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# HISTO-ANATOMICAL STUDY OF THE VEGETATIVE ORGANS OF *HERACLEUM SPHONDYLIUM* SSP. *SPHONDYLIUM*

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**Abstract:** This study presents the first comprehensive histo-anatomical characterization of the root, stem, and leaf of *Heracleum sphondylium* ssp. *shpondylium*. Transverse sections revealed key structural features across all organs. The root exhibited a circular outline and secondary growth, with a periderm composed of phellem, phellogen, and phelloderm, a cortex containing aerenchyma and secretory canals, and a wide zone of secondary phloem and xylem, both traversed by medullary parenchyma. The stem displayed a layered organization from epidermis to central pith, including a cortex with collenchyma and secretory canals, and collateral vascular bundles forming a ring that defined the transition to the central cylinder. The dorsiventral leaf anatomy featured a two-layered palisade parenchyma on the adaxial surface and spongy parenchyma on the abaxial side. Secretory canals were consistently present in the cortex and vascular regions of all organs, and long, simple, unicellular trichomes with a basal crown of thick-walled cells were found on the leaf surface. These anatomical structures, particularly the secretory canals and trichomes, likely play a key role in the biosynthesis and release of furanocoumarins.

**Keywords:** *Heracleum sphondylium* ssp. *sphondylium*., hogweed, secretory canals, trichomes, plant histo-anatomy

## 1. Introduction

The genus *Heracleum* is represented in Romania by *H. carpaticum* (endemic in Europe), *H. palmatum* (endemic in the Romanian Carpathians), and *H. sphondylium* with three subspecies: *montanum* (European alpine), *sphondylium* (Eurasian), and *sibiricum* (European). The hybrid *H. x rodnense* (*H. carpaticum* x *H. sphondylium* ssp. *sphondylium*) is present in the Rodna Mountains. There have also been reports in Romania of two species escaping from cultivation: *H. mantegazzianum* and *H. sosnowskyi* (Sârbu et al., 2013).

*H. sphondylium* ssp. *sphondylium*, commonly referred to as hogweed or brânca ursului, is a biennial or perennial plant. It has white, orange-bruising roots. In the first year, a taproot is present. As it ages, a network of external vegetative meristems and related secondary taproots grows, expanding the root crown. Therefore, the root expands and thickens considerably with the years. The plant has a hollow stem with a furrowed surface, reaching a height of 50-200 cm. The leaves have 5-7 broad, lobed, and toothed segments and a less swollen vagina. The white or rarely pink flowers are 3 to 10 mm in size and grow in large umbels (up to 15 cm in diameter) with 12 to 25 rays. The outer flowers of the umbellet are zygomorphic with highly uneven petals, of which the larger ones are directed outwards. The tiny fruits are schizocarps (of 2 mericarps), which can grow up to 1 cm in length. Fruit shape can be from elliptical to rounded, winged, and flattened. Each mericarp has six resin canals (approximately as long as the fruit), two on the inner or commissural face and four on the outer, dorsal face (Sheppard, 1991; Sârbu et al., 2013; Benedec et al., 2017; Matarrese et Renna, 2023).

Furocoumarins (bergapten, isopimpinellin, and heraclenin) and essential oil were found in the seeds and roots of different *H. sphondylium* subspecies. Monoterpenes, sesquiterpenes, and phenyl-propanoids composed manly the extracted essential oils (Matarrese et Renna, 2023).

Regarding the phenolic composition, high levels of rutin were described by Benedec et al. (2017) in the leaves and flowers of the *H. sphondylium* ssp. *sphondylium*. In terms of other flavonoids, roots contain ferulic and chlorogenic acids, leaves have quercitrin, and flowers contain quercetin.

