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The Promotive Effect of Different Concentrations of Marine Algae on Spinach Plants (*Spinacia oleracea* L.)

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A FIELD experiments were carried out during the winter seasons 2013/2014, 2014/2015 at the Experimental Station Farm of the Faculty of Agriculture, Alexandria University, at Abeis, Alex, governorate, A.R.E. to study the promotive effect of different concentrations of marine algae as biostimulantfertilization and mineral fertilization on growth and yield of Spinach, three of algae were applied in four rates (5%, 10%, 20% and 40%) and sprayed alone or in combination with others and recommended NPK as a control were applied . Plants were harvested after 50 days from planting and the characters were measured such as (plant height (cm), fresh and dry weights of leaves (%), total yield, total leaf's chlorophyll content, mineral contents of leaves and the accumulation of nitrate). All the tested treatments increased plant yield and quality in varying degrees. *Ulva lactuca* + Bread yeast 5% was the best treatment to increase the yield of Spinach plants in addition was recorded the lowest amount of nitrate in Spinach leaves. We can evaluate the effect of seaweed extract on nitrogen metabolism in root and leaves of Spinach plants cultivated in medium culture deficiency or richened of mineral elements.

Keywords: Algae, Biostimulant, Nitrate content, Spinach.

Spinach (*Spinacia oleracea* L.) is one of the vegetables having inherently high nitrate concentration and petioles have several fold higher concentration than leaf blades. High nitrate concentration is found in leafy vegetables practically under intensive nitrogen fertilization. Nitrogen is usually the most abundant element in plants. Intensive application of fertilizer caused an excess of nitrogen for crops. There is strong evidence that some of the excess nitrogen taken up by the plant is not converted to protein but remains as non-protein nitrogen. This is not only inefficient use of nitrogen by the plant but leads to inefficient use by the animal and the risk of ill effects on human beings eating the plant. Mordogan (2000) pointed out that spinach yield increased with the increases in N level, furthermore, increased the soluble N compounds in the plant. Paradiso *et al*. (2001) stated that leaf nitrate content increased in the last two weeks before harvest and was highest with the maximum N rate. Karaman *et al*. (2000) stated that the nitrate content in the vegetables were increased with increasing regional N use especially NO_3-N, accumulation nitrate of vegetables increased much more than NH_4- N, Wang *et al*. (2003).

Nitrate is a naturally occurring form of nitrogen and it is an integral part of the nitrogen cycle in the environment. Nitrate is formed from fertilizers, decaying plants, manure and other organic residues. It is found in the air, soil, water and food (particularly in vegetables) and it is produced naturally within the human body. Walker (1990) and Speijers (2003). It is also used as a food additive, mainly as a preservative and antimicrobial agent. Speijers (1996 and 2003) It is used in foods such as cheese and cheese products, raw and processed meats, edible casings, processed fish, fish products, spirits and liqueurs. Due to the increased use of synthetic nitrogen fertilizers and livestock manure in intensive agriculture vegetables and drinking water may contain higher concentrations of nitrate than in the past.

Soil fertility is diminishing gradually due to soil erosions, loss of nutrient, accumulation of salts and other toxic elements, water logging and un-balanced nutrient compensation. Organic wastes and bio-fertilizers are the alternate sources to meet the nutrient requirement of crops and to bridge the future gaps. Farming regions that emphasizing heavy chemical application led to adverse environmental, agricultural and health consequences. Many efforts are being exercised to combat the adverse consequences of chemical farming (Faheed and Abd-El-Fattah, 2008).

Algae and active bread yeast extract have a positive effect on fruit setting, yield and fruit quality (Hegab *et al.,* 2005 and Abd El-Motty *et al.,* 2010). Algae extract as a new bio-fertilizer containing N, P, K, Ca, Mg, and S as well as Zn, Fe, Mn, Cu, Mo and Co, some growth regulators, polyamines, natural enzymes, carbohydrates, proteins and vitamins applied to improve nutritional status, vegetative growth, yield and fruit quality in different orchard as well as vineyards (Abd El-Migeed *et al.,* 2004, Abd El-Moniem & Abd-Allah, 2008, Spinelli *et al.,* 2009 and Abbas, 2013).

