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Effect of Seaweed Extract, Chitosan and Salicylic Acid on Growth, Seed Yield and Some Pests Infestation of Pea Plant

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Abstract: To examined the role of the foliar spraying with seaweed extract, chitosan and Salicylic acid on growth, seed yield and seed quality of the three pea cultivars Master B, Entsar 1 and Entsar 2 a field experiment was designed in split plot design with three replicates at the experimental farm of Sadat City University during the two successive seasons of 2018/2019 and 2019/2020 where the three pea cultivars were allocated in the main plots and the foliar spray treatments were randomly distributed in the sub-plots. The results confirmed a wide differ among all tested cultivars in all growth, seed yield traits as well as the severity infections with leaf miner larvae and adult aphid. Master B cv. expressed the highest plant height, foliage fresh weight, foliage dry weight and seed yield/plot and fad. in both seasons. Master B cv. also, followed by Entsar 2 showed the lowest number of leaf larvae and adult aphid in both seasons Data showed that all used treatments significantly exceeded the control treatment in all growth and seed yield traits. Pea plants that sprayed with seaweed extract had the highest plant height, foliage fresh weight, foliage dry weight and seed yield/plot and fad in both seasons, followed by chitosan and salicylic acid in all traits. The lowest leaf miner larvae and adult aphid were obtained from pea plants that treated with seaweed extract in both seasons, followed by chitosan and salicylic acid. Results revealed that the three tested cultivars differ in their response to the different used treatments. The pea cultivar Master B that sprayed with seaweed extract had the highest plant height, foliage fresh weight, foliage dry weight and seed yield/plot and fad as well as the lowest number of: leaf miner larvae and adult aphid in both seasons.

Key words: Pea (*Pisum stivum*) · Cultivars · Growth stimulants · Yield and insect infections

INTRODUCTION

Pea (*Pisum sativum* L.) is one of the important vegetables in the world and ranks among the top 10 vegetable crops (Ref./in the world?). Pea is commonly used in human diet throughout the world and it is rich in protein (21-25%) [1]. Peas are important for their nutritional values as it is constituent of high amount of protein carbohydrates, vitamin A and B and has high levels of amino acids lysin and trypophan. Peas are source of minerals such as phosphorous, iron and zinc. It is a cheap source of protein and minerals especially important in developing countries where zinc deficiency is fifth major cause of diseases and even deaths [1]. There is a significant increase in the demand for peas in Egypt because of its nutritional value and its acceptance by the Egyptian consumer, especially during the fasting period in Ramadan each year, which leads to a significant increase in the price of peas.

Since there is a noticeable gap between production and consumption, the government resorts to importing as a quick solution to bridge that gap. However, the Egyptian plant breeder has taken upon himself to develop fundamental solutions to this problem represented in many points such as increasing the area planted with the crop by planting it in new or marginal lands, As well as

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breeding new varieties with high productivity, such as Entsar 1 and 2, or importing distinctive varieties from abroad, such as Master B. These steps often coincide with improving agricultural practices and processes that lead to an increase in the yield, such as service operations, irrigation, control of fungal and insect pests and Weeds, in addition to developing the composting process using organic and biological alternatives that are safe for consumer health, such as seaweed extract, chitosan and salicylic acid.

Seaweed extract is a new generation of natural organic fertilizer highly nutritious and encourage faster germination of seeds and enhance yield and resistant ability of several crops [2]. It was found that that seaweed extract has been found to contain growth stimulators such as auxins, gibberellins and cytokinin. The extract also comprises growth promoting hormones (IAA and IBA), trace elements (Fe, Cu, Zn, CO2, Mo, Mn and Ni), vitamins and amino acids [3]. Seaweeds are an important source of organic matter and fertilizer nutrients. Seaweed extracts have been used in agriculture as soil conditioners or plant stimulators. They are applied as foliar spray and are able to enhance plant growth, improving yield and productivity of several crops [4]. Gollan and Wright [5] found an enhancement effect of algae extract on pea plant growth characteristics may be attributed to the auxin content of the algae extract which has an effective role in cell division and enlargement. This leads to increase he shoot growth, leaves number and plant dry weight.

