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ECOPHYSIOLOGICAL RESPONSES AND GENETIC VARIANTS IN SOME SPECIES OF THE GENUS *zygophyllum*

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ABSTRACT

Five *Zygophyllum spp.* representing five different localities (habitats) in South Sinai were comparatively studied from ecophysiological and genetical points of view . The obtained results demonstrated that under ecological conditions prevail the southern Sinai (drought) the physiological responses and genetic varients of the *Zygophyllum spp.* under study are very coincided with such conditions. However, the studied species display slightly variations in their physiological and genetic characteristics due to the role of ecotypification of these species in their various habitats.
Keywords : *Zygophyllum spp.*,Ecology, Physiology, Genetics, South Sinai .

INTRODUCTION

The genus *Zygophyllum* is widely distributed in the Egyptian deserts.It included a number of species distinguishable by their different morphological, cytological characteristics as well as ecological adaptations.

The *Zygophyllum* species constituents zygophylin (28 % in leaves .018 % in stems and 0.26 % in fruits). Quinivinic acid (0.36 % in leaves, 0.47 % in stems and 0.31 % in fruits). Flavonoides e.g.; Kaempferol – 3 – rutinoside. All *Zygophyllum* species are leaf and stem succulent and unpalatable therefore, they are not suitable for neither grazing nor cutting for fuel. However, from economical potentialities, it has good reputation in folk medicine as a remedy for rhaumatism, gout, cough, asthma, hypertension, flatulent colic and as diuretic. The juice expressed from the fresh leaves and stems are used as abrasive cleanser and as a remedy for the treatment of certain skin diseases (Batanouny *and Ezzat* 1971;El-gamal, *et .al.*,1995; Rizk and El-Ghazaly, 1995; and Batanouny 1999).

At present, the description and conservation of *Zygophyllum spp.* depend, to a large extent, on their morphological, ecophysiological traits and photochemical composition. However, it is necessary to get an appropriate discrimination criteria which could provide an adeqate information about their genetic divergence. The matter which help the plant programmers to establish gene bank as well as avoiding of mislabeling and duplication of *Zygophyllum* collections . For this purposes, many workers used the electrophoretic techniques as an important tool for genotype identification for many cultivars , among them Abd El-Salam *et.al.*, (1998) , Abou El-Enain *et.al.*, (1999) and El-Fikey *et.al.*, (2002).

Therefore,this work was performed as an attempt to elucidate the discrimination of *Zygophyllum spp.*, collected from South Sinai, from ecological, anatomical, physiological and genetic points of view.

MATERIALS AND METHODS

Sampling:

Five ***Zygophyllum spp.*** were collected at (January - 2003) from different sites located in South Sinai, Table (1), and Figure (1).

Table (1): Growth form, habitat type and site of collection of the studied five *Zygophyllum species* .

Zygophyllum spp.	Groth Form / habitat type	Location
1- *Z. Simplex*	Mat-shaped ann.and bi - plant of wide ecological amplitude	EL Naqab
2- *Z. album*	Shrub of wide ecological amplitude	Ras- Sudr
3- *Z. coccineum*	Shrub of Xerophytic habitats	Nuwieba
4- *Z. decumbens*	Shrub of Xerophytic habitats	Taba
5- *Z. dumosum*	Shrub of Xerophytic habitats	Sant-kathrine

Also, the five associated soil profiles were dug wide open to the depths where the greatest amount of plant roots occurred. Accordingly nine soil samples were collected and then air - dried, crushed, sieved through 2 mm sieve and subjected to analysis.

Soil analysis:

- Particle size distribution was carried out by the dry sieving method of Kilmer and Alexander (1949).
- Soil reaction was determined in the soil paste using a Beckman bench type pH- meter, Richards (1954).
- Total salinity (EC_e) in the soil saturation extract was determined conductimetrically, Richards (1954).
- Cationic and anionic compositions of the soil extract were accomplished following the methods described by Richards (1954) and Jackson (1962).

Preparation of protein amino acids:

Defatted plant powder (0.1gm) was extracted first with 80% ethyl alcohol, dried and dissolved in 10ml of 6N HCL in a sealing tube, Awapora (1948). The mixture was hydrolyzed at 110 °C for 24 hours, filtered and the hydrolyzed protein amino acids were obtained by evaporation of the hydrolyzate to dryness. The residue was washed with distilled water. The volume of the filtrate was adjusted to 100ml using distilled water. Preparation of amino acids for injection in GLC was performed by the method described by Lamin and Gehrke (1966).

Crude protein content:

Total protein was calculated by multiplying the total nitrogen by 6.25. The total nitrogen was determined by using the modified micro-Keldahl method of Peach and Tracey (1965).

Photosynthetic pigment:

Photosynthetic pigment was calculated quantitatively by using equation chlorophyll (a) = $10.3\ E_{663} - 0.918\ E_{644}$ = µg/g, chlorophyll (b) = $19.7\ E_{644} - 3.870\ E_{663}$ = µg/g and carotenoids = $4.2\ E_{452.5}$ − {0.0264 chlorophyll(a) + 0.4260 chlorophyll (b)} = µg/g as mentioned by Metzner *et al.*, (1965).

