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# Algal application as a biological control method of root-knot nematode *Meloidogyne incognita* on cucumber under protected culture conditions and its impact on yield and fruit quality

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## Abstract

This study was conducted during seasons 2016 and 2017 to control the root-knot nematode *Meloidogyne incognita* in cucumber, cultivated in infected soil, using some algal treatments under greenhouse conditions, at Kaha Farm, Qalubia Governorate, Egypt. Six algal treatments were tested: two foliar applications of *Spirulina* and *Amphora*, two drenched soil applications of *Spirulina* and *Amphora*, two treatments using *Spirulina*, as spraying and drenching, and *Amphora*, as spraying and drenching, in addition to the Rugby nematicide (10% Ebufos, at the rate of 5 g/m<sup>2</sup>) and control. Rugby was applied by a soil prepared in its experimental units. After 15 days from transplanting, the algal extract treatments were applied twice monthly for 3 months. The same concentration (2 g/l) of both types of algae was applied in both foliar and drench treatments. The control was sprayed only by water. The results indicated that the soil drenched with *Amphora* or *Spirulina* extracts had significant increments in vegetative growth, yield, and fruit quality. In contrast, the control plants had the lowest values in all criteria. *Amphora* (sprayed with soil drenched) treatment gave 2.5 and 2.69 folds the control in marketable yield in 2016 and 2017 seasons, respectively. The combination of sprayed and soil drenched with *Amphora* was more effective in nematode's control or in enhancing plant resistance for nematode as shown at most nematode parameters, especially the rate of nematode reproduction factor (RF), which reached 0.42 and 0.45 in both seasons, respectively. It had insignificant differences compared with the nematicide. Therefore, using algae for the biological control of root-knot nematodes is recommended, especially in sustainable agriculture for maintaining the soil and improve fertility.

**Keywords:** Algae, Root-knot nematode, *Meloidogyne incognita*, Control, Cucumber, Yield and fruit quality

## Background

Cucumber, *Cucumis sativus* L., is one of the cash crops grown worldwide. Plant nematodes cause a great damage in agriculture. In Egypt, they have become dangerous pests in most crops, especially in the newly reclaimed sandy soil. Cucumber is highly susceptible to root-knot nematodes (Sikora and Fernández 2005).

Synthetic nematicides are one of the most effective methods to manage root-knot nematodes. However, their negative potential impact on the environment and ineffectiveness after prolonged use have led to a total ban or restricted use of most nematicides and an urgent need for safe and effective options. Biological control promises to be one of very important alternative methods. Nonchemical and eco-friendly strategies for nematode management such as soil management, organic amendments, fertilization, biological control, and heat are a major economic and environmental acceptable methods to control root-knot nematodes (Collange et al. 2011). *Spirulina* is a type of blue-green alga

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that is rich in proteins, vitamins, minerals, carotenoids, and antioxidants that can help the plants to produce a good yield and resist the nematodes (Shawky et al. 2009).

Youssef and Ali (1998) mentioned that three species of blue-green algae: *Anabena oryzae*, *Nostoc calcicola*, and *Spirulina* sp. together achieved the highest significant reduction in the number of galls and egg masses of *Meloidogyne incognita* infecting cowpea and improved plant growth. Culture of the *Microcoleus vaginatus* (alga) inhibited hatching of *M. incognita* eggs and killed second stage juveniles (Khan et al. 1997). Sharma and Gaur (2008) showed that the extracts and exudates of five cyanobacteria made a significant inhibition of *M. incognita*. Shawky et al. (2009) recommended the bio-organic application of nematicides against the root-knot nematode in cucumber to reduce the soil and plant polluting by chemical nematicides. Aqueous extract of *Spirulina platensis* enhanced the control of *M. incognita* and achieved the highest reduction in the number of the second-stage juveniles in soil, numbers of galls, developmental stages, females, egg masses, and egg numbers/egg mass in roots of cucumber. In addition, all applications significantly improved vegetative growth and increased the yield of cucumber. Besides, they enhanced the soil biological activity, total cyanobacterial count, CO<sub>2</sub> evolution, dehydrogenase and nitrogenase activities, and available nitrogen, phosphorus, and potassium that resulted in increasing of soil electrical conductivity and soil pH reduction. Youssef and Eissa (2014) found that *Nostoc*, *Anabaena*, *Oscillatoria*, *Aulosira*, and *Lyngbya* enriched paddy field soil by fixing atmospheric nitrogen and supply vitamin B complex and growth-promoting substances.

