COMPARATIVE VEGETATIVE ANATOMY AND FRUIT MICROMORPHOLOGY OF THE GENUS KUNDMANNIA SCOP. AND THEIR TAXONOMICAL SIGNIFICANCE

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Abstract

Kundmannia Scop. is a genus of the Apiaceae (Umbelliferae) family which consists of 463 genera and ca. 3500 species. Kundmannia belongs to the Oenantheae tribe and are well represented in Spain, France, Italy, Turkey, Syria and North Africa where they grow on calcareous or siliceous soils. In this study; fruit micromorphology and the vegetative anatomy of the Kundmannia species growing in Turkey were compratively examined for the first time. The anatomical sections were taken from the stem, ray, leaf and and petiole of the plants for taxonomic deduction. Various tissues of taxa were given comparative microanatomical measurements according to the anatomical studies, lignified or non-lignified medullary rays in stems, stomata types (diacytic and anomocytic), stomata index ratio (0.58 - 0.7), fruit shape and fruit epidermal surface are considered as important anatomical characters. In the fruit micromorphology, the cells on the mericarp surface have seen as striate and rugulate while the surface between the two ribs in the species have observed rugulate or striate. Fruit micromorphology and vegetative anatomy of the genus Kundmannia has not been studied before. However, the anatomical structure of the genus has been elucidated in this study, for the first time.

Introduction

The family Apiaceae (Umbelliferae) is the third largest family in terms of genera in Turkish Flora. It is also the eighth largest family with approximately 455 species, and 33% of these are endemic (Davis *et al.* 1988, Guner *et al.* 2000, Uruşak and Kızılarslan 2013). The genus *Kundmannia* Scop. is represented by five species which are distributed in Spain, France, Italy, Turkey, Syria and North Africa. This genus distributing in the flora of Turkey is described by the study of *K. syriaca* Boiss. and *K. anatolica* Hub.-Mor. and these are endemic to Turkey.

Apiaceae species have specific odours because they have secretory cavities (vittae), which are schizogenous oil ducts with resin, oil, or mucilage. They are found in the roots, petioles, stems, leaves, and fruits (Metcalfe and Chalk 1979, Pluinkett *et al.* 2014). In the family Apiaceae, morphological and anatomical fruit characteristics and the number of vittae have reliable diagnostic importance (Uruşak and Kızılarslan 2013). Morphological and anatomical studies have increased over the last few years (Selvi *et al.* 2013, Bakiş and Babaç 2014, Bayırlı *et al.* 2014, Ozcan and Eminagaoglu 2014, Satil *et al.* 2015, Çavuşoğlu and Karaferyeli 2015). However studies on the vegetative anatomy and fruit micromorphology of this genus have not been found. Metcalfe and Chalk (1950) and Watson and Dallwitz (2000) explained the anatomical characteristic properties of the family Apiaceae.

The purpose of this study is to give a detailed account of the micromorphological (fruit) and anatomical features (stem, ray and lamina) of the *Kundmannia* species growing in Turkey. It is expected that it will provide an important information to the taxonomic studies as monograph and will contribute for further investigation in future.

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Materials and Methods

Plant specimens were collected from their type localities. Information about the plant material is given in Table 1. Species collected were examined in the laboratory and were investigated using the relevant literature (Hedge and Lamond 1972, Davis *et al.* 1988). The specimens were dried according to standard herbarium techniques and stored in the Tunceli University Herbarium, Turkey.

Table 1. Collection details of specimens used for micromorphological and anatomical studies.

Species	Collection areas and collector's number
Kundmannia syriaca	Adana, between Feke and Himmetli, Göksu passage, rocky cracks, 750 m, 10.06.2012, Paksoy 1088.
Kundmannia anatolica	Antalya, Akseki, 2-3 km before Güneykaya village, rocky cracks, 640 m, 04.06.2012, Paksoy 1080.

Anatomical studies were carried out on specimens preserved in 70% alcohol. Cross sections of stem and leaves (lamina and petiole) were stained with phloroglucinol - HCl solutions (Yakar-Tan 1982) and chlorophyll in leaves was removed with chloral hydrate. Stomatal density on abaxial and adaxial surfaces of the leaf was counted under a light microscope (LM). Stomatal index was calculated according to the method of Meidner and Mansfield (1968). Stomatal terminology and the leaf epidermal terminology were based on the classification proposed by Dilcher (1974) and Wilkinson (1979), respectively. Measurements and photographs were taken using Olympus BX 51 and Nikon Eclipse E200 binocular LM. Fruit micromorphology is studied by Tabletop scanning electron microscopy (SEM). For SEM, fruits were fixed on aluminum stubs using double-sided adhesive. The SEM micrographs were taken in a NeoScope JCM–5000 at an accelerating voltage of 10 kV.

