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Research Paper

Systematic significance of leaf epidermal features in *Brassicaceae* of Swat, Pakistan

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Abstract

Brassicaceae is taxonomically diverse and economically important plant lineage. Species identification and taxa relationship in this group often poses taxonomic problems particularly in temperate geographic regions. The present work was aimed at testing the evolutionary and systematic potential of leaf epidermal microscopic characters in Brassicaceae of Swat Pakistan. Epidermal preparations and microscopy are based on specimens of Brassicaceae collected from wild populations of the plants from different areas of the district. In total 27 species in 21 genera of the family were subjected to epidermal preparations. Ordinary epidermal cells ranged from polygonal (often), to pentagonal, hexagonal and tubular in shape (rare). Size of ordinary epidermal cells ranged from 40-373.5 µm. Three different types of stomata were examined in the studied species i-e anomocytic, staurocytic and anisocytic. Anomocytic stomata were observed only in Euclidium syriacum. Staurocytic type of stomata were found only on the adaxial surface of Nasturtium officinale. Size of stomata ranged from 15-40 µm. Size of stomatal pore ranges form (2-5) µm. Size of stomatal complex ranged between (40-200 µm). Only non-glandular trichomes were observed i-e stellate, unicellular, branched, T-shaped and Y-shaped. In 7 species unicellular non-glandular trichomes and in 6 species branched trichomes were present. In 4 speciesY-shaped trichomes were seen. Stellate trichomes were found in (Capsella bursapastoris and Alyssum desertorum). T-shaped trichomes were present in (Savignya parviflora). Size of trichomes ranged from (62.5-700) µm. In conclusion the micromorphologic features of leaf epidermis were sufficiently significant to distinguish closely related species.

Introduction

The *Brassicaceae* (Syn. *Cruciferae*) commonly known as the mustard family, is among the economically most valuable families, containing many edible oil yielding, vegetable and fodder plants. The family is large and contain about 338 genera and 3709 Species (Franzke *et al.*, 2010). In

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Pakistan, it is represented by 92 genera and 250 species (Jafri, 1973), with most of the species distributed in the upper temperate and alpine regions of the country. Members of the family are mostly dominant in cool temperate regions of the Northern hemisphere (Hedge, 1976) extending to the subtropics with little representation in the tropics as well. The family is most diverse in the Mediterranean region, South West Asia and Western North America (APG III, 2009). This family contains a considerable diversity of cultivated food crops such as radish, mustard, cress, kale, cauliflower, cabbage, broccoli etc. Many species, particularly those belonging to the genus *Brassica*, are used to produce edible and medicinal oils (Eruca oil) and animal fodder and oil cakes. A wide range of plants in this family are common ornamentals, including *Mathiola*, *Erysimum* (wallflower), *Iberis* (Candytuft), and *Lunaria* (money plant). However, the most promising plant among the angiosperm i-e *Arabidopsis thaliana* (the thale cress), the model organism belongs to this family. Members of this family are predominantly herbs, mostly perennial in the colder region, having (Glucosinolate compounds) sulphur contents in all parts. The most distinguishing features of the family are the presence of 4 cross shaped petals and 6 stamens which are arranged in a tetradynamous pattern and the presence of characteristic siliqua or silicula fruit (APG III, 2009).

Epidermal characters have potential taxonomic significance and are helpful as an additional taxonomic character (Stace, 1965; Baronova, 1992). Metcalfe and Chalk (1957) studied the anatomy of the family Brassicaceae and determined the diagnostic anatomical characteristics as epidermal cell types, stomata type and the arrangement of the sclerenchymatic cells around the vascular bundles of the leaves. Each individual stomatal orientation is important but their pattern of distribution and presence or absence on upper and lower epidermis provide us an important taxonomic key (Khalik et al., 2005). Stace (1980) reported 31 different types of stomata among cotyledonous plants. Anatomical features of leaf epidermis such as stomata, trichomes and other characters are useful anatomical tools (Stace, 1980). All the non-reproductive organs, leaf is the most commonly used in plant taxonomy and leaf epidermis is of prime importance in solving taxonomic problems parallel with cytology (Stace, 1965, 1984). Inamdar and Rao (1983) studied the taxonomic significance of trichomes in 35 species of the family Brassicaceae. The trichomes are eglandular only which are classified into unicellular, bicellular and multicellular on the basis of number of cells. Khalik et al. (2005) studied in detail the morphology and systematic significance of trichomes in 82 species of Brassicaceae from Egypt. They classified the trichomes into 12 different types, having both unicellular and multicellular and eglandular and slightly glandular forms, and has discussed that trichomes characters were useful to distinguish Brassicaceae members at tribe, genus and below levels. A key to species based on trichome morphology has also been provided. Ancev and Goranova (2006) studied the diversity of trichomes on leaves and fruits of 18 members of tribe Alysseae from Bulgaria and reported 4 different trichomes; simple, 2-5-armed stalked, stellate and dendride type. They found that the trichomes may be smooth or tuberculate. The trichome characters could distinguish between closely related species like Alyssum pirincium and A. cuneifolium. Doaigey et al., (2013) studied the epidermal micromorphology of 34 species of Brassicaceae form Saudi Arabia using LM and SEM microscopy and divided these species into four groups based on the features of eglandular trichomes. They distinguished the species by providing key based on trichome, stomata and cell shape and wall morphology.

