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## Comparative anatomic analysis of leaves of *Lonicera tatarica* and *Lonicera microphylla*

The study of anatomical structure of vegetative organs of plants allows to estimate biological and ecological features of species and to reveal features of structure, which allow to carry out identification of taxa at microscopic level. This is especially important for taxonomically close species. The paper presents the results of a comparative anatomical study of leaves of *Lonicera tatarica* and *Lonicera microphylla*. Leaf samples were collected in the summer period of 2024, fixed in Strauss-Fleming's solution, surface preparations and transverse sections were made manually. The results of the studies allowed us to establish that both honeysuckle species are characterized by light leaves, of dorso-ventral type with differentiated mesophyll into columnar and spongy tissues. Stomata are few, of anomocytic type, localized mainly on the underside of the leaf. In both species, small rounded druses of calcium oxalate are present. Differences between the species were found in the shape of leaf epidermal cells, the number of columnar and spongy tissue layers, the shape of conductive bundles, the number of calcium oxalate druses and the presence of simple trichomes.

**Keywords:** *Lonicera tatarica*, *Lonicera microphylla*, anatomical structure, leaf, petiole, comparative study.

### Introduction

Nowadays, the study of fruit plants is of great importance as sources of food, vitamins and biologically active substances [1, 2]. In Karaganda region, the genus *Lonicera* L. (family Caprifoliaceae Juss.) is a promising object for use as a fruit crop and object for green building.

Anatomo-morphological structure of plants in the comparative aspect is an indicator of biological features and ecology of species, and also allows to establish affinity between taxa [3, 4], to act as diagnostic signs of medicinal plant raw materials [5, 6].

In Karaganda region *Lonicera tatarica* L. and *Lonicera microphylla* Willd. ex Schult are of interest as ornamental, food and medicinal plants. Thus, *Lonicera tatarica* is characterized by resistance to climatic conditions, weakly damaged by diseases and pests, suitable for wide application in green building [7], during flowering as an excellent mellifer, berries contain a complex of vitamins and minerals [8], suitable in cooking, and weaving as a source of medicinal raw materials [9]. The fruits of *Lonicera microphylla* have antioxidant and tonic properties [10].

As a study of biological properties of both species of honeysuckle, the objective was to investigate the anatomical parameters of the leaf and to identify the diagnostic characters of the species.

### Experimental

Leaf samples of both species of *Lonicera* L. (Fig. 1) were collected in the Karkaraly mountains (Karkaraly district, Karaganda region) in June 2024. Species identification was carried out at the Department of Botany of the University. The plant specimens were deposited in the herbarium fund of the Faculty of Biology and Geography of Karaganda Buketov University.

Dry leaves (leaves plates and petioles) were soaked in Strauss-Fleming mixture (distilled water: ethyl alcohol 96 % — glycerol 40 % in the ratio 1:1:1), transverse sections and surface preparations were prepared using a blade [11].

The description of micropreparations was carried out in accordance with the methodological guidelines [12, 13]. During description, attention was paid to cell structure, shape, presence of trichomes and inclusions, shape of stomata, shape and localization of conductive bundles.



Figure 1. *Lonicera tatarica* and *Lonicera microphylla* in fruiting phase, Karkaraly Mountains

### Results and Discussion

The cells of the upper and lower sides of the leaf of *L. tatarica* are rounded or slightly elongated (Fig. 2), the walls are slightly curved. Rounded druzes of calcium oxalate are translucent on both sides. Stomata are oval in shape, anomocytic type, localized mainly on the lower side of the leaf.

