

## PETIOLE ANATOMY OF SOME LAMIACEAE TAXA

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### Abstract

In this study, anatomical structures of the petiole of 7 taxa viz., *Glechoma hederacea* L., *Origanum vulgare* L., *Scutellaria salviifolia* Benth., *Ajuga reptans* L., *Prunella vulgaris* L., *Lamium purpureum* L. var. *purpureum*, *Salvia verbenaca* L., *Salvia viridis* L., *Salvia virgata* Jacq., belonging to the *Lamiaceae* family were examined and compared. In all the studied taxa, some differences were found in the petiole shape, arrangement and number of vascular bundles, hair types and the presence of collenchyma. *G. hederacea*, *S. virgata* and *O. vulgare* consist of a total of 3 vascular bundles, with a big bundle in the middle of the petiole and a single small vascular bundle in each corner. *P. vulgaris* has 5 vascular bundles. *S. verbenaca* has a total of 11 vascular bundles, with a big bundle positioned in the middle. *L. purpureum* L. var. *purpureum* consists of 4 vascular bundles. *S. salviifolia* has 3 vascular bundles. *A. reptans* has a total of 9 vascular bundles, with 1 big bundle in the middle. *S. viridis* consists of 7 vascular bundles. Petiole has glandular and eglandular hairs. Eglandular hairs consist of capitate hairs, whereas peltate hairs are only found in *S. salviifolia*.

### Introduction

The Lamiaceae is a large family. Most of the species have great importance due to their economic values. Lamiaceae is represented by about 258 genera and 3500 species in the world (Duarte & Lopes, 2007). Turkey is accepted as a gene center for this family (Başer, 1993). With their pleasant fragrance, many species of Lamiaceae have been used as herbal teas in Turkey. Many of species are used as raw material in cosmetic industry. Some species are traditionally used as medicinal plants (Baytop, 1984). Some *Ajuga* L., and *Salvia* L., species are cultivated as ornamental plants (Baytop, 1984; Özdemir & Şenel, 2001; Akçin *et al.*, 2006).

Rich chemical contents of the Lamiaceae species have been investigated by many researchers (Werker, 2006; Kaya & Kutluk, 2007). The morphology and anatomy of some species have been studied (Tahir *et al.*, 1995; Özdemir & Şenel, 2001; Baran & Özdemir, 2006). The most important features of Lamiaceae taxa are glandular hairs distributed in vegetative and reproductive organs (Werker, 2006). These hairs are source of etheric oils and their structures have been examined anatomically and micromorphologically (Hanlidou *et al.*, 1991; Vrachnakis, 2003; Kaya *et al.*, 2007). In recent years, anatomical characters have been used in taxonomy (Agbagwa & Ndukwu, 2004; Kharazian, 2007). The structure of petiole shows differences between genera and species. Thus, useful petiole anatomic characters are determined in designated taxonomical structures of some species (Olowokudejo, 1987; Shaheen, 2007; Eric *et al.*, 2007). Anatomical structures of the petiole are very important in family Lamiaceae (Metcalf & Chalk, 1972). The main object of this study was to investigate the anatomical structures and hair micromorphologies of petioles of 7 Lamiaceae taxa.

### Materials and Methods

Plant materials were collected from different locations in Turkey (Table 1). Samples were fixed in 70% alcohol for anatomical studies. All measurements were realized with an ocular-micrometer on a light microscope (Tables 2, 3). Transverse section preparations of petioles were prepared manually. The photographs of petioles and hairs were taken with a Nikon FDX-35 microscope.

For scanning electron microscopy, dried petioles were mounted on stubs using double-sided adhesive tape. Samples

were coated with 12.5- 15 nm of gold. Coated leaves were examined and photographed with a JMS-6400 Scanning Electron Microscope.

### Result and Discussion

***Glechoma hederacea* L.:** Petiole is sulcate with obtuse margins (Fig. 1A). In transverse section, the adaxial and abaxial epidermises of the petiole consist of single layer cell of rectangular and oval cells. Epidermis cells are  $17.75 \pm 3.42 \times 14.75 \pm 2.48 \mu$ . Both epidermises are covered with a smooth cuticle. Collenchyma is 2-3 layers under both epidermises in the middle part of the petiole. Multilayered collenchyma is present at the margins. Chlorenchyma cells are especially seen at the abaxial side. Vascular bundles are located in the parenchyma tissue and this tissue occupies large parts of the petiole. Petiole consists of a total of 3 vascular bundles, with a big bundle in the middle of the petiole and a single small vascular bundle in each corner. The big bundle is arc-shaped. Vascular bundles are surrounded by bundle sheath cells. Median vascular bundle is surrounded by parenchyma cells (Table 2, Fig. 2).

