Full Length Research Paper

Foliar epidermal anatomy of some ethnobotanically important species of genus *Ficus* Linn.

Kiran Yasmin Khan¹, Mir Ajab Khan¹, Mushtaq Ahmad¹, Ghulam Mujtaba Shah², Muhammad Zafar¹, Rabia Niamat¹, Mamoona Munir¹, Arshad Mehmood Abbasi¹, Hina Fazal¹, Paras Mazari¹ and Nighat seema¹.

¹Department of Plant sciences, Quaid-i-Azam University, Islamabad, Pakistan. ²Department of Botany, Government Post Graduate College No. 1, Abbottabad, Pakistan.

Accepted 5 January, 2011

The main objective of this paper is the detailed morphological study of leaf epidermis of Genus *Ficus* (Moraceae). The study based on the micro characters of leaf epidermis of some species of *Ficus* collected from Pakistan. The study revealed the many interesting epidermal features of leaves that have not previously been reported before and has been conducted first time. No published details are available of such comprehensive foliar anatomical studies of following species of genus *Ficus* L. species has been conducted first time. Note the measurements, shape and type of the leaf epidermal cells including pavement cells, stomata and shape of guard cell pairs, trichomes and ornamentation of cuticular membrane. Pavement cells are often polygonal and irregular. Among the species the *F. virens* has larger pavement cells as well as stomata as compare to other species. Whereas there are little variations represented in qualitative and quantitative characters among the species of same genus. Stomata are paracytic except in *F. nerrifolia* and *F. callosoa* and are restricted on abaxial surface. Trichomes present in some species mostly are unicellular and non-glandular with bulbous base. Cuticular membrane is mostly smooth and pubescent. And in some species peltate glands also observed.

Key words: Ficus, anatomy, trichome, stomata, pavement cells and species.

INTRODUCTION

Moraceae (mulberry family), a family of dicotyledonous plants, containing 1500 species in 53 genera distributed mainly in warm regions of the world. This family includes the genus *Ficus* (figs), represented about 700 species and one of largest genera of higher plants (Scott, 1996). According to Janzen (1979), the genus *Ficus* makes up the most distinctive of the widespread genera of tropical plants. According to Ghafoor, (1985) nearly 1000 species, distributed in tropics and subtropics. Represented in Pakistan by 24 species, of which 11 are native. Anatomical studies have shown that foliar characters too are strictly comparable over a wide taxonomic range, to

to those of foliar organs and so guite reliable. It plays very important role in identification of incomplete plants, e.g., sterile specimens, archaeological remains and fragmentary fossils of all the non-reproductive organs (Stace, 1965). Although foliar epidermal morphological studies may not themselves be sufficient as taxonomic evidence. they could in conjunction with anatomical features serve as good taxonomic tools for delimiting taxa (Watson, 1962). The plant epidermis is a multifunctional tissue playing important roles in water relations, defense and pollinator attraction. This range of functions is performed by a number of different types of specialized cells, including pavement cells, stomata with associated cells and trichomes. These various cells show different degrees of morphological specialization. It is imperative to therefore to attempt a search for epidermal characters

^{*}Corresponding author. E-mail: kiranbaloch1@yahoo.com.

that may be of taxonomic utility. Literature on the epidermal morphology of *Ficus* is relatively rare though the taxonomic value of epidermal morphology is well documented in botanical literature for several other groups of angiosperms (Stace, 1965). Several reports were published on the epidermal structures and stomatal ontogeny by Karatela and Gill (1984a, b, 1985) who stressed their usefulness in plant taxonomy.

The study of the epidermal surfaces of Ficus revealed a number of important micro morphological characters, and these characters exhibit interesting interspecific variations that are of taxonomically significant (Sonibare et al., 2005). The lithocyst is a big epidermal cell tat goes into the subjacent tissues are characteristic of Moraceae. Acanthaceae or Cucurbitaceae (Fahn, 1985). Trichomes and hydathodes are other characters found on the epidermis and other organs of plants that can serve as good taxonomic tools. They have been employed both for classification and identification purposes by many systematists (Theobald et al., 1979; Rollins, 1993; Potgieter and van Wyk, 1999; Dickison, 2000; Batterman and Lammers, 2004). De Bary (1884) and Renner (1907) have also discussed hydathodes in Ficus. In Ficus diversifolia, hydathodes are scattered over the leaf lamina. They open to the adaxial surface via a circular, slightly depressed epidermal pad studied with 20-30 water pores (Lersten and Peterson, 1974).

Stomatal guard cells are essential to keep one particular component inside the plant i-e water. However, they must also allow the gaseous exchange essential for photosynthetic activity. Stomata and associated epidermal cells an important source of taxonomic characters. The pattern and frequency of stomata on any leaf surface are under tight genetic control, but may be modified by environmental parameters such as the availability of CO₂ (Croxdale, 2000; Glover, 2000). The plant epidermis consists of three main cell types: pavement cells, guard cells and their subsidiary cells that surround the stomata and trichomes, otherwise known as leaf hairs. The present study confined to leaf epidermal features of same tropical and sub tropical species of Genus Ficus (Moraceae). Some species have investigated first time for anatomical characters that might be useful plant biologist to this genus at global level.

