Wulfenia **24** (2017): 205–220

Wulfenia
Mitteilungen des
Kärntner Botanikzentrums
Klagenfurt

Structural features of the seed coat in Caucasian representatives of *Minuartia* (Caryophyllaceae)

Sofia G. Zaychenko & Alexander S. Zernov

Summary: Seeds of 18 species of *Minuartia* L. growing in the Caucasus were studied by means of scanning electron microscope (SEM) as well as light microscope (LM). Main focus was put on characteristics of the ultrastructure of the seed surface as well as on the anatomical structure of all layers of the seed coat. Several types of seed coat structure were identified. The most important characters for systematic purposes were identified: the microrelief of the outer cells of the exotesta due to the specific structure of the cuticular layer, the type of the curvature of the anticlinal exotesta walls, the so-called 'cell junctions', the shape of the thickenings of the exotesta periclinal cell walls and the presence or absence of pronounced endotesta and tegmen layers. In total, these features turned out to be species-specific for each member of the genus. In addition, a link between molecular phylogenetic data and the structure of seed coat was established.

Keywords: Caryophyllaceae, *Minuartia*, seeds, seed coat anatomy, Caucasus, systematics

The genus *Minuartia* L. (Alsinoideae Fenzl (Bittrich 1993), Caryophyllaceae Juss.) includes about 175 species (McNeill 1962; Rechinger 1988; Halliday 1993; Kamari 1997; Rabeler et al. 2005; Hernández-Ledesma et al. 2015). Their natural habitat is generally confined to the Arcto-Alpine zone of the northern hemisphere: Europe, especially in the Mediterranean region, the Irano-Turanian region and Caucasus. The genus is represented by 22 species in the Caucasus (Lazkov 2012). Taxonomic accounts of the genus were provided by Mattfeld (1922) and McNeill (1962, 1963). Both authors distinguish the genus relying on structural features of reproductive organs such as the presence of three stylodia as well as a capsule opening with three valves.

At present time, much attention is paid to the systematic status of *Minuartia* within Caryophyllaceae. Relations between the taxa within the genus are also subject to numerous discussions. In the last decade, significant changes in the understanding of the monophyletic nature of the genus have arisen as a result of numerous molecular phylogenetic studies (Harbaugh et al. 2010; Greenberg & Donoghue 2011; Dillenberger & Kadereit 2014; Hernández-Ledesma et al. 2015). Phylogenetic data disagree with the ideas based on the studies of morphological and anatomical features of the genus. During the last century, features of various parts of the shoot as well as the features of the flower in particular, various morphometric parameters were generally used for systematic purposes in the genus. Usually, the capsules, pericarp and exocarp surface structures are used as taxonomic features of the reproductive part for the differentiation of Caryophyllaceae (Devyatov 1991, 1996; Devyatov & Tajsumov 1999). The importance of using seed micromorphology at generic and specific levels in Caryophyllaceae has been recognized by many authors (Kozhanchikov 1975; Crow 1979; Celebioğlu et al. 1983; Yildiz & Çirpici 1998; Yildiz 2002; Minuto et al. 2006; Fawzi et al. 2010; Amini et al. 2011; Minuto et al. 2011). So far, a detailed description of the seed micro-morphology structural features of *Minuartia*

Table 1. The material used in the work and its origin.

Specimen	Voucher specimen
M. biebersteinii	MW: Georgian SSR, at the foot of Mount Kazbek. 08/1995. Anonymus
M. lineata	MHA: Azerbaijan SSR. The Nagorno-Karabakh AO. Hadrut region, Mount Ziyarat. 1966. Gogina
M. acuminata	MW: Nakhichevan. The top of the ridge in the upper reaches of the river Haiza. 07/20/1937. Zemtsova
M. akinfievii	MW: Armenia, the vicinity of Mount Yerevan. 1931. Karyagin
M. glomerata	MW: Crimea. Surroundings of the village Reservnoe, on the edge of the rocky outcrops. 05/31/2015. Zaychenko
M. meyeri	MW: Expedition to study the flora of the Nigrian region of Armenia. 06/11/1934. Anonymous
M. setacea	MHA: Karachay-Cherkessia. Road Ullu-kan-Uchkulan. 07/11/1976. Gogina
M. sclerantha	MW: Inter pag. Migri et eustodiam Alidara adfl. Araxem. 18/05/1934. Anonymous
M. wiesneri	MW: Azerbaijan SSR. Multi-cereal-euphorbia deserts on the hills of Turpacz. 09/27/1983. Kuvaev, Shelgunov
M. oreina	MHA: Karachay-Cherkessia. Zelenchuksky district, the upper reaches of the Aksaut River, the gorge of the Kti-Teberda River. 08/18/1979. Gogina
M. hybrida	MW: Georgia, Tbilisi. Rocky talus above the botanical garden. 06/01/1910. Voronov
M. aizoides	MW: Dagestan, right bank of the river Mularchay, alpine meadow, height 2710 m. 08/06/1989. Amirkhanov
M. circassica	MW: Teberda Reserve, Dombai, Mussa-Achitara. 08/31/1980. Gogina
M. rhodocalyx	LE: Western Caucasus. Caucasian State Reserve and its security zone. On the slopes on the eastern spur of Mount Chuba. 08/17/1929. Leskov & Rusaleev
M. imbricata	MW: North Caucasus. Krasnodarskiy krai. 1937. Kats
M. inamoena	MW: Karachay-Cherkessia. Karachaevsky district, gorge of the river Kukyurtlu, left side, height 2900 m, about 500 m from the edge of the glacier. 08/03/2009. Zernov
M. brotherana	MW: Georgia. Kazbegi. 08/01/1985. Khokhryakov
M. verna	MW: Karachay-Cherkessia. Zelenchuksky district, the Abishir-Ahuba ridge, the southern slope of the Tserkovniy Pass, a carbonate scree, a height of 2700. 07/08/2013. Zernov

exists for only a few species, mainly from Iran, Iraq and Turkey (Yildiz 2002; Mostafavi et al. 2013; Al-Saadi & Al-Taie 2014). There are no anatomical data in previous studies. Only five among the studied species inhabit the Caucasus. The relevance of this work is directly related to the refinement of the correlation between molecular genetic data on the systematics of *Minuartia* and the anatomical and morphological structural features of the seed coat as well as the clarification of the possibility of using the seed coat structure as a species-specific feature.

Materials and methods

Samples were collected in the field as well as obtained from the herbaria MW, MHA and LE. Field material was collected in the territory of the Teberda Nature Reserve in August 2015. Features of the seed coat surface were studied using a JSM scanning electron microscope (SEM) at Moscow State University. Anatomical examinations of the seed coat were performed on transverse sections by the use of toluidine blue staining. Further studies of the sections were carried out using an OLYMPUS CX 41 digital microscope with a visualization system. The terminology of Bittrich

(1993) was used to describe the surface of the exotesta. The seeds of most species were analyzed two to three times from the same accession. The possibility of variability within a species was preliminarily excluded based on a visual analysis of at least 20 seeds taken from 3–4 plants. The studied species and samples are presented in Table 1.

Results

Seed morphology. Table 2 shows the main morphological characteristics of the seeds of the 18 species studied. There was no strong deviation in terms of morphological structure according to the results of a visual assessment. Based on the assumption of a similar seed structure, the description of the cell surface ultrastructure (microrelief), the shape of the exotesta cells and the degree of anticlinal cell wall curvature, which determines the nature of cell boundaries, was determined using photographs obtained with SEM.

Seed shape. The kidney-shaped form is predominant among the seeds of Caucasian representatives of *Minuartia*. Only the form of the seeds of *M. glomerata* (Fig. 2a) and *M. acuminata* (Fig. 2g) was characterized as elliptical. The presence of a cristate-fimbriate edge along the back (dorsal) of the seed was noted for *M. rhodocalyx*, *M. imbricata*, *M. inamoena* and *M. brotherana* (Figs 4d, g, j; 5a). According to the taxonomy proposed by McNeill (1962, 1963), the above-mentioned species belong to the section *Spectabiles* ser. *Laricinae*. Another important feature is the surface of the back of the seed. It can be convex, concave and can also form an edge along the lateral surface of the seed. A brief description of the seeds of the studied species is given in Table 3. The most common surface form is convex. *Minuartia acuminata* (Fig. 1i), *M. glomerata* (Fig. 2c) and *M. aizoides* (Fig. 3l) have a concave surface of the back of the seed.

