

# Comparative anatomical study of underground and aboveground organs in *Ferula tadshikorum* Pimenov under natural and introduced environments

*Dilovar T. Khamraeva*

Tashkent Botanical Garden, Academy of Sciences of the Republic of Uzbekistan, 232 B Bogishamol St., Tashkent, 100053, Uzbekistan

*Dinara N. Tukhtaeva*

Research and Development Institute of Forestry, Tashkent region, 111104, Uzbekistan

*Olim K. Khojimatov*

Tashkent Botanical Garden, Academy of Sciences of the Republic of Uzbekistan, 232 B Bogishamol St., Tashkent, 100053, Uzbekistan

*Rainer W. Bussmann*

Department of Botany, State Museum of Natural History, Erbprinzenstrasse 13, 76133 Karlsruhe, Germany;  
Department of Ethnobotany, Institute of Botany and Bakuriani Alpine Botanical Garden, Ilia State University, 1 Botanical St., Tbilisi, Georgia

A comparative study was conducted on the anatomical structure of the roots and leaves of *Ferula tadshikorum*, a rare plant species. The study included two groups: virginile plants introduced at the age of 4 years (located in the Tashkent Botanical Garden) and natural individuals aged 12–14 years (found in the Surkhandarya region, Babatag ridge). The main objective was to understand how the plant's anatomy changes with age and its adaptation to different environments. Common characteristics were identified in the leaves of both groups of plants. These included the isolateral palisade type of mesophyll, a rounded triangular shape around the central vein, and the presence of 2-layer elongated palisade tissue on the flattened lateral parts from the adaxial side. Regarding the root structure, significant differences were observed between the natural and introduced plants. In the natural samples, the woody part of the root exhibited groups of libriform cells, which seemed to be influenced by the edaphotype (soil conditions) and age of the plant. These findings suggest that the conductive system of the roots develops differently depending on the age and environment. The introduction of *Ferula tadshikorum* in the Tashkent Botanical Garden proved to be successful, as evident from the results. The leaves of the introduced plants showed adaptive characteristics, such as a thickened outer wall of the epidermis, collenchyma strands, bast fibers, and the isolateral palisade type of mesophyll. These adaptations are likely to help the plant cope with its new environment. In the roots of both natural and introduced plants, an important adaptive function was observed. The parenchymal cells were filled with starch grains, and there was an abundance of secretory ducts. This suggests that the root acts as an accumulating organ for organic substances, aiding the plant's survival and growth. In conclusion, the study provided valuable insights into the anatomical changes of *Ferula tadshikorum* at different stages of its life and in different environments. The observed adaptations in the leaves and roots contribute to the plant's ability to thrive in varying conditions.

Academic editor: R. Yakovlev | Received 17 July 2023 | Accepted 30 October 2023 | Published 12 January 2024

<http://zoobank.org/96087989-9B5D-46AC-99B9-8005EE25A9AE>

**Citation:** Khamraeva DT, Tukhtaeva DN, Khojimatov OK, Bussmann RW (2024) Comparative anatomical study of underground and aboveground organs in *Ferula tadshikorum* Pimenov under natural and introduced environments. Acta Biologica Sibirica 10: 9–29.  
<https://doi.org/10.5281/zenodo.10475286>

## Keywords

Concentric circles, leaf, mesophyll, root, secretory duct, starch grains, virginile plant, *Ferula tadshikorum* Pimenov, Tashkent Botanical Garden

## Introduction

The determination of the patterns of these changes is important for developing the theory of ontogenesis, taxonomy, morphology, and phylogeny of plants. In the ontogeny of an individual, in most cases, changes in the anatomical structure of the shoot and root are visible, from which more conservative and more labile characters are distinguished. The degree of stability and variability of the same structures is not the same in representatives of different taxa and is determined by both internal causes and environmental factors (Barykina and Gulenkova 1985).

To identify the ecological amplitude of leaf adaptability, the analysis of its anatomical structure for local and introduced species is most often used in natural or new growing conditions. When plants in new climatic conditions, the evolutionary-adaptive features of the species are revealed due to morphological-anatomical and physiological-biochemical rearrangements, which primarily affect the leaf, since the main life processes of the plant occur in the leaf and the necessary organic substances are synthesized in this organ. In this sense, plant adaptation to new conditions is closely related to leaf structure and is aimed at maintaining its functions at the highest possible level for specific growing conditions (Sedelnikova 2000; Padmavathi and Chetty 2019; Bondar et al. 2021).

Very little attention has been paid to studies on the comparative study of the structure of the vegetative organs of specific plant species from natural and introduced habitats, and the main works are devoted to the study of certain plant organs from one place of growth for taxonomy, floristics, and ecology (Barykina and Alyonkin 2017).

Research intensity of the genus *Ferula* L. in botany, molecular biology, chemistry, and ethnobotany increases from year to year, since many of its representatives are of great practical importance as fodder, medicinal, essential oil and melliferous plants (Chen and Liu 1989; Fan et al. 2015; Elghwaji et al. 2017; Panahi et al. 2018; Eser and Yoldas 2019; Akalın et al. 2020; Tuncay et al. 2023).

Safina and Pimenov (1984) established taxonomic characters and features in the structure of fruits to solve several issues of evolutionary morphology and anatomy of genus *Ferula* species, common in Kazakhstan.

Akhmetova et al. (2015) analyzed the anatomical characteristics of the leaf blades of *Ferula iliensis* Krasn. ex-Korovin of different ages from three different populations growing in the eastern part of the Zailiysky Alatau (Kazakhstan). In all plants from the populations studied, a single type of leaf blade formation and xero-mesomorphic structure was determined; the presence of multilayered palisade tissue in the leaf mesophyll and more trichomes underside of the leaf were noted. The authors noted that differences in the anatomical structure of the leaf are mainly quantitative in nature and are associated with the environmental conditions of the plants.

S. Rakhimov and G.R. Denisova (2017) investigated the underground part of *Ferula tadshikorum* and revealed the formation of the root system. According to the authors, at the initial stages of ontogenesis, due to the thickening of hypocotyl and the basal parts of the polycyclic rosette shoot, a root is formed, which develops for 8–25 years. In the last year of life, all the reserve plant substances are spent on the development of a elongated generative shoot, and the plant, after the growing season, completes its development cycle.

However, there is lack of information on the anatomical structure of the vegetative or generative organs of *Ferula tadshikorum* from natural habitats, we previously published the data on the comparative morphological and anatomical structure of leaf blades and petioles of *Ferula tadshikorum* of individuals of different ages of the virginile period from the conditions of introduction (Khamraeva et al. 2021).

Currently, due to the increased interest in obtaining raw materials (gum-resin) from the roots of *Ferula tadshikorum* rare species by entrepreneurs in Uzbekistan, the creating and expanding plantations are extremely important (Khojimatov and Khamraeva 2018; Khojimatov et al. 2019; Khojimatov et al. 2021; Halkuzieva et al. 2022). Therefore, the study of structure of the roots and leaves in individuals of both natural and introduced habitats can reveal the result of plant adaptation to various environmental factors, and the information obtained on the structure of underground and aboveground organs complements the ecological characteristics of plants and also improves the understanding of the range of their plasticity.

The aim of this study was a comparative anatomical study of structure of the roots and leaves of natural and introduced specimens of *Ferula tadshikorum* in connection with ontogenetic characteristics of development.

## Materials and methods

*Ferula tadshikorum* is a perennial monocarpic plant, standing at a height of 1.5–1.8 m and notable for its strong garlic scent. It predominantly grows in Uzbekistan's middle belt of mountains, particularly in the southern regions of Surkhandarya and Kashkadarya. The plant can be found on various terrains such as loess and fine earth-gravelly slopes, limestones, variegated flowers, along dry river valleys, and terraces, at altitudes ranging from 1400 to 1800 m a.s.l.

For centuries, Central Asia has utilized the resin derived from the roots of *Ferula tadshikorum* for medicinal purposes. It has been traditionally employed to treat coughs, flatulence, convulsions, and even to inhibit the development of conditions like atherosclerosis, cataracts, and nervous and mental disorders. Additionally, the plant is known for its potent anthelmintic properties (Rakhmonov 2017). Due to its ecological significance, *Ferulata dshikorum* has been listed in the Red Book of the Republic of Uzbekistan with a status of 3 (Makhmudov 2019).