Radwan et al. (2012) reported that bio-products made a significant reduction in the root galls and the numbers of juveniles in the soil. Cyanobacteria application on tomato plants had an effect in the prevention of root-knot nematode *M. incognita* and gave a harmful effect of bio-mass on gall induction (Pushparaj et al. 2000).

Wu et al. (1997) reported the role of betaines in alkaline extracts of the marine brown alga, *Ascophyllum nodosum*, in suppression and decreasing the fecundity of the tomato root-knot nematode, *M. incognita*. Furthermore, cyanobacterium could be used as a protection agent against *M. incognita* in tomato, as reported by Pushparaj et al. (2000). *A. nodosum* extracts in the soil decreased the infection of tomato plants by root-knot nematodes and reduced the number of eggs (Massa 2010).

Enan et al. (2016) reported that the foliar application of algal extract, using *Spirulina*, produced significantly higher values of photosynthetic pigments, vegetative growth, sugar content, quality, minerals contents in leaves, and root yield on the sugar beet. Also, algal extracts promote vegetative growth and crop yield and

quality and in addition increase the tolerance to stress conditions (Ghalab and Salem 2001). Moreover, they increase the photosynthetic pigments and improve total carbohydrates, starch, amino acids, and proteins (Yassen et al. 2007). Algae were used on many plants as growth stimulants such as cucumber (Ahmed and Shalaby 2012) and tomato (El-Sayed et al. 2018).

*Amphora* contained high protein, lipid, and carbohydrate besides the high amounts of essential amino acids (Chtourou et al. 2015). *Amphora* had antioxidant activities (Lee et al. 2008).

The present study was carried out to evaluate the efficacy of two types of algae through different application methods to manage the root-knot nematode *M. incognita* infecting cucumber plants, and as growth promoters under greenhouse conditions.

## Material and methods

This study was carried out during the two successive seasons of 2016 and 2017 under greenhouse conditions at a highly *M. incognita*-infected soil at the experimental farm of Kaha Vegetable Research Farm, Horticulture Research Institute, Agriculture Research Center (ARC), Qalubia Governorate, Egypt, whereas the initial nematode populations were 13,752 and 14,124 second-stage juveniles/250 g soil in the first and second seasons, respectively.

## Plant materials

Cucumber, Hesham hybrid F<sub>1</sub> (Seminis Seed Company), seeds were sown on the fifth of February in 2016 and eighth of February in 2017 in seedling trays with 84 cells, filled with a mixture of peat moss and vermiculite at the ratio of 1:1 (v/v). Three hundred grams of ammonium sulfate, 400 g of calcium superphosphate, 150 g of potassium sulfate, 50 ml of nutrient solution, and 50 g of a fungicide were added for each 50 kg of the peat moss in the greenhouse nursery. Seedlings were transplanted to a greenhouse, on March 9, 2016, and March 12, 2017. Seedlings were transplanted into two rows in the bed (row 7 m length and 1.0 m width). The space between the plants was 0.50 m. The experimental unit contained 15 plants in each row. The conventional agricultural practices, i.e., irrigation, fertilization, weeding, and pest control, followed the standard commercial practices and were carried out as recommended by the Ministry of the Agriculture in Egypt, for cucumber production.

## Time of application

The experimental treatments, each with three replicates, were as follows: sprayed with *Spirulina platensis*, soil

drenched with *S. platensis*, sprayed with *Amphora coffeaeformis*, soil drenched with *A. coffeaeformis*, sprayed and soil drenched with *S. platensis*, sprayed and soil drenched with *A. coffeaeformis*, soil drenched with contact action nematicide, Rugby (organophosphates group 10% Ebufos), as a granule at the rate of 5 g/m<sup>2</sup>, as well as control. Two weeks after transplanting, the first foliar application was executed by spraying the cucumber plants, using alga extracts once biweekly, which continued for 3 months. The soil addition (drench) treatments were performed at the same time, while the Rugby nematicide was performed by a soil prepared in its experimental units. The recommended concentration of both types of alga (2 g/l) was applied in both foliar or soil drench treatments. The control plant was sprayed only with water in all times.

