

Systematic Significance of Seed Coat Morphology in *Silene* L. s. str. (Sileneae-Caryophyllaceae) from Korea

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Seeds of eight species of *Silene* s. str. in Korea were examined with a dissecting stereomicroscope and a scanning electron microscope (SEM) to evaluate the systematic significance of seed coat microstructure. The seeds of *Silene* s. str. were characterized by triangular or reniform shape, planed or concave dorsal surface in outline, tuberculate or colliculate testa, and sinuated cell margin. Based on the degree of grooves and invagination at dorsal surface, the seeds of the examined taxa of *Silene* s. str. fall into two distinct groups: (I) dorsal grooves inconspicuous, surface planed (*Silene jenseensis* and *S. repens*), (II) dorsal grooves conspicuous, surface concave (six remaining taxa). Within each group, seeds of all species may be further distinguished based on shape, size, and testa surface sculpturing. A key to the different species or groups of species is provided, and the systematic significance between/among the taxa is also briefly discussed.

Keywords: *Silene* s. str., Caryophyllaceae, seed micromorphology, SEM, systematics

The genus *Silene* L. belongs to the tribe Sileneae DC sensu Bittrich of Caryophyllaceae, and comprises about 700 species which are mainly distributed in temperate regions of the Northern Hemisphere (Chowdhuri, 1957; Melzheimer, 1988; Bittrich, 1993; Greuter, 1995).

The circumscription of the genus has long been controversial (Bittrich, 1993; Greuter, 1995, and references therein). Linnaeus (1754) divided *Silene* s. lat. into four genera. *Lychnis* L. and *Agrostemma* L. were recognized by having five styles. *Silene* s. str. and *Cucubalus* L. were diagnosed by trilocular capsules with three styles. *Silene* was further distinguished by the presence of a corona. The genus has been greatly expanded during the past decades. (Chowdhuri, 1957; Greuter, 1995). At the infrageneric level, a lumping trend has also been prevalent, from the multi-level hierarchical classification of Rohrbach (1869) to Chowdhuri's (1957) use of only section and subsection. Nevertheless, *Silene* in this wide circumscription has been tentatively subdivided into more than 20 sections, but knowledge of this large genus is still incomplete (McNeill, 1978; Bittrich, 1993; Greuter, 1995; Oxelman and Lidén, 1995).

Nine species of *Silene* s. str. are reported from Korea, two of which (*Silene fasciculata* Nakai, *S. takesimensis* Uyeke et Sakata) are known to be

endemic to the region (Lee, 1980, 1996). Recently, the systematic study of *Silene* s. str. in Korea was initiated by the senior author of this report. It was felt that the distinction of some taxa (*S. takesimensis*, *S. fasciculata*, *S. oliganthea* Nakai) was still unclear (Hong and Han, in prep.).

Surface features of seed coats are surprisingly little affected by environmental conditions under which a plant grows (Barthlott, 1984). Studies on seed morphology with SEM have revealed taxonomically-useful microcharacters, such as cellular arrangements (Clark and Jernstedt, 1978), cell shape (Canne, 1979; Oh, 1996), and the protrusions and patterns of the radial walls (Carolin, 1980). In Caryophyllaceae, seeds are relatively small (usually less than 3 mm long), and the testa is often variously sculptured (Bittrich, 1993). A few SEM studies of seed morphology for the taxa in Caryophyllaceae, including *Arenaria* L. (Wofford, 1981; Wyatt, 1984), *Sagina* L. (Crow, 1979), *Silene* (for a few local taxa, Melzheimer, 1977), and Korean *Melandrium* Roehl. (Chung and Lee, 1988), have been carried out.

Seed morphology of *Silene* has been briefly described in several manuals and floras (Martin and Barkley, 1961; Kitamura and Murata, 1963; Lee, 1980; Berggren, 1981, etc.). The seeds of *Silene* were first examined by Rohrbach (1868), who illustrated the dorsal and lateral surfaces of the seeds of 28 species. Shape and structure of the testa cells in the genus *Silene* by SEM have received considerable

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attention lately, especially in the works of Melzheimer (1977) and Candau and Talavera (1979) concerning the *Silene* sect. *Erectorefractae* Chowdhuri (8 species examined by SEM). According to Melzheimer (1977), the seed morphology of *Silene*, especially the form of testa cells, was found to be of great diagnostic value mainly for separating taxa at the species level.

Although the seed characters of *Silene* s. str. have been used systematically, there are no comparative SEM studies for the Korean species. The major purpose of the present study was to describe the amount of variation within the Korean species of *Silene* and to assess the systematic value of seed microcharacters in determining their interrelationships.