Color. As a general rule, the seeds are dark-brown due to the presence of tannins in the exotesta cells. Only the species of ser. *Laricinae* with reddish cristate-fimbriate edge have a light brown color.

Exotesta cell shape. An elongated form of cells predominates among the taxa studied. A polygonal shape is found in *M. acuminata* (Fig. 1h), *M. glomerata* (Fig. 2b), *M. wiesneri* (Fig. 2e) and *M. verna* (Fig. 5e). The precise shape of the cell could not be determined for species from ser. *Laricinae*, i.e. *M. rhodocalyx*, *M. imbricata*, *M. inamoena* and *M. brotherana* (Figs 4e, h, k; 5b). Therefore, we decided to call this shape irregular.

Cell border shape. A serrate cell border shape that ensures a tight closure between cells is typical of the Caucasian representatives of *Minuartia*. The degree in which this feature is manifested may significantly vary among species, but it is always possible to clearly observe the presence of a serrate junction, except *M. circassica* whose cellular connections are represented by straight and even seams.

Exotesta cell surface structure. Usually the surface of the cells has verrucose or verrucose-reticulated ornaments, which are derivatives of the cuticular layer. *M. sclerantha* has the most unsculptured surface. The cuticle forms a thin layer that covers the exotesta cells (Fig. 3k). Representatives of section *Spectabiles* ser. *Laricinae* are characterized by a wrinkled surface formed by a thin cuticular layer.

Sizes. Sizes range between 1515 × 1117 µm to 428 × 250 µm. Representatives of section *Spectabiles* ser. *Laricinae* have the largest seeds, while the smallest seeds are inherent to section *Minuartia*.

Table 2. Morphological seed structure characteristics.

Species name	Seed shape	Color	Cell shape	Cell border shape	Microrelief
M. biebersteinii	Kidney-shaped	Brown	Elongated	Serrate	Verrucose
M. lineata	Kidney-shaped	Brown	Elongated	Serrate	Verrucose
M. acuminata	Elliptical	Brown	Polygonal	Serrate	Verrucose
M. akinfievii	Kidney-shaped	Brown	Elongated	Serrate	Verrucose
M. glomerata	Elliptical	Brown	Polygonal	Serrate	Verrucose
M. meyeri	Kidney-shaped	Brown	Elongated-polygonal	Serrate	Verrucose
M. setacea	Kidney-shaped	Brown	Elongated	Serrate	Verrucose
M. sclerantha	Kidney-shaped	Brown	Elongated-polygonal	Serrate	Smooth
M. wiesneri	Kidney-shaped	Brown	Polygonal	Serrate	Verrucose
M. oreina	Kidney-shaped	Brown	Elongated	Serrate	Verrucose-reticulated
M. hybrida	Kidney-shaped	Brown	Elongated	Serrate	Verrucose
M. aizoides	Kidney-shaped	Brown	Elongated-polygonal	Serrate	Verrucose-reticulated
M. circassica	Kidney-shaped	Brown	Elongated-polygonal	Straight	Verrucose-reticulated
M. rhodocalyx	Kidney-shaped	Light brown	Irregular	Serrate	Wrinkled
M. imbricata	Kidney-shaped	Light Brown	Irregular	Serrate	Wrinkled
M. inamoena	Kidney-shaped	Light Brown	Irregular	Serrate	Wrinkled
M. brotherana	Kidney-shaped	Light Brown	Irregular	Serrate	Wrinkled
M. verna	Kidney-shaped	Brown	Polygonal	Serrate	Verrucose

Table 3. Seed back surface type.

Species name	Seed shape	Seed back surface
M. biebersteinii	Kidney-shaped	Convex
M. lineata	Kidney-shaped	Convex, forming an edge on the lateral surface of the seed
M. acuminata	Elliptical	Concave, forming an edge on the lateral surface of the seed
M. akinfievii	Kidney-shaped	Convex
M. glomerata	Elliptical	Concave
M. meyeri	Kidney-shaped	Convex
M. setacea	Kidney-shaped	Convex
M. sclerantha	Kidney-shaped	Convex
M. wiesneri	Kidney-shaped	Convex
M. oreina	Kidney-shaped	Convex
M. hybrida	Kidney-shaped	Convex
M. aizoides	Kidney-shaped	Concave
M. circassica	Kidney-shaped	Convex
M. rhodocalyx	Kidney-shaped	Cristate-fimbriate edge
M. imbricata	Kidney-shaped	Cristate-fimbriate edge
M. inamoena	Kidney-shaped	Cristate-fimbriate edge
M. brotherana	Kidney-shaped	Cristate-fimbriate edge
M. verna	Kidney-shaped	Convex

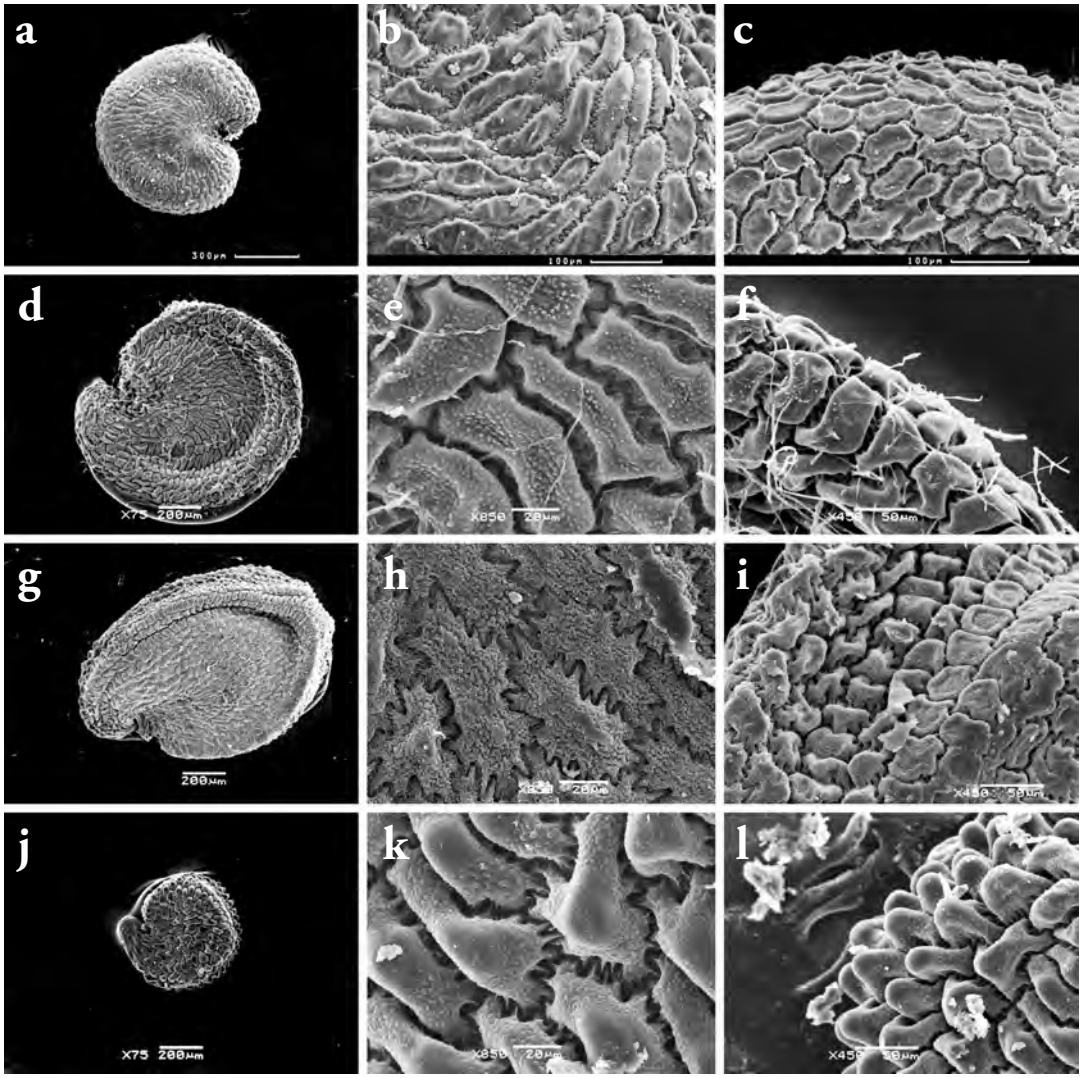

Figure 1. a, b, c – *M. biebersteinii*; d, e, f – *M. lineata*; g, h, i – *M. acuminata*; j, k, l – *M. akinfievii*. a, d, g, j – general view of the seeds; b, e, h, k – exotesta cells on the lateral surface of the seed; c, f, i, l – seed back surface.

