

**Morphological, anatomical and cytological investigation on alpine *Lamium cymbalariifolium* endemic to Turkey**Canan ÖZDEMİR<sup>1</sup> & Pelin BARAN<sup>2</sup><sup>1</sup>Celal Bayar University, Faculty of Art and Science, Department of Biology, Muradiye Campus, 45030 Manisa, Turkey cozdemir13@gmail.com<sup>2</sup>Uşak University, Faculty of Science and Art, Department of Biology, 1 Eylül Campus, D2 Academic Block, 64200 Uşak, Turkey

\*Corresponding Author: pelin.baran@usak.edu.tr

**Abstract**

In this study, some morphological characters, anatomical features and chromosome numbers of alpine *Lamium cymbalariifolium* Boiss. (Lamiaceae) endemic to Turkey are firstly described in detail. *Lamium cymbalariifolium* is an east mediterranean element growing on north-facing limestone screes at an altitude of 2100-2280m in the Southwest Anatolia. The investigated species is closely allied to *L. microphyllum* and *L. sandrasicum* which are other alpine endemic species to Turkey. Taxonomically significant characters for *L. cymbalariifolium* are tried to be pointed out. Morphologically, the corolla tube length, the upper lip length, lobe length of the upper lip, the lower lip length and lobe length of the lower lip, the length of pistil, filament, anther and the seed dimensions of the species are firstly reported in the present study. Anatomically, spring and autumn woods of the root, which are forming annual rings are clearly distinguishable. Stem has a thin collenchymatous layer at the corners whereas does not bear any extraxilar sclerenchyma tissue. Leaf is bifacial. Prismatic crystals occur in leaf and generative organs. Pollen type is trizonocolpate and pollen shape is subprolate. Cross-section of seed is triangular. Glandular hairs are classified into two main types, peltate and capitate. Furthermore, capitate glandular hairs are separated into two types, as type I and type II. The chromosome number is determined as  $2n=18$ . The results are presented with photographs, illustrations and tables.

**Keywords :** Anatomy, cytology, glandular hairs, Lamiaceae, *Lamium cymbalariifolium*, morphology.**Abbreviations:** S.D. Standard deviation, Min. minimum, Max. maximum.**Introduction**

The type genus of Lamiaceae is *Lamium* L. (Harley, 2003). The genus *Lamium* comprises about 40 species of herbaceous annuals and perennials occurring from North Africa to Eurasia (Mennema, 1989; Malberley, 1997). The distribution area of the genus reaches from Western Europe to Eastern Asia, including Northern Africa, approximately between 65 and 30° Northern latitude. Outside the natural area a small number of taxa are introduced by man and sometimes naturalized in Greenland and Iceland, in the America's, Australia and Tropical and South Africa. The centre of diversity of the genus is obviously found in the Irano-Turanian and the Mediterranean regions (Mennema, 1989). In Turkey, about 30 *Lamium* species naturally exist and approximately 23 taxa including varieties and subspecies are endemic. *Lamium cymbalariifolium* Boiss. is one of endemic alpine species of Turkey (Mill, 1982; Davis et al. 1988; Güner et al. 2000). The taxon is categorized as LR(cd) endemic (Ekim et al. 2000). Some *Lamium* species have a widespread usage in officinal and folk medicines (Bisset 1994; Bremness 1995; Baytop 1999; Cui et al. 2003) and show antiproliferative activities against bacteria (Roussis et al. 1996; Trouillas et al. 2003; Matkowski and Piotrowska, 2006; Yalçın et al. 2007; Paduch et al. 2007) and are considered useful remedies in variety of diseases in Anatolia, some parts of Europe and China (Bremness 1995; Baytop 1999; Cui et al. 2003; Özyaydin et al. 2006) and traditionally used as food in some countries (Bremness 1995; Flamini et al. 2005; Cui et al. 2003). In addition, they are deemed ornamental and well

suited to a variety of growing conditions (Rudy 2004). A taxonomical revision of the genus *Lamium* which is mainly based on the study of herbarium collections has been made by Mennema in 1989. A few studies on floral forms (Lord, 1980; 1982), chromosome numbers (Gill, 1983), systematic implications of pollen morphology (Abu-Asab & Cantino, 1994) of some *Lamium* species and a revision (Park & Kim, 1995) are also available in the literature. There has not been any detailed morphological and anatomical study belonging to *Lamium* species, except the recently done studies on endemic *Lamium lycium* Boiss. (Baran & Özdemir, 2009), endemic *L. moschatum* Miller var. *rhodium* (Gand.) R. Mill (Baran & Özdemir, 2011) and *L. truncatum* Boiss. (Celep et al. 2011). The present study which contributes to future systematic studies on the genus is a detailed morphological, anatomical and cytological research on *L. cymbalariifolium* which is one of endemic alpine species of genus *Lamium*.

**Results****Morphological properties**

Figure 1 shows view (Fig 1A), flowers (Fig 1B-E), habit (Fig 1F), flowered stem (Fig 1G), fruits (Fig 1H-J) and seeds of *L. cymbalariifolium* (Fig 1K-L). Figure 2 shows drawings of view (Fig 2A), leaf (Fig 2B), inflorescence (Fig 2C), calyx (Fig 2D), flower (Fig 2E) and seed (Fig 2F) of the taxon. The plant species is a caespitose perennial which have a taproot

covered by a very thin cork layer and creeping rootstocks reaching 30 cm in length and lying under stones (Fig 1F;2A). The stem is slender, clearly quadrangular and ascending (Fig 1F,G;2A). The stem is almost glabrous. The leaves are cymbal-like in appearance, clearly reniform in shape and dark green, brownish to purplish in colour. The leaf apex is obtuse. The leaf base is deeply cordate to cuneate. The leaves bear (3 -)5 - 7 crenations and venation reticulate-pinnate or -palmate (Fig 1A,G;2B,C). The leaf indumentum is glabrous or sparsely pubescent. The petiole is glabrous. The bracts are as same as leaves and sparsely pubescent. The verticillasters are 2-flowered and crowded near apex (Fig 1G;2C). The bracteoles are acicular in shape, sparsely pubescent (Fig 2D). The calyx has 5 triangular nearly equal teeth and the teeth are 2/3 fold the tube. The tube is nearly glabrous whereas the teeth are puberulent or pubescent (Fig 1I;2D). The bilabiate corolla is of a rose-pink upper lip and a lighter tube and a lower lip with purple stripes. The corolla tube is slightly curved or straight, villous and without annulus. The upper lip is deeply bifid forming two lobes that may be indented and the indumentum is glandular-pilose. The median lobe of lower lip is of transversely reniform parts and each part is shallowly emarginate (Fig 1A-E,G;2E). The stigma is bifid. The stamens are didynamus and the anther thecae are divaricate and hairy. The nutlets are triquetrous and dark brownish or brownish-olive in colour (Fig 1K-L;2F). Table 1 shows morphological measurements of the investigated taxon.