Total carbohydrate:

Total carbohydrate content was estimated as the method described by **Dubois *et al.*, (1951).**

Anatomical studies :

Fresh materials of five-studied *Zygophyllum spp.* (internode NO.4 from the stem) were taken and fixed in fromaline : acidic acid : ethyle alchohol (F.A..A.) 70% .Then samples were washed in 50% ethanol and dehydrated in ethanol-colroform according to the method outlined Esau (1960) . After complete dehydration the specimens were embedded in pure Paraffin wax (m.p.56 °C) .Sections transverse 20-30 µm in thickness were cut on a rotary microtome. Safrain and fast green method of staining was used, after staining the sections were mounted in canada balsam, dried and examined using Image processing and analysis system, Fukui (1988a). To calculate the cortex aria of stems the follow equation of calculation of cortex of stems 100-100ab/AB Beakbene (1941).

A*B The two dimention of the stem.

a*b The two dimention of the vascular bundance.

Biochemical analysis:

Protein electrophoresis

SDS- polyacrylamide gel electrophoresis (SDS-PAGE) was performed for total proteins of the five *Z.spp.* according to the method of Laemmli (1970), as modified by Studier (1973).

Isozymes electrophoresis.

Native – polyacrylamide gel electrophoresis (Native-PAGE) was used to identify isozymes variation among the five *Z.spp.* studied .Three isozymes (α Amylase, Protease and Malate dehydrogenase), were extracted from the plants. These isozymes were separated in 9% polyacrylamide gel electrophoresis according to Stegemann *et al.*, (1985).

After electrophoresis, the gels were stained according to their enzyme system with the appropriate substrate and chemical solution. Then, incubated at 37° C in a dark room for complete staining. The staining gels were carried out according to (Vallejos, 1983, Jonathan and Wendel, 1990 and Heldt, 1997), for – Amylase, Protease and Malate dehydrogenase, respectively.

Gel analysis:

All gels resulted from protein and isozyme electrophoresis were scanned using Gel Doc-2001 Bio-Rad system. The densitometric scanning of the bands were performed on three directions. Each band is recognized by its length, width and intensity. Accordingly, relative amount of each band could be quantified and scored.

RESULTS AND DISCUSSION

Ecological setting:

The studied ***Zygophyllum* species** were recorded from different habitat types located in the southern Sinai ; EL- Naqab , Ras-Sudr, Nuwieba , Taba, and Sant- kathrine were selected in the South Sinai region. This region is represented by a minor basin of the Gulf of Aqaba and the Gulf of Suez drainage systems, wadi deposits and distinguished summits. Stratigraphically, the basin and wadi deposits are occupied by the sediments belonging to recent deposits of gravelly coarse sand over basic rocks ,Dams& Moore (1981). Climatologically, this region is characterized by extreme aridity where low and erratic winter rainfall of 16 mm (Nuwieba) and 52 mm (EL - Naqab) per year prevails. However Sant- Kathrine due to its height receives ample rainfall, which has produced wadis and water courses. The maximum temperature 37 °C was recorded during the summer months, while temperature in winter months attains its minimum value of 11° C.The recorded maximum relative humidity varies from 45% to 56% and the minimum mean daily evaporation ranges between 4.6 mm and 11mm, Shabana (1998). It is taken into account that most of the wadis in this region have a relatively rich natural plant cover including xerophytic and halophytic species.

Concerning the soil nature of the studied sites, data presented in Tables (2&3) indicate that soil depth varies according to the selected sites , where it reaches 60 cm depth in Sudr and EL Naqab sites changing to very shallow (20 cm depth) in the other studied sites. Soils are generally gravelly coarse - textured soils with no variation with depths. Soil reaction ranged between slightly alkaline and alkaline, where pH values varied from 7.5 to 8.6 . EC fluctuates in narrow range, where it ranges from 0.26 to 2.70 ds m^{-1} . However , these values differ from one site to the other and even in the entire depths especially those of moderate depth . In general, the obtained values were under the critical levels of salinity .On the other hand, the soils of the collected studied plant were salt -free. The cationic ions were dominated by Ca^{++} ion followed by Mg^{++} and Na^{+} ions, whereas the K ion was the least. The anionic ions were dominated by SO_4^{-} and /or HCO_3^{-} ions followed by Cl ion. Therefore, the excepted salt composition in these soils is $CaSO_4$ or Ca $(HCO_3)_2$. The discrimination of the salt composition and its variation in the soils is completely dependent on the parent rock from which these soils are formed.

Commenting on the former presentation, it is taken into consideration that the ecological setting of the studied sites is, to some extent, comparable . However, minor variations were observed between them due to the differences in their elevations.

Table (2): Granulometric analysis of the soil profiles associated with the studied *Zygophyllum Spp.*

Locations & *Zygophyllum spp.*	Depth/ m	2-1mm	1-0.5mm	0.5-0.25mm	0.25-0.125mm	0.125-0.063m	<0.063mm	*Texture class*
El-Naqab *Z.simplex*	0-20	18.6	10.7	18.2	0.14	48.0	4.34	Fine sand
	20-40	20.5	18.4	24.5	0.69	16.5	18.9	Coarse sand
	40-60	16.2	18.6	32.3	0.66	16.5	15.7	Coarse sand
Ras-Sudr *Z.album*	0-20	20.9	19.6	16.1	21.7	15.2	6.5	Coarse sand
	20-40	26.5	23 7	13 8	10.6	10.1	15.3	Coarse sand
	40-60	23.5	23.2	15 5	11.9	10.4	15.5	Coarse sand
Nuwieba *Z.coccineum*	0-20	41.4	28.9	12.4	5.94	1.98	9.25	Coarse sand
Taba *Z.decumbens*	0-20	51.6	30.9	13.8	2.93	0.36	0.28	Coarse sand
Sant-Kathrine *Z.domosum*	0-20	21.3	29.7	18.5	9.76	4.72	5.99	Coarse sand

