LEAF ARCHITECTURE

Leaf architectural studies have been made in 53 genera and 100 species belonging to the families Oleaceae, Salvadoraceae, Apocynaceae, Asclepiadaceae, Buddlejaceae, Loganiaceae, Gentianaceae and Menyanthaceae. The genera and species studied in different families are as under:

1) Oleaceae

6 genera and 28 species

11) Salvadoraceae

1 genus and 2 species

111) Apocynaceae

20 genera and 30 species

iv) Asclepiadaceae

16 genera and 20 species

v) Buddlejaceae

1 genus and 5 species

vi) Loganiaceae

2 genera and 2 species

vii) Gentianaceae and Menyanthaceae

7 genera and 13 species

Family-wise description of leaf architecture follows:
(1) OLEACEAE

A family of 29 genera and 600 species chiefly of temperate and tropical regions (Willis, 1973). The family is of considerable economic importance. Most of the genera are grown as important ornamentals.

The leaf architecture has been studied in 6 genera and 28 species. Leaves are simple, mostly opposite, but alternate in *Jasminum floridum*, *J. humile* and *J. odoratissimum*. In *Jasminum auriculatum*, *J. bignoniaceum*, *J. cordifolium*, *J. flexile*, *J. floridum*, *J. humile*, *J. mesnyi*, *J. officinale*, *J. primulatum* and *Olea primuliana* leaves are pinnately compound (Fig. 1.1: B, C, E-G, I, K; Pl. 1.1: B; 1.2: A-D). The leaves/leaflets are symmetrical with a symmetric or an asymmetric base in *J. bignoniaceum* and *J. mesnyi* (Fig. 1.1: K; Pl. 1.2: D) expanded on the exmedial side of the short or long petiole. Leaves/leaflets are ovate, broad-ovate, ovate-oblong or nearly orbicular (Fig. 1.1: A-L; Pls. 1.1: A-C; 1.2: A-D; 1.3: A-D) with an acute (Fig. 1.1: A, B, D, E, G, I, J, L; Pls. 1.1: A-C; 1.2: C; 1.4: A-C), acuminate (Fig. 1.1: H, K; Pl. 1.3: C, D), obtuse (Fig. 1.1: C, F; Pls. 1.2: A, B, D; 1.4: B) or rounded (Pl. 1.3: A) apex and an acute (Fig. 1.1: A, D, H-L; Pls. 1.1: A-C; 1.2: B-D; 1.3: D), obtuse (Fig. 1.1: B, E-G; Pls. 1.2: A; 1.3: A, C) or cordate (Fig. 1.1: C) base. Margin is mostly entire (Fig. 1.1: A-L; Pls. 1.1: A-C; 1.2: A-D; 1.3: A) or crenate in *Jasminum dichotomum* (Pl. 1.3: B)
or serrate in Nyctanthes arbor-tristis (Fl. 1.3:D). In Osmanthus illicifolius margin is spiny (Fl. 1.3:C). The texture of the leaves is either chartaceous or coriaceous. Glands are not observed in any part of the leaf. The qualitative leaf features of the species are charted in Table 2.

**Venation**

Venation pattern conforms to pinnate camptodromous type with festooned brochidodromous secondaries which originate from the primary vein (mid-vein), do not merge into the margin, but upturn and join together in a series of prominent arches forming the brochidodromous secondaries and have a set of secondary loops outside the brochidodromous secondaries (Hickey and Wolfe, 1975) or multiarcuate wherein secondary veins form a coarcuate infra-marginal vein break up into a series of small arching loops and form a zone between the infra-marginal vein and the margin (Melville, 1976) in all the species (Fig. 1.1;A-L; Fls. 1.1; A-C; 1.2;A-D; 1.3;A-D).

**Major Venation Pattern**

The primary vein or midrib is the thickest vein of the leaf which departs from petiole and traverses straight (Fig. 1.1;I-L; Fls. 1.1;A-C; 1.2;B-D; 1.3;A-D) or markedly curved (Fig. 1.1;A-H; Fls. 1.2;A; 1.3;B,C). The thickness of the primary vein gradually decreases towards the apex. The
size of the primary vein is determined midway between the leaf apex and base as to the ratio of vein width (VW) to leaf width (LW). The size of the primary vein is moderate in all the species. The next smaller size class of veins are the secondary veins (2° veins) whose origin may be on either side of the primary vein in an alternate (Fig. 1.1: A, B, F, H, J-L; Pls. 1.1: A; 1.2: B, C; 1.3: A-C) or sub-opposite fashion (Fig. 1.1: C-E, G, L-K; Pls. 1.1: B, C; 1.2: A, D; 1.3: D). The number of 2° veins on either side of the primary vein varies from species to species irrespective of leaf size (see Table 3). The angle of divergence of secondary veins measured between the branch and continuation of the source vein (primary vein) above the point of branching is also not a constant feature and vary in different species and sometimes even in the same species from base to apex. The angle of divergence of secondary veins is acute at the base and wide acute at the apex (Fig. 1.1: A-L; Pls. 1.1: A-C; 1.2: A-D; 1.3: A-D). Relative thickness of secondary veins is moderate in all the species. The course of secondaries is abruptly curved. The secondary veins do not merge into the margin, but turn upwards and form arches with super adjacent secondaries with acute, right or obtuse angle (Pls. 1.1: A-C; 1.2: A-D; 1.3: A-D, at arrows).

Intersecondary veins are intermediate in thickness between the second and third orders, originating from the primary vein, interspersed among the secondaries having a
course parallel to them. Composite intersecondary veins are observed in all the species (Pls. 1.1:A-C; 1.2:A,C,D; 1.3:A-D, at arrow heads).

The next finer veins are the ternaries which arise from the primary and secondary veins having no definite pattern of angle of origin. Predominant angles of origin are: acute right angle (AR), obtuse right angle (OR), right angle obtuse (RO), right angle right angle (RR), right angle acute (RA), (see Table 2). The pattern of tertiary veins is random reticulate wherein angles of anastomoses vary (Hickey, 1973) or scalariform where intercostal areas are bridged at regular intervals by transverse veins either at right angles or with a regular orientation and have the appearance of rungs on a ladder (Melville, 1976) (Pls. 1.1:A-C; 1.2:A-D; 1.3:A-C). But in Osmanthus illicifolius admedian ramification of tertiary veins branching into higher order veins without rejoining the secondary veins towards the leaf axis (Hickey, 1973) or pendulous type with branching veins lying free in an intercostal area or an areolus attached at their distal ends and appearing to be pendulous from a sub-marginal vein or costal vein (Melville, 1976) (Pl. 1.3:C) is observed. The major veins are multiseriate in all the species.
Minor Venation Pattern:

The next finer veins are the quarternaries ($4^o$) which comprises the veins originating from the tertiaries and those of equal size from lower order veins. The veins originating from these and those of equal size from lower orders are the quaternaries ($5^o$) etc. The highest vein order is resolved up to sixth degree in most of the species, but in *Jasminum flexinale* and *Nyctanthes arbortristis* up to seventh degree and in *Jasminum auriculatum* up to fifth degree (see Table 3).

Marginal Ultimate Venation:

Marginal ultimate venation is mostly incomplete, i.e. freely ending veinlets are directly adjacent to the margin (Hickey, 1973) or marginal vein simple and incomplete, i.e. marginal vein broken, linking some of the excurrent veins, but leaving others free (Melville, 1976) (Pl. 1.4D,E) or looped (Hickey, 1973). Here the major portion of the marginal ultimate venation recurved to form loops or marginal vein simple and irregular, i.e. marginal vein irregular formed by the random union of veins adjacent to the leaf edge (Melville, 1976) (Pl. 1.4E,G).

Veinlets:

The freely ending ultimate veins of the leaf of the same order which occasionally cross areoles are veinlets. The
velnlets are either simple or branched. Simple vein endings are linear or curved (Pl. 1.7:A). The branched ones may divide dichotomously once or twice and branches may be symmetrical or asymmetrical. The veiinlets are usually uniseriate or biseriate, rarely multiseriate as in Osmanthus illicifolius. The vein endings vary in number irrespective of areole size. The veiinlets whether uni-, bi- or multiseriate without terminal tracheids are known as free vein endings. They are observed in all the species (Pl. 1.7:A). In most of the species where areoles are devoid of vein endings, a loop like structure is seen either by union of veins (Pl. 1.5:E, F,H), veins and tracheids (Pls. 1.5:C; 1.6:1-2,D,G,H) or tracheids (Pl. 1.6:E,F). The loops may be oval, triangular, squarish, circular or rectangular. Rarely the veiinlets are situated laterally along the primary vein in Jasminum flexile (Pl. 1.7:C).

Areolation:

The areoles are the smallest areas of the leaf tissue surrounded by the major veins which taken together form a contiguous field over most of the area of leaf. Well developed areoles are observed in most of the species, in which meshes are of relatively consistent size and shape. In some species imperfect, areoles of irregular shape and variable size are observed. The arrangement of the areoles is oriented having a similar alignment or pattern within particular blocks or
domains (Pls. 1.4iH, I; 1.5iA,B,D). The shape of the areoles may be quadrangular (Pl. 1.5iC), rectangular (Pl. 1.4iJ), pentagonal (Pl. 1.5iD), polygonal (Pl. 1.5iA,B) or irregular (Pls. 1.4iH; 1.5iB). The size of the areoles is not constant, but varies in different species and even within the same species. The number and size of areoles, vein endings, absolute vein-islet number, absolute vein termination number vary in different species (see Table 3).

Tracheids:

The veinlets are associated with terminal tracheids, which increase in cell diameter and are extraordinarily variable in shape, size and nature (Pls. 1.7iB-I; 1.8iA-I). The tracheids may be uniseriate, biseriate or multiseriate. Uniseriate tracheids are either long or isodiametric (Pl. 1.7iB-F). They may be uniseriate superimposed (Pl. 1.8iA,C), biseriate juxtaposed uniseriate superimposed (Pls. 1.7iH,X; 1.8iB,C), multiseriate juxtaposed (Pl. 1.8iF-H), multiseriate juxtaposed superimposed (Pl. 1.8iI) or multiseriate juxtaposed uniseriate superimposed (Pl. 1.8iD). The tracheids are normally present at vein tips, but rarely they are present laterally along 5° veins in Jasminum nitidum (Pl. 1.9iB). In most of the species tracheiodal nodules lie lateral and parallel or in groups along the veins, mostly at the apical region (Pl. 1.4iA-C, at arrows). Their occurrence decreases basipetally.
Isolated tracheids:

Tracheids either uni- or biseriate lie free and disjunct in the mesophyll of the areole. Sometimes tracheids are connected with the veinlets by an extension cell (Pl. 1.9:H). Such type of tracheids are also regarded as isolated as they are not connected by tracheary elements. Isolated tracheids are observed in Jasminum flexinale, J. nitidum, J. sambac and Nyctanthes arbortristis (Pl. 1.9:D, E, G).

Isolated Vein Endings:

These are either uniseriate or multiseriate vein endings with terminal tracheids lying free and disjunct in the areole (Pl. 1.9:F). These are common in most of the species.

Extension cells:

These are parenchymatous cells which have failed to differentiate into either sieve or tracheary elements and adjoin isolated tracheids with a vein (Pl. 1.9:H). They may be uniseriate or biseriate. They are observed in Jasminum nitidum and Nyctanthes arbortristis.
Bundle sheath cells:

All category of veins are jacketed by parenchymatous bundle sheath cells (Pls. 1.6\(\text{B-H}\); 1.8\(\text{A-I}\); 1.9\(\text{A-I}\)). The thickness of the bundle sheath varies from primary to higher order veins. In all the species bundle sheath ensheaths veins. The shape of the bundle sheath cells may be either round, oval, isodiametric or rectangular.

Tooth architecture:

The tooth of the margin is non-glandular with spherulate apex in *Jasminum dichotomum* (Pl. 1.3\(\text{B}\)), cassidate apex in *Nyctanthes arbortristis* (Pl. 1.3\(\text{D}\)) and spinose in *Osmanthus illicifolius* (Pl. 1.3\(\text{C}\)) (Hickey, 1979). The origin of the principle vein configuration of the tooth is direct, where secondary veins and their branches run straight into the tooth as a continuation of the laminar venation. The accessory veins are those of higher order. The principal vein of the tooth are looped.
Fig. 1.1 (A-L) : Showing leaf forms of :

A - *Jasminum arborescens*
B - *J. auriculatum*
C - *J. cordifolium*
D - *J. flexile*
E - *J. floridum*
F - *J. officinale*
G - *J. humile*
H - *J. nitidum*
I - *J. revolutum*
J - *J. rigidum*
K - *J. megyi*
L - *Olea dioica*
Fig. 1.1 (A-L)
Plate 1.1 (A-C): Direct photographs of cleared leaves showing architecture of:

A) *Olea europaea*

B) *O. primuliana*

C) *Phillyrea angustifolium*

(A: 4.75 X; B: 2.5 X; C: 2.7 X)
Plate 1.1 (A-C)
Plate 1.1
Plate 1.2 (A-D): Direct photographs of cleared leaves showing architecture of:

A) *Jasminum flexile*

B) *J. flexinale*

C) *J. primum

D) *J. bignoniaceum*

(A: 1.7 X, B: 2.2 X, C: 2 X, D: 5.3 X)
Plate 1.2 (A-D)
Plate 1.3 (A-D): Direct photographs of cleared leaves showing architecture of:

A) *Jasminum sambac*

B) *J. dichotomum*

C) *Nyctanthes arbor-tristis*

D) *Osmanthus illicifolius*

(A, B = 1.8 X; C = 1.33 X; D = 8.2 X)
Plate 1.3 (A-D)
Plate 1.4 (A-I): Showing cleared leaf apices, marginal ultimate venation and areoles in:

A, I) *Jasminum primulimum*

B) *J. sembae*

C) *J. officinale*

D) *J. flexile*

E) *J. floridum*

F) *J. humile*

G) *J. revolutum*

H) *J. flaxinale*

(A, C = 100 X; B, D-I = 40 X)

l - loop;

tn - tracheidal nodules
Plate 1.4  (A-I)
Plate 1.5 (A-H) Showing areoles and loop formation in:

A) Jasminum nitidum

B, C, H) J. primulinum

C) J. auriculatum

D) Nyctanthes arbortristis

E) Jasminum malabaricum

F) Scherbera sweitenoides

(A-D: 40 X; E-G: 100 X; H: 155 X)

1 - loop
Plate 1.5 (A-H)
Plate 1.6 (A-H): Showing loop formation in:

A) *Nyctanthes arbortristis*
B, F) *Schertera sycitendipes*
C-E) *Jasminum flexile*
G) *J. flexinale*
H) *J. rigidum*

(A, H - 250 X; B, C, F - 260 X; D - 335 X; E - 130 X; G - 460 X)

bs - bundle sheath cells;
l - loop
Plate 1.7 (A-I): Showing vein endings in:

A) Hyctanthes arbortrigis

B-D)

E) Jasminum flexile

E) J. nitidum

I) Scherbera obtusangulata

(A-200 X;  B - 815 X;  C, E - 780 X;  
D - 655;  F - 530 X;  G, H - 460 X;  
I - 420 X)
Plate 1.7 (A-I)
Plate 1.8 (A-I): Showing vein endings in:

A) *Jasminum nitidum*

B, C, F, I) *J. flexile*

D, G) *J. flexinale*

E, H) *J. floridum*

(A = 495 X; B = 340 X; C = 610 X;
D = 670 X; E = 500 X; F = 380 X;
G = 600 X; H = 545 X; I = 230 X)

bs = bundle sheath cells
Plate 1.8 (A-I)
Plate 19 (A-I) : Showing special features of venation in:

A, C-F) *Jasminum flexile*

B) J. floridum

G) *J. malabaricum*

H) *J. nitidum*

(A, D, I = 260 X; B = 680 X;
C = 410 X; E = 270 X;
F = 290 X; G = 340 X;
H = 560 X)

bs = bundle sheath cells;
ec = extension cell;
it = isolated tracheid;
ive = isolated vein ending.
Plate 1.9 (A-I)
The family Salvadoraceae comprises 3 genera and 12 species (Willis, 1973).

The leaf architecture has been studied in Salvadora oleoides and S. persica. The leaves are simple and opposite. The leaves are ovate, ovate-elliptic, lanceolate, ovate-lanceolate, elliptic-lanceolate with acute apex (Pl. 1.10: A) and obtuse base. Margin is entire (Pl. 1.10:B). The texture of the leaf is coriaceous in both the species. The qualitative leaf features are charted in Table 4.

**Venation** :

Venation pattern conforms to pinnate camptodromous type with festooned brochidodromous secondaries (Hickey and Wolfe, 1975) or multiarcurate (Melville, 1976).

**Major venation pattern** :

Secondary veins diverge uniformly at a moderate acute or wide acute angle from the straight or markedly curved primary vein in alternate or nearly sub-opposite fashion and form arches with superadjacent secondaries with acute, obtuse or right angle. The number of secondary veins on either side of the primary vein vary from species to species irrespective of leaf size (see Table 5). Composite inter-secondary veins are observed.
Tertiary veins originate at acute, obtuse or right angles from the secondaries and form random reticulate (Hickey, 1973) or scalariform pattern (Melville, 1976).

**Minor venation pattern**:

Higher order venation can be resolved up to sixth degree in both species.

Marginal ultimate venation is looped (Hickey, 1973) or marginal vein simple and irregular (Melville, 1976) (Pl. 1.10:B).

**Veinlets**:

The veinlets of the sixth order end in the areoles either with or without branching (Pl. 1.10:E). The veinlets may be biseriate or multiseriate (Pl. 1.10:E-H). The veinlet number vary irrespective of areole size. In both the species where areoles are devoid of vein endings a loop-like structure is seen which is formed by the union of veins or tracheids and veins (Pl. 1.10:D,E).

**Areolation**:

Areoles are quadrangular, pentagonal, polygonal or irregular in shape. Areoles are well developed and oriented (Pl. 1.10:C). The size of the areole is not constant, but varies in different species and even in the same species. The number and size of areoles, vein endings, absolute vein-islet
numbers and absolute vein termination numbers in thousand vary in both the species.

**Tracheids**

Tracheids are bisericate or multiseriate (Pl. 1.10: R-H). Tracheids are bi- or multiseriate elongated juxtaposed (Pl. 1.10: G), juxtaposed superimposed contiguous (Pl. 1.10: F, H). Isolated tracheids, isolated vein endings, extension cells are absent in both the species. Tracheidal nodules are observed throughout the lamina of both the species.

**Bundle sheath cells**

Parenchymatous bundle sheath cells ensheath all category of veins in varied thickness from primary to higher order veins. The shape of the sheath cells may be oval, round or elongated.
<table>
<thead>
<tr>
<th>Name of the species</th>
<th>Salvador oleoides</th>
<th>S. persica</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Shape</strong></td>
<td>ovate-lanceolate, lanceolate</td>
<td>ovate, ovate-elliptic</td>
</tr>
<tr>
<td><strong>Apex</strong></td>
<td>acute</td>
<td>acute</td>
</tr>
<tr>
<td><strong>Base</strong></td>
<td>obtuse</td>
<td>obtuse</td>
</tr>
<tr>
<td><strong>Margin</strong></td>
<td>entire</td>
<td>entire</td>
</tr>
<tr>
<td><strong>Texture</strong></td>
<td>coriaceous</td>
<td>coriaceous</td>
</tr>
<tr>
<td><strong>Primary vein size</strong></td>
<td>moderate</td>
<td>moderate</td>
</tr>
<tr>
<td><strong>Intersecondary vein</strong></td>
<td>composite</td>
<td>composite</td>
</tr>
<tr>
<td><strong>Predominant tertiary vein origin angle</strong></td>
<td>RO, AR, OR</td>
<td>RA, RO, AR</td>
</tr>
<tr>
<td><strong>Marginal ultimate venation</strong></td>
<td>looped</td>
<td>looped</td>
</tr>
<tr>
<td><strong>Venation pattern</strong></td>
<td>festooned brochidodromous type</td>
<td>festooned brochidodromous type</td>
</tr>
</tbody>
</table>
### Table 5: Showing Species-Wise Numerical Data on the Venation Pattern of Salvadoraceae

<table>
<thead>
<tr>
<th>Name of the species</th>
<th>Salvadora oleoides</th>
<th>S. persica</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Leaf area in mm²</strong></td>
<td>815</td>
<td>1020</td>
</tr>
<tr>
<td><strong>No. of 2° veins along one side of midrib</strong></td>
<td>6-10</td>
<td>6-9</td>
</tr>
<tr>
<td><strong>Range of angle between 1° and 2° veins</strong></td>
<td>40°-65°</td>
<td>45°-65°</td>
</tr>
<tr>
<td><strong>No. of areoles per mm²</strong></td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td><strong>Veinlets entering areoles per mm²</strong></td>
<td>10</td>
<td>8</td>
</tr>
<tr>
<td><strong>Vein ending terminations per mm²</strong></td>
<td>22</td>
<td>21</td>
</tr>
<tr>
<td><strong>Average size of areole in mm²</strong></td>
<td>1.0</td>
<td>1.0</td>
</tr>
<tr>
<td><strong>Absolute vein-islet No. in thousands</strong></td>
<td>0.815</td>
<td>1.020</td>
</tr>
<tr>
<td><strong>Absolute vein terminations in thousands</strong></td>
<td>17,930</td>
<td>21,420</td>
</tr>
<tr>
<td><strong>Highest vein order (in degrees)</strong></td>
<td>6°</td>
<td>6°</td>
</tr>
</tbody>
</table>
Plate 1.10 (A-H): Showing apex, marginal ultimate venation, areoles and vein endings

A-H - *Salvadora persica*

( A - 55 X; B, C - 50 X; D - 145 X; E - 115 X; R-H - 290 X )
Plate 1.10 (A-H)
The family Apocynaceae comprises about 180 genera and 1500 species (Willis, 1973). The members are predominantly tropical and a few temperate. The family embraces diverse habits from twining shrubs, erect tough lianes to trees.