In 2018, experiments on *Ferula tadshikorum* were initiated at the Laboratory of Introduction of Medicinal Plants in the Tashkent Botanical Garden, which operates under the Institute of Botany of the Academy of Sciences of the Republic of Uzbekistan. These experiments, along with phenological observations of the experimental plants, have been ongoing since then (Khamraeva et al. 2019; 2021).

To study the anatomical structure of leaves and roots of virginile plants of different ages, researchers collected samples from both the experimental sites of the Tashkent Botanical Garden and the natural growth areas in the Babatag ridge of Surkhandarya region in 2022. The collected samples were preserved in 70% ethyl alcohol. The transverse sections of the leaf plate and petiole, as well as the basal part of the root, were stained with methylene blue and enclosed in glycerol-gelatin using a conventional method (Barykina, Chubatova 2005). The cross sections were then photographed using a Canon A 2300 camera. We performed the statistical processing of

quantitative data using MS Excel according to Zaitsev (1990). For each anatomical parameter of the leaf blade, petiole, and root, the arithmetic mean was calculated based on 30 values.

Abbreviations in figure captions: AbE – abaxial epidermis; AdE – adaxial epidermis; B – bark; BF – bast fibers; BR – bast ray; C – cork; Ca – cambium; Ch – chlorenchyma; Cl – collenchyma; Hy – hypodermis; L – libriform; P – palisade; Par – parenchyma; Ph – phloem; Phl – phellogen; SD – secretory duct; SP – spongy parenchyma; T – trichome; Vs – vessels.

## Result

### Surkhondarya region. Virginile plant, 12–14 years old

Plant with rosette leaves, pubescent below, petiolate, large blade, more than 50 cm long, about 30 cm wide, broadly triangular in outline, segments of first order twice or thrice pinnately dissected; terminal lobes large, more than 20 cm long, 7–9 cm wide, lanceolate or ovate-lanceolate, pointed, crenate along the margin.

**Root.** The root is a taproot, differentiated into the following parts: closer to the surface, it is cylindrical, covered with annular folds for a short distance (2–3 cm), then passes into a thickened, tuberous part, and narrows sharply towards the apical part. The root neck is densely covered in the remains of vascular bundles of leaf petioles from previous years of vegetation. Outside, the root is covered with a dark brown, cracked, and easily lagging cork (Fig. 1a). Along the periphery of the root, a multilayer cork is formed from narrow tabular cells. Closer to it, the bark parenchyma is tortuous due to contractility. The secondary bark is multilayered, contains numerous secretory ducts, which are arranged in circles throughout the phloem (Figs 1b, c; Table 1). Around the epithelial cells of the ducts, there are lining cells filled with starch grains. The cambium is multilayered. Due to the increased contractile activity of the root, the conductive bundles are tortuous. The phloem is multilayered, around the xylem there are strands of libriform (Figs 1d, e). The vessels are arranged in radial chains; each group contains up to 4 to 5 large and several small vessels. In the thickened, clubbed part of the root, strong parenchymalization was observed, with the filling of almost all parenchymal cells with starch grains.

Indicators		Surkhondarya region	Tashkent Botanical Garden
Epithelial cells of large secretory duct	height	13.02±0.28	23.04±0.44
	width	28.50±0.5	39.36±0.91
	quantity	8.30±0.15	7.40±0.16
Cavity diameter of large secretory ducts		64.92±0.63	63.66±1.78

**Table 1.** Quantitative characteristics of the root of *F. tadshikorum*,  $\mu\text{m}$  (here and then  $n=30$ )

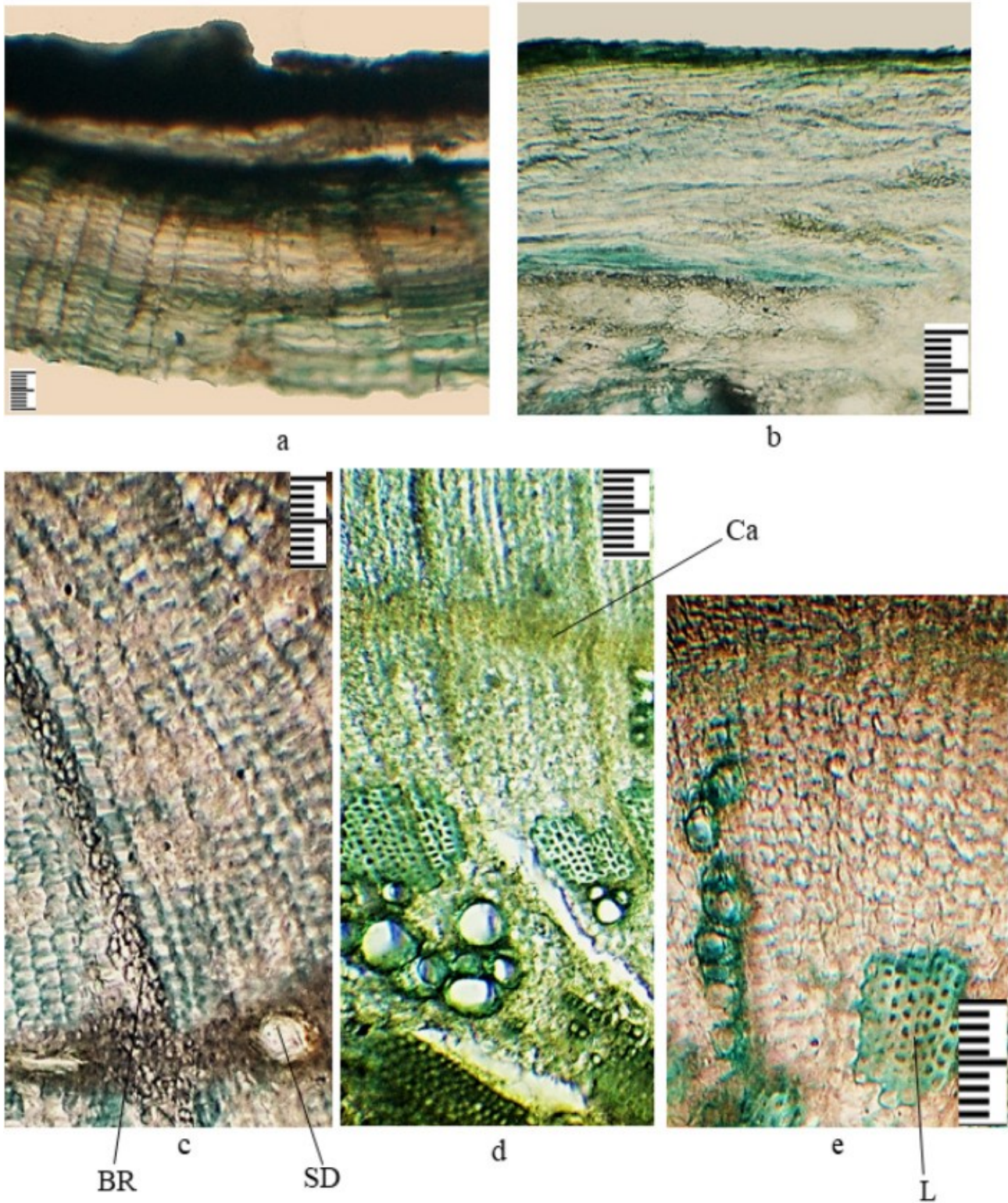
**Leaf.** On the transverse section, the leaf is lamellar, round-triangular in the region of the central vein, and biconvex on the sides in the places of vascular bundles (Fig. 2a). On the abaxial side, the surface is pubescent with simple trichomes, strongly ribbed, while on the adaxial side, slightly wavy (Figs 2b, c). The abaxial and adaxial epidermises are single-layered, the outer walls of the cells of the adaxial epidermis are the most thickened, and both epidermises are covered with a dentate cuticle (Table 2). The region of the central leaf vein is characterized by the following features:

- under the abaxial and adaxial epidermis, there is a single-layer hypodermis of smaller, round-oval cells;
- in the ribs, there are strands of collenchyma from the abaxial side up to 6–14 rows, adaxial – 3–8 rows;
- on the abaxial side above the medium and small bundles, the hypodermis and/or groups of



collenchyma cells break off from the other leaf tissue, as a result of which rather wide air cavities are formed, and on the adaxial part they are not numerous;

- the aquifer parenchyma consists of thin-walled round oval cells of up to 42–45 layers, of various sizes, it has air cavities.



