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Morphological Study of Salicornieae (Chenopodiaceae) Native to Iran

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Abstract: The tribe Salicornieae Dum. belonging to the subfamily Salicornioideae Kostel (Chenopodiaceae Vent.) includes halophyte plants. These 5 genera and 6 species are distributed in different habitats of Iran. Members of this tribe have reduced vegetative parts, scale like leaves and articulated stems. In this study quantitative and qualitative morphological characters for 46 accessions of Salicornieae were evaluated. Vegetative characters are not sufficient to distinguish these taxa. Our results indicated that vegetative form, globular buds, plant color, stem base disarticulation and presence of node at the base of inflorescence are diagnostic character in this tribe. Besides floral arrangement in inflorescences, bracts shape, color and shape of seeds are important key features in members of Salicornieae in Iran. Seed coat ornamentations and its hairs and color are found to be helpful in distinguishing these taxa. Quantitative characters show variation too, but as they were not significant, they can not help to resolve the taxonomic problems of this tribe in Iran. Based on studied morphological characters, an identification key for members of this tribe in Iran is provided.

Key words: Salicornieae, Chenopodiaceae, micro-morphology, Iran

INTRODUCTION

The tribe Salicornieae belongs to the subfamily Salicornioideae Kostel. (Chenopodiaceae Vent.), includes halophyte plants. These 5 genera and 6 species are distributed in different habitats of Iran (Hedge, 1997). Members of the tribe Salicornieae Dumort., are characterized by their distinctive reduced leaves, which may be modified to form an articulated, photosynthetic stem (de Frain, 1912). These specialized plants generally have spike-like compound inflorescences, comprised of paired cymules of tiny flowers that are sessile within succulent free or fused bracts (Kuhn *et al.*, 1993). Each cymule has (1-) 3 (-12) flowers.

The Salicornieae are among the most salt-tolerant plant and frequently occur in saline areas associated with coast lines, tidal floodways and salt lakes (Wilson, 1980; Davy *et al.*, 2001; Asri, 1998). These halophytes are globally distributed, being found on every continent with the exclusion of Antarctic (Kuhn *et al.*, 1993). Reduced morphology of Salicornieae limits the availability of easily recognized diagnostic characters at the tribal and generic levels. Furthermore the Salicornieae exhibit considerable phenotypic variation at the population level (Wilson, 1980; Davy, 2001; Freitag *et al.*, 2001) and taxonomic confusion is exaggerated by the occurrence of species complex (Wilson, 1980) and polyploids (Shepherd and Yan, 2003). The succulent vegetative morphology may also be modified when dried, limiting the use of herbarium material. As a result, the sub familial

and inter-generic relationships of the Salicornioideae are not fully clarified (Shepherd *et al.*, 2005).

Although few vegetative and floral features are diagnostic in the Salicornioideae, seed characters have been recognized as potentially useful at both the generic and specific levels (Wilson, 1980). In some respects mature fruits and seeds of the Salicornioideae are more convenient to use as taxonomic characters than vegetative and floral features. They are less inclined to exhibit variability in the field, are frequently retained on perennial plant for months or even years and are likely to remain relatively unmodified upon drying (Shepherd, 2005).

There have been few global taxonomic treatments of the Salicornieae (Kadereit *et al.*, 2003) and to our knowledge there has been no detailed survey of examining morphological characters of the tribe in Iran. The populations of tribe Salicornieae in Iran are studied for the first time.

MATERIALS AND METHODS

In this study 46 Accessions of different salty marshlands of Iran were collected (Fig. 1 and Table 1). At collecting site altitude from sea level were measured and soil sample were gathered. Stomata were taken from fresh material. We use methyl green for studying this part. Seeds were gathered from dried specimens. Seed length and width were measured by digital caliper for 50 seeds per species. Seed coat ornamentation was studied by



Fig. 1: Distribution map of accessions of the tribe Salicornieae in Iran

SEM (JEOL model JXA-140). Pollen grain gathered from fresh flowering taxa, were studied without acetolysis. Pollen diameter was measured for 100 grain of each accession (by light microscopy). Observed morphological characters and their states are shown at Table 2. Soil Electric conductivity was evaluated by Electrolytic Conductivity measuring set model MC-1-Marv. All collected samples are deposited at Herbarium of Alzahra University.

