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Response of Prickly Oil Lettuce (*Lactuca scariola* L.) To Uniconazole and Irrigation with Diluted Seawater

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Abstract: This study was carried out during winter seasons 2005/2006 and 2006/2007 in the greenhouse of the National Research Centre, Dokki, Giza, Egypt in order to investigate the response of growth, yield and seed quality of prickly oil lettuce (*Lactuca scariola* L.*)* to irrigation with different concentrations of diluted seawater (control (fresh water), 2500 and 5000 ppm) and spraying with growth retardant uniconazole (zero, 150 and 300 ppm). The plant height, dry weight and seed yield / plant (g) were significantly affected by increasing saline water in irrigation water. The results indicated that increasing of salinity up to 5000 ppm resulted in a decrease in the previous traits. Such reduction in seed yield due to increasing saline water estimated by 36% in comparison to the treatment received normal irrigation (control). Seed oil % was not affected by increasing salinity level and no trend was observed. Application of uniconazole gradually decreased the plant height and dry biomass by increasing uniconazole concentration from Zero to 300 ppm. On the other hand, seed yield was significantly increased by increasing uniconazole concentration from zero to 150 and/or 300 ppm, estimated by 46.98 and 81.88%, respectively. Proline ((μ mol g^{-1} fresh weight) was more accumulated in fresh leaves at high saline water and also the highest values of osmotic potential was noticed under the same conditions. The results also show that proline and osmotic potential contents were the highest at high salinity and uniconazole concentrations. Fatty acids contents were less affected in concern saline water or unicnazole treatments, however oleic acid was decreased at low or high salinity, whereas, linolenic acid was decreased by increasing uniconazole concentration up 300 ppm and also when irrigated with moderate saline water.

Key words: Diluted seawater · prickly oil lettuce · *Lactuca scariola* L. · growth · yield · proline · fatty acids

INTRODUCTION

The increase of salinity in arid and semi arid lands has become a problem of great concern in agriculture. It is believed that about 7% of the total surface over the world is salt affected [1]. Egypt, like other developing countries of the arid and semi arid regions faces four major problems namely, high rate of population increase, limited natural resources of good quality water, existence of salt affected land, shortage of food and feed. Therefore, studies should be oriented towards solving these problems and in particular to the management of salt affected lands [2]. The fresh water resources available for agriculture are declining quantitatively and qualitatively. Therefore, the use of lower quality supplies will inevitably be practiced for irrigation purposes to maintain economically viable agriculture. A biotic environmental stresses among which salt stress is considered as one of the most prevalent are the main cause of yield reduction in plants. In saline environments plants are directly exposed to osmotic stress resulting from a low external water potential induced by high salt concentration in the soil [3]. Therefore, plant show accumulation of osmotically active substances to high levels to create a water potential to facilitate inward water movement, either by uptake of inorganic ions from the external medium or by synthesis of organic solutes [4, 5].

Recent researches revealed that growth regulating substances, which in minute quantities are capable of modifying the metabolism of plant quantitatively and qualitatively.

Furthermore, many investigators reported that growth retardant can mitigate the effect of salt stress and promote salt tolerant [6-8]. Also, amongst

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various triazoles developed as plant growth regulators, uniconazole was found to be the most effective stress protection. Uniconazole is one of triazole growth retardant of anitgibberelllin nature [9, 10]. These compounds are known to play an important role in overcoming various environmental stresses [11, 12].

Prickly oil lettuce (*Lactuca scariola* L.) is an erect annual herb belonging to the family Asteraceae. It has been cultivated in Upper Egypt since ancient times to its higher content of oil. Nowadays, it was cultivated in limited area in Upper Egypt in winter season by intercropping with other crops, so it may be good idea to increase its cultivated area in the newly reclaimed sandy soils, especially in South valley, Sinai and also in Western Nobaryia regions. The problems of extending of cultivation in these areas are not only confined to shortage of water but also to the component of salinity of underground water.

Therefore, the main objective of this study was to evaluate the effect of different concentrations of uniconazole on growth, yield and seed quality of prickly oil lettuce plants grown under different levels of diluted seawater.

