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## A Morphological and Anatomical Study of an Annual Grass *Eremopyrum* (Poaceae) in Iran

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**Abstract:** *Eremopyrum* (Poaceae) is distributed in various habitats in Iran. This genus is an Irano touranian floristic element and is represented by five species in Iran. In the morphological study 55 quantitative and qualitative characters were evaluated in 26 populations. It was observed that length of lowermost and uppermost glumes and its awn, spike length and lodicules lengths are diagnostic characters. In anatomical studies, the cross sections of the leaf blade and dorsal epidermis were examined and the stomata index was calculated. Characters as frequency of short and silica cells, number of macro hairs, prickles, subsidiaries, shape and number of vascular bundles are diagnostic. Inter- and intra- specific variation in *Eremopyrum* is evaluated.

**Key words:** *Eremopyrum*, poaceae, morphology, anatomy, Iran

### INTRODUCTION

The genus *Eremopyrum* (Ledeb.) Jaub. and Spach belongs to tribe Triticeae Dumort., (Poaceae). It is represented by five annual species in Iran: *E. bonaepartis* (Spreng.) Nevski, *E. confusum* Melderis, *E. distans* (K. Koch) Nevski, *E. orientale* (L.) Jaub and Spach and *E. triticeum* (Gaertner) Nevski (Bor, 1970). These are widely distributed in dry areas from Morocco in the West to western China in the East (Bor, 1970; Frederiksen, 1991). According to the records in the Flora Iranica, these xerophytic weed species are of open, stony slopes, steppe and semi-desert habitats. The genus grows extensively at North Eastern and North Western parts of Iran. *Eremopyrum bonaepartis* is an important native pasture species. This is the first study of this genus in Iran. Frederiksen (1991) gave systematic notes and the distribution of the genus.

Chromosome base number is  $x = 7$ . Diploid and tetraploid ( $2n = 14$  and  $28$ ) position has been observed (Sakamoto 1967, 1972 and 1973). The anatomical structure of this genus has not been studied. Morphological characters are capable in distinguishing different taxa of tribe Triticeae (Baum 1976; Frederiksen 1991; Keshavarzi *et al.*, 2002; Terrell, 1993).

Anatomical studies of grasses have provided some important diagnostic features in coastal and inter-coastal parts (Metcalf, 1960; Ogundipe and Olatunji, 1992; Keshavarzi *et al.*, 2002). Due to the importance of tribe Triticeae and wheat relatives, the purpose of this paper was to investigate the morphological and anatomical

properties of *Eremopyrum* in Iran. Frederiksen (1991) consider 4 species in this genus but there were mentioned 5 species for flora of Iran. The aim of this study was to consider inter and intra species variations.

### MATERIALS AND METHODS

*Eremopyrum* accessions were collected from different localities in Khorasan, Azerbaijan, Quazvin, Semnan and Tehran provinces (Table 1). 10 individuals were gathered at each sampling site. Specimens are deposited in the herbarium of the faculty of Science at Alzahra University.

The plant samples were identified according to Frederiksen (1991) and Bor (1970). Fresh samples were used for morphological measurements (all ten individual of a population). Thirty two quantitative and 32 qualitative characters of vegetative and reproductive parts of specimens were evaluated (Table 2 and 3). We use basal spikelet of each individual for measurements. Samples were fixed in FAA for anatomical studies. We study the anatomical characters of dorsal epidermis in coastal and inter-coastal regions and also leaf transverse sections. The length and width of the stomata were measured with an ocular micrometer using the dorsal leaf epidermis. Totally 34 quantitative and qualitative anatomical characters were evaluated and measured for all 10 individuals of each population (Table 4).

