

Palynological study of the genus *Linum* in Iran (a taxonomic review)

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Abstract. *Linum* is the main genus of the Flax family widely distributed across the world with about 230 species. In the present study, pollen grains of 15 taxa from four sections of this genus were examined with scanning electron microscope and light microscope. The taxa primarily differed in polar and equatorial shape and their axis length, patterns of exine sculpturing and types of surface fine structure. Pollen shapes in the equatorial and polar view were circular, angular or elliptic. The applied ANOVA test for palynological characters showed a significant difference ($p < 0.001$) of some quantitative characters. Sculpture characters on the basis of their form, size, number, distribution, and fine structure showed variations at different taxonomic levels and were useful in the identification of species, subspecies and varieties in the genus. A significant positive/negative correlation was seen between some characters in the pollen grains. Palynological data, however, cannot show the species relationship in the sections; they are of taxonomic importance and may be used for differentiation of the species, subspecies and varieties.

Key words: *Linum*, micromorphology, pollen, SEM, taxonomy.

Introduction

The family *Linaceae* is composed of 22 genera (Vromans 2006) and approximately 300 species (Hickey 1988; Heywood 1993). The most important genus in the family is *Linum*, with about 230 species (Heywood 1993). The genus *Linum* is traditionally divided into five sections: *Linum*, *Linastrum*, *Cathartolinum*, *Dasylinum*, and *Syllinum* (Winkler 1931). About 22 *Linum* species and infraspecific taxa grow wild in Iran (Sharifnia & Assadi 2001) and are classified into these five sections (Rechinger 1974).

Palynology is the study of pollen grains produced by seed plants and the data provided by it are regarded as

important taxonomic criteria (Moore & al. 1991). Both morphological characteristics of the pollen and micromorphological characters of the pollen grains have been used in the classifications of plant species (Stace 1989).

Pollen morphology of the genus *Linum* was examined by several researchers but neither of these studies was thorough. There are no exhaustive reports on pollen morphology of the genus *Linum* including all its species (Erdtman 1952; Kuprianova & Alyoshina 1978; Nair & Sharma 1980; Moore & al. 1991). In a more detailed study carried out by Rogers & Xavier (1971), 25 to 30 species of the subgenus *Linum* were investigated. Four species, *L. stelleroides* Planch. from China, *L. hologynum* Rchb. from Southeast Europe,

and *L. marginale* Cunn. and *L. monogynum* Forst from Australia and New Zealand, have multiaperturate pollen. Of these, pollen morphology of *L. stelleroides* from China differs from the others and, along with the differences in habit and floral morphology, seems to indicate that the multiaperturate pollen may have arisen independently in that species. There is no evidence that would strongly support the idea that *L. stelleroides* is on an evolutionary route leading either to or beyond the other three species.

The present study concerns pollen micromorphology of 15 *Linum* taxa from four sections of this genus obtained with the help of scanning electron microscope (SEM) and light microscope (LM), in order to show the species relationships and whether such data can differentiate the *Linum* species of Iran.

Material and methods

Plant samples

Plant specimens of the 15 *Linum* species, subspecies and varieties were collected from natural populations in different regions of Iran in the spring of 2010-2011. Details of localities and voucher numbers are given in Table 1. The vouchers have been deposited in the Herbarium of the Shahid Beheshti University of Tehran, Iran (HSBU).

Palynological study

Pollen grains were obtained from mature buds of 15 *Linum* species and infraspecific taxa. For each taxon, three plant specimens were used and from each plant specimen at least three to four pollen grains were investigated and prepared for scanning electron microscopy and light microscopy, applying the prolonged acetolysis procedure of Erdtman (1960).

For LM, including size measurements, the pollen grains were mounted in glycerin jelly and sealed with paraffin. The polar (P) and equatorial (E) shape and length and P/E ratios were determined under the light microscope ($\times 1000$). Four replications were used for character measurements.

For scanning electron microscopy, the pollen grains were transferred directly to double-sided tape-affixed stubs and vacuum-coated with gold in Biorad E5200 auto sputter coater and examined and photographed with a Camscan MV2300 scanning electron microscope at 10 kV. The sculpturing types, dimensions and their fine structure, colpi dimensions, and apocolpium and mesocolpium length were studied. The terminology in this paper corresponds to that used by Moore & al. (1991).

Statistical analysis

For grouping of the studied species and varieties on the basis of the pollen characteristics, the obtained da-

Table 1. Palynologically studied taxa of the genus *Linum*.

