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Full Length Research Paper

Taxonomic treatment of medicinally important arboreal flora of tropical and subtropical region based on leaf epidermal anatomical markers

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Identification of medicinally important arboreal flora is important for sustainable and effective utilization. However phenotypic markers are insufficient for correct identification. Therefore present study was conducted for taxonomic evaluation of the selected sixteen tropical and subtropical medicinally important arboreal species, belonging to four orders and six angiosperm families based on leaf epidermal anatomical markers. The experiment was performed in the molecular taxonomy Laboratory, Department of Botany, Lahore College for Women University, Jail Road Lahore, Pakistan. The results of present investigations showed substantial variation in size, shape and arrangement of epidermal cells, stomata and trichomes. Eight different shapes of stomata; anomcytic, anomotetracytic, actinocytic, amphianisocytic, brachyparacytic, anisocytic, amphicyclocytic and stauricytic were observed. Anomotetracytic was the most dominant type of stomata observed in trees of tropical and subtropical region. Staurocytic stomata were found in *Erythrina subrosa* L. The variation was also found in number and shape of subsidiary cells and silica bodies. The present study concluded that leaf epidermal anatomical markers could be utilized significantly in delimiting the closely related taxa of medicinally important arboreal flora of tropical and sub tropical regions.

Key words: Leaf epidermal anatomy, anomyctic, anomotetracytic, actinocytic, amphian isocytic, brachyparacytic, anisocytic, amphicyclocytic, trichomes.

INTRODUCTION

Eastern medicinal system principally based on the medicinal properties of various plants species. Out of about 258,650 species of higher plants reported from the world; more than 10% are used to cure ailing communities (Shinwari, 2010). Majority of the people rely on medicinal plants to find treatment for various diseases. As herbal medicines has almost no side effect. Fortunately tropical and subtropical region is rich for the arboreal species. There are number of tree species (*Ficus infectoria, Syzgium cumini, Albizzia lebbek)* used in the preparation of various compositions of herbal medicines. However the sustainable utilization of plants depends on proper identification. Most of the taxonomic information of plants accumulated so far is based solely upon morphometry. As most of the taxonomist were agreed that similarities and dissimilarities of plants could measure by using morphological markers (Stace, 1980). Although morphological characters are directly exposed to environment, cause change in morphology and leads toward the taxonomic confusions. Morphometery could not provide solution of complex taxonomic problems. Therefore, taxonomist involved other biological techniques like leaf epidermal anatomy, cytology, electrophoresis as taxonomic tools (Stace, 1980).

Anatomical markers of leaf epidermis were found to be an important tool for taxonomic evaluation (Gilani et al., 2002; Yousaf et al., 2008) on inter and intra generic level. The epidermis is a system of cells, variables in structure and function that together constitute the covering of the plant body in the primary state (Esau, 1960). The

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characteristics of leaf epidermis are being increasingly important in resolving existing taxonomical confusions (Metcalfe and Chalk, 1960; Stace, 1980). As leaf epidermis anatomical features such as stomata, trichomes and other markers have been proved useful anatomical tool of potential taxonomical importance (Jones, 1986; Baranova, 1992). Hence, anatomical features are of particular value to identify small parts of plant materials (Stace, 1980). Hence, there is no information available on utilization of leaf epidermis anatomical marker for the taxonomical studies of tropical and subtropical arboreal flora. Therefore, present investigations were made to identify the leaf epidermal markers that can be utilized for the solution of existing taxonomic problem of tree species which overlap in most of their morphological characters and to evaluate the relationship of the critical taxa.

MATERIALS AND METHODS

Plant material

For leaf epidermal anatomical studies fresh leaves from living specimens were used. Fresh material of different tree species were collected from the different localities of Pakistan.

Isolation of leaf epidermis

For isolation of leaf adaxial and abaxial epidermis leaves were soaked for 3 to 4 days. Time of soaking was varied according to the texture of leaves. Epidermal samples were prepared according to the modified method of cotton (1974) who followed clerk's (1960) technique .The fresh leaves were placed in test tube filled with 88% lactic acid kept in hot boiling water for about 3 to 4 h. Lactic acid is used to soften the tissue of leaf due to which its peeling off is made possible.

Preparation of slides

A sharp blade is used for peeling of leaf material. The epidermis was cut across the leaf and scrapped away together with the mesophyll cells until only the epidermal layer of the leaf remained on the tile .Slides of both abaxial and adaxial sides of leaves were prepared and observed under the light microscope (model: Meiji techno).

Photographs of slides

Microphotographs were taken by using CCD digital camera (model: canon Pc1200 attached with MD lens MA151/30/73opter) fitted on light microscope (model: meiji techno). Identification of anatomical character was made by using power at high power plan 40×/0.65,∞/0.17,F=200,WD=0.5) and at lower power plan is 10×/0.25,∞/0.17,F=200,WD=7.3). These micrographs were useful for identification and differentiation of epidermal cells on the basis of microscopic features.

Statistical analysis

Data was evaluated by co-relation matrix and cluster analysis to determine inter specific relationship. Dendrogram was constructed on the basis of unweight pair group method with arithmetic average (UPGMA).The computer software SPSS.V 11.0 was use for this purpose.

RESULT AND DISCUSSION

Key for identification of species is shown in Table 1. The role of anatomical data in traditional taxonomy has been long recognized since the variation within the species, genera or a family is usually reflected in anatomical features as well (Metcalf, 1960). Leaf epidermal anatomical features such as shape of stomata, types of trichome and other characters are useful taxonomical tools .The present study dealt with the diversity in the types and shape of leaf epidermal features in tree species. Although studies on gross morphology and wood anatomy of the plant have proved valuable information for the identification of the plants yet this identification criteria would be incomplete without foliar epidermis morphology (Kadiri, 2006).

Trichomes are highly variable appendages of the epidermis including glandular and non-glandular, blunt, hairs and scales types. They are found on all parts of plant in leaf, stomata and root (Cowan, 1950). Trichomes vary widely in structure intra and interspecifically. These are sometimes remarkably uniform and may be used for taxonomic purposes. Same observations were made by Metcalfe and Chalk (1950) for dicotyledons. In the tree species, trichomes were observed on both abaxial and adaxial surface. Glandular trichomes are originated from the intracellular region. These types of trichomes were found on abaxial and adaxial surface of *Eucalyptus camaldulensis*, *Erythrina subrosa*, *Dalbergia sisso*, *Calliistemone lanceolatus and Melia azedarachta* (Figures 4, 5, 6, 7, 8, 9, 10, 11, 24 and 25). *Ficus religiosa* found different from all other species as no trichome was found on adaxial leaf surface (Figures 2 and 3). Shaheen et al. (2009) investigated the diversity and distribution of glandular and non-glandular trichomes within the genus *Sida* L. (Malvaceae). They recorded sellate, peltate, long stalked, short stalked and capitate trichomes. But they did not study other anatomical markers like cells types; arrangement of epidermal cells various types of stomatal types and silica bodies etc. Glandular trichomes were present only on adaxial surface in *Murraya koenigii* and was absent from abaxial surface (Figures 20 and 21). Single cell, non-glandular trichome are originated from the cell wall is another important anatomical feature which can be utilized for the delimitation of various taxas of dicotyledons .Single cells, non-glandular trichome were present on *A. lebbeck* (Figures 16 and 17) and *Murraya paniculata* (Figures 22 and 23*)* single cells. In *Cassia fistula* trichome present on abaxial surface and were absent from abaxial surface (Figures 10 and 11). Mbagwu et al. (2007) utilized trichomes for characterization of four species of the genus *Citrus* (Rutaceae). Non-glandular trichomes with

Table 1. key for identification of species.

