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Epidermal study of medicinal plants with special reference to identification, adulteration and authentification of crude leaf drugs

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Abstract

Leaf epidermal cells are important tool for the identification, adulteration and authentification of crude leaf drugs. Therefore, for baseline data generation, leaf epidermal cell complex of 20 medicinal plants of family, Apocynaceae was studied. Epidermal cells were polygonal (5-6 sided) in twelve species on adaxial, three species on abaxial and in rest, 5-7 sided were dominant on both surfaces, straight anticlinal walls were dominant on both the surfaces, curved condition is predominant on adaxial but wavy to sinuate is predominant on abaxial surface. Smooth surface was dominant on both surfaces, followed by undulated. The epidermal cell frequency differed a lot within the same surface and also on both the surfaces of the same leaf of the species. Highest frequency is dominated at middle zone on both surface. The costal cells are present on both surfaces, as they are trapezoidal, were dominant on abaxial and polygonal predominant on both surfaces. Anticlinal walls were straight on both surfaces. The thick walls were dominated on abaxial, outer wall flat, surface smooth on both surfaces. The epidermal and costal cells study is the pharmacognostic baseline data generation of 20 species of Apocynaceae.

Key words: Apocynaceae, Epidermis, Micromorphology, Costal cells, Adulteration

Introduction

The epidermal cell complex forms the outermost layer of plant body. It consists of epidermal cells, costal cells, and special epidermal cells (expect stomatal complex). Epidermal cells are confined to inter-costal areas, the costal cells are overlying on the veins and are structurally distinct from the other epidermal cells while the special epidermal cells are intermixed with costal cells or epidermal cells. These are often distinct due to their conspicuous shape, size and contents. The epidermis plays a vital role in identification of genuine drugs as well as adulteration check at interspecific and intrageneric level of market crude drugs, because vender sells the same drug with different name and many drugs under same name, to differentiate and also authenticate the drug is done based on leaf epidermal characters. Hence, epidermis on various organs of plants play a vital role in identification of the medicinal taxa. The quick and cheaper technique of identification, adulteration and authentification of crude leaf drugs is possible only by epidermal study of medicinal plants. In this context, a thorough investigation of medicinal plants of Apocynaceae has been carried out.

However, plant leaves by virtue of their position, shape and texture, they get exposed to various environmental conditions, hence, leaf epidermis play a vital role as it interacts with the exiting environment and get acclimatized to the exposed environment with modification within the epidermis (Sharma, 1972 and Madhavan *et al.,* 2009).

The leaves, bark, wood are the best accumulators of pollutants, specifically the heavy metals in them due to which not only productivity decreases, but also become accumulators, resulting into modification in biochemical and epidermal alteration in them. Therefore, plants are the

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indicators of pollution and they interact with the environment and modify themselves to suit to the changed environment (Srivastava *et al.,* 1980; Gupta and Ghouse, 1987; Seema *et al.,* 1991; Venkateshwar *et al.,* 1991 and Venkateshwar and Jeelani, 1992).

The foliar epidermal studies in Apocynaceae has been studied by several workers, However, different authors gave different names to the single type of cells or vice versa. The information regarding the various aspects of epidermal cell complex and costal cells, like shape of cells, anticlinal walls, surface characters, papillate nature, orientation, arrangement and distribution is negligible. Hence, to provide concrete data on medicinal plant epidermal cell complexes of leaf has been evaluated by micro-morphological study of leaf epidermal cell complex.

Material and Methods

Twenty species of the family Apocynaceae were investigated, which were collected from various places and their uses were depicted in Table 1. About 20 mature leaves/plant/location x5 plants were collected and fixed in carnou's fixative (Johansen, 1940), after two days, the fixative was replaced by 70% alcohol for preserving the material.

The baseline data on epidermis is chiefly based on microscopic study. The standard techniques were employed, the scraping method (Leelavathi, 1976) and "Triple acid method" (Ramayya and Rajagopal, 1968) which gave satisfactory results (Plates 1-7) for the preparation of epidermal peelings.

The peels were prepared from base, apex, middle lamina and margin locations of (Plates 8) the leaf, for both the surfaces, (Prabhakar *et al*., 1982). Sixteen slides/speices were prepared. Description of epidermal cell complex is followed (Plates 8-7). The microphotographs were taken by using olympus microscopic camera attached to the microscope. Plates 1 to 5 were prepared with four epidermal peels in each plate.

Results and Discussion

The epidermal cells were described as penta or hexagonal in *Alstonia scholaris*, *Ervatamia dirvaricata* (Kapoor *et al.,* 1969); *Holarrhena pubescens*, *Plumeria alba*, *P. rubra, Wrightia tinctoria*, *W. tomentosa* (Chandra *et al.,* 1969); *Rauwolfia canescens*, *Rauwolfia serpentina*, *Vallaris solanacea* (Sharma *et al.,* 1970); *Carissa carandas* (Kapoor and Mitra, 1979). *Plumeria rubra* (Trivedi and Upadhyay, 1977); and tetra to hexagonal in *Allamanda cathartica* (Kapoor and Mitra, 1979), However, Trivedi and Upadhyay (1973, 1977) described them as hexagonal on adaxial of *Alstonia scholaris*, *Rauwolfia canescens*, *Rauwolfia serpentina*, *Plumeria alba*, *Holarrhena pubescens*, *Wrightia tomentosa* and on abaxial of *Ervatamia dirvaricata* and irregular shapes in *Cascabela thevetia*, *Catharanthus roseus*, *Wrightia tinctoria*, *Vallaris solanacea*. Different authors mentioned different names to the same type of cells, and same name to two types of cells in the species and also differed on individual surfaces. But presently observed specific type of polygonal anisodiametric cells were maximum, followed by few polygonal isodiametric, dominant on both the surfaces and the cells were 5 to 7 sided (Tables 2 and 3; Plates 1-5).

