

Epidermal Studies in some Members of *Andropogoneae* (*Poaceae*)

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Abstract

The aim of this study was the micromorphology of the members of *Andropogoneae* (*Poaceae*) and to prepare the identification key using micromorphological features. Members of *Andropogoneae* were collected from the Navasari and surrounding areas, a district of Gujarat and scrutinized for detailed micromorphological study. Epidermal studies pertain to epidermal cell size, epidermal frequency, stomatal frequency and index, presence of micro-hairs, papillae, its frequency and types of silica bodies of 5 taxa belonging to tribe *Andropogoneae* of family *Poaceae* have been presented. All the 5 studied members show the variation in the arrangement of the epidermal cells in both the epidermises. The observed uniformity in certain epidermal characters and presence or absence of prickles, silica bodies, papillae and stomatal pattern helps in identification of presently studied 5 taxa

Keywords: *Andropogoneae*, micromorphology, micro-hairs, papillae, prickles, silica bodies

Introduction

Grasses covers about one fifth of the earth and has a great economic significance and ecological dominance (Shantz, 1954). Varieties of grass-dominated ecosystems are present on the surface of the mother Earth and grasslands are the signature of the family all over the globe. Taxonomically family *Poaceae* is one of the largest family having the 908 genera and more than 11,000 species (GPWG, 2001; Tzevlev, 1989). Epidermal micromorphology of the leaves is used in emphasizing the interrelationships and segregation into major clades. Metcalfe (1960), Stebbins and Khush (1961) have supported the earlier grouping for various taxa from different families of plant systematics. Epidermal micro characters are quite important to delineate the different taxa in terms of phylogenetic and taxonomic considerations. Angiosperms leaves are the most suited and studied for these purpose (Watson 1990). Several authors recognized the importance of these epidermal features in recognizing the different taxa belonging to different families (Inamdar *et al*, 1988; Parveen *et al*, 2000; Edeoga and Ikem, 2001; Stebbins and Khush, 1961; Rammayya and Rao 1987). More importantly this study may be useful to locate the markers present within the circumscription of the micromorphology (Hilu, 2007). Clayton and Renvioze (1986); Amarasinghe and Watson (1990) and Hilu (2006, 2004) utilized epidermal characters for understanding the interrelationship of the various taxa.

Indeed the foliar epidermal characters of the angiosperms depict the sufficient diversity of details due to its genetic and environmental makeup. *Poaceae* is the most widely observed family from arctic to seashore and from

wetlands to arid region. Micromorphological characters are valuable for systematic studies in the family *Poaceae*. Numerous reports on foliar anatomy and micro morphology are used for the delimiting the different groups and specifically subfamilies or tribes in family *Poaceae* (Brown 1958, Ellis 1987, Davlia and Clarke 1990, Amarasinghe and Watson, 1990). Hilu (1984), Clayton and Renvioze (1986), Whang and Kim (1994) have described the efficacy and characterization of epidermal studies for the members of the grasses.

Besides only epidermal characters of leaf and stem, other features like micro-hairs, papillae and silica bodies have been considered of significance in segregating the taxa at various levels. For example, the absence of microhairs is characteristic in *Pooideae* (Clayton and Renvioze, 1986). Jhonston and Watson (1976) described the presence of microhairs as a universal characteristic feature of the non-festucoid grasses. Character variability and ornamentation of the leaf epidermis are also of taxonomic value (Prat, 1932; Metcalfe, 1960; Amarasinghe and Watson, 1990), being especially useful for the characterization of the larger groups, particularly subfamilies and tribes.

Not only leaf but, the floral bracts of the grasses and its microscopic characters have been used to assess systematic relationships, as well as evolutionary trends (Acedo and Lamas, 2001). In the case of *Eragrostis* microhairs consider as characterization of lemmas and palae of grasses analyzing the value of their microcharacters for an infrageneric classification (Thomasson, 1986; Amarasinghe and Watson, 1990).

Earlier silica bodies and their structure and composition have been used for the differentiation between the various

grasses from the world. Leaf epidermal studies are important in segregating the different broad groups within the grasses particularly tribes and subfamilies and even certain extent the genera (Ellis 1987, Davalia and Clark 1990). Whang *et al.* (1998) has described and given variation and morphometric analysis of different species of *Oryza*. Krishnan *et al.* (2000) has utilized for the identification of grasses from the Tamilnadu, India and Lu and Liu (2003a) given the comparison of Chinese and American grasses.