S/N	Characteristics	Key
1a.	Trichome is present on Abaxial and adaxial surface.	3a
1b.	Trichomes are absent from abaxial and adaxial surface.	2a
2a.	Length of long cell ranges from 109.2 to 78.9 µm.	*Mangiferaindica*
2b.	Length of long cell ranges from 79.8 to 39.1 µm.	*Syzgiumcumini*
3a.	Non-glandular type trichomes are present on Abaxial and adaxial	4a
3b.	Glandular trichomes are present on Abaxial and adaxial surface	9a
4a.	Silica bodies are present on Abaxial and adaxial surface.	8a
4b.	Silica bodies are absent on Abaxial and adaxial surface.	5a
5a.	Smooth and single layered cells are present.	6a
5b.	Wavy and double layered cells are present.	*Albezialebbeck*
6a.	Single cells non-glandular trichomes are originated from the cell wall	7a
6b.	Single cells non-glandular trichomes are originated from the Intracellular cells	*Murrayakoenigii*
7a.	Width of stomata cavity ranges from 18.0 to 7.0 µm.	*Acaciaarabica*
7b.	Width of stomatal cavity ranges from 20.2 to 11.3 µm.	*Cassiafistula*
8a.	Anomotetracytic stomata are present.	*Murrayapaniculata*
8b.	Anomocytic stomata are present	*.Azadirachtaindica*
9a.	Polygonal epidermal cells are present on both sides.	10a
9b.	Rectangular cells are present on both sides.	*Euclayptuscamaldulensis*
10a.	Stomata are present on both sides.	11a.
10b.	Stomata are present only on adaxial surface.	*Erythrinasubrosa*
11a.	Number of trichome/unit area is 5 to 6.	*Calliistemonelanceolatus*
11b.	Number of tricome /unit area is 1 to 2.	12a
12a.	Length of subsidiary cells 54.5 to 25.2 µm.	*Dalbergiasisso*
12b.	Length of subsidiary cells 110.2 to 85.4 µm.	*Ficusinfectoria*

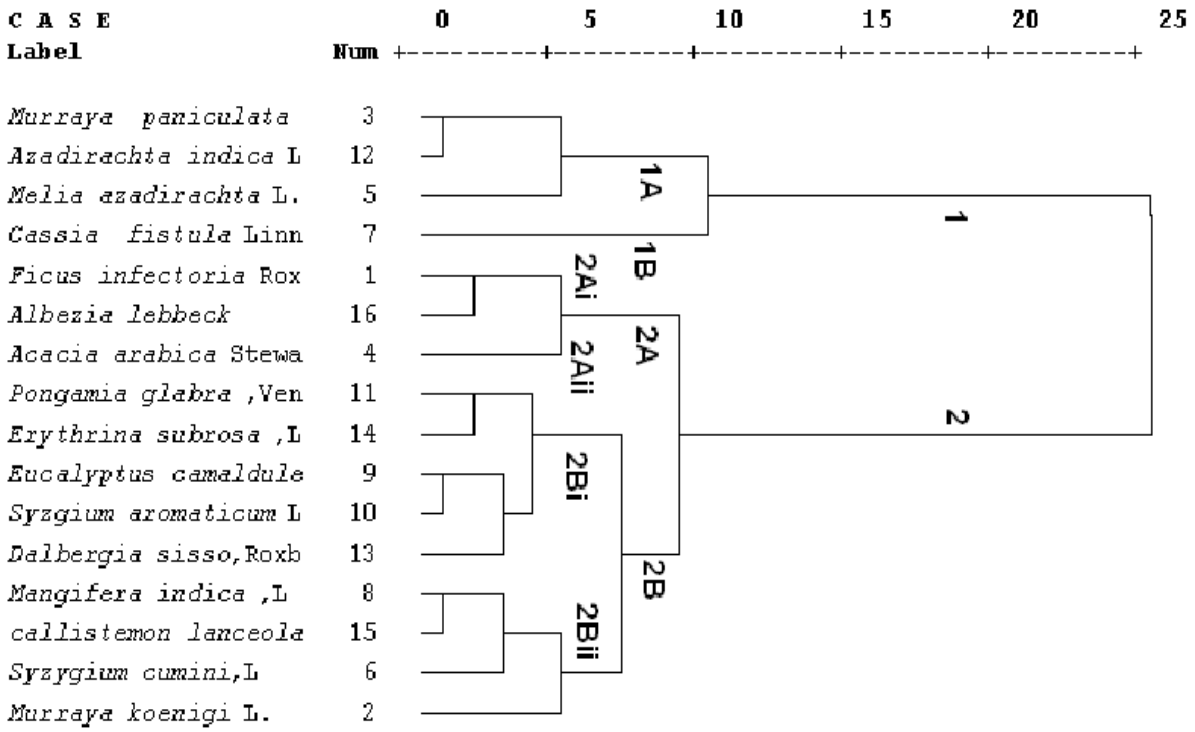

Figure 1. Cluster analysis of the some selected medicinally important arboreal flora of tropical and subtropical regions based on leaf epidermal anatomy.

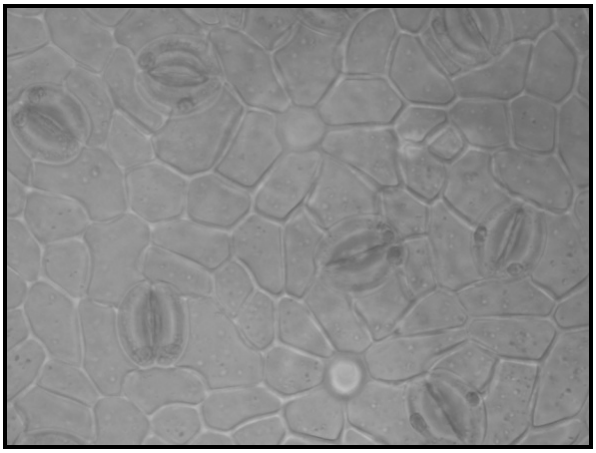

Figure 2. Leaf epidermal anatomy of *Ficus infectoria*, L Abaxial.

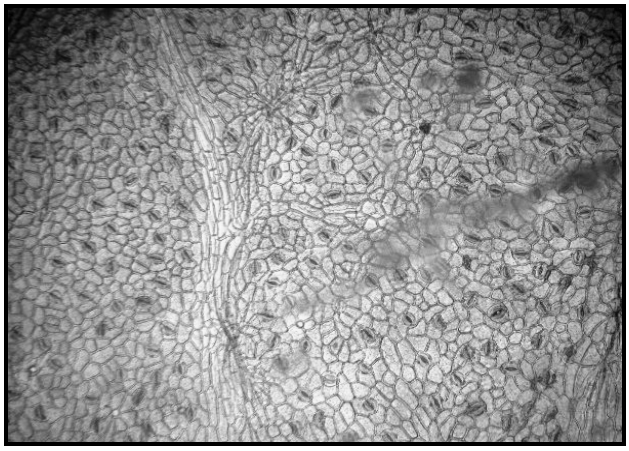

Figure 3. Leaf epidermal anatomy of *Ficus infectoria*, Adaxial side.

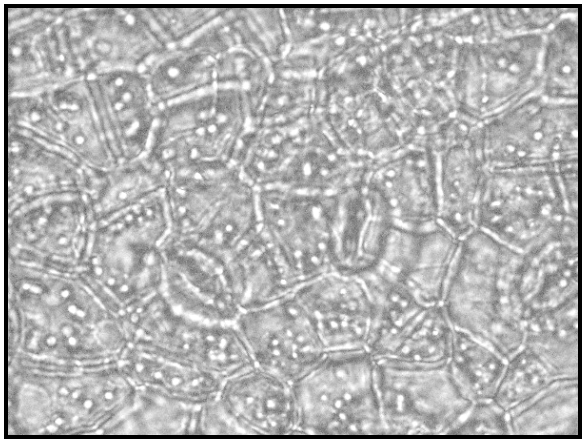

Figure 4. Leaf epidermal anatomy of *Eucalyptus camaldulensis* Dehnh Abaxial.

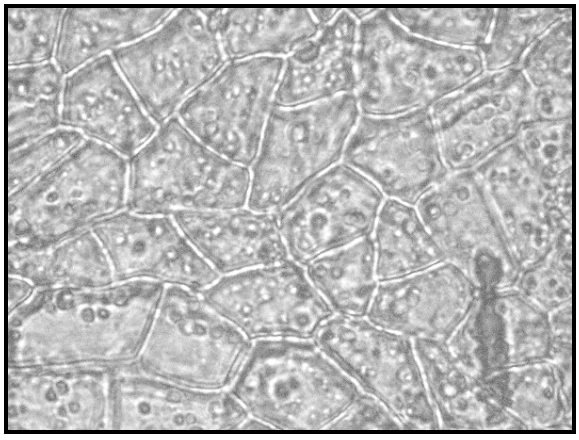

Figure 5. Leaf epidermal anatomy of *Eucalyptus camaldulensis* Adaxial side.

2 to 3cells originated from the cell wall found to be the unique character of tree species. These trichomes were of blunt in shape. These were recorded from abaxial and adaxial surface of *Pongamia glabra*, *Azadirachta indica,* and *Sygzgium aromarticum* (Figures 30, 14, 15, 26 and 27). Trichome were entirely absent from *S. cumini* (Figures 28 and 29) and *M. indica* (Figure 19).