MATERIALS AND METHODS

A pot experiments were carried out during the winter seasons 2005/2006 and 2006/2007 in the greenhouse of the National Research Center, Dokki, Giza, Egypt in order to investigate the effect of different concentrations of uniconazole (zero, 150 and 300 ppm) on growth, yield and seed quality of prickly oil lettuce (*Lactuca scariola* L.) plants grown under different levels of diluted seawater (control (fresh water), 2500 and 5000 ppm). Growth retardant uniconazole, chemically known as (E)-1-(P-chlorophenyl)-4,4-dimethyl-2(1,2,4-triazole-1-yl)-1-peten-2-ol[kindly supplied by the Scientific Bureau of Sumitomo Chemical Co. Ltd., Zamalik, Cairo, Egypt].

Plastic pots (40 cm diameter and 40 cm depth) were filled by 20 kg clay soil and arranged at factorial experiments in complete randomize design with 10 replicates for each treatment. The analysis of soil used was carried out following the methods described by Jackson [13] and data presented in Table 1.

Seeds of prickly oil lettuce (local variety) mainly were collected from Upper Egypt (Esna region) were sown at November 20 in the two successive winter seasons. Thinning was practiced at 30 days after planting to leave 2 plants / pot till harvest. Phosphorus and potassium fertilizers were added before sowing at a rate of 6.0 and 3.0 g/ pot of calcium super phosphate (15.5% P_2O_5) and potassium sulphate (48-50% K_2O), respectively. Nitrogen fertilizer was applied as two equal portions at a rate of 0.60 g/pot for each in form of ammonium nitrate (33.5% N) at 30 and 45 days after planting. At 45 days after planting irrigation with saline water was started. Portable water available in the experimental site was used to dilute seawater (mainly obtained from Suez Gulf) in order to get water with different salts concentrations. Chemical analysis of the diluted seawater (12.5 and 25%) was carried out according to the procedures applied by the U.S Salinity Laboratory Staff [14] and results are presented in Table 2. Plants were irrigated with adequate amount of either tap water (control treatment) or diluted seawater at the level of 65% of total holding capacity by weighing the pots for each treatment regularly and after three consecutively irrigations with saline water, extra amount of fresh water was added for leaching purpose.

Table 1: Physical and chemical properties of the soil

Soil properties	Values
Coarse sand %	6.50
Fine sand %	38.00
Silt %	37.00
Clay %	18.50
Texture	Clay
pH	7.90
E.C (dSm^{-1})	0.97
Organic matter %	1.89
Ca CO_3 %	0.72
N (ppm)	153.00
P (ppm)	3.50
K (ppm)	263.00
Cl^- (ppm)	85.20
Ca^{++} (ppm)	36.00
Mg^{++} (ppm)	7.20
Na^+ (ppm)	63.00

Table 2: Chemical analysis of the diluted seawater for irrigation in the greenhouse

	Seawater concentration%	
Characters	12.5	25.0
pH	8.0	8.0
TDS ($mg\ L^{-1}$)	6.0	13.5
Na ($mg\ L^{-1}$)	1910.0	3660.0
K ($mg\ L^{-1}$)	54.6	117.0
Ca ($mg\ L^{-1}$)	96.0	160.0
Mg ($mg\ L^{-1}$)	216.0	420.0
HCO_3 ($mg\ L^{-1}$)	597.0	683.0
Cl ($mg\ L^{-1}$)	3690.0	7460.0

At 60 and 90 days after planting total soluble solids % in the cell sap was estimated by using Hand Refractmeter and the corresponding values of osmotic potential (atm.) were then obtained from tables given by Gusev [15]. At the same two growth stages, proline accumulation ((μ mol g^{-1} fresh weight) in fresh leaves was estimated according to Bates *et al.* [16]. At 90 days after planting a representative sample was taken for determining the plant height (cm) and dry weight/plant (g). At harvest time, seed yield (g/plant) and 1000-seed weight (g) were estimated. Seed oil percentage was estimated by using Soxhlet apparatus and petroleum ether 40-60°C as a solvent according to the method described by A.O.A.C. [17]. Fatty acids composition of oil was also determined by using Gas Liquid Chromatography, the methyl esters were prepared according to Stahl [18] using Benzene: Methanol: Sulphuric acid (conc.) as a ratio of (10:86: 4).