Chitosan, a common name to a deacetylated form of chitin, is natural biodegradable materials derived from crustaceous shells, whose main attribute s corresponds to its polycationic nature [6]. Chitosan is of keen interest now a days and used in seed, leaf, fruit and vegetable coatings, as fertilizer and in controlled agrochemical release, to increase plant productivity [7]. Malerba and Cerana [8] confirmed that the beneficial effect of Chitosan (CHI) on plant growth may be attributed to the promoting effects on nutrients uptake and nutritional status especially nitrogen, potassium and phosphorous which are essential for plant growth. Chitosan have insecticidal and disease preventive actions, also it helps plants to stimulate stress tolerance.

Salicylic acid (SA) is a plant growth regulator known as an endogenous marking molecule, which is implicated in different physiological processes, like growth regulation, photosynthesis, stomatal behavior, nutrient uptake and mechanisms of tolerance to abiotic stresses [9]. Salicylic acid is an endogenous growth regulator with phenolic nature, which participates in regulation of several physiological processes in plants, such as stomatal closure, ion uptake, inhibition of ethylene biosynthesis and reduce transpiration [10]. SA can increase protein content and reduced adverse effects of salinity [11].

According to the previous descriptions the main objectives of this study were to examined the role of seaweed extract, chitosan and salicylic acid as safe alternatives for mineral fertilizers on increase growth and seed yield of the three pea cultivar Entsar 1, Entsar 2 and Master B under sand soil conditions.

Determined the effectiveness of seaweed extract, Chitosan and Salicylic acid on reduce the infections of aphid and leaf miner in pea plants.

MATERIALS AND METHODS

To examine the impact of some plant stimulants seaweed extract (20ml/l), chitosan (250ppm) and salicylic acid (50ppm) compared with control (tab water) on the growth, seed yield and tolerance to leaf miner and aphid of the three pea cultivars; i.e. Master B, Entsar 1 and Entsar 2 under Menofiya Governorate conditions. For this purpose a field experiment designed in a split plot design with three replicates was conducted at the experimental farm of Sadat City University, El-Menofya Governorate, Egypt during 2018/2019 and 2019/2020 seasons where the three pea cultivars were allocated in the main plots and the three used treatments and the control were randomly distributed in the sub-plots. This experiment included 12 treatments which were the combinations between three cultivars and four treatments as used as foliar applications.

Seeds of the three pea cultivars were sown on November 5th and 4th in both seasons, respectively. The plot size was four ridges each ridge was five meters long and 70 cm apart. Seeds were sown on the two side of the ridge at 20 cm hill spacing with one seed per hill. The plot area was 14 m^2 (including 196 plants). The wet planting method called (Herati) was used and all the other cultural practices (irrigation, fertilization and weed control) were followed as recommended. All treatments were applied as a foliar spray three times: the first one was in beginning of flowering, the second one was 10 days after the first on and the third was 10 days after the second one. Soil physical and chemical properties of the experimental site and analysis of irrigation water are presented in Table 1 and 2.

Table 1: Soil physical and chemical properties of the experimental site (Average two seasons)

Physical properties											
Sand %				Silt %			Clay %			Texture	
83				10			7			Sandy loamy	
Chemical properties											
		Soluble salts (mg/100g)									
pH	EC dS/m	Na^+	K^+	Ca^{++}	Mg^{++}	Cl^-	HCO_3^{--}	CO_3^{--}	SO_4^-	O.M	CO_3%
8.17	1.114	3.3	0.18	1.2	1.2	7.4	1.9	0	3.4	0.35	1.7

Table 2: Analysis of irrigation water

												Fe	Zn	Mn	Cu
pH	EC dS/m	Ca^{++}	Mg^{++}	Na^+	K^+	CO_3^{--}	HCO_3^{--}	Cl^-	SO_4^-	SAR	TDS	---------- ppm ----------			
7.98	1.26	4.2	2.6	6.5	0.2	0	4	7	2.5	3.5	896	0.11	0.18	<0.01	<0.01

Studied Characters Were

Vegetative Growth Traits: Five random plants from each experimental plot were randomly taken at 90 days after seed sowing to determine plant height (m), foliage fresh weight (g) and foliage dry weight (g)

Seed Yield Traits: Seed yield was measured as the total harvested seeds over all season and seed yield per faddan (tons) was calculated by converting the yield of the experimental plot (kg) to faddan.

