RESULTS AND OBSERVATIONS

3 RESULTS AND OBSERVATIONS

3.1 Gmelina arborea Roxb. (Verbenaceae)

Section I

3.1.1 General morphology

Gmelina arborea Roxb. is an unarmed, moderate sized deciduous tree commonly known as 'Malay bush beech'. In Gujarat it is known as 'Sivan' (fig. 1).

The branchlets and young parts are clothed with fine white pubescence. The leaves are simple, petiolate, broadly-ovate, acuminate, entire and glabrous above when mature. The leaves have a cordate or sometimes truncate base and shortly cuneate apex. The petiole is 10-20 cm long, cylindric and glandular at the top (Cooke, 1905; Shah, 1978).

According to the classification of Longmann and Jenik (1974), considering leaf retention and shedding in tropical trees, this deciduous tree shows periodic growth. The shedding of leaves occurs much before the bud opening. The life span of the leaves is about 6-7 months. New leaves develop in the summer months of May-June and defoliation begins in the next winter months of January-February. For the period of March-April the tree is without any leaves. Flowers appear in this period or sometimes along with the development of young leaves.

3.1.2 Sequential elongation of the petiole

Generally the length of a mature petiole in Gmelina arborea varies from 10-20 cm. But among the 20 labelled young leaves of 1.5 cm long petiole, the maximum length observed in the petioles of the mature leaves was 14.4 cm, so this measurement of the longest petiole has been considered typical in this study. The pattern of elongation is uniform in all the petioles studied.

The petiole ceased elongation within one month after the date of labelling. The elongation of the basal, middle and distal region ceased almost at the same time (fig.3). The elongation of the middle region is more than that of the other two regions. The basal and distal regions almost elongate equally.

A petiole is considered young from the time it could be clearly distinguished from the lamina upto the cessation of its elongation. The young petioles of 0.3, 0.6, 2.5 and 7 cm long are respectively considered as y_1 to y_4. A fully elongated petiole is considered as mature stage I. The leaves collected after each subsequent month of mature stage I are identified as mature stage II to VI based on their life span of 6-7 months. The various aspects of general vasculature and developmental and structural phases of procambium, cambium and phloem investigated in the petiole are based on detailed studies undertaken in these stages of the petiole.

3.1.3 Internode - node - petiole vascular continuum

This part deals with the following aspects of the leaf trace system, which is a developmental and functional continuum between internode, node and the leaf,

(i) nodal anatomy

(ii) nodal vasculature

(iii) petiole vasculature

(iv) leaf architecture

The growing shoot of Gmelina arborea produces petiolate leaves in an opposite, decussate manner (fig. 2), each pair separated at maturity by a clearly defined internode. A young stem is rectangular and the young leaves have conduplicate ptyxis. Because of the discreteness of the vascular system at the fourth node from the shoot apex the leaf trace petiolar vascular continuum has been studied in this node. At this node though the petiolar part is distinct from the lamina, the junction of the lamina and the petiole is not yet fully distinguished.

3.1.3.1 Nodal anatomy

The internodal primary vascular system consists of about 50 vascular strands peripherally arranged in a rectangular manner. They are either collateral or phloic. The vascular region of the internode associated with the formation of the leaf trace consists of five discrete strands (fig.4) of which

(i) 3 large and 2 small are collaterals (fig.5) or (ii) 2 large and 2 small are collaterals with one phloic strand (fig.6) or (iii) 4 large are collaterals with phloic strand (fig.7). Of these strands especially the large ones in many cases show their composite structure indicating gradual approximation and amalgamation of two or more strands. Figure 4 depicts the internode - node - petiole continuum in one plane.

3.1.3.2 Nodal vasculature

Further in the node, these five discrete strands A, B, C, D, E (figs. 5 - 7) through a series of approximations and amalgamations form two composite strands ABC and DE (fig. 8). Further, they form a single composite leaf trace ABCDE (fig. 9) with a single gap. Thus the leaf trace in Gmelina arborea is unilacunar with a composite strand (fig. 4) made up of five vascular strands.

The leaf trace subsequently trifurcates into a median strand M and two laterals L and R (fig.4). They are organized in the form of a shallow arc (fig. 10). The median strand M lies in the centre of the arc and the two laterals L and R are on either side of the median strand.

