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Anatomical Structure of the Vegetative and Generative Organs of *Haplophyllum Perforatum* (Rutaceae) Growing in the Conditions of Uzbekistan

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ABSTRACT— It is known that morphological and anatomical analysis is an important method for identifying medicinal plant materials (MPR). To date, not all types of pharmacopoeial plants have an optimal description of microscopic diagnostic signs. This article presents the study of the morpho-anatomical structure of the vegetative and generative organs of the medicinal, alkaloid and essential oil plant Haplophyllum perforatum, growing in the conditions of Uzbekistan, and identifies characteristic diagnostic signs, and also determines the localization of biologically active substances. The revealed features are species-specific diagnostic signs and can be used as additional diagnostic signs for the identification of species and plant materials.

KEYWORDS: anatomy, vegetative and generative organs, *Haplophyllum perforatum,* Uzbekistan.

1. INTRODUCTION

In Uzbekistan grow 16 species of the genus Haplophyllum Juss. (whole leaf). Their ecological optimum is adyrs and foothills. In the desert area there are 7 species, some of them marked only in limited areas [1], [2]. Species of the genus Haplophyllum Juss. (Rutaceae), are of interest as sources of alkaloids, essential oils, coumarin compounds, and other substances with valuable pharmacological properties [3]. Representatives of Haplophyllum contain broad-acting alkaloids and can be used for diseases of the digestive and respiratory organs, as well as narcotic [4]. In terms of the number of isolated alkaloids and their structural diversity, the species of the genus Haplophyllum are unique, since they contain new representatives of almost all known varieties of quinoline alkaloids, which are rich in plants of the Rutaceae family, as well as peculiar quinoline alkaloids that are not found in plants of other genera of this family [5], [6]. Valuable alkaloids of the Haplophyllum species are found mainly in the leaf and in small amounts in the stem, and also accumulate in the ovaries and are localized in the seeds [2]. The literature contains information about the location of CB in the leaf, shoot bark, and pericarp of the mature fruit of R. graveolens and their structure [7], [8]. The plant Haplophyllum perforatum is a rich source of biologically active substances such as alkaloids, terpenoids and others [10]. A review of materials on the study of coumarins, flavonoids, and lignans of five plant species of the genus Haplophyllum (family Rutaceae) is carried out. It was found that each studied plant species produces coumarins of a certain structural type, and the qualitative and quantitative composition of coumarins varies depending on the ecological- geographical, soil-climatic conditions, as well as the growing season of plant organs. Some structural features and methods for the identification and detection of coumarins on TLC have been revealed. A possible scheme for the biogenesis of coumarins in the H. obtusifolium Ledeb plant is proposed. Based on the analysis of the obtained data and taking into account the scheme of coumarin biogenesis for obtusin, obtusifol, and tusifolidiol, an alternative structure was proposed [9]. The species is widespread in the territory of the Republic of Uzbekistan. The insecticidal activity of this

plant was also studied [11]. Haplophyllum perforatum punctured are a huge source of secondary compounds. Many substances perform protective functions, participate in growth processes and ensure plant resistance to biotic and abiotic stresses. The compounds have antimicrobial, toxic effect against insects, growth-stimulating properties.

It is known that the modern practice of plant growing, growth regulators are considered as an ecologically clean and economically profitable way to increase the productivity of agricultural crops, allowing the use of energy-saving technologies and maximizing the physiological capabilities of plants [1, 2]. They not only increase the overall yield, but also have a positive effect on product quality. Recently, much attention has been paid to herbal preparations that increase the adaptive properties and immunity of agricultural crops, have antimicrobial properties [3], [4]. To date, not all types of pharmacopoeia plants have an optimal description of microscopic diagnostic signs. We have studied the morpho-anatomical structure of some medicinal plants in the conditions of Uzbekistan and identified characteristic diagnostic signs, and also determined the localization of biological active substances [21], [22]. In this regard, the study of the anatomical structure of the aboveground organs of the medicinal plant H. perforatum growing in the conditions of Uzbekistan is of scientific interest. This determines the relevance and novelty of our research. Our aim of the research work is to study the anatomical structure of the vegetative and generative organs of the medicinal species H. perforatum, to identify the structural features and localization of biologically active substances growing in the natural conditions of Uzbekistan.

