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## Changes in Quaternary Ammonium Compounds, Proline and Protein Profiles of Certain Halophytic Plants under Different Habitat Conditions

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**Abstract:** The ecophysiological adaptive responses of some halophytes (*Avicennia marina*, *Atriplex farinosa*, *Limonium axillare*, *Nitraria retusa*, *Arthrocnemum glaucum* and *Halocnemum strobilaceum*) and wide ecological amplitude species (*Salsola tetrandra*) were studied under different habitat conditions of two regions (Red Sea and Sinai Peninsula) during winter and summer seasons. The changes in plant quaternary ammonium salts, proline, total soluble protein and protein pattern in relation to habitat conditions were discussed. Both of halophytes and species of wide ecological amplitude accumulated higher contents of quaternary ammonium compounds, proline and total soluble protein. Salt stress in the most studied species was associated with the formation of certain adaptive stress proteins, which may play a role in protecting the membranes.

**Key words:** Osmotic adjustment, salinity, halophytes, species of wide ecological amplitude, protein pattern

### Introduction

Salinity is an important environmental stress from which natural and cultivated plant suffer. High salinity is extremely heterogenous in space and time at any environment and many plants use windows of low salinity at all levels of organization (Orcutt and Nilsen, 2000). Environmental stresses, such as extreme temperature, water availability and excess salinity in the soil, limit the growth and production yield of crops. These stresses trigger the development of mechanisms, which regulate or counteract the stress-induced effects in plant tissues (Tramontano and Jouve, 1997). The osmotic effects of salinity on water availability well strongly inhibit growth of related species and varieties (Neumann *et al.*, 1997). The littoral salt marshes are subjected to maritime influences, i.e. periodic flooding with seawater, seawater spray, seawater seepage, etc. Their formation takes place through the silting up of lagoons or shoreline areas protected by sand or single bars. They can be found, in general, if any of the following physiographic conditions is fulfilled; the presence of estuaries, the shelter of spots, off-shore barrier island and large or small protected bays with shallow water, while the inland salt marshes are far from the reach of the marine influence (Zahran, 1982). The Red Sea coastal zone of Egypt is situated within a region of arid climate (Ayyad *et al.*, 1993). The vegetation of the Red Sea coast is dominated by small number of halophytes which are not only well adapted to this environment but they also exhibit a remarkable ability to exploit auxiliary energy sources provided by tides to perform mineral cycling and food transport (Odum and Fanning, 1973) to compensate for the energy cost copying

with the stress factors. Sinai Peninsula can be divided into two climatic zones according to Ayyad and Ghabbour (1986); arid and hyperarid zones. Generally, the amount of rainfall in Sinai decreases from the northeast to the southeast. Plants subjected to water-stress showed a decrease in the large molecular weight fraction of proteins and an increase in the smaller molecular weight fraction (Stutte and Todd, 1967; La Rosa *et al.*, 1989 and Moons *et al.*, 1995). Proline accumulation could represent a compensatory mechanism for better plant survival during a period of drought-stress (Ibara-Caballero *et al.*, 1988). One of its roles is a reservoir of nitrogen and carbon source (Fukataku and Yamada, 1984). An interesting correlation between the capacity of some plants to accumulate specific amino acids and their ability to resist or tolerate a particular environmental stress was observed by Stewart and Larher (1980). Proline is one of the most common compatible osmolytes involved in counteracting the effects of osmotic stress in plants. Genetically engineered crop plants that over produce proline could therefore gain the ability to tolerate environmental stresses (Yoshihara *et al.*, 1997). Glycinebetaine is believed to serve as a nontoxic solute for cytoplasmic osmoregulation and a protectant against the damaging effects of salt on proteins and membranes (Csonka and Hanson, 1991; Rhodes and Hanson, 1993 and Takabe *et al.*, 1998). The transformants that produce small amounts of glycinebetaine have shown to confer salt tolerance (Hayashi *et al.*, 1997 and Takabe *et al.*, 1998). Jefferies *et al.* (1979) showed that high salinities and the water stress of tissues of some halophytes resulted in an accumulation of sorbitol, proline, quaternary ammonium compounds

and  $\alpha$ -amino-N depending on the species. Most of halophytes which are grown under stress synthesize betaine in their chloroplast and accumulate it as an osmoprotectant (Makela *et al.*, 1998). It has been observed that *Limonium* species adapted to dry, sandy or rocky soils tend to accumulate glycinebetaine, whereas *Limonium* species colonizing salt marshes tend to accumulate B-alaninebetaine (Rhodes and Hanson, 1993). At the whole plant level, accumulation of Glycinebetaine has been correlated with growth under stress in some species (Colmer *et al.*, 1995) and with maintained nitrogen fixation under osmotic stress (Riou and Le Rudulier, 1990). Salinity results in reduced the synthesis of RNA, DNA and proteins (Rausser and Hanson, 1966). In addition, several investigators reported that salinity increased the amino acid contents of summer weight of the plants. The present investigation was carried out to study the changes in quaternary ammonium compounds, proline and protein profiles of some halophytic plants under different habitat conditions during winter and summer seasons.

#### Materials and Methods

The plant materials used in the present investigation were obtained from the selected species which collected from their natural habitats. They can be classified into three main ecological groups: (1) Crynohalophytes (excretives), these include: (I) *Avicennia marina* (Forssk) Vierh. (Fig. 1) which collected from 94 km south of Mersa Alam (Fig. 2) at Lat 21° 6'14" N and Long. 17° 6'80" E, (II) *Atriplex*

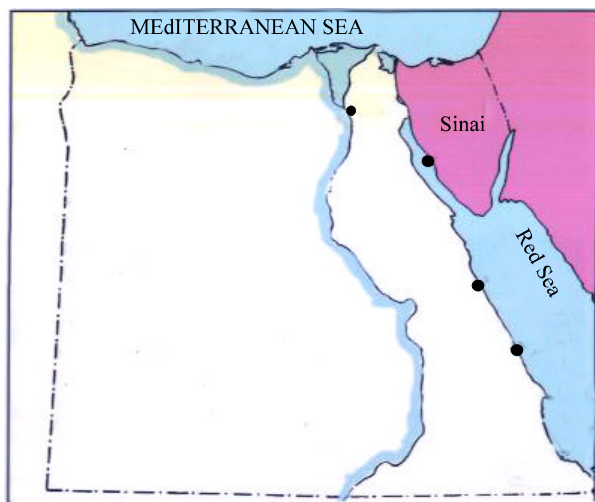


Fig. 1: Location map showing the sites from which the concerned plant species were collected; Ras Sudr, SW Sinani, Safaga and Mersa Alam on the Red Sea littoral zone

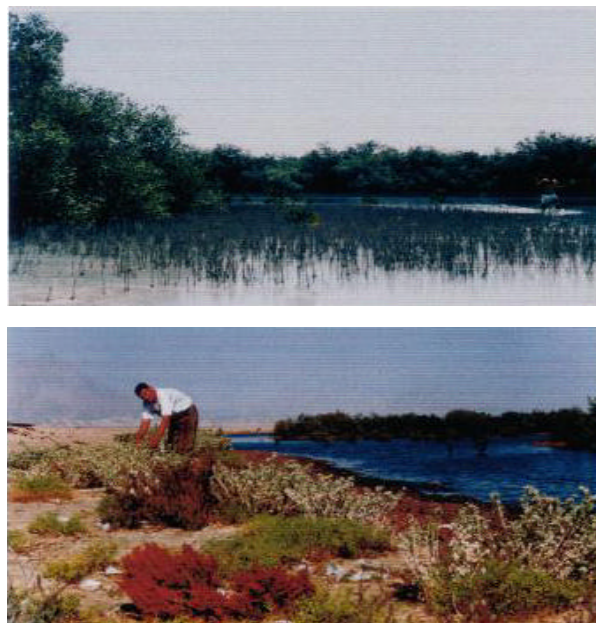


Fig. 2: The above general view shows *Avicennia marina* (Forssk.) Vierh community type with pneumatophores in the foreground whereas the lower one shows *A. marina* in the fare site and *Atriplex farinosa* (Forssk.) in the shoreline followed inland by other constituents including *Halopeplis perfoliata* (Forssk.)

