

THE MORPHOLOGICAL AND ANATOMICAL INVESTIGATION OF TWO ENDEMIC *CROCUS* L. (IRIDACEAE) SPECIES OF TURKEY

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Abstract

The paper reports morphological and anatomical features of *Crocus asumaniae* Mathew and *Crocus mathewii* Kerndorff et Pasche, endangered plants. Transvers sections of plant parts stem, scape and leaf were investigated. Characteristically, style is dividing into 3 slender yellow to orange branches, flowering takes place in autumn and is hysteranthous. In anatomical studies, the walls of root endodermal cells of both species show thickenings completely and hairs are found on the abaxial epidermis of leaf. Presence of sclerenchymatic tissues around the vascular bundles of the leaf is a characteristic feature.

Introduction

Turkey has a rich plant diversity with a high ratio of endemism (Ozturk, 2008). Iridaceae family has got a lot of endemic species. This large and diverse family, which especially scattered in the southern hemisphere, contains about 80 genera. Due to its beautiful flowers, this family is planted in parks and gardens as ornaments (Baytop, 1984; Kandemir *et al.*, 2012). The family includes several economically important genera such as *Crocus*, *Iris* and *Gladiolus*. *Crocus* genus has got 126 taxa which are distributed in the worldwide (Anon., 2010). 79 taxa (and one hybrid) of these taxa show distribution in Turkey and 31 are endemics (Güner *et al.*, 2012). Among these endemic species *C. asumaniae* and *C. mathewii* have been recorded under the category of endangered (EN) species by IUCN due to limited distribution (Ekim *et al.*, 2000). There are a lot of works on different endemics in Turkey (Ozturk *et al.*, 1992; Celik *et al.*, 1995; Uysal *et al.*, 2011). In this study, morphological and anatomical characteristics of the two endangered species *C. asumaniae* and *C. mathewii* have been investigated as there has not been any detailed study on these *Crocus* species.

Materials and Methods

Both plant specimens were collected in the month of November 2014 from Alacadağ Mountain located in the Antalya province between Finike and Kaş, in the South-West part of Turkey. Herbarium samples of *C. asumaniae* and *C. mathewii* are conserved in the herbarium of CBU, Botanical Research Laboratory. Morphological illustrations of the plant samples were made from freshly collected plants and morphologic measurements on root, stem and leaf of the same plant samples were performed. For anatomical investigation plant specimens were fixed in 70% ethanol and paraffin infiltrated tissues (Algan, 1981). The sections were also taken by hand and stained with safranin reactive. The slide preparations were photographed with motorized microscope in Celal Bayar University, Botanical Research Laboratory. Anatomical measurements were made by using ocular micrometer. Minimum, maximum, mean values were recorded and standard deviations calculated (Table 1).

Results

Morphological findings

***C. mathewii*:** The corm tunic is reticulate-fibrous. Leaves 4-10, developed at flowering time, 1-2 mm wide, grey-green, have hairs are constricted, arranged in two rows of abaxial shape. Prophyll present. Bracteole exist. Throat of perianth violet, segments 2.5-3 x 1-1.5 cm, the inner vaguely shorter and narrower than the outer, ovate to obovate, white or very pale lilac-blue. Filaments 3-4 mm long, white; anthers about 15 mm long, yellow. Style divided into 3 orange-red branches, each branch 7-10 mm long, less than half as long as the perianth segments, usually exceeding but sometimes equalling (Fig. 1A, C)

***C. asumaniae*:** The corm tunic is fibrous, fibres slender and slightly reticulate. Leaves 6, with tips just visible at anthesis, 0.5-1 mm broad. Prophyll is present. Bracteole is present and smaller than bract. Flowers generally whitish. Perianth segments are 2.2-3 cm length and 0.6-1 cm wide, acute, whitish or slightly lilac. Throat of perianth whitish and glabrous. Come as to male organs; filaments are 4-6 mm, white or pale yellow, glabrous; anthers 1.1-1.7 cm, yellow. Style dividing into 3 orange-red branches, each 1.3-1.8 cm and as long as perianth segments (Fig. 1B, D).

Anatomical findings

***C. mathewii*: Root:** At the top of the cross-section of the root has a single layered epidermis which is composed of irregular shaped cells. Exodermis which is located below the epidermis is arranged in a single row. Cortex formed 6-7 cell layers, 15-32.5µ diameter. The wall thickenings of the endodermal cells are thick. Under the endodermis a single layered pericycle is observed. There are a large metaxylem and 3 or 4 xylem strands are existent in the vascular tissue. (Fig. 2A, B).