#### Algae extract

The ready-made algal extract was obtained from Algal Biotechnology Unit, National Research Centre (NRC), Giza, Egypt. The blue-green alga, *Spirulina platensis*, belonging to Cyanophyta, and *Amphora coffeaeformis* were massively produced at the Algal Biotechnology Unit, NRC in continuous cultures. Algal extracts were prepared as previously described by Enan et al. (2016).

The alga *A. coffeaeformis* was locally isolated from the Egyptian irrigation water. Previous studies concerned with this alga focused on fish and animal nutrition. Regarding plant nutrition, recent studies are running on its high contents of polysaccharides, amino acid, and phytohormones. *Amphora* supposed to have a positive effect on the plant nutrition that overcomes the adverse effect of biotic stress as it contents have amino acids, proteins, and plant hormones.

#### Studied characteristics

##### Horticultural characteristics

###### A. Vegetative growth characteristics

Vegetative growth characters were recorded 75 days after transplanting of three plants randomly chosen from each plot as follows: (a) plant length (cm), (b) average internode length (cm), (c) number of leaves per plant, (d) stem diameter (cm), (e) number of branches/plant, (f) plant fresh weight (g), (g) number of days to flowering, (h) fruit set percentage, and (i) leaf area (cm<sup>2</sup>).

Leaf area (cm<sup>2</sup>) was expressed as the mean leaf area in square centimeters, using the fresh weight method. The leaves were cleaned from dust and then weighed to the nearest 0.001 g. Therefore, 20 disks of known area were separated as weight

$$\text{leaf area cm}^2 = \frac{\text{fresh weight of one leaf No.10}}{\text{fresh weight of 20 disks}} \times 20 \times \text{area of disk}$$

where the area of a disk is about 1.0 cm.

###### B. Yield and its components

Yield and its components are as follows: (a) total weight of fruits /plant, (b) total number of fruits/plant, (c) early yield (kg/plant), (d) early yield (number/plant), (e) total yield (ton/greenhouse), (f) marketable yield, and (g) percentage of unmarketable yield.

###### C. Fruit characteristics

Fruit characteristics are as follows: (a) fruit length (cm), (b) fruit diameter (cm), and (c) average fruit weight (g).

#### Chemical analyses

- Plant dry weight (g) was measured as the weight of the same plants used for plant fresh weight after being dried out in an oven with driven hot air at 70 °C until a constant weight is achieved.
- Plant dry matter percentage was measured by this equation:

$$\text{Plant dry matter}\% = \frac{\text{Plant dry weight (g)}}{\text{Plant fresh weight (g)}} \times 100$$

- Fruit dry matter (%) was determined by allowing 100 g of fruit fresh weight to dry in an oven at 70 °C till a constant weight is achieved.
- Total soluble solids (TSS %) were measured in fruit juice by using a hand refractometer, according to the methods of AOAC (2012). Five fruits were taken at random from each treatment for this test.
- For determination of leaf pigments, chlorophyll content, which used the fifth leaf from the shoots' growing tip, was measured by a SPAD 501 leaf chlorophyll meter (Yadava 1986).
- Total nitrogen in leaves was assayed in milligrams per gram dry weight by the micro-Kjeldahle method indicated by Pregl (1945).
- Potassium concentrations in leaves, in mg/g dry weight, were determined using a flam-photo metrically, according to the method described by Brown and Lilleland (1946).

- h. Total phosphorus (%) was determined calorimetrically, according to the method described by Murphy and Riely (1962), as modified by John (1970).

**Nematode estimation**

The initial population density in the infested field was estimated by modified sieving and Bearman’s plate technique (Viglierchio and Schmitt 1983). At the end of the season, plants were uprooted; roots were washed by tap water and stained by an acid fuchsine (Franklin and Goodey 1959). Numbers of galls, egg masses, and eggs/plant root were recorded on roots stained with red acid fuchsine.

**Experimental design and statistical analysis**

The experiments’ layouts were randomized complete blocks design, with three replications. Data were statistically analyzed, using analyses of variance (ANOVA) with the Stat soft statistical package (MSTATC) software program (Michigan State University, East Lansing, MI, USA). Probabilities of significance among treatments and means compared with the least significant difference (LSD) ( $P \leq 0.05$ ) were used to compare means within and among treatments according to Gomez and Gomez (1984).