For grouping the species and populations different methods of cluster analysis including single linkage, UPGMA and Ward as well as ordination base on Principal Component Analysis (PCA) were performed on

Table 1: Voucher details of *Eremopyrum* sampled in this study

Species	Variety	Herb. No.	Address
<i>E. bonaeapartis</i>	var. <i>bonaeapartis</i>	6AU	Quazvin, Kouhin
	var. <i>bonaeapartis</i>	52AU	Quazvin, Loushan
	var. <i>bonaeapartis</i>	28AU	Khorassan, Sarakhs, after Baig
	var. <i>bonaeapartis</i>	39AU	Khorassan, Robatesang, Nasar
	var. <i>bonaeapartis</i>	34AU	Khorassan, Torbateheydariye near Abasabad
	var. <i>bonaeapartis</i>	57AU	Azerbaijan, Tabriz, Khajeh
	var. <i>bonaeapartis</i>	64AU	Semnan, Bastam
	var. <i>bonaeapartis</i>	66AU	Tehran, Karaj, Eshtehard
	var. <i>sublanuginosum</i>	22AU	Khorassan, Sarakhs, Chahak
	var. <i>sublanuginosum</i>	23AU	Khorassan, 12 km after Mozdouran
<i>E. confusum</i>	var. <i>confusum</i>	1AU	Khorassan, Bojnourd to Shirvan, Sisab
	var. <i>confusum</i>	3AU	Khorassan, Sarakhs, 14 km after Mozdouran
	var. <i>confusum</i>	10AU	Khorassan, 15 km Mozdouran
	var. <i>confusum</i>	67AU	Khorassan, Sarakhs, Mozdouran
	var. <i>glabrum</i>	8AU	Khorassan, 15 km Mozdouran
<i>E. distans</i>		17AU	Khorassan, Torbateheydarieh after Bayg
		18AU	Khorassan, Sarakhs, Chahak
		19AU	Khorassan, Sarakhs, 12 km after mozdouran
		60AU	Semnan
		62AU	Azerbaijan, Tabriz to Ahar
<i>E. orientale</i>		18239FM	Khorassan, Taibad
		11AU	Khorassan, Bojnourd, Beshghardash
		15AU	Khorassan, 120 km S. bojnourd
		63AU	Azerbaijan, Tabriz to Ahar
<i>E. triticeum</i>		13254TH	Azerbaijan, Between Tabriz and Bazargan
		29191EV	Azerbaijan, Allah Abad

Table 2: Studied quantitative morphological characters of *Eremopyrum* in Iran

No.	Character	No.	Character
1	Spike length	17	Lemma width
2	Spike width	18	Lemma Awn length
3	Blade length	19	Lemma Nerve No.
4	Blade width	20	Lemma hairs length
5	Ligule length	21	Palae length
6	Upper Glume length	22	Palae width
7	Upper Glume width	23	Palae Gap depth
8	Upper Glume Awn length	24	Palae Nerve No.
9	Upper Glume Nerve No.	25	Palae apical lobes No.
10	Upper Glume Hairs length	26	Lodicules length
11	Lower Glume length	27	Lodicules width
12	Lower Glume width	28	Caryopsis length
13	Lower Glume Awn length	29	Caryopsis width
14	Lower Glume Nerve No.	30	Length of corona
15	Lower Glume hairs length	31	Floret No.
16	Lemma length	32	Culm length

Table 3: Studied qualitative morphological characters of *Eremopyrum* in Iran

No.	Character	No.	Character
1	Spike hair Type (Smooth 0, Hairy 1, both type 2)	10	Glume Hair (Smooth 0, hairy 1, both type 2)
2	Spike Shape (Elliptic 0, rectangular-elliptic 1, narrow rectangular 2, wide rectangular 3)	11	Frequency of Glume Hair (Dense 0, sparse 1)
3	Ligule Texture (Membranous 0, No membranous 1)	12	Shape of Glume Apex (awned 0, unawned 1)
4	Glume Base Shape (Connate 0, No connate 1)	13	Lemma Texture (Coriaceous 0, noncoriaceous 1)
5	Glume Texture (Coriaceous 0, Noncoriaceous 1)	14	Lemma Color (Darker at base 0, others 1)
6	Glume Color (pale green 0, Dark green 1)	15	Lemma Shape (Lanceolate 0, angustiform 1, curved 2)
7	Glume Shape (Lanceolate 0, angustiform 1, curved 2)	16	Lemma nerve condition (no nerved 0, nerved 1)
8	Glume nerve condition (no nerved 0, nerved 1)	17	Convergence of Lemma Nerves (Presence 0, absence 1)
9	Convergence of Glume Nerves (present 0, absence 1)	18	Lemma Hair (Smooth 0, hairy 1, both type 2)

