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Adaptive Responses of Some *Zygophyllum* Species in Wadi Hagul, Egypt

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Three species, *Zygophyllum coccineum* L., *Zygophyllum decumbens* Delile and *Zygophyllum simplex* L., (Zygophyllaceae) were collected from three localities of Wadi Hagul in Egypt to study their adaptive responses to different environmental conditions. Plant analyses (degree of succulence, water content, total pigments, proline, antioxidant enzymes, total phenols, ascorbic acid, hydrogen peroxide and malondialdehyde) were estimated to study their roles in the adaptive responses of those plants under their natural drought conditions. Results have revealed that xerophytes can further be divided into two groups depending on their drought tolerance strategies. The first group includes perennial plants (*Zygophyllum coccineum* and *Zygophyllum decembens*) that able to tolerate and resurrect following desiccation into photosynthetically active states in a short period of time, the other is the dessication avoiding group; annual or biennial plant (*Zygophyllum simplex*), which possess an improved water-storing organ, efficient water conduction within the plant body and/or a combination of these features. Moreover, plants in this group (true xerophytes) are thought to have a unique system to cope with stress tolerance, since they are able to defend their photosynthetic apparatus and other fragile cellular components from composite stresses in the harsh environments in which they are found.

Key words: Drought, succulence, *Zygophyllum*, Wadi Hagul.

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Introduction

Most plants are exposed to water stress due to extreme soil water deficits in arid and semi-arid environments. The survival of land plants in such areas relies on the availability of water and their adaptation under stress (Kramer, 1984). Drought resistance is a complex trait involving several interacting properties (Khan, 2003 and Scholz *et al.*, 2012).

In arid and semi-arid regions, seasonal variations may lead to differential physiological responses in plants inhabiting such environments. Many studies have been cited on plant adaptations to high temperature, and salt stresses (Joyce *et al.,* 1984; Murakeozy *et al.,* 2003 and Kusaka *et al.*, 2005).Ecophysiological studies have been investigated in elucidating plant function and identifying traits that are adaptive to specific environmental conditions (Ackerly*et al*., 2000).

Wadis are the most widespread ecosystems in the mountainous desert of the world (Fossati*et al*., 1999). The wadi system is an extreme case of a temporary inundated ecosystem in which the duration of flooding is shorter than the dry period (Evenari, 1985). Various habitats can be identified in a wadi: channels, bars, banks, etc. (Abdel Rahman and Batanouny, 1965). A wadi system embraces all the biotypes and related biocenases found in arid or hyper arid zones (Salama*et al*., 2012).

Zygophyllum species represent a group of succulent plants which are drought resistant or salt tolerant living under severe dry climatic conditions (Batanouny and Ezzat, 1971). Succulent plants can survive under the severe water stresses depending on the stored water in their tissues (Maximov, 1929). They can reuse the large quantities of water they save (Nobel, 1977).

Therefore, in the present study we focus on the mechanisms of the adaptive responses of the studied *Zygophyllum*species in WadiHagul, Egypt (*Z. coccineum*,*Z. simplex* and *Z. decumbens*) in order to understand the eco-physiological adaptation traits by these plants. Accordingly, degree of succulence, water content, total pigments, proline, antioxidant enzymes, total phenols, ascorbic acid, hydrogen peroxide and malondialdehyde were estimated to investigate their roles in adaptation.

Z. coccineum L. is a succulent Shrub, up to 75 cm; leaflets 2, bright green, glabrous, cylindrical, at least 10 mmlong; capsule 8-10 mm, long and apex obtuse.*Z. simplex* L. is a glabrous, mat-shaped sappy herb with simple cylindrical leaves, 9-10 mm, long ; yellow flowers;capsule minute, only 2 mm. deeply 5-parted.*Z. decumbens* Delile is a shrublet with prostrate or decumbent branches; leaves of one pair of broadly obovate, flattened, fleshy leaflets; flowers small, capsule pear-shaped, deeply 5-parted, apex entire (Boulos , 2000).

Study area:

WadiHagul is located in the northern portion of the Eastern Desert of Egypt within Cairo-Suez district, delimited by latitudes 29°48'28" - 29°57'43" N. and longitudes 32°09'32" - 32°17'27" E (Fig. 1). It is a hydrographic wadi located in Suez Governorate (Egypt), the study location is situated 381 km north east of the approximate center of Egypt and 112 km east of the capital Cairo. It is one of the main drainage lines dissecting Gebel Ataqa (C.817m) when the tributaries of the southwestern and western slopes drain into this wadi (Zahran and Willis, 1992).