Number of Leaf Miner (*Liromiza congesta*) and Aphid and (*Aphis gossypii*): Monitoring of *L. congesta* and *A. gossypii*, numbers was conducted throughout from three weeks after sowing until the end of the fruiting stage in the three pea cultivars plots. Sampling was carried out at weekly interval early in the morning before the insect pests adults tend to be more active [12]. Number of leaf miner and aphid were assessed by leaf random sampling. For sampling, 30 leaves representing top, middle and bottom canopy were picked from each of the three cultivars for each treatment were randomly selected per plot.. Leaf samples were kept in separate paper bags properly labeled with plot number. The leaves were then brought to the laboratory on the same day where leaf miner larvae and adult aphid were counted.

Statistical Analysis: Results were expressed as mean. The data were analyzed by using two-way ANOVA followed by LSD test through SPSS 16 (version 4). The treatments means were compared using least significant difference (LSD) at significant levels of 5% as described by Gomez and Gomez [13].

RESULTS AND DISCUSSIONS

Plant Growth

Cultivars Effects: The obtained data in Table 3 confirmed the wide differ among all tested cultivars in all growth traits. Master B cv. expressed the highest plant height (89.74 and 92.12cm), foliage fresh weight (79.81 and 79.93 g) and foliage dry weight (29.28 and 28.90g) in both seasons, respectively followed by Entsar 2 in all previous traits. In contrast, Entsar 1 cv showed the lowest plant height (72.55 and 70.74 cm), foliage fresh weight (54.45 and 52.98 g) and foliage dry weight (24.31 and 22.92 g).

Our results confirmed the presence of the significant differences among the three pea cultivars in all growth traits. The wide diversity among pea genotypes in growth traits were observed before by; Datta and Das [14] in plant height.

Treatments Effect: Data in Table 3 showed that all used treatments significantly exceeded the control treatment in all growth traits i.e. plant height, foliage fresh and dry weight in both seasons of this study. Pea plants that sprayed with seaweed extract had the highest plant height (94.91 and 99.46 cm), foliage fresh weight (76.76 and 80.68 g) and foliage dry weight (33.80 and 35.50g) in both seasons, respectively followed by chitosan and salicylic acid in all growth traits. on the other side, pea plants under the control treatment had the lowest plant height (65.40 and 61.48cm), foliage fresh weight (52.32 and 48.92g) and foliage dry weight with averages of 20.71 and 18.33g in the first and second seasons, respectively.

Table 3: Effect of pea cultivars, foliar application treatments and their interactions in pea plants growth traits during 2018/2019 and 2019/2020 growing seasons

		Plant height (cm)		Foliage fresh weight (g)		Foliage dry weight (g)	
Factors		1st Season	2nd Season	1st Season	2nd season	1st Season	2nd Season
Cultivar							
Master B		89.74	92.12	79.81	79.93	29.28	28.90
Entsar 1		72.55	70.74	54.45	52.98	24.31	22.92
Entsar 2		79.75	80.76	60.16	60.41	27.40	27.04
LSD 5%		1.14	1.35	1.32	1.15	0.94	1.01
Treatments							
Seaweed extract		94.91	99.46	76.76	80.68	33.80	35.50
Chitosan		85.57	87.27	69.09	70.70	29.34	29.64
Salicylic acid		76.84	76.63	61.05	57.47	24.13	21.69
Control		65.40	61.48	52.32	48.92	20.71	18.33
LSD 5%		2.09	3.79	3.81	4.53	1.83	2.31
Cultivar	Treatments						
Master B	Seaweed extract	99.79	104.44	89.64	94.08	35.47	37.20
	Chitosan	93.96	98.59	84.46	88.90	32.28	33.95
	Salicylic acid	85.97	90.55	75.12	70.88	25.50	23.03
	Control	79.24	74.91	70.01	65.87	23.85	21.42
Entsar 1	Seaweed extract	89.14	93.57	67.82	71.41	31.83	33.48
	Chitosan	72.27	68.28	54.01	50.78	23.44	21.02
	Salicylic acid	67.02	63.13	50.00	46.84	21.80	19.41
	Control	61.76	57.98	45.98	42.90	20.16	17.80
Entsar 2	Seaweed extract	95.80	100.36	72.83	76.54	34.11	35.81
	Chitosan	90.48	94.94	68.82	72.44	32.28	33.95
	Salicylic acid	77.53	76.22	58.03	54.70	25.09	22.62
	Control	55.19	51.53	40.96	37.98	18.11	15.78
LSD 5%		3.61	1.70	1.32	1.32	0.63	0.63