3.1.3.2.1 Median strand

The median strand M is large and collateral and it remains mostly discrete upto the petiole base. But it gives rise to a single phloic strand M2 on the adaxial

side of the petiole (fig.4).

3.1.3.2.2 Lateral strands

During their traverse along with the median, they frequently divide. A large collateral may bifurcate into two or more small collaterals or may give rise to a collateral and one or more phloic strands. The laterals on either side of the median divide almost equally giving rise to 8 strands. At a distance of about 40-90 um from the locus of formation of the unilacunar leaf trace the corticals are formed from the lateral strands (fig.4.).

3.1.3.2.3 Cortical strands

They are formed before the separation of the petiole from the stem axis. In the petiole they are located on either side of its adaxial groove. The two cortical strands do not develop at the same level.

At a distance of 40 um from the locus of formation of the unilacunar leaf trace, the large collateral R4 bifurcates giving rise to R5 and R6 (fig. 4). The collateral R6 on the abaxial side near R2 at the open end of the crescent traverse away from the main vascular ring into the cortex (figs.11-13). Further at a distance of 90 um from the locus of formation of the single leaf trace the collateral L5 bifurcates to form two collaterals L7 and L8. The L7 lying close to L3 separates from the main vascular ring and becomes

the cortical strand of the other side (fig. 4).

At a distance of 240 um from the locus of formation of the unilacunar leaf trace the petiole vascular system alongwith the axillary bud vasculature diverges from the main axis and the vascular strands are organized and distinctly separate from the vascular system of the internode.

3.1.3.3 Petiole vasculature

The young petiole in transection is almost circular with an adaxial groove. Throughout the petiole the vascular strands are discrete. The vascular system includes large and small collateral and phloic strands. The median located on the abaxial side of the petiole is the largest. It remains mostly discrete from the basal to the distal region on the petiole. But it does give rise to a few vascular strands. They are M1, M3, M4 towards the adaxial side. The median strand therefore is not much involved in subsequent additional vascularization of the petiole (fig.4). The laterals frequently divide and involve in the subsequent development of the vascularization of the petiole from its base onwards.

3.1.3.3.1 Arrangement of the vascular strands

The vascular strands in the basal region of the young petiole are arranged in a crescent with incurved open ends separated by a large interfascicular parenchymatous region. The bundles are widely spaced with the large median strand in

the centre of the crescent on the abaxial side of the petiole and the laterals adaxial to it.

In the middle and the distal region of the petiole the vascular strands form a horse-shoe pattern. Two cortical strands one on either side of the groove extend from the base to the distal region of the petiole (fig.14).

3.1.3.3.2 Number and size of the vascular strands

In the basal region of the petiole there are 9 collateral and 8 phloic strands which develop into 9 collateral and 11 phloic ones in the middle region and 13 collateral and 19 phloic ones in the distal region of the petiole. As figure 4 indicates the vascular system thus developed in the petiole is the result of bifurcations, approximations and amalgams of the different vascular strands in their varying combinations. The number of strands increases from the basal to the distal region in the petiole (fig.15).

At the extreme distal end of the petiole, the median along with the two adaxially placed collaterals separate from the vascular ring and traverse into the midvein. The rest of the laterals on either side of median arch out and separate as the basal secondary veins. The corticals extend along the margin of the lamina.

As the median is discrete in the entire petiole its area

of xylem and phloem tissues were calculated in the three different regions of the petiole. Its area decreases from the base upwards (fig.16). The procambial tissue is considered as a part of the phloem region.

3.1.4 Young petiole vasculature

To study the young petiole vascualture the leaf at the third visible node from the shoot tip, with a 2.5 cm long petiole was used.

3.1.4.1 Arrangement of vascular strands

In transection the petiole appears circular with an adaxial groove. For the convenience of the study, the petiolar region has been equally divided into the basal, middle and distal regions. In the basal region the vascular system forms an open ring with a conspicuous adaxial parenchymatous region (fig 17). Towards the middle region they form a closed ring (fig.18). The vascular strands on the adaxial side, located at the extreme end of open vascular ring in the petiole base converge towards one another during their course of traverse into the middle region of the petiole and occupy the open part of the vascular system. Thus the open ring form of vascular system in the basal region of the petiole gradually becomes closed towards the middle region. This arrangement of vascular strands continues in the distal region of the petiole (fig.19).

