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Some Active Ingredients, Total Protein and Amino Acids in Plants Produced from Irradiated *Ambrosia maritima* Seeds Growing under Different Soil Salinity Levels

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ABSTRACT

This study was carried out at the National Center for Radiation Research and Technology during two successive seasons, of 2007/2008 and 2008/2009, respectively in pots 30 cm in diameter. The aim of this experiment was to study the response of dampsisa (*Ambrosia maritima* L.) seeds that exposed to different doses of radiation (0, 20, 40, 80 Gy) after planting in soils contain mixtures of salts. The dose rate was 0.89 and 0.87 rad sec⁻¹. The salts used were NaCl, CaCl₂ and MgSO₄ in ratio 2:2:1 with concentrations 2000, 4000 and 6000 ppm. Irradiated and un- irradiated seeds were sown in, sand-loamy, soil with mixture of salts. Also, a group of irradiated and un-irradiated seeds were sown in normal soils without salt and serve as a control, all pots irrigated with tap water until field capacity. It was observed that saline condition decreased ambrosin, protein and amino acids trend, in dampsisa shoots. While, the results obtained refer to increasing proline concentration separated as a result of uses γ -rays and salinity treatments. The extreme sensitivity of the metabolic processes of proline synthesis and degradation themselves may be of benefit by regulating metabolic processes which adversely affected by stress. So, it was concluded that γ -rays improve plant growth and increase its chemical components under saline stress condition.

Key words: *Ambrosia maritima*, amino acids, protein, minerals, ambrosin, dampsin

INTRODUCTION

Dampsisa (*Ambrosia maritima* L.) is one of the wild plants present in Egypt and different African countries of Nile valley. It belongs to subfamily Tubuliflorae, that represent a branch of family Compositae from flowering plants (Evans, 1996). Shoeb and El-Emam (1976) stated that ambrosin and dampsin considered as the most active ingredients in dampsisa plants. Ambrosin was synthesized and described by Grieco *et al.* (1982), they attribute it to a group of natural products known as psuedo-guaianolides. Drinking decoctions of dampsisa were the most commonly used remedy for schistosomiasis in upper Egypt (Kloos *et al.*, 1982). Aqueous methanol extract of dampsisa plant had hepatoprotective and antioxidant actions in drug that induce hepatotoxicity in rats (Ahmed and Khater, 2001). Gamma irradiation considered as a valuable tools in many purposes, from which, developing varieties that economically and agriculturally important and have high productivity potential. Moreover, gamma irradiation increases plant resistance to unfavorable

conditions such as salinity, water stresses, cold and in some condition insects or diseases (Jain *et al.*, 1998). The many mutant varieties which are resistant to diseases, cold, salt and with high quality, have been developed (Jain *et al.*, 1998). The increase in some of amino acids in irradiated *Arachis hypogaea* and *Phaseolus vulgaris* seeds was attributed to the breakdown of proteins (Hussein, 1998). Release of free amino acids following radiation treatment was reported in wheat grains (Srinivas *et al.*, 1972); this might be due to autolysis or proteolysis. Similar results were obtained in gamma irradiated red gram (*Cajanus cajan*) proteins (Nene *et al.*, 1975). It was cleared that amino acids were continually synthesized during water stress (Prat and Fathi-Ettai, 1990; Guerrier and Bourgeois-Chaillou, 1994; Sundaresan and Sudhakaran, 1995). The present investigation introduces a study of the response of gamma irradiated *A. maritima* seeds that grown in different levels of soil salinity and the changes that takes place in their chemical components. The main objective of this study was estimation of active ingredients (ambrosin and damsin) in flowering stage of damsisa shoots as affected by radiation treatments, salinity levels and both of them, to report the changes that take place according to aforementioned treatments and whether affects their amounts.

MATERIALS AND METHODS

Damsisa seeds (*A. maritima* L.) were obtained from the Agriculture Research Center, Ministry of Agriculture, Dokki, Egypt. Seeds were irradiated with different doses of gamma rays (0, 20, 40 or 80 Gy). The irradiation process was carried out in the National Center for Radiation Research and Technology (NCRRT) using Cesium 137 as a source of gamma rays. The dose rate was 0.89 and 0.87 rad sec⁻¹, during the two successive seasons, respectively. Seeds were sown in the first of October of 2007/2008 and 2008/2009 seasons. The soil chemical analysis was determined according to Jackson (1973). The organic matter content was determined according to Walkley and Black method (Black, 1982). Data of soil analysis were shown in Table 1.