Earlier reports suggest that there may be variation amongst the species and genera but, it cannot be assumed that it will be true for of the herbs and grasses from different habit and habitat (Joshi and Janarthanam, 2004). In view of the above mentioned reports and the importance of the foliar epidermal studies for the grasses, the results depicted herein are the potential source for taxonomic considerations. Perusal of literature also depicted the lack of information on these taxa for the detail studies

Materials and methods

The present work describes the micromorphological characters of the 5 taxa belonging to tribe Andropogoneae collected from the Navasari district of Gujarat. Collected plants are preserved and mounted on the herbarium sheets, submitted to the BARO Herbarium (Department of Botany, Faculty of Science, The Maharaja Sayajirao University of Baroda, Vadodara, Gujarat, India). Voucher details of the sample examined in this study is given in Tab. 1. For micromorphological study, the leaves from the middle of the culms (3rd and 5th) were used throughout the preparation. The peels were made by scraping pieces of fresh or softened dried leaves (Glycerin : Water mixture) with the help of safety razor blade and stained it with the saffranin or Delafield's Haematoxylin stain and phenol to be mounted in glycerin (Hilu & Randall, 1984; Johnston and Watson, 1976).

Adaxial and abaxial leaf surfaces from all the five taxa were studied at $\times 400$ magnification and individual cells were identified and measured by micrometer. 20-25 peels were made from each species of several dozen of leaves. All the peels were examined and the representative areas were photographed using Leica research microscope using $\times 40$ objective. Final counts of different cells (average of 50 observations) summarized in the tables.

Results

Tab. 1. Voucher details of Sample examined in this study

No.	Species Name	Collected Area	Herbarium No. in BARO
1	<i>Cymbopogon jwarancusa</i> (Jones.) Schult.	Matwad, Navasari	RJD/35
2	<i>Cymbopogon schoenanthus</i> (L.) Spr.	Vansda, Navasari	RJD/43
3	<i>Ischaemum pilosum</i> (Klein ex Willd.) Wt.	Aru, Navasari	RJD/37
4	<i>Rottboelia exaltata</i> Linn.f.	Aru and Vansda, Navasari	RJD/40 & 48
5	<i>Sorghum halepense</i> (L.) Pers.	Samapore, Navasari	RJD/54

The species wise details of the epidermal complex and epidermal ornamentation are as under:

1. *Cymbopogon jwarancusa* (Jones.) Schult.

Syn. Andropogon jwarancusa Jones.

Long cells broadly oblong, of constant width, 15-20 \times 4-5 μ , usually sinuous walled, sparsely papillate, contiguous. The exodermic papillae small, 2 \times 2 μ , one per each cell. Short cells absent in the intercostal region. Prickle hairs present, variable in size, i.e. upper epidermis with 40-50 \times 4-5 μ where as lower epidermis with 7-8 \times 4-5 μ . The leaf was amphistomatic & Stomata of upper and lower epidermis usually in a single row, usually alternate with the interstomatal cells, equal in width to the interstomatal cells, 6-10 \times 6-8 μ , subsidiary cells usually dome shaped, quite simple & calculated stomatal index for the upper epidermis & lower epidermis was 0.33 & 0.31 respectively. Silica bodies longitudinal, dumbbell shaped. Micro-hairs usually bicellular type, slender with the apical cell (12 \times 1 μ) equal to the basal cell (10 \times 2 μ), 7-8 \times 2-3 μ in lower epidermis & 20-22 \times 2-3 μ in upper epidermis having Width/Length ratio 0.25 & 0.09 μ respectively. (Fig. 1 a & b)

2. *Cymbopogon schoenanthus* (L.) Spr.

Syn. Andropogon schoenanthus L.

Long cells narrowly oblong, of constant width, 35-40 \times 4-5 μ , usually sinuous walled, irregularly interspersed with short cells, densely papillate. The exodermic papillae small, 6-8 per cell, variable in size, i.e. 1 \times 1 μ present in the upper epidermis & of 2 \times 2 μ found in the lower epidermis. Short cells present solitary, an associated with silica-containing cells. Prickle hairs present in the upper epidermis only and are of 10-15 \times 4-5 μ . The leaf was amphistomatic and stomata of upper and lower epidermis usually in a single row, usually alternate with the interstomatal cells, 8-9 \times 5-6 μ , equal in width to the interstomatal cells, subsidiary cells usually triangular shaped, Stomata are quite simple & calculated stomatal index for the upper epidermis & lower epidermis was 0.10 & 0.24 respectively. Silica bodies longitudinal, dumbbell shaped. Bicellular Micro-hairs usually slender with the apical cell (7 \times 2 μ) equal to the basal cell (7 \times 2 μ), 14-15 \times 2-3 μ with Width/Length ratio 0.14. (Fig. 1 d)

3. *Ischaemum pilosum* (Klein ex Willd.) Wt.