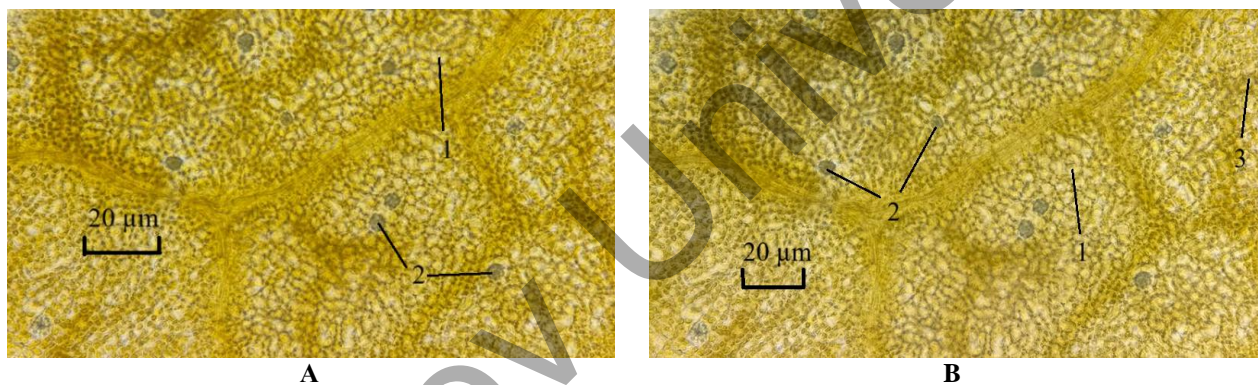


Figure 2. Micropreparation of the leaf of *Lonicera tatarica*, fragments of preparations with surface: A — upper epidermis, B — lower epidermis; 1 — epidermis cells, 2 — druzes; 3 — stomata

Cells of upper and lower epidermis of *Lonicera microphylla* leaf are characterized by elongated or rounded shape, with straight and distinctly thickened walls (Fig. 3). Stomata are localized on the lower side of the leaf, rounded, anomocytic type. Trichomes are placed along the central vein; they are simple, 1-2-celled.

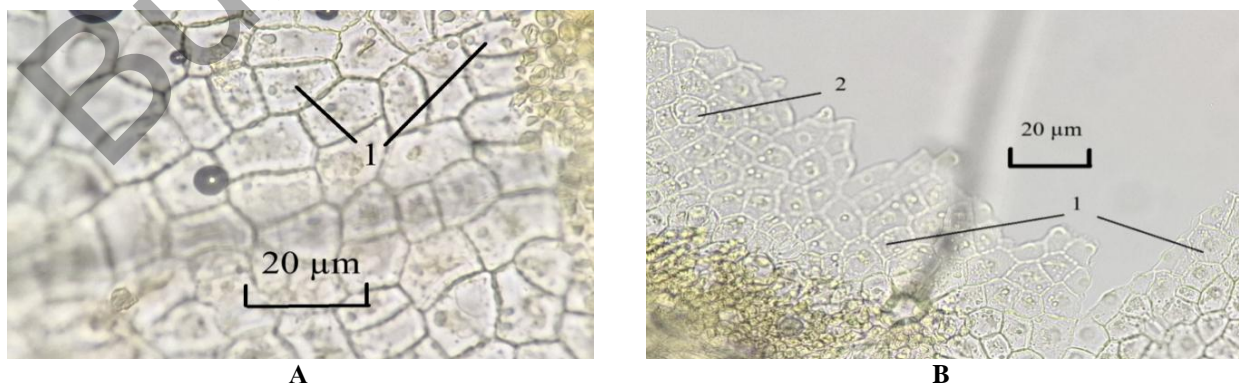


Figure 3. Micropreparation of the leaf of *Lonicera microphylla*, fragments of preparations with surface: 1 — epidermis cells, 2 — stomata

On the transverse section (Fig. 4) the leaf of *L. tatarica* is of dorso-ventral type, the mesophyll is indistinctly differentiated into palisade and spongy tissues. A single-layered epidermis is located on both sides. Its cells are rounded in shape, with a clearly visible layer of cuticle on the outer side. In the area of the main and lateral veins, areas of collenchyma are located under the epidermis, with mesophyll between the veins. Under the upper epidermis there are 1-2 layers of palisade tissue, on the lower side there are cells of spongy tissue with developed intercellular layers. Conductive bundles of elliptical shape, collateral, closed type, reinforced by sections of sclerenchyma. Numerous druzes of calcium oxalate are localized in the mesophyll.

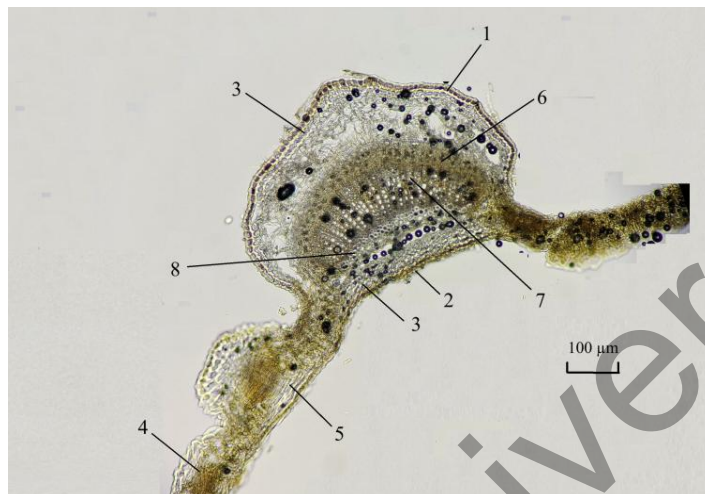


Figure 4. Transverse section of a leaf of *Lonicera tatarica*.