Cultivars X Treatments Effects: Results in Table 3 revealed that the three tested cultivars differ in their response to the different used treatments. The pea cultivar Master B that sprayed with seaweed extract had the highest plant height (99.79 and 104.44cm), foliage fresh weight (89.64 and 94.08g) and foliage dry weight (35.47 and 37.20g) in both seasons, respectively followed by Entsar 2 under the same treatment and both Master B and Entsar 2 under chitosan. On the other hand, Entsar 2 under the control treatment expressed the lowest plant height (55.19 and 51.52 cm), foliage fresh weight (40.96 and 37.98g) and foliage dry weight (18.11 and 15.78g) in the first and second seasons, respectively.

Our results indicated that all used treatments significantly exceeded the control and the three tested cultivars differ in their response to the different used treatments. The pea cultivar Master B that sprayed with seaweed extract had the highest plant height, foliage fresh weight and foliage dry weight. seaweed extract contain several auxins which has an effective role in cell division and enlargement leads to increase he shoot growth, leaves number and plant dry weight [5]. Also, Attememe [15] reported that seaweed extract contains macronutrients (N, P and K) which are very essential for growth and development of the plant. Our results are in the same lines with this of Sivasankari *et al.* [16] who showed that aqueous extract of seaweeds promoted the seedling growth including the parameters of shoot length, root length, fresh weight and dry weight of Vigna sinensis. Similar results were obtained by [16] Sivasankari *et al.* and Youssef *et al.* [17] in Cawpea, Rathore *et al.* [18] in Soybean, Hamed [19] in Phaseolus vulgaris, Oancea *et al.* [20], Sutharsan *et al.* [21] in tomato, Mohammed and Marwa Hamdoon [22], Nawar *et al.* [23] and Elsharkawy *et al.* [24] in pea and Mafakheri and Asghari [25] in Trigonella foenum-graecum.

The increase in pea growth under chitosan may due to chitosan can improve physiological processes in plant [26]. Khan *et al.,* [27] also, indicated that chitosan significantly affected growth attributes of pea. The role of chitosan in promoting plant growth were showed before by Ghoname *et al.* [28] in sweet pepper, Farouk *et al.* [26] in radish plant, Chookhongkha *et al.* [29] in chili plant, Fawzy *et al.* [30] in garlic and Mondal *et al.* [31] in mungbean.

In this study salicylic acid also improve all pea growth traits. salicylic acid (SA) is an effective endogenous plant growth regulator. It plays physiological

roles in plant including growth and development, thermogenesis, flower induction, nutrients uptake, ethylene biosynthesis, stomata movement, photosynthesis and enzymes activity [32, 33]. In the previous studies the foliar treatment with salicylic acid significantly increase all vegetative parameters (plant height, fresh and dry weights, leaves number and leaves area) in pea plants [34-37]. Similar results were obtained by Nour *et al.* [38] in snap bean, Sakr *et al.* [39] in soybean and Rady *et al.* [40] in common bean.

Seed Yield

Cultivars Effects: Results in table 4 showed a wide differ among all tested cultivars in seed yield and yield components traits. The pea Master B had the highest seed yield per plot (14.40 and 14.59 kg) and fad. (4.32 and 4.38 ton) in the two studied seasons, respectively followed by Entsar 2. On the other side, Entsar 1 cv showed the lowest seed yield per plot (11.63 and 11.51 kg) and fad. (3.49 and 3.45 ton) in both seasons, respectively.

In this study seed yield/fad differ significant in all tested genotypes and Master B cv. showed excellent seed yield/fad. The differences between pea genotypes may due to the wide genetic diversity of these genotypes [41, 42].

Treatments Effect: All used treatments significantly increased seed yield compared to control (Table 4). The highest seed yield/plot (15.82 and 16.23 kg) and seed yield/fed (4.75 and 4.87 ton) were obtained under the foliar spraying with seaweed extract in both seasons, respectively followed by chitosan then salicylic acid.