MATERIAL AND METHODS

Anatomy

Fresh samples for the type and shape of leaf epidermal cell studies exact localities and voucher specimen details are presented in Table 1. Specimen were collected from the field and prepared according to the modified method of Clark's (1960). The leaves were placed in test tubes and treated with 88% lactic acid, in water bath (Model, Memmert-91126-FRG, Germany) at 100 °C for 20-40 min. They were then removed from the test tube into a Petri dish. A drop of lactic acid was used to soften the tissue of leaf due to which

it's peeling off is made easy. Slides of both abaxial and adaxial sides of leaves were prepared and observed under light microscope. Microphotographs were taken by using CCD digital camera (Model: DK 5000) fitted on Leica light Microscope (model: DM 1000). These micrographs were useful for identification and differentiation of epidermal cells on the basis of microscopic features.

Statistical analyses

Cluster analysis and principle component analysis (PCA) are two techniques commonly used in numerical classification. Measurements were taken for the length and width of stomata, trichome and pavement cells with the help of scale fitted in light microscope (Model: MX 5200H Meiji). The anatomical measurements were compiled on recording sheets. Mean figures were entered into o Microsoft Excel spreadsheet and the raw data then coded to allow analysis using Statistica 7.9 software.

RESULTS AND DISCUSSION

The Foliar epidermal anatomy is one of the most noteworthy taxonomic characters from the systematic point of view and the taxonomic studies of a number of families are made on the basis of leaf epidermis (Bhatia, 1984; Stace, 1984; Jones, 1986; Baranova, 1972). Although taxonomists lately realized the importance of microscopic features of the epidermis, taxonomic monographs are now considered incomplete without them (Rejdali, 1991). In the present study, foliar epidermal anatomy of 22 species in which 13 native and 9 cultivated species of Ficus L. was investigated. Both qualitative and quantitative characteristics of adaxial and abaxial foliar epidermis of Ficus L. were evaluated. During this study the use of light microscopy has made it possible in depth to evaluate leaf surface features such as shape of pavement cells, stomata and trichomes types.

The studies proved very rewarding and resulted in exploration of valuable intergeneric and interspecific variations in the configuration of foliar epidermal morphology that can be used as an important supportive taxonomic tool to demarcate many species of genus *Ficus* under study. Anatomical studies revealed clear cut differences in size and shapes of stomata, epidermal cells, presence of macro-and micro-hair, their size and type etc. Microcharacters of taxonomic significance obtained from selected features of the epidermal leaf surfaces, using light microscope are presented in Tables 2 and 3.

Pavement cells

Polygonal shape of pavement cells are presented in *F. palmata, F. religiosa, F. racemosa, F. bengalensis, F. carica, F. nerrifolia, F. hispida, F. auriculata, F. johannis, F. sarmentosa, F. subincisa, F. benjamina, F. tsiela,*

Table 1. List of *Ficus* L. species investigated, with location, status, distribution and voucher numbers.

S/n	Species	Locality	Status in Pakistan	Distribution worldwide	Voucher number
01	Ficus elastica Roxb.	Islamabad	Introduced	Nepal, China and cultivated everywhere.	KK-11
02	F. microcarpa L.f.	Islamabad	Introduced	Australia, S.China, Sri Lanka, New Caledonia, Ryukyu Islands and cultivated in N. Africa.	KK-12
03	F. lacor Ham.	Islamabad	Introduced	N. Australia, S.E Asia, Burma, India	KK-13
04	F.pumila L.	Abbottabad	Introduced	China, Japan	KK-14
05	<i>F.tsiela</i> Roxb.	larkana	Introduced	India, Sri Lanka, Maldive.	KK-15
06	F.callosa Willd.	Abbottabad	Introduced	Indomalaysia, South, Central and Maharashtra Sahyadris.	KK-16
07	F.virgata Roxb	Poonch	Introduced	Nepal, Somalia, S.Egypt, Peninsula, India.	KK-17
80	F.benjamina L.	Islamabad	Introduced	N. tropical Australia, U.S.A, Malaysia, Indonesia, China	KK-18
09	F. lyrata Warb.	Islamabad	Introduced	Tropical Africa.	KK-19
10	F. palmata Forrsk.	Islamabad	Wild	Nepal, Somalia, S.Egypt, Peninsula, India.	KK-20
11	F. religiosa L.	Islamabad	Wild	Bangladesh, Cylon, Thailand, China and cultivated in N. Africa, U.S.A, Middle East.	KK-21
12	F. racemosa Roxb.	Islamabad	Wild	Thailand, India, sri Lanka, Malaysia, Australia, Indonesia.	KK-22
13	F. bengalensis L.	Rawalpindi	Wild	India, Bangladesh and cultivated in tropics.	KK-23
14	F. carica L.	Swat	Wild	N. Africa, Europe, Russia, Middle East	KK-24
15	F.subincisa Buch.	Abbottabad	Wild	Burma, India, S.E Asia, N.Australia.	KK-25
16	F. foveolata Wall.	Poonch	Wild	N. India, Bangladesh, Burma, China.	KK-26
17	F. virens Dryand.	Islamabad	Wild	N. Australia, S.E Asia, Burma, India.	KK-27
18	F. nerrifolia J.E. Sm. in Ress	Poonch	Wild	Temperate subhimalyan region , India.	KK-28
19	F. hispida L. f.	Muzafarabad	Wild	India, New guinea, Malaysia, N. Australia.	KK-29
20	F.auriculata Wall.	Nathiagali	Wild	China, Japan, Burma, Bhutan and India.	KK-30
21	F. johannis Boiss.	Abbottabad	Wild	Iran, Afghanistan.	KK-31
22	F. sarmentosa Buch.Ham	Abbottabad	Wild	N. India, Bangladesh, Burma	KK-35

