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Research Article

SYSTEMATIC SIGNIFICANCE OF FOLIAR EPIDERMAL CHARACTERS IN FIVE SPECIES OF *EREMOGONE* FENZL (CARYOPHYLLACEAE)

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ABSTRACT. The aim of the present work is to provide an account on the foliar epidermal morphological features of *Eremogone* species in Turkey, as a contribution to future research. Foliar epidermal morphology of five species of the *Eremogone* were investigated using Light Microscope (LM). Different morphological features including; epidermal cell shape, epidermal cell wall shape, size of epidermal cell, size of stomata and size of trichome were observed. The study revealed that the foliar epidermal morphology of the studied *Eremogone* species were in all cases diacytic and amphistomatic. The epidermal cell edges of *Eremogone* taxa with longitudinal cells are straight. *Eremogone acerosa*, *Eremogone acutisepala*, *Eremogone ledebouriana* characterized by unicellular trichomes, but these are not observed in other taxa of genus *Eremogone*. In all species, the stomatal density of the lower epidermis is higher than that of the upper epidermis. Our results show that the micro-morphology of foliar epidermal morphological characters play an important role in definition of taxa at species levels.
Keywords: *Eremogone, Caryophyllaceae, Foliar, Epidermis Morphology, Light Microscopy*

INTRODUCTION

The genus *Eremogone* Fenzl is one of the members of Caryophyllaceae native to Türkiye. The genus *Eremogone* consists of 20 taxa with 2 subspecies in Turkey, 13 of which are endemic [1, 2]

Secondary metabolites and various compounds obtained from Caryophyllaceae family plants have anti-cancer, antimicrobial, antioxidant, antibacterial, antifungal and antiseptic effects, as well as therapeutic properties such as cytotoxic, analgesic and antipyretic [3–6]. They are also used as ornamental plants and for defence against insects [7].

The family Caryophyllaceae Juss. has been widely studied by several researchers based on morphology [8–15], pollen morphology [11, 16–26], seed morphology [27–36], molecular systematics [37–44]; however, leaf epidermal morphology studies of Caryophyllaceae family taxa are limited [45–47]. There is no article on leaf epidermis anatomy of *Eremogone* species.

Very few published studies involving members of the family Caryophyllaceae deal with the morphological characteristics of the leaf epidermis and their systematic

relationships within the same genus. Taxonomically, the genus is known to have of synonyms and a number of the species are considered to be data incomplete hence, the identification of the species is problematic [48].

Eremogone acerosa (Boiss. & Heldr.) Ikonn., *Eremogone acutisepala* (Hausskn. ex F.Williams) Ikonn., *Eremogone gypsophiloides* (L.) Fenzl, *Eremogone ledebouriana* (Fenzl) Ikonn., *Eremogone pseudacantholimon* (Bornm.) Holub studied for systematic characterization with their leaf epidermal anatomical characters. This study aims to contribute to multidisciplinary studies by analysing the leaf epidermal morphology of *Eremogone* taxa in detail.

MATERIALS AND METHODS

For the present study, samples of five taxa of *Eremogone* were collected at various times from different parts of Turkey by one of the authors (M. Armağan). A list of taxa and full collector information are provided in Table 1.

Plant samples collected to examine the leaf epidermis surface were followed with modifications to the protocols of Clarke [49] and Nazir [50]. The dried leaves were kept in water to soften them. The leaves were placed in a tube filled with 70% lactic acid and nitric acid 30% kept hot in boiling water bath (Model, Nüve BM30) for about 50-60 min. Transparent leaf pieces be poured into separate petri dishes and washed several times with water to remove chemicals. The abaxial and adaxial surfaces of the washed transparent leaf pieces were gently separated with the help of a clean needle and brush, and the abaxial and adaxial surfaces were placed on separate slides. 1 drop of lactic acid was dropped on them and they were covered with a coverslip so that no bubbles remained. The prepared samples were left to dry by applying nail polish to the coverslip edges to become a permanent preparation.