**Figure 1.** The main root anatomical structure of the virginile *Ferula tadshikorum* in natural populations (12–14 years old). **a** – part of the periderm, **b** – detail of the secondary bark, **c** – secretory ducts in the bark parenchyma, **d** – part of the central cylinder, **e** – detail of the cambial and xylem parts. Scale bar 100 μm.

The mesophyll is isolated palisade. In the area of the central vein of the leaf on the adaxial side, the palisade parenchyma is two-layered, under the epidermis the cells of the palisade are more elongated and the second layer is short. On the abaxial side, there is 2-3-layer palisade tissue, but the cells are shortened. In the flattened lateral parts of the leaf, the palisade parenchyma is two-layered, which is on the adaxial side with elongated cells, and on the leaf. abaxial part under the epidermis, the cells are short, almost rounded, the second layer of columnar cells (Fig. 2d; Table 2). Between them, there is a 2–3 layer of spongy tissue of large horizontally elongated cells (Table 2).

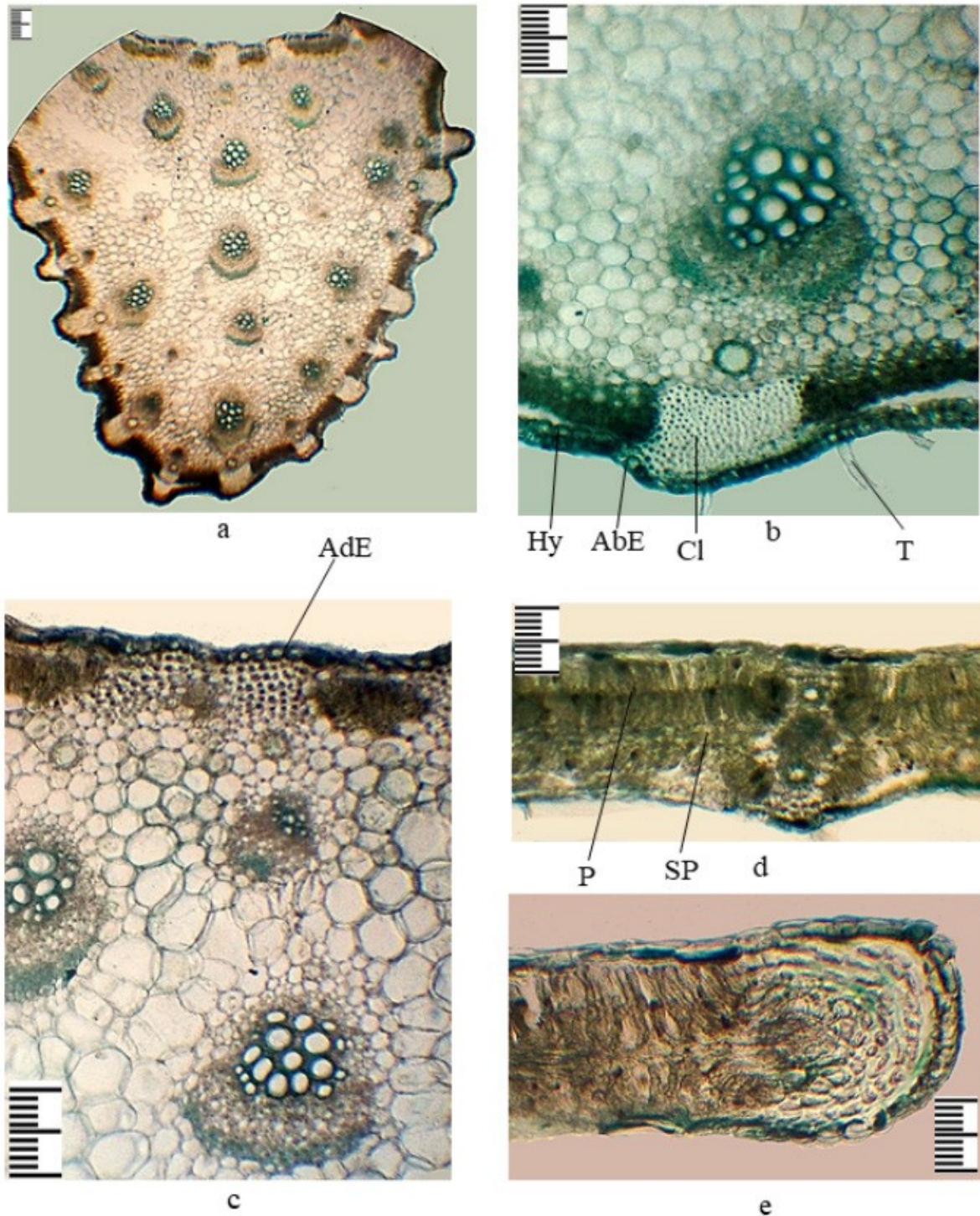
Conductive bundles of collateral type. The central vein is multi-bundled, represented by several large, medium, and small peripheral and parallel-arranged three central conductive bundles. The peripheral bundles are located opposite the collenchyma strands; their size corresponds to the size of the bundle. There are bast fibers above the phloem of large and medium bundles, larger bundles have more multilayer ones. The central vein conductive bundles on the phloem side have one secretory duct, which has up to 9–11 epithelial cells on the abaxial side, and up to 8–10 on the adaxial side (Table 2). At the edge of the leaf, secretory ducts are located on the abaxial part with 6–7 epithelial cells (Fig. 2e).

Indicators		Surkhandarya region	Tashkent Botanical Garden
The thickness of the cell outer walls	abaxial epidermis	6.50±0.3	6.36±0.22
	adaxial epidermis	10.32±0.45	7.08±0.46
Abaxial epidermis	height	30.18±0.49	32.16±0.6
	width	26.76±0.86	35.76±0.59
Adaxial epidermis	height	29.88±0.69	26.34±0.75
	width	31.08±0.76	42.66±1.18
Palisade tissue on the abaxial part	height	41.52±1.03	36.60±1.46
	width	15.36±0.63	11.22±0.51
Palisade tissue on the adaxial part	height	54.24±1.6	54.0±1.80
	width	14.70±0.45	14.04±0.55
Spongy parenchyma thickness		70.50±1.71	50.58±0.89
Epithelial cells of large secretory duct	height	13.26±0.35	11.28±0.55
	width	22.92±0.46	21.30±0.83
	quantity	10.80±0.16	13.10±0.27
Epithelial cells of small secretory duct	height	12.42±0.63	10.92±0.33
	width	17.34±1.8	17.94±0.68
	quantity	9.30±0.31	8.90±0.23
Cavity diameter of ducts	large	52.32±1.54	70.32±1.21
	small	36.24±0.99	33.30±1.4

**Table 2.** Quantitative characteristics of the leaf blade of *F. tadshikorum*, μm

There are numerous conductive bundles of different sizes in the lateral parts of the leaf. Bast fibers are absent above the lateral bundles. Above and below the large lateral bundles there is one secretory duct and small collenchyma cells. On the abaxial side, the conductive bundles and collenchyma strands are larger than on the adaxial side. Large secretory ducts have up to 8–10 epithelial cells, and small up to 6–7.

**Petiole. The base part.** On the transverse section, the petiole is rounded-oval. The epidermis is single-layered, larger-celled than the single-layered hypoderm (Fig. 3a). The petiole consists of 7–8 layers of chlorophyll-bearing tissue, and the other cells are rounded oval, thin-walled, parenchymal (Fig. 3b).



**Figure 2.** The leaf anatomical structure of the virginile *Ferula tadshikorum* in natural populations (12–14 years old). **a** – detail of the central part of the leaf, **b** – detail from the abaxial part of the central vein, **c** – detail from the adaxial part of the central vein, **d** – lateral part, **e** – the edge of the leaf. Scale bar 100  $\mu\text{m}$ .