Table 3: Continued

No.	Character	No.	Character
19	Frequency of Lemma Hair (Dense 0, Sparse 1)	26	Palae Apex (gapped1, no gapped 0)
20	Shape of Lemma Apex (Awned 0, unawned 1)	27	Caryopsis Color (brown 0, yellow 1)
21	Palae Texture (Membranous 0, no membranous 1)	28	Lodicules Texture (Membranous 0, no membranous 1)
22	Palae Nerve condition (nerved 1, no nerved 0)	29	Lodicules Shape (Oblong 0, Widely triangular 1, Rectangular-elliptic 2)
23	Palae Color (pale green 0, dark green 1)	30	Lodicules covering (haired 1, Glabrous 0)
24	Convergence of Palae Nerves (Presence 0, Absence 1)	31	Caryopsis Shape (Angustiform 0, short to wide rectangular 1, rectangular
25	Palae Hair (Smooth 0, Short and hard hairs 1)	32	Rachis Fragility (fragile 0, hard 1)

Table 4: Studied Anatomical Characters for species of *Eremopyrum* in Iran

Quantitative Characters

No.	Character	No.	Character
1	Short cell No. IC	11	Stomata width IC
2	Prickle No. IC	12	Stomata diameter IC
3	Macro hair IC	13	Silica cell No C
4	Stomata No.IC	14	Prickle No. C
5	No. of stomata band IC	15	Macro hair C
6	Long cell length IC	16	Long cell width C
7	Long cell width IC	17	Long cell length C
8	Short cell length IC	18	Silica cell width C
9	Short cell width IC	19	Silica cell length C
10	Stomata length IC		

Qualitative Characters

	Character		Character
1	Short cell (Single 0, Single and twin 1)	9	Frequency of Prickles (Few 0, Numerous 1, Mild 2)
2	Frequency of short cell (/Mild 2, numerous 1, few 0)	10	Macro Hair (Present 1, Absent 0)
3	Long cell width (Even 1, noneven 0)	11	Frequency of Macro hairs (Mild 2, Numerous 1, Few 0)
4	Thickening of long cell (Mild 2, Thick 1, Thin 0)	12	Shape of subsidiaries (Domeshape 1, Parallel sided 0)
5	Shape of long cell (Sinusian 1, even 0)	13	Silica Cell Shape (Rectangular 1, Nonrectangular 0)
6	Silica Cell (Single 0, Twin 1, both 2)	14	Silica Cell Walls (Curvaceous 1, no curvaceous 0)
7	Frequency of silica cell (Mild 2, Numerous 1, Few 0)	15	Short cell shape (Rectangular 1, Nonrectangular 0)
8	Prickle (present 1, Absent 0)		

C = Coastal, IC = Intercostal

standardized data. Squared Euclidean distance was used as a measure of similarity in cluster analysis. Uni-Variate and multivariate statistical analyses used SPSS ver. 9 software.

## RESULTS

**Quantitative morphological characters:** From studied characters, lemma and glume nerve number, palae nerve and lobe numbers show no variation. None variant features are omitted from further analysis. By factor analysis, it was shown that 7 factors are responsible for more than 72% of variation in *Eremopyrum* species. We

choose the first three factors that made 53% of variations. The first factor which causes more than 29% of variation comprises characters as lemma awn Length, length of lowermost and uppermost lemma, length of lemma hair, awn length of lowermost and uppermost lemma, have the most important role in morphological variation in *Eremopyrum* species in Iran. As second factor there are spike width and length and ligules length. Length of caryopsis and width of lemma take part in third factor and cause 9% of variation. There is a great correlation between length of awn in lowermost and uppermost glume, length of lowermost and uppermost glume, length of upper glume hairs and hairs of lemma.

Dendrogram based on quantitative characters reveals clear separation of taxa. UPGMA cluster was in concordance with Ward method. In this Diagram *E. distans* have a separate branch. *Eremopyrum bonaepartis* var *bonaepartis* and *E. orientale* comprise another distinct branch. There are confusions between *E. confusum* var *glabrum* and *E. bonaepartis* var *sublanuginosum* and *E. confusum* var *confusum*. PCA diagram shows also these relations between taxa (Fig. 1). As it could be seen there is a clear separation in *E. distans* and *E. orientale* due to 32 quantitative morphological characters.