MATERIALS AND METHODS

We examined eight Korean taxa of *Silene* s. str. *S. foliosa* Maxim. occurring in North Korea, Japan and Manchuria was not included in this study due to the lack of samples. Materials used for this study were mainly obtained from herbarium specimens deposited in GWN, KHUS, SKK, SNU, SNUA (Abbreviations after Holmgren et al. 1990, cf. Table 1). We also obtained some field-collected seeds. Fully matured and undamaged seeds were selected under dissecting stereomicroscope at 30X magnification for size and color determination.

For SEM observations, seeds were attached to aluminum stubs with double-sided cellophane tape and coated with gold in an ion sputter coater (JEOL JFC-1100). The samples were then observed with JEOL JSM-35 or JSM-5200 SEM at 15 KV or 25 KV, respectively, and photographed with Kodak TMX 120 film. The terminology mainly follows that of Stern (1992) for general morphology.

RESULTS AND DISCUSSION

General Morphology of Seeds

Seeds of the Korean species of *Silene* s. str. are obliquely triangular or nearly reniform in outline. The ventrally located hilum, associated with a distinct notch, is median or subterminal. The seeds range from 0.5 to 1.9 mm in length, and are brown, reddish-brown or dull reddish-brown in color. Color or size variation appears to be primarily related to the number and position of ovules developing within an ovary (Tomb, 1974). Nevertheless, in each species of *Silene* s. str., there is a typical range of size and shape (Table 2, Figs. 1-16). Invagination occurs along the dorsal ridges forming dorsal grooves (Figs. 2, 4, 6, 8, 10, 12, 14, and 16). The degree and the depth of dorsal grooves are correlated with the degree of septa within a capsule. The lateral faces are typically flat or shallowly concave.

The testa cell in all species appears to be composed of cells having inter-digitated walls (i.e., sinuated cell margins). The cogwheel-like cells vary in outline from orbicular to elongate. The elongate cells occur most abundantly on the lateral surface of the seed, and are oriented toward the hilum. The testa cell tends to become more elongated near the hilum. However, very small, rounded cells form a band around the funicle. The surface of the cells are colliculate in most species, but seeds of *S. repens* Patr. have a slightly tuberculate surface (Figs. 7 and 8). The cogwheel-like testa cells are similar to those of some species of *Arenaria* L. (Wofford, 1981; Wyatt, 1984) and *Sagina* L. (Crow, 1979). But the detailed microsculpturing of *Silene* seeds are distinct from those of *Arenaria* and *Sagina* in having usually rounded rather than concave dorsal parts.

Table 1. List of specimens examined of *Silene* s. str. in Korea. *: specimens used for SEM micrographs.

<i>S. armeria</i> L.: Gwangweon: Mujangbong, 13 Jul 1998, <i>Han</i> 713 (KHUS); Mt. Gongjak, 4 Sep 1979, <i>Lee</i> s.n. (GWN); Hongcheon, 4 Sep 1977, <i>Yun</i> s.n. (GWN); Mt. Yonghwa, 18 Sep 1976, <i>Yun</i> s.n. (GWN). Kyungbuk: Isl. Ullung, 7 Aug 1961, <i>Lee</i> s.n. (SNUA). Kyungnam: Masan, 7 Aug 1980, <i>Lee</i> s.n. (SNUA). Seoul: Koryo Arbor., in 1935, <i>Do & Sim</i> 2020*, 2021 (SNU).
<i>S. koreana</i> Kom.: Gwangweon: Mt. Gongjak, 18 Aug 1998, <i>Han</i> 878 (KHUS). Pyongnam: Mt. Namglin, in 1935, <i>Do & Sim</i> 2068 (SNU)*.
<i>S. macrostyla</i> Maxim.: Hambuk: Mt. Gomu, Anonymous 445 (SKK)*.
<i>S. repens</i> Patr.: Hambuk: Mt. Gwanmohong, in 1936, <i>Do</i> 2030, 2032, 2057 (SNU). Hamnam: Geumbiryone, in 1934, <i>Do & Sim</i> 2028*, 2029 (SNU). Hwanghae: Mt. Jangsu, in 1935, <i>Do</i> 2031 (SNU).
<i>S. jensisensis</i> Willd.: Gwangweon: Mt. Gari, 19 Aug 1998, <i>Han</i> 820 (KHUS); Mt. Seolak, 25 Sep 1996, <i>Lee & Cho</i> s.n. (SNUA)*.
<i>S. fasciculata</i> Nakai: Cheju: Isl. Cheju, Mt. Halla, 17 Jul 1935, <i>Do</i> 2012 (SNU)*; 6 Aug 1998, <i>Hong</i> 806 (KHUS).
<i>S. takesimensis</i> Uyeki et Sakata: Kyungbuk: Isl. Ullung, 10 Aug 1989, <i>Jeong</i> 8800 (SKK)*; 14 Jul 1998, <i>Park</i> 714 (KHUS).
<i>S. oliganthea</i> Nakai: Gwangweon: Mt. Gari, 19 Aug 1998, <i>Han</i> 819 (KHUS)*. Hamnam: Shinhung-gun, in 1935, <i>Jeong & Nakai</i> 446 (SKK).