### Anatomical properties

**Root:** In the cross-section, 21 - 35 layered peridermis at the outermost layer, 20 - 25 layered parenchymatous cortex, 1-layered endodermis, phloem region, 1 - 5 layered cambium and xylem at the innermost layer respectively can be visible. In young roots epidermis exists at the outermost layer and a large parenchymatous pith region is located at the innermost. Any extraxilar sclerenchyma tissue does not exist. Vessels in xylem are arranged irregularly. 1-layered secondary xylem rays and 1 - 20 layered primary pith rays are located in xylem. Spring and autumn woods which consist of annual rings are clearly distinguishable (Fig 3A, Table 2).

### Stem

The stem cross-section is quadrangular to roundish (Fig 3B). Epidermis is formed by oval, squarish, or rectangular shaped cells. 1 - 2 layered plate or angular collenchyma is located at the corners (Fig 3C). 5 - 9 layered parenchymatous cortex is located under epidermis. Any extraxilar sclerenchyma tissue does not exist. Phloem and 2 - 4 layered vascular cambium are distinguishable. Mainly 4 major vascular bundles occur. Tracheae show orderly linear and radial arrangement. A parenchymatous pith region with intercellular spaces is located at the innermost layer (Fig 3B, Table 2).

### Petiole

Epidermis at the outermost layer, is formed by cells nearly oval, quadrangular or circular (Fig 4D). Epidermis bears glandular hairs and stoma cells. Parenchyma with circular cells and intercellular spaces is 4 - 8 layered under epidermis. Parenchyma cells bear large and small prismatic crystals and a small number of chloroplasts. However the cells located close to epidermis are richer in point of chloroplasts. 1 layered discontinuous collenchyma adjacent to epidermis is located at the corners. There are two large and one small

collateral vascular bundles in the center and also one small bundle at the petiolar wings (Fig 3D, Table 2).

### Leaf

Diacytic stomata and some starch grain containing cells occur in both adaxial and abaxial epidermis. Palisade parenchyma is 3 - 4 layered and spongy parenchyma is 3 - 6 layered (Fig 4A). Mesophyll cells are getting smaller and roundish around the small median bundle in the cross-section of leaf midrib (Fig 4B). Mesophyll cells bear many large and small prismatic or needle-shaped crystals (Fig 4A-B, Table 2).

### Calyx

Abaxial epidermis is usually thicker than the adaxial. Both epidermis bear glandular hairs and stoma cells. 7 - 9 layered parenchyma is of elongated or flattened cells and large intercellular spaces. Parenchyma cells contain many plastids, large starch grains and crystals (Fig 4C, Table 2).

### Corolla

Abaxial epidermis is usually thicker than the adaxial. Epidermis cells contain a small number of starch grains. Parenchyma with large intercellular spaces is 9 - 11 layered. Parenchyma cells contain plastids, starch grains and prismatic or needle-shaped crystals (Fig 4D, Table 2).

### Generative Organs

The cross-section of pistil is nearly elliptical at the median level. An epidermis at the outermost layer, a central sclerenchyma axis and two small lateral vascular bundles and parenchyma with small starch grains, respectively, take part (Fig 4E). Each anther is of two thecae and each theca is of two pollen sacs. Tapetum at the innermost layer of pollen sac covers pollen grains (Fig 4F). Filament consists of an epidermis with small starch grains, a central small vascular bundle and parenchyma with chloroplasts, large starch grains and prismatic or needle-shaped crystals (Fig 4G). Pollen type is trizonocolpate, which is classified according to Moore et al. (1991). Pollen shape is subprolate, which is classified according to Punt et al. (2007). The exine ornamentation is reticulate (Fig 4I-K, Table 3). Seed consists of an embryo at the innermost layer, 6 - 20 layered endosperm, 3 - 6 layered parenchyma with flattened cells, 1 layered seed coat with quadrangular or rectangular cells and sclerenchyma layer containing very large prismatic crystals and a cork layer that consists of longitudinally elongated cells containing large starch grains at the outermost layer, respectively. Both sclerenchyma which is giving general shape of seed, and cork layer take part at the outermost two layers, and serve as protective tissues for seed (Fig 4H).

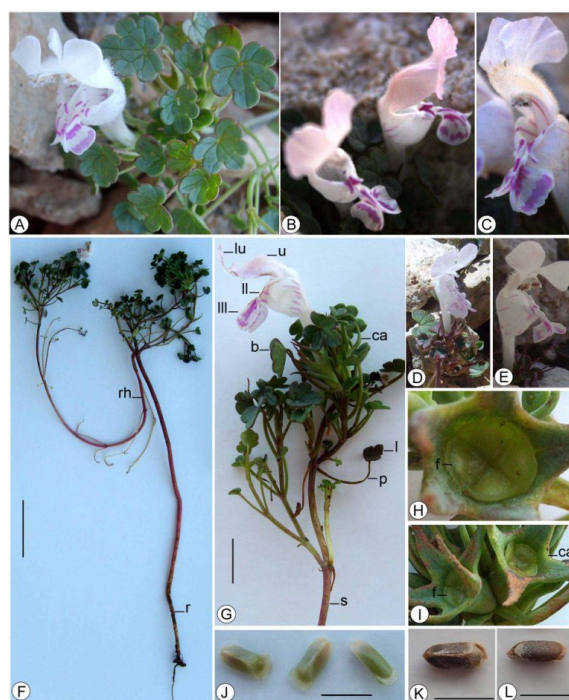
### Trichome properties

Glandular hairs consist of two main types as capitate and peltate. Capitate glandular hairs are divided into two types, as type I and II, according to Werker (1993)'s classification. The capitate glandular hairs bear 1, 4 head cells and 1 or 2 stalk cells (Fig 5A-C). Type I capitate hairs bear a round shaped head and secreted drops on the cuticle (Fig 5A-B) while type II capitate hairs bear a slightly elongated head with only 1 cell and a broken cuticle (Fig 5C). Peltate glandular hairs (Fig 5D-O) bear a large head consisting of 4,