Collenchyma strands on peripheral large conductive bundles up to 15–18 rows, over small ones up to 5–6 rows. The petiole is represented by numerous conductive bundles, the outer circle consists of large, medium, and small bundles, and the other bundles are also of various sizes, placed in the center, forming a longitudinal line in 8 rows (Fig. 3c, d). All conductive bundles are of collateral type. Above the phloem of the bundles are bast fibers. The xylem of the bundles consists of small and large vessels. A secretory duct is located between the collenchyma and the conductive bundles;



its size corresponds to the size of the bundles. In small bundles, secretory ducts have up to 8–9 epithelial cells, and in large ones up to 10–11. Near the large xylem of the peripheral bundles, there are small secretory ducts among 2–3, which are with 6–8 epithelial cells. The parenchyma of the petiole abundantly contains secretory ducts with 6–8 epithelial cells. There are air-bearing cavities in the parenchyma of the petiole.

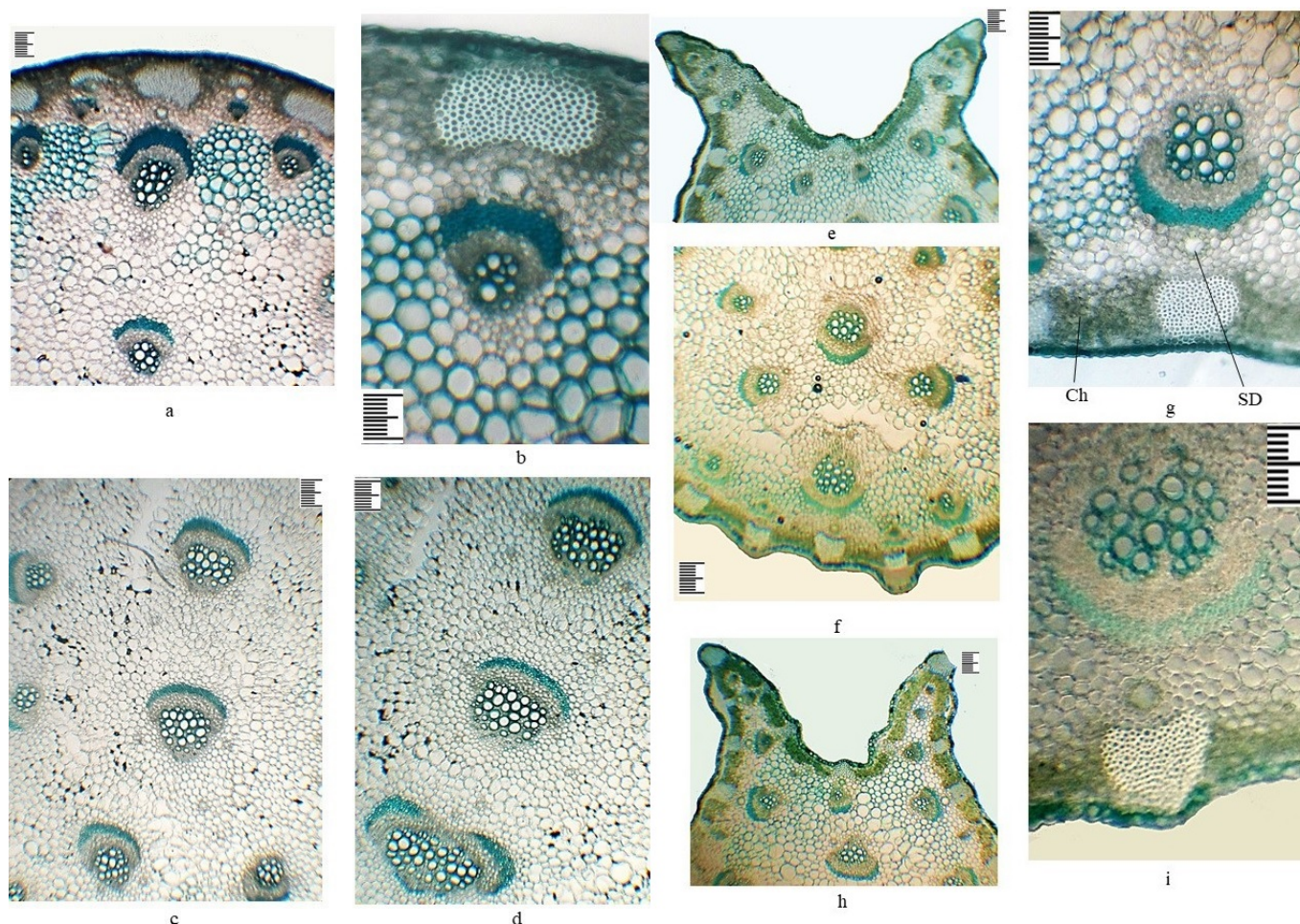
**The middle part.** On the cross section of the petiole of a rounded-ovoid shape, tapering to the adaxial part, two curved outgrowths are formed (Figs 3e–g). At the ends of the outgrowths there are 2–3 conductive bundles, above and below them are collenchyma strands and a secretory duct. The anatomical structure is almost similar to the base part of the petiole, but there are slight differences (Table 3). In the petiole from the abaxial to the adaxial part, the bundles are located in the center in 7 rows. The surface is slightly ribbed and pubescent with simple trichomes.

The chlorophyll-bearing parenchyma is 5–7 layered. The collenchyma over large bundles has up to 13–17 rows and over small up to 5–7 rows. The secretory ducts in large bundles have up to 10–11 epithelial cells, and in small ones up to 8 (Table 3).

**The upper part.** The structure is almost similar to the middle part, but there are some differences (Figs 3h, i). The surface is strongly ribbed. The conductive bundles in the center of the petiole are arranged in 4 rows. The chlorophyll-bearing parenchyma is 4–5 layered. The collenchyma over large bundles has up to 12–14 rows, small up to 5–6 rows. Large secretory ducts have up to 9–10 epithelial cells and small ones have up to 7–8 cells.

Indicators		Surkhandarya region	Tashkent Botanical Garden
The thickness of the outer wall of the epidermis		7.26±0.41	7.02±0.26
Height of the epidermis		27.06±0.83	25.02±1.19
Cavity diameter of large vessels		41.88±0.89	36.06±0.91
Epithelial cells of large secretory duct	height	15.12±0.57	13.26±0.49
	width	21.78±0.76	23.82±0.58
	quantity	10.40±0.16	13.40±0.33
Cavity diameter of large secretory ducts		37.56±1.14	53.82±1.91

**Table 3.** Quantitative characteristics of the middle petiole part of *F. tadshikorum*,  $\mu\text{m}$



**Figure 3.** The petiole anatomical structure of the virginile *Ferula tadshikorum* in natural populations (12–14 years). **a–d** – details of the marginal and central parts of the petiole base, **e–g** – details of the adaxial and abaxial sides of the middle part of the petiole, **h–i** – details of the adaxial and abaxial sides of the upper part of the petiole. Scale bar 100  $\mu\text{m}$ .

### **Botanical Garden. Virginile plant, fourth year of vegetation.**

The morphological description of the plant's 4 years of vegetation in the virginile age state is given in our previously published work (Khamraeva et al. 2023).

**Root.** The root is rod-shaped, cylindrical, covered with annular folds for a short distance, and thin in the apical zone. The root neck has remnants of conductive bundles of leaf petioles from previous years of vegetation. The cork of the root is brown, cracking and easily falling behind. Along the periphery of the root, a multilayer cork is formed from narrow tabular cells (Fig. 4a). The secondary bark is multilayered (Fig. 4b). Above the phloem there are numerous secretory ducts, which are with 6 epithelial cells (Fig. 4c; Table 1). There are cells around the epithelial cells of the ducts.