Tab. 2. Comparative micromorphology of five members of Andropogoneae

Species name Character	<i>Cymbopogon jwarancusa</i> (Jones.) Schult.	<i>Cymbopogon schoenanthu</i> (L.) Spr.	<i>Ischaemum pilosum</i> (Klein ex Willd.) Wt.	<i>Rottboelia exaltata</i> Linn.f.	<i>Sorghum halepense</i> (L.) Pers.
	Long cell				
Size (μ)	15-20×4-5	35-40×4-5	30-40×6-7	35-40×10-12	20-25×6-7
Shape	Rectangular	Rectangular	Rectangular	Hexagonal	Rectangular
Periclinal Wall	Sinuuous	Sinuuous	Sinuuous	Smooth, sinuous & inflated	Sinuuous
	Short cell				
Intercostal Zone	Absent	Present	Present	Present	Absent
	Inter-stomatal cell				
Size (μ)	10-15×4-5	10-15×4-5	10-20×6-7	27-30×14-15	12-15×10-12
	Silica bodies				
Shape	Dumbbell	Dumbbell	Dumbbell	Cross - Dumbbell	Dumbbell
	Exodermic cell				
Prickle size (μ)	L.E.- 7-8 × 4-5 U.E.- 40-50 × 4-5	L.E. -absent U.E. -10-15 × 4-5	Absent	Absent	Absent
	Bicellular Microhair				
-size (μ)	L.E. 7-8 × 2-3 U.E. 20-22 × 2-3	14-15 × 2-3	9-10 × 3-4	12-13 × 3-4	10-12 × 2-3
-W/L ratio	L.E. - 0.25 U.E. - 0.09	0.14	0.30	4.0	4.5
Papillae	Present	Present	Absent	Absent	Absent
	Stomata				
Cuticularization	Sunken	Simple	Simple	Sunken	Simple
Stomatal index of L.E.	0.31	0.24	0.10	0.28	0.20
Stomatal index of U.E.	0.33	0.10	0.31	0.17	0.21

Syn. Andropogon pilosus Klein ex Willd.

Long cells narrowly to broadly oblong, 30-40 × 6-7 μ , of constant width, usually sinuous walled, cells smooth, contiguous or irregularly interspersed with short cells. Short cells present, solitary, without an associated silica-containing cell. Prickle hairs absent. The leaf was amphistomatic and stomata of lower epidermis usually in a single row, of upper and lower epidermis usually in a single row and sometimes in two rows, usually alternate with the interstomatal cells, rarely scattered; equal in width to the interstomatal cells, 12-13 × 7-8 μ , subsidiary cells usually triangular in shape and show prominent silica deposition at the tip of the cell, quite simple & calculated stomatal index for the upper epidermis & lower epidermis was 0.31 & 0.10 per mm respectively. Silica bodies longitudinal, dumbbell shaped. Micro-hairs mostly bicellular, 9-10 × 3-4 μ , slender with the apical cell (15 × 2 μ) much longer or equal to the basal cell (10 × 3 μ) having 0.30 Width/Length ratio. (Fig. 1 g)

4. *Rottboelia exaltata* Linn.f.

Long cells hexagonal, of constant width, 20-25 × 6-7 μ , usually smooth-slightly sinuous-inflated walled, cells smooth, contiguous or irregularly interspersed with short cells. Short cells present in the intercostal zone, solitary, without an associated silica-containing cell. Silicified prickle hairs present at the margin of the leaves with prominent basal supporting cells. The leaf was amphistomatic and stomata of upper and lower epidermis usually in a single row, usually alternate with the interstomatal cells, 12-13 × 12-13 μ , equal in width to the interstomatal cells, subsidiary cells usually dome shaped, sunken and calculated stomatal index for the upper epidermis & lower epidermis was 0.17 & 0.28 per mm respectively. Silica bodies longitudinal, cross-dumbbell shaped. Bicellular micro-hairs usually slender, 12-13 × 3-4 μ , with the apical cell (5 × 3 μ) much longer than the basal cell (9 × 2 μ) having Width/Length ratio 4.0. (Fig. 1 c, e & f)

5. *Sorghum halepense* (L.) Pers.

Syn. Andropogon halepensis Brot.

Holcus halepensis L.

