

CUTICULAR AND EPIDERMAL STUDIES IN SOME SPECIES OF *TERMINALIA* L.

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ABSTRACT

Foliar cuticle and epidermis of six species of *Terminalia* L. (Combretaceae) have been studied under light and scanning electron microscopes. The genus is characterised by the presence of typically compartmented trichomes with striated surface. Stomata are mostly anomocytic; rarely diacytic and paracytic types are also present. Stomatal abnormalities like single guard cell, degenerated guard cells, cytoplasmic connections between two stomata and contiguous stomata have been recorded. Domatia are present in *T. catappa* (lobetiform) and *T. muelleri* (marsupiform). Cuticular striations are present in *T. arjuna* and *T. oliveri*. Cuticular and epidermal features have been found to be useful in distinguishing the genus *Terminalia* from the other genera of Combretaceae as also in the separation of various species of the genus.

INTRODUCTION

Terminalia Linn. (family Combretaceae) is represented by about 250 species, distributed all over the tropics (WILLIS, 1966). Some twelve species have been reported by HOOKER (1879) from India, mainly confined to plains and lower hills. They are mostly large trees with considerable economic importance, the fruits of *T. chebula* are used in dyeing and tanning and also in medicine; the wood of several species is used as a good timber and excellent fuel; the leaves are good fodder for cattles; the leaves of *T. tomentosa* are used for feeding tussah silk worm; the kernel of *T. catappa* is edible.

Though the genus is so much important from economic point of view, it is not well defined taxonomically, and considerable difficulty arises while separating the species of this genus from those of *Combretum*. In view of the above lacuna in our knowledge about taxonomic circumscription of this important genus, the present investigations which deal with the scanning and light microscopic studies of leaf surface and cuticle of the six species have been undertaken.

MATERIAL AND METHODS

The material of the species presently investigated consists of mature and healthy leaves collected from the plants growing in the gardens of the National Botanical Research Institute, Lucknow. Cuticles were separated from leaves by mechanical peeling (scraping with a safety-razor blade), or by maceration with 10-30 per cent nitric acid (AHMAD, 1974). The cuticles were washed with water, stained with aqueous safranin, mounted in pure glycerine and coverslip was ringed with canada balsam. The species investigated are: *Terminalia arjuna* Bedd., *T. bellerica* Roxb., *T. catappa* Linn., *T. chebula* Retz., *T. muelleri* Benth. and *T. oliveri* Brandis.

Sample preparation for Scanning Electron Microscopy—Small strips (1 cm square) were trimmed by razor edge from the leaves of different species. The material was dehydrated in ethanol series and dried in oven at 60°C for about 24 hrs. After drying, small pieces of leaf (ca 0.5 cm square) were cut from the strips. Two pieces of the material, one facing abaxial surface and the other adaxial upwards were mounted side by side on the specimen