Glandular and eglandular hairs are evident on both epidermises. Eglandular hairs are multicellular and long or unicellular. Glandular hairs are of capitate types. Capitate hairs consist of 1-2 stalk cells and 1-3 head cells (Table 3, Figs. 11, 12, 26 A).

***Origanum vulgare* L.:** Petiole shape is broadly sulcate with obtuse margins (Fig. 1B). The adaxial and abaxial epidermises of the petiole consist of single layer cell of oval, rectangular and  $16.75 \pm 2.09 \times 12.5 \pm 3.34 \mu$  cells. The adaxial and abaxial epidermises are covered with an undulate cuticle layer. Collenchyma is 2 layered under the adaxial epidermis, 3-4 layered under the abaxial epidermis and 3 layered at the corners. The median vascular bundle is multi-lobed and broadly arc-shaped. There is a single small vascular bundle in each corner. Four layered parenchyma cells with chloroplasts are located between the collenchyma layers and the vascular bundles at the corners. Vascular bundles are surrounded by a bundle of sheathed cells (Table 2, Fig. 3).

The adaxial side in this species is larger than the others. Hair types are glandular and eglandular. Eglandular hairs are multicellular (5- 9) and long hairs. The glandular hairs include capitates types. Capitate hairs have 1-2 stalk cells and 1-2 head cells (Table 3, Figs. 13, 14, 26 B).

**Table 1. Localities of studies Lamiaceae taxa.**

Taxa	Locality
<i>Glechoma hederacea</i> L.	A6 Ordu: Boztepe, road side, 450 m.
<i>Origanum vulgare</i>	A6 Ordu: Boztepe, road side, 400 m.
<i>Scutellaria, salviifolia</i>	A5 Amasya: Karaman Mountain, 550 m.
<i>Ajuga reptans</i>	A6 Ordu: Aybastı, Perşembe Plateau, road side, 1500 m.
<i>Prunella vulgaris</i>	A6 Samsun: Kurupelit, 150 m.
<i>Lamium purpureum</i> L. var. <i>purpureum</i>	A6 Samsun: Kurupelit, 100 m.
<i>Salvia verbenaca</i>	A6 Ordu: Perşembe, 15 m.
<i>Salvia viridis</i>	A6 Samsun: Bafra, vicinity of Derbent Dam, 350 m.
<i>Salvia virgata</i>	A6 Samsun: Bafra, Gelemen Farm, 100m.

**Table 2. Anatomical characteristics of studied Lamiaceae taxa.**

Taxa	Petiole shape	Cuticle structure	Number of collenchyma layer			Chlorenchyma	Number of vascular bundle		Vascular bundle shape
			Ab.	Ad.	Corner		Middle	Corner (pair)	
<i>Glechoma hederaceae</i>	Sulcate with obtuse margins	Smooth	2 (3)	2 (3)	4-5	+	1	1	Arc-shaped
<i>Origanum vulgare</i>	Broadly sulcate with obtuse margins	Undulate	3-4	2	3	+	1	1	Broadly arc-shaped, multi-lobed
<i>Scutellaria salviifolia</i>	Broadly sulcate with flat adaxially	Undulate	1	1	2	+	1	1	Arc-shaped
<i>Ajuga reptans</i>	Narrowly sulcate with long and acute margins	Smooth	1	1	1-2	+	1	4	Big arc-shaped
<i>Prunella vulgaris</i>	Narrowly and acutely sulcate	Smooth	2-3	1	5-6	-	1	2	Big vascular bundle, 2-lobed
<i>Lamium purpureum</i> var. <i>purpureum</i>	Broadly sulcate with obtuse margins	Smooth	1		2-3	+	2	1	Spherical
<i>Salvia verbeneca</i>	Sulcate with acute margins	Undulate	4-5	1-2	6	+	1	5	Broadly big arc-shaped, multi-lobed (10-13)
<i>Salvia viridis</i>	Flat adaxially with erect margins	Undulate	2	1	1-2	+	1	3	Big arc-shaped, 2 lobed
<i>Salvia virgata</i>	Flat and obtusely adaxially sulcate	Undulate	3	1	5-7	+	1	1	Big vascular bundle, 4-lobed

