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RESEARCH ARTICLE

Seed morphology of *Silene commelinifolia* Boiss. complex (Caryophyllaceae Juss.)

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Abstract

Silene (Caryophyllaceae) is a large genus with about 700 species distributed worldwide. *Auriculatae* is the largest section of this genus with 21 endemic species represented in Iran. Seed micromorphology of *S. commelinifolia, S. lucida, S. nurensis* and *S. eremicana* from 18 populations was studied by the light microscopy. As a result, 6 populations were adopted for further SEM studies. A great variation was observed in seed shape and size, lateral and peripheral surfaces of seeds, and shape of testa cells edge. Totally, 16 quantitative and qualitative seed features were measured and evaluated. Seed size varied from 1.28 × 1.43 mm in Alvand population of *S. eremicana*, to 1.97 × 1.50 mm in Bozghosh population of *S. lucida*. PAST and SPSS software was applied to demonstrate the species relationships. *S. eremicana* and *S. lucida* were closely grouped, which is in accordance with their morphological similarities. Seed micromorphology revealed that the species, which are similar to *S. commelinifolia* are clearly separated from each other. Therefore seed morphology is of taxonomic importance in the studied group.

Keywords: *Silene*, seed, micromorphology, SEM, Iran

Introduction

Silene L. (Caryophyllaceae Juss.) is a large genus with worldwide distribution, containing about 700 species. These species are mainly hermaphrodite, although a few species are dioecious or gynodioecious (Bari 1973; Greuter 1995). These are annual, biennial or perennial herbs. The section *Auriculatae* (Boiss.) Schischkin is the largest within this

Table 1. Voucher details of studied *Silene* taxa.

Nr	Taxon	Locality	Code	SEM
1	*S. commelinifolia* var. *ovatifolia*	West Azerbaijan, Piranshahr to Naghadeh, Gerd Kashaneh, Lik Bin Village, Landi Sheykhan Mountain, 36 41 7.5 N, 45 26 27.1 E, 2400 m, 02.07.2010, A. Gholipour, 890277		
2	*S. commelinifolia* var. *ovatifolia*	Tehran, Darakeh mountain, 35 49 37.3 N, 51 22 47.3 E, 1925 m, 20.06.2008, A. Gholipour, 8768	Com12	*
3	*S. commelinifolia* var. *ovatifolia*	West Azerbaijan, Urmia, Anhar, Marmisho, Solok, 37 29 0.33 N, 44 45 0.22 E, 2327 m, 20.07.2011, A. Gholipour, 900832		
4	*S. commelinifolia* var. *commelinifolia*	Mazandaran, Baladeh, Kamarbon, Gosfandsarai-e chai khaksar, 36 14 16.1 N, 51 22 17.1E, 2852 m, 07.07.2011, A. Gholipour, 900624		
5	*S. commelinifolia* var. *commelinifolia*	Tehran, Haraz Road, Polur, 35 48 899 N, 52 01 643 E, 2405 m, 10.06.2007, A. Gholipour, 8637		
6	*S. commelinifolia* var. *commelinifolia*	West Azerbaijan, Urmia, Anhar, Marmisho, 37 29 03.2 N, 44 36 24.7 E, 3007 m, 02.07.2012, A. Gholipour, 91312		
7	*S. commelinifolia* var. *commelinifolia*	Hamadan, Alisadr cave, 29.06.2010, A. Gholipour		
8	*S. commelinifolia* var. *commelinifolia*	Tehran, Touchal, 35 52 572 N, 51 24 131 E, 2700 m, 23.06.2008, A. Gholipour, 8771	Com9	*
9	*S. commelinifolia* var. *commelinifolia*	Tehran, Dizin, Gajerah, Velayatroud village, 36 03 N, 51 23 E, 2500 m, 15.07.2008, A. Gholipour		
10	*S.* cf. *commelinifolia*	Ardabil, km 30 Ardabil to Kivi, before Neor lake, 38 00 549 N, 48 55 225 E, 2590 m, 16.07.2011, A. Gholipour, 900701		
11	*S.* cf. *commelinifolia*	Ardabil, Neor lake, 09.08.2008, A. Gholipour		
12	*S.* cf. *commelinifolia*	East Azerbaijan, Sarab, Shalgoon Village, Bozqush Mountain, 37 45 54 N, 47 35 31 E, 2650–3000 m, 08.07.2012, A. Gholipour, 91387	Com3	*
13	*S. eremicana*	Hamadan, Alvand Mountain, Ganjnameh, 34 43 475 N, 48 25 039 E, 2800 m, 26.06.2007, A. Gholipour, 86105	ere	*
14	*S. lucida*	East Azerbaijan, Sarab, Shalgoon village, Bozqush Mountain, 37 45 54 N, 47 35 31 E, 2650–3000 m, 08.07.2012, A. Gholipour	Luc2	*
15	*S. lucida*	Gilan, Kelachay, Rahim Abad, Eshkevarat, Chakol, Boza kuh, 2800–3100 m, 29.06.2007, A. Gholipour, 86139		
16	*S. lucida*	Piranshahr to Naghadeh, km 5, Zarkanan and Silveh villages, Kuh-e Sepiarez, 36 50 59.9 N, 44 58 24.8 E, 2820 m, 01.07.2010, A. Gholipour, 890257		
17	*S. nurensis*	Chaharmahal and Bakhtiari, Farsan, Kuhrang, Zardkuh, 32 18 704 N, 50 08 574 E, 3300–3400 m, 29.07.2008, A. Gholipour, 8782	Nur1	*
18	*S.* cf. *commelinifolia*	Lorestan, Azna, Daretakht, Oshtorankuh, 33 20 522 N, 49 20 427 E, 2535 m, 09.08.2008, A. Gholipour, 8787		