The vascular system includes discrete collateral and

phloic strands. The latter are randomly distributed among the collaterals. One or two layers of parenchyma cells containing starch grains ensheath the vascular system (fig.20). Parenchyma cells flanking the phloem of the cortical strands also contain starch grains.

3.1.4.2 Number and size of the vascular strands

The number of collateral and phloic strands in the three different regions of the petiole varies. There are 12 large and small collateral and 15 phloic strands in the basal region, but there are 2 more phloic strands in the middle region and in the distal region 11 collateral and 14 phloic strands are present. The median collateral is the largest (fig.24) and remains mostly discrete throughout. As previously mentioned the area of the median strand decreases from the base upwards.

3.1.4.3 Cortical strands

In the basal and distal regions there is a single collateral cortical strand on either side of the adaxial groove (figs. 21 and 23), but in the middle region it forms a collateral and phloic strand (fig.22).

3.1.5 Vascular system in the mature petiole

Two petioles, one 14.4 cm long which ceased elongation and second 17 cm long collected one month after the cessation of elongation have been studied. They are represented here as

MI and MII respectively.

3.1.5.1 Nodal anatomy

A mature node is also rectangular. The trace strands have been identified by their primary xylem groups which project into the pith. The vascular region of the internode associated with the formation of the leaf trace consists of four strands A, B, C and D (fig.25).

3.1.5.2 Nodal vasculature

Further in the node, these four strands through approximation and amalgamation form two composite strands AB and CD (figs. 26-28). Further they form a single strand ABCD (fig. 29). The leaf trace trifucates into a median M and two laterals L and R (fig. 31). The median strand is formed first (fig. 30). Further up the strands divide and form petiolar vascular system (figs. 32 and 33). Cortical strands are also formed (fig. 33).

3.1.5.3 Petiole vasculature

Transection of the petiole shows a single layer of epidermis with tabular or barrel shaped cells. The hypodermis consists of 3-4 layers of collenchyma cells. The pith and the cortex are delimited by collateral and phloic strands which extend along the entire petiole.

3.1.5.3.1 Arrangement of the vascular strands

The vascular strands are discrete in the basal and distal regions of the petiole and its arrangement remains similar to that in a 2.5 cm long petiole. In the middle region of the petiole due to the lignification of the cells within and between the adjacent strands, they do not appear discrete (fig. 34) and form a cylinder with an adaxial opening of parenchyma tissue.

3.1.5.3.2 Number of the vascular strands

In MI the basal region shows 26 vascular strands, 14 collateral and 12 phloic strands which organize into 18 collateral and 12 phloic ones in the middle region and 16 collaterals and 12 phloic strands in the distal region.

In MII there are 40 vascular strands in the basal region which includes 17 collaterals and 23 phloic strands. In the middle region there are 15 collaterals and 18 phloic strands and in the distal region there are 14 collaterals and 10 phloic strands.

It may be mentioned here that the number of vascular strands in the basal, middle and distal regions may vary in different petioles but the range of variations is not significant.

3.1.5.3.3 Cortical strands

In MI at the base, the cortex on one side shows 2

collaterals 'a' and 'c' and a phloic strand 'b' (fig. 35) and on the other side there is a single collateral (fig. 36). Towards the middle region the three strands organize into a single collateral (fig. 37). In the distal end of the petiole there are two collaterals 'a' and 'b' (fig. 38).

In MII there is a collateral 'a' and pholic strand 'b' on one side of the cortex (fig. 39) and a collateral (fig.40) on the other side of the cortex in the basal region. In the middle region there is a collateral 'a' and a small group of protophloem fibres on one side (fig. 41) and two collaterals 'a' and 'b' on the other side of the cortex (fig. 42). And in the distal region 3 collaterals 'a', 'b' and 'c' on one side of the cortex (fig. 43) and on the other side there is a collateral strand (fig. 44).

The number of cortical strands, like that of the vascular strands varies in different petioles in the three different regions but the range of variations is not significant.

