Acta Biologica Sibirica
Vol 10 (2024): Acta Biologica Sibirica, 1573–1588
Articles

Anatomy of leaves of Siberian species of the genus Lilium in conditions of Western Siberia forest zone

Tatiana N. Belaeva Siberian Botanical Garden, National Research Tomsk State University, Tomsk, Russia
Liu Si Siberian Botanical Garden, National Research Tomsk State University, Tomsk, Russia
Alina N. Butenkova Siberian Botanical Garden, National Research Tomsk State University, Tomsk, Russia
Alexey S. Prokopyev Siberian Botanical Garden, National Research Tomsk State University, Tomsk, Russia

Siberia harbors four lily species. The number of Siberian lily populations has declined in recent years due to habitat disturbance caused by anthropogenic impact, including tourism, urban expansion, economic activities, and active harvesting of flowering plants for bouquets. The aim of this study was to conduct a comparative analysis of the leaf anatomical structure of three Siberian species of the genus *Lilium* (*L. pensylvanicum* Ker-Gawler, *L. pilosiusculum* (Freyn) Miscz. and *L. pumilum* DC.), successfully introduced into the Siberian Botanical Garden of Tomsk State University, in order to identify their adaptive potential, including water stress tolerance and insolation in culture. The values of indicators from different species were compared by one-way analysis of variance (ANOVA). Reliable differences were revealed between the studied species in 13 quantitative parameters of the anatomical structures: the number and size of epidermal cells, leaf thickness, thickness of the lower and upper epidermis, mesophyll, columnar mesophyll, and the width of the cells of the upper mesophyll layer. In the conditions of the forest zone of Western Siberia, the studied species exhibit different water stress tolerance: from xerophyte *L. pumilum* to mesophyte *L. pensylvanicum* and more broad-leaved mesophyte *L. pilosiusculum*. Among the studied lilies, *L. pumilum* is a sun-loving plant, *L. pilosiusculum* is a shade-tolerant plant, and *L. pensylvanicum* occupies an intermediate position. This modern anatomical study of lilies in the forest zone of Western Siberia was conducted for the first time, which made it possible to obtain original quantitative characteristics of the epidermis and mesophyll of the leaf blades of plants brought into culture from natural habitats in Northern Asia.

Acta Biologica Sibirica 10: 1573–1588 (2024)

doi: 10.5281/zenodo.14329047

Corresponding author: Alina N. Butenkova (sbg-flowers@yandex.ru)

Academic editor: R. Yakovlev | Received 11 November 2024 | Accepted 5 December 2024 | Published 11 December 2024

http://zoobank.org/9C008162-9F2E-480B-827C-8499047F898C

Citation: Belaeva TN, Si L, Butenkova AN, Prokopyev AS (2024) Anatomy of leaves of Siberian species of the genus *Lilium* in conditions of Western Siberia forest zone. Acta Biologica Sibirica 10: 1573–1588. https://doi.org/10.5281/10.5281/zenodo.14329047

Keywords

Adaptive potential, leaf anatomy, *Lilium*, Siberian rare species, water stress tolerance, biodiversity conservation, insolation

Introduction

The genus *Lilium* L. (family Liliaceae) comprises 105 species of polycarpic bulbous plants distributed in the northern hemisphere, mainly in temperate and subtropical regions of Europe, Asia and North America (Baranova 1999).

The center of origin for the genus is probably East Asia (South-West and Central China). Lily species are bulbous geophytes, predominantly mesophytes, thriving in forested, well-watered habitats (Baranova 1999; Songyun and Tamura 2000). V.K. Negrobov (1981) identified two primary categories of ecological adaptation within the genus *Lilium*: mesophytic and hygromesomorphic (typical of the majority of North American lily species) and xeromorphic (some European and Northeast Asian species). M.V. Baranova (1990) categorized lilies as a specialized life form of polycarpic perennial bulbous plants, some of which are adapted to unfavorable temperature conditions.

The taxonomy of the genus *Lilium* as a whole and its individual species is under development in connection with the accumulation of modern data from molecular genetic studies. The currently accepted concept of the phylogenetic and taxonomic system of the genus is developed based on an analysis of genetic, geographic, and morphological data (Pelkonen and Pirttilä 2012), according to which the genus is divided into 7 main groups.

Lily species and their cultivars are valuable ornamental plants (Sorokopudova 2019; Zhou et al. 2021). The growing popularity of lilies is due to the development of new hybridization methods and breeding successes (Pelkonen and Pirttilä 2012). Over the past 50 years, lilies have been distributed worldwide as incredibly popular garden and cut flowers (Tuyl et al. 2018).

Siberia harbors four lily species: *L. buschianum* Lodd., *L. pensylvanicum* Ker-Gawler, *L. pumilum* DC., and *L. pilosiusculum* (Freyn) Miscz. (Vlasova 1987). One species grows in the Tomsk region: *Lilium pilosiusculum* (Zverev 2014). Three species are cultivated in the Siberian Botanical Garden: *L. pensylvanicum*, *L. pilosiusculum* and *L. pumilum*.

Siberian lily species represent a valuable source for selection due to their hardiness and resistance to viral and fungal diseases (Kireeva et al. 1992). Siberian lily species are used in folk medicine in China, Tibet, Mongolia, Siberia, and the Far East and are promising for the development of novel pharmaceuticals (Bokov et al. 2019). For centuries, bulbs of *L. pilosiusculum* and other Siberian species have been eaten raw, boiled, fried, dried, and used as a spice, coffee substitute, and flour (Telyatiev 1985; Sokolov 1994; Ståhlberg and Svanberg 2010).

The number of Siberian lily populations has declined in recent years due to habitat disturbance caused by anthropogenic impact, including tourism, urban expansion, economic activities, and active harvesting of flowering plants for bouquets. Additionally, digging for bulbs with subsequent replanting in garden plots has contributed to this decline (Polyakov 2017; Trofimova 2020; Senchik and Malikova 2009). One of the basic strategies for preserving plant genetic diversity *exsitu*is their cultivation in botanical garden collections. Plant cultivation is important for the conservation of species biodiversity, the creation of reserve potential with subsequent reintroduction into natural populations, the development of cultivation principles for wider use in regional landscaping and the production of new sources of medicinal raw materials.