genus, representing 21 endemic species in Iran (Melzheimer 1980).

According to Bittrich (1993), seeds of Caryophyllaceae are small or very small (0.4–3 mm long), black, brown or nearly white, reniform, pyriform, or orbicular, and mostly laterally compressed. Different seed morphological studies had revealed the taxonomic importance of seed characters in *Silene* (Yildiz & Cirpici 1998; Hong *et al.* 1999; Fawzi *et al.* 2010). Chowdhuri (1957) found that seeds morphology in *Silene* is of

Table 2. Qualitative and quantitative characteristics of seed of taxa studied of *Silene*.

Nr	Character	States / Units
1	seed shape	round-reniform (0), symmetrical-reniform (1), cordate-reniform (2), asymmetrical-reniform (3), semi-circle reniform (4)
2	peripheral surface of seed	flat (0), concave (1), convex (2)
3	lateral surface of seed	flat (0), concave (1), convex (2)
4	testa cells edge	v-shaped (0), undulate (1), smooth (2), sinuate (3)
5	shape of peripheral cells	elongated polygonal (0), polygonal (1)
6	trichomes at hillum	determinate (0), indeterminate (1)
7	seed Length	mm
8	seed Width	mm
9	seed length to width ratio	
10	testa cell length	µm
11	testa cell width	µm
12	testa cell length to width ratio	
13	width of peripheral surface	mm
14	hilum region length	µm
15	hilum region width	µm
16	hilum length to with ratio	

taxonomic importance on the section level. Melzheimer (1987) considered testa cells shape studied by SEM for *Silene* species separation. Seed morphology of some *Silene* species in Pakistan was studied by Ghazanfar (1983). Numerical analysis of seed features by Zareh (2005) showed that these features are more diagnostic whenever used with a set of macromorphological data. Camelia (2011) studied some *Silene* and recorded that most seeds are uniform but testa cells shape is informative.

Seed morphological studies in this genus can provide additional taxonomic information to help taxa distinguishing. This is the first seed morphological studies for *S. commelinifolia* Boiss., *S. lucida* Chowdh., *S. nurensis* Boiss. et Hausskn., and *S. eremicana* Stapf.

Material and methods

Eighteen populations of 4 *Silene* (namely *S. commelinifolia, S. lucida, S. nurensis,* and *S. eremicana*) growing in Iran were studied at the beginning, and then seeds from 6 populations were used for further SEM and statistic analyses (Tab. 1). The specimens were collected from nature, and later the vouchers were deposited at the Herbarium of Shahid Beheshti University (HSBU) and Payame Nour University (PNUSH).

Beforehand, the samples were studied by a Dino-Lite Pro hand stereomicroscope. For SEM studies, selected samples were directly transferred by fine pipette to a metallic stub using double-sided adhesive tape, and then coated with gold in a sputtering chamber (Sputter Coater BALTEC, SCDOOS). The coating was restricted to 100 Å. The SEM examination was carried out on EM32000 KV25 microscope. The measurements were calculated on the base of 10–20 readings from each specimen by use of ImageTool ver. 3.