**Table 3. Hair characteristics of studied Lamiaceae taxa.**

Taxa	Glandular hairs			Eglandular hairs	
	Capitate		Peltate	Unicellular	Multicellular
	Head cell	Stalk cell			
<i>Glechoma hederaceae</i>	1-3	1-2	-	+	++
<i>Origanum vulgare</i>	1-2	1-2	-	+	++
<i>Scutellaria salviifolia</i>	1-2	1-2	+	+	++
<i>Ajuga reptans</i>	1	1	-	-	+
<i>Prunella vulgaris</i>	1-2	1	-	-	++
<i>Lamium purpureum</i> var. <i>purpureum</i>	1-3	1	-	-	++
<i>Salvia verbeneca</i>	1-2	1-6	-	+	++
<i>Salvia viridis</i>	1-2	1	-	+	++
<i>Salvia virgata</i>	1-2	1	-	+	++

++ = Dense, + = Few, - = Absent

***Scutellaria salviifolia* Benth:** Petiole is broadly sulcate with flat adaxially. Margins are obtuse (Fig. 1C). Both epidermises cells are single layered, small, rectangular and  $15.55 \pm 1.45 \times 12.8 \pm 2.44 \mu$  cells. Epidermis cells are covered with an undulate cuticle layer. Collenchyma is single layered under both the epidermises and 2 layered at the corners. Chlorenchymatic cells are seen at the corners of petiole. Petiole has a single bundle in the middle and a small single bundle in each corner, a total of 3 vascular bundles. The median bundle is arc-shaped and surrounded by parenchyma cells at the abaxial side (Table 2, Fig. 4).

Glandular and eglandular hairs are evident on both the epidermises. Eglandular hairs are multicellular long hairs or rarely unicellular. The glandular hairs include peltate and

capitate types. Capitate hairs have unicellular or multicellular head cells (Table 3, Figs. 15, 26 C).

***Ajuga reptans* L.:** Petiole shape of this species is narrowly sulcate with long and acute margins (Fig. 1D). Both epidermises cell are single layered, oval or rectangular. Epidermis is covered with a smooth cuticle. Collenchyma is single layered under both epidermises and 1-2 layered at the corners. Narrow and long margins of petiole are filled with chlorenchyma cells (1-2 layered). *A. reptans* has a total of 9 vascular bundles, with a big arc-shape bundle in the middle. In each corner, there are 2 small and 2 big vascular bundles. Both big and small vascular bundles are surrounded by bundle sheath cells. Median vascular bundle has scleranchymatic cells on the phloem (Table 2, Fig. 5).