It was found that the essential oil from seeds of *H. sphondylium* ssp. *ternatum* is rich in 1-octanol and octyl butyrate and presents cytotoxic activity against tumor cells (Maggi et al., 2014). Other studies reported the antibacterial, antifungal, and antioxidant biological activities of the different plant extracts (methanol, ethanol, and aqueous) from *H. sphondylium* (Matarrese et Renna, 2023). Furthermore, the vasorelaxant property of the dichloromethane extract was also revealed (Senejoux et al., 2013). The fruit of *H. sphondylium* ssp. *montanum* had a yield of the essential oil of 4.39%, while the root presented a yield of 0.22%. However, the aerial parts of the same taxa had a noticeably lower yield 0.19%. The primary chemicals found in *H. sphondylium* ssp. *montanum* roots essential oil was composed of myristicin (13.8%) and octyl acetate (57.1%), while the fruits showed a rich profile, with myristicin (8.9%) and octyl acetate (73.7%) predominating. The aerial portions, on the other hand, showed a balanced distribution, with the primary essential oil chemicals being octyl acetate (56.1%), myristicin (24.2%), and hexyl butyrate (4.1%) (Kose et al., 2025).

Regarding the histo-anatomical structure of the organs in the case of *H. sphondylium*, Bradley and Fell (1966) described the tissues of the ripe fruits, while Laczkó-Zöld et al. (2023) presented the secretory tissues from various phases of fruit maturity. Bicchi et al. (1990) showed that the two different types of secretory structures (secretory canals and vittae) in *H. sphondyhum* ssp. *sphondylium* fruits store different chemicals, highlighting that chemical variations are linked to histological diversity. Arora et al. (1982) described the characteristics of the leaf epidermal trichomes in *H. sphondylium* using a scanning electron microscope (SEM).

Given that other descriptions of the anatomy of *H. sphondylium* have not been found in the scientific literature, the aim of the study was the detailed anatomical characterization of the vegetative organs of *H.*

*sphondyhum* ssp. *sphondylium*. This microscopic investigation is the initial phase of a larger project that aims to clarify the possible structures involved in bioactive substances secretion in case of this taxa.

## 2. Materials and methods

*H. sphondylium* ssp. *sphondylium* plants were harvested from the spontaneous flora in Sovata area, Romania (N46° 39.010' E24° 58.603'), and after identification, a voucher specimen (Voucher number: FS/1607/2022) was stored at the Department of Pharmacognosy, George Emil Palade University of Medicine, Pharmacy, Science and Technology of Târgu Mureş, Romania.

Before section preparation, root, stem, and leaf samples were preserved in 60% ethanol for the anatomical study. According to Tanase et al. (2017), ruthenium red and iodine green were used to stain rehydrated plant parts. A Canon EOS 250D (Canon Inc., Japan) camera and a light microscope (Ceti Topic-T, Belgium) were used to capture photographs.

## 3. Results and discussion

**Root structure**

The cross-sectional outline of the *H. sphondylium* root was circular (**Fig. 1 A, B, and C**), and the root displayed a secondary structure.