F. pumila, F. lacor and F. microcarpa as shown in (Figure 1 and 2 C, L, E, N, H and Q), from both adaxial and abaxial surface while in F. foveolata and F. virens adaxial surface is polygonal and abaxial surface is irregular as shown in (Figures 2D and M). Both polygonal and irregular shapes of epidermal cells are represented in most of the

species of genus *Ficus* (Sonibare *et al.*, 2005). In *F.callosa* undulating type of cells observed on adaxial surface as shown in (Figure 1 G). Isodiametric cell shape observed on abaxial surface of *F. virgata* (Figure 2O). Whilethe measurements of all species are given in Table 3. Distribution of length and width of pavement cells

on both sides has shown in Figures 6 and 7.

Stomata

The stomata are restricted to the abaxial surface of lamina (hypostomatic) except in *F. callosa* in

 Table 2. Anatomical Characterization of leaf epidermis of genus Ficus L. based on (LM).

C/m	Charles	Pavement cell shape	Type of stomata	Trichomes type		Cuticular m	embrane
S/n	Species	Adaxial /Abaxial	Adaxial/Abaxial	Adaxial	Abaxial	Adaxial	Abaxial
01	F. elastica	Polygonal to isodiametric/ irregular	Absent / Paracytic	Absent	Absent	Smooth	Smooth
02	F. microcarpa	Polygonal/ Polygonal	Absent / Paracytic	Absent	Absent	Smooth	Smooth
03	F. lacor	Polygonal/ Polygonal	Absent / Paracytic	Absent	Absent	Smooth	Smooth
04	F. pumila	Polygonal/polygonal	Absent / Paracytic	bulbous base, Unicellular and non glandular	Unicellular, thin and non glandular	Smooth	Scabrous
05	F. tsiela	Polygonal/ Polygonal	Absent / Paracytic	Absent	Absent	Smooth	Smooth
06	F. callosoa	Undulating/Polygonal	Anomocytic / Absent	Absent	Absent	Smooth	Slightly pubescent
07	F. virgata	Polygonal/isodiametric	Absent / Paracytic	Absent	Absent	Smooth	Smooth
80	F. benjamina	Polygonal/ Polygonal	Absent / Paracytic	Absent	Absent	Smooth	Smooth
09	F. lyrata	Polygonal/ Polygonal to irregular	Absent / Paracytic	Absent	Absent	Smooth	Smooth
10	F. palmata	Polygonal/ Polygonal	Absent / Paracytic	Bulbous base, prickle and non glandular	Unicellular and non glandular	Scabrous	Pubescent
11	F. religiosa	Polygonal/ Polygonal	Absent / Paracytic	Absent	Absent	Smooth	Slightly smooth
12	F. racemosa	Polygonal/ Polygonal	Absent / Paracytic	Absent	Unicellular, thin and non glandular	Slightly glabrous	Slightly glabrous
13	F. bengalensis	Polygonal/ Polygonal	Absent / Paracytic	Absent	Unicellular and non glandular	Smooth	Smooth
14	F. carica	Polygonal/ Polygonal	Absent / Paracytic	Prickle, unicellular and non-glandular	Thin and dense	Scabrous	Pubescent
15	F. subincisa	Polygonal/ Polygonal	Absent / Paracytic	Multicellular, falcate shaped	Bicellular and glandular	Scabrous	Scabrous
16	F. foveolata	Polygonal/Irregular	Absent / Paracytic	Absent	Absent	Smooth	Pubescent
17	F. virens	Polygonal/Irregular	Absent / Paracytic	Absent	Absent	Smooth	Slightly scabrous
18	F. nerrifolia	Polygonal/ Polygonal	Absent / Anomocytic	Absent	Absent	Smooth	Smooth
19	F. hispida	Polygonal/ Polygonal	Absent / Paracytic	Absent	Unicellular, prickle, bulbous base and non- glandular	Scabrous	Scabrous
20	F. auriculata	Polygonal/ Polygonal	Absent / Paracytic	Unicellular, bulbous base and non- glandular	Unicellular and non glandular	Smooth	Pubescent
21	F. johannis	Polygonal/ Polygonal	Absent / Paracytic	bulbous base, thin and unicellular	Thin, unicellular	Scabrous	Pubscent to Scabrous
22	F. sarmentosa	Polygonal/ Polygonal	Absent / Paracytic	Unicellular and non glandular	Unicellular and non glandular	Smooth	Pubescent