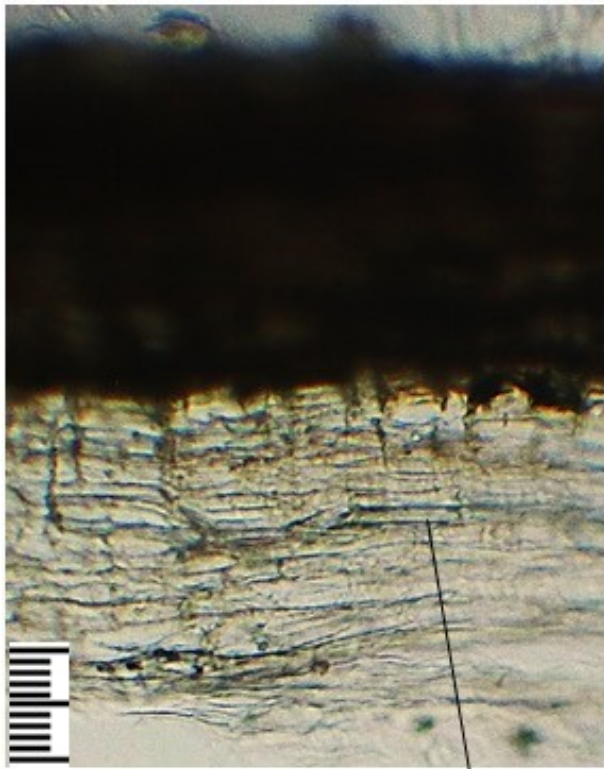
The conductive bundles are arranged in separate circles; as a result, concentric groups of bundles are formed with a xylem in the center and a phloem around it (Fig. 4d). Cambium is bundled, multi-layered (Fig. 4e). Between the circles of conductive bundles, the bark parenchyma is abundantly filled with starch grains. The vessels are arranged in radial chains; in each group there are up to 5–8 large and several small vessels.

**Leaf.** On the cross section, the leaf is lamellar, rounded-triangular in the area of the central vein, and biconvex on the sides in the places of conductive bundles (Fig. 5a). On the abaxial side, the

surface is less wavy than in natural individuals, covered with simple trichomes, and on the adaxial side it is ribbed, and there are rare awlshaped unicellular trichomes. The abaxial and adaxial epidermis are single layered, their outer walls are almost uniformly thickened, and both epidermises are covered with a serrated cuticle (Figs 5b, c; Table 2). The area of the central vein of the leaf is characterized by the following signs:

- under the abaxial and adaxial epidermis there is a single layer hypoderm of smaller, rounded-oval cells;
- the collenchyma strands are located in the ribs from the abaxial side of 7–12 rows, adaxial – 5–9 rows;
- on the abaxial side, above the medium and small bundles, the hypoderm and/or groups of collenchyma cells break off from the central leaf tissue, resulting in several narrow air-bearing cavities, and on the adaxial part they are single;
- the aquifer parenchyma consists of thin-walled round oval cells of up to 37–40 layers, of various sizes, and it has air-bearing cavities.

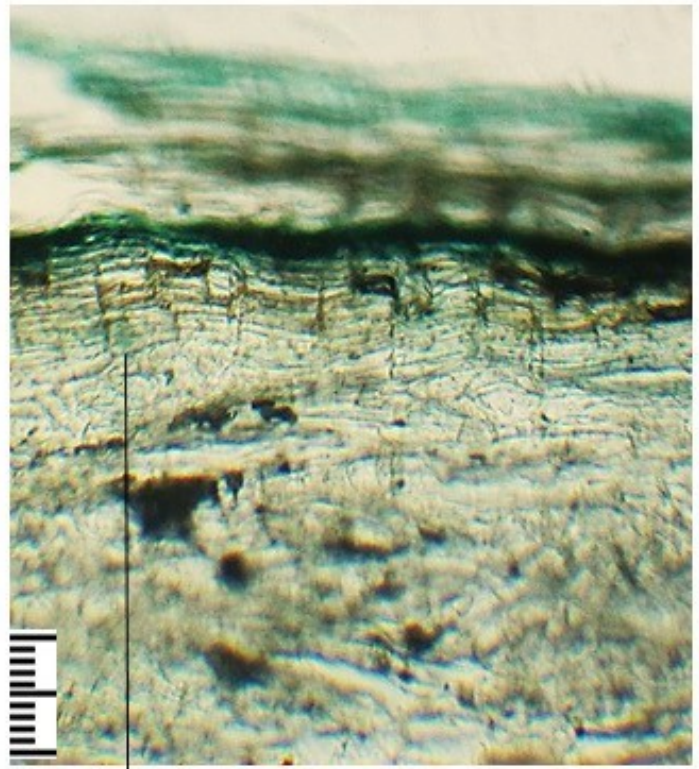
The mesophyll is an isolateral palisade. In the area of the central vein of the leaf on the adaxial side, the palisade parenchyma is 2-layered, the layer under the epidermis consists of more elongated cells, and the second layer is in some places more or less shortened. From the abaxial part, there is 2–3-layer palisade tissue, a layer under the epidermis of elongated cells, and the other 1–2 layers of shorter, almost rounded cells. In the flattened lateral parts of the plate, the palisade parenchyma 2-layered; from the adaxial side of elongated cells, and the abaxial part under the epidermis, the cells are short, almost rounded, the second layer of columnar cells (Fig. 5d; Table 2). Between them, there is a 2–3-layer spongy parenchyma of horizontally elongated cells (Table 2).



B

a

C

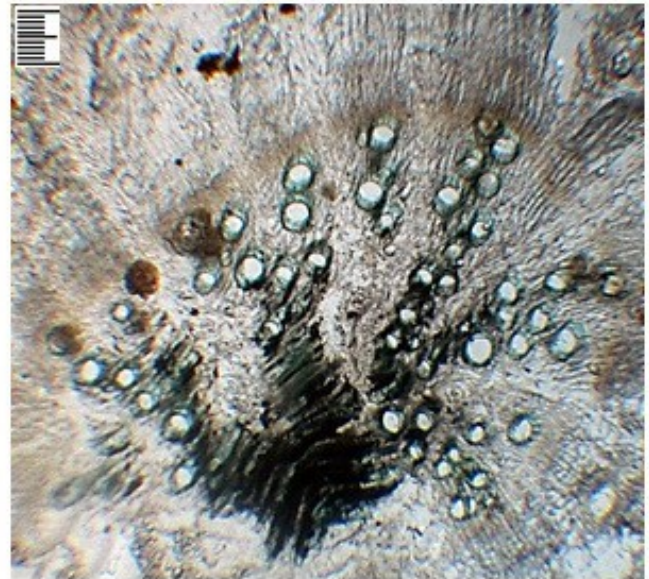


Phl

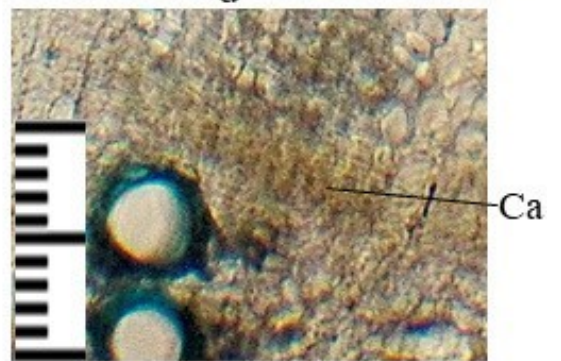
b



c



d



Ca

e

**Figure 4.** The main root anatomical structure of the virginile *Ferula tadshikorum* under culture conditions (4th year of vegetation). **a** – part of the periderm, **b** – detail of the secondary bark, **c** – secretory ducts in the bark parenchyma, **d** – part of the concentric circle of vascular bundles, **e** – detail of the cambial part. Scale bar 100  $\mu\text{m}$ .

Conductive bundles of collateral type. The central vein is multi-bundles, represented by several large, medium, and small peripheral and parallel three central conductive bundles. The peripheral bundles are located opposite the collenchyma strands; their size corresponds to the size of the bundle.