Table (3): Some chemical analysis of the soil profiles associated with the studied *Zygophyllum spp*

Species	Location	Depth/ Cm	pH	EC_e ds/m^{-1}	Na^+	K^+	Ca^{++}	Mg^{++}	CO_3^-	HCO_3^-	Cl^-	SO_4^-
					mg /100g							
Z simplex	El-Naqab	0-20	7.9	0.72	1.17	0.24	5.0	3.0	2.4	Tr	2.75	4.44
		20-40	7.7	1.19	2.19	0.49	8.0	4.5	3.0	Tr	2.50	2.79
		40-60	8.12	1.09	2.13	0.24	8.5	2.5	2.4	Tr	3.00	7.79
Z album	Ras-Sudr	0-20	8.3	0.60	2.90	0.33	3.5	1.5	2.4	Tr	4.00	1.2
		20-40	8.6	1.36	10.1	0.12	15.0	1.5	1.8	Tr	1.00	14.8
		40-60	7.9	2.70	2.13	0.17	23.0	6.0	1.8	Tr	1.75	27.6
Z coccineum	Nuwieba	0-20	7.8	0.60	0.87	0.42	5.0	2.5	3.6	Tr	2.5	2.89
Z.decumbens	Taba	0-20	7.5	0.70	1.02	0.31	4.5	3.0	3.0	Tr	2.5	3.33
Z.dumosum	Sant-Kathrine	0-20	8.1	0.26	0.17	0.14	3.5	0.5	1.8	Tr	1.25	1.26

Morphology of the studies species

(1) *Zygophyllum simplex* .L (Forssk).

Glabrous, mat-shaped sappy herb with simple cylindrical leaves, 9-10 mm, long and yellow flowers. Capsule minute, only 2 mm, across, deeply 5-parted annual, biennial, completing its life cycle in two seasons, very common " in the deserts of Egypt (D.), the Red Sea coastal region (R.), Gebel Elba and surrounding mountains (G.E.), and Sinai proper, i.e. South of El-Tih desert (S) Tackholm (1974). Samples of *Z.simplex* were collected from El-Naqab rocky grounds.

(2) *Zygophyllum Album* (L.E.)

Shrubby. branches and leaves blue-green, mealy pubescent. leaflets 2, obovoid, about 7mm long. Flowers small (Frut.), very common in the Oases of the libyan desert (O.), the Mediterranean Coastal Strip from El-Sallum to Rafah (M.), All the deserts of Egypt (D.), the Red Sea-Coastal region (R.), Sinai proper (S.), In both deserts and salt marshes, often sand-accumulating, growing cushion. shape, frequently reaching over 1m.in height, Tackholm (1974). Samples of *Z. album* were collected from Ras-Sudr, coastal dry salt marsh.

(3) *Zygophyllum coccineum* (L)

Shrub, up to 75 cm. Leaflets2, bright green, glabrous, cylindrical, at least 10 mm. Long, capsule 8-10 mm. Long, apex. Obtuse, Tackholm (1974). Samples of *Z. coccineum* were collected from Wadi-Watir Nuwieba.

(4) *Zygophyllum decumbens* Del.

Shrubet 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. Rare (r.), the Arabian desert east of Nile(Da.),i.R.,5.Tackholm (1974). Samples of *Z. dcumbens* were collected from the roky slopes of Wadi Taba.

(5) *Zygophyllum dumosum* Boiss

More woody than any other species, up to 75 cm height . Leaflets 2, mealy-pubescent, cylindrical, about 10 mm. Long.Petals narrow, longer than in any other species. Capsule 5.parted, the 5 carpels strongly compressed with broadly winged margin ,Tackholm (1974). Samples of *Z. dumosum* were collected from Sant-Kathrine,

Anatomical studies :

Table (4) and Figure (2) summarized the differences in stem anatomy among the five *Zygophyllum species* .The results clearly obvious that four main anatomical variants were recorded in the stem of *Z.spp.* of different habitats.The first outline in across section was tert in all *Zygophyllum species* under study. The second cutical width was (1 µ) in three *Z.spp. (Z. simplex, Z. decumbens and Z. dumosum).* while the other two *Zygophyllum spp. (Z. album and Z. coccineum*) were (2 µ) width. From the table, it is clear that epiderms cosists of one layer of radial cells and also show that the highest percentage area was 12.4 % and 11.8 % in (Fig.1 .B) *Z. dumosum and Z. coccineum*, respectivly. Whereas the lowest percentage area was 4.3 % in *Z. simplex*.The results indicated that different variants in the percentage area of the third layer (cortex) among *Z.spp.* under study, where the highest values were 83.8 % and 89.2 % in *(Z. album and Z. coccineum*), respectivly. While the lowest one was 81.1 % in *Z. simplex*. The results also obvious that the chlorenchyma was abundancet in all *Z.spp.*but the parenchyma was absent in two *Z.spp. (Z. simplex , Z. decumbens)* and present in other three *Z.spp.* On the other hand, there were small varians in percentage area of (vascular cylender) which ranged between 10.5 % and 16.2 %.This study of the stems showed that *(Z. decumbens)* is rather hard than the other *Z.spp.* ,where the *Z. coccineum*, is soft and more jucy (Table 4) .

From the former results, it could differentiate clearly between the different ecotypes of *Z.spp.* These ecotypes can be the product of genetic responses of the *Z.spp.*of their habitats. Also the rsults have demonstrated that ecological differences play an important role in the ecotypification of these genus *in various habitats of* Egypt.