Another important anatomical marker is presence of silica bodies. Mostly two, round and irregular shapes of silica bodies were found in the present investigation. Round shape silica bodies are present on abaxial and adaxial surface of *M. paniculata, D. sisso*, *E. subrosa* (Figures 22, 23, 8, 9, 6 and 7). Whereas round shape of silica bodies were present only on abaxial surface of *A. indica* and *S. cumini* and were not found on adaxial surface (Figures 14, 15, 28 and 29). Irregular shape of silica bodies is present on abaxial and adaxial surface *S. aromarticum* and *M. azedarachta* (Figures 26, 27, 24 and 35). These Irregular silica bodies were only recorded on abaxial surface of *E. camaldulensis* and *P. glabra* (Figures 4, 5 and 30). Silica bodies were absent from *A. arabica, A. lebbeck, C. lanceolatus, C. fistula, F. infectoria, M. indica* and *M. Koenigii* (Figures 18, 12, 13, 10, 11, 2, 3, 19, 20 and 21).

Different types of stomata were first of all studied by Stresburger (1886), followed by Vesque (1889), recognized four broad categories of stomata based on the presence and arrangement of accessory cells as well as their mode of development. Stomatal patterning is related to ordered placement of stomata on the leaf surface. The pattering process involves the selection of undifferentiated cells to become stomata (Vesque (1889). It is not concerned with the physical events of their differentiation. The opening pores are present on the surface of leaf epidermis bounded by two specialized epidermal cells called the guard cells, by which change in shape bring about the opening and closure of the

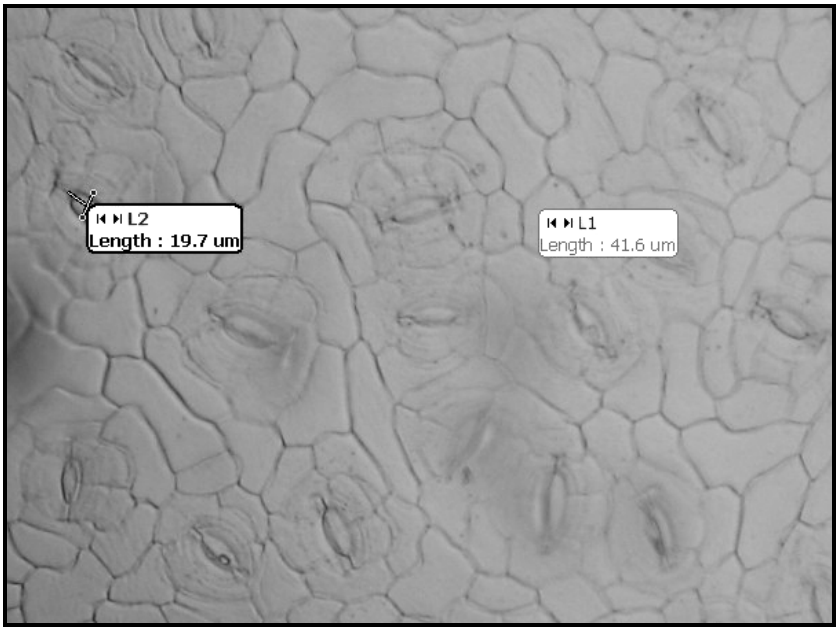

Figure 6. Leaf epidermal anatomy of *Erythrina subrosa*, L Abaxial.

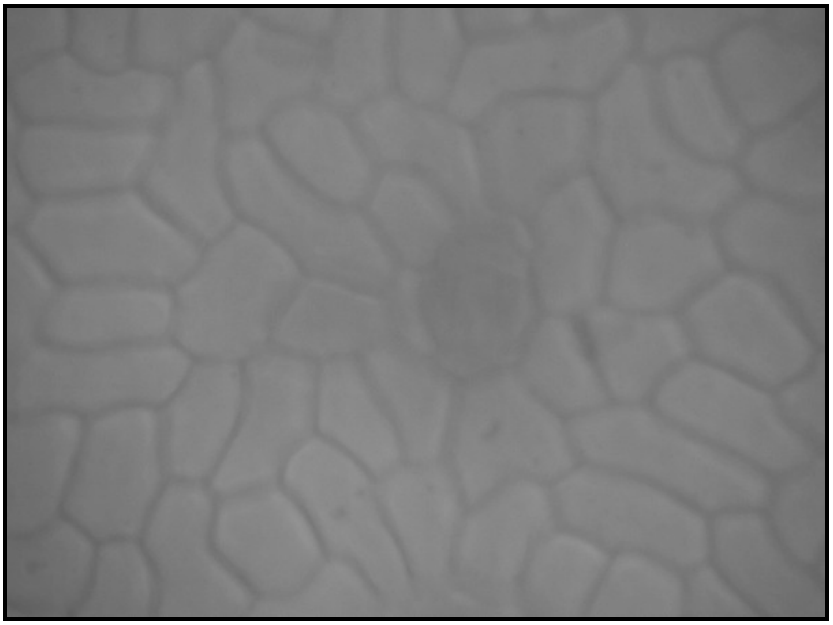

Figure 7. Leaf epidermal anatomy of *Erythrina subrosa***,** Adaxial side .

aperture. The features appearances play a role in the opening and closing of the stomatal pore (Stalfelt, 1956). On the basis of arrangement the guard epidermal cell neighboring the guard cell more than 25 main types of stomata in dicot have been recognized (Metcalfe, 1979).

In present study nine different types of stomata were recognized like anomocytic, Amphianisocyic, Brachyparacytic, Anomotetracytic, Anisocytic, Amphicyclocytic, Ammphianisocytic, actinocytic and staurocytic. Ahmad et al. (2009) recognized seven (anisocytic, amphianisocytic, axillocytic, anomotetracytic, actinocytic, diacytic and staurocytic) types of stomata. They just emphasis on stomata anatomical marker but they ignore other anatomical character. Anomocytic stomata were present on both abaxial and adaxial surface in *C. lanceolatus* and *A. indica*. This type of stomata is only present on abaxial surface of *F. infectoria.* Perveen et al. (2007) recognized six types of

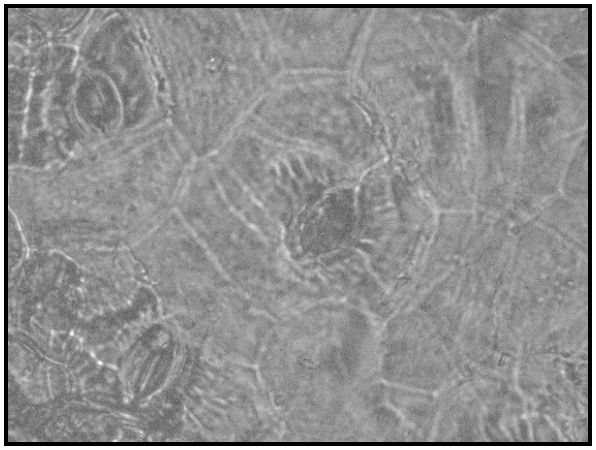

Figure 8. Leaf epidermal anatomy of *Dalbergia sisso*, Roxb Abaxial.

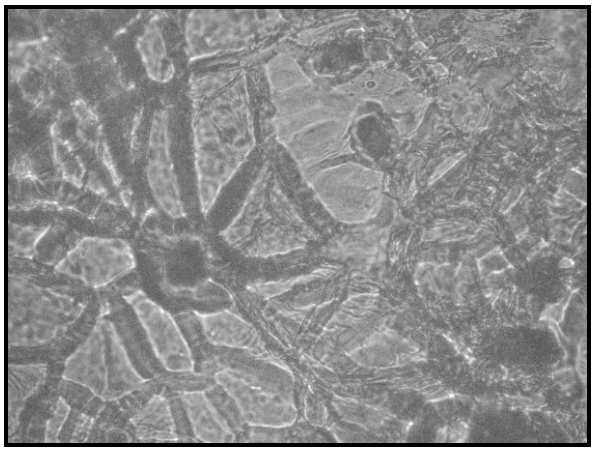

Figure 9. Leaf epidermal anatomy of *Dalbergia sisso*, Adaxial side.