Taxa	Locality	Collector	Voucher number
Sect. <i>Linum</i>			
<i>L. austriacum</i> L.	Markazi, Saveh to Hamadan, after Nobaran 1654m	Talebi	HSBU 2011102
<i>L. glaucum</i> Boiss. Nöe in Boiss.	Kurdistan, Sanandaj, Kani Mushkan, 1678m	Talebi	HSBU 2011155
<i>L. nervosum</i> Waldst & Kit. var. <i>bungei</i> (Boiss.) Sharifnia	Mazenderan, 90 km Karaj to Chalous, 2193 m	Talebi	HSBU 2011129
<i>L. nervosum</i> Waldst & Kit. var. <i>nervosum</i>	Mazenderan, 90 km Karaj to Chalous, 2193 m	Talebi	HSBU 2011130
<i>L. usitatissimum</i> L. var. <i>usitatissimum</i>	Markazi, Saveh to Salafchegan, Saleh Abad, 1320 m	Talebi	HSBU 2011165
<i>L. bienne</i> Miller	Guilan, Rasht, Saravan Park, 150 m	Talebi	HSBU 2011128
Sect. <i>Dasylinum</i> Planch.			
<i>L. densiflorum</i> P.H. Davis	West Azarbaijan, Urmia, Silvana 1648 m	Talebi	HSBU 2011194
Sect. <i>Syllinum</i> Griseb.			
<i>L. mucronatum</i> subsp. <i>armenum</i> (Bordzil.) P.H. Davis	Azerbaijan, Salmas, Ghoshchi, 1557 m	Talebi	HSBU 2011140
<i>L. mucronatum</i> subsp. <i>orientale</i> (Boiss.) P.H. Davis	Zanjan, 90 km Abhar to Zanjan, 1839	Talebi	HSBU 2011132
<i>L. mucronatum</i> Bertol. subsp. <i>mucronatum</i>	Hamedan, Avaj, 2350 m	Talebi	HSBU 2011196
<i>L. mucronatum</i> subsp. <i>assyriacum</i> P.H. Davis	Khuzestan, Izeh, Atabaki Park 350 m	Talebi	HSBU 2011164
<i>L. album</i> Ky. ex Boiss.	Kurdistan, 20 km Sanandaj to Kamyaran, 1329 m	Talebi	HSBU 2011114
Sect. <i>Linastrum</i> (Planch) H. Winkler			
<i>L. tenuifolium</i> L.	Kalibar, Arasbaran Protected Area, Vinagh 1200 m	Talebi	HSBU 2011197
<i>L. corymbulosum</i> Reichenb.	Guilan, Rodbar, Darestan jungle, 654 m	Talebi	HSBU 2011127
<i>L. strictum</i> L. var. <i>spicatum</i> Pers.	Khuzestan, Masjed Suleiman, Andika 535 m	Talebi	HSBU 2011193

ta were standardized (mean = 0, variance = 1) and used for multivariate analysis, including UPGMA (Unweighted Paired Group using Average method) and Principal Coordinate Analysis (PcoA) (Podani 2000).

One-way ANOVA and least-significant difference tests were applied to assess the significance of quantitative palynological difference between the studied species and Pearson's coefficient of correlation determined between quantitative palynological characters. NTSYS ver. 2 (1998) and SPSS ver. 9 (1998) software were used for statistical analyses.

Results

Palynological characters of the studied taxa (SEM and LM) are presented in Table 2. In all studied taxa the pollen shape in polar view was circular (except in *L. densiflorum* which was concave-triangular).

Equatorial view of pollen in some taxa, such as *L. austriacum*, *L. densiflorum* and *L. mucronatum* subsp. *orientale* was circular, while in the other it was el-

liptic (e.g. Fig. 1). The elliptic shapes showed various marginal forms, including acuminate-obtuse (in *L. strictum* var. *spicatum*, *L. mucronatum* subsp. *mucronatum*, *L. mucronatum* subsp. *armenum*, *L. album*, *L. nervosum* var. *nervosum*, and *L. nervosum* var. *bungei*), acuminate-acute (*L. corymbulosum*), emanate (*L. tenuifolium*, *L. mucronatum* subsp. *assyriacum* and *L. bienne*), and truncate (*L. glaucum* and *L. usitatissimum* var. *usitatissimum*).

Diameter of the equatorial axis in pollen grains varied from 35.70 μm in *L. mucronatum* subsp. *assyriacum* to 68.22 μm in *L. austriacum*, while size of the polar axis varied from 40.30 μm in *L. densiflorum* to 71.05 μm in *L. tenuifolium*.

With regard to aperture, the pollen grains in the *Linum* species were trizonocolpate, with three long and boat-shaped grooves with pointed ends. The colpi were situated in the equatorial zone of the pollen. Variations in size of the pollen colpi were observed in the *Linum* species. The smallest colpi was found in *L. mucronatum* subsp. *armenum*, 25.12 μm long, while the largest colpi (56.91 μm) was found in *L. austriacum*.

Table 2. Selected palynological characters of the studied *Linum* taxa, with combined SEM and LM. Sample values are given in parenthesis.

