are presented as the average value. The separated oil was dried using anhydrous sodium sulfate. Oil percentage (%) and oil yield (L/ha) were calculated. Oil percentage was calculated using the following formula:

Essential oil percentage = (oil volume in the graduated tube/sample dry weight) × 100.

The dried oil was stored at 4 °C until GC-MS analysis in dark vials.

The active ingredients of ajwan essential oil were identified by GC-MS analysis using a Perkin Elmer (model: Clarus 580/560 S, Tokyo, Japan) equipped with four capillary columns (30 m × 0.25 mm ID and film thickness 0.25 μm).

Physiological and biochemical determinations

Leaf pigments. Samples of ajwain leaves were collected at the flowering stage for photosynthetic pigments determination as described by Dere et al. (1998) using methanol (96%). Leaf chlorophyll and carotenoids were calculated and expressed in mg/g FW (fresh weight).

Total carbohydrates, free amino acids, and protein determination. Total carbohydrates in leaves at the harvest stage were determined following the method previously described by Alayafi et al. (2025) using anthrone reagent. Total free amino acids in fruits were estimated according to the procedure outlined by Yemm and Cocking (1955) using the ninhydrin reagent technique. Total protein (%) was determined using the micro-Kjeldahl method, with a nitrogen-to-protein conversion factor of 6.25 (Jones 1931).

Total polyphenols. The ground fruits (1 g) samples were stirred with 50 mL of 80% methanol and macerated for two days at room temperature. After thoroughly removing solvents, the extract was kept below 4 °C for total phenol determination according to McDonald et al. (2001). Total phenols content was determined spectrophotometrically at 765 nm and presented as mg gallic acid equivalent/g dry weight.

Nutrient estimation

For nutrient estimation, a 0.5 g sample of dried leaves collected at the harvest stage was digested using sulfuric and perchloric acids to determine the nutrient content Piper (1967) and Jackson (1978). Nitrogen (N) was determined using the micro-Kjeldahl method according to Black et al. (1965) and presented as a percentage (%). Phosphorus (P) was assessed colourimetrically as described in the Jackson (1978) method using stannous chloride, phosphomolybdate-sulfuric acid, and was calculated in percentages (%). Potassium (K) (%) was determined using atomic absorption according to Christain (1969).

Statistical analysis

The data obtained were analysed using a split-plot design with three replications. Data was analysed through two-way analysis of variance (ANOVA) using the COSTAT program (Tokyo, Japan). The Duncan's multiple range test evaluated the means at $P \leq 0.05$ and $P \leq 0.01$.

RESULTS

Plant growth

The effect of nitrogen-fixing bacteria soil application and seaweed extracts foliar spray, and their

https://doi.org/10.17221/266/2025-PSE

Table 1. Growth traits of ajwain plants in response to two strains of nitrogen-fixing bacteria, seaweed extracts, and their interaction during 2022/2023 and 2023/2024 seasons

		Plant height (cm)		Number of branches/plant		Plant fresh weight (g)		Plant dry weight (g)	
		1st season	2nd season	1st season	2nd season	1st season	2nd season	1st season	2nd season
Nitrogen-fixing bacteria (NFB)									
Un-inoculated (NFB0)		75 28 ± 1.38^{b}	76.46 ± 1.18^{c}	11.83 ± 0.32^{b}	13.08 ± 0.25^{b}	42.38 ± 0.41^{c}	48.73 ± 0.22^{b}	20.79 ± 0.20^{c}	24.55 ± 0.12^{c}
Azotobacter chroococcum (NFB1)		80.65 ± 0.56^{a}	82.71 ± 1.27^{b}	12.61 ± 0.26^{a}	14.05 ± 0.31^{a}	44.98 ± 0.76^{b}	49.82 ± 0.45^{a}	22.10 ± 0.42^{b}	25.15 ± 0.20^{b}
Azospirillum lipoferum (NFB2)		82.14 ± 0.77^{a}	85.56 ± 1.70^{a}	13.14 ± 0.24^{a}	14.47 ± 0.37^{a}	47.13 ± 1.09^{a}	50.76 ± 0.81^{a}	23.51 ± 0.61^{a}	26.09 ± 0.37^{a}
Seaweed extracts (SWE)									
Without (SWE0)		76.78 ± 1.70^{b}	76.42 ± 1.16^{c}	11.72 ± 0.24^{b}	13.03 ± 0.23^{c}	42.39 ± 0.46^{c}	48.63 ± 0.23^{b}	20.78 ± 0.21^{c}	24.54 ± 0.14^{b}
250 mg/L (SWE1)		81.31 ± 0.98^{a}	84.99 ± 1.64^{a}	13.22 ± 0.23^{a}	14.69 ± 0.37^{a}	46.89 ± 0.99^{a}	50.85 ± 0.48^{a}	23.32 ± 0.59^{a}	25.75 ± 0.33^{a}
500 mg/L (SWE2)		79.98 ± 0.92^{a}	83.31 ± 1.47^{b}	12.64 ± 0.28^{a}	13.89 ± 0.24^{b}	45.21 ± 0.93^{b}	49.83 ± 0.73^{a}	22.30 ± 0.50^{b}	25.51 ± 0.33^{a}
NFB × SWE									
	SWE0	70.17 ± 0.72^{e}	72.00 ± 0.58^{e}	10.92 ± 0.22^{e}	12.33 ± 0.33^{d}	41.06 ± 0.50^{d}	47.91 ± 0.32^{c}	20.29 ± 0.25^{e}	24.19 ± 0.19^{d}
Un-inoculated	SWE1	78.33 ± 1.30^{cd}	78.90 ± 0.90^{d}	12.50 ± 0.14^{bcd}	13.67 ± 0.22^{c}	43.62 ± 0.22^{cd}	49.23 ± 0.21^{bc}	21.39 ± 0.15^{de}	24.82 ± 0.09^{cd}
	SWE2	77.33 ± 0.88^{d}	78.47 ± 0.87^{d}	12.08 ± 0.68^{cd}	13.25 ± 0.38^{cd}	42.45 ± 0.32^{d}	49.05 ± 0.29^{bc}	20.69 ± 0.29^{e}	24.65 ± 0.19^{cd}
	SWE0	79.60 ± 0.86^{bcd}	78.40 ± 0.83^{d}	11.83 ± 0.22^{de}	13.33 ± 0.36^{cd}	42.50 ± 0.24^{d}	48.74 ± 0.64^{bc}	20.69 ± 0.26^{e}	24.64 ± 0.32^{cd}
A. chroococcum	SWE1	81.63 ± 1.04^{abc}	86.50 ± 1.04^{b}	13.25 ± 0.29^{ab}	14.83 ± 0.65^{ab}	47.08 ± 1.06^{b}	50.79 ± 0.25^{b}	23.27 ± 0.53^{bc}	25.63 ± 0.12^{b}
	SWE2	80.73 ± 0.96^{abcd}	83.23 ± 0.96^{c}	12.75 ± 0.43^{bcd}	14.00 ± 0.14^{bc}	45.36 ± 0.65^{bc}	49.93 ± 0.69^{bc}	22.35 ± 0.33^{cd}	25.19 ± 0.35^{bc}
	SWE0	80.57 ± 0.61^{abcd}	78.87 ± 0.70^{d}	12.42 ± 0.08^{bcd}	13.42 ± 0.22^{cd}	43.60 ± 0.76^{cd}	49.24 ± 0.16^{bc}	21.37 ± 0.34^{de}	24.80 ± 0.08^{cd}
Lipoferum	SWE1	83.97 ± 0.88^{a}	89.57 ± 0.30^{a}	13.92 ± 0.22^{a}	15.58 ± 0.44^{a}	49.97 ± 0.76^{a}	52.55 ± 0.99^{a}	25.31 ± 0.29^{a}	26.79 ± 0.53^{a}
	SWE2	81.87 ± 1.73^{ab}	88.23 ± 0.50^{ab}	13.08 ± 0.22^{abc}	14.42 ± 0.44^{bc}	47.83 ± 1.61^{ab}	50.50 ± 1.70^{b}	23.85 ± 0.52^{b}	26.69 ± 0.25^{a}
***P*-value**									
NFB		< 0.001***	< 0.001***	< 0.001***	< 0.001***	< 0.001***	0.0039**	< 0.001***	< 0.001***
SWE		< 0.001***	< 0.001***	< 0.001***	< 0.001***	< 0.001***	0.0018**	< 0.001***	< 0.001***
NFB × SWE		0.0302*	0.0486*	0.9520^{ns}	0.7573^{ns}	0.2411^{ns}	0.5279^{ns}	0.0191*	0.0470*

Data (means ± standard error); *$P \leq 0.05$; **$P \leq 0.01$; ***$P \leq 0.001$; ns – non-significant difference. Mean values sharing the same lower-case letter for NFB, seaweed, and their interactions in the same column are not significantly different at $P \leq 0.05$ from Duncan's test

https://doi.org/10.17221/266/2025-PSE

interaction on the growth traits of ajwain plants is presented in Table 1. The plant height and branch number traits showed the highest values following NFB2 compared to uninoculated plants. Treated plants with SW1 presented the highest plant height and branch number/plant. Concerning the interaction, the treatment of NFB2 × SWE1 showed the tallest plants (83.9 and 89.6 cm for both seasons, respectively) with the highest branch number (13.9 and 15.6 for both seasons, respectively) compared with the other treatments. Also, treated plants with NFB2 showed the maximum fresh and dry weights compared to NFB0 plants. Concerning seaweed treatments, SWE1 exhibited the highest fresh and dry weights, and the NFB2 × SWE1 treatment gave both seasons the highest fresh and dry weights.