During the two successive seasons, four levels of salinity (0, 2000, 4000, 6000 ppm) were obtained by adding the mixture of sodium chloride, calcium chloride and magnesium sulphate at ratio of 2:2:1 by weight and were mixed with soil in each pot. Plants were fertilized according to the recommendations of the Egyptian Ministry of Agriculture and Land reclamation (150 kg calcium super phosphate/fed, 100 kg potassium sulphate/fed and 100 kg ammonium sulphate/fed).

The pots (30 cm in diameter) were filled with 5 kg sand-loamy soil obtained from the farm of (NCRRT). In the two successive seasons, pots were divided into 4 groups. The first group consists of non irradiated control seeds and other three doses of irradiated seeds by 20, 40 and 80 Gy and sown in normal soil (control). The first group was also serving as control for salinity groups. Three

Table 1: Average of some physico-chemical and mechanical analysis of experimental soil during two successive seasons of 2007/2008 and 2008/ 2009

Properties	Values	Properties	Values
pH	7.40	Ca ⁺² (meq L ⁻¹)	8.40
Ec (dS m ⁻¹)	2.10	Mg ⁺² (meq L ⁻¹)	2.70
HCO ₃ ⁻	1.65	O.M (%)	1.10
Cl ⁻ (meq L ⁻¹)	12.00	Clay (%)	12.80
SO ₄ ⁻ (meq L ⁻¹)	8.45	Sand (%)	60.00
Na ⁺ (meq L ⁻¹)	10.35	Silt (%)	27.20
K ⁺ (meq L ⁻¹)	0.36	Texture class	Sandy loam

sets as mentioned above in the first group were reply for salt treatments which were 2000, 4000, and 6000 ppm. The irradiated seeds were divided into four groups according to soil salinity used in planting (0, 2000, 4000, 6000 ppm).

Ten seeds were germinated in pots with 0, 2000, 4000 and 6000 ppm and left to grow then plants were thinned to three plants per pot, all plastic pots were irrigated with tap water until field capacity.

The produced plants were air dried and then milled for extraction of ambrosin and damsins in shoots of flowering stage and HPLC analysis were carried out according to Slacanin *et al.* (1988) and Amin (1990).

- Total protein percent was obtained by multiplying the values of nitrogen concentration by 6.25, Nitrogen was estimated using micro Kjeldahl method of AOAC (1990)
- Amino acids analysis were done in amino acids laboratory at NCRRT according to method of Winders and Eggum (1966), after preparing the sample by weighing 100 mg powdered sample in glass tube containing 10 mL of 6 N HCl, the tube was sealed and kept in an oven at 110°C for 24 h for complete digestion as method of AOAC (1990). The system used for analysis was High performance amino acid analyzer (HPLC), Biochroma 20 (auto sampler version) pharmacia Biotech at NCRRT. Data analysis of chromatogram was done by Chromatography Data System Tutorial and user's Guide-version 6.7
- Determinations of K, Ca, Mg and Na were carried out in the dry material of damsisa plant shoots (Vegetative, Flowering and Fruiting stages). The wet digestion of 0.5 g plant material with sulphuric and perchloric acids was carried out on shoots as reported by Piper (1947). Flame photometer apparatus (CORNING M 410) was used to determine K and Na, atomic absorption spectrophotometer (GBC, 932 AA) was used to determine Ca and Mg

RESULTS AND DISCUSSION

Ambrosin and damsins: Some active ingredients of *A. maritima* shoots during flowering stage were shown in Table 2. It was observed that gamma radiation increase ambrosin percentage in damsisa shoots produced from seeds treated by 20, 40 or 80 Gy doses. The highest level of salinity 6000 ppm was noticed to increase ambrosin percentage in shoots produced from different doses of radiation as compared by normal, control. Meanwhile, all treatments used at different levels of salinity either irradiated or un-irradiated control for salinity mostly produced shoots having high

Table 2: Average percentage of active ingredients as affected by γ -rays and soil salinity in *A. maritima* L. shoots at flowering stage during 2008/2009 season

Salinity (ppm)	Dose (Gy)									
	Ambrosin					Damsin				
	0	20	40	80	Mean A	0	20	40	80	Mean A
0	0.41	0.39	0.41	0.67	0.47	2.17	1.43	1.07	0.97	1.41
2000	0.45	0.56	0.41	0.43	0.46	1.43	0.81	1.50	0.97	1.18
4000	0.41	0.65	0.52	0.53	0.53	0.89	0.89	1.67	0.90	1.09
6000	0.57	0.49	0.43	0.59	0.52	0.81	0.90	0.97	0.90	0.90
Mean b	0.46	0.52	0.44	0.55	1.32	1.01	1.30	0.94		
LSD 5%	A = NS B = NS A*B = NS					A = 0.33 B = 0.26 A*B = 0.52				

A: Salinity, B: Irradiation, A*B: Interaction and LSD: Least significant difference

ambrosin percentage than the control set, same findings were obtained by Khater *et al.* (2002), on damsisa plants irrigated with saline water levels. The opposite is true in percentage to damsins content, except for 40 Gy dose. The results declare that shoots produced from seeds treatment by 40 Gy contain more damsins percentage than its corresponding control in different salinity sets.