Species	Pollen shape (e)	Pollen shape (p)	Sculpture type	Sculpture dimensions (μm)	Equatorial length (μm)	Polar length (μm)	P/E (μm)	Colpus length (μm)	Apocolpium (μm)	Mesocolpium (μm)
<i>L. nervosum</i> var. <i>nervosum</i>	Elliptic-obtuse	Circular	Polygonal & circular gemmate-pilate	0.8×0.73	54.51	57.02	1.04	42.59	41.06	38.82
<i>L. nervosum</i> var. <i>bungei</i>	Elliptic-obtuse	Circular	Gemmate & baculate	1.19×0.85	52.83	54.31	1.03	44.57	45.72	40.57
<i>L. corymbulosum</i>	Elliptic-acute	Circular	Gemmate & baculate & pilate	1.09×1.07	43.17	65.60	1.51	55.32	23.64	33.70
<i>L. austriacum</i>	Circular	Circular	Gemmate	1.49×1.38	68.22	60.20	0.88	56.91	65.55	52.44
<i>L. album</i>	Elliptic-obtuse	Circular	Pilate	1.12×1.15	44.30	57.63	1.30	43.26	46.28	37.84
<i>L. glaucum</i>	Elliptic-truncate	Circular	Clavate	1.04×1.13	55.49	53.04	0.95	48.75	23.39	47.30
<i>L. bienne</i>	Elliptic-emanate	Circular	Small & large gemmate	0.81×0.66	44.44	42.97	0.97	30.90	32.75	29.95
<i>L. densiflorum</i>	Circular	Triangular-concave	Small & large gemmate	1.30×1.13	41.85	40.30	0.96	32.57	35.25	29.74
<i>L. usitatissimum</i> var. <i>usitatissimum</i>	Elliptic-truncate	Circular	Small & large gemmate	1.17×0.86	50.40	50.61	1.00	38.84	42.60	40.27
<i>L. strictum</i> var. <i>spicatum</i>	Elliptic-obtuse	Circular	Small & large gemmate	1.03×0.93	42.88	46.21	1.07	27.52	41.33	33.20
<i>L. tenuifolium</i>	Elliptic-emanate	Circular	Gemmate	1.38×1.34	55.10	71.05	1.28	55.27	42.27	40.56
<i>L. mucronatum</i> subsp. <i>mucronatum</i>	Elliptic-obtuse	Circular	Small & large gemmate	1.45×1.32	42.95	46.40	1.08	40.56	39.49	29.94
<i>L. mucronatum</i> subsp. <i>assyriacum</i>	Elliptic-emanate	Circular	Baculate-pilate	1.34×1.28	35.70	64.5	1.80	52.34	13.13	26.34
<i>L. mucronatum</i> subsp. <i>orientale</i>	Circular	Circular	Gemmate	1.22×1.24	51.90	46.52	0.89	39.44	41.62	34.69
<i>L. mucronatum</i> subsp. <i>armenum</i>	Elliptic-obtuse	Circular	Gemmate	1.09×1.04	43.53	47.55	1.09	25.12	24.35	22.05

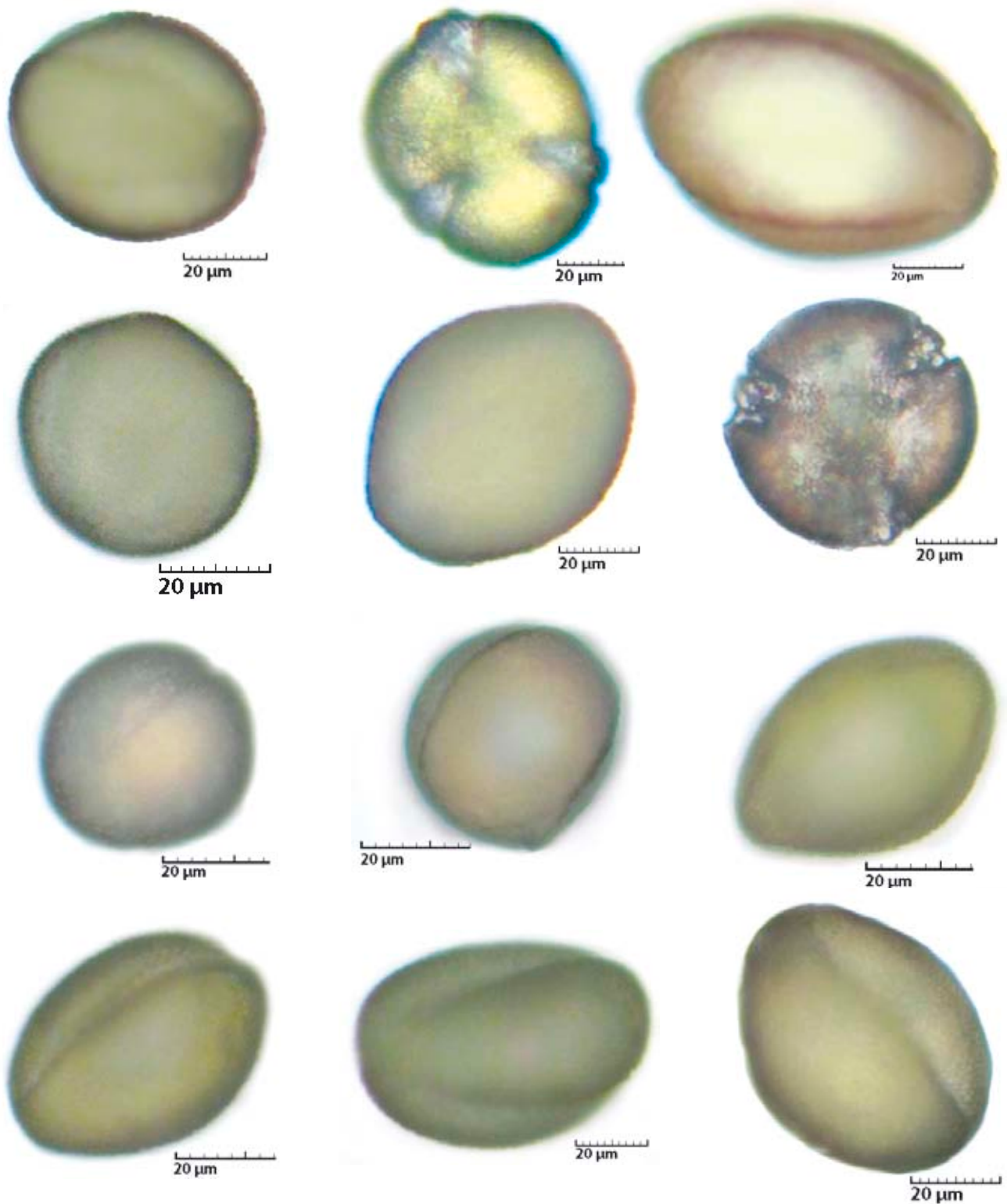


Fig. 1. Pollen morphology of the studied taxa of genus *Linum*.