Table 3. Summary of quantitative epidermal characteristics of Genus Ficus L. based on (LM)

Cassias	Pavement cells	Stor	mata	Trichomes		
Species	Length (μm)	Width (μm)	Length (μm)	Width (μm)	Length (μm)	Width (μm)
Ficus elastica	15.5(12.5-20) / 24.5(22.5-25)	14(12.5-17.5) / 28.5(25-32.5)	25.5(22.5-27.5)	10.5(7.7-12.5)	Absent	-
F. microcarpa	60(47.5-75) / 25.5(20-37.5)	44.5(50-70) / 23(20-25)	18(15-20)	13(10-17.5)	Absent	-
F. lacor	21.5(15-25) / 27(22.5-30)	18(12.5-20) / 42(25-67.5)	11.5(10-12.5)	11(10-12.5)	Absent	-
F .pumila	31(20-42.5) / 28(22.5-35)	22.5(17.5-30) / 25(15-32.5)	10.5(7.5-12.5)	8.25(6.25-10)	86.5(62.5-120)	12(10-15)
F .tsiela	30.5(22.5-42.5) / 33(22.5-42.5)	16.5(12.5-20) / 19.5(15-25)	27(22.5-30)	20(17.5-22.5)	Absent	-
F. callosoa	55.5(50-62.5) / 55(50-60)	38.5(27.5-50) / 38.75(27.5-50)	25(20-27.5)	22(20-23.75)	Absent	-
F. virgata	25(17.5-32.5) / 21(17.5-22.5)	18(17.5-20) / 13(10-15)	11.25(10-12.5)	11.26(10-12.5)	Absent	-
F.benjamina	13(10-15) / 17.5(12.5-22.5)	14(7.5-17.5) / 11.5(7.5-17.5)	16.25(13.75-17.5)	11(10-15)	Absent	-
F. lyrata	23.5(20-27.5) / 21.5(15-25)	20(15-27.5 / 24(20-25)	23(17.5-27.5)	14.5(12.5-20)	Absent	-
F. palmata	39(25-50) / 25.5(20-37.5)	34(25-40) / 23(20-25)	11.25(10-12.5)	8.25(6.25-10)	225(75-300)	40(20-62.5)
F. religiosa	37.5(12.5-50) / 44.5(30-67.5)	29.5(25-42.5) / 43(35-57.5)	24(20-27.5)	15(15-15)	Absent	-
F. racemosa	31.5(22.5-40) / 22.5(17.5-25)	23(12.5-27.5) / 9.5(7.5-12.5)	11.5(10-12.5)	4.6(10-12.5)	200(150-250)	31.5(20-50)
F. bengalensis	71.5(62.5-87.5) / 24(17.5-35)	60.5(50-77.5) / 55.5(48-60)	21.5(20-22.5)	20(15-25)	115(75-150)	19.5(12.5-25)
F. carica	36.5(32.5-45) / 28(20-37.5)	40.5(27.5-50) / 25.5(20-30)	14.25(11.25-17.5)	11.37(10-13.75)	267.5(200-500)	30(20-57.5)
F. subinciosa	27.5(17.5-32.5)/35(20-42.5)	38.5(27.5-45) / 28(20-45)	19.5(17.5-22.5)	14.5(12.25-20.25)	350.5(27.5-550)	37.5(20-62.5)
F. foveolata	33.5(25-37.5) / 23.5(17.5-37.5)	25(20-30) / 19.5(17.5-20)	11.5(10-12.5)	11.5(10-12.5)	Absent	-
F. virens	72(65-77.5) / 18(17.5-22.5)	55(47.5-60) / 15.5(12.5-17.5)	20.5(16.25-25)	16.5(12.5-20)	Absent	-
F. nerrifolia	29.5(25-37.5) / 33.5(25-42.5)	23(20-27.5) / 20.5(15-30)	20.83(20-22.5)	14.16(12.5-15)	Absent	-
F. hispida	31(25-47.5) / 30.5(25-45)	20.5(17.5-25) / 22(17.5-25)	15(12.5-17.5)	18.75(12.5-25)	11.5(7.5-15)	90.5(75-102.5)
F.auriculata	34(30-40) / 31.5(25-35)	19.5(15-22.5) / 15(10-17.5)	19.5(17.5-22.5)/	11.37(10-13.75)	245(150-375)	39(30-45)
F. johannis	41.5(37.5-47.5) / 34.5(25-50)	28.5(22.5-32.5) /18(15-22.5)	14.25(11.25-17.5)	12(11.25-12.5)	162.5(150-200)	50(30-50)
F. sarmentosa	34(27.5-47.5) / 24(17.5-35)	24(22.5-25) / 17(12.5-20)	21.5(20-22.5)	20(15-20)	115(75-127.5)	16.5(10-20)

which stomata present on adaxial surface as shown in (Figure 2P). *F.elastica* has no stomata on its adaxial surface (Valenzuela, 1998) and it is also shown in (Figure .A). The taxonomic significance of stomata distribution and morphology of different species of genus *Ficus* was studied by Sonibare et al. (2005). Large number of stomata present in most of the species, including *F.*

bengalensis, F. nerrifolia, F. hispida, F. auriculata, F. johannis, F. virens and F.sarmentosa but there are few in F. palmata, F. religiosa F. racemosa and F. carica. Stomata are mostly of paracytic type except in F. nerrifolia and F. callosa as they have anomocytic type. While the mean length and width of all species given in Table 3. Distribution of length and width of stomatal cell also shown in

Figure 4.