The obtained data were statistically analyzed as factorial experiments in complete randomize design according to Snedecor and Cochran [19] and the combined analysis was done according to Steel and Torrie [20], the treatments means were compared using LSD test and 5% of probability.

RESULTS AND DISCUSSION

Growth and yield: Data presented in Table 3 show that increasing salinity concentration in irrigation water significantly decreased growth and dry biomass production of prickly oil lettuce, however the plants received moderate saline water (2500 ppm) increased the plant height in comparison to plants irrigated with fresh water. This means that the oil lettuce plants seem to be more tolerant to low salinity level. On the other hand, increasing salt concentration in irrigation water up to 5000 ppm resulted in a reduction in plant height estimated by 16.2% in comparison to control plants. The stimulatory effect of moderate salinity on the growth of some halophytic plants was reported by Ashour *et al.* [2]. This may be also attributed to the low plant growth which it's adversely effects of a specific ion concentration exceeds their thresholds and become toxic. Salts may be also reducing plant growth by reducing the water potential or by interfering with nutrient uptake. These results are also in agreement with those obtained by Abo El-Kheir *et al.* [8], Ahmed [21] and Mekki *et al.* [22]. The retardation in growth also my be attributed mainly to the osmotic inhibition of water absorption. Salinity increases the amount of work necessary to counteract osmotic and ionic stresses for normal cellular maintenance, as a consequence, these are less energy left for growth requirements [23].

Table 3: Growth and yield of prickly oil lettuce in response to irrigation with different diluted seawater concentrations

Seawater ppm	Plant height cm	Dry weight (g/plant)	Seed yield (g/plant)	1000-seed weight g	Seed oil %
Control	65.42	17.05	5.09	1.86	36.01
2500	68.71	16.72	4.43	1.75	35.93
5000	57.61	11.60	3.26	2.00	36.62
LSD at 5%	3.77	1.76	0.42	0.16	---

Dry matter production was also significantly decreased due to further increasing salinity in irrigation water (Table 3). There is no significant differences between the treatment received moderate saline water and the treatment received normal irrigation, whereas, the plants irrigated with high saline water (5000 ppm) produced lesser dry matter than control treatment, such reduction estimated by 32%. On the other hand, the increase in dry matter production in treatment received moderate saline water was insignificant compared to the control plants. In general the plant biomass is dependent absolutely on the growth of plants, the stress caused by the ions concentrations allows the water gradient to decrease, making it more difficult for water and nutrients to move through the root membrane [24]. In this concern, accumulation of salts in the root zone affect plant performance through creation of water deficit and disruption of ions homeostasis [25] which in turn cause metabolic dysfunctions. These results were also confirmed by Heakal *et al.* [26].

Seed yield / plant was significantly decreased due to increasing salinity in irrigation water (Table 3). The plants irrigated with fresh water had the highest seed yield/plant (5.09 g), whereas, the lowest was recorded with treatment received high saline water (3.26 g). The reduction in seed yield due to high saline water estimated by 36% compared to the treatment received normal irrigation. At the same time, a slight reduction in seed yield was observed when plants irrigated with low saline water (4.43 g). The reduction in seed yield due to salinity may be attributed to the depression in the vegetative growth. In this concern, El-Saidi [23] pointed that the presence of sodium in irrigation water increases the exchangeable sodium in the colloidal system of the soil and this results is the deterioration of soil physical properties which in turn affect the plant growth and productivity. Similar confirmation was reported by Abo-El-Kheir *et al.* [8] Ahmed [21] and Mekki *et al.* [22]. Data presented in Table 3 also show that 1000- seed weight was significantly affected by irrigation with saline water, increasing of salinity in irrigation water resulted in an increase in seed index and the differences between two salinity levels were significant. The increase of 1000-seed weight under high saline water my be attributed to more translocation of total carbohydrates content with increasing salinity levels and these results were confirmed by Munns and Termaat [3], who suggested that carbohydrates are more accumulated as a result of decrease in utilization.

Seed oil percentage was not affected by irrigation with saline water; data in Table 3 indicated that the control and other two salinity treatments approximately had the same oil percentages, this meaning that increasing salinity concentrations in irrigation water did not affect on seed oil content of prickly oil lettuce. On the contrary, Mekki *et al.* [22] on sunflower pointed that increasing salinity level up to 8000 ppm of NaCl in irrigation water resulted in a decrease in seed oil content by 24.09%.