Fragment in the area of the central vein: 1 — lower epidermis, 2 — upper epidermis, 3 — collenchyma, 4 — spongy mesophyll, 5 — palisade mesophyll, 6 — phloem, 7 — xylem, 8 — sclerenchyma

The transverse section of *Lonicera microphylla* is also flat, dorso-ventral on the transverse section (Fig. 5), not clearly differentiated into palisade and spongy tissue. On both sides, the leaf is surrounded by a 1-layer epidermis composed of oval cells and a thick layer of cuticle. The columnar tissue is arranged in 2 layers and the spongy tissue in 2-3 layers. Conducting bundles are oval curved, collateral, closed type.

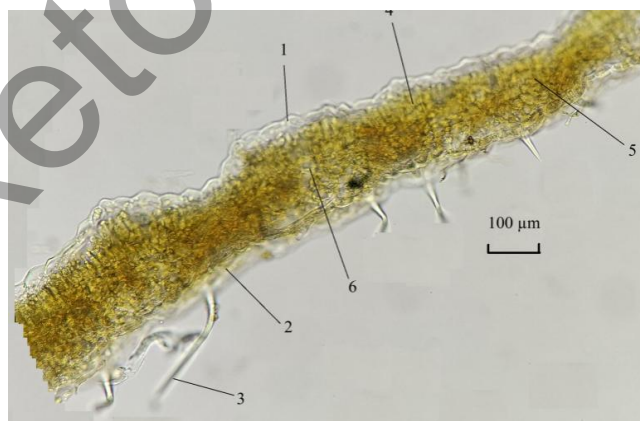


Figure 5. Transverse section of a leaf of *Lonicera microphylla*.

Fragment in the area of the central vein: 1 — lower epidermis, 2 — upper epidermis, 3 — collenchyma, 4 — spongy mesophyll, 5 — palisade mesophyll, 6 — phloem, 7 — xylem, 8 — sclerenchyma

The leaf petiole of Tatar honeysuckle is broadly ovate, slightly curved in cross section (Fig. 6). Along the perimeter there is a single-layer epidermis with a thick layer of cuticle. Beneath the epidermis lies a multilayered collenchyma. In the center of the petiole lies a vascular-fiber bundle, collateral, closed type, reinforced by sclerenchyma strands. The mesophyll zone is thin, composed of thin-walled parenchyma cells, with large intercellular cells and small druzes of calcium oxalate.

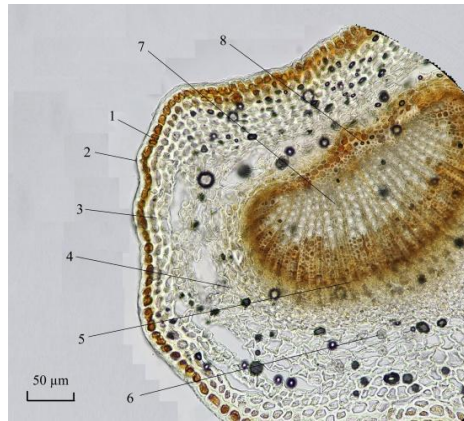


Figure 6. Transverse section of *Lonicera tatarica* leaf petiole: 1 — epidermis, 2 — cuticle, 3 — collenchyma, 4 — mesophyll, 5 — phloem, 6 — druses, 7 — xylem, 8 — sclerenchyma

The leaf petiole of *Lonicera microphylla* on a transverse section is curved (Fig. 7). Rounded epidermal cells with cuticle, adjacent to 2–6-layer collenchyma, lie along the perimeter. The conducting bundle is narrow, elongated, collateral, of closed type, surrounded by sections of sclerenchyma. Numerous druses of calcium oxalate are placed around the central vein under a layer of lamellar collenchyma. Few simple trichomes are noted.



Figure 7. Transverse section of a leaf of *Lonicera microphylla*. Fragment in the area of the central vein: 1 — epidermis, 2 — trichomes, 3 — collenchyma, 4 — calcium oxalate druses, 5 — central conductive bundle.

Comparison of anatomical features of the leaf of both species of honeysuckle allowed us to identify characteristic features (Table).