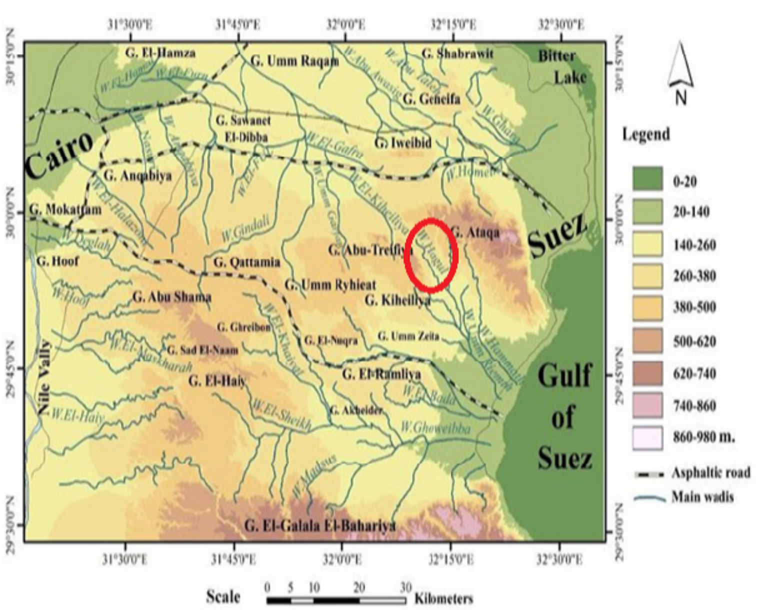

Fig.(1). Map of Cairo-Suez road showing the location of the studied site Wadi Hagul.

Materials and Methods

Plant materials used in the present investigations are**:** The plant materials used in the present study were collected from three sites along wadi Hagul, each of them represent one stream of the wadi. The first studied plant is *Zygophyllumcoccineum* which is collected from upstream midstream and downstream sites along the study area, *Z. simplex*was collected from two upstream and midstream, while *Zygophyllumdecumbens*was collected from midstream and downstream.

Soil analysis

Soil samples were collected seasonally during dry and wet seasons at the surface (0-20 cm) and sub-surface (20-40 cm) depths. Some physical and chemical parameters of such soil samples were analyzed. Particle size (%) was determined by the dry sieving to separate soil fractions according to Wentworth scale (Udden,1914; Wentworth, 1922;Krumbein and Sloss, 1963 and Ryan *et al*., 1996). Soil reaction (pH) was determined in the soil solution 1:1 using a portable pH-meter (ModelIonlab pH level 1) (Richards, 1954). Electrical conductivity (EC) of the soil extracts was determined as described by Richards (1954) and expressed as mmhos. The anions and cations (Cl^- and K^+) of the soil extracts were analyzed following the method described by Richards(1954) and Rayan (1996) and their values expressed as meq Calcium carbonates was determined volumetrically using Collin's Calcimeter (Piper, 1950).

Ecophysiological analysis:

Degree of succulencewascarried out andcalculated according to Dehan and Tal (1978) and Ahmed and Shalaby (1985).The photosynthetic pigmentswere determined using spectrophotometric method (Metzner*et al.* 1965).Free proline contentwas estimated photometrically in an acid ninhydrin assay (Bates *et al.* 1973). For the determination ofoxidative enzyme activities(superoxide dismutase (SOD), catalase (CAT), peroxidase (POX), ascorbic acid oxidase (ASO) and polyphenol oxidase (PPO)),were extracted from fresh materials following Guerrier and Strullu(1990),CAT, POX and PPO activities were determined according to Kar and Mishra (1976), SOD activity was determined followingMarklund andMarklund(1974), while ASO activity was measured according to

Diallinas *et al.* (1997).Total phenolswere determined following Malik and Singh(1980) whileascorbic acidwas estimated by the method of Mukherjee and Choudhury (1983), hydrogen peroxidecontentwas determined according to Velikova *et al.* (2000) and lipid peroxidation as indicated bymalondialdehyde (MDA)was estimated according to Hodges *et al.* (1999).