One can judge about the shape of the seed by the numerical expression of the length to width ratio. This ratio is greater than the one for most species indicating a kidney-shaped form. The value is less than one for elliptical seeds (*M. glomerata*, *M. acuminata*). The cell sizes vary depending on the location on the seed itself. The longest cells are on the side surface of the seed, the smallest and shortest are in the hilum area. On average, the cells on the lateral surface are always more elongated than the cells on the surface of the back.

Seed coat anatomy

The main anatomical structural features of exotesta cells are presented in Table 4. A typical outline of the structure of the seed coat can be noted for all of the studied species. The seed coat consists of three layers: the external layer (exotesta), the intermediate layer (endotesta) and the internal layer (tegmen). The exotesta is represented by full-fledged cells with well-defined and often heavily thickened cell walls in all examined species.

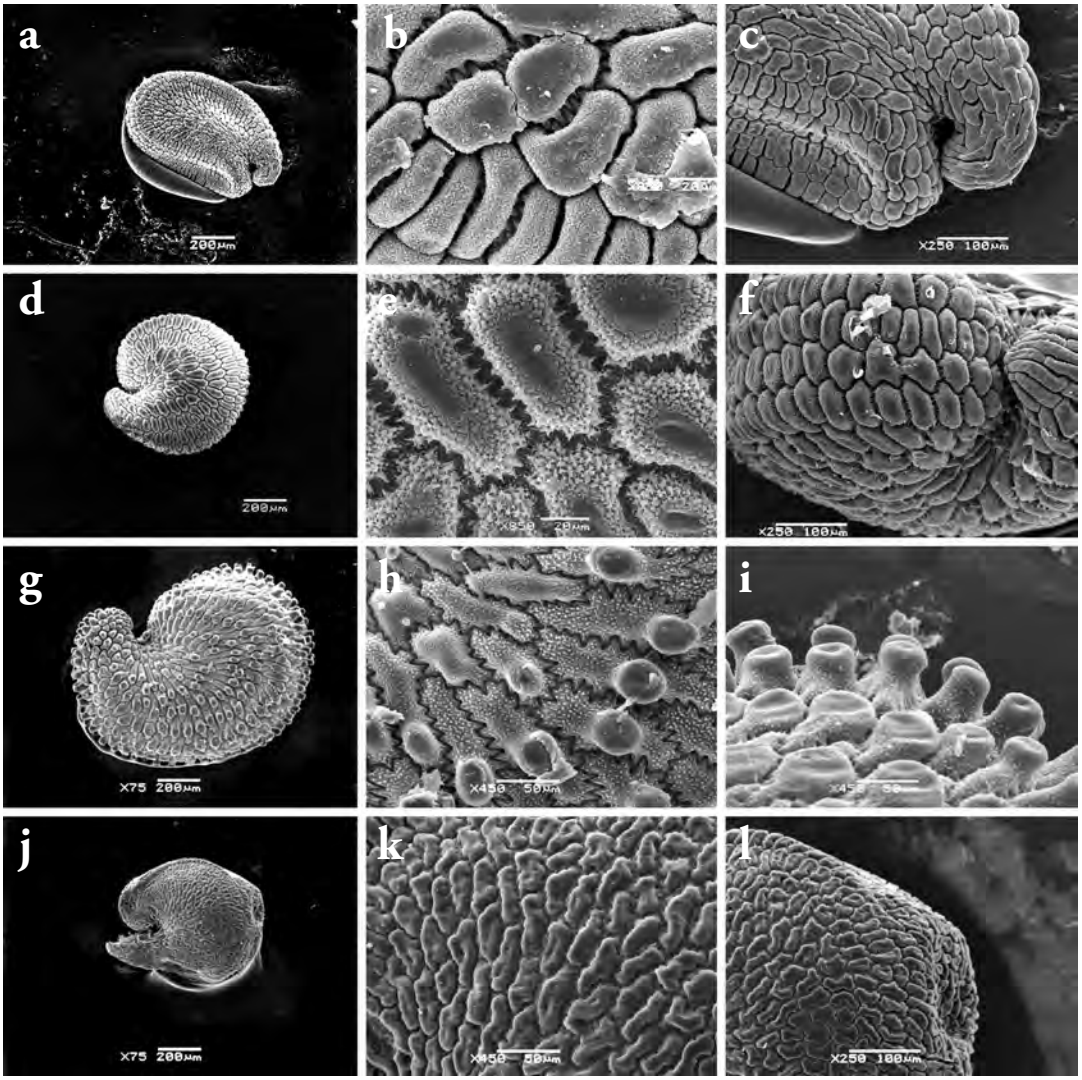

Figure 2. a, b, c – *M. glomerata*; d, e, f – *M. meyeri*; g, h, i – *M. setacea*; j, k, l– *M. sclerantha*. a, d, g, j – general view of the seeds; b, e, h, k – exotesta cells on the lateral surface of the seed; c, f, i, l – seed back surface.

Exotesta. In all cases, the exotesta is represented by a single layer of cells. The cells are usually filled with tannins, which give the seeds a dark color. The nature of the thickening of the external anticlinal cell wall varies. A strongly thickened cellulose wall of conical shape is typical of *M. acuminata* (Fig. 6c), *M. akinfievii* (Fig. 6d), *M. meyeri* (Fig. 7b) and *M. oreina* (Fig. 8c). The rare cylindrical form is typical of *M. setacea* (Fig. 7c). A trapezoidal exotesta cell structure is typical of the section *Acutiflorae*: *M. biebersteinii* (Fig. 6a) and *M. lineata* (Fig. 6b). A papillate thickening was observed in *M. hybrida* (Fig. 8d) and *M. verna* (Fig. 9b). In *M. aizoides* (Fig. 7a), the outer periclinal wall is strongly thickened, but does not form any outgrowths. Therefore it is of planar shape. Depending on the location of the cells on the seed, different types of cell wall thickenings are observed for some species. Strong differences between the cells of the lateral surface and the cells of the back were observed in section *Spectabiles* ser. *Laricinae*. Exotestal cells of the back form long thin-walled outgrowths which constitute the cristate-fimbriate edge of the seed. These outgrowths are nothing but a highly vertically elongated cell wall. Such cells