*farinosa* (Forssk) which collected from the southern extension of the Red Sea coast, (III) *Limonium axillare* (Forssk.) Ktze (= *Statice axillaris*) (a and b) which collected from two sites differing in their location to the Red Sea coast. The first site (a) lies 32 km south of Mersa Alam (Lat 24° 48' 771" N and Long 35° 245" E). The second one (b) existing 70 km south of Mersa Alam (Lat. 24° 30' 43" N and Long 35° 08' 44" E), (IV) *Nitraria retusa* (Forssk.) Asch. (a and b) which collected from two desert locations; (a) delta portion of Wadi Sudr in SW coast of Sinai Peninsula (N 29° 37' 483" E 32° 41' 543") and (b) the Red Sea coast 75 km south of Safaga city (N 26° 39' 657" E 33° 56' 320") (Fig. 2) (2) Succulent halophytes, which includes: (I) *Halocnemum strobilaceum* (Pallas) M. Bieb. that collected from delta portion of Wadi Sudr (N 29° 37' 483" E 32° 41' 543"), (II) *Arthrocnemum glaucum* (Del.) Ung. Sternb. Which collected from its natural population growing at 94 km of Mersa Alam (N 24° 21' 613" E 35° 17' 680"). (3) Species of wide ecological amplitude, which includes: *Salsola tetrandra* (Forssk.) (a and b) (Fig. 3) and collected from two different zones from the delta of Wadi Sudr (N 29° 37' 483" E 32° 41' 543"). The climatic data of the studied areas were obtained from the nearest meteorological stations to the selected localities covering

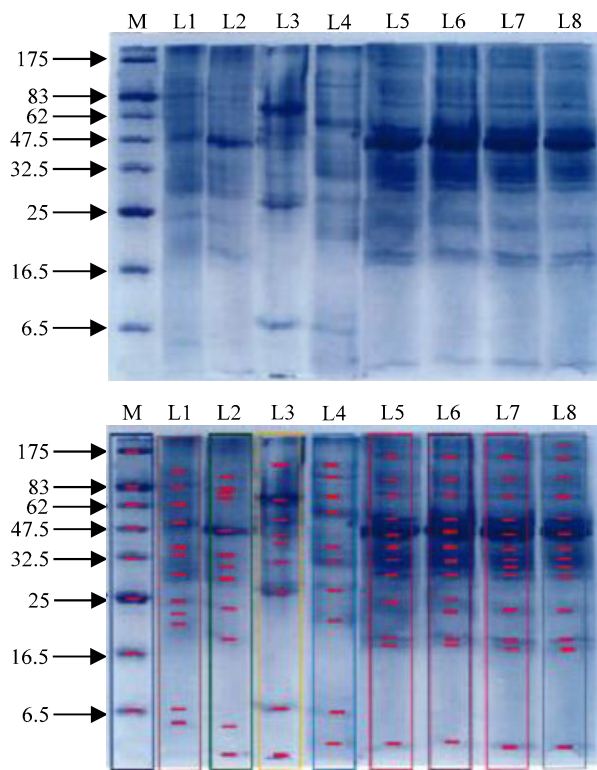


Fig. 3: The upper general view shows a part of the Delta of Wadi Sudr with hummocky zone built by *Salsola tetrandra* (Forssk.) which is seen in the lower close-up-view

the period from January 1999 to December 1999. The monthly mean values of climatic particulars were tabulated. The methods adopted in the present investigation for plant sampling and analyses could be summarized as follows : Extraction, identification and quantification of quaternary ammonium by Plasma Desorption Mass Spectrometry (PDMS) as followed by Bonham *et al.* (1995). Proline content was determined as described by Bates *et al.* (1973). The total soluble proteins were determined spectrophotometrically using the Bio-Rad protein assay that described by (Bio-Rad Technical Bulletin 1051, 1977). The protein banding patterns of the studied species were extracted and detected using the method described by Laemmli (1970).

### Results and Discussion

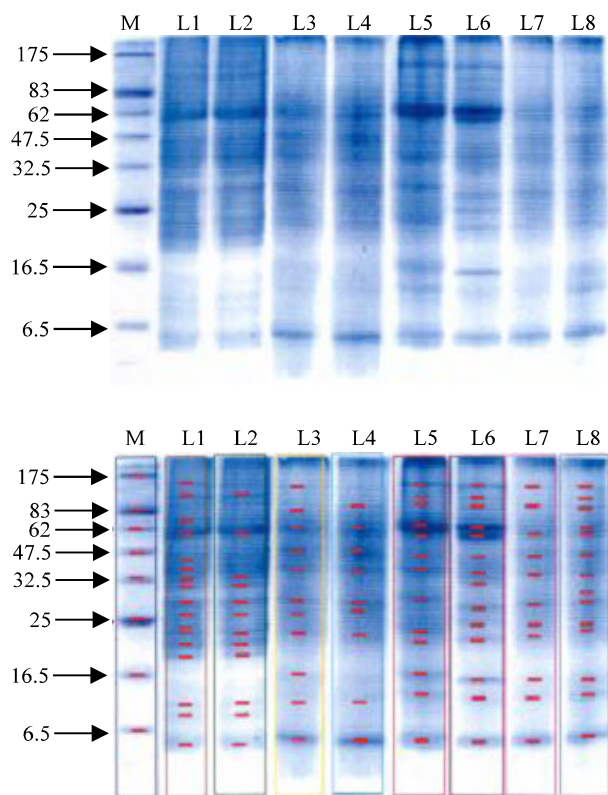
Climatic data of the studied areas are shown in Table 1 covering the period from January 1999 to December 1999. The data indicate the intensity of atmospheric drought to which desert plants are subjected. Regarding the climate of the Deltoid portion of Wadi Sudr which exists in south west of Sinai, it was characterized by its great variability. The period from May to August is completely rainless, its monthly values ranged from 2.4 to 6.6 mm during the



Crynohalophytes  
Red Sea  
M: Marker  
L1: *Avicennia marina* (summer)  
L2: *Avicennia marina* (winter)  
L3: *Atriplex farinosa* (summer)  
L4: *Atriplex farinosa* (winter)  
L5: *Limonium axillare* (a) (summer)  
L6: *Limonium axillare* (a) (winter)  
L7: *Limonium axillare* (b) (summer)  
L8: *Limonium axillare* (b) (winter)

Fig. 4: Seasonal fluctuations in protein banding patterns of the studied species under different habitat conditions

period of April and December reaching the maximum value of 9.9 mm during January (Table 1). The irregularity of rainfall is pronounced and this represents a common factor of all arid regions (Strahler, 1951). There are wide variations in air temperature of the south west Sinai area throughout the whole year. Data in Table 1 indicate that the summer months (June, July and August) were the hottest months while those of winter (January, February, November and December) were the coldest ones. The monthly maximum mean temperature reached a maximum of 31.9°C in June. Relative humidity of the Wadi Sudr area recorded its minimum value of 45% in July (1999).