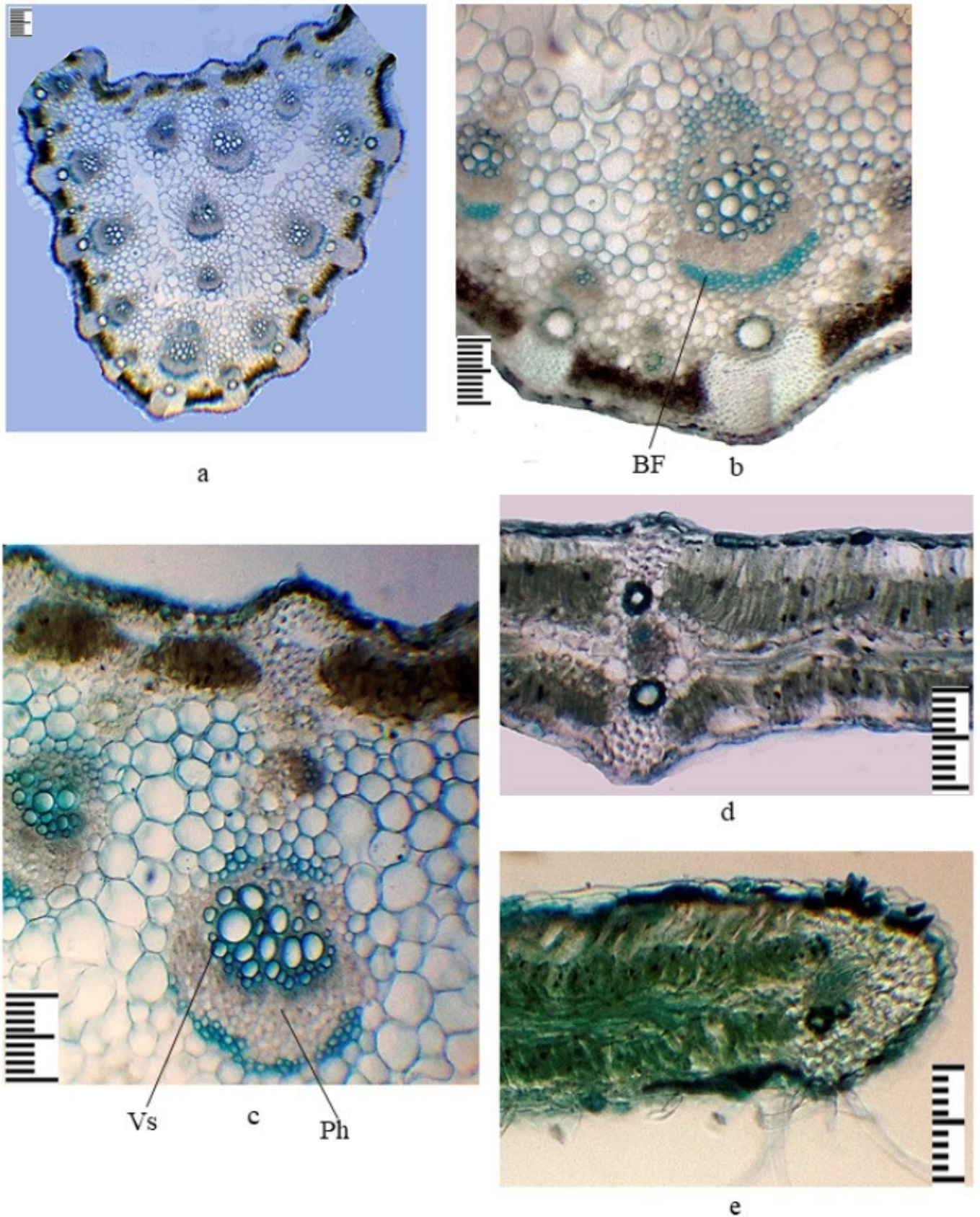
Above the phloem of large and medium bundles, there are bast fibers, and in large bundles, they are more multilayered. The vascular bundles of the central vein on the phloem side have a secretory duct, up to 12–14 epithelial cells on the abaxial side, and up to 8–10 epithelial cells on the adaxial side (Table 2). At the edge of the leaf, there are secretory ducts in the abaxial part with 6–7 epithelial cells (Fig. 5e).

In the lateral parts of the leaf, there are numerous conductive bundles of different sizes. There are no bast fibers above the lateral bundles. Above and below the large lateral bundles there is one secretory duct and collenchyma strands. On the abaxial side, the vascular bundles and strands of the collenchyma are larger than on the adaxial side. Large secretory ducts have up to 12–13 epithelial cells, small up to 9–10.

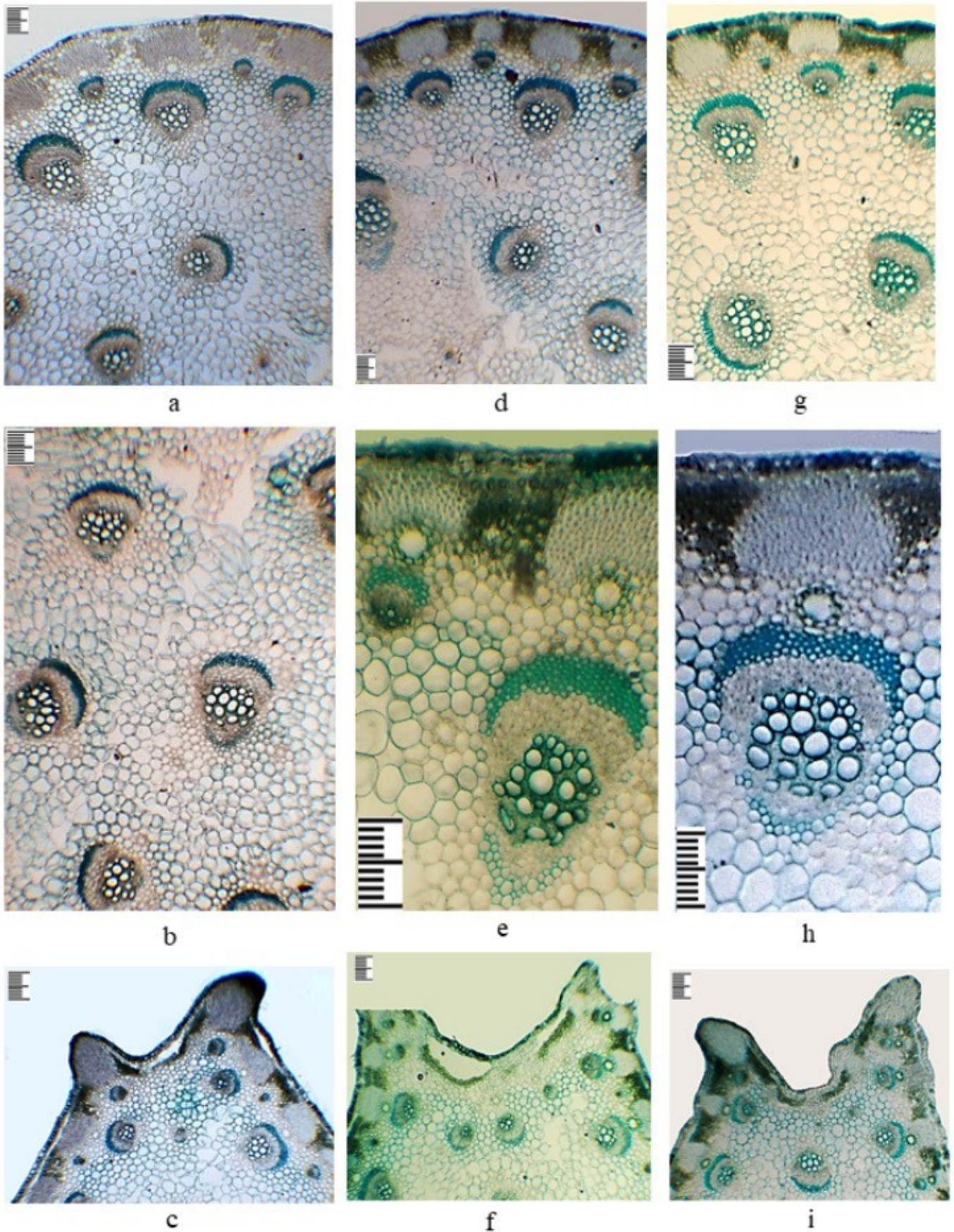
**Petiole. The base part.** On the cross section, the petiole is round-triangular in shape, on the adaxial part there are two outgrowths, at their ends there is a collenchyma strand (Fig. 6a). The epidermis is single-layered, there are simple trichomes on the surface, and under it there is a single-layered hypodermis, more small-celled, chlorophyll-bearing. The hypodermis is an 8-layer chlorophyll-bearing parenchyma, and the rest of the round-oval, thin-walled, parenchymal cells. Collenchyma strands in large peripheral vascular bundles have up to 17–19 rows, and over small bundles up to 7–8 rows. The petiole is represented by numerous vascular bundles, the outer circle is made up of large, medium, and small bundles, and the other bundles are also of various sizes, placed in the center, forming a longitudinal line in 8 rows (Figs 6b, c). All conductive bundles are of collateral type. Above the phloem of the bundles are bast fibers. The xylem of the bundles consists of small and large vessels. Between the collenchyma and the conductive bundle, there is a secretory duct its size corresponds to the size of the bundle. In small bundles, secretory ducts have up to 10 epithelial cells, and in large bundles up to 12–14. Near the large xylem of the peripheral bundles, there are small secretory ducts, including 2–3, which contain 6–8 epithelial cells. In the petiole parenchyma, there are abundant secretory ducts with 6–8 epithelial cells. In the parenchyma of the petiole, there are air cavities.