Nowadays, braed yeast (*Saccaromycescervisiae)* as a natural bio-stimulant appeared to induce an important influence on growth and yield of many crops, since it has various basic function, i.e. CO_2 production as well as formation of alcohol, acids and esters (Martinez-Anoya *et al.,* 1990). Active dry yeast is a natural safety bio-fertilizer, it is considered as a natural source of cytokinins that stimulates cell division and enlargement as well as the synthesis of protein and nucleic acid (Mady, 2009). Spraying Valencia orange trees with active bread yeast either once on March or August or twice at both dates was favorable in improving growth, fruit set, number of fruits and yield as well as fruit weight and volume (Hegab *et al.,* 2005). Ahmed and Ragab (2002) supported the beneficial effect of yeast on nutritional status of Picual olive trees.

In addition, bio-organic is very safe for human, animal and environment to get lower pollution and reduce soil salinity via decrease mineral usage fertilization as well as saving fertilization cost. The present study aimed to assess the effect of these biostimulants on yield and growth of *Spinaciaoleracea.*

Materials and Methods

The seaweeds used in this study were *Ulva lactuca* (class: *Chlorophyceae*) and *Jainarubens* (class: *Rhodophyceae*). They were collected from the coastal area of Abu-Kir, Alexandria-north of Egypt. The samples were washed thoroughly with seawater to remove all the unwanted impurities, adhering sand particles and epiphytes. The samples were placed separately in polythene bags, kept inside an ice box and transported to the laboratory. Samples were washed thoroughly using tap water to remove surface salt and spread on blotting paper to remove excess water. Samples were air-dried (25°C) during 2-4 days followed by dry at 60°C for 12 hours.

Dried seaweeds were hand crushed and powdered with coffee-grinder. Algae were heated with sterile distilled water in a ratio 1:100 (w/v) at 60°C for 45 minutes, and then the extracts were filtrated through a filter paper and stored at 4°C for further experimental studies. The filtrate were 10^{-2}g of dried seaweeds per milliliter (g DSW ml^{-1}) extracts. The filtrate were considered as 100% seaweed extracts and different concentrations of seaweed liquid extract (SLE) were prepared by diluting of these extracts with distilled water. Such concentrations 5%, 10%, 20% and 40% were used in our experiment (Bhasle *et al*., 1975 and Anisimov *et al*., 2013). The major components of the used algae are shown in Table 1.

TABLE 1. Physic-chemical composition and mineral contents of *Ulva lactuca*and *Jainarubens*extracts .

Parameters	***Ulva lactuca***	***Jainarubens***
Color	Green	Red
pH	6.63	6.68
Ash%	37.15	42.40
Moisture content%	11.76	9.95
Protein%	15.65	6.396
Carbohydrate%	29.60	6.146
Lipid%	0.82	1.014
Nitrogen(mg/kg)	474	523
Phosphorus(mg/kg)	93	288.53
Potassium (mg/kg)	1570	1454
Sodium (mg/kg)	2065	4400
Manganese (mg/kg)	37.7	149.44
Calcium (mg/kg)	1892	3776.1
Magnesium (mg/kg)	3334	3952.2
Iron (mg/kg)	228.6	45
Zinc (mg/kg)	42.16	123
Copper (mg/kg)	5.7	4.1
Cobalt (mg/kg)	0.6	1.0
Cadmium (mg/kg)	0.27	0.157
Chromium (mg/kg)	0.87	0.051
Nickel (mg/kg)	5.51	6.0
Lead (mg/kg)	1.0	6.1

Bread yeast was applied in four rates (5%, 10%, 20% and 40%) and sprayed alone or in combination with seaweed treatments. The chemical composition of bread yeast are shown in Table 2.

TABLE 2. Chemical composition of *Saccharomyces cerevisiae* (bread yeast) (after, Nagodawithana, 1991).

Protein	**47%**	**Nucleic acids**	**8%**
Carbohydrates	33%	Lipids	4%
Minerals	8%		
Approximate composition of vitamins (mg/kg)			
Thiamine	6-100	Biotin	1.3
Riboflavin	35-50	Cholin	4000
Niacin	300-500	Folic acid	5-13
Pyridoxine HCl	28	Vit-B_{12}	0.001
Pantothenate	70		
Approximate composition of minerals (mg/kg)			
Na	0.12	Cu	8.00
Ca	0.75	Se	0.10
Fe	0.02	Mn	0.02
Mg	1.65	Cr	2.20
K	21.00	Ni	3.00
P	13.50	Va	0.04
S	3.90	Mo	0.04
Zn	0.17	Sn	3.00
Si	0.03	Li	0.17

In the field this study was conducted during the two winter seasons of 2013/2014, 2014/2015 at the Experimental Station Farm of the Faculty of Agriculture, Alexandria University, at Abeis, Alex, governorate, A.R.E.

