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Application of seaweed and salicylic acid on qualitative and qualitative traits of field pumpkin, *Cucuorbita pepo* L. in Khorramabad, Iran

Narges Kamali Omidi, Ali Khorgami*, Kazem Taleshi

Department of Agriculture, Khorramabad branch Islamic Azad University, Khorramabad, Iran

** Corresponding author's E-mail: ali-Khorgamy@yahoo.com*

ABSTRACT

The present study was conducted to investigate the effect of different levels of foliar application of salicylic acid and seaweed in 2017-18 cropping year in Khorramabad City, Iran with a temperate climate at 11478 meters above sea level. The study was carried out in a factorial arrangement based on completely randomized block design with three replications. Factors comprised density application of seaweed at three levels (control, seeds of wisdon and seaweed spraying) and salicylic acid spraying at four levels (control, one, two, and three times salicylic acid spraying). The traits evaluated include: seed yield, the weight of 1000 seeds, fruit yield, fruit number, the seed number, oil yield, essence yield, protein yield and phenolic compounds. The results showed that the effect of seaweed on all treatments were significantly different, while in the case of phenolic compounds, the differences were not significant. The effect of salicylic acid on all treatments were significant except for fruit yield and phenolic compounds. Seed yields led to the highest performance at a3 (781.92 kg h^{-1}) while lowest at a1 (494.54 kg h^{-1}) compared to other levels. The salicylic acid spraying at s4 (712.41 kg h^{-1}) exhibited the highest, while s1 (587.72 kg h^{-1}) displayed the lowest compared to other levels. Interaction of seaweed and salicylic acid were not significant. Seed yield was highest at a3s4 (825.00 kg h^{-1}) while lowest at a1s1 (448.17 kg h^{-1}). In conclusion, Pumpkin as drug crop is suitable for planting in Khorramabad region, Iran.

Keywords: Growth promoting, Fertilizer, Seed-Coating, Protein, Oil.
Article type: Research Article.

INTRODUCTION

The increasing approach towards using medicinal plants clarifies the global significance of cultivating and producing these plants; at present, the demand for medicinal plants as consumable products in health and pharmaceutical industries are increasing (Hecel & Sustrikova 2006). Given the needs of the society, cultivating medicinal plants is now a main branch of agriculture as well as a main source of raw materials' extraction and production for producing the existing drugs. Field pumpkin is one of the most important medicinal plants. Field pumpkin is a one-year plant belonging to Cucurbitaceae, also called cucurbits family. It has a major role in treating prostate gland, urinary tract burning, and atherosclerosis (Balouchi 2013). The oil obtained from field pumpkin contains valuable materials including unsaturated fatty acid, vitamin A, vitamin E, minerals, phytosterols, carotenoids, protochlorophylls. The most important fatty acids accounting for nearly 90% of the oil content include linoleic, oleic, and palmitic acids. Linoleic acid accounts for 50% of fatty acids (Fruhwirth & Hermetter 2008). Given its antibacterial benefit, it is widely used for treating earaches, fevers, bronchitis, urinary tract diseases and infections, and prostate (Mayor *et al.* 2011). Sustainable agriculture based on using organic and biological fertilizers and with the purpose of removing or reducing chemical inputs is a desirable method for overcoming these problems. Soil fertility has a major role on the yield of cucurbits; it is of high significance to apply fertilizers in soils lacking nutrients.