A= *L. mucronatum* subsp. *armenum*, B= *L. mucronatum* subsp. *orientale*, C= *L. mucronatum* subsp. *mucronatum*, D= *L. austriacum*, E= *L. glaucum*, F= *L. usitatissimum* var. *usitatissimum*, G= *L. nervosum* var. *nervosum*, H= *L. nervosum* var. *bungei*, I= *L. corymbulosum*, J= *L. bienne*, K= *L. strictum* var. *spicatum*, L= *L. album*.

The ANOVA test for palynological characters applied to the 15 *Linum* taxa showed significant differences ($p < 0.001$) in some characters (e.g. Table 3), namely, in the size of sculpture length and width, as well as in the polar and equatorial axis length (see Fig. 2).

The exine sculpturing types in the studied species were gemmate, pilate, baculate, or clavate. A gemmate type, with sculpturing elements longer than $1\ \mu\text{m}$ and contracted at the base, was observed in *L. mucronatum* subsp. *mucronatum*, *L. mucronatum* subsp. *orientale*, *L. tenuifolium*, *L. austriacum*, *L. corymbulosum*, *L. usitatissimum* var. *usitatissimum*, *L. nervosum* var. *nervosum*, *L. nervosum* var. *bungei*, *L. mucronatum* subsp. *armenum*, *L. bienne*, and *L. strictum* var. *spicatum*.

A pilate type, with elements showing swollen or knob-like heads higher than $1\ \mu\text{m}$ occurred in *L. album*, while in *L. mucronatum* subsp. *armenum* two

forms of large size and small size occurred. A uniform size was observed in *L. nervosum* var. *nervosum* and *L. corymbulosum*.

A baculate type with rod-shaped sculpturing elements higher than $1\ \mu\text{m}$ was observed in *L. corymbulosum* and *L. nervosum* var. *bungei*, while a clavate type with club-shaped sculpturing elements higher than $1\ \mu\text{m}$ occurred in *L. glaucum*.

The pollen grains were echinated or granulated at the surface of sculpturing. The number and decoration of these fine structures varied in the studied taxa. They occurred as a central large-granulated fine process in *L. bienne*, and as echinated fine structures in *L. mucronatum* subsp. *mucronatum* and *L. usitatissimum* var. *usitatissimum*.

Granulated processes arranged in a ring structure and without a central process were observed in *L. austriacum*, *L. album* and *L. strictum* var. *spicatum* (e.g. Fig. 3).

Table 3. ANOVA test of the quantitative palynological characters.

Character	Similarity	Sum of Squares	df	Mean Square	F	Sig.
Sculpture length	Between groups	3.046	14	0.218	1.851	0.077
	Within groups	3.526	30	0.118		
	Total	6.572	44			
Sculpture width	Between groups	2.636	14	0.188	1.344	0.241
	Within groups	4.204	30	0.140		
	Total	6.840	44			
Equatorial length	Between groups	3779.701	14	269.979	5.510	0.001
	Within groups	1469.984	30	48.999		
	Total	5249.685	44			
Polar length	Between groups	3545.555	14	253.254	16.738	0.001
	Within groups	453.914	30	15.130		
	Total	3999.469	44			
P/E	Between groups	3.815	14	0.273	14.558	0.010
	Within groups	.562	30	1.872E-02		
	Total	4.377	44			
Colpus length	Between groups	3452.472	14	246.605	6.009	0.001
	Within groups	1231.175	30	41.039		
	Total	4683.647	44			
Sculpture distance	Between groups	3.879	14	0.277	3.870	0.001
	Within groups	2.148	30	7.159E-02		
	Total	6.027	44			
Colpus width	Between groups	624.315	14	44.594	15.475	0.010
	Within groups	86.450	30	2.882		
	Total	710.765	44			
Apocolpium size	Between groups	6921.159	14	494.369	4.061	0.010
	Within groups	3652.474	30	121.749		
	Total	10573.633	44			
Mesocolpium size	Between groups	2406.177	14	171.870	15.416	0.001
	Within groups	334.455	30	11.149		
	Total	2740.632	44			