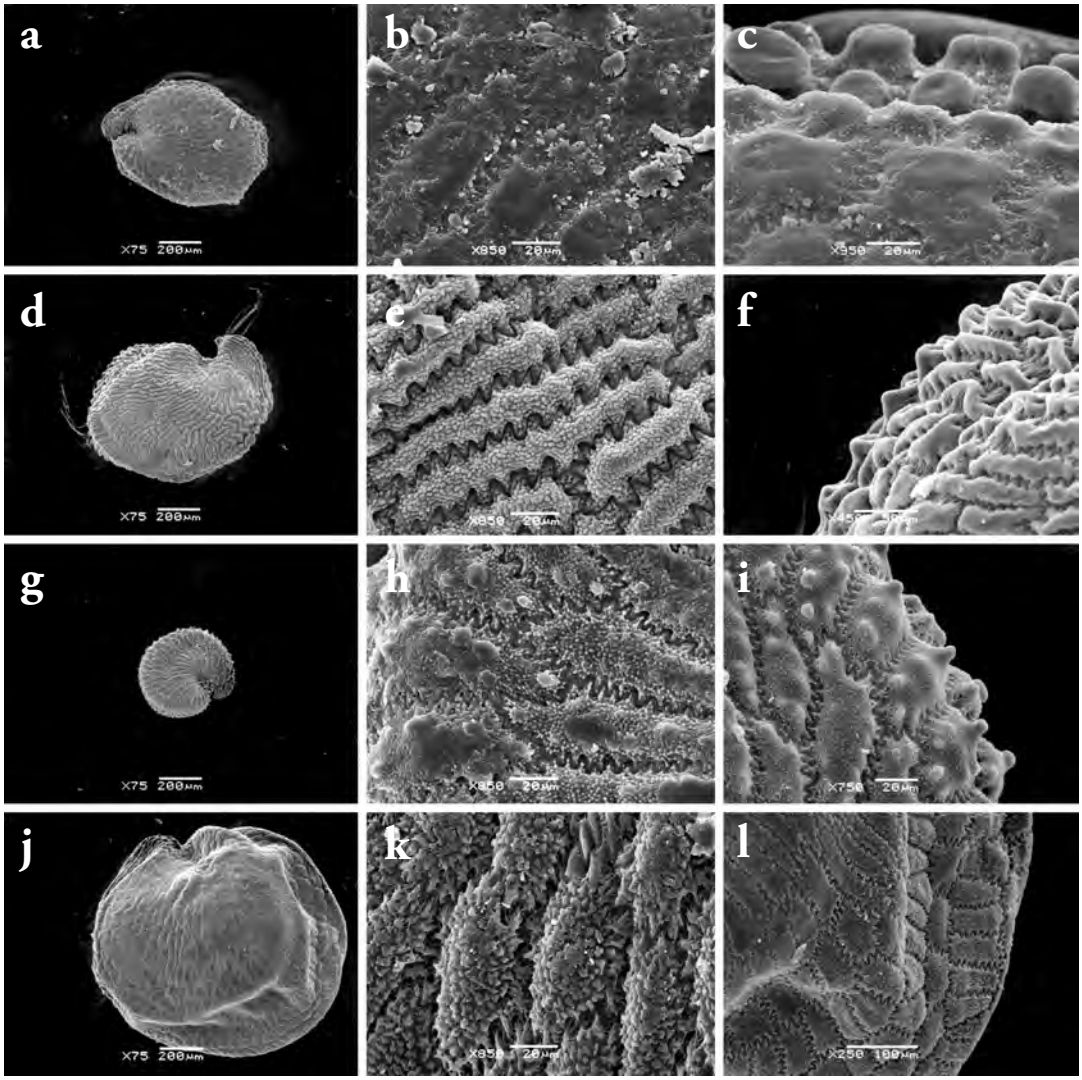

Figure 3. a, b, c – *M. wiesneri*; d, e, f – *M. oreina*; g, h, i – *M. hybrida*; j, k, l – *M. aizoides*. a, d, g, j – general view of the seeds; b, e, h, k – exotesta cells on the lateral surface of the seed; c, f, i, l – seed back surface.

are free of contents. Apparently, this structure of exotestal cells, which form a kind of 'wing' around the kidney-shaped seed, serves to ensure a better volatility of the diaspores. There is no thickening of the outer cell wall on the lateral surface of the seeds with a cristate-fimbriate edge. As a result, the cell shape is irregular and resembles a dome, but even so, the exotesta periclinal outer wall acquires a crumpled appearance. Another species with different types of exotesta cells is *M. circassica* (Fig. 9b, c). The exotesta of the back is represented by thick-walled conical cells and the exotesta on the lateral surface is represented by flat-shaped cells. A similar structure is also present in *M. wiesneri* (Fig. 8a, b) differing in the trapezoidal shape of the cells typical of the exotesta on the back of the seed.

Cuticle. The cuticle on the surface of *Minuartia* seeds can form various structures. The simplest cuticle was observed in *M. rhodocalyx*, *M. imbricata*, *M. inamoena*, *M. brotherana* (Fig. 9a) and also in *M. sclerantha* (Fig. 7d). The seeds of these plants have a cristate-fimbriate edge. In these

Table 4. Anatomical structure characteristics of seed coat.

Species name	The shape of the outer periclinal wall of cells located on the back of the seed	The shape of the outer periclinal wall of cells located on the lateral surface of the seed	Cuticle structure
M. biebersteinii	Trapezoid	Trapezoid	Micro-papillae
M. lineata	Trapezoid	Trapezoid	Micro-papillae
M. acuminata	Conical	Conical	Micro-papillae
M. akinfiewii	Conical	Conical	Warty micro-papillae
M. glomerata	Trapezoid	Dome-shaped	Micro-papillae
M. meyeri	Conical	Conical	Micro-papillae
M. setaceae	Cylindrical	Cylindrical	Micro-papillae
M. sclerantha	Trapezoid	Trapezoid	Thin cuticular layer
M. wiesneri	Trapezoid	Planar	Cuticular layer with shapeless glomerates
M. oreina	Conical	Conical	Trichome-like micro-papillae
M. hybrida	Papillate	Papillate	Micro-papillae
M. aizoides	Planar	Planar	Trichome-like Micro-papillae
M. circassica	Conical	Planar	Trichome-like Micro-papillae
M. rhodocalyx	Winged	Wrinkled dome-like	Thin cuticular layer
M. imbricata	Winged	Wrinkled dome-like	Thin cuticular layer
M. inamoena	Winged	Wrinkled dome-like	Thin cuticular layer
M. brotheriana	Winged	Wrinkled dome-like	Thin cuticular layer
M. verna	Papillate	Papillate	Micro-papillae

species, the cuticle is represented by a thin layer covering the exotesta cells. *Minuartia wiesneri* has a cuticle that forms lumps and clots on the seed surface. All other species are characterized by a cuticle of a more complex structure. Vertical outgrowths of the cuticle, the so-called micropapillae are typical of most representatives. Micropapillae can be of different shapes. For example, they can look like small verucose micropapillae (*M. akinfievii*) or they can be long, hair-like, trichome-like micropapillae (*M. oreina*, *M. aizoides*, *M. circassica*) as well as simple micropapillae (*M. biebersteinii*, *M. acuminata*, *M. glomerata* and others).

Endotesta and tegmen. The inner layers of the seed coat in mature seeds are usually represented by strongly compressed and poorly discernible cells. Single-layer endotesta and tegmen can only be observed in seeds of *M. rhodocalyx*, *M. imbricata*, *M. inamoena* and *M. brotherana* (Fig. 9a).

Discussion

The structure of seed coat in 18 species of *Minuartia* was examined in detail. Significant differences in the morphological and anatomical structure of different representatives were discovered.