Some physical and chemical characteristics of the experimental soil atFaculty of Agriculture, Alexandria University are shown in Table 3.

Three of algae were applied in four rates (5%, 10%, 20% and 40%) as biostimulant and NPK fertilization (control) were used in the present study. Commercial Spinach cultivar (Baldy) was used in this experiment .Seeds of spinach were sown on 25th of November, in both seasons. Three replicates per treatment (each replicate contained 25 treatments). NPK fertilization was carried out according to the recommendations for commercial production of spinach plant as outlined by Ministry of Agriculture and Land Reclamation-Arab Republic of Egypt. The NPK treatment consisted in ammonium nitrate (33% N) at the rate of 5 kg 100 m^{-2}, calcium super phosphate at the rate of 6, 25 kg 100 m^{-2}, potassium sulphate (48% K_2O) at the rate of 1.25 kg 100 m^{-2}.

TABLE 3. Soil's physical and chemical properties of the experimental sites in the tow winter seasons of 2013/2014 and 2014/2015 before cultivation.

Properties	Winter 2013/2014	Winter 2014/2015
Physical properties		
Sand %	32.8	31.9
Silt %	25. 2	25.8
Clay %	42. 0	42.3
Soil texture	Clay loam	Clay loam
Chemical properties		
pH	8.01	7.95
E.C. ($dS.m^{-1}$)	3.01	3.02
Soluble cations (meq/ l)		
Ca^{++}	2.41	2.30
Mg^{++}	2.00	1.90
Na^{+}	2.73	2.65
K^{+}	0.46	0.36
Soluble anions (meq/ l)		
HCO_3^{-}	2.20	2.20
Cl^{-}	2.00	1.90
SO_4^{--}	3.40	3.11
Total N %	0.196	0.15
Available P ppm)	0.337	0.286

First application of biostimulant was done after two weeks from planting the second and the third applications were added every 15 days from the first dose.

Plants were harvested after 50 days from planting, ten plants were randomly chosen from the different treatments and their growth was measured based on plant height (cm), fresh and dry weight of leaves , total yield, total leaf's chlorophyll content (mg 100g f.w.) according to the method described by Yadava (1986), mineral contents of leaves according to A.O.A.C. (1990) and the content of nitrate were determined at High Institute of public Health, Environmental Health Research, Analysis and Studies unit, Alexandria University.

Treatment means were separated and compared using the L.S.D test at 0.05 level of significance according to Snedecor and Cochran (1980).The statistical analysis was performed using CoStat software package for Windows.

Results and Discussions

Vegetative growth

With regard to the data presented in Table 4 *Jainarubens + Ulva lactuca* 40% had significantly higher value of plant height in two seasons. On the other hand, *Jainarubens* 5% resulted in shorter plant height. From the results observed in Table 4 it is clear the high number of leaves per plant in both seasons was significantly by *Jainarubens* 20%, the minimum number of leaves was 5.66 and 6.33 leaf/ plant produced by *Ulva lactuca* 5% treatment during the first and second seasons, respectively.

From the above results, it could be concluded that *Jainarubens*and *Ulva lactuca*as a bioorganic fertilizers for growing spinach plants. Seaweed products contain growth regulators (auxins, cytokinins and gibberellins), amino acids and mineral nutrients, that accordingly affect plant growth and division positively effect on increasing the plants height. This may be due to the improving effect of such treatments on nutritional status of the plants, which reflected on increasing plants height. The previous results are agreed with those obtained by Abbas (2013) and Al-Shakankery *et al.* (2014) who reported that the growth and yield of maize plants increased when used marine algae as biofertilizers.

TABLE 4. Plant height (cm) and number of leaves/plant of Spinach plants affected by different concentrations of marine algae and bread yeast as biostimulant, during the two winter seasons of 2013/2014 and 2014/2015.