Table 1. Epidermal characteristics of different species of *Terminalia*

Traits	Surface	<i>T. ajuna</i>	<i>T. belerica</i>	<i>T. catappa</i>	<i>T. chebula</i>	<i>T. muelleri</i>	<i>T. oliveri</i>
Epidermal cell							
(a) Size (μm)	Upper	37 × 22	68 × 48	42 × 28	34 × 20	34 × 23	39 × 24
	Lower	37 × 23	54 × 36	33 × 21	38 × 23	37 × 24	36 × 25
(b) frequency/mm ²	Upper	..	500—675	1550—1950	1725—2425	1875—2520	1600—2200
	$\bar{x} \pm s$..	1220 ± 118.32	1765 ± 119.9	2103 ± 190.7	2178 ± 172.8	1873 ± 203.7
	C.V.	..	9.69	6.79	9.06	7.93	10.8
Stomata—	Lower	..	450—750	1825—2100	1450—2250	1300—1875	1550—1875
	$\bar{x} \pm s$..	650 ± 97	1957 ± 111.9	1693 ± 212.2	1581 ± 210	1732 ± 90.7
	C.V.	..	14.9	5.72	12.5	13.28	5.23
(a) Size (μm)	Upper	..	36 × 28	24 × 17	28 × 18	28 × 21	27 × 18
	Lower	..	27 × 23
(b) frequency/mm ²	Upper	..	200—350	325—625	200—525	250—550	375—625
	$\bar{x} \pm s$..	244 ± 44.19	506 ± 73.8	378 ± 94	425 ± 91	475 ± 73.6
	C.V.	..	18.11	14.58	24.9	21.4	15.00
(c) Stomatal index	Upper	..	27.29	20.54	18.25	21.18	21.52
	Lower	..	23.53
Trichome							
(a) Size ..	Lower & Upper (min—mean—max)	..	176—364—592	192—459—1104	192—310—544	112—422—896	128—288—448
	Upper	..	3—7	1—5	2—9	2—11	2—5
	$\bar{x} \pm s$..	5 ± 1.57	2 ± 1.20	4 ± 1.6	5 ± 2.7	3 ± 1.19
(b) frequency/mm ² (intercostal)	C.V.	..	31.57	50.17	40.17	54.28	31.45
	Lower	..	1—7	14—27	1—13	3—17	4—10
	$\bar{x} \pm s$..	4 ± 1.8	18 ± 3.6	3.9 ± 3.4	7 ± 3.3	6 ± 2.35
C.V.	..	46.31	19.43	97.59	48.5	34.2	

R = Range ; \bar{x} = mean ; S = Standard deviation ; C.V. = Coefficient of variation.

stubs using double-sided adhesive tape. Specimens were then coated with a thin conductive film of gold about 200Å° in an ion sputter coater (JFC-1100). Coated specimens were examined with a JEOL-35C Scanning Electron Microscope at an accelerating voltage of 10 kV and a tilt of 30° incident to the electron beam at an aperture 100 µm. The image was observed at a magnification range from 200—5,000 and photographs were taken on ORWO 120 roll film.

Quantitative measurements of the epidermal characters of individual species are given in table 1. All observations are based on the average of at least 25 random readings in each case. The length of hair is expressed in table as the minimum observed, the mean and the maximum observed length.

OBSERVATIONS

A comparative account of the epidermal characters of six species of *Terminalia* Linn. is given below :

Lower epidermis—Intercostal cells irregular, sinuous-walled (sinuations 'U'-shaped) in *T. chebula*, *T. arjuna* and *T. belerica* (Text-figs. 1, 3, 7) ; slightly sinuous-walled in *T. oliveri* (Text-fig. 11) ; elongate or isodiametric, arcuate or straight-walled in *T. muelleri* and *T. catappa* (Text-figs. 5, 9). Hair-scars present in some intercostal cells.

Costal cells elongate, straight-walled, arranged in rows (Text-fig. 15) ; in *T. arjuna* and *T. catappa* cells over minor veins slightly sinuous-walled. Hair-scars common.

Marginal cells polygonal or isodiametric, straight-walled or arcuate, elongate, smaller than costal cells, circular hair-scars common in *T. chebula* (Text-fig. 13). Characteristic, ragged holes present in *T. catappa* (Pl. 2, Fig. 36).