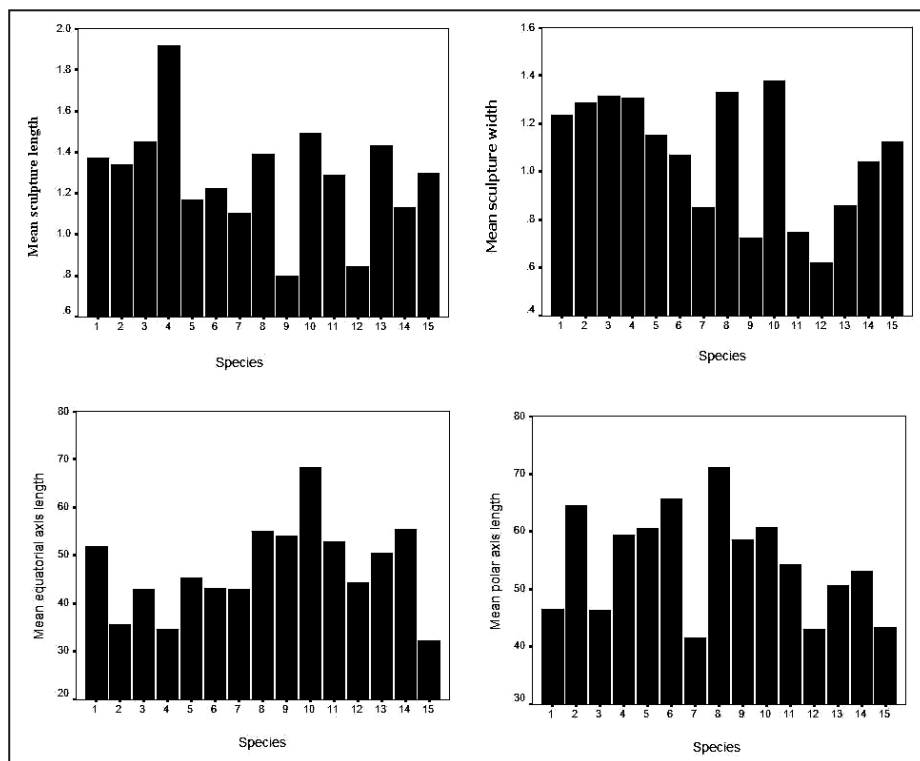


Fig. 2. Diagram of the ANOVA test applied to the studied taxa on the basis of palynological characters.

Abbreviations: 1= *L. mucronatum* subsp. *orientale*, 2= *L. mucronatum* subsp. *assyriacum*, 3= *L. mucronatum* subsp. *mucronatum*, 4= *L. mucronatum* subsp. *armenum*, 5= *L. album*, 6= *L. corymbulosum*, 7= *L. strictum* var. *spicatum*, 8= *L. tenuifolium*, 9= *L. nervosum* var. *nervosum*, 10= *L. austriacum*, 11= *L. nervosum* var. *bungei*, 12= *L. bienne*, 13= *L. usitatissimum* var. *usitatissimum*, 14= *L. glaucum*, 15= *L. densiflorum*.

The UPGMA dendrogram and the PcoA plot of palynological characters (Figs 4, 5) did not group together the four *L. mucronatum* subspecies and the two varieties of *L. nervosum*, which morphologically are very similar. Therefore, although these data cannot show the species relationships in the sections, they are of taxonomic importance and may be used for differentiation of the species, subspecies and varieties.

Significant correlations, positive/negative, were seen between some characters in the pollen grains. For example, there was a positive significant correlation ($r=0.35$, $p<0.05$) between the polar/equatorial (P/E) ratio with colpus length, while the P/E ratio had a negative significant correlation ($r=-0.34$, $p<0.05$) with colpus width. Positive significant correlations ($p<0.01$) occurred between the polar axis length and colpus length ($r=0.69$) and also with sculpture distance ($r=0.41$). Similarly, positive significant correlations ($p<0.01$) occurred between the equatorial axis length and colpus width ($r=0.42$) with apocolpium ($r=0.46$) and also with mesocolpium ($r=0.71$). Therefore, the changes occurring in these pollen characteristics during diversification of species seem to have been programmed in the process of joint evolving of characters. Usually, such changes are adaptive in nature which is yet to be determined in the studied taxa.

Discussion

On the basis of the shape and number of apertures on pollen surface, the plant species were divided into two main groups: basal angiosperms and eudicots. This pattern was partially confirmed with DNA sequencing and molecular phylogenetic reconstruction, using initially the chloroplast gene *rbcL*, followed by combined multi-gene analyses (Chase & al. 1993; Soltis & al. 2003).

In all studied taxa of the genus *Linum*, pollen grains were trizonocolpate, with three long grooves in the equatorial zone, and with a monomorphic or polymorphic process on their surfaces. Pollen apertures are not placed randomly on the surface of the pollen grain, but usually have a very definite placement with respect to the grain pole and equator, defined by its position in the pollen tetrad (Harley & Baker 2001).

Phylogenetically, palynologists have focused their attention on the aperture (Kuprianova 1948; Nair 1974). The pollen grains of approximately 140 species of the genus *Linum* have been examined. Most had tricolpate pollen, but nineteen species have been found to have multi-aperturate pollen. These were distributed among three of the five sections (namely *Linum*, *Linastrum* and *Syllinum*) usually recognized in the genus, and almost certainly were the result of three independent evolutionary