Considering the importance of Siberian representatives of the genus *Lilium* for landscaping and selection, as well as the need to preserve them in natural conditions, the relevance of an indepth study of their biological and anatomical-morphological features in nature and culture is beyond doubt.

The structural parameters of the leaf, as an organ of transpiration and photosynthesis, are the most informative indicators in comparative studies of plants from different ecological groups, allowing us to assess their ecological plasticity and adaptive potential (Gamaley 2004; Khramchenkova 2016). During the introduction stage, quantitative parameters of the leaf epidermis and mesophyll undergo specific changes within the reaction norm, which define them as dynamic systems that allow the species to adapt to new ecological conditions (Sedelnikova 2002). The area, thickness and type of leaf anatomy are less correlated with the ecological properties of plants compared to structural indices of the photosynthetic tissues (Ivanova 2014).

Limited data on the anatomical structure of leaves of Siberian lilies are reported in works by E.P. Nemchenko and V.C. Novikov (1979), M.V. Baranova (1990), O.A. Sorokopudova (2003), S. Güven et al. (2014). E.P. Nemchenko and V.S. Novikov (1979) analyzed the anatomical structure of leaf blades of 15 lily species, cultivated in the Moscow State University Botanical Garden, which, in their opinion, not only supplemented the diagnostics of taxa, but also made it possible to judge the family relationships within the genus. Common features characteristic of the leaf blades of the studied species were established: a single-layer epidermis covered with a layer of cuticle; stomata without accompanying cells, located in the overwhelming majority of species on the lower side, the presence of bladder-like cells in the epidermis, and a collateral type of vascular bundles. The studied lily species are subdivided into a polymorphic group of narrow-leaved and more broad-leaved species, which include *L. pumilum* and *L. pensylvanicum*, and a group of broad-leaved lilies, which includes *L. martagon*.

M.V. Baranova (1990) divided all lily species into three groups depending on the profile of the abaxial epidermal cell walls: 1) deeply, irregularly sinuous; 2) uniformly wavy; 3) straight linear or barely wavy. The author emphasized that the structure of the epidermal tissue can be used not only to characterize lily species, but also to establish close relations between larger intrageneric subdivisions.

O.A. Sorokopudova (2003) studied the anatomy of leaf blades of 11 species and 13 varieties of lilies grown at the Novosibirsk Zonal Station and in the Central Siberian Botanical Garden (Novosibirsk). She considers the differences in the anatomical structure of lily species in Moscow and the Novosibirsk region to be unimportant and explains them by the polymorphism of the species, different agroclimatic conditions in the places of introduction and classified *L. pumilum* as the most xero- morphic species of the genus.

Over the past 20 years, the anatomy of some tropical lily species, which are rare endemic and valuable medicinal plants, has been studied with the aim of developing the principles of their cultivation in open and closed ground (Dhyani et al. 2009; Embaturova and Korchagina 2011; Paltiyan-Bugtong et al. 2023). Such studies are also important for the identification of plant materials (Dhyani et al. 2009). S. Güven et al. (2014) studied the anatomy of vegetative organs of 8 lily species in Turkey and found hypostomate leaves with anomocytic stomata in all studied species.

Unfortunately, these publications do not provide a detailed quantitative assessment of various anatomical parameters of leaf different lily species, with the exception of the number and size of stomata and the thickness of the leaf blade.

In general, all authors point to the limitations of research and the need to ac- cumulate data on the anatomy of lily species in various growing conditions, taking into account the prospects for their use in plant growing and the importance of developing scientifically based technologies for their cultivation in different regions. The aim of this study was to conduct a comparative analysis of the leaf anatomical structure of three Siberian species of the genus *Lilium*, successfully introduced into the Siberian Botanical Garden of Tomsk State University, in order to identify their adaptive potential, including water stress tolerance and insolation in culture.

This study will enhance identification of structural adaptation of plants during introduction with regard to their potential use in landscaping and medicine, and concerns related to biodiversity conservation. Such studies are of particular importance in connection with predicted climate changes in the natural habitats of plants, as well as assessments of the possibility of adaptation of species in various natural and climatic zones upon introduction.

This modern anatomical study of lilies in the forest zone of Western Siberia was conducted for the first time, which made it possible to obtain original quantitative characteristics of the epidermis and mesophyll of the leaf blades of plants brought into culture from natural habitats in Northern Asia.

Materials and methods

The study was conducted in 2023–2024 in the Siberian Botanical Garden of Tomsk State University, in the forest natural-climatic zone of Western Siberia. The climate of Tomsk is sharply continental, exhibiting a prolonged winter season, a return of spring cold, early autumn frosts, and a brief, yet warm, summer period (Koshinsky 1982).

The mean annual air temperature is –0.8 °C (Pilnikova 1993). The precipitation levels (on average 517 mm per year) indicate that Tomsk and its environs are located in the zone of moderate humidity (Koshinsky 1982). The study focused on three Siberian species of the genus *Lilium*.

Lilium pilosiusculum (Freyn) Miscz., section Martagon Duby (Baranova 1990; Pelkonen and Pirttilä 2012), is a Siberian-Central Asian endemic with a disjunctive range (Galanin et al. 2008). The species is distributed throughout Siberia, North Mongolia, and North-West China (Xinjiang), thriving in both mountainous and lowland forest areas (Vlasova 1987; Songyun and Tamura 2000). M.V. Baranova (1990) reported its occurrence in the Urals and the European part of Russia (Vyatka and Kama River basins). The species grows in coniferous and mixed forests, birch groves, mountain forests, forest glades, and in herbaceous and subalpine meadows (Vlasova 1987). The ecological optimum of the species on the soil moisture gradient is 61.5 (Korolyuk 2006), which is greater than that of the previous species, but also corresponds to the moisture content of dry and fresh meadows and forests according to the ecological scale of L.G. Ramensky (1956) (range 53–63).