It was found that damsins percentage increase by 4.9, 87.6 and 19.7% above its corresponding control at 2000, 4000 and 6000 ppm soil concentration. Sangwan *et al.* (2001) stated that the production of essential oil not only depends upon the metabolic state of the source tissues but also may be integrated with the stress factors. Razmjoo and Sabzalian (2008) indicated that increased salinity significantly reduced essential oil content of chamomile. Ashraf and Harris (2004) also showed that oil content in the seed of medicinal plant, bishops weed (*Ammolei majus*), was decreased consistently with increase in external salt levels. Meanwhile, Agastian *et al.* (2000) noticed that all major processes such as photosynthesis, protein synthesis and energy and lipid metabolism are affected, during the onset and development of salt stress within a plant, Baher *et al.* (2002), on basil, Hendawy and Khalid (2005) on *Salvia officinalis* and Khalid (2006) on *Ocimum americanum*.

Protein: There are fluctuations on protein% in damsisa shoots at the three different stages of growth during two seasons as shown in Table 3. But it was clear that shoots of plants that grown in soil under 4000 or 6000 ppm salt concentration mostly had the highest percentage of protein as

Table 3: Total protein percentage as affected by different doses of γ -rays and soil salinity in shoots of *A. maritima* L. shoots during 2007/2008 and 2008/2009

Salinity (ppm)	Dose (Gy)									
	2007/2008					2008/2009				
	0	20	40	80	Mean A	0	20	40	80	Mean A
Vegetative stage										
0	10.81	14.94	16.44	17.38	14.89	14.06	20.31	19.75	16.94	17.77
2000	10.50	18.88	18.19	16.00	15.89	20.69	12.56	12.13	21.94	16.83
4000	10.25	24.88	22.88	22.38	20.10	31.31	16.75	18.63	26.81	23.38
6000	8.81	18.25	17.50	36.13	20.17	20.00	17.06	39.00	26.44	25.63
Mean b	10.09	19.23	18.75	22.97		21.52	16.69	22.38	23.03	
LSD 5%	A = 0.39	B = 0.48	A*B = 0.9			A = 0.71	B = 0.95	A*B = 1.9		
Flowering stage										
0	12.44	29.75	20.75	16.38	19.83	20.50	14.81	23.56	19.44	19.58
2000	8.69	30.56	18.56	18.13	18.98	15.81	23.31	26.38	18.81	21.08
4000	6.25	25.38	25.81	16.63	18.52	16.44	21.38	25.88	22.50	21.55
6000	16.13	26.25	35.19	31.63	27.30	23.31	11.44	16.38	24.44	18.89
Mean b	10.88	27.99	25.08	20.69	19.02	17.74	23.05	21.30		
LSD 5%	A = 0.73	B = 0.53	A*B = 1.1			A = 0.80	B = 0.61	A*B = 1.22		
Fruiting stage										
0	12.13	8.63	33.63	27.19	20.40	23.19	23.00	20.81	16.44	20.86
2000	9.50	30.94	26.75	21.19	22.10	17.50	22.25	26.56	20.13	21.61
4000	22.25	18.75	19.50	22.50	20.75	22.56	25.88	14.00	16.19	19.66
6000	31.75	19.69	16.00	14.44	20.47	24.63	17.06	20.31	18.69	20.17
Mean b	18.91	19.50	23.97	21.33	21.97	22.05	20.42	17.86		
LSD 5%	A = 0.65	B = 0.57	A*B = 1.13			A = N.S	B = 0.73	A*B = 1.46		

A: Salinity, B: Irradiation, A*B: Interaction, LSD: Least significant difference

compared by control normal soil. At vegetative stage, the mean of protein percentage was 20.10% and 23.38% for plants grown at 4000 ppm soil concentration comparing to unstressed control (14.89%, 17.77%) in control set for the first and second season, respectively. In contrast to the above results, a progressive and consistent decrease in the percent of total protein were found. El-Mogy (1999) showed that the moderate and higher treatments of soil salinity (0.2, 0.4%) tended to decrease the nitrogen percentage and protein content of *A. maritima* leaves compared to control. Whereas, the maximum content of protein was obtained when using soil salinity of 0.1%, same finding on *A. maritima* was obtained by El-Sanafawy (2000). Salinity affecting both water absorption and biochemical processes such as N_2 and CO_2 assimilation and protein synthesis (Delfine *et al.*, 1999).

