

Morphological and Anatomical Investigations on Turkish Endemic *Hieracium lasiochaetum* (Bornm. & Zahn) Sell & West (Asteraceae)

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ABSTRACT

In this study, morphological and anatomical characteristics of *Hieracium lasiochaetum* (Bornm. & Zahn) Sell & West, an endemic species, were investigated. Morphological features like leaf shape, stylus colour and indumentum were found to be as taxonomically important in this species. In anatomical investigations, cross-sections of the root, stem, leaf and petiole and the surface sections of leaves were examined. It has been observed that stem and petiole have a distinct collenchyma layer. The leaf is dorsiventral (bifacial) and hypostomatic. Stoma cells are anomocytic. Additionally, seeds and trichomes of the species were examined by scanning electron microscope (SEM). The primary sculpture of the achenes appears to be reticulate type. Trichomes type are simple or stellate eglandular and glandular.

Key Words: Anatomy, morphology, Hieracium lasiochaetum, scanning electron microscopy (SEM).

INTRODUCTION

The genus *Hieracium* L.(Asteraceae) is a large genus well known for its apomictic members and its taxonomic complexity [1]. Nearly 10.000 mostly apomictic species of *Hieracium* have been described up to now [2]. According to the monography by Zahn [3,4], the genus *Hieracium* is divided into four subgenera. Two of them, *Hieracium* and *Pilosella* L., are reported in Flora of Turkey [5]. Sell & West [6] recognized these two subgenera as a distinct genus. The genus *Hieracium* is represented by 113 species and 68 of them are endemic in Turkey [2, 7-10]. Further, *Hieracium lasiochaetum* (Bornm.& Zahn) Sell & West is an endemic taxon to Turkey [7].

Some species of *Hieracium* have antibiotic properties and are used against nephritis, gout, dyspna, cardiac oedema and diarrhoea [11,12]. In addition, *Hieracium* species are an important source of nectar for honeybees [9].

In recent years, the genus *Hieracium* has been the subject of chemical, ecological, taxonomical, genetical, karyological and molecular studies [11, 13-18]. However, there are limited information about anatomical studies on *Hieracium* species. Metcalfe & Chalk [19] reported that the anatomical diversity was commonly observed in the structure of leaves in species belonging to the Asteraceae. Milan et al. [20] investigated leaf morphology and anatomy of some Asteraceae species. They determined that the secretory structures and various types of glandular or covering trichomes are of great taxonomical interest in the Asteraceae family.

Achenes features were also widely used for the classification of Asteraceae [21-23]. Zhu et al [4] reported that the fruit surface sculpturing may provide useful data for the classification of this family. However, the anatomical and micromorphological properties of *H. lasiochaetum*, which is endemic taxon to Turkey, have not been studied. Therefore, the aim of this study was to investigate the micromorphological and anatomical properties of *H. lasiochaetum*.

MATERIALS AND METHODS

Plant samples were collected from natural populations in Kocadağ (Samsun) between May-July 2007. The specimens were dried according to standard herbarium techniques and stored at the Ondokuz Mayıs University Herbarium (OMUB). Taxonomical identification of the plants were done according to Flora of Turkey [5]. Fresh samples were used for morphological measurements. Samples were collected from the following locations:

A5 Samsun: Kocadağ, northern slopes, shrubs, 980 m, 05.vi.2007, Akçin 1283; A5 Samsun: Kocadağ, northeast, woodland, 878 m, 29.v.2007, Akçin 1295.

For anatomical analysis, some samples were fixed within 70 % alcohol. Anatomical investigations were performed on the cross-section of root, stem, petiole and leaves and the surface sections of leaves taken manually. The materials were fixed with glycerine-gelatine [24]. Anatomic structures were measured using a ocular micrometer determining maximum and minimum boundaries. All measurements were determined on at least 30 specimens. The microscopical photographs of the samples were taken with an Nikon Coolpix P4 digital camera.

For scanning electron microscopy, dried fruit and leaf samples were directly mounted on stubs using double-sided adhesive tape and sputter-coated with gold. The prepared specimens were examined and photographed on a JEOL-JSM 6400 Scanning Electron Microscope. Seed morphology of the plant was assessed according to Barhlott [25,26] and Garg & Sharma [27].

RESULTS

Morphological Properties (Figure 1)

Stem was 15-45 (-50) cm tall, with numerous long, simple eglandular hairs and stellata and short glandular hairs above. Basal leaves 30-85 (-100)x 10-35 mm, mostly elliptic, sometimes lanceolate or ovate, acute to obtuse, usually denticulate, sometimes with teeth in lower half, cuneate at base and with petiole. Cauline leaves 0-2, usually linear-lanceolate. Capitula 2-6. Peduncles up to 45(-50) mm, with numerous stellata and glandular and long simple eglandular hairs. Involucre 9-13(-14) mm, phyllaries linear-lanceolate, acute or obtuse, with stellata and numerous shorter glandular hairs and simple eglandular hairs. Ligules were glabrous. Stylus was yellow. Pappus biseriate, pappus hairs rigid, dirty white.