A significant number of scientists consider *L. pilosiusculum* (Freyn) Miscz. as a subspecies (or variety) of the polymorphic Eurasian species *L. martagon* L., which differs from the typical European taxon. Its leaves differ in a narrower lanceolate, linear lanceolate (rather than wide lanceolate) shape. The arachnoid pubescence is persistent on its bracts, the upper part of the young generative shoot, and the outer perianth tepals (Baranova 1990; Songyun and Tamura 2000; Pelkonen and Pirttilä 2012). The study of the genome of *L. martagon* L. var. *pilosiusculum* by Chinese scientists (Zhou et al. 2018) supports the conclusion that the taxon is distinct.

The species is included in the Red Data Books of Transbaikal Territory (Polyakov 2017), the Republic of Sakha (Yakutia) (Danilova 2017), Khanty-Mansiysk Autonomous Region (Vasin and Vasina 2013), and Yamalo-Nenets Autonomous Region (Ektova and Zamyatin 2010).

For TSU SibBG collection, the plant was harvested in the vicinity of Anikino settlement (Tomsk district). It is cultivated at the expositions of rare and ornamental plants of TSU SibBG in illuminated areas. The species is resistant to cultivation.

Lilium pensylvanicum Ker-Gawler, also known as *L. pensylvanicum* or Daurian, section Pseudolirium Wilson (Baranova 1990; Pelkonen and Pirttilä 2012), is a Siberian-East Asian species native to both Siberia and East Asia. The species is distributed in Central and Eastern Siberia, extending from the Yenisei River basin to the Kamchatka Peninsula, the Kuril Islands, and Sakhalin Island in the Far East. Its northernmost locality is in the lower reaches of the Lena River, situated at 71°N. Outside of Russia, it is found in North-East Mongolia, North-East China, Japan (Honshu and Hokkaido islands), and the Korean peninsula. The species grows in floodplain and forest meadows, on the edges of larch and birch forests, in sparse bushes, and on gravels. In the mountain forest belt, it inhabits mountain meadows (Govorina et al. 1986; Vlasova 1987). The ecological optimum of the species on the soil moisture gradient is 55.5 (Korolyuk 2006), which corresponds according to the ecological scale of L.G. Ramensky (1956) (range 53–63) to the moisture content of dry and fresh meadows and forests, that is, to drained habitats of the forest zone.

The polymorphism of the species is evidenced by variations in floral characteristics, including coloration, the size of flower-bearing shoots, the pubescence of the stem and flowers, the cross-sectional shape of the stem, the presence or absence of the anisotropic underground portion of the shoot, the structure of scales, and even the time of flowering (Baranova 1990).

The species is included in the Red Data Books of Krasnoyarsk Territory (Stepanov 2022), the Republic of Buryatia (Anenkhonov 2023), Sakha (Yakutia) (Danilova 2017), Irkutsk Region (Trofimova 2020), Transbaikal Territory (Polyakov 2017), Jewish Autonomous Region (Rubtsova and Schlotthauer 2019), Kamchatka Territory (Chernyagina 2018), and Magadan Region (Kondratyev 2019).

In 2015, the Siberian Botanical Garden enlarged its collection with plants from the Transbaikal Territory, which exhibited distinctive, brick-red flowers. The species is cultivated at the exposition of rare plants in small plots in illuminated areas. The species is resistant to cultivation.

Lilium pumilum Delile, also known as dwarf lily, is native to Asia. It is classified within the section Sinomartagon Comber (Baranova 1990; Pelkonen and Pirttilä 2012).

The species grows in the Siberian and East Asian regions. It is distributed in Middle and Eastern Siberia, and in the Far East (Vlasova 1987). The western limit of its distribution is located in the Krasnoyarsk Territory, Khakassia, and Tuva. Outside of Russia, it is found in Mongolia, North-East China, and the Korean Peninsula (Vlasova 1987; Songyun and Tamura 2000). The species is confined to mountainous and foothill areas, including forest glades, steppe meadows, dry stony and rubbly slopes, screes, rocks, and meadow steppes and shrub thickets (Vlasova 1987; Baranova 1990). The ecological optimum of the species on the soil moisture gradient is the lowest among the studied species – 54 (Korolyuk 2006).

According to O.A. Sorokopudova (2019), drought tolerance of *L. pumilum* is due to low stature, the presence of small and dense semi-tunicate bulbs with a small number of scales, and obliquely vertically arranged leaves. V.N. Negrobov (1981) refers *L. pumilum* to xerophytes, while A.M. Zarubin et al. (1990) refers this species to xeromesophytes.

It is included in the Red Data Books of the Republics of Tyva (Ondar and Shaulo 2019) and Khakassia (Ankipovich 2012), Krasnoyarsk Territory (Stepanov 2022), Transbaikal Territory (Polyakov 2017), Irkutsk Region (Trofimova 2020), Amur Region (Senchik and Malikova 2020), Jewish Autonomous Region (Rubtsova and Schlotthauer 2019), and Khabarovsk Territory (Voronov 2019).

In 2016, the species was brought to the Siberian Botanical Garden from the Republic of Khakassia (vicinities of Lake Belyo). It is cultivated at the exposition of rare plants in small plots with a total area of about 2 m^2 in a sunny area with drained soils.

The species is drought-resistant.

The materials employed for the study were the stem leaves of three lily species collected during the mass flowering stage. The leaves were taken from the middle portion of the shoot. For analysis of the epidermal structures, the formed leaf were used.

The anatomical structure of leaves was studied using conventional techniques (Vehov at al. 1980) and as outlined by K. Ezau (Ezau 1980a; Ezau 1980b) and A.A. Pautov (2003; 2012). The stomatal index was calculated using the formula proposed by A. Kästner (1972). The stomatal types were classified using the method by M.A. Baranova (1985).

The methodological basis for assessing the ecological adaptations of plants to various growing conditions was the publications of N.K. Bordman (1977), T.K. Goryshina (1979), T.J. Givnish (1988), L.A. Ivanova (2014).