The family is locally represented by a large number of species including those used as wild vegetables and medicinal herbs. Majority of the species are superficially similar because of the presence of corymbose inflorescence and often variously divided leaves. These make the species taxonomically problematic and pose difficulties in authentic identifications. The microscopic features of the leaf epidermis provide substantial data for making identification keys and taxonomic

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confirmations. Review of literature shows that to date there exist no studies on the micromorphological characteristics of the *Brassicaceae* in Pakistan.

Materials and Methods

Taxa sampling and field collections

The present work is based on members of *Brassicaceae* family collected from different ecological regions of district Swat. Collections were made from February to September 2015 from wild populations of *Brassicaceae* in the study area. At least five specimens of each species were collected from different localities. Field information was recorded for each specimen and were properly dried, preserved and then mounted on standard herbarium sheets according to methods described by (Judd *et al.*, 2007). The voucher specimens of each species were deposited in the herbarium of University of Swat (SWAT) for future reference (Table 1). Species were identified using published flora (Jafri, 1973; Cheo *et al.*, 2001) and with the help of expert taxonomist.

Preparation of leaf epidermal peel for microscopic examination

For epidermal preparations, representative samples 1 to 2 cm were cut from the midportion of mature foliage leaves. The fresh leaves were placed in a test tube filled with 88% lactic acid, kept in water bath and boiled at 100°C for 30 to 40 minutes. For preparing abaxial leaf epidermis, the leaf was placed upside down with the abaxial surface beneath towards the slide and the adaxial surface above away from slide surface. Sharp razor blade was used to remove the upper epidermis and mesophyll tissue, remaining only the lower (abaxial) epidermis. The adaxial (upper epidermis) was prepared using the same procedure. The prepared epidermis was mounted on a clean glass slide using lactic acid or glycerol.

Microhistological photographs of both epidermal surfaces were taken using a camera (Lucida infinity 1.5) fixed with x20 objective lens of the microscope. Observations of the slides were done using Compound Nikon microscope using 10x, and 40x objectives. For each epidermis at least two slides were prepared. Measurements of the quantitative and qualitative features were done for all representative slides. At least 10 measures of each character were taken for authenticity and statistical analysis. Preparative techniques were followed after Clark (1960) and Ullah et al. (2011). Terminology was used in accordance with Khalik (2005).

Rresults and Discussion

The present investigation on leaf micromorphological features are based on 27 species belonging to 21 genera of the family *Brassicaceae* from Swat Pakistan. The largest genera are *Thlaspi* and *Lepidium* represented by 3 species each, while *Sisymbrium* and *Rorippa* each have two species. The remaining genera are represented by one species each (Table 1). We noticed considerable variation in anatomical characters of leaf epidermis among different species of *Brassicaceae*. Micromorphological studies showed variation in ordinary epidermal cells, trichomes and stomata etc in various species of the family.

Epidermal Cells

The ordinary epidermal cells were irregular to polygonal and sometimes isodiametric, pentagonal and tubular in shape. In the present work, majority of the species have undulating wall morphology, some have sinuous wall and, in some species, straight walls were also reported. Similar findings have also been reported by (Ahmad *et al.*, 2010; Doigey *et al.*, 2013). In *Sisymbrium irio* and *Cardamine loxostemonoides* isodiametric cells were also observed. The cell walls are either thin or

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thick with warty or beaded appearance or rarely straight. In *Lepidium apetalum* the ordinary epidermal cells are irregular to polygonal and wall morphology is undulating to straight. Ahmad *et al.*, (2010) reported polygonal to tubular epidermal cells in *Lepidium apetalum* (Fig. 1).