Figure 1. *H. lasiochaetum.* A. general appearance, B. capitula, C. flower, D. involucre, E. achene.

Achene Morphology (Figure 2, 3)

Achenes are ovoid or obovoid form and dark brown having axial length and breadth of 3-5 mm. Primary sculpture appears to be reticulate type having more or less rectangular cells. Epidermal cells of the species examined are longitudinally elongated. The anticlinal walls of epidermal cells are more or less straight and raised.





Figure 2-3. Achene morphologies of *H. lasiochaetum.* 2. Pappus and general view of achene, 3. Achene surface.



Figure 4. Cross-section of the root. p) Pith, per) Periderm, pr) Parenchyma, sc) Secretory canals, x) Xylem.

Root (Figure 4)

A transverse section taken from the root was observed as follows. Root is perennial and shows secondary growth. Periderm is 3-6 layered. Cortex is multilayered and parenchymatic. Parenchymatic cells are $37.0-62.5x \ 27.5-42.5 \ \mu m$ (Table 1). Secretory reservoirs are present within cortex cells. The cells of endodermis and pericyl are not distinguishable. Cambium cells are 2-3 layered and consists of rectangular cells. Vascular bumdles are collateral type. Rays are distinguishable. The pith containing parenchymatic cells was filled root centre.

		Breadth		Length (µm)		
		(µm)				
		Min.	Max.	Min.	Max	
ROOT	Periderma cells	20.0	25.0	42.5	50.0	
	Parenchyma cells	37.0	62.5	27.5	42.5	
	Diameter of trachea	12.5	27.5	-	-	
STEM	Epiderma cells	12.5	17.5	10.0	12.5	
	Collenchyma cells	10.0	12.5	10.0	20.0	
	Parenchyma cells	17.5	25.0	22.5	30.0	
	Endoderma cells	12.5	17.5	12.5	20.0	
	Diameter of trachea	15.0	20.0	-	-	
	Diameter of pith cells	40.0	120.0	-	-	
LEAF	Upper epidermis cells	22.5	50.0	20.0	32.5	
	Palisade parenchyma	22.5	27.5	42.5	55.0	
	cells					
	Spongy parenchyma	30.0	42.5	32.5	62.0	
	cells					
	Diameter of trachea	10.0	17.5	-	-	
	Lower epidermis cells	20.0	37.5	17.5	30.0	
	Stomata cells	25.0	30.0	37.5	47.5	
PETIOLE	Abaxial cells	12.5	17.5	15.0	22.5	
	Cortex cells	27.5	97.5	25.0	70.0	
	Collenchyma cells	12.5	25.0	20.0	35.0	
	Diameter of trachea	12.5	20.0	-	-	
	Adaxial cells	12.5	20.0	12.5	22.5	

Stem (Figure 5-9)

A transverse section taken from the middle part of the stem was observed (Figure 5,6). The epidermis is composed of a single layer of almost rectangular cells. There are glandular or abundant simple and stellata eglandular trichomes on the epidermis (Figure 7-9). Eglandular trichomes are generally with cuticular micropapillae (Figure 8). Glandular hairs have a short unicellular stalk and head (Figure 9). The lamellar collenchyma tissues consisting of 4-5 layers of rectangular cells and are located under the epidermis. Parenchyma cells are 2-3 layered and are 17.5-25.0x22.5-30.0 µm (Table 1). Endoderma is distinguishable. Collateral vascular bundles are arranged in a more or less regular ring. Phloem and xylem members are clear. Cambium is distinguishable and 1-2 layered. The pith consists of large hexagonal or polygonal parenchymatic cells.

 Table
 1. Anatomical measurements of H.



Figure 5-6. Cross-section of the stem. c) Collenchyma, e) Epiderma, p) Pith, ph) Phloem, pr) Parenchyma, x) Xylem, vb) Vascular bundles.





Figure 7-10. Hair types of the *H. lasiochaetum.* 7. Stellata hairs of the stem, 8. Eglandular hairs of stem, 9. Eglandular and glandular hairs of stem, 10. Scanning electron photograph of eglandular hairs of leaf. gh) Glandular hairs, egh) Eglandular hairs.

Leaf (Figure 10-13)

In transversal section, leaves are covered by a thick cuticular layer on both surfaces (Figure 11, 12). The upper and lower epidermis consists of a single row rectangular cells. There were long and simple eglandular hairs on both epidermis. Eglandular trichomes with cuticular micropapillae, unbranched and consist of elongated cells (Figure 10). Leaf is bifacial and hypostomatic. Palisade parenchyma cells are usually one or sometimes two layered. There are 2-3 layers of isodiametric spongy parenchymatic cells (Figure 11). The midrib region is well developed and forms projecting parts towards the outside. The xylem towards the upper surface and the phyloem towards the lower surface. Vascular bundles are surrounded by a parenchymatic bundle sheath (Figure 12). The anomocytic type stomata is present on the lower epidermis (Figure 13). Measurements about leaf anatomy are presented in Table 1.





