Diversity in foliar stomatal types in some members of Verbenaceae

*Shubhangi N. Ingole and **G.V. Patil

*Department of Botany B.R.D.I.K. College, Badnera, Amravati-444 602
**Amravati University. Amravati-444 602

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The present investigation incorporates the study of foliar stomatal complex in 20 taxa belonging to 12 genera of the family Verbenaceae. Both amphistomatic and hypostomatic leaves have been observed. In majority of genera more than two types of stomata are found on same surface of the same leaf. However, in most cases a particular type predominates the other type. Abnormal stomatal types have been observed in some taxa which are recognized as contiguous tomata, half stomata, arrested stomata and few others. Hence, diversity is observed not only in the external morphological features of species but also in stomata. These patterns are species specific of which some are predominant.

Key-words—Stomatal types, Abnormal stomata, Verbenaceae.

INTRODUCTION

THE study of morphology and ontogeny of the stomatal complexes in leaves has been one of the fruitful areas of research. Several classifications and types are recognized in stomatographic studies of flowering plants by Vesque (1889), Florin (1931, 1933), Smith (1935), Metcalfe and Chalk (1950, 1961), van Cotthem (1970), Dilcher (1974), Baranova (1981, 1986, 1992), Christophel *et al.* (1996) and others.

Different types of stomata have been reported on the same surface of an organ in diverse angiospermic families (Tognini 1897, Loftfield 1921, Metcalfe & Chalk 1950, Sen 1958, Pant & Kidwai 1964, Paliwal 1965, Pant & Mehra 1965, Inamdar & Patel 1971, Bahadur *et al.* 1971). The epidermal characters are used for distinguishing certain groups of plants (Stace 1965, Ramayya & Rajagopal 1968).

Study of foliar stomatal complex in 20 taxa belonging to 12 genera of Verbenaceae is presented here.

MATERIAL AND METHOD

The genera and species collected from different habitats and localities including Melghat forest area of Amravati district (Maharashtra) are listed in Table 1. The taxa are arranged according to Bentham and Hooker system.

To study the stomatal complex, epidermal peeling of leaves was done mechanically by forcep or by scrapping with the help of razor blade, hard and difficult materials were peeled off after treating with 5% aqueous sodium hydroxide. Cleared parts were washed with distilled water and treated with 2% acetic acid for 1-2 hours to neutralize residual sodium hydroxide. Finally, the prepared peels were stained with 1% aqueous safranin followed by mounting in 50% glycerine and ringed with tar or nail polish.

For each species, analysis of stomatal complex was made from random samplings of 10-different peels on either surfaces. Characters of adjacent epidermal cells were also studied.

Epidermal peels were studied from mature leaves of both fresh and herbarium materials. The latter were softened by soaking in warm water. Stomatal classification is based on the morphological classification proposed by Baranova (1992).

OBSERVATION

Present study includes the examination of amphistomatic and hypostomatic leaves of the taxa. It