3.1.4 Leaf architecture

The basic axis of orientation in the leaf is towards the base (downward) termed as basal and the curvature of the leaf elements is convex and curved away from the centre of the axis. The organization of the leaf is simple consisting of a single lamina. Generally the lamina and the leaf base are symmetrical but also sometimes asymmetric. The leaf is wide

ovate (fig. 45). The apex is short, acuminate with the tip acute and margins markedly concave (fig. 45). The leaf base is acute and decurrent with the margin extending downward along the petiole at a gradually decreasing angle. Generally the leaf margin is entire and smooth without noticeable projections. But in some leaves margin shows one or two dentations. The texture of the leaf is chartaceous. Basilaminar nectaries are present. The petiole is inflated with a pulvinus base.

3.1.4.1 Secondary veins

The venation is pinnate with a single primary vein (mid vein) serving as an origin for the higher order venation (fig. 45). It is the camptodromous type of venation where the secondary veins do not terminate at the margin of the leaf but join together in a series of prominent arches (fig. 46) and so classified as brochidodromous. The primary vein is moderate. The course of vein is straight lacking any noticeable curvature or change.

The angle of divergence of secondary veins measured between the branch and the continuation of the source vein above the point of branching is acute and moderate. This is nearly uniform for all secondaries. The course of secondary veins is curved bending in an arc. It is uniformly curved with the arc gradually increasing in its degree of curvature. The lowermost secondary veins branch out on the exmedial

side. The secondary veins at the margin loop joining the super adjacent secondary veins at an acute angle enclosed by 3o arches (fig. 46).

3.1.4.2 Tertiary veins

The tertiary vein originates on the admedial and the exmedial side of the secondaries and are at right angles. These tertiary veins at times anastomose with other tertiary or secondary veins, the pattern of anastomosing being random reticulate (fig. 47). They are percurrent, i.e., tertiaries from the opposite secondaries are joining. The course of tertiary veins shows variations from simple unbranched to forked nature giving rise to third order ramifications. It may be convex with the middle portion of the vein curving away from the centre of the leaf. Its relationship to the mid vein is noticed to be oblique tending in an obtuse angle. This relationship remains approximately constant for all the tertiary veins.

3.1.4.3 Quarternary and quinternary veins

In the higher order venation the vein orders are distinct. The quarternary veins are thick and wide and their course is relatively randomly oriented. The quinternary veins are thin with a random course.

3.1.4.4 Higher order venation

The marginal ultimate venation is incomplete with some

freely ending veinlets directly adjacent to the margin (fig. 48) and others looped, i.e., the major portion of it is recurved to form loops.

The veinlets are branched (fig. 47) giving rise to ramification by dichotomizing twice or thrice. At times they do not dichotomize but remain simple, curved and unbranched (fig. 47). The areoles are imperfect with meshes of irregular shape. They show random arrangement with no preferred orientation (fig. 47).

Plate 1

Fig. 1. Gmelina arborea in the month of September.

Fig. 2. A twig showing opposite, decussate arrangement of leaves.


1


2

Fig. 3.