Table 2. Seed characteristics of Korean species of *Silene s. str.*

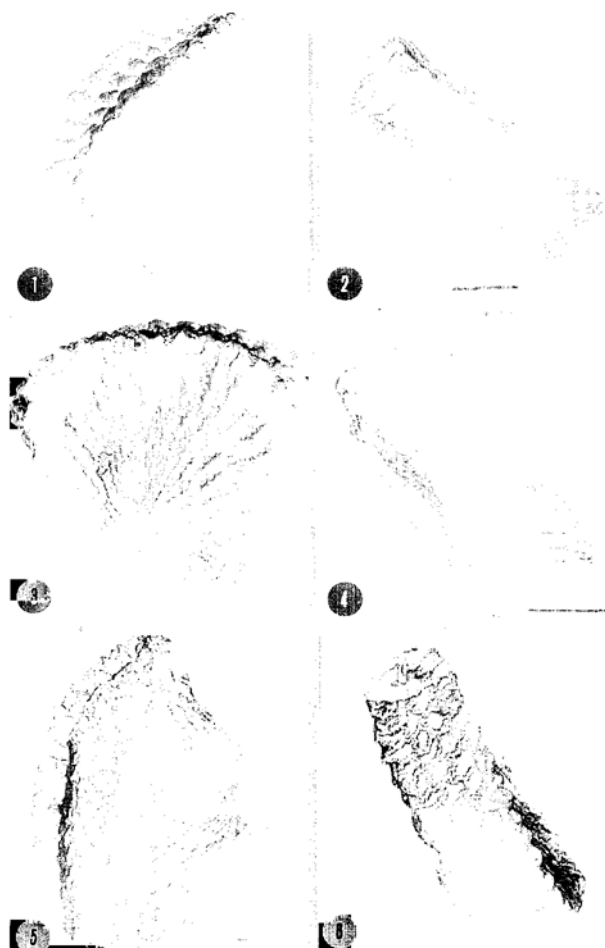
Toxon	Seed Length, Width (mm)	Lateral outline	Lateral cell number in row	Ventral cell number in row	Dorsal groove	Testa
<i>S. armeria</i>	0.87-1.00, 0.79-0.88	triangular	3	3	conspicuous	colliculate
<i>S. koreana</i>	0.73-0.95, 0.67-0.88	depressed obovate	3-4	3	conspicuous	colliculate
<i>S. macrostyla</i>	1.16-1.31, 0.99-1.16	depressed obovate	4	4	conspicuous	colliculate
<i>S. repens</i>	0.81-0.86, 0.65-0.73	reniform	4-5	5	inconspicuous	tuberculate
<i>S. jensiseensis</i>	0.77-0.91, 0.69-0.81	depressed obovate	3-4	4	inconspicuous	colliculate
<i>S. fasciculata</i>	0.80-0.81, 0.73-0.85	depressed obovate	4	4-5	conspicuous	colliculate
<i>S. takesimensis</i>	1.46-1.61, 1.20-1.33	asymmetrically reniform	3-4	3	conspicuous	colliculate
<i>S. oliganthella</i>	1.53-1.86, 1.09-1.26	reniform	4	4-5	conspicuous	colliculate

Seed features of the examined taxa may be divided into two distinct types (I, II; see below). The categories of the two types are primarily characterized by