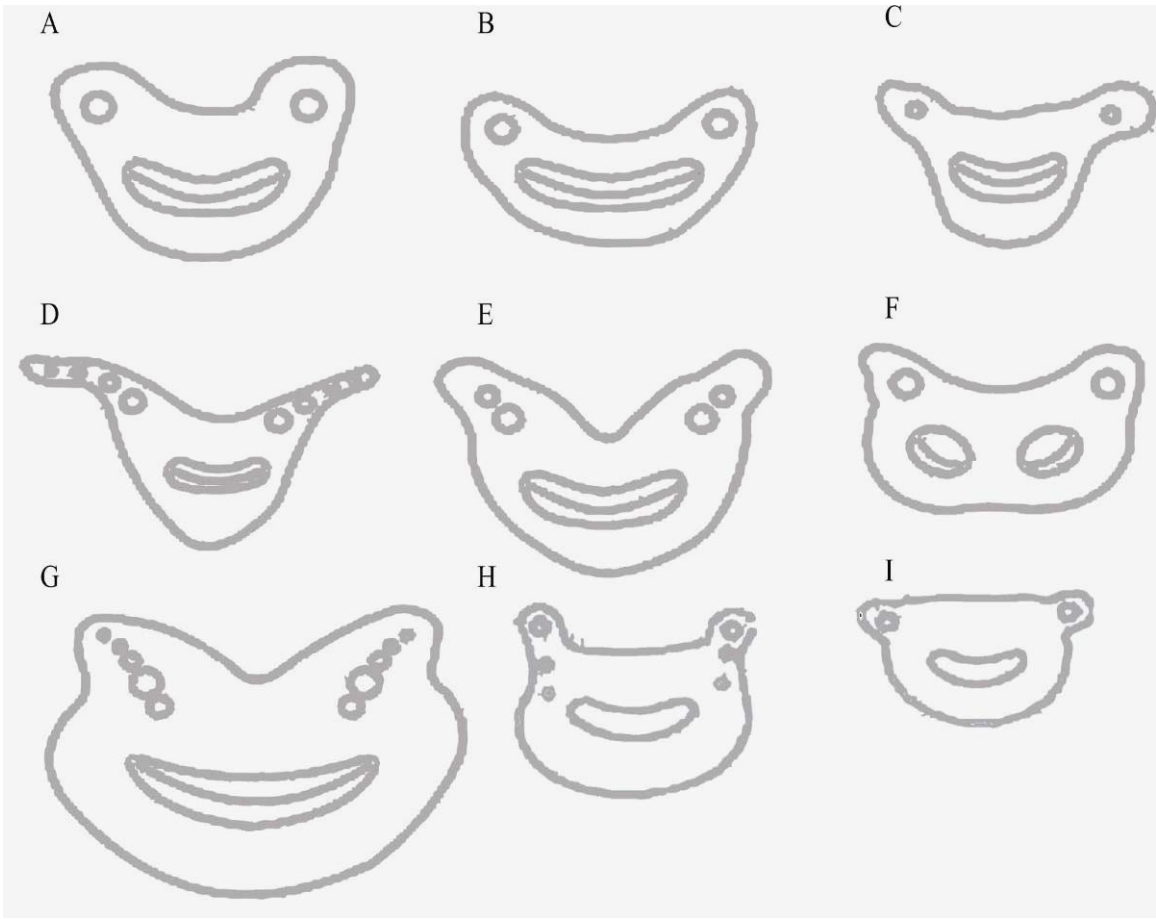
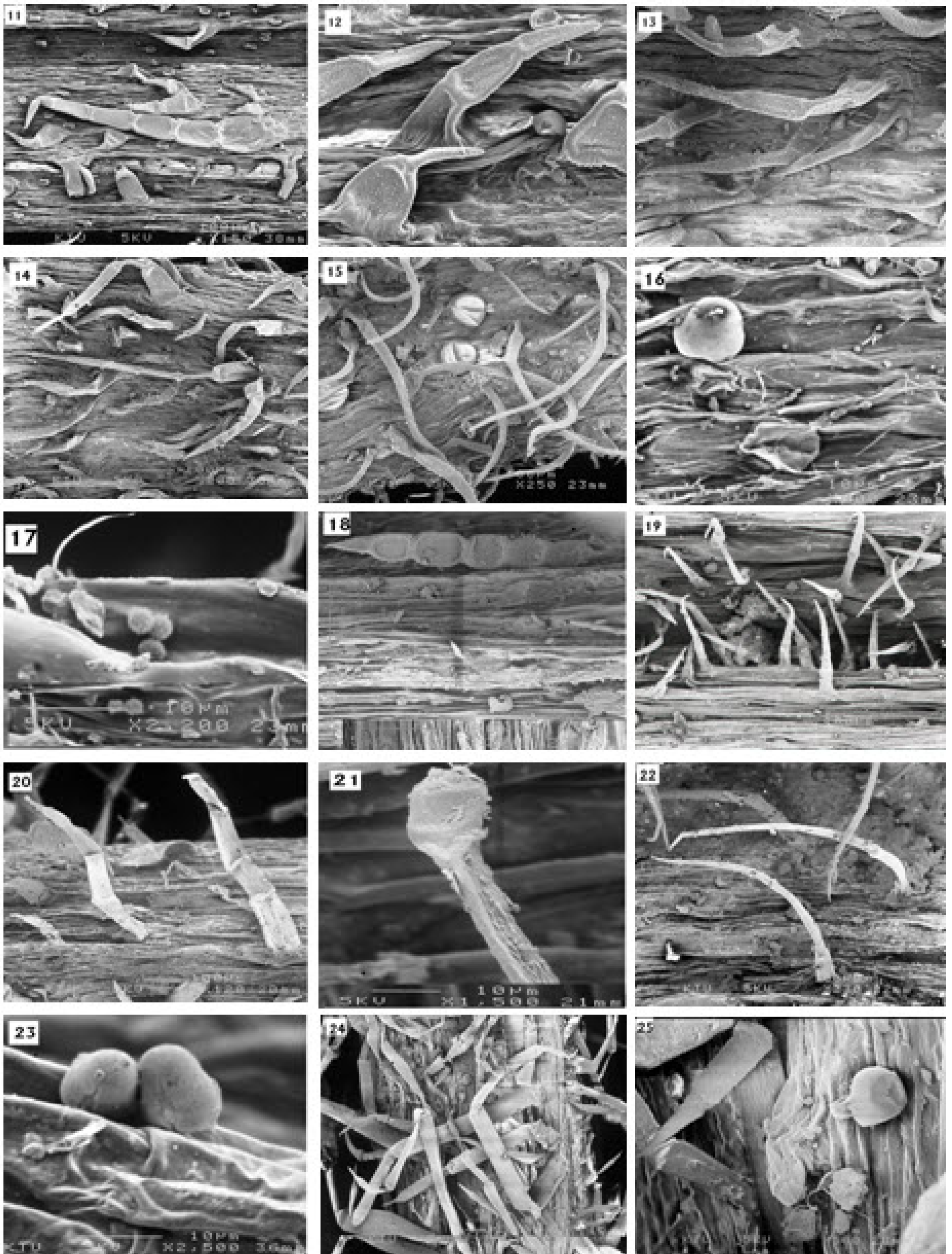