The anticlinal walls of epidermal cells were described as straight on both surface of *Allamanda cathartica*, *Alstonia scholaris* (Kapoor *et al.,* 1969); *Plumeria alba*, *P. rubra*, *Wrightia tinctoria* (Chandra *et al.,* 1969); *Alstonia scholaris*, *Rauwolfia canescens*, *Rauwolfia serpentina*, *Carissa carandas*, *Cascabela thevetia*, *Plumeria alba, P.rubra, Holarrhena pubescens* (Trivedi and Upadhyay, 1977); *Cascabela thevetia* (Fjell, 1983); while sinuate on both surfaces of *Carissa carandas*, *Cascabela thevetia* (Kapoor *et al.,* 1969); *Catharanthus pusillus*, *Catharanthus roseus* (Sharma *et al.,* 1970), *Rauwolfia serpentina*, *Ervatamia dirvaricata*, *Wrightia tinctoria*, *W. tomentosa*, *Catharanthus roseus* and *Vallaris solanacea* (Trivedi and Upadhyay, 1973 and 1977).

Further, anticlinal walls were reported as adaxially straight and abaxially sinuate in *Holarrhena pubescens* (Chandra *et al.,* 1969); *Rauwolfia canescens, Rauwolfia serpentina Vallaris solanacea* (Sharma *et al.,* 1970) and vice versa in *Wrightia tomentosa* (Chandra *et al*., 1969), *Ervatamia dirvaricata* (Kapoor *et al*., 1969). The anticlinical walls are dominated with straight walled in all the species except in *Cascabela thevetia, Holarrhena pubescens*, where they are curved to wavy and curved to straight in *Wrightia tomentosa* and wavy in *W. tinctoria*, sinuate in *Aganosma calycina*, on both the surfaces. Futher, straight to curved dominated in all the species except in *Holarrhena pubescens*, *Rauwolfia canescens*, *Rauwolfia serpentina*, *Vallaris solanacea*, are with wavy to sinuate and sinuate to wavy in *Aganosma calycina*, *Wrightia tinctoria*, curved to wavy in *Allamanda cathartica*, *Catharanthus roseus*, *Wrightia tomentosa* on abaxial surface (Tables 2 and 3; Plates1-5).

Anticlinal walls of foliar epidermal cells are more conspicuously undulated on abaxial than on the adaxial surface (Cutter, 1977). But an opposite condition was also reported by Leelavathi (1976) and Kumar and Ramayya (1983) in *Parkinsonia acleata, Wagatea spicuta Humbodtia brunonia and Indigofera tinctoria.*

Aniclinal walls of foliar epidermal cells are usually straight in tree members and sinuate in herbs (Rajagopal. 1973) but Leelavathi (1976) reported sinuate anticlinal walls in some tree members and straight anticlinial walls in herbaceous members which differed by the study of Rajagopal (1973). Similarly, in Apocynaceae, herbaceous members, the anticlinal walls are straight to curved (adaxially) while in shrubs, the anticlinal walls are straight on adaxial and wavy to sinuate on abaxial surface. In tree members and also in shrubs, it was observed that the adaxial anticlinal walls differ with abaxial surface (Table 3; Plates 1-5). Surface of epidermal cells are more striated on adaxial surface. than on abaxial surface. The striation on both surfaces of *Acocanthera venenata*, *Allamanda cathartica*, *Plumeria acutifolia*, *P.alba, P. rubra*, *Rauwolfia serpentina* and on adaxial of *Catharanthus pusillus*; abaxial of *Holarrhena pubescens* and reticulation on adaxial of *Carissa carandas*, *Vallaris solanacea* and in rest smooth surface was observed (Tables 2 and 3; Plates 1-5). This texture helps in prevention of diseases.