**Table 1.** Morphological measurements of *Lamium cymbalariaifolium*.

	Min.-Max. (cm)	Mean±S.D* (cm)		Min.-Max. (cm)	Mean±S.D* (cm)
<b>Root</b>					
Root length	14.00-60.00	33.73±17.05	<b>Corolla</b>		
<b>Stem</b>			Lobe of upper lip	0.40-0.65	0.53±0.08
Stem length	13.00-49.00	25.79±10.80	Lower lip length	0.65-1.50	1.11±0.29
<b>Leaf</b>			Lobe of lower lip	0.30-0.50	0.36±0.08
Leaf length	00.35-01.10	00.65±00.24	Upper Filament	0.70-1.10	0.93 ± 0.13
Leaf width	00.50-01.40	00.77±00.32	Lower Filament	0.90-1.60	1.30±0.22
<b>Petiole</b>			Anther length	0.13-0.25	0.17±0.04
Petiole length	00.70-02.70	01.78±00.70	Pistil length	2.80-3.90	3.33±0.34
<b>Calyx</b>			<b>Bract</b>		
Calyx length	1.25-1.90	1.52±0.22	Bract length	0.30-0.85	0.54±0.14
Teeth length	0.45-1.10	0.75±0.18	Bract width	0.50-1.00	0.75±0.16
Calyx tube	0.50-0.90	0.67±0.13	<b>Bract stick</b>	1.00-1.90	1.39±0.29
<b>Corolla</b>			<b>Bracteol</b>	0.70-1.65	1.01±0.26
Corolla length	2.70-4.50	3.57±0.52	<b>Seed</b>		
Tube length	1.60-3.15	2.28±0.44	Seed length	0.40-0.55	0.46±0.04
Upper lip length	0.90-1.75	1.33±0.24	Seed width	0.18-0.25	0.22±0.03

\* S.D. : Standard Deviation.



**Fig 1.** View (A), flowers (B-E), habit (F), flowered stem (G), fruits (H-J), seeds (K-L) of *Lamium cymbalariaifolium* (Scale Bar 5 cm for F, 1 cm for G, 5 mm for J-L, K. Ventral surface L. Dorsal surface, r. root, rh. rhizome, s. stem, p. petiole, l. leaf, b. bract, ca. calyx, u. upper lip of corolla, lu. lobe of the upper lip, ll. lower lip of corolla, lll. lobe of the lower lip.

6, 7, 8 or 10 cells and 1 central cell and an additional stalk 1 or 2 celled (Fig 5H,J,M) or without stalk (Fig 5D-F).

#### Cytological properties

Somatic chromosome number of *Lamium cymbalariaifolium* was counted as  $2n=18$  at the root tip mitosis (Fig 6).

#### Discussion

Our morphological findings of *L. cymbalariaifolium* were generally consistent with the morphological description of the taxon given in the Flora of Turkey (Mill, 1982), with some

exceptions of the numerical data. Mill (1982) reported the stem as 12 - 20 cm, petiole 0.6 - 2 cm, leaf length 0.17 - 0.6 cm, leaf width 0.2 - 0.7 cm, bracteol 0.9 - 1.1 cm, calyx 1.1 - 1.3 cm and corolla 2.6 - 3.5 cm, while we determined them as 13 - 49 cm, 0.7 - 2.7 cm, 0.35 - 1.1 cm, 0.5 - 1.4 cm, 0.7 - 1.65 cm, 1.25 - 1.9 cm and 2.7 - 4.5 cm, respectively (Table 1). The findings show that the upper limits of characters mentioned above seem to be enhanced. This may be resulted depending on number of the collected plant samples and some changes in seasonal conditions for years. The perennial taxa of the genus *Lamium* possess more or less woody rhizomes which are lacking in the annual taxa (Mennema, 1989). *L. cymbalariaifolium* which is a perennial taxon bears