RESULTS

Ecological features: Members of tribe Salicornieae are found in soils with electric conductivity from 17 to 150 Mm cm^{-1} (Table 1). The highest electric conductivity (150) belongs to a population of *Salicornia europaea* from western coast of Uromiyeh Lake (Azerbaijan). The lowest electric conductivity (17) belongs to populations of *Salicornia europaea* and *Halocnemum strobilaceum* from Khajeh salt mine in Azerbaijan. As a whole Salicornieae populations of Iran are found in a wide range of soil EC.

Plants of this tribe are found in a wide range of altitude (0-1600) from sea level in Iran (Table 1). Except *Arthrocnemum macrostachyum* which is limited to Persian Gulf Coast, at south of Iran, other taxa of central parts of Iran and Azerbaijan are distributed at different altitude till 1600 m. *Halocnemum strobilaceum* are distributed in disturbed salt marshes, the coast of salty lakes and rivers

and make a great patches of vegetation. There are no other plants, where Salicornieae members grow in Iran. In western coast of Uromiyeh Lake, Coast of Talkheroud River and Mighan desert we can visit large marshes of *Halocnemum strobilaceum*. Populations of *Halostachys belangeriana* are also take part in making large patches of vegetation in coast of salt marshes and salty rivers and in the rivers estuaries. Populations of genus *Salicornia* comprise the first band of vegetation near salty rivers and lakes. After this band there is a layer of *Halocnemum strobilaceum*. Members of Salicornieae are found in different salty habitats of Iran (Fig. 2).

Macro morphology: We have studied all macro morphological characters for 64 accessions. We have put the average values of each taxon in Table 3. Some no variant characters as articulated stems, phyllotaxy, number of flowers, perianth parts and stamens were eliminated. The most variation is shown in length and diameter of inflorescence parts and bract shape.

Micromorphology: Pollen grains were studied by light microscopy. We found that all pollen grains are spherical (Fig. 3). Pollen grains of this tribe were all polyaperturate although they show some differences in their diameter (Table 4).

Seeds of members of tribe Salicornieae in Iran are very different from each other. Seeds are globular and elliptical. Their color is from pale yellow-brown to dark

Table 1: Character of sampled accessions of tribe Salicornieae native to Iran

Taxon	Address	Herb. No.	Collector	Altitude m.s.l	E.C. (Mm cm ⁻¹)
<i>Arthrocnemum macrostachyum</i> (Moric) C. Koch	Boushehr, boushehr	8313	Keshavarzi	0	-
<i>Halocnemum strobilaceum</i> (Pall.) M. Bieb	Golestan, Incheboroun	8292	Keshavarzi	20	-
	Azerbaijan, Uromiyeh Lake	8261	Zare	1300	98.0
	Azerbaijan, Sarab, Asbforoshan	8375	Zare	-	46.5
	Azerbaijan, Marand, Ivaghli	8381	Zare	1050	32.9
	Azerbaijan, Marand, Gapleg	8348	Zare	800	48.8
	Azerbaijan, Marand, 125 km to Maku	8347	Zare	800	40.5
	Azerbaijan, Islami Island, Khaseban	8366	Zare	1350	75.3
	Azerbaijan, Khajeh	8361	Zare	1500	17.7
	Azerbaijan, Tabriz, 35 km to Ahar	8341	Zare	1470	70.2
	Qoum, Qoum	8271	Moazeni	823	115.0
	Markazi, Arak, Mighan desert	8383	Zare	1600	50.6
	Fars, Shiraz	8291	Zare	1493	-
	Boushehr, boushehr	8200	Moazeni	-	-
<i>Halostachys belangeriana</i> (Moq.) Botch	Azerbaijan, Marand, Ivagli	8380	Zare	1050	32.9
	Qoum, Hoze Sultan Lake	8221	Moazeni	823	45.2
	Tehran, Varamin	8282	Moazeni	83	-
	Isfahan, Varzaneh	8390	Zare	1498	35.5
<i>Microcnemum coralloides</i> (Loscos and Pardo) Buen	Markazi, Arak, Mighan desert	8201	Zare	1600	52.6
<i>Salicornia europaea</i> L.	Golestan, Incheboroun	8291	Keshavarzi	20	-
	Azerbaijan, Uromiyeh Lake	8292	Zare	1300	150.0
	Azerbaijan, Sarab, Asbforoshan	8372	Zare	1400	42.5
	Azerbaijan, Sarab, Asbforoshan	8373	Zare	1400	45.3
	Azerbaijan, Sarab, Asbforoshan	8374	Zare	1400	46.5
	Azerbaijan, Marand, Ivagli	8346	Zare	1050	32.9
	Azerbaijan, Marand road, 44 km to Khoy	8349	Zare	800	82.8
	Azerbaijan, Islami Island, Khaseban	8369	Zare	1350	70.1
	Azerbaijan, Islami Island, Khaseban	8367	Zare	1350	78.5
	Azerbaijan, Khajeh	8362	Zare	1500	20.4
	Azerbaijan, Khajeh	8363	Zare	1500	17.7
	Azerbaijan, Khajeh	8364	Zare	1500	17.7
	Azerbaijan, Khajeh	8365	Zare	1500	17.7
	Qoum, Qoum	8272	Moazeni	823	-
	Markazi, Saveh	8352	Keshavarzi	1000	54.0
	Markazi, Arak, Mighan desert	8384	Zare	1600	74.2
	Fars, Shiraz, Fassa bridge	8392	Zare	1493	-
	Fars, maharlou Lake	8393	Zare	1493	-
	Fars, maharlou Lake	8394	Zare	1493	-
	Isfahan, Varzaneh	8395	Zare	1498	94.0
	Isfahan, Varzaneh	8386	Zare	1498	95.3
	Isfahan, Varzaneh	8389	Zare	1498	95.0
<i>Salicornia persica</i> Akhani	Fars, Maharlou Lake	8395	Zare	1493	-
	Fars, Maharlou Lake	8396	Zare	1493	-
	Isfahan, Varzaneh	8387	Zare	1498	96.0
	Isfahan, Varzaneh	8388	Zare	1498	96.0