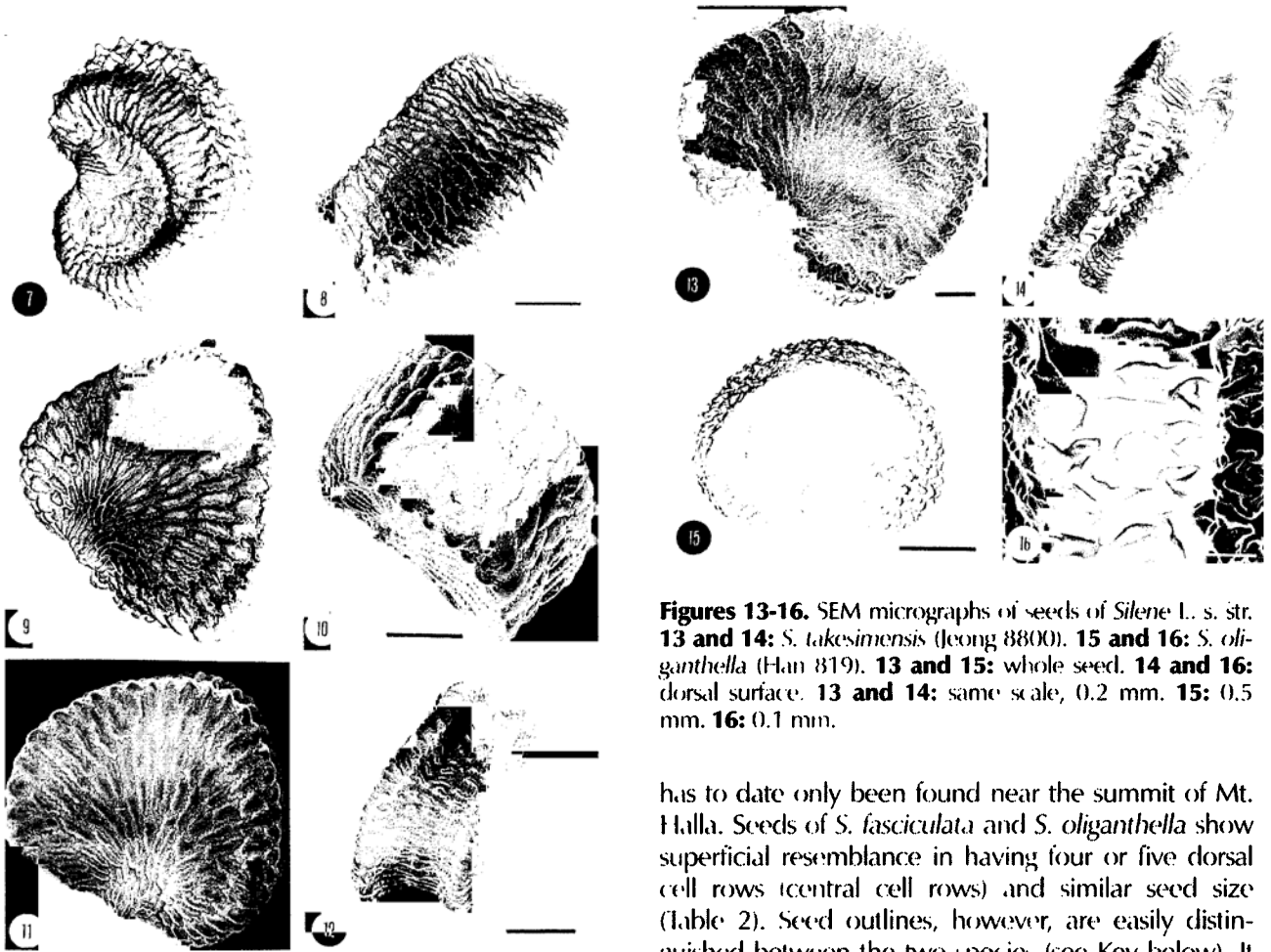
the distinctiveness of dorsal grooves and the degree of invagination of the dorsal surface. Within each type, seed size, color, shape and testa characters may further characterize each species. We have not chosen to describe seed morphology of each species in detailed, but micrographs (Figs. 1-16) are presented to show variations in each species. Descriptions are restricted to the minimum necessary to facilitate or to discuss the systematic appraisal of each species.

Type I: Dorsal grooves are not distinct, and dorsal surfaces are planar. The seeds of two species, *S. jensiseensis* Willd. and *S. repens*, are included in this type. Seeds of *S. jensiseensis* show basically colliculate cell surfaces and depressed obovate outlines (Figs. 9 and 10). In contrast, those of *S. repens* are slightly tuberculate with spines (9-18 μm long) on the cell surface and reniform in outline (Figs. 7 and 8), which is unique with respect to other Korean *Silene* taxa. Chung and Lee (1988) studied the seed morphology of the Korean *Melandrium*, and suggested that the seeds of *Silene s. str.* could be distinguished from those of *Melandrium* (crassuloid seed type) in having a 'saginoïd seed type (obliquely triangular in outline and concave)'. However, in the present study, both types are found in Korean *Silene s. str.* It is also noteworthy that the tuberculate testa cells of *S. repens* were similar to those of *Melandrium* (cf. Chung and Lee, 1988; Hong and Han, unpublished data). Thus, further comparative investigation between *Silene* and *Melandrium* is needed.