Results and Discussion

Comparative anatomical characters of stem, ray and leaf and fruit micromorphology of *Kundmannia* species are given in Tables 2, 3, 4 and 5, respectively. Moreover, comparative leaf epidermal surface among the taxa are illustrated on Table 6, and biometric measures of the taxa are illustrated on Table 7.

Table 2. Comparative stem anatomy of Kundmannia species.

	Surface		Co	ortex		Inter	Medullary	
Taxa	of stem	Collen-	Parenchyma	Number of sec	cretory canal	fascicular	rays	Phloem
		chyma	(Chlorenchyma)	Collenchyma	Parenchyma	region		
K. syriaca	Straight	1 - 8	1 - 2	Present	Present	2 - 4 row	Lignified	2 - 5
K. anatolica	Straight	6 - 9	2 - 4	Absent	Present	3 - 4 row	Unlignified	3 - 5

General anatomical characters: Stem anatomy: In transverse section of the stem, the outermost single-layered epidermis is covered by a thin layer of cuticle (Fig. 1). The stem is glabrous, roundish and straight. The cortex consists of collenchyma, parenchyma and endodermis. Underneath the epidermis is arranged alongside collenchyma and parenchyma. Collenchyma is 3 - 9 layered while parenchyma is 2 - 5 layered and chlorenchymatic. Secretory canals of various sizes are observed within the cortex. Endodermis is unclear. Vascular bundles were arranged in a ring. Interfascicular region was sclerenchymatic with 8 - 12 rows. The xylem portions of the

bundles were embedded in pith. Pith cells were parenchymatic and polygonal or orbicular in shape. The number of vascular bundles arranged in a ring are 21 - 25. Interfascicular region was located in between vascular bundles. Phloem was 2 - 5 layers and consisted of irregular and squashed cells. Cambium cells were not clear. Pith region occurred in the center of the stem composed of large orbicular or polygonal parenchymatic cells (Table 2).

Table 3. Comparative ray anatomy of Kundmannia species.

	Surface		Corte	X		Inter	Medullary	
Taxa	of stem	Collen- chyma	Parenchyma (Chloren-		ber of ry canal	fascicular region	rays	Phloem
			chyma)	Collen- chyma	Paren- chyma	•		
K. syriaca	Straight	1 -8	1 - 2	Present	Present	2 - 4 row	Lignified	2 - 5
K. anatolica	Straight	6 - 9	2 - 4	Absent	Present	3 - 4 row	Lignified	3 - 5

Table 4. Comparative lamina anatomy of Kundmannia species.

	Meso-	Palisade	Spongy			Mid	dle vasc	ular bundl	le	
Taxa	phyll	layer	layer	Coller	nchyma	Paren	chyma	Bundle	Xylem	Phloem
	type			Ad.	Ab.	Ad.	Ab.	sheat	fibers	fibers
K. Syriaca	Bifacial	2	2 - 4	1 - 2	2 - 3	2 - 4	2 - 5	Absent	Absent	Present
K. Anatolica	Bifacial	2	3 - 6	1 - 2	1 - 2	3 - 5	5 - 7	Absent	Present	Present

Table 5. Comparative petiole anatomy of Kundmannia species.

Taxa	Number of	Adaxial	Abaxial	Number of	Secretory
	vascular bundle	surface	surface	collenchyma cluster	canals
K. syriaca	10	Straight	Convex	11 - 15	Present
K. anatolica	9	Straight	Convex	13 - 17	Present

Table 6. Characteristics of the leaf epidermis of Kundmannia species under LM.

Characters	К.	syriaca	К. а	natolica
Characters	Adaxial	Abaxial	Adaxial	Abaxial
Anticlinal cell wall	Straight	Undulate	Straight	Undulate
Stomata type	Diacytic anomocytic	Diacytic anomocytic	Diacytic anomocytic	Diacytic anomocytic
Stomata length (μ)	24.9 ± 2.1	29.9 ± 2.1	28.4 ± 2.0	31.4 ± 1.7
Stomata width (μ)	18.5 ± 2.4	22.5 ± 3.9	15.1 ± 2.0	18.9 ± 1.3
Number of stomata (1 mm ²)	59 ± 17	100 ± 18	106 ± 16	167 ± 18
Number of epidermis cells	353 ± 17	390 ± 28	406 ± 27	300 ± 19
Stomata index	14.3	20.4	20.7	35.8
Stomata index ratio		0.7	1	0.58