**Qualitative morphological characters:** The results of qualitative morphological characters are shown in Table 5. One of the most distinct differences between *Eremopyrum* species in Iran is Length of Spike (Fig. 2). *E. triticeum* has the shortest spike and *E. bonaepartis* var *bonaepartis* have the longest one.

Factor analysis shows that the first 5 factors cause more than 97% of variation in these taxa. Spike shape, the frequency of glume and lemma hairs are the most important qualitative factors (46% of variation). Caryopsis and ligules shape cause more than 20% of variation (2nd factor). The presence of hairs in glume, spike and lemma are responsible for 16% of variation (3rd factor).

**Anatomical observations:** In coastal region, the walls of long cells were even (*E. distans*, *E. orientale*, *E. confusum* var. *glabrum* and *E. triticeum*) or uneven (*E. bonaepartis*, *E. confusum* var. *confusum*). Silica cells were in pair or single in all species. *E. triticeum* has only single silica cells (Table 6). Except *E. distans* and *E. bonaepartis* var. *sublanuginosum* other species have prickles. Subsidiaries cells are dome shapes in *E. triticeum* and *E. distans* and *E. orientale* but these are parallel sided in *E. bonaepartis* and *E. confusum* (Fig. 3 and 4).

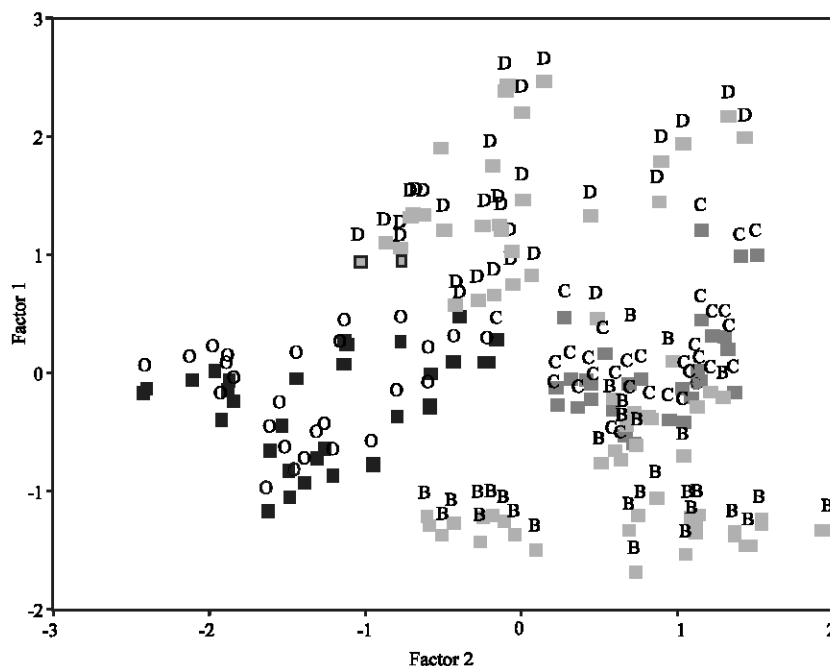


Fig. 1: PCA diagram for *Eremopyrum* Species in Iran by Using Quantitative Characters (Abbreviations: B: *E. bonaepartis*, C: *E. confusum*, D: *E. distans*, O: *E. orientale*)

Table 5: Results of evaluation of qualitative morphological characters in *Eremopyrum* of Iran

Taxon	Spike		Shape of			Shape of					Caryopsis Shape	Lodicules Covering	Rachis Fragility
	hair type	Spike Shape	Glume Shape	Glume Hair	Glume Apex	Lemma Shape	Lemma Hair	Lemma Apex	Palea Apex	Lodicules Shape			
<i>E. triticeum</i>	2	0	0	2	0	0	1	1	0	1	0	0	0
<i>E. bonaepartis</i> var. <i>bonaepartis</i>	0	3	0	0	0	0	0	0	1	2	1	2	1
<i>E. bonaepartis</i> var. <i>sublanuginosum</i>	1	3	0	1	0	0	1	0	1	2	1	2	1
<i>E. confusum</i> var. <i>confusum</i>	1	3	0	1	1	0	1	0	1	2	1	2	1
<i>E. confusum</i> var. <i>glabrum</i>	0	3	0	0	1	0	0	0	1	2	1	2	1
<i>E. distans</i>	1	2	1	1	1	1	1	1	1	0	1	0	1
<i>E. orientale</i>	2	1	2	2	1	2	2	1	1	1	1	1	1