Yield and yield-related traits

Likewise, plant growth characteristics and fruit yield were also affected by the application of NFB and SWE treatments as compared with control plants (Table 2). The weight of 1 000 fruits, the number of umbels per plant, and fruit yield were significantly improved with the application of NFB2, except that the number of umbels per plant showed a non-significant difference for the first season. On the other hand, SWE1 increased 1 000 fruit weight, the number of umbels per plant and fruit yield. The combined application of NFB2 × SWE1 markedly enhanced fruit characteristics and total fruit yield, which gave 27.6% and 32.7% higher fruit yield per plant than control plants for the first and second seasons, respectively.

Essential oil

Similarly, oil percentage and yield traits also improved following NFB and SWE applications (Table 3). All strains of NFB have enhanced total oil yield, either per plant or hectare. The most notable effect was recorded with NFB2 inoculation, which increased oil percentage by 20.37% and 23.23% for the first and second seasons, respectively. Foliar application of SWE positively influenced oil production. The application of SWE1 increased the oil percentage by 17.86, 23.35, 7.54, and 8.7% over control and SW2 for the first and second seasons, respectively. Maximum oil production was recorded with the combined application of NFB2 × SWE1.

Essential oil components

The major detected essential oil components were α-thujene, phellandrene, α-pinene, cis-sabinene, β-pinene, α-terpinene, p-cymene, γ-terpinene, limonene, terpinene-4-ol and thymol (Table 4). Results also showed that thymol, γ-terpinene, p-cymene, and ß-pinene were the main oil components. All used NFB strains recorded the highest thymol percentage compared with NFB0, and the most pronounced effect was recorded with NFB2. Foliar application of SWE increased thymol content. The application of SWE1 increased thymol content by 24.57%. The combined application of NFB2 and SWE1 recorded the highest thymol (52.30%) and the lowest γ-terpinene (28.81%).

Photosynthetic pigments

The enhanced results of plant growth characteristics and fruit yield were attributed to the increase in photosynthetic pigments content (Table 5), which was reflected in plant photosynthesis activity and metabolism. NFB2 inoculation and SWE1 foliar spray significantly improved total chlorophyll and carotene content, except for total chlorophyll in the second season. In this regard, the most pronounced effect was related to NFB2 × SWE1 treatment.

Total carbohydrates

Table 6 presents the effect of NFB soil application and SWE foliar spray and their interaction on the carbohydrate content of ajwain plants. Total carbohydrates significantly recorded the highest value affected by NFB2 compared to uninoculated plants. Treated plants with SWE1 significantly contained higher total carbohydrates relative to the control in both seasons. Concerning the interaction, the NFB2 × SWE1 treatment gave the highest carbohydrate level, which produced 28.85% and 26.53% higher carbohydrate content compared to the control in the first and second seasons, respectively.

Total phenols

The phenolic content in ajwain leaves was enhanced following NFB soil application compared to the uninoculated plants (Table 6), where NFB2 soil treatment recorded the highest values in this respect. About SWE foliar applications, the untreated plants exhibited the lowest phenolic content (20.87

https://doi.org/10.17221/266/2025-PSE

Table 2. Yield traits of ajwain plants in response to two strains of nitrogen-fixing bacteria, seaweed extracts , and their interaction during the 2022/2023 and 2023/2024 seasons

		Number of umbels/plant		Weight of 1 000 fruits (g)		Fruit yield/plant (g)	
		1st season	2nd season	1st season	2nd season	1st season	2nd season
Nitrogen-fixing bacteria (NFB)							
Un-inoculated (NFB0)		55.33 ± 1.17^{b}	57.67 ± 0.67^{c}	0.782 ± 0.013^{c}	0.815 ± 0.013^{c}	5.10 ± 0.10^{c}	5.16 ± 0.11^{c}
Azotobacter chroococcum (NFB1)		59.89 ± 0.59^{a}	62.00 ± 1.03^{b}	0.880 ± 0.030^{b}	0.899 ± 0.030^{b}	5.57 ± 0.16^{b}	5.72 ± 0.18^{b}
Azospirillum lipoferum (NFB2)		60.45 ± 0.60^{a}	64.78 ± 1.43^{a}	0.936 ± 0.043^{a}	0.976 ± 0.045^{a}	5.83 ± 0.21^{a}	6.04 ± 0.22^{a}
Seaweed extracts (SWE)							
Without (SWE0)		56.11 ± 1.33^{b}	57.89 ± 0.68^{c}	0.772 ± 0.012^{b}	0.801 ± 0.010^{c}	5.03 ± 0.09^{c}	5.11 ± 0.10^{c}
250 mg/L (SWE1)		60.33 ± 0.71^{a}	64.11 ± 1.40^{a}	0.925 ± 0.036^{a}	0.970 ± 0.034^{a}	5.87 ± 0.17^{a}	6.06 ± 0.20^{a}
500 mg/L (SWE2)		59.22 ± 0.76^{a}	62.44 ± 1.32^{b}	0.900 ± 0.036^{a}	0.919 ± 0.038^{b}	5.60 ± 0.17^{b}	5.75 ± 0.18^{b}
NFB × SWE							
	SWE0	51.00 ± 0.58^{f}	55.67 ± 0.88^{e}	0.756 ± 0.012^{c}	0.779 ± 0.014^{e}	4.96 ± 0.13^{d}	5.02 ± 0.15^{c}
Un-inoculated	SWE1	58.00 ± 0.58^{de}	59.00 ± 1.00^{d}	0.804 ± 0.017^{c}	0.858 ± 0.007^{de}	5.32 ± 0.17^{cd}	5.39 ± 0.21^{c}
	SWE2	57.00 ± 1.15^{e}	58.33 ± 0.67^{d}	0.785 ± 0.031^{c}	0.809 ± 0.005^{e}	5.03 ± 0.20^{d}	5.08 ± 0.17^{c}
	SWE0	58.33 ± 0.88^{cde}	58.67 ± 0.67^{d}	0.774 ± 0.020^{c}	0.799 ± 0.022^{e}	5.01 ± 0.15^{d}	5.07 ± 0.19^{c}
A. chroococcum	SWE1	61.33 ± 0.88^{ab}	65.33 ± 0.88^{b}	0.946 ± 0.037^{ab}	0.973 ± 0.005^{bc}	5.96 ± 0.16^{ab}	6.13 ± 0.13^{ab}
	SWE2	60.00 ± 0.58^{abcd}	62.00 ± 0.58^{c}	0.919 ± 0.026^{b}	0.926 ± 0.047^{cd}	5.73 ± 0.15^{bc}	5.96 ± 0.15^{b}
	SWE0	59.00 ± 0.58^{bcde}	59.33 ± 0.67^{d}	0.787 ± 0.032^{c}	0.824 ± 0.008^{e}	5.11 ± 0.24^{d}	5.24 ± 0.22^{c}
A. lipoferum	SWE1	61.67 ± 0.88^{a}	68.00 ± 0.58^{a}	1.024 ± 0.038^{a}	1.080 ± 0.042^{a}	6.33 ± 0.16^{a}	6.66 ± 0.12^{a}
	SWE2	60.67 ± 1.20^{abc}	67.00 ± 1.5^{ab}	0.997 ± 0.049^{ab}	1.023 ± 0.064^{ab}	6.05 ± 0.11^{ab}	6.21 ± 0.09^{ab}
***P*-value**							
NFB		< 0.001***	< 0.001***	< 0.001***	< 0.001***	< 0.001***	< 0.001***
SWE		< 0.001***	< 0.001***	< 0.001***	< 0.001***	< 0.001***	< 0.001***
NFB × SWE		0.0402*	0.0121*	0.0457*	0.0422*	0.0494*	0.0464*

Data (means ± standard error); *$P \leq 0.05$; **$P \leq 0.01$; ***$P \leq 0.001$; ns – non-significant difference. Mean values sharing the same lower-case letter for NFB, seaweed, and their interactions in the same column are not significantly different at $P \leq 0.05$ from Duncan's test

https://doi.org/10.17221/266/2025-PSE

Table 3. Fruit and essential oil yield of ajwain plants in response to two strains of nitrogen-fixing bacteria, seaweed extracts, and their interaction during 2022/2023 and 2023/2024 seasons