Table

**Comparative anatomical features of the leaf of *Lonicera tatarica* and *Lonicera microphylla***

Characteristics	<i>Lonicera tatarica</i>	<i>Lonicera microphylla</i>
Cells of leaf epidermis	Rounded or slightly elongated	Elongated or rounded
Drusae of the leaf lamina	+	-
Stomata	Anomocytic, localized on the lower side	Anomocytic, localized on the lower side
Trichomes on the leaf lamina	-	Simple
Type of leaf on transverse section	Dorso-ventral	Dorso-ventral
Mesophyll	Differentiated into palisade and spongy tissues	Differentiated into palisade and spongy tissues
Number of layers of columnar tissue	1-2	2
Number of layers of spongy tissue	2-3	2-3
Shape of conductive bundles	Elliptical	Oval
Shape of petiole on transverse section	Broadly ovate, slightly curved	Narrow-ovate, strongly curved
Druses	Few	Numerous

Thus, differences in the shape of leaf epidermis cells, presence of trichomes and druses of calcium oxalate, number of layers of columnar and spongy tissue, shape of petiole on transverse section were noted.

### Conclusion

Thus, the anatomical structure of the leaves of *Lonicera tatarica* and *Lonicera microphylla* was studied. It was determined that the leaves are light-type, with a dorsoventral structure in cross section, and petioles reinforced with areas of sclerenchyma. The mesophyll of the leaf blade is differentiated into columnar and spongy tissues. The stomata are small, few in number, anomocytic, and usually located on the lower side of the leaf. Round calcium oxalate crystals are present in the structure of the leaf blade and petiole.

Distinctive features of the leaves of both species are the shape of the epidermal cells, the number of layers of columnar and spongy tissue (*Lonicera microphylla* has more layers), the shape of the conducting bundles (broad and narrow-ovoid), the number of calcium oxalate crystals and the presence of simple trichomes on the underside of the leaf in *Lonicera microphylla*, in the absence of trichomes in the structure of *Lonicera tatarica*.

The data obtained confirm the xeromesophytic structure of the leaves and also show differences between plants for species identification based on vegetative organs.

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### Author contribution

The manuscript was written through contributions of all authors. All authors have given approval to the final version of the manuscript. CRediT: **Gavrilkova E.A.** — Investigation, Methodology, Plant material collection, Writing-review & editing; **Tleukenova S.U.** — Conceptualization, Data curation; **Ageev D.V.** — Plant anatomy; **Ramazanov A.K.** — Data curation, visualization.

### Conflict of Interest

Authors declare no conflict of interest.

### References

- 1 De Cortes Sanchez-Mata, M., Matallana-Gonzalez, M. C., & Morales, P. (2016). The contribution of wild plants to dietary intakes of micronutrients (I): vitamins. *Mediterranean Wild Edible Plants*. Springer Nature.
- 2 Maikhun, R. K., Semua, R. I., Singh, A., & Nautiyal, M. C. (1994). Wild fruits as a contribution to sustainable rural development: a case study from the Garhwal Himalaya. *Int J Sustain. Devel. World Ecol.*, 1, 56–68.
- 3 Albrechtova, J. (2003). *Plant anatomy in environmental studies*. Prague: Charles University.
- 4 Lobo, S. M., & Krishnakumar, G. (2014). Studies on ecological anatomy of the mangrove fern *Acrostichum aureum* L. *Int Journal of Plants, Animal & Environmental Science*, 4(1), 195–200.
- 5 Duschanova, G., Shakhmurova, G., Ravshanova, M., & Maimataeva, A. (2023). Diagnostic signs of aboveground and underground organs of *Ferula tenuisecta* Korov. in natural conditions. *Web of Conferences*, 421, 04005. <https://doi.org/10.1051/e3sconf/202342104005>
- 6 Nuzhyna, N., Maliarenko, V., & Syvets, H. (2022). Features of the root and rhizome anatomical structure of *Potentilla alba* L. as a diagnostic sign of the raw materials. *Bulletin of Taras Shevchenko National University of Kyiv, series biology*, 2(89), 10–13. <https://doi.org/10.17721/1728.2748.2022.89.10-13>
- 7 Nosirov, S. S., Rakhimova, N. K., & Matismoilov, A. Kh. (2023). Seed Germination and Vegetative Propagation of Promising Decorative *Lonicera tatarica* L. (Caprifoliaceae Juss.), Introduced into the Tashkent Botanical Garden (Uzbekistan). *American Journal of Plant Sciences*, 14, 496–505. <https://doi.org/10.4236/ajps.2023.144033>
- 8 Popoviciu, D. R., & Negreanu-Pirjol, T. (2023). Health benefits of antioxidant bioactive compounds in the fruits and leaves of *Lonicera caerulea* L. and *Aronia melanocarpa* (Michx.) Elliot. *Antioxidants*, 12, 951. <https://doi.org/10.3390/antiox12040951>