brown. Studying seeds with stereo microscopy we found that they have some differences in their seed coat ornamentations so we use Scanning Electro Micrographs to study these differences. As it could be seen in Fig. 4 and 5 ornamentations of these taxa are in form of tuberculate (*Arthrocnemum macrostachyum*), Granular (*Halocnemum strobilaceum*) and smooth (*Halostachys belangeriana*). Two species of *Salicornia* show some differences in distribution patterns of seed coat hairs. These hairs are in form of hooked or smooth (Fig. 4 and 5). Seeds length and width and the ratio of these characters show differences through members of this tribe in Iran (Table 4).

Epidermis which covers the stems of taxa was studied. We found that Stomata distribution pattern is of anemocytic type. Some characters as Frequency of stomata per leaf area, Length and width of guard cells and features of epidermis cell walls show some differences (Table 5). The lowest stomata index is found in *Microcnemum coralloides* and the highest stomata Index is found in *Holostachys belangerian*. Different populations of *Salicornia* in Iran show the highest variation. There are some special papillae on the epidermis surface of *Halostachys belangeriana*, *Arthrocnemum macrostachyum* and *Halocnemum strobilaceum*. These papillae are absent in other Iranian members of this tribe (Fig. 6).

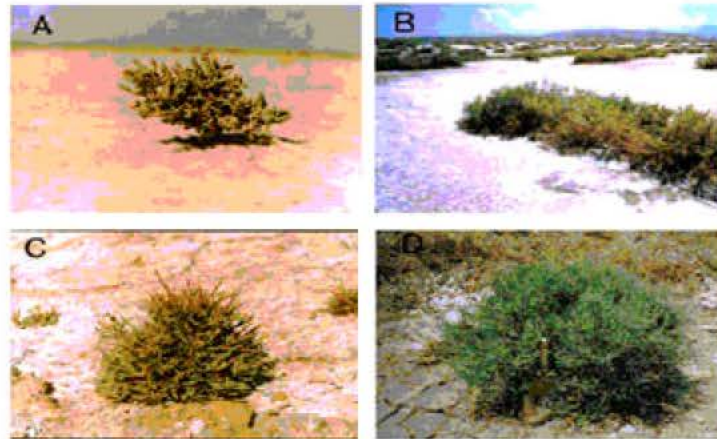


Fig.2: Members of tribe Salicornieae in different habitats of Iran A and B: *Halocnemum strobilaceum*, C and D: *Salicornia europaea*