The leaf architecture has been studied in 20 genera and 30 species. Leaves are simple, mostly opposite, but alternate in Cerbera manghas, C. odoratum, Plumeria rubra and Thevetia peruviana or whorled in Alseamanda cathartica, A. neriifolia, A. violacea, Alstonia scholaris, Nerium indica, Bauwolfia serpentina and R. tetraphylla. The leaves are symmetrical with a symmetric or asymmetric base. They are ovate, wide ovate, elliptic, obovate, oblong, elliptic-oblong, oblong-elliptic, elliptic-lanceolate, lanceolate, oblong-lanceolate, ovate-oblong (Fig. 1.2:A-M; Pls. 1.11:A-C; 1.12:A-C) with an acute (Fig. 1.2:A,D,E,H,L,M; Pls. 1.11:A; 1.13:B), acuminate (Fig. 1.2:B,C,F,J,J; Pls. 1.11:B; 1.12:A, D), mucronate (Pl. 1.12:C) or obtuse (Fig. 1.2:C,K; Pls. 1.11:C; 1.13:A); apex and an acute (Fig. 1.2:B,D,F,J,K,M; Pls. 1.11:A,B; 1.12:A,C), obtuse (Fig. 1.2:A,C,H,L; Pl. 1.11:C) or cordate (Fig. 1.2:E,L) base. Margin is entire in all the species (Fig. 1.2:A-M; Pls. 1.11:A-C; 1.12:A,C,D).

Texture of the leaves is chartaceous or coriaceous. Glands are observed at the junction of the petiole and lamina in
Holarrhena antidysenterica and Nerium indicum and at the base of the petiole in Aganosma caryophyllata, Alsemanda cathartica, A. nerifolia, A. violacea, Ervatamia divaricata, Plumeria rubra. The qualitative leaf features of the species are charted in Table 6.

Venation:

The venation pattern conforms to pinnate camptodromous type with festooned brochidodromous secondaries (Hickey and Wolfe, 1975) or multiarcurate (Melville, 1976) (Fig. 1.2iA-E, G-M; Pls. 1.11iA-C; 1.12iA) in all the species, except in Catharanthus rosillus and C. roseus where it is eucamptodromous (Fig. 1.2iF; Pl. 1.12iC). In eucamptodromous venation pattern, secondaries upturn and gradually diminish apically inside the margin and are connected to the superadjacent secondaries by a series of cross veins without forming prominent marginal loops (Hickey, 1973) or simple curvilinear. Here, the secondaries curve gradually towards the margin and often form marginal or sub-marginal veins (Melville, 1976).

Secondaries diverge uniformly at a moderately acute angle from the straight or markedly curved primary vein in an alternate (Fig. 1.2iC,E-I,M; Pls. 1.11iB,C; 1.12iA,C) or nearly opposite or sub-opposite fashion (Fig. 1.2iD,E,J-L Pl. 1.11iA) and curve upwards and form arches with
superadjacent secondaries with acute, right angle or obtuse angle (Pls. 1.11: A-C; 1.12: A, D, at arrows). The number of 2° veins on either side of the primary vein vary from species to species irrespective of the leaf size (see Table 7). Intercostal areas are fairly regular in size and shape, but contain composite intersecondary veins (Pls. 1.11: B, C; 1.12: A-C, at arrow heads). Intramarginal vein is observed in Nerium indicum (Pl. 1.13: F). Intramarginal vein is closely parallel to the leaf margin with the secondary veins fused to it; probably as a result of the fusion and straightening of the exmedial brochidodromous secondary arch segments, it appears to be an independent vein (Hickey, 1973) or marginal vein simple and linear, which is situated close to the leaf edge without any other veins extending beyond it and formed by linking the ends of all the excurrent veins at the margin (Melville, 1976).

The tertiary veins arise from the secondaries having no definite pattern of angles of origin. Predominant tertiary vein angles of origin are right angle right angle (RR), right angle acute (RA), right angle obtuse (RO), acute acute (AA), acute right angle (AR), obtuse right angle (OR), acute obtuse (AO) or obtuse obtuse (OO) in all the species studied except in Alstonia scholaris, where predominant tertiary vein origin angle is not seen (see Table 6). The pattern of tertiary veins is either random reticulate (Pl. 1.11: A), orthogonal reticulate (Hickey, 1973)
or scalariform (Melville, 1976) (Pls. 1.11; B, C; 1.12; A, B, D) or transverse ramified (Pl. 1.12; C) where branching is oriented across intercostal area (Hickey, 1973) or dendroid, i.e. regularly or irregularly dichotomous veins occupying an areole and attached to the areolar veins at one point (Melville, 1976). But admedian ramified in Alstonia scholaris (Hickey, 1973) or pendulous type (Melville, 1976).

**Minor venation pattern**

The higher order venation can be resolved mostly up to sixth degree (6°), but in *Catharanthus roseus* and *C. roseus* up to fourth degree and *Catharanthus major*, *C. variegata*, *Kopsia fruticosa*, *Nerium indicum* and *Thevetia peruviana* up to fifth degree and in *Chromemorphe macrophylla* up to seventh degree (see Table 7).

**Marginal ultimate venation**

Marginal ultimate venation is incomplete (Pl. 1.13; B, C) or looped (Pl. 1.13; D) (Hickey, 1973) or marginal vein simple and incomplete or irregular (Melville, 1976), fimbriate (Pl. 1.13; E) where higher order veins fused into a vein running just inside the margin (Hickey, 1973) or marginal vein simple, arcuate, i.e. marginal vein formed of arcing veins linking the ends of the excurrent veins (Melville, 1976) (see Table 6).
Veinlets:

The fourth, fifth, sixth and seventh order veinlets end in the areoles either with or without branching. The branched ones divide once or twice dichotomously and symmetrically or asymmetrically. The veinlets may be uniseriate (Pl. 1.16:B), biseriate (Pls. 1.14:E,F; 1.15:B,D,F; 1.16:C) or multiseriate (Pls. 1.14:D; 1.15:C). The veinlet number vary irrespective of areole size. In all the species uniseriate, bis- or multiseriate free vein endings are observed (Pls. 1.14:F; 1.15:C). Occasionally, a vessel with a single scalariform perforation plate is present at the vein tip along with the tracheids in Catharanthus major (Pl. 1.18:L, at arrow). Rarely, in a multiseriate veinlet some of the elements fail to differentiate into tracheary elements (Pl. 1.18:K, at arrow). In most of the cases where areoles are devoid of vein endings a loop-like structure is seen which is formed either due to the union of tracheids (Pls. 1.14:B; 1.15:D), veins and tracheids (Pls. 1.14:A; 1.15:B,E) or veins (Pls. 1.14:B,D,F; 1.15:A,D). Rarely, the loop is seen at the vein tip (Pl. 1.16:A).

Areolation:

The areoles in most of the species are either well developed or imperfect. But in Alstonia scholaris, Catharanthus pusillus and C. roseus, the areolation is
lacking. The arrangement of the areoles is either random (Pl. 1.14:A,B,E) or oriented (Pls. 1.12:B; 1.14:C,D,F).

The shape of the areoles may be quadrangular, pentagonal, polygonal or irregular. The size of the areoles is not constant, but varies in different species and even within the same species. The number and size of areoles, vein endings, absolute vein-islet numbers, absolute vein termination numbers vary in different species (see Table 7).

Tracheids:

Tracheids are present mostly at the vein endings, but sometimes along their lateral sides (Pl. 1.17:C,D,F).

Tracheids are either uniseriate, biseriate or grouped (Pls. 1.16:I; 1.17:E). Tracheids may be uniseriate long (Pl. 1.16:D), biseriate isodiametric juxtaposed (Pl. 1.16:E), biseriate long juxtaposed (Pl. 1.16:G), 'V'-shaped with isodiametric superimposed (Pl. 1.17:B), uniseriate superimposed biseriate juxtaposed (Pl. 1.16:J), and 'T'-shaped (Pls. 1.16:F; 1.17:A). In most of the species tracheidal nodules lie lateral and parallel or in bunches along the lateral side of the veins or mostly on the apical region (Pl. 1.16:H,K,L). Their occurrence decreases basipetally.
Isolated tracheids:

The isolated tracheids may be solitary (Pls. 1.14:F, 1.18:D,F) or grouped which lie free and disjunct in the mesophyll (Pl. 1.18:C,E). Isolated tracheids are observed in most of the species.

Isolated vein endings:

These are uniseriate or biseriate vein endings with terminal tracheids lying free and disjunct in the areole (Pl. 1.17:C-J). These are common in most of the species.

Isolated free vein endings:

These are uniseriate or biseriate vein endings without terminal tracheids lying free and disjunct in the areole, noticed in *Allemnda neriifolia, Catharanthus roseus*, *C. roseus* and *Trachelospermum jasminoides* (Pl. 1.18:A,B).

Extension cells:

These are parenchymatous cells which adjoin isolated tracheid with a vein (Pl. 1.18:G,I) or a vein with another vein (Pl. 1.18:J) or a vein with a vein ending (Pl. 1.18:H). Extension cells are observed in most of the species.
Parenchymatous bundle sheath cells ensheath all category of veins from primary to higher order (Figs. 1.15: D-F; 1.16: B, C). The bundle sheath cells may be either round, oval, isodiametric or rectangular.
Fig. 1.2 (A-M): Showing leaf forms of:

A - Aganosma caryophyllata
B - Allamanda perfolia
C - A. violacea
D - Alstonia scholaris
E - Catharanthus major
F - C. pusillus
G - C. variegata
H - Ichnocarpus frutescens
I - Parsonia spiralis
J - Cerbera odollum
K - Plumeria rubra
L - Trachelospermum jasminoides
M - Vallaris solanacea
Plate 1.11 (A-C): Direct photographs of cleared leaves showing architecture of:

A) *Thevetia peruviana*

B) *Rauwolfia tetraphylla*

C) *Caryissa congesta*

(A-3 X; B = 1.9 X; C = 2.5 X)
Plate 1.11 (A-C)
Plate 1.12 (A-D)  Direct photographs of cleared leaves showing architecture, areole and apex in:

A) Ervatamia divaricata
B) Chonemorpha macrophylla
C) Catharanthus roseus
D) Wrightia tinctoria

(A = 1.4 X; B, D = 3 X; C = 3.5 X)
Plate 1.12 (A-D)
Plate 1.13 (A-F): Showing cleared leaf apices, marginal ultimate venation and intramarginal vein in:

A) Catharanthus major
B, C) Trachelospermum Jasminoides
D) Allemanda nerifolia
E) Kopsia fruticosa
F) Nerium indicum

(A-C, E, F = 50 X; D = 120 X)

l - loop
tn - tracheidal nodules
Plate 1.13 (A-F)
Plate 1.14 (A-F) : Showing areoles of cleared leaves in :

A) Wrightia tinctoria
B) Trachelospermum jasminoides
C) Nerium indicum
D) Kopsia fruticosa
E) Plumeria rubra
F) Thevetia peruviana

( A - 55 X; B-E - 50 X; F - 300 X )

it - isolated trachoid
l - loop
Plate 114
Plate 1.15 (A-F): Showing areoles and loop formation in:

A) *Allamanda cathartica*

B, D, E) *Trachelospermum jasminoides*

C) *Allamanda neriifolia*

F) *Nerium indicum*

( A - 50 X; B, C - 125 X; D - 130 X; 
E - 310 X; F - 120 X )

bs - bundle sheath cells

l - loop
Plate 10.15 (A-F)
Plate 1.16 (A-L): Showing vein endings and tracheidal nodules in:

A) **Alamanda nerifolia**

B, C, K) **Trachelospermum jasminoides**

D) **Catharanthus major**

E) **Vallaris solanacea**

F, G) **Plumeria rubra**

H) **Catharanthus roseus**

I) **Rauwolfia tetraphylla**

J) **R. serpentina**

K) **Wrightia tinctoria**

(A - 230 X; B, C, H - 240 X; D - 700 X; E - 560 X; F, G - 450 X; I - 340 X; J - 260 X; K - 380 X; L - 340 X)

bs - bundle sheath cells
l - loop

tn - tracheidal nodules
Plate 1.16 (A-L)
Plate 1.17 (A-J): Showing vein endings and isolated vein endings in:

A-D, F) Reuvolfia tetraphylla
E, I) Trachelospermum jasminoides
G) Allemanda nerifolia
H, J) Thevetia peruviana

(A = 330 X; B = 335 X; C = 430 X;
D = 540 X; E = 340 X; F = 410 X;
G = 230 X; H, J = 290 X; I = 115 X)

ive - isolated vein ending
Plate 1.17 (A-J)
Plate 1.18 (A-L) : Showing special features in:

A, J) Catharanthes roseus
B, H) Trachelospermum jasminoides
C, D, I) Rauvolfia tetraphylla
E) P. serpentina
F) Allemada cathartica
G) Thevetia peruviana
K, L) Catharanthes major

( A = 450 X; B = 230 X; C = 270 X;
D = 250 X; E = 130 X; F = 300 X;
G = 360 X; H = 290 X; I = 380 X;
J, K = 370 X; L = 430 X )

ec - extension cell
ife - isolated free vein ending
it - isolated tracheid
(iv) ASCLEPIADACEAE

The family Asclepiadaceae comprises 130 genera and 2000 species mostly tropical and subtropical. The members are predominantly erect or twining shrubs or perennial herbs. Sometimes fleshy and with reduced non-functional or obsolete leaves.

The leaf architecture has been studied in 16 genera and 20 species. Leaves are simple and opposite. The leaves are symmetrical with a symmetric or asymmetric base, expanded on the exmedial side of the short or long petiole. Leaves are ovate, broadly ovate, oblong, obovate, oblong-lanceolate, obovate-oblong, elliptic-oblong, lanceolate, linear-lanceolate, orbicular, ovate-lanceolate (Fig. 1.3; A-K; Pls. 1.19; A-C; 1.20; A-D; 1.21; A) with an acute (Fig. 1.3; A,F,H,J; Pls. 1.19; A; 1.20; A), obtuse (Fig. 1.3; I), acuminate (Fig. 1.3; C,E,G,K; Pls. 1.19; B,C; 1.20; B-D), mucronate (Pl. 1.21; A,C) or rounded (Fig. 1.3; B,D) apex and an acute (Fig. 1.3; A,F,K; Pl. 1.19; A), obtuse (Fig. 1.3; C,D,G,I; Pls. 1.19; C; 1.20; A, B; 1.21; A), cordate (Fig. 1.3; I; Pls. 1.19; B; 1.20; C) or lobate (Fig. 1.3; B,E,H; Pl. 1.20; D) base. Margin is entire in all the species (Fig. 1.3; A-K; Pls. 1.19; A-C; 1.20; A-D; 1.21; A,C-F). The texture of the leaf is either chartaceous or coriaceous. The glands are observed at the junction of the lamina and petiole in Calotropis gigantea, C. procera, Dasydes
volubilis and Holostemma annularium. The qualitative leaf features of the species are charted in Table 8.

Venation:

Venation pattern conforms to pinnate emptodromous type with festooned brochidodromous secondaries (Hickey and Wolfe, 1975) or multiarcuate (Melville, 1976) in most of the species (Fig. 1.3: A-D, F, G, I-K; Pls. 1.19: A-C; 1.20: A,B; 1.21: A). But in some species actinodromous type, where three or more primary veins diverge radially from a single point (Hickey, 1973) or palmatipinnate, i.e. intermediate between palmate and pinnate, with distal part of the leaf pinnate and a basal or suprabasal pair of pinnated major veins extending 1/3-2/3 of the length of the lamina (Melville, 1976) (Fig. 1.3: E, H; Pl. 1.20: C, D) is observed.

Major venation pattern:

Secondary veins diverge uniformly at a moderately acute angle from the straight or markedly curved primary vein in an alternate (Fig. 1.3: B, E, H, I; Pls. 1.19: A-C; 1.20: A, C, D; 1.21: A), nearly opposite or sub-opposite fashion (Fig. 1.3: A, C, D, F, G, J, K; Pl. 1.20: B) and curve upwards and form arches with superadjacent secondaries with acute, right angle or obtuse angle (Pls. 1.19: A-C; 1.20: A-D; 1.21: A, at arrows). The primary vein size is either stout or moderate. The number of 2° veins on either side of the primary vein
vary from species to species irrespective of leaf size (see Table 9). Relative thickness of the secondary veins is moderate. The course of the secondaries is mostly abruptly curved or sinuous in Pentatropis capensis (Pls. 1.21:C; 1.22:B). Intercostal areas are fairly regular in size and shape, but contain composite intersecondary veins (Pls. 1.19:B; 1.20:B; 1.21:A, at arrow heads). Tertiary veins originate at right angle, acute or obtuse angles from the secondaries (see Table 8) and form random or orthogonal reticulate (Hickey, 1973) or scalariform (Melville, 1976) (Pls. 1.19:A; 1.20:B); admedial ramified (Hickey, 1973) or pendulous (Melville, 1976) in Pentatropis capensis and Tylophora indica or percurrent (Hickey, 1973) where tercaries from the opposite secondaries join or regular scalariform pattern (Melville, 1976), i.e. bridging veins straight or nearly so and regularly spaced in Asclepias curassavica, Dregea volubilis, Gynema sylvestre, Holostemma annularium, Leptadenia hastata and Telosma pallida (Pls. 1.19:A,B; 1.20:A,C,D).

Minor venation pattern :

Mostly the higher order venation can be resolved up to sixth degree, but in Pentatropis capensis and Oxystelma secamone up to fifth degree and in Dregea volubilis and Leptadenia hastata up to seventh degree (see Table 9).
Marginal ultimate venation:

Mostly the marginal ultimate venation is either incomplete (Pl. 1.21:F) or looped (Pl. 1.21:D,E) (Hickey, 1973) or marginal vein simple incomplete or irregular (Melville, 1976) (see Table 8).

Veinlets:

The fifth, sixth and seventh order veinlets end in the areoles either with or without branching. The veinlets may be uniseriate (Pl. 1.22:F) or biseriate (Pls. 1.22:C,E; 1.23:D). The veinlet number varies irrespective of areole size. Uniseriate or biseriate free vein endings are observed in all the species. In most of the cases where areoles are devoid of vein endings, a loop-like structure is seen which is formed either due to the union of tracheids (Pl. 1.23:F,G,I), veins (Pls. 1.22:A-E; 1.23:A,D,E) or veins and tracheids (Pl. 1.23:B,C,F,H).

Areolation:

Quadrangular, pentagonal, polygonal or irregular areoles are well developed or imperfect and oriented to some extent as domains or blocks (Pl. 1.22:A-F). The size of the areoles is not constant, but varies in different species and even in the same species. Venation characters show variations in areole size, number of veinlets entering per areole and
the organization of terminal vein endings in different species (see Table 9).

**Tracheids**: 

Tracheids are present mostly at the vein endings, but sometimes along the lateral side of the vein endings (Pl. 1.24:H-J). Rarely, some of the elongated tracheids are situated laterally at the junction of the veins (Pl. 1.26:H). Tracheids are either uniseriate (Pl. 1.24:A-C,F,G), biseriate (Pl. 1.25:A-I) or grouped into a bunch (Pl. 1.26:A-C). Tracheids may be uniseriate solitary lateral (Pl. 1.24:F,G), uniseriate terminal isodiametric (Pl. 1.24:A). Uniseriate tracheids are at right angles to the vein (Pl. 24:B,C), obliquely oriented juxtaposed (Pl. 1.25:A), biseriate elongated (Pl. 1.25:B), juxtaposed contiguous and juxtaposed superimposed (Pl. 1.25:C,D), isodiametric juxtaposed (Pl. 1.24:D) or isodiametric superimposed (Pl. 1.24:E).

Rarely, in *Dregae volubilis* vessel elements with two perforation plates are present along the third degree and fourth degree veins (Pl. 1.26:E,F). In most of the species, tracheidal nodules lie lateral and parallel or in bunches along the lateral sides of the veins or mostly on apical region (Pls. 1.21:B, at arrow). Their occurrence decreases basipetally. Sometimes, the tracheids lie along the lateral sides of the fourth degree veins (Pl. 1.26:G).
Isolated tracheids:

Tracheids either uniseriate or biseriate which lie free and disjunct in the mesophyll are called isolated tracheids. Sometimes, the isolated tracheids may be connected with the free vein ending by extension cells. Isolated tracheids are observed in most of the species (Pl. 1.27:A,B,H). In some cases the tracheid is isolated and lie near the vein (Pl. 1.26:D).

Isolated vein endings:

These are uniseriate or biseriate vein endings with terminal tracheids lying free and disjunct in the areole. These are common in most of the cases (Pls. 1.23:A; 1.26:J; 1.27:C-E,I).

Isolated free vein endings:

These are uniseriate or biseriate vein endings without terminal tracheids lying free and disjunct in the areole. In *Leptadenia seyalica* and *Pergularia daemia* isolated free vein endings are noticed (Pl. 1.27:F).

Extension cells:

These are parenchymatous cells which either adjoin isolated tracheid with a vein (Pl. 1.27:H), a vein with
another vein (Pl. 1.27;i) or with a vein ending (Pl. 1.27;i).
Extension cells are observed in most of the species.

**Bundle sheath cells:**

Parenchymatous bundle sheath cells ensheath all category of veins varied in thickness from primary to higher order veins (Pl. 1.25;i). The bundle sheath cells may be either round, isodiamic or rectangular in shape.
Fig. 1.3 (A-K): Showing leaf forms of:

A - *Asclepias curassavica*
B - *Calotropis procera*
C - *Cryptolepis buchanani*
D - *Cryptostegia grandiflora*
E - *Pergularia daemia*
F - *Hemideinaeus indicus*
G - *Pentatropis capensis*
H - *Holostemma annularium*
I - *Leptadenia seychelica*
J - *Tylophora indica*
K - *T. termis*
Plate 1.19 (A-C): Direct photographs of cleared leaves showing architecture of:

A) *Oxystelma secamone*

B) *Hologetema annularium*

C) *Leptodenia hastata*

( A - 6 X; B, C - 1 X )
Plate 1.19 (A-C)
Plate 1.20 (A-D) : Direct photographs of cleared leaves showing architecture of:

A) *Cymnema sylvestre*

B) *Leptadenia reticulata*

C) *Telosma pallida*

D) *Dreges volubilis*

( A - 1X; B - 1.7 X;  
C - 1X; D - 3.2 X )
Plate 1.20 (A-D)
The family Loganiaceae comprises 7 genera and 130 species mostly of tropical and a few of warm temperate regions (Willis, 1973).

