



Full Length Article

Micromorphological Investigation of Foliar Anatomy of Genera *Aconogonon* and *Bistorta* of Family Polygonaceae

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ABSTRACT

Leaf epidermal studies have been carried out on six species belonging to two genera of the family Polygonaceae. The use of light microscopy has made possible in depth to study leaf surface features such as shape of epidermal cells, stomatal pattern, their distribution on adaxial and abaxial leaf surface and trichomes types. Epidermal cell shapes are variable but generally polygonal. Six different stomatal patterns are reported for *Aconogonon* (Meisn.) Reichenb. and *Bistorta* Adans. Variation among glandular and non glandular trichomes was also noted. Cyclocytic stomata are recorded first time in *Aconogonon alpinum* (All.) Schur. This anatomical study has taxonomic importance, on the basis of which identification keys are prepared.

Key Words: Polygonaceae; Staurocytic stomata; Cyclocytic; Peltate trichome; Spheroidal trichomes

INTRODUCTION

Polygonaceae is a cosmopolitan family containing approximately 1,200 species from 48 genera (Freeman & Reveal 2005; Sanchez & Kron, 2008). They are group of morphologically different herbs, shrubs, small trees or climbers characterized by simple leaves with covering ochrear stipules, unilocular ovary and endospermic seeds (Hutchinson & DaLziel, 1954; Brummitt, 1992). The family is generally considered to be comprised of 30 to 49 genera. They are distributed worldwide from the tropics to the arctic, but most of the species are concentrated in the northern temperate region (Heywood, 1978).

Metcalf and Chalk (1950) gave comprehensive report on foliar anatomy of the Polygonaceae. Lersten and Curtis (1992) examined foliar anatomy of genus *Polygonum* and described numerous anatomical features of leaves, their variations and distribution patterns, among 153 species of *Polygonum* sensu lato representing all sections of the genus. Ayodele and Olowokudejo (2006) made comparative studies on the leaf epidermal features of five tribes including 19 species of family Polygonaceae in West Africa.

In Pakistan, the family is represented by 19 genera and 103 species (Qaisar, 2001). Genus *Aconogonon* (Meisn.) Reichenb. is distributed from North to South Asia, Japan and North America and in Pakistan represented by 5 species, while *Bistorta* Adans, distributed in North America, Europe and Asia, represented in Pakistan by 7 species (Qaisar, 2001).

In this study we included six species, which belong to two genera of the family. The main objectives of this study was to add more information about leaf anatomy so as to

differentiate two genera of on the basis of epidermal cells, stomatal type and foliar trichomes (glandular & non-glandular), both qualitatively and quantitatively. In addition, the aim was to determine whether there is any difference regarding epidermal cells, stomata and trichome types at upper and lower epidermis and which type is most prevalent in two genera of Polygonaceae.

MATERIALS AND METHODS

Dried leaves of representative specimen from Quaid-i-Azam University herbarium, Islamabad, Pakistan, of two genera of Polygonaceae, listed in Table I, were used for anatomical studies. Dried leaves were placed in boiling water for few minutes to soften until they became unfolded and ready for epidermal scrapping. Leaf samples were prepared according to modified method of Cotton (1974) who followed the method of Clark (1960). The leaves were placed in 88% lactic acid kept hot in boiling water bath (Model, Memmert-91126-FRG, Germany) for 30 to 40 min for softening the tissue and facilitates scraping the leaf surface with scalpel. Slides of both abaxial and adaxial surface of leaf were prepared and mounted in fresh 88% lactic acid. Both qualitative and quantitative micromorphological foliar characteristics were observed, microhistological photographs of both surfaces were taken by Nikon (FX-35) Camera equipped light microscope.

RESULTS AND DISCUSSION

Our study included shape determining of leaf epidermal cells, stomatal types and trichomes (glandular &

non-glandular) on adaxial and abaxial surface both quantitatively and qualitatively. Epidermal features have provided significant supplementary proof and are of taxonomic importance (Stace, 1965). The characteristics of leaf epidermis of *Aconogonon* (Meisn.) Reichenb. and *Bistorta* Adans. of Polygonaceae under LM observations are listed in Table II and III.