In general, 16 qualitative and quantitative seed characteristics were analyzed (Tab. 2). To reveal species relationships we applied cluster analysis, principal components analysis (PCA) and principal coordinate analysis (PCO) plotting. For multivariate analysis, the mean

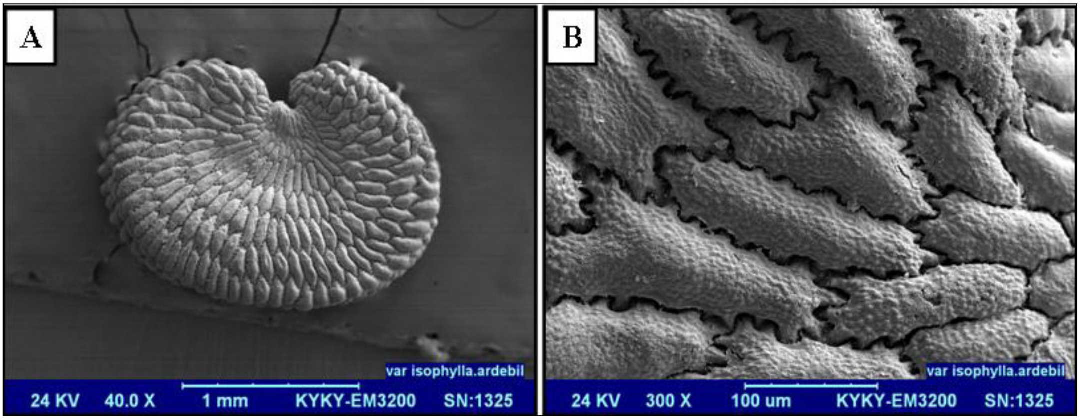

Fig. 1. SEM micrographs of seed surface in *Silene* cf. *commelinifolia* (Bozqush population): **A** – lateral surface; **B** – testa cells shape.

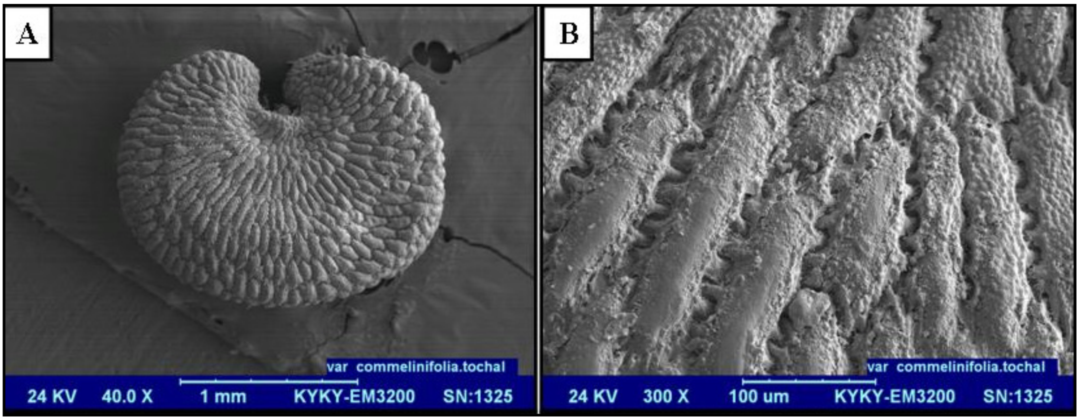

Fig. 2. SEM micrographs of seed surface in *Silene commelinifolia* var. *commelinifolia* (Touchal population): **A** – lateral surface; **B** – testa cells shape.