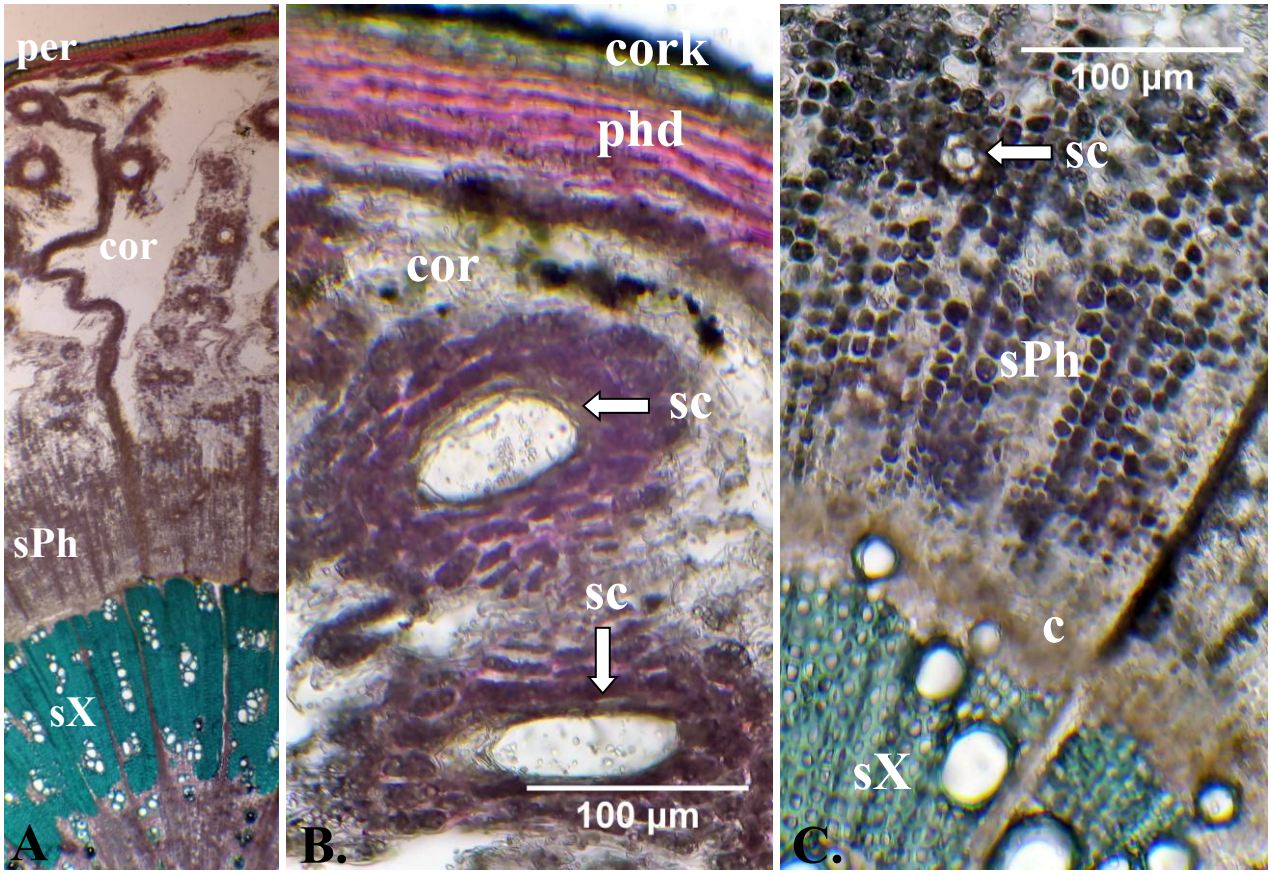


**Fig. 1.** The root structure of *Heracleum sphondylium* ssp. *sphondylium*: A. general view; B. the secondary cortex and the primary cortex with the secretory canals; C. the secondary phloem with secretory canals and the secondary xylem.

**Abbreviations: per**-periderm; **cork**-phellem; **phd**-phelloderm; **cor**-cortex; **sc**-secretory canal; **sPh**-secondary phloem; **c**-cambium; **sX**-secondary xylem (photographs by Erzsébet Domokos).

The following tissues, were identified, arranged from the outside of the root to the inside: the periderm or secondary cortex, which is made up of the phellogen, presented a thick layer of suber (phellem) and tabular cork cells (phelloderm); the cortex, which contained numerous secretory canals and aerenchyma; a broad zone of secondary phloem, also containing secretory canals (considerably smaller in diameter than those found in the cortex); and secondary xylem, approximately equal in width to the secondary phloem. Both the secondary phloem and xylem were radially traversed by medullary parenchyma. Similar root structures were described by Betekhtina et al. (2018) for *H. sosnowskyi* and Lee (1965) for *H. mantegazzianum*.

**Stem structure**

The stem has a circular-costate cross section. The early stages of transition between primary and secondary structures were visible (**Fig. 2 A, B, and C**). The following primary tissues could be identified from the outside to the inside of the stem: the epidermis, composed of large cells covered by a cuticle; long, irregularly distributed unicellular trichomes on the epidermis; the thin cortex with thick layers of collenchyma beneath the epidermis, next to the ribs; cortex containing also a few secretory canals above the vascular bundles; the collateral vascular bundles of various sizes arranged in a circle, demarcating the boundary between the cortex and the central cylinder; numerous secretory canals within the central cylinder (**Fig. 3**); and the central pith (central cylinder which lacks medullar parenchyma).