The connection between the structure of the seed coat and the classification based on the morphological features of plants. The systematics of the genus developed on the basis of morphology is based in most cases on features such as shape of the leaves, presence of pubescence on various parts of the shoot, number of veins in the leaves, petals and sepals and ratio of length of sepals to corolla leaves. The structure of the seeds in this system was not taken into account,

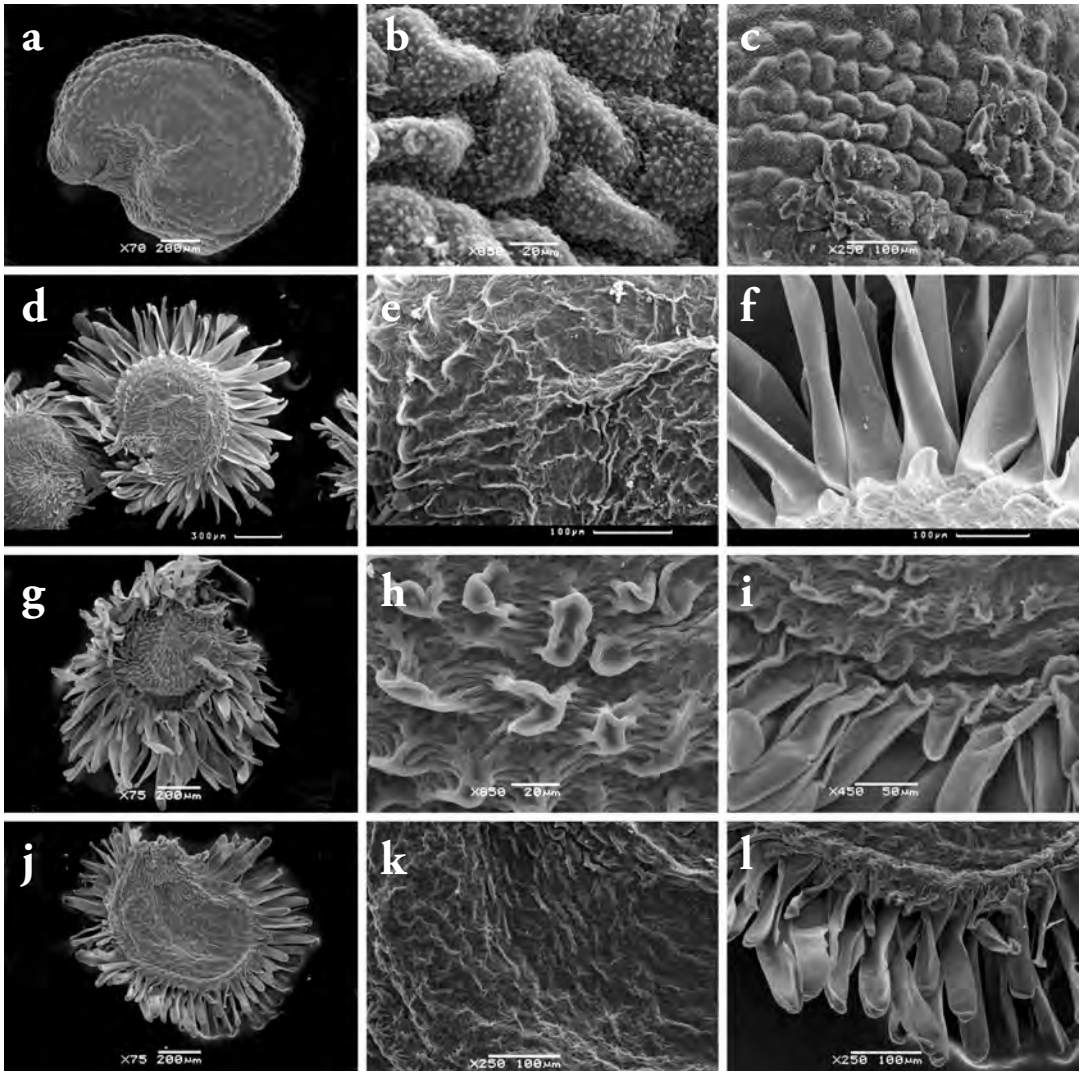

Figure 4. a, b, c – *M. circassica*; d, e, f – *M. rhodocalyx*; g, h, i – *M. imbricata*; j, k, l – *M. inamoena*. a, d, g, j – general view of the seeds; b, e, h, k – exotesta cells on the lateral surface of the seed; c, f, i, l – seed back surface.

except for the mentioning of the specific cristate-fimbriate edge on diaspores in representatives of section *Spectabiles*. According to the results of this work, it can be argued that the nature of the structure of the seed coat, especially the anatomical study of the seed coat, does not support the traditional classification of the genus. A unified structural plan of the seed coat can be observed only in several groups. These groups include representatives of the section *Acutiflorae*, *M. biebersteinii* and *M. lineata*. It can be noted that these species retained their sister status in the system based on molecular genetic data (Dillenberger & Kadereit 2014). The second group with a unified seed coat organization plan contain species from section *Spectabiles* ser. *Laricinae*. However, *M. circassica* has a very different testa structure than other members of this section. A general plan cannot be isolated for species in section *Minuartia*.

The connection between molecular data on systematics of *Minuartia* and testa structure. After the publication of the phylogenetic classification of the genus (Dillenberger & Kadereit 2014), the problem of finding well-established systematic features characterizing the taxa arose.

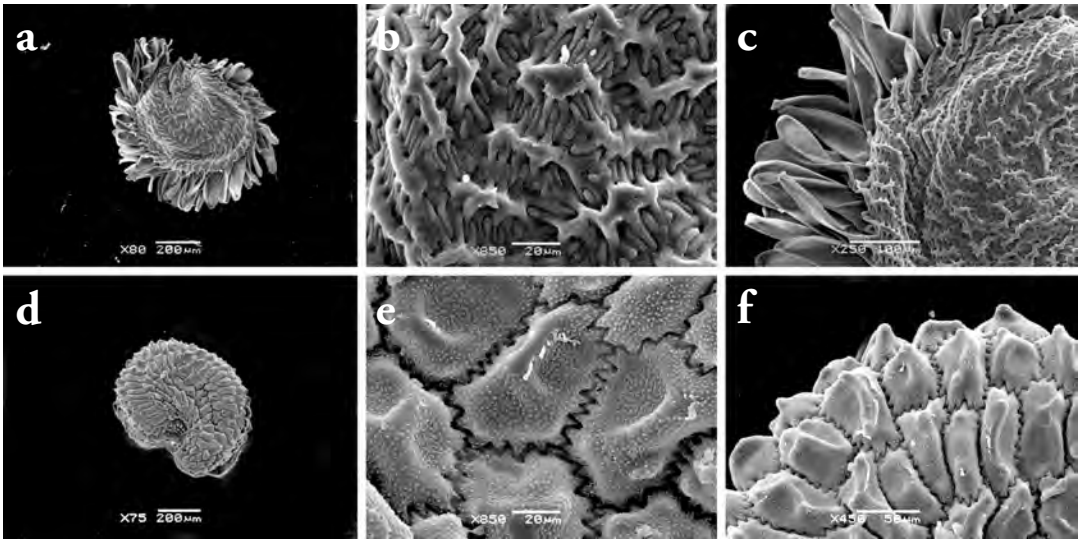

Figure 5. a, b, c – *M. brotherana*; d, e, f – *M. verna*. a, d, g, j – general view of the seeds; b, e, h, k – exotesta cells on the lateral surface of the seed; c, f, i, l – seed back surface.