Scape: Shape of scape is star like. Outer part of scape is covered by thick cuticle. Epidermis consisting of isodiametric cells is single-layered, 10-25 µ. Cortex is composed of ovoidal parenchymatous cells which are 20-55 µ in diameter and has thin wall. Vascular bundles are located both in periphery and central part of stem. Two big vascular bundles are located in the pith of *C. mathewii*, other medium-sized bundles are surrounding in the form of a ring in the central part of the aerial stem and other small vascular bundles in the peripheral part (Fig. 2C, D, Table 1).

Table 1. Anatomical measurements of *C. asumaniae* and *C. mathevii*.

	Width (μ)					Length (μ)						
	Min.-Max.		Mean \pm S.D			Min.-Max.		Mean \pm S.D				
<i>C. asumaniae</i>												
Root												
Epidermis cell	10.00	-	25.00	16.87	\pm	5.30	7.50	-	17.50	11.56	\pm	3.25
Cortex cell (Diam.)	25.00	-	50.00	10.92	\pm	8.57						
Trachea (Diam.)	5.00	-	15.00	9.92	\pm	3.38						
Scape												
Epidermis cell	10.00	-	20.00	15.31	\pm	3.64	10.00	-	20.00	15.62	\pm	3.47
Cortex cell (Diam.)	12.50	-	42.50	25.97	\pm	8.23						
Trachea (Diam.)	7.50	-	17.50	12.50	\pm	3.27						
Leaf												
Adaxial Cuticle	2.50	-	5.00	3.21	\pm	0.98						
Adaxial Epidermis	12.50	-	17.50	15.83	\pm	2.58	10.00	-	15.00	12.50	\pm	2.23
Abaxial Epidermis	10.00	-	17.50	12.22	\pm	2.31	7.50	-	15.00	10.55	\pm	2.42
Palisade parenchyma	25.00	-	52.50	41.56	\pm	6.79	10.00	-	17.50	13.43	\pm	2.29
Spongy parenchyma	20.00	-	57.50	36.73	\pm	8.47						
Trachea	5.00	-	12.50	8.61	\pm	2.29						
Abaxial cuticle	2.50	-	5.00	3.90	\pm	1.06						
<i>C. mathevii</i>												
Root												
Epidermis cell	12.50	-	25.00	17.27	\pm	4.10	12.50	-	22.50	15.68	\pm	3.18
Cortex cell (Diam.)	15.00	-	32.50	23.38	\pm	5.29						
Trachea (Diam.)	10.00	-	17.50	14.06	\pm	2.65						
Scape												
Epidermis cell	10.00	-	25.00	18.75	\pm	5.72	12.50	-	22.50	19.31	\pm	4.34
Cortex cell (Diam.)	20.00	-	55.00	35.40	\pm	6.63						
Trachea (Diam.)	7.50	-	20.00	13.36	\pm	3.72						
Leaf												
Adaxial Cuticle	1.25	-	3.75	2.39	\pm	0.83						
Adaxial Epidermis	11.25	-	22.50	16.25	\pm	3.49	15.00	-	23.75	18.06	\pm	3.27
Abaxial Epidermis	10.00	-	16.25	13.40	\pm	2.62	15.00	-	21.25	18.29	\pm	2.11
Palisade parenchyma	17.50	-	37.50	26.56	\pm	6.43	10.00	-	13.75	11.79	\pm	1.88
Spongy parenchyma	11.87	-	22.50	15.89	\pm	3.41						
Trachea	7.50	-	10.00	8.95	\pm	1.22						
Abaxial cuticle	2.50	-	4.37	3.62	\pm	1.11						

S.D.: Standard Deviation



Fig. 1. The general appearance of *C. mathewii* (A-C) and *C. asumaniae* (B-D), in the natural habitat; a: corm, b: scape, c: flower, d: corm tunic, e: stamen, f: leaves, g: style, h: tepal. Scale bars: 1 cm.

Leaf: There is a nearly rectangular keel in the middle of the leaf which has two lateral arms, with their margins recurved towards the keel. Pale stripe running axially along the centre of the leaf is the characteristic feature. Abaxial and adaxial cuticle have thickened, abaxial cuticle thicker than adaxial cuticle (Table 1). Palisade and spongy parenchyma are slightly distinguished on the mesophyll of arms. Palisade parenchyma cells consisted

of 1 or 2 layered cells. Spongy parenchyma cells are positioned on abaxial surface. Vascular bundles are positioned in single line and the sclerenchymatic cells are existed at the phloem pole of vascular bundles (Table 1; Fig. 2E, F). Two big vascular bundles exist at both of keel corners and junctions of keel and arms. Hair like papillae are present on both ends abaxial surface of arms and at the keel corner (Fig. 2).