**Results and discussion**

**Algal effects on horticultural characteristics**

**Effect of algal treatments on vegetative growth and flowering**

Data presented in Tables 1 and 2 showed that the plant and internode length, number of branches and leaves per plant, plant fresh and dry weight, stem diameter, and leaf area of cucumber plants had significant increases than the characteristics of the control in both seasons.

Different algal treatments had insignificant differences in most vegetative characteristics compared with Rugby (chemical nematicide) treatment, except *Amphora* (spray + soil drench) treatment, which demonstrated a significant superiority in most vegetative characteristics in both seasons compared with the Rugby nematicide and control. These results may be due to some secondary metabolite products such as the hormone components, indoles, cytokinins, gibberellins, brassinosteroids, and other compounds, which are considered as plant growth regulators including amino acids, peptides, and polyamines in *Amphora* in high concentrations than in *Spirulina*. This conclusion is consistent with that of Lee et al. (2008), in addition to a higher amount of different antimicrobial and nematicidal actions such as antioxidants, polyphenols, flavonoids, and some enzymes as record by Chtourou et al. (2015).

Finally, all biological treatments showed good effects in the confrontation with chemical nematicide, while *Amphora* (spray + soil drench) had a significant increment in plant vigor and gave an excellent impact compared with chemical nematicide (Rugby). These results agree with that of Radwan et al. (2012).

Insignificant differences were recorded among the plants regarding the chlorophyll SPAD index, which was measured before treatments’ implementation; in contrast, significant increments were observed in this parameter after application compared with the control plants, whereas *Amphora* (spray + soil drench) improved the chlorophyll content compared with the Rugby treatment. The foliar application of algal extract, using *Spirulina*, produced significantly higher values of photosynthetic pigments (chlorophyll a and b and carotenoids). These results are likely due to the high contents of algae and their extract of cytokinins and similar substances. The growth promoters’ components in algae

**Table 1** Effect of algal treatments of cucumber Hesham hybrid compared with control and Rugby nematicide on vegetative growth under infested soil with root-knot nematodes in 2016 and 2017 seasons

Treatments	2016						2017					
	Plant length cm	Number		Stem diameter mm	Internode length cm	Leaf area cm <sup>2</sup>	Plant length cm	Number		Stem diameter mm	Internode length cm	Leaf area cm <sup>2</sup>
		Branches	Leaves					Branch	Leaves			
Control	231	16	45	16.5	10.3	187	248	15	44	17.6	9.40	196
Rugby 10G	345	24	61	25.5	9.8	286	378	28	62	28.2	10.6	298
Spirulina spray	290	21	52	24.7	12.1	278	322	24	47	26.9	11.8	286
Spirulina drench	315	21	54	26.2	13.5	256	363	24	55	27.1	12.1	274
Amphora spray	320	24	54	25.3	12.8	294	345	26	57	27.6	12.5	302
Amphora drench	335	23	56	25.3	14.2	312	370	27	60	26.6	12.7	321
Spirulina spray + drench	335	28	63	26.4	13.7	278	365	26	59	27.4	14.0	287
Amphora spray + drench	375	31	66	27.6	13.9	338	387	33	64	27.9	14.2	323
LSD at t <sub>0.05</sub>	56	1.9	5.3	2.10	1.10	43	42	2.1	4.8	2.20	1.40	38

**Table 2** Effect of algal treatments of cucumber Hesham hybrid compared with control and Rugby nematicide on plant fresh and dry weight, dry matter percentage, chlorophyll SPAD index, number of days to flowering, and fruit set percentage under infested soil with root-knot nematodes in 2016 and 2017 seasons

