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EFFECT OF SALINITY ON BIOMASS PRODUCTION AND ACTIVITIES OF SOME KEY ENZYMATIC ANTIOXIDANTS IN KOCHIA (*KOCHIA SCOPARIA*)

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Abstract

Soil salinity is a major constraint to food production due to its negative impact on crop yield. Kochia (*Kochia scoparia*) is a salinity-resistant plant that can widely be used as emergency forage for livestock by using saline waters and soils in desert ecosystems. In order to investigate physiological mechanism, antioxidants activity and potential production of Kochia in response to different levels of salinity, an experiment was performed in a split plot based on randomized complete block design with three replications. Saline waters (5.2, 10.5 and 23.1 dS m^{-1}) and three Kochia ecotypes (Birjand, Borujerd and Sabzevar) were allocated as main and sub plots, respectively. The results showed that salinity did not impose any significant effect on dry matter production but relative water content (RWC) and seed yield decreased by salinity stress. In general, no positive correlation coefficient was observed between dry matter production and physiological and biochemical parameters except superoxide dismutase (SOD) at 23.1 dS m^{-1}. There was no significant difference among ecotypes in dry matter production and seed yield. Sabzevar ecotype showed the highest proline, total phenol content and peroxidase (POX) activity. Ascorbate peroxidase (APX), catalase (CTA), and superoxide dismutase (SOD) activity was higher in Borujerd ecotype, while highest soluble sugar, glutathione reductase (GR) activity and DPPH - radical scavenging activity was observed in Birjand ecotype. According to these results, Kochia has a reliable tolerance to elevated levels of salinities up to 23 dS m^{-1} and it seems that it can control oxidative stress by continuing growth.

Introduction

Salinity is one of the most important environmental factors limiting crop production of marginal agricultural soils in many parts of the world. It is estimated that about a third of the world's cultivated land affected by salinity. *Kochia scoparia* (L., Schrad) is a highly, drought and salinity resistant plant widely used as emergency forage for livestock (Gul *et al*., 2010). Kochia can establish on saline soils, not only to produce protective short-lived vegetation coverage, but also is being used as an alternative forage crop, especially in regions faced with forage shortage (Jami Al-Ahmadi & Kafi 2006). Kochia also has high forage yield potential; Kafi *et al*., (2010) reported an annual forage yield up to 11 ton ha^{-1}.

Salinity can affect growth and yield of most crops, high salinity is known to cause both hyper ionic and hyper osmotic effects in plants, leading to membrane disorganization, increase in activated oxygen species production and metabolic toxicity (Joseph & Jini, 2011). Reactive oxygen species (ROS) are highly reactive and, in the absence of any protective mechanism, can seriously disturb normal metabolism through oxidative damage toward pigments, lipids, proteins and nucleic acids (Molassiotis *et al*.,

2006). Noreen & Ashraf (2009) reported that differential salt tolerance of the radish cultivars was not found to be associated with higher antioxidant enzyme activities, and some other key metabolites, so they cannot be used as potential selection criteria for salt tolerance in the six cultivars of radish examined in their study. In this work, we examined the activities of some antioxidative enzymes, responsible for detoxifying ROS, in Kochia plants grown under salinity condition.

Material and Methods

Three Kochia (*Kochia scoparia*) ecotypes were selected from three different regions of Iran, (Borujerd, relatively cold without salinity problem; Birjand arid, warm, and relatively salty and Sabzevar warm with salinity of soils and water resources). This field study was conducted in 2008 at the Salinity Research Station (36°15´N, 59°28´E) of Faculty of Agricultural Ferdowsi University of Mashhad, Iran. The annual maximum and minimum temperatures were 42 and -27.8°C, respectively. The experiment was established as a split plot based randomized complete block design with three replications. Saline waters (5.2, 10.5, and 23.1 dS m^{-1}) and three Kochia ecotypes were allocated as main and sub plots, respectively. The source of irrigation water for low-level of salinity was the water pumped from a deep well near the site (Table 1). For the remaining two higher levels of salinity, water was transferred by tankers from ground sources in the same within a distance of 5 km. Chemical analysis of the water resources in terms of the three levels of salinity is shown in Table 1. Low salinity level (5.20 dS m^{-1}) played the role of control because previous experiments showed no negative effects of this level of salinity on Kochia growth. Amount of water that used for irrigation controlled by volume counter at each plot.