		Fruit yield/ha (kg)		Essential oil (%)		Essential oil yield/ha (L)	
		1st season	2nd season	1st season	2nd season	1st season	2nd season
Nitrogen-fixing bacteria (NFB)							
Un-inoculated (NFB0)		566.85 ± 11.16^{c}	573.59 ± 11.75^{c}	3.73 ± 0.08^{c}	3.83 ± 0.15^{c}	21.17 ± 0.67^{c}	21.96 ± 0.95^{c}
Azotobacter chroococcum (NFB1)		618.53 ± 17.97^{b}	635.30 ± 20.23^{b}	4.06 ± 0.13^{b}	4.28 ± 0.16^{b}	25.25 ± 1.43^{b}	27.44 ± 1.75^{b}
Azospirillum lipoferum (NFB2)		647.70 ± 22.85^{a}	670.71 ± 24.70^{a}	4.49 ± 0.18^{a}	4.72 ± 0.24^{a}	29.31 ± 2.08^{a}	32.11 ± 2.63^{a}
Seaweed extracts (SWE)							
Without (SWE0)		558.11 ± 10.12^{c}	567.84 ± 11.29^{c}	3.75 ± 0.07^{c}	3.81 ± 0.10^{c}	20.95 ± 0.61^{c}	21.68 ± 0.90^{c}
250 mg/L (SWE1)		652.34 ± 18.85^{a}	673.04 ± 22.24^{a}	4.42 ± 0.21^{a}	4.70 ± 0.25^{a}	29.11 ± 2.16^{b}	31.97 ± 2.53^{a}
500 mg/L (SWE2)		622.63 ± 18.73^{b}	638.71 ± 20.49^{b}	4.11 ± 0.12^{b}	4.32 ± 0.17^{b}	25.67 ± 1.32^{a}	27.86 ± 1.85^{b}
NFB × SWE							
	SWE0	550.77 ± 14.55^{d}	557.59 ± 17.06^{c}	3.64 ± 0.15^{d}	3.69 ± 0.24^{d}	20.07 ± 1.32^{c}	20.56 ± 1.35^{c}
Un-inoculated	SWE1	590.59 ± 19.28^{cd}	598.65 ± 22.91^{c}	3.81 ± 0.12^{cd}	3.99 ± 0.36^{cd}	22.53 ± 1.32^{c}	23.75 ± 1.55^{c}
	SWE2	559.18 ± 21.68^{d}	564.52 ± 18.91^{c}	3.75 ± 0.18^{cd}	3.81 ± 0.24^{d}	20.91 ± 0.65^{c}	21.57 ± 1.98^{c}
	SWE0	556.15 ± 15.14^{d}	563.26 ± 21.14^{c}	3.69 ± 0.12^{cd}	3.81 ± 0.12^{d}	20.50 ± 0.51^{c}	21.50 ± 1.41^{c}
A. chroococcum	SWE1	662.59 ± 17.36^{ab}	680.78 ± 14.75^{ab}	4.33 ± 0.24^{b}	4.64 ± 0.20^{b}	28.73 ± 0.23^{b}	31.67 ± 2.07^{b}
	SWE2	636.85 ± 16.14^{bc}	661.85 ± 16.20^{b}	4.17 ± 0.12^{bc}	4.40 ± 0.24^{bc}	26.53 ± 1.01^{b}	29.14 ± 1.62^{b}
	SWE0	567.41 ± 26.74^{d}	582.68 ± 24.84^{c}	3.93 ± 0.00^{bcd}	3.93 ± 0.21^{cd}	22.29 ± 1.05^{c}	22.98 ± 2.08^{c}
Lipoferum	SWE1	703.85 ± 17.50^{a}	739.68 ± 13.26^{a}	5.12 ± 0.12^{a}	5.48 ± 0.12^{a}	36.07 ± 1.74^{a}	40.48 ± 0.25^{a}
	SWE2	671.85 ± 12.39^{ab}	689.77 ± 9.74^{ab}	4.41 ± 0.12^{b}	4.76 ± 0.12^{b}	29.57 ± 0.35^{b}	32.86 ± 1.22^{b}
***P*-value**							
NFB		< 0.001***	< 0.001***	< 0.001***	< 0.001***	< 0.001***	< 0.001***
SWE		< 0.001***	< 0.001***	< 0.001***	< 0.001***	< 0.001***	< 0.001***
NFB × SWE		0.0494*	0.0464*	0.0445*	0.0378*	0.0059**	0.0019**

Data (means ± standard error); *$P \leq 0.05$; **$P \leq 0.01$; ***$P \leq 0.001$; ns – non-significant difference. Mean values sharing the same lower-case letter for NFB, seaweed, and their interactions in the same column are not significantly different at $P \leq 0.05$ from Duncan's test

https://doi.org/10.17221/266/2025-PSE

Table 4. Essential oil constituents of ajwain plant as influenced by nitrogen fixing bacteria (NFB), seaweed extracts (SWE) and their combinations

Component (%)		RT	T1	T2	T3	T4	T5	T6	T7	T8	T9
1	α-thujene	6.48	0.449	–	0.282	0.366	0.308	0.263	0.328	0.193	0.204
2	α-pinene	6.72	0.483	0.393	0.393	0.470	0.323	0.204	0.520	0.187	0.254
3	cis-sabinene	6.78	0.449	0.409	0.390	0.366	0.215	0.263	0.255	0.193	0.204
4	β-pinene	8.21	8.709	7.235	7.658	9.435	5.088	4.421	3.088	3.987	4.548
5	D-limonene	8.82	0.256	0.253	0.235	0.190	0.222	–	0.275	0.125	0.143
6	α-terpinene	9.44	0.652	0.568	0.498	0.479	0.445	0.434	0.443	0.223	0.334
8	p-cymene	9.89	22.355	24.676	22.531	20.692	20.743	21.411	21.001	12.379	11.473
7	α-phellandrene	9.97	0.357	0.364	0.282	0.352	0.337	0.590	0.365	0.209	0.242
9	γ-terpinene	11.08	48.27	38.79	41.95	49.25	37.85	43.26	39.85	28.81	32.04
10	trans-sabinenehydrate	11.46	0.495	0.815	0.906	0.137	0.100	0.816	0.482	–	–
11	Terpinen-4-ol	13.91	0.735	1.118	1.083	0.856	0.314	0.305	0.308	–	–
12	2-Caren-4-ol	14.06	0.442	0.125	0.152	0.363	0.215	0.129	0.193	0.129	–
13	Thymol	17.40	15.79	24.57	23.63	15.89	26.41	25.45	24.21	52.31	46.49
Total identified			99.44	99.324	99.99	98.85	92.57	97.54	91.32	98.74	95.93

RT – retention time; T1 – NFB0 × SW0; T2 – NFB0 × SW1; T3 – NFB0 × SW2; T4 – NFB1 × SW0, T5 – NFB1 × SWE1; T6 – NFB1 × SW2; T7 – NFB2 × SW0; T8 – NFB2 × SW1; T9 – NFB2 × SW2; NFB0 – un-inoculated; NFB1 – *Azotobacter chroococcum*; NFB2 – *Azospirillum lipoferum*; SW0 – untreated with SWE; SWE1 – 250 mg/L of SWE; SWE2 – 500 mg/L of SWE

and 22.79 mg GAE/g DW for the first and second seasons, respectively), whereas SWE1 treatment significantly recorded the highest phenolics (23.8 and 25.5 mg GAE/g DW for the first and second seasons, respectively) level. The highest phenol values of 25.7 and 27.6 mg GAE/g DW were given by the NFB2 × SWE1 treated plants for the first and second seasons, respectively.

Total free amino acids

The total free amino acid content in ajwain leaves was significantly affected following NFB soil application compared to the uninoculated plants (Table 6), as NFB2 treatment significantly recorded the highest values in this respect. About SWE foliar applications, the untreated plants exhibited the lowest amino acids content (0.173 and 0.179 g 100/g for the first and second seasons, respectively), whereas SWE1 treatment significantly recorded the highest amino acids level (0.281 and 0.287 g/100 g for the first and second seasons, respectively). The SWE2 application ranked second in this respect. The highest amino acid (0.345 and 0.352 g/100 g for the first and second seasons, respectively) content was obtained by the NFB2 × SWE1 application.

Protein content

Ajwain plants treated with NFB2 showed an enhancement in their protein values as compared with NFB0 and NFB1 soil treatments (Table 6). Among SWE foliar spray treatments, SWE2 was the most effective concentration in enhancing protein content in ajwain plants, while SWE1 ranked the second in this respect. The highest protein content (11.75% and 12.83% for the first and second seasons, respectively) was given by the NFB2 × SWE2 application.