Also, in gamma radiation treatments the 20 or 40 Gy dose of radiation produced the highest protein concentration as compared by un-irradiated control. Also, significant changes were observed as a result of sowing irradiated seeds in 4000 or 6000 ppm and producing plants having highest protein percentage mainly at first and second stages of growth at 2007/2008 season and at second stage of growth at 2008/2009 season. Prasad *et al.* (1997) found that both NH_4 and NO_3 -N increased with increasing salinity up to 5 dS m^{-1} . The effect of NaCl salinity in presence or absence of the irradiated lupin (*Lupinus termis* L.) seeds with gamma rays, on nitrogen assimilation and ion uptake was investigated by Khodary (2004). Significant decreased in the contents of protein and amino acids, were observed upon exposure (0, 500, 1000, 2000, 3000 ppm). On the other hand, in seeds irradiated with gamma rays (10, 25, 100 Gy) these nitrogenous fractions were increased after NaCl treatments, the effect was more pronounced particularly with 25 Gy. Also, Hussein (2010) observed that treating seeds before sowing by gamma rays (50-250 Gy) increased total protein contents in the produced seedlings of mungbean seeds. Radiation caused oxidative injury by accelerating free radical generation in living systems. The primary damage induced by ionizing radiation is modified in enzymatic repair processes (Alikamanoglu *et al.*, 2007). It was previously shown that gamma irradiation significantly influences the cell metabolism and protein synthesis in plant meristem cells (Casarett, 1968). According to the results obtained in the present study, it was observed that increased gamma dosage caused a reduction of total protein concentration. However, plants irradiated at relatively low dosage (10, 20 Gy) displayed a higher total protein concentration compared to their non-irradiated counterparts. This result demonstrated that there was a direct correlation between gamma dosage and protein content. Gamma irradiation caused inhibition of tissue culture growth along with failure of RNA and subsequently of protein synthesis (Bajaj, 1970). This accounts for the lower protein concentration in plants irradiated at high dosage (70 Gy).

Amino acids: Data in Table 2 and 3 demonstrated the effect of salinity treatment and γ -rays on essential amino acids (threonine, cystine, methionine, valine, isoleucine, tryrosine and phenylalanine) and non essential amino acids (aspartic, serine, glutamic, proline, glycine, alanine, histidine and arginine) in *A. maritima* plants in the two successive seasons. The concentration of amino acids in *A. maritima* plants at flowering stage were shown in Table 4 and 5 during 2007/2008 and 2008/2009 season respectively as affected by different levels of salinity and doses of gamma rays. It was observed that the amino acid pool increased by salinity in shoot produced from seeds planted in soil with salt concentration 2000, 4000 or 6000 ppm during two seasons 2007, 2008 except 4000 ppm level of salinity at 2007 season where it's value less than control. The results obtained were similar with several reports of increase level of free amino acid pool during salt treatment in different plant species (Muthukumarasamy *et al.*, 2000; Wang and Nii, 2000). Concerning amino acids concentration as affected by γ -rays, it was observed

Table 4: Amino acids concentration (mg g⁻¹ DW) as affected by different doses of γ -rays and soil salinity *A. maritima* L. shoots at flowering stage of during 2007/2008 season

Salinity levels																

Control (ppm)				2000 (ppm)				4000 (ppm)				6000 (ppm)				

Dose (Gy)																