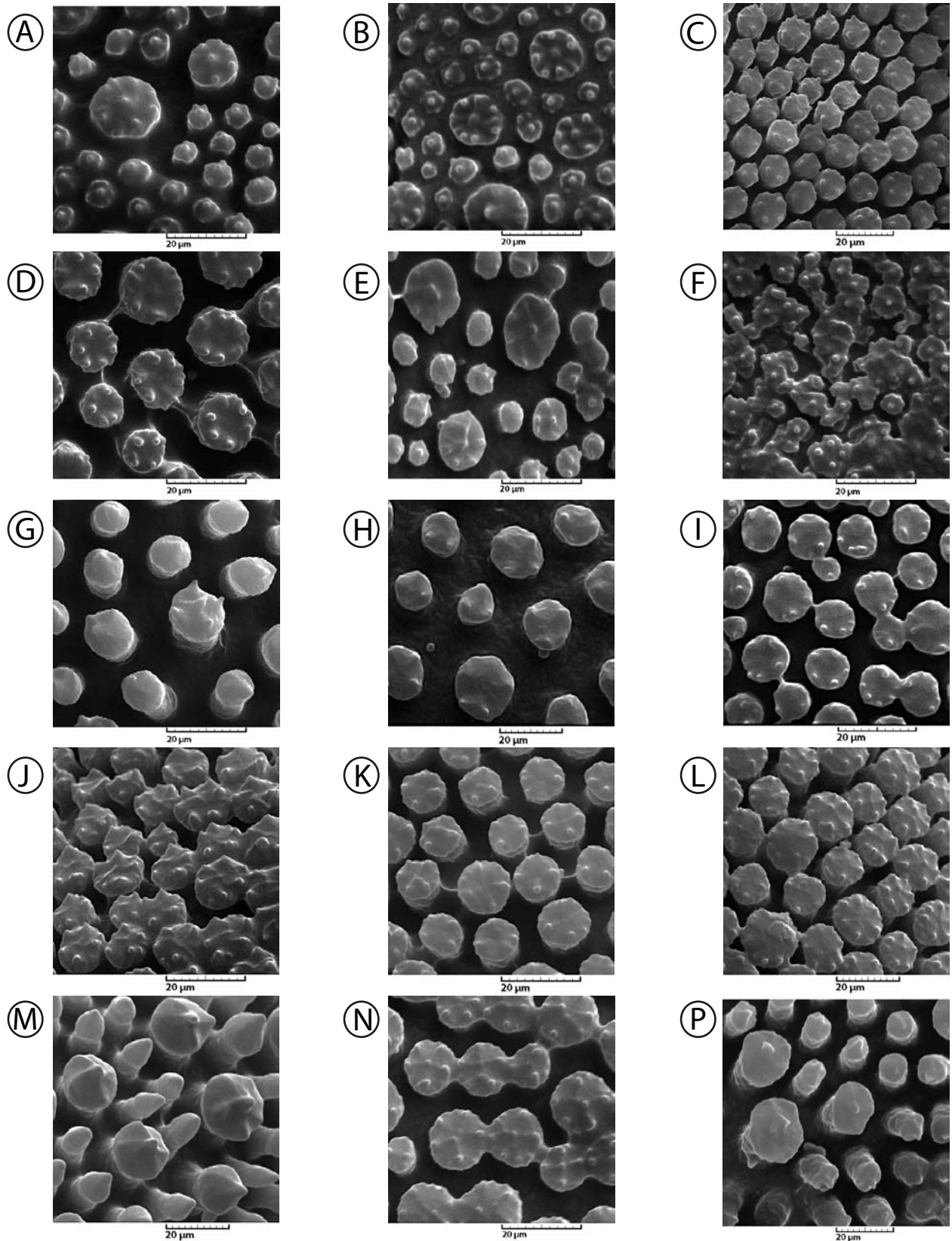


Fig. 3. Electronic micrograph of pollen surface exine sculpturing.

A= *L. nervosum* var. *bungei*, B= *L. nervosum* var. *nervosum*, C= *L. glaucum*, D= *L. austriacum*, E= *L. usitatissimum* var. *usitatissimum*, F= *L. bienne*, G= *L. corymbulosum*, H= *L. tenuifolium*, I= *L. strictum* var. *spicatum*, J= *L. densiflorum*, K= *L. album*, L= *L. mucronatum* subsp. *mucronatum*, M= *L. mucronatum* subsp. *armenum*, N= *L. mucronatum* subsp. *orientale*, P= *L. mucronatum* subsp. *assyriacum*.

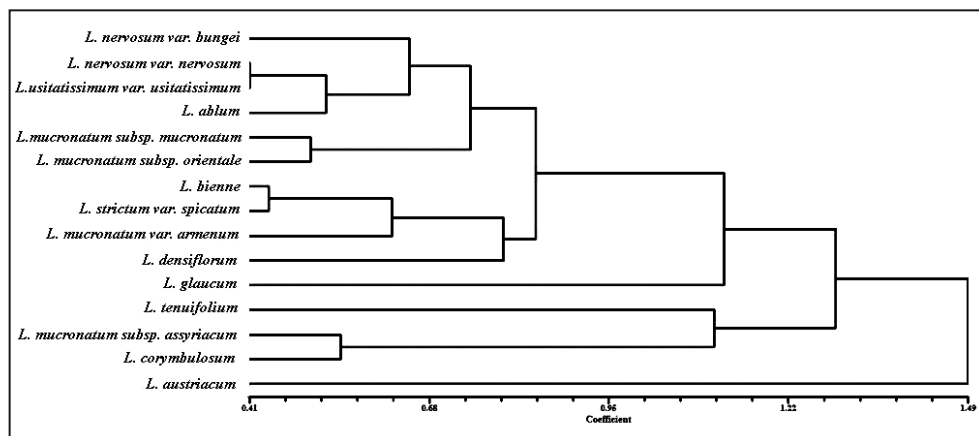


Fig. 4. UPGMA tree based on palynological characters.