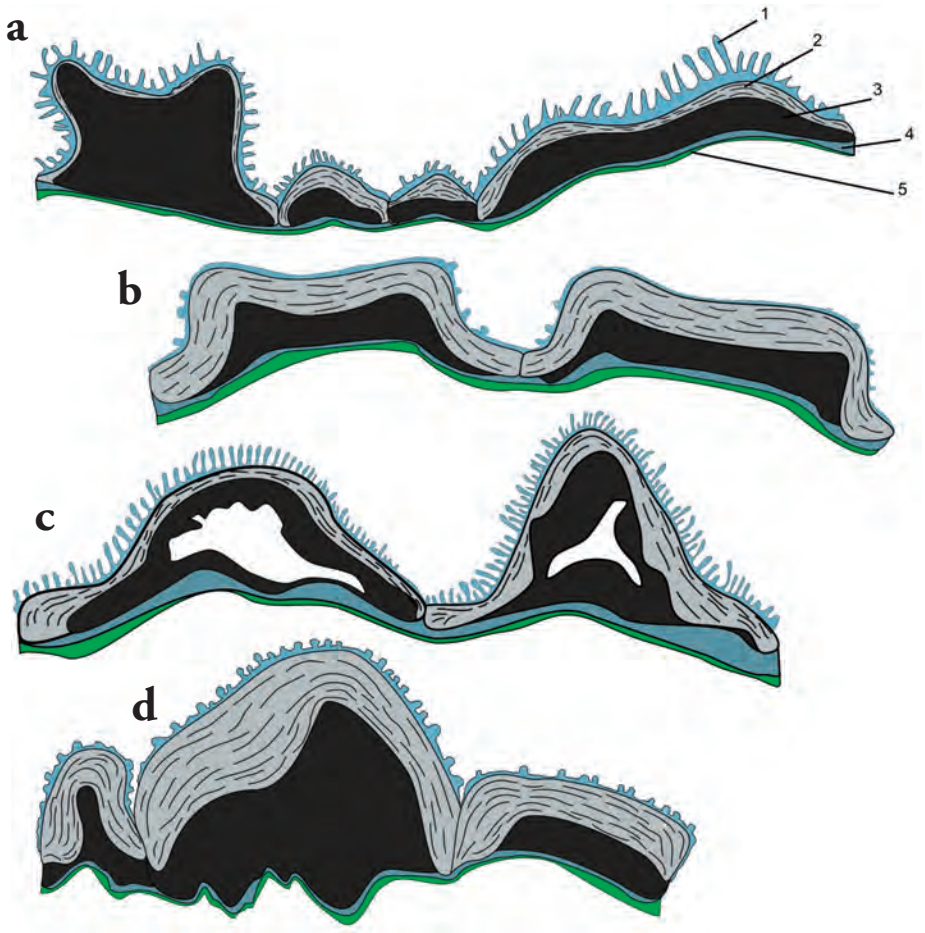

Figure 6. Testa structure. a – *M. biebersteinii*; b – *M. lineata*; c – *M. acuminata*; d – *M. akinfievii*. 1 – cuticular layer; 2 – exotesta cell walls; 3 – cell filled with tannins; 4 – compressed endotestal cell layer; 5 – obliterated tegmen.

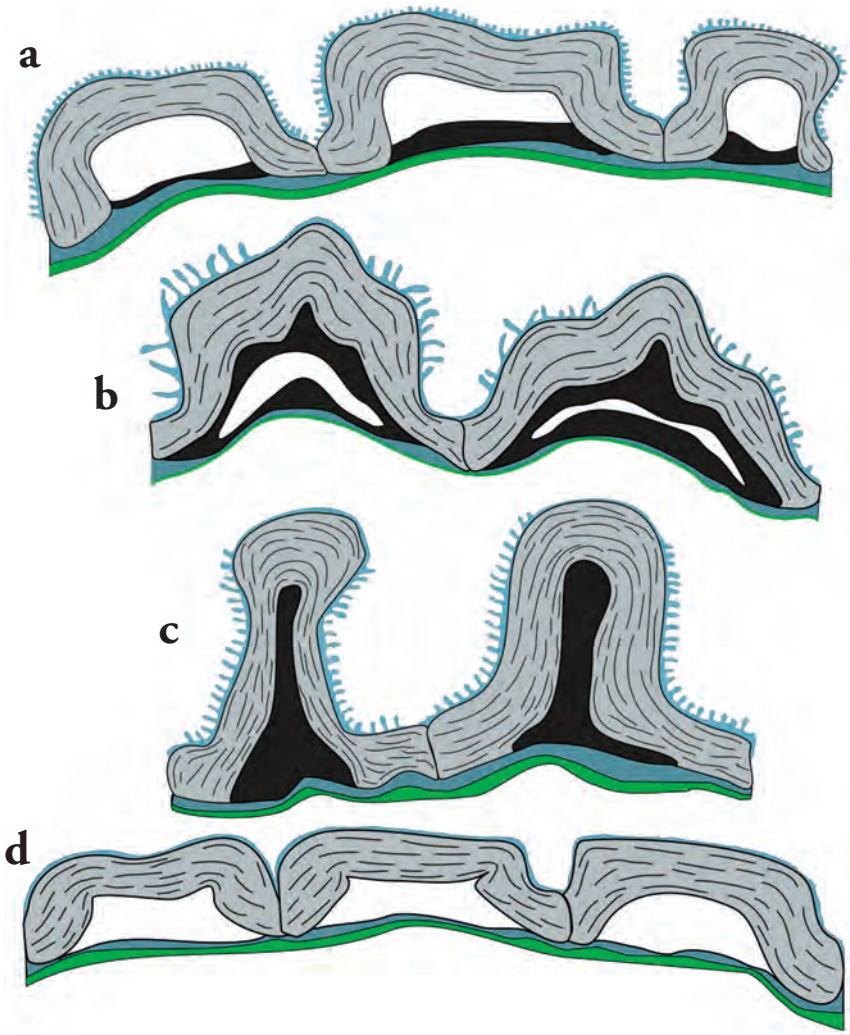

Figure 7. Testa structure. a – *M. glomerata*; b – *M. meyeri*; c – *M. setacea*; d – *M. sclerantha*.

Specific morphological features were not revealed for most of the new genera proposed by the authors. However, it can be noted that some of the species examined in this work have a clear pattern in the structure of the seed coat which does not fit into the old system of the genus. According to the old classification, *M. hybrida* and *M. verna* belonged to section *Minuartia* and section *Tryphane*, respectively. Molecular data indicate the sister status of these species. The morphology of these plants is different: *M. hybrida* is an annual plant covered with rare glandular hairs, with narrow leaves only in the upper part, petals are twice or ⅓ shorter than the calyx, sepals are lanceolate, pointed and the capsule is equal to the calyx in length. *Minuartia verna* is perennial with a lignified lower part of the shoot, usually the leaves and peduncle are covered with glandular hairs, the sepals are ovoid with a short point and the petals are equal to or are ¼ longer than the calyx. The structure of their seed coat is significantly different from the other studied representatives and is very similar to each other. The brown coloured seeds of *M. hybrida* and *M. verna* have a kidney-shaped form and a serrated border between cells. The anatomical structure of the testa can serve as the significant feature for these species. The presence of a specific thickening of exotestal cells, which looks like papillae, is only typical of *M. hybrida* and *M. verna*.

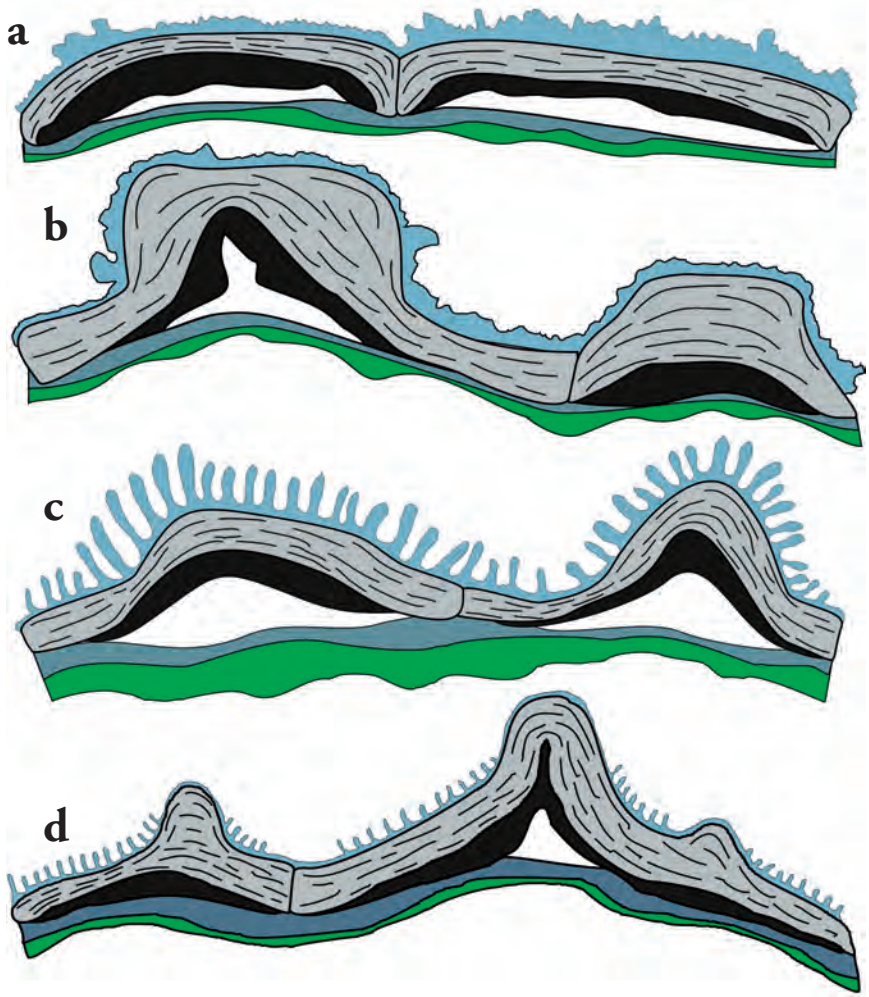

Figure 8. Testa structure. a, b – *M. wiesneri* (a – section in the region of the lateral surface of the seed, b – section in the area of the back of the seed); c – *M. oreina*; d – *M. hybrida*.