Aconogonon. In this genus, irregular and undulate epidermal cells were restricted to abaxial surface, showing its mesomorphic nature (Stace, 1965), while adaxial surface was with polygonal cells (Table III). There was variation in shape of epidermal cells on both leaf surfaces (Stace, 1965). Largest epidermal cells were noted in *A. rumicifolium* i.e., 60-125 × 35-45 μm, while length of *A. alpinum* (35-65 × 15-25 μm) smallest in the genus (Table II).

Metcalf and Chalk (1950) recorded that stomata in Polygonaceae are nearly always anomocytic type in the

family Polygonaceae, infrequently paracytic as in genus *Triplaris*. However four types of stomata were recorded in genus *Aconogonon*. Stomata were restricted to abaxial surface in *A. rumicifolium* and *A. alpinum* (Table III). In *A. tortuosum* leaves were amphistomatic. Staurocytic was the most widespread type of stomata recorded in *A. tortuosum* and in *A. rumicifolium* (Fig. 1a). The staurocytic stomata type was introduced by Van Cotthem (1970). Kapoor *et al.* (1971) recorded anomocytic stomatal pattern in *A. rumicifolium*. *A. alpinum* was unique in having cyclocytic stomata with striations (Fig. 1b). Inamdar (1971) reported striated stomata in *Persicaria chinensis*. Ayodele and Olowokudejo (2006) reported cyclocytic stomata in *P. salicifolia* ssp. *salicifolia*. Quantitatively diameter of all stomatal types was same i.e., 10-15 μm, while stomatal length varies from 12.5-25 μm (Table II).

Presence of glandular and non-glandular trichomes

Fig. 1a-m. Distribution of various stomata and trichomes in two genera of family Polygonaceae. Microscopic magnification for a, b, d, f, h & m 1000X, for c, k & l, 100X; for e, g, i, & j 400X. For description of figures, see text