Crynohalophytes

A. Red Sea

M: Marker

L1: *Nitraria retusa* (a) (summer)

L2: *Nitraria retusa* (a) (winter)

L3: *Nitraria retusa* (b) (summer)

L4: *Nitraria retusa* (b) (winter)

B. Sinai Peninsula

L5: *Nitraria retusa* (a) (summer)

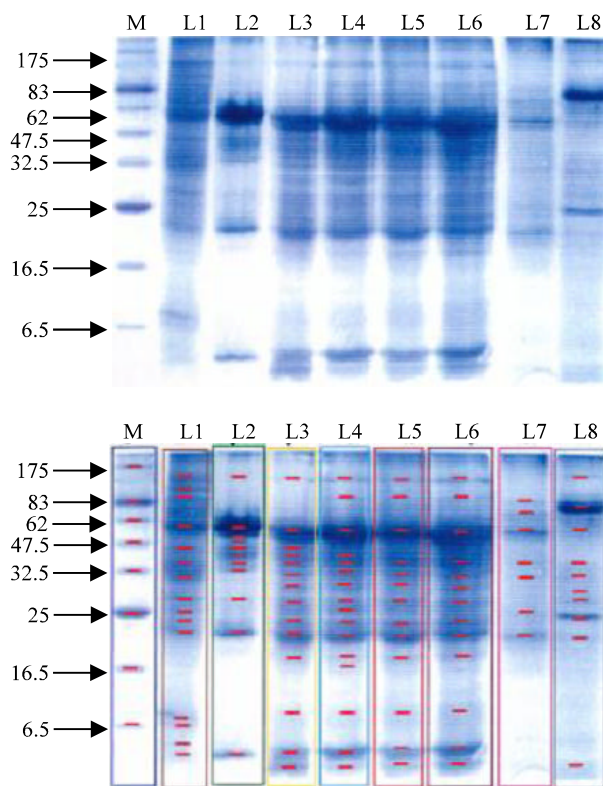
L6: *Nitraria retusa* (a) (winter)

L7: *Nitraria retusa* (b) (summer)

L8: *Nitraria retusa* (b) (winter)

Fig. 5: Seasonal fluctuations in protein banding patterns of the studied species under different habitat conditions

Concerning the climate of the southern area of the Red Sea coast, data in Table 1 indicate that there are wide variations in the rainfall and relative humidity throughout the year. The months March, April and May were the most rainy and humid. According to the climatic records of the Red Sea coastal zone, the mean maximum temperature reached the highest value of 42.1 in August while the mean minimum temperature was recorded in January, 1999 (13.77°C). The climate of the Red Sea coastal area of Egypt is arid. Many investigators (Zahran



Succulent halophytes

A. Red Sea

M: Marker

L1: *Arthrocnemum glaucum* (summer)

L2: *Arthrocnemum glaucum* (winter)

B. Sinai Peninsula

L3: *Halocnemum strobilaceum* (summer)

L4: *Halocnemum strobilaceum* (winter)

Species of wide ecological amplitude Sinai Peninsula

L5: *Salsola tetrandra* (a) (summer)

L6: *Salsola tetrandra* (a) (winter)

L7: *Salsola tetrandra* (b) (summer)

L8: *Salsola tetrandra* (b) (winter)

Fig. 6: Seasonal fluctuations in protein banding patterns of the studied species under different habitat conditions

and Willis, 1992 and Abd El-Ghani, 1998) have described the climate of the Red Sea coast. Concerning the quaternary ammonium salts of the Crynohalophytes of the Red Sea coastal area, data presented in Table 3 reveal that the osmoprotectant glycinebetaine was accumulated in higher level during summer compared with that recorded in winter in *Avicennia marina* (310 and 280 n mole g<sup>-1</sup> D.wt.). *Atriplex farinosa* exhibited a reverse behaviour, where it accumulated the highest amount of

Table 1: Monthly variations of rainfall, maximum and minimum temperatures and relative humidity of Sinai Peninsula and Red Sea regions during the period from January 1999 to December 1999

Months	Sinai Peninsula				Red Sea			
	Rainfall mm/month	Temperature °C		Relative Humidity (%)	Rainfall mm/month	Temperature °C		Relative Humidity (%)
		Maximum	Minimum			Maximum	Minimum	
January	9.90	19.57	8.33	68.00	0.00	24.35	13.77	50.00
February	9.20	17.57	7.87	65.00	0.00	25.08	14.81	51.21
March	6.60	19.63	10.13	65.00	0.04	25.82	17.00	55.00
April	2.40	22.83	10.77	48.00	0.08	31.05	20.30	57.70
May	0.00	25.82	12.00	49.00	0.05	34.72	25.50	56.80
June	0.00	31.91	18.71	50.00	0.00	38.36	25.00	50.70
July	0.00	29.82	14.00	45.00	0.00	40.00	27.14	53.00
August	0.00	31.20	18.90	53.00	0.00	42.10	29.12	52.30
September	10.00	28.30	17.92	56.00	0.00	36.68	26.90	51.60
October	5.00	26.00	14.30	60.00	0.00	33.21	22.58	51.50
November	5.10	17.00	7.90	68.00	0.00	29.26	21.55	53.91
December	6.60	13.30	3.40	73.00	0.00	26.24	18.41	53.76

glycinebetaine during winter.  $\beta$ -alaninebetaine compound was detected in both *Limonium axillare* (a) and (b) during winter (38 and 30 n mole  $g^{-1}$ D.wt., respectively). In addition to glycinebetaine both *Nitraria retusa* (a) and (b) of the Red Sea coast accumulated trigonelline compound during winter (9.26 and 1.413 n mole  $g^{-1}$ D.wt., respectively). *Nitraria retusa* (a) and (b) of the Sinai Peninsula accumulated very low contents of glycinebetaine during winter (16.428 and 6.667 n mole  $g^{-1}$ D.wt.) compared with those attained by the other plant species. The succulent halophytes species from both Red Sea and Sinai Peninsula areas attained their maximum values during summer which decreased markedly during winter season.

Quaternary ammonium compounds such as glycinebetaine,  $\beta$ -alaninebetaine, trigonelline act as non toxic (compatible) osmoticum preferentially located in the cytoplasm and chloroplasts of plant cells and also may act as enzyme protectants (Wyn Jones and Storey, 1981). These organic solutes are accumulated in halophytes and in a wide range of glycophytic plant species. The accumulation of quaternary ammonium compounds was highest in the salt tolerant wild species (Baburina and Shevyakova, 1988). Betaines act as osmoregulatory substances, maintaining the osmotic balance with environment (Rhodes and Hanson, 1993).

It is evident that the compatible osmotic solute, glycinebetaine can alter the thermodynamic properties of membranes perhaps by direct interacting with phosphatidylcholine moieties (Rudolph *et al.*, 1986). Moreover, Wyn Jones and Gorham (1983) have discussed the possible roles of glycinebetaine in modulating the flux of  $Na^+$  from the cytoplasm to the vacuole. They reported that quaternary ammonium salts may specifically modulate the activity of membrane carrier proteins or channels. In addition, betaine compounds stabilize proteins and membranes when present at high concentration and so

can be used to raise cytoplasmic osmotic pressure in stressed cells without deleterious effects, since glycinebetaine is predominantly localized in the cytoplasm of leaf cells of halophytic chenopods *Suaeda maritima* and *Atriplex gmelini* leaves. Moreover, Rhodes and Hanson (1993) reported that the genes of betaine synthesis are nuclear encoded and are induced by salt/water stress.

$\beta$ -alaninebetaine is one of the quaternary ammonium salts which was recorded in both *Limonium axillare* (a) and (b) during winter in response to relatively high total soluble salts (TSS) of the associated soils (Table 2). In plumbaginaceae family, various species accumulated  $\beta$ -alaninebetaine compounds under salt stress which may serve as an osmoprotectant (Hanson *et al.*, 1994).  $\beta$ -alaninebetaine synthesis does not have  $O_2$  requirements, which may be rational for the observation that *Limonium* species adapted to dry, sandy or rocky soils tend to accumulate glycinebetaine, whereas *Limonium* species colonizing salt marshes tend to accumulate  $\beta$ -alaninebetaine (Hanson *et al.*, 1991). Interestingly, substituting  $\beta$ -alaninebetaine for glycinebetaine with some species of plumbaginaceae has not narrowed their adaption, as many  $\beta$ -alaninebetaine accumulators are found in dry saline environments as well as the high altitudes.  $\beta$ -alaninebetaine thus appears to be effective over a border ecological spectrum than glycinebetaine (Rhodes and Hanson, 1993). Both of *Nitraria retusa* (a) and (b) from Sinai Peninsula region accumulated glycinebetaine during winter where trigonelline compounds were not found in these species. Glycinebetaine was induced as a result of high total soluble salts in the soil of both *Nitraria retusa* (a) and (b) (Table 2). Trigonelline is a pyridine betaine, functions as a cell cycle regulators and accumulates in response to various stresses. It is believed to act as an osmoregulator. It is preferentially located in the cytoplasm and plays