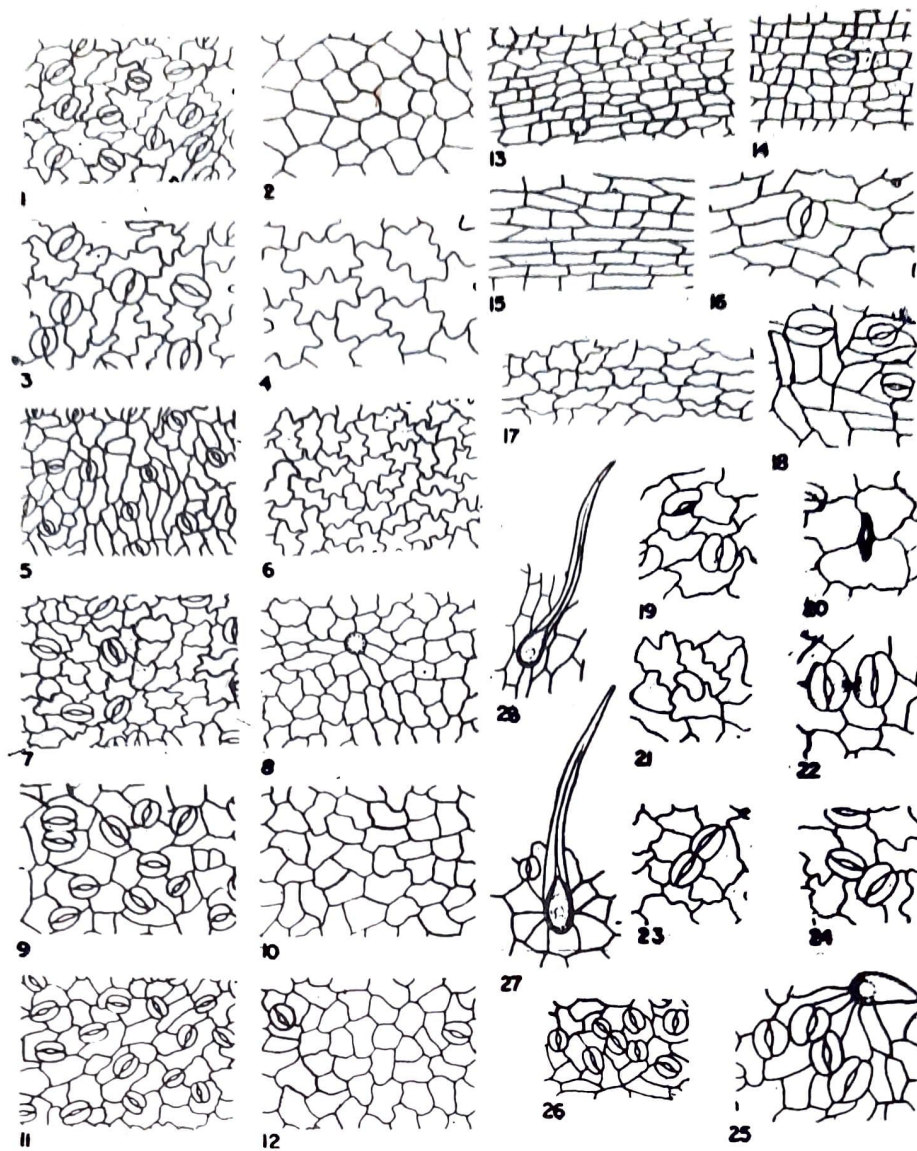
Cuticular striations present in *T. oliveri* and *T. arjuna* (Pl. 1, Fig. 31).

Stomata restricted to intercostal areas, rarely present on minor veins also (Text-figs. 16, 18) ; anomocytic, rarely diacytic and paracytic. Several stomatal abnormalities present : Single guard cell and aborted guard cells (Text-figs. 19-21 ; Pl. 2, Fig. 35) common in *T. belerica* ; contiguous or twin stomata (both superposed and juxtaposed) (Text-figs. 23-25 ; Pl. 1, Fig. 26 ; Pl. 2, Fig. 33) common in *T. chebula* and *T. arjuna* ; cytoplasmic connections between two adjacent stomata (Text-figs. 22 ; Pl. 2, Fig. 34) common in *T. muelleri* and *T. chebula* and rare in *T. arjuna* ; stomatal abnormalities rare in *T. catappa*.

Glandular hairs absent. Non-glandular hairs dense in costal areas, sparse in intercostal areas (dense in costal and marginal areas in *T. chebula*). Simple, unicellular, typically compartmented with a cellulose membrane more or less conically or convexly arched outwards towards the filamentous distal end, sometimes appearing to be bicellular owing to its peculiar construction, somewhat bulbous, short, basal part and thick-walled elongated apical part ; hair base 5-8 celled forming circular poral rim ; walls thick, lumen narrow (Text-figs. 27, 28). Trichome surface striated in all species in different patterns ; striations typically twisted in *T. oliveri* (Pl. 1, Fig. 29) and straight in *T. muelleri* (Pl. 1, Fig. 30).