Type II: Dorsal grooves are distinct, and dorsal surfaces are concave. Testa cells are colliculate. The seeds of six species, *S. takesimensis* Uyeki et Sakata, *S. macrostyla* Maxim., *S. koreana* Kom., *S. fasciculata* Nakai, *S. armeria* L., and *S. oliganthella* Nakai, are included in this type. Seed sizes of *S. takesimensis*, *S. macrostyla* and *S. oliganthella* are larger than 1 mm,



Figures 1-6. SEM micrographs of seeds of *Silene* L. s. str. **1 and 2:** *S. armeria* (Do 2020). **3 and 4:** *S. koreana* (Do & Sim 2068). **5 and 6:** *S. macrostyla* (Anonymous 445). **1, 3, and 5:** lateral view of whole seed. **2, 4, and 6:** dorsal surface. **1 and 2:** same scale, 0.2 mm. **3 and 4:** same scale, 0.2 mm. **5 and 6:** same scale, 0.2 mm.



Figures 7-12. SEM micrographs of seeds of *Silene* L. s. str. **7 and 8:** *S. repens* (Do & Sim 2028). **9 and 10:** *S. jensisensis* (Lee & Cho s.n.). **11 and 12:** *S. fasciculata* (Do 2012). **7, 9, and 11:** lateral view of whole seed. **8 and 10:** plane dorsal surface. **12:** dorsal surface. **7 and 8:** same scale, 0.2 mm. **9 and 10:** same scale, 0.2 mm. **11 and 12:** same scale, 0.2 mm.

Figures 13-16. SEM micrographs of seeds of *Silene* L. s. str. **13 and 14:** *S. takesimensis* (Jeong 8800). **15 and 16:** *S. oliganthea* (Han 819). **13 and 15:** whole seed. **14 and 16:** dorsal surface. **13 and 14:** same scale, 0.2 mm. **15:** 0.5 mm. **16:** 0.1 mm.

while those of the three other species are usually under or slightly over 1 mm (Table 2).

Seeds of *S. takesimensis* and *S. oliganthea* show superficial resemblance, especially in size and lateral outline, but they can be distinguished by differences in ventral cell numbers. Additionally, *S. oliganthea* can be easily separated from *S. takesimensis* by its radical leaves and large flowers, usually less than 10 in number. In habitat, *S. takesimensis* occurs only on the cliffs of mountain slopes on Ullung Island, while *S. oliganthea* is found mostly in alpine regions of the northern part of Korea and in a few localities of the central part of Korea (e.g. Sorak Mts., Gari Mt., etc., Hong and Han, unpublished data).

S. fasciculata, an extremely rare Korean endemic,

has to date only been found near the summit of Mt. Halla. Seeds of *S. fasciculata* and *S. oliganthea* show superficial resemblance in having four or five dorsal cell rows (central cell rows) and similar seed size (Table 2). Seed outlines, however, are easily distinguished between the two species (see Key below). It is also noteworthy that *S. fasciculata* is not easily distinguished from *S. oliganthea* in habit or habitat (i.e. usually high mountain summits). Thus, further taxonomic investigation between the two species is needed.

In addition, *S. armeria* and *S. koreana* are similar in shape and the number of dorsal cell rows, although the two taxa show different geographical distributions. The seeds of these two species are very similar in all characters, except for the slight difference in outline and testa cell length. *S. armeria* was introduced from Europe and has been naturalized in many localities of Korea (Lee, 1980). Seeds of *S. koreana* are characterized by depressed, obovate outlines with subterminal hilum and one deeply-sunk row among three dorsal cell rows (Figs. 3 and 4).

In conclusion, on the basis of the seed characters, especially shape, surface, and size, the Korean taxa of *Silene* s. str. can be distinguished to some extent. A key to the species of *Silene* s. str. in Korea based on seed features is as follows:

Key to the Korean Taxa of *Silene* s. str. Based on Seed Characters

1. Dorsal grooves inconspicuous, dorsal surface plane (Type I)
 2. Surface colliculate, outline depressed obovate *S. jensiseensis*
 2. Surface tuberculate, outline reniform *S. repens*
1. Dorsal grooves conspicuous, dorsal surface concave (Type II)
 3. Seed lengths 1.1-1.9 4
 3. Seed lengths 0.6-1.0 5
 4. Outline reniform *S. oliganthea*, *S. takesimensis*
 4. Outline obovate *S. macrostyla*
 5. Outline triangular *S. armeria*
 5. Outline obovate *S. fasciculata*, *S. koreana*

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