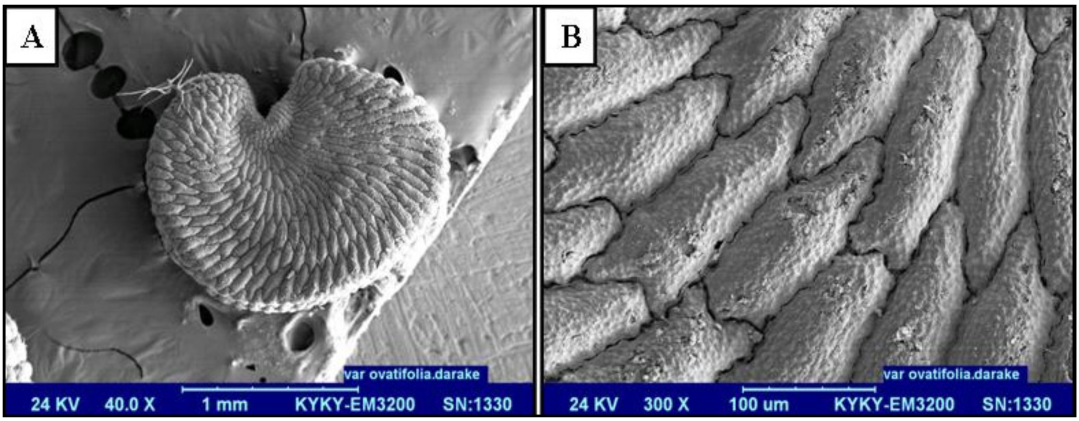

Fig. 3. SEM micrographs of seed surface in *Silene commelinifolia* var. *ovatifolia* (Darake population): **A** – lateral surface; **B** – testa cells shape.

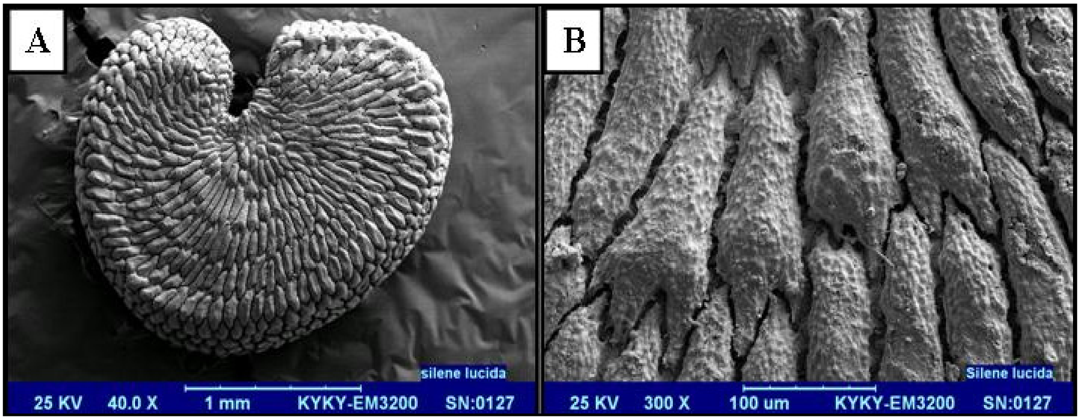

Fig. 4. SEM micrographs of seed surface in *Silene lucida* (Bozqush population): **A** – lateral surface; **B** – testa cells shape.

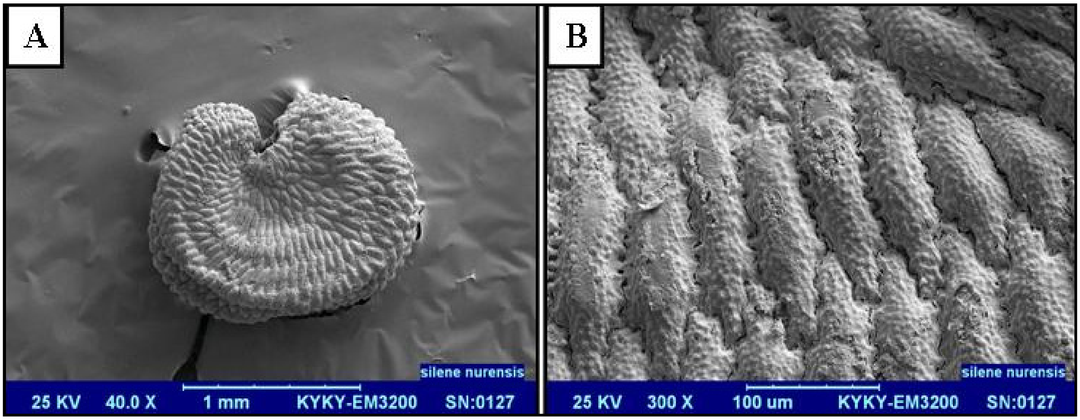

Fig. 5. SEM micrographs of seed surface in *Silene nurensis* (Zardkuh population): **A** – lateral surface; **B** – testa cells shape.