Nutrient content

The results depicted in Table 7 indicate the content of nutrients accumulated in ajwain leaves due to NFB and/or SWE treatments. The content of N, P, and K was enhanced following NFB applications as compared with un-inoculated treatments, and NFB2 presented the highest values in this respect. SW1 and SW2 foliar applications caused an elevation in nutrient content, reaching its greatest values following SW1 application, except for N% that was mostly improved with SW2. In terms of the interaction, the treatment of NFB2 × SW1 showed the maximum nutrient values in ajwain leaves, except

https://doi.org/10.17221/266/2025-PSE

Table 5. Photosynthetic pigments of ajwain leaf in response to two strains of nitrogen fixing bacteria, seaweed extracts, and their interaction during 2022/2023 and 2023/2024 seasons

		Chlorophyll *a*		Chlorophyll *b*		Total chlorophyll		Carotenoids	
		(mg/g FW)							
		1st season	2nd season	1st season	2nd season	1st season	2nd season	1st season	2nd season
Nitrogen fixing bacteria (NFB)									
Un-inoculated (NFB0)		0.514 ± 0.018^{c}	0.557 ± 0.013^{c}	0.251 ± 0.006^{b}	0.338 ± 0.019^{c}	0.765 ± 0.018^{c}	0.894 ± 0.023^{c}	0.230 ± 0.002^{c}	0.247 ± 0.003^{c}
Azotobacter chroococcum (NFB1)		0.561 ± 0.027^{b}	0.604 ± 0.017^{b}	0.332 ± 0.025^{a}	0.482 ± 0.031^{b}	0.893 ± 0.048^{b}	1.086 ± 0.045^{b}	0.246 ± 0.005^{b}	0.264 ± 0.006^{b}
Azospirillum lipoferum (NFB2)		0.637 ± 0.032^{a}	0.668 ± 0.028^{a}	0.351 ± 0.029^{a}	0.529 ± 0.038^{a}	0.988 ± 0.055^{a}	1.193 ± 0.065^{a}	0.256 ± 0.006^{a}	0.279 ± 0.007^{a}
Seaweed extracts (SWE)									
Without (SWE0)		0.508 ± 0.017^{b}	0.555 ± 0.010^{b}	0.251 ± 0.005^{b}	0.342 ± 0.020^{b}	0.759 ± 0.014^{c}	0.897 ± 0.026^{b}	0.230 ± 0.003^{c}	0.246 ± 0.003^{c}
250 mg/L (SWE1)		0.623 ± 0.031^{a}	0.650 ± 0.029^{a}	0.349 ± 0.024^{a}	0.513 ± 0.038^{a}	0.972 ± 0.052^{a}	1.164 ± 0.062^{a}	0.255 ± 0.006^{a}	0.280 ± 0.007^{a}
500 mg/L (SWE2)		0.581 ± 0.031^{a}	0.624 ± 0.005^{a}	0.334 ± 0.029^{a}	0.490 ± 0.033^{a}	0.915 ± 0.054^{b}	1.113 ± 0.053^{a}	0.247 ± 0.005^{b}	0.264 ± 0.006^{b}
NFB × SWE									
	SWE0	0.494 ± 0.039^{e}	0.545 ± 0.028^{d}	0.248 ± 0.012^{b}	0.275 ± 0.011^{d}	0.742 ± 0.029^{d}	0.820 ± 0.039^{e}	0.225 ± 0.003^{d}	0.238 ± 0.003^{d}
Un-inoculated	SWE1	0.539 ± 0.011^{cde}	0.567 ± 0.027^{d}	0.255 ± 0.009^{b}	0.371 ± 0.029^{c}	0.794 ± 0.019^{d}	0.938 ± 0.003^{d}	0.235 ± 0.003^{cd}	0.254 ± 0.004^{cd}
	SWE2	0.508 ± 0.043^{de}	0.558 ± 0.017^{d}	0.251 ± 0.011^{b}	0.367 ± 0.017^{c}	0.759 ± 0.042^{d}	0.925 ± 0.027^{d}	0.230 ± 0.003^{d}	0.248 ± 0.003^{cd}
	SWE0	0.504 ± 0.041^{de}	0.556 ± 0.004^{d}	0.252 ± 0.011^{b}	0.369 ± 0.028^{c}	0.756 ± 0.033^{d}	0.925 ± 0.031^{d}	0.231 ± 0.006^{d}	0.248 ± 0.003^{cd}
A. chroococcum	SWE1	0.597 ± 0.024^{bc}	0.643 ± 0.030^{bc}	0.388 ± 0.020^{a}	0.555 ± 0.014^{ab}	0.985 ± 0.028^{bc}	1.198 ± 0.043^{bc}	0.259 ± 0.001^{ab}	0.284 ± 0.007^{b}
	SWE2	0.583 ± 0.061^{bcd}	0.613 ± 0.029^{cd}	0.355 ± 0.044^{a}	0.522 ± 0.023^{b}	0.938 ± 0.104^{c}	1.135 ± 0.031^{c}	0.248 ± 0.004^{bc}	0.261 ± 0.006^{c}
	SWE0	0.525 ± 0.008^{cde}	0.563 ± 0.015^{d}	0.253 ± 0.010^{b}	0.382 ± 0.026^{c}	0.778 ± 0.004^{d}	0.945 ± 0.038^{d}	0.233 ± 0.007^{d}	0.253 ± 0.004^{cd}
Lipoferum	SWE1	0.734 ± 0.027^{a}	0.740 ± 0.027^{a}	0.404 ± 0.005^{a}	0.615 ± 0.006^{a}	1.138 ± 0.031^{a}	1.355 ± 0.020^{a}	0.272 ± 0.004^{a}	0.301 ± 0.005^{a}
	SWE2	0.651 ± 0.024^{b}	0.700 ± 0.002^{ab}	0.397 ± 0.051^{a}	0.589 ± 0.021^{a}	1.048 ± 0.027^{ab}	1.279 ± 0.025^{ab}	0.263 ± 0.003^{a}	0.284 ± 0.001^{b}
***P*-value**									
NFB		< 0.001***	< 0.001***	< 0.001***	< 0.001***	< 0.001***	< 0.001***	< 0.001***	< 0.001***
SWE		< 0.001***	< 0.001***	< 0.001***	< 0.001***	< 0.001***	< 0.001***	< 0.001***	< 0.001***
NFB × SWE		0.0426*	0.0359*	0.0494*	0.0479*	0.0042**	0.0031**	0.0446*	0.0255*

Data (means ± standard error); *$P \leq 0.05$; **$P \leq 0.01$; ***$P \leq 0.001$; ns – non-significant difference. Mean values sharing the same lower-case letter for NFB, seaweed, and their interactions in the same column are not significantly different at $P \leq 0.05$ from Duncan's test; FW – fresh weight

https://doi.org/10.17221/266/2025-PSE

Table 6. Total carbohydrates, total phenols, amino acids, and protein content of ajwain plants in response to two strains of nitrogen fixing bacteria, seaweed extracts, and their interaction during 2022/2023 and 2023/2024 seasons