It is worth mentioning that the anatomical features of stem of the recognized ecotypes exhibited considerable differences, which appear also to be distinctive. These differences seem to be the product of the adaptive mechanisms to environment (Batanouny *et al.,* 1991 and Khafagi *et al.*, 1996.)

Protein amino acids:

The results in Table (5) show the protein amino acids composition of the studied five *Zygophyllum* species .From obtained results ,it is obvious that Aspartic and Glutamic acids (acidic - amino acid) tend to be present in pronounced amounts than the other protein amino acids, since they represent as a storage form of nitrogen Beevers (1976) and Miflin (1981). However, *Z. album* and *Z. decumbens* attain the highest amount as compared to the other studied species. In case of basic amino acids, Lysine recorded the highest amount than the other basic amino acids and also accompanied with the two above mentioned *Zygophyllum* species. Essential adaptive responses in the adjustment of the succulent xerophyte *Z. coccineum,* it is only species that appears the basic amino acid Araginine in addition to the aromatic amino acid such as Proline, phenylalanine and Tyrosine in large amounts as well as sulfur amino acid Cystine and methionine were also prominent. These may be regarded as a sign of the presence of resistant proteins (21.6 201,141 and 4.3 mg/100g) respectively. In this regard,some authors references have already reported that under severe drought, plant organs

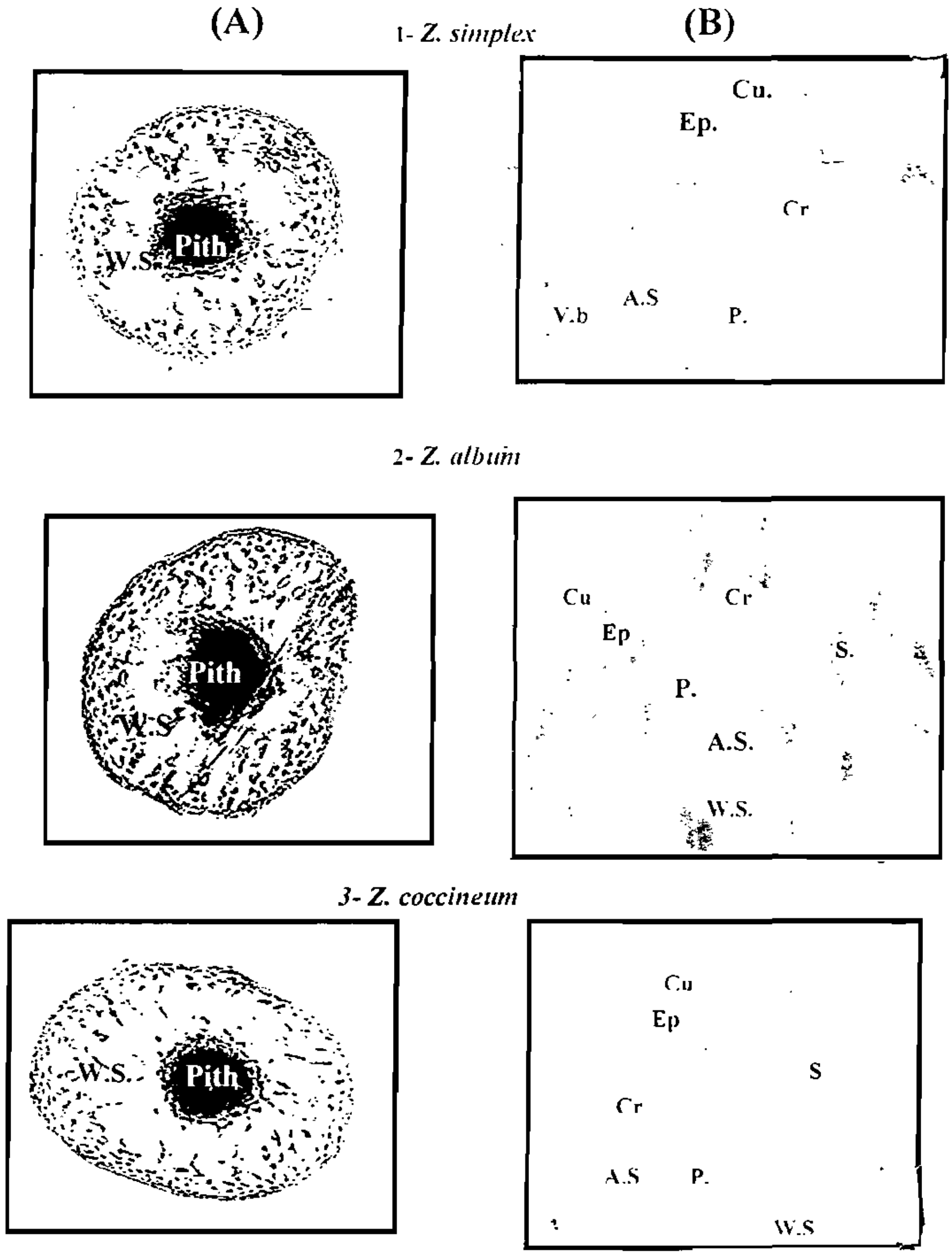
Table (4): Anatomical features of stem of five *Zygophyllum spp.*, under different habitat conditions