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Nowadays, applying chemical fertilizer has significantly increased for compensating the lack of soil nutrients and high yield. However, in numerous cases, applying these chemicals has resulted in environmental pollutions, ecological damages, and increased production costs (Salehe *et al.* 2014).The agronomic management of applying different types of fertilizers affects qualitative and quantitative yield of field pumpkin. In recent years, given its capabilities for being used in organic agriculture and sustainability, seaweed essence has become popular (Roth & Goyne 2004). Since seaweed essence contains growth hormone substances including cytokinin and auxin as well as minerals such as iron, copper, zinc, cobalt, molybdenum, manganese, nickel, vitamins and amino acids, it has a positive effect on the plant growth. It has been reported that the foliar application of seaweed essence has significantly improved the quality and quantity of fenugreek medicinal plant (Mafakheri 2017). This substance exists in the plants in small quantities; it functions as an antioxidant in the plants and results in the removal of free radicals in the plants (Khaled *et al.* 2007).Salicylic acid can have a protective and defensive role in plants subject to environmental stresses such as drought stress; this acid can increase the plant resistance against such stresses (Eiasu *et al*. 2012). However, the reports indicate that the foliar application of salicylic acid increases resistance to stress and reduces the harmful effect of oxidative stresses at different phases of the plant growth (Bideshki & Arvin 2010). In sustainable agricultural systems, any agricultural improvement is required to result in the increased production and reduced environmental damages that ultimately bring about increased resistance. In one of these methods, the application of biological stimuli can increase the effect of mineral fertilizers. In recent years, given its capabilities for being used in organic agriculture and sustainability, seaweed essence has become popular (Roth & Goyne 2004). Unlike chemical fertilizers, the essence obtained from seaweed is non-poisonous and prevents environmental damages; it does not bring about pollution for humans, animals, and birds (Sibi *et al.* 2016). The bioactive substances extracted from seaweed are largely applied in agricultural and garden products all over the world; different studies have reported the positive useful effects of these bioactive substances especially the increased qualitative and quantitative yield of different products (Craigie 2011). The liquid extracts obtained from seaweed have become highly significant for the foliar spraying of many products including different types of grass, grains and cereals, flowers, and plant species. Seaweed essence contains primary and secondary nutrients. Amino acids, vitamins, and growth hormone substances such as cytokinin, auxin, and Abscisic acid (Thambiraj *et al.* 2012; Alamet *et al.* 2013). Given the significance and popularity of medicinal plants (especially over the recent years), giving due attention to the strategies of sustainable agriculture and minimizing the application of chemical fertilizers and applying organic fertilizers, and the increased severity of environmental stresses in crops, the present study has been conducted to investigate the response and reaction of field pumpkin to different levels of foliar application of salicylic acid and application of seaweed in Khorramabad.

MATERIALS AND METHODS

The present study was conducted to investigate the effect of different levels of foliar application of salicylic acid and seaweed in the crop year of 2017-18 in Khorramabad City, Iran with a temperate climate at 11478 meters above sea level and the geographical coordinates of 33° 29' 16" North and 48° 21' 21" East as well as the annual precipitation of 52506 mm. The study was conducted in a factorial arrangement based on completely randomized block design with three replications. The factors investigated include salicylic acid spraying (S) at four levels including control (S1), i.e., spraying with purified water, one-time (S2), two times (S3) and three times acid spraying (S4), as well as the application of seaweed (A) at three levels including control (A1) i.e., foliar spraying with purified water (A1), seed inoculation with seaweed (A2), and foliar spraying with seaweed (A3). Before the tillage, the sample of the soil was randomly collected from the farm, and its physical and chemical characteristics were measured (Table 1). Tillage was conducted at the beginning of spring as spring tillage. Disc plowing was performed to crush the clods. Then, the farm was flattened using trowel. The furrows were then made, and streams and stacks were created, then the farm was irrigated for one time. The seeds were soaked in lukewarm water for 24 hours and were then placed on a wet cloth; the seeds were followed by soaking in Benomyl fungicide for a few minutes. Each plot included six five-meter line at a distance of 1 m and planting was conducted at the depth of 3-5 cm. The distance between the plants on the planting row was determined to be 50 cm, to prevent intermixture of treatments for each 4 terrace 2 unplanted line of 100 cm were considered. According to the environmental temperature, Pumpkin was planted in Khoramabad on the second half of May and immediately irrigated.

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Table 1. The results of physical and chemical analysis of the soil used in the present study.

Soil texture	Farm humidity (%)	Lime (%)	Organic carbon (%)	Total nitrogen (%)	Absorbable phosphorous (Mg kg^{-1})	Exchangeable potassium (Mg kg^{-1})	Electrical conductivity (dS m^{-1})	acidity
Clay-silt	22.5	15.9	0.98	1.01	6.9	355	0.55	7.9

When the plant was harvested, the qualitative and quantitative traits of the plant were measured including seed yield, the weight of 1000 seeds, fruit yield, fruit number, the seed number per fruit, oil yield, essence yield, protein yield and phenolic compounds. The data obtained were analyzed using SAS 9.0, Mean comparison was also performed using Duncan's test and software Excel was applied for drawing the diagrams.