Sampling and harvest procedure: For fresh biomass used for assays, samples were taken from fully matured leaves chosen randomly. All measurements with fresh leaves carried out at the beginning of anthesis. Youngest fully expanded leaves sampled for membrane stability index (MSI) and RWC, carried in the ice for immediate determination. Samples for biochemical determination kept frozen at -80°C, until determinations. At maturity stage, harvested plants dried in an oven at 70°C until constant mass reached and total biomass and seed yield were measured.

Leaf membrane damage was determined by recording the electrical conductivity of leaf leakages (Sairam *et al*., 2002). Leaf relative water content was estimated according to Smart & Bingham (1974). Extraction and estimation of leaf proline was conducted according to the procedures described by Bates *et al*., (1973). Soluble sugars were determined based on the method of phenol-sulfuric acid (Dubois *et al*., 1956).

Table 1. Main chemical properties of the waters and soil (0-30 cm) at the study site.

	Na	Ca	Mg	K (meq.l^{-1})	SO_4	HCO_3	Cl	EC dS.m^{-1}
Water 1	32.50	8.60	9.20	0.23	15.00	2.40	34.40	5.20
Water 2	67.10	16.40	22.20	0.38	25.00	3.00	75.60	10.50
Water 3	179.80	27.00	46.80	0.31	56.10	3.20	172.40	23.10
Soil	31.10	10.60	10.20	0.75	31.30	1.80	26.80	5.80

Enzyme assays: Leaf fresh materials (0.1 g) powdered in liquid nitrogen, and homogenized in 1 ml of 0.1 M potassium phosphate buffer, pH 7.8 containing 1 mM ethylenediaminetetraacetic acid (EDTA) by a homogenizer into microtube. Insoluble materials were removed by Beckman refrigerated centrifuge at 12000 *g* for 20 min at 4°C and supernatant used as the source of enzyme extraction. One hundred microliters of supernatant was aliquots to microtubes for APX, CAT, SOD, POX and GR activity and stored at -80°C until assay enzyme activities. All the steps of antioxidant determination carried out at 4°C. APX (EC 1.11.1.11) activity was determined according to Yamaguchi *et al.* (1995). CAT (EC 1.11.1.6) activity assayed by measuring the initial rate of hydrogen peroxide disappearance according to Velikova *et al*., (2000). SOD (EC 1.15.1.1) activity assayed by monitoring the inhibition of the photochemical reduction of nitroblue tetrazolium, based on the method of Yu & Rengel (1995). POX (EC 1.11.1.7) activity estimated based on the method described by Srinivas *et al*., (1999). GR (EC 1.6.4.2) activity was measured according to Lee & Lee (2000). For determination of DPPH (1,1-diphenyl-2-picrylhydrazyl) radical scavenging activity we used Abe *et al*., (1998) method. Total phenolic content assessed by using the Folin-Ciocalteau phenol reagent method (Singleton & Rossi, 1965).

The data compiled were submitted to an analysis of variance (ANOVA) and the differences between the means were compared by LSD test ($p\leq 0\ 05$).