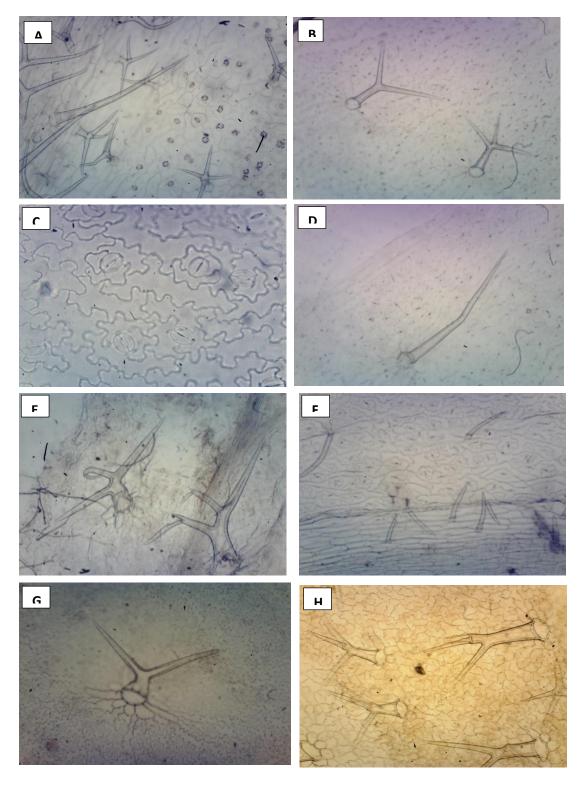


Figure 1. A-B. Arabis pterosperma, dendroid and simple hairs. C-D. Arabidopsis thaliana, stomata and simple trichomes. E. Malcolmia africana, dendroid trichome. F. Lipidium apetalum simple hairs. G. Euclidium syriacum Y-shaped trichome. H. Neslia apeculata stalked 2-3 armed trichome.

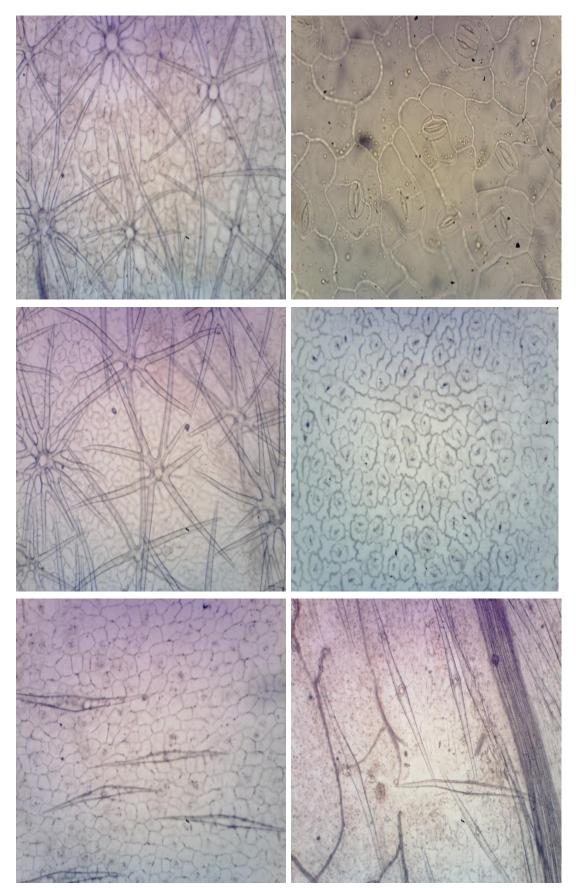


Figure 1: I-K. Alyssum desertorum stellate trichome. J. Sisymbrium irio anisocytic stomta. L. Thlaspi perfoliatum stomata. M-N. Savigna parviflora T-shape trichome.

Stomata

Metcalfe and Chalk (1979) reported anisocytic type of stomata in *Brassicaceae* family. Stomata were present on both the surfaces of the species and is anisocytic in all species, intermixed in *Euclidium syriacum* with anomocytic stomata. In *Nasturtium officinale* DC. Stomata is staurocytic on the adaxial surface. In most of the species' stomata were abundant to common but in some species stomata were rare. Mousavi and Sharifi-rad, (2014) studied the micromorphological characters of *Cardaria draba* and observed anisocytic type of stomata which is similar to our findings. Ahmad *et al.*, (2010) have reported amphianisocytic type stomata in *Lepidium apetalum* which is different from our result because we observed only anisocytic type of stomata. We also have found mostly anisocytic type of stomata, similar to the above works. Doigey *et al.* (2013) have reported that anomoytic stomata also occur in *M. africana* but we have not observed in our specimen (Fig. 1).

Trichomes

Trichomes were observed in 13 species while 14 species were found galbrous having no trihomes. Trichomes were observed on both the surfaces of the same species in Arabidopsis thaliana, Arabis pterosperma, Alliaria petiolata, Cardamine loxostemonoides, Capsella bursa-pastoris, Cardaria draba, Euclidium syriacum, Lepidium pinnatifidum, Lepidium apetalum, Malcolmia africana, Neslia apiculata, Sisymbrium orientale and Alyssum desertorum (Table 2). The trichomes are eglandular mainly, rarely glandular clavate type were observed. The trichomes were unicellular, stellate, Y-shaped, T-Shaped, branched hairs (dendroid type), clavate and multicellular glandular (Table 2; Fig. 1). In most of the species, trichomes were common, rare and in some species trichomes were abundant. Metcalf and Chalk (1957) reported mainly single-celled and very rarely secretary trichomes, the non-glandular trichomes were y-shaped, T-shaped, shield-like and simple. We observed in Cardaria draba non glandular unicellular and branched hairs trichomes. Mousavi and Sharifi-Rad, (2014) studied the micromorphological characters of Cardaria draba and observed non glandular unicellular trichomes which is same to our result because in our result. Ancev and Goranova (2006) reported 4 types of trichomes in tribe Alysseae, which were simple, dendroitic, stalked and stellate. Our observations are corroborated with their findings, Khalik (2005) reported different types of trichomes in Brassicaceae family two main types of trichome are observed non-glandular unicellular trichomes which may be simple, hooked, vasculate, appressed medifixed and trifixed, Yshaped, branched (3-4 fids), dendroid, stellate and clavate and 2. glandular trichomes which is consists of a multicellular stalk and a head of more cells and unicellular which is in the terminal part globular or slightly club-shaped which is different from our result because in our result only nonglandular trichomes were reported.