RESULTS AND DISCUSSION

Seed yield (kg h^{-1})

The results of variance analysis indicated that the effects of seaweed and salicylic acid spraying on seed yield were significant, while their mutual effects did not exhibit significant differences ($p < 0.01$). The highest seed yield was related to the effect of seaweed A3 (781.92 kg ha^{-1}), while the lowest to A1 (494.54 kg ha^{-1}). The highest effect of salicylic acid spraying on seed yield was associated with S4 (721.41 kg ha^{-1}), while the lowest with S1 (587.72 kg ha^{-1}; Table 2). Salicylic acid spraying not only moderates the effects of drought via maintaining more moisture and reducing transpiration during the growing season, but also increases the aerial organs growth and dry weight (Shakirova & Bezrukova 1997). Salicylic acid spraying results in the increased dry weight of the plant which is attributed to the increased net photosynthesis, Rubisco carboxylation efficiency, and increased photosynthetic enzyme activities. Finally, the increased dry weight results in the increased yield (Fariduddin *et al.* 2003). Bayat *et al.* (2010) reported that applying salicylic acid spraying resulted in the increased corn seed yield.

Table 2. Results of analysis of variance of qualitative and quantitative traits in pumpkin.

Source of change	Degree of freedom	Seed yield	Weight of 1000 seeds	Fruit yield	Fruit number	Seed number per fruit	Oil yield	Essence yield	Protein yield	Phenolic compounds
Replication	2	309387.8	1664.1	152084444.4	0.64	367985.8	82401.9	21.7	214385.5	1.97 ns
Seaweed	2	247798.3**	14900.1**	428497777.8**	0.31**	1286587.1**	59628**	15.6	16500.2**	3.22 ns
Salicylic	3	25964.5*	892.6**	23518518.5 ns	0.05**	96177.6**	6429.8**	1.6*	1748.5*	0.03 ns
S×A	6	6992.6 ns	451.9 **	8225185.2 ns	0.01 ns	18192.7 **	1597.2 ns	0.40 ns	445.8 ns	2.29 ns
Error	22	6072.6	34.2	9606869	0.006	2688.6	1494.9	0.36	373.3	1.15
Coefficient of variation		12.1	3.6	8.450	2.7	3.8	12.3	12.8	12.2	1.60

Note: *, ** and ns indicate significant level at 0.05, 0.01 and non-significant, respectively.

Weight of 1000 seeds

Based on the results of variance analysis, using seaweed and salicylic acid spraying and their mutual effects exhibited significant effects on the weight of 1000 seeds of field pumpkin ($p < 0.01$; Table 2). The highest weight was related to seaweed A3 (193.03 g), while the lowest yield to seaweed A1 (122.88 g). The highest effect of salicylic acid on the weight of 1000 seeds was related to S4 (171.47 g), while the lowest to S1 (150.66 g; Table 2). Khan *et al.* (2010) reported that the application of salicylic acid results in the increased growth rate and photosynthesis of the plant, and will subsequently results in an increased yield.

Fruit yield

The results of variance analysis of the present study indicated that the effect of seaweed on fruit yield was significant ($p < 0.01$). The effects of salicylic acid spraying and the mutual effects of seaweed and salicylic acid

spraying on fruit yield were not significant ($p > 0.01$). The highest fruit yield was related to the effect of seaweed A3 (42833 kg ha^{-1}), while the lowest yield to the effect of seaweed A1 (30900 kg ha^{-1}). The highest effect of salicylic acid spraying on fruit yield was found in treatment S4 (38178 kg ha^{-1}), while the lowest in S1 (35067 kg ha^{-1}; Table 2). The more desirable the plant growth in its growth period, the more appropriate the plant extent. The plant weight during its growth period is affected by the density of nutrients and the photosynthesized products made in the plant. Thus, given the existence of growth hormones, applying seaweed will result in the increased density of nutrients in the leaves and the promoted plant yield. Seaweed can affect the absorption process and movement of nutrients in the plant (Sunarpi *et al*. 2010). The dry weight of the plant in the treatment of applying seaweed was increased. The presence of growth stimulus factors (such as indole-3-acetic acid, indole-3-butyric acid, gibberellic acid, micro and macro elements, vitamins, amino acids) in the seaweed essence results in the improved growth of the plants (Shahbazi *et al.* 2015). It has been reported that applying seaweed essence has a positive effect on the total root volume in Norway spruce and grape trees, elevating them which is in line with the findings of other studies conducted on the positive effect of seaweed on the root growth of other plants (Calvo *et al*. 2014; Arioli *et al.* 2015). It seems that the time and method of applying seaweed essence exhibit significant effects on the root volume. Applying seaweed essence in the vegetative stage of the plant (in comparison with the reproductive stage) and using the essence on the foot of the root (in comparison with the spraying) have resulted in the development of safflower's roots, leading to the increased volume of the roots and fruit yield (Sibi *et al.* 2016).