		Total carbohydrates (%)		Total phenols (mg GAE/g DW)		Amino acids (g/100 g)		Protein content (%)	
		1st season	2nd season	1st season	2nd season	1st season	2nd season	1st season	2nd season
Nitrogen fixing bacteria (NFB)									
Un-inoculated (NFB0)		12.79 ± 0.18^{c}	13.64 ± 0.20^{c}	20.89 ± 0.28^{b}	22.73 ± 0.28^{c}	0.171 ± 0.007^{c}	0.178 ± 0.007^{c}	8.02 ± 0.23^{b}	8.36 ± 0.26^{c}
Azotobacter chroococcum (NFB1)		14.23 ± 0.37^{b}	15.38 ± 0.39^{b}	22.79 ± 0.60^{a}	24.14 ± 0.51^{b}	0.232 ± 0.020^{b}	0.234 ± 0.021^{b}	8.46 ± 0.23^{b}	9.13 ± 0.30^{b}
Azospirillum lipoferum (NFB2)		14.84 ± 0.34^{a}	15.80 ± 0.36^{a}	23.45 ± 0.69^{a}	25.55 ± 0.65^{a}	0.284 ± 0.021^{a}	0.292 ± 0.021^{a}	10.58 ± 0.45^{a}	11.17 ± 0.58^{a}
Seaweed extracts (SWE)									
Without (SWE0)		12.95 ± 0.25^{c}	13.89 ± 0.24^{c}	20.87 ± 0.32^{c}	22.79 ± 0.25^{c}	0.173 ± 0.008^{c}	0.179 ± 0.009^{c}	8.15 ± 0.27^{b}	8.33 ± 0.25^{b}
250 mg/L (SWE1)		14.72 ± 0.43^{a}	15.77 ± 0.42^{a}	23.86 ± 0.71^{a}	25.52 ± 0.70^{a}	0.281 ± 0.023^{a}	0.287 ± 0.023^{a}	9.44 ± 0.46^{a}	10.02 ± 0.49^{a}
500 mg/L (SWE2)		14.19 ± 0.33^{b}	15.17 ± 0.43^{b}	22.40 ± 0.44^{b}	24.11 ± 0.48^{b}	0.233 ± 0.020^{b}	0.239 ± 0.020^{b}	9.48 ± 0.60^{a}	10.30 ± 0.66^{a}
NFB × SWE									
Un-inoculated	SWE0	12.27 ± 0.27^{d}	13.23 ± 0.32^{e}	20.48 ± 0.44^{c}	22.25 ± 0.31^{a}	0.148 ± 0.005^{f}	0.156 ± 0.005^{f}	7.44 ± 0.31^{c}	7.67 ± 0.41^{e}
	SWE1	13.13 ± 0.23^{c}	14.19 ± 0.28^{cd}	21.35 ± 0.62^{c}	23.42 ± 0.58^{a}	0.193 ± 0.005^{d}	0.199 ± 0.005^{d}	8.19 ± 0.25^{bc}	8.48 ± 0.41^{de}
	SWE2	12.98 ± 0.23^{c}	13.51 ± 0.26^{de}	20.84 ± 0.43^{c}	22.53 ± 0.32^{a}	0.172 ± 0.006^{e}	0.179 ± 0.006^{e}	8.44 ± 0.44^{bc}	8.92 ± 0.25^{d}
A. chroococcum	SWE0	13.02 ± 0.47^{c}	13.95 ± 0.37^{cde}	20.76 ± 0.43^{c}	22.55 ± 0.33^{a}	0.169 ± 0.004^{e}	0.167 ± 0.006^{ef}	8.00 ± 0.22^{bc}	8.23 ± 0.24^{de}
	SWE1	15.22 ± 0.32^{a}	16.38 ± 0.11^{ab}	24.46 ± 0.74^{ab}	25.45 ± 0.70^{a}	0.305 ± 0.005^{b}	0.311 ± 0.005^{b}	9.13 ± 0.43^{b}	10.02 ± 0.31^{c}
	SWE2	14.46 ± 0.16^{b}	15.81 ± 0.23^{b}	23.16 ± 0.18^{b}	24.41 ± 0.58^{a}	0.221 ± 0.008^{c}	0.225 ± 0.009^{c}	8.25 ± 0.22^{bc}	9.14 ± 0.33^{d}
Lipoferum	SWE0	13.57 ± 0.23^{c}	14.48 ± 0.29^{c}	21.38 ± 0.80^{c}	23.58 ± 0.29^{a}	0.203 ± 0.004^{d}	0.213 ± 0.004^{cd}	9.00 ± 0.36^{b}	9.10 ± 0.16^{d}
	SWE1	15.81 ± 0.21^{a}	16.74 ± 0.27^{a}	25.77 ± 0.25^{a}	27.68 ± 0.67^{a}	0.345 ± 0.008^{a}	0.352 ± 0.008^{a}	11.00 ± 0.40^{a}	11.57 ± 0.55^{b}
	SWE2	15.14 ± 0.05^{ab}	16.19 ± 0.07^{ab}	23.21 ± 0.48^{b}	25.38 ± 0.49^{a}	0.305 ± 0.004^{b}	0.312 ± 0.004^{b}	11.75 ± 0.39^{a}	12.83 ± 0.38^{a}
***P*-value**									
NFB		< 0.001***	< 0.001***	< 0.001***	< 0.001***	< 0.001***	< 0.001***	< 0.001***	< 0.001***
SWE		< 0.001***	< 0.001***	< 0.001***	< 0.001***	< 0.001***	< 0.001***	< 0.001***	< 0.001***
NFB × SWE		0.0466*	0.0194*	0.0440*	0.1052^{ns}	< 0.001***	< 0.001***	0.0259*	< 0.001***

Data (means ± standard error); *$P \leq 0.05$; **$P \leq 0.01$; ***$P \leq 0.001$; ns – non-significant difference. Mean values sharing the same lower-case letter for NFB, seaweed, and their interactions in the same column are not significantly different at $P \leq 0.05$ from Duncan's test; GAE – gallic acid; DW – dry weight

https://doi.org/10.17221/266/2025-PSE

Table 7. Nitrogen (N), phosphorus (P), and potassium (K) of ajwain plants in response to two strains of nitrogen fixing bacteria, seaweed extracts, and their interaction during 2022/2023 and 2023/2024 seasons

		N		P		K	
		(%)					
		1st season	2nd season	1st season	2nd season	1st season	2nd season
Nitrogen fixing bacteria (NFB)							
Un-inoculated (NFB0)		1.28 ± 0.04^{b}	1.34 ± 0.04^{c}	0.215 ± 0.005^{c}	0.227 ± 0.005^{c}	2.13 ± 0.04^{c}	2.28 ± 0.03^{b}
Azotobacter chroococcum (NFB1)		1.35 ± 0.04^{b}	1.46 ± 0.05^{b}	0.237 ± 0.007^{b}	0.252 ± 0.006^{b}	2.37 ± 0.08^{b}	2.63 ± 0.11^{a}
Azospirillum lipoferum (NFB2)		1.69 ± 0.07^{a}	1.79 ± 0.09^{a}	0.268 ± 0.010^{a}	0.282 ± 0.008^{a}	2.54 ± 0.11^{a}	2.77 ± 0.12^{a}
Seaweed extracts (SWE)							
Without (SWE0)		1.30 ± 0.04^{b}	1.34 ± 0.04^{b}	0.219 ± 0.006^{c}	0.235 ± 0.008^{b}	2.13 ± 0.05^{b}	2.26 ± 0.04^{b}
250 mg/L (SWE1)		1.51 ± 0.07^{a}	1.60 ± 0.08^{a}	0.258 ± 0.009^{a}	0.270 ± 0.009^{a}	2.51 ± 0.10^{a}	2.76 ± 0.11^{a}
500 mg/L (SWE2)		1.52 ± 0.10^{a}	1.65 ± 0.10^{a}	0.243 ± 0.011^{b}	0.257 ± 0.010^{a}	2.40 ± 0.09^{a}	2.65 ± 0.11^{a}
NFB × SWE							
	SWE0	1.19 ± 0.05^{c}	1.23 ± 0.07^{e}	0.208 ± 0.007^{a}	0.214 ± 0.007^{a}	2.10 ± 0.10^{c}	2.21 ± 0.08^{c}
Un-inoculated	SWE1	1.31 ± 0.04^{bc}	1.36 ± 0.06^{de}	0.227 ± 0.008^{a}	0.241 ± 0.005^{a}	2.17 ± 0.07^{c}	2.34 ± 0.03^{c}
	SWE2	1.35 ± 0.07^{bc}	1.43 ± 0.04^{d}	0.211 ± 0.008^{a}	0.225 ± 0.006^{a}	2.13 ± 0.07^{c}	2.28 ± 0.05^{c}
	SWE0	1.28 ± 0.04^{bc}	1.32 ± 0.04^{de}	0.215 ± 0.005^{a}	0.233 ± 0.009^{a}	2.14 ± 0.14^{c}	2.26 ± 0.06^{c}
A. chroococcum	SWE1	1.46 ± 0.07^{b}	1.60 ± 0.05^{c}	0.259 ± 0.003^{a}	0.269 ± 0.008^{a}	2.58 ± 0.06^{ab}	2.93 ± 0.09^{ab}
	SWE2	1.32 ± 0.04^{bc}	1.46 ± 0.05^{d}	0.237 ± 0.009^{a}	0.255 ± 0.006^{a}	2.39 ± 0.10^{bc}	2.69 ± 0.16^{b}
	SWE0	1.44 ± 0.06^{b}	1.46 ± 0.03^{d}	0.233 ± 0.013^{a}	0.258 ± 0.008^{a}	2.15 ± 0.07^{c}	2.32 ± 0.09^{c}
Lipoferum	SWE1	1.76 ± 0.06^{a}	1.85 ± 0.09^{b}	0.289 ± 0.002^{a}	0.299 ± 0.011^{a}	2.78 ± 0.08^{a}	3.02 ± 0.09^{a}
	SWE2	1.88 ± 0.06^{a}	2.05 ± 0.06^{a}	0.281 ± 0.008^{a}	0.290 ± 0.006^{a}	2.69 ± 0.07^{a}	2.97 ± 0.06^{ab}
***P*-value**							
NFB		< 0.001***	< 0.001***	< 0.001***	< 0.001***	< 0.001***	< 0.001***
SWE		< 0.001***	< 0.001***	< 0.001***	< 0.001***	< 0.001***	< 0.001***
NFB × SWE		0.0259*	0.0259*	0.0789^{ns}	0.7766^{ns}	0.0450*	0.0142*

Data (means ± standard error); *$P \leq 0.05$; **$P \leq 0.01$; ***$P \leq 0.001$; ns – non-significant difference. Mean values sharing the same lower-case letter for NFB, seaweed, and their interactions in the same column are not significantly different at $P \leq 0.05$ from Duncan's test

https://doi.org/10.17221/266/2025-PSE

for N% since NFB2 × SW2 treatment resulted in the highest values in both seasons.