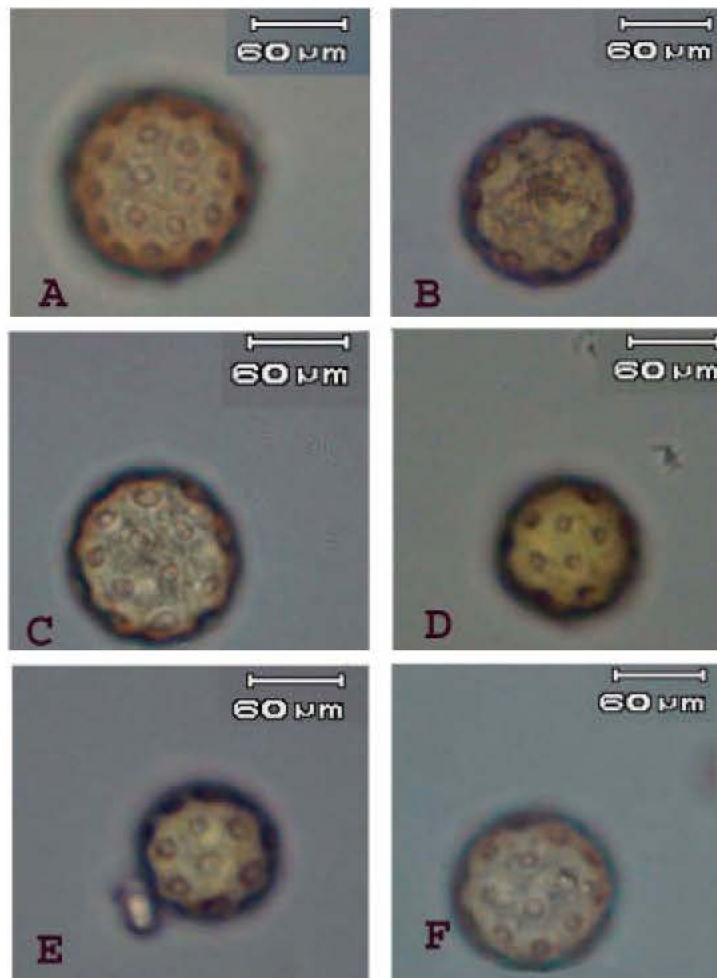


Fig.3: Pollen Grains of Salicornieae in Iran A; *Salicornia europaea*, B; *Salicornia persica*, C, D and E *Halocnemum strobilaceum* F; *Halostachys belangeriana*

Table 2: Evaluated morphological character of Salicomeae in Iran

Qualitative Ch.	State of Ch.	Quantitative Ch.	Scale
Life duration	Perennial (P) Annual (An)	Plant height	cm
Vegetative form	Shrub (Sh) Herb (H)	Canopy diameter	cm
Woody base	Present (+) Absent (-)	Root diameter	mm
Stem base branched	Present (+) Absent (-)	Internodes diameter	mm
Articulated stems	Present (+) Absent (-)	No. of clustering	-
Globular buds	Present (+) Absent (-)	No. of cluster per node	-
Phyllotaxy	Opposite Non-opposite	Flower no.	-
Hairs	Present (+) Absent (-)	Perianth no.	-
Smell	Present (+) Absent (-)	Length of inflorescence parts	mm
Salt crystals	Present (+) Absent (-)	Length of terminal Infl.	mm
Color	Yellow-green Green-orange Pale Green Glaucous	Length of lateral Infl.	mm
Inflorescence	Cymous Non cymous	Diameter of terminal Infl.	mm
Arrangement of flower	Linnear Triangular	Diameter of lateral Infl.	mm
Bracts shape	Cup shape Ob pyramid Peltate	Length of Infl. Base	mm
Exertion of stamens	Present (+) Absent (-)	No. of stamens	-