**Leaf structure**

The leaf cross-section at the level of the midrib presented the following structure (**Fig. 4. A and B**): an epidermis composed of polygonal and isodiametric cells covered by a cuticle; hypodermic collenchyma cords; secretory canals located both above and below the collateral vascular bundles, as well as within the fundamental tissue. The mesophyll is characteristic to the dorsiventral leaf, with a palisade parenchyma (columnar cells in two layers) located on the adaxial (upper) surface of the leaf, while spongy parenchyma on the abaxial (lower) side (**Fig. 5.**). Long trichomes were observed on the adaxial epidermis. The shape and the structure of the trichomes were in accordance with the description of Arora et al. (1982): simple, unicellular, and have 10-12 tiny and thick-walled crown cells surrounding them at the base.

According to Weryszko-Chmielewska and Chwil (2017), scanning electron microscopy examinations in case of *H. sosnowskyi* stem and leaves, showed that furanocoumarin crystals were present on the surface of the trichomes and other epidermal cells, but also in parenchyma cells. Additionally, they discovered that lipids, essential oil, polysaccharides, tannins, and furanocoumarins are present in these trichomes. The majority of furocoumarins are released by trichomes and epidermal cells. The release is concentrated in subepidermal cells and on the surface of trichomes. Additionally, the furanocoumarins are extruded from epidermal cells and go through the cuticular layer, forming crystals on the epidermis. Strong fluorescence from the trichomes, which are most prevalent on the abaxial leaf surface, suggests that furanocoumarins are present in their secretions (Weryszko-Chmielewska and Chwil, 2014; Bruni et al., 2019).