Fig. 1. Petiole shapes of studied taxa. A- *Glechoma hederaceae*, B- *Origanum vulgare*, C- *Scutellaria salviifolia*, D- *Ajuga reptans*, E- *Prunella vulgaris*, F- *Lamium purpureum* var. *purpureum*, G- *Salvia verbenaca*, H- *S. viridis*, I- *S. virgata*.



Figs. 2-10. Transverse section of petioles of studied taxa. Fig. 2. *G. Hederaceae*. Fig. 3. *O. vulgare*. Fig. 4. *Scutellaria salviifolia*. Fig. 5. *A. reptans*. Fig. 6. *P. vulgaris*. Fig. 7. *L. purpureum* var. *purpureum*. Fig. 8. *Salvia verbenaca*. Fig. 9. *S. viridis*. Fig. 10. *S. virgata*.



Figs. 11-25. Scanning electron micrographs of petiole hairs of studied taxa. Figs. 11, 12. *G. hederaceae*. Figs. 13, 14. *O. vulgare*. Fig. 15. *Scutellaria salviifolia*. Figs. 16, 17. *A. reptans*. Fig. 18. *P. vulgaris*. Fig. 19. *L. purpureum* var. *purpureum*. Figs. 20-21. *Salvia verbenaca*. Figs. 22, 23. *S. viridis*. Figs. 24, 25. *S. virgata*.

There are few glandular and eglandular hairs on the petiole. Eglandular hairs are long. Glandular hairs are capitated types (Table 3, Figs. 16, 17, 26 D).

***Prunella vulgaris* L.:** Petiole is narrowly and acutely sulcate (Fig. 1E). Epidermis cells are single layered and are arranged regularly in both sides. Epidermis is covered with a smooth cuticle. Collenchyma is 2-3 layered in abaxial side, single layered in adaxial side and 5-6 layered at the corners of the petiole. Collenchyma cells are  $23.5 \pm 4.04 \times 20.25 \pm 3.17 \mu$ . Parenchyma cells cover a large area. *P. vulgaris* consists of a big vascular bundle with 2-lobed in the middle of the petiole. In each corner it has a small and a big bundle, a total of 5 vascular bundles. Vascular bundle is collateral type. Median vascular bundle is surrounded by parenchyma cells (Table 2, Fig. 6).

Hair types are glandular and eglandular. Eglandular hairs are multicellular (9-10) and long. The glandular hairs are capitate types and have 1-2 head cells (Table 3, Figs. 18, 26 E).

***Lamium purpureum* L. var. *purpureum*:** Petiole is broadly sulcate with obtuse margins. Abaxial side is flat (Fig. 1F). Adaxial and abaxial epidermises are single layered. Both epidermises are covered with smooth cuticle. Petiole has single layered collenchyma in abaxial side, 2-3 layered collenchymas at the corners. There are 3-4 layered chlorenchyma cells at the corners, 1-2 layered chlorenchyma cells at the margins. Petiole of this species consists of 2 big vascular bundles in the middle and a small single bundle in each corner. Middle and small vascular bundles are surrounded by parenchyma cells (Table 2, Fig. 7).