Table 1: Medicinal importance of 20 species of family apocynaceae

S. No.	Name of the plant	Habit	Habitat	Part used	Purpose/uses
1.	*Acocanthera venenata*. G.Don.	T	OC	Milky juice	Poisoning , Poisonous juice Applied on arrows for hunting
2.	*Aganosma calycina*.DC. Mr.W.B;M;H	C	OC		As substitute of *Alstonia scholaris*
3.	*Allamanda cathartica*.L	C	OC	Leaf juice / Bark	Purgative, emetic, cathartic, Hydrogogue in ascited. High does poisonous antidote to snake bite.
4.	*Alstonia scholaris* (L)R.Br	T	OC/W	Milky juice, leaves	Ulcers, ear-ache, women after confinement
5.	*Carissa carandas* .L.	T	W	Leaves	intermittent fever.
6.	*Carissa spinarum*	T	W	Roots	Rheumatism
7.	*Catharanthus pusillus* (Murroy)G.Don.	H	OC/W	Dried plant	Cattle poisoning, Anticancer
8.	*Catharanthus roseus* (L). G.Don.	H	OC/W	Entire plant	Diabetes, Anticancer
9.	*Cascabela thevetia* (L) Lippoid	S	OC/W	Entire plant	Urethral discharges, acrid, lastringent, worms, skin diseases, leucoderma, piles
10.	*Ervatamia dirvaricata* (L) Burkill	S	W/OC	Root	Astringent, digestible, diseases of the blood
11.	*Holarrhena pubescens* (Buch-Hand)Wallich ex.Don.	T	OC/W	Leaves	Astringent, galactogogue, gonic, aphrodisiac, chronic bronchitis, lumbago, urinary discharges, boilsregulate menstruation.
12.	*Nerium indicum* Miller	S	OC/W	Leaves , roots	Opthalmia, powerful repellents,
13.	*Plumeria acutifolia* Poiret.,	T	OC/W	Leaves	Anti-inflammatory, rheumatism, abortion, thirst and cough
14.	*P.alba Sp.*	T	OC/W	Leaves	
15.	*P.rubra*.L.	T	W/OC	Leaves	
16.	*Rauwolfia canescens* L	S	W/OC	Leaves	Removal of opacities of the cornea andRoot used in hypertension
17.	*Rauwolfia serpentina* (L) Bebth ExKurz	S	OC/W	Root	
18.	*Vallaris solanacea* (Roth) Kuntze.g	S	OC/W	Milky juice	Old sores, sinuses
19.	*Wrightia tinctoria* (Roxb) R.Br.	T	W	Bark	Menstrual and renal complaints
20.	*Wrightia tomentosa* Roemer&Schulter	T	W		

OC-ornamental and Cultivated; W-Wild, W.B= West Bengal (Botanical garden)

Table 2: Characters of epidermal cells and freqency (mm^2) in the family apocynaceae adaxial surface

S.No.	Name of the plant	Cell sides	Mostly	Few	ALCW	LB	LM	LA
1.	*Acocanthera venenata*.G. Don.	5-7	PAD	RID	ST,ST-CD	1326.4951	1380.4804	1295.6464
2.	*Aganosma calycina*.DC. Mr.W.B;M;H	5-7	PAD	RID	SEU	1357.3439	1341.9196	1311.0708
3.	*Allamanda cathartica*.L	5-7	PAD	RID	ST,ST-CD	555.2770	570.7014	570.7014
4.	*Alstonia scholaris* (L)R.Br	5-7	PAD	RID	ST	1095.1297	1758.3772	1295.6464
5.	*Carissa carandas* L.	5-6	PAD	RID	ST	879.1886	979.4470	902.3252
6.	*Carissa spinarum*	5-6	PAD	RID	ST,ST-CD	2583.5807	2545.0198	2641.4211
7.	*Catharanthus pusillus* (Murroy) G.Don.	5-6	PAD	RID	ST,ST-CD	1634.9975	1804.6296	1665.8312
8.	*Catharanthus roseus* (L). G.Don.	5-6	PAD	RID	ST,ST-CD	1195.3881	1349.6317	1395.9047
9.	*Cascabela thevetia* (L) Lippoid	5-6	PAD	RID	CD-WY	1388.1926	1326.4951	1419.0413
10.	*Ervatamia dirvaricata* (L)Burkill	5-7	PAD	RID	ST,ST-CD	13311.0708	1141.4028	1110.5541
11.	*Holarrhena pubescens* (Buch-Hand)Wallich ex.Don.	5-7	PAD	RID	ST	14113.2191	1426.7535	1241.6612
12.	*Nerium indicum* Miller	5-6	PID	RID	ST	1850.9235	1889.4844	1712.1042
13.	*Plumeria acutifolia* Poiret	5-7	PAD	PAD	ST	2120.8498	215941.07	1943.4697
14.	*P.alba Sp.*	5-6	PAD	PAD	ST	663.2476	902.3252	791.6463
15.	*P.rubra*.L.	5-6	PAD	PID	ST	2113.1377	2113.1132	2097.6807
16.	*Rauwolfia canescens* L	5-7	PAD	PID	ST	817.4912	902.3252	837.9283
17.	*Rauwolfia serpentina* (L) Bebth ExKurz	5-6	PAD	PID	ST	802.0668	809.7790	825.2034
18.	*Vallaris solanacea* (Roth) Kuntze.g	5-6	PAD	PID	ST-CD	832.9155	1079.7053	1203.0863
19.	*Wrightia tinctoria* (Roxb)R.Br.	5-6	PAD	PID	WY	1079.6929	1110.5541	1033.4204
20.	*Wrightia tomentosa* Roemer & Schulter	5-6	PAD	PID	CD-ST	1943.4472	2020.5715	1948.4826

CD= Curved; ST= Straight; PAD= Polygonal anisodiametric; PID= Polygonal, isodiametric; RID = Rectangular isodiametric SEU = Sinuate; WY= Way; LB= Leaf base; LM= Leaf midrib/middle area; LA= Leaf apex regions, ALCW: Anticlinal cell wall

Table 3: Characters of epidermal and cell freqency (mm^2) in the family apocynaceae, abaxial