Cultivars X Treatments Effects: The response of the three pea cultivars significantly differed under the different treatments in all yield traits during the two growing seasons (Table 4). The pea cultivar Master B that sprayed with seaweed extract showed the highest seed yield per plot (17.00 and 17.42 kg) and fad. (5.10 and 5.23 ton) in both seasons, respectively followed by Master B and Entsar 2 that sprayed with chitosan. On the other hand, Entsar 2 under the control treatment expressed the lowest seed yield per plot (7.60 and 7.28 kg) and fad. (2.28 and 2.18 ton) in both seasons, respectively.

In the present study all used treatments significantly increased yield and yield components traits compared to control. The pea cultivar Master B that sprayed with seaweed extract had the highest yield measurements. Seaweed components consider a great source of macro and micronutrients, essential fatty acids, amino acids, vitamins, cytokinins, auxins like growth promoting substances affecting cellular metabolism in treated plants leading to enhance growth and yield [43]. Our results are in agree with those of Mohammed and Hamdoon [22] and Elsharkawy *et al.,* [24] who showed that spraying pea plants with algae extract at 10 or 15 % increased significantly seed yield of pea.

Also our result indicated that chitosan resulted in a large increase in seed yield compared to control. Khan *et al.* [27] and Zakiullah *et al.* [44] indicated that chitosan significantly affected seed yield and its attributes of pea.

Salicylic acid also, increase seed yield/fad. of pea plant. In this respect Gad El-Hak *et al.* [34]; Chame *et al.* [45] and also, EL-Afifi *et al.* [35] found that the maximum increase fresh seeds weight/pod, number of fresh seeds/pod, green pod yield and seeds weight/dry pod was observed in the pea plants treated with SA.

Severity of Leaf Miner and Aphid

Cultivars Effects: The presented data in Table 5 indicated that the severity infections with leaf miner larvae and adult aphid significantly differ in the three tested cultivars. Master B was the most tolerant cultivar to both leaf miner and aphid where it showed the lowest number of leaf larvae (5.33 and 5.41) and adult aphid (5.98 and 6.06) in both seasons, respectively followed by Entsar 2. In the contrast of this Entsar 1 cultivar was the most susceptible cultivar to both leaf miner and aphid where it showed the highest number of leaf larvae (7.35 and 7.21) and adult aphid (8.14 and 8.00) in both seasons, respectively.

Our results revealed that, the severity infections with leaf miner larvae and adult aphid significantly differ in the three tested cultivars. Master B was the most tolerant cultivar to both leaf miner and aphid. Similar results was obtained before by Greveniotis *et al.* [42] who tested five cultivars of peas and they found a large differ among genotypes in their tolerance to common insects and disease infections and Vermio cultivar was the most tolerance genotype to white fly, aphid and leaf miner also, the same genotype showed moderate tolerance to rust disease in all tested locations.

Treatments Effects: All used treatments significantly decreased infection of both leaf miner and aphid in pea plants compared with control (Table 5). The lowest leaf miner larvae (4.10 and 4.19) and adult aphid (4.59 and 4.69) were obtained from pea plants that treated with seaweed extract in both seasons, respectively followed by chitosan

Table 4: Effect of pea cultivars, foliar application treatments and their interactions on seed yield/plot and faddan of pea during 2018/2019 and 2019/2020 growing seasons

		Seed yield/plot (kg)		Seed yield/fad (ton)	
Factors		1st Season	2nd Season	1st Season	2nd Season
Cultivar					
Master B		14.40	14.59	4.32	4.38
Entsar 1		11.63	11.51	3.49	3.45
Entsar 2		12.89	13.03	3.87	3.91
LSD 5%		0.39	0.71	0.23	0.43
Treatments					
Seaweed extract		15.82	16.23	4.75	4.87
Chitosan		14.50	14.72	4.35	4.42
Salicylic acid		11.99	11.93	3.60	3.58
Control		9.58	9.31	2.87	2.79
LSD 5%		0.89	1.03	0.54	0.62
Cultivar	Treatments				
Master B	Seaweed extract	17.00	17.42	5.10	5.23
	Chitosan	15.80	16.21	4.74	4.87
	Salicylic acid	12.89	12.99	3.86	3.90
	Control	11.90	11.75	3.57	3.52
Entsar 1	Seaweed extract	15.20	15.59	4.56	4.68
	Chitosan	11.91	11.75	3.57	3.53
	Salicylic acid	10.19	9.81	3.05	2.95
	Control	9.25	8.90	2.77	2.67
Entsar 2	Seaweed extract	15.27	15.67	4.59	4.70
	Chitosan	15.79	16.20	4.74	4.86
	Salicylic acid	12.89	12.99	3.87	3.90
	Control	7.60	7.28	2.28	2.18
LSD 5%		1.09	1.33	0.66	0.8

and salicylic acid. On the other side pea plants under the control treatment had the highest number of leaf miner larvae (8.40 and 8.13) and adult aphid (9.30 and 9.02) in the first and second seasons, respectively.