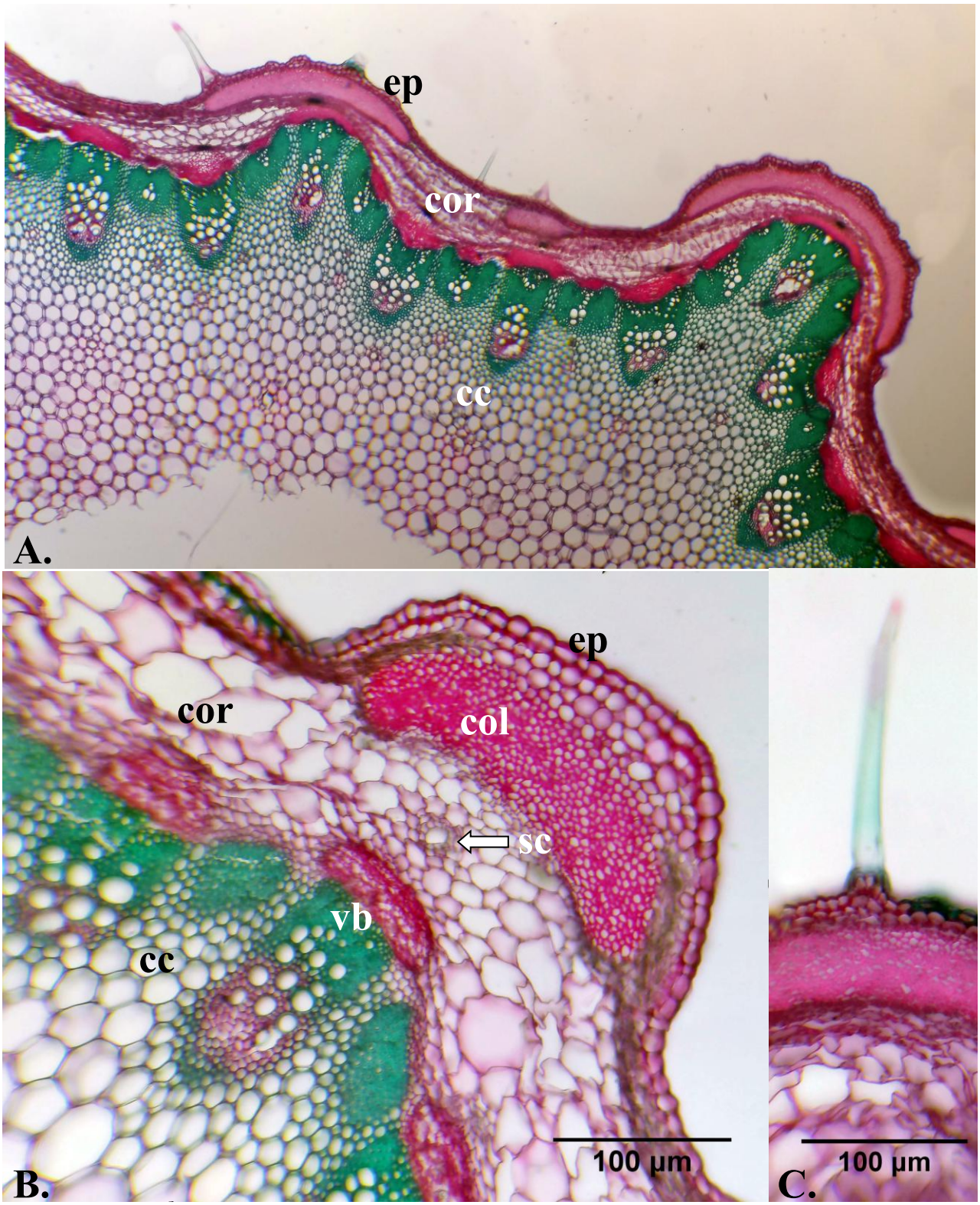


**Fig. 1.** The stem structure of *Heracleum sphondylium* ssp. *sphondylium*: A. general view; B. the epidermis, the cortex, and the vascular bundles from the central cylinder; C. unicellular trichome on the stem epidermis surrounded by crown cells

**Abbreviations: ep**-epidermis; **cor**-cortex; **cc**-central cylinder; **col**-collenchyma; **sc**-secretory canal; **vb**-vascular bundle (photographs by Erzsébet Domokos).

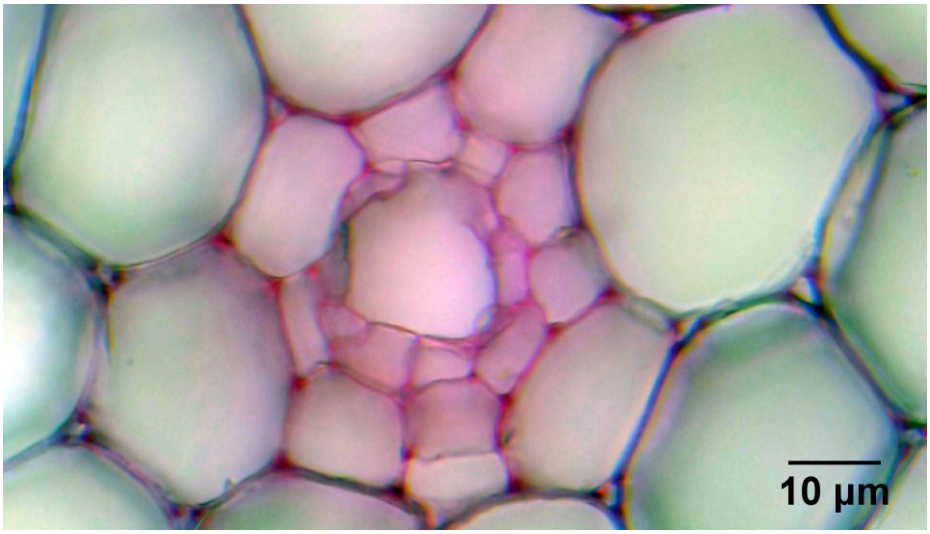


**Fig. 3.** Secretory canal with epithelial cells from the stem's central cylinder in case of *Heracleum sphondylium* ssp. *sphondylium* (photograph by Erzsébet Domokos).