S.No.	Name of the plant	Cell sides	Mostly	Few	ALCW	LB	LM	LA
1.	*Acocanthera venenata*.G.Don.	5-6	PAD	PID	WY-SEU	1295.6314	1318.7830	1349.6613
2.	*Aganosma calycina* DC. Mr.W.B;M;H	5-6	PAD	PID	DEU-WY	10.4204	1033.4204	940.8861
3.	*Allamanda cathartica* L	5-7	PAD	PID	CD-WY	686.3841	655.5354	616.9755
4.	*Alstonia scholaris* (L)R.Br	5-7	PID	PAD	ST	3655.5739	3925.5003	335.47989
5.	*Carissa carandas* L.	5-6	PAD	PAD	ST-CD	547.5648	542.1405	524.4283
6.	*Carissa spinarum*	5-7	PAD	PID	ST-CD	3462.7694	3354.7989	3655.5739
7.	*Catharanthus pusillus* (Murroy)G.Don.	5-7	PAD	PID	ST-CD-WY	1218.5247	1249.3589	1079.6929
8.	*Catharanthus roseus* (L).G. Don.	5-7	PAD	PID	CD-WY-SE	1195.3881	1033.4323	1318.9356
9.	*Cascabela thevetia* (L) Lippoid	5-7	PAD	PID	ST-CD	2699.2634	2845.7949	2660.7025
10.	*Ervatamia dirvaricata* (L) Burkill	5-7	PAD	PID	CD-ST	1187.8133	994.8713	1048.8566
11.	*Holarrhena pubescens* (Buch-Hand)Wallich ex.Don.	5-7	PAD	PID	WY-SE	1064.2810	987.1592	1048.8566
12.	*Nerium indicum* Miller	5-7	PAD	PID	ST/CD	3300.8136	3362.5110	3347.0867
13.	*Plumeria acutifolia* Poiret	5-7	PID	PAD	ST,ST-CD	1557.8606	1573.2850	154.2362
14.	*P.alba Sp.*	5-7	PAD	PAD	ST-CD	718.7753	856.6521	709.5267
15.	*P.rubra.*L.	5-7	PAD	PAD	ST-CD	2506.4589	2313.6544	2352.2153
16.	*Rauwolfia canescens* L	5-7	PAD	PAD	WY-SE	164.7977	1295.6465	1218.5247
17.	*Rauwolfia serpentina* (L) Bebth ExKurz	5-7	PAD	PAD	WY-SE	1341.9195	1349.6317	1380.4804
18.	*Vallaris solanacea* (Roth) Kuntze.g	5-7	PAD	PAD	WY-SE	1024.7201	1025.7201	1203.1002
19.	*Wrightia tinctoria* (Roxb)R.Br.	5-7	PAD	PAD	SEU	701.8085	740.4694	717.3229
20.	*Wrightia tomentosa* Roemer & Schulter	5-7	PAD	PAD	CD-WY	78.6492	79.2812	76.4843

CD=Curved; PAD= Polygonal anisodiametric; PID= Polygonal isodiametric; SEU= Sinuate ; ST= Straight; WY= Wavy; ALCW: Anticlinal Cell wall

Table 4: Characters of costal cell as observed in the family apocynaceae adaxial and abaxial

S.No.	Name of the plant	Type of costal cells					
		Cell	Mostly	Few	Cell	Mostly	Few
1.	*Acocanthera venenata* G.Don.	TR	TRN	5SN	TRN	TRL	-
2.	*Aganosma calycina*.DC.Mr.W.B;M;H	TR	TRL	TRN	TRN	TRL	TRN
3.	*Allamanda cathartica* L	TRL	TRL	TRN	PAD	PAD	PADN
4.	*Alstonia scholaris* (L)R.Br	PAD	5SN	TRN	TRN	TRN	TRL
5.	*Carissa carandas* L.	PAD	5SN	PADN	TRL	TRL	TRN
6.	*Carissa spinarum*	-	-	-	-	-	-
7.	*Catharanthus pusillus* (Murroy)G.Don.	TRL	TRL	TRN	PAD	5SN	TRN
8.	*Catharanthus roseus* (L).G.Don.	TRN	TRN	-	TRL	TRN	-
9.	*Cascabela thevetia* (L) Lippoid	TRL	TRL	-	PAD	PID	PADN
10.	*Ervatamia dirvaricata* (L)Burkill	TRL	TRL	-	TRL	TRL	-
11.	*Holarrhena pubescens* (Buch-Hand) Wallich ex.Don.	TRL	TRL	TRN	PAD	5SN	PID
12.	*Nerium indicum* Miller	TRN	TRN	SQ	SQ	SQ	TRN
13.	*Plumeria acutifolia* Poiret	TRN	TRN	SQ	PAD	PAD	TRN
14.	*P.alba Sp.*	PAD	5-6	TRN	TRN	TRN	SQ
15.	*P.rubra*.L.	TRL	TRL	TRN	PAD	5-6	PADN
16.	*Rauwolfia canescens* L	-	-	-	-	-	-
17.	*Rauwolfia serpentina* (L) Bebth ExKurz	PAD	5-6	TRN	PAD	5-6	TRN
18.	*Vallaris solanacea* (Roth) Kuntze .g	TRN	TRN	SQ	TRN	TRN	SQ
19.	*Wrightia tinctoria* (Roxb)R.Br.	TRL	TRL	TRN	TRL	TRL	TRN
20.	*Wrightia tomentosa* Roemer&Schulter	TRL	TRL	TRN	TRL	TRL	TRN

Sq=Squarish; PAD= Polygonal anisodiametric; PADN= Polygonal anisodiametric non-linear; TRN= Trapezoidal non-linear; TRL= Trapezoidal linear cells.