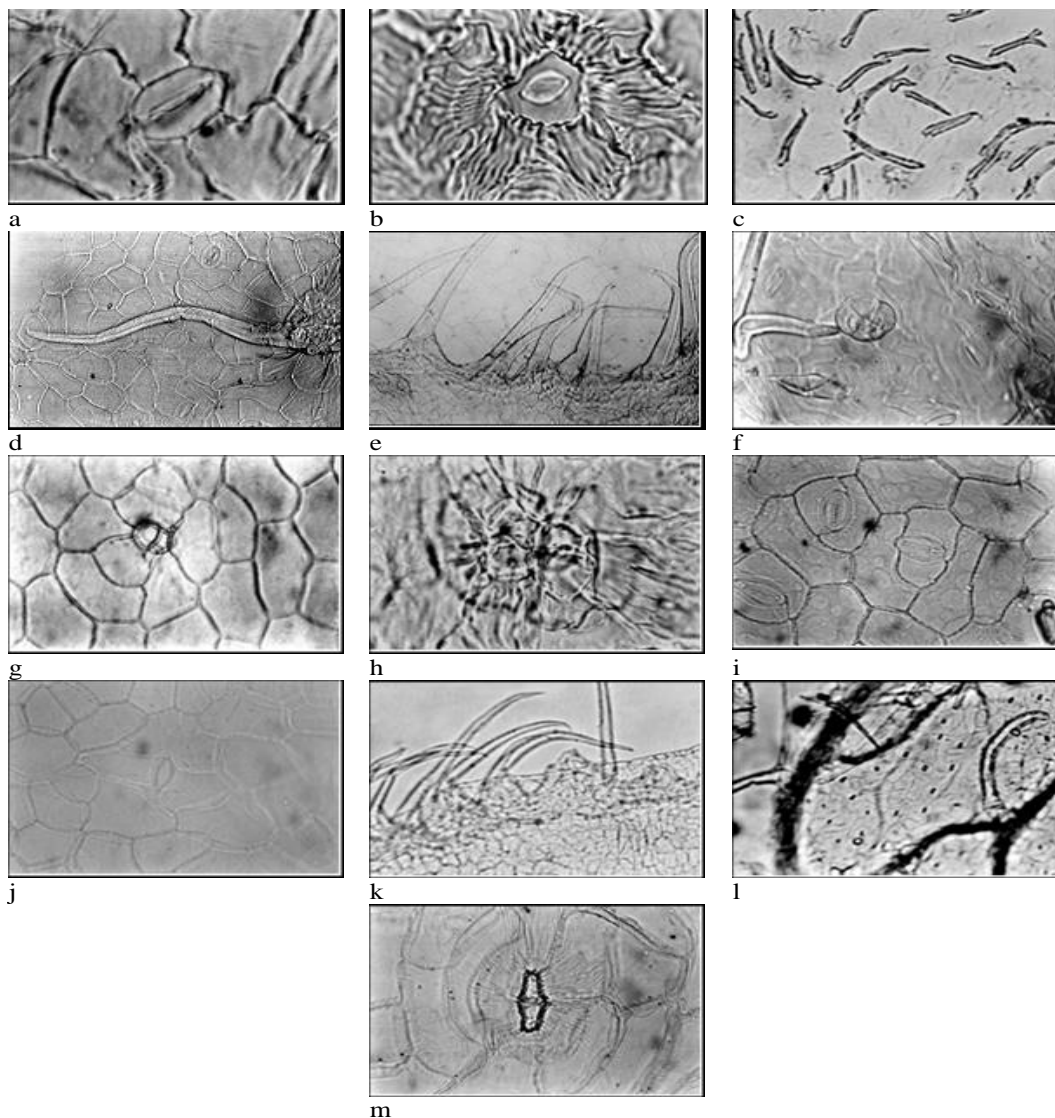


Table I. List of species investigated, with location of voucher specimens arranged in alphabetical order of taxa

Species	Locality	District	Voucher No.
<i>Aconogonon alpinum</i>	Chambhari (Stony soil)	Muzaffarabad	99632
	Dir	Dir	41998
<i>A. rumicifolium</i>	Mount Makra (Northern side)	Hazara	940044
	Mount Makra (Southern side)	Hazara	92943
<i>A. tortuosum</i>	Naltar	Gilgit	116866
	Ghanche Nallah (rocky soil)	Skardu	63901
<i>Bistorta affinis</i>	Deosai (rocky soil)	Skardu	66348
	Satpura pass	Baltistan	39025
<i>B. amplexicaulis</i>	Suden gali	Muzaffarabad	09315
	Mekay near Parachinar	Kurram agency	09322
<i>B. vivipara</i>	Deosai top	Skardu	67809
	Mount Makra (Southern side)	Hazara	96758

Table II. Quantitative leaf epidermal characteristics of *Aconogonon* and *Bistorta* (Polygonaceae)

Species	Ordinary epidermal cells		Stomata		Trichomes			
	Adaxial	Abaxial	Adaxial	Abaxial	Non glandular		Glandular	
					Length (µm)	Width (µm)	Length (µm)	Width (µm)
<i>Aconogonon</i>								
<i>A. alpinum</i>	35-65/ 25-70	15-25/ 15-35	Absent/ 20-25	Absent/ 10-15	Absent	Absent	Two-celled peltate: Absent/25-50 Four-celled peltate: Absent/30-40	Two-celled peltate: Absent/15-35 Four-celled peltate: Absent/10-20
<i>A. rumicifolium</i>	60-125/ 35-50	35-45/ 20-25	Absent/ 12.5-25	Absent/ 10-15	160-250/ 90-160	25-35/ 10-20	Two-celled peltate: 15-30 20-25	Two-celled peltate: 10-25/15-20
<i>A. tortuosum</i>	25-70/ 30-45	20-35/ 25-30	15-25/ 20-25	10-15/ 10-15	55-315/ 65-250	10-20/ 10-20	Spheroidal: 10-15/10-15	Spheroidal: 5-10/5-10
<i>Bistorta</i>								
<i>B. affinis</i>	40-70/ 25-50	20-30/ 20-35	Absent/ 20- 25	Absent/ 10-15	Absent	Absent	One-celled peltate: 10-25/10-20	One-celled peltate: 10-15/7.5-10
<i>B. amplexicaulis</i>	35-60/ 25-70	20-35/ 15-35	20-25/ 15-25	10-15/ 10-20	175-195/ 70-145	20-30/ 10-15	Two celled-20/15-20	Two-celled peltate: 15/5
<i>B. vivipara</i>	40-75/ 35-90	30-45/ 20-45	30-35/ 25-30	10-20/ 15-20	Absent	Absent	Two-celled peltate: 15-20/15-20/	Two-celled peltate: 15- 20/10-20