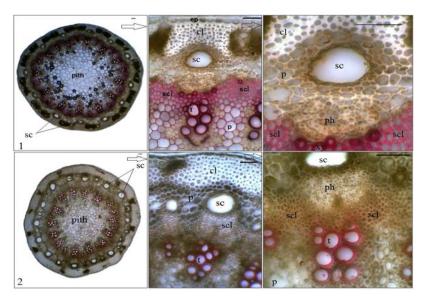


Fig. 1. Stem anatomy of *Kundmannia* species. 1. *K. syriaca*, 2. *K. anatolica*. ep: epidermis, cl: collenchyma, p: parenchyma, sc: secretory canal, ph: phloem, t: trachea, scl: sclerenchyma (Scale bar 50 μm).

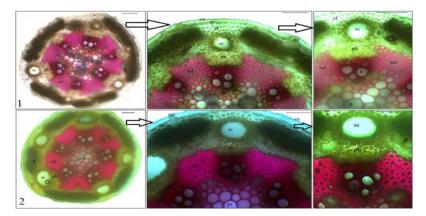


Fig. 2. Ray anatomy of Kundmannia species. 1. K. syriaca, 2. K. anatolica (Scale bar 50 μm).

Ray Anatomy: In transverse section of the ray, the outermost single-layered epidermis is covered by a thin layer of cuticle (Fig. 2). The stem is glabrous, roundish and straight. Epidermis layer contains epistomatic type stomata cells. The cortex consists of 2 - 4 layered collenchyma, and 4 - 7 layered chlorenchyma. Secretory canals are embedded into cortex. Endodermis layer is unclear. Vascular bundles were 6 in number and arranged in a ring. İnterfascicular region was sclerenchymatic with 8 - 12 rows. The xylem portions of the bundles were embedded in pith. Pith cells were parenchymatic and polygonal or orbicular in shape. Interfascicular region was located in between vascular bundles. Phloem was 5 - 9 layered and consisted of irregular and squashed cells. Cambium cells were not clear. Pith region of the stem was composed of large orbicular or polygonal parenchymatic cells some of which were lignified (Table 3).

Lamina anatomy: The transverse section of the leaf shows a thin cuticle on the upper and lower epidermis (Fig.3). The epidermal cells are isodiametric and rectangular, oval or cuboidal in shape. These cells are rich in terms of crystals (druse type) (Fig. 4). Amphistomatic type of stomata are present on both surfaces of the leaf. In surface section, the epidermal cell walls are undulate or straight and stomata type is diacytic ranunculous (anomocytic) type (Fig. 4). The stomatal index is 14.3 - 20.7 on upper surface and 20.4 - 35.8 on lower surface. The stomatal index ratio is between 0.58 and 0.7. Mesophyll consists of 2 layered palisade and 2 - 6 layered spongy parenchyma cells. Drusen crystals found that epidermis cells are intensive whereas leaf mesophyll cells are sparse (Fig. 3, Tables 4, 6).

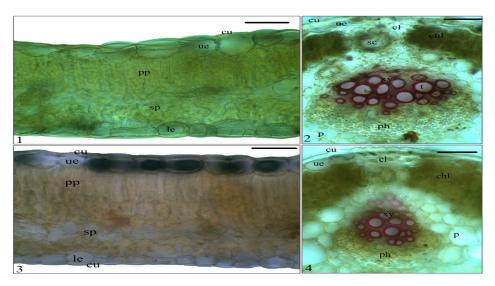


Fig. 3. Lamina anatomy of Kundmannia species. 1 - 2. K. syriaca, 3 - 4. K. anatolica (Scale bar 50 μm).

Petiole anatomy: A cross section of the petiole shows that the adaxial surface is slightly straight and abaxial surface is convex and rounded. The single layered epidermis is composed of oval and rectangular cells. Collenchyma clusters are situated under epidermis. There are 2 - 4 layered chlorenchyma cells between the clusters. Petiole is composed of hexagonal or polygonal parenchymatic cells. which includes numerous secretory canals but without crystals. Crescent shaped vascular bundles are found in the centre of petiole. Xylem elements are located toward the adaxial surface of petiole (Table 5, Fig. 5).

Fruit micromorphology: Fruits polachenarium, terete, oblong to linear cylindrical not compressed, 10 furrowed, stylopodium conical $3.5 - 5.6 \times 0.8 - 1.2$ mm. Surface is glabrous and brown. Surface epidermis includes stomata cells. The cells on the dorsal surface form a striate and rugulate pattern. The surface between the two ribs in the species is rugulate or striate (Fig. 7).