The thickest epidermal cell walls observed on both the surfaces of *Nerium indicum* , thick on both surfaces of *Carissa carandas*, *Carissa spinarum*, *Ervatamia dirvaricata*, *Rauwolfia canescens* and thin on both the surface of *Aganosma calycina*, *Alstonia scholaris, Cascabela thevetia*, *Plumeria acutifolia*, *Rauwolfia serpentina, Vallaris solanacea*, *Wrightia tinctoria, Wrightia tomentosa* and in rest of the species, there is no correlation in between adaxial and abaxial surfaces. However, if the epidermal cells are thicker on adaxial than on abaxial, it will be thin or vise versa, observed in rest of the species (Tables 2 and 3, Plates 1-5).

The outer wall outgrowths led to form projections, which appear like varicose, and papillate, were observed on both the surfaces but flat walls observed in many species on adaxial surface expect on both the surfaces of *Allamanda cathartica, Holarrhena pubescens, Wrightia tomentosa* and on adaxial of *Alstonia scholaris, Catharanthus pusillus, Plumeria acutifolia, Rauwolfia serpentina* and adaxial of *Acocanthera venenata*, *P. alba* where they were convex. Further, on adaxial of *Carissa spinarum, Plumeria* alba, *Vallaris solanacea, Wrightia tinctoria* and on abaxial of *Plumeria acutifolia* and on both surfaces of *Plumeria rubra, Rauwolfia canescens* where they are papillate, verrucose on abaxial of *Alstonia scholaris,* and in rest of the species, flat outer walls are observed, (Tables 2 and 3, Plates 1-5; 8.8 J&K).

Epidermal cell frequency shown maximum variation within the surface and in both the surfaces of the same species in all the taxa expect in *Carissa spinarum*, *Rauwolfia serpentina*. But in some species, the cell frequency is medium throughout the leaf except on apex where there is low frequency on adaxial of *Plumeria rubra* and abaxial of *Aganosma calycina* and low on base, medium throughout the leaf on adaxial of *Allamanda cathartica*, were observed. Thus, there is no frequency correlation within the same surface or in both the surfaces of rest of the species. However, on the middle portion of the leaf, high frequency observed in ten species on adaxial and nine species on abaxial surface. Further, medium

frequency also observed on the middle portion of the leaf in eight species on adaxial and six species on abaxial surface. (Tables 2 and 3; Plates 1-5).

There is no information available regarding the orientation and arrangement and distribution of foliar epidermal cells in Apocynaceae but in the present study of Apocynaceae, it was revealed that the epidermal cells are variously oriented and irregularly arranged on leaf leaving costal occupancy.

The shape of the costal cells is mostly polygonal anisodiametric on adaxial surface of *Alstonia scholaris, Carissa carandas*, *Plumeria acutifolia*, *Rauwolfia serpentina* and on abaxial of *Alstonia scholaris*, *Catharanthus pusillus*, *Cascabela thevetia*, *Holarrhena pubescens*, *Plumeria alba*, *Plumiria rubra*, *Rauwolfia serpentina*. The trapezoidal linear cells on both surfaces of *Acocanthera venenata*, *Aganosma calycina*, *Vallaris solanacea*, on adaxial of *Catharanthus roseus*, *Plumeria acutifolia*, *Nerium indicum*; Tropezoidal non-linear cell on adaxial of *Allamanda cathartica*, *Catharanthus pusillus*, *Cascabela thevetia*, *Holarrhena pubescens*, *Plumiria rubra*, on both surfaces of *Ervatamia dirvaricata*, *Wrightia tinctoria*, *W. tomentosa*. Further on abaxial of *Carissa carandas*, *Catharanthus roseus*, whereas squarish cells observed in *Nerium indicum* (Table 5, Plate 8, A-F).

Anticlinal walls of the costal cells are straight or straight to curved in all the taxa except in *Aganosma calycina* where they are sinuate on both the surfaces and on abaxial of *Acocanthera venenata*, *Allamanda cathartica*, *Catharanthus roseus*, *Ervatamia dirvaricata*, *Holarrhena pubescens*. While wavy to sinuate on abaxial of *Rauwolfia serpentina*. *Vallaris solanacea*, *Wrightia tinctoria*, *Wrightia tomentosa* (Table 4; Plates1-5; 8. J&K).

The cell walls were thin in both the surfaces of *Catharanthus pusillus*, *Carissa carandas*, *Catharanthus roseus*, *Cascabela thevetia*, *Ervatamia dirvaricata*, *Holarrhena pubescens*; thin on adaxial and thick on abaxial of *Vallaris solanacea*, *Rauwolfia serpentina*, *Plumeria acutifolia, P. alba*, *P. rubra*, *Wrightia tinctoria*, *Wrightia tomentosa* but in *Nerium indicum,* opposite and in rest no costal cells were observed on both surfaces. The costal cells were smooth on both surfaces of all the taxa except in *Acocanthera venenata*, *Allamanda cathartica* and adaxial of *Plumeria rubra* were they are striated. (plates1-5). The outer wall of the costal cells is flat on both the surfaces and they are parallely arranged and irregulary oriented in all the species except in *Carissa spinarum* and *Rauwolfia canescens* where there were no costal cells (Table 4; Plates 1-5).