Results and Discussion

Dry matter: Dry matter production remained statistically unchanged in different levels of salinity, but it declined markedly at 23.1 dS m^{-1} (Table 2). Biomass production of Sabzevar ecotype was higher than tow other ecotypes but the changes in dry matter production were not statistically significant ($p\leq 0.05$) among salinity levels and ecotypes. In the present work, average dry matter production at anthesis stage was 12 $ton.h^{-1}$, that is considerable yield in saline condition and is in agreement with other reports (Salehi *et al.*, 2009; Kafi *et al*., 2010). Stimulating the growth and increase biomass production under 10.5 dS m^{-1} observed in all ecotypes (Table 2). These findings are in agreement with those of Ashraf & Harris (2004) on halophytic grasses and Kafi *et al*., (2010) on Kochia who under lined the stimulating effect of moderate salinity on the growth of halophytes. Munns & Tester (2008) pointed out that in a saline soil; halophytes could use ions for osmotic adjustment and decrease cost of salinity stress.

Seed yield: Seed production exhibited a significant ($p\leq 0.05$) decline under salinity treatments and reaching the minimum rate in the plants treated by 23.1 dS m^{-1} (Table 2). There were no significant ($p\leq 0.05$) differences among ecotypes in seed yield, but Birjand ecotype produced the highest seed yield (2.3 $ton.h^{-1}$) (Table 2). Interaction between salinity and ecotypes showed that increased salinity cause seed yield loss but it was not significant ($p\leq 0.05$) (Table 2). Seed production capacity of Kochia is acceptable, 2.90 $ton.ha^{-1}$ (Kafi *et al*., 2010). Previous work by Steppuhn *et al*., (1993) indicated that Kochia displays a reliable tolerance to salinity after establishment. In our experiment, increased salinity up to 10.5 and 23.1 dS m^{-1} decreased 7.8 and 24.9% seed production, respectively. In spite of dry matter production, effect of high level of salinity on seed yield was significant. Significant correlation between dry matter and seed yield was observed (Table 4). Because of gradual accumulation of toxic ions in shoots, most of biomass production occurred during vegetative growth stage that salt accumulation was low, but seed production was formed mainly at the end of plant life that high rate of salt accumulated. Therefore, the negative effect of salinity on seed production was more pronounced than dry matter production. No significant differences among ecotypes in seed yield production showed that seed production capacity was not different amongst ecotypes.

Table 2. Effect of salinity on dry matter (ton.h^{-1}), seed yield (ton.h^{-1}), membrane stability index (MSI) (%), relative water content (RWC) (%), proline content (mg.gdw) and soluble sugars (mg.gdw) in different Kochia ecotypes.

	Salinity	Ecotypes			Means
		Birjand	Borujerd	Sabzevar	
Dry matter (ton.h^{-1})	5.2	12.80	11.68	12.40	12.29
	10.5	11.79	12.20	13.53	12.51
	23.1	11.18	11.70	10.87	11.25
	Means	11.92	11.86	12.27	
	LSD 0.05	Salinity: 2.80	Ecotype: 1.66	Salinity × Ecotype: 2.88	
	P value	Salinity: 0.249^{ns}	Ecotype 0.850^{ns}	Salinity × Ecotype: 0.619^{ns}	
Seed yield (ton.h^{-1})	5.2	2.77	2.08	2.38	2.41
	10.5	2.00	2.32	2.33	2.22
	23.1	2.00	1.78	1.65	1.81
	Means	2.26	2.06	2.12	
	LSD 0.05	Salinity: 0.35	Ecotype: 2.18	Salinity × Ecotype: 0.64	
	P value	Salinity: 0.012**	Ecotype: 520^{ns}	Salinity × Ecotype: 0.197^{ns}	
MSI (%)	5.2	54.40	23.73	34.27	37.47
	10.5	19.53	27.50	30.05	25.69
	23.1	23.11	19.35	31.28	24.58
	Means	32.34	23.53	31.86	
	LSD 0.05	Salinity: 8.28	Ecotype: 6.97	Salinity × Ecotype: 12.07	
	P value	Salinity: 0.002**	Ecotype: 0.029*	Salinity × Ecotype: 0.002**	
RWC (%)	5.2	75.38	73.06	76.43	74.95
	10.5	75.27	68.90	73.77	72.65
	23.1	71.22	68.16	71.37	70.25
	Means	73.96	70.04	73.86	
	LSD 0.05	Salinity: 4.86	Ecotype: 2.27	Salinity × Ecotype: 3.92	
	P value	Salinity: 0.002**	Ecotype: 0.003**	Salinity × Ecotype: 0.587^{ns}	
Proline (mg.gdw)	5.2	1.95	1.40	2.28	1.87
	10.5	2.05	2.88	2.96	2.63
	23.1	2.76	5.29	7.56	5.20
	Means	2.25	3.19	4.27	
	LSD 0.05	Salinity: 0.21	Ecotype: 0.58	Salinity × Ecotype: 0.99	
	P value	Salinity: 0.001**	Ecotype: 0.001**	Salinity × Ecotype: 0.001**	
Soluble sugars (mg.gdw)	5.2	85.32	71.40	68.97	75.23
	10.5	67.01	65.86	52.80	61.89
	23.1	70.09	77.29	100.52	82.63
	Means	74.14	71.51	74.10	
	LSD 0.05	Salinity: 7.60	Ecotype: 4.12	Salinity × Ecotype: 7.14	
	P value	Salinity: 0.001**	Ecotype: 0.317^{ns}	Salinity × Ecotype: 0.001**	