DISCUSSION

The obtained results in this study revealed that all nitrogen fixing bacteria strains significantly increased the vegetative growth parameters of plant height, number of branches, fresh and dry weights as compared to the control in both seasons. The stimulating effect of NFB on ajwain vegetative growth may be attributed to the phytohormones released by N fixers such as indole-3-acetic acid (IAA) (Taghavi et al. 2009) which increases cell elongation, cell division and differentiation in plants (El-Serafy et al. 2021), indole-3-butyric acid (IBA) and tryptophol that indirectly promote plant growth (Mahdy et al. 2024). Also, yield related traits of number of umbels/plant, fruit yield/plant, and weight of 1 000 fruits were stimulated following nitrogen fixing bacteria as compared to the control ajwains.

It is well known that nitrogen application impact the physiological processes, increases photosynthesis activities and produces more assimilate, biomass, and eventually yield (Ghilavizadeh et al. 2013, El-Serafy et al. 2021, Alayafi et al. 2025). A significant increase in the crop yield following NFB that can fix about 20–200 kg N/ha, promote plant growth and productivity of roselle plants (Bahgat et al. 2023). Free-living N_2-fixing bacteria e.g. *Azotobacter* and *Azospirillum* increases available N in the soil and releases phytohormones of gibberellic acid (GA3) and IAA, which could stimulate plant growth, nutrient absorption, and photosynthesis, leading to an increase in the plant productivity (Sumbul et al. 2020, Din et al. 2021). The highest and significant increase in 1 000 fruit weight and fruit yield was obtained by *Azospirillum lipoferum* than *Azotobactor chroococcum* strains and this may be attributed to the detection ability of bacteria strains to root exudates components, or to poor adaptation to root exudates (Banchio et al. 2008). Higher oil percentage was obtained by ajwain plants which received *Azospirillum lipoferum* and *Azotobactor chroococcum* soil application and this may be due to their simultaneous effect on the uptake of nitrogen, increases the synthesis of secondary metabolites, and thus increasing the amount of essential oil content in the plant as reported by Yogita et al. (2013), and Chahal et al. (2017) in ajwain.

In current investigation, ajwains plants sprayed with SWE showed an enhancement in their vegetative growth traits. This positive effect of SWE can be attributed to its composition of natural growth regulators (cytokinins and auxins) which stimulate plant growth *via* increasing the number of metabolic events; cell division and enlargement which in turn increases growth traits (Prasad et al. 2010). The role of cytokinins in improving overall growth through promoting the development of lateral buds and vascular tissues (Wu and Lin 2000). Additionally, the presence of carbohydrates in the SWE is closely related to the stimulation of lateral buds for growth and differentiation (Youssef et al. 2022). The enhancement in herb dry weight in this study may be due to increase in vegetative growth, which may be reflected in the increase in photosynthesis and the availability of organic nutrients which led to an increase in the plant dry weight (Attememe 2009, Spinelli et al. 2010). It has been found that seaweed extract stimulated root growth consequently more water and nutrients from the soil, causing an increase in the yield (Mancuso et al. 2006; Alam et al. 2013).

Importantly, seaweed foliar application led to an increase in the yield and oil content in ajwain plants. The SWE contains various polysaccharides, nutrients, hormones, betaines, and sterols (Khan et al. 2009). The presence of carbohydrates in the SWE composition is associated with the synthesis of plant secondary metabolites, including essential oils (Elansary et al. 2016) which support the current findings. The beneficial effects of SWE on the essential oil content improvement have been reported in several aromatic species (Tawfeeq et al. 2016, Ghatas et al. 2021). The highest and significant increase in growth and yield was obtained by SW1 than SW2, however, higher doses did not yield additional benefits and, in some cases, caused negative effects. This aligns with Liu et al. (2024), who reported that excessive fertilisation reduces nutrient absorption efficiency and may lead to leaf senescence. Moreover, overuse of seaweed extract may restrict root growth and increase susceptibility to stress and disease, such as root rot and leaf drop (Shireen et al. 2020, Liu et al. 2024). Higher nutrients can disturb internal nutrient balance, consuming energy inefficiently and reducing productivity (El-Serafy et al. 2023).

The results obtained revealed that NFB soil supplementation increased chlorophyll, carbohydrates, and total phenolic compounds in ajwain leaves as compared to the control. Nitrogen fixing bacteria produce growth promoting substances resulting in more efficient absorption of nutrients, which are

https://doi.org/10.17221/266/2025-PSE

the main components of photosynthetic pigments and consequently the chlorophyll and carbohydrates content as well as N, P and K content (Kahil et al. 2017). Leaf pigments play an important role in the phenols and flavonoids formation that contribute to the antioxidant activity of sage plants (Amer et al. 2019). It has been found that plant growth promoting bacteria induced the synthesis of specific phenolic compounds (Kandoliya and Vakharia 2013) which in agreement with current results.

Similarly, total free amino acids and protein content in ajwain plants showed an enhancement following NFB application as compared to the control. These results are in accordance with findings of Płaza et al. (2021) who reported that *Azotobacter* and *Azospirillum* are capable of biological N fixation by reducing the N to ammonia which is necessary for the synthesis of amino acids that contributed to protein synthesis in plants. NFB application increased the N, P, and K percent in leaves as compared to the control. It is well known that biofertilisation inoculation increased root growth, then water and nutrients uptake (Singh and Singh 2016). *Azotobacter* and *Azospirilum* soil application increased macro and micronutrient absorption by plant roots (Adeel et al. 2014). The considerable potential of N fixation by these bacteria is due to their ability to produce the enzyme nitrogenase, which makes it possible to improve N acquisition efficiency (Zhiyong et al. 2024).

Additionally, foliar application of SWE significantly enhanced photosynthetic pigment concentrations and total carbohydrate content, indicating improved photosynthetic activity (Mughunth et al. 2024). These effects may be attributed to the presence of bioactive compounds in seaweed, such as betaines, amino acids, vitamins, and minerals, which stimulate metabolic processes and enzymatic activities (Ali et al. 2021). Furthermore, the increase in chlorophyll content and improved leaf development suggests that seaweed extract acts as a natural biostimulant, promoting plant growth and productivity under various conditions (Mughunth et al. 2024).

The application of seaweed extract has been found to significantly enhance the accumulation of phenolic compounds, amino acids, and proteins in plants. The increase in phenolic content is likely due to the activation of specific biosynthetic pathways involved in the production of secondary metabolites, particularly phenolics (Kocira et al. 2022). Furthermore, the elevated levels of amino acids and proteins can be attributed to improved nitrogen uptake and assimilation, as N is a key element in the synthesis of amino acids and the subsequent formation of proteins. Seaweed extract enhances nitrogen metabolism in plants, thereby facilitating the biosynthesis of nitrogenous compounds, which ultimately supports increased amino acid and protein production (Mohamed et al. 2023). These findings are in agreement with previous studies that highlight the role of seaweed extract in promoting plant growth and metabolic activity through the improvement of nutrient availability and assimilation processes (Ali et al. 2021, 2023). Seaweed foliar application enhances the uptake of essential nutrients such as N, P, and K due to the presence of bioactive compounds that stimulate root activity and improve nutrient absorption. Studies have reported increased nutrient efficiency and uptake in treated plants which contributes to better growth and metabolic performance (Turan and Köse 2004).

Interestingly, NFB or SWE treatments changed the percentages of main components detected in essential oil in this experiment. The alterations in the essential oil compounds proportion can be influenced by various factors, e.g. plant genotype, climatic conditions, growth practices, and harvest time (Preedy 2015, Youssef et al. 2022). In the present experiment, the constituents of ajwain essential oil responded differently to the application of NFB and SWE extracts have shown general improvement in nutrient acquisition capabilities and an improvement in the plant growth.

Seaweed application has been reported to increase in nitrogen acquisition and the transcription studies showed that this was due to an overexpression of the BnNRT1.1/BnNRT2.1 and BnSultr4.1/BnSultr4.2 genes which encode root transporters associated with the uptake (Billard et al. 2014). Nitrogen may be effective in accelerating the enzyme activities that are involved in the conversion of γ-terpinene to thymol. Increasing the application of seaweed extract increased the percentage of thymol and decreased γ-terpinene and p-cymene. Therefore, nitrogen seems to stimulate the biosynthesis of thymol at the expense of γ-terpinene and p-cymene, since these substances are precursors of thymol (Omer et al. 1999).

In conclusions, the effects of incorporated NFB soil supplementation and exogenous SWE foliar application on the growth and productivity of ajwain plants were evaluated in this study. NFB can biologically fix nitrogen and produce biological active substances e.g. auxin, gibberellin, vitamin B, and biotin etc., in the rhizosphere leading to an enhancement in the plant

https://doi.org/10.17221/266/2025-PSE

growth. SWE has important growth-promoting substances, including auxins, cytokinins, gibberellins, and several nutrients which are essential for plant growth and production. NFB with foliar SWE applications had a marked improvement in growth and yield as well as the composition of *Trachyspermum ammi* L. essential oil, in particular the *Azospirillum lipoferum* × *SW*E at 250 mg/L treatment. This combined application (NFB2 × SWE1) was more effective in improving the fruit yield per plant by 27.6% and 32.7% for the first and second seasons, respectively. Also, this treatment resulted in the highest conservation rate of γ-terpinene to thymol, as this treatment exhibited the highest thymol content and least γ-terpinene and P-cymene proportion. Therefore, this sustainable and ecofriendly application is recommended to improve the productivity and quality of ajwain aromatic species with high essential oil quality.