The prepared slides were studied using Ivymen System Microscope. Leaf epidermis characters were then photographed using Borox Digital Camera. Under microscopy the following characters were observed epidermal cell length, epidermal cell width, stomata length, stomata width, number of stomata and epidermal cell for stomatal indexes, and trichome length, trichome base width. The qualitative and quantitative characters are summarized in (Tables 2 and 3), respectively. The leaf epidermis cells were examined in detail and microphotographs were recorded. The length and width of epidermal cells, stomata and trichomes were measured at least 20-30 times for statistical analyses. The mean, minimum, maximum and standard error values of the numerical data were recorded using Paleontological Statistics (PAST 4.03) software programme. The number of epidermal cells and stomata per unit area were determined and stomatal index was calculated according to the formula of [51] was used.

$$\text{Stomatal index} = \frac{\text{Number of stomata per unit area}}{\text{Number of stomata per unit area} + \text{number of epidermal cells per unit area}} \text{ x } 100$$

Eqn. 1

Table 1. *List of the species investigated, with location*

Taxon Name	Localities	Altitude (m)	Collection Date	Habitat	Phyto geographic Region
E. acerosa	Konya: Halkapınar, northwest of Aydos	2500	8.07 2022	Step	------- Endemic
E. acutisepala	Erzurum: Aşkale, Kop mountain pass	2055	24.07 2022	Stony (lime/limestone) slopes steppe	Irano-Turanian Endemic
E. gypsophiloides	Tunceli: Pülümür, East of Kırklar	2320	30.06 2022	Grassland pasture	Irano-Turanian
E. ledebouriana	Niğde: Ulukışla, between Darboğaz and Madenköy	1855	7.07 2022	Limestone gravelly steppe	------- Endemic
E. pseudacantholimon	Tunceli: between Hiver	2140	9.08 2022	Serpantine steppe	İrano – Turanian

RESULTS

Qualitative and quantitative results were obtained in the study in which five species of the *Eremogone* genus were examined. Qualitative results are detailed in Table (2-3), while microphotographs are provided in Fig. 1-2.

Although the epidermal cell lengths and widths of the analysed taxa are different, their shapes are the same in all taxa. The epidermal cell edges of *Eremogone* taxa with longitudinal cells are straight. The largest cell lengthwise observed on upper epidermis *Eremogone gypsophiloides* (190-570=349,7 ± 20,5 μm) and width in *Eremogone pseudacantholimon* (17–25,6=21,1± 0,5 μm). The largest cell lengthwise observed on lower *epidermis Eremogone gypsophiloides* (178-458=300±12 μm) and width in *Eremogone acutisepala* (17-49=28,3 ±2,42 μm). The shortest cell lengthwise observed on upper epidermis *Eremogone pseudacantholimon* (126-396=233,9 ±13 μm) and width in *Eremogone ledebouriana* (15-21=18,7± 0,5 μm). The shortest cell lengthwise observed on lower epidermis *Eremogone acerosa* (79-234=154,6 7 ± μm) and width in *Eremogone ledebouriana* (13-22=17,5± 0,5 μm).

Stomata were observed on the lower and upper epidermis surfaces of all examined taxa. It has been determined that five species have diacytic stoma type.The largest stomatal length (28-37=33.8 0.8 μm-29-37=33 0.5μm) and width (25.3-35.6=32.3 0.7μm-27-37032 0.8μm) in the upper and lower epidermis, respectively, were examined in *E. pseudacantholimon.* Angular trichomes were present in *E. acerosa, E. acutisepala and*

E. ledebouriana, while no trichome structure was observed in *E. gypsophiloides and E. pseudacantholimon*. According to stomatal index calculation, stomatal density of the lower epidermis was higher than the upper epidermis in all species.

Table 2. *Quantitative foliar epidermal features within the genus Eremogone species*