Table 2: Chemical composition of soil profiles associated with the studied plant species (Hassanein *et al.*, 2003)

Plant species and localities	Habitat	Depth	FC%		CaCO <sub>3</sub> %		OC%		Humus %		Avail. P <sup>33</sup> mg 100 g <sup>-1</sup> dry soil		Avail N. mg 100 g <sup>-1</sup> dry soil	
			W	S	W	S	W	S	W	S	W	S	W	S
I. Crynohalophytes														
A. Red Sea	Estuaries and coastal strip of the Red Sea 94	0-20	3.960	0.790	81.200	92.450	0.072	0.090	0.124	0.145	0.060	0.390	0.175	0.138
<i>Avicennia marina</i>	km south of Mersa Alam	20-50	30.900	25.700	8.300	36.560	0.240	0.180	0.314	0.310	0.062	0.032	0.140	0.181
<i>Atriplex farinosa</i>	a. Salt-affected, inland extension of the coastal plain (32 km south of Mersa Alam)	50-70	28.130	26.100	30.900	44.010	0.204	0.170	0.351	0.300	0.070	0.210	0.230	0.190
	b. Inland extension of the coastal plain (70 km south of Mersa Alam)	0-20	7.270	2.610	1.450	17.240	0.060	0.036	0.103	0.083	0.002	0.104	0.250	0.160
<i>Limonium axillare</i>	a. Coastal (7 km south of Safaga)	20-50	2.800	3.780	5.400	13.920	0.024	0.048	0.041	0.103	0.002	0.268	0.370	0.500
	b. Inland (200 m west of the shoreline)	50-70	2.500	0.440	2.300	7.890	0.031	0.060	0.030	0.083	0.001	0.112	0.270	0.330
		0-20	1.780	1.240	22.000	12.470	0.132	0.050	0.227	0.144	0.052	0.140	0.230	0.160
		20-50	1.480	3.720	18.100	17.040	0.060	0.080	1.103	0.090	0.070	0.080	0.300	0.210
		50-70	1.670	2.910	17.700	14.110	0.048	0.070	0.083	0.165	0.070	0.069	0.200	0.300
<i>Nitraria retusa</i>	a. Coastal (7 km south of Safaga)	0-20	7.730	3.230	18.900	14.940	0.024	0.060	0.041	0.103	0.012	0.028	0.225	0.430
		20-50	1.320	2.050	19.110	18.910	0.036	0.048	0.062	0.083	0.011	0.016	0.325	0.310
		50-70	1.270	1.990	18.700	17.000	0.120	0.0630	0.206	0.090	0.052	0.011	0.275	0.250
	b. Inland (200 m west of the shoreline)	0-20	2.000	8.260	21.600	16.830	0.096	0.120	0.165	0.186	0.040	0.205	0.550	0.350
		20-50	1.140	3.600	15.370	14.540	0.012	0.110	0.021	0.103	0.020	0.160	0.570	0.340
		50-70	1.580	3.810	16.470	15.190	0.048	0.060	0.083	0.062	0.032	0.004	0.300	0.330
B. Sinai Peninsula	a. Coastal (Leveled head of Delta Wadi Sudr)	0-20	19.780	4.500	41.400	86.600	0.240	0.072	0.413	0.124	0.160	0.275	0.400	0.156
<i>Nitraria retusa</i>		20-50	13.600	1.690	69.100	98.100	0.080	0.060	0.141	0.103	0.110	0.300	0.236	0.100
		50-70	2.290	6.620	83.700	75.200	0.060	0.048	0.107	0.083	0.200	0.320	0.108	0.024
	b. Inland extension of the Delta of Wadi Sudr.	0-20	4.500	2.490	85.100	98.700	0.090	0.060	0.162	0.103	0.210	0.230	0.110	0.056
		20-50	1.910	1.330	93.800	95.400	0.102	0.048	0.175	0.083	0.163	0.170	0.072	0.028
		50-70	18.220	1.370	74.000	94.400	0.090	0.036	0.162	0.062	0.110	0.350	0.120	0.048
II. Succulent halophytes														
A. Red Sea	94 km south of Mersa Alam	0-20	3.960	0.790	81.200	92.450	0.072	0.090	0.124	0.145	0.060	0.390	0.175	0.138
<i>Arthrocnemum glaucum</i>		20-50	30.900	25.700	8.300	36.560	0.240	0.180	0.314	0.310	0.062	0.032	0.140	0.181
		50-70	28.130	26.100	30.900	44.010	0.204	0.170	0.351	0.300	0.070	0.210	0.230	0.190
B. Sinai Peninsula	The closest zone of the Delta of Wadi Sudr to the Gulf of Suez	0-20	1.910	7.500	85.400	82.500	0.120	0.084	0.266	0.289	0.150	0.250	0.220	0.120
<i>Halocnemum strobilaceum</i>		20-50	1.670	7.530	92.800	90.200	0.060	0.012	0.103	0.021	0.350	0.252	0.1700	0.016
		50-70	1.360	4.690	95.700	87.500	0.070	0.024	0.124	0.041	0.188	0.290	0.136	0.092
III. Species of Wide Ecological amplitude														
Sinai Peninsula	a. Coastal (Leveled head of Delta Wadi Sudr).	0-20	19.780	4.500	41.400	86.600	0.240	0.072	0.413	0.124	0.160	0.275	0.400	0.156
<i>Salsola tetrandra</i>		20-50	13.600	1.690	69.100	98.100	0.080	0.060	0.141	0.103	0.110	0.300	0.236	0.100
		50-70	2.290	6.620	83.700	75.200	0.060	0.048	0.107	0.083	0.200	0.320	0.108	0.024
	b. Inland extension of the Delta of Wadi Sudr.	0-20	0.830	5.300	83.500	79.900	0.680	0.036	0.144	0.062	0.250	0.225	0.420	0.160
		20-50	6.110	2.700	79.400	95.400	0.090	0.060	0.165	0.103	0.100	0.365	1.310	0.048
		50-70	2.330	2.660	83.900	93.500	0.060	0.072	0.103	0.124	0.113	0.250	2.300	0.002