Amino acids	0	20	40	80	0	20	40	80	0	20	40	80	0	20	40	80
Aspartic	1.42	1.96	2.36	1.26	1.72	1.06	2.02	1.78	1.18	1.64	1.92	1.94	1.54	1.50	1.82	1.58
Threonine	0.70	1.18	1.30	0.60	0.74	0.48	0.94	0.82	0.50	0.88	0.96	1.04	0.90	0.82	0.94	0.74
Serine	0.86	1.14	1.38	0.70	0.88	0.62	1.14	1.02	0.64	0.98	1.10	1.18	1.06	0.94	1.10	0.90
Glutamic	1.90	2.92	3.36	1.56	1.98	1.24	2.60	2.32	1.44	2.36	2.58	2.68	2.36	2.24	2.50	2.06
Proline	6.60	16.22	20.70	3.12	4.98	1.98	16.00	11.68	1.38	6.92	9.50	11.80	14.90	8.24	9.80	6.50
Glycine	0.98	1.32	1.30	0.80	1.04	0.64	1.06	0.98	0.84	1.04	1.14	1.16	1.14	0.94	1.06	0.86
Alanine	1.16	1.40	1.86	1.10	1.40	0.94	1.52	1.40	0.88	1.36	1.54	1.66	1.62	1.30	1.52	1.20
Cystine	0.06	0.12	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.04	0.00	0.00	0.00	0.00	0.00	0.00
Valine	0.62	0.90	1.16	0.60	0.80	0.48	0.84	0.74	0.52	0.76	0.84	0.96	0.90	0.76	0.84	0.62
Methionine	0.02	0.04	0.22	0.00	0.06	0.00	0.02	0.02	0.00	0.02	0.08	0.18	0.04	0.08	0.02	0.04
Isoleucine	0.38	0.66	0.84	0.34	0.48	0.28	0.54	0.44	0.28	0.46	0.48	0.62	0.58	0.50	0.54	0.38
Leucine	1.12	1.44	1.86	0.90	1.22	0.78	1.34	1.18	0.74	1.26	1.38	1.54	1.50	1.18	1.38	0.98
Tryrosine	0.46	0.68	0.96	0.32	0.52	0.26	0.66	0.50	0.24	0.56	0.68	0.74	0.72	0.48	0.66	0.38
Phenylalanine	0.64	0.92	1.20	0.44	0.66	0.36	0.82	0.66	0.38	0.68	0.82	0.86	0.90	0.62	0.82	0.52
Histidine	0.28	0.72	0.86	0.20	0.30	0.16	0.52	0.32	0.12	0.28	0.42	0.40	0.54	0.30	0.40	0.34
Lysin	0.96	1.50	1.58	0.84	1.10	0.70	1.20	1.04	0.72	1.14	1.22	1.30	1.24	1.02	1.20	0.88
Arginine	0.76	3.14	2.90	0.72	1.34	0.60	1.46	1.18	0.52	1.08	1.40	1.28	1.66	1.08	1.30	1.56
Total mg g ⁻¹	18.90	36.30	43.80	13.50	19.20	10.60	32.70	26.10	10.40	21.50	26.10	29.30	31.60	22.00	25.90	19.50

Table 5: Amino acids concentration (mg g⁻¹ DW) as affected by different doses of γ -rays and soil salinity in *A. maritima* L. shoots at flowering stage during 2008/2009 season

Salinity levels																

Control (ppm)				2000 (ppm)				4000 (ppm)				6000 (ppm)				

Dose (Gy)																

Amino acids	0	20	40	80	0	20	40	80	0	20	40	80	0	20	40	80
Aspartic	2.3	2.0	2.0	1.86	1.92	2.06	2.1	1.9	2.1	2.38	2.18	1.92	2.02	1.90	1.78	1.66
Threonine	1.4	1.1	1.3	1.08	1.06	1.20	1.1	1.1	1.3	1.42	1.28	1.08	1.12	1.04	0.98	0.90
Serine	1.3	1.1	1.2	1.06	1.04	1.20	1.1	1.1	1.2	1.32	1.24	1.08	1.14	1.08	1.02	0.98
Glutamic	3.2	2.7	2.9	2.60	2.62	2.94	2.7	2.7	3.0	3.30	3.00	2.58	2.76	2.64	2.52	2.26
Proline	2.2	13.0	17.0	14.10	13.10	19.60	18.0	12.0	17.0	7.90	12.80	12.40	16.50	15.40	11.90	10.60
Glycine	1.4	1.1	1.3	1.06	1.04	1.24	1.1	1.1	1.3	1.30	1.16	1.02	1.02	0.98	0.92	0.88
Alanine	1.6	1.2	1.2	1.44	1.32	1.74	1.5	1.5	1.3	1.98	1.74	1.42	1.48	1.40	1.28	1.28
Cystine	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Valine	1.2	1.0	1.0	1.02	0.98	1.34	1.0	1.0	1.1	1.56	1.20	0.94	1.06	0.96	0.9	0.84
Methionine	0.1	0.2	0.1	0.14	0.04	0.46	0.1	0.2	0.2	0.38	0.22	0.04	0.24	0.12	0.02	0.14
Isoleucine	0.9	0.9	0.8	0.74	0.74	0.98	0.7	0.7	0.9	1.46	0.96	0.68	0.92	0.76	0.72	0.60
Leucine	1.8	1.6	1.6	1.5	1.44	2.02	1.5	1.6	1.7	2.30	1.74	1.54	1.78	1.44	1.30	1.28
Tryrosine	1.2	1.4	1.0	0.88	0.6	2.06	0.8	0.8	1.1	2.14	1.18	1.22	1.64	0.78	0.42	0.66
Phenylalanine	1.4	1.2	1.1	1.02	1.02	1.48	1.0	1.0	1.2	1.84	1.32	1.12	1.30	0.98	0.92	0.80