Treatments	2016							2017						
	Plant weight (g)		Dry matter (%)	SPAD index		No. of days to flowering	Fruit set%	Plant weight (g)		Dry matter (%)	SPAD index		No. of days to flowering	Fruit set%
	Fresh	Dry		Before	After			Fresh	dry		Before	After		
Control	618	53.46	8.65	39.6	42.7	21	58	675	59.46	8.8	40.2	43.1	22	52
Rugby 10G	830	77.19	9.30	40.6	48.7	21	71	812	71.19	8.8	41.2	47.7	21	69
Spirulina spray	690	61.66	8.94	40.1	46.3	22	67	711	64.70	9.1	40.6	46.8	21	70
Spirulina drench	745	66.01	8.86	39.8	49.5	22	66	765	71.01	9.3	41.0	49.4	22	68
Amphora spray	722	66.65	9.22	40.4	49.8	21	69	734	71.19	9.7	40.7	48.7	21	71
Amphora drench	822	74.88	9.11	39.8	50.8	20	72	824	83.6	10.1	40.8	49.3	21	69
Spirulina spray + drench	802	75.23	9.38	39.9	49.7	21	69	789	70.23	8.9	41.5	48.2	22	68
Amphora spray + drench	845	81.79	9.70	40.4	53.4	21	75	839	80.4	9.5	40.9	51.7	21	72
LSD at t <sub>0.05</sub>	42.0	7.30	0.36	NS	3.60	NS	3.4	33.2	8.50	0.48	NS	2.3	NS	2.2

may cause these influences in cucumber plants. Similar results were obtained by Enan et al. (2016) on sugar beet and El-Sayed et al. (2018) on tomato.

Insignificant differences were found among all treatments over the control, in the number of days to flowering in both seasons. This indicated that all algal treatments had no effect on earliness. Fruit set percentage showed a significant enhancement in the biological treatments, specially *Amphora* (spray + soil drench), which had 75 and 72%, while the chemical nematicide gave 71 and 69% in the first and second seasons, respectively. Ahmed and Shalaby (2012) recorded the seaweed extracts of green and red algae or commercial seaweed extract (Algreen), as suitable applications to improve vegetative growth and yield of cucumber. Furthermore, similar results were obtained by Youssef and Ali (1998) on cowpea, Shawky et al. (2009) on cucumber, and El-Sayed et al. (2018) on tomato.

Data in Table 3 indicated significant differences among all treatments and control in early and total yields as a number and weight of fruits per plant or per greenhouse, in addition to the marketable yield and the percentage of unmarketable yield in both seasons. The fruit yield was significantly higher in the average fruit weight and the number of fruits per plant by using *Amphora* (spray + soil drenched). For instance, the total marketable yield with *Amphora* (spray + soil drench) was 2.5 and 2.69 folds the control in 2016 and 2017 seasons, respectively.

Also, the cucumber marketable yield of *Amphora* (spray + soil drench) treatment gave 1.18 and 1.11 folds the Rugby nematicide treatment in the first and second seasons, respectively. While the marketable yield of cucumber plants treated with *Spirulina* (spray + soil drench) gave 2.17 and 2.37 folds the control in both seasons, respectively. The cucumber plants treated with

Rugby nematicide had 1.02 and 0.98 folds the *Spirulina* (spray + soil drench) in both seasons 2016 and 2017, respectively. But *Amphora* soil drenched treatment had 4.436 tons per greenhouse, with about 256-kg increase in margin than the cucumber plants treated with Rugby nematicide, which gave 4.180 tons in the first season and 4.615 tons by a margin of 2 kg than cucumber plants treated with Rugby nematicide, which gave 4.612 tons in the second season. These results may be due to the effects of other growth factors in the composition of algae extract such as phytohormones, prohormones, amino acids, and fatty acids, in addition to the algae extraction which is considered as a large storage of mineral nutrients. All algal extraction components increased the photosynthetic pigments, maximizing the storage of carbohydrates and starch and increasing the cucumber yield and its components. Furthermore, algae contain some antioxidants, polyphenols, flavonoids, and other compounds which resist the nematodes, limiting their spread and reducing their harmful effects as reported by Shawky et al. (2009).

The treatments included both foliar and drench applications at the same time such as *Amphora* and *Spirulina* spray with drench which had the best effect. Spraying algae had effects of encouraging to achieve the growth of plants by more compounds such as prohormones or phytohormones, and effects of amino acids, fatty acids, and other protein compounds such as lipids and peptides and mineral nutrition in chelate compounds that led to increased plant growth and produced healthy plants, which reflect on high yield and better quality. In addition, the lethal effects gave a harmful effect to the second generation of nematodes, followed by decreasing the juveniles and reducing the gall and egg formation, which resulted from the presence of compounds similar