Table 2: Continue

Plant species and localities	Habitat	pH		TSS (ppm)		Soluble anions (meq L <sup>-1</sup> )				Soluble cations (meq L <sup>-1</sup> )							
		-----		-----		Cl <sup>-1</sup>		SO <sub>4</sub> <sup>-2</sup>		Na <sup>+1</sup>		K <sup>+1</sup>		Ca <sup>+2</sup>		Mg <sup>+2</sup>	
		W	S	W	S	W	S	W	S	W	S	W	S	W	S	W	S
I. Crynohalophytes																	
A. Red Sea	Estuaries and coastal	7.36	7.37	7.552	2528	98.0	77	0.817	0.800	870	45.7	1.97	1.05	27.6	17.5	52.4	57.5
<i>Avicennia marina</i>	strip of the Red Sea 94	7.24	7.05	14272	11520	190	100	0.833	0.817	217	152.0	6.92	10.3	40	38	60	42.5
<i>Atriplex farinosa</i>	km south of Mersa Alam	7.42	7.00	14016	10500	186	160	0.829	0.781	165	120.7	7.18	9.55	22.0	36	680	41.0
	a. Salt-affected, inland extension	7.33	7.44	2496	5037	22.6	180	0.842	0.797	12.2	83.0	0.50	2.00	25.0	15.0	56.4	27.0
	of the coastal plain (32 km south	7.35	7.61	2624	2765	26.4	100	0.808	0.66	14.8	57.0	0.74	1.59	20.5	6.5	27.5	39.5
<i>Limonium axillare</i>	of Mersa Alam)	6.75	7.54	2421	2112	24.5	70	0.758	0.68	11.9	44.0	0.65	1.15	21.6	6.0	24.9	40.0
	b. Inland extension of the coastal	7.05	7.26	2713.6	4077	440	115	0.885	0.804	499	61.9	1.80	1.18	21.6	19.1	88.4	41.8
	plain (70 km south of Mersa	7.44	7.52	5120	2387	65.4	72	0.813	0.809	63.5	30.5	1.04	0.89	23.5	19.0	2.5	51.8
	Alam)	7.71	6.57	2304	2011	32.0	68	0.607	0.751	26.1	25.0	0.64	0.77	11.0	17	26	30.5
	a. Coastal (7 km south of	7.35	7.01	9728	19136	128	330	0.833	0.813	130	239	5.38	17.9	26	38.5	50	61.5
	Safaga)	7.73	7.73	2816	3648	14.4	116	0.708	0.725	28.7	65.3	1.00	1.25	18	7.5	14	36.6
<i>Nitraria retusa</i>		7.45	7.6	2368	2911	36.0	100	0.70	0.651	27.8	55	0.95	1.05	13.6	6.7	8.4	24.0
	b. Inland (200 m west of the	7.45	7.18	1344	17664	16.0	300	0.733	0.800	10.4	222.0	0.56	1.51	12.0	62.5	26.0	97.5
	shoreline)	7.42	7.20	1536	19712	26.0	400	0.808	0.812	7.83	27.8	0.46	1.21	17.4	40.5	25.6	59.5
		7.40	7.32	1472	3558	14.6	106	0.817	0.812	9.6	52.2	0.54	0.97	19.8	21.5	21.2	42.5
B. Sinai Peninsula	a. Coastal (Leveled head of	7.22	7.19	85760	25664	3050	342	0.825	0.804	2871	165	28.2	8.97	49.0	39.5	185.0	60
	Delta Wadi Sudr)	7.71	7.54	19072	5472	470	100	0.820	0.790	348	43	15.4	1.03	28.0	14.5	68.0	34
<i>Nitraria retusa</i>		7.85	7.40	5376	11008	128	184	0.817	0.86	91	68.7	1.31	1.67	19.0	36.3	21.0	39.7
	b. Inland extension of the Delta	7.60	7.56	10694	48128	220	400	0.750	0.804	142	147	17.0	11.3	15.0	60.0	45	140
	of Wadi Sudr.	7.85	7.65	1792	3981	62	72	0.710	0.808	37	21.7	0.95	1.50	10.0	21.0	15	53
		7.77	7.70	3168	3827	105	70	0.742	0.775	61	20	1.85	1.51	9.6	10.5	19.4	38.5
II. Succulent halophytes																	
A. Red Sea		7.36	7.37	7552	2528	98.0	77	0.817	0.800	870	45.7	1.97	1.05	27.6	17.5	52.4	57.5
<i>Arthrocnemum glaucum</i>	94 km south of Mersa Alam	7.24	7.05	14272	11520	190	100	0.833	0.817	217	152.0	6.92	10.3	40	38	60	42.5
		7.42	7.00	14016	10500	186	160	0.829	0.781	165	120.7	7.18	9.55	22.0	36	680	41.0
B. Sinai Peninsula	The closest zone of the Delta	7.03	7.25	36480	4000	805	216.5	0.808	0.750	780	11.7	23.0	6.50	50.0	17.5	96.0	99.5
<i>Halocnemum strobilaceum</i>	of Wadi Sudr to the Gulf of Suez	7.83	7.62	1786	2579	75	50	0.708	0.67	48	11.3	0.95	1.18	11.0	3.25	14.0	26.75
III. Species of Wide Ecological amplitude																	
Sinai Peninsula		7.22	7.19	85760	25664	3050	342	0.825	0.804	2871	165	28.2	8.97	49.0	39.5	185.0	60
	a. Coastal (Leveled head of	7.71	7.54	19072	5472	470	100	0.820	0.790	348	43	15.4	1.03	28.0	14.5	68.0	34
<i>Salsola tetrandra</i>	Delta Wadi Sudr).	7.85	7.40	5376	11008	128	184	0.817	0.86	91	68.7	1.31	1.67	19.0	36.3	21.0	39.7
	b. Inland extension of the Delta	7.20	7.02	18368	60480	350	483	0.810	0.808	244	209	15.4	11.5	29.6	64.0	55.4	82
	of Wadi Sudr.	7.64	7.54	2579	2362	85	26	0.775	0.80	52	27.8	1.4	0.97	15.0	22.5	17.0	20.5
		7.53	7.51	1798	3245	65	68	0.804	0.775	31	10.4	1.03	1.20	18.6	10.5	2.6	30.5



Table 3: Seasonal fluctuations in glycinebetaine, trigonelline,  $\beta$ -alaninebetaine, proline and total soluble protein contents of the studied species under different habitat conditions

Plant species and localities	Habitat	Glycinebetaine (nmole g <sup>-1</sup> D.wt.)		Trigonelline (nmole g <sup>-1</sup> D.wt.)		B-alaninebetaine (nmole g <sup>-1</sup> D.wt.)		Proline ( $\mu$ mole g <sup>-1</sup> F.wt.)		Total soluble protein (ug g <sup>-1</sup> F.wt.)	
		Winter	Summer	Winter	Summer	Winter	Summer	Winter	Summer	Winter	Summer
I. Cynohalophytes											
A. Red Sea											
<i>Avicennia marina</i>	94 km south of Mersa Alam	280.000	310.000					1.7 $\pm$ 0.5	2.9 $\pm$ 0.4*	3006 $\pm$ 7.1	3421 $\pm$ 14.8*
<i>Atriplex farinosa</i>	94 km south of Mersa Alam	295.000	277.000					0.4 $\pm$ 0.1	1.7 $\pm$ 0.6*	754.4 $\pm$ 3.5	976.1 $\pm$ 3.7*
<i>Limonium axillare</i>	a. Coastal (32 km south of Mersa Alam)	-	-	-	-	38.000	18.000	2.2 $\pm$ 0.2	3.7 $\pm$ 0.6*	2211.4 $\pm$ 1.6	2285.4 $\pm$ 6.3*
	b. Inland (70 km south of Mersa Alam)	-	-	-	-	30.000	13.658	4.9 $\pm$ 0.4	2.55 $\pm$ 0.8*	1819.4 $\pm$ 14.6	1916.8 $\pm$ 2.8*
<i>Nitraria retusa</i>	a. Coastal (7 km south of Safaga)	2.778	4.167	9.260	7.240	-	-	8.5 $\pm$ 1.0	5.2 $\pm$ 0.8*	4056.5 $\pm$ 6.3	5241 $\pm$ 15.5**
	b. Inland (200 m west of the shoreline)	4.927	6.000	1.413	1.000	-	-	2.9 $\pm$ 0.4	1.9 $\pm$ 0.3*	4514.9 $\pm$ 9.4	2084.4 $\pm$ 20.7**
B. Sinai Peninsula											
<i>Nitraria retusa</i>	a. Coastal (Leveled head of Delta Wadi Sudr)	16.428	2.500	-	-	-	-	9.8 $\pm$ 0.07	12.5 $\pm$ 0.7*	3195.06 $\pm$ 5.7	2804.3 $\pm$ 5.9**
	Inland (Hummocky inland extension of Delta Wadi Sudr)	6.667	2.632	-	-	-	-	9.2 $\pm$ 7.1	0.4 $\pm$ 0.1**	3445.9 $\pm$ 1.9	2231.5 $\pm$ 3.5**
II. Succulent halophytes											
A. Red Sea											
<i>Arthrocnemum glaucum</i>	94 km south of Mersa Alam	279.000	330.000	-	-	-	-	1.1 $\pm$ 0.1	1.6 $\pm$ 0.5	2335 $\pm$ 7.1	3236.5 $\pm$ 3.4**
B. Sinai Peninsula											
<i>Halocnemum strobilaceum</i>	The closest zone of the Delta of Wadi Sudr to the Gulf of Suez	58.000	285.000	-	-	-	-	3 $\pm$ 0.1	0.8 $\pm$ 0.07**	2891.8 $\pm$ 2.6	1876.2 $\pm$ 5.4**
III. Species of wide ecological amplitude											
Sinai Peninsula											
<i>Salsola tetrandra</i>	a. Coastal (Leveled head of Delta Wadi Sudr)	275.000	300.000	-	-	-	-	2.9 $\pm$ 0.4	1.5 $\pm$ 0.4*	3236.5 $\pm$ 3.3	2198.9 $\pm$ 12.2**
	b. Inland (Hummocky inland extension of Delta Wadi Sudr)	130.000	125.000	-	-	-	-	1.9 $\pm$ 0.4	0.8 $\pm$ 0.07*	1709.5 $\pm$ 3.3	1136.5 $\pm$ 17.6*