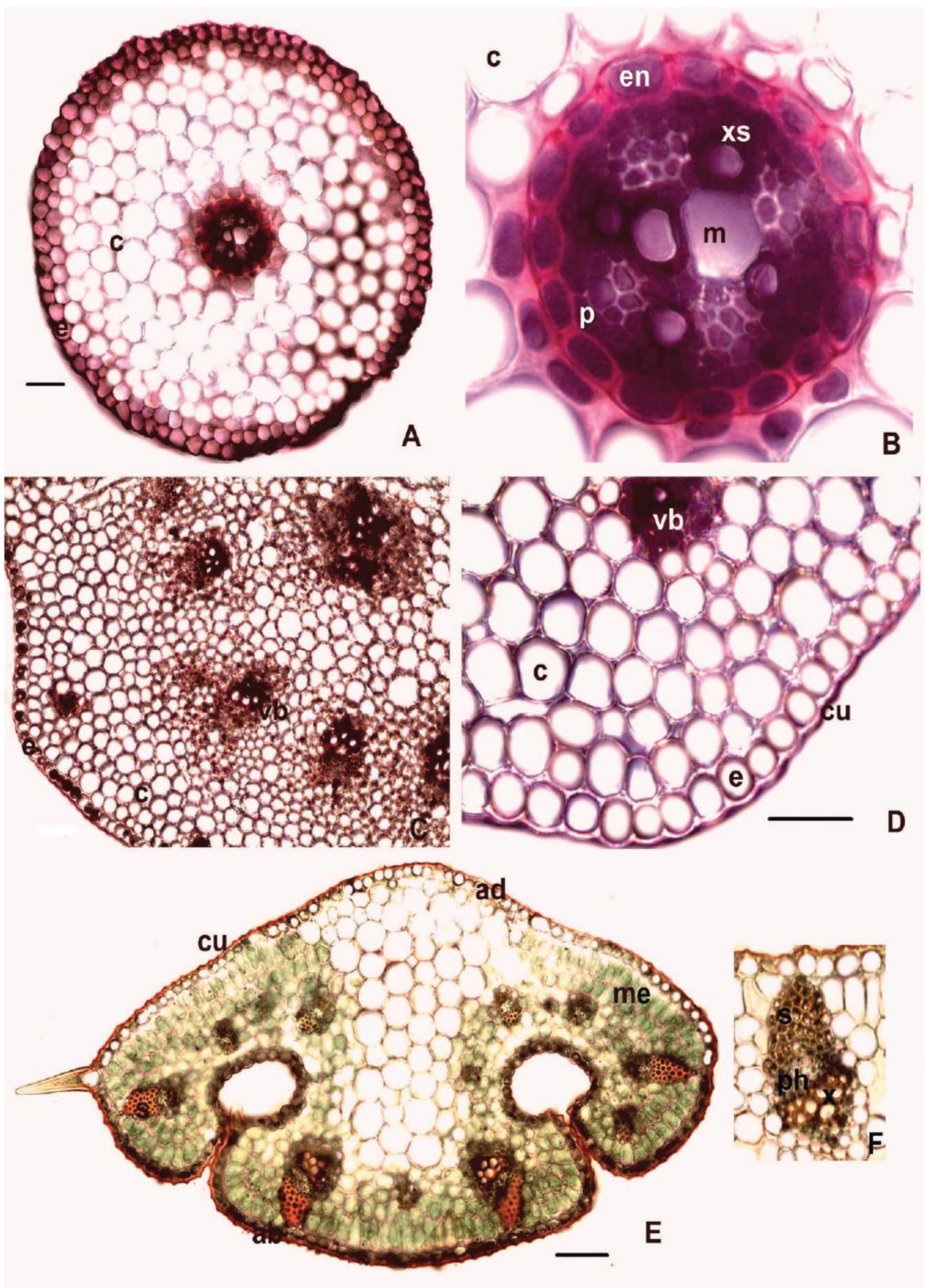


Fig. 2. Cross-section of root (A,B), scape (C,D), leaf (E,F) of *Casumania*. Scale bars: 50 μ .

ab: abaxial epidermis, ad: adaxial epidermis, cu: cuticle, c: cortex, e: epidermis, en: endodermis, m: metaxylem, me: mesophyll, p: pericycle, ph: phloem, s: sclerenchyma, x: xylem, xs: xylem strands.