Eglandular hairs are unicellular or multicellular (2-3). Glandular hairs are capitate types. Capitate hairs have 1 stalk cell and 1-3 head cells (Table 3, Fig. 19, 26 F).

***Salvia verbenaca* L.:** Petiole is sulcate with acute margins (Fig. 1G). Both epidermises cells are single layered, small and rectangular shaped. Epidermis cells  $19.75 \pm 3.4 \times 17.45 \pm 2.10 \mu$ m. Both adaxial and abaxial epidermises are covered with a cuticle layer with undulate. Collenchyma is 1-2 layered in adaxial side, 4-5 layered in abaxial side and 6 layered at the corners. Multi-lobed (10-13) and broadly arc-shaped big vascular bundle is seen in the median part of petiole. The lobes are arranged separately. In each corner there are 3 big and 2 small bundles. Bundle sheath is clear at big bundles. Multilayered sclerenchyma are seen on the phloem in median vascular bundles (Table 2, Fig. 8).

There are dense multicellular (1-5) long, short eglandular hairs and capitate glandular hairs. Capitate hairs have 1 -6 stalk hair and 1-2 head cells (Table 3, Figs. 20, 21, 26 G).

***Salvia viridis*:** Petiole is flat adaxially with erect margins (Fig. 1H). Adaxial and abaxial epidermises are single layered, rectangular or oval shaped. Both epidermises are covered with undulate cuticle layer. Collenchyma is 1 layered in adaxial side, 2 layered in abaxial side and 1-2 layered at the corners. *S. viridis* consists of a big arc-shaped vascular bundle with 2-lobed in the middle of the petiole. In each corner there are 1 big and 2 small bundles. Bundle sheath is clear at big bundles. Corners and side of the petiole are filled with chlorenchymatic cells (Table 2, Fig. 9).

Hair types are glandular and eglandular. Eglandular hairs are multicellular (1-5) and long or short. The glandular hairs include capitate types. Capitate hairs have 1 stalk cells and 1-2 head cells (Table 3, Figs. 22, 23, 26 H).

***Salvia virgata*:** Petiole shape of this species is flat and obtusely adaxially sulcate (Fig. 1I). Both epidermises cells are single layered, small and rectangular or oval shaped. There is undulate cuticle on both abaxial and adaxial epidermises. Collenchyma is single layered in adaxial side, 3 layered in abaxial side and 5-7 layered at the corners of the petiole. Multi-lobed (4), big vascular bundle is seen in the median part of the petiole. The lobes are arranged separately. There is oval shaped vascular bundle in each corner. There are chlorenchymatic cells around the vascular bundles in the corner. All vascular bundle are surrounded with bundle sheath (Table 2, Fig. 10).

Glandular and eglandular hairs are evident on both epidermises. Eglandular hairs are multicellular (1-4), long or short hairs. The glandular hairs are capitate types. Capitate hairs have unicellular or multicellular head cells (Table 3, Figs. 24, 25, 26 I).

In this study, anatomical structures of the petiole of 7 taxa belonging to the *Lamiaceae* family were examined and compared. All the studied taxa were found to have differences in the arrangement and a number of vascular bundles in the petioles, the hair types, the petiole shapes, presence of collenchyma and the structure of the epidermis.

*G. hederaceae* consists of a total of 3 vascular bundles, with a big bundle in the middle of the petiole and a single small vascular bundle in each corner. *O. vulgare* also has 3 vascular bundles in total, with a big bundle in the middle and a single small vascular bundle in each corner. *P. vulgaris* consists of a big vascular bundle in the middle of the petiole. In each corner it has a small and a big bundle, a total of 5 vascular bundles. *S. verbenaca* has a total of 11 vascular bundles, with a big bundle positioned in the middle. In each corner there are 3 big and 2 small bundles. *S. virgata* consists of a big bundle in the middle part of the petiole and a small bundle in each corner, a total of 3 vascular bundles. *S. viridis* has a total of 4 vascular bundles. There are 1 big bundle in the middle and 1 big and 2 small bundles in each corner of petiole. *L. purpureum* L. var. *purpureum* consists of 2 big vascular bundles in the middle and a small single bundle in each corner. *Scutellaria salviifolia* has a single bundle in the middle and a small single bundle in each corner, a total of 3 vascular bundles. *Ajuga reptans* has a total of 9 vascular bundles, with 1 big bundle in the middle. In each corner, there are 2 small and 2 big vascular bundles. The results showed us that within the studied taxa, there were many differences between the amount and arrangement of vascular bundles, as well as their shapes. Ozdemir & Senel (1999, 2001) showed the importance of the amount of vascular bundles and its arrangement within the petiole in the *Salvia* species. Maksymowych *et al.*, (1983) examined petiole anatomy of 26 herbaceous and ligneous taxa and showed that the vascular bundles were positioned differently in each and every different type.