stomata (anomocytic, paracytic, diacytic, parallelocytic, cyclocytic and anisocytic). In which anomocyticstomata are most dominant found in 54 taxa of 69 dicot species. Amphianisocytic stomata were present on both abaxial and adaxial surface of *P. glabra*. Yasmin et al. (2010) worked on the micro morphological investigation of foliar anatomy of *Fagopyrum Mill*., and Rumex. L. of Polygonaceae. They selected the specimens from Quaid-e-Azam University herbarium. They selected eleven species belonging to *Fagopyrum* Mill and Rumex. L. of the family Polygonaceae .They studied the anatomical features such as shape of epidermal cells, stomatal pattern , their distribution on abaxial and adaxial on the leaf surface and variation in trichomes types. Among of these species, they reported five different stomatal (Paracytic, pericytic, anomocytic, staurocytic, anisocytic). Although it was good piece of work but they emphasized only on shape of stomata and trichome types and ignored all these important characters (arrangement of epidermal cells, variation of trichomes and silica bodies etc .Which could be proved very important taxonomic tool for identification. Brachyparacytic stomata were present on both abaxial and adaxial surface in *A. lebbeck* and *E. camaldulensis* (Figures 16, 17, 4 and 5). Anomo tetracytic stomata were present on both abaxial and adaxial surface in *M. paniculata*, *S. cumini*, *M. azadirachta* and *S. aromaticum* (Figures 23, 24, 29, 30, 25, 26, 27 and 28). These stomata were present only on abaxial surface in *M. indica* (Figure 19). Nikolic et al. (2003) worked out a comparative analysis of stomata and leaf trichome characteristics in *Quercus robex* L. genotype variability of leaf trichome and stomata characteristics. Among these species, they recognized solitary glandular and uniseriate glandular hairs that were present on the adaxial leaf surface. Single glandular trichomes were observed in all genotype. But they ignored all other anatomical characters. But they studied the impact of unfavorable factors of the environment, particularly air pollution morpho anatomical changes of leaves in different tree species.

On abaxial surface, Actinocytic stomata were present in *S. aromarticum* (Figures 26 and 27). Staurocytic stomata are present on abaxial surface in *E. subrosa* (Figures 6 and 7). Anisocytic stomata were present on both abaxial and adaxial surface in *M. koenigi, D. sisso, C. fistula* and *A. arabica* (Figures 20, 21, 8, 9, 10, 11 and 18). Epidermal cells are quite variable in the configuration of the characteristic markers, which have been proven to be systematic value in cuticular characters, epidermis, stomata, subsidiary cells and trichome (Ellis, 1976). So the taxonomic significance of epidermal morphology is well documented in botanical literature. Considering the anatomical characters, five different types of epidermal cells were identified. Klimko et al. (2006) revealed the number of micro morphological characters at taxonomic level in selected taxa of the genus *Ficus* L. (Moraceae). Irregular type of cells was found in *P. glabra*, *S. cumini*, *S. aromarticum* and *A. lebbeck* (Figures 28, 29, 26, 27, 30, 16 and 17). Pentagonal epidermal cells are found in *A. indica* (Figures 16 and 17). Nikolic et al. (2007) investigated the pentagonal, rectangular and hexagonal types of cell and abundant stomatal types of both species; *Solanum macrocarpon* and *Solanum nigrum*.

Polygonal shape of epidermal cells was found in *M. koenigii, A. indica, F. infectoria*, *E. subrosa*, *D. sisso* and *C. lanceolatus* (Figures 8, 9, 12, 13, 10, 11, 2, 3, 19, 20 and 21). Ahmad et Al. (2010) worked out taxonomic diversity in epidermal cell of some sub-tropical plant species. For this purpose they selected 40 species belonging to 38 genera of 22 families. The shape and type of large epidermal cells among these varies from straight and polygonal .They concluded the large variation in epidermal cells configuration at different taxonomic level.But they ignored another anatomical characters that could provide an inter generic and intra generic variation in many species. Although there is a need

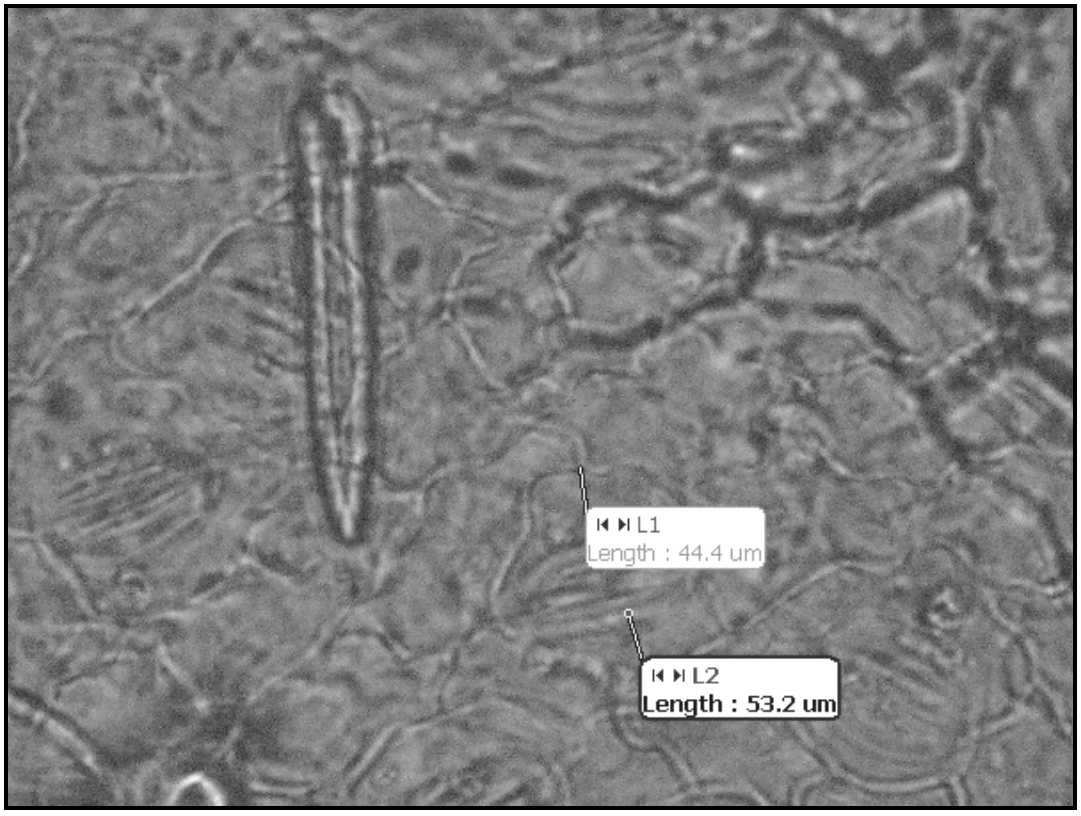

Figure 10. Leaf epidermal anatomy of *Cassia fistula* Linn. Abaxial.

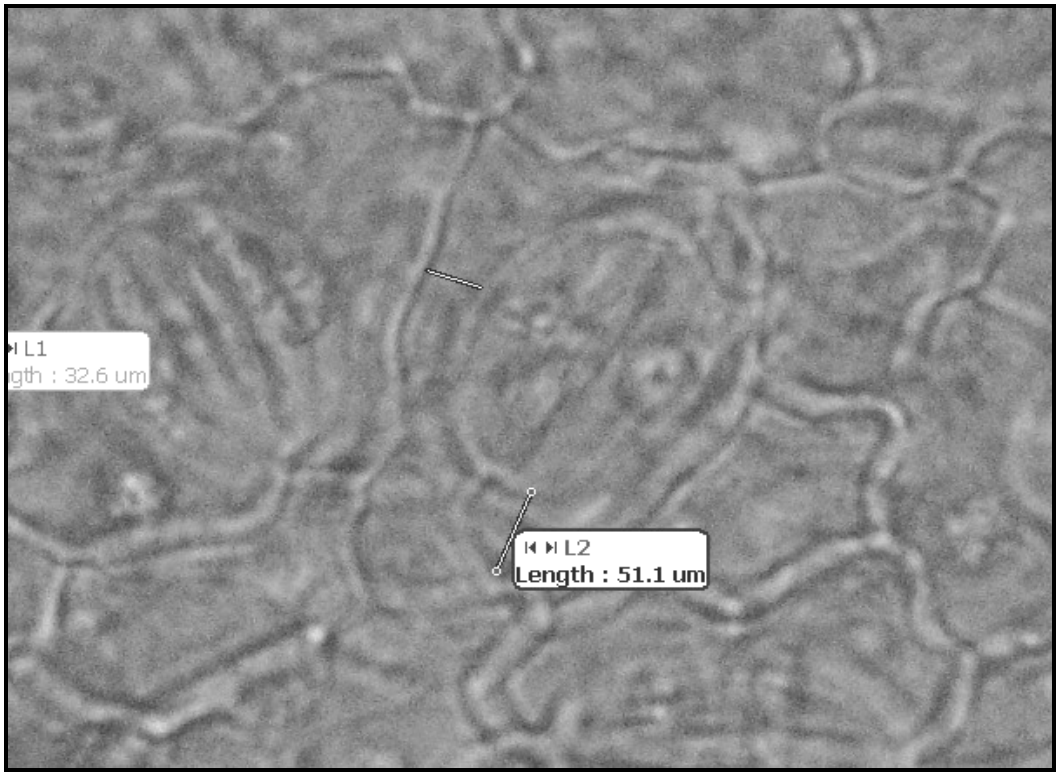

Figure 11. Leaf epidermal anatomy of *Cassia fistula* Linn. Adaxial side.