- 9 Oi, S., Wang, J., & Yang, Q. (2023). Pharmacological effects of genus *Lonicera* and its application in tumors of the digestive system. *Highlight in Science, Engineering and Technology*, 80, 489–495. <https://doi.org/10.54097/t6kjwz34>
- 10 Tang, Y. -R., Zeng, T., Zafar, S., Yuan, H. -W., Li, B., & Peng, C. -Y. (2018). *Lonicera* Flos: a review of chemical constituents and biological activities. *Digital Chinese Medicine*, 1, 173–188.
- 11 Ruzin, S. E. (1999). *Plant microtechnique and microscopy*. Oxford: Oxford University Press.
- 12 Crang, R., Lyons-Sobaski, Sh., & Wise, R. (2018). *Plant Anatomy. A concept-based approach to the structure of seed plants*. Springer Nature.
- 13 Rudall, P. J. (2006). *Anatomy of flowering plants. An introduction to structure and development*. Cambridge: Cambridge University Press.

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### ***Lonicera tatarica* және *Lonicera microphylla* жапырақтарының салыстырмалы анатомиялық талдауы**

Өсімдіктердің вегетативті мүшелерінің анатомиялық құрылымын зерттеу түрлердің биологиялық және экологиялық ерекшеліктерін бағалауға және таксондарды микроскопиялық деңгейде анықтауға мүмкіндік беретін құрылымдық ерекшеліктерді анықтауға мүмкіндік жасайды. Бұл әсіресе таксономиялық жақын түрлер үшін өте маңызды. Мақалада *Lonicera tatarica* және *Lonicera microphylla* жапырақтарының салыстырмалы анатомиялық зерттеу нәтижелері келтірілген. Жапырақ үлгілері 2024 жылдың жазында жиналды, Штраус-Флеминг ерітіндісінде тіркелді, беттік препараттар мен көлденең қималар қолмен жасалды. Зерттеу нәтижелері үшқаттар тұқымдасының екі түрі де бағаналы және кеуекті тәрізді ұлпаларға дифференциалданған мезофиллі бар дорзо-вентральды типтегі, жарық жапырақтарымен сипатталатынын анықтады. Жапырақ санылаулары аз, аномоциттік типті, негізінен жапырақтың төменгі жағында локализацияланған. Екі түрде де кальций оксалатының ұсақ дөңгелек друзалары бар. Түрлер арасындағы айырмашылықтар жапырақ эпидермисінің жасушалары түрінде, бағаналы және кеуек тәрізді тіндердің қабаттарының саны, өткізгіш шоктардың пішіні, кальций оксалатының друзаларының саны және қарапайым трихомалардың болуы.

*Кілт сөздер:* *Lonicera tatarica*, *Lonicera microphylla*, анатомиялық құрылымдар, жапырақ, қысқа шыбық, салыстырмалы зерттеу.

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### **Сравнительный анатомический анализ листьев *Lonicera tatarica* и *Lonicera microphylla***

Изучение анатомического строения вегетативных органов растений позволяет оценить биологические и экологические особенности видов, а также выявить особенности строения, которые позволяют проводить идентификацию таксонов на микроскопическом уровне. Это особенно важно для таксономически близких видов. В статье представлены результаты сравнительного анатомического исследования листьев *Lonicera tatarica* и *Lonicera microphylla*. Образцы листьев были собраны в летний период 2024 г., зафиксированы в растворе Штрауса-Флеминга, поверхностные препараты и поперечные срезы выполнены вручную. Исследования показали, что оба вида жимолостей характеризуются световыми листьями, дорзо-вентрального типа с дифференцированным мезофиллом на столбчатую и губчатую ткани. Устьица немногочисленные, аномоцитного типа, локализованы преимущественно с нижней стороны листа. У обоих видов присутствуют мелкие округлые друзы оксалата кальция. Отличия между видами выявлены в форме клеток эпидермиса листа, количестве слоев столбчатой и губчатой ткани, форме проводящих пучков, количестве друз оксалата кальция и присутствии простых трихом.

*Ключевые слова:* *Lonicera tatarica*, *Lonicera microphylla*, анатомические структуры, лист, черешок, сравнительное изучение.

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