Table III. Qualitative leaf epidermal characteristics of *Aconogonon* and *Bistorta* genera (Polygonaceae)

Species	Ordinary epidermal cells (Adaxial/Abaxial)	Stomata (Adaxial /Abaxial)	Trichomes (Adaxial /Abaxial)	
			Non-glandular (Adaxial /Abaxial)	Glandular (Adaxial /Abaxial)
Genus <i>Aconogonon</i>				
<i>A. alpinum</i>	Polygonal /Irregular	Absent/ Pericytic, cyclocytic	Absent/Absent	Two-celled and four-celled peltate/Two-celled and four celled peltate trichome
<i>A. rumicifolium</i>	Tetrahedral, polygonal and hexagonal/Irregular	Absent/ Staurocytic	Long hair like with broad base/ Same as on adaxial surface	Two-celled peltate/Two-celled peltate
<i>A. tortuosum</i>	Polygonal /Irregular	Staurocytic, anisocytic/ Staurocytic, anisocytic	uniseriate, long with swollen base/Same as on adaxial surface	Spheroidal/ Spheroidal
Genus <i>Bistorta</i>				
<i>B. affinis</i>	Polygonal/Polygonal	Absent/Paracytic	Absent	One-celled peltate/One-celled peltate
<i>B. amplexicaulis</i>	Polygonal/Polygonal	Anomocytic, staurocytic /Anomocytic, staurocytic	Hair like, with or without basal septa/Same as on adaxial surface	Two-celled peltate/Two-celled pelt.
<i>B. vivipara</i>	Polygonal/Polygonal	Paracytic/Paracytic	Absent	Two-celled peltate/Two-celled peltate

was an important feature for the identification and classification of taxa (Munsif *et al.*, 2007). Trichomes are widespread in Polygonaceae (Metcalfe & Chalk, 1950; Lersten & Curtis, 1992). Glandular and non-glandular both kinds of trichomes were noted in *Aconogonon* spp. (Table III). Thick uniseriate non-glandular hairs with swollen base

and tapering apex were seen in *A. tortuosum* and *A. rumicifolium* (Fig. 1c-f), which were absent in *A. alpinum*. Lersten and Curtis (1992) noted same kind of thick hairs in *A. tortuosum*, *A. rumicifolium* (Kapoor *et al.*, 1971 also reported their presence) and *A. alpinum*. Largest hair were recorded in *A. rumicifolium* on adaxial surface, which

ranged in size from 160-250 × 25-35 µm and smallest on abaxial surface of the same species i.e., 90-160 × 10-20 µm (Table II).

Among the glandular trichomes, common type were peltate, which were centrally two-celled in *A. rumicifolium* (Fig. 1g & Table II) and centrally two and four-celled peltate (Fig. 1g & h) trichomes were observed in *A. alpinum*, where they were more frequently distributed on adaxial surface. Spheroidal glands were seen only in *A. tortuosum* (Fig. 1f), while Lersten and Curtis (1992) observed only capitate trichomes. Mitchell (1971) recognized spheroidal glands as multicellular plate glands on abaxial surface of aquatic species of Polygonaceae.

Centrally two-celled peltate trichomes ranged in size from 10-50×10-35 µm, while four-celled 25-40 × 10-25 µm (Table II). Multicellular spheroidal glands were smaller in size than that of peltate trichome i.e., 10-15 × 5-10 µm.