Treatments	Plant height(cm)		N. of leaves /plant	
	Winter 2013/2014	Winter 2014/2015	Winter 2013/2014	Winter 2014/2015
Bread yeast 5%	38.66 F-J	39.33 F-J	6.00 BC	6.66 BC
Bread yeast 10%	39.66 E-I	40.33 E-I	6.00 BC	6.66 BC
Bread yeast 20%	36.66 G-J	37.33 G-J	7.00 A-C	7.66 A-C
Bread yeast 40%	39.33 E-J	40.00 E-I	6.33 A-C	7.00 A-C
Jainarubens 5%	33.00 J	33.66 J	6.66 A-C	7.33 A-C
Jainarubens 10%	41.00 D-H	41.66 D-H	7.66 AB	7.66 A-C
Jainarubens 20%	42.66 C-G	43.33 C-G	8.00 A	8.66 A
Jainarubens 40%	45.33 B-E	46.00 B-E	6.00 BC	6.66 BC
Ulva lactuca 5%	34.33 IJ	35.00 IJ	5.66 C	6.33 C
Ulva lactuca 10%	33.33 IJ	34.00 IJ	6.00 BC	6.66 BC
Ulva lactuca 20%	38.00 F-J	38.66 F-J	7.33 A-C	8.00 A-C
Ulva lactuca 40%	41.33 D-H	42.00 D-H	6.33 A-C	7.00 A-C
Jainarubens+ Bread yeast 5%	39.66 E-I	40.33 E-I	7.66 AB	8.33 AB
Jainarubens+ Bread yeast 10%	39.66 E-I	40.33 E-I	7.66 AB	7.66 A-C
Jainarubens+ Bread yeast 20%	41.33 D-H	42.00 D-H	6.33 A-C	7.00 A-C
Jainarubens+ Bread yeast 40%	36.00 H-J	36.66 H-J	7.00 A-C	7.66 A-C
Ulva lactuca + Bread yeast 5%	51.33 AB	52.00 AB	7.00 A-C	7.66 A-C
Ulva lactuca + Bread yeast10%	41.00 D-H	41.66 D-H	6.00 BC	6.66 BC
Ulva lactuca + Bread yeast20%	49.00 A-C	49.66 A-C	6.00 BC	6.66 BC
Ulva lactuca + Bread yeast40%	41.00 D-H	41.66 D-H	6.66 A-C	7.33 A-C
Jainarubens+ *Ulva lactuca* 5%	43.33 C-F	44.00 C-F	7.00 A-C	7.66 A-C
Jainarubens+ *Ulva lactuca*40%	43.00 C-G	43.66 C-G	7.33 A-C	8.00 A-C
Jainarubens+ *Ulva lactuca*20%	46.33 A-D	47.00 A-D	6.00 BC	6.66 BC
Jainarubens+ *Ulva lactuca*40%	52.33 A	53.00 A	7.33 A-C	8.66 A
RECOMMENTED N P K (CONTROL)	36.66 G-J	37.33 G-J	6.33 A-C	7.66 A-C

*Values followed by the same alphabetical letter(s) in common, within a particular group of means in each character, do not significantly differ, using Revised L.S.D test at 0.05 level of probability.

Total Yield

The results clarified the differences due to the studied factor on the total yield shown in Table 5. Results indicated that the total yield was significantly affected by different concentration of marine algae and NPK fertilization in both seasons, whereas the highest total yield was obtained using (*Jainarubens + Ulva lactuca* 40%) 2784 and 2829 g/m^2 in both seasons, respectively. However, the lowest total yield was obtained when *Jainarubens* 10%) treatment was applied, in both seasons. From the previous results, it is clear that yield was increased and reached the maximum by using *Jainarubens + Ulva lactuca* 40%. The increment in yield may explained due to the positive effect of alga *Jainarubens*40% in combination with *Ulva lactuca* 40% as bio-organic. The obtained results may confirm the previous work done by Abd El-Motty *et al.* (2010) who reported that yield of mango was increased by algae extracts.

TABLE 5. Total yield (g/m²) of spinach as affected by different concentrations of marine algae and bread yeast as biostimulant, during the two winter seasons of 2013/2014 and 2014/2015.