Domatia present in the axils of major costae and mid-rib in *T. muelleri* and *T. catappa*. In *T. muelleri* the domatia are marsupiform or pocket-shaped with broad distal opening and narrow proximal part ; hair-tuft present inside the domatia (Pl. 1, Fig. 32). In *T. catappa* domatia are labetiform or bowl-shaped. The pit is partially sunken in the leaf tissue.

Upper epidermis—Intercostal cells irregular or polygonal in shape, arcuate or



Text-figs. 1-28. Epidermal features of *Terminalia* L. Figs. 1-12. Lower and upper epidermis (Lower epidermis on the left and upper on the right. 1, 2, *T. arjuna* ; 3, 4, *T. belerica* ; 5, 6, *T. catappa* ; 7, 8, *T. chebula* ; 9, 10, *T. muelleri* & 11, 12, *T. oliveri*). Fig. 13. *T. chebula*, hair scar in marginal cells. Figs. 14-18. Costal cells (14, *T. muelleri* ; 15, 16, *T. belerica* ; 17, *T. catappa* ; 18, *T. muelleri*). Figs. 19-26. Stomatal abnormalities (19, 20, *T. belerica*, aborted guard cells ; 21, *T. belerica*, single guard cell ; 22, *T. arjuna*, cytoplasmic connection ; 23, 24, *T. chebula*, contiguous stomata ; 25, *T. arjuna*, contiguous stomata ; 26, *T. muelleri*, contiguous stomata). Figs. 27, 28, *T. muelleri*, non-glandular trichomes. (Figs. 1-17, 19-21, 26-28 $\times 400$; Figs. 18, 22-25 $\times 600$).

straight-walled in *T. arjuna*, *T. chebula*, *T. muelleri* and *T. oliveri* (Text-figs. 2, 8, 10, 12), sinuous-walled in *T. belerica* and *T. catappa* (Text-figs. 4, 6) ; cells bigger than lower epidermis in *T. belerica* and *T. catappa*, but slightly smaller than lower in other species. Hair-scars sparsely present.

Costal cells straight-walled, elongate, arranged in rows ; cells over minor veins in *T. belerica* and *T. catappa* slightly sinuous-walled (Text-fig. 17). Hair-scars common.

Stomata sparsely present on or along the major veins in *T. arjuna* and *T. muelleri* (Text-fig. 14) ; in *T. oliveri* stomata also sparsely present in intercostal areas (Text-fig. 12) ; anomocytic or diacytic ; stomata absent in *T. belerica*, *T. chebula*. In *T. catappa* stomata are common only around the marginal ragged holes.

Trichomes similar to those of lower epidermis in structure and distribution.

DISCUSSION

STACE (1965a) has pointed out the significance of situations of epidermal cell walls in distinguishing various species of *Terminalia*. Among the species presently investigated, *T. chebula*, *T. arjuna* and *T. bellerica* have lower epidermal cells with sinuous anticlinal walls while *T. catappa*, *T. muelleri* and *T. oliveri* have polygonal cells with straight walls.

Characteristic cuticular striations are present on the lower surface of *T. arjuna* and *T. oliveri*. Striations are also present on the trichome surface of different species, and, while they are twisted in *T. oliveri*, in other species they are parallel. Cuticular striations have also been reported by STACE (1961) in some other genera of Combretaceae.

There is some variation in the frequency and distribution of stomata among the investigated species; the stomatal frequency is highest in *T. arjuna* and lowest in *T. bellerica* (Table 1). Though the stomata are usually confined to the lower epidermis, in some species like *T. arjuna*, *T. muelleri* and *T. oliveri* they occur on the upper surface also. The stomatal size is quite variable; the largest stomata are recorded in *T. bellerica* ($36 \times 28 \mu\text{m}$) and smallest in *T. catappa* ($24 \times 17 \mu\text{m}$).