*Significant difference in probability level of 5%, **Significant differences in probability level of 1%, ns no significant difference in probability level of 5%, LSD: Least Significant Difference

Membrane stability index: MSI decreased by increased level of salinity in irrigation water. MSI in 10.5 and 23.1 dS m^{-1}, were 11.8 and 11.9 % lower than MSI at 5.2 dS m^{-1} (Table 2). Cellular injury increased in all ecotypes and all salinity levels, and the magnitude of increase was more pronounced in Birjand and Borujerd. Membrane stability index is one of the useful parameters to differentiate genotypes under salinity stress (Farooq & Azam, 2006). In spite of strong decrease in MSI under high level of salinity, Sabzevar maintained higher MSI compared to Birjand and Borujerd. Correlation between MSI with dry matter and seed yield in different levels of salinity and ecotypes did not show significant coefficient (Table 4). Ashraf & Ali (2008) reported that cell membrane permeability was an effective determinant of salt tolerance in canola cultivars, because it showed a positive association with the activities of different antioxidant enzymes such as SOD, CAT and peroxidase as well as with the degree of salt tolerance of the canola cultivars.

Relative water content (RWC): Leaf RWC declined 2.3 and 4.7% at 10.5 and 23.1 dS m^{-1} salinity compared to 5.2 dS m^{-1} (Table 2). The highest and lowest RWC observed in Birjand and Borujerd, respectively. In this study, it was noticed that leaf RWC declined under salinity stress conditions. Yang *et al*., (2009), have reported similar results in *Populus cathayana*. The rapid change in salt concentration caused decreased in leaf water potential (Munns & Tester, 2008). Result of correlation coefficient showed that RWC had positive relationship with MSI (Table 4).

Proline content and total soluble sugars: The leaf proline content was significantly increased in all ecotypes at all salinity levels. There was a linear increase in proline accumulation with increasing salt concentration in the growth medium. A more pronounced increase was observed in Sabzevar ecotype compared to other ecotypes. However, proline content increased by about 29.3, 73.5 and 69.8% the shoots of Birjand and Borujerd and Sabzevar in 23.1 dS m^{-1} compared to 5.2 dS m^{-1} salinity, respectively (Table 2). Salinity stress imposed a significant effect on total soluble sugars. Total soluble sugars were lower at 10.5 dS m^{-1} salinity against 5.2 and 23.1 dS m^{-1} in all ecotypes (Table 2).