Treatments	Total yield (g/m²)	
	Winter 2013/2014	Winter 2014/2015
Bread yeast 5%	2333 A-D	2392 A-D
Bread yeast 10%	1888 D-F	1948 D-F
Bread yeast 20%	2059 B-F	2133 B-F
Bread yeast 40%	1984 C-F	2029 C-F
Jainarubens 5%	1918 D-F	1977 D-F
Jainarubens 10%	1570 EF	1626 F
Jainarubens 20%	1925 D-F	1984 D-F
Jainarubens 40%	2177 A-F	2236 A-F
Ulva lactuca 5%	2355 A-D	2414 A-D
Ulva lactuca 10%	2266 A-D	2325 A-D
Ulva lactuca 20%	1795 D-F	1854 D-F
Ulva lactuca 40%	1881 D-F	1953 D-F
Jainarubens+ Bread yeast 5%	2007 B-F	2066 B-F
Jainarubens+ Bread yeast 10%	2392 A-D	2451 A-D
Jainarubens+ Bread yeast 20%	1903 D-F	1977 D-F
Jainarubens+ Bread yeast 40%	1570 EF	1692 EF
Ulva lactuca + Bread yeast 5%	2148 A-F	2208 A-F
Ulva lactuca + Bread yeast 10%	2296 A-D	2355 A-D
Ulva lactuca + Bread yeast 20%	2592 A-C	2644 A-C
Ulva lactuca + Bread yeast 40%	2622 AB	2681 AB
Jainarubens+ *Ulva lactuca* 5%	2362 A-D	2422 A-D
Jainarubens+ *Ulva lactuca* 10%	2296 A-D	2355 A-D
Jainarubens+ *Ulva lactuca* 20%	2207 A-D	2264 A-E
Jainarubens+ *Ulva lactuca* 40%	2784 A	2829 A
RECOMMENTED N P K (CONTROL)	2044 B-F	2103 B-F

*Values followed by the same alphabetical letter(s) in common, within a particular group of means in each column, do not significantly differ, using Revised L.S.D test at 0.05 level of probability.

Leaves dry weight

Data in Table 6 showed that there was significant effect of all different concentration of marine algae on leaves dry weight of spinach plants in both seasons compared with NPK fertilization. The best result was obtained with using (*Jainarubens* 10%), in both seasons. Similar results were achieved by Shaaban *et al.* (2010), found to have beneficial effects, such as increased crop yield, increased uptake of nutrient and resistance to stress conditions, when such algae applied to soil. Optimization of the nutrient fixing and nutrient toxicity, enabling every element to play its role a harmony with other nutrients, which leads, in turn, to the best dry matter accumulation.

Chemical composition

Total chlorophyll content

Data obtained in Table 6 showed that the highest value was obtained with *Jainarubens* + *Ulva lactuca* 20% in two seasons 48.33. On the other hand the lowest of the chlorophyll content with N P K (CONTROL). The important role of biostimuants on enhancing the leaf chlorophyll might be attributed to their action on increasing the availability of water and minerals the high chlorophyll content might have resulted from enhanced plant growth (Mady, 2009). *Sargassum*extract was effective in enhancing chlorophyll synthesis in *Zea mays* and *Phaseolus mungo* (Homme *et al*., 1992). Applying yeast to field bean plant increased content of chlorophyll a, b and total chlorophyll (Abbas, 2013).

Mineral content

Results of leaf mineral content as influenced by various applied treatments were presented in Tables 7 and 8 showed that all different concentration of marine algae on leaf's nitrogen content were significant compared with control treatment (NPK).Treatments with the same letter in common are not statistically differentvalue was obtained but the highest number was (*Jainarubens* 40%) 2.23 and 2.27, respectively in two seasons followed by (*Ulva lactuca* + Bread yeast 5%) 2.25 and 2.25 respectively in two seasons . However, the lowest leaf's nitrogen was obtained by(NPK). Data obtained in Table 7 showed that there was significant effects of marine algae treatments on leaf's nitrate accumulation % of Spanish plants. The highest accumulation of nitrate was (*Ulva lactuca* 40%) in two seasons. However, the lowest leaf's nitrate accumulation % was obtained by (*Jainarubens* + Bread yeast 5 % and *Ulva lactuca* + Bread yeast 5%), in both seasons.

From the above mentioned results it could be noticed that nitrogen content in the leaves was significantly affected by treatments in both studied seasons. The concentration of bio-organic (*Ulva lactuca* + Bread yeast 5%) improved nitrogen content in the leaves. These results are in agreement with those reported by Hegab *et al.* (2005) and El-Motty *et al.* (2010).

TABLE 6. Leaf's chlorophyll content (mg/100 g f.w) and leaf's dry matter content (%) of spinach plants as affected by different concentrations of marine algae and bread yeast as biostimulant during the two winter seasons of 2013/2014 and 2014/2015.