Table 1. Details of species collection and accession numbers.

S.	Species	Collection	Accession	Collector and locality
No		no.	no.	information
1	Alliaria petiolata (M.Bieb.) Cavara Ex. Grande	ASW.14	SWAT0203	Chail Madyan in moist places; Asma , Aziz, Dr. Zahid, Nasar
2	Alyssum desertorum Stapf	ASW.04	SWAT0220	Matta; Asma , Aziz, Dr. Zahid
3	Arabidopsis thaliana (L.) Heynh.	ASW.23	SWAT0201	Mountains West of Matta; Dr. Zahid
4	Arabis pterosperma Edgew.	ASW.21	SWAT0202	Miandam foot hills Dr. Zahid
5	Brassica rapa L.	ASW.16	SWAT0204	Odigram; Asma, Aziz
6	Capsella bursa-pastoris (L.) Medik.	ASW.20	SWAT0206	Marghazar, Kanju; Asma
7	Cardamine loxostemonoides O.E.Schulz	ASW.24	SWAT0205	Madyan; Asma and Dr. Zahid
8	Cardaria draba (L.) Desv.	ASW.12	SWAT0207	Matta; Asma , Aziz
9	Coronopus didymus (L.) Sm.	ASW.18	SWAT0208	Odigram; Asma, Aziz, Nasar

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10	Descurainia sophia (L.) Webb ex Prantl	ASW.22	SWAT0209	Aziz, Dr. Zahid
11	Euclidium syriacum (L.) R.Br.	ASW.08	SWAT0210	Barikot; A. Wahid and Z. Ullah
12	Goldbachia laevigata (M.Bieb.) DC.	ASW.15	SWAT0211	Matta; Asma, Dr. Zahid
13	Isatis costata C.A. Mey.	ASW.17	SWAT0212	Islampur; Asma , Aziz
14	Lepidium apetalum Willd.	ASW.09	SWAT0214	Matta; Asma , Aziz, Dr. Zahid,
15	Lepidium pinnatifidum Ledeb	ASW.19	SWAT0213	Kanju; Asma , Aziz
16	Lepidium sativum L.	ASW.02	SWAT0215	Odigram; Asma , Imran
17	Malcolmia africana (L.) R.Br.	ASW.07	SWAT0216	Matta; Asma , Aziz
18	Nasturtium officinale DC.	ASW.13	SWAT0218	Kanju, Matta; Asma , Aziz
19	Neslia apiculata Fisch., C.A.Mey. and Avé- Lall	ASW.06	SWAT0217	Madyan; Asma, Dr. Zahid, Ahmad
20	Rorippa montana (Wall. ex Hook. f. and Thomson) Small	ASW.10	SWAT0221	Chuprial Matta; Dr. Zahid
21	Rorippa islandica (Oeder) Borbás	ASW.05	SWAT0219	Matta; Asma , Aziz, Dr. Zahid
22	Savignya parviflora (Delile) Webb	ASW.03	SWAT0224	Barikot; Asma , Aziz, Dr. Zahid
23	Sisymbrium irio L.	ASW.27	SWAT0222	Madyan; Asma
24	Sisymbrium orientale L.	ASW.11	SWAT0223	Kanju; Asma , Aziz, Dr. Zahid
25	Thlaspi perfoliatum L.	ASW.26	SWAT0226	Miandam; Dr. Zahid
26	Thlaspi andersonii (Hook. f. and Thomson) O.E. Schulz	ASW.01	SWAT0227	Lalkoo; Dr. Zahid
27	Thlaspi arvense L.	ASW.25	SWAT0225	Jargo; Dr. Zahid

Table 2. Qualitative attribute of leaf blade microscopic features of *Brassicaceae* family, Swat, KPK.