Table 3. Comparison of the mean mutual effect of seaweed and salicylic acid on qualitative and quantitative traits in pumpkin.

seaweed	Salicylic acid	Seed yield (kg h^{-1})	Weight of 1000 seeds (g)	Fruit yield (kg h^{-1})	Fruit number per m^2	Seed number in fruit Per m^2	Oil yield (kg h^{-1})	Essence yield (kg h^{-1})	Protein yield (kg h^{-1})	Phenolic compounds (%)
control	Control	448.17e	109.93 e	29600f	2.60d	913.3e	220.22e	3.12e	107.74d	65.79b
Control	One time	461.67de	115.93 e	3000ef	2.63d	937.0e	226.24ed	3.24de	111.89d	65.88b
Control	Two times	462.67de	120.06 e	30800ef	2.63d	954.0e	227.35ed	3.45de	112.05d	66.23ab
Control	Three times	562.00cd e	145.60 d	33200de f	2.83c	1211.0d	276.38cd e	3.06cde	138.79cd	66.32ab
Seed inoculation	Control	598.33bc d	149.20 d	34133de f	2.83c	1223.3d	293.79bc d	4.28bcd	147.60bc	66.32ab
Seed inoculation	One time	605.67bc d	150.93 d	35733cd e	2.86bc	1298.0d	297.36bc d	4.33bcd	149.33bc	66.33ab
Seed inoculation	Two times	699.47ab c	171.03 c	37333cd	2.93abc	1418.3c	344.26ab c	5.13abc	173.68ab c	66.46ab
Seed inoculation	Three times	706.57ab c	181.00 b	38000cd	2.96abc	1467.0b c	347.50ab c	5.18abc	175.19ab	66.70ab
Acid spraying	Control	716.67ab	184.36 b	40267bc	3.00ab	1514.6b	351.81ab	5.25ab	177.58ab	67.04ab
Acid spraying	One time	768.33a	194.53 a	41200ab c	3.00ab	1702.0a	378.40a	5.74a	191.12a	67.64ab
Acid spraying	Two times	817.67a	197.89 a	4333ab	3.03a	1712.0a	398.70a	6.09a	202.89a	67.895ab
Acid spraying	Three times	825.00a	198.76 a	46533a	3.03a	1717.6a	406.10a	6.20a	205.72a	67.17a

Note: Each column having non-common letters indicates the significant difference at $p < 0.05$.

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Seed number per fruit

The results of the present study indicated that the effects of seaweed, salicylic acid, and their mutual effects on seed number per fruit were significant at $p < 0.01$. The highest seed number per fruit was related to the effect of seaweed A3 (1661.58), while the lowest to A1 (1006.92). The effect of salicylic acid spraying on the seed number per fruit was the highest in S4 (1464.11), while the lowest to S1 (1213.00; Table 2). Salicylic acid increased the number of reproductive units and seed filling and the number of seeds per plant increased in this way (Zamaninanejad *et al.* 2013). Seaweed and chemical fertilizer have also been effective on seed traits (number of seeds per plant and the weight of 1000 seeds). Generally speaking, the existence of sufficient nutrients in the soil will result in the improved nutrition of the plant, elevated efficiency of converting photosynthetic materials to dry substances, and thus the upraised number and the seed weights (Mukesh *et al.* 2013).