The variations in the expression of costal cells on midvein, lateral veins and on alveolar veins were of taxonomic significance in a family or in genus (Kumar, 1983). Additionally, based on traits of costal cells, distribution and orientation, six basic patterns and six combination patterns were prepared.

The basic patterns are:

I. Costal cells absent on both surfaces of *Carissa spinarum* and *Rauwolfia canescens.*

II. Costal cells present on primary vein on both surfaces of *Allamanda cathartica*, *Alstonia scholaris*, *Cascabela thevetia*.

III. Costal cells present on primary, secondary veins on both surface of *Catharanthus roseus*, *Catharanthus pusillus*.

IV. Costal cells present on both the surfaces of *Acocanthera venenata*, *Ervatamia dirvaricata*, *Rauwolfia serpentina*, *Vallaris solanacea*, *Wrightia tinctoria*. tincotoria.

V. Costal cells on 1^0, 2^0, 3^0 and 4^0 veins on both the surfaces of *Carissa carandas*, *Holarrhena pubescens*.

VI. Costal cells present on 1^0, 2^0, 3^0 on upper (adaxial) surface and 1^0, 2^0, 3^0, 4^0 on lower (abaxial) surface of *Nerium indicum*, *Plumeria acutifolia*, *Plumeria alba*, *Plumeria rubra* and *Wrightia tomentosa* (Table 4).

The baseline data of leaf epidermal cell characters are helpful in identification, adulteration check and authentification of medicinal plants of 20 species of family Apocynaceae.

1a. Anticlinal walls of epidermal cells straight on adaxial surface.

2a. Anticlinal walls wavy to sinuate on abaxial surface.

2b. Anticlinal walls are other than wavy to sinuate on abaxial surface

3a. Surface of epidermal cells undulated on abaxial surface - *Ravwolfia serpentina*

3b. Surface of epidermal cells not undulated surface – *Rauwolfia canescens*

4a. Epidermal out growths are present on abaxial surface.

5a. Epidermal cells smooth surfaced on adaxial surface - *Alstonia scholaris.*

5b. Epidermal cells other than smooth *i.e.* undulated on adaxial surface.

6a. Epidermal cells were pitted thickings-*Plumeria alba*

6b. Epidermal cell with out pittings - *Plumeria rubra.*

4b. Epidermal out growths were absent on abaxial surface.

7a. Epidermal cells smooth surfaced on adaxial surface - *Carissa carandas.*

7b. Epidermal cells other than smooth surfaced - *Plumeria acutifolia*

1b. Anticlinal walls of epidermal cells were other than straight on adaxial surface.

8a. Anticlinal walls of epidermal cells were sinuate to wavy on adaxial surface.

9a. Epidermal cells undulated on any surface.

10a. Epidermal cells undulated on both surfaces - *Acocanthera venenata*

10b. Epidermal cells undulated only on abaxial surface – *Holarrhena pubscens.*

9 b. Epidermal cells other than undulation on any surface.

11 a. Epidermal cells sinuated on both surfaces.

12 a. Epidermal cells outer wall flat – *Aganosma calycina.*

12 b. Epidermal cells outer wall pipillate – *Wrightia tenetoria.*

11 b. Epidermal cells sinuated only on abaxial surface -. *Vallaris solanacea*

8b. Anticlinal walls of epidermal cells were other than sinuate to wavy on abaxial surface.

13a. Epidermal cell walls curved to wavy on abaxial surface.

14a. Epidermal cells surface undulated on abaxial surface - *Allamanda cathartica*

14b. Epidermal cells surface other than undulation on abaxial surface.

15a. Anticlinal walls straight to curved - *Catharanthus roseus*

15b. Anticlinal walls other than straight to curved - *Wrightia tomentosa.*

13b. Epidermal cell walls other than curved to wavy on abaxial surface.

16a. Anticlinal walls curved to wavy on adaxial surface - *Cascabella thevetia*

16b. Anticlinal walls other than curved to wavy on adaxial *Cascabella thevetia.*

17a. Anticlinal cell walls curved to wavy on abaxial surface - *Ervatomia divaricata.*

17b. Anticlinal cell walls other than curved to wavy on abaxial surface

18a. Epidermal cells distributed though the adaxial surface - *Carissa spinarum.*

18b. Epidermal cells distributed through the leaf except 1 -5 venation.