Table 1. Plants collected from Amravati district, Maharashtra

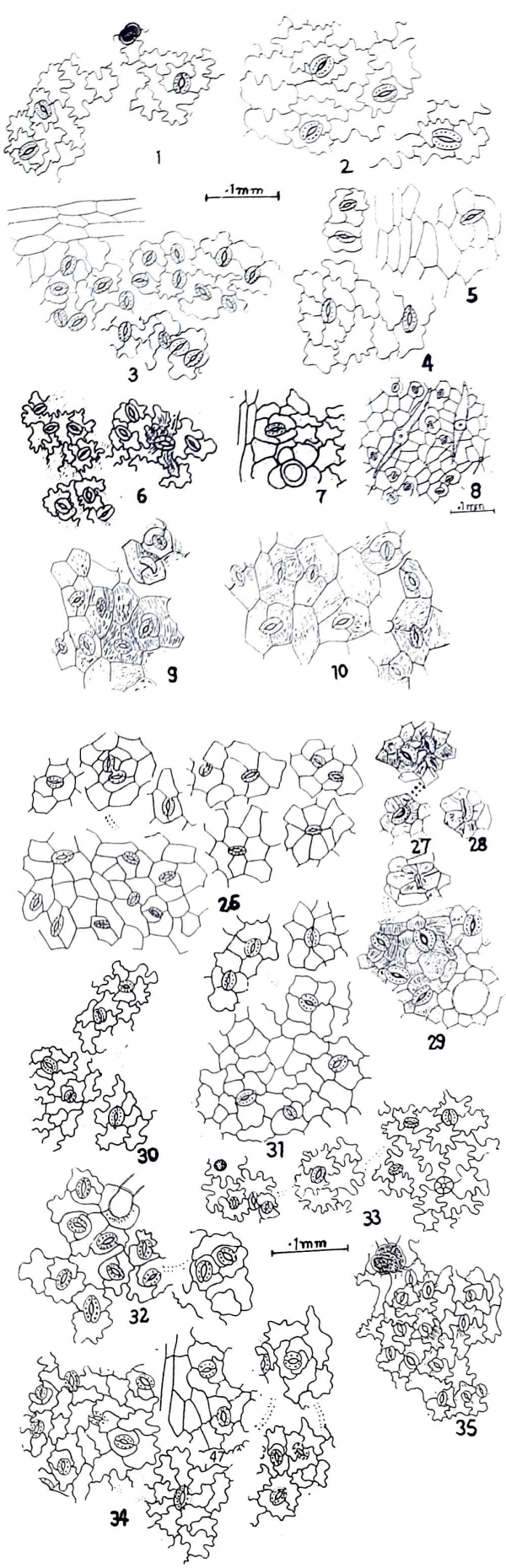
Name of Taxa	Habit
Lantana camara var. *aculeata*	Straggling shrub
Lantana camara var. *flava*	Small shrub
Lantana camara var. *nevea*	Shrub
Phyla nodiflora	Prostrate herb
Stachytarpheta jamaicensis	Herbaceous undershrub
Stachytarpheta mutabilis	Undershrub
Verbena bipinnatifida	Prostrate decumbent herb
Petrea volubilis	Liana
Duranta erecta (*Duranta repens*)	Bushy shrub with drooping Brancehs
Tectona grandis	Tree
Gmelina arborea	Tree
Gmelina philippensis	Thorny sprawling shrub with drooping branches
Vitex negundo	Small tree
Clerodendrum inerme	Straggling shrub
Clerodendrum multiflorum.	Scrambling shrub
Clerodendrum serraium	Shrub
Clerodendrum splendens	Scandent shrub
Clerodendrum viscosum	Gregarious shrub
Holmskioldia sanguinea	Straggling shrub
Nyctanthes arbor-tristis	Small tree

has been observed that majority of genera show more than one or two types of stomata on same surface of the same leaf, however, in some plants a particular type was found dominant. In *Lautana camara* var. *aculeata* diacytic and anomocytic stomata are found (Text figs 1, 2). In *L. camara* var. *flava* and *L. camara* var. *nivea* diacytic, anomocytic and anisocytic types (Text figs 8-10), in *Stachytarpheta jamaicensis* and *S. mutabilis* diacytic, anomocytic (Text figs 11-14), in *Verbena bipinnatifida* anomocytic and anisocytic (Text figs 15, 16), in *Petres volubilis* diacytic (Text fig 17), in *Dweanta erecta* anisocytic, anomocytic, tetracytic, actinocytic (Text figs 18-20), in *Gmelina arborea* anomocytic, anisocytic (Text fig 21), in G. *philippensis* anisocytic, anomocytic, actinocytic, tetracytic, diacytic (Text fig 26), in *Vitex negundo* anisocytic (Text fig 25), in *C. inerme* anomocytic (Text figs 27-29), in *C. multiflorum* anomocytic, anisocytic, actinocytic, tetracytic (Text figs 30, 31), in *C. serratum* diacytic (Text fig 32), in *C. splendens* diacytic, anomocytic (Text fig 33), in *Clerodendrum viscosum* anisocytic, diacytic, anomocytic (Text fig 35), in *Holmskioldia sanguinea* anisocytic, diacytic anomocytic, actinocytic (Text fig 34) and in *Nyctanthes arbor-tristis* anomocytic (Text figs 23-24), Stomatal types found in differential combinations on different surfaces of leaves are compiled (Table 2).

DICUSSION

Among the taxa studied, *L. camara* var. *aculeata, L. camara* var. *flava, L, camara* var. *nivea, P. nodiflora, S. jamaicensis, S. mutabilis, V. bipinnatifida* and *C. multiflorum* are characterized by amphistomatic leaves and *P. volubilis, D. erecta, T. grandis, G. arborea, G. philippensis, V. negundo, C. inerme, C serratum, C. splendens, C. viscosum, H. sanguinea* and *N. arbor-tristis* are characterized by hypostomatic leaves. Hypostomatic leaves are also reported by Sayeeduddin and Moinuddin (1939) in *H. sanguinea.* Shah and Mathew (1982) reported amphistomatic dorsiventral organs in *L. camara* var. *aculeata* and *L. camara* var. *nivea.* They observed anomocytic, diacytic, paracytic and anisocytic, tetracytic, cyclocytic and transitional forms between paracytic and diacytic types of stomata in *L. camara* and its varieties where different types of stomata occur on the same surface in different combinations. They found anisocytic dominant type. Present study carried on *L. camara* and its varieties show similar phenomenon where different forms of transitional types are observed on same surface of the organ. Predominant form is found as diacytic along with anomocytic and anisocytic types in *L. camara* var. *aculeata* whereas in *L. camara* var. *nivea* diacytic predominant type along with anomocytic and anisocytic types are found only on adaxial surface of lamina.