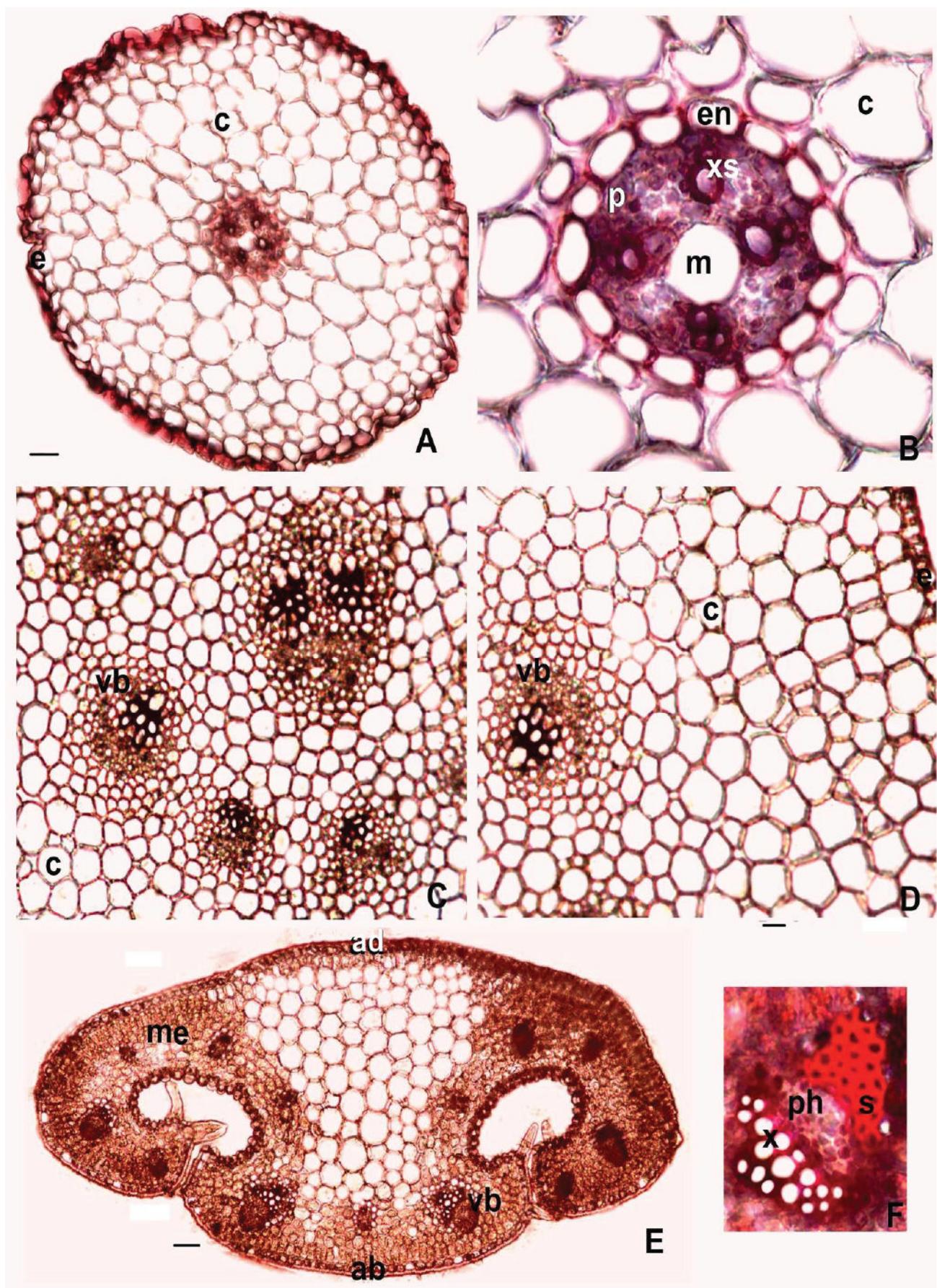


Fig. 3. Cross-section of root (A,B), scape (C,D), leaf (E,F) of *C. mathewii*. Scale bars: 25 μ .

ab: abaxial epidermis, ad: adaxial epidermis, cu: cuticle, c: cortex, e: epidermis, en: endodermis, m: metaxylem, me: mesophyll, p: pericycle, ph: phloem, s: sclerenchyma, x: xylem, xs: xylem strand, vb: vascular bundle.