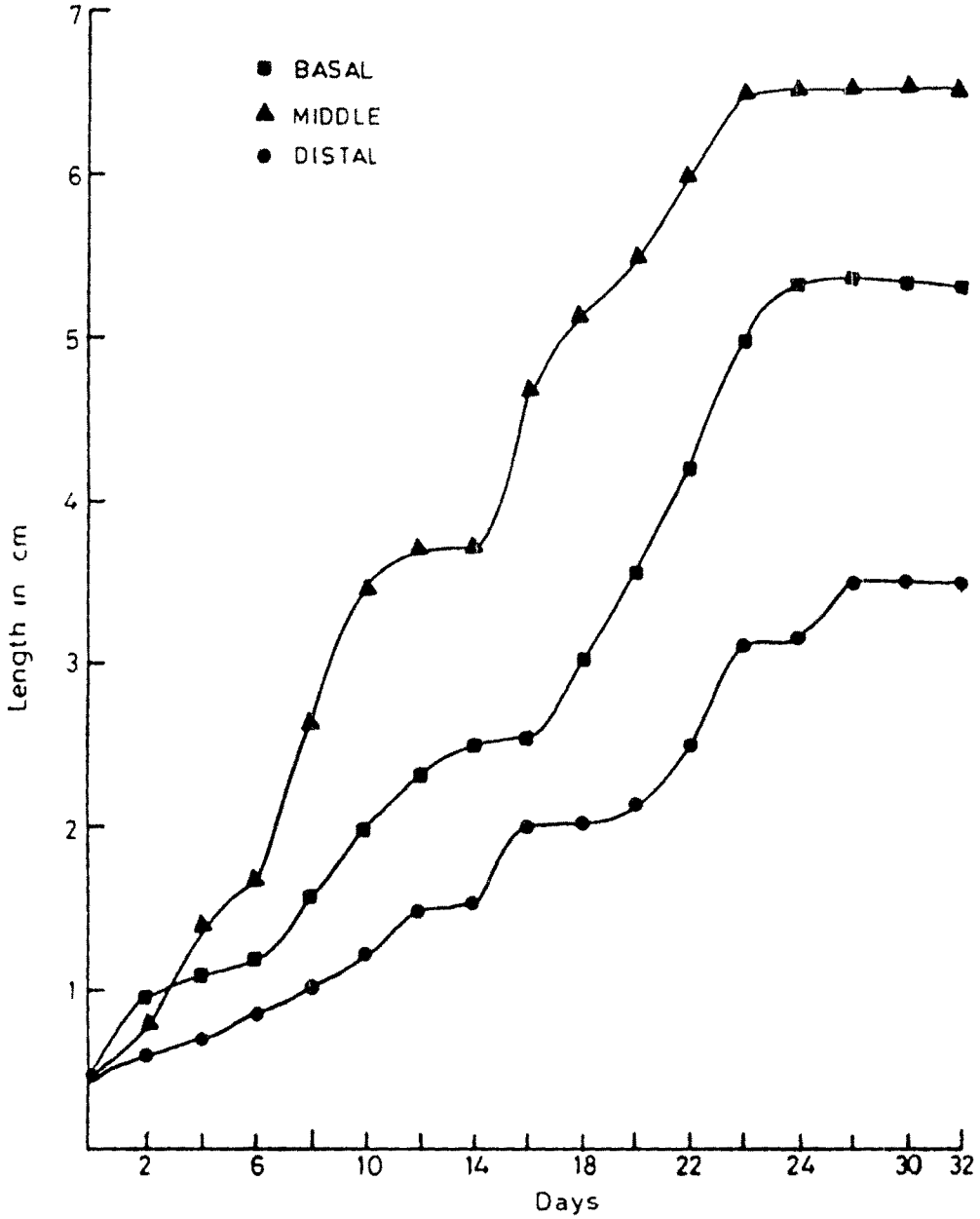
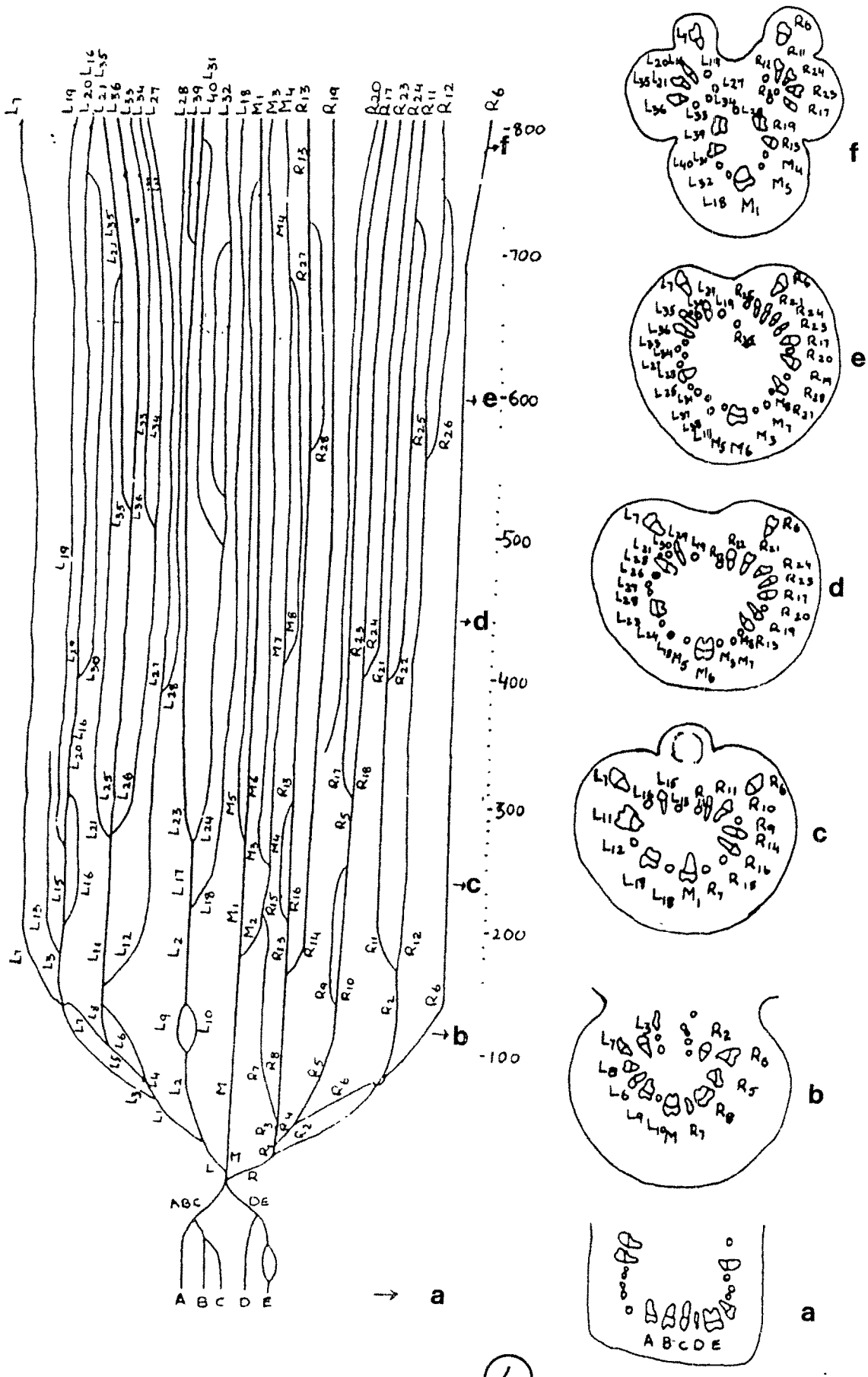