Statistical analysis and data confirmation

Data were statistically analyzed using the least significant difference (LSD) ($p<0.05$) between mean values for the results of dry and wet seasons to compare significant differences. The computations were done y using SPSS software Version (16.0) .Values presented are means ± standard deviation (SD) of three replicates.

Results

Soil characteristics

The soil texture of associated profile of the three studied locations was mainly formed of sand.Soil reaction (pH) was slightly alkaline (Tables 1and 2). In upstream location, the soil texture is sandy. Electric conductivity (E.C.) was high (1.1-1.4 mmhosL^{-1}) at the surface soil layer indicating more soluble salts in the surface layer. The soil pH ranged between almost neutral to slightly alkaline (7.75-8.5). Cations showed higher Ca^{+2} (2.94meq L^{-1}), Mg^{+2}(2.45meq L^{-1}), Na^{+}(6.33meq) and K^{+}(0.23meq L^{-1}) during dry season. Anions revealed higher Cl^{-} (8.33 meq L^{-1}) and HCO_3^{-} (1.66meqL^{-1})during dry season, while SO_4^{-2} was relatively low (3.81 meqL^{-1}), calcium carbonate was ranged between 15.5 to 5.7, attained comparatively higher values in dry season. In midstream location,the soil texture is sandy, soil reaction is tended tobe slightly alkaline (7.52-8.4), E.C. amounted to 2.36 mmhos during dry season in the upper layer. Cations prevailing were Na^{+} (12.9 meqL^{-1}), Ca^{+2}(6.24meqL^{-1}) followed by Mg^{+2}(4.16 meqL^{-1}) reached highest contents among all the studied soil profile during dry season, but K^{+}(0.35meqL^{-1}) is of low content.Prevailing anions are mostly Cl^{-}(19.5 meqL^{-1}) followed by SO_4^{-2} (9.1 meqL^{-1}). Soil texture at the downstream location isgravely at 20-40 cm, while 0-20 cm layer isalmostlysandy with gravel fractions. The E.C. ranged between 0.25 mmhos in upper layer (0-20 cm) to 0.14 mmhos at depth (20-40 cm) during dry season. The cation content decreased with increase depth till 40 cm during dry season. Anion of chlorides(0.95meqL^{-1}) surpassed sulphates (0.89 meqL^{-1}).

1. Degree of succulence and Water content

Both parameters were greater in plants collected in wet season than in plants collected in dry season.*Zygophyllum decumbens* collected during wet season from midstream location attained the highest degree of succulence (4.7) and water content (79.7) which showed significant difference from the same plant collected from the same location in dry season.(Table 3).

2. Photosynthetic pigments

Zygophyllum simplex collected from midstream location(during wet season) showed the highest total pigments(13.71mg /g f.wt of leaves),while *Zygophyllumcoccineum* collected from upstream location (during dry season)showed the lowest value (4.4 mg /g f.wt of leaves).(Table 4).

3. Proline

It has been shownin Table (5) that;proline content increased in dry season than in wet season showing significant difference. *Zygophyllum coccineum* collected from upstream location(during dry season) showed the highest proline content (115.97µg /g f.wt),while *Zygophyllum simplex* collected from midstream location (during wet season) showed the lowest value(43.62µg /g f.wt).It is noticeable that, the midstream location showed the lowest proline content in all species compared with other locations.

4. Antioxidant enzymes

The activity of these enzymes was significantly increased during dry season in the three studied species (Table 6).The activity of all enzymes decreased obviously in mid stream location in all three species.

The activity of SOD showed its highest value within *Zygophyllumdecumbens* in downstream location (35.87unit /mg protein), while the least value within *Zygophyllumcoccineum* in downstream location (3unit /mg protein).

The activity of CAT showed the highest value with *Zygophyllumdecumbens* in downstream location (149.17µmolH2O2 destroyed / g f.wt / min), while the lowestvalue is recorded with *Zygophyllum coccineum*in midstream location (3.7µmol H2O2 destroyed / g f.wt / min).

The activity of Peroxidase (POD) showed the highest value with *Zygophyllumdecumbens* in downstream location (84.3 g f.wt/min.), while the lowest value is recorded within *Zygophyllum coccineum* in midstream location (0.13 g f.wt / min.).