Table 4: Seasonal fluctuations in protein banding patterns of the studied species under different habitat conditions

Habitat			%							
Plant species			Red Sea							
			<i>Avicemia marina</i>		<i>Atriplex farinosa</i>		<i>Limonium</i> (a)		<i>Limonium</i> (b)	
Lanes			L1	L2	L3	L4	L5	L6	L7	L8
Rows	M.Wt.	Marker	Summer	Winter	Summer	Winter	Summer	Winter	Summer	Winter
1	186.0									6.61
2	175.0	12.48								
3	155.0									
4	130.0				12.33	10.78		7.82	8.15	7.90
5	112.0		9.91							
6	99.0			11.64		9.04				
7	88.0						7.86	8.60		8.27
8	83.0	17.69	10.59							7.22
9	78.0			10.45						
10	72.0		10.36	11.50	17.12	11.50	7.68	8.55	8.23	7.30
11	62.0	12.13								
12	61.0									
13	58.0					13.23				
14	52.0		10.91		14.61		9.74	11.48	10.85	8.54
15	47.5	10.41		14.98						
16	44.0				13.15		11.03	12.36	14.47	14.06
17	36.0				10.71	11.72	6.11	9.37	7.38	7.72
18	34.0		9.47							
19	32.5	10.42								
20	31.0		9.82	8.65	10.34	11.93	9.72	11.01	9.05	8.21
21	30.0			9.63					7.31	4.70
22	29.0		9.84	10.29			8.77	9.08	8.11	7.29
23	28.0				12.13	9.99	7.52			
24	25.0	16.80								
25	24.0		7.50				7.61	4.19		
26	23.0		5.92	7.69				3.95	6.43	5.82
27	21.0		7.64			9.54				
28	20.0						6.42	4.42	3.67	5.78
29	19.0			7.12			6.54	5.76	4.59	5.83
30	16.5	11.04								
31	6.5	9.02	3.92		6.68	6.33				
32	6.0		4.10	4.21						
33	5.0			3.84	2.93	5.95	3.18	3.07	3.74	3.95
Total number of bands			12	11	9	10	13	13	13	14

some other role such as the modification of membrane transport properties. However, it may also be a sensitive indicator of salt stress (Tramontano and Jouve, 1997). The contents of proline show wide variations among the different species under different habitat conditions (Table 3). It is clear that *Nitraria retusa* (a) of the Red Sea area attained higher values of proline contents during winter ( $8.5 \mu \text{mole g}^{-1}\text{F.wt.}$ ) reaching the maximum value among all the studied plants in *Nitraria retusa* (a) of the Sinai Peninsula during summer ( $12.5 \mu \text{mole g}^{-1}\text{F.wt.}$ ). The succulent halophytes of the two studied areas recorded relatively moderate amounts of proline content ranged between  $0.8$  to  $3 \mu \text{mole g}^{-1}\text{F.wt.}$ . *Salsola tetrandra* (a) and (b) of Sinai Peninsula attained their higher values of proline during winter ( $2.9$  and  $1.9 \mu \text{mole g}^{-1}\text{F.wt.}$ ). These values decreased significantly during summer to  $1.5$  and  $0.8 \mu \text{mole g}^{-1}\text{F.wt.}$  Proline is known to be a compatible solute, it accumulates in the cytoplasm without having

any detrimental effects on cytosolic enzyme activities and it is the least inhibitory of cell growth among all amino acids (Lutts *et al.*, 1996). Also, proline has been reported to play an important role in osmoregulation as a buffer against osmotic imbalance caused by high vacuolar ion concentration (Serrano and Gaxiola, 1994). Proline is accumulated by a number of different plants in response to both water stress and salinity (McCue and Hanson, 1990; Delauney and Verma, 1993). Proline accumulation could represent a compensatory mechanism for better survival during the period of drought stress and acted a role as an osmotic regulator (Nikolopoulos and Manetas, 1991). Thus the accumulation of proline in salt stressed plants is considered as one of the major physiological defense mechanisms of these plants against stressed habitats. *Limonium axillare* (b), *Nitraria retusa* (a) and (b) of the Red Sea area exhibited significant decreases in proline contents during summer, which

Table 5: Seasonal fluctuations in protein banding patterns of the studied species under different habitat conditions

Habitat		%									
Plant species		Red Sea					Sinai Peninsula				
Lanes	M.Wt.	Marker	<i>Nitraria retusa(a)</i>		<i>Nitraria retusa(b)</i>		<i>Nitraria retusa(a)</i>		<i>Nitraria retusa(b)</i>		
			L1	L2	L3	L4	L5	L6	L7	L8	
Rows	M.Wt.	Marker	Summer	Winter	Summer	Winter	Summer	Winter	Summer	Winter	
1	175.0	13.21									
2	135.0		6.10				8.44	8.52	5.66	4.45	
3	122.0				7.40						
4	108.0		7.45	11.84			6.85	6.04			
5	98.0										
6	86.0					9.09	6.98	6.42	7.31	5.43	
7	83.0	22.68			9.14						
8	81.0									6.12	
9	70.0		8.07				9.23				
10	62.0	13.17			13.60	19.76		12.55		7.91	
11	58.0		10.01	16.99			8.80	11.46	14.31	10.58	
12	47.5	10.99			13.01	13.77					
13	44.0		6.75				7.13	8.60	13.52	8.86	
14	36.0		7.85		11.99	12.80	8.47				
15	32.5	10.30	5.26	11.29				7.71	12.21	9.28	
16	31.0							8.31			
17	30.0		5.32	9.87							
18	28.0		6.71	12.43	10.53		8.88			5.25	
19	27.0					14.25		6.95	14.93	6.92	
20	26.0		6.35	8.73	8.53	8.38					
21	25.0	17.86									
22	24.0		6.24					6.18	10.38	7.20	
23	23.0		7.20	8.87	9.49	7.57	8.46			6.81	
24	22.0								7.44		
25	21.0		5.85	8.05			4.67	5.41			
26	20.0		4.26	5.42							
27	16.5	7.92			3.84		7.69				
28	15.0							4.22	2.71	5.67	
29	11.0		1.63	1.54	3.24	5.12	6.62	2.99	2.73	6.13	
30	8.0		1.75	1.24							
31	6.5	3.86								9.39	
32	5.0		3.19	3.70	9.23	9.25	7.79	4.64	8.80		
Total number of bands			17	12	11	9	13	14	11	14	

followed the effect of relatively high total soluble salts in soil during winter (Table 2). Csonka *et al.* (1988); Kishor *et al.* (1995) and Hassanein (2000) suggested that the accumulation of proline in tissues confers some salinity tolerance. Proline may have a significant role to metabolism other than just osmotic adjustment (Hare and Cress, 1997).