Various types of abnormal stomata have been observed in *S. jamaicensis, P. nodiflora, C. inerme, G. arborea, G. philippensis, H. sanguinea, N. arbor-tristis* and some others. These are categorized into major types as contiguous stomata, stomata connected by cytoplasmic bridge, stomata with single guard cell, degenerated stomata, stomata with arrested development, etc. Regarding stomatal type, in majority of genera more than two types of stomata are found



Text figs.1-35. Abaxial and adaxial laminar epidermis in surface view showing diverse stomatal types. Figs. 1, 3, 6, 8-9, 11-13, 15, 17-21, 23-30, 32-35. Abaxial epidermis. Text figs. 2, 4, 5, 7, 10, 12, 14, 16, 22, 31. Adaxial epidermis 1-2. *L. camara* var. *aculeata* showing variously oriented diacytic and anomocytic stomatal types. Diacytic stomata with unequal adjacent cells, one distinctly smaller, anomocytic stoma surrounded by 3 adjacent cells. 3-5. *L. camara* var *flava* showing intercostals and costal regions with diacytic and anomocytic stomata surrounded by 3-6 adjacent cells. 6-7. *L. camara* var. *nivea* showing diacytic, anomocytic and anisocytic types and occasional weak cuticular striations running from same. 8-10. *P. nodiflora* showing diacytic, anomocytic and paracytic types, adjacent cells in diacytic type, distinct with one of the cell arched; presence of cuticular strations, half stoma, medifixed adpressed trichomes also seen. 11-12. *S jamaicencis* showing diacytic, anomocytic, contiguous stomata with cytoplasmic bridge. 13-14. *S. mutabilis* showing anomocytic, anisocytic, half and arrested stomata. 17. *P. volubilis* showing diacytic stomata.18-20. *D. erecta* showing anisocytic, actinocyclic, anomocytic and tetracytic types. 21. *G. arborea* showing anomocytic and anisocytic types. 22. *T. grandis* showing a stomatic adaxial epidermis. 23-24. *N. arbortristis* showing anomocytic, half, abortive stomata and fine cuticular striations radiating from them. 25. *V. negundo* showing predominant anisocytic stomata. 26. *G. philippensis* showing anomocytic, actinocytic, tetracytic, diacytic and contiguous stomata with cytoplasmic bridge. 27-29. *C. inerme* showing stomata with single guard cell, contiguous stomata with cytoplasmic bridge, anomocytic, stomata, gland and strong cuticular striations. 30-31. *C. multiflorum* showing anomocytic, anisocytic, actinocytic, tetracytic types. 32. *C. serratum* showing diacytic types of stomata with equal or unequal, distinct, arched or similar adjacent cells. 33. *C. splendens* showing diacytic stomata with equal or unequal adjacent cells and anomocytic stomata. 34. *H. sanguinea* showing anisocytic, diacytic and actinocytic types. 35. *C. viscosum* showing anisocytic, diacytic, anomocytic types with thin cuticular striations running in adjacent cells along with prominent gland.

Table 2. Foliar stomatal types

Name of Taxa	Type of Leaf	Type of Stomata		Predominant Type		Abnormal/contiguous stomata	
		Abaxial	Adaxial	Abaxial	Adaxial	Abaxial	Adaxial
Lantana camara var. *aculeata*	Amphistomatic	DI, AM (SU) (3-4)	DI, AM (SE) (3-4)	DI	DI	–	–
Lantana camara var. *flava*	Amphistomatic	DI, AM, AN (SU) (3-6)	DI, AM (SE) (3-6)	DI	DI	–	Few contiguous
Lantana camara var. *nivea*	Amphistomatic	DI, AM (SU) (3-4)	DI, AN	DI, AM	DI equally predominant	Stomata with single guard cell	–
Phyla nodiflora	Amphistomatic	DI (S, D, U)	DI, PA, AM (SDU) (3)	DI	DI	–	Half stomata with single guard cell
Stachytarpheta jamaicensis	Amphistomatic	DI, AM (SU) (3)	DI, AM (SE) (3)	DI	DI	–	Abnormal pair of contiguous stomata connected by cytoplasmic bridge between adjacent cells.
Stachytarpheta mutabilis	Amphistomatic	DI, AM (SU) (3-4)	DI (SU)	DI	DI	–	–
Verbena bipinnatifida	Amphistomatic	AM (3-5)	AN, AM (3-5)	AM	AN	–	–
Petrea volubilis	Hypostomatic (D,U)	DI	–	DI	–	–	–
Duranta erecta	Hypostomatic	AN, AM, (4) TE, AC (4)	–	AN	–	Few contiguous	–
Tectona grandis	Hypostomatic						
Gmelina arborea	Hypostomatic	AM, AN (4-6)	–	AM	–	Contiguous stomata sharing common adjacent cells frequent	–
Gmelina philippensis	Hypostomatic	AN, AM (4-6) AC, TE, DI	–	AN	–	-Do-	–

Contd...