The cuticle is also represented by the same micropapillae structures. The combination of all of the above mentioned features is uniquely specific to *M. hybrida* and *M. verna*.

Two more species, *M. biebersteinii* and *M. lineata*, related to section *Acutiflorae*, possess a similar seed coat structure. The allocation of section *Acutiflorae* is due to a number of morphological features in the organization of their shoots: they have pointed sepals with three veins, the petals are inversely ovoid-elongated, 1.5 times longer than the sepals. Representatives of this section retained their close position in the molecular system of the genus. Seed structure also attests to their close relationship. The seeds of *M. biebersteinii* and *M. lineata* are kidney-shaped, brown, with verrucose ornaments, the cells are elongated and have serrated junctures between them. The exotesta cells have a strongly thickened trapezoidal wall, the cuticle forms micropapillae. The most atypical structure of the seed coat has *M. circassica*, which belonged to section *Spectabiles* ser. *Caucasicae* according to the old genus system. Molecular data show that *M. circassica* occupies a separate position and is included in a separate taxon, which Dillenberger & Kadereit (2014) proposed to call *Cherleria* L. The distinctive feature in the structure of *M. circassica* seeds is the

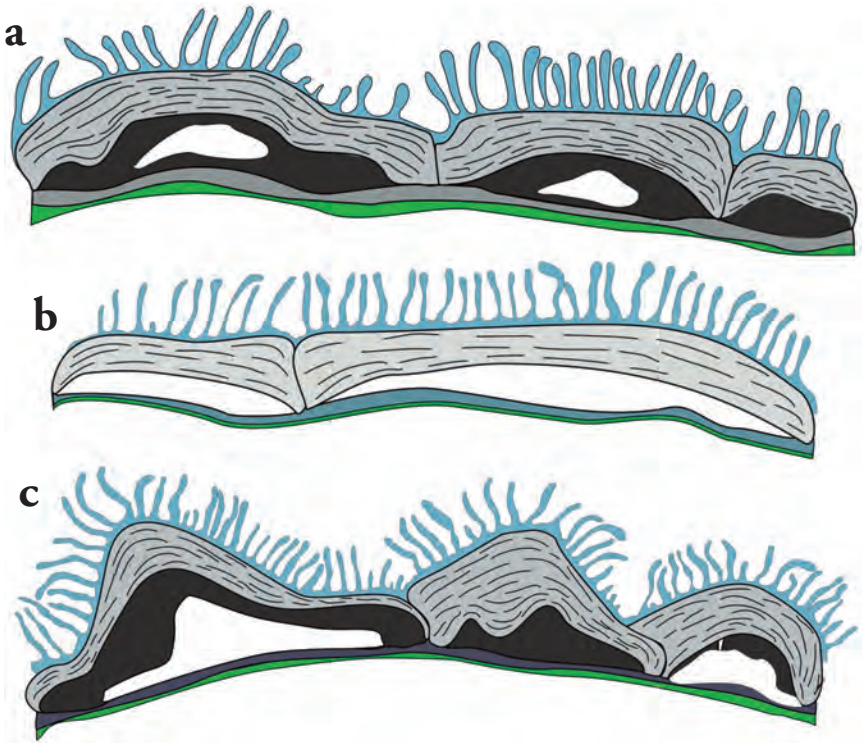

Figure 9. Testa structure. a – *M. aizoides*; b, c – *M. circassica*; (b – section in the region of the lateral surface of the seed, c – section in the area of the back of the seed).

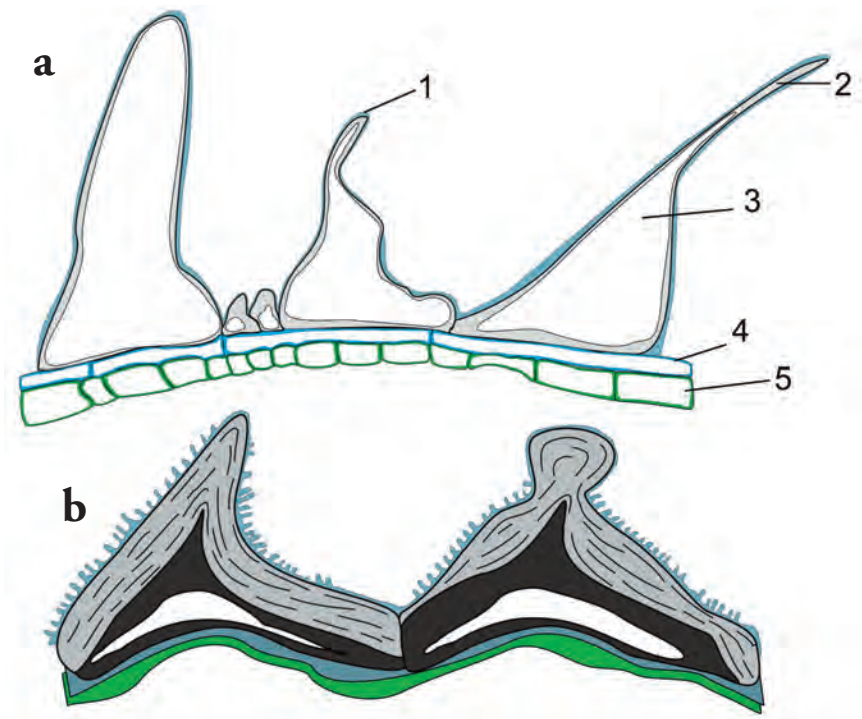

Figure 10. Testa structure. a – type of seed coat structure typical of section *Spectabiles* ser. *Laricinae* (*M. rhodocalyx*, *M. imbricata*, *M. inamoena*, *M. brotherana*); b – *M. verna*. 1 – cuticular layer; 2 – exotesta cell walls; 3 – exotesta cell cavity; 4 – endotesta; 5 – single-layered tegmen.

presence of direct connections between the cells, which indicates the presence of straight anticlinal walls in the exotestal cells, while all other studied representatives of *Minuartia* have anticlinal walls that are more or less distorted. In addition, it can be noted that there is a difference between the types of exotesta cells on the back (conical) and on the lateral surface (planar) of the seed. The cuticle forms very large trichome-like micropapillae which form a verrucose-reticulated ornament on the surface of the seed.

According to the system proposed by McNeill (1962, 1963), the subsection *Minuartia* included *M. akinfievii*, *M. meyeri*, *M. wiesneri* and *M. setacea*. All of the above-mentioned species retained their closely related positions in the phylogenetic classification of the genus and entered *Minuartia* s.str., *M. oreina* which was earlier referred to section *Plurinerviae* also entered *Minuartia* s.str. Currently, neither morphological nor karyotypic features clearly distinguish representatives of *Minuartia* s.str. Based on the results of this work, the existence of a unified structural plan for the seed coat in *Minuartia* s.str. cannot be clearly stated, but some trends can be observed. The presence of a massive, heavily thickened outer periclinal wall in the exotesta that may form into different shapes (conical, trapezoidal, dome-shaped) is typical of all species. The cuticle is represented by different structures, but it is always thick.