19a. Costal cells distributed on adaxial upto 2^0 order of veins only - *Catharanthus pusillus*

19b. Costal cells distributed on adaxial more than 2^0 order of venation - *Nerium indicum.*

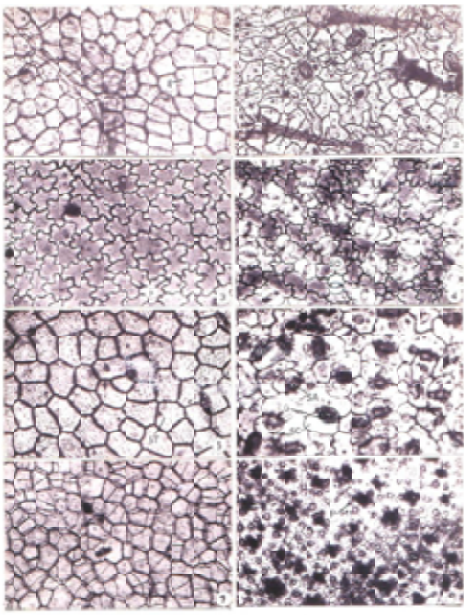

Plate No.1: Showing epidermis
Acocanthera venenata 1. Adaxial (15X10x) and 2. Abaxial (15X10x) surface. *Aganasoma calyxina* 3. Adaxial (10X20x) and 4. Abaxial (10X20x) surface. *Allamanda catharitica* 5. Adaxial (15X10x) and 6. Abaxial (10X20x) surface. E-Epidermal cell, SC-Subsidiary cell, G-Guard cell, SA-Stomatal aperture, ST-Striation, S-Sinuations, T-Trichome, P-Papillate.

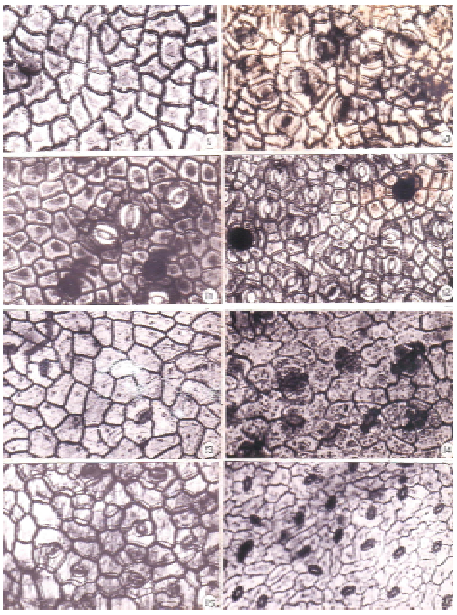

Plate No. 2: Showing epidermis
Carissa carandas 9. Adaxial (10X15x) and 10. Abaxial (10X45x) surface. *C. spinerum* 11. Adaxial (10X45x) & 12. Abaxial (10X45x) surface. *Catharanthus pusilla* 13. Adaxial (10X15x) and 14. Abaxial (10X45x) surface. *C. roseus.* 15. Adaxial (10X20x) & 16. Abaxial (10X20x) surface.

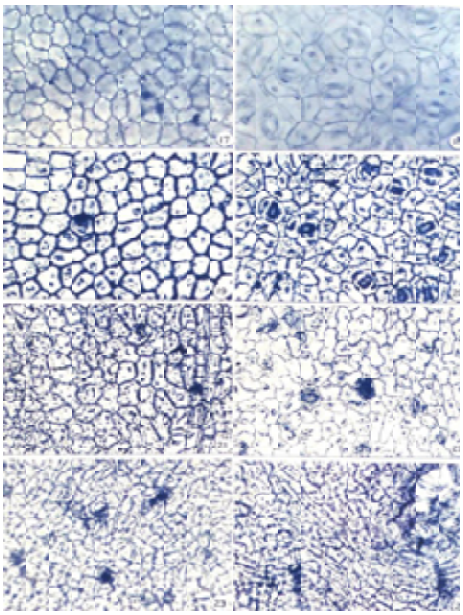

Plate No. 3: Showing epidermis
Cascabella thevetia 17. Adaxial (15X10x) and 18. Abaxial (15X10x) surface. *Ervatomia divericata* 19. Adaxial (15X10x) and 20. Abaxial (15X10x) surface. *Holarrhena pubescence* 21. Adaxial (10X20x) and 22. Abaxial (10X20) surface. *Nerium indicum* 23. Adaxial (10X20x) and 24. Abaxial (10X20x) surface.

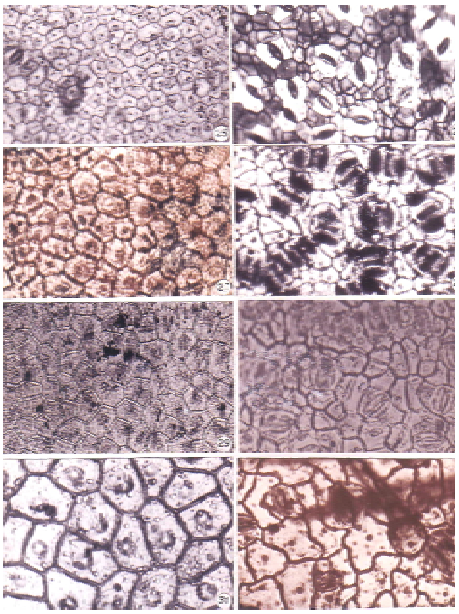

Plate No. 4: Showing epidermis
Plumerai acutifolia 25. Adaxial (10X10x) and 26. Abaxial (10X15x) surface. *P. alba.* 27. Adaxial (10X20x) and 28. Abaxial (10X20x) surface. *P. rubra* 29. Adaxial (10X15x) and 30. Abaxial (10X15x) surface. *Rauwolfia canescens.* 31. Adaxial (10X45x) and 32. Abaxial (10X45x) surface.