Compartmented combretaceous trichomes have been observed in all the investigated species and there is not much variation in their shape, size and distribution which could help in the separation of different species. However, they are very important from diagnostic point of view at generic level. Among the investigated species, *T. muelleri* and *T. catappa* can be distinguished from other species in having domatia on their lower surface; marsupiform domatia are present in *T. muelleri* and labetiform in *T. catappa*.

According to METCALFE AND CHALK (1957) the family Combretaceae is characterised by anomocytic stomata and typically compartmented combretaceous trichomes. This finds support from the present study also as all the investigated species have anomocytic stomata and compartmented non-glandular hairs. However, paracytic and diacytic stomata are also present, though sparsely in some species. According to STACE (1965b) the different species of *Terminalia* show lot of variation in their epidermal features. He, however, noted that anomocytic stomata and compartmented trichomes are invariably present in the genus. HOOKER (1879) subdivided twelve Indian species of this genus into two sections on the basis of fruit-wing characters. However, according to EXCELL (1954) the classifications of the genus into sub-genera or sections is not all satisfactory. The present study also lends support to the above view. There is considerable similarity between *Terminalia* and *Combretum* and it is often difficult to distinguish the two genera, especially the sterile material. However, the absence or presence of glandular trichomes in *Terminalia* and *Combretum* respectively is useful in distinguishing the two genera (STACE, 1965b).

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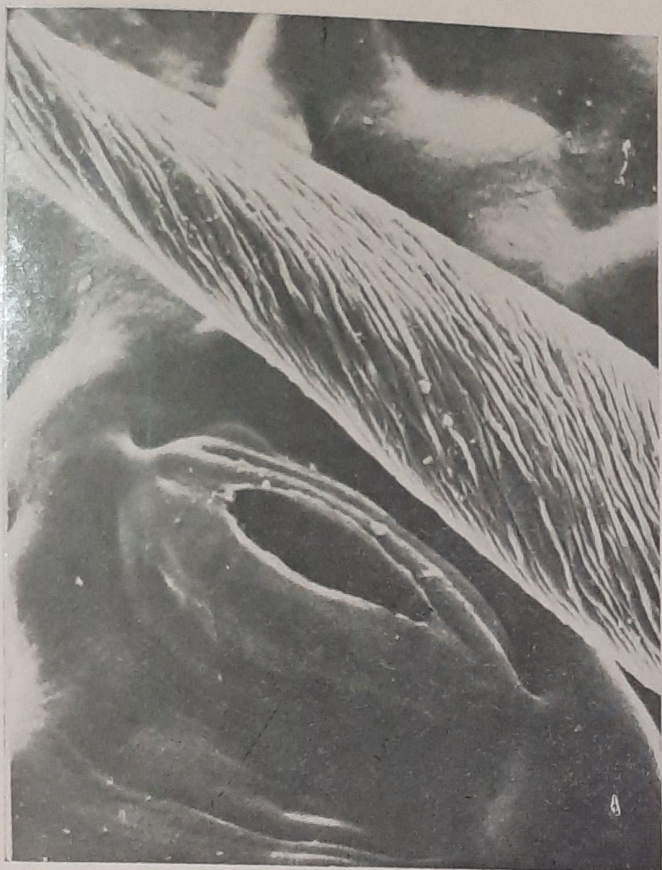
EXPLANATION OF PLATES

PLATE 1

- 29-32. Scanning electron micrographs of foliar surface.
- 29-30. Striations on trichome (29, *T. oliveri* ; 30, *T. muelleri*).
31. *T. arjuna*, cuticular striations.
32. *T. muelleri*, hair tuft inside domatia.
- (Fig. 29. $\times 2832$; Fig. 30. $\times 3480$; Fig. 31. $\times 6960$; Fig. 32. $\times 121$).

PLATE 2

- 33-36. Light micrographs of the foliar cuticle.
33. *T. chebula*, twin stomata.
34. *T. chebula*, cytoplasmic connection.
35. *T. belerica*, stomata with single guard cell.
36. *T. catappa*, Marginal ragged holes.
- (Figs. 33—34. $\times 672$; Fig. 35. $\times 504$; Fig. 36. $\times 336$).