Long cells broadly oblong, $20-25 \times 6-7 \mu$, of constant width, usually sinuous walled, cells smooth, contiguous. Short cells were totally absent in the intercostals region. Prickle hairs absent. The leaf was amphistomatic and stomata of lower epidermis usually in a single row; of the upper epidermis usually in two rows; usually alternate with the interstomatal cells, rarely scattered; $9-10 \times 6-7 \mu$, equal in width to the interstomatal cells, subsidiary cells usually triangular shaped and shows the presence of papillae in the center, quite simple & calculated stomatal index for the upper epidermis & lower epidermis was 0.21 & 0.20 per mm respectively.

Silica bodies longitudinal, dumbbell shaped. Microhairs usually bicellular, $10-12 \times 2-3 \mu$, slender with the apical cell ($10 \times 2 \mu$) much longer or equal to the basal cell ($5 \times 2 \mu$) with 4.5 Width/Length ratio. (Fig. 1 h)

Discussions

The present study revealed that micromorphological features of the leaf surfaces have a considerable value in identification. The observed characters study yielded data which can be considered as marker or biomarker. The present study recorded features which are in accordance with the earlier reports (Metcalf and Clifford, 1968; Renvoize, 1982; Davlia and Clark, 1990; Hilu, 1984). However, few features from the study are described in detail and some features are not reported earlier.

Analysis of the presently studied members of Tribe *Andropogoneae* depicted that, the long cells were of rectangular, smooth and sinuous type in all the members, except *Rottboelia* wherein cells were hexagonal, smooth and inflated type. The short cells were found only in *Rottboelia* and mostly alternating with long cells. The costal zone depicts presence of short cells alternating with silica cells in all the members.

The measured size of interstomatal cells was variable (Tab. 2). Stomatal nature and stomatal index has also been observed as well as calculated and presented in Tab. 2. Out of 5 members, only *Rottboelia exaltata* and *Cymbopogon jwarancusa* showed presence of sunken stomata and rest other with simple stomata.

In the terms of exodermic cells prickles and macrohairs were not recorded in almost all the members, except *Cymbopogon schoenanthus* and *Cymbopogon jwarancusa* in which prickles were recorded with variable size. The exodermic papillae were recorded from *Cymbopogon schoenanthus* and *Cymbopogon jwarancusa* only, with variable size (Tab. 2). All the five members depicted bicellular type microhairs, which is in accordance with the earlier report (Tateoka *et al.*, 1959; Renvoize, 1982). The largest width/

length ratio has been reported from *Sorghum halepense* and smallest from upper epidermis of *Cymbopogon jwarancusa*.

Cork cells showed the silica deposition of Dumbbell shapes in all taxa. Along with that only *Rottboelia exaltata* depicted Cross shaped. By and large, the morphophytes are the result of the silica deposition in the plant cell due to physiological and environmental factors (Whang *et al.*, 1998; Lu and Lui, 2003b).

Watson *et al.* (1985) and Clayton (1981) have described the interrelationship of the family Poaceae. In the present paper we have tried to understand the interrelationship between the micromorphological patterns among the five members of the Andropogoneae. On the basis of presence or absence of Prickles, Silica bodies, Papillae and Stomatal pattern presently studied 5 taxa can be classified as follows:

Prickles present, Silica body Dumbbell – Cross-shaped

Papillae present, Prickles laminar, Silica body Dumbbell shaped only

Stomata present only in the single file between two successive veins

- *Cymbopogon jwarancusa* (Jones.) Schult.

Stomata present in the two file between two successive veins

- *Cymbopogon schoenanthus* (L.) Spr.

Papillae absent, Prickles marginal, Silica body Cross-Dumbbell shaped

- *Rottboelia exaltata* Linn.f.

Prickles absent, Silica body Dumbbell shaped only

Two Stomata file present successively

- *Sorghum halepense* (L.) Pers.

Two Stomata file separated by 4-5 files of long cells

- *Ischaemum pilosum* (Klein ex Willd.) Wt.