For preparations, sections were cut from the central portion of the leaves using a freezing microtome (MZ-2). The section thickness was of 60–90 μm. Anatomical structures were examined on leaf cross-sections using a light microscope. Cross-sections were prepared from the central portion of the leaf, with five repetitions per plant, using leaves from five shoots; not less than 30 sections of each plant were analyzed (Zhukova and Minets 2020). The number of cells and stomata per 1 mm^2 of epidermis was determined after separation of skin in the middle part. Microscopic cross-sections of leaves and microscopic measurements were obtained using a Carl Zeiss Axio Lab A1 light microscope equipped with an AxioCam ERc 5s digital camera connected to a computer via the Axio Vision 4.8 software.

The obtained measurement data were processed using Statistica 8.0. The following parameters were identified: arithmetic mean M (Mean); standard error of the arithmetic mean m (SE); standard deviation σ; the coefficient of variation CV; minimum and maximum values Min–Max. Anatomical indicators were considered lowly variable with the coefficient of variation CV < 20%, moderately variable with CV = 20–40%, and highly variable with CV > 40% (Butnik and Timchenko 1987).

The indicators of different species were compared by the one-way ANOVA to determine the statistical significance of the differences (Zhukova and Minets 2020) using Statistica 8.0 (2008). Statistically significant differences were determined at $p< 0.05$.

Results and discussion

Green assimilating leaves of the studied lily species are located on an elongated generative shoot, with spiral (*L. pensylvanicum*, *L. pumilum*) or false verticillate (*L. pilosiusculum*) leaf arrangement. In the latter case, elongated internodes alternate with several significantly shortened ones, resulting in a whorled form of leaves on the shoot, with one leaf per node (Baranova 1999). As evidenced by available literature, the verticillate leaf arrangement in the genus *Lilium* is relatively constant and serves to distinguish related groups of species. Leaf venation is parallel (Baranova 1990).

Lilium pilosiusculum leaves are linear lanceolate or lanceolate. In the middle part of the stem, 11.7–15.0 cm long and 1.6–2.2 cm wide, acuminate, narrowed to the base, margins cartilaginous, dentate. *L. pensylvanicum* exhibits alternate leaf arrangement, leaves narrow lanceolate, 12.2–16.7 cm long and 1.3–1.9 cm wide. Leaves of *L. pumilum* are xeromorphic, numerous, narrowest among the studied species, linear, uninervate, 6.2–10.6 cm long and 0.1–0.3 mm wide in the middle part of the shoot (Fig. 1).

Lilium pensylvanicum exhibits the largest size of cells of adaxial and abaxial epidermis. The cells of adaxial epidermis are larger than those of abaxial epidermis, which is inherent to monocotyledonous plants (Nemchenko and Novikov 1979). The highest number of cells of adaxial epidermis per 1 mm^2 was found in *L. pumilum*, which 1.5–1.8 fold exceeds that in the other two species (see Suppl. material 1: Table 1). The bulliform (bubble-shaped) cells of the ground tissue were found above the veins and along the leaf margins in all species studied, and fringed formations could be observed along the leaf margins.

Stomata of the three species were quite large, with an average length and width of 78.0–105.3 µm and 55.4–60.5 µm, respectively (see Suppl. material 1: Table 1), and the type of stomatal apparatus was anomocytic, which is consistent with available literature data (Baranova 1990; Sorokopudova 2003). *L. pilosiusculum* and *L. pensylvanicum* leaves are hypostomatic, and *L. pumilum* leaves are amphistomatic.

The highest stomatal density, as expected, was found in *L. pumilum* (on average 65.6 stomata per mm^2 on abaxial epidermis and 39.7 stomata per mm^2 on adaxial epidermis) against an average of 40.8–44.3 stomata per mm2 on abaxial epidermis in the other species (see Suppl. material 1: Table 1). A significant number of stomata on adaxial and abaxial epidermis of *L. pumilum* ensures intensive transpiration that significantly reduces leaf temperature and likely permits photosynthesis at high air temperature. The stomata on adaxial and abaxial epidermis of leaves of *L. pumilum* are close in size, yet the average number of stomata on abaxial epidermis is 1.7 fold greater than that on adaxial epidermis. The location of stomata mainly on the abaxial side of the leaf is due to slower water loss during transpiration on this side compared to adaxial epidermis.

The conducted studies revealed reliable differences between the studied species in 13 quantitative parameters of the anatomical structures of the studied lily species: the number and size of epidermal cells, leaf thickness, thickness of the lower and upper epidermis, mesophyll, columnar mesophyll, and the width of the cells of the upper mesophyll layer. *L. pumilum* significantly differed from the other two species in the number of stomata on the lower epidermis, leaf thickness in the area of the central vein, *L. pilosiusculum* – in the values of the stomatal index, the thickness of the spongy mesophyll, and the length of the cells of the upper mesophyll layer (see Suppl. material 1: Table 1).

The average values of the abaxial stomatal index of leaves varied from 35.3 % to 42.1 % (see Suppl. material 1: Table 1), which is within the range reported for some Turkish species of the genus *Lilium* (26.5–44.8 %) (Güven et al. 2014). The adaxial stomatal index of *L. pumilum* averaged 39.8 %.

The largest abaxial stomata were found in *L. pensylvanicum*, and the smallest stomata were observed in *L. pilosiusculum*. The sizes of stomata within one species varied slightly (from 4.5 to 14.8 %) and, according to data by O.A. Sorokopudova (2003), are a good diagnostic marker for determining the basic ploidy level at early stages of ontogenesis.

The thinnest leaf and the smallest mesophyll thickness were found in *L. pilosiusculum*, and the thickest ones were observed in *L. pumilum* (Suppl. material 1: Table 1).

The leaves of both *L. pilosiusculum* and *L. pensylvanicum* are dorsoventral. The mesophyll consists of 6–8 layers, differentiated into columnar (1–2 layers) and spongy (5–6–7 layers) parenchyma. In cross-section, cells of spongy mesophyll are large, loose, with large intercellular spaces, rounded, oval, or irregularly shaped; this structure facilitates water transpiration (Fig. 1).