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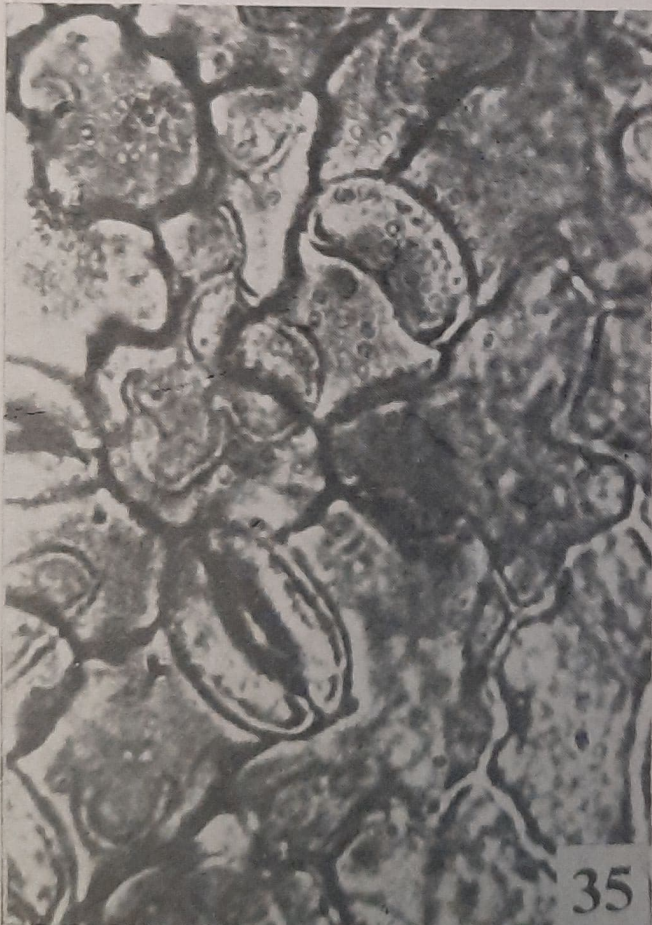
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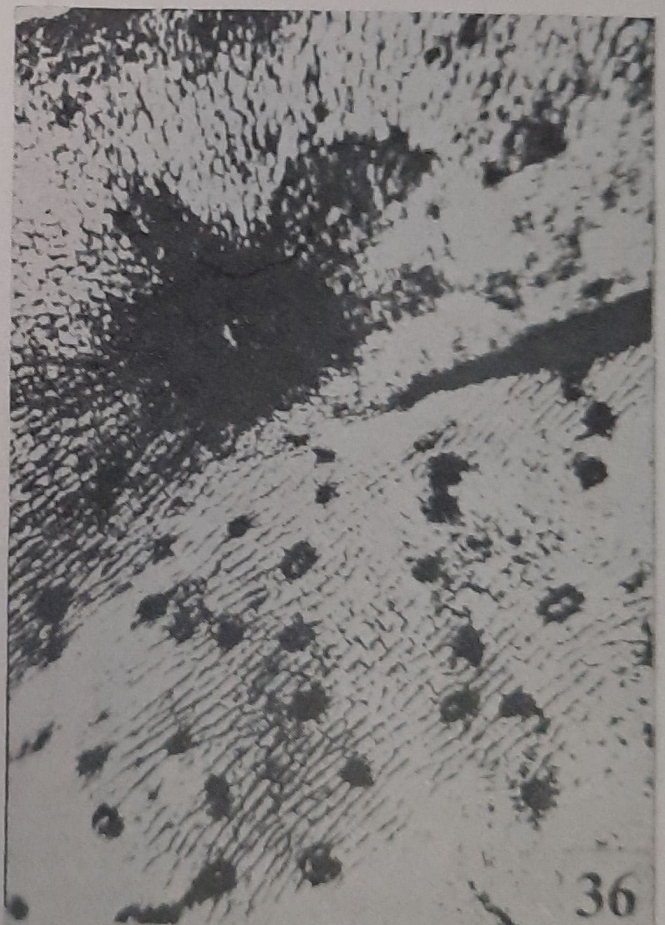
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