Treatments	Leaf's chlorophyll content (mg/100 g f.w)		Leaf's dry matter content (%)	
	Winter 2013/2014	Winter 2014/2015	Winter 2013/2014	Winter 2014/2015
Bread yeast 5%	39.00 AB	40.00 AB	22.33 J	22.66 K
Bread yeast 10%	38.66 AB	39.66 AB	21.71 L	21.65 N
Bread yeast 20%	39.00 AB	40.00 AB	23.44 G	23.42 G
Bread yeast 40%	39.33 AB	39.50 AB	24.00 E	25.00 A
Jainarubens 5%	41.66 AB	42.66 AB	24.00 E	24.22 D
Jainarubens 10%	40.33 AB	41.33 AB	26.00 A	25.00 A
Jainarubens 20%	41.33 AB	42.33 AB	24.66 C	23.66 F
Jainarubens 40%	39.00 AB	40.00 AB	22.00 K	22.22 L
Ulva lactuca 5%	39.00 AB	40.00 AB	24.33 D	23.66 F
Ulva lactuca 10%	39.66 AB	40.66 AB	25.00 B	24.66 B
Ulva lactuca 20%	41.00 AB	42.00 AB	24.33 D	24.45 C
Ulva lactuca 40%	40.33 AB	41.33 AB	23.33 H	23.21 I
Jainarubens+ Bread yeast 5%	39.33 AB	40.33 AB	23.00 I	23.33 H
Jainarubens+ Bread yeast 10%	38.00 AB	39.00 AB	23.66 F	22.00 M
Jainarubens+ Bread yeast 20%	35.66 AB	36.66 AB	23.66 F	23.33 H
Jainarubens+ Bread yeast 40%	46.33 AB	47.00 AB	21.00 N	21.33 O
Ulva lactuca + Bread yeast 5%	43.33 AB	44.33 AB	23.33 H	23.66 F
Ulva lactuca + Bread yeast 10%	44.33 AB	45.00 AB	23.00 I	23.00 J
Ulva lactuca + Bread yeast 20%	42.00 AB	43.00 AB	24.33 D	23.66 F
Ulva lactuca + Bread yeast 40%	41.66 AB	42.66 AB	23.66 F	23.66 F
Jainarubens+ *Ulva lactuca* 5%	41.00 AB	42.00 AB	24.33 D	23.00 F
Jainarubens+ *Ulva lactuca* 10%	41.66 AB	42.66 AB	23.33 H	23.65 F
Jainarubens+ *Ulva lactuca* 20%	48.33 A	48.33 A	23.66 F	23.66 F
Jainarubens+ *Ulva lactuca* 40%	46.33 AB	47.00 AB	23.33 H	23.92 E
RECOMMENTED N P K (CONTROL)	33.33 B	32.66 B	21.33 M	21.67 N

*Values followed by the same alphabetical letter(s) in common, within a particular group of means in each character, do not significantly differ, using Revised L.S.D test at 0.05 level of probability

With respect to nitrate accumulation, the application of seaweed extract increased the amount of nitrate in plant leaves. *Jainarubens* + Bread yeast 5 % and *Ulva lactuca* + Bread yeast 5% extracts were found more effective than *Jainarubens* + *Ulva lactuca* 40%. When seaweed biostimulant (*Jainarubens* + Bread yeast 5 % and *Ulva lactuca* + Bread yeast 5%) extract was incorporated in the medium culture, we noted an important decrease of nitrate accumulation. However, the incorporation of *Jainarubens* + *Ulva lactuca* 40% or *Ulva lactuca* 40% extracts in medium culture showed a remarkable increased of nitrate.

This decrease in nitrate accumulation might be due to the consequence of the presence of the high concentration of mineral elements that can inhibit the enzymatic activity. This finding was also reported by Esteban *et al*., 2004 as the most important enzyme activities within nitrogen metabolism, such as nitrate

reductase and nitrite reductase, which were affected negatively by the highest NH_4NO_3 rate applied to *Phaseolus vulgaris* plant at different levels.

TABLE 7. Leaf's nitrogen and nitrate content of spnach plants as affected by different concentrations of marine algae and bread yeast as biostimulant during the two winter seasons of 2013/2014 and 2014/2015.