The palisade parenchyma cells are typically of irregular shape, sometimes with finger-like outgrowths (referred to as branching cells, M.V. Baranova (1990)). The presence of finger-like outgrowths of palisade parenchyma cells in *L. martagon* was reported in S. Güven et al. (2014).

The leaves of *L. pumilum* are isolateral, covered with waxy plaque. Palisade tissue is found on both adaxial (1–2 layers) and abaxial (2 layers) sides (Fig. 1). The palisade parenchyma cells are elongated, rectangular. The intercellular spaces of spongy tissue are small.

The columnar mesophyll thickness in *L. pumilum* averages 188.2 μm, which is 1.4 fold greater than that in *L. pensylvanicum* and 1.7 fold greater than that in *L. pilosiusculum* (Suppl. material 1: Table 1). Leaf venation is curvinervate. Vascular bundles are surrounded by sclerenchyma and are provided with a multilayered parenchyma separating them from the mesophyll. Most epidermal parameters are weakly or moderately variable; the most variable are stomatal density (number of stomata per mm^2) of leaf abaxial epidermis (CV = 20.8–43.2%) and the cell size of abaxial epidermis (CV = 32.9–41.8%).

The following parameters vary least within the species: length (CV = 6.4–12.9 %) and width (CV = 4.5–6.6 %) of stomata, leaf thickness (CV = 7.7–11.4 %), leaf thickness in the vein area (CV = 6.8–17.5 %), mesophyll thickness (CV = 10–18.6 %) (Suppl. material 1: Table 1).

A comparison of the obtained results with available literature data (Nemchenko and Novikov 1979; Sorokopudova 2003) showed that Tomsk specimens of *L. pensylvanicum* and *L. pilosiusculum* showed a slightly thicker leaf compared to Moscow and Novosibirsk specimens: on average 552.3 μm vs. 448.8–453.0 μm and 411.2 μm vs. 363.0–382.0 μm, respectively. Tomsk specimens of *L. pumilum* (590.2 μm) were inferior to Moscow specimens (653.0 μm), but superior in this parameter to Novosibirsk specimens (456.0 μm).

The stomatal density of abaxial epidermis (65.6 pcs.) in *L. pumilum* growing in Tomsk is close to that growing in Novosibirsk (60.9 pcs.), but exceeds Moscow specimens 1.5 fold. Moscow and Novosibirsk specimens of *L. pensylvanicum* are virtually similar in stomatal density to Tomsk specimens (42.0–44.0 vs. 40.8 in Tomsk). The average values of stomatal density in Novosibirsk specimens of *L. pilosiusculum* amounted to 52.8 vs. 44.3 in Tomsk specimens. In Novosibirsk and Moscow, typical *L. martagon* was characterized by lower values of stomatal density (27 pcs.). Thus, the quantitative indices of stomata number and morphometric leaf thickness are variable and depend both on climatic and agrotechnical cultivation conditions and, probably, on the plant origin, especially in polymorphic species.

The study results indicate that *L. pilosiusculum* is a mesophyte and heliosciophyte (relatively large dorsoventral leaves, deeply convoluted epidermal walls, irregularly shaped palisade mesophyll cells, the thinnest leaf among the studied species, spongy mesophyll with developed intercellularity, etc.). As noted above, the stomatal density of this species is higher compared to the typical *L. martagon*. *L. pilosiusculum* requires regular moistening and enjoys partially shaded conditions. The species belongs to the most ancient mesophytic species of the genus *Lilium*, living in diffused light conditions (Baranova 1990).

Lilium pumilum exhibits the greatest xeromorphism and heliophyticity (the narrowest and isolateral leaves compared to other species, dense and small epidermal cells, maximum stomatal density of adaxial and abaxial epidermis). Mesophyll is characterized by tightly packed cells, developed columnar parenchyma and mechanical tissue, and poorly developed intercellular cells. These features of the anatomical structure of *L. pumilum* leaves are likely due to high insolation and lack of moisture, which contribute to the formation of adaptations that reduce transpiration rate and protect the leaf from overheating.

Lilium pensylvanicum is a heliophyte and mesophyte species with dorsoventral leaves, the smallest number of stomata on abaxial epidermis compared to other species, one or two-layered palisade mesophyll on the abaxial side of the leaf, large stomata, developed spongy mesophyll with well-defined intercellular cells, etc. The species exhibits a certain plasticity and some xeromorphic characters of the anatomical structure of leaves that reduce transpiration (wavy or straight epidermal cell walls, relatively thick leaves, thicker mesophyll compared to *L. pilosiusculum*), which allow plants to successfully adapt to conditions of temporary moisture deficiency in the forest-steppe zone.

Probably, mesophilic species of the genus *Lilium* in the process of adaptation to the cold and drier climate of Siberia, compared to the initial center of formation of the genus, acquired xeromorphic features. A similar phenomenon was noted by a number of Siberian scientists (Sobolevskaya 1977; Cheremushkina et al. 1992) for representatives of other generic complexes. Thus, according to K.A. Sobolevskaya (1977), species with a dual xeromesophilic nature, formed in the harsh conditions of the cryoxerophilic regime, had greater potential for adaptation to new environmental conditions and represent valuable material for introduction.