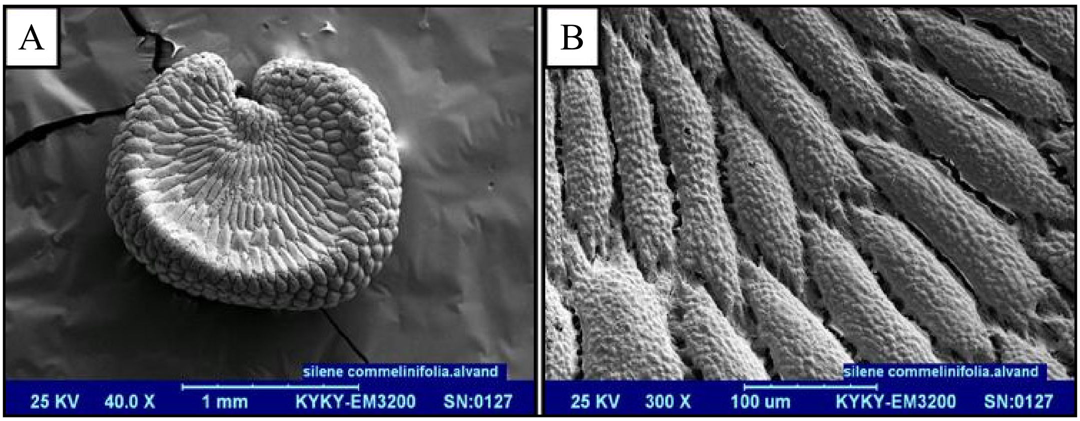

Fig. 6. SEM micrographs of seed surface in *Silene eremicana* (Alvand population): **A** – lateral surface; **B** – testa cells shape.

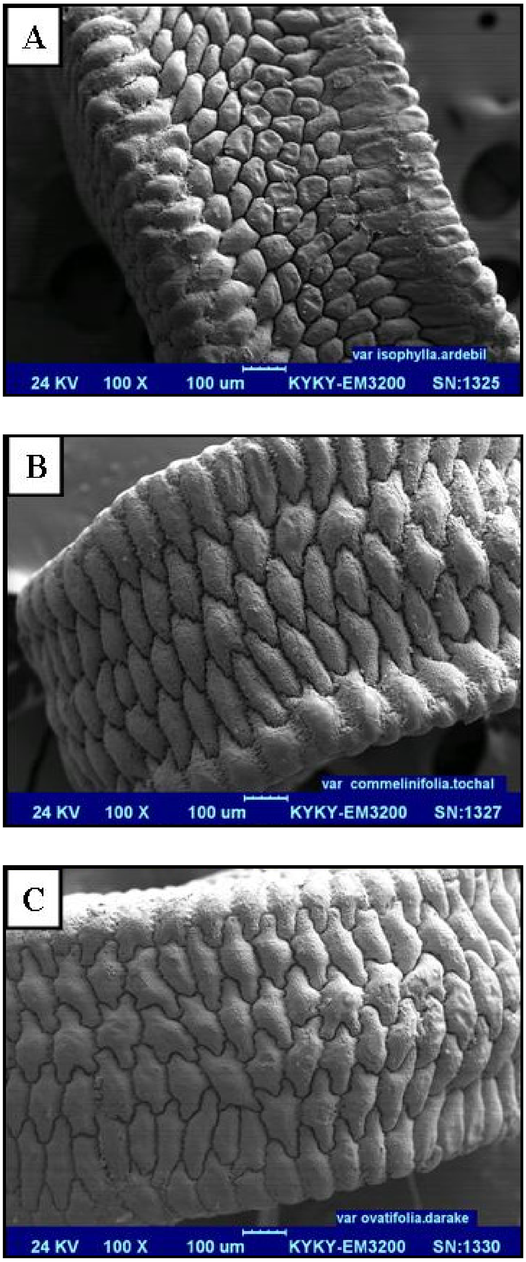

Fig. 7. SEM micrographs of seed surface in peripheral view: **A** – Bozqush population of *Silene* cf. *commelinifolia*; **B** – Touchal population of *S. commelinifolia* var. *commelinifolia*; **C** – Darake population of *S. commelinifolia* var. *ovatifolia*.