The leaf architecture has been studied in Cynoehamum metreola and Fagraea obovata. The leaves are simple and opposite and symmetrical with symmetric base. Leaves are obovate, ovate-elliptic or elliptic-lanceolate (Fig. 1.5; A, C) with rounded (Fig. 1.5; A) or acuminate (Fig. 1.5; C) apex and acute (Fig. 1.5; A) or obtuse base. Margin is entire in both the species (Fig. 1.5; A, C). The texture of the leaf is chartaceous in Cynoehamum metreola or coriaceous in Fagraea obovata. The qualitative leaf features are charted in Table 12.

**Venation**

Venation pattern conforms to pinnate ceptodromous type with festooned brochidodromous secondaries (Hickey and Wolfe, 1975) or multiauriculate (Melville, 1976) (Fig. 1.5; A, C).

**Major venation pattern**

Secondary veins diverge uniformly at a moderately acute or wide acute angle from the straight or markedly curved primary vein in nearly opposite or sub-opposite fashion.
(Fig. 1.5: A, C) and curve upwards to form arches with superadjacent secondaries with acute, obtuse or right angle (Fig. 1.5: A, C, at arrows). The primary vein size is moderate in Cynoctonum metreola or stout in Pachraea obovata. The number of 2^o veins on either side of the primary vein varies in both the species irrespective of the leaf size (see Table 13). Composite intersecondary veins are observed in both the species (Fig. 1.5: A, at arrow heads).

Tertiary veins originate at acute or right angles from the secondaries (see Table 12) and form random reticulate (Hickey, 1973) or scalariform pattern (Melville, 1976).

**Minor venation pattern**

Higher order venation can be resolved up to fifth degree in Cynoctonum metreola and up to sixth degree in Pachraea obovata.

Marginal ultimate venation is looped (Hickey, 1973) or marginal vein simple and irregular (Melville, 1976) (Fig. 1.5: F) in both the species.

**Veinlets**

The veinlets of the fifth and sixth order end in the areoles either with or without branching. The veinlets may be biserrate in Cynoctonum metreola (Fig. 1.5: F) or
multiseriate in *Fagraea obovata* (Fig. 1.5:B,E,G). The veinlet number vary irrespective of areole size. In both the species where areoles are devoid of vein endings a loop-like structure is formed by the union of veins or veins and tracheids (Fig. 1.5:F).

**Areolation:**

Areoles are quadrangular, pentagonal, polygonal or irregular. They are imperfect and arranged randomly in *Cynoctonum merreola*; well developed and oriented in *Fagraea obovata* (Fig. 1.5:B). The size of the areole is not constant, but varies in different species and even in the same species. The number and size of areoles, vein endings, absolute vein-islet numbers and absolute vein termination numbers vary in both species (see Table 13).

**Tracheids:**

Tracheids are either biseriate or multiseriate (Fig. 1.5:B,D). They are elongated and juxtaposed. Tracheids are rarely present in *Cynoctonum merreola*. Isolated tracheids are present in *Fagraea obovata* (Fig. 1.5:E).

**Bundle sheath cells:**

Parenchymatous bundle sheath cells ensheath all category of veins. The shape of the sheath cells may be oval, round or elongated.
### TABLE 12: SHOWING SPECIES-WISE QUALITATIVE LEAF FEATURES OF LOGANIACEAE

<table>
<thead>
<tr>
<th>Name of the species</th>
<th>Cynoctomma metrolea</th>
<th>Pagrusa obovata</th>
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</thead>
<tbody>
<tr>
<td><strong>Shape</strong></td>
<td>ovate-elliptic, elliptic-lanceolate</td>
<td>obovate</td>
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<tr>
<td><strong>Apex</strong></td>
<td>acuminate</td>
<td>rounded</td>
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<tr>
<td><strong>Base</strong></td>
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<td>acute</td>
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<td><strong>Margin</strong></td>
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<tr>
<td><strong>Texture</strong></td>
<td>chartaceous</td>
<td>coriaceous</td>
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<tr>
<td><strong>Primary vein size</strong></td>
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<tr>
<td><strong>Inter-secondary vein</strong></td>
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<td>composite</td>
</tr>
<tr>
<td><strong>Predominant tertiary vein origin angle</strong></td>
<td>OR, AR, RR</td>
<td>RR, RA, OR</td>
</tr>
<tr>
<td><strong>Marginal ultimate venation</strong></td>
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<td>looped</td>
</tr>
<tr>
<td><strong>Venation pattern</strong></td>
<td>festooned brochidodromous type</td>
<td>festooned brochidodromous type</td>
</tr>
<tr>
<td>Name of the species</td>
<td>Cynoctorum metrolea</td>
<td>Pagrusa obovata</td>
</tr>
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<td>---------------------</td>
<td>---------------------</td>
<td>------------------</td>
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<tr>
<td>Leaf area in mm²</td>
<td>925</td>
<td>3250</td>
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<td>No. of 2° veins along one side of midrib</td>
<td>6-10</td>
<td>12-18</td>
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<td>Range of angle between 1° and 2° veins</td>
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<td>No. of areoles per mm²</td>
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<td>Vein terminations per mm²</td>
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<td>Absolute vein-islet No. in thousands</td>
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<tr>
<td>Absolute vein termination No. in thousands</td>
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<td>Highest vein order</td>
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<td>6°</td>
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</table>
Fig. 1.5 (A-G): Showing leaf forms, areoles, marginal ultimate venation, vein endings and isolated tracheids in:

A,B,E,G - *Fagrea obovata*

C,D,F - *Cynostomum metreola*

( A,C - 1 X; B,E - 40 X; D,F,G - 140 X )

it - isolated tracheid
The families Gentianaceae and Menyanthaceae comprise 85 genera and 933 species, chiefly of arctic and alpine plains (Willis, 1973).

The leaf architecture has been studied in 7 genera and 13 species. Leaves are simple, mostly opposite, but alternate in 

Nymphoides cristatum and N. indicum. The leaves are symmetrical, expanded on the exmedial side of short or long petiole. Leaves are ovate, broadly ovate, ovate-oblong, linear, linear-oblong, ovate-lanceolate, elliptic-oblong or orbicular (Pls. 1.31:A,B; 1.32:A; 1.33:A) with an acute (Pl. 1.31:B), obtuse (Pl. 1.31:A), acuminate (Pl. 1.33:A) or rounded (Pl. 1.32:A) apex and an acute (Pls. 1.31:A,B; 1.32:A), obtuse, cordate (Pl. 1.33:A) or deeply cordate base. Margin is entire in all the species (Pls. 1.31:A,B; 1.32:A,C; 1.33:A). Texture of the leaves is either membranaceous, chartaceous or coriaceous. The glands are not observed in any part of the leaf. The qualitative leaf features of the species are charted in Table 14.

Venation :

Venation pattern conforms to acrodromous type where two or more primary or strongly developed secondary veins run in convergent arches towards the leaf apex. Arches are
not curved at base. Their position is basal with perfect development (Hickey, 1973) or convergate (Curvi-palmate) (Melville, 1976) in all the species (Pls. 1.31:A,B; 1.32:A; 1.33:A).

**Major venation pattern**:

The course of the primary vein is straight or markedly curved. The thickness of the primary vein is moderate or weak in Canacora diffusa, Centaurium centaurioides, Eracum pumilum, Gentianum quadrifaria, Hoppea dichotoma, Nymphoides cristatum and N. indicum. Secondary veins diverge uniformly from the primary vein at moderately acute angle in nearly opposite, sub-opposite or alternate fashion (Pls. 1.31:A,B; 1.32:A; 1.33:A). The number of 2° veins on either side of the primary vein vary from species to species irrespective of areole size (see Table 15). Relative thickness of the secondary veins is either weak or moderate. Mostly composite intersecondary veins are observed, except in Eracum pumilum, Gentianum quadrifaria and Hoppea dichotoma.

Tertiary veins originate at acute, obtuse or right angles from the secondaries (see Table 14) and form random reticulate (Hickey, 1973) or scalariform (Melville, 1976) pattern (Pls. 1.31:A,B; 1.32:A; 1.33:B).
Minor venation pattern:

Higher order venation can be resolved mostly up to fifth degree, occasionally up to fourth degree in *Canacore diffusa*, *Centaurium centaurioides*, *Exacum puniflorum*, *Gentianum quadrirarum* and *Hoppea dichotoma*.

Marginal ultimate venation:

Marginal ultimate venation is mostly looped (Hickey, 1973) or marginal vein simple and irregular (Pl. 1.32:C) (Melville, 1976), but incomplete in *Canacore diffusa* (Pl. 1.33:B) (Hickey, 1973) or marginal vein simple and incomplete (Melville, 1976) or fimbriate (Hickey, 1973) in *Enicostema byssopifolium* and *Exacum bicolor* or marginal vein simple and arcuate (Melville, 1976) (see Table 14).

Veinlets:

The veinlets of the fourth and fifth order end in the areoles either with or without branching. The veinlets may be uniseriate or biseriate (Pls. 1.32:B-E, 1.33:B-E). The veinlets number vary irrespective of areole size. In most of the species where areoles are devoid of vein endings a loop-like structure is formed due to the union of tracheids (Pl. 1.33:F), veins and tracheids (Pls. 1.33:D, 1.34:A,B) or veins (Pls. 1.32:C,D, 1.33:C).
Areoles are quadrangular, pentagonal, polygonal or irregular. They are imperfect and arranged randomly (Pls. 1.32:D,E; 1.33:B,C). Areoles are absent in Gentianum quadrifera and Hoppea dichotoma. The size of the areole is not constant, but varies in different species and even in the same species. The number and size of areoles, vein endings, absolute vein-islet numbers and absolute vein termination numbers vary in different species (see Table 15).

Tracheids:

Tracheids are either uniseriate, biseriate (Pl. 1.34:C-H) or grouped (Pl. 1.34:F,G). Tracheids may be long, isodiametric, juxtaposed or superimposed (Pl. 1.34:C-H). Mostly tracheidal nodules are restricted to apical region (Pls. 1.32:E; 1.33:G), but in Centaurium centaurioides, they are observed at basal region towards the margin. Isolated tracheids are present in Enicostema hyssopifolium (Pl. 1.34:I).

Bundle sheath cells:

Parenchymatous bundle sheath cells ensheath all category of veins. The bundle sheath cells may be oval, rectangular or round (Pls. 1.33:D,E; 1.34:B-D,G).
DISCUSSION AND CONCLUSIONS

According to Hickey and Wolfe (1975) the leaves of Gentianales are basically simple, margin entire, venation pinnate, secondary veins brochidodromous, tending to form an intramarginal vein, tertiary veins tending to be parallel to the secondaries and latex present. In the members of the order Gentianales studied the venation pattern mostly conforms to pinnate camptodromous type with festooned brochidodromous secondaries, but in some members of the Apocynaceae, Asclepiadaceae and in the members of the Gentianaceae, it is pinnate eucamptodromous, actinodromous and acrodromous respectively. According to Melville (1976) venation pattern mostly conforms to multiauricate or occasionally, curvipinnate (Apocynaceae), palmati-pinnate (Asclepiadaceae) or convergate type (Gentianaceae). Sehgal and Paliwal (1974) classified the leaves of the Euphorbiaceae as uni-, bi- or triveined on the basis of the number of strands entering the base of the leaf. The leaves of the order Gentianales fall under the univeined category. According to Elymale and Wylie (1944) the major veins include the primary, secondary and tertiary and in exceptional cases the quaternary veins may also become part of the major system. But those of subsequent order after tertiary veins constitute minor venation pattern. By having the secondary growth and the presence of vessels and sieve tubes with
sieve plates and companion cells major veins can be differentiated from the minor veins. The minor veins showed no detectable cambial activity and have tracheids alone in the xylem and phloem constituting the parenchyma only (Plymale and Wylie, 1944). Rarely in Drosa volubilis vessel elements with two perforation plates are observed along with the third or fourth degree veins and in Catharanthus major a vessel with single scalariform perforation plate is observed at the vein tip along with the tracheids.

Hickey (1973) classified the marginal ultimate venation into looped, fimbriate and incomplete, which is formed by the higher order veins. In Nerium indicum intramarginal vein is observed which is formed by the union of secondary veins and its branches only. There are no higher order veins beyond the intramarginal vein. Therefore, it becomes extremely difficult to classify the marginal ultimate venation in Nerium indicum according to Hickey (1973). It can very well be classified into marginal vein simple and linear type of Melville (1976). Marginal ultimate venation observed is either incomplete, looped or fimbriate (Hickey, 1973) or marginal vein simple and incomplete, irregular or arcuate (Melville, 1976).
Contradictory opinions are expressed in literature regarding the taxonomic significance of quantitative leaf features. According to Levin (1929) the veinlet number varies within the narrow limits and numbers for different species are sufficiently constant for use as valuable specific characters. Hall and Melville (1951, 1954) suggested that the number of veinlet terminations, either alone or in conjunction with other histological characters is of taxonomic value particularly in genera with only a small number of species. Gupta (1961) pointed out that the vein-islet numbers and veinlet termination numbers are inversely proportional to the area of the lamina. According to Lems (1964) the density of veins is independent of ultimate vein size and the species differ genetically with respect to the range of their vein densities. Verghese (1969) studied the venation pattern in some Scrophulariaceae and pointed out that the number of vein-islets and veinlets are more or less constant for a species. Nicely (1965) pointed out that significant variations within the same leaf as regards the size and shape of the areoles and number of vein endings in each vein-islet. Sehgal and Paliwal (1974) investigated the foliar venation in 150 species of Euphorbia complex and have arrived at the conclusion that the size of the areole cannot be of much significant taxonomic value, especially when the number of species is large, since the range of areole size (vein-islet number per unit area) for several species tend to overlap
considerably and in certain instances are practically identical. Nicely (1965), Sehgal and Paliwal (1974), Singh et al. (1976), Jain (1978) and Inamdar and Murthy (1978) concluded that there is no correlation between the size of the areole and number of veinlets and vein terminations in different species as well as in the same leaf. The present observations in some Gentianales are also in concurrence with the reports of Nicely (1965), Sehgal and Paliwal (1974), Singh et al. (1976), Jain (1978) and Inamdar and Murthy (1978). Since many conflicting opinions exist regarding the size of the areole and number of vein endings, the statistical data will be of no use. So these features cannot be taken into consideration for phylogenetic and taxonomic values. As the nearby areoles are of more or less equal size, vary in their number of vein endings. These number of vein endings are in no way connected to the size of the areole, where the areoles are devoid of less vein endings or none. The loop formation is commonly seen in the areole, which is formed either by the union of tracheids, veins and tracheids or veins and reduces the distance between the veins and probably helps in transport system. Loop formation is observed in all the species.

Fischer (1885) distinguished into principal and secondary or minor vein endings in dicotyledons. Principal vein endings are usually branched structures and secondary ones short often with a single tracheid. Strain (1933) studied
the vein endings in 118 species of miscellaneous collection of families of dicotyledons and categorized the vein endings on the basis of the number of terminal tracheids. Hickey (1973) classified the veinlets into none, simple or branched. Simple ones are either linear or curved. Branched ones divide once, twice or thrice dichotomously. Simple and branched veinlets are observed in the species studied. According to Strain (1933) the vein endings are in no way correlated with the taxonomic affinities of plants.

Mostly tracheids are observed at the terminal position of the veinlets, but sometimes they are along the lateral sides of the veinlets and along with the fourth and fifth degree veins, which increase in cell diameter and are extraordinarily variable in size, shape and nature. Uniseriate, biserial or grouped tracheids are observed in all the species. In dicotyledons the vein endings often contain only tracheids (Fahn, 1977). But in Catharanthus major the tracheids are contiguous with vessel elements at the tips of vein endings.

Kakkar and Paliwal (1972) observed the tracheidal nodules metamorphosed from bundle sheath cells in some members of the Euphorbiaceae at the apices of the lamina. In xerophytic forms dilated tracheidal elements arranged in large groups are located especially at the apices and near the margins (Sehgal and Paliwal, 1974). The tracheidal nodules have polymorphic names such as 'peculiar cells'
(Gilbert, 1881; Riberhorst and Zamura, 1965); 'mechanical cells' (Mangin, 1882); 'water storage cells' (Kny and Zimmermann, 1885); 'Lignified idioblasts' (Warming, 1909); 'storage tracheids' - speichertracheiden (Solereder and Mayer, 1930); 'water cells' (Pirwitz, 1931); 'tracheoidal idioblasts' (Foster, 1956); 'hybrid cells' (Rao, 1957); 'water storage tracheids' (Hendrickson, 1972); and 'sclerotracheoids' (Rao and Das, 1979). According to Olatunji and Mengim (1980) the occurrence of the tracheoidal elements in many unrelated plants may be due to convergent adaptation to xerophytic habit. In most of the genera tracheidal nodules are noticed at the apical region of the lamina and their occurrence decreases basipetally. But in Centaurium centauricoides, they are observed at basal region towards the margins and in Salvadora oleoides and S. persica throughout the lamina. These tracheidal nodules differ from the tracheids in form, arrangement and thickness. Most of the members studied of the Gentianales manifest tracheidal nodules and are xerophytic in habit except Oxystelma secamone which grows near marshy places. Therefore, in Oxystelma secamone correlation between the occurrence of tracheidal nodules and xerophytic habit cannot be made.

Lot of confusion exists in the earlier literature regarding vein endings, isolated vein endings, isolated free vein endings and isolated tracheids. Firstly, Kassapligil (1951) reported the occurrence of isolated veins in
dicotyledonous leaves. Later, the isolated disjunct veins were observed in *Circaeastr* (Foster and Arnott, 1960) and in Hawaiian Euphorbias (Herbst, 1971, 1972). The tracheidal elements, lying free in the areole have been described as free vein endings (Sehgal and Paliwal, 1974). Inamdar and Murthy (1981) studied the vein endings in some members of the Solanaceae and have rightly pointed out the distinction between isolated tracheids, isolated vein endings (with terminal tracheids) and isolated free vein endings (without terminal tracheids), which lie free and disjunct in the mesophyll of the leaf. These are observed in most of the species. Slade (1957, 1959) is of the opinion that the vein endings are caused by the rupture of the minor vascular network during development, expansion and enlargement of mesophyll cells of the leaf. This view is contradicted by Bray (1963) and Lersten (1965) who believe that the vein endings do not result by rupture, but there is a progressive differentiation of procambium from the ground meristem during expansion of the lamina. The view of Bray (1963) and Lersten (1965) seems to be justified. Hara (1962) reported some elongated cells termed as "extension cells" between one vein and vein ending of another vein in *Daphne pseudomazorrensis*. Inamdar and Murthy (1978) reported the presence of extension cells between two veins in *Natura innoxia* and *Solanum melongena* and in between a vein and tracheid in *Solanum surattense*. The parenchymatous extension cells adjoin
isolated tracheid with a vein or a vein with another vein or a vein with a vein ending. Sehgal and Paliwal (1974) designated the term 'ornamented' for the veins, which are surrounded by a parenchymatous bundle sheath. The parenchymatous bundle sheath cells surround all the category of veins varied in thickness from primary to higher order veins.

**TAXONOMIC SIGNIFICANCE**

Taxonomic problems centred around the order Gentianales are: (i) systematic position of the genus *Nyctanthes*, and (ii) family status of the families Menyanthaceae, Buddlejaceae and Pariplocaceae.

The systematic position of the genus *Nyctanthes* has been a matter of debate since Airy Shaw (1952) transferred it from the Oleaceae to the Verbenaceae. Stainton (1952) supported the view of Airy Shaw (1952) on gross anatomical features. This genus is treated within the Oleaceae by Eichler (1878), Bentham and Hooker (1876), Engler and Prantl (1895) and Takhtajan (1966). Embryological (Crete, 1963; Kapil and Vani, 1966), chemotaxonomical (Das and Rao, 1966), anatomical (Inamdar, 1967; 1968; Kundi and De, 1968; Murthy et al., 1978), floral morphological (Kabharpal and Tyagi, 1970), palynological (Saxena, 1975) and leaf architectural (Mohan and Inamdar, 1983) studies support the inclusion of the genus *Nyctanthes* within the Oleaceae.
Mohan and Inamdar (1983) have pointed out the similarities in the leaf architectural features between *Nyctanthes* and the Oleaceae. The similarities are: (i) shape, apex, base, margin and texture of leaf, (ii) pinnate camptodromous with festooned brochidodromous venation pattern, (iii) presence of intersecondary veins, (iv) marginal ultimate venation, (v) veins and veinlets, (vi) areoles, (vii) bundle sheath, and (viii) presence of isolated tracheids, extension cells and loops.

Patel et al. (1981a, 1981b) have supported the separation of the Menyanthaceae from the Gentianaceae and Buddlejaceae from the Loganiaceae on the basis of stomatal structure and ontogeny. However, the present leaf architectural studies reveal the similarities between families Menyanthaceae and Gentianaceae, and Buddlejaceae and Loganiaceae. Therefore, on the basis of leaf architectural studies family status to Menyanthaceae and Buddlejaceae may not be given.

Patel et al. (1983) did not support the separation of the Periplocaeae from the Asclepiadaceae on epidermal features. Leaf architectural studies of these two families also support this view.
The families Menyanthaceae - Gentianaceae, Buddlejaceae - Loganiaceae and Periplocaenae - Asclepiadaceae show similarities to a larger extent as regards qualitative leaf features; major and minor venation pattern (see Table 16).
HISTOCHEMISTRY OF LEAF EPIDERMIS

The histochemical localization of starch, insoluble polysaccharides, proteins and lipids is made in the leaf epidermis of 16 genera and 23 species belonging to five families of the Gentianales. Observations were restricted to fresh leaves. The starch grains present in the guard cells were counted and the average of fifteen readings were taken.