Oil yield

The results of variance analysis of the present study indicated that the effect of seaweed and salicylic acid on oil yield were significant ($p < 0.01$). However, the mutual effect of seaweed and salicylic acid spraying on oil yield was not significant ($p > 0.01$). The highest oil yield was related to the effect of seaweed A3 (383.75 kg ha^{-1}), while the lowest to A1 (242.79 kg ha^{-1}). The effect of salicylic acid spraying on oil yield was attributed to S4 (350.32 kg ha^{-1}), while the lowest to S1 (288.61 kg ha^{-1}; Table 2).

Salicylic acid spraying resulted in the increased plant yield, upraised levels of pure photosynthesis, Rubisco carboxylation efficiency, and photosynthetic enzyme activities (Fariduddin *et al.* 2003). Elevated oil yield as a result of salicylic acid spraying is likely associated with antioxidant response protecting the plant, and the upraised seed number subsequently resulted in the increased oil yield (Singh & Usha 2003).

Essence yield

The results of variance analysis of the present study indicated that the effect of seaweed on essence yield was significant ($p < 0.01$). The effect of salicylic acid spraying on essence yield was also significant at 5%. However, the mutual effect of seaweed and salicylic acid spraying on essence yield was not significant ($p > 0.05$). The highest essence yield was related to the effect of seaweed A3 (5.82 kg ha^{-1}), while the lowest to A1 (3.53 kg ha^{-1}). The effect of salicylic acid spraying on essence yield was the highest in S4 (5.23 kg ha^{-1}), while the lowest in S1 (4.21 kg ha^{-1}; Table 2). Other studies have also indicated that applying salicylic acid elevates the essence common sage which is a medicinal plant (Rowshan *et al.* 2010).

Protein yield

The results of variance analysis of the present study indicated that the effect of seaweed on protein yield was significant ($p < 0.01$). The effect of salicylic acid spraying on protein yield was also significant at $p < 0.05$. However, the mutual effect of seaweed and salicylic acid spraying on protein yield was not significant ($p > 0.05$). The highest protein yield was related to A3 (194.39 kg ha^{-1}), while the lowest to A1 (120.25 kg ha^{-1}). The effect of salicylic acid spraying on protein yield was the highest in S4 (176.74 kg ha^{-1}), while the lowest to S1 (144.39 kg ha^{-1}; Table 2). Some authors have reported that salicylic acid increases the production of secondary compounds in medicinal plants (Rowshan *et al.* 2010). Khan *et al.* (2010) reported that the application of salicylic acid results in the elevated growth rate and photosynthesis of the plant, and will subsequently leads to an upraised yield.

Fruit number

The results of variance analysis of the present study indicated that the effects of seaweed and salicylic acid spraying on fruit number was significant ($p < 0.01$). However, the mutual effect of seaweed and salicylic acid spraying on fruit number was not significant ($p > 0.05$). The highest fruit number was related to A3 (2.99), while the lowest to A1 (2.68). The effect of salicylic acid spraying on fruit number was the highest in S4 (2.98), while the lowest value to S1 (2.80; Table 2). Salicylic acid increases the number of reproductive units and seed filling. Hence, the number of seeds per plant elevates (Zamaninanejad *et al.* 2013). The effect of seaweed essence on improving plant growth is owing to the existence of a series of various compounds that are likely to affect one another at different densities. However, the interaction of these compounds are still unknown (El Gamal & Ahmed 2016).

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Phenolic compounds

The results of variance analysis of the present study indicated that the effects of seaweed, salicylic acid spraying, and also their mutual effects of on phenolic acid were not significant ($p > 0.05$). The highest phenolic acid was related to A3 (67.30), whereas the lowest to A1 (66.30). The effect of salicylic acid spraying on phenolic was the highest in S4 (66.81), while the lowest in S1 (66.66; Table 2). Salicylic acid, ortho-hydroxy, and benzoic acid belong to phenolic acids which can regulate the plant growth (Havate *et al.* 2010). The bioactive substances extracted from seaweed are largely used in agricultural and garden products all over the world. In most of the cases, its positive useful effects have been reported especially for ielevating the qualitative and quantitative yield of different products (Craigie *et al.* 2011).

CONCLUSION

The results of variance analysis of the present study indicated that the effect of seaweed was significant on all traits except for phenolic compounds. However, the effect of salicylic acid was not significant on all traits except for fruit yield and phenolic acids.

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