Zygophyllum Species	Location	Cuticle Thickness In µ	Epidermis-Depth µ	Cortex-Area %	Vacularbundle %
Z. Simplex	El-Naqab	1	13	87.0	13.0
Z. album	Ras-Sudr	2	15	87.1	12.9
Z. coccineum	Nuwieba	2	14	89.2	10.8
Z. decumbens	Taba	1	15	83.8	16.2
Z. dumosum	Sant-Kathrine	1	14	86.3	13.7

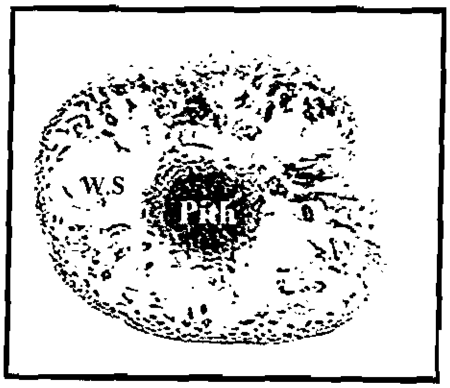
Table (5): Protein amino acid composition of different studied five *Zygophyllum spp.*

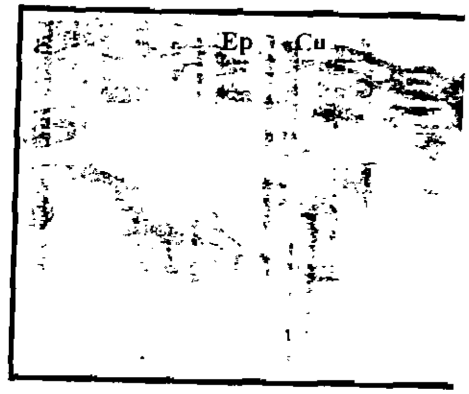
Zygophyllum spp / Locations	Acidic Amino acid gl100gµ		Basic Amino acid g/100gµ			Cycli Amino acid µg/100g			Aliphatic Amino acids µg/100g								S-Containing µg/100g	
	Aspartic (µg/ml)	Glutamic (µg/ml)	Lysine	Histadine	Arginin	Proline	Phenyl alanine	Tyrosine	Glycine	Alanine	Valine	Leucine	Iso - leucine	Thre-onine	Serine	α Amino butaric	Cystine	Methionine
Z. simplex El-Naqab	90.1	84.3	20.7	61.2	—	—	41.3	54.6	59.6	73.6	91.2	18.2	9.4	14 2	23.3	60 7	—	—
Z album Ras-Sudr	319 4	88 4	145.8	38.0	—	7.1	33.8	28.4	61.5	41.8	50.1	56.9	34.6	38.6	83 5	251 1	—	—
Z coccineum Nuwieba	111 4	79.3	61.7	82.3	8 9	21.6	201.2	141.2	70.3	51.6	84 6	90.1	40.3	92.6	80 3	201 3	4.3	3.1
Z decumbens Taba	229.8	280.6	163.6	142.4	—	—	119.6	56.7	118.5	115.7	136.11	178.8	101.8	106.3	117.1	277.4	—	2.1
Z dumosum Sant Kathrine	308 1	79.1	43.1	112.6	—	—	34.1	23.3	39.6	31.8	42.9	49.9	30.7	28.3	47 8	122 7	—	—

Figure (1)

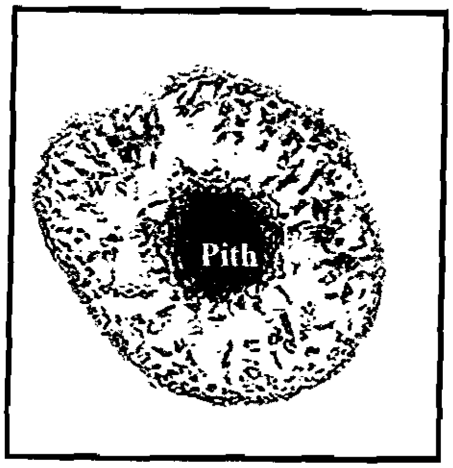

(A)
1- Z. simplex
(B)
W.S.
Pith
Cu.
Ep.
Cr
V.b
A.S
P.
2- Z. album
Pith
W.S
Cu
Cr
Ep
S.
P.
A.S.
W.S.
3- Z. coccineum
W.S.
Pith
Cu
Ep
S
Cr
A.S
P.
W.S

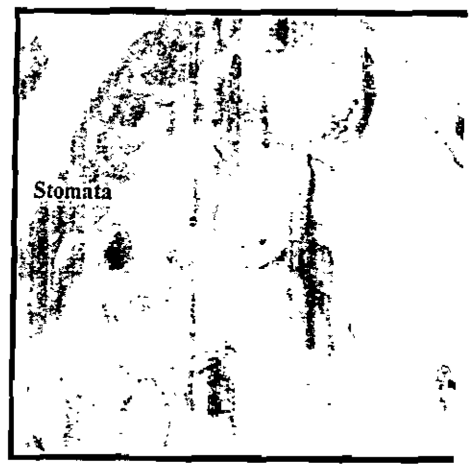
(A) *4- Z. decumbens* (B)





5- Z. dumosum





Figures (1& 2)

A : T.S. sections in the Zygophyllum stems (X50).
B : The epidermis and cortex of the stems (X250).
Cu:cutical , Ep. :epedermis , Cr :cortex A.S :air space ,
S.M. :Storage material ,P. : Parenchyma and Collenchyma
W.S: water storage cells, Stomata, pith & V.b: vascular bundle .

accumulate considerable quantities of certain amino acids e.g. phenylalanine, valine and prolines. Proline accumulation is regarded by some investigators to play a role as nitrogen reserve and by others to protect protoplasm from dryness. Kramer (1983) pointed out that proline accumulates by water stress because water stress stimulates its synthesis from glutamate by loss of feed back decrease of rate of proline oxidation and decreases its incorporation into protein.