Table 3: Micromorphological characters of the Iranian Salicornieae

Taxa	<i>Arthrocnemum macrostachyum</i>	<i>Halocnemum strobilaceum</i>	<i>Halostachys belangeriana</i>	<i>Microcnemum coralloides</i>	<i>Salicornia europaea</i>	<i>Salicornia persica</i>
Character						
Life Duration	P	P-An	P	An	An	An
Vegetative form	Shrub	Shrub	Shrub	Herb	Sh-H	Sh-H
Woody base	+	+	+	-	+-	+-
Stem base branched	+	+	+	-	+-	+
Articulated stems	+	+	+	+	+	+
Globular buds	-	+	-	-	-	-
Phyllotaxy	Opposite	Opposite	Opposite	Opposite	Opposite	Opposite
Hairs	-	-	-	-	-	-
Smell	-	-	-	-	-	-
Salt crystals	+	-	-	+	+	+
Color	Glaucous	Yellow to purplish green	Glaucous	Yellow to purplish green	Pale to dark purplish green	Pale to dark purplish green
Inflorescence						
Arrangement of flower	Linear	Linear	Linear	Linear	Triangular	Triangular
Bracts	+	+	+	+	-	-
Bracts shape	Cuplike	Peltate	ob-pyramid	Cuplike	-	-
Exertion of stamens	+	-	-	-	-	-
Plant height	26.25±9.46	23.45±16	7	112.5±75	32.31±10.72	35±7.07
Canopy diameter	28.75±2.5	16.04±14.25	2	122.5±63.96	46.54±34.3	50±14.14
Root diameter	4.25±0.95	3.5±1.55	1.5	4.75±0.5	3.67±0.86	4.5±0.7
Internodes diameter	2.22±0.15	2.35±0.93	1.2	2.2±0.5	2.48±0.8	5.5±0.7
No. of clustering	3±0.82	2.36±0.79	1	4	3.08±2.36	4
No. of cluster per node	5.25±2.06	3.4±0.9	2	3	3	3
Flower no.	3	3	3	3	3	3
Perianth no.	3	3	3	3	3	3
Length of inflorescence parts	2.95±0.1	2.83±0.65	1.9	0	0	2.2
Length of terminal Infl.	123±37.89	46.43±22.21	29	25.66±5.13	12.62±4.8	60±4.1
Length of lateral Infl.	92.5±40.3	34.31±14.92	179	21.33±6.35	9.13±4.08	50±5.1
Diameter of terminal Infl.	1.65±0.28	1.74±0.43	1.8	2.63±0.05	1.97±0.36	4.2±0.2
Diameter of lateral Infl.	1.42±0.66	1.74±0.43	1.4	2.63±0.06	1.98±0.36	4.2±1.3
Length of infl. Base	5.75±0.95	7.12±3.77	6.2	1.6±1.15	0	4.8±1.1
No. of stamens	1	1	1	1	1	1

+: Present; -: Absent

Table 4: Results of studying pollen grains and seeds in members of tribe Salicornieae in Iran

Taxon	Pollen diameter	Seed length	Seed width	Seed L/W
<i>Arthrocnemum macrostachyum</i>	20.50±0.7	1.18±0.07	0.74±0.06	1.59
<i>Halocnemum strobilaceum</i>	20.77±1.2	0.90±0.08	0.69±0.04	1.30
<i>Halostachys belangeriana</i>	18.66±1.52	0.83±0.02	0.56±0.02	1.48
<i>Salicornia europaea</i>	23.58±2.8	1.18±0.2	0.73±0.11	1.62
<i>Salicornia persica</i>	29.00±2.58	1.58±0.09	0.66±0.04	2.39

Table 5: Stem epidermis characters in members of tribe Salicornieae in Iran

Taxon	St. index	Length of guard cell	Width of guard cell
<i>Arthrocnemum macrostachyum</i>	76.15±6.25	30	30
<i>Halocnemum strobilaceum</i>	76.03±12.81	29.44±3	24.72±2.6
<i>Halostachys belangeriana</i>	79.22±26.6	30.00±2.5	26.66±3.8
<i>Microcnemum coralloides</i>	29	42.5	35
<i>Salicornia europaea</i>	61.24±17.85	28.38±3.17	25.00±3.53
<i>Salicornia persica</i>	51.68±19.29	33.12±9.86	26.25±8.29