Trichomes

Tricomes are observed on both surfaces in few species, including *F. carica* as shown in (Figure 2 B and J) *F. racemosa, F. subincisa, F. auriculata*.

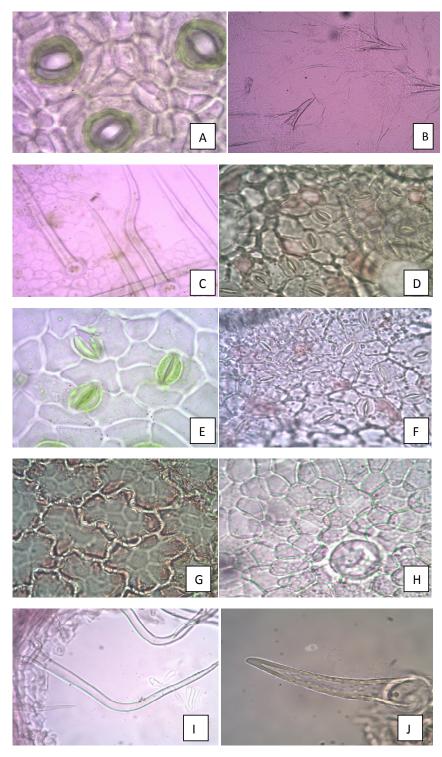


Figure 1. Characteritics of abaxial epidermal cells. A, dense stomata with pavement cells of F. elastica (40x). B, bulbous base prickle shape trichomes of F. carica (10x).C, long trichomes and pavement cells of F. palmata (20x). D. showing dense stomata, peltate glands and pavement cells of F. foveolata (20x). E, stomata and pavement cells of F. religiosa (40x). F, stomata and pavement cells of F. virgata (20x). G, showing undulating pavement cells of F. callosoa (40x). H, peltate gland with pavement cells of F. johannis (40x). I, unicellular, non glandular long trichome and bicellular glandular macrohair of F. subincisa (20x). J, unicellular bulbous base trichome of F. hispida (20x).

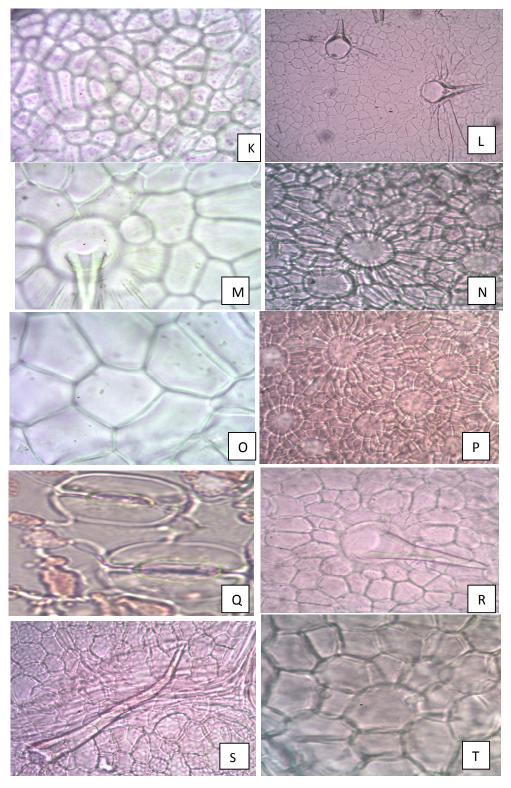


Figure 2. Characteristics of adaxial epidermal anatomy of Ficus. K. pavement cells of F.elastica (10 x). L, pavement cells and prickle shape trichomes of F.carica (10x). M, bulbous base trichome and pavement cells of F. palmata (40x). N, pavement cells and peltate glands of F. foveolata (20x). O, pavement cells of F. religiosa (40x). P, pavement cells and peltate glands of F. virgata (20x). Q, presence of adaxial stomata of F. callosoa (100x). R, bulbous base trichome with pavement cells of F. johannis (40x). S, show long thin unicellular trichome and pavement cells of F. subincisa (20x). T, pavement cells of F. hispida (40x).

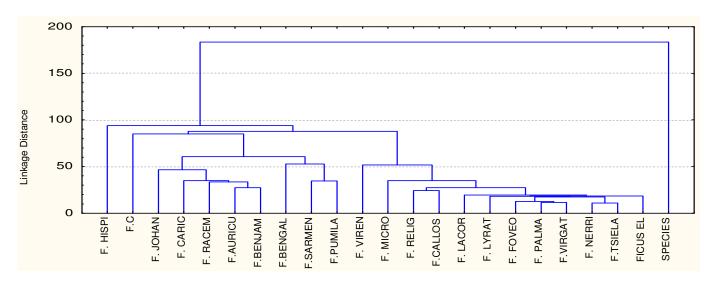


Figure 3. Cluster analysis of 22 variables.