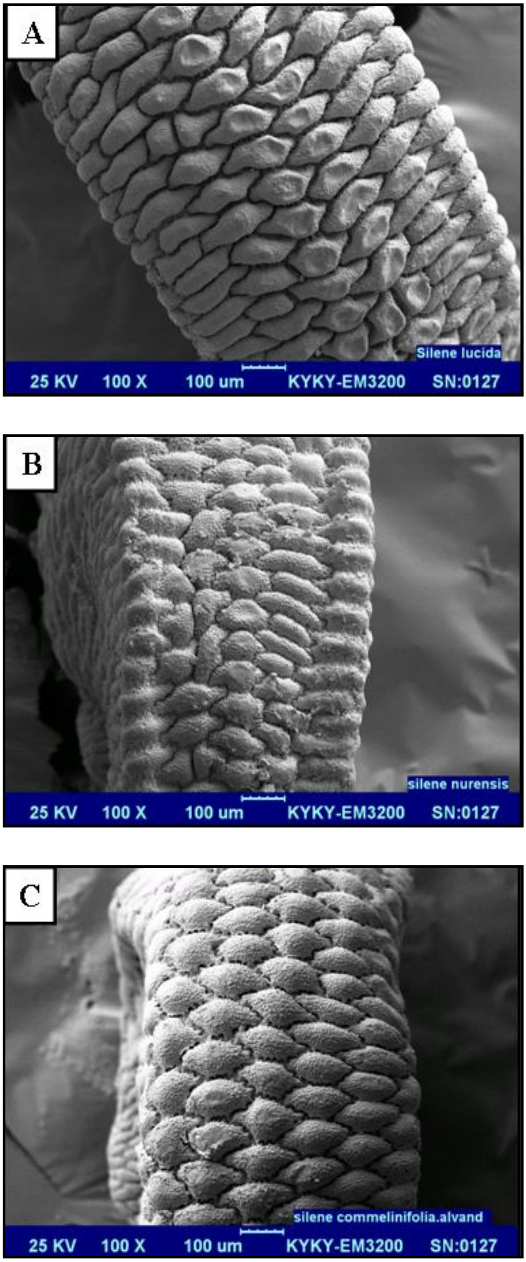

Fig. 8. SEM micrographs of seed surface in peripheral view: **A** – Bozqush population of *Silene lucida*; **B** – Zardkuh population of *S. nurensis*; **C** – Alvand population of *S. eremicana*.

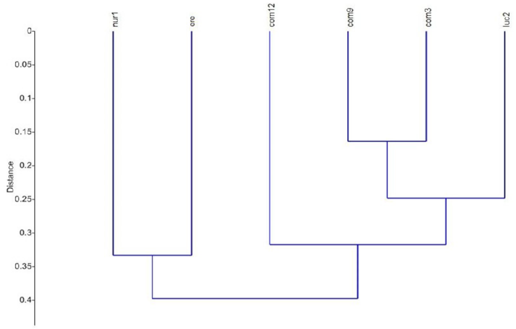

Fig. 9. Cluster analysis by UPGMA based on seed features: **com9** – *Silene commelinifolia* var. *commelinifolia*; **com12** – *S. commelinifolia* var. *ovatifolia*; **com3** – *S.* cf. *commelinifolia*; **nur1** – *S. nurensis*; **luc2** – *S. lucida*; **ere** – *S. eremicana*.

of the quantitative characters was used, while qualitative characters were coded as binary/multistate characters. Standardized variables (mean = 0, variance = 1) were applied in the statistical analysis. The average taxonomic distances and squared Euclidean distances were used as dissimilarity coefficients in the cluster analysis of seed morphological data. SPSS ver. 20 and PAST (Hammer *et al.* 2001) were used for statistical analysis. Cophenetic correlations were determined to fit the obtained dendrogram.

Results

Studied populations showed some differences in seed shape, size, and in lateral and peripheral surface characteristics. Main differences were related to such seed features as testa cells size and shape of their edges. General shape of seeds was round-reniform, symmetrical-reniform, cordate-reniform, asymmetrical-reniform and semi-circle reniform (Figs. 1–6).

Seed size varied from 1.28×1.43 mm in Alvand population of *S. eremicana* to 1.97×1.50 mm in Bozghosh population of *S. lucida*. Lateral seed surface is convex in Alvand population of *S. eremicana,* and flat in other studied populations (*S. lucida, S. nurensis, S.* cf. *commelinifolia* and *S. commelinifolia*). Peripheral surface of seeds is concave in Zardkuh population of *S. nurensis* and Bozghosh population of *S.* cf. *commelinifolia* (Fig. 7) and convex in Alvand population population of *S. eremicana* (Fig. 8).

Testa cells shape in studied populations was fusiform. The edges of testa cells were v-shaped in Bozqush population of *S.* cf. *commelinifolia* (Fig. 1 B) and Touchal population of *S. commelinifolia* var. *commelinifolia* (Fig. 2 B), while it was smooth and undulate in Darakeh population of *S. commelinifolia* var. *ovatifolia* (Fig. 3 B). Bozqush population of *S. lucida* (Fig. 4 B) and Alvand population of *S. eremicana* (Fig. 6 B) showed semi smooth testa cells edges. In Zardkuh population of *S. nurensis* testa cells edges were irregular and sinuate (Fig. 5 B).