Codes are as mentioned in Table 3

Table 6: Result of Qualitative characters study of dorsal epidermis in *Eremopyrum* species of Iran

Characters	<i>E. bonaepartis</i>		<i>E. confusum</i>			<i>E. triticeum-</i>	<i>E. distans</i>	<i>E. orientale</i>
	var. <i>bonaepartis</i>	var.	var. <i>confusum</i>	var. <i>glabrum</i>				
		<i>sublanuginosum</i>						
Short cell	0	0	1	0	0	0	0	1
Frequency of short cell	1	2	2	2	2	2	0	0
Silica cell	2	0	1	0	0	0	1	1
Frequency of silica cells	0	0	0	0	0	0	1	0
Prickles	1	1	1	1	1	1	0	1
Frequency of prickles	1	2	1	0	0	0	0	2
Macro hair	0	1	0	0	0	0	1	1
Frequency of macro hair	0	1	0	0	0	0	1	2
Shape of subsidiaries	1	0	0	0	0	1	1	1

Codes are as mentioned in Table 4

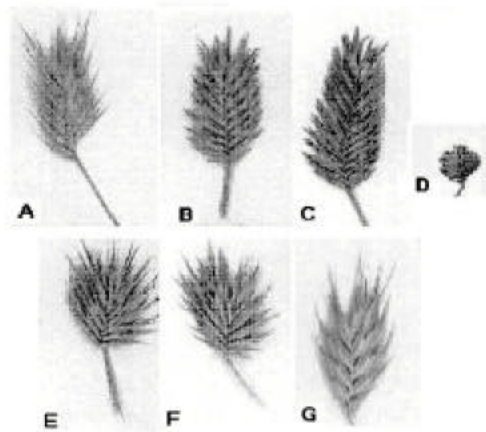


Fig. 2: Spike morphological Variation in *Eremopyrum* species of Iran. A = *E. distans*, B = *E. bonaepartis* var. *bonaepartis*, C = *E. bonaepartis* var. *sublanuginosum*, D = *E. triticeum*, E = *E. confusum* var. *glabrum*, F = *E. confusum* var. *confusum* and G = *E. orientale*

In inter-coastal region there are single short cells except in *E. orientale*. Walls of long cells are only even in *E. distans* and *E. triticeum* but both types are present in *E. orientale*, *E. bonaepartis* and *E. distans* (Fig. 3 and 4). Prickles are present in *E. bonaepartis* var. *bonaepartis*, *E. bonaepartis* var. *sublanuginosum*, *E. confusum* and *E. orientale*. There are macro hairs in *E. distans*, *E. bonaepartis* var. *sublanuginosum* and *E. orientale*. Stomata subsidiaries shape in *E. distans*, *E. orientale* and *E. triticeum* are dome shape. Parallel-sided subsidiaries are seen in other two species of *Eremopyrum*.

Factor analysis shows three first qualitative factors in dorsal epidermis causes more than 76% of variation. As First factor, frequency of prickles and thickening of long cells are important. Frequency of buliform cells (2nd factor) and presence of prickles (3rd Factor) have also important roles in *Eremopyrum* variations.

Factor analysis of quantitative characters of dorsal epidermis also shows that first three factors are more important causing more than 60% of variation. In first factor width and length of short cells in intercostal

region, stomata length, diameter of stomata opening has the major effects. Width of stomata, prickles number and length of long cells in coastal region, cause the variation in 2nd factor. The observed variation in 3rd factor is due to macro-hair number in inter-coastal and coastal regions.