Taxon			**Epidermal Cell Length (µm)**	**Epidermal Cell Width (µm)**	**Stomata Cell Length (µm)**	**Stomata Cell Width (µm)**	**Stomatal İndex**
Eremogone acerosa	Upper Epidermal Cell	Min.	170	15		16	
		Max.	331	28	34	29	
		Mean	247,1	20,2	27,1	21	25
		S. E.	4,7	0,5	0,1	1,4	
	Lower Epidermal Cell	Min.	79	17	22	21	
		Max.	234	31	34	28	
		Mean	154,5	23,7	29,1	24,1	30,7
		S. E.	7	0,7	1,23	0,8	
Eremogone acutisepala	Upper Epidermal Cell	Min.	140	14	24	19	
		Max.	380	27	32	30	
		Mean	264	20,7	28,4	26	19
		S. E.	2,7	0,7	0,4	0,6	
	Lower Epidermal Cell	Min.	81	17	26,8	21	
		Max.	274	49	34	28	
		Mean	155	28,3	30	23,5	32,6
		S. E.	6,4	2,42	0,6	0,5	
Eremogone gypsophiloides	Upper Epidermal Cell	Min.	190	15,9	30,5	19	
		Max.	570	25	37	23,3	
		Mean	349,7	19,5	33,8	21,7	28,5
		S. E.	20,5	0,55	0,4	0,2	
	Lower Epidermal Cell	Min.	178	15	20,7	21	
		Max.	458	24	31	27	
		Mean	300	19,5	25,4	24	39
		S. E.	12	0,7	0,4	2,4	
Eremogone ledebouriana	Upper Epidermal Cell	Min.	110	15	29	20	
		Max.	396	21	38	27	
		Mean	267	18,7	33	22,5	28,5
		S. E.	13	0,5	0,53	0,5	
	Lower Epidermal Cell	Min.	151	13	24	18	
		Max.	416	22	35	26	
		Mean	280	17,5	28,6	22,6	33,8
		S. E.	16	0,5	0,8	0,5	
Eremogone pseudacantholimon	Upper Epidermal Cell	Min.	126	17	28	25,3	
		Max.	396	25,6	37	35,6	
		Mean	233,9	21,1	33,8	32,3	30
		S. E.	13	0,5	0,8	0,7	
	Lower Epidermal Cell	Min.	113	15	29	27	
		Max.	258	29	37	37	
		Mean	170	21	33	32	37
		S. E.	7	1,1	0,5	0,8	

Table 3. *Quantitative data for foliar trichomes in Eremogone species*

		Trichome Length (µm)	**Trichome Base Width (µm)**
Eremogone acerosa	Min.	14	11
	Max.	64	41
	Mean	52	30,4
	S. E.	2,1	1,45
Eremogone acutisepala	Min.	66	30
	Max.	78	37
	Mean	72,4	32,6
	S. E.	1,19	0,6
Eremogone gypsophiloides	Min.	---	---
	Max.	---	---
	Mean	---	---
	S. E.	---	---
Eremogone ledebouriana	Min.	61	29
	Max.	114	50
	Mean	81	39
	S. E.	5,56	2,2
Eremogone pseudacantholimon	Min.	---	---
	Max.	---	---
	Mean	---	---
	S. E.	---	---

Leaf epidermis hair structure is important for the identification of Caryophyllaceae taxa at species level. Trichomes were observed on the margins of the leaf epidermis in three species, but not in the others.