UPGMA dendregram (Fig. 9), PCA (Fig. 10) and PCO (Fig. 11) plots of micro-morphological characters clearly separated studied taxa. UPGMA dendrogram showed two main clusters (Fig. 9). First cluster composed of two subsets composed of Zardkuh population of *S. nurensis* (*nur1*) and Alvand population of *S. eremicana* (*ere*). Second cluster composed of two subsets and contained Darakeh population of *S. commelinifolia* var. *ovatifolia* Melzh. (*com12*), Touchal population of *S. commelinifolia* var. *commelinifolia* (*com9*), Bozqush populations of *S. lucida* (*luc2*) and *S.* cf. *commelinifolia* (*com3*). Cluster analysis by Ward's method showed the same division.

Principal components analysis of seed micromorphological data revealed that two first components comprised about 70 % of the total variance. In the first component with about 46 % of total variance, seed morphological characters, including seed shape, testa cells edge, seed length, seed width, length/width ratio, width of peripheral surface, hilum region length, testa cell length, testa cell width and length/width ratio showed the highest correlation (>0.7). In the second component with about 24 % of total variance, lateral and peripheral surface had the highest correlation (>0.7). PCA indicated that seed characters (such as seed shape and size, lateral and peripheral surface, the size and edge of testa cells) are useful in identification and

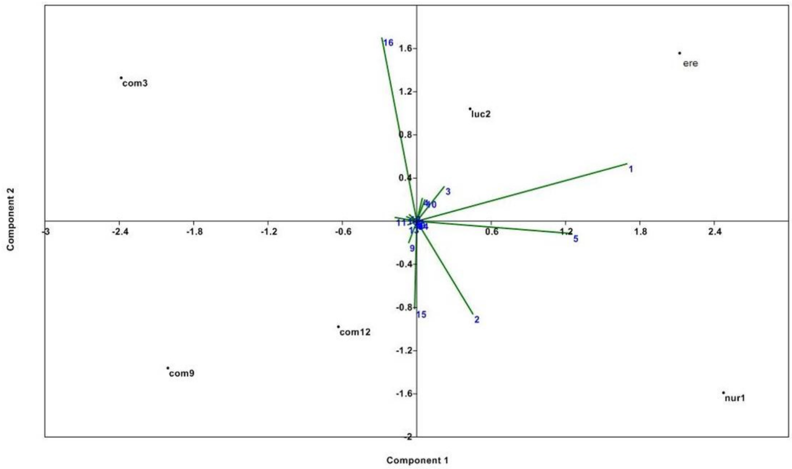

Fig. 10. PCA ordination scatter diagram of studied accessions based on seed features: **com9** – *Silene commelinifolia* var. *commelinifolia*; **com12** – S. *commelinifolia* var. *ovatifolia*; **com3** – *S.* cf. *commelinifolia*; **nur1** – *S. nurensis*; **luc2** – *S. lucida*; **ere** – *S. eremicana*.

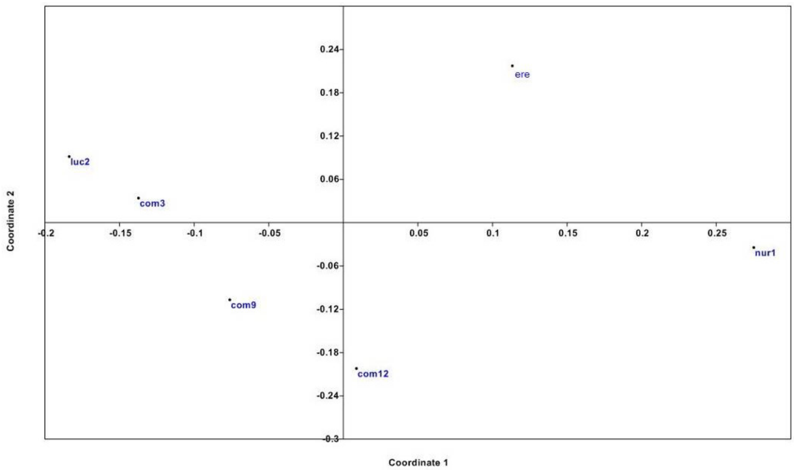

Fig. 11. PCO scatter diagram based on seed features in studied taxa. **com9** – *Silene commelinifolia* var. *commelinifolia*; **com12** – S. *commelinifolia* var. *ovatifolia*; **com3** – *S.* cf. *commelinifolia*; **nur1** – *S. nurensis*; **luc2** – *S. lucida*; **ere** – *S. eremicana*.

classification of the studied species and varieties (Fig. 10). Principal coordinate analysis plot showed taxa relationships and indicated the clear separation of taxa (Fig. 11). Cluster analysis, as well as PCA and PCO based on *Silene* seed morphology showed more or less similar outcomes.