2. Materials and methods

The objects of the study were wild plants Haplophyllum perforatum (M. Bieb.) Kar. & Kir. ex Vved. Is a perennial, green or bluish, herbaceous plant covered with pinpoint glands and is widespread throughout Central Asia (Kizylkum, Fergana Valley, Mirzachul, Zarafshan Valley, Karshi Steppe, Tien Shan, Pamir-Alai [8]. The material was collected in mid-May 2020 in the flowering phase on a sanitary protection zone, a clay slope growing in the Tashkent region, Kibray district, Ulugbek settlement - 41 ° 40'60.19 "N 69 ° 45'8361" (Fig. one). Simultaneously with the morphological description, the aboveground plant organs were fixed in 700 ethanol for anatomical study. The epidermis was examined on paradermal and cross sections. Each fabric was described - according to [23], [24]. Cross sections were made through the middle of the leaf, calyx, petals, and base of the petiole, stipule, pedicel, and stem. Hand- prepared preparations were stained with methylene blue, safranin, followed by gluing in glycerin-gelatin [25]. Micrographs were taken with a computer microphoto attachment with a Samsung ES70 digital camera and a Canon A123 digital camera under a Motic B1-220A -3 microscope.

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Figure - 1. Appearance of Haplophyllum perforatum in the flowering phase (photo Tulkin Tillaev).

3. Results and discussion

The leaves of Haplophyllum perforatum are glabrous, from broadly oblong to lanceolate, sharp or obtuse, gradually narrowing into a short petiole, entire. On the paradermal section, the outlines of epidermal cells are rectilinear-rounded, the projection is polygonal. The cells of the adaxial epidermis are larger than those of the abaxial epidermis. Leaves are amphistomatic. The stomata are located transversely to the longitudinal axis of the leaf. The shape of the stomata is round-oval. The stomata are most numerous on the abaxial side, less on the adaxial side. Stomatal guard cells on both sides of the leaf are almost the same length. The stomata are submerged. The stomata are anomocytic and hemiparacytic (Fig. 1). The leaf mesophyll on the transverse section of the isolateral-palisade type, which is represented by 2-3 rows of palisade cells on both sides of the leaf and a spongy layer of varying thickness between them. The epidermis is represented by one row of cells with a thick-walled cuticle layer. Palisade parenchyma is large and long 1-2 row, smaller and shorter - 3 row. The spongy parenchyma is round, small-celled, consists of 2-3 rows with cavities. The palisade parenchyma is chlorophyll-bearing; among the palisade parenchyma there are 3-4 secretory receptacles of the schizo-lysigenic type. Lateral vascular bundles are closed, collateral, numerous, with 3-4 small vessels (Fig. 1). The main vein protrudes on the abaxial side. Under the epidermis there is a lamellar 1-2 row collenchyma and a 4-5 row parenchyma. The main vein has 1 conductive bundle. The main conducting bundle is closed collateral, which consists of phloem and xylem (Fig. 1).

The leaf petiole on the cross section is heart-shaped, parenchymal-bundle type, consists of a single-row epidermis, two-three-row collenchyma, palisade parenchyma and one conducting bundle. The petiole protrudes on the abaxial side. Lamellar 2-3 row collenchyma is located under the epidermis (Fig. 1). At the edges of the petiole under the collenchyma, there are palisade chlorophyll-bearing parenchymal cells, consisting of 3 rows. There is 1 conductive bundle in the center of the petiole. The conducting bundle is closed collateral, heart-shaped and more sclerified, which consists of phloem and xylem. The arrangement

of the vessels of the conducting bundle is chain-like. There are no secretory receptacles (Fig. 1).