Flag leaf transverse sections show some homogeneity. All species show two layer bundle sheaths. Inner layer is complete and the cells are smaller than outer layer. Mesophyll has a radial differentiation. Shapes of transverse sections show some differences in *Eremopyrum* species (Fig. 5 and 6). Factor analysis of quantitative characters of transverse section shows the importance of three first factors. These factors cause more than 64% of variations. In First factor width of sclerenchyma, length of buliform cells, number of vascular bundles and length of sclerenchyma have the most important role. In second factor number of cells of outer layer of bundle sheath, cause the 20% of variation. Number of cells of inner layer of bundle sheath (3rd factor) makes the 14% of variation.

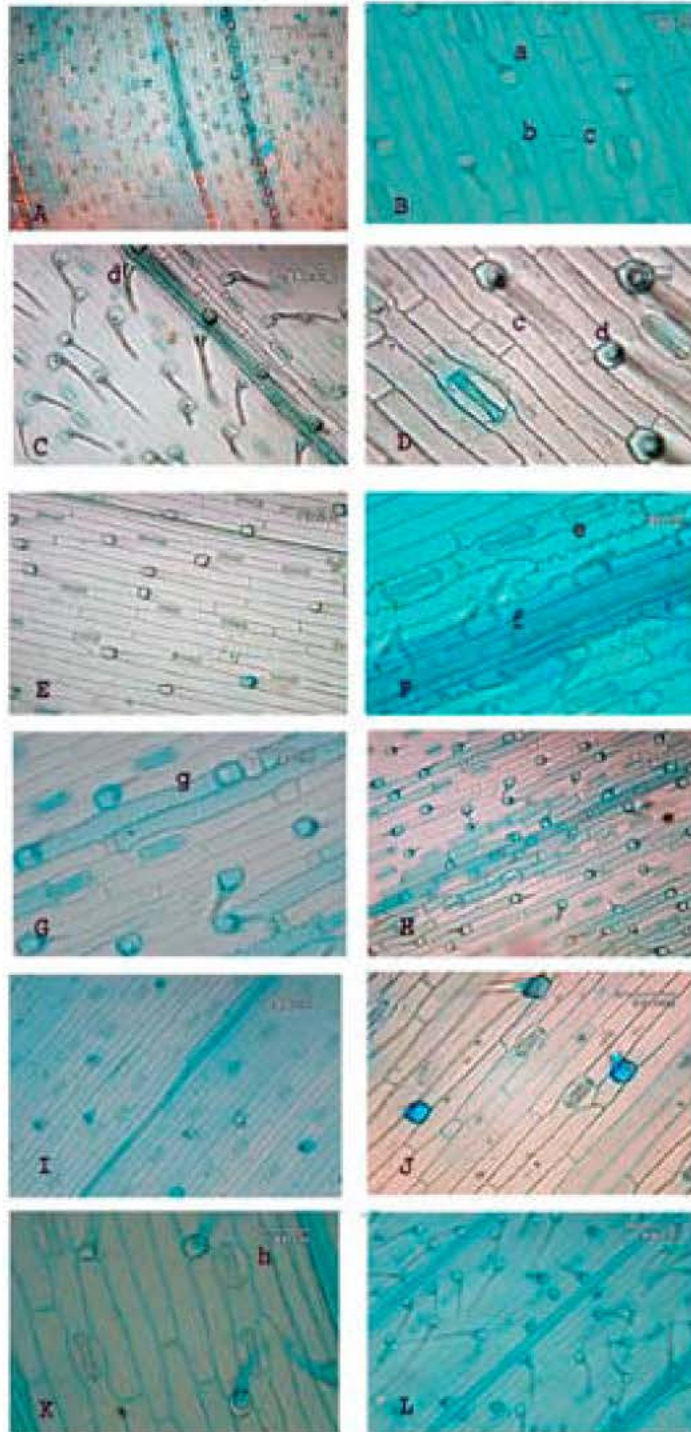


Fig. 3: Dorsal epidermis in *Eremopyrum* species of Iran. A and B = *E. bonaepartis* var. *bonaepartis*, C and D = *E. bonaepartis* var. *sublanuginosum*, E = *E. confusum* var. *glabrum*, F, G and H = *E. confusum* var. *confusum*, I and J = *E. triticeum*, K and L = *E. distans*. Abbreviations: a = Prickles, b = Subsidiaries, c = Long cell, d = Silica bodies, e = Intercoastal region, f = Coastal region, g = Long cell with sinuous walls, H = Dome shaped subsidiaries