Concerning, the effect of uniconazole on growth and yield, data in Table 4 show that spraying oil lettuce plants with uniconazole up to 300 ppm decreased plant height and a gradually decrease in plant height was noticed by increasing uniconazole concentration from zero to 150 and / or 300 ppm. Such reduction was estimated by 19.13 and 28.83%, respectively. The depression effect of uniconazole may be attributed to the inhibition of cell division or cell elongation through functioning as antiauxins and antigebberelline, where uniconazole is classified as growth retardant, such a retarding effect was due to reduction of internodes length [9, 27]. The retardation of plant height may be also attributed to inhibition of ent-kaurene oxidase, which catalyzes the sequential oxidations from ent-kaurene to kaurenoic acid in the early sequence of GA biosynthesis [28-30]. Similar observations were reported by Ramadan [31], Wang and Chen [32] and Bekheta *et al.* [33].

In this regard, dry matter accumulation was also significantly decreased by increasing uniconazole concentration up to 300 ppm, therefore the insignificant differences between untreated plants and plants treated with low uniconazole concentration was noticed. Increasing uniconazole concentration up to 300 ppm resulting in a decrease of dry matter accumulation estimated by 21.13%. The reduction in biomass production may be attributed to the retardation of growth due to the direct effect of uniconazole. The effect of uniconazole may prevent excessive vegetative growth and improve translocation on photosynthates from source to sink. Similar findings are in agreement with those obtained by Bekheta *et al.* [33], Imam *et al.* [34] and Mekki and El- Kholy [35].

Table 4: Growth and yield of prickly oil lettuce in response to uniconazole

Uniconazole ppm	Plant height cm	Dry weight (g/plant)	Seed yield (g/plant)	1000- seed weight g	Seed oil %
Zero (control)	76.07	16.75	2.98	1.97	36.83
150	61.52	15.42	4.38	1.77	35.23
300	54.14	13.21	5.42	1.87	36.57
LSD at 5%	3.77	1.76	0.42	0.16	---

Spraying oil lettuce plants by uniconazole significantly increased seed yield / plant (Table 4). A gradually increase of seed yield was noticed by increasing uniconazole concentration from zero to 150 and/or 300 ppm, estimated by 46.98 and 81.88%, respectively. This might be attributed to the fact that the growth retardant uniconazole improved photosynthetic activities with the crop canopy by improving the chlorophyll in the leaves and retaining a higher LAI during the sink development phase and in turn trapped more photosynthetically active radiation. The significant increase of seed yield due to uniconazole treatments are in agreement with the findings of El-Greedly and Mekki [9] and Rodrigues *et al.* [36]. The results in Table 4 also show that the seed index (1000- seed weight) was significantly affected due to uniconazole spraying, however, no trend was observed in seed index due to spraying with uniconazole in comparison to untreated plants. In this concern, seed oil percentage was not affected by uniconazole application and no trend was noticed, therefore a slight decrease in seed oil % at 150 ppm of uniconazole compared to untreated plants or the treatment received 300 ppm. On the contrary, El-Greedly and Mekki [9] pointed that spraying canola plants by 60 ppm of uniconazole produced the highest seed oil content.

Regarding, the interaction between salinity levels and spraying with uniconazole, data in Table 5 indicated that the growth and yield traits were significantly affected due to the used treatments. Plant height and dry matter accumulation were decreased under high saline irrigation and high uniconazole concentration. The lowest plant height and dry biomass production were recorded at high saline water and high uniconazole concentration. However, the plants received moderate saline water produced more dry biomass under spraying with low uniconazole treatment than that at high saline water and the same uniconazole treatment. Similar findings were reported by Abo El-Kheir *et al*, [7,8] on sunflower.