The stem erect, corymbose-branched, glabrous, 30-70 cm in height. The base of the stem on the cross section is rounded, bunch-type, more lignified. The epidermis is single-row, rounded-oval, with a thick outer wall. A lamellar 4-5 row collenchyma is located under the epidermis. Under the collenchyma there is a rounded-oval cortex parenchyma, which consists of 5-6 rows and is preserved throughout. In the collenchyma and the cortex part of the stem, at a certain section, one secretory receptacle of the schizo-lysigenic type is found, which has an outlet to the surface, through which the secretion evaporates. Groups of bast fibers are formed under the bark parenchyma. The phloem is extensive, located between the bovine parenchyma and the libriform. Wood takes up most of the stem. Vessels are large, small, rounded and oval, located radially; radial rays are single-row, elongated and short. Libriform is extensive, thin-walled. The core is not wide, represented by large and small rounded-oval cells, with cavities (Fig. 2).

The peduncle on the cross section is rounded, bundle type, lignified. The epidermis is single-row, rounded-oval, with a thin outer wall. A lamellar 1-2 row collenchyma is located under the epidermis. Under the collenchyma there is a rounded-oval cortex parenchyma, which consists of 5-6 rows and is preserved throughout. In the collenchyma and the cortex part of the peduncle, there is a secretory receptacle of the schizo-lysigenic type, which has an outlet to the surface. Groups of bast fibers are formed under the bark parenchyma. The phloem is extensive, located between the bovine parenchyma and the libriform. Wood takes up most of the peduncle. Vessels are large, small, rounded and oval, located radially; radial rays are single-row, elongated and short. Libriform is extensive, thin-walled. The core is not wide, represented by large and small rounded-oval cells, with cavities (Fig. 3).

The pedicel on the cross section is rounded, bundle type. The epidermis is single-row, rounded-oval, with a thin outer wall. A lamellar 1 row collenchyma is located under the epidermis. Under the collenchyma there is a rounded-oval cortex parenchyma, which consists of 5-6 rows and is preserved throughout. In the collenchyma and the cortex part of the peduncle, there is a secretory receptacle of the schizo-lysigenic type, which has an outlet to the surface. Groups of bast fibers are formed under the bark parenchyma. The phloem is extensive, located between the bovine parenchyma and the libriform. Vessels are large, small, round and oval. Libriform is extensive, thin-walled. The core is not wide, represented by large and small rounded- oval cells, with cavities (Fig. 3).

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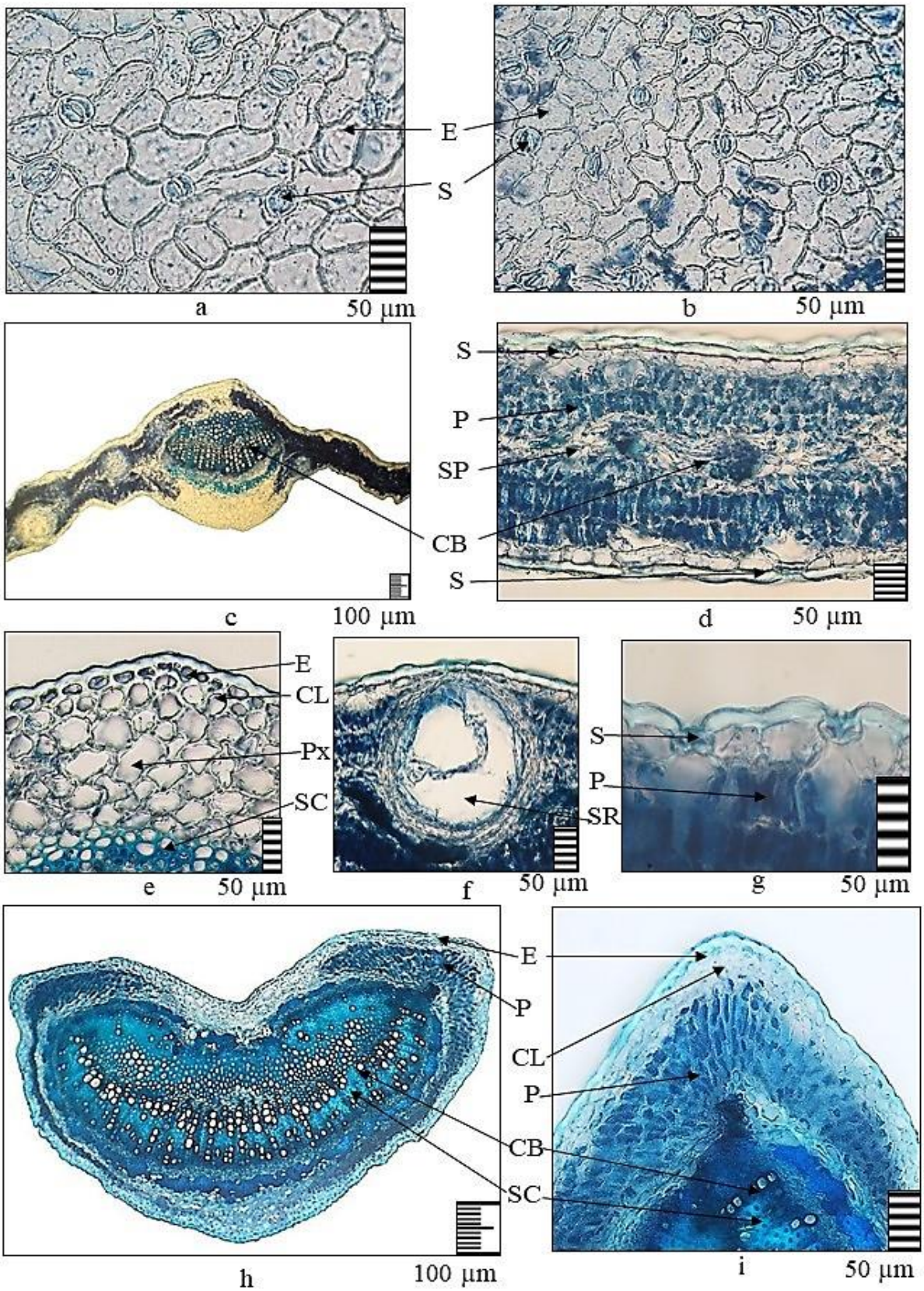