Starch:

Starch is present in both the guard cells of the 23 species investigated (Pl. 2.1: A-L) and the epidermal cells of Buddleja globosa, Rytianthes arboristriata, and Thevetia peruviana. Starch in the form of spherical bodies (Pl. 2.1: A-L) is distributed either around the nucleus and at the polar region (Pl. 2.1:B-D), at polar region (Pl. 2.1:A,H), along the guard cell wall and on the periphery (Pl. 2.1:G, L-L) or randomly distributed (Pl. 2.1:E,F). The number of starch grains in guard cells vary in different species. The number of starch grains in both the guard cells may be equal.
They are observed in the subsidiary cells of *Asclepias curassavica* (Pl. 2.1:C) only. The range in number of starch grains vary from a minimum of 10 in *Catharanthus roseus* to a maximum of 30 in *Salvadora persica* (see Table 17).

**Insoluble polysaccharides**: 

The epidermal and guard cells develop cherry red colour with this test. Insoluble polysaccharides vary considerably in their number, size and distribution in guard cells of different species. They are in the form of spherical granules (Pl. 2.2:A-D) observed in the guard cells of all the species studied. They are present in the epidermal cells of *Ervatamia divaricata* (Pl. 2.2:D, at arrows), *Buddleja globosa*, *Calotropis gigantea*, *C. procera* and *Thręsia peruviana*. PAS positive bodies are observed in the subsidiary cells of *Aganosma caryophyllata* (Pl. 2.2:A, at arrows) and *Pergularia deamia*.

**Cellulose**: 

The epidermal cells of all the species respond quickly to this test and develop blue colour (Pl. 2.2:E,F). The subsidiary and epidermal cells showed similar reaction in *Allamanda neriifolia* and *Tylephora indica* (Pl. 2.2:E,F), but in other species the subsidiary cells stained light yellowish.
in colour. A paracytic stomatal complex may be identified by its subsidiary cells being of light blue colour, in contrast to the dark blue epidermal cells (Pl. 2.2:K,F). This test is very useful to separate subsidiary cells from other epidermal cells as they react differently and develop dissimilar colour.

**Proteins**: 

Proteins stained blue in colour and appear as small droplets in the guard cells of all the species studied (Pls. 2.2:G-I; 2.3:A-C). They are also found in the subsidiary cells of Buddleja globosa, Plumeria rubra, Salvadora oleoides and S. persica (Pl. 2.3:C, at arrows). They are found in the epidermal cells of Agosoma caryophyllata, Alemena cathartica, A. neriifolia, Calotropis gigantea, C. procera, Catharanthus roseus, Jasminum sambac and Nyctanthes arbor-tristis (Pls. 2.2:G,I; 2.3:A,B, at arrows).

**Lipids**: 

Lipids appear in the form of small shining bodies in the guard and epidermal cells of all species (Pl. 2.3:D-I). Lipids are abundant in the epidermal cells of Alemena cathartica, A. neriifolia, Evatemia divaricata, Jasminum auriculatum, Pergularia daemia, Plumeria rubra and Salvadora persica (Pl. 2.3:D-H, at arrows). They are also observed in the subsidiary cells of Pergularia daemia, Thevetia peruviana and Tylophora indica (Pl. 2.3:G,I, at arrows).
TABLE 17: SHOWING THE OCCURRENCE OF STARCH GRAINS IN THE GUARD CELLS OF DIFFERENT SPECIES.

<table>
<thead>
<tr>
<th>No.</th>
<th>Name of the species</th>
<th>No. of starch grains</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Aganosma camphylata</td>
<td>15</td>
</tr>
<tr>
<td>2</td>
<td>Allemaria cathartica</td>
<td>20</td>
</tr>
<tr>
<td>3</td>
<td>A. neriifolia</td>
<td>22</td>
</tr>
<tr>
<td>4</td>
<td>A. violacea</td>
<td>19</td>
</tr>
<tr>
<td>5</td>
<td>Asolepis curassavica</td>
<td>25</td>
</tr>
<tr>
<td>6</td>
<td>Buddleia globosa</td>
<td>16</td>
</tr>
<tr>
<td>7</td>
<td>Calotropis gigantea</td>
<td>17</td>
</tr>
<tr>
<td>8</td>
<td>C. procera</td>
<td>20</td>
</tr>
<tr>
<td>9</td>
<td>Catheranthus pusillus</td>
<td>12</td>
</tr>
<tr>
<td>10</td>
<td>C. roseus</td>
<td>10</td>
</tr>
<tr>
<td>11</td>
<td>Ervatania divaricata</td>
<td>14</td>
</tr>
<tr>
<td>12</td>
<td>Holarrhena antidysenterica</td>
<td>18</td>
</tr>
<tr>
<td>13</td>
<td>Jasminum auriculatum</td>
<td>16</td>
</tr>
<tr>
<td>14</td>
<td>J. officinale</td>
<td>18</td>
</tr>
<tr>
<td>15</td>
<td>J. sambac</td>
<td>14</td>
</tr>
<tr>
<td></td>
<td>Common Name</td>
<td></td>
</tr>
<tr>
<td>---</td>
<td>-----------------------------------</td>
<td>---</td>
</tr>
<tr>
<td>16</td>
<td><em>Leptadenia reticulata</em></td>
<td>16</td>
</tr>
<tr>
<td>17</td>
<td><em>Nyctanthes arbortristis</em></td>
<td>16</td>
</tr>
<tr>
<td>18</td>
<td><em>Pargularia daemia</em></td>
<td>18</td>
</tr>
<tr>
<td>19</td>
<td><em>Flumeria rubra</em></td>
<td>22</td>
</tr>
<tr>
<td>20</td>
<td><em>Salvadora oleoides</em></td>
<td>28</td>
</tr>
<tr>
<td>21</td>
<td><em>S. persica</em></td>
<td>30</td>
</tr>
<tr>
<td>22</td>
<td><em>Thevetia peruviana</em></td>
<td>26</td>
</tr>
<tr>
<td>23</td>
<td><em>Tylophora indica</em></td>
<td>21</td>
</tr>
</tbody>
</table>
Plate 2.1 (A-L) : Showing starch in :

A - *Agamoea caryophyllata*
B - *Alectra cathartic*
C - *Asclepia curassavica*
D - *Buddleia globosa*
E - *Calotropis procera*
F - *Catharanthus roseus*
G - *Jatropha curculorum*
H - *J. officinale*
I - *J. sempervirens*
J - *Niptanthos arbortristis*
K - *Salvadora persica*
L - *Thevetia peruviana*

( A-I = 1440 X; B = 1320 X; C,D = 400 X;
E = 1000 X; F = 1560 X; G, H = 1600 X;
J,L = 1080 X )
Plate 2.2 (A-D) : Showing insoluble polysaccharides in:

A - *Agaronga caryophyllata*
B - *Allemanda nerifolia*
C - *Catharanthus roseus*
D - *Ervatamia divaricata*
E, F - Showing cellulose in:
E - *Allemanda nerifolia*
F - *Tylophora indica*
G-I - Showing proteins in:
G - *Allemanda cathartica*
H - *Calotropis gigantea*
I - *Catharanthus roseus*

(A - 1200 X; B, D, I - 1000 X; C - 1600 X;
E, G, H - 1040 X; F - 350 X)
Plate 2.2 (A-D)
Plate 2.3 (A-C): Showing proteins in:

A - *Jasminum sambac*
B - *Nyctanthes arbortristis*
C - *Plumeria rubra*

D-I: Showing lipids in:

D - *Aganosma caryophyllata*
E - *Erramenia divaricata*
F - *Jasminum auriculatum*
G - *Fargularia desmia*
H - *Plumeria rubra*
I - *Tylophora indica*

(A - 385 X; B, C, F-H - 1080 X; D = 1040 X; E = 1240 X; I = 1440 X)
DISCUSSION AND CONCLUSIONS

Piecemeal information is available regarding the histochemistry of leaf epidermis. Meidner and Mansfield (1968) stated that the stomatal complex histochemistry is incompletely known. The contents of stomatal apparatus along with the epidermal cells by localization of various substrates are studied in 16 genera and 23 species during the present investigation. The number, size and distribution of the starch grains vary in different species. Starch and insoluble polysaccharides are present in guard cells of all the species studied and in the epidermal cells of Buddleja globosa, Nyctanthes arbor-tristis, Calotropis gigantea, G. procera and Thevetia peruviana. The starch grains are observed only in the subsidiary cells of the Nyctanthes arbor-tristis. In the subsidiary cells of Aganosma carvophyllate and Pergularia daemia PAS positive bodies are found. Cutter (1978) reported that except in the aquatic species of Helonepaeus and Ranunculus with sub-merged leaves, the epidermal cells do not contain well developed plastids in angiosperms, but in the epidermal cells of Buddleja globosa, Nyctanthes arbor-tristis and Thevetia peruviana the starch grains are observed. Hinchman (1973) opined that the polysaccharides ratio vary considerably depending on the species or cultivar, the location and maturity of the tissues and other
factors. In the epidermal cells of *Buddleia globosa*, *Calotropis gigantea*, *C. procera* and *Thevetia peruviana* PAS bodies number is lesser than the guard cells. In *Phaseolus mungo* Malik and Sethi (1975a, 1975b), Sethi and Malik (1974a, 1975) and Bhatia and Malik (1977) localized different enzymes in the guard cells, epidermal cells as well as trichomes, and investigated the role of enzymes in stomatal mechanism.

In monocots Tikku et al. (1978) pointed out that the subsidiary cells can be distinguished from the other epidermal cells with *I_2KI-H_2SO_4* test. Murthy and Inamdar (1980) suggested that the same thing is true for dicotyledons also.

The present observations support the view of latter authors. However, it is difficult to say whether the subsidiary cells are mesogenous or perigenous in origin. Meldner and Mansfield (1968) observed large oil droplets in guard cells of the evergreen leaves. Whatley (1972) observed that the subsidiary cells have chloroplasts and were capable of synthesizing starch and the presence of high amount of protein in guard cells. During the present studies, proteins are also observed in subsidiary and epidermal cells besides guard cells. In a fern, *Athroneris vallichiana* Patel et al. (1975) found that the subsidiary cells can be clearly distinguished from epidermal cells and guard cells by the absence of lipid bodies by using Sudan Black 'B'. Raju and Shah (1975) reported that the lipid bodies are restricted in the subsidiary cells of the mature leaves of some Zingiberaceae members. Vanwyk et al.
(1982) observed one spherical lipid body in the subsidiary cells of the genus Eugenia. The lipid bodies are observed in guard cells of all the species, in the epidermal cells of Allamanda cathartica, A. nerifolia, Brysotamia divaricata, Jasminum auriculatum, Pergularia daemia, Plumeria rubra and Salvadora persica and in the subsidiary cells of Pergularia daemia, Thevetia peruviana and Tylophora indica during the course of present investigation.

Murthy and Inamdar (1980) reported lipid bodies in the guard and epidermal cells of 12 species of the Solanaceae. These authors also observed lipid bodies in the subsidiary cells of Nicotiana tabacum, Lycopersicum esculentum, Solanum surattense and Physalis minima. Therefore, it cannot be generalised for all plants that the subsidiary cells can be clearly distinguished from epidermal and guard cells by the absence of lipid bodies.
Scanning electron microscopic studies of leaf epidermis have been made in 34 genera and 47 species belonging to the families Oleaceae, Apocynaceae, Asclepiadaceae, Buddlejaceae and Gentianaceae. The genera and species studied in different families are as under:

1) Oleaceae 5 genera and 7 species
2) Apocynaceae 13 genera and 15 species
3) Asclepiadaceae 10 genera and 10 species
4) Buddlejaceae 1 genus and 5 species
5) Gentianaceae 5 genera and 10 species

Species-wise description is given under each family.
OLEACEAE

1. *Jasminum auriculatum* : Stomata are uniformly distributed, but irregularly oriented over the abaxial epidermis of leaf. The cuticle is thick and shows tall ridges in between stomata. Short striae confined to each cell as well as radiating from stomata are clearly seen. Stomatal pore is narrow with slightly raised stomatal rim (Pl. 3.1:4).

2. *Jasminum sambac* : Stomata are not uniformly distributed over the abaxial leaf epidermis. The orientation of stomata is in any direction. Cuticular striae and folding are not seen. Stomatal and peristomatal rims are distinct. Stomata are raised. The stomatal aperture is long and narrow (Pl. 3.1:B, C).

3. *Hybanthes arbortristis* (Pls. 3.1:D-F; 3.2:A) : The abaxial leaf epidermis showing distribution of trichomes and stomata. Trichomes occur over or near the veins (Pl. 3.1:E). Stomata are uniformly distributed, but irregularly oriented in between the veins (Pl. 3.1:E). The cuticle is folded showing ridges around stomata. Short striae are confined to each epidermal cell. Stomata are sunken. Stomatal and peristomatal rims are distinct. The stomatal pore is small (Pl. 3.1:D). The unicellular conical trichomes have broad undulated (Pl. 3.1:F) or non-undulated (Pl. 3.2:A) base. The surface of the trichome is either smooth or tuberculate.
4: **Olea europaea** (Pl. 3.2:B) : Lower epidermis of leaf showing peltate trichome. The trichome is rotate in form which is divided into segments or rays. The trichome surface is smooth and not tuberculate.

5. **Olea prinulena** (Pl. 3.2:C, D) : Uniformly distributed, but irregularly oriented stomata occur on the lower surface of the leaf. The cuticle is uneven and coated with wax deposition. The paracytic nature of the stomata can be easily noticed. The stomatal and peristomatal rims are distinct. The stomatal pore is long and narrow (Pl. 3.2:D). The stomatal and peristomatal rims are covered with wax particles.

6: **Osmanthus illicifolius** (Pl. 3.2:E). There is a heavy deposition of wax on the cuticle of abaxial leaf epidermis. Stomata are sunken and appear to be covered by a membranous material. Anomocytic nature of the stomata is evident.

7: **Schrebera svietenoides** (Pl. 3.2:F) : The abaxial surface of leaf is uneven with cuticular striae and stomata. Cuticular striae are twisted around stomata. Stomatal rim is distinct, but the peristomatal rim is obscure. Stomatal pore is closed.
1. **Aganosma caryophyllata** (Pl. 3.3:A) : The cuticle of the abaxial surface of leaf is uneven which shows concentric cuticular striae around a stoma. The stomatal and peristomatal rims are distinct. Stoma is sunken with a very small aperture.

2. **Allamanda neriifolia** (Pl. 3.3:B, C) : Cuticular ridges, faint striae and uniformly but irregularly oriented stomata are seen on the lower surface of leaf. Paracytic nature of the stomata is clearly noticed. The stomatal and peristomatal rims are distinct. The stomatal pore is long and narrow.

3. **Allamanda violacea** (Pl. 3.3:D) : The abaxial surface of leaf is uneven. Cuticle is coated with wax particles. The stomatal rim is distinct. The stomatal aperture is lenticular, long and narrow.

4. **Catharanthus roseus** (Pl. 3.3:E) : The lower epidermis of leaf shows uneven cuticular surface and stoma with stomatal pore obscured by waxy material.

5. **Cerbera odollum** (Pl. 3.3:F) : The abaxial epidermis of leaf shows uneven surface and stoma flanked by wings of cuticular striae. The stomatal and peristomatal rims are raised. The stomatal aperture is long and narrow but partly obscured by waxy coating.
6: **Chenomorpha macrophylla** (Pl. 3.4;A,B) : Cuticular striae and stomata are seen on the abaxial surface of leaf. Stoma is flanked by wings of cuticular striae on either side. Cuticular striae over the other part of the surface appear like reticulately raised buttresses. The stoma is sunken with narrow and long lenticular aperture. The stomatal rim is raised. Peristomatal rim is also distinct.

7: **Helonhena antidyserterica** (Pl. 3.4;E) : The abaxial surface of leaf is with short cuticular striae around stomata as well as over epidermal cells and uniformly distributed, but irregularly oriented stomata. Stomata are sunken with narrow and long aperture. The stomatal and poristomatal rims are raised and distinct. The anticlinal epidermal cells are undulated. Paraecytic and anomocytic nature of stomata is distinctly noticed.

8: **Ichnoeparps frutecens** (Pl. 3.4;D,E) : Ridged cuticle, faint cuticular striae and uniformly but irregularly oriented stomata between cuticular ridges are seen over the abaxial leaf epidermis. Stoma is sunken with wide stomatal pore. The stomatal rim is raised. The peristomatal rim is also distinct.

9: **Herium indicum** (Pl. 3.4;F) : Stomatal crypt covered by means of unicellular trichomes are seen on the lower surface of leaf. Trichome surface is tuberculate. The cuticular surface is undulated and studded with wax particles (Pl. 3.5;A).
10: **Rauwolfia serpentina** (Pl. 3.5:B) : Simple uniseriate filiform trichome with rough and tuberculate surface is noticed on the abaxial surface of leaf. Tubercles are long and arranged in linear fashion.

11. **Rauwolfia tetraphylla** (Pl. 3.5:C) : The abaxial surface of leaf is not smooth. Cuticular striations are noticed over epidermal cells. Stomatal and peristomatal rims are distinct. Stomatal aperture is long and narrow. Paracytic nature of stomata is clearly seen.

12: **Erythania divaricata** (Pl. 3.5:D) : Wings of cuticular striae are seen radiating from the stoma over the subsidiary cells. Raised stomatal rim is distinct but not the peristomatal rim. Stomatal pore is wide, long and lenticular. Stoma is paracytic.

13: **Trachelospermum jasminoides** (Pls. 3.5:E,F; 3.6:A,B) : Conical trichomes and stomata are observed on the abaxial surface of leaf. Conical trichomes are raised over the epidermis. The surface of the conical trichome is tuberculate (Pl. 3.6:A). Tubercles are small and arranged more or less in a spiral fashion (Pl. 3.6:B). The cuticular surface is uneven and beset with minute particles of wax. Stomata are distributed uniformly but oriented irregularly in any direction. Raised stomatal rim is distinct. Peristomatal rim is not noticed. Stomatal aperture is wide and short.
14: Vallaris salanacea (Pl. 3.6:C,D) : Uniformly distributed but unevenly oriented stomata are noticed on the abaxial surface of leaf. The leaf surface is uneven with raised arched anticlinal epidermal cell walls and faint cuticular striae. Paracytic and anomocytic nature of the stomata is clearly seen. Stomata are sunken with raised stomatal and peristomatal rims. The stomatal aperture is short, narrow and lenticular.

15: Wrightia tinstoria (Pl. 3.6:E,F) : Stomata are uniformly distributed but irregularly oriented over the abaxial surface of leaf. The leaf epidermis is uneven showing sunken stomata, arched anticlinal walls and cuticular striae around stomata and over epidermal cells. Stomata are surrounded by raised stomatal and peristomatal rims. Stomatal aperture is lenticular and narrow to wide. Paracytic nature of the stomata is clearly noticed.
ASCLEPIADACEAE

1: Calotropis gigantea (Pl. 3.7A,B) : Heavy cuticular striae radiating from and around stomata; simple unicellular and simple uniseriate filiform trichome are seen over the abaxial surface of leaf. Thin walled nature of the trichomes can be clearly noticed. Stomata are widely spaced from one another. Stomata are sunken and surrounded by raised stomatal and peristomatal rims. Stomatal pore is long, narrow and lenticular.

2: Dracaena volubilis (Pl. 3.7C) : The abaxial leaf epidermis shows cuticular striae radiating from stomata. Cuticular striae are also seen around stomata and all over the leaf surface. Raised stomatal and peristomatal rims are distinct. Stomata are predominantly paracytic. Stomatal pore is long, narrow and lenticular.

3: Gymnema sylvestre (Pl. 3.7D) : Conical trichome and uneven surface showing cuticular striae radiating from trichome base are noticed over the lower surface of leaf. The trichome is bulbous at the base. The trichome surface is tuberculate. Tubercles are uniformly distributed over the trichome surface.

4: Homalodesmus indicus (Pl. 3.8A) : The abaxial leaf surface exhibits cuticular striae around stomata and over
epidermal cells. Cuticular striae are long and parallel. The orientation of stomata is in any direction. Stomata are uniformly distributed over the leaf surface. Stomata are sunken with distinct raised stomatal and peristomatal rims. The stomatal aperture is lenticular and narrow to wide.

5: Helotianthus annularis (Pl. 3.8:E, C) : Cuticular striae radiating from stomata are observed over abaxial surface of leaf. The stomata are surrounded by distinct stomatal and peristomatal rims. The stomatal aperture is narrow, long and lenticular. Cuticular striae are long and parallel.

6: Leptadenia reticulata (Pls. 3.8:D-F; 3.9:A) : The abaxial surface of leaf shows uniformly distributed trichomes and stomata. The surface of the cuticle is uneven and shows parallel long striations radiating from trichome bases or stomata (Pl. 3.8:E,F). Cuticular striae are heavy and present all over the surface sometimes reticulate forming crests and buttresses (Pl. 3.8:D,F). The conical shedding trichome surface is tuberculate and warty. Stomata are sunken with distinct stomatal and peristomatal raised rims. The stomatal aperture is lenticular, long and wide.

7: Maradania volubilis (Pls. 3.9:B-D) : Uneven cuticular surface and uniformly distributed, but irregularly oriented stomata are observed over the lower surface of leaf. Cuticular
striae are present all over the epidermis sometimes radiating from stomata and reticulate with fine crests and buttresses. Cuticular ridges also are distinct. Trichome surface is uniformly tuberculate (Pl. 3.9:D). Stomata are sunken with distinct raised stomatal and peristomatal rims. Stomatal pore is lenticular, long and narrow.

8: Oxystelma secamone (Pl. 3.9:E,F) : The abaxial surface of leaf shows undulating or loosely curled filaments of wax, sunken stoma and trichome. The trichome is uniformly tuberculate. Stoma is surrounded by raised stomatal and peristomatal rims. The stomatal aperture is lenticular, long and narrow.