The activity of ASO showed the highest value with *Zygophyllum decumbens* in downstream location (12.1mM of ascorbate oxidized/g f.wt/min.), while the lowest value is recorded with *Zygophyllum coccineum* in midstream location (0.17 mM of ascorbate oxidized/g f.wt/min).

The activity of PPO showed the highest value with *Zygophyllum coccineum*in upstream location (53.98g f.wt/min.), while the lowest value is recorded within the same species in midstream location (49.52g f.wt/min.).

5. Total phenols and Ascorbic acid

Total phenols show the same behavior as ascorbic acid, as the content increased in wet season than in dry one.*Zygophyllum decumbent* in midstream location had the highest total phenols content (1.71mg/g f.wt), while *Zygophyllum coccineum* in upstream showed the lowest content (0.48mg / g f.wt). *Zygophyllum coccineum* located in midstream revealed the highest ascorbic acid content (499.54mg/100g f.wt) and *Zygophyllum simplex* in upstream showed the lowest content (152.45mg/100g f.wt).

6. Hydrogen peroxide and Malondialdehyde contents

Zygophyllum decumbens collected from downstream location (during dry season) showed the highest hydrogen peroxide content among all plants (219.01mM/g f.Wt),while *Zygophyllum simplex* collected from midstream location (during wet season) showed the lowest value (111.37mM/g f.Wt), which in case of malondialdehyde showed the same behavior (0.103nmol/g f.wt), whereas *Zygophyllum coccineum* collected from upstream location (during dry season) showed the highest malondialdehyde content among all plants (0.95nmol/g f.wt).

Discussion

Soil characteristics

Soil factors play important role in plant growth, chemical composition and distribution of plants under different habitat conditions. In the present study soil samples supporting the growth of the different studied species, were collected from the sites where the three selected *Zygophyllum* species (*Zygophyllum coccineum*, *Zygophyllum decumbens* and *Zygophyllum simplex*) grown.

The three studied locations lie successively declining contours from the upstream to the downstream of Wadi Hagul. The inclination angle of the slope of the wadi course is quite noticeable and consequent features of soil erosion and deposition occur along the wadi course.

The soils of the studied areas are aridisols (dry, desert-like soils) or entisols (weakly-developed soils); characterized by weak profile development, coarse texture, low moisture content, low nutritional content and slightly alkaline or neutral reaction.(Zaki, 1995).

1. Degree of succulence and Water content

This may be due to the morphological and anatomical characteristics of this plant. This conclusion coincided with Waly*et al*., (2011), who studied the anatomical characteristics of the three studied *Zygophyllum* species in this work.Succulence is considered as mechanism through which plants are adapted to adverse environmental conditions including salinity and drought (Ahmed and Shalaby, 1985 and Morsy, 1996; 2002).

Species of the genus *Zygophyllum* are among the succulent xerophytes which can withstand the adverse conditions of the desert environments; shortage of water, scorching temperature and likely soil salinity (Kassas and Batanouny, 1984). Zaki (1995) indicated that *Zygophyllum coccineum* attained the lowest degree of succulence in summer. It might be associated with a decrease in moisture availability in which resulted in a decrease in total available carbohydrates and nitrogen uptake.

2. Photosynthetic pigments

All species showed decreasing in photosynthetic pigments in dry season, as plants lose their pigments due to drought conditions of their habitats and they may have another strategy to overcoming drought rather than pigment accumulation. These results are in accordance with Wingler *et al.* (1999), it is reported that the combined effects of heat and light stress on photosynthesis superimposed with drought will be more complex. Under these conditions, plants are known to lose chlorophyll (Havaux and Tardy, 1999). Moreover, drought not only causes dramatic loss of pigments, but also leads to disorganization of thylakoid membranes (Ladjal *et al.*, 2000).

3. Proline

Accumulation of the amino acid proline is one of the most frequently reported modifications induced by water deficit, salt, and waterlogging stresses in plants and is thought to be involved in stress tolerance mechanisms, although its precise role remains controversial (Gadallah, 1995; Lutts *et al*., 1999 and Yiu *et al*., 2009).

The plant free proline massive accumulated when the plants subject to drought stress, is due to decrease inproline dehydrogenase activity is decreased and weaken in proline oxidation; drought suppressed the protein synthesis, decreased proline utilization and increasedproline accumulation in plants (Zhan *et al*., 2011).