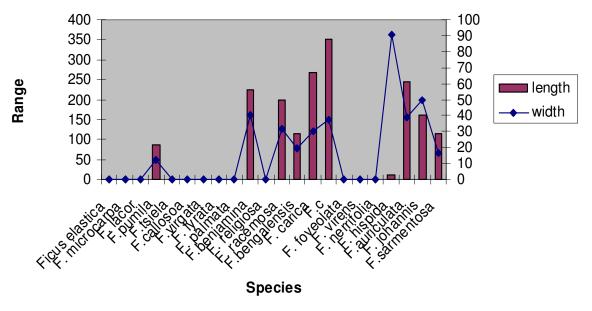


Figure 4. Distribution of length and width of stomatal cells of Ficus.

F. johannis and F. sarmentosa and in F. palmata as shown in (Figure 1 C and L), while in F. bengalensis and F. hispida (Figure. H) trichomes observed only on abaxial surface. Whereas in F. religiosa F. nerrifolia and F. virens trichomes are absent as shown in (Figure E and N). The taxonomic value of homes in angiosperm is well documented in the botanical literature (Theobald et al., 1979). The nature of the trichomes in the genus seems to be more reliable than their mere presence or absence. In present study, trichomes present in some species while absent in others. And in the species where the trichomes present observed on both surfaces of leaf. Most of the

species have unicellular but the *F. subincisa* has multicellular microhairs observed shown in (Figure 1I). Distribution of length and width of trichome shown in Figure 5. Trichome type has been found to be of little diagnostic value in the genus *Ficus*. (Sonibare et al., 2005). Their qualitative characters given in Table 2 while quantitative in Table 3.

Cuticular membrane

The cuticular membrane of mostly species were smooth,

	Table 4. Correlation	analysis	of 8	parameters	of some	Ficus species
--	----------------------	----------	------	------------	---------	---------------

	Stomatal	Var2	Trichome	Var4	Pavement	Var6	Var7	Var8
Stomatal	1							
Var2	0.27796356	1						
Trichome	-0.210907613	-0.15567	1					
Var4	-0.2432497	0.454594	0.55309	1				
Pavement	0.510218874	0.268371	0.013355	-0.00298	1			
Var6	0.449582713	0.180363	0.236891	0.027456	0.900994	1		
Var7	0.07944487	0.335464	-0.01753	0.04465	0.214039	0.118611	1	
Var8	-0.074348336	0.030948	-0.16394	-0.19361	0.03956	0.121434	0.61118	1

Marked correlations are significant at p < 0.05000.

(glabrous) including *Ficus elastica*, *F. microcarpa*, *F. lacor F. tsiela*, *F. virgata*, *F. benjamina*, *F. lyrata*, *F.bengalensis and F. nerrifolia*. The outer surface of the cuticle is smooth to weakly undulate in most species of *Ficus* (Sonibare *et al.*, 2005). *While* scabrous membrane observed in *F. palmata*, *F. johannis*, *F. carica*, *F. subincisa* and *F. hispida*. While abaxial surface was pubescent in *F. palmata*, *F. carica*, *F. foveolata*, *F. auriculata*, *F. johannis* and *F. sarmentosa*.

Statistical analyses

Correlation analysis

Table 4 shows the correlation coefficient of the eight quantitative parameters. It is observed that there is highly significant correlation between stomatal lengths and pavement cell length of adaxial side, trichome length and trichome width, pavement cell length of adaxial and pavement cell width of adaxial, pavement cell length of abaxial and pavement cell width of abaxial side.

Principle component analysis (PCA)

PCA is a method used for the reducing the dimensions of the original data. As presented in Table 5. Factor 1 explained the 64.86% of the total variance and shows significant values of *F. elastica* (0.848), *F. lacor* (0.88), *F. pumila* (0.641), *F. virgat* (1.077), *F. lyrata* (1.189), *F. palmata* (1.00), *F. racemosa* (0.98) and *F. foveolata* (0.751). These species predicted positive factor loading due to their resemblance in many morphological qualitative features i.e shape and cuticular membrane of leaf and climatic conditions are also same as following species grow at interactive temperature. All these species have highly significant values and are strongly associated and provide some strength to the existing classification. *F. microcarpa* (-0.833), *F. callosa* (-1.781), *F. religiosa* (-0.74) and *F.*

bengalensis (-1.767) predicted negative values of the species of factor 1 as their climatic conditions are same. And resemble morphologically as all these species are large trees. On the basis of anatomical characters these species are found to be closely related.