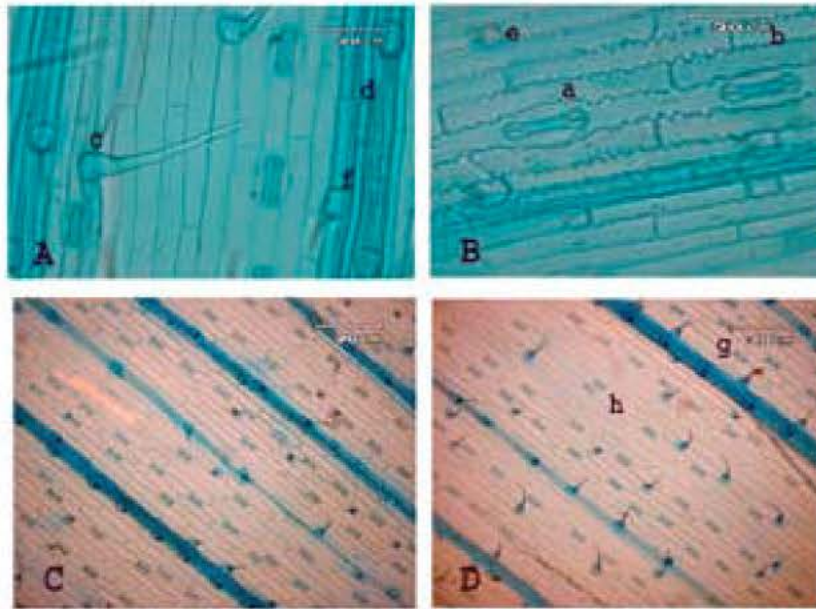


Fig. 4: Dorsal epidermis in *Eremopyrum orientale*. A and B = Bojnourd population, C and D = Beshghardash population. Abbreviations: a = Subsidiaries, b = Long cells, c = Macro hair, d = Silica bodies, e = Short cell, f = Prickle, g = Coastal region, h = Intercoastal region

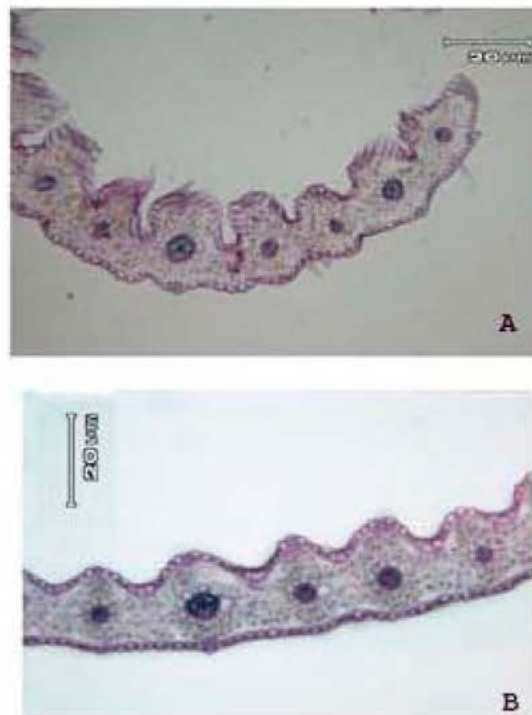


Fig. 5: Different shapes of transverse sections in *Eremopyrum* species. A = *E. orientale*, B = *E. confusum* var. *confusum*