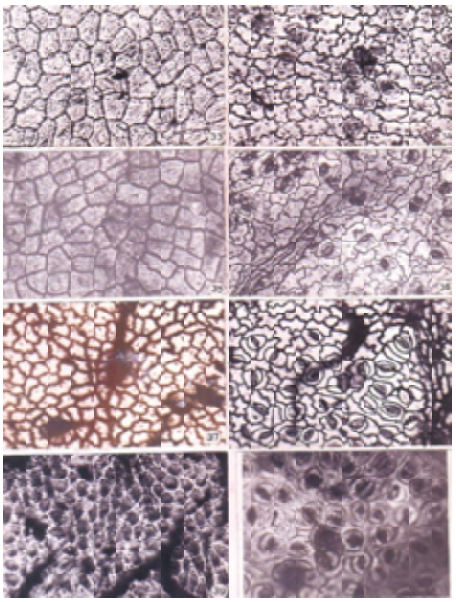

Plate No. 5: Showing epidermis
Rauwolfia serpentina. 33 Adaxial (10X15x) and 34. Abaxial (15X10x) surface. *Vallaris solanacea.* 35. Adaxial (10X15) and 36. Abaxial (15X10x) surface. *Wrightia tinctoria.* 37. Adaxial (10X45x) and 38. Abaxial (10X45x) surface. *W. tomentosa* 39. Adaxial (10X10x) and 40. Abaxial (10X20x) surface

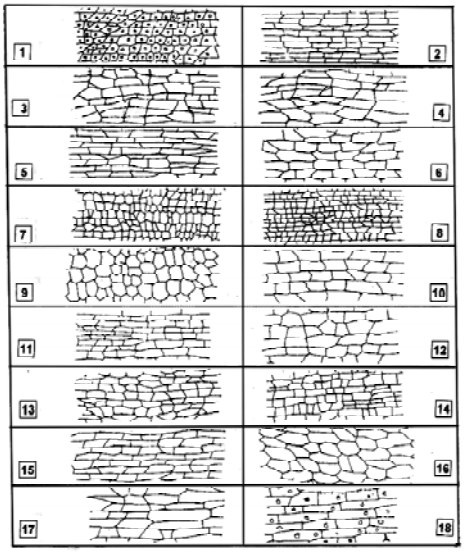

Plate No. 6: Diagrammatical representation of costal cell distribution pattern
1-6 (marked places, other than this are distribution pattern of epidermis); 7- epidermal peels are taken from different locations of the leaf, LB- leaf base, LL- leaf lamina, MD- midrib area, LM- leaf margin, AX- leaf apex. A to L types of epidermal cells. (A- curved anticlinal walls and squarish in shape, B- curved and U shaped anticlinal wall, C- U, V anticlinal wall, D- quadrilateral trapezoidal non-linear, E- polygonal linear cells, F- polygonal non-linear cells, G and H pitted anticlinal wall, I-reticulate surface of the cell, J- spinous, K- papillate, L- nature of epidermal cell)

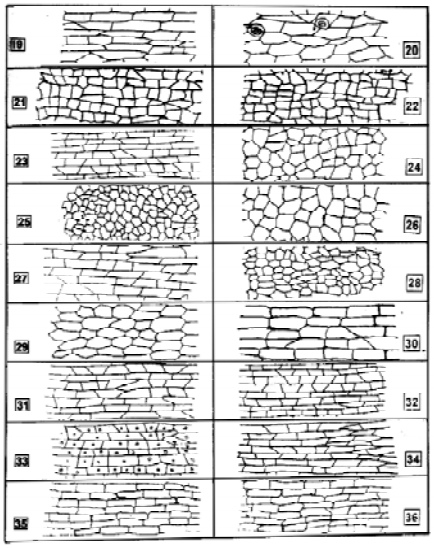

Plate No. 7: Costal cells

Acocanthera venenata 1. Upper, 2. Lower, *Aganosma calycina* 3. Upper (10X45x), 4. Lower; *Allamanda cathartica*, 5. Upper 6 lower (20X45x) *Alstonia scholaris* 7. Upper, 8 lower (10X45x), *Carissa carandas* 9. Upper, 10 lower; *Catharanthus pusillus* 11.upper, 12 lower (15X45x) *Catharanthus roseus* 13. Upper, 14. Lower (10X45x), *Cascabela thevetia* 15 upper 16 lower (10X45x); *Ervatamia dirvaricata* 17. Upper 18. Lower (15X45x).

Plate No. 8: Epidermal cell pattern

Holarrhena pubescens 19. Upper, 20 lower. Nerium indicum 21. Upper. 22. Lower (20X45x) *Plumeria acutifolia* 23. Upper 24. Lower (8X45X25X45x) *P.alba* 25. Upper, 26. Lower (8X40x). *P.rubra* 27 upper. 28 lower (10X20x) *Rauwolfia serpentina* 29. Upper 30. Lower (10X45) *Vallaris solanacea* 31. Upper 32. Lower (10X45x) *Wrightia tinctoria* 33. Upper 34 lower (20X45x) *Wrightia tomentosa* 35. Upper 36. Lower (20X45x)

Conflict of interest

The authors declare no conflict of interest.

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