Sr. No.	Species	EpiC shape	EpiC. wall Morph.	Stomata Type	Stomatal distribution	Trichomes	Trichomes distribution
1	Alliaria petiolata	Pol/Ir	Undulating, Sinuous (ad)	Anisocytic, Anomocytic	Abundant	_	
2	Alyssum desertorum	Ir /pol	Undulating and straight	Anisocytic	Abundant	Stellate, Multicellular Glandandular	Abundant
3	Arabidopsis thaliana	Ir/pol	Undulating and sinuous	Anisocytic	Common	Simple, clavate, Y- shaped, dendroid type	Common
4	Arabis pterosperma	Pol/ir	Undulating and straight	Anisocytic	Common, rare (Ad)	Simple hairs, clavate, Y-shaped, dendroid type	Abundant
5	Brassica rapa	Pol/ir	Undulating and sinuous	Anisocytic	Common	—	
6	Capsella bursa -pastoris	Ir/ pol	Undulating and straight	Anisocytic	Abundant	Simple hairs, Stellate	Abundant
7	Cardamine loxostemonoides	Ir/pol /Iso	Undulating	Anisocytic	Common	Simple hairs, T- shaped	Rare
8	Cardaria draba	Ir/pol	Undulating	Anisocytic	Abundant	Simple hairs, dendroid type	Abundant
9	Coronopus didymus	Ir /pol	Undulating	Anisocytic	Abundant	_	
10	Descurainia sophia	Ir /pol	Undulating and sinuous	Anisocytic	Abundant	_	

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11	Euclidium syriacm	Ir/pol	Undulating and sinuous	Anisicytic and anamocytic	Abundant	Y-shaped, Stellate, dendroid type	Rare
12	Goldbachia laevigata	Ir/pol	Undulating and straight	Anisocytic	Abundant		
10	-	7 / 1	77 1 1	4	C		
13	Isatis costata	Ir/pol	Undulating and straight	Anisocytic	Common		_
14	Lepidium	Ir/pol	Undulating	Anisocytic	Abundant	Simple hairs	Common
	pinnatifidum		and sinuous				
15	Lepidium apetalum	Ir/pol	Undulating and straight	Anisocytic	Abundant	Simple hairs	Abundant
16	Lepidium virginicum	Ir /pol	Undulating and straight	Anisocytic	Abundant	_	
17	Malcolmia africana	Ir/pol	Undulating and sinuous	Anisocytic, Anomocytic	Abundant	Dendroid type and Y-shaped	Common
18	Nasturtium officinale	Ir /pol	Undulating	Anisocytic	Abundant		_
19	Neslia apiculata	Ir /pol	Undulating and sinuous	Anisocytic	Abundant	Dendroid type	Common
20	Rorippa montana	Ir /pol	Undulating and straight	Anisocytic	Common		-
21	Rorippa islandica	Ir/pol	Undulating and straight	Anisocytic	Abundant		_
22	Savignya parviflora	Ir /pol	Undulating	Anisocytic	Abundant	T-shaped	Abundant
23	Sisymbrium irio	Ir/pol /Iso	Undulating	Anisocytic	Common		
24	Sisymbrium orientale	Ir /pol	Undulating	Anisocytic	Abundant	Simple hairs	Abundant
25	Thlaspi perfoliatum	Ir/pol	Undulating	Anisocytic	Common		_
26	Thlaspi andersonii	Ir/pol	Undulating	Anisocytic	Common		_
27	Thlaspi arvense	Pol /ir	Undulating	Ansicoytic	Common		

 $Key: Ir = irregular, Pol = Polygonal, Iso = Isodiametric, EpiC = Epidermal Cell, Ad = Adaxial \ surface$