Discussion

The results of present study (Figs. 1–6) are in accordance with several previous works, reported that the most *Silene* species have generally reniform and small seeds (Yildiz & Cirpici 1998; Zareh 2005; Fawzi *et al.* 2010).

We found that such micromorphological features as shape, size, lateral and peripheral surface of seeds, the size and edge of testa cells can be effectively applied for separation of studied species and varieties. For example, rounded reniform seed shape with v-shaped testa cells edge were the seed features for *S. commelinifolia*.

Studied taxa are clearly separated, what confirms previous morphological findings of Atazadeh *et al.* (2014). Although species are separated from each other, there is evident relationship between *S. eremicana* and *S. lucida*. Both varieties of *S. commelinifolia* (var. *ovatifolia* and var. *commelinifolia*) showed a high similarity in their seed features.

Atazadeh *et al.* (2014) pointed the unique position of *S.* cf. *commelinifolia* populations (such as Bozqush population) defined on the base of morphological and karyotype studies, and suggested existence there of new subspecies. The results of present study are in accordance with that conclusion. Populations of *S.* cf. *commelinifolia* in present study showed similarity in their seed micromorphology (*i.e.* shape, size, lateral and peripheral surface of seeds, the size and edge of testa cells) and probably represent new subspecies of *S. commelinifolia*.

References

Atazadeh N., Keshavarzi M., Sheidai M., Gholipour A. 2014. Morphological and karyotype diversity in populations of four *Silene* species (Caryophyllaceae). *Acta Biol. Szegediensis* **58 (1)**: 27–37.

Bari E.A. 1973. Cytological studies in the genus *Silene* L. *New Phytol.* **72**: 833–838.

Bittrich V. 1993. Caryophyllaceae. In: **Kubitzki K., Rohwer J., Bittrich V.** (eds), The families and genera of vascular plants. **Vol. 2**: 206–236.. Springer verlag, Berlin.

Camelia I. 2011. Aspects regarding seeds morphology and germination peculiarities at some taxa from *Silene* L. genera. *J. Plant Develop.* **18**: 5–10.

Chowdhuri P.K. 1957. Studies in the genus *Silene*. *Notes Royal Bot. Gard. Edinburgh* **22**: 221–287.

Fawzi N., Fawzy A., Mohamed A. 2010. Seed morphological studies on some species of *Silene* L. (Caryophyllaceae). *Int. J. Bot.* **6 (3)**: 287–292.

Ghazanfar S.A. 1983. Seed characters as diagnostic in the perennial sections of the genus *Silene* L. (Family Caryophyllaceae). *Pakistan J. Bot.* **15 (1)**: 7–12.

Greuter W. 1995. *Silene* (Caryophyllaceae) in Greece: A subgeneric and sectional classification. *Taxon* **44 (4)**: 543–581.

Hammer Q., Harper D.A.T., Ryan P.D. 2001. PAST: Paleontological statistics software package for education and data analysis. *Palaeontol. Electron.* **4 (1)**: art. 4. http://palaeo-electronica.org/2001_1/past/issue1_01.htm

Hong S.P., Han M.J., Kim K.J. 1999. Systematic significance of seed coat morphology in *Silene* L. s. str. (Sileneae – Caryophyllaceae) from Korea. *J. Plant Biol. (Korea)* **42**: 146–150.

Melzheimer V. 1980. Caryophyllaceae. In: Rechinger K.H. (ed.), Flora Iranica. **Nr. 163**: 353–508. Akademische Druck-U, Verlagsanstalt, Graz.

Melzheimer V. 1987. On the taxonomic position of *Silene thebana* (Caryophyllaceae). *J. Plant Syst. Evol.* **155**: 251–256.

Yildiz K., Cirpici A. 1998. Seed morphological studies of *Silene* L. from Turkey. *Pakistan J. Bot.* **30 (2)**: 173–188.

Zareh M.M. 2005. Seed diversity among certain species of Caryophyllaceae in Egypt. *Pakistan J. Biol. Sci.* **8 (5)**: 714–720.