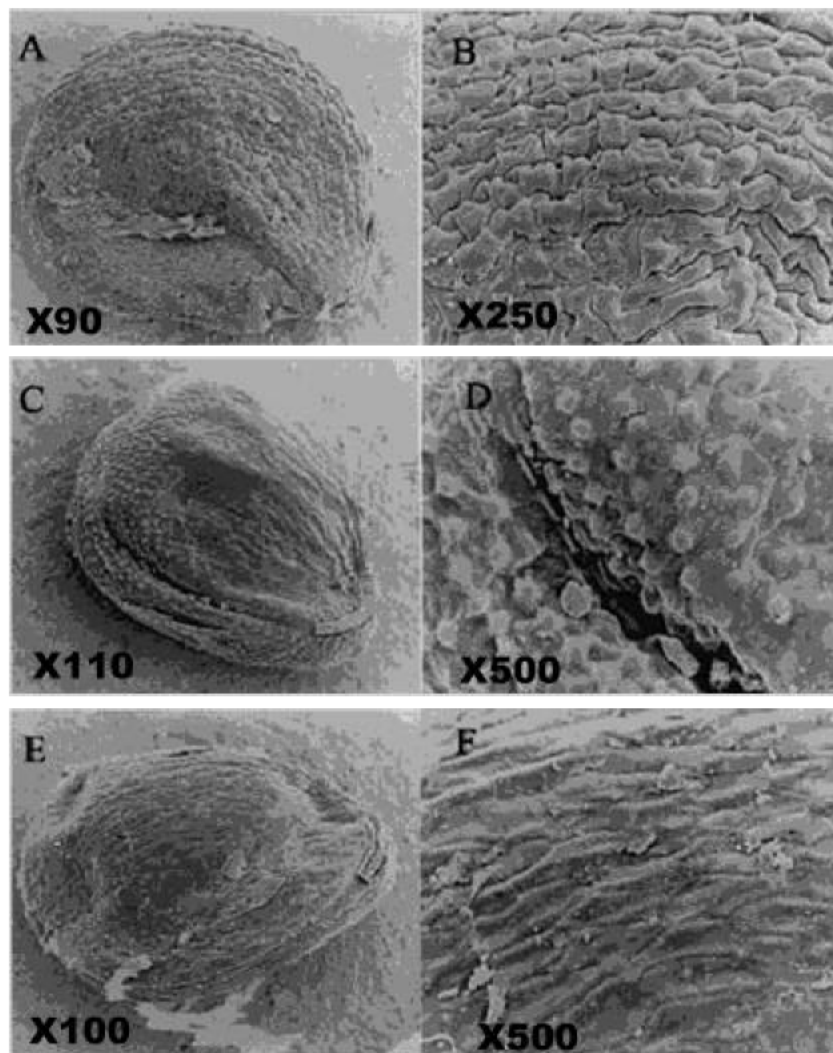


Fig. 4: Scanning electron micrograph of salicornieae seeds of Iran showing seed coat ornamentations. *Arthrocnemum macrostachyum* (A and B); *Halocnemum strobilaceum* (C and D); *Halostachys belangeriana* (E and F)

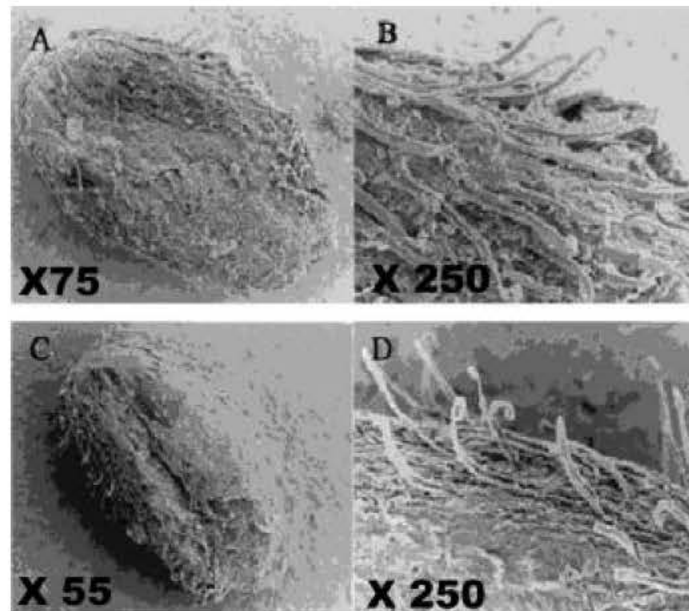


Fig. 5: Scanning electron micrograph of salicornieae seeds of Iran showing seed coat ornamentations of *Salicornia europaea* (A and B) and *S. persica* (C and D)