9: Pentatropis oapensis (Pl. 3.10:A-C) : The abaxial leaf epidermis shows uneven surface and uniformly distributed but irregularly oriented sunken stomata (Pl. 3.10:A). The cuticle shows ridges and striae random or radiating from stomata. Cuticular striae are long and parallel. Stomata are surrounded by distinct raised stomatal rim. The peristomatal rim is not distinct. The stomatal aperture is lenticular long and narrow to wide. Sometimes the stomatal aperture becomes obscure due to waxy material (Pl. 3.10:C).
Tylophora indica (Pls. 3.10:D, 3.11:A-F): Uniformly distributed conical trichomes and sunken stomata are observed over the lower surface of leaf (Pl. 3.11:A,E). The cuticle shows ridges and striae either random or concentric around stomata or radiating from trichome bases or stomata. Cuticular striae are long and parallel (Pl. 3.10:D). Sometimes, cuticular striae appear to be twisted like a twine (Pl. 3.10:B). The trichome base is bulbous either spherical or radiating. The trichome surface is tuberculate (Pl. 3.10:B, C, F). Tubercles are uniformly distributed over trichome surface (Pl. 3.10:C). The stomatal and peristomatal rims are distinct and raised. Paraecytic nature of the stomata is clearly seen. The stomatal pore is long, narrow and slit-like or lenticular.
BUDDLEJACEAE

1: *Buddleja asiatica* (Pl. 3.12:A) : Branched trichomes with smooth surface is noticed on the lower surface of leaf.

2: *Buddleja globosa* (Pl. 3.12:B;C) : The abaxial surface of leaf manifests ridged cuticle and uniformly distributed but irregularly oriented sunken stomata. Raised stomatal and peristomatal rims are distinct (Pl. 3.12:C). Paracytic and anisocytic nature of the stomata is clearly noticed. Stomatal aperture is lenticular and narrow to wide.

3: *Buddleja madagascariensis* (Pl. 3.12:D) : Branched candellabra type of trichomes with smooth surface are observed on the lower surface of leaf.

4: *Buddleja lindleyana* (Pl. 3.12:E) : Note the Candellabra type of trichome with smooth surface on the abaxial leaf surface.

5: *Buddleja officinalis* (Pl. 3.12:F) : Branched trichomes with smooth surface occur on lower surface of leaf.
**GENTIPLACEA**

1: *Ceanora diffusa* (Pl. 3.13:A) : The abaxial epidermis of leaf shows uneven cuticular surface studded with wax particles and a stoma. Raised stomatal and peristomatal rims are observed. Stomatal aperture is narrow, long and slit-like.

2: *Enicostema hyssopifolium* (Pl. 3.13:B,C) : Cuticular ridges randomly distributed and uniformly but irregularly oriented stomata are observed on the lower surface of leaf. Cuticular striae are short sometimes radiating from guard cells of a stoma (Pl. 3.13:C). Raised stomatal and peristomatal rims are seen. The stomatal aperture is slit-like or lenticular and narrow to wide.

3: *Exacum astropurpureum* (Pl. 3.13:D) : The adaxial surface of leaf becomes uneven due to cuticular ridges. Cuticular ridges appear crest or buttress-like.

4: *Exacum bicolor* (Pl. 3.13:E) : The abaxial surface of leaf shows cuticular striae radiating from guard cells and a stoma. The raised stomatal and peristomatal rims are noticed. The stomatal aperture is narrow and slit-like.

5: *Exacum pedunculatum* (Pl. 3.13:F) : Cuticular 3-4-armed ridges with radiating striae over each epidermal cells are seen on the lower surface of leaf.
6: *Exacum papilum* (Pl. 3.14A) - Uneven cuticular surface with short parallel striae and widely spaced stomata are observed on the lower surface of leaf. The stomata are oriented in any direction. The stomatal and peristomatal rims are seen. The stomatal aperture is slit-like or lenticular and narrow to wide.

7: *Exacum tetragonum* (Pl. 3.14B, C) - The adaxial surface of leaf shows polygonal epidermal cells with cuticular parallel striae radiating from the base of the trichome. Trichome surface is minutely tuberculate.

8: *Hoppea dichotoma* (Pl. 3.14D) - Stoma with narrow slit-like aperture, stomatal and peristomatal rims and cuticular parallel striae radiating from guard cells are observed on the lower surface of leaf.

9: *Nymphoides cristatum* (Pl. 3.14E) - The adaxial surface of leaf shows raised stoma. Raised stomatal rim is distinct, but peristomatal rim is absent. The stomatal aperture is long and lenticular.

10: *Nymphoides indicum* (Pl. 3.14F) - The abaxial surface of leaf reveals cuticular ridges as well as short parallel striae and uniformly distributed but irregularly oriented raised stomata. The stomatal rim is noticed but not the peristomatal one. The stomatal aperture is long and lenticular.
Plate 3.1 (A-F): Scanning electron micrographs showing leaf epidermis in:

A - *Jasminum auriculatum*

B, C - *J. sambac*

D-F - *Myrtannthia arbortristis*

(A - 500 X; B - 1100 X; C - 5500 X; D-F - 1000 X; E - 200 X)
Plate 3.1 (A-F)
Plate 3.2 (A-F): Scanning electron micrographs showing leaf epidermis in:

A - **Mycenthes arbortristis**

B - **Olea europaea**

C, D - **O. primuliana**

E - **Oggenthus illicifolius**

F - **Schrebera swietenoides**

( A = 2000 X; B, E = 500 X; C, F = 1000 X; D = 5000 X )
Plate 3.2 ( A-F )
Plate 3.3 (A-F) : Scanning electron micrographs showing leaf epidermis in :

A - Aganosma caryophyllata

B, C - Allamanda neriifolia

D - A. violacea

E - Catharanthus roseus

F - Gerbera odollum

( A, E, F = 2000 X; B = 500 X; C = 5000 X; D = 3000 X )
Plate 3.3 (A-F)
Plate 3.4 (A-F): Scanning electron micrographs showing leaf epidermis in:

A, B - *Chonemorpha macrophylla*

C - *Holarrhena antidysenterica*

D, E - *Ichnocarpus frutescens*

F - *Nerium indicum*

(A - 2000 X; B - 5000 X; C, F - 500 X;

D - 550 X; E - 2800 X)
Plate 3.5 (A-F): Scanning electron micrographs showing leaf epidermis in:

A - Nerium indicum
B - Neuwolvia serpentina
C - R. tetraphylla
D - Erythrina diversica
E, F - Trachelospermum jasminoides

(A - 5000 X; B, C - 2000 X; D - 2200 X;
E - 2800 X; F - 100 X)
Plate 3.5 (A-F)
Plate 3.6 (A-F): Scanning electron micrographs showing leaf epidermis in:

A, B - *Trachelospermum jasminoides*

C, D - *Vallaris solanacea*

E, F - *Wrightia tinctoria*

(A, C, E = 500 X; B, D = 5000 X; F = 2000 X)
Plate 3.6 ( A-F )
Plate 3.7 (A-D): Scanning electron micrographs showing leaf epidermis in:

A, B - *Calotropis gigantea*

C - *Dregea volubilis*

D - *Gynema sylvestre*

(A - 550 X; B - 2200 X; C - 2400 X; D - 1500 X)
Plate 3.7 (A-D)
Plate 3.8 (A-F): Scanning electron micrographs showing leaf epidermis in:

A - *Hemideumus indicus*

B, C - *Holostemma annularium*

D-F - *Leptadenia reticulata*

(A - 1000 X; B, F - 900 X; C, E - 2200 X; D - 1100 X)
Plate 3.8 (A-F)
Plate 3.0 (A-F): Scanning electron micrographs showing leaf epidermis in:

A - *Leptadenia reticulata*

B-D - *Marcadenia volubilis*

E-F - *Oxystelma secamone*

(A - 900 X; B - 550 X; C - 2200 X; D - 5500 X; E, F - 5000 X)
Plate 3.10 (A-D) : Scanning electron micrographs showing leaf epidemics in :

A-C - Pentatropis capensis

D - Tylophora indica

(A - 550 X; B - 2200 X;
C - 1100 X; D - 2000 X)
Plate 3.10 (A-D)
Plate 3.11 (A-F): Scanning electron micrographs showing leaf epidermis in:

A-F: *Tylophora indica*

(A, E = 200 X; B, D, F = 1000 X; C = 2000 X)
Plate 3.12 (A-E): Scanning electron micrographs showing epidermis in:

A - Buddleja asiatica
B, C - B. globosa
D - B. madagascariensis
E - B. lindleyana
F - B. officinalis

(A, C, E = 1000 X; B = 200 X; D, F = 500 X)
Plate 3.13 (A-F): Scanning electron micrographs showing leaf epidermis in:

A - C. scora *diffusa*

B, C - *Enicostema hyssopifolium*

D - *E. scopum atropurpureum*

E - *E. bicolor*

F - *E. pedunculatum*

(A - 3400 X; B - 200 X; C - 1000 X; D - 500 X; E - 2400 X; F - 2000 X)
Plate 3.14 (A-F): Scanning electron micrographs showing leaf epidermis in:

A - *Eucalyptus pumila*

B, C - *E. tetragonia*

D - *Hoppea dichotoma*

E - *Nymphoides cristata*

F - *N. indica*

(A - 500 X; B, F - 550 X; C - 220 X;
D - 2000 X; E - 5000 X)
DISCUSSION AND CONCLUSIONS

Valuable contribution in the field of scanning electron microscopic study of leaf epidermis is presented by Rollins and Benerjee (1975, 1979) and Wilkinson (1979). Here, the stomata lie almost at the same level as the epidermis or sunken below the epidermis, sometimes lying in a stomatal crypt as in *NERIUM INDICUM* or rarely slightly raised above the epidermis. Stomata are uniformly distributed but irregularly oriented. Wilkinson (1979) reported 5 principal types of cuticular foldings vis.: (i) striae, (ii) filigree, (iii) reticulate, (iv) ridges, and (v) wrinkles. The cuticle show ridges, foldings, striae, crests and buttresses. Cuticle may and may not be covered by wax. Cuticular striations are long or short, heavy or light, parallel or twisted or radiating. Rao (1982) reported cuticular ridges and furrows in *BUNIAERUCAGO* and *CARDAMINE AFRICANA*.

According to Shiv Raj and Bidinger (1980) the grass leaf surface ornamentations are classified into five types, vis.: (i) band type, (ii) flowery type, (iii) lotus type, (iv) open type, and (v) closed type on the basis of special structures found at the end wall of epidermal cells in cultivars of *SORGHUM*. Taxa studied of Gentianales do not fall under any of these categories.
Kozlowski (1976) reported the occurrence of wax around the stomatal apertures in the leaves of Betula and *Cercis canadensis*. Epicuticular leaf waxes have been classified on a morphological basis (see Wilkinson, 1979). Rao (1982) observed wax deposition on the pericarp epidermis of *Brassica juncea* and *Raphanus sativus*. Waxy deposition is observed in the leaf epidermis of some taxa studied here. In *Catharanthus roseus*, *Cartera odorata* and *Pentatropis microphylla*, the stomatal pore becomes obscure due to waxy secretion.

Wilkinson (1979) used the terms "stomatal rim" and "peristomatal rim" to describe the morphology of the stomatal apparatus. Wilkinson (1979) reported peristomatal rim in *Schinopsis marginata*, *Rhus thrysiflora* and *Buchanania obovata*. Rao (1982) observed 2-3 peristomatal rims in *Brassica juncea*, *Cochlearia cochlearioides*, *Capsella burse-pastoris*, *Iberis umbellata*, *Malconia maritima*, *Raphanus sativus* and *R. sativus* var. *caudatus*. During the course of present studies stomatal rim is observed in all the species studied. The peristomatal rim is either present or absent.

Patel and Inamdar (1972) have emphasized the taxonomic significance of trichomes in Gentianales on the basis of light microscopic studies. Rollins and Banerjee (1975, 1979),
and Inamdar and Rao (1983) made scanning electron microscopic studies of trichomes in the Brassicaceae. According to the latter authors, trichome surface ornamentation can be useful for identification of species in Brassicaceae. Scanning electron microscopic studies made during the present investigation reveal that the trichomes are either thin or thick walled with smooth or tuberculate surface. Scanning electron microscopic study reveals features of leaf epidermis which cannot be seen under the light microscope. It is suggested that detailed features of cuticle, presence or absence of wax, trichome ornamentation, occurrence of stomatal and peristomatal rims and position of the stomata may be of taxonomic value.
DEVELOPMENT, ULTRASTRUCTURE AND SECRETION OF EXTRAFLORAL NECTARIES

1) *Holarrhena antidysenterica* :

A group of ten to twenty extrafloral nectaries is located in a shallow groove on the adaxial surface at the junction of the lamina and the petiole (Pl. 4:1:A). There is no definite pattern of arrangement of nectaries. The nectaries on young leaves are translucent yellowish which later turn brown or black on older leaves. Each nectary is differentiated into a long head and a short stalk without vascular supply (Pl. 4:1:F). The head is differentiated into an outermost layer of palisade-like epithelial cells, surrounding the sub-epithelial ground tissue. The length and diameter of nectaries vary from 1.0-1.5 mm and 0.2-0.4 mm respectively.
The initiation of the nectaries takes place when the leaf primordia are about 650 μm long. A small group of protoderm cells of leaf primordia become papillate and differentiate as nectary meristems on the adaxial surface (Pl. 4.1aB). Simultaneously, the sub-protoderm cells beneath the nectary meristems also undergo divisions to form cushion-like protruberances. Each leaf primordium has a number of 10-15 nectary meristems which differentiate into nectaries during further course of development. The nectary meristems are densely cytoplasmic with prominent nuclei. The divisions in epidermal cells of the meristem are frequently anticlinal and seldom oblique. Sub-epidermal cells divide in various planes. As a result of active divisions in the sub-epidermal tissue, the nectary grows upwards and becomes elongated (Pl. 4.1c). At this stage the epidermal cells show radial elongation, while the cells of the ground tissue of the nectary remain isodiametric. Later, the epidermal cells show marked radial elongation and undergo rapid anticlinal and/or oblique divisions in order to cope up with the increasing bulk of the ground tissue. Now the elongated epidermal nectary cells become palisade-like and form an epithelium of the nectary (Pl. 4.1d). The cells in epithelial layer are densely cytoplasmic and distinctly nucleated (Pl. 4.1e). The cells of the ground tissue show elongation on maturity. Meanwhile, the stalk also undergoes differentiation.
of the protoderm cells rapidly divide anticlinally, but since the cells thus produced soon start elongation and vacuolation, they appear narrower and shorter than the overlying palisade layer and form the epidermis of the stalk. The ground cells of the basal region remain isodiametric (Pl. 4.11F).

In cross-section the head of the nectary appears as an outermost layer of palisade-like epithelial cells, which surrounds the isodiametric cells of the sub-epithelial ground tissue (Pl. 4.21B).

ULTRASTRUCTURE

Ultrastructure of nectaries is described in four stages, viz.: (i) young, (ii) pre-secretion, (iii) secretion, and (iv) mature.

1) Young (Leaves 5-15 mm long):

The epithelial cells of the young nectary contain granular dense cytoplasm and a few irregularly shaped large vacuoles with an electron dense material. The epithelial cell walls are very thin with closely appressed plasmalemma (Pl. 4.31B, at arrow heads). Very thin cuticle is present on the outer periphery of the upper tangential wall of the epithelial cells (Pl. 4.31A,C). Plasmodesmata traverse between radial walls of the epithelial cells as well as the inner tangential wall of the epithelial and adjacent sub-epithelial cells (Pl. 4.31A). Careful
observations revealed that there are no plasmodesmata on the outer tangential wall of the epithelial cells. In oblique sections, many longitudinally cut microtubules are observed near the radial walls of the epithelial cells at the apical region (Pl. 4.5A,B).

Nucleus is generally situated more towards the basal part of the epithelial cell containing almost spherical single nucleolus (Pl. 4.3A). The nucleus has several conspicuous peripheral heterochromatic bodies (Pl. 4.3A).

The dense cytoplasm contains Golgi bodies, endoplasmic reticulum (ER), mitochondria, plastids and numerous ribosomes. ER occur generally in the form of ribosome studded cisternae; rough ER (Pl. 4.3C). The cisternae are much flattened and in section appear as double lines. The ER cisternae in the form of short segments occur in cytoplasm near the cell walls. Rough ER shows linear pattern of polyribosome aggregates (Pls. 4.3B; 4.4B,C). The ground cytoplasm is rich with free ribosomes and aggregates often in circular and spiral pattern of polyribosomes (Pls. 4.4C; 4.5A,C, at arrows).

Only a few Golgi bodies appear in the form of a stack of more or less compressed closed 5-8 cisternae with associated vesicles. They frequently occur in the cytoplasm near to the cell walls (Pls. 4.3A,B; 4.4B; 4.5D). Golgi bodies are occasionally observed revealing a degree of activity which may be related to residual wall synthesis.
In sections plastids vary in shape and size considerably. Most of them appear as either almost round, oval or elongated (Pls. 4.3;A,B; 4.4;A,C). Plastids are often found close to the nucleus and cell walls (Pl. 4.3;A). Plastid interior is occupied by dense granular stroma, starch grains and a few poorly developed lamellae (Pls. 4.3; A,B; 4.4;A,C). Plastids contain electron dense material (osmiophilic material) either scattered in the stroma or associated with lamellae (Pl. 4.4;A,C). Starch grains are often surrounded by osmiophilic material (Pl. 4.3;A). Starch grains roughly oval in shape appear as electron translucent regions in micrographs.

Mitochondria are common organelles distributed throughout the cytoplasm. They are variable in shape and appear globular, elongated or oval (Pls. 4.3;A-C; 4.4;C). Mitochondria contain less developed cristae and frequently show electron translucent spaces in matrix (Pls. 4.3;A-C; 4.4;C; 4.5;D).

Globoid lipid bodies are seen in the cytoplasm at apical region of the epithelial cells, which show electron translucent spaces in them (Pls. 4.3;C; 4.5;C).

Only few irregularly shaped large vacuoles appear in the cytoplasm. The vacuoles are located at both the radial ends of the epithelial cells and often store either granular or electron dense material of various amounts (Pls. 4.3;A,C; 4.4;C; 4.5;D).
Pre-secretion (Leaves 1.6-2.5 cm long):

The epithelial cells after reaching their maximal size, undergo a series of ultrastructural changes until the stage of secretion. The amount of the cellular contents is increased and the volume of the vacuole is relatively decreased compared to the preceding stage (Pl. 4.6:1). The epithelial cells contain granular dense cytoplasm with abundant cell organelles viz., ER, Golgi bodies, mitochondria, plastids and ribosomes.

The epithelial cell walls are thick with undulated or wavy plasmalemma (Pl. 4.6:B-D). The plasmalemma withdraws from the cell walls forming the plasmalemma invaginations. Plasmalemma invaginations are more frequent along the radial walls. These invaginations thus form the extraplasmic space between the cell wall and plasmalemma (Pl. 4.6:B-D). At many places along the cell walls dense staining regions are found in extraplasmic space (Pl. 4.6:B,C, at arrow heads). The dense material appears to be the result of accumulation of dictyosome vesicles. The cuticle is considerably thick lying on the outer tangential wall of the epithelial cell. The thick cuticle consists of a moderately electron-dense matrix, apparently composed of cutin and a fine fibrillar network of electron-dense material (Pl. 4.6:A). The indentations between the pectin-cellulosic wall and cutinized layer are also observed.
The nucleus is generally situated more towards the basal part of the epithelial cell containing spherical single nucleolus (Pl. 4,6; A). The nucleus has conspicuous peripheral heterochromatic bodies.

ER and the Golgi bodies are the dominant cell organelles and occupy much of the area in the cytoplasm (Pl. 4,6; B-F). The ER cisternae appear in the form of long profiles, frequently aligned parallel to the cell walls (Pls. 4,6; D,E; 4,7; A,C) and also distributed throughout the cytoplasm. ER occur in the form of typical ribosome studded cisternae, rough ER and swelling of its elements (Pl. 4,4; D). Distended ER elements are observed in the vicinity of the plasmalemma; some of them being attached to it. Occasionally, ER cisternae appear as whorls in the cytoplasm (Pl. 4,6; E). The ground cytoplasm is rich with free ribosomes and aggregates often in circular and spiral patterns of polyribosomes (Pls. 4,6; C,D; 4,7; B, at double arrows).

The density of Golgi bodies with hypertrophied cisternae is high in the cytoplasm at this stage. They appear in the form of a stack of more or less compressed closed 5-6 cisternae with associated peripheral tubules and vesicles arranged parallel to each other (Pl. 4,6; B-D,F). They are distributed throughout the cytoplasm and often appear close to the cell walls (Pl. 4,6; B,F). Golgi vesicles appear to be produced in large numbers into surrounding cytoplasm from the swollen rim of cisternae. The Golgi vesicles appear to fuse
with or encircled by plasmalemma (Pl. 4.6:C, at arrows), presumably depositing the secretory material into the extraplastmic space.

In sections, plastids vary greatly in shape and size. Most of them appear as either round, oval, elongate or dumbbell-shaped (Pl. 4.6:1A-D). Plastids are situated mostly near the cell walls distributed throughout the cytoplasm. Significantly, plastid interior is occupied by large number of starch grains with various sizes and shapes (Pl. 4.7:1A-D). The electron dense material (osmiophilic material) is reduced considerably and surrounded by thylakoids and starch grains in the young stage (Pl. 4.7:1A-D). Occasionally, the electron dense material is still seen surrounding the starch grains at this stage (Pl. 4.7:1D).

The number of mitochondria is increased compared to young stage. They are variable in shape and appearing globular, elongate or oval with well developed cristae. Still some of the mitochondria show electron translucent spaces (Pl. 4.7:1A-D). Mitochondria are seen near the plasmalemma invaginations (Pl. 4.6:1C).