In the present study, a 2.7-fold increase was observed in leaf proline content of 23.1 dS m^{-1} compared to 5.2 dS m^{-1}. In general, total correlation showed the negative relationship between proline content with MSI and RWC (Table 4). There were no significant relationship found between soluble sugar and RWC (Table 4). Nasir Khan *et al*., (2007) reported that the parallel increase in proline content with the electrolyte leakage suggested that proline might be involved in the osmotic adjustment of salinized plants. Under salt stress, increase in proline content might be caused by induction of proline biosynthesis or decrease in oxidation of proline to glutamate or enhancement in protein turnover. Similarly, proline supply energy required for compartmentation of ions in vacuole (Amirjani, 2010). Therefore, osmolyte production in plant, require energy that cause decreased biomass production. In present experiment, we did not found any positive correlation between proline and antioxidants activity in Kochia (Table 4).

Ascorbate peroxidase: APX activities exhibited a decline under 10.5 dS m^{-1} salinity compared with 5.2 and 23.1 dS m^{-1} salinity in Birjand and Borujerd ecotypes. Borujerd ecotype showed the highest APX activities among ecotypes (Table 3).

Catalase activity: The catalase activity exhibited a considerable decline under 10.5 dS m^{-1} salinity stress compared with other salinity levels. Borujerd showed generally less CAT activity than Birjand and Sabzevar (Table 3). Interaction between salinity levels and Kochia ecotypes showed different patterns. In Sabzevar CAT activity declined by increased salinity levels but in Borujerd and Birjand the lowest CAT activity was obtained in 10.5 dS m^{-1} salinity (Table 3).

Superoxide dismutase activity: The SOD activity measured in leaves at anthesis subjected to salt stress showed a highest activity under 10.5 dS m^{-1} salinity (Table 3). The SOD activity was highest in the fully expanded leaves of Borujerd and was lowest in Sabzevar (Table 3). Interaction between salinity and ecotype showed that in Sabzevar with increasing levels of salinity SOD activity decreased but in Birjand and Borujerd activity of SOD at 10.5 dS m^{-1} was more than other salinity levels (Table 3).

Table 3. Effect of salinity on ascorbate peroxidase (APX), catalase (CTA), superoxide dismutase (SOD), peroxidase (POX), glutathione reductase (GR), DPPH - radical scavenging activity and total phenol in different Kochia ecotypes.