The total soluble protein contents in all cynrohalophytes of the Red Sea area except for *Nitraria retusa* (b) attained significant increases during summer. *Nitraria retusa* (b) of the Red Sea area showed the highest value of total soluble protein (4514.9  $\mu\text{g g}^{-1}\text{F.wt.}$ ) during winter followed by highly significant decrease to 2034.4  $\mu\text{g g}^{-1}\text{F.wt.}$  during summer. It is of interest to notice that *Nitraria retusa* (a) of the Red Sea recorded the highest value of total soluble protein among all the studied plants (5241  $\mu\text{g g}^{-1}\text{F.wt.}$ ) during summer. From Table 3 it is apparent that both of *Nitraria retusa* (a) and (b) of the Sinai Peninsula accumulate higher values of total soluble protein during winter (3195.6 and 3445.9  $\mu\text{g g}^{-1}\text{F.wt.}$ ,

respectively). The total soluble protein contents in both *Salsola tetrandra* (a) and (b) of Sinai Peninsula showed higher values during summer (2198.9 and 1136.5  $\mu\text{g g}^{-1}\text{F.wt.}$ , respectively) than those showed in winter (3236.5 and 1709.5  $\mu\text{g g}^{-1}\text{F.wt.}$ , respectively). Plants frequently produce a number of unique proteins as a part of their response to environmental stresses (Orcutt and Nilsen, 2000). Salinity also, induced distinct protein changes, which may be either qualitatively or quantitatively (Ramagopal, 1987; Hurkman and Tanaka, 1987).

Several proteins induced by salt stress are also induced by water stress and chilling and the signal transduction pathways may be very similar for all these stresses (Wu *et al.*, 1997). There were variations among the studied plants in protein configuration. It has been shown in the present work that protein pattern of *Avicennia marina* (Fig. 4 and Table 4) during winter period using one dimensional SDS-PAGE, was characterized by the presence of eleven protein bands which were associated

Table 6: Seasonal fluctuations in protein banding patterns of the studied species under different habitat conditions

Habitat		%								
Plant species	Red Sea				Sinai Peninsula					
	<i>Arthrocnemum gilaucum</i>		<i>Halocnemum strobilaceum</i>		<i>Salsola tetrandra</i> (a)		<i>Salsola tetrandra</i> (b)			
Lanes		L1	L2	L3	L4	L5	L6	L7	L8	
Rows	M.Wt	Marker	Summer	Winter	Summer	Winter	Summer	Winter	Summer	Summer
1	175.0	11.39								
2	138.0		9.41	4.93						5.61
3	130.0		8.49		2.33	4.42	5.18	3.97		
4	88.0		8.08			2.98	5.43	4.55		
5	83.0	29.36							10.38	
6	73.0									32.22
7	64.0								15.15	
8	62.0	11.58								
9	56.0		16.62	35.70	12.13	24.45	24.68	24.83	19.23	11.24
10	51.0			9.62						
11	50.0									
12	47.5	8.38								
13	44.0		10.27	7.34	10.92					
14	40.0			6.26	6.09	5.86				
15	38.0				7.26					
16	35.0		7.75	5.97		4.50	11.76	10.15	11.98	6.31
17	32.5	7.63		5.65		6.27				
18	31.0		8.39		6.55	6.63	7.56	7.78	10.11	5.94
19	29.0				6.44	6.83	6.33	6.49		6.50
20	27.0		5.76	4.45						
21	26.0				7.95		8.22	7.63		8.11
22	25.0	23.78	3.86			9.17			11.09	
23	24.0		4.52							12.09
24	23.0		10.02	14.63	8.72	6.46	6.99	6.37		
25	21.0				11.50	9.13	8.18	7.26	22.05	7.04
26	18.0				4.08	2.41	3.13	3.47		
27	16.5	5.76				0.77				
28	8.0		2.83		2.52	1.75	4.23	5.89		
29	6.5	2.13	2.35							
30	5.0		0.92	5.45	7.58	6.36	5.99	7.47		
31	4.0		0.72		5.93	2.01	2.32	4.14		4.96
Total number of bands			15	10	14	16	13	13	7	10

with high total soluble salts in its soil (Table 2). This number of bands were increased during summer to twelve bands with the appearance of new protein bands and the disappearance of others in response to dryness of summer and salt stress. Water stress during the summer may induce the accumulation of stress responsive proteins of molecular weights 112.0, 83.0, 52.0, 34.0, 24.0, 21.0 and 6.5 kD some of these proteins were thought to belong to dehydrins group (25-60 kD) or aquaporins (25.0-30.0 kD) (Arora *et al.*, 2000). Aquaporins are groups of membrane bound proteins that regulate membrane water permeability in cells and may influence water movement among tissues and organs. The permeability of membranes to water depends on the number of aquaporins in the membrane and their effectiveness at transporting water (Maurel, 1997). Aquaporins selectively transport water and do not co-transport any other ions. Therefore, these proteins are important for turgor maintenance and water distribution within and among cells but not so important for ion homeostasis. Dehydrins

are predicted to act as stabilizers (chaperon-like) that prevent or reduce the denaturation of other cellular micromolecules under dehydrative conditions (Campbell *et al.*, 1998). Also they have a protective role in the survival of cells under water stress due to their function as ion trap in dehydrating cells sequestering ions as they became concentrated (Close, 1996). Thus the new synthesizing protein bands may be induced for stress adaptation during summer while the disappearance of other bands may be attributed to the degradation of some proteins (Vierling, 1991).

Nine protein bands were detected for *Atriplex farinosa* (Fig. 4 and Table 4) which ranged from (130.0-5.0 kD) during summer. It is of interest to note that the *de novo* synthesized protein bands during summer (52.0 and 44.0 kD) have disappeared during winter. These bands may belong to dehydrin group (25-60 kD). These proteins have an important role to plant cell (Close, 1996 and Maurel, 1997). On the other hand, during winter *Atriplex farinosa* accumulated more protein bands in response to the

highest total soluble salts in the associated soil (Table 2), which increased to ten bands ranging from 130.0 to 5.0 kD with the appearance of three new protein bands of molecular weights 99.0, 58.0 and 21.0 kD. In this respect, Godoy *et al.* (1990) and Vierling and Nguyen (1992) found that responses of plant under salt stress, being characterized by the synthesis of high and low molecular weight proteins.

Protein banding pattern of *Limonium axillare* (a) attained the same number of protein bands during both seasons. During summer a new protein band (28.0 kD) was induced for the acclimation of this plant to both drought and high total soluble salt stresses during the dry period (Tables 1 and 2). This induced protein may belong to dehydrin group (25-60) or aquaporins (25-30) which have an important role in plant cell (Maurel, 1997). *Limonium axillare* (b) attained thirteen bands of proteins during summer of molecular weights ranging from (155.0-5.0 kD) and increased to fourteen bands during winter with molecular weights ranging from (186.0-5.0 kD). The new induced protein (186.0 kD) during winter may be one of the adaptive responses of *Limonium axillare* (b) to salinity conditions.

Acclimation of *Nitraria retusa* (a) (Fig. 5 and Table 5) to the adverse conditions of drought and salinity in summer has induced seventeen protein bands of molecular weights ranging from 135.0 to 5.0 kD, which decreased to twelve bands during winter. During summer five new protein bands appeared (135.0, 70.0, 44.0, 36.0 and 24.0 kD), which may include dehydrins or aquaporins. These proteins may be important in plant adaptation to the desiccation and ionic effects of saline conditions (Yamaguchi-Shinozaki *et al.*, 1992).

A marked reduction in the number of protein bands was observed in *Nitraria retusa* (a) grown on coastal regions; 7 km south of Safaga compared with *Nitraria retusa* (b) grown at 200 m west of the shoreline of Red Sea. Nine bands were recorded during winter of molecular weight ranging from 86.0 to 5.0 kD which increased to eleven one with molecular weights ranged from 122.0 to 5.0 kD during summer. In response to elevation in total soluble salts in soil (Table 2) and dry conditions of summer (Table 1) in *Nitraria retusa* (b) new proteins were induced with molecular weights of 122.0; 83.0, 28.0 and 16.5 kD. In this concern Zhao and Harris (1992) reported that a large number of genes are induced and accumulated in response to salinity stress, many of which have a known function to help these plants to accommodate stress habitats.