BASAL
MIDDLE
DISTAL
Length in cm
1
2
3
4
5
6
7
2
6
10
14
18
22
24
30
32
Days

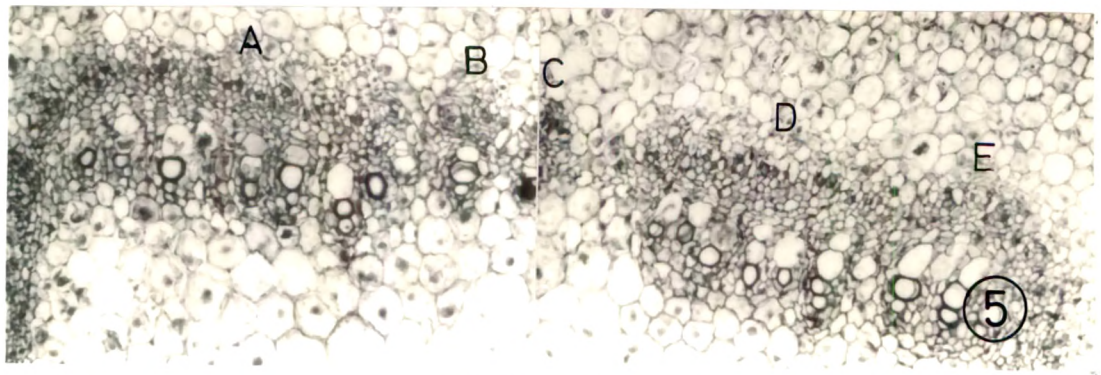
Fig. 4.