**Table 2.** Anatomical measurements of *Lamium cymbalariifolium*.

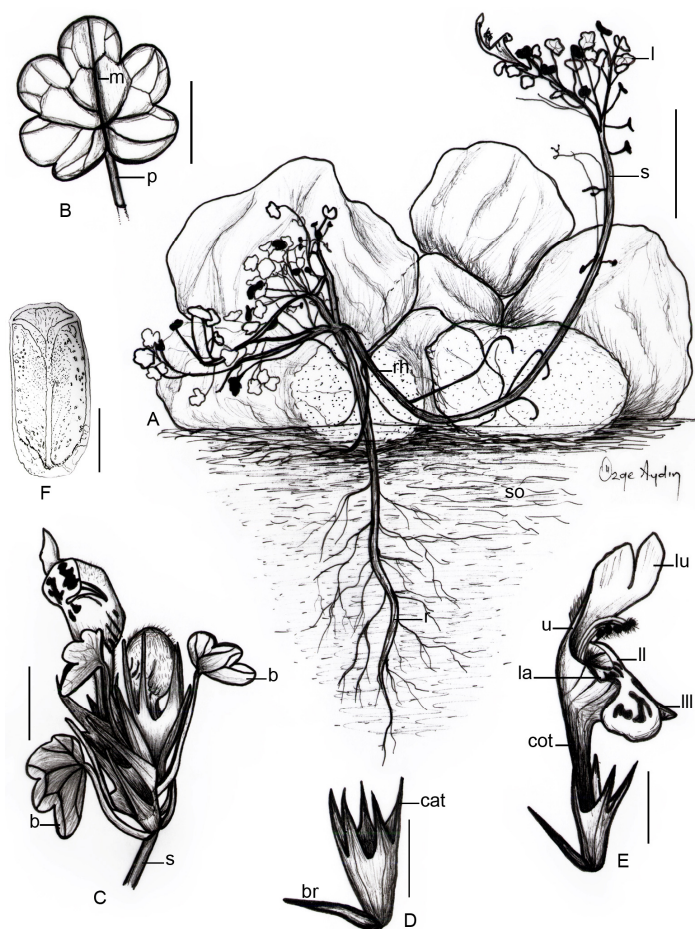
	Width (µm)		Length (µm)	
	Min. - Max.	Mean ± S.D.*	Min. - Max.	Mean ± S.D.*
<b>Root</b>				
Epidermis cell	26.50-79.40	43.85±18.10	34.40-58.20	43.67±8.46
Peridermis cell	26.50-100.60	53.6 ±27.04	23.80 -42.40	29.79 ±5.80
Parenchyma cell	10.60-100.60	50.58±28.93	07.90 -42.40	23.10 ±09.28
Vessel diameter	10.60-79.40	49.74±26.36		
<b>Stem</b>				
Epidermis cell	13.20-47.60	33.30 ±13.32	21.20 -42.40	31.57 ±8.42
Parenchyma cell	23.80 -100.60	61.41±28.79	15.90-100.60	57.00±29.94
Collenchyma cell	13.20-39.70	24.85±7.84	15.90-37.10	28.32±5.12
Vessel diameter	11.60 -52.90	33.71 ±16.18		
Pith cell diameter	21.20-132.40	78.15 ±37.86		
<b>Petiole</b>				
Cuticle thickness	5.30-10.60	6.95±1.90		
Adaxial Epidermis cell	10.60-63.50	36.74±16.27	21.20-68.80	43.52±15.66
Abaxial Epidermis cell	10.60-47.60	25.16±11.82	18.50-47.60	30.88±10.68
Parenchyma cell diameter	15.90-100.60	55.42±30.40		
Vessel diameter	5.30-18.50	12.07±4.79		
<b>Leaf</b>				
Adaxial cuticle thickness	4.20-13.20	8.68±2.46		
Abaxial cuticle thickness	5.30-13.20	8.62±2.58		
Adaxial epidermis cell	13.20-116.50	58.06±35.88	15.90-52.90	33.53 ±12.71
Abaxial epidermis cell	15.90-105.90	49.41±27.24	21.20-58.20	36.72±11.21
Spongy cell dimension	26.50-95.30	58.37±22.70		
Palisade cell	26.50-58.20	43.76±9.61	42.40-95.30	71.29±18.41
Vessel diameter	6.40-18.50	10.74±3.11		
<b>Calyx</b>				
Adaxial epidermis cell	15.90-105.90	49.10±33.80	18.50-52.90	34.42±13.28
Abaxial epidermis cell	10.60-84.70	52.13±23.00	21.20-63.50	41.32±14.29
Parenchyma cell	23.80-68.80	43.17±12.37	23.80-84.70	51.50±19.64
Vessel diameter	6.40-18.50	12.55±3.81		
<b>Corolla</b>				
Adaxial epidermis cell	15.90-47.60	31.12±9.65	15.90-52.90	33.09±12.83
Abaxial epidermis cell	13.20-68.80	35.08±16.77	18.50-47.60	30.68±9.20
Parenchyma cell	13.20-47.60	28.15±9.78	10.60-39.70	22.15±8.65
Vessel diameter	5.30-15.90	10.74±2.74		

\* S.D. : Standard Deviation

rhizomes as reported for perennial *L. lycium* (Baran & Özdemir, 2009) in contrast to annual *L. moschatum* var. *rhodium* (Baran & Özdemir, 2011). The anatomical analysis given in this work provides the first detailed description of *L. cymbalariifolium*, which is comparable with findings of Metcalfe and Chalk (1972), some *Lamium* species (Baran & Özdemir, 2009; Baran & Özdemir, 2011; Celep et al. 2011) and some other investigated Lamiaceae members (Çobanoğlu, 1988; Çobanoğlu et al. 1992, Özdemir & Şenel, 1999; 2001; Kaya & Başer, 2002; Uysal, 2002; Baran & Özdemir, 2006; Dinç & Öztürk, 2008). According to Metcalfe and Chalk (1972), pith rays of Lamiaceae family are 2 - 12 or more rowed and quite heterogeneous in structure. The root cross-section of *L. cymbalariifolium* pointed out 1 - 20 rowed heterogeneous pith rays. The pith rays are 1-4 rowed for both *L. lycium* (Baran & Özdemir, 2009) and *L. truncatum* roots (Celep et al. 2011) and 1-2 rowed for *L. moschatum* var. *rhodium* root (Baran & Özdemir, 2011). The old root of the investigated taxon bears a thick peridermis and a centre filled with xylem elements while the young root bears an epidermis and a parenchymatous pith (Fig 3A). However, the old root of *L. lycium* has a parenchymatous centre in contrast to the young root which is filled with xylem elements (Baran & Özdemir, 2009). The root center of annual

*L. moschatum* var. *rhodium* points out primary xylem elements (Baran & Özdemir, 2011). The root center which is filled with primary xylem is also reported in some Lamiaceae members (Çobanoğlu, 1988; Çobanoğlu et al. 1992; Özdemir & Şenel, 1999; Uysal, 2002; Baran & Özdemir, 2006) in contrast to some others (Özdemir & Şenel, 2001). The characteristic feature of Lamiaceae family is a quadrangular stem and a well-developed collenchyma as supporting tissue at the corners of stem and a developed sclerenchyma tissue surrounding the vascular tissue (Metcalfe & Chalk, 1972). A thin collenchyma occurred at the corners of quadrangular stem and even in petiole of *L. cymbalariifolium* (Fig 3B-D). However, any extraxilar sclerenchyma is hardly seen in the cross-sections of neither stem nor root of *L. cymbalariifolium* (Fig 3B). Thicker and more protruding collenchyma is reported for both *L. lycium* (Baran & Özdemir, 2009) and *L. truncatum* stems (Celep et al. 2011). Endodermis is distinguishable in the root but not in the stem of *L. cymbalariifolium*. However endodermis is visible in root and





**Fig 2.** Drawings belonging to *Lamium cymbalariifolium*  
 A. View, B. Leaf, C. Inflorescence, D. Calyx, E. Flower, F. Seed , (Scale Bar 5cm for A, 5mm for B, 1 cm for C-E, 2 mm for F), r. root, rh. rhizome, s. stem, p. petiole, l. leaf, m. midrib, b. bract, b. bracteol, cat. calyx teeth, cot. corolla tube, la. lateral appendage, u. upper lip of corolla, lu. lobe of the upper lip, ll. lower lip, ll. lobe of the lower lip, so. soil.

stem of *L. lycium* (Baran & Özdemir, 2009) and stem of *Sideritis galatica* Bornm. (Kaya & Başer, 2002).