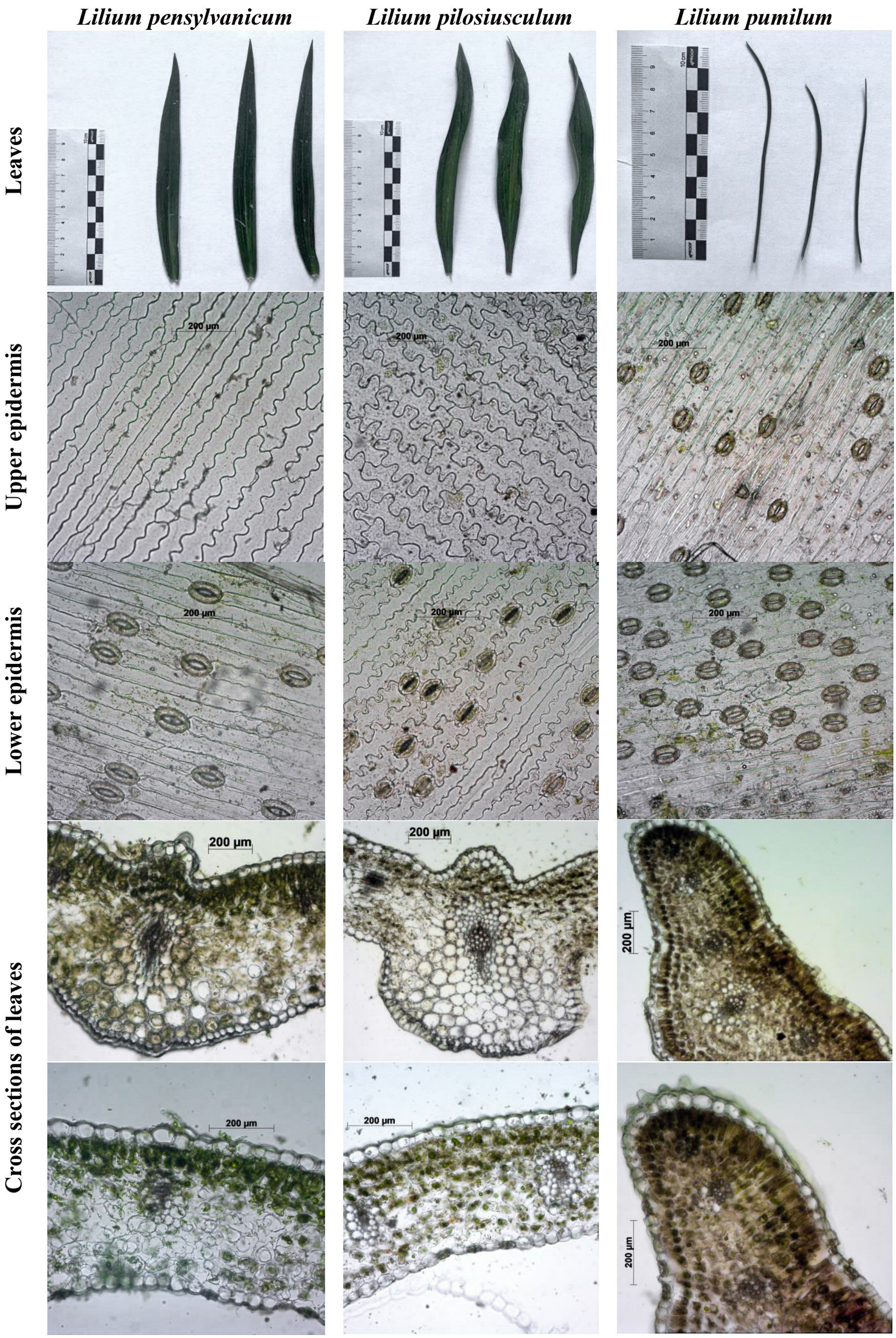
Lilium pensylvanicum
Lilium pilosiusculum
Lilium pumilum
Leaves
Upper epidermis
Lower epidermis
Cross sections of leaves
200 µm

Figure 1. *Appearance and anatomy of leaves of species of the genus Lilium*

Conclusions

The conducted studies revealed reliable differences between the studied species in 13 quantitative parameters of the anatomical structures of the studied species of lilies.

In the conditions of the forest zone of Western Siberia, the studied species exhibit different water stress tolerance: from xerophyte *L. pumilum* to mesophyte *L. pensylvanicum* and more broad-leaved mesophyte *L. pilosiusculum*. Among the studied lilies, *L. pumilum* is a sun-loving plant, *L. pilosiusculum* is a shade-tolerant plant, and *L. pensylvanicum* occupies an intermediate position. In this ecological series, there is a reliable decrease in the number of cells of the adaxial epidermis per unit area, a decrease in the thickness of the leaf and mesophyll, as well as the thickness of the spongy mesophyll.

In the forest zone of Western Siberia, *L. pumilum* can be recommended for cultivation in open and sunny areas, *L. pilosiusculum* is appropriate for cultivation in partially shaded areas, and *L. pensylvanicum* thrives in both open and slightly shaded areas.

The obtained results are of particular interest in connection with predicted climate changes in the natural habitats of plants, as well as assessment of the possibility of adaptation of species in different natural and climatic zones during introduction.

Acknowledgement

The study was performed within the framework of a state assignment of the Ministry of Science and Higher Education of the Russian Federation (project No. FSWM- 2020-0019).

References

Anenkhonov OA (Ed.) (2023) Red Data Book of the Republic of Buryatia: Rare and endangered species of plants and mushrooms. Constant Publishing House, Belgorod, 342 pp. [In Russian]

Ankipovich ES (Ed.) (2012) Red Book of the Republic of Khakassia. Rare and endangered species of plants and fungi. Nauka, Novosibirsk, 286 pp. [In Russian]

Baranova MA (1985) Classification of morphological types of stomata. Botanical Journal 70(12): 1585–1595. [In Russian]

Baranova MV (1990) Lilies. Agropromizdat, Leningrad, 384 pp. [In Russian]

Baranova MV (1999) Bulbous plants of the Liliaceae family (geography, biomorphological analysis, cultivation). Nauka, St. Petersburg, 229 pp. [In Russian]

Bokov DO, Luferov AN, Krasnyuk II(Jnr), Bessonov VV (2019) Ethno-pharmacological review on the wild edible medicinal plant, *Lilium martagon* L. Tropical Journal of Pharmaceutical Research 18(7): 15591564. https://doi.org/10.4314/tjpr.v18i7.29

Boardman NK (1977) Comparative Photosynthesis of Sun and Shade Plants. Annual Review of Plant Physiology 28: 355–377. https://doi.org/10.1146/annurev.pp.28.060177.002035