Acknowledgement. The author would like to thank the Deanship of Scientific Research at Tanta University for supporting this work.

REFERENCES

Adeel S., Rehman F.U., Hanif R., Zuber M., Haq E.U., Muneer M. (2014): Ecofriendly dyeing of UV-irradiated cotton using extracts of *Acacia nilotica* Bark (Kikar) as source of quercetin. Asian Journal of Chemistry, 26: 830–834.

Alam M.Z., Braun G., Norrie J., Hodges D.M. (2013): Effect of *Ascophyllum* extract application on plant growth, fruit yield and soil microbial communities of strawberry. Canadian Journal of Plant Science, 93: 23–36.

Alayafi A.H., Dahab A.A., El-Sheshtawy A.A., Sharma A., Elhakem A., Youssef S.M., El-Serafy R.S. (2025): Stimulatory effect of *Delonix regia* flower extract in protecting *Syzygium cumini* seedlings from salinity. Plants, 14: 875.

Ali N.S., Gad M.M., Abdul-Hafeez E.Y. (2023): Evaluating the efficiency of organic manures and seaweed extract on the improvement of growth and productivity of *Foeniculum vulgare* Mill. plants. Assiut Journal of Agricultural Sciences, 54: 91–107.

Ali E.F., Al-Yasi H.M., Issa A.A., Hessini K., Hassan F.A.S. (2022a): Ginger extract and fulvic acid foliar applications as novel practical approaches to improve the growth and productivity of Damask rose. Plants, 11: 412.

Ali E.F., Issa A.A., Al-Yasi H.M., Hessini K., Hassan F.A.S. (2022b): The efficacies of 1-methylcyclopropene and chitosan nanoparticles in preserving the postharvest quality of Damask rose and their underlying biochemical and physiological mechanisms. Biology, 11: 242.

Ali O., Ramsubhag A., Jayaraman J. (2021): Biostimulant properties of seaweed extracts in plants: implications towards sustainable crop production. Plants, 10: 531.

Al-Saif A.M., Ahmed M.E.M., Taha M.A., Sharma A., El-Sheshtawy A.A., Abouelsaad I.A., El-Serafy R.S., Mahdy R.M. (2024): Preharvest applications improve the postharvest storage and quality of tomato fruits by enhancing the nutritional value and antioxidant system. Horticulturae, 10: 1248.

Alshallash K.S., Mohamed M.F., Dahab A.A., Abd El-Salam H.S., El-Serafy R.S. (2022): Biostimulation of *Plectranthus amboinicus* (Lour.) Spreng. with different yeast strains: morphological performance, productivity, phenotypic plasticity, and antioxidant activity. Horticulturae, 8: 887.

Amer H.M., Salem S.H., Salaheldin S., Hussein M.S., Abd El-Fatah S.I. (2019): The growth, chemical composition and evaluation of antimicrobial activity of *Salvia officinalis* oil under partial substitution of mineral NPK fertilizer by bio-fertilizer. Middle East Journal of Applied Sciences, 8: 457–468.

Attememe J.Y.A. (2009): The effect of humic acid and seaweed extracts on growth, chemical characteristics of *Rosmarinus officinalis* L. The 6th Scientific Conference, Biology Department, College of Education, Tikrit University, 1–17.

Bahgat A., Dahab A.A., Elhakem A., Gururani M.A., El-Serafy R.S. (2023): Integrated action of rhizobacteria with *Aloe vera* and moringa leaf extracts improves defense mechanisms in *Hibiscus sabdariffa* L. cultivated in saline soil. Plants, 12: 3684.

Bairwa R., Sodha R.S., Rajawat B.S. (2012): *Trachyspermum ammi*. Pharmacognosy Reviews, 6: 56–60.

Banchio E., Bogino P.C., Zygadlo J., Giordano W. (2008): Plant growth promoting rhizobacteria improve growth and essential oil yield in *Origanum majorana* L. Biochemical Systematics and Ecology, 36: 766–771.

Billard V., Etienne P., Jannin L., Garnica M., Cruz F., Garcia-Mina J.M., Yvin J.C., Ourry A. (2014): Two biostimulants derived from algae or humic acid induce similar responses in the mineral content and gene expression of winter oilseed rape (*Brassica napus* L.). Journal of Plant Growth Regulation, 33: 305–316.

Black C.A., Evans D.D., Ensminger L.E. (1965): Methods of Soil Analysis. Madison, American Society of Agronomy.

Chahal K.K., Dhaiwal K., Kumar A., Kataria D., Singla N. (2017): Chemical composition of *Trachyspermum ammi* L. and its biological properties: a review. Journal of Pharmacognosy and Phytochemistry, 6: 131–140.

Choudhary S., Shivran A.C., Choudhary R.R., Singh R., Daroga S.P. (2017): Effect of nitrogen and plant growth regulators on growth and productivity of Ajwain (*Trachyspermum Ammi* L. Sprague). International Journal of Current Microbiology and Applied Sciences, 6: 396–400.

Christain G.D. (1969): Medicine, trace elements and atomic absorption spectroscopy. Analytical Chemistry, 41: 24–40.

Dere S., Günes T., Sivaci R. (1998): Spectrophotometric determination of chlorophyll-*A*, *B* and total carotenoid contents of some algae species using different solvents. Turkish Journal of Botany, 22: 13–17.

Din I., Khan H., Khan N.A., Khil A. (2021): Inoculation of nitrogen fixing bacteria in conjugation with integrated nitrogen sources

https://doi.org/10.17221/266/2025-PSE

induced changes in phenology, growth, nitrogen assimilation and productivity of wheat crop. Journal of the Saudi Society of Agricultural Sciences, 20: 459–466.

Elansary H.O., Yessoufou K., Shokralla S., Mahmoud E.A., Skalicka-Woźniak K. (2016): Enhancing mint and basil oil composition and antibacterial activity using seaweed extracts. Industrial Crops and Products, 92: 50–56.

El-Serafy R.S., El-Sheshtawy A.A. (2020): Effect of nitrogen fixing bacteria and moringa leaf extract on fruit yield, estragole content and total phenols of organic fennel. Scientia Horticulturae, 265: 109209.

El-Serafy R.S., El-Sheshtawy A.A., Abd El-Razek U.A., Abd El-Hakim A.F., Hasham M.M.A., Sami R., Khojah E., AlMushhin A.A.M. (2021): Growth, yield, quality, and phytochemical behavior of three cultivars of quinoa in response to moringa and *Azolla* extracts under organic farming conditions. Agronomy, 11: 2186.

El-Serafy R.S., El-Sheshtawy A.A., Dahab A.A. (2023): Fruit peel soil supplementation induces physiological and biochemical tolerance in *Schefflera arboricola* L. grown under heat conditions. Journal of Soil Science and Plant Nutrition, 23: 1046–1059.

El-Serafy R.S., El-Sheshtawy A.A., Dahab A.A., Al-Ashkar I. (2021): Can yeast extract and chitosan-oligosaccharide improve fruit yield and modify the pharmaceutical active ingredients of organic fennel? Industrial Crops and Products, 173: 114130.

Ghatas Y., Ali M., Elsadek M., Mohamed Y. (2021): Enhancing growth, productivity and artemisinin content of *Artemisia annua* L. plant using seaweed extract and micronutrients. Industrial Crops and Products, 161: 113202.

Ghilavizadeh A., Darzi M.T., Hadi M.H.S. (2013): Effects of biofertilizer and plant density on essential oil content and yield traits of ajowan (*Carum copticum*). Middle East Journal of Scientific Research, 14: 1508–1512.

Jackson W.A. (1978): Nitrate acquisition and assimilation by higher plants: processes in the root system. In: Nielsen D.R., MacDonald J.G. (eds.): Nitrogen in the Environment. London, Academic Press, 45–88.

Jones D.B. (1941): Factors for converting percentages of nitrogen in foods and feeds into percentages of protein. Circular No. 183. United States Department of Agriculture, Washington, 1–22.

Kahil A.A., Hassan F.A.S., Ali E.F. (2017): Influence of bio-fertilizers on growth, yield and anthocyanin content of *Hibiscus sabdariffa* L. plant under Taif region conditions. Annual Research Review in Biology, 17: 1–15.

Kandoliya U.K., Vakharia D.N. (2013): Induced resistance and phenolic acid accumulation in biological control of chickpea wilt by *Pseudomonas fluorescens*. Asian Journal of Bio Science, 8: 184–188.