Many scientists have also examined other families petiole vascular bundles. Heneidak *et al.*, (2007) examined 15 different types of the *Fabaceae* family and showed the importance of the shape of the petioles, the features of the epidermal cells, fibres, crystal types, secretion elements, hairs and the anatomy of the vascular bundles. Other scientists have studied with the *Rubiaceae* family and have examined the anatomy of the petioles, the structure of the vascular bundles and the hair types and showed the importance it has in terms of taxonomic classification (Kocsis *et al.*, 2004).

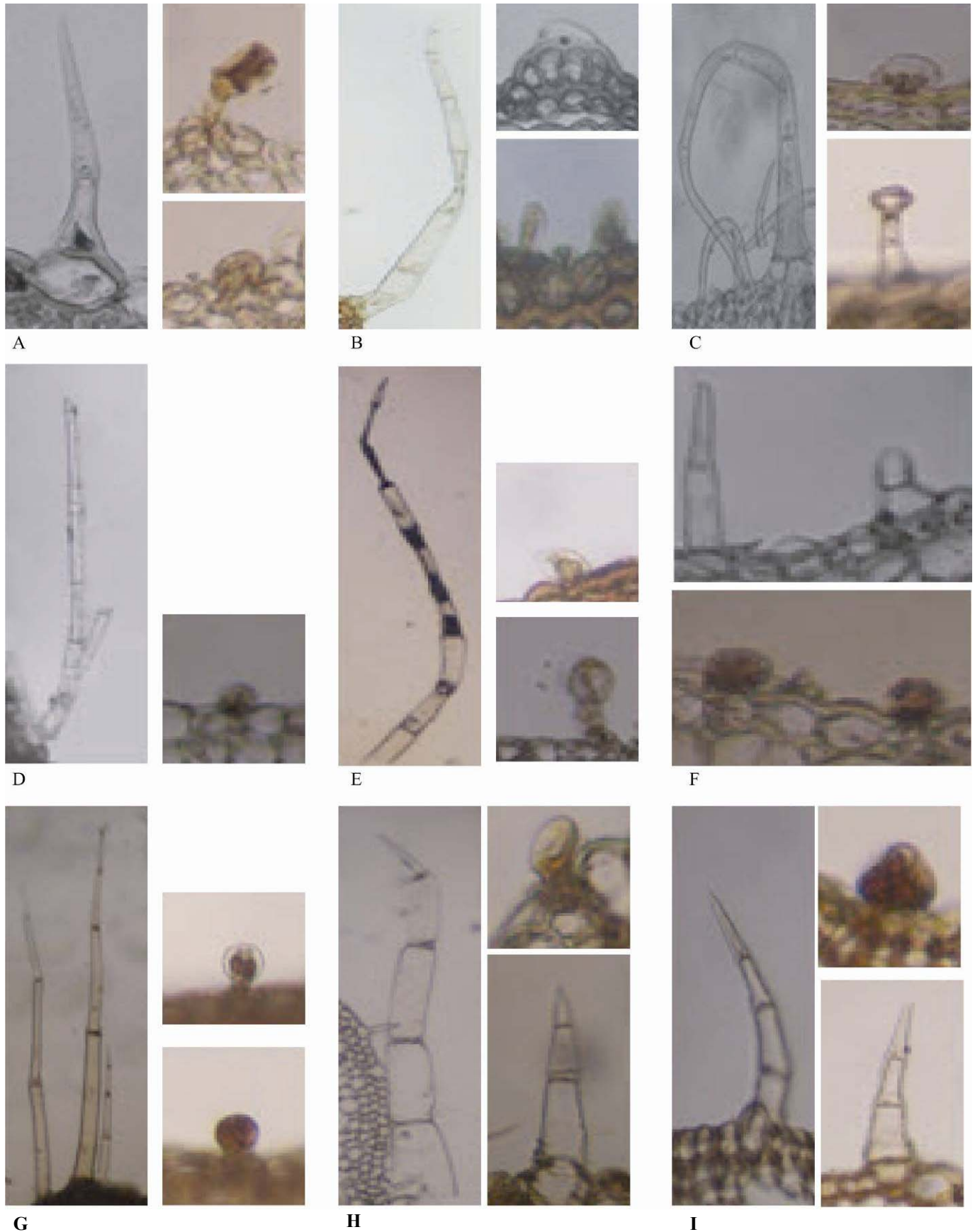


Fig. 26. Glandular and eglandular hairs of petiole of studied taxa. A- *G. hederaceae*, B- *O. vulgare*, C- *Scutellaria salviifolia*, D- *A. reptans*, E- *P. vulgaris*, F- *L. purpureum* var. *purpureum*, G- *Salvia verbenaca*. H- *S. viridis*. I- *S. virgata*.

Petiole shapes showed distinct differences among the examined types. The petioles of *G. hederaceae* look like sulcate with obtuse margins. *O. vulgare* has broadly sulcate petiole with obtuse margins. The adaxial side of *P. vulgaris* is broader. *S. verbenaca* has petioles in the form of a sulcate with

acute margins. The abaxial side of *L. purpureum* var. *purpureum* is flat. The petiole of *S. salviifolia* is broadly sulcate with flat adaxially and *S. virgata* has flat and obtusely adaxially sulcate petiole. The petiole of *S. viridis* is flat adaxially with erect margins. *A. reptans* petiole looks like a

narrowly sulcate with long and acute margins. Olowokudejo (1987) studied the petiole shapes of 46 taxa belonging to the *Cruciferae* family and reported the differences in the petiole shapes. This study suggested that determining the difference in the anatomical characteristics of the petioles can be a more useful way of taxonomic classification.

There are glandular hairs on the vegetative and generative organs of the plants belonging to the *Lamiaceae* family (Werker, 2006). The species belonging to this family are able to be characterised by the presence of these secretion hairs. Glandular hairs are generally peltate and capitate hairs. Peltate hairs have short stalk with the head cell containing a lot of cells. Capitate hairs have a stalk which consists of many cells or a single cell. In addition, the head cell of capitate hairs consists of either two or single cells (Hanlidou *et al.