**Table 3** Effect of algal treatments of cucumber Hesham hybrid compared with control and Rugby nematicide on early and total, marketable yield and unmarketable yield percentage under infested soil with root-knot nematodes in 2016 and 2017 seasons

Treatments	2016						2017					
	Yield/Plant		Early Yield/Plant		Marketable yield (ton/ Greenhouse)	Unmarketable (%)	Yield/Plant		Early Yield/Plant		Marketable Yield (ton/ Green house)	Unmarketable (%)
	Number	Weight (kg)	Number	Weight (g)			Number	Weight (kg)	Number	Weight (g)		
Control	21	2.150	2.21	215.3	1.970	8.38	20	2.095	3.1	312.3	1.906	9.01
Rugby 10G	37	4.431	5.30	634.9	4.180	5.66	41	4.874	5.5	653.8	4.612	5.38
Spirulina spray	27	3.229	5.00	598.0	3.107	3.76	30	3.214	4.8	578.2	3.423	5.28
Spirulina drench	34	4.375	5.21	670.5	4.102	6.25	36	3.405	5.3	648.5	4.158	5.61
Amphora spray	30	3.695	5.62	692.4	3.462	6.31	34	4.464	5.3	695.9	4.248	4.84
Amphora drench	38	4.714	5.83	722.9	4.436	5.89	41	4.647	5.6	662.0	4.615	4.78
Spirulina spray + drench	35	4.505	5.81	747.7	4.280	5.00	41	4.626	5.4	625.1	4.510	4.98
Amphora spray + drench	42	5.168	6.24	767.5	4.923	4.74	45	4.981	5.9	721.5	5.136	4.56
LSD at t 0.05	3.8	0.311	1.23	46	1.120	1.43	4.1	0.521	1.11	55	1.44	1.87

to nematicides such as flavonoids, polyphenols, phenylalanine, and antioxidants. This effect is confirmed by Youssef and Eissa (2014) who reported that *Azolla*, *Anabaena symbiosis* Nostoc, *Anabaena*, *Oscillatoria*, *Aulosira*, and *Lyngbya* play an important role of fixing atmospheric nitrogen and supplying vitamin B complex and growth-promoting substances.

On the other hand, the percentage of unmarketable yield was decreased by *Amphora* (spray + drench)

treatment, followed by *Spirulina* drench in the first season giving 3.76% compared with the percentages of unmarketable yield of control plants that gave 8.38% and 9.1% in both seasons, respectively. These results may be due to the highest amounts of amino acids and total protein contents in both algae. These results agree with those of Ahmed and Shalaby (2012) who recorded the highest amounts of amino acid, fatty acid, and polypeptides in algae extract, which improved the fruit quality.

**Table 4** Effect of algal treatments of cucumber Hesham hybrid compared with control and Rugby nematicide on fruit characters, total soluble solids, and NPK under infested soil with root-knot nematodes in 2016 and 2017 seasons

Treatments	2016								2017							
	Fruit weight (g)	Fruit (cm)		T.S.S	Fruit dry matter (%)	Minerals			Fruit weight (g)	Fruit		T.S.S	Fruit dry matter (%)	Minerals (%)		
		Length	Diameter			N	P	K		Length	Diameter			N	P	K
Control	102.4	14.6	2.2	4.2	5.2	1.414	0.20	1.13	104.75	13.8	2.4	4.6	4.9	1.325	0.23	1.24
Rugby 10G	119.8	17.6	2.8	4.2	4.8	3.257	0.46	3.09	118.878	17.21	2.9	4.6	4.7	3.39	0.39	2.78
Spirulina spray	119.6	17.8	2.8	4.4	4.6	2.114	0.30	4.12	120.467	16.98	3	4.5	4.7	2.451	0.32	3.96
Spirulina drench	128.7	17.8	2.7	4.2	4.7	3.052	0.447	2.97	122.361	17.4	2.9	4.5	4.8	3.292	0.39	2.37
Amphora spray	123.2	17.5	3.1	4.2	4.5	3.283	0.335	3.62	131.294	17.5	3.2	4.5	4.7	3.415	0.36	3.54
Amphora drench	124.0	18.4	3.2	4.3	4.6	3.433	0.540	4.91	118.22	17.2	3.2	4.4	4.8	3.79	0.48	4.34
Spirulina spray + drench	128.7	17.8	3.1	4.3	4.4	2.534	0.415	3.63	115.756	18.1	2.8	4.6	4.9	3.14	0.45	3.48
Amphora spray + drench	123.0	18.8	3.2	4.6	4.2	4.216	0.542	3.51	122.295	17.8	2.9	4.6	4.8	4.156	0.56	4.68
LSD at t 0.05	7.400	1.32	0.4	NS	NS	0.654	0.121	0.54	12.45	2.31	0.3	NS	NS	0.461	0.10	0.514