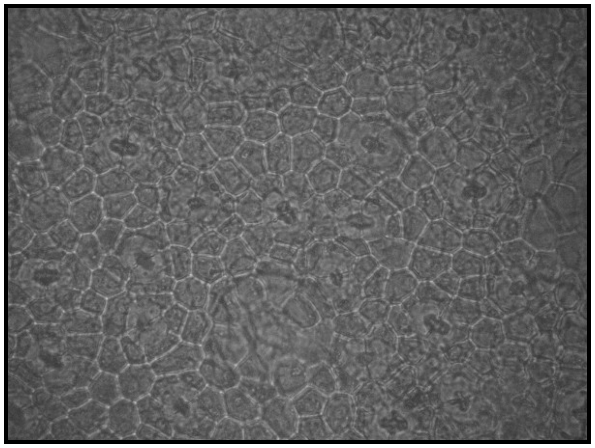

Figure 12. Leaf epidermal anatomy of *Callistemon lanceolatus*, DC Abaxial.

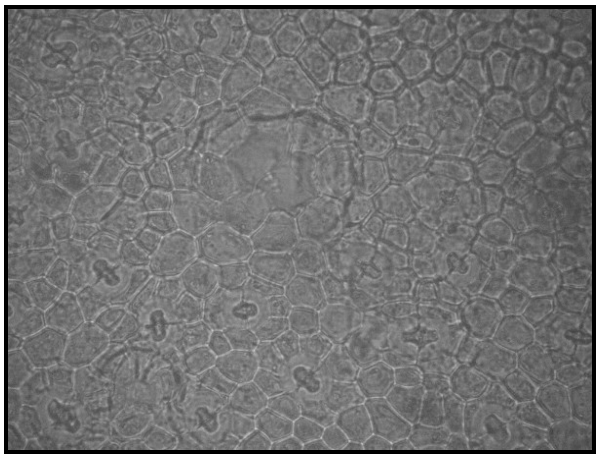

Figure 13. Leaf epidermal anatomy of *Callistemon lanceolatus*, Adaxial side.

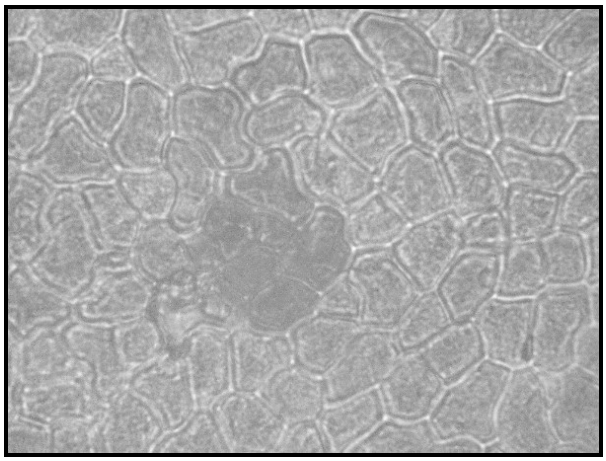

Figure 14. Leaf epidermal anatomy of *Azadirachta indica* L. Abaxial.

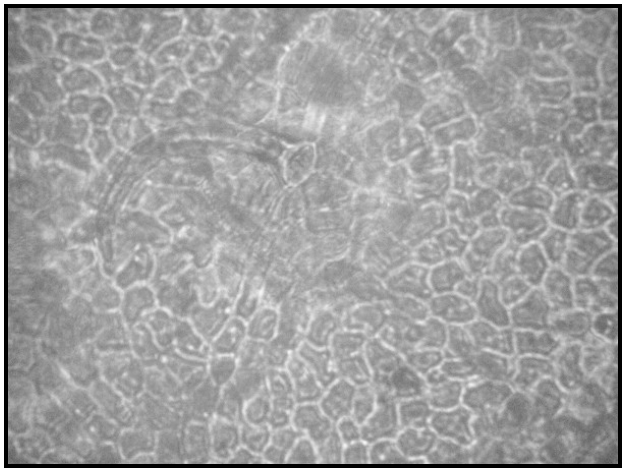

Figure 15. Leaf epidermal anatomy of *Azadirachta indica* L., Adaxial side.

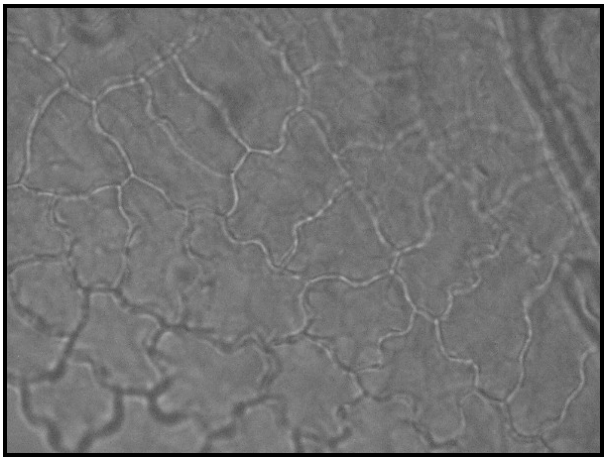

Figure 16. Leaf epidermal anatomy of *Albezia lebbeck* Abaxial.

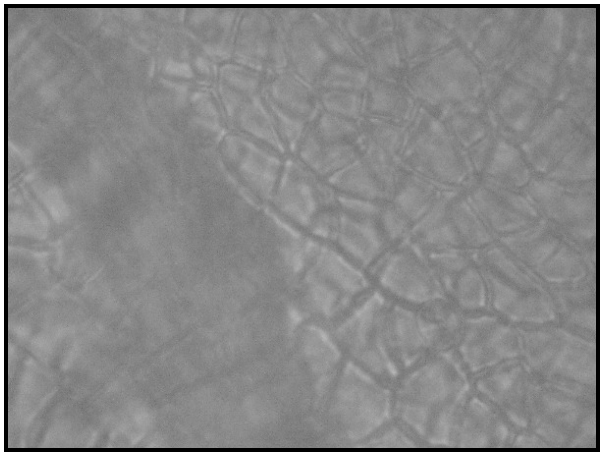

Figure 17. Leaf epidermal anatomy of *Albezia lebbeck* Adaxial side.

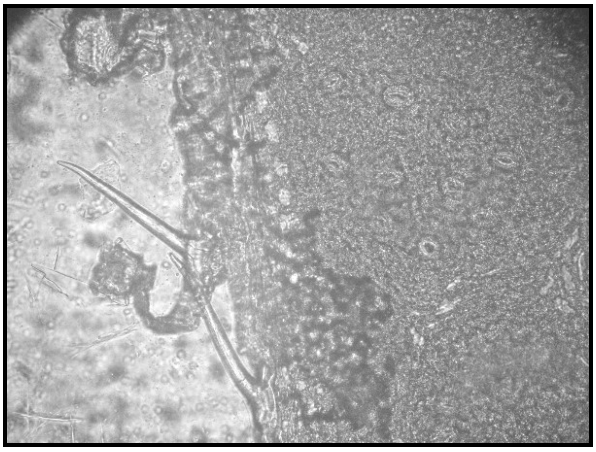

Figure 18. Leaf epidermal anatomy of *Acacia arabica*, Stewart (Abaxial).

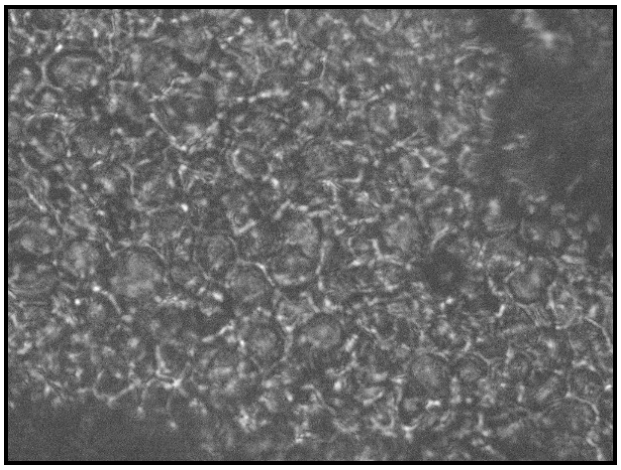

Figure 19. Leaf epidermal anatomy of *Acacia arabica*, Stewart (*Magnifera indica* (Abaxial).