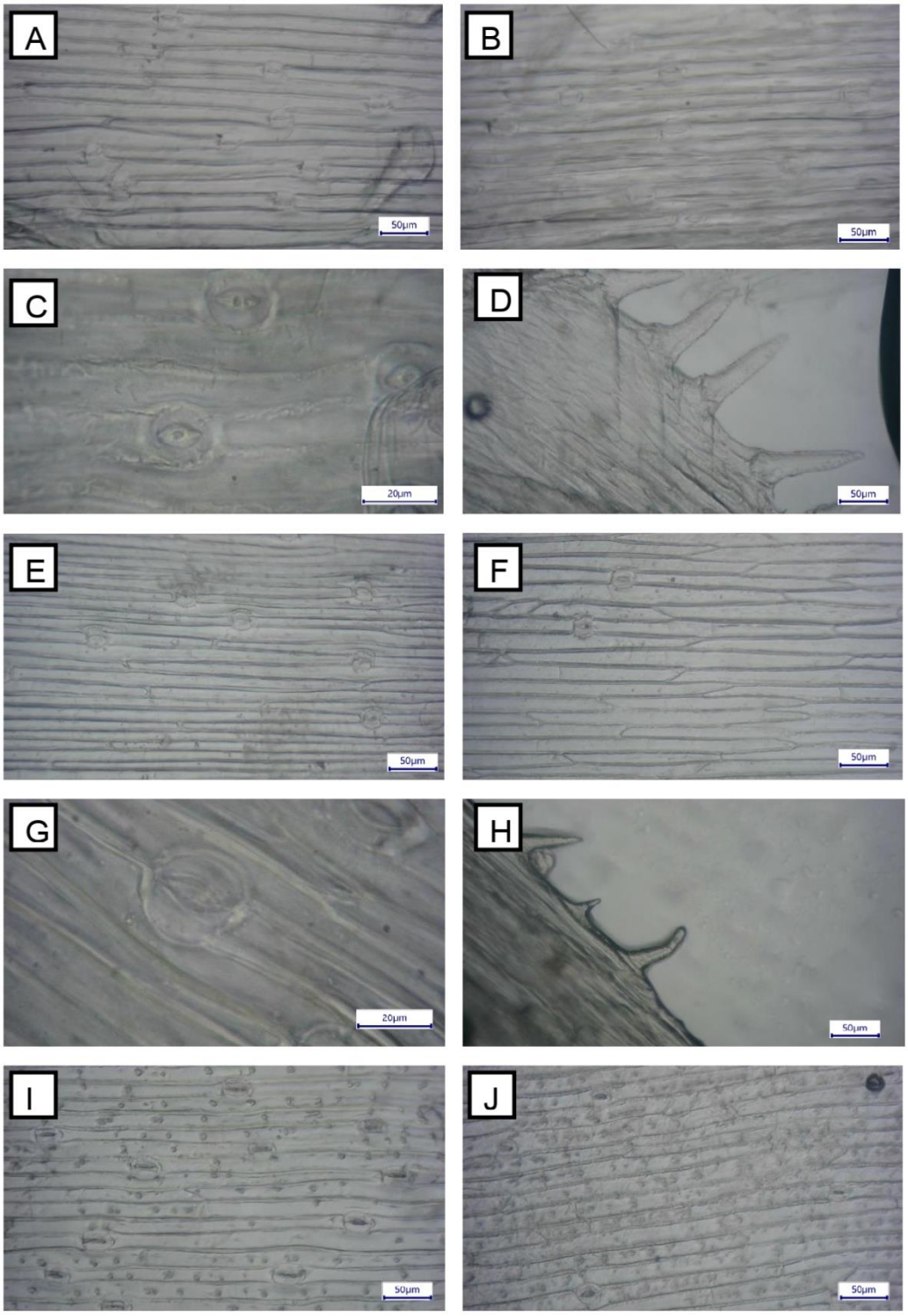


Fig. 1. *Light micrographs of shape of epidermal cell, stomata and trichome of Eremogone acerosa (A) Upper epidermal cell (B) Lower epidermal cell, (C) Stomata (D) Trichome, Eremogone acutisepala (E) Upper epidermal cell (F) Lower epidermal cell, (G) Stomata (H) Trichome, Eremogone gypsophiloides (I) Upper epidermal cell (J) Lower epidermal cell*