The volume of the vacuole is relatively reduced compared to young stage (Pl. 4.6:1A,B). The vacuoles contain electron dense material (osmiophilic material) in high amounts (Pl. 4.6:1A).
iii) **Secretion** (Leaves 2.6-8.0 cm long)

At this stage, the epithelial cells contain dense cytoplasm and numerous small vacuoles.

The cell wall microfibrils of the epithelial cells become loose and the plasmalemma invaginations occur along the radial walls into the cytoplasm forming the extraplasmic space. The area of the extraplasmic spaces is increased at this stage and the accumulation of secretory material is seen in between plasmalemma and radial walls of the epithelial cells (Pls. 4.21C; 4.81A-D; 4.91A; 4.101A-C). In sections, the secretory material is accumulated on either side of the radial walls in extraplasmic spaces throughout the length of the epithelial cells. This secretory material consists of multivesicular and multilamellar structures (Pls. 4.81D,E; 4.101A,C). In sections, the multilamellar structures appear as concentric whorls (Pl. 4.81E). The cuticle, overlying the outer tangential wall of epithelial cells becomes detached from the underlying cellulosic wall forming sub-cuticular space. The sub-cuticular space is filled with secretory material (Pls. 4.91A; 4.111C,D). The extraplasmic spaces appear to be intercellular channels in transsections of the epithelial cells of the nectary. The epithelial cells are not closely packed, but have small vertical intercellular channels at the junction of three or four adjacent cells (Pl. 4.21E, at arrows).
The nucleus generally occupies a more or less median position of the cell (Pls. 4.8; A; 4.9; A). Nucleus contain single nucleolus which is roughly spherical in shape (Pls. 4.8; A; 4.9; A; 4.10; B). The nucleus has conspicuous peripheral heterochromatic bodies.

The dense cytoplasm of the epithelial cells also contains active Golgi bodies and ER elements. The ER cisternae appear in the form of long profiles aligned parallel to the cell walls (Pl. 4.8; B, D) and also found distributed throughout the cytoplasm (Pl. 4.8; C). ER is studded with ribosome aggregates known as rough ER (Pl. 4.8; B-D). Distended ER elements are observed in the vicinity of the plasmalemma; some of them being attached to it (Pl. 4.8; D). Free ribosomes can be seen in the ground cytoplasm. The number of free ribosomes are less compared to preceding stage.

Numerous, very active Golgi bodies are seen in the cytoplasm. They appear in the form of stack of more or less compressed 6-10 cisternae with associated peripheral vesicles arranged parallel to each other (Pls. 4.8; B, C; 4.9; A-C; 4.10; B, C). Golgi vesicles appear to be originating in large numbers into surrounding cytoplasm from the swollen rim of cisternae. Some of the Golgi vesicles from the cisternae appear in a chain-like arrangement (Pl. 4.8; C, at arrow head). Frequently, Golgi vesicles are seen in the extraplasmic spaces along the radial walls (Pl. 4.8; D, at arrows).
The epithelial cells show increase in the volume of vacuome compared to preceding stages. A large number of small vacuoles are distributed throughout the cytoplasm, which are filled with or without electron dense material (Pls. 4.8IA; 4.9IA). The number of vacuoles containing electron dense material (osmiophilic material) is reduced considerably compared to the earlier stages. Nearly small vacuoles often show fusion to form large vacuoles (Pls. 4.8IA, 4.9IA, at broad arrows). Some of the vacuoles contain granular material and myelin-like bodies inside (Pls. 4.8B; 4.9A; 4.10D). Frequently vacuoles containing granular material appear close to the radial walls, which are distended (Pls. 4.8B; 4.9A; 4.10A,C).

iv) Mature stage (Leaves 8.0-12.0 cm long): After the cessation of secretion, the volume of the vacuome increases compared to the ground cytoplasm. The epithelial cells of the nectary become more vacuolated (Pls. 4.2IB; 4.12IA). The empty appearance of epithelial cells is due to the cytoplasm being predominantly confined to parietal zone by the large vacuoles which occupies much of the cell volume (Pl. 4.12IA). The cytoplasm of the epithelial cells becomes opaque with less amount of cell organelles. At this stage, compartmentation between the cytoplasm and vacuole is lost, and organelle and membrane definition become obscured (Pl. 4.12IB). Lipid bodies are present (Pl. 4.12IA) and some membrane elements and organelles could still be
Plate 4.1 (A-F): Showing light micrographs of the extrafloral nectary:

A - Extrafloral nectaries situated at the junction of the lamina and distal part of the petiole on adaxial side.

B, C - Cross-section of the petiole showing the nectary meristems.

D - Longisection of the nectary showing the outer elongated epidermal cells and inner ground tissue.

E - Longitudinal section of young nectary showing palisade-like epithelial cells (epidermal cells) containing dense cytoplasm and nuclei, note vacuolated subepithelial cells (ground tissue).

F - Longitudinal section of the nectaries having short stalk and elongated head containing palisade-like outer epidermal cells and inner ground tissue.

(A = 450 X;  B = 210 X;  C = 145 X;
D = 1440 X;  E = 240 X)

e - epithelial cells;  h - head;  la - lamina;
n - nectary;  nm - nectary meristems;  s - stalk;
se - subepithelial cells.
Plate 4.2 (A-E): Light micrographs of extrafloral nectary:

A - Longitudinal section of nectary showing epithelial cells containing dense cytoplasm and vacuolated sub-epithelial cells, note the nucleus with prominent nucleolus.

B - Cross-section of the nectary showing outer palisade-like epidermal cells and inner isodiamic ground tissue.

C - Longitudinal section of nectary showing vacuolated epithelial cells, note the highly vacuolated sub-epithelial cells.

D - Longitudinal section of mature nectary showing highly vacuolated epithelial and sub-epithelial cells.

E - Cross-section of epithelial cells of the nectary showing the intercellular spaces among three or four cells (extraplasmic spaces, at arrows)

( A = 575 X; B = 620 X; C = 720 X;
  D = 240 X; E = 1240 X )

e = epithelial cells; se = subepithelial cells.
Plate 4.2 (A-E)
Plate 4.2
Plate 4.3 (A-C): Electron micrographs showing longitudinal sections of young nectaries:

A - Epithelial and subepithelial cells showing cell contents, note the electron dense material in the plastids and vacuoles and plasmodesmal connections between adjacent epithelial cells and adjacent epithelial and subepithelial cells.

B - An upper portion of the epithelial cell showing plastids with electron dense material, electron translucent mitochondria and polyribosomes.

C - An upper portion of the epithelial cell showing thin cuticle on the outer tangential wall, note the mitochondria and lipid bodies.

(A - 5500 X; B - 26000 X; C - 20000 X)

c - cuticle; cw - cell wall; d - dictyosome; e - epithelial cell; l - lipid body; m - mitochondria; n - nucleus; nu - nucleolus; p - plastid; pd - plasmodesmata; rer - rough endoplasmic reticulum; se - subepithelial cells; v - vacuoles.
Plate 4-3 (A-C)
Plate 4.4 (A-D): Electron micrographs showing longitudinal sections of epithelial cells:

A - A plastid showing electron dense material (osmiophilic material) around the thylakoids.

B - An upper portion of the epithelial cell showing enlarged cuticle on the outer tangential wall, note the dictyosome and rough ER.

C - Middle portion of the epithelial cell showing the rough ER near the cell wall, note the electron dense material in plastid and electron translucent mitochondria.

D - Middle portion of the epithelial cell showing swollen rough ER cisternae parallel to the cell wall.

( A - 93400 X; B, C - 34800 X; D - 63000 X )

c - cuticle; cw - cell wall; d - dictyosome;
m - mitochondria; p - plastid; pl - plasmalemma;
rer - rough endoplasmic reticulum; v - vacuole.
Plate 4.4 ( A-D )
Plate 4.5 (A-D): Electron micrographs showing the longitudinal sections of epithelial cells of the nectary:

A, B - An upper portion of the epithelial cells near radial walls showing the longitudinally cut microtubules, note the polyribosomes.

C - Middle portion of the epithelial cell showing the globular lipid bodies with electron translucent spaces, note the polyribosomes.

D - Middle portion of the epithelial cell showing the electron dense material (osmiophilic material) the vacuoles.

(A, B = 57400 X; C = 34000 X; D = 15200 X)

cw - cell wall; d - dictyosome; l - lipid; mt - microtubules; rer - rough endoplasmic reticulum; v - vacuole.
Plate 4.5 (A-D)
Plate 4.6 (A-F): Electron micrographs showing the longitudinal sections of epithelial cells:

A - A row of epithelial cells showing granular dense cytoplasm, note less vacuoles in cytoplasm, and plastids with starch grains.

B - Middle portion of the epithelial cell showing numerous dictyosomes, plastids loaded with starch grains, note the undulations of plasmalemma along radial walls.

C - Plasmalemma showing invaginations along the radial wall forming extraplasmic space containing vesicles and electron dense material, note the vesicles, mitochondria and dictyosomes with peripheral vesicles in the vicinity of plasmalemma.

D - Middle portion of the epithelial cell showing extraplasmic space, note the long profiles of rough ER parallel to the cell wall and dictyosomes with vesicles.

E - Lower portion of the epithelial cell showing long profiles of rough ER, note the concentric whorls of rough ER and dictyosomes.

F - Middle portion of the epithelial cell showing densely populated dictyosomes with peripheral vesicles, note numerous vesicles in the cytoplasm.

( A = 2200 X; B = 7000 X; C = 42400 X;
D, F = 20800 X; E = 16500 X )

c = cuticle; cw = cell wall; d = dictyosome;
m = mitochondria; n = nucleus; nu = nucleolus;
p = plastids; pl = plasmalemma; rer = rough endoplasmic reticulum; v = vacuole; vi = vesicles.
Plate 4.6 (A-F)
Plate 4.7 (A-D): Electron micrographs showing the longitudinal sections of the epithelial cells:

A-D - Plastids showing large starch grains occupying much of the stroma and less electron dense material, note the profiles of RER along the radial walls, mitochondria and dictyosomes.

(A = 15600 X; B = 24700 X; C = 18500 X; D = 19500 X)

d = dictyosomes; m = mitochondria; p = plastid; per = rough endoplasmic reticulum; s = starch grains.
Plate 4.7 (A-D)
Plate 4.8 (A-E): Electron micrographs showing the longitudinal sections of epithelial cells:

A - Epithelial cells showing numerous small vacuoles and accumulation of secretory material in the extraplasmic space along the radial walls, note the less number of starch grains in the plastids and highly vacuolated subepithelial cells.

B - Lower portion of epithelial cell showing large vacuole containing fibrillar material, note the secretory material in extraplasmic space.

C - Middle portion of the epithelial cell showing densely populated dictyosomes with peripheral vesicles and rough ER, note the secretory material in the extraplasmic space.

D - A large extraplasmic space showing the secretory material containing multi-vesicular and multilamellar structures.

E - An enlarged portion of secretory material showing concentric rings of multilamellar structures.

( A = 5000 X; B = 8400 X; C = 6300 X; D = 29000 X; E = 99200 X )

cw - cell wall; d - dictyosomes; e - epithelial cells; ml - multilamellar bodies; n - nucleus; nu - nucleolus; p - plastid; pl - plasmalemma; rer - rough endoplasmic reticulum; se - subepithelial cells; sm - secretory material; v - vacuole.
Plate 4.9 (A-C) : Electron micrographs showing the longi-sections of epithelial cells:

A - Epithelial cells showing high amount of secretory material in extraplasmic space along the radial walls, note the plastids with a few starch grains, high vacuolation, cuticle on the outer tangential wall and nucleus with nucleolus.

B - Middle portion of the epithelial cell showing vacuoles with no electron dense material, note the dictyosomes with peripheral vesicles, lipid body and RER.

C - Middle portion of the epithelial cell showing plastids with thylakoids, dictyosomes, note the lack of electron dense material and a few starch grains in plastids.

( A = 2700 X; B = 33200 X; C = 15200 X )

c - cuticle; cv - cell wall; d - dictyosomes; l - lipid; m - mitochondria; n - nucleus; nu - nucleolus; p - plastid; pl - plasmalemma; rer - rough endoplasmic reticulum; sm - secretory material; v - vacuoles; vi - vesicles.
Plate 4.9 (A-C)
Plate 4.10 (A-D): Electron micrographs showing longitudinal sections of the epithelial cells:

A - Middle portion of the epithelial cell showing the secretory material in extraplasmic space along the radial walls, note the vacuoles with less amount of electron dense material.

B - An epithelial cell showing nucleus with a prominent nucleolus, note the secretory material in extraplasmic space along the radial walls; plastids with a few small starch grains.

C - Lower portion of the epithelial cell showing the secretory material along the radial walls in extraplasmic space and vacuoles with electron dense fibrillar material.

D - Middle portion of the epithelial cell showing vacuoles with less amount of electron dense material, note the mitochondria and dictyosomes.

( A, B - 7200 X; C - 6800 X; D - 16000 X )

ew - cell wall; d - dictyosome; m - mitochondria;
n - nucleus; nu - nucleolus; p - plastid; pl - plasmalemma; sm - secretory material; v - vacuole.
Plate 40.10 (A-D)
Plate V.11 (A-D): Electron micrographs showing the longiseetions of epithelial cells:

A - Middle portion of the epithelial cell showing the cell organelles.

B - Middle portion of the epithelial cell showing the high amount of secretory material in extraplasmic space and vacuole with dense fibrillar material.

C, D - An upper portion of the epithelial cells showing the outer tangential wall and secretory material in subcuticular space.

(A = 15600 X; B = 12400 X; C = 32400 X;
D = 25300 X)

C = cuticle; cw = cell wall; d = dictyosome;
p = plastid; pl = plasmalemma; rer = rough endoplasmic reticulum; sm = secretory material;
v = vacuole.
Plate 411 (A-D)
Plate 4.12 (A-D) : Electron micrographs showing longitudinal sections of epithelial cells :

A - Epithelial cells showing high vacuolation. Vacuoles contain electron dense material i.e. cytoplasmic debris, note the secretory material along the radial walls.

B - Middle portion of the epithelial cell showing the dissolution of cell organelles in cytoplasm.

C, D - Upper portion of the epithelial cells showing large amount of secretory material in subcuticular space, note the detachment of cuticle from the cell wall in figure D.

( A = 4300 X; B = 7200 X; C = 12400 X; D = 20000 X )

c - cuticle; cd - cytoplasmic debris; cw - cell wall; sm - secretory material; v - vacuole.
Plate 4.13 (A-D) : Light micrographs of epithelial cells of nectary showing histochemical localization of Acid phosphatase activity (A), Periodic acid/Schiff's reagent (B), Proteins (C) and Lipids (D).

( A = 360 X; B, C = 420 X; D = 380 X )
Plate 4.13 (A-D)
11) Pumera rubra

Each leaf bear a group of ten to fifteen extrafloral nectaries on the adaxial side at their base (Fl. 4.13:A). There is no definite pattern of arrangement of nectaries. Each nectary is differentiated into a long head and a short stalk without vascular supply. The head of the nectary containing an outermost layer of epithelial cells surrounds the sub-epithelial ground tissue (Fl. 4.14:B-D). The length and diameter of the nectaries vary from 0.8-1.2 mm and 0.2-0.5 mm respectively.

The nectary of the leaf follow the same pattern of development as in Holorrhena antidysenterica.

ULTRASTRUCTURE

Young stage (Leaves 5-12 mm long) :

The epithelial cells of the young nectary contain granular dense cytoplasm.

Nucleus with almost a single spherical nucleolus lies more or less in the centre of the epithelial cells (Fls. 4.14; B; 4.15:A,B). The nucleus has several conspicuous peripheral heterochromatin bodies (Fl. 4.15:A-C).
Epithelial cell walls are very thin with closely appressed plasmalemma (Pls. 4,15: A-C, 4,16: A). Cuticle is present on the outer periphery of the upper tangential wall of the epithelial cells (Pls. 4,14:B, 4,15:A,B, 4,16:E). Plasmodesmata traverse the lower tangential wall of the epithelial cells and adjacent sub-epithelial cells and the wall between adjacent epithelial cells (Pls. 4,17:B,D).

Careful observations revealed that there are no plasmodesmata on the outer tangential wall of the epithelial cells. Rough ER and mitochondria lie in proximity of the plasmodesmata (Pl. 4,17:B).

The epithelial cells contain dense cytoplasm and few large vacuoles at both the poles of the epithelial cells. The cytoplasm contain ER, Golgi bodies, mitochondria, plastids and numerous ribosomes.

ER generally occurs in the form of ribosome studded cisternae, i.e. rough ER (Pls. 4,15:A,C, 4,16:A,B). The cisternae appear much flattened which in sectional view appear as double lines (Pl. 4,16:A,B). The ER cisternae in the form of short and long segments occur in the cytoplasm near the cell walls (Pl. 4,16:A,B) and also throughout the cytoplasm (Pl. 4,16:A). Rough ER shows linear pattern of polyribosome aggregates (Pl. 4,16:A,B). Rough ER cisternae lie nearer or in close contact with the plasmodesmata which is often clear (Pl. 4,17:B). Often long profiles of rough ER cisternae appear close or in contact with the plastids.
The ground cytoplasm is rich with free ribosomes and aggregates often in circular and spiral pattern of polyribosomes (Pl. 4.16:A-B, at arrows).

Only few Golgi bodies are seen in the cytoplasm and appear in the form of a stack of more or less compressed 5-6 cisternae with associated peripheral tubules and vesicles arranged parallel to each other (Pl. 4.16:B). They occur frequently near the cell walls, nuclei and occasionally distributed throughout the cytoplasm. Golgi vesicles appear to be originating from the swollen rim of cisternae into the surrounding cytoplasm (Pl. 4.16:B).

In sections plastids vary in shape and size. Most of them appear as either almost oval, elongate, dumbbell-shaped or erratic (Pls. 4.15:A-C; 4.16:B). Plastids are situated mostly near the nucleus and cell walls (Pl. 4.15:A-B). Plastid interior is occupied by a few starch grains (Pl. 4.15:A). Starch grains appear as electron translucent regions in micrographs and roughly round in shape.

Mitochondria are common organelles distributed throughout the cytoplasm. They are variable in shape and appear as globular, elongate or oval in shape (Pl. 4.16:A). Mitochondria contain well developed cristae (Pl. 4.16:A).
Large vacuoles with irregular shapes appear in the cytoplasm. These vacuoles are situated at both the ends of epithelial cells (Pl. 4.15:A,B). Apparent fusion among the vacuoles has been noticed (Pl. 4.15:C, at arrow). The vacuoles contain granular material. Small globoid lipid bodies are seen in the cytoplasm (Pls. 4.15:A,C, 4.16:A).

**Pre-secretion** (Leaves 1.3-3.5 cm long)

The epithelial cells after reaching their maximal size contain very dense cytoplasm with cell organelles, viz. ER, Golgi bodies, mitochondria, plastids and ribosomes.

The epithelial cell walls are thick with undulated plasmalemma along the radial walls (Pls. 4.17:C, 4.18:A-D). Frequently plasmalemma invaginates into the cytoplasm along the radial walls (Pl. 4.18:A-C). Multivesicular structures (paramural bodies) are often seen in the plasmalemma invaginations, along the radial walls, where the initial development of the lumen occurs by dissolution of middle lamella of the cell wall (Pl. 4.18:A-C, at arrows). Many vesicles are filled with electron dense granular material, which are observed in the invaginations of plasmalemma outside the cytoplasm (Pl. 4.18:A-C). In the cytoplasm, near the multivesicular structures RER, SER, mitochondria and similar vesicles are often seen (Pl. 4.18:B,C).

Infrequently, small lipid bodies and vacuoles are seen near the plasmalemma invaginations (Pl. 4.18:B). Cuticle thickness
is increased on the outer tangential wall of the epithelial cell. The thick cuticle consists of a moderately electron-dense matrix, apparently composed of cutin and a fine fibrillar network of electron dense material (Pl. 4,19:D).

The amount of ER is more compared to early stage and occupies much of the cytoplasm. The ER cisternae appear in the form of long profiles frequently aligned parallel to the cell walls and also distributed throughout the cytoplasm (Pls. 4,16:C-D; 4,17:A-C). ER cisternae are studded with ribosome aggregates, i.e. rough ER (Pls. 4,16:C; 4,17:A,C). Some of the rough ER cisternae often appear as concentric whorls in the cytoplasm near the cell walls (Pls. 4,17:A; 4,20:B). Occasionally, some of the concentric whorls of ER surrounded by a portion of cytoplasm presumably forms pseudovacuole (Pls. 4,17:A; 4,20:B).

The edges of the parallelly arranged ER cisternae are swollen and some show constrictions (Pl. 4,17:A,C). Near these swollen edges there are many vesicles whose topography and arrangement suggest that they have budded off from the ER cisternae (Pl. 4,16:C; 4,17:A,C). These vesicles are seen close to and in contact with the plasmalemma (Pl. 4,17:C) and occasionally the swollen edges themselves are continuous with the plasmalemma (Pls. 4,17:C; 4,18:B). In addition to the ER, electron translucent SER tubules (Pl. 4,16:C,D) occur in the cytoplasm. SER tubules were sometimes in close
contact with the plasmalemma. These SER tubules are distributed throughout the cytoplasm. Numerous free ribosomes appear in cytoplasm and sometimes they occur as aggregates of polyribosomes.

Active Golgi bodies are seen at this stage in the cytoplasm. They appear in the form of a stack of more or less compressed closed 5-6 cisternae with associated peripheral vesicles and tubules arranged parallel to each other (Pls. 4.16:C,D; 4.17:A; 4.18:D). They are abundantly distributed throughout the cytoplasm. Golgi vesicles appear to be originating in large number into the surrounding cytoplasm (Pls. 4.16:C,D; 4.17:A; 4.18:D). In the cytoplasm Golgi bodies with peripheral vesicles are appearing close to the plasmalemma. Some of these vesicles of Golgi bodies are often close and in contact with the plasmalemma (Pl. 4.18:D, at curved arrow).