**The middle part.** The structure of the middle part is similar to that of the base. Differences relate only to the reduction in tissue size (Fig. 6 d–f). From the abaxial to the adaxial part of the petiole, the vascular bundles are located in the center in 7 rows (Table 3). The parenchyma 6–7 layered. The collenchyma over large bundles has up to 13–14 rows and over small bundles up to 6–9 rows. Above small bundles, there are secretory ducts of up to 10 epithelial cells, over large ones of up to 12–14 (Table 3).

**The upper part.** The structure is the same as the previous parts (Figs 6 g–i). There are the following differences: the cells of the epidermis are filled with chlorophyll grains. From the outer to the inner part of the petiole, the vascular bundles are arranged in 5 rows. The chlorophyll-bearing parenchyma is 5 to 6 layered. The collenchyma over large bundles has up to 10–13 rows, and small 6–8 rows. Large secretory ducts have up to 10–12 epithelial cells, and small ones have up to 9 cells. From the study, we determined that in the anatomical structure, the leaf and root of virginile plants, both from natural and introduced habitats, have several similar features, as well as some distinctive features.



**Figure 5.** The leaf anatomical structure of the virginile *Ferula tadshikorum* under culture conditions (4th year of vegetation). **a** - detail of the central part of the leaf, **b** - detail from the abaxial part of the central vein, **c** - detail from the adaxial part of the central vein, **d** - lateral part, **e** - the edge of the leaf. Scale bar 100  $\mu\text{m}$ .



**Figure 6.** The petiole anatomical structure of the virginile *Ferula tadshikorum* under culture conditions (4th year of vegetation). **a-c** - detail of the abaxial, central and adaxial parts of the petiole base; **d-f** - detail of the abaxial, central and

adaxial parts of the middle part of the petiole; **g-i** – detail of the abaxial, central and adaxial parts of the upper part of the petiole. Scale bar 100  $\mu\text{m}$ .

Analyzing the anatomical structure of the root of virginile *Ferula tadshikorum* of different years of life, it was revealed that the underground organs have a strong parenchymalization and filling of parenchymal cells with starch grains, as well as numerous secretory ducts in the thickened part of the root. We can emphasize that the structure of the root of the virginile plant *Ferula tadshikorum* is closely related to the duration of its life cycle and monocarpicity. Since nutrients accumulate in the roots throughout the entire life cycle and in the last year of life they are consumed to develop a shoot with a complex inflorescence.

In virginile individuals of *Ferulata dshikorum* at the age of 4 years, the root exhibits distinct concentric groups of bundles, with the cambium displaying a bundle structure. However, in natural individuals at 12–14 years of age during the virginile phase of development, the cambial line starts to level off within the root, beginning with the internal centroxylem bundles and progressing to the peripheral ones. As a result, the xylem and phloem, which belong to different circles of the conductive complex, come closer together. Consequently, the secretory ducts in the phloem assume a circular position relative to the central cylinder. Additionally, groups of libriform cells develop in the woody part of the root, providing strength to the underground organs as they grow in hard rocks.

These changes in the anatomical structure of the root were also noted in *Ferula iliensis* Krasn. Ex Korovin, with consideration of the plants' age and the functional differentiation of this organ, as reported by Safina and Isaeva (1981). Moreover, similar polycentric abnormal root structures were found in the desert monocarpic species *Ferula varia* (Schrenk) Trautv. and *Ferula foetida* (Bunge) Regel, as documented by Sharipova (2017).

In the leaf blade of virginile *Ferula tadshikorum* individuals of different ages and habitats, several common features were identified:

- A rounded triangular shape is observed in the cross-section around the central vein, accompanied by a single-layer hypoderm, numerous bundles, collenchyma strands, bundles of bast fibers, and secretory ducts above the phloem, along with a multilayer aquifer parenchyma in the center.
- The mesophyll exhibits an isolateral palisade type.
- In the flattened lateral parts on the adaxial side of the leaf, a 2-layer elongated palisade parenchyma and a 2–3-layer spongy parenchyma of horizontally elongated cells are present, with no bast fibers above the lateral vascular bundles.

Moving to the petiole of virginile specimens, the general structure follows a similar scheme, but with a decrease in the volume of the main tissues such as conductive bundles, chlorophyll-bearing parenchyma, and collenchyma strands from the base to the upper part. The overall anatomical characteristics of the petiole are as follows:

- Under the epidermis, a smaller-celled single layer hypodermis is present.
- Circles of collenchyma and chlorophyll-bearing parenchyma alternate in the petiole.
- Peripheral conductive bundles of various sizes, along with central ones in multiple rows, are visible.
- The central part of the petiole consists of thin-walled parenchymal cells.
- Peripheral bundles are equipped with a secretory duct on the phloem side and have 2–3 small



secretory ducts on the xylem side.

These observations provide valuable insights into the anatomical features of *Ferula tadshikorum*, shedding light on the root and leaf structures and their changes during different stages of development and in diverse habitats.

According to Tyurina et al. (1976), *Ferula capsica* M.B., a plant commonly found in Southeastern Altai, possesses xeromorphic features in its leaf. These features include the isolateral palisade type of mesophyll, a high percentage of palisade, and a thickened cuticle, indicating its adaptation to arid conditions.

On the other hand, Butnik et al. (2009) observed specific leaf characteristics in *Ferula foetida*, a desert species growing in the Kyzylkum desert of Uzbekistan. These characteristics include the isolateral palisade type of mesophyll, the presence of collenchyma strands above and below vascular bundles, and secretory ducts under the central bundles. A comparative analysis of literature and their own data led to the conclusion that species within the same genus, but from different ecological habitats, can develop similar leaf structures, which is an adaptive evolution in the transformation of the assimilation apparatus. The isolateral palisade type of mesophyll, being a light xeromorphic leaf structure, is typical for plants in well-lit and arid environments. As a result of adaptation to environmental factors, this feature is found not only in the representative of mountain habitats, *Ferula tadshikorum*, but also in the desert species, *Ferula foetida*. Hence, it can be inferred that in plants during their virginile age state, both in natural and introduced individuals, the isolateral palisade type of mesophyll is a genetically fixed trait, possibly due to the historical development of the species itself and its systematic position. This assumption is supported by their previously published data on the leaf structure of introduced juvenile and immature plants with isolateral mesophyll palisade (Khamraeva et al., 2021).

In the structure of the leaf and root of virginile *Ferula tadshikorum* plants at different ages and habitats, the distinctive features are more quantitative than qualitative, as indicated in Table 4.

In conclusion, based on the data obtained, it is recommended to cultivate *Ferula tadshikorum* in new locations to conserve natural populations and facilitate industrial harvesting of its raw materials from plantations. Our successful experiments on introducing this valuable medicinal plant in the Tashkent Botanical Garden support this recommendation.

The comparative analysis of introduced plants with natural individuals revealed shared anatomical characteristics in the roots and leaves of juvenile plants, indicating structural preservation. The main adaptive features in the leaves include a thickened outer wall of the epidermis, collenchyma strands, bast fibers, and the presence of isolateral palisade mesophyll. Meanwhile, in the roots, strong parenchymization and the presence of starch grains in all parenchymal cells, along with an abundance of secretory ducts, play crucial roles.

These distinctive features in the roots and leaves can also be utilized for species identification, taxonomy, and understanding the phylogeny of the *Ferula* genus. Therefore, they provide valuable insights for further research and conservation efforts.