4. Antioxidant enzymes

To mitigate the oxidative damage initiated by ROS (during dryseason), *Zygophyllum* spp. developed a complex defense antioxidative system including low molecular mass antioxidant , hydrogen peroxide, proline as well as antioxidant enzymes; ASO, SOD, CAT, PPO and POD.

SOD is the front line enzyme in ROS attack since it rapidly scavenges superoxide, one of the first ROS to be produced, dissimulating it to oxygen and H_2O_2 (Bowler, 1992).However, this reaction only converts one ROS to another, and H_2O_2 also needs to be destroyed since it promptly attacks thiol proteins.

The major enzymatic cellular scavengers of H_2O_2 are catalase and ascorbate peroxidase (Noctor, 1998). They have however different affinities for this ROS and seem to have different cellular roles in H_2O_2 scavenging. SOD controls the first threshold of the water–water cycle of antioxidant system (Asada, 1999 and Zhu *et al.,* 2009a). It plays a key role in quenching active oxygen (Fu and Huang, 2001), working as catalyzing the dismutation of O_2^- into H_2O_2 which are eliminated by CAT, POD and other antioxidant enzymes.

Catalases and peroxidases (CAT and POD) play an essential role in scavenging for H_2O_2 toxicity. The combined action of CAT and SOD converts the toxic superoxide radical (O_2^-) and hydrogen peroxide (H2O2) to water and molecular oxygen (O_2), thus averting the cellular damage under unfavorable conditions like drought stress (Noctor *et al*., 2000; Reddy *et al*., 2000 and Chaitanya *et al*., 2002).

The activities of CAT (9.35U·g-1·min-1) and POD (257.53 U·g-1·min-1) were higher in *Caraganakorshinskii*compared to the other four desert plants (*Calligonum mongolicum, Haloxylon ammodendron, Nitraria sphaerocarpa and Tamarix ramosissima*) and increase in CAT and POD activity helps to overcome the damage of tissue metabolism by reducing the toxic level of H_2O_2 (Zhou *et al*., 2013).

Regarding the activities of the antioxidant enzymes it is apparent that, there is a direct relation between activities of those enzymes and the climatic conditions and edaphic factors of soil (Table 2), the midstream location was rich in the percentage of silt and clay, cations and anions as compared with other upstream and downstream locations.

A common effect of drought stress is the disturbance between the generation and quenching of reactive oxygen species (ROS) (Smirnoff, 1998).

5. Total phenols and Ascorbic acid

The accumulation of phenolic compounds varies strongly with the growth state, development and responses to environmental stresses and is a result of balance between biosynthesis and further catabolism (Korkina, 2007).

Ascorbic acid (ASC) is the primary water-soluble antioxidant in plants and animals (Levine *et al*., 1995 and Siesand Stahl, 1995).Ascorbate is a major soluble redox molecule with pivotal roles in allowing several metabolic pathways towork properly. ASC regenerates other metabolites, among which tocopherols, from oxidative damages and protects the catalytic site of a number of enzymes (e.g., hydroxylases) from irreversible oxidation, possibly caused by reactive oxygen species (ROS) in both animal and plant cells. It can be used as substrate or enzyme cofactor in various biological reactions (Lodge, 2008 and De Gara *et al*., 2010).

Enzymatic ASC oxidation mainly occurs in plants through the reactions catalyzed by ascorbate oxidase (AOX) and ASC peroxidase (APX), two typical plant enzymes. AOX is an apoplastic enzyme involved in cell elongation (Takahama and Oniki, 1994). The down-regulation of this enzyme causes a shift in the apoplastic ASC pool toward its reduced state; it also increases plant yield during water deficit, through a carbon flux re-allocation, but does not determine a significant ASC increase in the investigated tissues (Garchery *et al*., 2013).The ascorbic acid content of plants is observed to be reduced by water stress (Abdel-Kader, 2001; Manivannan *et al*., 2007 and Nair *et al*., 2008). Water stress caused significant reduction of the ascorbic acid of the leaves of *Gongronem alatifolium* (Osuagwu,2012).

Generally, increasing in total phenols and ascorbic acid during wet season may be due to prepare for the dry season.

6. Hydrogen peroxide and Malondialdehyde

It has been shown that hydrogen peroxide and malondialdehyde content increased in dry season than in wet season showing significant difference. The midstream location showed the lowest hydrogen peroxide and malondialdehyde content in all species compared with other locations.