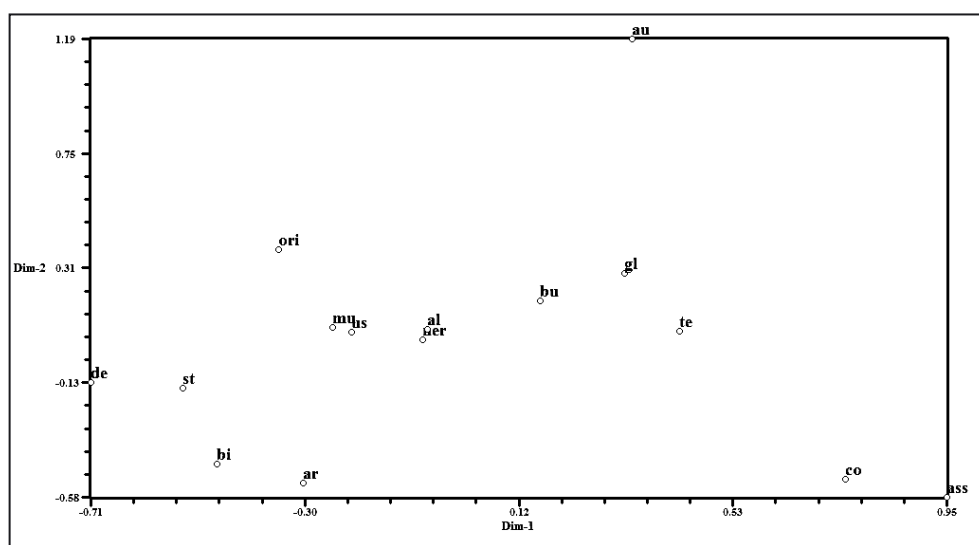


Fig. 5. PcoA plot of the studied taxa on the basis of palynological characters.

Abbreviations: **bu**= *L. nervosum* var. *bungei*, **ner**= *L. nervosum* var. *nervosum*, **gl**= *L. glaucum*, **au**= *L. austriacum*, **us**= *L. usitatissimum* var. *usitatissimum*, **bi**= *L. bienne*, **co**= *L. corymbulosum*, **te**= *L. tenuifolium*, **st**= *L. strictum* var. *spicatum*, **de**= *L. densiflorum*, **al**= *L. album*, **mu**= *L. mucronatum* subsp. *mucronatum*, **ar**= *L. mucronatum* subsp. *armenum*, **ori**= *L. mucronatum* subsp. *orientale*, **ass**= *L. mucronatum* subsp. *assyriacum*.

routes. There is evidence that the multiaperturate condition may have evolved twice in each of two of these subgenera. It is possible that species with multiaperturate pollen may have evolved from those with triaperturate pollen on as many as five different occasions in this genus (Rogers & Xavier 1972).

On the basis of the pollen shape, the angiosperms pollen has been grouped under several classes by the P/E ratio, such as oblate, oblate-spheroidal, spheroidal, and elliptic (Walker & Doyle 1975).

The polar view of pollen grains in all studied taxa was similar, except for the species *L. densiflorum*. However, the equatorial view of the pollen grain was different and varied between and among the studied sections, appearing as circular or elliptic. Elliptic shape was found in three different forms.

The evolutionary trends affecting pollen morphology can be described. They include increase in grain size, shortening of the colpi, increase in the number of apertures, and increase in the process of polymor-

phism. Basic or primitive pollen grains in the species of all five sections of the genus *Linum* are comparatively small (*ca.* 50 μ m) and have three elongated colpi with few processes on their surfaces; they have thin exine (*ca.* 2.0 μ m) and truncate monomorphic or moderately polymorphic processes. The total variation of these evolutionary developments has resulted in pollen grains up to 125 μ m in diameter, with exine up to 9.0 μ m thick, with up to 24 or more apertures and a distinct process of polymorphism (Xavier & al. 1980).

In order to measure the pollen size, the polar and equatorial axis of the pollen grains was measured. On the basis of these characters, the smallest and biggest pollen grains were found in *L. densiflorum* and *L. austriacum*, respectively. The *Linum* pollen is rather large, when compared to most angiosperms (Xavier & al. 1980).

On the basis of several taxonomical reports on this genus in Iran (Parsa 1951; Rechinger 1974; Sharifnia &

Assadi 2001) and on samples collected by the authors (Talebi & al. 2012), it was found out that the species *L. austriacum* is widely distributed in different regions, while *L. densiflorum* is restricted to small regions in the northwest of Iran. Apparently, in addition to ecological parameters, oval fertilization and seed production turned out to be important factors necessary for the distribution and succession of each species. Correlation of pollen size and pollen tube growth rate has been positive (Gore & al. 1990; Manicacci & Barrett 1995). Varis (2009) has found that in *Pinus sylvestris* germinated pollen grains were larger in size than non-germinated. Thus the size of pollen grains is considered to be an indication of pollen viability and the proportion of large grains has been used to estimate pollen performance (Dufay & al. 2008). For example, in the species *Erythronium grandiflorum* Pursh., plants producing larger pollen grains sired more seeds than plants with smaller pollen grains (Cruzan 1990).

Pollen size was found to be an index to chromosome numerical variations (aneuploidy and polyploidy) and has proved useful in cytopalynological studies (Saraswathyamma & al. 1995). In *Vinca rosea*, size variation of pollen grains has been reported between diploid and induced tetraploid plants (Dnyansgar & Sudhakaran 1972).

In the present study, different types of sculpturing were seen on the surface of pollen exine. The exine surface patterns often serve as supplementary factors to the apertural form in reaching the taxonomic and phylogenetic conclusions (Erdtman 1952; Nair 1974). Pollen surface decoration was in a clavate, pilate and gemmate to baculate form, which were seen individually or in combination with the surface types mentioned with various sizes and varying intervals on the outer surface of the exine.

Numerous infrageneric classifications of *Linum* have been published. In 1837 Reichenbach published the first theory of a natural system for grouping the species of this genus in sections (Diederichsen & Richards 2003). Winkler (1931) provided the basis for the five sections of *Linum*. This system is usually recognized in most regional floristic and taxonomic works. Yuzepchuk (1949) distinguished nine sections with further taxonomic subdivisions in the genus.