Figure - 1. The structure of the epidermis, mesophyll of the leaf (a-g) and the leaf petiole (h-i) of H. perforatum: a - adaxial epidermis; b - abaxial epidermis; c-d - leaf mesophyll; e- epidermis and collenchyma in the main vein of the leaf; f - secretory receptacle in the leaf mesophyll; g - submerged stomata, h - general view of leaf petiole, i - epidermis, collenchyma and palisade parenchyma. Legend: CB - conducting beam,

CL - collenchyma, E – epidermis, P - palisade parenchyma, Ph - phloem, S - stomata, SC - sclerenchyma, SP - spongy parenchyma, SR - secretory receptacles. Magnification – 50-100 micron.

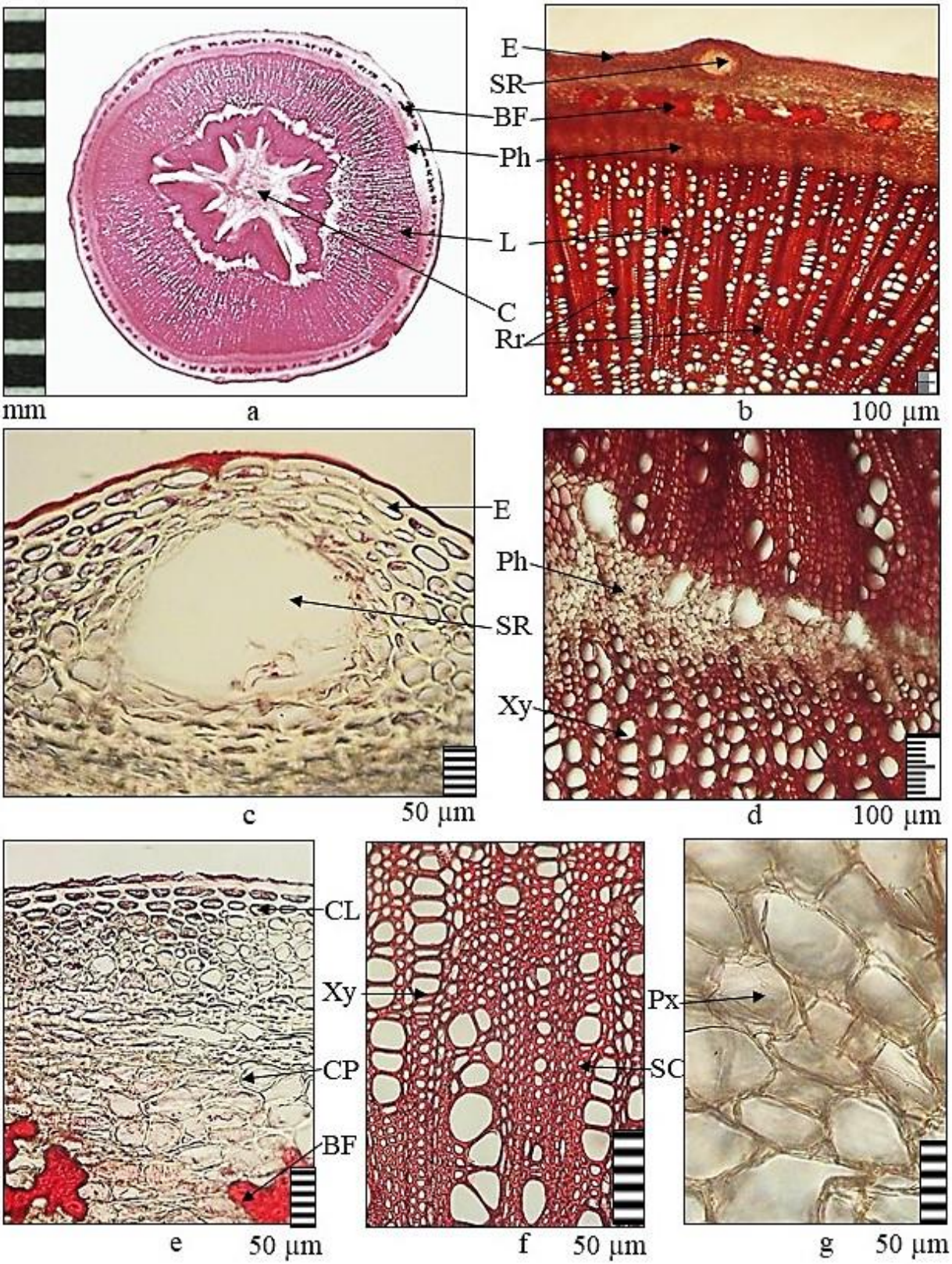

Figure - 2. The structure of the stem of H. perforatum:

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a-b - general view of the stem; c - secretory receptacle in the stem; d-f - libriform and radial rays; e - cortex parenchyma; g - core. Legend: BF - bast fibers, C – core, CB - conducting bundle, CL - collenchyma, CP - cortex parenchyma, E – epidermis, L - libriform, Ph - phloem, Px - parenchyma, RL - radial rays, SR - secretory receptacle, SC - sclerenchyma, Xy - xylem, Magnification – 50-100 micron.