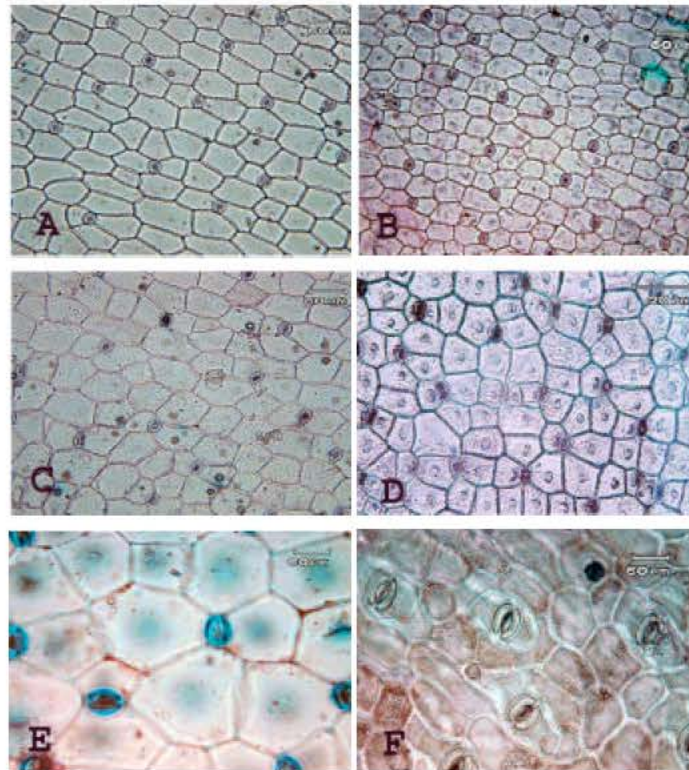


Fig. 6: Stomata distribution pattern in members of tribe salicornieae in Iran A and C; *Salicornia persica*, B; *Salicornia europaea*, D; *Halocnemum strobilaceum*, E; *Halostachys belangeriana*, F; *Arthrocnemum macrostachyum*

DISCUSSION

In this project macro and micro morphological characters were studied. The values of characters in separating taxa of this tribe in Iran were evaluated. Results show that characters as vegetative form and duration of life, arrangements of flowers in inflorescence and bract shape are diagnostic. Seed coat ornamentation is also important and all these can be used in preparing an efficient identification key for members of this tribe in Iran:

- 1 a. Annual herb, woody at base or not 2
b. Perennial herb, woody at base 4
- 2 a. Small plants, not branching, 5-10 cm height, Flower with linear arrangement in inflorescence, Seed coat vesiculate *Microcnemum coralloides*.
b. small herb to large bushes, branching, 8-50 cm height, Flowers with triangular arrangement are immersed in inflorescence, Seed coat with Hooked hairs. 3
- 3 a. Inflorescence branches 3-5, middle flower doesn't exceed the upper segment, Rectangular, Seed length 0.7-1.2 mm. *Salicornia europaea*.
b. Inflorescence branches 5-8, middle flower attached to the upper segment, with truncate end, pentagonal shape, Seed length 0.7-1.6 mm. *Salicornia persica*.
- 4 a. Small branching shrubs, Having opposite globular buds, yellow to green, Inflorescence without pedicel, with peltate bracts, Seed coat ornamentations granular *H. strobilaceum*.
b. Big branching shrubs, without opposite globular buds, glucose, Inflorescence pedicelate, without peltate bracts, Seed coat ornamentations is not granular 5
- 5 a. Bracts ob-pyramidal, not attached to each other, Flower arrangement linear, free, Stamens are not exerted, Seed coat almost smooth *H. belangeriana*
b. Bracts cup shape, attached to each other, Flower are immersed in inflorescence axis, free, Stamens are exerted, Seed coat tuberculate *A. macrostachyum*

Present study shows that there is a very low variation in populations of *Halocnemum strobilaceum* and *Halostachys belangeriana* in Iran where there is a very high variation in morphological features of populations of *Salicornia* especially *S. europaea*. It seems common and general methods of morphological studies are not capable of revealing this kind of variation, so it is

better to use molecular approaches for evaluating the genetic variation in this taxon for future studies.

Vegetative characters are not sufficient to separate these taxa so the necessity of using features of reproductive parts is distinct. Seed characters which are less affected by climate and habitat conditions provide very useful characters in this tribe.

Our results of studying seed coat ornamentations are in congruence with Shepherd *et al.* (2005) although we have not such high species richness in this tribe in Iran. It seems Seed coat features are more efficient in separation of Genus in this tribe. Results of studying pollen grains of tribe Salicornieae are in congruence with Akhani (2003), he believes pollen grains of *Salicornia persica* is larger than *S. europaea* due to the polyploidy in this species.

Due to the wide distribution of steppe region and salty soils in Iran and the wide range of salt tolerance of members of Salicornieae, it seems that they are good choices for desert improving and management and also as fodder supply. In traditional medicine of Iran, *Salicornia* has also an edible and officinal importance.

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