Secretory canals were densely found in stem (cortex, parenchyma and phloem), lamina (median vein, phloem and mesophyll) and petiole (whole parenchyma tissue) of *Kundmannia* type members as well.

Biometrical measures of various tissues composing the vegetative organs of *Kundmannia* taxa were taken, and no significant differences were found between the two taxa (Table 7). In stem anatomy it is observed that the medullary rays consist of lignified cells in *K. syriaca* while the cells are unlignified in *K. anatolica* (Table 2, Figs 1, 6). No significant differences were found

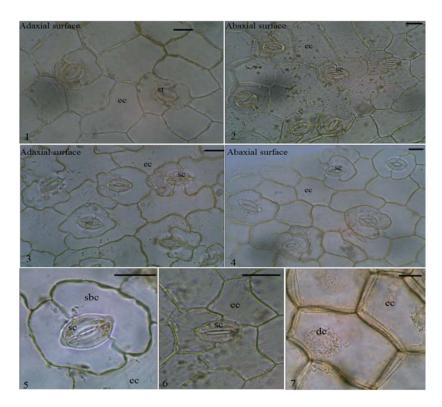


Fig. 4. Leaf epidermal surface of *Kundmannia* species. 1 - 2. *K. syriaca*, 3 - 4. *K. anatolica*. 5. Diacytic stomata. 6. Anomocytic stomata (rare). 7. Druse crystals seen in leaf epidermal cells (Scale bar 20 µm).

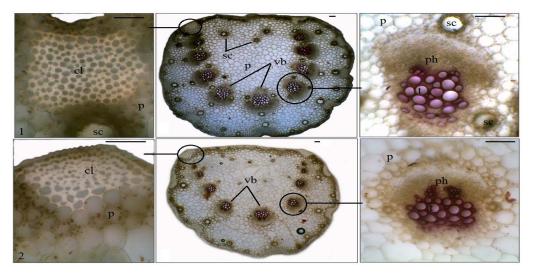
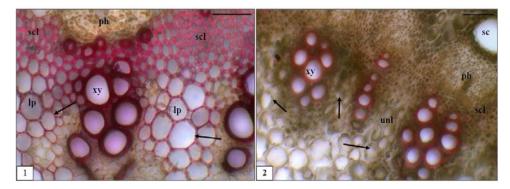


Fig. 5. Petiole structures in cross section of *Kundmannia* species. 1. *K. syriaca.* 2. *K. anatolica* (Scale bar = $50 \mu m$).



(Parenchyma cells are shown by arrow)

Fig. 6. Interfascicular region in the stem of *Kundmannia* species. 1. *K. syriaca*, 2. *K. anatolica*. (Scale bar = $50 \mu m$).

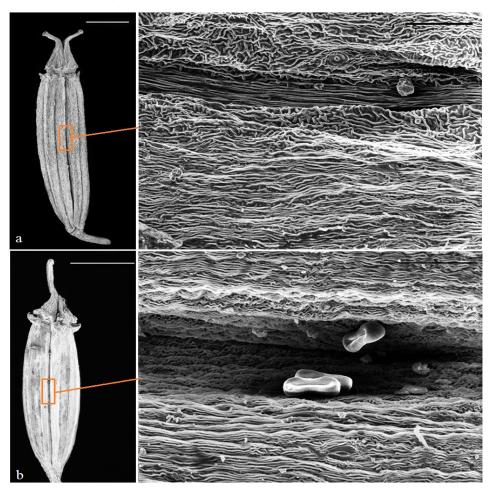


Fig. 7. Fruit micromorphology of Kundmannia species. a. K. syriaca, b. K. anatolica.

Table 7. Comparative micro-anatomical measurements of various tissues of Kundmannia species.