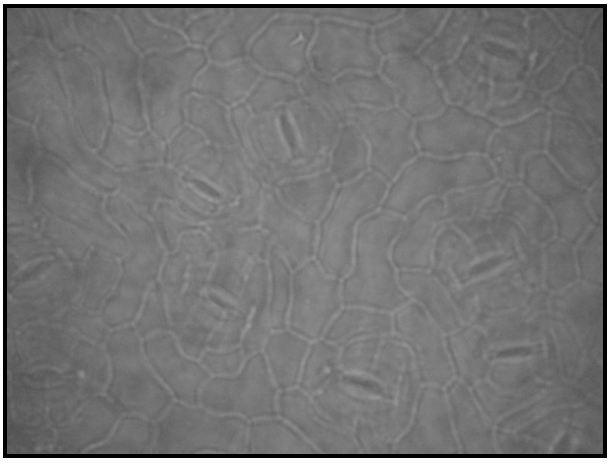

Figure 20. Leaf epidermal anatomy of *Murraya koenigii***,** Adxial sides (left and right).

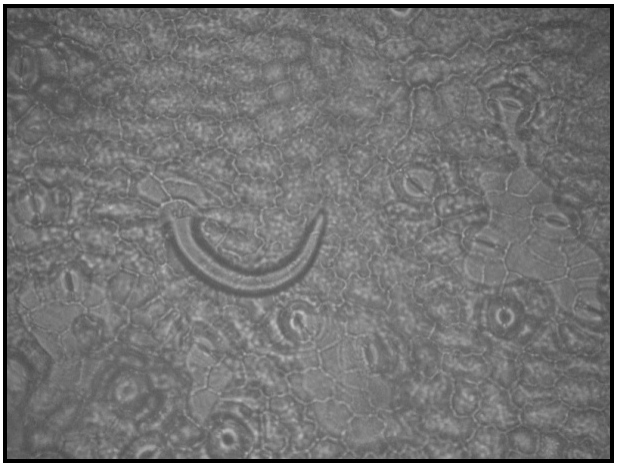

Figure 21. Leaf epidermal anatomy of *Murraya koenigii*, Adxial sides.

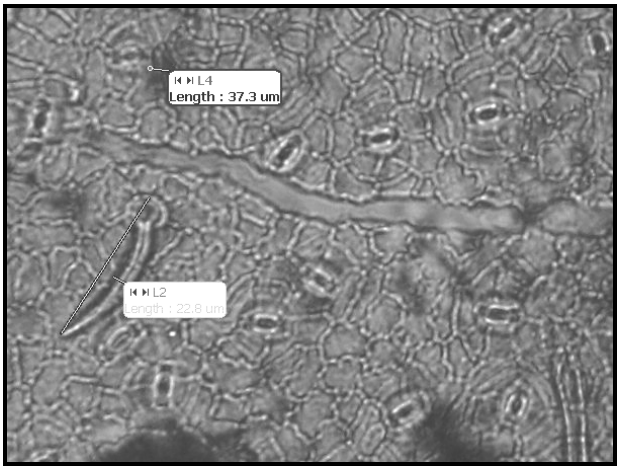

Figure 22. Leaf epidermal anatomy of *Murraya paniculata*, Adaxial.

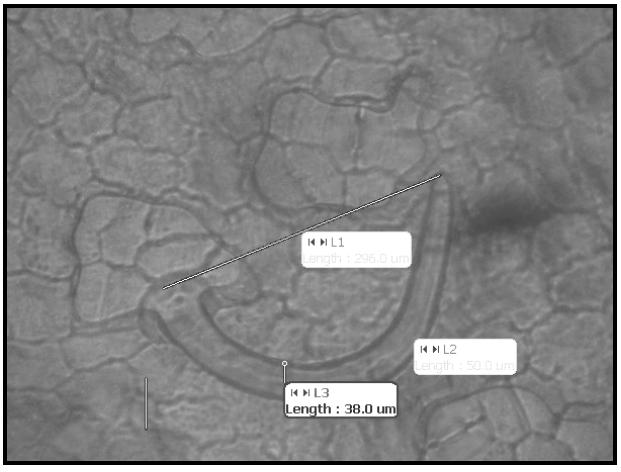

Figure 23. Leaf epidermal anatomy of *Murraya paniculata*, Adxial sides.

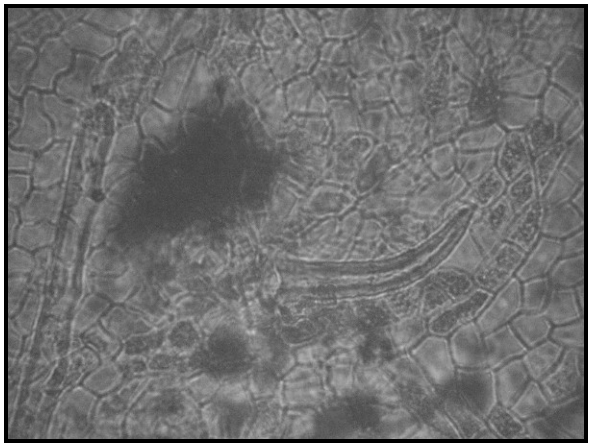

Figure 24. Leaf epidermal anatomy of *Melia azedarachta*, L Abaxial.

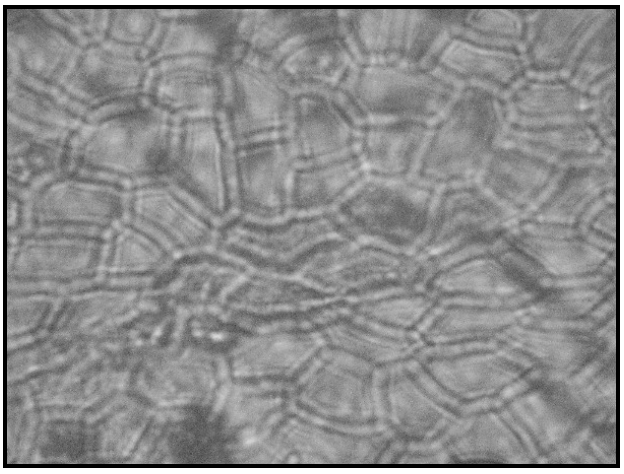

Figure 25. Leaf epidermal anatomy of *Melia azedarachta*, Adxial sides.

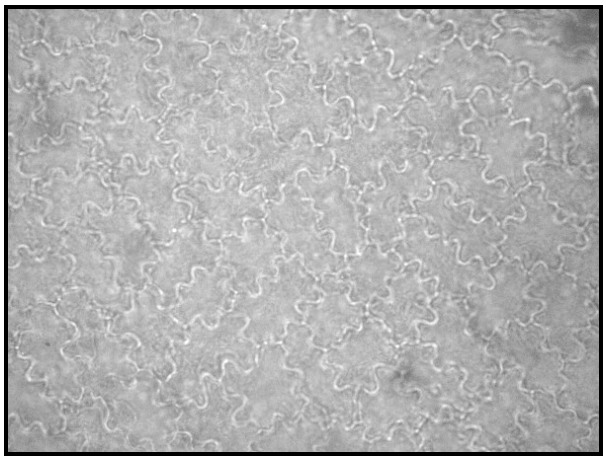

Figure 26. Leaf epidermal anatomy of *Sygzgium cuminii*, L, Abaxial.