Table 5: Response of growth and yield of oil lettuce to irrigation with saline water and uniconazole treatments

Seawater ppm	Uniconazole ppm	Plant height cm	Dry weight (g/plant)	Seed yield (g/plant)	1000-seed weight g	Seed oil %
Control	Zero	80.00	20.71	3.34	1.97	37.18
	150	61.13	17.05	4.92	1.87	36.73
	300	55.13	13.41	7.02	1.74	34.39
2500	Zero	81.13	16.38	3.06	1.74	35.81
	150	66.38	17.58	4.92	1.61	32.95
	300	58.63	16.20	5.32	1.91	39.03
5000	Zero	67.10	13.17	2.55	2.19	37.49
	150	57.06	11.63	3.30	1.83	36.09
	300	48.66	10.00	3.94	1.97	36.29
LSD at 5%		6.64	3.05	0.72	0.27	---

Seed yield/ plant and 1000-seed weight were also significantly affected due to irrigation with saline water and uniconazole treatments. Data in Table 5 show that under saline irrigation treatments, seed yield was gradually increased by increasing uniconazole concentration up to 300 ppm. Plants grew under normal irrigation produced the highest seed yield (7.02 g) when plants received higher uniconazole concentration. At the same time, plants irrigated with moderate saline water produced seed yield reached to 5.32 g at the same conditions of spraying. This means that the oil lettuce plants tended to be more tolerant to moderate salinity and also to be less affected at high saline irrigation, just when spraying with growth retardants. Seed oil content was only increased as plants received moderate salinity level and high uniconazole treatment (Table 5).

Proline and Osmotic Potential: In general, the phenomenon of free proline accumulation in plants exposed to diverse environment stresses has considerable eco-physiological significance. Proline has also been known to accumulate in the leaves of many higher species subjected to salt stress [37]. The present data in Table 6 show that proline was more accumulated in oil lettuce leaves when it exposed to high saline water at 60 and 90 days plant age. In this concern, Delauney and Verma [38] pointed that the level of proline overproduction during salt stresses is assumed to be very important because it is recognized that it influence not only the osmotic potential but also minimizes the effect of salt damage. Also, the accumulation of proline was reported to serve as nitrogen storage compound and protecting of cellular structure [39]. At the same time, data indicated that osmotic potential was only significantly affected due to irrigation with saline water at 90 days plant age, while

Table 6: Proline ((μ mol g^{-1} fresh weight) and osmotic potential (Atm.) of prickly oil lettuce in response to saline water and uniconazole treatments

		Proline (μ mol g^{-1} fresh weight)		Osmotic potential (Atm.)	
		Days after planting			
Saline water ppm	Uniconazole ppm	60	90	60	90
Control	Zero	5.22	6.23	6.48	8.36
2500	150	6.24	8.52	7.38	8.36
5000	300	7.60	8.28	7.71	11.03
Mean		**6.35**	**7.68**	**7.19**	**9.24**
Control	Zero	6.77	6.47	6.54	8.76
2500	150	11.74	9.70	7.05	9.68
5000	300	6.72	5.39	8.09	10.70
Mean		**8.41**	**7.19**	**7.23**	**9.71**
Control	Zero	6.83	6.48	7.69	7.93
2500	150	9.51	7.76	6.54	10.71
5000	300	11.87	17.47	7.33	12.72
Mean		**11.07**	**10.57**	**7.19**	**10.45**
Mean values of uniconazole:	Zero	6.27	6.39	6.90	8.35
	150	9.16	8.66	6.99	9.58
	300	10.39	10.38	7.71	11.48
LSD at 5%	S	2.03	2.26	NS	0.33
	U	2.03	2.26	0.26	0.33
	SxU	3.52	3.91	0.44	0.57

S: salinity, U: uniconazole

at 60 days plant age it was insignificant, however, increasing salinity level in irrigation water increased the osmotic potential and reached to maximum under high saline water at 90 days growth stage. Osmotic adjustment of nonhalophytes growing under saline conditions was achieved mainly by active accumulation of salts, soluble organic compounds [5]. Veerangagouda *et al.* [40] also reported that the tolerance of plants to salinity by osmotic adjustment were at high concentration of NaCl, they suggested that reducing sugars and other solutes contributes more to the solute potential and therefore maintained positive turgor leading to tolerance. This finding is important for the plants breeders that enable them to isolate new genetic variability and consequently select or synthesize new varieties to be more tolerant to salt stress. According to Greenway and Munns [41] the response of different plants to salt stress depends on the degree of their tolerance, as well as, type level and duration of osmotic substrate.