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Table 3. Quantitative characteristics of leaf surface features; epidermal cells, stomata and trichomes of Brassicaceae from Swat Pakistan.									n.	
Species Name	OECL (µm)	OECW(µm)	SL(µm)	SW(µm)	SPL(µm)	SCL(µm)	SCW(µm)	SuW(µm)	TrL(µm)	TrW(µm)
Species Name	Min-Max; Mean(±SE)	Min-Max; Mean(±SE								
A. desertorum	87-147;	37-75;	25-30;	10-25;	12-20;	100-200;	50-112;	25-50;	520-600;	30-50;
A. aeseriorum	111.5(±9.9)	58(±6.4)	$27(\pm 0.9)$	$18(\pm 2.5)$	16(±1.3)	137.5(±17.8)	80(±11.6)	38.5(±4.3)	556(±15.0)	$42(\pm 3.7)$
A	45-85;	25-75;	15-25;	10-15;	5-12;	67-100;	45-57;	27-62;		
A. petiolata	67.5(±8.4)	$46.5(\pm 9.5)$	$20(\pm 1.8)$	$13(\pm 1.2)$	9(±1.3)	35(±5.5)	$52(\pm 2.2)$	$40(\pm 6.1)$		
4	40-115;	37-70;	25-30;	10-25;	10-15;	75-130;	50-112;	27-75;	180-730;	20-100;
A. pterosperma	84(±13.8)	56(±5.6)	28(±0.9)	17(±2.5)	13(±0.9)	95(±11.1)	74.5(±11.1)	55.5(±12.6)	450(±99.1)	64(±16.3)
A . 1111	87-175;	30-72;	17-25;	10-15;	5-12;	55-85;	35-95;	20-50;	150-550;	20-60;
A. thaliana	$127(\pm 14.4)$	$47(\pm 7.1)$	$21(\pm 1.3)$	$12(\pm 1.2)$	$9.5(\pm 1.2)$	$70.5(\pm 5.3)$	$57.5(\pm 10.4)$	$32.5(\pm 5.2)$	348(±65.6)	$42(\pm 6.6)$
D nana	65-373;	93-150;	15-30;	10-15;	7-15;	77-115;	77-112;	30-75;		
B. rapa	167(±30.1)	126.2(±9.9)	$21(\pm 2.6)$	$12(\pm 1.2)$	11.5(±1.3)	99.5(±7.6)	94(±5.8)	$50(\pm 8.6)$		
C. bursa-pastoris	100-150;	32-52;	17-25;	10-20;	5-10;	40-70;	42-62;	20-55;	150-630;	30-90;
C. bursa-pasioris	118.8(±9.4)	44.5(±3.5)	22(±1.5)	15(±1.6)	8.5(±1)	52.5(±5.2)	52(±3.9)	30(±6.3)	388(±96.6)	56(±10.8)
C. didymus	55-175;	17-57;	20-25;	10-20;	7.5-12;	45-80;	45-55;	25-62;		
C. alaymus	102(±26.4)	$35(\pm 7.5)$	22(±0.9)	15(±2.2)	10.5(±0.9)	61(±6.82)	50(±1.8)	37(±6.9)		
C. draba	112-250;	62-162;	30-37;	10-25;	15-22;	100-200;	75-190;	52-82;	210-300;	50-70;
C. araba	188.5(±27.1)	101(±17.3)	34(±1.3)	17(±2.5)	19(±1.27)	155.5(±17.3)	133(±20.0)	68.5(±5.0)	268(±17.1)	58(±3.7)
C. loxstemonoides	50-162;	37-87;	17-25;	10-20;	7-15;	60-150;	45-75;	15-50;	150-500;	40-60;
C. ioxsiemonoides	95(±19.7)	$60(\pm 8.4)$	$22(\pm 1.5)$	14(±1.9)	11.5(±1.3)	93(±15.6)	56(±5.6)	34(±6.3)	310(±64.0)	$48(\pm 3.7)$
D. Sophia	75-145;	22-55;	20-27;	10-20;	10-15;	87-135;	45-82;	17-37;		
D. зорни	109(±11.97)	42(±5.7)	24(±1.3)	16(±1.9)	12.5(±1.1)	111(±10.4)	63.5(±7.1)	29(±3.6)		
E. syriacum	100-212;	42-85;	25-40;	15-25;	15-25;	80-162;	50-87;	50-80;	200-450;	50-100;
E. syrtacum	131(±20.6)	57.5(±7.9)	32.5(±2.5)	21(±1.9)	20(±1.8)	117(±13.9)	64.5(±8)	65.5(±5.5)	326(±44.5)	$72(\pm 8.6)$
G. laevigata	125-250;	42-75;	27-37;	15-20;	12-20;	87-200;	75-140;	30-72;		
O. taevigata	192.5(±25.5)	60.5(±6.3)	32.5(±1.8)	18(±1.2)	17.5(±1.4)	139(±18.5)	110.5(±11.1)	46(±8.1)		
I. costata	55-120;	45-75;	22-30;	10-20;	10-15;	75-125;	72-115;	25-45;		
1. Costata	84(±11.97)	55.5(±5.5)	25.5(±1.5)	16(±1.9)	12(±0.9)	92(±9.9)	90(±6.9)	34(±3.4)		
L. apetalum	62-112;	45-75;	22-27;	10-20;	12-20;	37-60;	50-75;	25-50;	62-150;	15-22;
E. aperanim	93(±9.16)	59.5(±5.4)	$24.5(\pm 0.9)$	15(±1.6)	16(±1.3)	48.5(±3.7)	64.5(±4.2)	35(±4.6)	110(±16.0)	18.5(±1.3)
L. pinnatifidum	50-100;	25-70;	20-27;	10-20;	10-17;	10-17;	45-75;	15-50;	350-570;	20-40;
L. pumanjiann	$76.5(\pm 8.0)$	44.5(±7.8)	24(±1.3)	33.5(±1.9)	14(±1.3)	14(±1.3)	58(±5.1)	33.5(±6.1)	448(±35.6)	28(±3.7)
L. virginicum	70-185;	55-100;	27-37;	15-25;	12-22;	75-150;	70-100;	25-75;	_	
	156.5(±21.8)	73(±7.6)	32.5(±1.8)	21(±1.9)	17.5(±1.8)	115(±13.3)	79.5(±5.4)	46(±8.3)		
M. africana	50-80;	35-50;	27-40;	10-20;	20-30;	60-167.5;	80-167;	47-80;	200-550;	80-100;
1.1. cy i cana	66.5(±4.9)	44(±3.2)	31.5(±2.3)	15(±1.6)	25.5(±1.7)	130.5(±19.3)	118.5(±15.5)	63(±5.6)	348(±59.6)	90(±4.5)
N. apiculata	75-125;	22-37;	30-37;	10-20;	15-20;	110-175;	50-75;	25-37;	150-250;	30-60;
т. арисиша	104(±8.3)	30(±3.2)	$33.5(\pm 1.7)$	16(±1.9)	18(±0.9)	144(±14.1)	63.5(±4.3)	33(±2.4)	198(±16.6)	46(±5.1)