Khan W., Rayirath U.P., Subramanian S., Jithesh M.N., Rayorath P., Hodges D.M., Critchley A.T., Craigie J.S., Norrie J., Prithiviraj B. (2009): Seaweed extracts as biostimulants of plant growth and development. Journal of Plant Growth Regulation, 28: 386–399.

Kocira S., Pérez-Pizá M.C., Bohata A., Bartos P., Szparaga A. (2022): Cold plasma as a potential activator of plant biostimulants. Sustainability, 14: 495.

Lawless J. (1992): The Encyclopedia of Essential Oils. Element Book. Ltd. Long Mead, Shoftesbury, Dorest.

Liu J., Wang D., Yan X., Jia L., Chen N., Liu J., Zhao P., Zhou L., Cao Q. (2024): Effect of nitrogen, phosphorus and potassium fertilization management on soil properties and leaf traits and yield of *Sapindus mukorossi*. Frontiers in Plant Science, 15: 1300683.

Mahdy R.M., Al-Saif A.M., Ahmed M.E.M., El-Bary T.S.A., Sharma A., El-Sheshtawy A.A., El-Serafy R.S., El-Ghany T.S.A. (2024): Evaluation of two different methods of fulvic acid application (seed priming and foliar spray) on growth, yield, and nutritional quality of pea (*Pisum sativum* L.). Plants, 13: 3380.

Mancuso S., Azzarello E., Mugnai S., Briand X. (2006): Marine bioactive substances (IPA extract) improve foliar ion uptake and water stress tolerance in potted *Vitis vinifera* plants. Advances in Horticultural Sciences, 20: 156–161.

McDonald S., Prenzler P.D., Antolovich M., Robards K. (2001): Phenolic content and antioxidant activity of olive extracts. Food Chemistry, 73: 73–84.

Mohamed S.M., Shalaby M.A., El-Shiekh R.A., El-Banna H.A., Emam S.R., Bakr A.F. (2023): Metabolic syndrome: risk factors, diagnosis, pathogenesis, and management with natural approaches. Food Chemistry Advances, 3: 100335.

Moussa M.M., Mansour M.M.F., El-Sharnouby M., Hassan F.A.S. (2024): Foliar application of bee honey or ginger extract enhanced *Salvia officinalis* L. growth and productivity by improving phytohormones, antioxidants, chlorophylls, and nutrients. Agronomy, 14: 1819.

Mughunth R.J., Velmurugan S., Mohanalakshmi M., Vanitha K. (2024): A review of seaweed extract's potential as a biostimulant to enhance growth and mitigate stress in horticulture crops. Scientia Horticulturae, 334: 113312.

Mukherjee A., Patel J. (2020): Seaweed extract: biostimulator of plant defense and plant productivity. International Journal of Environmental Science and Technology, 17: 553–558.

Naghdi Badi H.A., Lotfi Zadeh M., Qavami N., Mehrafarin A., Khavazi K. (2013): Response of quantity and quality yield of valerian herb (*Valeriana officinalis* L.) to application of phosphorus bio/chemical fertilizers. Journal of Medicnal Plants, 12: 25–37.

Omer E.A. (1999): Response of wild Egyptian oregano to nitrogen fertilization in a sandy soil. Journal of Plant Nutrition, 22: 103–114.

Omer E.A., Said-Al Ahl H.A.H., El Gendy A.G. (2014): Yield and essential oil of Ajwain (*Trachyspermum ammi*) plant cultivated in saline soil of north Sinai in Egypt. Journal of Essential Oil Bearing Plants, 17: 469–477.

Piper C.S. (1967): Soil and Plant Analysis. 2nd Edition. Bombay, Asia Publishing House.

Płaza G., Jałowiecki Ł., Głowacka D., Hubeny J., Harnisz M., Korzeniewska E. (2021): Insights into the microbial diversity and structure in a full-scale municipal wastewater treatment plant with particular regard to Archaea. PLoS One, 16: e0250514.

https://doi.org/10.17221/266/2025-PSE

Prasad K., Das A.K., Oza M.D., Brahmbhatt H., Siddhanta A.K., Meena R., Eswaran K., Rajyagur M.R., Ghosh P.K. (2010): Detection and quantification of some plant growth regulators in a seaweed-based foliar spray employing a mass spectrometric technique sans chromatographic separation. Journal of Agricultural and Food Chemistry, 58: 4594–4601.

Preedy V.R. (2015): Essential Oils in Food Preservation, Flavor and Safety. Cambridge, Academic Press.

Pruthi J.S. (2001): Minor Spices and Condiments, Crop Management and Post-Harvest Technology. Delhi, Indian Council of Agricultural Research, 124–133.

Sathyanarayana E., Hadole S.S., Ghawade S.M. (2017): Effect of nutrient levels on nutrient uptake by Ajwain crop (*Trachyspermum ammi* L. Sprague). International Journal of Chemical Studies, 5: 1987–1990.

Shireen F., Nawaz M.A., Xiong M., Ahmad A., Sohail H., Chen Z., Abouseif Y., Huang Y., Bie Z. (2020): Pumpkin rootstock improves the growth and development of watermelon by enhancing uptake and transport of boron and regulating the gene expression. Plant Physiology and Biochemistry, 154: 204–218.

Singh G., Singh S.S. (2016): Effects of FYM, biofertilisers and inorganic fertilizers on yield and nutrients uptake of European dill (*Anethum graveolens* L.). International Journal of Agricultural Science and Research, 6: 301–304.

Sivropoulou A., Papanikolaou E., Nilolaou C., Kokkini S., Lanaras T., Arsenakis M. (1996): Antimicrobial and cytotoxic activities of origanum essential oils. Journal of Agricultural and Food Chemistry, 44: 1202–1205.

Spinelli F.S., Fiori G.K., Noferini M.L., Sprocatti M.D., Costa G.R. (2010): A novel type of seaweed extract as a natural alternative to the use of iron chelates in strawberry production. Scientia Horticulturae, 125: 263–269.

Sumbul A., Ansari R.A., Rizvi R., Mahmood I. (2020): *Azotobacter*: a potential bio-fertilizer for soil and plant health management. Saudi Journal of Biological Sciences, 27: 3634–3640.

Taghavi S., Garafola C., Monchy S., Newman L., Hoffman A., Weyens N., Barac T., Vangronsveld J., van der Lelie D. (2009): Genome survey and characterization of endophytic bacteria exhibiting a beneficial effect on growth and development of poplar trees. Applied and Environmental Microbiology, 75: 748–757.

Tawfeeq A., Culham A., Davis F., Reeves M. (2016): Does fertilizer type and method of application cause significant differences in essential oil yield and composition in rosemary (*Rosmarinus officinalis* L.)? Industrial Crops and Products, 88: 17–22.

Turan M., Köse C. (2004): Seaweed extracts improve copper uptake of grapevine. Acta Agriculturae Scandinavica, Section B-Soil and Plant Science, 54: 213–220.

Tursun A.O. (2022): Effect of foliar application of seaweed (organic fertilizer) on yield, essential oil and chemical composition of coriander. PlosOne, 17: e0269067.

Viuda-Martos M., Mohamady M.A., Fernández-López J., AbdElRazik K.A., Omer E.A., Pérez-Alvarez J.A., Sendra E. (2011): *In vitro* antioxidant and antibacterial activities of essentials oils obtained from Egyptian aromatic plants. Food Control, 22: 1715–1722.

Wu Y.T., Lin C.H. (2000): Analysis of cytokinin activity in commercial aqueous seaweed extract. Gartenbauwissenschaft, 65: 170–173.

Yemm E.W., Cocking E.C. (1955): The determination of amino acids with ninhydrin. Analyst, 80: 209–213.

Yogita R., Nikam T.D., Dhumal K.N. (2013): Effect of foliar application of plant growth regulators on growth, yield and essential oil components of Ajwain (*Trachyspermum ammi* L.). International Journal of Seed Spices, 3: 34–41.

Youssef S.M., El-Serafy R.S., Ghanem K.Z., Elhakem A., Abdel Aal A.A. (2022): Foliar spray or soil drench: microalgae application impacts on soil microbiology, morpho-physiological and biochemical responses, oil and fatty acid profiles of Chia plants under alkaline stress. Biology, 11: 1844.

Zarshenas M., Moein M., Samani S., Petramfar P. (2014): An overview on Ajwain (*Trachyspermum ammi*) pharmacological effects; modern and traditional. Journal of Natural Remedies, 14: 98–105.

Zhiyong S., Yaxuan G., Yuanyuan W., Xiang Y., Xu G., Zhenhong L., Zhenyu L. (2024): Nitrogen-fixing bacteria promote growth and bioactive components accumulation of *Astragalus mongholicus* by regulating plant metabolism and rhizosphere microbiota. BMC Microbiology, 24: 377–381.

Received: June 15, 2025
Accepted: July 23, 2025
Published online: August 24, 2025