Cryohalophytes from Sinai Peninsula which include *Nitraria retusa* from two sites (coastal and inland) showed that in summer *Nitraria retusa* (a) attained

thirteen protein bands of molecular weights ranging from 135.0 to 5.0 kD, these bands increased to fourteen bands in winter. The increment in the number of protein bands during winter may be due to the high level of total soluble salts in soil where it reached its highest amount during this period 85760 ppm at (0-20 cm depth) (Table 2). These results indicated that salt stress induced *de novo* synthesis of stress proteins, suggesting that plant cells were able to monitor different levels of stress intensity and modulate gene expression accordingly these results agreed in part with those of Cherry, (1994). *Nitraria retusa* (b) showed eleven bands of protein during summer which increased to fourteen ones of molecular weights ranging from 135.0 to 6.5 kD in winter. Three new protein bands were induced during winter of molecular weights 81.0, 62.0 and 28.0 kD, some of these proteins were thought to belong to dehydrins group (25-60 kD) or aquaporins (25.0-30.0 kD). Generally, *Nitraria retusa* (b) showed a decrease in the number of bands and mostly an increase in their intensities compared with those detected in *Nitraria retusa* (a). This indicates the diversity of these plants with their different habitats.

Succulent halophytes from Red Sea region as represented by *Arthrocnemum glaucum* (Fig. 6 and Table 6) attained ten protein bands of molecular weights ranging from 138.0 to 5.0 kD during winter, increased to fifteen ones of molecular weights ranging from 138.0 to 4.0 kD in summer which was associated with relatively high amount of TSS (Table 2). During summer new responsive proteins were induced (130.0, 88.0, 31.0, 25.0, 24.0, 8.0, 6.5 and 4.0 kD), some of these proteins may belong to dehydrins (25-60 kD) or osmotin (25 kD). Close (1996) reported that induction of dehydrins is a general stress response rather than a salt specific response. On the other hand, Osmotin (a type of responsive protein) might play an important role in osmotic adjustment to the cells and adaptive mechanism for salt and drought stresses, either by facilitating rapid accumulation of proline and glycinebetaine localized in the cytoplasm and acts a non toxic osmoticum following osmotic adjustment to occur without perturbing metabolic function (Moons *et al.*, 1995; Orcutt and Nilsen, 2000) or by providing certain metabolic alterations in the cells, which may be helpful in osmotic adjustment (Kononowicz *et al.*, 1993).

*Haloacnemum strobilaceum* (Fig. 6 and Table 6) (Sinai Peninsula) attained fourteen protein bands during summer which increased to sixteen ones ranging between 130.0 to 4.0 kD in winter. During winter some important protein bands were induced like osmotin (25.0kD). Ericson and Alfinito, (1984) reported that upon salinity stress, osmotin can be increased enough to account for 10% of the total protein. In addition to these proteins another kinds of

molecular weight proteins 88.0, 35.0, 32.0 and 18.0 kD were observed. These proteins may be induced in response to the high level of TSS (Table 2) during this period which may include aquaporins, dehydrins and other unknown proteins which enable the plant species to accommodate with this stress conditions (Close, 1996).

Species of wide ecological amplitude which include both *Salsola tetrandra* (a) and (b) (Fig. 6 and Table 6). *Salsola tetrandra* (a) attained the same number of protein bands during both seasons with no difference in their molecular weights, which ranged from 130.0 to 4.0 kD. On the other hand, *Salsola tetrandra* (b) showed a higher number of protein bands during winter (ten bands) compared to those attained during summer (seven bands). During summer one of the most adaptive responsive protein was appeared which may be osmotin (25.0 kD) and was associated with the highest amount of total soluble salts in the soil (Table 2). Osmotin may represent induced protein that can be accumulated specifically in response to low water potential and are putatively involved in the maintenance of cellular homeostasis under prolonged stress (Ericson and Alfinito, 1984). Osmotin appeared also to provide osmotic adjustment to the cell by facilitating the accumulation of solutes and providing metabolic alteration in the cells which may be helpful in osmotic adjustment (Singh *et al.*, 1987).

It is of interest to mention that the crynhalophytes *Avicennia marina*, *Atriplex farinosa*, *Limonium axillare* (a) and (b) are characterized by the presence of 4 common salt adaptive proteins of molecular weights 5.0, 21.0-24.0, 28.0-31.0 and 72.0 kD, while *Nitraria retusa* (a) and (b) grown in Red Sea and Sinai are characterized also by the presence of 4 common salt adaptive proteins of molecular weights 5.0, 11.0, 22.0-24.0 and 28.0-30.0 kD. Succulent halophytes, *Arthrocnemum glaucum* and *Halocnemum strobilaceum* share 4 common salt adaptive proteins, their molecular weights are 4.0-5.0, 23.0, 40.0-44.0 and 56.0 kD. In addition, species of wide ecological amplitude which are represented by *Salsola tetrandra* (a) and (b) grown in Sinai are characterized by the presence of 5 common salt adaptive proteins of molecular weights 4.0-5.0, 21.0-24.0, 29.0-31.0, 35.0 and 56.0 kD.

These results suggest that the production of these stress adaptive proteins may represent a common mechanism which enables halophytes to withstand the harmful effect of salinity. Some of the salt adaptive proteins may be involved in many functions related to some aspects of protein metabolism such as stabilizing native proteins (Anderson and Guy, 1991), preventing aggregation of denaturated proteins (Sheffield *et al.*, 1990; Ellis and Van der Vies, 1991), refolding stress denaturated proteins (Lee *et al.*, 1995), participating in the folding and assembly of

nascent polypeptides into functional forms (Boston *et al.*, 1996), facilitating translocation across membrane (Kang *et al.*, 1990) and targeting non-native and folding incompetent polypeptides for degradation (Chiang *et al.*, 1989). These functions fall under a generalized version of the molecular chaperone concept (Li *et al.*, 1999).

It is evident from the obtained results that creating a favorable water potential gradient is the most challenging factor in the adjustment of the different species to various stress conditions. In this respect, the behavior of halophytes and species of wide ecological amplitude showed different mechanisms in their adjustment to different habitat conditions. The climate of the studied areas is obviously hyper arid. There are wide fluctuations in air temperature, both diurnally and seasonally and scanty rainfall occurs during winter. The variability of rainfall is a feature of both monthly and yearly records.

Quaternary ammonium salts include glycinebetaine,  $\beta$ -alaninebetaine and trigonelline which are compatible solutes with the cytoplasm can be accumulated in response to low water availability. This was confirmed by the finding that most of the studied species accumulated high levels of glycinebetaine in response to the high total soluble salts and low moisture availability of the associated soil.  $\beta$ -alaninebetaine, one of the important compatible solutes, was recorded only in *Limonium axillare* which was associated with the highest value of total soluble salts. Trigonelline, also one of the quaternary ammonium salts, was accumulated in addition to glycinebetaine in *Nitraria retusa* (Red Sea) in response to the relatively high total soluble salts during winter. *Nitraria retusa* (Sinai Peninsula) did not accumulate trigonelline which may be explained as a result of the high temperature and climatic aridity in the Red Sea, compared to the climate of Sinai Peninsula. Total protein and proline contents in most of the studied species tended to be accumulated under both salt and drought stresses. Cymnhalophytes tended to accumulate higher values of proline, but the highest was recorded by *Nitraria retusa* grown on the coastal region of Delta Wadi Sudr ( $12.5 \mu \text{mol g}^{-1} \text{F.wt}$ ) during summer. Such accumulation may be related to more favorable conditions for carrying out metabolic processes. One of the most outstanding features in the adjustment of the studied species under different habitat conditions was the biosynthesis of certain proteins which may play principal roles in the building up of osmotic potential. Regarding protein electrophoresis during both summer and winter in all the studied species (halophytes and species of wide ecological amplitude), some protein bands disappeared and others appeared which may be important in plant adaptation to both salinity and drought stresses. Water stress during summer induced the accumulation of

responsive proteins of molecular weights (25-60 kD) which were thought to belong to dehydrins groups. Also, salt stress in most of the studied species was associated with the biosynthesis of a protein with molecular weight 25 kD which might be osmotin. Osmotin play an important role in osmotic adjustment. Some of the induced proteins might be aquaporins which are a group of membrane bound proteins (25-30 kD) that affect water transport through membrane and not ions, their abundance in the tonoplasm makes them important for water and ionic balance between the cytoplasm and the vacuole.

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