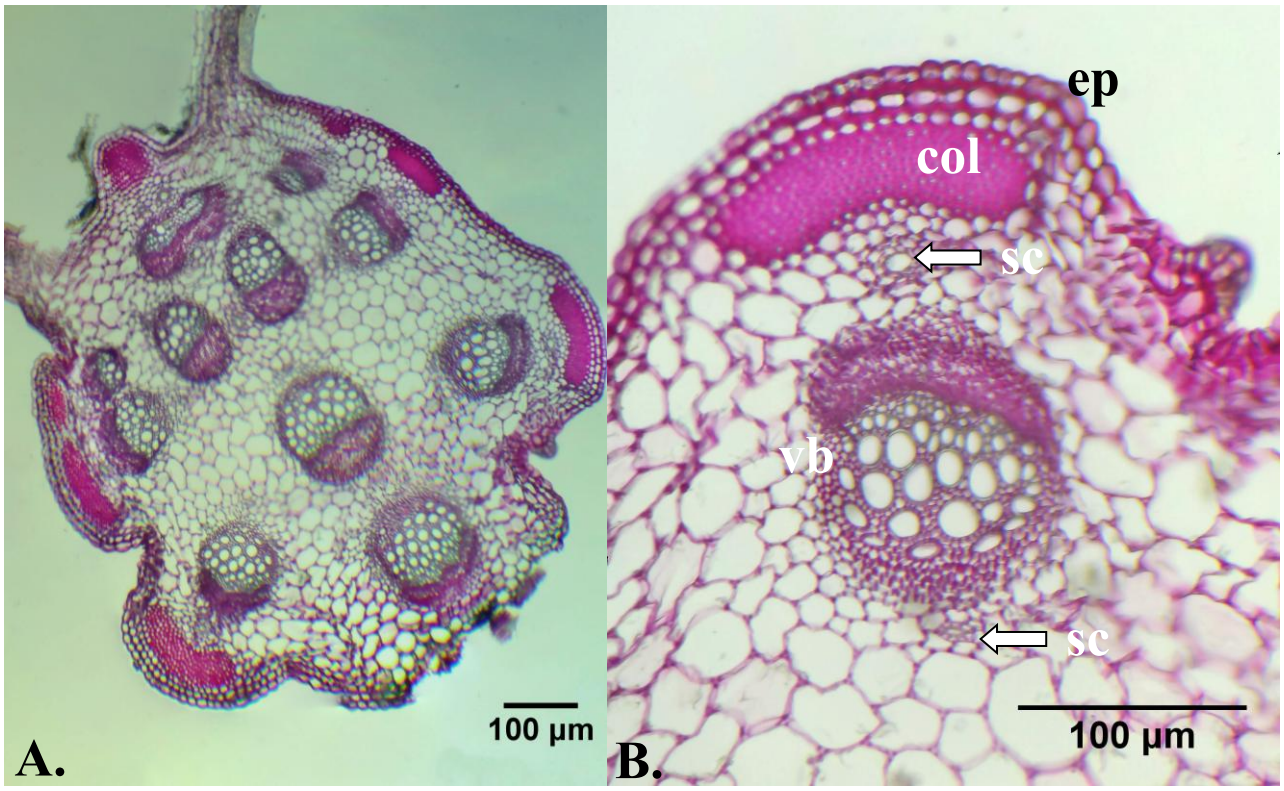


**Fig. 4.** Leaf cross-section at midrib in case of *Heracleum sphondylium* ssp. *sphondylium*: A. general view; B. secretory canals near the vascular bundle.

**Abbreviations: ep**-epidermis; **col**-collenchyma; **sc**-secretory canal; **vb**-vascular bundle (photographs by Erzsébet Domokos).


**Fig. 5.** Dorsiventral leaf structure of *Heracleum sphondylium* ssp. *sphondylium* with a large unicellular trichome on the lower epidermis (photograph by Erzsébet Domokos).

## Conclusions

This study provides the first detailed histo-anatomical description of the root, stem, and leaf of *Heracleum sphondylium* ssp. *sphondylium*. It was found that secretory canals are present in the cortex and secondary phloem of the root, in the cortex and parenchymal tissues of central cylinder from the stem, while in the leaf these secretory structures are located near the vascular bundles and in the fundamental parenchyma. These secretory tissues, along with the trichomes on the stem and leaf surfaces, likely play an important role in the production and secretion of furanocoumarins.

## Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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