The systematic treatment of the genus is still in the process of discussion, while pollen morphology of the genus could not confirm any of the earlier proposed arrangement of the species in sections (Grig-

oryeva 1988). Taxonomy and classification of the genus *Linum* have changed with time. Many researchers had classified the *Linum* species either on the basis of morphological characters, or place of origin (Linnaeus 1857; De Candolle 1904). Others taxonomists grouped the *Linum* species on the basis of their chromosome numbers (Kikuchi 1929; Chennaveeraiah & Joshi 1983; Gill 1987). The amplified fragment length polymorphism (AFLP) based on phylogeny of 17 species of *Linum* was not compatible with the traditional sections (e.g. Winkler 1931) of the species (Vromans 2006). Karyotype numbers did not reflect the phylogenetic relationships among the *Linum* species. For example, an analysis based on RAPD data indicated that *L. decumbens* ($2n=30$) was clustered with *L. grandiflorum* ($2n=16$), but not with the other 75 species with which it shares the same chromosome number (e.g. *L. angustifolium* and *L. usitatissimum*; Fu & al. 2002)

The studied taxa have shown a distinct range of palynological variations as they were differentiated from each other in the UPGMA tree and PcoA plot. The result of this study does not confirm the species classification in sections according to *Flora Iranica* (Rechinger 1974), which was based on morphological characters. For example, in this study were investigated the pollen grains of six taxa of the section *Linum*, namely *L. usitatissimum* var. *usitatissimum*, *L. glaucum*, *L. bienne*, *L. austriacum*, *L. nervosum* var. *nervosum*, *L. nervosum* var. *bungei*, but in the UPGMA tree or PcoA plot these taxa were quite distanced and had strongly varying palynological data.

The range of variations in phenotypic characters used to distinguish the species often overlaps within the section *Linum* (Diederichsen & Richards 2003). To resolve this problem, Ockendon & Walters (1968) proposed to lump the species into groups, e.g. the *Linum perenne* group, and to declare as preliminary the exact determination of the species until a revision of the genus was completed. The *Linum perenne* group consists of *L. leonii*, *L. austriacum* and five subspecies of *L. perenne*. In addition to the chromosome number, heterostyly, ecology, and breeding system, the pollen characters are taxonomically more reliable than the quantitative morphological characters in this group (Ockendon 1971). In the UPGMA tree or PcoA plot, the species *L. austriacum* is differentiated from the other members of section *Linum* and also from other species. The results of this study have confirmed the Ockendon (1971) classification of the *Linum perenne* group.

Talebi & al. (2012) have examined the seed micromorphology of eleven taxa of the genus *Linum*. The results of the study have shown that section *Linum* is very heterogeneous in seed traits and thus an anisotropic group was organized whose members differed in seed characters. Thus some species were different in seed shape, color, size and types of surface sculpturing from other members and so formed a sister group for the other species in this section.

Variations in leaf and stem anatomy show that the section *Linum* is a heterogeneous group (Sharifnia & Albouyeh 2002), while seed coat anatomy, and pollen and leaf ultrastructure have supported this idea (Moroz & Tsymbalyuk 2005; Optasyuk 2006; Svetlova & Yakovleva 2006). Some botanists divide the section *Linum* into two independent sections including such species as: *L. usitatissimum* and *L. bienne* ($2n=30$) and *L. grandiflorum* ($2n=16$) and *Adenolinum* for example *L. austriacum*, and *L. perenne* and *L. leonii* ($2n=18$) (Yuzepchuk 1949; Egorova 1996).

The same is valid for the section *Linastrum*, in which the species *L. tenuifolium*, *L. corymbulosum* and *L. strictum* var. *spicatum* do not club together in the palynological study.

Linum mucronatum was reported as a very variable species (Ozcan & Zorlu 2009) and the palynological study has confirmed this observation. Four subspecies of *Linum mucronatum* are strongly distanced in the UPGMA tree. Besides in pollen characters, the four subspecies of *L. mucronatum* have differed from each other in morphological features. For example, there were differences between *L. mucronatum* subsp. *armenum* and *L. mucronatum* subsp. *orientale* in some key characteristics of leaf and stem morphology, petal colors, habitat, and features of inflorescence. A seed micromorphological study has confirmed the differentiation of these subspecies and it was also possible to delineate both subspecies from this section by their seed cell shape and the wall structures in SEM (Ozcan & Zorlu 2009).

L. mucronatum subsp. *assyriacum* differed from *L. mucronatum* subsp. *mucronatum* in some morphological characters of the basal leaf shape and form, stem branching and phytogeographical distribution (Sharifnia & Assadi 2001).

In the flora of Turkey, *Linum mucronatum* subsp. *gypsicola* var. *papilliferum* was referred to the species *L. pamphylicum* subsp. *papilliferum* (Hub.-Mor. & Reese) Yilmaz & Kaynak comb. & stat. nov. on the ba-

sis of some morphological features of flowering stems, basal and cauline leaves (Yilmaz & Kaynak 2006).

The data of our palynological study do not support the species classification in traditional sections, but support the assumption that the *Linum perenne* group is in section *Linum*. Furthermore, palynological characters are taxonomically important and useful in identification and distinction of species and infraspecific taxa in this genus, especially of the two varieties of *L. nervosum* and also of the species *L. austriacum* from *L. glaucum* which are morphologically very similar.

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