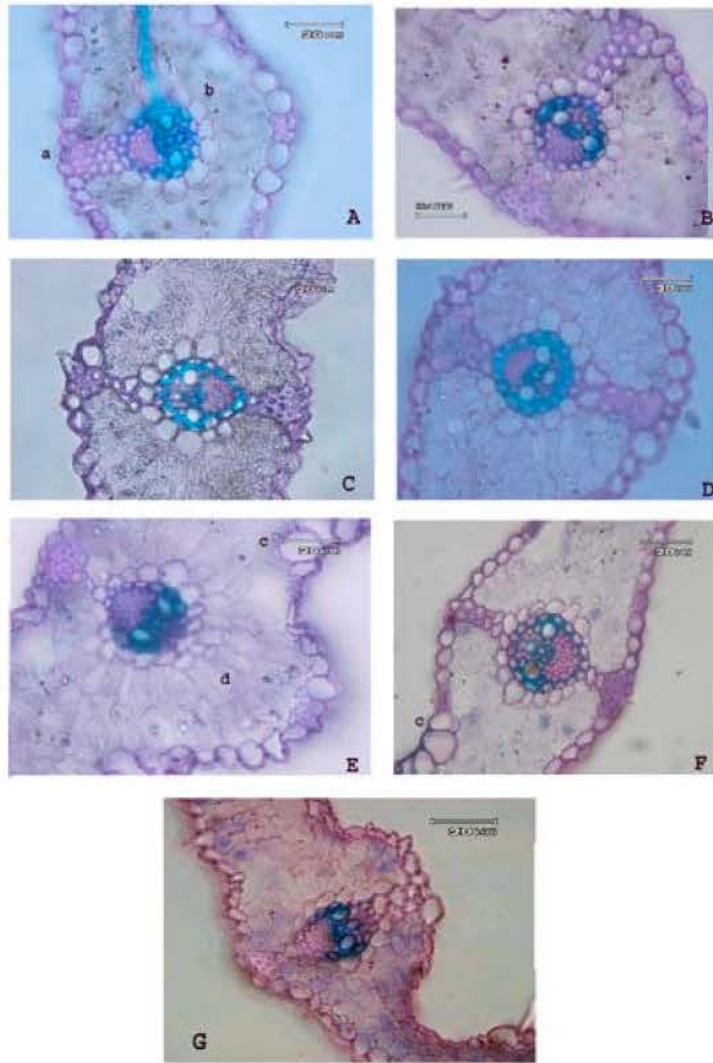


Fig. 6: Mid rib differences among *Eremopyrum* species of Iran. A = *E. bonaepartis* var *subalnuginosum*, B = *E. bonaepartis* var *bonaepartis*, C = *E. confusum* var *confusum*, D = *E. confusum* var *glabrum*, E = *E. orientale*, F = *E. distans*, G = *E. triticeum*. Abbreviations: a = Grid of Scleranchyma, b = outer layer of bundle sheath, c = bulliforme cells and d = differentiated mesophyl

#### DISCUSSION

There are a lot of significant differences in *Eremopyrum* species. Due to factor analysis awn of glume and lemma, spike shape, caryopsis shape, hair in glume and lemma, the presence of bristles in lodicules and the gap of the palae tip are important diagnostic characters. We can use them in providing an efficient identification key for *Eremopyrum* species in Iran.

Frederiksen (1991) stated that *Eremopyrum* has only 4 species in world. She put *E. confusum* as a separate subgroup of *E. bonaepartis*. We found that *E. bonaepartis* are distinguished from *E. confusum* in Iran

by awn position. Some reproductive morphological characters as length of spike, glume and lemma, hair and gap of the tip of palea and the size of lodicules in *E. confusum* are more than *E. bonaepartis*. Spike shape is very efficient feature in separating *Eremopyrum* species in Iran. In identification key of *Eremopyrum* species of Iran, some characters as glume awn, length of glume, presence of prickles and macro-hairs, shape of glume and lemma are very important. Accessions of *E. orientale* show some variation in morphology and anatomy. They should be studied further to find whether there are any genetic variations.

Anatomical characters are efficient in distinguishing some morphologically similar species group as *E. orientale*, *E. distans* and *E. triticeum*. *E. confusum* and *E. bonaepartis* are anatomically distinct. Some differences in number of stomata rows and vascular bundles were shown. Factor analysis shows that length of buli-form cells, number of vascular bundles, number of cells of inner and outer bundle sheaths, frequency of prickles, frequency of short cells, width and length of short cells in inter-coastal region and number of prickles are diagnostic anatomical characters.

Morphological and anatomical variations of *Eremopyrum* species were studied. The values of characters were evaluated. We find five distinct *Eremopyrum* species in Iran. Due to the observed differences, it seems inefficient to place *E. confusum* as a separate subgroup of *E. bonaepartis*. Further study should focus the genetic variation of some taxonomic confusing species as *E. confusum* and *E. bonaepartis*.

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