Vascular cambium is distinguishable in root and stem of *L. cymbalariifolium* (Fig 3B) as reported in *L. lycium* (Baran & Özdemir, 2009) and *L. moschatum* var. *rhodium* (Baran & Özdemir, 2011), in herbaceous stems of *Stachys yildirimli* M. Dinç (Dinç & Öztürk, 2008), and in stems of some *Salvia* species (Çobanoğlu, 1988; Özdemir & Şenel, 2001) and in contrast to stems of *L. truncatum* (Celep et al. 2011), *Sideritis galatica* (Kaya & Başer, 2002) and some other *Salvia* species (Özdemir & Şenel, 1999; Baran & Özdemir, 2006). A pith hollow does not occur in the stem centre of *L. cymbalariifolium* (Fig 3B). Vascular bundle structure in petiole of Lamiaceae species may be important in point of taxonomy (Metcalf & Chalk, 1972). The petiole cross-section, as illustrated in Figure 3D, points out same structure

as well as petioles of *L. lycium* (Baran & Özdemir, 2009), *L. moschatum* var. *rhodium* (Baran & Özdemir, 2011), *L. truncatum* (Celep et al. 2011), *L. album* and *L. takesimense* taxa (Park & Kim, 1995). Palisade parenchyma of mesophyll is 3-4 layered for *L. cymbalariifolium* leaf while it is 2-3 layered for *L. lycium* leaf (Baran & Özdemir, 2009), 1 or 3 layered for *L. moschatum* var. *rhodium* leaf (Baran & Özdemir, 2011), 1 layered for *L. truncatum* leaf (Celep et al. 2011). The cross-section of the leaf midrib of *L. cymbalariifolium* points out a concave shape making the leaf gain “cymbalariiform” characteristic (Fig 1A,G; 2B-C; 4B). Anatomical structures of pistil, filament and anther of *L. cymbalariifolium* (Fig 4E-G) are consistent with those of *L. amplexicaule* L. (Lord, 1980; 1982), *L. lycium* (Baran & Özdemir, 2009), *L. moschatum* var. *rhodium* (Baran & Özdemir, 2011). Glandular hairs can be visible throughout the plant especially on inflorescence (Fig 5A-O). Two main types of glandular hairs, as capitate and peltate, are distinguished in the family (Werker, 1993; Baran et al. 2010a;b) as earlier reported in *L. lycium* (Baran & Özdemir, 2009), *L. moschatum* var. *rhodium* (Baran & Özdemir, 2011) and *L. truncatum* (Celep et al. 2011). Two different types of capitate hairs, as type I and type II, are detected. The type I capitate hairs in our work (Fig 5A-B) correspond to type I capitate glandular hairs described by Werker (1993), Ascensão et al. (1995), Ascensão & Pais (1998), Bisio et al. (1999) and to type II capitate glandular hairs described by Serrato-Valenti et al. (1997). The type II capitate hairs in our work (Fig 5C) correspond to type II capitate glandular hairs described by Werker (1993).

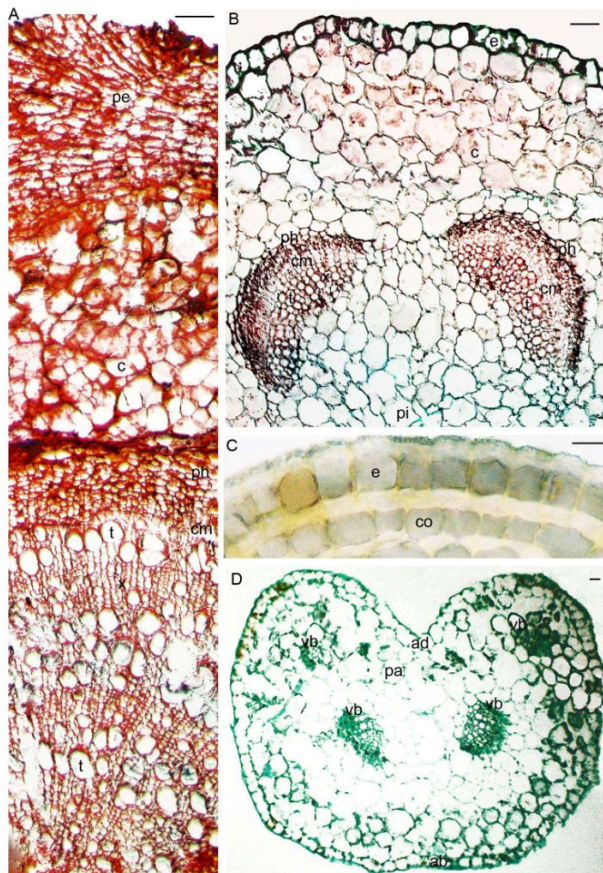
In addition to the peltate hairs with a four-celled head (Fig 5E-F,K), as reported earlier for *Lamium galeobdolon* (L.) L. (Uphof & Hummel, 1962) and *L. truncatum* (Celep et al. 2011), the peltate hairs with more than 4-celled and raised head upon epidermis (Fig 5H,J,M), as described earlier by Corsi and Bottega (1999) are also found in *L. cymbalariifolium*. Those two kinds of peltate hairs are also reported for *L. lycium* (Baran & Özdemir, 2009) and *L. moschatum* var. *rhodium* (Baran & Özdemir, 2011). As a result of the present study, morphological description of endemic *L. cymbalariifolium* which is a perennial herbaceous species has been expanded contributing to the knowledge of the Flora of Turkey (Mill, 1982). We determined that stem, petiole, bracteol, calyx, corolla length and leaf dimensions of *L. cymbalariifolium* increased.