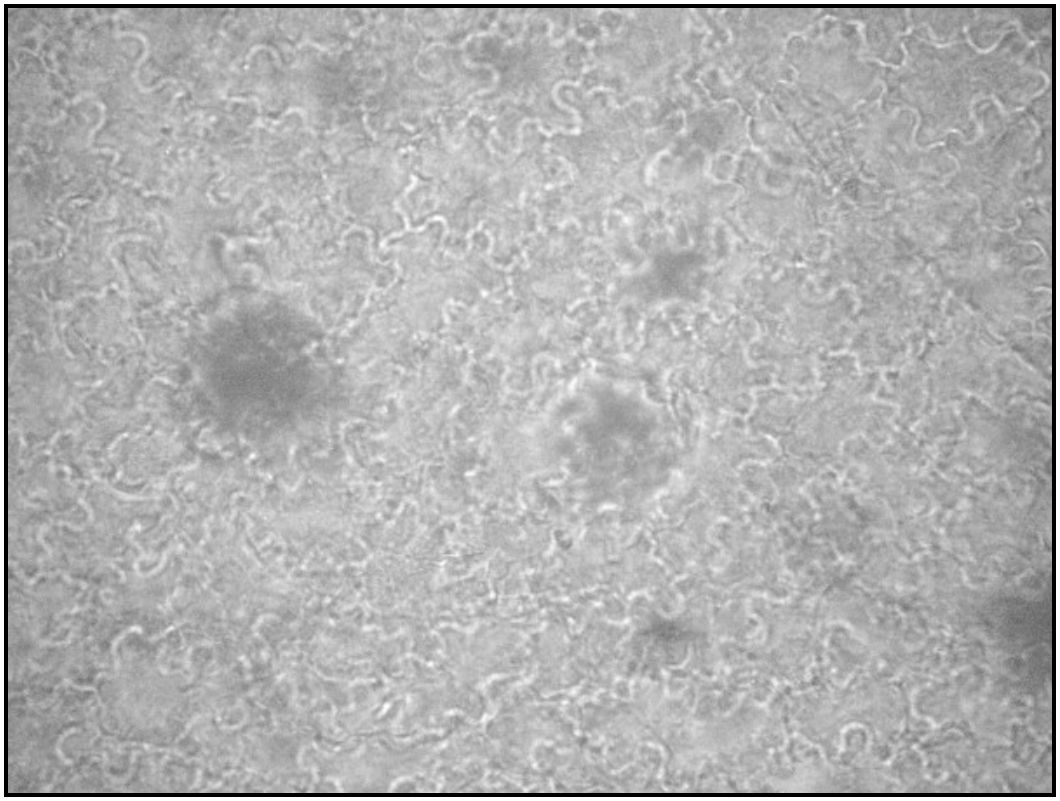

Figure 27. Leaf epidermal anatomy of *Sygzgium cuminii*, L, Adxial sides.

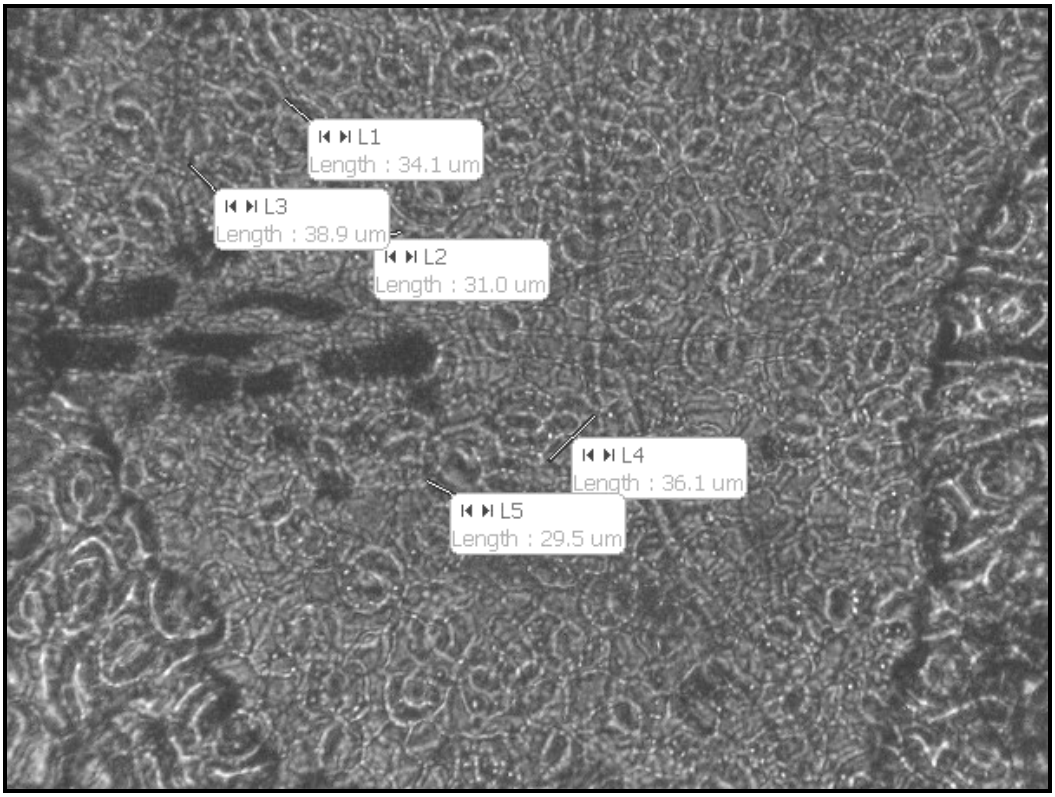

Figure 28. Leaf epidermal anatomy of *Syzygium cumini*, L Abaxial.

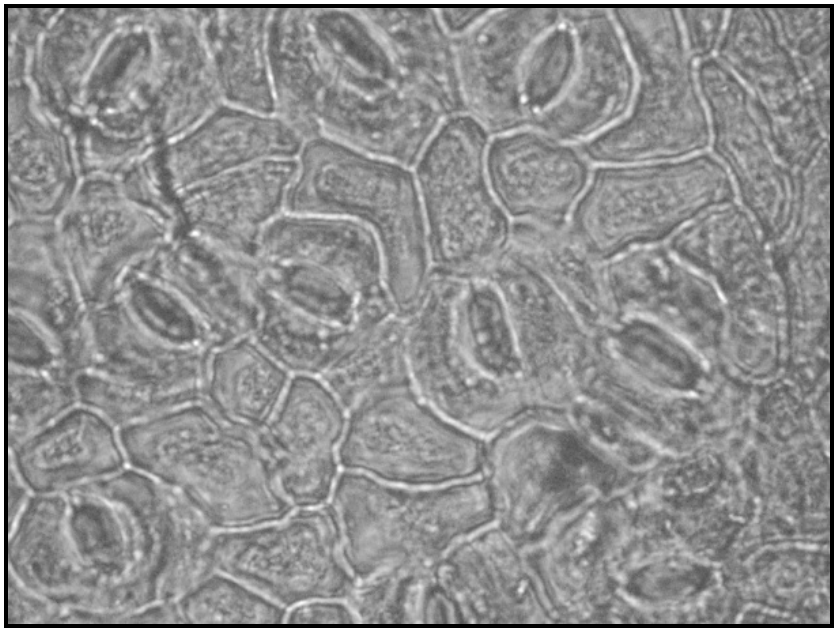

Figure 29. Leaf epidermal anatomy of *Syzygium cumini*, Adxial sides.

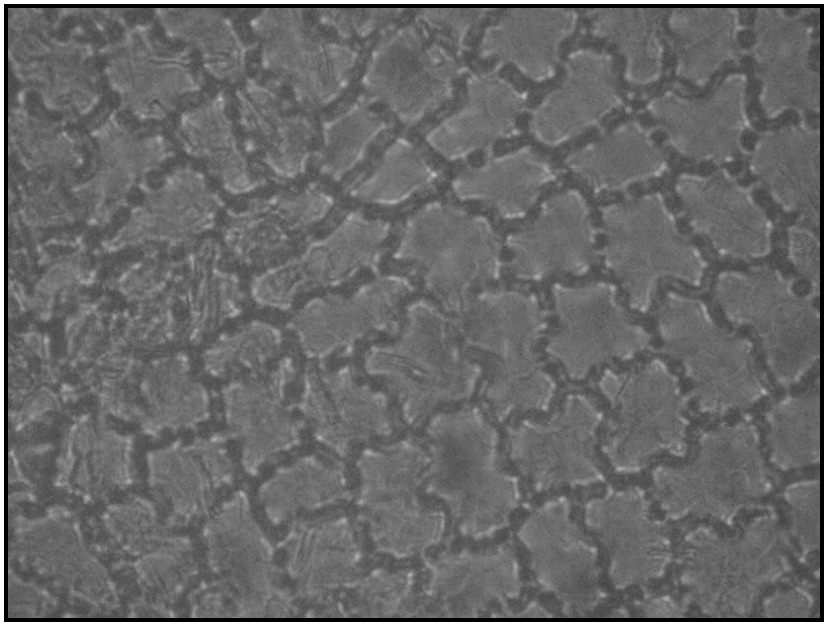

Figure 30. Leaf epidermal anatomy of *Pongemia glabra* Vent, Roxb (Abaxial side).

of more comprehensive work on the shape and type of leaf epidermal cells for their further elaboration on a taxonomic characters. Farooqui et al. (1989) studied cuticular characters in three species of *Dalbergia.* They distinguished the papillate and non papillate cuticular characters in two species. They just emphasized on the epidermal cells and ignored other anatomical characters. Rectangular shape of epidermal cells is found in *M. paniculata, M. azedarachta, E. camaldulensis* and *A. arabica* (Figures 8, 9, 24, 25, 4, 5, 18, 22 and 23). Hexagonal shape of epidermal cells was found *C. fistula* (Figures 10 and 11).