Malonyldialdehyde (MDA) is a product of lipid peroxidation- content has been considered as an indicator of oxidative damage (Neto *et al.,* 2006).

The formation of MDA was considered as a measure of lipid peroxidation that was induced by a high water stress level **(Jones, 1999)**. MDA, a decomposition product of polyunsaturated fatty acid hydroperoxides, has been utilized very often as a suitable biomarker for oxidative stress. Usually the increase in lipid peroxidation was simultaneously accompanied by an increase in hydrogen peroxide levels.

Zygophyllum decembens, in this studyshowed highcapacity to adapt to the surrounding circumstances; by increasing some antioxidant enzymes (ASO), (SOD), (CAT) and (POD), and hydrogen peroxide during dry season and degree of succulence during wet season. Meanwhile, *Zygophyllum coccineum* increased proline, PPO and malondialdehyde during dry season and ascorbic acid during wet season, whereas, *Zygophyllum simplex* increased total pigments and total phenols during wet season.

Table (1). Granulimetric analysis of soil profiles associated with the studied species

Associated plants	Depth	Very fine gravel (%)	Very coarse sand (%)	Coarse sand (%)	Medium sand(%)	Fine sand (%)	Very fine sand(%)	Silt & Clay (%)	Texture
		(4-2 mm)	(2-1 mm)	(1-0.5 mm)	(0.5-0.25 mm)	(0.25-0.125 mm)	(0.125-0.063 mm)	(<0.063 mm)	
Zygophyllumcoccineum	0-20	12.89	8.66	16.22	26.72	21.89	9.7	3.92	Sandy
Zygophyllum simplex	20-40	7.29	11.96	24.95	40.59	11.7	2.11	1.4	Sandy
Zygophyllumcoccineum *Zygophyllumdecumbens*	0-20	2.25	4.28	16.98	28.9	27.64	14.91	5.04	Sandy
Zygophyllum simplex	20-40	3.83	5.25	11.55	33.6	32.27	8.85	4.65	Sandy
Zygophyllumcoccineum *Zygophyllumdecumdens*	0-20	21.89	19.77	23.64	17.09	9.88	4.15	3.58	Sandy
	20-40	51.27	24.66	13.12	5.45	3.57	1.19	0.74	Gravely

Table (2). Soil reaction, electrical conductivity and chemical analysis of soil pastes of wadi Hagul.

Location	Associated plants	Depth	pH		EC mmhos/cm		soluble anions meqL[-1]						Soluble cations meqL[-1]									
							So4 ?		Cl[-]		HC O3[-]		Mg[++]		Na[+]		Ca[++]		K[+]			
			Dry	Wet	Dry	Wet	Dry	Wet	Dry	Wet	Dry	Wet	Dry	Wet	Dry	Wet	Dry	Wet	Dry	Wet	Dry	Wet
Upstream	*Zygophyllum coccineum*	0-20	774	8.5	138	1.05	3.81	5	83	4.1	1.4	0.8	2.45	0.84	81	2.8	2.94	2.04	0.23	0.2	15.5	7.9
	Zygophyllum simplex	20-40	775	8.5	112	0.87	3.24	1.4	42	3.3	1.44	0.9	1.54	0.53	433	11	312	2.42	0.13	0.1	81	5.7
Midstream	*Zygophyllum coccineum* *Zygophyllum decumbens*	0-20	752	8.4	234	1.8	5.59	9.1	14	19.5	3.74	2.1	4.14	1.98	12.9	5.4	424	5.4	0.35	0.2	19.3	17.3
	Zygophyllum simplex	20-40	798	8.4	0.25	0.2	0.73	2.9	0.9	1.4	0.78	0.5	0.54	0.12	0.95	0.4	0.78	0.29	0.17	0.1	14.9	13.1
Downstream	*Zygophyllum coccineum* *Zygophyllum decumbens*	0-20	794	8.3	0.25	0.12	0.89	8.2	1	0.43	0.78	0.42	0.4	0.24	0.94	0.4	0.81	0.41	0.15	0.1	41.7	40.4
		20-40	80.9	8.5	0.88	0.14	0.34	2.7	0.7	0.22	0.4	0.34	0.21	0.73	0.48	0.1	0.3	0.15	0.11	0.1	51.2	48.7

Table (3). Seasonal fluctuations inDegree of succulence of the studied.