Name of Taxa	Type of Leaf	Type of Stomata		Predominant Type		Abnormal/contiguous stomata	
		Abaxial	Adaxial	Abaxial	Adaxial	Abaxial	Adaxial
Vitex negundo	Hypostomatic	AN	–	AN	–	Few contiguous stomata sharing adjacent cells frequent	
Clerodendrum inerme	Hypostomatic	AM	– (4-6)	AM	–	Single guard cell, few with abortive guard cells. Few contiguous lying side by side with cytoplasmic bridge, one of the stoma of pair, having unequal guard cells	–
Clerodendrum multiflorum	Amphistomatic	AM, AN (3-5)	AM, AN (3-5) AC, TE (8)	AM	AM	–	–
Clerodendrum serratum	Hypostomatic	DI (DUE)	–	DI	–	–	–
Clerdendrum splendens	Hypostomatic	DI, AM (SUE) (3-4)	–	DI	–	–	–
Clerodendrum viscosum	Hypostomatic	AN, DI (S,U) AM (3-4)	–	AN	–	–	–
Holmskioldia sanguinea	Hypostomatic	AN, DI (SU) AM, AC (4-5) (8)	–	AN	–	Few contiguous without cytoplasmic bridge	–
Nyctanthes arbor-tristis	Hypostomatic	AM (3-6)	–	AM	–	Half stoma with single guard cell and abortive stomata frequent	–

AM = Anomocytic, AN= Anisocytic, DI = Diacytic, PA = Paracytic, AC = Actinocytic, TE = Tetracytic, S = adjacent cells similar to adjoining epidermal cells, U = adjacent cells unequal, D = adjacent cells distinct, E = adjacent cells equal, (letters and numbers in bracket indicating characters of adjacent cells and number of adjacent cells in anomocytic

on same surface of same leaf in same species, however, many times particular type predominates the other type. Pant and Kidwai (1964) reported the occurrence of more than one type of stomata with numerous variations in the organization of the stomatal apparatus in the leaves of *P. nodiflora*. The diacytic is found as predominating type and rests as paracytic (10%) anisocytic (4%) or anomocytic (3%). Interestingly they observed transitional stomata between the diacytic and paracytic types. They also noticed abnormalities including contiguous stomata, stoma with a single guard cell and arrested stomata. The similar findings with diverse stomatal types in same leaf and same surface in *P. nodiflora* are also noticed in present work, along with abnormal stomata with a single guard cells, comparative variations are noticed more on adaxial surface. However, the predominant stomatal type on both surfaces is diacytic.

The presence of several types of stomatal apparatus in the same leaf was examined by Tognini as early as in 1897, shortly after the publication of the first classification of stomatotypes by Vesque (1889). Variation in the organization of the subsidiary cells in same leaf has also been observed by later workers (Metcalfe & Chalk, 1950; Pant & Kidwai, 1964; Inamdar, 1969; den Hartog & Bass, 1978; Shanmukh Rao & Ramayya, 1983; Padmini & Shanmukh Rao, 2001).

Baranova (1992) used the term "heterostomatic" for the taxa showing several types of stomata in a single plant. Pant and Banerji (1965) opined that even if variation is seen in the organization of stomatal complex on the leaves of a single plant, in some cases the instability itself or the range of instability afford a basis for systematic distinction between the taxa. Stebbins and Khush (1961) concluded that, despite the variability, there is characteristically a predominant stomatotype for each family, and the stomata provide a useful aid in the recognition of relationships among plants. Baranova (1992) emphasized that the taxonomic value of such leaf epidermal characters is distinct in different taxa. It is concluded that morphological classification of stomata is best suited on their appearance in the mature leaf. Fourteen morphological types of stomata are now recognized, anomocytic, anisocytic, paracytic, diacytic, actinocytic, encyclocytic (cyclocytic), tetracytic, pericytic, dermocytic, polocytic, staurocytic, hemiparacytic, laterocytic and stephanocytic. Intermediary forms occur between these types but it is not always easy to distinguish these types. Baranova (1992) outlined difficulties encountered in the utilisation of such data. She stated a basic difficulty in stomatography i.e. that they show some genetically uncontrolled variability. Sometimes the amplitude of variation can serve as basis for the systematic differentiation of taxa.

In present work, diacytic, anomocytic, anisocytic, paracytic, tetracytic, actinocytic types are found along with abnormal stomata in some taxa e.g. contiguous stomata, half stomata, arrested stomata and few others. The study suggests that species show diversity not only in external morphological features but also in stomata located on leaves. These patterns are species specific as some of the stomata types are predominant in particular taxa.

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