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N. officinale	50-100; 70(±8.5)	25-57; 46.5(±5.8)	22-37; 29.5(±2.7)	10-25; 20(±2.7)	15-20; 17.5(±1.1)	50-87; 71(±7.4)	50-80; 65(±5.5)	17-50; 34(±5.9)	_	
R. islandica	62-175; 111(±18.2)	37-75; 54(±6.6)	27-32; 29(±1)	10-20; 16(±1.9)	12-17; 15.5(±0.9)	100-175; 127(±13.5)	72-125; 86(±9.9)	25-75; 48.5(±8.4)	_	
R. montana	50-100; 72.5(±8.3)	32-55; 45(±4.3)	25-30; 27.5(±1.1)	10-20; 14(±1.9)	15)20; 18.5(±1)	42-100; 63.5(±10.2)	50-100; 67(±8.6)	25-62; 46(±6.1)	_	
S. irio	67-145; 102.5(±14.7)	37-87; 62.5(±8.8)	22-27; 24.5(±0.9)	10-25; 17(±2.5)	7-15; 11.5(±1.3)	62-100; 84(±6.8)	42-80; 58.5(±6.4)	22-42; 32(±3.7)	_	
S. orientale	45-75; 61.5(±5.3)	20-45; 31.5(±4.7)	17-25; 21(±1.3)	5-15; 11(±1.9)	7-15; 11.5(±1.3)	47-80; 62(±6.6)	47-82; 62(±7.9)	20-62; 39(±7.9)	450-650; 220(±35.2)	40-100; 68(±11.,6)
S. parviflora	50-125; 93.5(±12.4)	37-55; 46.5(±3.3)	20-25; 23(±0.9)	10-20; 15(±1.6)	7-12; 9.5(±0.9)	67-95; 78.5(±4.7)	57-115; 80(±11.5)	20-50; 31(±5.6)	400-700; 534(±53.4)	20-40; 30(±3.2)
T. andersonii	157-245; 197(±14.9)	47-87; 65.5(±7.9)	20-25; 22.5(±1.1)	15-25; 19(±1.9)	7-15; 11.5(±1.3)	72-105; 89.5(±6.6)	50-75; 61.5(±4.3)	37-50; 42(±2.4)	_	
T. arvense	145-175; 160.5(±5.61)	50-75; 60(±4.3)	22-30; 26(±1.3)	10-20; 16(±1.9)	10-17; 13.5(±1.3)	100-200; 137(±18.8)	50-150; 89(±17.2)	17-62; 49.5(±8.3)	_	
T. perfoliatum.	50-92; 73.5(±7.9)	37-75; 58.5(±6.6)	20-25; 22.5(±1.1)	10-20; 15(±1.6)	10-17; 13.5(±1.3)	47-85; 66(±6.7)	37-62; 52.5(±4.4)	17-50; 30.5(±5.4)	_	

Key to the abbreviations: OECL: Ordinary Epidermal Cell Length; OECW: OrdinarJy Epidermal Cell Width; SL: Stomata Length; SW: Stomatal width; SPL: Stomatal Pore Length; SCL: Stomatal complex Length; SCW: Stomatal complex Width; SuW: Subsidiary Cell Width; TrL: Trichome Length; TrW: Trichome Width

Conclusion

Result of the present investigation revealed that considerable variation exist in micromorphological characters in the leaf epidermis among the taxa studied. In particular greater diversity was observed in the trichomes, stomata, and ordinary epidermal cells type, shape, size and distribution. These microscopic characters can be utilized to separate closely related species among the family. Non glandular hairs are a characteristic of the family and were observed in some species. Stomata in most members are Anisocytic, sometimes staurocytic and in some cases anomocytic. Epidermal anatomy was useful for authentic determination of the problematic taxa e.g in *Lepidium pinnatifidum* and *Lepidium apetalum* unicellular trichomes are present whereas in *Lepidium virginicum* trichomes were absent. In *Sisymbrium orientale*, unicellular trichomes were present whereas in *Sisymbrium irio*, trichomes were absent. It is concluded that leaf micromorphological characters are useful in systematics and give better results when supplemented by morphology. However, these characters alone could not give higher resolution and hence these should be associated with other data from morphology, palynology etc to give better results.