	Salinity dS m^{-1}	Ecotypes			Means
		Birjand	Borujerd	Sabzevar	
APX (unit.g^{-1}dw)	5.2	214.47	219.11	100.46	178.01
	10.5	131.58	141.77	137.23	136.86
	23.1	180.55	193.93	133.51	169.33
	Means	175.53	184.94	123.73	
	LSD 0.05	Salinity: 82.89	Ecotype: 52.34	Salinity × Ecotype: 90.66	
	P value	Salinity: 0.236^{ns}	Ecotype: 0.054*	Salinity × Ecotype: 0.317^{ns}	
CAT (unit.g-1dw)	5.2	461.95	265.57	621.31	449.61
	10.5	215.19	148.03	339.36	234.20
	23.1	394.97	576.66	186.67	386.10
	Means	357.37	330.09	382.45	
	LSD 0.05	Salinity: 567.20	Ecotype: 1069.00	Salinity × Ecotype: 1852.00	
	P value	Salinity: 0.329^{ns}	Ecotype: 0.934^{ns}	Salinity × Ecotype: 0.330^{ns}	
SOD (unit.g^{-1}dw)	5.2	187.61	242.80	222.39	217.60
	10.5	398.04	429.99	174.05	334.03
	23.1	207.94	209.49	157.70	191.71
	Means	264.53	294.09	184.71	
	LSD 0.05	Salinity: 70.26	Ecotype: 56.18	Salinity × Ecotype: 97.31	
	P value	Salinity: 0.001**	Ecotype: 0.010**	Salinity × Ecotype: 0.028*	
POX (unit.g^{-1}dw)	5.2	19.40	23.66	21.68	21.58
	10.5	23.33	17.57	27.28	22.72
	23.1	24.45	22.53	19.88	22.29
	Means	22.39	21.25	22.95	
	LSD 0.05	Salinity: 5.37	Ecotype: 4.09	Salinity × Ecotype: 7/08	
	P value	Salinity: 0.831^{ns}	Ecotype: 0.662^{ns}	Salinity × Ecotype: 0.061^{ns}	
GR (unit.g-1dw)	5.2	89.72	64.56	85.96	80.08
	10.5	119.63	47.14	76.22	81.00
	23.1	53.12	100.77	64.66	72.85
	Means	87.49	70.82	75.61	
	LSD 0.05	Salinity: 83.23	Ecotype: 41.21	Salinity × Ecotype: 71.38	
	P value	Salinity: 0.893^{ns}	Ecotype: 0.673^{ns}	Salinity × Ecotype: 0.192^{ns}	
DPPH (%)	5.2	24.35	21.55	12.19	19.36
	10.5	16.51	14.16	16.67	15.78
	23.1	26.72	27.28	25.05	26.35
	Means	22.53	20.99	17.97	
	LSD 0.05	Salinity: 5.75	Ecotype: 4.13	Salinity × Ecotype: 7.16	
	P value	Salinity: 0.001**	Ecotype: 0.088^{ns}	Salinity × Ecotype: 0.092^{ns}	
Total phenols (mg galic.g^{-1}dw)	5.2	7.34	7.13	8.01	7.49
	10.5	8.03	7.22	11.24	8.83
	23.1	7.63	7.80	6.38	7.27
	Means	7.67	7.38	8.54	
	LSD 0.05	Salinity: 1.78	Ecotype: 1.19	Salinity × Ecotype: 2.07	
	P value	Salinity: 0.030*	Ecotype: 0.129nd	Salinity × Ecotype: 0.016**	

*Significant difference in probability level of 5%, **Significant differences in probability level of 1%, ns no significant difference in probability level of 5%, LSD: Least Significant Difference

Table 4. Correlation coefficients among dry matter, seed yield, membrane stability index (MSI), relative water content (RWC), proline content, soluble sugar Ascorbate peroxidase (APX), catalase (CTA), superoxide dismutase (SOD), peroxidase (POX), glutathione reductase (GR), DPPH - radical scavenging activity and total phenol in saline condition in Kochia.

	Dry matter	Seed yield	MSI	RWC	Proline	Soluble sugar	APX	CAT	SOD	POX	GR	DPPH	Total phenol
Dry matter	1	0.60**	0.14^{ns}	0.03^{ns}	-0.21^{ns}	-0.35^{ns}	-0.15^{ns}	0.18^{ns}	0.19^{ns}	-0.11^{ns}	-0.17^{ns}	-0.15^{ns}	0.41*
Seed yield		1	0.41*	0.29^{ns}	-0.51**	-0.28^{ns}	-0.18^{ns}	0.12^{ns}	0.11^{ns}	-0.36^{ns}	-0.34^{ns}	-0.25^{ns}	0.26^{ns}
MSI			1	0.41*	-0.13^{ns}	0.29^{ns}	-0.01^{ns}	0.13^{ns}	-0.27^{ns}	-0.34^{ns}	-0.45*	0.04^{ns}	-0.02^{ns}
RWC				1	-0.42*	-0.12^{ns}	-0.24^{ns}	-0.18^{ns}	-0.08^{ns}	0.06^{ns}	0.16^{ns}	-0.27^{ns}	0.04^{ns}
Proline					1	0.60**	0.10^{ns}	-0.11^{ns}	-0.32^{ns}	0.21^{ns}	-0.04^{ns}	0.40*	-0.20^{ns}
Soluble sugar						1	0.39*	0.04^{ns}	-0.41*	0.03^{ns}	-0.11^{ns}	0.50**	-0.56**
APX							1	0.67**	-0.40*	0.31^{ns}	0.15^{ns}	0.67**	-0.25^{ns}
CAT								1	-0.20^{ns}	0.08^{ns}	0.29^{ns}	0.35^{ns}	0.15^{ns}
SOD									1	-0.37*	0.20^{ns}	-0.36^{ns}	0.01^{ns}
POX										1	0.48**	0.30^{ns}	0.19^{ns}
GR											1	0.09^{ns}	0.04^{ns}
DPPH												1	-0.26^{ns}
Total phenol													1