C. asumaniae

Root: Also in these species, root epidermis can be seen single layered, cells generally oval-shaped and thinly walled, $10,5-25 \times 7-17,5 \mu$. Similarly, exodermis tissue which is seen under the epidermis, is a one layered too. The cortex is 5-6 layered, oval-shaped, parenchymatic with intercellular spaces. Cells $25-50 \mu$ in diameter. Endodermal cells with wall thickenings. Pericycle cells thin-walled. There is a single metaxylem on the median part of vascular cylinder and xylem strands are surrounded of the vascular cylinder (Fig. 3A, B).

Scape: Form of scape is polygonal. Exterior surface of the scape is coated with thick cuticle. Epidermis is one-layered, $10-20 \mu$. There is multi-layered cortex below the epidermis. Cells of cortex are thin walled, parenchymatous and has intercellular spaces. Vascular bundles are numerous and more scattered. But at the edges bundles are smaller than centers (Fig. 3C, D).

Leaf: Leaves of *C. asumaniae* have a median rectangular keel and two lateral arms, with their margins recurved towards the keel. Abaxial and adaxial cuticle are very thick, $2,5-5 \mu$ in size. *C. asumaniae* leaves are bifacial. Palisade parenchyma cells are usually two layered. Spongy parenchyma cells are generally located under palisade parenchyma at abaxial surface. Major bundles are occurring at the corners of keel and towards the arm margin. Hairs are present on adaxial surface of arms (Fig. 3E, F).

Discussion

Morphologically corm tunic, color of perianth and style-branch are detailed as characteristic features among bulbous plant species. Some morphological characters of many *Crocus* species have not been given by researchers (Davis, 1984; Güner *et al.*, 2000), such as the presence of hairs on leaves. Characteristically, hairs were observed to be more intense in *C. mathewii*. In this study, the detailed anatomical examinations were given. Therefore, this examination supplies the first detailed description of *C. asumaniae* and *C. mathewii* and a comparison with some other investigated members of *Crocus* genus have been presented. Exodermis is present in both of species. Likewise Akan *et al.*, (2007) have observed exodermis in *C. leichtlinii* (D. Dewar) Bowles. Both species' root endodermal cell wall is thick. This property has also been mentioned in the cross-section of root of *C. chrysanthus* (Herbert) Herbert, *C. fleischeri* Gay, *C. leichtlinii* (D. Dewar) Bowles (Özdemir *et al.*, 2004, 2013; Akan *et al.*, 2007). This thickening is distinctive feature for *Crocus* species. There is a single metaxylem in the centre of the vascular cylinder of both species. Özdemir *et al.*, (2004) have observed same features in the transverse sections of *C. danfordiae* Maw. and *C. fleischeri*.

In the transverse sections of scape, vascular bundles are dispersed in the peripheral and median parts. Some researchers have observed same features in *C. flavus* Weston subsp. *flavus*, *C. pallasii* Goldb. subsp. *turcicus* B. Mathew and *C. cancellatus* Herbert subsp. *damascenus*

(Herbert) B. Mathew (Akan & Eker, 2004; Özdemir *et al.*, 2006). But it was stated that vascular bundles are placed as one rings in the median parts of stem of *C. pulchellus* Herbert and *C. aeriis* Herbert (Özyurt, 1978; Özdemir & Akyol, 2005).

Rudall & Mathew (1990) have defined the leaf anatomy of some species and they have noticed that the leaves of majority of species have a distinguished cross-sectional outline with a median square or rectangular keel and two lateral arms. It is possible that these features are taxonomical important. In cross section of leaves of *C. asumaniae* and *C. mathewii* contain a median rectangular keel and two lateral arms with their margins recurved towards the keel. Leaves are bifacial. According to Rudall & Mathew (1990), *Crocus* leaf is morphologically more homologous with the unifacial than bifacial leaves of most other Iridaceae. Hair like papillae are usually situated on abaxial sides of arm ends and corner of keel of *C. mathewii*, same feature has been observed in *C. graveolens* Boiss. and Reuter (Rudall & Mathew, 1990). But *C. asumaniae* has hairs on adaxial surface of arm. Hair-like papillae are present in *C. olivieri* Gay subsp. *istanbulensis* Mathew, *C. candidus* E. D. Clarke, *C. pallasii* Goldb. subsp. *pallasii*, *C. cancellatus* Herbert subsp. *mazziaricus* (Herbert) Mathew, and *C. pulchellus* (Satil & Selvi, 2007). Vascular bundles are collateral type in both species. The xylem is head for the adaxial surface and the phloem head for the abaxial surface. The phloem poles of larger bundles and smaller bundles have also sclerenchymatous cells as caps. As a result, These anatomical features can be used as taxonomical characters in the identifying the species.

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