Treatments	Leaf's nitrogen content (%)		Leaf's nitrate content (%)	
	Winter 2013/2014	Winter 2014/2015	Winter 2013/2014	Winter 2014/2015
Bread yeast 5%	2.07 AB	2.09 A	0.193 D-F	0.21 I
Bread yeast 10%	1.93 AB	1.94 AB	0.19 EF	0.22 G-I
Bread yeast 20%	1.95 AB	1.96 AB	0.22 D	0.23 G
Bread yeast 40%	2.03 AB	2.04 AB	0.36 A	0.34 B
Jainarubens 5%	2.06 AB	2.07 A	0.135 G	0.19 JK
Jainarubens 10%	2.06 AB	2.06 A	0.193 D-F	0.216 G-I
Jainarubens 20%	2.21 A	2.21 A	0.193 D-F	0.20 J
Jainarubens 40%	2.23 A	2.27 A	0.22 D	0.243 F
Ulva lactuca 5%	2.11 A	2.11 A	0.153 FG	0.213 HI
Ulva lactuca 10%	2.03 AB	2.04 AB	0.136 G	0.123 L
Ulva lactuca 20%	2.00 AB	2.00 AB	0.358 A	0.336 BC
Ulva lactuca 40%	1.99 AB	1.99 AB	0.358 A	0.373 A
Jainarubens+ Bread yeast 5%	1.99 AB	2.00 AB	0.071 H	0.076 M
Jainarubens+ Bread yeast 10%	2.10 A	2.10 A	0.165 FG	0.18 K
Jainarubens+ Bread yeast 20%	2.13 A	2.13 A	0.18 EF	0.19 JK
Jainarubens+ Bread yeast 40%	2.12 A	2.12 A	0.24 CD	0.246 F
Ulva lactuca + Bread yeast 5%	2.25 A	2.25 A	0.071 H	0.076 M
Ulva lactuca + Bread yeast 10%	2.12 A	2.12 A	0.18 EF	0.186 K
Ulva lactuca + Bread yeast 20%	2.22 A	2.22 A	0.22 D	0.22 G-I
Ulva lactuca + Bread yeast 40%	2.16 A	2.16 A	0.22 D	0.226 GH
Jainarubens+ *Ulva lactuca* 5%	2.22 A	2.22 A	0.18 EF	0.18 K
Jainarubens+ *Ulva lactuca* 10%	2.10 A	2.10 A	0.22 D	0.22 G-I
Jainarubens+ *Ulva lactuca* 20%	2.23 A	2.23 A	0.27 C	0.26 E
Jainarubens+ *Ulva lactuca* 40%	2.23 A	2.23 A	0.311 B	0.325 C
RECOMMENTED N P K (CONTROL)	1.77 B	1.76 B	0.311 B	0.31 D

*Values followed by the same alphabetical letter(s) in common, within a particular group of means in each column, do not significantly differ, using Revised L.S.D test at 0.05 level of probability.

The data in Table 8 clearly indicated that NPK fertilization (control treatment) had higher leaf's P content % than the all different concentration of marine algae during both seasons. For example, in 2013 season, NPK (control) contained 0.55 and 0.56%, respectively also, the higher leaf's K content % was reported in *Ulva lactuca* + Bread yeast 10% followed by *Ulva lactuca* + Bread yeast 5% during both seasons. These results are in agreement with those reported by Stino *et al*. (2009). This organic fertilization also facilitate the growth of plants by improving the uptake of nutrients and production of some phytohormones. The objective of this research was to provide the potential advantages of biostimulants as organic matter on growth and development of plants.

TABLE 8. Leaf's potassium and phosphorus content (%) of spnach plants as affected by different concentrations of marine algae and bread yeast as biostimulant, during the two winter seasons of 2013/2014 and 2014/2015.