Furthermore, numerical morphological data belonging to the anther, filament, pistil, corolla tube, upper lip, upper lip lobes, lower lip, lower lip lobes and seed have been firstly reported in the present study (Table 1). On the other hand, the present study which is comparable with some *Lamium* species and other Lamiaceae members in anatomical aspect contributes to anatomical data of the family pointing out that any extraxilar sclerenchyma tissue does not exist (Table 2,3; Fig 3-5). Cytologically, it is outstanding that somatic chromosome number of the taxon (Fig 6) does not differ from that of *L. moschatum* var. *rhodium* (Baran & Özdemir, 2011). However, further investigation with the other *Lamium* species may illuminate whether any taxonomical implication is present or not within the genus *Lamium* or in the family. The anatomical and cytological findings in the present study supply the first data available for *L. cymbalariifolium* in the literature.

**Table 3.** Pollen measurements of *Lamium cymbalariifolium*

Pollen	Min.	-	Max.	Mean	±	S.D.*
Polar Axis (P)	28.60	-	41.60	36.83	±	03.13
Equatorial Diameter (E)	23.40	-	33.80	29.55	±	03.31
P/E Ratio	00.92	-	01.61	01.26	±	00.18

\* S.D. : Standard Deviation.



**Fig 3.** Protective, supportive, vascular and ground tissues of the root (A), stem (B,C) and petiole (D) of *Lamium cymbalariifolium*  
e. epidermis, pe. periderm, c. cortex parenchyma, co. collenchyma, ph. phloem, cm. cambium, x. xylem, t. trachea, sw. spring wood, aw. autumn wood, pi. pith, pih. pith hollow (Scale Bars 63µm for A,B,D, 17 µm for C).

## Materials and Methods

### Materials

Plant samples have been collected from natural populations of the given location at different dates. Some samples have been used for morphological examination, some have been dried as herbarium samples. All of the herbarium samples which are collected at the same date take the same number. The samples have been collected from the following location: C 2 Antalya: Elmalı, Sedir Araştırma Ormanı, Kızlar Sivrisi, 1953m, 27 July 2008, *Baran 218*. C 2 Antalya: Elmalı, Sedir Araştırma Ormanı, Kızlar Sivrisi, 1933m, 26 June 2009, *Baran 270*.

### Morphological examination method

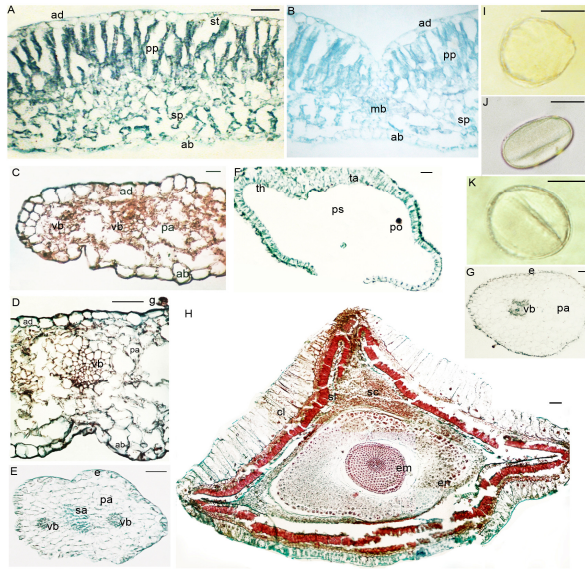
The taxonomical description of the specimens followed Mill (1982). Morphological measurements have been based on 15-20 plant specimens. Morphological original drawings of the

vegetative and generative organs of the species have been made. Photographs of both the plant species in the field and the herbarium samples and their parts have been taken by Sony digital camera.

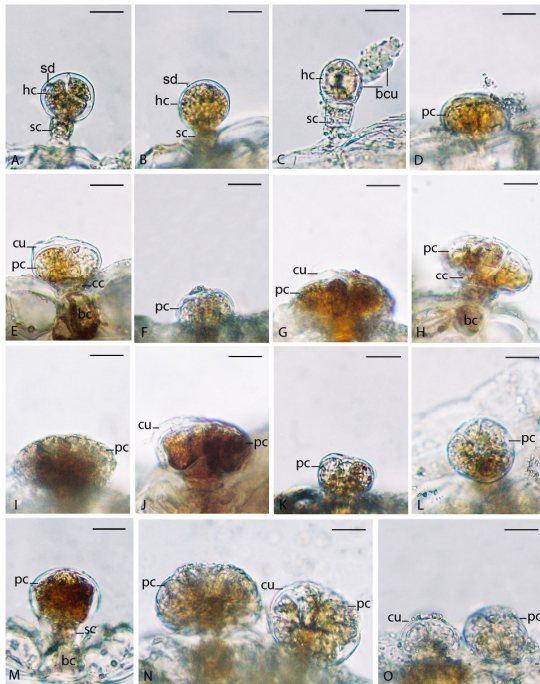
### Anatomical examination method

Anatomical studies have been carried out on the samples kept in alcohol 70%. The paraffin method (Algan, 1981) using microtome has been applied for preparing the anatomical cross-sections of root, stem, petiole, leaf, corolla, anther and seed. Safranin O and Fast green has been used for dyeing the cross-sections. Handle-blade sections have been taken for calyx, pistil, filament and stem collenchyma tissue, dyeing with Sartur reactive (Baytop, 1981). Anatomical measurements have been based on 30 cells. Classification of glandular hairs followed Werker (1993). Pollen slides have been prepared using the technique of Wodehouse (1965). Measurements have been based on 30 pollen grains.