Butnik AA, Timchenko OV (1987) Structure of the epidermis of leaves of species of the family Chenopodiaceae. Botanical Journal 72(8): 1021–1030. [In Russian]

Cheremushkina VA, Dneprovsky YuM, Grankina VP, Sudobina VP (1992) Rhizome onions of Northern Asia: biology, ecology, introduction. Science, Novosibirsk,156 pp. [In Russian]

Chernyagina OA (Ed.) (2018) Red Data Book of Kamchatka Territory. Volume 2. Plants. Kamchatpress Publishing House, Petropavlovsk-Kamchatsky, 388 pp. [In Russian]

Danilova NS (Ed.) (2017) Red Data Book of the Republic of Sakha (Yakutia) Vol. 1: Rare and endangered species of plants and mushrooms. Reart Publishing House, Moscow, 412 pp. [In Russian]

Dhyani A, Bahuguna YM, Semwal DP, Nautiyal BP, Nautiyal MC (2009) Anatomical features of *Lilium polyphyllum* D. Don ex Royle (Liliaceae). Journal of American Science 5(5): 85–90.

Ektova SN, Zamyatin DO (Eds) (2010) Red Data Book of the Yamalo-Nenets Autonomous Okrug: animals, plants, fungi. Basko Publishing House, Yekaterinburg, 307 pp. [In Russian]

Embaturova EYu, Korchagina AV (2011) Morphological and anatomical study of *Lilium philippinense* Baker as a promising species for introduction into culture. VSU Bulletin, Geography, Geoecology series 1: 84–86. [In Russian]

Ezau K (1980a) Anatomy of seed plants: in 2 books. Translation from English. Book 1. Mir, Moscow, 224 pp. [In Russian]

Ezau K (1980b) Anatomy of seed plants: in 2 books. Translation from English. Book 1. Mir, Moscow, 560 pp. [In Russian]

Galanin AV, Belikovich AV, Khrapko OV (2008) Flora of Dauria. Volume I. Dalnauka, Vladivostok, 184 pp. [In Russian]

Gamaley YuV (2004) Transport system vascular plants. St. Petersburg State University Publishing house, St. Petersburg, 424 pp. [In Russian]

Givnish TJ (1988) Adaptation to Sun and Shade: A Whole-plant Perspective. Australian Journal of Plant Physiology 15: 63–92.

Goryshina TK (1979) Plant ecology. Higher school Publishing house, Moscow, 368 pp. [In Russian]

Govorina TP, Ivanova VP, Astankovich LI, Vlasova NV, Yarina OA, Danilova NS (1986) Lily of Pennsylvania – *Lilium pensylvanicum* Ker-Gawl. In: Sobolevskaya KA (Ed.) Biological features of Siberian plants in need of protection. Nauka, Novosibirsk, 107–121. [In Russian]

Güven S, Okur S, Demırel M, Coskuncelebi K, Makbul S, Beyazoğlu O (2014). Pollen morphology and anatomical features of *Lilium* (Liliaceae) taxa from Turkey. Biology 69(9): 1122–1133. https://doi.org/10.2478/s11756-014-0416-2

Ivanova LA (2014) Adaptive features of leaf structure in plants of different ecological groups. Ecology 45(2): 107–115. https://doi.org/10.7868/S0367059714020024 [In Russian]

Kästner A (1972) Blattepidermis-Strukturen bei Carlina. Flora 161(3): 225–255. [In German]

Khramchenkova OM (2016) Leaf as an organ of photosynthesis. Manual. Desna Poligraf Publishing house, Chernigov, 32 pp. [In Russian]

Kireeva MF, Korshikova NG, Ivanova NV, Martynova VV (1992) Results and prospects of lily breeding at the IV Michurin All-Russian Research Institute of Horticulture. In: Sidyakin YuP (Ed.)

Lilies. Lev Tolstoy Publishing House, Tula, 1517. [In Russian]

Kondratyev AV (Ed.) (2019) Red Data Book of Magadan Region. Rare and endangered species of animals, plants and fungi. Okhotnik Publishing House, Magadan, 356 pp. [In Russian]

Korolyuk AYu (2006) Ecological optimums of plants in the south of Siberia. Botanical studies of Siberia and Kazakhstan 12: 3–28. [In Russian]

Koshinsky SD (Ed.) (1982) Tomsk Climate. Gidrometeoizdat, Leningrag, 176 pp. [In Russian]

Negrobov VN (1981) Morphological and biological characteristics of lilies, their introduction and selection. PhD Thesis, Botanical Garden of the USSR Academy of Sciences, Moscow, Russia. [In Russian]

Nekhaichenko DV, Koksheeva IM, Kislov DE (2014) Variability of epidermal structures of the *Hydrangea paniculata* (Hydrangeaceae) leaf under culture conditions. Vestnik KrasSAU 12: 52–57. [In Russian]

Nemchenko EP, Novikov VS (1979) Anatomical structure of leaves some species of *Lilium* L. Biological sciences 6: 5358. [In Russian]

Ondar SO, Shaulo DN (Eds) (2019) Red Data Book of the Republic of Tyva (animals, plants and mushrooms). World Publishing House, Voronezh, 560 pp. [In Russian]

Paltiyan-Bugtong JC, Lumpio RG, Napaldet JT (2023) Morpho-anatomy and mathematical modelling in *Lilium philippinense* Baker from Cordillera Central Range, Philippines. Tropical Life Sciences Research 34(3): 217–235. https://doi.org/10.21315/tlsr2023.34.3.12

Pautov AA (2012) Morphology and anatomy of vegetative organs of plants: text-book. St. Petersburg University Publishing House, St. Petersburg, 336 pp. [In Russian]

Pautov AA (Ed) (2003) Questions of comparative and ecological anatomy of plants. St. Petersburg University Publishing House, St. Petersburg, 220 pp. [In Russian]

Pelkonen V-P, Pirttilä A-M (2012) Taxonomy and phylogeny of the genus *Lilium*. Floriculture and Ornamental biotechnology 6(2): 1–8.