8			K. syriaca	iaca					K. anatolica	tolica		
Tissues		Width (µm)			Length (µm)	m)		Width (µm)	(m.		Length (µm)	mm)
	Min.	Max.	Mean ± Sd	Min.	Мах.	Mean ± Sd	Min.	Max.	Mean ± Sd	Min.	Max.	Mean ± Sd
Stem												
Epidermis layer	14.6	27.4	20±3.2	2	91	3	14.3	28.1	20.4 ± 3.2	,	3	11
Epidermis cells	14.5	25.5	18.2 ± 3.3	9.01	19.97	16.1 ± 3.2	15.2	27.9	21.7±3.4	=	27.6	16.4±4.2
Collenchyma layers	30.4	82.6	63.1±12.2		1	9	87.4	119.6	102.2 ± 8.8		1	
Secretory canal diameter(in	13.3	36.08	25.7±5.5	17.1	34.2	26.5±4.2	Absent					
collenchyma)												
Secretory canal diameter (in	28.3	64.6	53.2±11.4	18.9	58.45	49.2±5.9	35	87.5	63.8±18	27.5	85	61.2 ± 16.6
chlorenchyma)												
Interfascicular region	45.58	102.5	67.3±14.6		1		25	06	54.1±17			
Trache cells	11.4	36.1	23.8 ± 6.3	11.4	34.2	23.3±5.9	12.4	39.4	23.4 ± 8.2	12	41.1	25.5±8.3
Pith region	810.9	972.3	890.7±52				069	950	821 ± 83.6	,		
Ray												
Epidermis layer	18.7	45.7	27.8 ± 10.6	•	ī	í	17.8	25	21.3 ± 2.0	,	,	
Epidermis cells	15.7	36.8	23.6±6.2	14.3	19.6	16.8±1.5	12	29.4	21 ± 5.9	13	24.1	19.9±2.9
Collenchyma layers	19	36.1	28.8±5.7	,			13.1	35.1	23.3 ± 8.6			
Secretory canal diameter (in	15.2	41.8	26.2±10	13.3	39.9	23.2±8.6	34.6	85.6	60 ± 16.9	23.04	57.6	41.8 ± 12.04
parencyhma)												
Interfascicular region	30.1	75.1	55.5±14.6				30.3	81.6	53.4±21.1	,	,	
Trache cells	5.2	28.4	21.4 ± 5.3	5.3	28.5	21.3 ± 5.0	12.9	27.2	19.5±4.9	10.7	29.4	19.2 ± 5.01
Pith region	95	182	131.6±284		a i	1	86.4	155.5	130.5 ± 19.8			1
Lamina												
Upper (adaxial) epidermis cells	21.4	9.79	41 ± 13.02	16.2	43.7	30.4 ± 8.5	18.6	67.3	39.3 ± 14.1	16.1	33.07	25±4.7
Lower (abaxial) epidermis	18.4	54.5	33.4±11.3	17.3	32.6	25.4 ± 5.01	9.4	33.1	21.2±5.5	12.04	28	19.5±4.3
cells												
Mesophyl layers	121.7	230.5	157±34.1	e.	r	·	157.4	288.3	201.2 ± 33.3		9	E
Palisade parenchyma	5.8	14.3	9.6±2.4	24.4	68.4	44.3±10.6	9	Ξ	8.4 ± 1.2	25.8	40.7	32.6±4.6
Spongy parenchyma	12	35.6	19.5±6.2	11.6	27.6	18.2±3.8	11.2	29.4	20±5.5	9.4	25.4	16±3.4
Petiole												
Epidermis cells	7.5	30	19±6.3	12.5	32.5	22.8±5.9	9.4	30.8	18.2 ± 7.8	9.4	24.5	14.6±4.4
Parenchyma cells	20.4	99.2	53±20.4	17	85.6	45.6±21.6	25.4	127.2	63.6±27.3	26.3	109.9	59.7±20.7
Collenchyma layers	52.6	167.3	127±31.5				13.8	105.1	62.3 ± 28.1			
Secretory canal diameter	28.7	92.6	64.2±15.1	24	92.6	62.5±18	16.7	83.7	47.7±20.1	16.5	84.2	45.6±19.4
Trache cells	8.9	53.4	29.9±13.4	9.3	54.2	28.2±13.1	9.5	41.04	23.5±10.5	9.5	32.2	20.9±7.02

between taxa in ray and petiole anatomy. Stoma index rates of the leaves create differences among the taxa. The stoma index rate in *K. syriaca* was calculated to be 0.7 while it came out as 0.58 in *K. anatolica* (Table 6). Metcalfe and Chalk (1950) characterized stoma type of Apiaceae family as anomocytic, while Watson and Dallwitz (2000) characterized it as anomocytic or parasitic. Yet the stoma type of *Kundmannia*, a type of Apiaceae family, is considered to be anomocytic. Nevertheless, it was observed that it was densely diasidic (Fig. 4). Significant characters which could be used in divisions of taxa in fruit micromorphology were found. It is observed that fruit type of *K. syriaca* is oblong-cylindrical, while that of *K. anatolica* is prismatic-cylindrical (Fig. 7). On the fruit epidermal surface, rib epidermal surface of *K. syriaca* was observed to be rugulate, and it is striate among the ribs, while in *K. anatolica*, it is rugulate and striate above the ribs, and striate among the ribs (Fig. 7).

In conclusion, the following characters were found to be important for distinguishing (?) *Kundmannia* taxa: the state of parenchyma cells in interfascicular region in the stem, leaf stoma types, stoma index rates, fruit type and fruit surface morphology.

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