Table 5: Continued

Amino acids	Salinity levels															
	Control (ppm)				2000 (ppm)				4000 (ppm)				6000 (ppm)			
	Dose (Gy)															
	0	20	40	80	0	20	40	80	0	20	40	80	0	20	40	80
Histidine	0.8	0.5	0.6	0.54	0.48	0.60	0.5	0.5	0.6	0.86	0.68	0.44	0.56	0.48	0.44	0.36
Lysin	1.6	1.2	1.4	1.22	1.12	1.36	1.2	1.2	1.4	1.48	1.32	1.16	1.22	1.12	1.04	1.00
Arginine	2.7	2.0	2.4	2.06	1.52	2.24	2.1	1.4	1.9	2.60	2.02	1.48	1.96	1.86	1.40	1.26
Total mg g ⁻¹	25.0	32.0	37.0	32.40	30.00	42.50	37.0	30.0	37.0	34.20	34.00	30.10	36.70	32.90	27.50	25.50

that 20, 40 and 80 Gy dose increased levels of amino acids pool, markedly above normal control (un-irradiated and unstressed control) during two season of experiment. Also, treating seeds by γ -rays then sowing at different salinity levels mostly accumulate total amino acids pool as compared to its corresponding controls. The results revealed that glutamic acid (3.22 mg g⁻¹) was the prominent amino acid followed by arginine (2.68 mg g⁻¹) and aspartic acid (2.26 mg g⁻¹) in herb of *A. maritima* plants. Similar results were reported by Al-Jassir (1992) on black cumin, Hussein (2010) on mungbean; Swailam (2009) on sesame and Hussein and Atia (2009) on mushroom, they observed that amino acids increased above the control values.

In the first season 2007, it was observed that, in control, 2000 and 4000 ppm, total amino acid increased in plants produced from irradiated seeds by 20, 40 or 80 Gy above its corresponding control and above the normal control (unstressed and un irradiated). Meanwhile, at the second season (2008), the control and 2000 ppm only significant supremacy were observed above its corresponding control and above normal control. In other word, 6000 ppm in the first season and 4000, 6000 ppm salinity levels decrease total amino acids concentration. It was mentioned that these levels of salinity mainly at 6000 ppm inclusive marked decline in proline concentration, as compared by its corresponding control. It is well known that proline content in the leaves of many plants get enhanced by several stresses including salt stress (Lee and Liu, 1999; Hernandez *et al.*, 2000) and gamma radiation. Ali (1991) in coincidence with our results obtained, the author stated that free amino acids other than proline increased significantly with the rise of salinization level. Proline in particular was frequently recorded to be considerably accumulated more than any other free amino acids in stress plants. There are some exceptions during two seasons, in 4000 and 6000 ppm salinity set during 2008 season.

Proline serves as a storage sinke for carbon and nitrogen and a free-radical scavenger. Khodary (2004) investigated the effect of NaCl in presence or absence of gamma irradiation on seeds of *Lupinus termis*. The author determined protein, amino acids, nucleic acids and nitrate, potassium and phosphorus uptake. Significant decreases in the contents of protein, amino acids and nucleic acids were observed upon NaCl exposure (0, 500, 1000, 2000, 3000 ppm). On the other hand, in seeds irradiated with gamma rays (10, 25, 50, 100 Gy), these nitrogenous fractions were increased after NaCl treatments, particularly with 25 Gy. Proline may contribute to osmotic adjustment at the cellular level. Many investigators recorded an accumulation of amino acids especially proline in plant exposed to salt stress (Perez-Alfocea *et al.*, 1993; Rhodes and Hanson, 1993; Zidan and Al-Zahrani, 1994; Aziz and Larher, 1995; Hartzendorf and Rolletschek, 2001; Xiong and Zhu, 2002; Moghaieb *et al.*, 2004; Eraslan *et al.*, 2007; Costa *et al.*, 2008).

It is clearly shown that salinity stress increased proline content in shoots. These results indicated that the increasing in proline levels at high salinity concentration might be one of the earliest metabolic responses triggered in the translocation pathway that links the perception of many environmental stresses to the elicitation of physiological responses at the cellular level (Hare *et al.*, 1996).

Many studies suggested that proline is a protective agent of enzyme and membrane (Solomon *et al.*, 1994) and as an intracellular structure (Mudgal *et al.*, 2010) or a storage compound of carbon and nitrogen for rapid recovery from stress (Jager and Meyer, 1977). Moreover, Maiti *et al.* (2000) demonstrated that proline increased in all barley genotypes with the increase in salt stress. Also, proline is regarded as a source of energy and nitrogen for recovering tissue. So, it increased under stress levels and considered an osmotic adjuster (Costa *et al.*, 2008).