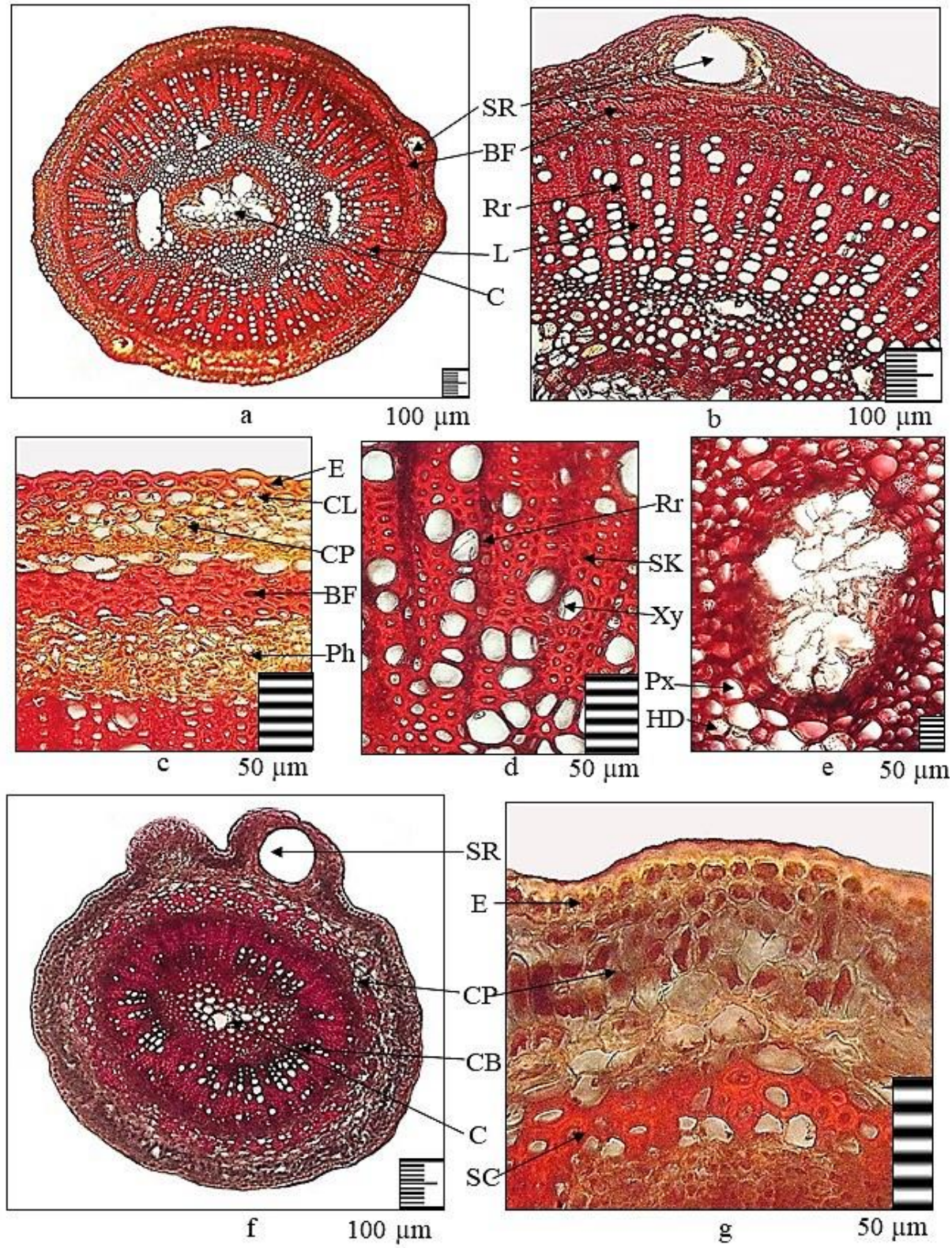

Figure - 3. The structure of the peduncle (a-e) and pedicel (f-g) of H. perforatum:

a-b - general view of the peduncle, c-g - cortex parenchyma, d - libriform and radial rays; e – core, f - general view of the pedicel. Legend: BF- bast fibers, C – core, CB - conducting bundle, CL - collenchyma, CP - cortex parenchyma, E – epidermis, L - libriform, Ph - phloem, Rr - radial ray, Px - parenchyma, SC - sclerenchyma, SR – secretory receptacle, Xy – xylem. Magnification – 50-100 micron.

4. Conclusions

Thus, the anatomical structure of the vegetative and generative organs of H. perforatum has been studied. The following diagnostic signs were revealed: in the leaf - rectilinear-rounded outline of epidermal cells; submerged stomata of anomocytic and hemiparacytic types; isolateral-palisade type of leaf mesophyll; palisade chlorophyll-bearing parenchyma; secretory receptacles of the schizo-lysigenic type; in the petiole-parenchymal-fascicular type of structure; lamellar collenchyma is located under the epidermis; at the edges of the petiole, the palisade parenchyma is chlorophyll-bearing; the conducting bundle is closed collateral and more sclerified; there are no secretory receptacles; the stem, peduncle and peduncle are similar in structure, bundle type and more lignified; the secretory receptacle of the schizo-lysigenic type, they are located closer to the epidermis in order to evaporate the secret; the radial rays are elongated and short; the libriform is extensive, the core is not wide. Biological active substances are localized in secretory receptacles and parenchymal cells of the cortex. The revealed features are species-specific diagnostic signs and can be used as additional diagnostic signs for the identification of species and plant materials.

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