Table 7: Fatty acids composition of prickly oil lettuce in response to saline water and uniconazole treatments

Seawater ppm	Uniconazole ppm	Fatty acids composition % Palmitic	Stearic	Oleic	Linoleic	Linolenic
Control	Zero	8.97	4.50	24.88	60.31	1.61
2500	150	12.10	5.91	25.64	54.35	2.00
5000	300	7.85	4.71	26.43	58.85	2.16
Mean		**9.64**	**5.04**	**25.65**	**57.84**	**1.92**
Control	Zero	13.22	5.01	24.01	55.33	2.43
2500	150	13.12	5.13	21.17	58.45	2.13
5000	300	13.41	4.78	22.21	58.40	1.20
Mean		**13.25**	**4.97**	**22.46**	**57.39**	**1.92**
Control	Zero	13.98	5.09	22.44	56.34	2.15
2500	150	13.88	4.76	20.19	59.34	2.13
5000	300	12.94	6.52	22.32	56.68	1.54
Mean		**13.60**	**5.46**	**21.65**	**57.45**	**1.94**
Mean	Zero	12.0	4.87	23.78	57.33	2.06
values of	150	13.03	5.27	22.33	57.38	2.09
uniconazole	300	11.40	5.34	23.65	57.98	1.63

Regarding, the effect of uniconazole treatments on proline accumulation and osmotic potential, data in Table 6 indicated that a significant effect was noticed due to spraying with uniconazole. Increasing the uniconazole concentration up to 300 ppm resulted in a significant increase in proline accumulation at 60 and 90 days plant age in comparison to untreated plants. The results also revealed that osmotic potential was gradually increased by increasing uniconazole concentration at two growth stages (Table 6). These results are in harmony with the findings of Abo El-Kheir *et al.* [7, 8] on sunflower. Data also show that proline accumulation and osmotic potential were significantly affected by the interaction between salinity x uniconazole treatments, proline was more accumulated in oil lettuce leaves at high salinity level and high uniconazole concentration in both growth stages. At the same time, osmotic potential was also increased at all irrigation treatments and high uniconazole concentration, however, under high salinity level the osmotic potential was higher than other treatments. These results are in agreement with findings of Badr [42] who reported that the highest values of osmotic potential were recorded when sunflower plants was subjected to high salt stress and untreated by paclobutrazole.

Fatty Acids Composition: Gas liquid chromatographic analysis cleared that the oil of prickly oil lettuce seeds contained palmitic (16:0), stearic (18:0) as saturated fatty acids and oleic (18:1), linoleic (18:2) and linolenic (18:3) as unsaturated fatty acids (Table 7). The results indicated that the two major components of total fatty acids are oleic and linoleic acids in oil of prickly oil lettuce and that previously reported by Mekki *et al.* [43]. In general, the all fatty acids were less affected by the treatments used, however, palmitic acid was increased under moderate or high saline water in comparison to control plants, stearic acid seem to be the same values. Oleic acid was decreased by using moderate or high saline water in comparison to the treatment had normal irrigation, at the same conditions linoleic and linolenic acids had the same values. This means that the prickly oil lettuce plants were not affected by salinity and it was to be more tolerant to high salinity. Also the changes of fatty acids content may be attributed mainly to genetic variability for each crop.

In this concern, spraying plants with uniconazole concentration did not affect on changes of fatty acids content, so increasing the uniconazole concentration, palmitic, stearic, oleic and linoleic had no response to uniconazole concentration, whereas, increasing uniconazole concentration up to 300 ppm decreased the linolenic content. In general, the plants received normal irrigation and 150 or 300 ppm of uniconazole recorded the highest values of oleic acid, while linoleic acid was increased under normal irrigation and untreated with uniconazole or when plants subjected under moderate saline water and 150 ppm of uniconazole. Linolenic acid was only decreased by increasing uniconazole concentrations; it was reached to the lowest content at moderate saline water and high uniconazole concentration (Table 7). The presence of linolenic acid in vegetable oils at low content is desirable, because the presence of this acid at higher content in the oil makes it easily oxidized to give unpleasant odors. Similar findings were pointed by Mekki and El- Kholy [35] on oil seed rape.

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