Cultivars X Treatments Effects: Results in Table 5 indicated that the infection of leaf miner and aphid in the three tested cultivars differ significantly according to the different used treatments. The pea cultivar Master B that sprayed with seaweed extract had the lowest number of: leaf miner larvae (3.88 and 4.08) and adult aphid (4.37 and 4.57) in both seasons, respectively followed by Master B under chitosan and Entsar 1 and 2 under seaweed extract. On the other hand, Entsar 1 under the control treatment expressed the highest number of: leaf miner larvae (9.91 and 9.54) and adult aphid (10.82 and 10.44) in the first and second seasons, respectively.

In this study, the lowest infections with leaf miner larvae and adult aphid were obtained from pea plants that treated with seaweed extract. Sosnowski *et al.* [46] showed that use of seaweed extracts increases the tolerance of plants to abiotic stresses (salinity, drought, or extreme temperatures) and biotic stresses such as insect pests (white fly and leaf miner) and fungus diseases. Also, Mafakheri and Asghari [25] showed that foliar applications of seaweed decrease the infections of aphid, white fly and leaf miner in Trigonella foenum-graecum under open field conditions.

As for chitosan effect of insect infections Malerba and Cerana [8] confirmed that chitosan have insecticidal and disease preventive actions, also it helps plants to stimulate stress tolerance. In the study of Badawy *et al.* [47] reported that growth and larval vitality of oleander aphid (Aphis nerii) and cotton leaf worm (Spodoptera littoralis) were severely affected by chitosan. While, Sahab *et al.* [48] reported that chitosan incorporated with polyacrylic acid offers excellent potential in managing attack of common pests like cotton aphid and beetles in soybean cultivation.

In this study salicylic acid reduced the infections of leaf miner and aphid in pea plant compared to control and this may due to the phenolic nature of salicylic acid that repelled the insect pests. Salicylic acid is an endogenous growth regulator with phenolic nature, which participates in regulation of several physiological processes in plants, such as stomatal closure, ion uptake, inhibition of ethylene biosynthesis and reduce transpiration [10].

Table 5: Effect of pea cultivars, foliar application treatments and their interactions in the severity of pea plants leaf miner and aphid during 2018/2019 and 2019/2020 growing seasons

		Number of leaf miner larvae		Adult aphid numbers	
Factors		1st Season	2nd Season	1st Season	2nd Season
Cultivar					
Master B		5.33	5.41	5.98	6.06
Entsar 1		7.35	7.21	8.14	8.00
Entsar 2		5.87	5.81	6.56	6.50
LSD 5%		1.07	0.78	1.16	1.13
Treatments					
Seaweed extract		4.10	4.19	4.59	4.69
Chitosan		5.05	5.15	5.67	5.77
Salicylic acid		7.17	7.10	8.01	7.94
Control		8.40	8.13	9.30	9.02
LSD 5%		0.89	0.83	0.93	0.87
Cultivar	Treatments				
Master B	Seaweed extract	3.88	4.08	4.37	4.57
	Chitosan	3.99	4.19	4.48	4.68
	Salicylic acid	6.27	6.31	7.02	7.06
	Control	7.17	7.06	8.05	7.94
Entsar 1	Seaweed extract	4.37	4.57	4.87	5.07
	Chitosan	7.17	7.06	8.05	7.94
	Salicylic acid	7.94	7.66	8.83	8.54
	Control	9.91	9.54	10.82	10.44
Entsar 2	Seaweed extract	4.06	3.93	4.55	4.42
	Chitosan	3.99	4.19	4.48	4.68
	Salicylic acid	7.30	7.33	8.18	8.21
	Control	8.14	7.80	9.02	8.68
LSD 5%		1.39	1.31	1.46	1.37

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