Plastids in sections vary in shape and size. Most of them appear as either elongated, dumbbell-shaped or oval shaped (Pl. 4.17:A). Plastids are situated mostly near the cell walls, nucleus and throughout the cytoplasm. Occasionally, a cluster of plastids are seen in the proximity of nucleus at the lower side of the epithelial cells. No marked difference is observed in plastid interior, occupied by few starch grains (Pl. 4.17:A).
The number of mitochondria is increased compared to young stage. They are variable in shape and appear globular, elongate or oval shaped with well developed cristae. Some of the mitochondria show electron translucent spaces in their matrix (Pls. 4.17:A; 4.18:A). Close to some mitochondria, where their enveloping double membrane is open, a row of small vesicles is detected (Pl. 4.16:D, at arrows). In some mitochondria the dissolution of cristae can be seen (Pls. 4.16:C; 4.17:C).

The volume of the vacuole is reduced considerably. Only few small vacuoles are distributed in the cytoplasm containing granular material (Pl. 4.18:B). Infrequently, some of the vacuoles occur close to the plasmalemma invaginations.

**Secretion stage** (Leaves 3.6-15.0 cm long):

At the stage of secretion the epithelial cells contain granular dense cytoplasm and less vacuoles. The epithelial cells show increase in the volume of the ground cytoplasm, containing ER, Golgi bodies, mitochondria, plastids, ribosomes etc. The cytoplasm of the epithelial cells shows dense population of cell organelles.

The epithelial cell wall microfibrils become loose, and the middle lamella of the cell wall along the radial wall
of the epithelial cells gets dissolved forming a lumen in
the wall between adjacent epithelial cells (Figs. 4.18;E;
4.19;A-C,F). The secretory material is deposited in the
cell wall lumen between the radial walls of this adjacent
epithelial cells and in the sub-cuticular space (Figs.
4.19;A-C,F; 4.20;A; 4.21;A-D; 4.22;A-C). Simultaneously,
the cuticle shows detachment from the upper tangential wall
of the epithelial cell (Pl. 4.19;D). In the cytoplasm many
multivesicular bodies are often seen (Figs. 4.19;E,F; 4.20;A;
4.21;B-D). Infrequently, besides multivesicular structures,
multilamellar structures do occur in the plasmalemma
invaginations (Pl. 4.19;A). These multivesicular structures
disperse in the cell wall microfibrils and pass through it
into the wall lumen (Figs. 4.18;E; 4.19;A; 4.21;B, at arrows).
Often some of the vesicles appear at the outer periphery of
the cell wall (at the boundary of lumen) where the secretory
material is deposited (Figs. 4.19;E; 4.21;C,D, at arrows).
Occasionally, multivesicular structures appear at the lower
tangential wall of the epithelial cell (Pl. 4.20;B). In the
proximity of plasmalemma invaginations, occasionally
mitochondria, myelin-like bodies and ribosomes are seen
(Pl. 4.21;B). At maximum secretion stage accumulation of
secretory material in the lumen along the radial walls of
the epithelial cells and in the sub-cuticular space is seen
in high amounts (Figs. 4.14;C,D; 4.20;A; 4.21;A,C,D; 4.22;A-E).
Some of the plasmalemma invaginations lack the multivesicular
bodies. Presumably these vesicles containing secretory material might pass through the cell wall into the lumen where the secretory material is deposited. This shows that the secretion takes place via vesicles from the cytoplasm to outside of the cell.

ER is the dominant cell organelle at the stage of secretion in the cytoplasm and occupies much of its volume. Two types of endoplasmic reticulum (ER) are observed: (i) typical ribosome studded cisternae rough ER, and (ii) electron translucent ramifying smooth endoplasmic reticulum (SER) tubules. At the stage of secretion ER show dilations and appears to be vesiculated. These vesicles are seen throughout the cytoplasm (Figs. 4.18 F; 4.19 D, E; 4.20 A; 4.21 A; 4.22 A). Some of the vesicles are partly filled with deposits of electron dense material. These vesicles are seen close to an in contact with the plasmalemma (Fig. 4.18 E, at curved arrow). Some of the SER tubules are seen in the vicinity of plasmalemma and occasionally in contact with it. Many vesicles are seen in the plasmalemma invaginations, whose topography suggest that they are vesiculated ER (Figs. 4.18 E; 4.19 A; 4.20 A; 4.21 B-D). Some of the vesicles are seen in the middle lamellae of cell wall and near the vicinity of the cell wall lumen along the radial walls of the epithelial cells (Figs. 4.18 E; 4.19 A). These vesicles are probably transporting the secretory material from the cytoplasm to outside the cell by
exocytosis. Rough ER, SER tubules and vesicles derived from the ER are seen in close proximity of the plasmalemma invaginations and often in close contact with them (Pls. 4.19; C, F).

Golgi bodies are less developed at the secretion stage compared to the preceding stages. Golgi bodies appear in the cytoplasmas in the form of stack of 5-8 cisternae with associated peripheral vesicles (Pls. 4.19: B; 4.21: D). Golgi vesicles appear to be produced in large numbers into the surrounding cytoplasm (Pl. 4.21: D). Golgi bodies with peripheral vesicles appear close to the plasmalemma invaginations (Pl. 4.21: D). Besides the vesiculate ER, Golgi vesicles are probably playing a role in transporting the secretory material by exocytosis, i.e. from the cytoplasm to outside the cell wall.

In sections plastids vary in shape and size. Most of them appear as either elongate, dumbbell-shaped or oval. Plastids are situated mostly near the cell wall and nucleus. No marked difference is seen in plastid interior, occupied by a few starch grains. Some plastids are without starch grains.

Numerous mitochondria are observed in the cytoplasm, with well developed cristae. These mitochondria appear as oval, round or elongate (Pls. 4.19: B, D-F; 4.20: A, B; 4.21: B; 4.22: B). Often mitochondria are observed in the vicinity of
plasmalemma invaginations along with ribosomes and myelin-like bodies. At this stage there is increase in number of mitochondria as compared to preceding stages.

Globular lipid bodies are observed in the cytoplasm at the apical region. Myelin-like bodies are seen near the plasmalemma invaginations (Pl. 4,21:B).

Before the secretion ceases, the ground cytoplasm of the epithelial cell becomes vesiculated, occupying much of the cell volume. The volume of the vacuole increased. Vacuoles of various shapes and sizes containing granular material appear in cytoplasm (Pl. 4,22:A). Numerous globoid lipid bodies are seen throughout the cytoplasm (Pl. 4,22:A).

High amount of secretory material is observed between the radial walls of the epithelial cells and in the subcuticular space which consists of variously sized droplets containing granular material (Pl. 4,22:A-C). Cuticle bursts off due to the pressure exerted by the high amount of underlying secretory material. The nature and topography of secretory material outside the cuticle is same to that in subcuticular space and in between cell walls (Pl. 4,14:C,D).

Mature stage (Leaves 15.0-20.0 cm long):

After the cessation of secretion the cytoplasm of the
epithelial cells becomes opaque and the volume of the vacuole increases considerably. Preceding to mature stage many autophagic vacuoles are seen in the cytoplasm. Many multi-vesicular bodies are seen in the invaginations of the tonoplast membrane into the vacuole (Pl. 4.22; D). At maturation, the vacuoles occupy much of the cell volume leaving only a thin layer of peripheral cytoplasm along the cell walls (Pl. 4.23; A, B, D). The cytoplasm of the epithelial cells show less amount of cell organelles. Only a few short segments of ER cisternae occur in the cytoplasm near the cell walls (Pls. 4.22; D; 4.23; B, C). The density of dictyosomes also decreases and appear in the form of stack of 3-4 cisternae with attached peripheral vesicles. Lipid bodies are numerous in the cytoplasm. At later stages of development cytolytic processes and death of the cells occur.
Plate 4.14 (A-D): Showing the light micrographs of the extrafloral nectary:

A - Extrafloral nectaries situated on adaxial side at the base of the petiole.

B-D - Longitudinal sections of the nectary showing palisade-like epidermal (epithelial) cells and ground tissue (sub-epithelial cells)

B - Part of the nectary showing elongated epithelial cells with cuticle on outer tangential walls.

C - Showing the detached cuticle and secretory material in subcuticular space.

D - Showing the secretory material outside the cuticle and in subcuticular space, note the detached cuticle from the outer tangential walls of the epithelial cells.

(A - 21 X; B - 450 X; C - 575 X; D - 225 X)

c - cuticle; e - epithelial cells; n - nectary;
p - petiole; se - subepithelial cell; sm - secretory material.
Plate 4.15 (A-C): Electron micrographs showing longisections of epithelial cells:

A - An enlarged epithelial cell from 'B' showing prominent cuticle on the outer tangential wall, nucleus with a nucleolus, and ground cytoplasm with distribution of vacuoles, elongated and variously shaped plastids, mitochondria, endoplasmic reticulum, golgi bodies and lipid bodies.

B - Radially elongated and tangentially adjacent row of epithelial cells showing median nuclei and nucleolus, vacuoles grouped at the two radial ends and the other cell organelles.

C - Middle portion of the epithelial cells showing thin radial walls with an appressed plasmalemma, nuclei, plastids, sER, lipid bodies and vacuoles apparently fusing (arrows)

( A - 6300 X; B - 3900 X; C - 7600 X )

c - cuticle; cw - cell wall; d - dictyosomes; l - lipid bodies; m - mitochondria; n - nucleus; nu - nucleolus; p - plastid; pl - plasmalemma; rer - rough endoplasmic reticulum; v - vacuole.
Plate 4.16 (A-E) Contd....

E - Upper portion of the epithelial cell showing the cuticle lying on the upper tangential wall, note the closely appressed plasmalemma with cell wall and dilated smooth ER.

(A - 26000 X; B - 39600 X; C - 20500 X; D - 27400 X; E - 22100 X)

c - cuticle; cw - cell wall; d - dictyosome;
l - lipid; m - mitochondria; p - plastid;
pl - plasmalemma; rer - rough endoplasmic reticulum; ser - smooth endoplasmic reticulum;
v - vacuoles; vi - vesicles.
Plate 4.16 (A-E) Contd...
Plate 4.16 (A-E) : Electron micrographs showing longitudinal sections of epithelial cells:

A - Upper portion of epithelial cell showing grouped mitochondria with well developed cristae, RER, dictyosomes and polyribosomes (arrows), note the lipid body.

B - Epithelial cell showing dictyosomes with attached vesicles, cisternal ER running parallel with an elongated plastid, mitochondria with wider cristae, distributed in ribosomal rich cytoplasm.

C - Middle portion of the epithelial cell showing vesicular and rough ER, dictyosomes forming large vesicles, and mitochondria with electron translucent matrix.

D - Middle portion of the epithelial cell showing the well developed tubular ER, note the vesiculate structures attaching to the mitochondria (at arrows).
Plate 4-16 ( A-E )
Plate 4,17 (A-D): Electron micrographs showing longitudinal sections of epithelial cells:

A - Epithelial cell showing concentric whorls of extensively developed ER and many secretory vesicles.

B - Lower tangential wall showing the plasmodesmatal connections between adjacent epithelial and subepithelial cell.

C - Middle portion of epithelial cell showing the well developed ER, note the mitochondria showing the electron translucency; constrictions in the ER cisternae.

D - Radial wall adjoining the epithelial cells showing plasmodesmata and associated vesicles forming parasmal body (PB); SER can be seen extensively along the wall.

(A, B = 45100 X; C = 16400 X; D = 26000 X)

cw - cell wall; e - epithelial cell;
m - mitochondria; p - plastid; pb - parasmal body; pd - plasmodesmata; pl - plasmalemma;
rer - rough endoplasmic reticulum; se - subepithelial cell; ser - smooth endoplasmic reticulum; sv - secretory vesicle; v - vacuole; vi - vesicles.
Plate 4, 17 ( A-D )
Plate 4.18 (A-E) : Electron micrographs showing longitudinal sections of epithelial cells :

A - Radial wall showing plasmalemma invagination with a secretory vesicle and undulation of plasmalemma, note the loosening of the cell wall microfibrils and formation of lumen.

B - Plasmalemma showing extensive invaginations into cytoplasm with associated vesicles on radial wall, note the formation of cell wall lumen (arrows). Electron translucent mitochondria, SER, lipid body and secretory vesicles near the plasmalemma.

C - Plasmalemma invagination showing various sizes and shapes of vesicles and radial wall with lumens (arrows).

D - Radial wall showing the radial extension of lumen along the centre of cell wall, note the dictyosomes with peripheral vesicles near the wall and fusion of vesicle with the plasmalemma (curved arrow).

E - Radial wall showing widely separated plasmalemma into cytoplasm with vesicles, note the arrangement of vesicles on either side of the plasmalemma (arrows).

( A - 50400 X; B - 26600 X; C - 57600 X; D - 38500 X; E - 35700 X )

cv - coated vesicle; cw - cell wall; d - dictyosomes; l - lipid; m - mitochondria; pb - paramural body; pl - plasmalemma; ser - smooth endoplasmic reticulum; sv - secretory vesicle; v - vacuole; vi - vesicles.
Plate 4-18 (A-E)
Plate 4.19 (A-F) Electron micrographs showing longitudinal sections of epithelial cells:

A - Radial wall showing numerous spherical secretory vesicles in the cell wall, note the concentric whorls of membranous structures in the cell wall along the secretory vesicles.

B - Radial wall showing the accumulation of secretory material in the lumen, note the undulation of plasmalemma, dictyosomes attached with vesicles, ER and mitochondria near the plasmalemma.

C - Radial wall of the epithelial cell showing the accumulation of secretory material in cell wall lumen, note large paramural body with vesicles. ER close to the paramural body.

D - Upper tangential wall showing the separation of cuticle from cell wall, note the SER and mitochondria near the plasmalemma.

E - Upper tangential wall showing detached cuticle from cell forming subcuticular space filled with secretory material, note the secretory vesicles on the outer side of the cell wall (arrows). Multivesicular bodies and SER near the site of secretion in the cytoplasm.

F - Apical portion between two cells showing large lumen formed by the separation of the radial walls and cuticle, note the deposition of secretory material in the lumen, multivesicular bodies near the plasmalemma in the cytoplasm and SER.

(A - 57600 X; B - 28200 X; C - 47600 X; D - 18800 X; E - 20000 X; F - 22100 X)

c - cuticle; cw - cell wall; d - dictyosome; lu - lumen; m - mitochondria; ml - multilamellar body; mb - multivesicular body; pb - paramural body; pl - plasmalemma; rer - rough endoplasmic reticulum; ser - smooth endoplasmic reticulum; sm - secretory material; sv - secretory vesicle.
Plate 4, 19 (A-F)
Plate 4.20 (A,B): Electron micrographs showing longitudinal sections of epithelial cell:

A - Apical position of the cells showing the detached cuticle from cell wall with the deposition of secretory material in subcuticular space.

B - Lower portion of the epithelial cell showing large concentric whorls of RER surrounding a vacuole (pseudo-vacuole), mitochondria with well developed cristae, note the parasmural body on the lower tangential wall.

(A - 15540 X; B - 47250 X)

c - cuticle; cw - cell wall; d - dictyosome; m - mitochondria; pb - parasmural body; pl - plasmalemma; psv - pseudovacuole; rer - rough endoplasmic reticulum; sm - secretory material.
Plate 4.20
Plate 4.21 (A-D): Electron micrographs showing the longitudinal sections of the epithelial cells:

A - Cells showing separation of radial walls and detachment of cuticle from outer tangential walls, note the distribution of secretory material in the lumen in between the cell walls and extensive network of SER in the cytoplasm.

B - Part of epithelial cell showing myelin-like bodies close to the radial wall, note peramural body, mitochondria and vesicles near the wall region.

C, D - Radial walls of the cells showing the deposition of high amount of secretory material in the wall lumen between two cells, note the secretory vesicles on the inner and outer periphery of the cell wall (arrows).

( A - 5900 X; B - 38200 X; C - 20500 X; D - 26600 X )

c - cuticle; cw - cell wall; m - mitochondria; mb - myelin-like bodies; pb - paramural bodies; pl - plasmalemma; ser - smooth endoplasmic reticulum; sm - secretory material; sv - secretory vesicle.
Plate 4.21 (A-D)
Plate 4.22 (A-D): Electron micrographs showing the longitudinal sections of the epithelial cells:

A - Upper portion of the epithelial cell showing the secretory material in subcuticular space, note the numerous secretory vesicles, lipid bodies in the cytoplasm.

B - Upper portion of the epithelial cell showing the variously sized numerous droplets of secretory material in the subcuticular space.

C - Upper portion of the epithelial cell showing detached cuticle and deposition of secretory material all around the cells.

D - Middle portion of the epithelial cell showing the electron translucent ground cytoplasm, autophagic vacuoles (av) with multivesicular bodies, lipid bodies and mitochondria.

( A - 10800 X; B - 9800 X; C - 4900 X; D - 34000 X )

av = autophagic vacuoles; c = cuticle; cw = cell wall; l = lipid bodies; m = mitochondria; mvb = multivesicular bodies; sm = secretory material; sv = secretory vesicles.
Plate 4.23 (A-D): Electron micrographs showing longitudinal sections of epithelial cells:

A - A portion of the epithelial cell showing a large vacuole occupying much of the cell volume with a thin peripheral cytoplasm.

B - Part of the epithelial cell showing large lipid bodies and small fragments of ER near the radial wall.

C - Cell showing the oval lipid bodies, mitochondria and large vacuoles.

D - Radial wall of the cell showing the deposition of the secretory material and cell with large vacuoles with a thin peripheral cytoplasm.

( A - 7900 X; B - 34000 X; C - 45100 X; D - 4800 X )

cw - cell wall; l - lipid bodies; m - mitochondria; rer - rough endoplasmic reticulum; sm - secretory material; v - vacuole.
Plate 4.23 (A-D)
Plate 4.24 (A-D): Light micrographs of the epithelial cells of the nectary showing the histochemical localization of:

A - Acid phosphatase
B - PAS reaction
C - Proteins
D - Lipids

( A - 145 X; B - 500 X; C - 560 X; D - 360 X )

c - cuticle; sm - secretory material.
Plate 4.24 (A-D)
The nectary starts secreting when the leaf is still young, about 5 mm long. Secretion continues during leaf maturation.

Secreted substances within the epithelial cells and those secreted on the nectary were detected with histochemical stains. The secreted material contains lipidic compounds, insoluble polysaccharides and proteins (Fls. 1, 13; D; 4, 24; D). A rough estimation of relative amounts of secreted material reveals a gradient from epithelial cells which secrete the largest amount of all types of materials (Table 18).

At different stages of leaf development materials are secreted by epithelial cells. Lipids are produced continuously from young stage to mature stage. Protein production is greatest when the leaves are about 2.6-12.0 cm long in both the species. Polysaccharides are produced along with the lipids continuously (Table 19).

Lipids appeared as tiny globules in epithelial cells stained with Sudan Black "B" (Fls. 1, 13; D; 4, 24; D, at arrows). The lipidic material appeared in high amounts was detected at the stage of secretion until the secretion ceases. This lipidic material is seen in the secretory material, which is deposited in the sub-cuticular space (in between cuticle and cell walls) below the cuticle and outside the cuticle.
(Pl. 13A, 24A, at arrows). Lipidic material in high amounts was detected in secretory material of *Plumeria rubra* (Pl. 24D), while less in *Holarrhena antidysenterica*. Proteins appeared in the epithelial cells as tiny particles stained blue in colour with mercuric bromophenol (Pls. 13B, 24C, at arrows). Proteins are seen along with lipids in the secretory material. Polysaccharides are the next dominant compounds appeared in the secretory material along with lipids which stained red with PAS reaction appeared both in the epithelial and the secretory material in the sub-cuticular space and outside the cuticle (Pls. 13E, 24B, at arrows).

**Acid Phosphatase activity**

The results of Gomori test indicate that high acid phosphatase activity occurs in the epithelial cells of the nectary (Pls. 13A, 24A). The reaction to the Gomori test is less pronounced in the epithelial cells of the young nectary and becomes more distinct towards the stage of secretion. The epithelial cells of the nectary show high acid phosphatase activity compared to sub-epithelial cells. The lower activity observed in sub-epithelial cells may be related to the occurrence of a vacuole which is revealed in the stages studied.
### TABLE 18

**SHOWING RELATIVE AMOUNTS OF SUBSTANCES SECRETED BY EPITHELIAL CELLS**

<table>
<thead>
<tr>
<th>Stain</th>
<th>Compounds stained</th>
<th>Colour</th>
<th>Epithelial cells of Holarrhena</th>
<th>Epithelial cells of Plumeria</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sudan Black B</td>
<td>Lipids</td>
<td>Blue</td>
<td>+++</td>
<td>+++</td>
</tr>
<tr>
<td>Nile Blue</td>
<td>Fats, oils, waxes, volatile terpenes</td>
<td>Pink</td>
<td>++</td>
<td>++</td>
</tr>
<tr>
<td></td>
<td>Acidic lipids</td>
<td>Blue</td>
<td>++</td>
<td>++</td>
</tr>
<tr>
<td>PAS</td>
<td>Insoluble polysaccharides</td>
<td>Red</td>
<td>+</td>
<td>++</td>
</tr>
<tr>
<td>Bromo-phenol Blue-HgCl</td>
<td>Proteins</td>
<td>Blue</td>
<td>++</td>
<td>+++</td>
</tr>
<tr>
<td>I₂KI</td>
<td>Starch</td>
<td>Brown-black</td>
<td>++</td>
<td>+</td>
</tr>
</tbody>
</table>

**N.B.** Amount of material:  
+ = small; ++ = medium;  
+++ = large.
### TABLE 19: SHOWING SECRETORY SUBSTANCES IN EPITHELIAL CELLS AT DIFFERENT STAGES OF DEVELOPMENT

<table>
<thead>
<tr>
<th>Stage</th>
<th>Secreted material</th>
<th>Lipids</th>
<th>Polysaccharides</th>
<th>Proteins</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Nolarrhena antidysenterica</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Young stage (leaf length 5-15 mm)</td>
<td>+</td>
<td>†</td>
<td>-</td>
<td></td>
</tr>
<tr>
<td>Presecretion (leaf length 1.6-2.5 cm)</td>
<td>+</td>
<td>+</td>
<td>†</td>
<td></td>
</tr>
<tr>
<td>Secretion (leaf length 2.6-8.0 cm)</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td></td>
</tr>
<tr>
<td>Mature (leaf length 8.0-12.0 cm)</td>
<td>+</td>
<td>†</td>
<td>†</td>
<td></td>
</tr>
<tr>
<td><strong>Plumeria rubra</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Young stage (leaf length 5-12 mm)</td>
<td>+</td>
<td>†</td>
<td>†</td>
<td>†</td>
</tr>
<tr>
<td>Presecretion (leaf length 1.3-3.5 cm)</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td></td>
</tr>
<tr>
<td>Secretion (leaf length 3.6-15.0 cm)</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td></td>
</tr>
<tr>
<td>Mature stage (nearly mature leaf)</td>
<td>+</td>
<td>†</td>
<td>†</td>
<td>†</td>
</tr>
</tbody>
</table>

+ = present;  - = absent;  † = traces.
The course of development of the endoplasmic reticulum and Golgi bodies are seemingly more active and their number is greater in epithelial cells of the nectary at developmental stages prior to secretion. In *Holarrhena antidysenterica* both the ER elements and Golgi bodies are active at the stage of secretion. The process of swelling of the ER elements at the time of secretion is compatible. In *Plumeria rubra*, at the stage of secretion the ER elements are more active than the Golgi bodies which were very sparse. The vesicles derived from the Golgi and ER cisternae, suggested fusion of vesicles with the plasmalemma would result in an enlargement of its surface. It is possible that the multivesicular (paremural bodies) structures associated with the plasmalemma which are most evident in epithelial cells of *Plumeria rubra* at the stage of secretion, represent the enlargement of cell surface. Multivesicular structures are not seen in the *Holarrhena antidysenterica*. There is no indication as to how the plasmalemma returns to its normal size. It is suggested that the numerous vesicles bleb off from the ER cisternae and Golgi bodies involved in the secretion of *Holarrhena antidysenterica*, while in *Plumeria rubra* many of multivesicular bodies contain vesicular structures, which bleb off from ER cisternae involved in the secretion.
In *Bolarrhena antidysenterica* towards the stage of secretion there is a decrease in the amount of osmiophilic material present in plastids and vacuoles which suggested that probably this material might have been transported from the cell to the outside into the extraplastic space.