*significant difference in probability level of 5%, ** significant difference in probability level of 1%, ns no significant difference in probability level of 5%

Peroxidase activity: Result showed no significant effects due to salinity and ecotype in peroxidase activity.

Glutathione reductase activity: Result showed no significant effects due to salinity in glutathione reductase activity, however, the lowest activity of GR obtained at 23.1 dS m^{-1} (Table 3). GR activities of Birjand ecotype was enhanced at much higher rate than two other ecotypes (Table 3). GR activities of Birjand ecotype were higher at 10.5 dS m^{-1} but in Borujerd ecotype the activity of GR decline at this level of salinity.

DPPH - radical scavenging activity: The high and low DPPH - radical scavenging activity in leaves were found at 23.1 and 10.5 dS m^{-1}, respectively (Table 3). The DPPH - radical scavenging activity exhibited no considerable difference among ecotypes (Table 3) and also no interactions between treatments was significant ($p \leq 0.05$).

Recent works showed that salt tolerance is closely related to the efficiency of antioxidant enzymes (Arbona *et al*., 2003; Ashraf, 2009; Nawaz & Ashraf, 2010; Joseph & Jini, 2011). SOD and phenol content showed higher activity under 10.5 dS m^{-1} salinity. The high activity of CAT was observed at 5.2 dS m^{-1} salinity and higher activity of APX, POX, GR and DPPH - radical scavenging was obtained at 23.1 dS m^{-1} salinity. The increase dry matter accumulation (220 kg ha^{-1}) at 10.5 dS m^{-1} compared to 5.2 dS m^{-1} salinity and increase activity of SOD and phenol content in this level of salinity may suggest the existence of an effective scavenging mechanism to remove ROS at this level of salinity. However, we did not find any strong positive correlation between antioxidants activity and dry matter production (Table 4). One of the major effects of salinity is the peroxidation of lipids and loss of membrane integrity due to these ROS activity (Joseph & Jini, 2011). However, in the present study negative correlations were observed between MSI and different antioxidants in salt condition.

Total phenols: There were significant differences in phenol content among salinized plants at anthesis stage of the of Kochia ecotypes. Phenol content at 10.5 dS m^{-1} was higher than the other salinity levels. Total phenols concentration was not affected by ecotypes (Table 3). Phenol content was higher in Birjand and Sabzevar ecotypes and lower in Borujerd at 10.5 dS m^{-1} (Table 3). Phenols constitute a part of cellular solutes and provide a reducing environment stress (Singh, 2004). Loss of membrane integrity due to these ROS activity by salinity was reported (Joseph & Jini, 2011). Thus, phenol accumulation in salt tolerant plants could be a defense mechanism for scavenging the free radicals of oxygen and preventing cell membrane damage during stress (Singh, 2004). Results of this study showed that at 10.5 dS m^{-1} salinity, phenol content and dry matter accumulation higher than other levels of salinity. These results concur with the findings of Singh (2004) that reported increase content of phenol in chickpea under salinity stress.

Conclusion

Results indicate no positive significant correlation between antioxidative enzymes and biomass production on Kochia in present levels of salinity. This result indicated that possibly Kochia is capable to manage high levels of salt toxicity. Similarly, total biomass production in different levels of salinity was not significantly decrease up to 23.1 dS m^{-1}. According to these results, Kochia is a reasonable tolerant plant to elevated levels of salinity and seems that it is capable to produce reasonable biomass and seed at these levels of salinities.

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