The second factor explained 81.47% of the total variance and have highly significant factor values of species including F. elastica Roxb. (-1.062), F. lacor Ham. (-1.41), F.callosa Willd. (-1.27), F. palmata Forrsk. (-0.706), F. religiosa L. (-1.49), F. foveolata Wall. (-0.596)), F. tsiela Roxb. (-0.69) and F. nerrifolia J.E.Sm in Ress. (-0.723), all these species are highly correlated and strongly associated with stomatal width, trichome width and pavement cell width of abaxial and adaxial sides. F. benjamina L. (1.128), F. racemosa Roxb. (1.127), F. bengalensis L. (0.701), F. carica L. (1.11), F. subincisa Buch. (1.31), F. hispida. L.f(1.629), F. auriculata Wall. (1.18) and F. johannis Boiss. (1.109) shows positive correlation among them as their climatic conditions are almost same and have resemblance in morphological characters as F. carica L. and F. johannis Boiss have same shape of leaves and are closely related species. Following parameters are good taxonomic tools for the identification and classification of species. This analysis also proved that foliar anatomical studies taxonomically significant to delimit taxa.

Cluster analysis

Cluster analysis produces a hierarchical classification of taxa based on the similarity matrix. As presented in Figure 5 dendrogram was constructed on the basis of length and width of epidermal cells, length and width of stomata, trichomes on adaxial and abaxial surface in order to establish relationship among different taxa as Group 1 has 12 species including *F. elastica* Roxb., *F. tsiela* Roxb., *F. nerrifolia* J.E.Sm., *F. virgata* Roxb., *F. palmata* Forrsk., *F. foveolata* Wall., *F. lyrata* warb., *F. lacor* Ham, *F. callosa* Willd., *F. religiosa* L., *F. microcarpa* L.f. and *F. virens* Dryand. as all these have morphological

Table 5. Principal Component Analysis (PCA) of Plant Parameters.

Species	Factor 1	Factor 2		
Ficus elastica	0.8	-1.06		
F. microcarpa	-0.83	-0.40		
F. lacor	0.88	-1.41		
F. pumila	0.64	-0.19		
F. tsiela	0.06	-0.69		
F. callosoa	-1.78	-1.27		
F. virgata	1.07	-0.32		
F. lyrata	1.18	-0.36		
F. palmata	1.00	-0.70		
F. benjamina	0.01	1.12		
F. religiosa	-0.74	-1.49		
F. racemosa	0.98	1.12		
F. bengalensis	-1.76	0.70		
F. carica	-0.04	1.11		
F. subincisa	-0.25	1.31		
F. foveolata	0.75	-0.59		
F. virens	-2.45	-0.46		
F. nerrifolia	0.19	-0.72		
F. hispida	-0.28	1.62		
F. auriculata	0.39	1.18		
F. johannis	-0.019	1.10		
F. sarmentosa	0.12	0.42		

^{*} loading higher than 0.5 are italicized.

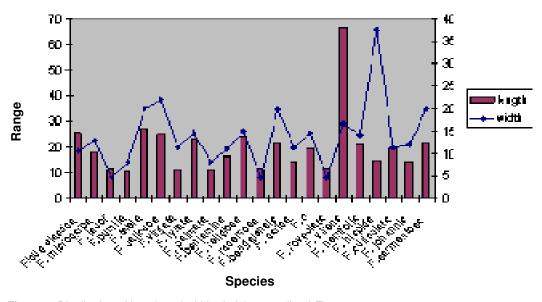


Figure 5. Distribution of length and width of trichome cells of Ficus

resemblance. In second Group there are 9 species in which *F. pumila* L. is closely related with *F. sarmentosa* Buch. Ham. as both are climbers, *F. bengalensis* L, *F. benjamina* L. is closely related with *F. auriculata* Wall. as

both are morphologically shrubby trees. *F.* racemosa Roxb., *F.* carica *L.* and *F.* johannis Boiss. and *F.* subincisa Buch. (indicated with *F.c* in Figures 4 to 7 and Figure 3) closely related species as their morphological

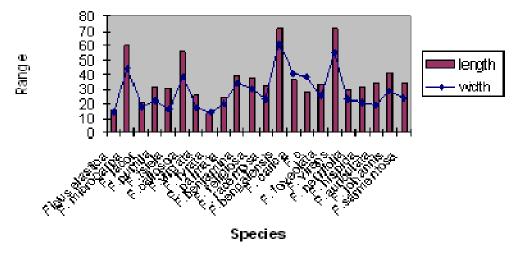


Figure 6. Distribution of length and width of pavement cells on adaxial side.

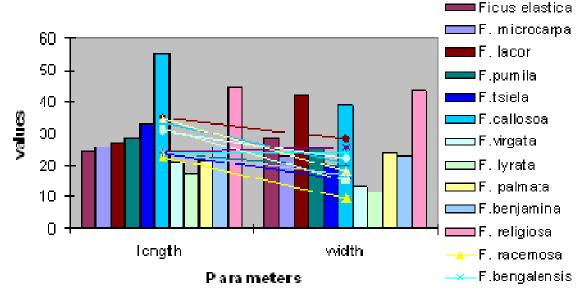


Figure 7. Distribution of length and width of pavement cells of adaxial side.

appearance of leaves have resemblance and are small size trees; and *F.hispida* L.f. variable forms an separate linkage as it is morphological different from all other species as well as differ in qualitative parameters of anatomical studies.