On the basis of the cell arrangement, species were separated into different category. Smooth and single layered cells separate *A. arabica C. fistula*, *D. sisso, E. subrosa, F. infectoria, M. indica and S. cumini* (Figures 6, 7, 19, 10, 11, 18, 8, 9, 3, 28 and 29). Smooth and double layered cells are present in *A. indica, C. lanceolatus,*

Table 2. Correlation of leaf epidermal anatomical characters.

Leaf epidermal anatomical characters	1	2	3	4	5	6	7	8	9	10	11	12
1	1											
2	0.44	1										
3	0.80	0.43	1									
4	0.28	0.14	0.50	1								
5	0.24	0.13	0.38	0.25	1							
6	0.07	0.43	0.24	0.07	0.37	1						
7	0.01	-0.44	0.12	0.22	-0.12	-0.11	1					
8	0.37	-0.34	0.10	0.31	-0.07	0.03	0.66	1				
9	0.00	0.29	0.03	0.11	0.14	0.06	0.48	0.27	1			
10	-0.07	0.32	-0.07	0.18	-0.01	0.08	0.28	-0.03	0.75	1		
11	-0.24	0.10	-0.17	-0.05	0.29	0.43	-0.23	-0.01	-0.35	-0.15	1	
12	-0.21	0.06	-0.12	0.02	0.28	0.44	-0.36	-0.10	-0.44	-0.23	0.94	1

1: Length of long cell, 2: Width of long cell, 3: Length of short cell, 4: Width of short cell, 5: Length of trichome cell, 6: Width of trichome cell, 7: Length of stomatal cavity, 8: Width of stomatal cavity, 9: Length of subsidiary cell, 10: Width of subsidiary cell, 11: Length of silica bodies, 12: Width of silica bodies.

E. camaldulensis and *M. azedarachta* (Figures 14, 15, 12, 13, 4 and 5). Wavy and double layered cells are present in *M. paniculata, P. glabra, S. aromarticum* and *A. lebbeck* (Figures 22, 23, 26, 27, 30, 16 and 17). Leaf epidermal characters like trichome types, difference of origin, variation in stomata types, and arrangement of cells and presence of silica bodies were considered important features for the characterization of tree species. A dendogram was constructed on the basis of all the anatomical characters observed on both abaxial and adaxial sides of trees species from Lahore District (Figure 1). At 100% linkage distant of 16 species were divided into large subcluster 1 and 2. Cluster 1 is further divided into 1A containing *M. paniculata* L. and *M. azedarachta* L. and 1B containing *C. fistula* Linn. at 42%. At 38% 2 group divided into two groups 2A and 2B. However, at 22% distant divided into further two groups 2Ai containing *F. infectoria* Roxb.sana and 2Aii containing *A. arabica*, Stewart. At 30% distant 2B is divided into 2Bi and 2Bii. However, at 18% distant divided 2Bi containing *D. sisso*, Roxb, *E. camaldulensis* Dehnh and *P. glabra* Vent, Roxb. At 24% distant divided 2Bii containing *M. koenigii* L.

At 16% distant divided 2Bii containing *M. indica* L. and *S. cumini* L. At 12% distant divided two species containing *A. lebbeck* L. (bent) and *E. subrosa* L. At 4% distant divided three species containing *A. indica* L., *C. lanceolatus* DC and *S. aromarticum* L. The difference is negligible and these species consider similar on the basis of epidermal anatomical characters. Co-relation in trees species of leaf epidermal anatomical characters was calculated by using Microsoft Excel software and represented in Table 2. Ranges of co-relation ranges from 0.00 to 0.94. Length of silica bodies and width of silica bodies showed closely maximum relationship at 94%. Similarly, least correlation co-relation was found between length of subsidiary cell and length of long cell. Length of short cell and width of short cell show relationship at 50%. Length of stomatal cavity and width of stomatal cavity show close relation at 66%. Length of subsidiary cell and width of subsidiary cell show relationship at 75%. Width of trichome cell and width of short cell show co-relation at 43%. Length of trichome and width of trichome cell showed co-relation at 37%. Width of subsidiary cell and width of short cell show relationship at 32%. Length of subsidiary cell and length of trichome cell show co-relation at 0.14. Width of trichome and width of short cell show co-relation at 0.07. Length of subsidiary cell and length of stomatal cavity showed co-relation at 0.06. Lenfth of subsidiary cell and length of short cell show relationship at 0.03. Co-relation of tree species from order Fabales, Sapindales and Myrtales based on leaf epidermal anatomical character was calculated by using Microsoft Excel software and represented in Table 3.

Range co-relation varies from 0.02 to 0.97. *M. indica* and *M. paniculata* of family Anacardiaceae and Rutaceae, respectively showed maximum co-relation index 0.97. Similarly least co-relation was found between *A. indica* and *F. infectoria* (0.02). *A. indica* and *M. indica* were two more closely related species with similarity index of 0.96. Two important species of *Murraya* found in Lahore district are *M. koenigii* and *M. paniculata* are quite different from each other on the basis of leaf epidermal anatomical characters. The Co-relation index of these two species is 0.13. Mrytaceae economically and medicinally is important family. Two important species of this family *S. cumini* and *S. aromaticum* were included in the present investigation and it was found that co-relation between these two species based on leaf epidermal anatomical character is 0.60. *C. fistula* and *A. arabica*.

Table 3. Co-relation of Tree species from order Fabales, Sapindales and Myrtales based on Leaf epidermal anatomical characters.

Species names	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
1	1															
2	0.13	1														
3	0.65	0.16	1													
4	0.06	0.97	0.03	1												
5	0.22	0.94	0.02	0.96	1											
6	0.41	0.61	0.59	0.54	0.52	1										
7	0.80	0.39	0.82	0.28	0.34	0.59	1									
8	0.69	0.18	0.54	0.14	0.24	0.67	0.51	1								
9	0.63	0.69	0.41	0.67	0.76	0.78	0.69	0.60	1							
10	0.65	0.73	0.61	0.65	0.70	0.56	0.86	0.37	0.76	1						
11	0.43	0.88	0.22	0.88	0.93	0.60	0.50	0.37	0.82	0.80	1					
12	0.73	0.53	0.36	0.51	0.66	0.67	0.65	0.69	0.93	0.68	0.76	1				
13	0.81	0.40	0.55	0.36	0.48	0.76	0.67	0.87	0.85	0.60	0.62	0.91	1			
14	0.50	0.82	0.26	0.80	0.86	0.42	0.61	0.19	0.75	0.88	0.88	0.64	0.49	1		
15	0.31	0.90	0.35	0.86	0.84	0.49	0.59	0.07	0.69	0.87	0.85	0.51	0.37	0.93	1	
16	0.47	0.44	0.36	0.44	0.51	0.78	0.41	0.85	0.75	0.41	0.52	0.77	0.85	0.30	0.24	1

Key: 1. *Murraya koenigi*; 2. *Murraya paniculata*, 3: *Ficus infectoria*, 4: Mangifera indica, 5; Azadirachta indica, 6; *Melia azadirachta* 7. *Eucalyptus camaldulensis*; 8. *Callistemon lanceolatus*, 9; *Syzygium cumini*, 10. *Syzgium aromaticum*, 11. *Albezia lebbeck*; 12. *Dalbergia sisso*, 13. *Erythrina subrosa*; 14. *Pongamia glabra*, 15. *Acacia arabica* 16. *Cassia fistula* and *Murraya paniculata* shows maximum.

Although, belonging to same family but they have quite different anatomical characters therefore index of correlation is only 0.24. It is concluded that medicinally important arboreal flora could be easily identify if penotypical markers and leaf epidermal markers utilized in close association. The important leaf epidermal markers for this group are size, shape and distribution of epidermal cells, stomata and trichomes. Stomatal type is an important marker which could be effectively utilized for the taxonomic purpose.

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