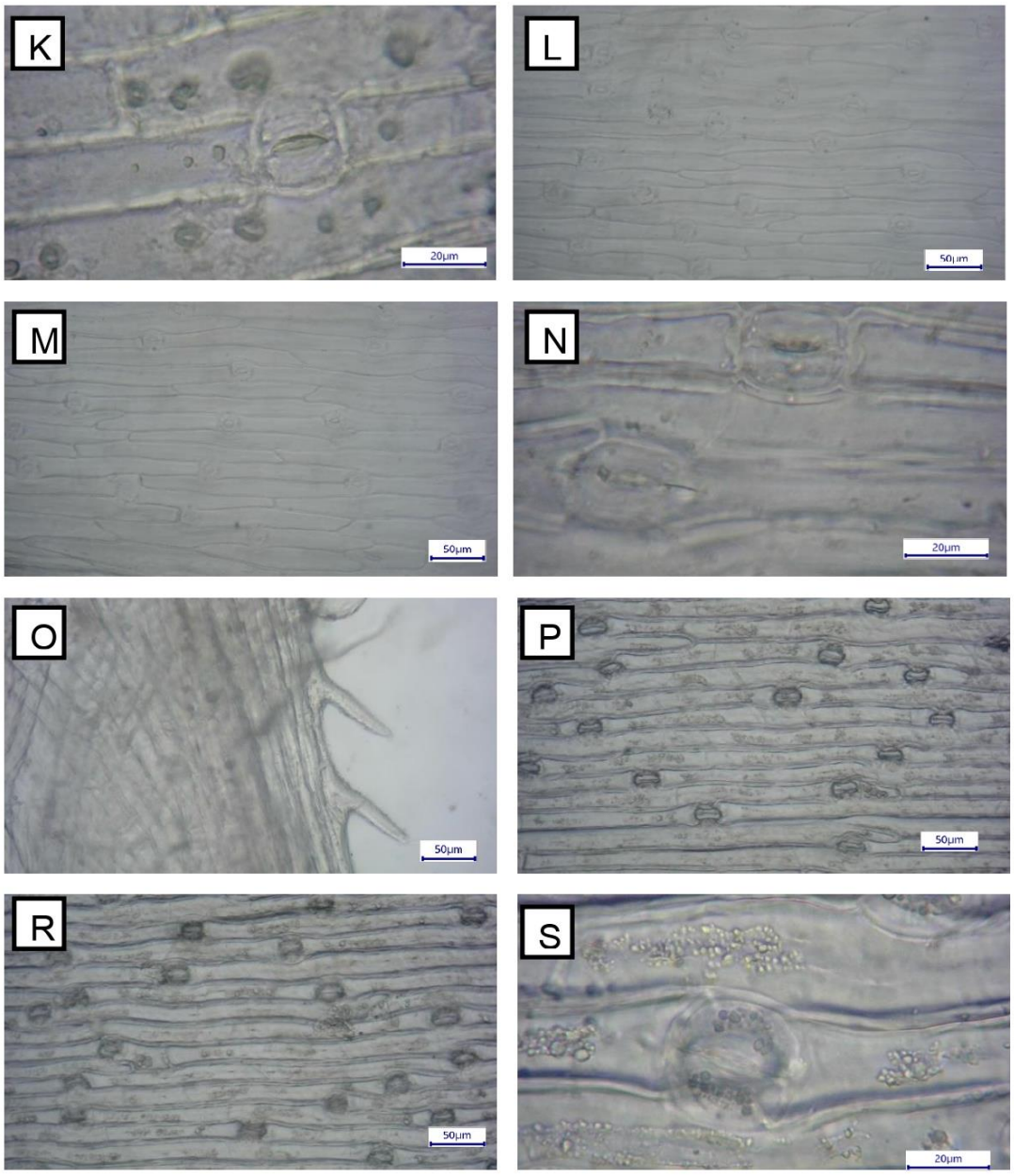


Fig. 2. *Light micrographs of shape of epidermal cell, stomata and trichome of Eremogone gypsophiloides (K) Stomata, Eremogone ledebouriana (L) Upper epidermal cell (M) Lower epidermal cell, (N) Stomata (O) Trichome, Eremogone pseudacantholimon (P) Upper epidermal cell (R) Lower epidermal cell, (S) Stomata*

DISCUSSION

Zaman and Park [52] investigated *Silene takesimensis* and reported that the epidermis surface has micro and macro hairs. The *Eremogone* species studied are amphistomatic, meaning that stomata are located on both the upper and lower surfaces of the leaves. This result is in agreement with studies [10, 47, 53].

Ullah et al. [26] reported 8 of 14 species has unicellular, multicellular and non-glandular trichomes, 12 species has diacytic stomata and two species, *Silene pseudo-*

verticellata and *Vaccaria hispanica*, had anomocytic stomata. All of the *Eremogone* species analysed by us had diacytic stomata, while trichomes were observed in three species.

In their study, Ullah et al. [47] examined the leaf epidermal anatomy of 18 species belonging to the Alsinoideae (Caryophyllaceae) subfamily using light microscopy (LM). Members of Alsinoideae consist of taxonomically complex genera such as *Arenaria*, *Cerastium*, *Minuartia*, *Sagina* and *Stellaria*. *Eremogone* consists of species that were previously subgenera of *Arenaria* and *Minuartia.* Ullah et al. observed the shape of the epidermal cells to be irregular, wavy or tetragonal, anticlinal cell wall as undulate, sinuate or straight. Also observed diacytic, paracytic, anomocytic and anisocytic stomata. Diacytic stomata were reported in a number of genera (*Arenaria*, *Lepyrodiclis*, *Minuartia*, *Sagina* and *Stellaria*). The epidermal cell walls of the *Eremogone* species examined have a straight shape, while all have diacytic stomata.

CONCLUSION

The genus *Eremogone*, dominated by species previously classified as subgenera of *Arenaria* and *Minuartia*, is taxonomically problematic. For this reason, many molecular and morphological studies have been done and continue to be done in the classification of the genus. This study, which analysed the leaf epidermis micromorphology of *Eremogone* taxa, consisted of measurements of epidermis cell length and width, stomatal length and width, stomatal density and trichome length and trichome base width. Anatomical characters are constant since they are not affected by environmental variability [54]. The results obtained can be used for morphological distinction of the genus.

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