Plant species	Location	Degree of succulence		Water content	
		Dry	**Wet**	**Dry**	**Wet**
Zygophyllum coccineum	Upstream	3.36±0.02	3.64±0.12 (s)	70.22±0.20	73±0.22 (s)
	Midstream	4.08±0.01	4.46±0.02 (s)	75.48±0.08	77.58±0.08 (s)
	downstream	3.57±0.05	3.79±0.03	72.03±0.38	73.6±0.18 (s)
Zygophyllum simplex	Upstream	2.92±0.07	2.98±0.04	65.82±0.78	66.7±0.18
	Midstream	3. ±0.01	3.24±0.24	68±1	69.67±0.58 (s)
Zygophyllum decumbens	Midstream	4.04±0.04	4.68±0.43 (s)	75.27±0.25	79.73±0.13 (s)
	downstream	2.05±0.01	3.25±0.13 (s)	51.13±0.32	67.58±1.9 (s)

Table (4). Seasonal fluctuations in total pigments (mg / g fresh f.wt.) of the studied species.

Plant species	Location	Chl a mg / g fresh weight of leaves		Chl b mg / g fresh weight of leaves		Carotenoids mg / g fresh weight of leaves		Chl a / chl b		Total pigments mg / g fresh weight of leaves	
		Dry	Wet	Dry	Wet	Dry	Wet	Dry	Wet	Dry	Wet
Zygophyllum coccineum	Upstream	2.42±0.22	5.65±0.25 (s)	0.44±0	2.13±0.34 (s)	1.54±0.22	2.57±0.25 (s)	5.5±0.5 (s)	2.69±0.37	4.4±0.38	10.34±0.38 (s)
	Midstream	6.41±0.27	7.85±0.13 (s)	1.69±0.13	2.35±0.13 (s)	2.49±0.34	3.08±0 (s)	4±0.38	3.35 ±0.12	10.93±1.25	13.27±0.25 (s)
	Downstream	5.87±0.13	6.42±0.94	1.61±0.13	1.71±0.35	2.35±0.13	2.39±0.36	3.65±0.25	3.8±0.51	9.83±0.13	10.51±1.54 (s)
Zygophyllum simplex	Upstream	3.37±0.13	6.16±0.44 (s)	1.61±0.13	2.05±0.55	1.94±0.06	2.93±0.13 (s)	2.09±0.08	3.11 ±0.61 (s)	6.89±0.13	11.15±1.11 (s)
	Midstream	6.78±0.17	7.85±0.13 (s)	1.93±.08	2.35±0.13	2.42±0	3.52±0 (s)	3.8±0.56	3.35±0.22	11.15±0.46	13.71±0.13 (s)
Zygophyllum decumbens	Midstream	6.09±0.25	6.85±0.57 (s)	3.4±0.41	2.05±0.51	3.3±0	3.08±0.44	1.88±0.34	3.85±0.35 (s)	12.69±0.71	12.98±2.59
	Downstream	1.54±0	2.05±0.13	0.22±0	0.51±0.13	3.01±0.13	3.01±0.13	7±0 (s)	4.17±1.04	4.77±0.13	5.57±0.13

Table (5). Seasonal fluctuations in Proline (µg / g f.wt.) of the studied species.

Plant species	Location	Proline µg / g fresh weight	
		Dry	**Wet**
Zygophyllum coccineum	Upstream	115.97±1.68 (s)	82.91±0.97
	Midstream	53.88 ±1.27 (s)	50.2±0.19
	Downstream	113.74±0.94 (s)	78.06±1.91
Zygophyllum simplex	Upstream	88.06±2.08 (s)	81.28±2.15
	Midstream	46.1 ±0.6 (s)	43.62±0.66
Zygophyllum decumbens	Midstream	77.57±0.95 (s)	62.49±1.21
	Downstream	111.78±0.88 (s)	77.09±1.30

Table(6). Seasonal fluctuations inantioxidant enzymes,SOD (unit /mg protein), CAT (µmol H2O2 destroyed / g f.wt./ min) , Peroxidase (g f.wt./ min), ASO (mM of ascorbate oxidized / g f. wt. / min.) and ppo (g f. wt. / min.) of the studied species.