In *Plumeria rubra*, the vesicles containing the secretory material seem to be passing through the cell wall into the wall lumen and sub-cuticular space where the secretory material is depositing.

A decrease in the amount of starch in plastids during the stage of secretion is noted in the epithelial cells of the nectaries of both the species. This may perhaps be taken as an indication that the starch accumulated in the secretory cells is a source for some of the secreted material.
DISCUSSION AND CONCLUSIONS

Linnaeus (1735) introduced the term 'nectary' (nectarium). Caspary (1848) divided the nectaries into floral and extrafloral. Delpino (1868-75) considered the two classes of nectaries from the functional point of view and proposed the terms nuptial and extranuptial nectaries. Nuptial nectaries are those occurring within the flowers and are directly associated with pollination, and extranuptial nectaries are those occurring on the outer parts and on vegetative organs and are not directly associated with pollination. Elias and Gelband (1976) used the terms floral and extrafloral nectaries for nuptial and extranuptial nectaries. Fahn (1979) currently considered the same terms floral and extrafloral nectaries proposed by Elias and Gelband (1976).

According to Metcalfe (1938) it is clearly evident that the extrafloral nectaries consist of aggregated glandular trichomes and the hairs occur singly elsewhere on plant surface have a common origin (see Malvaceae; Ragnarsg, 1960; Schnell et al., 1963 and Pyttna, Sterculiaceae; Arbo, 1972).

Schumann (1891) used the term collectors for certain secretory structures on the interpetiolar stipules of Puntara lanceolata, but the same structures as glandular shaggy hairs were described by Solereder (1908). In Solereder's (1908) glossary, the term 'collectors' refers to large hairs
secreting mucilage and found on buds. Similar structures have been described and termed as 'squamellae' (Ramayya and Bahadur, 1968), 'nectarthode' (Lewis, 1968) and 'colleters' (Lersten, 1974a, 1974b). These are glandular multicellular structures consisting of elongated or reduced axis with oval to elongated glandular epidermal cells. They are found on the leaves, stipules and budscales in many families of dicotyledons and secrete various mucilages, gums and resins. Lewis (1968) described the secretory glands which are located in pairs along the adaxial surface of the midrib, singly at the tips and notches of the serrated margins, on secondary veins and fine reticulations as well as at the apices of the stipules. Ramayya and Bir Bahadur (1968) described that the secretory glands are arranged in rows at the base of the adaxial surface of the leaves of Allemanda cathartica and Tabernaemontana divaricata secrete a yellowish-brown resinous substance. Von Faber (1912) described the secretory dendroid trichomes or colleters in Pavetta. Colleters in the Rubiaceae occur exclusively on the adaxial surface or the margin of the stipules bearing finger-like structures with elongate axial cells and a palisade-like epidermis (Lersten, 1974). Lersten and Horner (1967) and Horner and Lersten (1968) in Psychotria basteriophila and Miller et al. (1983) in Psychotria kirkii described secretory dendroid trichomes which occur on the adaxial surface of the stipules in the shoot tips of leaf nodulated
Rubiaceous species and have been reported to be involved in the maintenance of a colony of bacterial endophytes present in the shoot tip throughout the entire life of the host.

In *Holarrhena antidysenterica* and *Plumeria rubra*, the structures are resembling the colleteria, but their occurrence suggest that they are extrafloral nectaries. These nectaries occur adaxially at the junctions between the lamina and the distal end of the petiole in *Holarrhena antidysenterica* and at the base of the petiole on the adaxial surface in *Plumeria rubra*. These structures secrete whitish-brown material. The structure, ontogeny of the nectaries of *Holarrhena antidysenterica* and *Plumeria rubra* as well as the manner of secretion using light and transmission electron microscope has been studied.

Previous investigations showed that the light and ultrastructural characteristics of the secretory cells of various types of floral and extrafloral nectaries were similar (Mabesvari, 1954; Wrischer, 1962; Figier, 1966, 1971; Fahn and Rachmilevitz, 1970; Elias, 1972; Rachmilevitz and Fahn, 1973, 1975; Wargin et al., 1975; Elias and Gelband, 1977, and Durkee, 1982). The structural similarity was evident mainly at the stage of secretion. In all the cases the secretory cells are distinguished by the occurrence of reduced vacuole and increased volume of dense cytoplasm rich in ribosomes, mitochondria and ER elements. At the beginning
of the stage of secretion the ratio of cytoplasmic volume to vacuole was the highest, and the ER occupied most of the cytoplasmic volume which often appeared mainly in the form of densely-packed cisternae. Throughout the stage of secretion, swelling of the rough and lamellar ER followed by a formation of smooth or partly rough vesicles, was noted and were observed in the vicinity of the plasmalemma or attached to it. It was suggested that they may contain sugar solution which would be secreted by means of their fusion with the plasmalemma. Observations made on a variety of nectaries (Schmeplf, 1964a, 1964b; Byrne, 1966a, 1966b; Findley and Mercer, 1971) support the view that the above mentioned ultrastructural features are characteristic of nectar secreting cells in general, and can, therefore, be used for identification of secretory cells of nectaries. On the basis of this view, the ultrastructure of the epithelial cells of the nectaries of Holarrhena antidysenterica and Plumeria rubra indicate that these are secretory in nature.

In Holarrhena antidysenterica, the process of swelling of the ER elements was evident in the epithelial cells at the stage of secretion. Many vesicles, derived from ER, were observed in the vicinity of plasmalemma and closely associated with it. Well developed Golgi bodies were noted in the epithelial cells at the developmental stages preceding to secretion as well as at the stage of secretion. In the secretory cells of the other nectaries studied (Byrne, 1966a,
Golgi bodies were seemingly more active with greater numbers at the developmental stages preceding to secretion than at the stage of secretion. Rachmilevitz and Fahn (1975) observed numerous active Golgi bodies in the secretory cells of the *Tropaeolum majus* at the secretion stage. On the basis of the present observations, it is suggested that the Golgi bodies take part together with ER elements in transport of the secretory material to the extraplasmic space, i.e. by exocytosis.

In *Plumeria rubra*, long profiles of ER cisternae occupy much of the cytoplasm. Some of the rough ER cisternae appear as concentric whorls, which surround the portion of cytoplasm, presumably forming the 'pseudovacuole' at the preceding to secretion stage. Kristen (1977) suggested that there appear to be two types of vacuoles, i.e. pseudovacuoles that are enclosed by rough ER cisternae and which play a role in protein carbohydrate accumulation in placental papillae of *Aptenia cordifolia* and *Platythra hauckeliana*. Kristen (1977) assumes that in contrast to the storage vacuoles the 'pseudovacuoles' play an important role in the storage of protein than polysaccharide. At the stage of secretion many vesicles, mainly originating from ER, appear in the vicinity of plasmalemma, and in the plasmalemma invaginations, whose topography suggest their origin from ER. Some of the vesicles are seen in the middle lamella of cell wall and near the
vicinity of cell wall lumen along the radial walls of the epithelial cells. Rough ER, SER tubules and vesicles derived from the ER are seen in the close proximity of the plasmalemma invaginations and often in close contact with them. These vesicles of ER origin, are probably transporting the secretory material from the cytoplasm to outside the cell by exocytosis. Golgi bodies with peripheral vesicles appear close to the plasmalemma invaginations. It is suggested that at the secretion stage besides the vesiculate ER, Golgi vesicles are probably playing a role in transporting the secretory material by exocytosis. Unselman and Healey (1974) suggested that in the secretory trichomes of *Sparbitis nil*, the coated vesicles and the rough endoplasmic reticulum are active in secretory export.

In *Holarrhena antidysenterica*, the plasmalemma withdraws from the cell wall forming plasmalemma invaginations (extraplastic space). Invaginations of the plasmalemma forming the extraplastic space have been commonly observed in secretory trichomes (Anselumken, 1964; Schmepf, 1965, 1969a; Horner and Lersten, 1968; Perrin, 1970; Wall, 1970; Akers et al., 1978). According to Littge (1971) an enlarged plasmalemma which forms the extraplastic space, is the functional structure in respect to intensive short distance transport and facilitate secretion by allowing intimate contact of the membrane with mitochondria and ER.
The extraplasmic spaces appear to be intercellular channels in transsections of the epithelial cells of the nectary. The epithelial cells are not closely packed, but have small vertical intercellular channels at the junction of three or four adjacent cells in cross-sections. Such channels have also been seen between epidermal cells in the resin-secreting structures of *Roupulus deltoides*. In that species the spaces have been implicated as a pathway for movement of the secretory product to the cuticle (Curtis, 1974).

In *Plumeria rubra* multivesicular structures (parasmural bodies) are seen in the plasmalemma invaginations, along the radial walls where the initial development of the lumen occurs by dissolution of middle lamella of the cell wall. In the cytoplasm Golgi bodies with peripheral in vesicles appear close to the plasmalemma and some vesicles are in contact with the plasmalemma. It is, therefore, possible that vesicles developing from the Golgi bodies carry lytic enzymes for dissolution of the cell wall and thus forming lumen in between adjacent epithelial cells. According to Gilliland et al. (1976) the dictyosome or ER originated vesicles fuse with the plasmalemma to form parasmural bodies. Subsequently, the swelling and dissolution of middle lamella occurs. Fahn and Banayoun (1976) observed many multivesicular structures in the invagination of the plasmalemma outside the cytoplasm, whose vesicles were filled with electron dense
granulated material in the initial stages of dust development. In the vicinity of these multivesicular structures, Golgi bodies and similar vesicles were seen in the cytoplasm which carry lytic enzymes for the dissolution of the middle lamella. According to De Halac (1980) the paramural bodies as well as multivesicular bodies, associated with plasmalemma or free in cytoplasm, may be involved in cell wall synthesis and secretion.

Wollenweber et al. (1971) studied the ultrastructural changes occurring in the cells of the bud glandular trichomes of some Alnus species and observed the sub-cuticular spaces in the regions above the anticlinal walls, where the cuticle together with the cuticular layer splits from the pecto-cellulosic cell wall at the stage of secretion. During the course of secretion the secretory substance accumulates in the sub-cuticular space. In many instances secreted fluid accumulates at the surface of gland cells between the cell wall and cuticle and eventually the cuticle breaks liberating the fluid (Lüttge, 1971). Thin cuticle which covers the walls of the protoderm of the young developing stipules is being stretched and lifted off the collateral cell walls by the pressure of the heterogenous material being stretched through the cell wall (Miller et al., 1983). Shimony et al. (1973) observed an amorphous substance, apparently the polysaccharides between the cuticle and cell walls in the upper region of the secretory cells. The cuticle is already detached even in relatively young glands.
In *Holarrhena antidysenterica* the cuticle together with the cuticular layer overlying the outer tangential wall of the epithelial cell splits from the pecto-cellulosic cell wall forming the sub-cuticular space filled with secretory material at the stage of secretion. During the course of secretion the volume of the secretory material increases and probably the cuticle is burst off due to the pressure exerted by the secretory material. In *Plumeria rubra*, the sub-cuticular space is formed by the splitting of the cuticle together with cuticular layer from the outer tangential wall of the epithelial cell forming the sub-cuticular space at the developmental stages preceding to secretion. At the secretion stage the secretory material started accumulating in the sub-cuticular space and the cuticle lifts off from the cell wall. At later stages, the cuticle bursts off due to the pressure exerted by the secretory material.

Numerous plasmodesmata were found to connect secretory or excretory cells of glands with each other (Lütge and Kräpf, 1969), as well as the gland cells with the surrounding parenchyma (Ziegler and Lütge, 1966; Thomson et al., 1969). Numerous plasmodesmata were observed connecting the protoplasts of the cells of the nectariferous tissue (Fahn and Rachmilevitz, 1970; Findley and Mercer, 1971; Wergin et al., 1975; Gunning and Hughes, 1976). Both in *Holarrhena antidysenterica* and *Plumeria rubra* nectaries the pre-secretory
material is transported mainly in the symplast. Plasmodesmata have been observed connecting the protoplasts of the adjacent epithelial cells and adjacent sub-epithelial and epithelial cells. Here the pre-nectar flows through the plasmodesmatal pathway.

According to Lyshede (1980) mitochondria with swollen tubules probably indicate high rate of respiration. It is worth mentioning that these cells are believed to be heavily involved in transport processes (Hansen and Day, 1980). At the secretion stage and pre-secretion stage many mitochondria are observed in the epithelial cells of the nectaries of Holarrhena antidysenterica and Plumeria rubra. It may be possible that high energy level required for the synthesis and transport of the secretory material is derived from abundant mitochondria present in the epithelial cells at the stage of secretion.

Plastids are most often considered to produce lipophilic substances (Wooding and Northcote, 1965; Amelunxen and Arbeiter, 1967; Amelunxen and Gronau, 1969; Heinrich, 1970; Vassilyev, 1970; Fahn and Benyoum, 1976; Deil and McComb, 1977; Akers et al., 1978; Wörner and Fahn, 1981). In Holarrhena, osmiophilic material occurs in the plastids, surrounding the thylakoids and starch grains in the young stage of the epithelial cells. The osmiophilic material diminishes towards the stage of secretion. Probably
this osmiophilic material is transported from the plastids to the extraplasmic space where the secretory material is deposited. Werker and Fahn (1981) suggested that the chloroplasts of the photosynthesizing cells in *Impala viscosa*, peripheral reticulum might be functioning in transfer of the osmiophilic material to the chloroplast envelope; it is transferred from there to ER which has connections with the plastid envelope and then through the ER to the plasmalemma.

Amelunxen (1965) mentioned the possibility of the essential oil being synthesized in the ground cytoplasm. As the osmiophily of the vacuolar content diminishes concurrently with the development of sub-cuticular space it is possible that the vacuolar content is also associated with oil production. Wollenweber et al. (1971) observed strong osmiophilic material inside the large irregularly shaped vacuoles in the cells of the bud glandular trichomes of some *Alnus* species. In *Epilobium*, the vacuoles containing electron dense material at the pre-secretion stage and the latter diminishes towards the stage of secretion. At the stage of secretion the secretory material is deposited in the extraplasmic space along the radial walls and the sub-cuticular space. It seems that the osmiophily of the vacuolar content diminishes towards the stage of secretion. Probably this osmiophilic material might have been transported
from the vacuoles to the plasmalemma and finally accumulates in the extraplasmonic space where the secretory material is deposited.

Due to variety of chemical compounds produced and the numerous ultrastructural changes occurring during secretion, identification by cytochemical methods with the electron microscope of the specific cell compartments involved in the production of each compound in the epithelial cells has not been attempted.

On the basis of the present observations in Holarrhena and Plumeria, it is suggested that the lipids secreted from the epithelial cells are produced by tubular ER and plastids. Tubular ER was suggested to be involved in the biosynthesis of terpenes in various secretory hairs (Amelunxen, 1965; Amelunxen and Arbeiter, 1969; Schnepf, 1969a, 1969b, 1969c, 1969d, 1972; Schnepf and Klasova, 1972; Heinrich, 1973; More and Mollenhauer, 1974; Gunning and Steer, 1975; Werker and Fahn, 1981). In addition to ER, plastids are most often considered to produce lipophilic substances (Wooding and Northcote, 1965; Amelunxen and Arbeiter, 1967; Amelunxen and Gronau, 1969; Heinrich, 1970; Vassilyev, 1970; Fahn and Bojesen, 1976; Dell and McComb, 1977; Akers et al., 1978; Werker and Fahn, 1981).
Golgi bodies are reported to produce both acid and neutral polysaccharides (Gunning and Steer, 1975; Fahn, 1979) in mucilage secreting hairs of carnivorous plants (Schnepf, 1960, 1961, 1974) and in mucilage ducts (Bouchet and Deyson, 1974). Involvement of ER in polysaccharide production has been suggested for some secretory structures (Horner and Larsen, 1968; Bouchet and Deyson, 1974; Werker and Kislev, 1978). On the basis of present observations, it is concluded that the polysaccharides are apparently produced by Golgi bodies.

The RER was described commonly to the bulk production of protein (Gunning and Steer, 1975; Werker and Vaughan, 1976). In Helianthus and Plumeria, RER is found in the epithelial cells at the preceding stage of secretion as well as at the stage of secretion. But in Plumeria, RER is in smaller amounts than SER.

Elimination of the secretory material from the secretory cells to the outside of the nectary:

Schnepf (1964, 1969) studied the septal nectaries of Gasteria and some other Liliaceae and suggested that nectar secretion may be based on molecular processes at the plasmalemma. To support this view he found evidence, in the very great enlargement of the plasmalemma, as a result of development of wall protuberances towards the stage of
nectar secretion. Reed et al. (1971) discussed the possibility of active transport of sugars within the nectary of *Abutilon*.

Fahn and Rachmilevitz (1970, 1975) and Rachmilevitz and Fahn (1973) suggested that nectar accumulated in the ER of secretory cells of *Lonicera* and *Vinea*, is transported to the plasmalemma by vesicles of ER origin and is eliminated from the cytoplasm by fusion of the membranes of the vesicles with the plasmalemma. This view was supported by Belin-Depoux and Clair-Massulajtys (1975) who studied the extrafacial nectaries of *Almurites moluccana*.

Observations were made on the floral nectaries of *Diplodaria* and *Helleborus* (Eyne, 1966), *Cynoglossum officinale* (Tacina, 1973), extrafacial nectaries of *Vicia faba* (Figier, 1966, 1971) and in the septal nectaries of some *Bromeliaceae* (Bacus and Schnepf, 1975). These authors expressed a similar view, but attributed the function of elimination to vesicles of Golgi bodies.

In *Tropaeolum* and *Musa*, Rachmilevitz and Fahn (1975) and Fahn and Benouaishe (1979) respectively observed that active Golgi bodies occur in addition to the very well developed ER at the stage of nectar secretion and thus suggested that in the nectaries of these plants both the ER and the Golgi apparatus may take part in nectar secretion.
Heinrich (1975a) localized different phosphatases in the secretory epithelial cells of septal nectaries of *Aloe* and found high activity of ATPase, nucleoside diphosphatase and glucose 6-phosphatase in the ER of these cells and frequent absence of activity in their plasmalemma and hence suggested that ER plays an important role in nectar transport. On the basis of labelling experiments in *Aloe* nectaries, Heinrich (1975b) suggested that the nectar is secreted by vesicles, but was not sure whether they were ER vesicles or Golgi vesicles.

At the secretion stage, the autoradiographs of secretory cells of nectaries of *Lonicera*, fed with tritiated sucrose show the most evident labelling in the ER and vesicles of ER origin (Fahn and Rachmilevitz, 1975).

The present observations suggest that in *Holarrhena*, the secretory material is eliminated from the cytoplasm of the epithelial cells to extraplasmic and sub- cuticular spaces by the vesicles of ER origin and Golgi bodies and in *Plumeria*, the secretory material accumulates in the ER and is transported from the cytoplasm by vesicles of ER origin to the cell wall lumen and sub- cuticular space. Both the cases support the view that at least nectar is eliminated from the cytoplasm of nectar secreting cells by vesicles which fuse with the plasmalemma originating either from ER or Golgi bodies or from both.
Fahn and Rachmilevitz (1970) and Rachmilevitz and
Fahn (1973) observed many multivesicular structures between
the plasmalemma and cell wall in Lonicera and Vinea. These
multivesicular structures are participating in the
pre-nectar transport. Kalman and Gulyas (1974) suggested
that in the extrafloral nectaries of Riginae communis the
fluid is actually transported by multivesicular structures.
Belin-Depoux and Clair-Macxulajtys (1975) concluded that in
addition to symplastic transport, apoplastic transport takes
place by the presence of multivesicular structures in
petiolar nectaries of Alyssites. On the basis of present
observations, it is suggested that in addition to symplastic
transport, apoplastic transport takes place in the extra-
floral nectaries of Holarrhena and Plumeria.