Data presented in Table 4 indicated that *Amphora* (spray + drench) treatment showed a significant increase in most of the fruit characteristics. Regarding mineral concentrations, nitrogen, phosphorus, and potassium, *Amphora* (spray + drench) showed a significant increase compared to Rugby and control for this trait. The results could be due to the algae's large amounts of mineral nutrients (Marrez et al. 2014). On the other hand, data showed insignificant differences between all treatments and control for the total soluble solid and fruit dry matter. These results agree with those of Youssef and Eissa (2014) who reported an increase of 10–30% in paddy rice and suppression in weed growth caused by *Azolla*. Furthermore, Marrez et al. (2014) reported that *Spirulina* is a rich source of potassium (K) and contains considerable amounts of Ca, Cu, Fe, Mg, Mn, P, and Zn; hence, it increases uptake and accumulation of these elements in plants. This in turn explains the significant increase of vegetative growth and yield and its components (Enan et al. 2016).

**Effect of algal treatments on nematode control**

Data presented in Table 5 summarize significant differences in all nematode parameters at different treatments compared with control. Control (untreated) plants exhibited a high gall formation in the two seasons; on the other hand, Rugby nematicide and *Amphora* (spray + drench) treatments caused the lowest gall numbers compared to the control plants. Differences in numbers of galls in the first season and egg masses in

both seasons between Rugby and *Amphora* (spray + drench) were insignificant. Applications of various algal treatments suppressed *M. incognita* population on cucumber. The effectiveness of the treatments was expressed by reducing the number of galls on the root system and the final nematode number in soil (final population). These results may be due to the huge amount of secondary metabolites and bionematicides, which are produced by algae (*Amphora* and *Spirulina*) such as phenols, antioxidant, polyphenols, and flavonoids. A similar conclusion was mentioned by Gervick et al. (2001) that the blue-green alga, such as *S. platensis*, produce components that had a nematicidal effect. *Amphora* may produce high amounts of these products than *Spirulina*, which explains its superiority in nematode resistance. Similarly, Khan et al. (1997) reported that the efficacy of culture filtrates of the cyanobacterium against egg hatching and mortality of *M. incognita* was dependent on its concentration and period of exposure. The high mortality percentages recorded for nematode juveniles exposed to the algal culture filtrates may be due to the presence of some phenolic compounds and mineral salts that facilitate and accelerate the rate of penetration of algal byproducts through a snail's skin, hence increasing their harmful effects (Mahmoud 2001). Moreover, Radwan et al. (2012) concluded that microbial products are one of the components in integrated root-knot nematode management on tomatoes. In this respect, Youssef and Ali (1998) and Pushparaj et al.

**Table 5** Effect of algal treatments of cucumber Hesham hybrid compared with control and Rugby nematicide on the galls, egg masses, eggs, rate of buildup, and number of larvae in infested soil with root-knot nematodes in 2016 and 2017 seasons

Treatments	2016						2017					
	No. of larvae in 100 g Soil	No. of galls/ root system	No. of egg mass/root system	No. of egg/ egg mass	No. of eggs/ root system	Reproduction factor (RF)	No. of larvae in 100 g Soil	No. of galls/ root system	No. of egg mass/root system	No. of eggs/ egg mass	No. of eggs/ egg system	Reproduction factor (RF)
Control	14,622	409	112	39	4368	1.035	14,516	345	198	40	7920	1.05
Rugby 10G	3890	48	21	6	126	0.275	4294	55	26	7	182	0.31
<i>Spirulina</i> spray	13,751	301	98	32	3136	0.97	13,909	291	105	37	3885	1.01
<i>Spirulina</i> drench	7396	128	62	28	1736	0.52	8277	150	72	27	1944	0.60
<i>Amphora</i> spray	13,800	254	87	26	2262	0.98	13,472	277	98	33	3234	0.98
<i>Amphora</i> drench	5373	149	53	22	1166	0.38	7178	112	43	19	988	0.52
<i>Spirulina</i> spray + drench	8400	126	56	19	1064	0.59	8184	132	52	30	1290	0.59
<i>Amphora</i> spray + drench	5940	77	33	9	297	0.42	6127	98	29	8	232	0.45
LSD at t <sub>0.05</sub>	1682	33	20	5	78	0.14	1509.6	26	17	4	89	0.15