The values of the 8 quantitative parameters that characterize each of the 22 species of genus *Ficus* subjected to principle component analysis (PCA) are the same as for the cluster analysis. The result shows the degree of affinity among the species within the genus. The grouping observed as a result of the principal component analysis (PCA) provides some strength to the existing classification (Berg, 1989; Corner, 1965;

Ramirez 1977).

Conclusion

The differences were observed in the abaxial and adaxial surfaces of the same species while most of anatomical characters are stable in most of the species. It is possible that each species respond to its environment in particular way by modifying the certain features to improve its adaptation. Statistical analysis also supports the existing classification.

It is concluded that same characters discover in this

study are very useful and interesting at global level to identify and differentiate the different species of genus Ficus.

ACKNOWLEDGEMENT

The authors wish to thank Quaid-i-Azam University, Islamabad. Pakistan for supporting this work.

REFERENCES

- Baranova M (1972). Systematic anatomy of the leaf epidermis in the Magnoliaceae and some related families. Taxo., 21: 447–469.
- Batterman MRW, Lammers TG (2004). Branched foliar trichomes of Lobeliodeae (Campanulaceae) and the infrageneric classification of Centropogon. Syst. Bot., 29(2): 448-458.
- Berg CC (1989). Classification and distribution of *Ficus*. Experientia, 45: 605-611
- Bhatia RC (1984). Foliar Epidermal Studies of *Heliotropium supinum* L. Folia Geobotanica and Phytotaxonomica, 19(4): 381-385.
- Clark J (1960). Prepration of leaf epidermis for topographic study. Stain Technol., 35: 35–39.
- Corner EJH (1965). Checklist of *Ficus* in Asia and Australasia with keys to identification. Gdns. Bull. Singapore, 21: 1-186.
- Croxdale JL (2000). Stomatal patterning in angiosperms. Am. J.Bot., 87: 1069–1080.
- De-Bary A (1884). Comparative Anatomy of Vegetative Organs of the Phanerogams and Ferns. English Translation by Bower FO and Scott DH. Oxford. Oxford University Press.
- Dickison WC (2000). Integrative Plant Anatomy. San Diego: Harcourt Academic Press.
- Fahn A (1985). Plant Anatomy. Oxford: Pergamon Press.
- Ghafoor A (1985). Moraceae EN, Ali SI (Eds.). Fl. Pak., 171: 1-54.
- Glover BJ (2000). Differentiation in plant epidermal cells. J. Exp. Bot., 51: 497–505.
- Janzen DH (1979). How to be a Figure. Ann. Rev. Ecol. Syst., 10: 13-15
- Jones JH (1986). Evolution of the Fagaceae: the implications of foliar features. Ann. Missouri Bot. Garden, 73: 228–275.

- Karatela YY, Gill SL (1984a). Epidermal structures and stomatal ontogeny in some Gymnosperms from Nigeria. Feddes Rep. 95(5-6): 351-354.
- Karatela YY, Gill SL (1984b). Structure and ontogeny of some Nigerian ferns. Nova Hedwigia, 40: 477-486.
- Lersten HR, Peterson WH (1974). Anatomy of hydathodes and pigment disks in leaves of *Ficus diversifolia* (Moraceae). Bot. J. Linn. Soc., 68: 109-113
- Potgieter MJ, Wyk AI (1999). Leaf anatomy of the South African lcacinaceae and its taxonomic significance. S. Afr. J. Bot., 65: 153-163
- Ramirez WB (1977). A new classification of *Ficus*. Ann. Missouri Bot. Garden, 64: 296-310.
- Rejdali moh FLS (1991). Leaf micromorphology and taxonomy of North African species of *Sideritis* L. (Lamiaceae). Bot. J. Linn. Soc., 107: 67–77.
- Renner O (1907). Beitrage zur anatomie und systematic der Artocarpeen und Conocephalen, insbesondere der Gattung *Ficus*. Bot. Jahrb., 39: 319-448.
- Rollins RC (1993). The Cruciferae of Continental North America. Stanford University Press.
- Scott T (1996). Concise encyclopedia biology. p. 77
- Sonibare MA., Jayeola AA, Egunyomi A, Murata J (2005). A survey of epidermal morphology in *Ficus* Linn. (Moraceae) of Nigeria. Bot. Bull. Acad. Sin., 46: 231-238.
- Stace CA (1965). The significance of leaf epidermis in the taxonomy of Combretaceae I. A general review of the tribal, generic and specific characters. Bot. J. Linn. Soc., 59: 229-250.
- Stace CA (1984). The taxonomic importance of leaf surface. In: Heywood VH, Moore DM, eds. Current concepts in plant taxonomy. London: Academic Press, 67–94.
- Theobald WL, Krahulik JL, Rollins RC (1979). Trichome description and classification. *In* C.R. Metcalfe and L. Chalk (eds.), Anatomy of the Dicotyledons, 2nd edition, Clarendon Press, Oxford, 1: 40-53.
- Valenzuela JL (1998). Ficus Epidermal Structures: A Tool for Introducing Leaf Anatomy. University of California Press on behalf of the National Association of Biology Teachers. 60(3):216-219.
- Watson L (1962). The taxonomic significance of stomatal distribution and morphology I. Epacridaceae. N. Phytol., 61: 36-40.