Figure 13. Surface section of the leaf. st) Stomata

Petiole (Figure 14)

Petiole is covered by ovoidal epidermal cells. There are many long and simple hairs on epidermis (Figure 14). The collenchyma is located under the epidermis. Collenchyma consists of 3-4 layered cells at the corners of petiole, whereas it is 1-2 layered at the other parts. Parenchymatic cortex is present under collenchyma. Cortex cells are 27.5-97.5x25.0-70.0 µm (Table 1). The air cavity is formed on the adaxial side. There is a large vascular bundle in the median region of the petiole. Apart from the large central bundle, the petiole contains two lateral bundles of similar sizes, four small bundles half as large as the latter and more than ten small bundles. Vascular bundles are collateral and covered by parenchymatic bundle sheath. The xylem is located towards upper side of petiole and the phloem towards lower side of petiole. Cambium is distinguishable.



Figure 11-12. Cross-section of the leaf. c) Collenchyma, le) Lower epidermis, ph) Phloem, pr) Parencyhma, pp) Palisade parenchyma, sp) Spongy parenchyma, ue) Upper epidermis, x) Xylem.



Figure 14. Cross-section of the petiole. ab)Abaxial epidermis, ac) Air cavity, ad) Adaxial epidermis, vb) Vascular bundles.

DISCUSSION

In this study, the morphological and anatomical characteristics of *H. lasiochaetum*, which is a endemic taxon were examined. The results obtained from morphological studies showed many similarities compared with the Flora of Turkey [5]. However, some morphological characters of this taxon were not given in the Flora of Turkey, such as achene colour, achene micromorphology and details of the pappus hairs. These characters were obtained in this study. In additionally, morphological characters such as indumentum, leaf shape and stylus colour were used as taxonomic characters in determining the species. However, our studies indicated that indumentum characters was effected by different ecological factors.

Micromorphological studies on fruit and seed structure plays an important role in systematics of Asteraceae family [27,28]. Bremer [29] reported that the micromorphology of fruit can be used for characterization and its characters are valuable specially at generic and specific level. It was also determined that the fruit micromorphology of some *Hieracium* species are used in the delimination of species [27]. Our studies indicated that the primary sculpture of *H. lasiochaetum* achenes was reticulate. The results obtained from morphological studies were generally similar with the results of Garg & Sharma [27].

Metcalfe & Chalk [19] explained the general anatomical characteristics of the family Asteraceae in their article. However, the anatomical properties given in this study provide the first detailed description of the endemic species *H. lasiochaetum.* In this research, it was found that this species had a secondary root structure.

Metcalfe & Chalk [19], Heywood [30] and Bremer [31] have described the presence of secretory structures in especially in roots of Asteraceae. It was also determined that *Mikania glomerata* Spreng (Asteraceae) had secretory ducts and *Porophylum ruderale* (Cass) (Asteraceae) had secretory cavities [20]. Castro et al. [32] used the secretory structures as parameters for identification of genera of Asteraceae. In our study, we determined that *H. lasiochaetum* has secretory canals within root cortex parenchyma tissue. Although some recent studies on Asteraceae reported the existence of endodermis in the root [33-35], the endodermis have not been distinguishable in roots of *H. lasiochaetum*.

Observations of the stem transverse section showed that stem has a distinct collenchyma layer. Distinguishable endodermis was present between cortex and vascular tissue. Özörgücü et al. [36] reported that endodermis is usually distinguishable in stem of Asteraceae. Vascular bundles were characterized by into more or less regular ring in stem. The pith cells were large and parenchymatic.

This species has dorsiventral (bifacial) leaves. However, the mesophyll of the studied species varied according to the number of cell layers composing the palisade and spongy parenchyma. Metcalfe & Chalk [19] reported that there were generally bifasial mesophyll in the family Asteraceae and the anatomical diversity was commonly observed in the structure of leaves of species belonging to the Asteraceae. Sajo & Menezes [37] observed that the Asteraceae species *Vernonia sessilifolia* Less and *V. linearis* Spreng had dorsiventral mesophyll, whereas *V. psilophylla* D.C. had isobilateral mesophyll. Our results basically agree with the characteristics pointed out by Metcalfe & Chalk [19]. In this research, it was found that *H. lasiochaetum* had anomocytic stomata. Stomata cells occurred on the lower epidermis. Metcalfe & Chalk [19] reported that there were both anomocytic and anisocytic stomata in the family Asteraceae.

Transversal sections of the petiole showed that the vascular units of *H. lasiochaetum* were surrounded by orbicular parenchymatous cells (endodermis). Melo-de Pinna & Menezes [35] observed that the endodermis clearly involves the vascular unit as an sheath at the central vascular unit region of petiole of *Ianthopappus corymbosus* Roque & Hind (Asteraceae). Lotocka & Geszprych [38] determined that the very large air cavity was formed on the adaxial side of petiole in *Rhaponticum carthomoides* (Wild) Iljin (Asteraceae). In this study, the air cavity was formed on the adaxial side of the petiole of *H. lasiochaetum*. Our results are consistent with the results of Lotocka & Geszprych [38].

The results of this study exhibited that *H. lasiochaetum* has some characteristic morphological and anatomical features. These results could be provide significant data in solving some taxonomical problems of *Hieracium* species.

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