From results of the present work, , it could be also concluded that the amino acids; arginine, proline, cystine and methionine were detected and disappeared in plant species according to differences in habitats conditions. This could be attributed to stimulation of synthesis of new type of protein or proteins that were not being made under some habitats, through activation of the ribosome messenger system or by the formation of functional polysomes, Morcus *et al.*, (1966). Possible differences in the formation of the adaptive protein type in different plant species exist under different habitats. The synthesis of protein types rich in certain amino acid or contain specific amino acids could be the key to survival of certain plant species under specific climatic or edaphic conditions.

Pigment content:

Contents of chlorophyll a, b and carotenoids in the fresh leaves of the studied *Zygophyllum species* are tabulated in Table (6) . From the table, it is obvious that *Z.spp.*have an amount of chlorophyll (a,b) ranged in narrow scale , where *Z. album* has the largest amount of chlorophyll a, b at Ras-Sudr than the other species, where it attains (3.68 and 4.19 µg/g), respectively, followed by both of *Z. simplex* (2.64 and 1.22 µg/g). *Z. coccineum,* (2.58 and 1.5 µg/g) and *Z. dumosum* (2.35 and 1.12 µg/g) Meanwhile, *Z. decumbens* recorded the lowest values of pigment (1.90 and 0.586 µg/g). On the other hand, the cartenoids pigment recorded the highest value in *Z. simplex* (0.852 µg/g), whereas the lowest value was recorded in *Z. dummsum* at Sant–Katherine (0.12 µg/g). The differences in chlorophyll a,b and carotenoids amounts in the five studied *Z.spp.* may be attributed to differences in habitat conditions from which their samples were colleted.

Table (6): Some chemical constituents of the studied five *Zygophyllum spp* .

Zygophyllum species	Location	Chlorophyll a (µg/g)	Chlorophyll b (µg/g)	Carotenoids (µg/g)	Carbohydrat (µg/100g d wt)	Protein (µg/100g.d.wt)
Z. simplex	El-Naqab	2.64	1.22	0.852	59.0	6 45
Z. album	Ras-Sudr	3.68	4.19	0.278	47.1	7 81
Z coccineum	Nuwieba	2.58	1.51	0.529	45 0	9 71
Z. decumbens	Taba	1.90	0 986	0 560	48.5	10 2
Z dumosum	Sant-Kathrine	2.35	1 12	0 12	77.5	12.6

In this respect, Al–Tantawy (1983), and Abd El-Maksoud (1987) reported on some desert plants grown under different habitats concluded that xerophytes attained higher concentrations of chlorophyll and carotenoids due to their adaptive mechanisms under dry conditions. This view was held also by Nour El-Din *et al.*, (1999), who reported that response attainment of high levels of total photosynthetic pigment is an adaptive to moisture stressed habitats through which better water use efficiency may be accelerated..

Total carbohydrate contents:

Carbohydrates content varied according to response to different habitat conditions in south Sinai, Table (1) .The results illustrated in Table (6) noticed that accumulation of total carbohydrates attained the highest level in *Z. dumosum* (77.5 μg/100g) than the others species under stucompared to their stufied species.Ebad *et al.*, (1991) concluded that carbohydrates tended to increase in halophytic plants during the rainy season. Such increase is related to more favorable conditions for carrying out metabolic processes.

Crude protein contents:

The results in Table (6) reveal that accumulation of total protein attained high levels in all *Zygophyllum spp.* In *Z. dumosum,* it reached to the maximum value 12.6μg/100g dry Wt., while the minimum value was recorded by *Z. simplex* 6.45 μg/100g dry Wt.. In this connection, Ahmed and Girgis (1979) showed the importance of nitrogen intermediates as osmotically active ingredients in plant metabolism. They also concluded that xerophytes depend, to a large extent, on the accumulation of organic intermediates in building up their osmotic pressure. Nour El-Din (1995) reported that the adaptive responses of desert plants under different habitat conditionsand concluded that in dry desert habitats true xerophytes attained higher concentrations of crude protein under different habitat conditions.

Protein electrophoresis

Total protein banding patterns based on SDS-PAGE for the five *Zygophyllum spp.* under study are illustrated in Table (7) and Figure(3) .

Table (7) : Protein analysis of five *Zygophyllum spp* under different habitats.

Band No.	low Mol. Marker	RF.	1	2	3	4	5	Mol. M. Bands
1	97.40	0.16					+	76.8
2	66.20	0.18	+	+	+	+	+	66.9
3		0.22					+	63.5
4		0.27		+			+	583
5		0.33					+	52.25
6	45.00	0.39					+	47.3
7		0.43		+		+	+	43.31
8		0.45					+	41.2
9		0.48		+			+	38 2
10		0.53	+	+	+	+	+	34.7
11	31.00	0.56					+	32.5
12		0.64	+	+	+	+	+	28.27
13		0.68	+	+				26.50
14		0.72					+	24.91
15	21.50	0.78	+	+	+			22.48
16	14.40	0.89	+	+	+	+	+	16.38

{1,2,3,4 and 5} *Zygophyllum spp.* see (Table 1)