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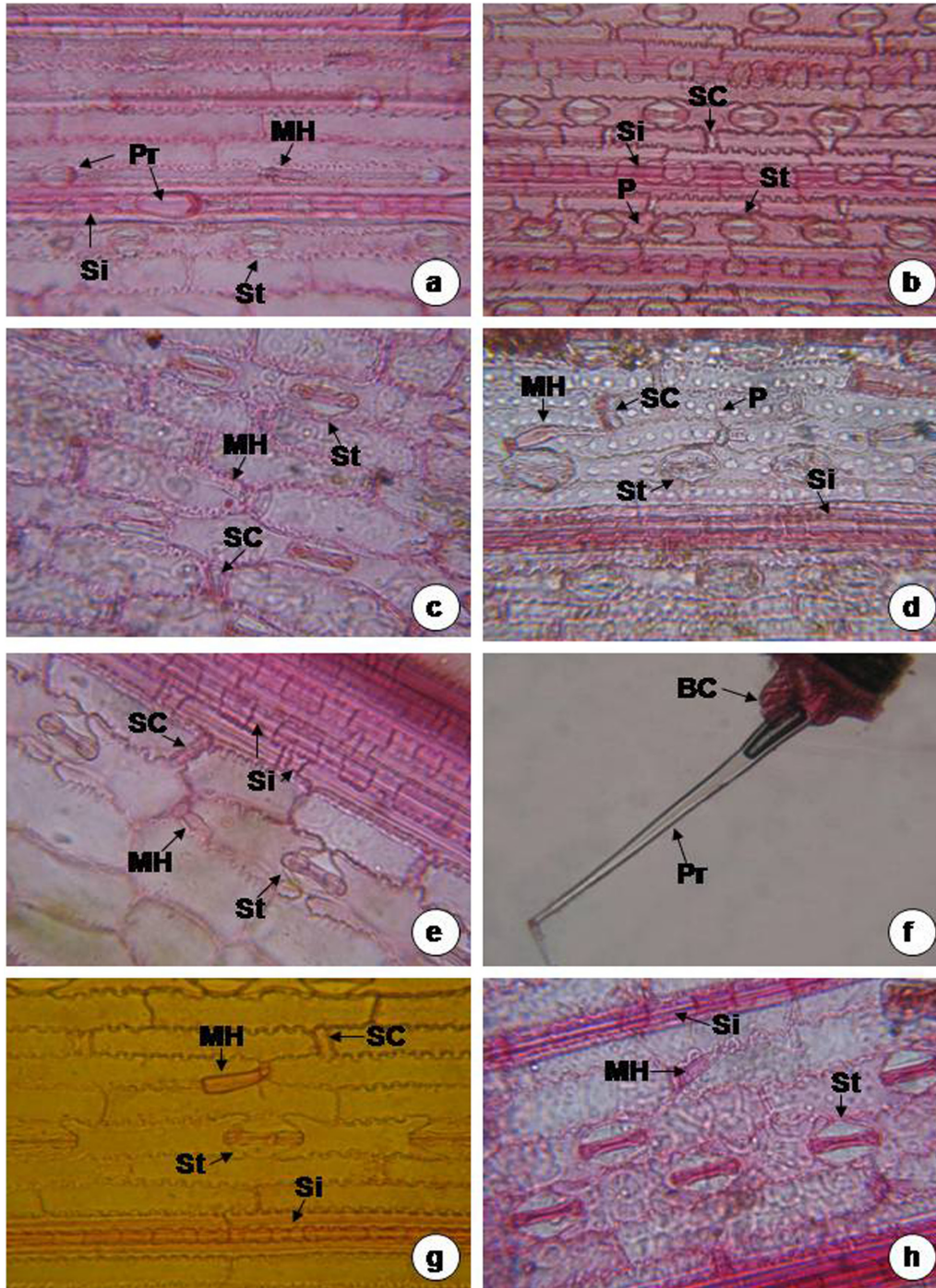


Fig. 1.

- a. Lower Epidermis of *Cymbopogon juarancusa* (Jones.) Schult. showing Stomata, Micro-hairs, Silica bodies & Prickles ($\times 400$).
 - b. Upper Epidermis of *Cymbopogon juarancusa* (Jones.) Schult. showing Stomata, Silica bodies & Short cells ($\times 400$).
 - c. Lower Epidermis of *Rottboelia exaltata* Linn.f. showing Stomata, Micro-hairs & Short cells ($\times 400$).
 - d. Lower Epidermis of *Cymbopogon schoenanthus* (L.) Spr. showing Stomata, Micro-hairs, Silica bodies, Papillae & Short cells ($\times 400$).
 - e. Upper Epidermis of *Rottboelia exaltata* Linn.f. showing Stomata, Micro-hairs & Silica bodies ($\times 400$).
 - f. Margin of leaf of *Rottboelia exaltata* Linn.f. showing Silicified Prickle with its Basal supporting cells ($\times 100$).
 - g. Lower Epidermis of *Ischaemum pilosum* (Klein ex Willd.) Wt. showing Stomata, Micro-hairs, Silica bodies & Short cells ($\times 400$).
 - h. Upper Epidermis of *Sorghum halepense* (L.) Pers. showing Stomata, Micro-hair & Silica bodies ($\times 400$).
- (Legends: Si: Silica bodies, St: Stomata, SC: Short cell, MH: Micro-hair, BC: Basal cells, Pr: Prickle, P: Papillae).