**Table 6** Means of galls, egg mass, and egg number reductions compared with control at different treatments in 2016 and 2017 seasons

Treatments	2016			2017		
	Gall reduction%	Egg masses reduction%	Eggs reduction%	Gall reduction%	Egg masses reduction%	Eggs reduction%
Rugby 10G	88.26	81.25	97.12	84.05	86.86	97.70
Spirulina spray	26.40	12.50	28.20	15.65	46.97	50.95
Spirulina drench	68.70	44.64	60.26	56.52	63.64	75.45
Amphora spray	37.89	22.32	48.21	19.70	50.51	59.16
Amphora drench	63.56	52.68	73.30	67.50	78.28	87.52
Spirulina spray + drench	69.19	50.00	75.64	61.74	73.74	83.70
Amphora spray + drench	81.17	70.53	93.20	71.59	85.35	97.07

(2000) reported that the blue-green algae reduced the number of galls and egg masses of the root-knot nematode-infected cowpea and tomato plants, respectively.

Variation in the number of eggs between the two treatments, Rugby and *Amphora* (spray + drench), in the first season was significant. Egg production fitness was always correlated with untreated plants (control), while this ability decumbently degenerated with the Rugby and *Amphora* (spray + drench).

With respect to algal treatments in Table 5, the numbers of galls, egg masses, and eggs in addition to RF values, obtained through the two seasons, greatly differed and the differences were statistically significant than that of the control. In contrast, control plants supported the highest rates of nematode reproduction (RF), while the Rugby treatment supported the lowest rate. Reproductive values of *Meloidogyne* spp. greatly varied among all the treatments and control. The nematodes reproduced efficiently on the control plants, whereas reproductions on the treated plants were not efficient.

Control plants exhibited a high degree of nematode infection, egg-mass formation, and egg deposition; this indicated that the final nematode population (14,622 and 14,516) increased over the initial nematode populations (13,752 and 14,124). Reproductive values of nematodes were significantly different between control plants and all treatments. RF values of the Rugby were 0.31 and 0.275, while they were 1.05 and 1.035 on the control in the two seasons, respectively. Some of the tested algal treatments possessed nematicidal effects that may be released in the soil. In addition, these treatments of algae may produce compounds, such as asetamide, hexamethyl, methoxyphenyl, phenol, and others, which are normally toxic to the root-knot nematode. All the tested products significantly reduced the numbers of juveniles in soil and galls, females, and egg masses on roots. Also, these products suppressed egg hatching.

Data presented in Table 6 show percentages of reduction in gall, egg mass, and egg numbers of the six algal

treatments and chemical nematicide (Rugby) compared with the control. Data were confirmed at all parameters, whereas percentages of reduction in gall numbers in the first and the second seasons for Rugby nematicide were 88.26 and 84.05%, recording the highest gall reduction in both seasons, respectively. These results are near to those of *Amphora* (spray+ drench) treatments, which were 81.17 and 71.59%. Concerning *Amphora* (spray + drench) treatment, percentages of reduction of egg masses were 70.53 and 85.35 and egg numbers were 93.20 and 97.07%, while the percentages of reduction in Rugby nematicide were 81.25 and 86.86 for egg masses and 97.12 and 97.07 for eggs in both seasons, respectively.

These results confirm that the bio-management could be an alternative strategy to the chemical approach. These results agree with that of Sharma and Gaur (2008) who studied the effects of extracts and exudates of five cyanobacteria, autotrophic blue-green algae, on the hatching of *M. incognita*.

## Conclusion

This study demonstrated that the most tested algal treatments significantly reduced root-knot nematode numbers, specially *Amphora* (spray + soil drench) treatment that showed the highest suppression effects on the root-knot nematode. Furthermore, this treatment enhanced plant growth than the other applications. The foliar and soil drench application should be used.

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