It also stabilizes sub cellular structures (membranes and proteins) and buffers cellular redox potential under stress (Bohnert and Jensen, 1996; Chen and Murata, 2002). The extreme sensitivity of the metabolic processes of proline synthesis and degradation themselves may benefit by regulating metabolic processes which are adversely affected by stress (Hare and Cress, 1997).

Hanafy Ahmed *et al.* (2002) suggested that, the high values of total free amino acids concentration under saline soil conditions may be induced as a result of reducing the rate of incorporation of free amino acid into protein. In addition, the authors mentioned that the osmotic adjustment within the cytoplasm might be maintained by synthesis of compatible solutes such as amino acids which have deleterious effects on metabolism and growth at high concentrations.

Minerals: Plant growth and the accumulation of Na⁺, K⁺, Ca²⁺ and Mg²⁺ were considered to be the most important factors related to the specific effects of ions under salt stress, Mineral investigated in *Ambrosia maritima* shoots were shown in Table 6. Salinity stress disturbs the uptake and accumulation of essential nutrients. Generally Na⁺ and K⁺ percentage increased with increasing salinity levels in soil. In the first stage of growth, Na percentage increased to 0.32% in plant having 2000 ppm, 0.26% for 4000 ppm and 0.37% at 6000 ppm soil percentage. In contrast, in control Na percentage was 0.24% at vegetative stage. But in case of K the untreated plants gave 0.34% while its values in plants grown in salty soil increase to be 1.02, 0.80 and 1.07%, for the stressed control in 2000, 4000 and 6000 ppm, respectively. Similar results have been found in number of studies as on wheat Begum *et al.* (1992), Ashraf and Rauf (2001) on maize and Hussain *et al.* (2009) on black seeds.

Also, treating seeds before sowing by gamma radiation generally increase Na percentage in damsisia plant during vegetative stage of growth as compared by its corresponding control or un-irradiated and unstressed control. Meanwhile, at flowering and fruiting stages, decreases in Na percentage were observed with some exception mainly on 40 Gy and rarely at 80 Gy where Na percentage was show similar increase as in the first stage of growth.

K⁺ percentage was higher in plants produced from seeds treated by 40 Gy and sown in soil having different salinity levels or normal soil as compared by its corresponding control. These results are in agreement with Izzo *et al.* (1996). It was observed that Na⁺ and K⁺ decreased at high salt concentration (6000 ppm) at fruiting stage, as compared by its corresponding control or normal control.

It was evident from data in the first and second stages of growth, that Ca percentage enclose more or less about 1% as affected by salinity stress in control set and in 2000 or 4000 ppm sets, while at 6000 ppm, the stressed control had 1.64% in vegetative stage. Concerning the Ca²⁺ percentage at the fruiting stage the control was 1.59%, decreased to 1.22, 1.28 and 1.07% under

Table 6: Mineral percentage as affected by different doses of γ -rays and soil salinity in *A. maritima* L. shoots during 2008/2009 season