Key to *Aconogonon* species:

1a: Epidermal cells on adaxial leaf surface tetrahedral, hexagonal and polygonal, only two-celled peltate trichomes present.
1. *A. rumicifolium*.

1b: Epidermal cells on adaxial leaf surface polygonal, peltate trichomes two, four-celled and spheroidal type
2.

2a: Non-glandular trichomes absent, stomata pericytic and cyclocytic.
2. *A. alpinum*.

2b: Non glandular trichomes present, stomata anisocytic and staurocytic.
3. *A. tortuosum*.

***Bistorta*.** The comprehensive survey made by Lersten and Curtis (1992) on foliar anatomy of *Polygonum* (Polygonaceae), genus *Bistorta* was studied under one of the section of *Polygonum*. The epidermal cells were polygonal, smooth and thick walled on adaxial and abaxial leaf surfaces (Table III). Kapoor *et al.* (1971) noted polygonal cells in *B. affinis*. Smallest epidermal cells in length were seen in *B. affinis* abaxial surface i.e., 25-50 µm, while width of cells in *B. affinis* abaxial surface and *B. amplexicaulis* abaxial surface was same i.e., 20-35 µm (Table II).

In *B. affinis* leaves were hypostomatic, also observed by Kapoor *et al.* (1971). Inamdar (1971) reported the amphistomatic leaves, while other species of the genus under observation were amphistomatic, with few stomata on adaxial surface (Table III). Three different stomatal types were noted in *Bistorta* spp. (Fig. 1a, i & j), most common of which was paracytic type. Kapoor *et al.* (1971) and Inamdar (1971) observed paracytic stomata along with anomocytic and anisocytic type in *B. amplexicaulis*. Mitchell (1971) reported anomocytic and anisocytic stomatal types in *B. vivipara*. Quantitatively stomata were of same size in *B. affinis* abaxial surface and *B. amplexicaulis* adaxial surface i.e., 20-25 × 10-15 µm (Table II).

Thick walled unicellular and bicellular non-glandular trichomes were seen in *B. amplexicaulis* (Fig. 1k & l), larger in size on adaxial surface and smaller on abaxial surface.

Mitchell (1971) noted the presence of unicellular hair in *B. vivipara*. Haraldson (1978) provided most comprehensive description of non-glandular trichomes in Polygonaceae. Lersten and Curtis (1992) recorded only glandular trichomes in *B. amplexicaulis*.

Two types of peltate trichomes were seen in *Bistorta* spp. (Table III). In *B. vivipara* two-celled peltate trichomes were seen with long spreading striations (Fig. 1m). However, Ayodele and Olowokudejo (2006) recorded eglandular trichomes covered with striae in *Oxygonum sinuatum*. Mitchell (1971) observed two-celled peltate trichome without striations in *B. vivipara*. One-celled peltate trichomes were seen in *B. affinis* (Kapoor *et al.*, 1971, reported their presence). Quantitative data indicated that in *B. vivipara*, length and width of trichome was nearly same i.e., 10-25 × 10-22.5 µm (Table II).

Key to *Bistorta* species on the basis of foliar anatomy:

1a: Non-glandular trichomes in the form of hair like structure, which was basally septate or not, stomata anomocytic and staurocytic.
1. *B. amplexicaulis*.

1b: Non-glandular trichomes absent, paracytic stomata present
2.

2a: One-celled peltate trichomes present, leaves hypostomatic.
2. *B. affinis*.

2b: Two celled peltate trichomes present, leaves amphistomatic.
3. *B. vivipara*.

CONCLUSION

This foliar anatomical study revealed a number of vital micromorphological and anatomical characters, which were non-decisive. However, there were some differences, which were helpful in the recognition of taxa. New types of stomata are reported in present work such as cyclocytic, staurocytic and pericytic types. Results showed that different genera cannot be distinguished on the basis of trichomes type. Quantitative data served as diagnostic tool to some extent but it could not be used to distinguish different taxa of Polygonaceae.

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