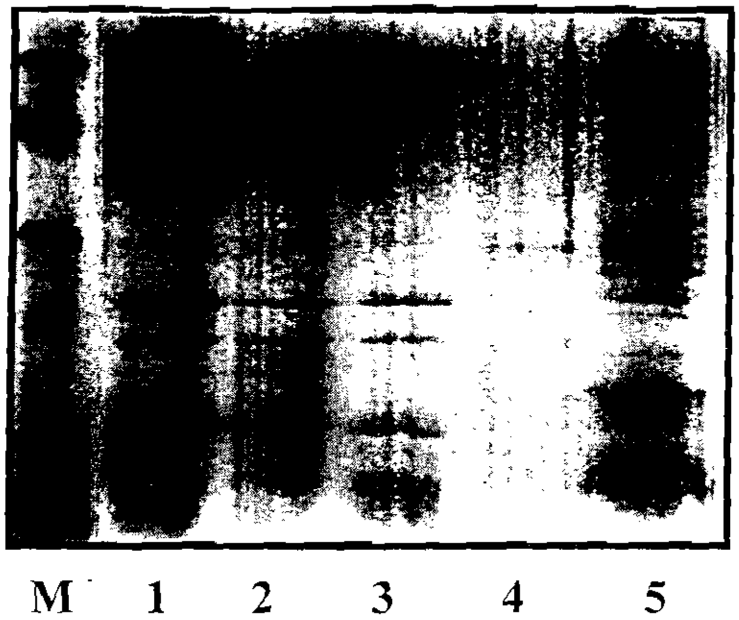


Figure (3) : Zymogram of protein banding pattern among five *Zygophyllum spp.*

The total number of bands was 16, which were not necessarily present in all the studied species .The bands were detected at approximately molecular weights ranged between 76.8 and 16.38 kDa. The resulted profiles comprise twelve polymorphic bands, which were present in some species and absent in the others. Whereas the other four monomorphic bands were with approximately molecular weights of 66.9, 34.7, 28.27 and 16.38 KDa. However, some specific bands, which could be used to distinguish some species from the other were detected. For instance, *Z. dumosum* has seven positive specific bands with MWs 76.80, 63.5, 52.25,47.3,41.2, 32.5 and 24.91 KDa., whereas also *Z. simplex and Z. album* have the same unique band with MW of 26.5 KDa. The results also revealed that the maximum number of bands was 14 bands and observed in *Z. dumosum* ,whereas the minimum one was 5 bands and recorded in both *Z. coccineum and Z. decumbens* . This finding is completely alined with those obtained by El Rabey *et al.,* (2002) who found a discrimination between 15 barley cultivars when used SDS-protein profiles for studing the genetic relationship between these cultivars.Furthermor, Table (8) shows the highest similarity matrix between *Z. album and Z. coccineum* of about 65.3%.On the other hand the lowest similarity matrixwas shown between *Z. simlpex and Z. dumosum* of about 30.3% .

Table (8) :Similarty matrixof protein banding pattern among five *zygophyllum spp.* under different habitats.

Z.spp.	1	2	3	4	5
1	100	51.1	61.3	38.3	30.3
2	51.1	100	65.3	62.9	48.1
3	61.3	65.3	100	64.1	41.6
4	38.3	62.9	64.1	100	43.3
5	30.3	48.1	41.6	43.3	100

Isozymes electrophoresis

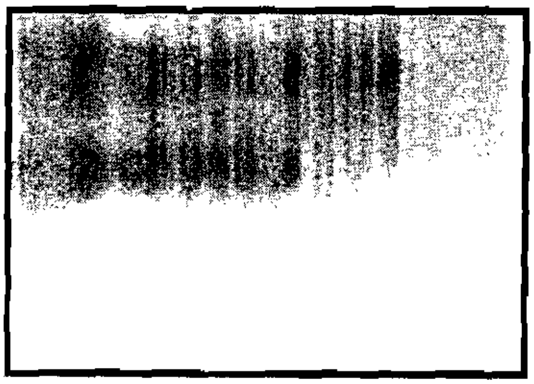
In the present study three isozyme systems : (α- Amylase, Protease and Malate dehydrogenase) were studied. The obtained results are as follows:

α- Amylase :

Banding patterns of α-Amylase isozyme for the studied *Zygophyllum spp.* are demonsterated in Table (9) and Figure (4), where the banding indicate that this iso-enzyme revealed low polymorphic level, where its pattern gives two bands expressed among the studied *Z.spp.* The first band was scored as a common band in all *Zygophyllum spp.* with differences in intensity between the studied species . while the second band was distinguishable for three *Z.spp. (Z. simlpex, Z. album and Z. coccineum)* and this band is absent in other *species (Z. decumbens and Z. dumosum)*

Table (9) : Electrophoretic patterns of α-amylase isozyme of five *Zygophyllum spp.*

Z.spp.	1	2	3	4	5
Amylase 1	+	+	+	+	+
Amylase 2	+	+	+	-	-



1 2 3 4 5

Figure (4):Zymogram of α –amylase banding pattern among five *Zygophyllum spp*

Protease :

Shown in Table (10) and Figure (5) the banding patterns of Protease for the five studied *Zygophyllum species* under study.According to the presence or absence of bands, this iso-enzyme was identifed by three bands. The last band was monomorphic band expressed in all *Z.spp.*, with differences in bands intensity between the five studied *Z.spp.* Meanwhile, other two bands proved to be polymorphic bands distinguishing the *Z. simplex and Z. coccineum* species.