12–14 years of life, Surkhandarya region, Babatag	Fourth year of vegetation, Tashkent Botanical Garden
Root	
In the xylem, there are groups of cells from the libriform	Absent
Leaf blade	
The outer walls of the cells of the abaxial epidermis are most thickened	Uniform thickening of the cell walls of both epidermis
In the area of the central vein, the collenchyma strands are more layered on the abaxial part up to 7–14 rows, which is why the surface is strongly ribbed	In the area of the central vein collenchyma strands are more layered on the adaxial part up to 5–9 rows, as a result, the surface is ribbed, and there are rare awl-shaped unicellular trichomes

In the area of the central vein on the abaxial side, there are ruptures under the epidermis, which is why wide air-bearing cavities are formed	Narrow air-bearing cavities are formed
In the area of the central vein, the aquifer parenchyma reaches 42–45 rows	It reaches 37–40 rows
On the abaxial side, the palisade cells are more elongated, and the length-width ratio of length to width is 2.8:1	On the abaxial side, the palisade cells are less elongated, but the ratio of length to width is 3.3:1
Petiole	
The shape of the cross section in the lower part is rounded-oval, and in the other parts two outgrowths on the adaxial part	The shape of the cross section is rounded- triangular in all parts, and there are two outgrowths on the adaxial part

**Table 4.** Distinctive features in the structure of the root and leaf of the *Ferula tadshikorum* at different age and habitats

## References

- Akalın E, Tuncay H, Olcay B, Miski M (2020) A New *Ferula* (Apiaceae) species from Southwest Anatolia: *Ferula pisidica* Akalın & Miski. *Plants* 9(740): 1–11. <https://doi.org/10.3390/plants9060740>
- Akhmetova A, Mukhitdinov N, Ydyrys A (2015) Anatomical indicators of the leaf structure of *Ferula iliensis*, growing in the eastern part of Zailiyskiy Alatau (Big Boguty Mountains). *Pakistan Journal of Botany* 47(2): 511–515.
- Barykina RP, Gulenkova MA (1985) Ontogenetic anatomy and its importance for taxonomy and phylogeny. *Moscow University Biological Sciences Bulletin* 10(6): 82–92. [In Russian]
- Barykina RP, Chubatova NV (2005) A Large Workshop on the Ecological Anatomy of Flowering Plants. Association of Scientific Publications, Moscow, 77. [In Russian]
- Barykina RP, Alyonkin VYu (2017) Comparative anatomy of vegetative organs in some species of *Coldenia l.* and *Tiquilia pers.* (Boraginaceae) in relation to their ecology, life forms and systematic position. *Moscow University Biological Sciences Bulletin* 122 (2): 55–67. [In Russian]
- Bondar YuV, Zerkal SV, Hetko NV, Rysin SL (2021) Comparative anatomical structure of the leaf *Rhododendron catawbiense* Michx. and *Rhododendron brachycarpum* D. Don, growing under conditions of N.V. Tsitsin main botanical garden of the Russian Academy of Sciences. *Samara Journal of Science* 10(1): 33–40. <https://doi.org/10.17816/snv2021101104>[In Russian]
- Butnik AA, Ashurmetov OA, Nigmanova RN, Begbaeva GF (2009) Ecological anatomy of desert plants of Central Asia. *Herbs. Tashkent*, 155 pp. [In Russian]
- Chen X-Y, Liu Q-X (1989) Chemotaxonomic studies in *Ferula* of China on the basis of flavonoids. *Acta Phytotaxonomica Sinica* 27(3): 184–189. [In Chinese]
- Elghwaji W, ElSayed AM, ElDeeb KS, El-Sayed AM (2017) Chemical composition, antimicrobial and antitumor potentiality of essential oil of *Ferula tingitana* L. Apiaceae grow in Libya. *Pharmacognosy Magazine* 13(51): 446–451. [https://doi.org/10.4103/pm.pm\\_323\\_15](https://doi.org/10.4103/pm.pm_323_15)
- Eser N, Yoldas A (2019) Identification of heat-resistant chemical components of *Ferula elaeochoytris* root extracts by gas chromatography mass spectrometry. *Tropical Journal of Pharmaceutical Research* January 18 (1): 55–60. <https://doi.org/10.4314/tjpr.v18i1.9>
- Fan C, Li X, Zhu J, Song J, Yao H (2015) Endangered Uyghur medicinal plant *Ferula* identification through the second internal transcribed spacer. *Evidence-Based Complementary and Alternative Medicine. Special Issue*: 1–6. <https://doi.org/10.1155/2015/479879>

- Halkuzieva MA, Khamraeva DT, Bussmann RW (2022) Bio-morphological properties of *Ferula tadshikorum* Pimenov and *Ferula foetida* (Bunge) Regel under plantation conditions. *Plant Science Today* 9(sp3): 79–84. <https://doi.org/10.14719/pst.1863>
- Khamraeva DT, Grabovec NV, Bussmann RW, Khojimatov OK (2021) Leaf morphological and anatomical structure of pregenerative individuals of *Ferula tadshikorum* in *ex situ* conditions. *Acta Biologica Sibirica* 7: 193–210. <https://doi.org/10.3897/abs.7.e63714>
- Khamraeva DT, Tukhtaeva DN, Khojimatov OK, Bussmann RW (2023) *Ferula tadshikorum* Pimenov – introduction, chemical composition and use in folk medicine. *Ethnobotany Research and Applications* 25(24): 1–10. <https://doi.org/10.32859/era.25.24.1-10>
- Khamraeva DT, Khojimatov OK, Uralov AI (2019) Growth and development of *Ferula tadshikorum* Pimenov in culture. *Acta Biologica Sibirica* 5(3): 172–177. <https://doi.org/10.14258/abs.v5.i3.6588>
- Khojimatov O, Khamraeva D (2018) Studying the structure of the pericarp of the fruits of the valuable medicinal plant *Ferulata dshikorum* and prospects for its cultivation in Uzbekistan. *Uzbek Biological Journal* 4: 45–48.
- Khojimatov OK, Khamraeva DT, Makhmudov AV, Khujanov AN (2019) Instructions for the seed cultivation of *Ferula tadsnikorum* Pimenov in the conditions of southern Uzbekistan. MUXR press, Tashkent, 44 pp.
- Khojimatov OK, Bussmann RW, Khamraeva DT (2021) Some aspects of morphobiology, conservation of resource potential, crop cultivation and harvesting of raw materials of promising *Ferula* species. *Ethnobotany Research & Applications* 22: 31. <https://doi.org/10.32859/era.22.31.1-8>
- Makhmudov AV (2019) *Ferula tadshikorum* Pimenov. Red Data Book of the Republic of Uzbekistan. *Plants* 2: 95–96. [In Russian]
- Padmavathi NV, Chetty MK (2019) Morpho-Anatomical studies of *Atalantia racemosa* Wight ex Hook., an important medicinal plant of Chittoor Dt. of Andhra Pradesh, India. *International Journal of Bio-Pharma Research* 8(2): 2491–2499. <https://doi.org/10.21746/ijbpr.2019.8.2.3>
- Panahi M, Banasiak Ł, Piwczyński M, Puchałka R, Kanani MR, Oskolski A, Modnicki D, Miłobędzka A, Spalik K (2018) Taxonomy of the traditional medicinal plant genus *Ferula* (Apiaceae) is confounded by incongruence between nuclear rDNA and plastid DNA. *Botanical Journal of the Linnean Society* 188: 173–189. <https://doi.org/10.1093/botlinnean/boy055>
- Rakhimov S, Denisova GR (2017) Some features of underground organs of *Ferula tadshikorum* M. Pimen. (*Ferula* L.). *Journal Bulletin of ASAU* 8 (154): 86–90. [In Russian]
- Safina LK, Pimenov MG (1984) *Ferula* of Kazakhstan. Science, Alma-Ata, 110 pp. [In Russian]
- Safina LK, Isaeva SN (1981) Anatomical analysis of the vegetative organs of two endemic species of *Ferula*. *Tidings of the Academy of Sciences of the Kazakh SSR* 6: 8–16. [In Russian]
- Sedelnikova LL (2000) Anatomical-morphological structure of leaf blade in the representatives of the genus *Crocus* L. in connection with introduction in Siberia. *Bulletin of the Main Botanical Garden* 179: 69–77. [In Russian]
- Tuncay HO, Akalın E, Dođru-Koca A, Eruçar FM, Miski M (2023) Two New *Ferula* (Apiaceae) Species from Central Anatolia: *Ferula turcica* and *Ferula latialata*. *Horticulturae* 9(144): 1–15. <https://doi.org/10.3390/horticulturae9020144>



Tyurina EV, Guskova IN, Valutskaya AG (1976) Apiaceae of Southern Siberia as a material for introduction. Nauka Publ., Novosibirsk, 256 pp. [In Russian]

Zaitsev GN (1990) Mathematics in experimental botany. Nauka, Moscow, 296 pp. [In Russian]