Pilnikova Z N (Ed.) (1993) Scientific and Applied Climate Reference Book of the USSR. Series 3. Long-term data. Tomsk, Novosibirsk, Kemerovo Provinces and Altai Territory. Gidrometeoizdat, St.-Petersburg, 20(16), 717 pp. [In Russian]

Polonsky VI, Polonskaya DE, Kozlovskaya TV (2013) Medicinal plant materials of the Krasnoyarsk forest-steppe and its identification based on the anatomical characteristics of leaf epidermal cells. Vestnik KrasSAU 5: 86–91. [In Russian]

Polyakov OA (Ed.) (2017) Red Data Book of the Transbaikal Territory. Plants. House of Peace, Novosibirsk, 384 pp. [In Russian]

Ramensky LG (1956) Ecological assessment of forage lands based on vegetation cover. State Publishing House of Agricultural Literature, Moscow, 240 pp. [In Russian]

Rubtsova TA, Schlotthauer SD (Eds) (2019) Red Data Book of the Jewish Autonomous Region. Rare and endangered species of plants and mushrooms. Birobidzhan Publishing House, Birobidzhan, 267 pp. [In Russian]

Sedelnikova LL (2002) Biomorphology of geophytes in Western Siberia. Nauka, Novosibirsk, 308 pp. [In Russian]

Senchik AV, Malikova EI (Eds) (2020) Red Data Book of the Amur Region: Rare and endangered species of animals, plants and mushrooms. Far Eastern State Agrarian University Publishing house, Blagoveshchensk, 499 pp. [In Russian]

Sobolevskaya KA (1977) Ways and methods of introducing plants of natural flora in Siberia. Introduction of plants of Siberia. Nauka, Novosibirsk, 3–28. [In Russian]

Sokolov PD (Ed.) (1994) Plant resources of Russia and adjacent countries. Flowering plants, their chemical composition, use: Families ButomaceaeTyphaceae. Nauka, St. Petersburg, 271 pp. [In Russian]

Songyun L, Tamura MN (2000) *Lilium* Linnaeus. In: Wu Z, Raven PH, Hong D (Eds) Flora of China: in 25 vol. Vol. 24: Flagellariaceae through Marantaceae. Science Press, Beijing and Missouri Botanical Garden Press, St. Louis, 135.

Sorokopudova OA (2003) Some anatomical and morphological characteristics of species and varieties of the genus *Lilium*. Bulletin of the Main Botanical Garden 185: 100104. [In Russian]

Sorokopudova OA (2019) Lilies in culture. All-Russian Selection and Technological Institute of Horticulture and Nursery of the Russian Academy of Agricultural Sciences Publishing House, Moscow, 186 pp. [In Russian]

Ståhlberg S, Svanberg I (2010) Gathering food from rodent nests in Siberia. Journal of Ethnobiology 30 (2): 184–202. https://doi.org/10.2993/0278-0771-30.2.184

Stepanov NV (Ed.) (2022) Red Data Book of Krasnoyarsk Territory: in 2 volumes. Vol. 2. Rare and endangered species of plants and fungi: in 2 parts. Siberian Federal University Publishing House, Krasnoyarsk, 762 pp. [In Russian]

Telyatyev VV (1985) Useful plants of Central Siberia. Vost.-Sib. Publishing House, Irkutsk, 383 pp. [In Russian]

Trofimova SM (Eds) (2020) Red Data Book of the Irkutsk Region. Republican Printing House, Ulan-Ude, 552 pp. [In Russian]

Tuyl van JM, Arens P, Shahin A, MarasekCiołakowska A, Barba-Gonzalez R, Kim HT, Lim K-B (2018) Lilium. Ornamental Crops 11: 481–512. https://doi.org/10.1007/978-3-319-90698-0_20

Vasin AM, Vasina AL (Eds) (2013) Red Data Book of the Khanty-Mansiysk Autonomous Okrug – Yugra: animals, plants, mushrooms. 2nd edition. Basko Publishing House, Yekaterinburg, 460 pp. [In Russian]

Vehov VN, Lotova LI, Filin VR (1980) Workshop on the anatomy and morphology of higher plants. Moscow State University Publishing House, Moscow, 196 pp. [In Russian]

Vlasova NV (1987) *Lilium* L. – Lily. In: Malyshev LI, Peshkov GA (Eds) Flora of Siberia. Vol. 4: Araceae-Orchidaceae. Science, Novosibirsk, 96–99. [In Russian]

Volkova SA, Pshennikova LM (2011) The leaf epidermis in species of the genus *Dasiphora* (Rosaceae). Turczaninowia, 14(2): 123–129. [In Russian]

Voronov BA (Ed.) (2019) Red Data Book of Khabarovsk Territory: rare and endangered species of plants, mushrooms and animals. Mir Publishing House, Voronezh, 604 pp. [In Russian]

Zarubin AM, Ionycheva MP, Telyatnikov MYu, Astankovich LI (1990) Dwarf lily *Lilium pumilum* Delile. Biological basics of rare protection and disappearing Siberia plants. Central Siberian botanical garden Publishing House, Novosibirsk, 4558. [In Russian]

Zhou J, An R, Huang X (2021) Genus *Lilium*: A review on traditional uses, phytochemistry and pharmacology. Ethnopharmacology 270: 113852. https://doi.org/10.1016/j.jep.2021.113852

Zhukova AA, Minets ML (2020) Biometrics: manual. In 3 hours. Part 2. Basic data analysis techniques. BSU, Minsk, 151 pp. [In Russian]

Zverev AA (2014) *Lilium* L. – Lily. In: Revushkin AS (Ed) Key of plants of the Tomsk region. Tomsk State University Publishing House, 338. [In Russian]

Supplementary material 1

Table1. Characteristics of the anatomical structure of leaves of three Siberian lilies

Authors: Tatiana N. Belaeva, Liu Si, Alina N. Butenkova, Alexey S. Prokopyev Data type: table

Link: http://journal.asu.ru/biol/article/view/16541/14159