Treatments	Leaf's phosphorus content (%)		Leaf's potassium content (%)	
	Winter 2013/2014	Winter 2014/2015	Winter 2013/2014	Winter 2014/2015
Bread yeast 5%	0.33B-F	0.34 C-F	2.11 A-C	2.12 C-G
Bread yeast 10%	0.35 B-E	0.36 B-E	2.03 BC	2.10 D-G
Bread yeast 20%	0.38 BC	0.39 BC	2.08 A-C	2.13 C-G
Bread yeast 40%	0.37 B-D	0.363 B-D	2.14 A-C	2.22 A-E
Jainarubens 5%	0.32C-G	0.33 C-G	2.08 A-C	2.12 C-G
Jainarubens 10%	0.30 D-G	0.31 D-G	2016 A-C	2.16 B-F
Jainarubens 20%	0.286 E-G	0.293 E-G	2.24 A-C	2.19 B-F
Jainarubens 40%	0.26 G	0.26 G	2.16 A-C	2.13 C-G
Ulva lactuca 5%	0.38 BC	0.38 BC	1.98 BC	1.97 E-G
Ulva lactuca 10%	0.41 B	0.42 B	1.95 C	1.93 G
Ulva lactuca 20%	0.38 BC	0.39 BC	1.97 BC	1.95 FG
Ulva lactuca 40%	0.38 BC	0.39 BC	1.98 BC	1.97 E-G
Jainarubens+ Bread yeast 5%	0.28 FG	0.28 FG	2.14 A-C	2.21 A-E
Jainarubens+ Bread yeast 10%	0.29 D-G	0.306 D-G	2.18 A-C	2.20 B-F
Jainarubens+ Bread yeast 20%	0.34B-F	0.35 C-F	2.40 A	2.41 AB
Jainarubens+ Bread yeast 40%	0.33B-F	0.34 C-F	2.24 A-C	2.38 A-C
Ulva lactuca + Bread yeast 5%	0.29 D-G	0.303 D-G	2.31 AB	2.29 A-D
Ulva lactuca + Bread yeast 10%	0.28 FG	0.293 E-G	2.39 A	2.46 A
Ulva lactuca + Bread yeast 20%	0.283 E-G	0.29 E-G	22.26 A-C	2.32 A-D
Ulva lactuca + Bread yeast 40%	2.30B-F	0.29 E-G	2.30 AB	2.34 A-D
Jainarubens+ *Ulva lactuca* 5%	0.34B-F	0.353 C-F	2.19 A-C	2.25 A-D
Jainarubens+ *Ulva lactuca* 10%	0.36 B-D	0.366 B-D	2.07 A-C	2.14 C-G
Jainarubens+ *Ulva lactuca* 20%	0.34B-F	0.35 C-F	2.19 A-C	2.15 B-G
Jainarubens+ *Ulva lactuca* 40%	0.39 BC	0.39 BC	2.18 A-C	2.24 A-D
RECOMMENTED N P K (CONTROL)	0.55 A	0.56 A	2.30 AB	2.33 A-D

*Values followed by the same alphabetical letter(s) in common, within a particular group of means in each column, do not significantly differ, using Revised L.S.D test at 0.05 level of probability

Conclusion

The present study is an important step towards the utilization of the extracts of the seaweeds to improve yield and growth of spinach plants. The presence of nitrogen, phosphorus, potassium and some trace elements in seaweeds make an excellent choice as biostimulant. All the tested treatments increased plant yield and quality in varying degrees. *Ulva lactuca* + Bread yeast 5% was the best treatment to increase the yield of Spinach plants in addition was recorded the lowest amount of nitrate in Spinach leaves. The nitrate accumulation was not dependent on the richness of seaweed extract in mineral element particularly nitrogen.

As perspective, we believe that it would be beneficial to carry out more research including characterization of growth hormones and others metabolites. We can also evaluate the effect of seaweed extract on nitrogen metabolism in root and leaves of spinach plants cultivated in medium culture deficiency or richened of mineral elements.

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تأثيرالتركيزات المختلفة من الطحالب البحرية على نباتات السبانخ

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أجريت هذه التجربة الحقلية خلال موسمى الشتاء ٢٠١٤/٢٠١٣ و ٢٠١٥/٢٠١٤ على الترتيب في محطة أبيس للبحوث الزراعية بمزرعة كلية الزراعة ، جامعة الإسكندرية ، لدراسة تأثير تركيزات مختلفة من الطحالب البحرية كتسميد عضوى وهى:
(Bread yeast-Jainarubens-Ulva lactuca) مقارنة مع التسميد المعدني على نمو وإنتاجية السبانخ، وقد طبقت أربعة تركيزات من كل طحلب (٥٪، ١٠٪، ٢٠٪ و ٤٠٪) ورشها وحدها أوتخلط مع بعضها ، وإستخدام التسميد المعدنى من معدلات NPK الموصى بها من وزارة الزراعة .وقد تم حصاد النباتات بعد ٥٠ يوما من الزراعة وتم قياس بعض الصفات مثل (ارتفاع النبات سم ،المادة الجافة للأوراق ٪، المحصول الكلي، محتوى الأوراق للكلوروفيل ، محتوى النيتروجين والبوتاسيوم والفوسفور وتراكم النتراتفى الأوراق . (وقد أظهرت النتائج ان طحلبى Ulvalactuca (Bread yeast + ٥٪) مختلطين معا قد أعطى أفضل النتائج من حيث كمية المحصول و إنخفاض كمية النترات بالأوراق .