Salinity levels	Stage														
	Vegetative stage					Flowering stage					Fruiting stage				
	Dose (GY)														
	0	20	40	80	M	0	20	40	80	M	0	20	40	80	M
Calcium															
0	1.04	1.03	1.01	0.83	0.98	0.86	0.87	0.93	0.84	0.88	1.59	1.43	1.01	1.04	1.27
2000	1.00	1.42	1.19	1.22	1.21	1.07	1.07	0.88	0.95	0.99	1.22	1.34	1.23	1.47	1.32
4000	0.92	0.95	1.22	1.14	1.06	1.33	0.91	1.24	1.35	1.21	1.28	1.26	1.46	0.87	1.22
6000	1.64	1.15	0.85	0.87	1.13	1.16	1.13	0.87	0.81	0.99	1.07	1.09	0.97	1.42	1.14
M	1.15	1.14	1.07	1.02		1.11	1.00	0.98	0.99		1.29	1.28	1.17	1.20	
Potassium															
0	0.34	0.95	1.02	0.80	0.78	1.63	0.46	1.62	0.14	0.96	1.19	1.21	0.58	1.04	1.01
2000	1.02	0.56	1.19	1.07	0.96	1.31	1.34	1.38	0.36	1.10	0.95	1.41	1.24	1.62	1.31
4000	0.80	1.09	1.45	1.14	1.12	1.35	0.95	1.65	1.16	1.28	1.43	1.28	1.46	1.60	1.44
6000	1.07	1.19	1.12	1.14	1.13	1.50	1.33	1.19	1.50	1.38	1.11	1.04	0.77	0.73	0.91
M	0.81	0.95	1.20	1.04		1.45	1.02	1.46	0.79		1.17	1.24	1.11	1.14	
Sodium															
0	0.24	0.26	0.29	0.26	0.26	0.29	0.15	0.31	0.15	0.23	0.34	0.31	0.15	0.22	0.26
2000	0.32	0.32	0.19	0.36	0.30	0.39	0.34	0.48	0.14	0.34	0.41	0.19	0.54	0.34	0.37
4000	0.26	0.31	0.41	0.29	0.32	0.31	0.22	0.39	0.32	0.31	0.24	0.17	0.24	0.29	0.24
6000	0.37	0.49	0.37	0.39	0.41	0.34	0.36	0.32	0.29	0.33	0.22	0.20	0.17	0.10	0.17
M	0.30	0.35	0.32	0.33		0.33	0.27	0.38	0.23		0.30	0.22	0.28	0.23	
Magnesium															
0	0.66	0.62	0.41	0.36	0.52	0.45	0.42	0.46	0.56	0.47	0.65	0.39	0.32	0.31	0.42
2000	0.36	0.43	0.58	0.48	0.46	0.45	0.45	0.55	0.46	0.48	0.54	0.36	0.35	0.37	0.41
4000	0.56	0.4	0.43	0.39	0.45	0.46	0.51	0.50	0.51	0.50	0.46	0.35	0.43	0.38	0.41
6000	0.51	0.45	0.34	0.31	0.40	0.36	0.31	0.30	0.26	0.31	0.45	0.32	0.32	0.39	0.37
M	0.52	0.48	0.44	0.39		0.43	0.42	0.45	0.45		0.53	0.36	0.36	0.36	

M: Median

the effect of 2000, 4000 and 6000 ppm soil salinity, respectively. The reduction in Ca was observed and similar to that, respectively El-Tarawy (1976) on *Matthiola incana*, El-Makawy (1999) on *Dimorphotheca ecklonis* and *Callistephus chinensis*, Khan (1992), Al-Harbi (1995) and Ashraf and Khanum (1997) on wheat and El-Sanafawy (2000) on *Ocimum basilicum* and *Ambrosia maritima* who reported that Ca% in plants increased with increasing soil salinity levels. Regarding the effect of gamma rays on Ca percentage in shoots of damsisa plants, it was observed that, in 2000 ppm salinity level increased Ca percentage at the first and second stage of growth. Also, the dose 40 and 80 Gy in 4000 ppm increased Ca percentage. While at fruiting stage, it was noticed a reduction in Ca percentage. These results are in agreement with those obtained by El-Tarawy (1976) on *M. incana*, El-Makawy (1999) on *Dimorphotheca ecklonis* and *Callistephus chinensis* and El-Sanafawy (2000) on *Ocimum basilicum* and *A. maritima* who reported that Ca% in plants increased with increasing soil salinity levels. The contrast was take place at flowering stage except 80 Gy. It was noticed that set treated by different doses of radiation and sown in soil, having 4000 ppm salt concentration, produced plants having high Ca²⁺ percentage as compared by its corresponding control, Table 6.

Concerning Mg^{2+} percentage in the shoots of *A. maritima* plants, the data indicated that decreases were detected with increasing salinity concentration in soils during the three stages of growth as compared by unstressed and un-irradiated control as shown in Table 4. Concerning the effect of γ -rays, it was observed that at the first stage of growth that all radiation dose decreased $Mg\%$ comparing with normal control (unstressed and un-irradiated). But, the opposite takes place at 2000 ppm set; all plants produced from 20, 40 or 80 Gy doses had higher $Mg\%$ than its corresponding control at vegetative and flowering stage. The highest percentage of Mg^{2+} as compared by those in control set were in 40 and 80 Gy dose at control set. Meanwhile, at 4000 and 6000 ppm, $Mg\%$ decreased in plant produced from radiation dose (20, 40, 80 Gy) as compared by its corresponding control at vegetative stage. The results are in accordance with those of El-Tarawy (1976) on *M. incana*, El-Makawy (1999) on *D. ecklonis* and *C. chinensis* and El-Sanafawy (2000) on *Ocimum basilicum* and *Ambrosia maritima*, who stated that $Mg\%$ in the leaves decreased with increasing soil salinity levels. Also, similar results were obtained by Youssif *et al.* (2010) on *Tetragonia tetragonoides* in shoots. $Mg\%$ in shoots with different doses of radiation comparing with stressed control.

CONCLUSION

It was concluded that gamma rays can alleviate harmful effect of soil salinity and this appears in an increase of most amino acids, proteins and some chemical components of damsoya plants that grow under salinity stress condition.

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