The greatest difference in the structure of the seed coat can be seen in section *Spectabiles* which includes *M. rhodocalyx*, *M. imbricata*, *M. inamoena*, *M. brotherana* and *M. aizoides*. Phylogenetic studies have revealed the closely related position of *M. brotherana* and *M. aizoides*. However, the thickening of the outer periclinal wall in all of the exotesta cells in *M. aizoides* has an atypically flat structure. The outer wall forms papillous structures in other species. This group has a separate position in the molecular tree and, according to the authors, is treated as a new taxon. It would be interesting to study the anatomical features of the seed coat structure in a larger group of representatives of *Minuartia* as well as closely related taxa, and also to compare these data with molecular genetic studies to accurately establish the connection between carpological features and systematic position of the taxa.

Acknowledgements

We gratefully acknowledge Professor Dmitriy Sokoloff for discussion of the manuscript and a valuable advice.

The study was performed in the framework of a government order to Moscow State University (AAAA-A16-116021660045-2) and was funded by Grant # 14-50-00029 'Scientific basis of the national biobankdepository of the living systems' from Russian Science Foundation (RNF).

References

Al-Saadi S.A.A.M. & Al-Taie S.S. (2014): Seed micromorphological of some species *Minuartia* L. (Caryophyllaceae) in Iraq. – Int. J. Agriculture Sci. **6**(1): 393–398.

Amini E., Zarre S.H. & Assadi M. (2011): Seed micromorphology and its systematic significance in *Gypsophila* (Caryophyllaceae) and allied genera. – Nordic J. Bot. **29**: 660–669.

Bittrich V. (1993): Caryophyllaceae. – In: Kubitzki K., Rohwer J.G. & Bittrich V. [eds]: The families and genera of vascular plants. Vol. 2, Flowering plants Dicotyledons: Magnoliid, Hamamelid, and Caryophyllid families: 206–236. – Berlin: Springer.

Celebioğlu T., Favarger C. & Huynh K.L. (1983): Contribution a la micromorphologie de la testa des graines du genre *Minuartia* (Caryophyllaceae) I. sect. *Minuartia*. – Adansonia **4**: 415–435.

Crow G. E. (1979): The systematic significance of seed morphology in *Sagina* (Caryophyllaceae) under scanning electron microscopy. – Brittonia **31**: 52–63.

Devyatov A. G. (1991): O tipah vskryvaniya korobochek v tribe *Lychnideae* Fenzl in Endlicher (Caryophyllaceae). – Byull. Moskovsk. Obshch. Isp. Prir., Otd. Biol. **5**: 64–69. [In Russian]

Devyatov A. G. (1996): Ob osnovnyh napravleniyah ehvolyucii plodov v semejstve gvozdichnyh. – Materialy IX Moskovskogo soveshchaniya po filogenii rastenij. M: 44–47. [In Russian]

Devyatov A. G. & Tajsumov M. A. (1999): Raznoobrazie ul'trastuktury poverhnosti plodov korobochek podsemejstva *Silenoideae* sem. Caryophyllaceae. – In: Tr. VI Mezhdunar. konf. po morfologii rastenij pamyati I. G. i T. I. Serebryakovyh. M.: 73–75. [In Russian]

Dillenberger M. S. & Kadereit J. (2014): Maximum polyphyly: Multiple origins and delimitation with plesiomorphic characters require a new circumscription of *Minuartia* (Caryophyllaceae). – Taxon **63**(1): 64–88

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

Greenberg A. K. & Donoghue M. J. (2011): Molecular systematics and character evolution in Caryophyllaceae. – Taxon **60**: 1637–1652.

Halliday G. (1993): *Minuartia* L. – In: Tutin T. G., Burges N. A., Chater A. O., Edmondson J. R., Heywood V. H., Moore D. M., Valentine D. K., Walters S. M. & Webb D. A. [eds]: Flora Europaea. Vol. 1 [2nd ed.]: 152–160. Cambridge: Cambridge University Press.

Harbaugh D. T., Nepolroeff M., Rabeler R. K., McNeill J., Zimmer E. A. & Wagner W. L. (2010): A new lineage based tribal classification of the family Caryophyllaceae. – Int. J. Pl. Sci. **171**: 185–198.

Hernández-Ledesma P., Berendsohn W.G, Borsch T., Von Mering S., Akhani H., Arias S., Castañeda-Noa I., Eggli U., Eriksson R., FloresOlvera H., Fuentes-Bazán S., Kadereit G., Klak C., Korotkova N., Nyffeler R., Ocampo G., Ochoterena H., Oxelman B., Rabeler Adriana Sanchez R. K., Schlumpberger B. O. & Uotila P. (2015): A taxonomic backbone for the global synthesis of species diversity in the angiosperm order Caryophyllales. – Willdenowia **45**(3): 281–383.

Kamari G. (1997): *Minuartia* L. – In: Strid A. & Tan K. [eds]: Flora Hellenica. Vol. 1: 170–191. – Königstein: Koeltz Scientific Books.

Kozhanchikov V. I. (1975): Morfologicheskie priznaki semyan *Caryophyllaceae* i vozmozhnye puti ih ehvolyucii. – Bot. Zhurn. **52**(7): 1277–1286. [In Russian]

Lazkov G. A. (2012): *Minuartia*. – In: Takhtajan A. L. [ed.]: Konspekt flory Kavkaza. Tom. 3(2): 165–170. – St. Petersburg: St. Peterburgskogo Univ. [In Russian]

Linnaeus C. (1753): Species plantarum. Tom. I. – Holmiae: Salvius.

Mattfeld J. (1922): Geographisch-genetische Untersuchungen über die Gattung *Minuartia* (L.) Hiern. – Repert. Spec. Nov. Regni Veg. Beih. **15**.

McNeill J. (1962): Taxonomic studies in the Alsinoideae I: Genetic and infrageneric groups. – Monthly Notes Roy. Bot. Gard. Edinburgh **24**: 79–155.

McNeill J. (1963): Taxonomic studies in the Alsinoideae II: A revision of the species in the Orient. – Monthly Notes Roy. Bot. Gard. Edinburgh **24**: 241–404.

Minuto L., Fior S., Roccotiello E. & Casazza G. (2006): Seed morphology in *Moehringia* L. and its taxonomic significance in comparative studies within the Caryophyllaceae. – Pl. Syst. Evol. **262**: 189–208.

Minuto L., Roccotiello E. & Casazza G. (2011): New seed morphological features in *Moehringia* L. (Caryophyllaceae) and their taxonomic and ecological significance. – Pl. Biosyst. **145**: 60–67.

Mostafavi G., Assadi M., Nejadsattari T., Sharifnia F. & Mehregan I. (2013): Seed micromorphological survey of the *Minuartia* species (Caryophyllaceae) in Iran. – Turkish J. Bot. **37**(3): 446–454.

Rabeler R. K., Hartman R. L. & Utech F. H. (2005): *Minuartia*. – In: Flora of North America Editorial Committee [eds]: Flora of North America, vol. 5: 116–136. – Oxford: Oxford University Press.

Rechinger K. H. (1988): *Minuartia* L. – In: Rechinger K. H. [ed.]: Flora Iranica. Vol. 163: 28–53. – Graz: Akademische Druck- und Verlagsanstalt.

Yildiz K. (2002): Seed morphology of Caryophyllaceae species from Turkey (North Anatolia). – Pakistan J. Bot. **34**: 161–171.

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

Addresses of the authors:

Sofia G. Zaychenko
Lomonosov Moscow State University
Faculty of Biology
Department of Higher Plants
Leninskie Gory 1 (12)
119234 Moscow
Russia
E-mail: zaychenko_so@mail.ru

Alexander S. Zernov (corresponding author)
Lomonosov Moscow State University
Faculty of Biology
Department of Higher Plants
Leninskie Gory 1 (12)
119234 Moscow
Russia
Moscow Region State University
Faculty of Biology and Chemistry
Department of Botany and Applied Biology
Radio str. 10a
105005 Moscow
Russia
E-Mail: zernov72@yandex.ru