References

- Ahmad, K., Khan, M.A., Ahmad, M., Shaheen, N and Nazir, A. 2010. Taxonomic diversity in epidermal cells of some sub-tropical plant species. *International Journal of Agriculture and Biology.*, 12: 115_118.
- Ančev, M., & Goranova, V. 2006. Trichome morphology of eleven genera of the tribe Alysseae (Brassicaceae) occurring in Bulgaria. *Willdenowia*, 36(1), 193-204.
- APG, III .2009. An update of the Angiosperm Phylogeny Group classification of the orders and families of flowering plant. *Botanical Journal of the Linnean Society*. *161*, 105-121.
- Baronova, M. 1992. Principles of comparative stomatographic studies of flowering plants. *Botanical Review*. 58: 1-9.
- Cheo, T.Y., L.L. Lu., G. Yang., I. Al-Shehbaz and V. Dorofeev (2001). Brassicaceae Burnett. In: Z.Y, Wu., P.H. Raven (eds). Flora of China, Vol 8 Brassicaceae through Saxifragaceae. Science Press Misspuri Botanical Garden, Beijing/St.Louis.
- Clark, J. 1960. Preparation of leaf epidermis for topographic study. Stain. Technol., 35: 35-39.
- Doaigey, A., El-Habashy, I., Al-Watban, A., Milagy, A.E., Al-Sahli, A., Siddiqui, M., Al-Whaib and M. El-Zaidy 2013. Epidermal characteristics of 34 species of Brassicaceae growing in desert of Saudi Arabia. Wulfenia, 20(9): 202-221.
- Franzke, A., Lysak, M.A., Al-Shehbaz, l.A., Koch, M.A. and Mummenhoff, K. 2010. Cabbage family affairs: the evolutionary history of *Brassicaceae*. *Trened in Plants Society*, 839: 1-9.
- Hedge, I.C. 1976. A systematic and geographical survey of the old world *Cruciferae*. In:Vaughan, J.G., MacLeod, A.J. and Jones, B.M.G. (eds), The Biology and Chemistry of the *Cruciferae*. pp. 1-45. Academic Press, New York, USA.
- Inamdar, J. A. & Rao, N. V. 1983: Light and scanning electron microscopic studies on trichomes of some *Brassicaceae*. Feddes Repert. **94:** 183-190.
- Jafri, S.M. 1973. Flora of West Pakistan. No. 55 *Brassicaceae*. Department of Botany, University of Karachi.

- Judd, W.S., C.S. Cambell., E.A. Kellog., P.F. Stewens and M.J. Donoghue. 2002. "Plant Systematics". A phylogenetic approach. Sinauer Associates, Inc. Publishers, Sunderland, Massachusetts USA.
- Khalik, K. A. 2005. Morphological studies on trichomes of Brassicaceae in Egypt and taxonomic significance. Acta Botanica Croatica, *64*(1), 57-73.
- Metacalfe, C.R. and Chalk, L. 1979. Anatomy of dicotyledons, systematic. Anatomy of leaf and stem. *Volume 1.2ndED*. Claremont press, Oxford.
- Metcalfe, C.R and Chalk, L. 1957. Anatomy of the Dicotyledons (Leaves, Stem and wood in relation to taxonomy with notes on economic uses, *Volume*. *1*, pp. 79-87. Oxford University Press, Amen House, London.
- Metcalfe, C.R, Chalk, L. 1950. Anatomy of the Dicotyledons, Volume I. Clarendon Press, Oxford.
- Mousavi, S.M., and J. Sharifi-Rad. 2014. Anatomical, morphological and micromorphological study of seed, trichome and stomata of *Cardaria draba L. Desv (Brassicaceae)* in Sistan, Iran. *International Journal of Biosciences*, 5(11); 63-969.
- Stace, C. 1980. Plant taxonomy and biosystematics. Edward Arnold publisher Ltd, London. Esau, K and Clark. (1960). Preparation of leaf epidermis for topographic study. Stain. Technol., *35*:35-39.
- Stace, C.A. 1965. Cuticular studies as an aid to plant taxonomy. Bull. of the British Museum (Nat. Hist.) *Botany*, *4*: 1-78.
- Stace, C.A. 1984. The taxonomic importance of the leaf surface. In V. H Herwood, D. M. Moore (eds.), Current concepts in Plant taxonomy Systematic association special Volume. 25. Academic Press London and Orlando.
- Ullah, Z. M.A. Khan., M. Ahmad., M. Zafar and K. Ullah. 2011. Systematic implications of foliar epidermis in Andropogoneae (Poaceae) from Hindukush-Himalayas Pakistan. Journal of Medicinal Plants Research. 5(6), 949-957.