*, 1991; Ascensao *et al.*, 1995; Valenti *et al.*, 1997). Tahir *et al.*, (1995) examined the morphology of the leaf surface of 13 species belonging to the *Lamiaceae* family and reported the presence of sesil glandular hairs. In our study, the only species that showed a small amount of sesil hair was *S. salviifolia*. Rapisarda *et al.*, (2001) studied *Nepetha sibthorpii* (*Lamiaceae*) and showed the glandular hairs micromorphology characteristic to be beneficial. Similarly, Bosabalidis & Tsekos (1984) stressed the importance that hair structures have on *Origanum* species. Kaya *et al.*, (2007) examined the peltate and capitate hairs of *Nepeta congesta* var. *congesta* and showed the amount of head and stalk cells present. Vrachnakis (2003) reported the presence of big peltate hairs in *Origanum dictamnus* and the capitate hairs were found in variety types. In this study, we found capitate hairs to be found with 1-2 stalk cells and 1-head cell; 2 head cells and 1 stalk cell. All the examined species petioles consisted of capitate hairs, whereas peltate hairs were only found in *S. salviifolia*.

Furthermore, there was an undulate cuticle on the surface of the epidermis of the species *O. vulgare*, *S. verbenaca* and *Scutellaria salviifolia*. The other studied species had smooth cuticle on the epidermis. According to the results, it is observed that the differences in the species have an important effect. All the taxa examined contained collenchyma in the petioles. However, there were differences in the amount of collenchyma layer and the position. *O. vulgare*, *P. vulgaris* and *S. verbenaca* contains collenchyma on both sides of their petioles with the abaxial side containing a lot more. The surfaces of the abaxial and adaxial of *S. salviifolia* and *A. reptans* contain a single row. *L. purpureum* var *purpureum* contains only one row on the abaxial part. Collenchyma was present in the corners of all the species.

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(Received for publication 16 February 2009)