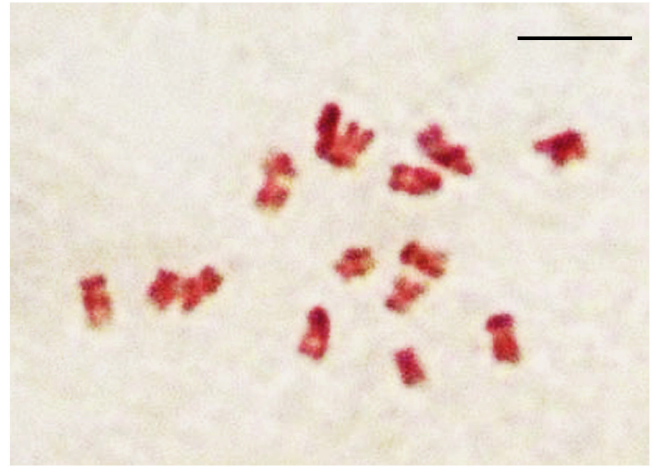




**Fig 4.** Anatomical properties of the leaf (A), midrib (B), calyx (C), corolla (D), pistil (E), anther (F), filament (G), seed (H) and polar (I) and equatorial (J-K) view of pollen of *Lamium cymbalariaifolium*, ab. abaxial epidermis, ad. adaxial epidermis, cl. cork layer, e. epidermis, em. embryo, en. endosperm, pa. parenchyma, mb. median bundle, vb. vascular bundle, pp. palisade parenchyma, sa. sclerenchyma axis, sc. seed coat, sl. sclerenchyma layer, sp. spongy parenchyma, st. stoma, g. glandular hair, th. theca, ta. tapetum, ps. pollen sac, po. pollen (Scale Bars 35  $\mu$ m for A-G, 100  $\mu$ m for H, 17  $\mu$ m for I-K).



**Fig 5.** Glandular trichome types of *Lamium cymbalariaifolium* Type I capitate glandular hairs (A-B), Type II capitate glandular hairs (C), Peltate glandular hairs (D-O). hc. head cell, cu. cuticle, sc. stalk cell, sd. secreted drop, bc. base cell, bcu. broken cuticle, pc. periphery cell, cc. central cell (Scale Bars: 17  $\mu$ m)



**Fig 6.** Somatic chromosomes of *Lamium cymbalariaifolium* (Scale Bar 4  $\mu$ m).

Classification of pollen type and shape followed Moore et al. (1991) and Punt et al. (2007) respectively. Microscopic examinations have been made on the research microscope OLYMPUS BX50 with attachment of camera.

#### Cytological examination method

Cytological study has been carried out according to the following steps. Seeds have been germinated in sterilized petri dishes. Then root tips have been pretreated with saturated solution of  $\alpha$ -mono-bromonaphtalene (16 h) and fixed in a mixture of 3:1 ethanol and acetic acid for 24 h. Root tips have been hydrolyzed with 1 N HCl for 10 min at 60°C in an oven, then stained with Feulgen reagent for 1 h in darkness and finally squashed in 45% acetic acid. Squash techniques followed Elçi (1994) and Gönüz et al. (2009). Cytological analysis has been made on the research microscope Olympus BX50 and photographs have been taken on Leica DW 3000 with the camera Leica DFC 295.

#### Acknowledgements

We would like to thank Celal Bayar University, The Unit of Scientific Research Projects for financial support.

#### References

- Abu-Asab MS, Cantino PD (1994) Systematic implications of pollen morphology in subfamilies Lamioideae and Pogostemonoideae (Labiatae). *Ann Mo Bot Gard.* 81:653–686
- Algan G (1981) Bitkisel dokular için mikroteknik, Fırat Üniv. Fen-Ed. Fak. Yayınları Bot. No:1, İstanbul
- Ascensão L, Marques N, Pais MS (1995) Glandular trichomes on vegetative and reproductive organs of *Leonotis leonurus* (Lamiaceae). *Ann Bot-London* 75:619–626
- Ascensão L, Pais MS (1998) The leaf capitate trichomes of *Leonotis leonurus*: histochemistry, ultrastructure and secretion. *Ann Bot-London* 81:263–271