Table (10) : Electrophoretic patterns of Protease isozyme of five *Zygophyllum spp*

Z.spp.	1	2	3	4	5
Protease-1	-	+	+	+	+
Protease-2	+	+	-	+	+
Protease-3	+	+	+	+	+

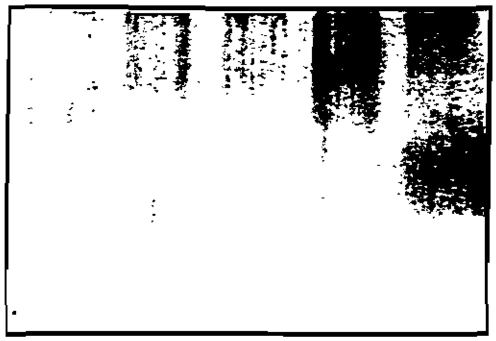


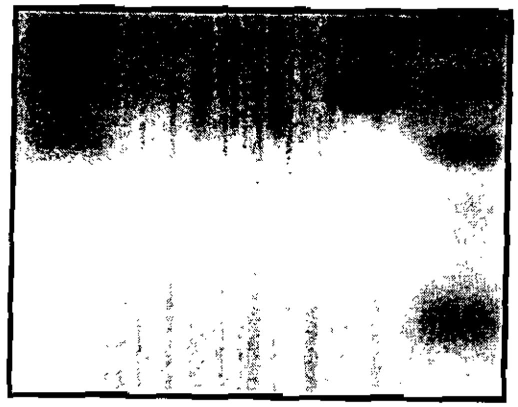
Figure (5) : Zymogram of protease banding pattern among five *Zygophyllum spp*

Malate dehydrogenase (Mdh):

Electrophoretic patterns of (Mdh) isozymes are represented in Table (11) and Figure (6). The results revealed that a high polymorphism in bands number.Meanwhile, differences in bands intensity were also noticed between and within all of the species. Total five bands were identified for the studied species, which were present in some species and absent in others, where the two bands of number one and two scored as common bands expressed in all species, while three remained bands were polymorphic bands. On the other hand, bands number four and five were present only in *Z. dumosum* and could be considered as positive markers of this species. From this result *Z. simplex and Z. coccinum* and also *Z. album and Z. decumbens* have the same trend expressed of this iso-enzyme,while *Z.dumosum* has the hightest expression of this iso-enzyme.

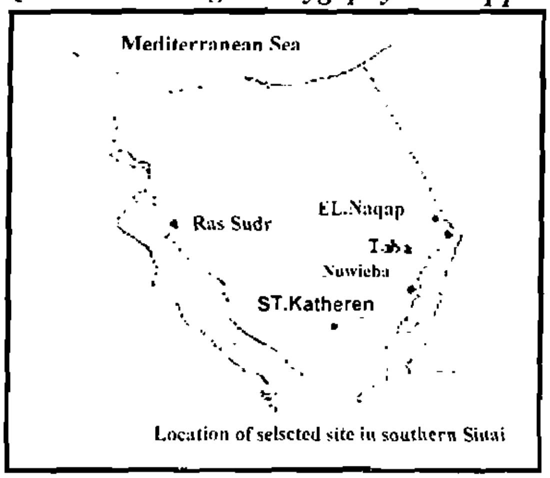
Table (11) : Electrophoretic patterns of malate dehydrogenase isozyme of five *Zygophyllum spp.*

Z.Spp	1	2	3	4	5
1–Mdh	+	+	+	+	+
2–Mdh	+	+	+	+	+
3–Mdh	+	-	+	-	+
4–Mdh	-	-	-	-	+
5–Mdh	-	-	-	-	+



1 2 3 4 5

Figure (6) : Zymogram of malate dehydrogenase banding patterns among five *Zygophyllum spp.*



Location of selscted site in southern Sinai

In general, the results of the above three isozymes indicate that these isozymes were identified in the five studied *Zygophyllum spp*. Moreover, the malate dehydrogenase isozyme generates sufficient polymorphic profile followed by protease isozyme. Meanwhile, α– amylase isozyme produced a minimum value of polymorphic profile. This indicate that these isozymes could be quietly used to discriminate the studied *Zygophyllum spp.* in proper manner.

Commenting on the overall results, it may be concluded that under ecological conditions prevailing the southern Sinai (drought), the physiological responses and genetic variants of the studied five *Zygophyllum spp.* are very coincid with these ecological conditions. However, the noticeable differences between these species with respect to their physiological and genetic characteristics might be rendered to the role of ecotypification of such species in their various habitats.

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الاستجابات الفسيولوجية- البيئية والتباين الوراثي في بعض أنواع جنس الرطريط (*Zygophyllum*)

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خمسة أنواع من جنس الرطريط (*Zygophyllum*) تم جمعها من خمسة بيئـــات مختلفة من جنوب شبة جزيرة سيناء وهم موضع الدراسة المقارنة الحالية والتي تنـــاولت النواحي الفسيولوجية والوراثية .

من النتائج التي تم الحصول عليها يتضح أنة تحت الظـــروف البيئيـــة الجفافيـــة السائدة في جنوب سيناء نجد أن الاستجابات الفسيولوجية والاختلافات الوراثية لنباتات هـذا الجنس تواكب تماما هذه الظروف , على الرغم مما أظهرته هذه الأنواع موضـع الدراســـة من تباين في الاستجابات الفسيولوجية والخصائص الوراثية والتي قد تعزى إلى اتجاه هذه الأنواع إلى تطوير نماذج بيئية (Ecotypes) متباينة لها تحت الظروف البيئية المختلفة .