Plant species	Location	SOD unit /mg protein		CAT µmol H2O2 destroyed / g fresh weight / min		peroxidase gram fresh weight /min.		ASO mM of ascorbate oxidized / gfresh wt / min.		ppo gram fresh weight / min.	
		Dry	Wet	Dry	Wet	Dry	Wet	Dry	Wet	Dry	Wet
Zygophyllum coccineum	Upstream	19.2±0.66 (s)	8.13±0.31	45.27±0.85 (s)	36.27±0.64	81.17±0.21 (s)	67.77 ±0.57	7.65±0.17 (s)	4.23±0.23	53.98±0.37 (s)	51.05±0.19
	Midstream	12.9 ±0.3 (s)	5.2 ±0.36	9.3±0.36 (s)	3.7±0.62	16.33 ±0.4 (s)	0.13 ±0.003	0.4±0.51	0.17 ±.01	49.89±0.22	49.52 ±0.1
	Downstream	32.73±0.35 (s)	3 ±0.3	35.7±0.61 (s)	28.23±1.37	34.13±0.5 (s)	29.94 ±0.15	3.08±0.15 (s)	2.13±0.018	52.89±0.24 (s)	50.39 ±0.20
Zygophyllum simplex	Upstream	24.53±13.45 (s)	18.37±0.55	24±1 (s)	21.8 ±0.26	83.84±0.24 (s)	58.5±0.11	8.68±0.29 (s)	7.73 ±0.21	53.71±0.26 (s)	52.86 ±0.2
	Midstream	5.1±0.2	4.5±0.44	22.13 ±1.06 (s)	13.93±0.31	65.68 ±0.69 (s)	1.28±0.014	4.52 ±0.48 (s)	3.62±0.16	52.15±0.076 (s)	51.61 ±0.26
Zygophyllum decumbens	Midstream	14.87±0.38 (s)	6.67±0.4	50.83±0.21 (s)	26.3±1.13	11.51±0.31 (s)	4.88±0.19	6.04±0.13 (s)	0.6±0.011	51.67±0.24	51.6±0.1
	Downstream	35.87±0.42 (s)	27.9±0.5	149.17±1.04 (s)	77.07±1.1	84.3±0.34 (s)	73.97±0.55	12.1±0.28 (s)	0.9±0.007	53.02±0.12 (s)	52.06±0.29

Table (7). Seasonal fluctuations intotal phenols mg / g f.wt,ascorbic acid mg/100g fresh wt,hydrogen peroxide mM/g f.Wt and malondialdehyde n mol/ g f.wtof the studied species.

Plant species	Location	Totalphenols mg/ g freshweight		Ascorbic acid mg/100g freshwt		Hydrogen peroxide mM/g fresh Wt		Malondialdehyde nmol/ g freshweight.	
		Dry	Wet	Dry	Wet	Dry	Wet	Dry	Wet
Zygophyllumcoccineum	Upstream	0.48±0.13 (s)	1.07±0.04	213.82±1.63	315.53±1.7 (s)	165.04±1.39 (s)	161.86±0.43	0.95±0.002 (s)	0.72±0.02
	Midstream	1.13±0.03	1.71±0.02	458.03±8.99	499.54±2.13 (s)	±1.64 138.06 (s)	±1.45 121.72	0.41±.002 (s)	0.17±0.002
	Downstream	0.92±0.01	1.22±0.04	277.40±6.7	±1.46 464.29 (s)	±1.58 160.39 (s)	149.35±4.17	0.59±.001 (s)	0.38±0.001
Zygophyllum simplex	Upstream	0.93±0.05	1.40±0.04	152.45±5.38	201.05±0.27 (s)	194.11±1.35 (s)	111.37±0.57	0.45±0.004 (s)	0.21±.003
	Midstream	1.48±0.01	1.96±0.02	265.52±6.71	±7.44 337.61 (s)	154.55±2.03 (s)	82.56±1.21	0.29±0.006 (s)	0.103±0.002
Zygophyllumdecumbens	Midstream	0.89±0.04	2.28±0.09	273.38±4.34	301.72±1.4 (s)	214.96±0.79 (s)	169.61±1.63	0.63±0.005 (s)	0.13±0.002
	Downstream	0.76±0.02	1.34±0.06	211.17±1.84	233.67±0.63 (s)	219.01±0.86 (s)	197.98±1.61	0.75±0.003 (s)	0.5±0.002

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