- Baran P, Özdemir C (2006) The morphological and anatomical characters of *Salvia napifolia* Jacq. (Lamiaceae) in Turkey. *Bangladesh J Botany* 35:77–84
- Baran P, Özdemir C (2009) The morphological and anatomical properties of *Lamium lycium* (Lamiaceae), endemic to Turkey. *Nord J Bot.* 27:1–9
- Baran P, Aktaş K, Özdemir C (2010a) Structural investigation of the glandular trichomes of endemic *Salvia smyrnea* L. *S Afr J Bot.* 76:572–578
- Baran P, Özdemir C, Aktaş K (2010b) Structural investigation of the glandular trichomes of *Salvia argentea* L. *Biologia* 65(1):33–38
- Baran P, Özdemir C (2011). Morphological, anatomical and cytological investigation of endemic *Lamium moschatum* Miller var. *rhodium* (Gand.) R. Mill. *Biologia* 66(3):439–447
- Baytop A (1981) Bitkisel drogların anatomik yapısı, İstanbul University Publications, İstanbul
- Baytop T (1999) Türkiye’de bitkilerle tedavi (Geçmişte ve Bugün), Nobel Tıp Kitapevleri, İstanbul
- Bisio A, Corallo A, Gastaldo P, Romussi G, Ciarallo G, Fontana N, De Tommasi N, Profumo P (1999) Glandular trichomes and secreted material in *Salvia blepharophylla* Brandege ex Epling grown in Italy. *Ann Bot-London* 83:441–452
- Bisset NG (1994) Herbal drugs and phytopharmaceuticals: A handbook for practice on scientific basis, Scientific Publishers, Stuttgart
- Bremness L (1995) The complete book of herbs, Dorling Kindersley Publishing, London
- Celep F, Kahraman A, Atalay Z, Doğan M (2011) Morphology, anatomy and trichome properties of *Lamium truncatum* Boiss. (Lamiaceae) and their systematic implications. *Aust J Crop Sci.* 5(2):147–153
- Corsi G, Bottega S (1999) Glandular trichomes of *Salvia officinalis*: new data on morphology localization and histochemistry in relation to function. *Ann Bot-London* 84:657–664
- Cui SY, Chen XG, Hu Z (2003) Identification and determination of ecdysone and phenylpropanoid glucoside and flavonoids in *Lamium maculatum* by capillary zone electrophoresis. *Biomed Chromatogr.* 17:477–482
- Çobanoğlu D (1988) *Salvia palaestina* Bentham’ın (Lamiaceae) morfolojik ve sitolojik özellikleri. *Doğa Bilim Dergisi:Biyojoloji* 12:215–223
- Çobanoğlu D, Özel S, Evren H (1992) *Salvia trichoclada* Bentham (Lamiaceae)’nin morfolojik özellikleri. XIth National Biology Congress, Haziran Elazığ, pp. 83–89
- Davis PH, Mill RR, Tan K (1988) Flora of Turkey and the east aegean islands (suppl.1), Edinburgh Univ. Press, Edinburgh
- Diñç M, Öztürk M (2008) Comparative morphological, anatomical and palynological studies on the genus *Stachys* L. sect. *Amblesia* Bentham (Lamiaceae) species in Turkey. *Turk J Bot.* 32:113–121
- Ekim T, Koyuncu M, Vural M, Duman G, Aytaç Z, Adıgüzel N (2000) Red data book of Turkish plants (Pteridophyta and Spermatophyta), Foundation for Turkish Nature Conservation and Van Centinential University Press, Ankara
- Elçi Ş (1994) Sitogenetikte araştırma yöntemleri ve gözlemler, 100. Yıl Üniversitesi Yayınları, Van
- Flamini G, Cioni PL, Morelli I (2005) Composition of the essential oils and in vivo emission of volatiles of four *Lamium* species from Italy: *L. purpureum*, *L. hybridum*, *L. bifidum* and *L. amplexicaule*. *Food Chemistry* 91:63–68
- Gill LS (1983) Cytotaxonomic studies of the tribe Stachydeae (Labiatae) in Canada. *Willdenowia* 13:175–181
- Gönüz A, Kesercioğlu T, Akı C (2009) Sitotaksonomide temel ilkeler, Onsekiz Mart Üniversitesi, Çanakkale
- Güner A, Özhatay N, Ekim T, Başer KHC (2000) Flora of Turkey and the east aegean islands (suppl. 2), Edinburgh University Press, Edinburgh
- Harley RM (2003) Validation of the Lamiaceae (Labiatae). *Kew Bulletin* 58:765–766
- Kaya A, Başer KHC (2002) Endemik bir tür: *Sideritis galatica* Bornm. (Labiatae). In: Başer KHC, Kırımer N (eds) Proceedings of the XIVth symposium on plant originated crude drugs, Eskişehir, 2002
- Lord E (1980) An anatomical basis for the divergent floral forms in the cleistogamous species, *Lamium amplexicaule* L. (Labiatae). *Am J Bot.* 67:1430–1441
- Lord E (1982) Morphogenesis in *Lamium amplexicaule* L. (Labiatae) with a model for the evolution of the cleistogamous flower. *Bot Gaz.* 143:63–72
- Malberley DJ (1997) The plant book: A portable dictionary for the vascular plants, Cambridge University Press, Cambridge
- Matkowski A, Piotrowska M (2006) Antioxidant and free radical scavenging activities of some medicinal plants from the Lamiaceae. *Fitoterapia* 77:346–353
- Mennema J (1989) A taxonomic revision of *Lamium* (Lamiaceae), Leiden Botanical Series, Leiden
- Metcalfe JR, Chalk L (1972) Anatomy of the dicotyledons, vol 2, Clarendon Press, Oxford
- Mill RR (1982) *Lamium* L. In: Davis PH (ed) Flora of Turkey and the east aegean islands, vol 7, Edinburgh Univ. Press, Edinburgh
- Moore PD, Webb JA, Collinson ME (1991) Pollen analysis, 2nd edn. Blackwell Science Ltd, United Kingdom
- Özaydın S, Dirmenci T, Tümen G, Başer KHC (2006) Plants used as analgesic in the folk medicine in Turkey. In: Ertuğ F (ed) ICEB 2005: Proceedings of the 4th international congress of ethnobotany, İstanbul, August 2005. Ege Publications, p 167
- Özdemir C, Şenel G (1999) The morphological, anatomical and karyological properties of *Salvia sclarea* L. *Turk J Bot.* 23:7–18
- Özdemir C, Şenel G (2001) The morphological, anatomical and karyological properties of *Salvia forskahlei* L. (Lamiaceae) in Turkey. *J Econ Taxon Bot.* 19:297–313
- Paduch R, Wojciak-Kosior M, Matysik G (2007) Investigation of biological activity of *Lamium albi* flos extracts. *J Ethnopharmacol.* 110:69–75
- Park SJ, Kim YS (1995) A taxonomic study on the genus *Lamium* (Lamiaceae) in Korea. *Korean J Plant Taxon.* 25:103–122
- Punt W, Hoen PP, Blackmore S, Nilsson S, Le Thomas A (2007) Glossary of pollen and spore terminology. *Rev Palaeobot Palyno.* 143:1–81
- Roussis V, Chinou I, Perdetzoglou D, Loukis A (1996) Identification and bacteriostatic activity of the essential oil of *Lamium garganicum* L. subsp. *laevigatum* Arcangeli. *J Essent Oil Res.* 8:291–293
- Rudy MR (ed) (2004) A comparative study of ground cover *Lamium*. *Plant Evaluation Notes* 23:1–4
- Serrato-Valenti G, Bisio A, Cornara L, Ciarallo G (1997) Structural and histochemical investigation of the glandular trichomes of *Salvia aurea* L. leaves and chemical analysis of the essential oil. *Ann Bot-London* 79:329–336



- Trouillas P, Calliste CA, Allais DP, Simon A, Marfak A, Delage C, Duroux JL (2003) Antioxidant, anti-inflammatory and antiproliferative properties of sixteen water plant extracts used in the Limousin countryside as herbal teas. *Foodchemistry* 80:399–407
- Uphof JCT, Hummel K (1962) Plant hairs. In: Zimmermann W, Ozenda PG (eds) *Encyclopedia of plant anatomy*, Gebrüder Borntraeger, Berlin
- Uysal İ (2002) *Stachys cretica* L. subsp. *smyrnea* Rech Fill. endemik taksonunun morfolojisi, anatomisi ve ekolojisi üzerinde arařtırmalar. *Ekoloji Çevre Dergisi* 11:16–20
- Werker E (1993) Function of essential oil-secreting glandular hairs in aromatic plants of the Lamiaceae. *Flavour Frag J.* 8:249–255
- Wodehouse RP (1965) *Pollen grains*, Hammer Press, New York
- Yalçın FN, Kaya D, Kılıç E, Özalp M, Ersöz T, Çalış İ (2007) Antimicrobial and free radical scavenging activities of some *Lamium* species from Turkey. *Hacettepe University Journal of the Faculty of Pharmacy* 27:11–22