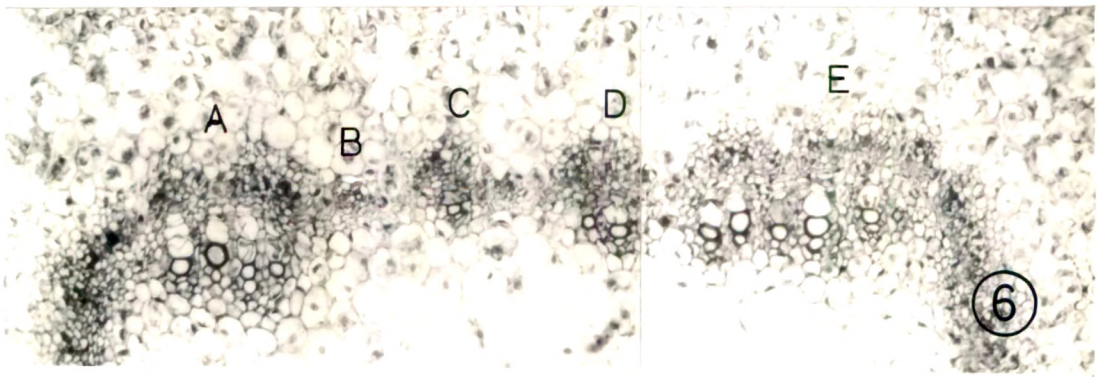

-800
-700
-600
-500
-400
-300
-200
-100
f
e
d
c
b
a
ABC
DE
A B C D E
f
e
d
c
b
a
A B C D E

4

Plate 2

Figs. 5 - 8.


A
B
C
D
E
5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B
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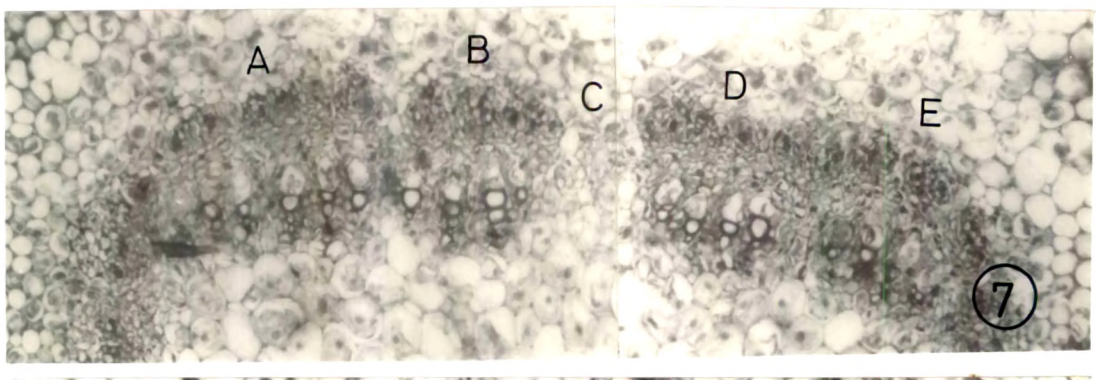
6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D
E
7

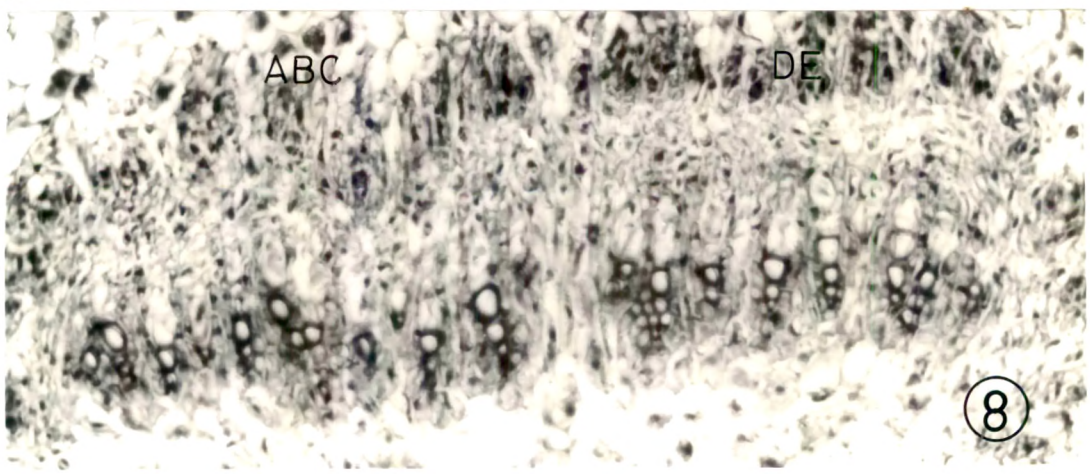
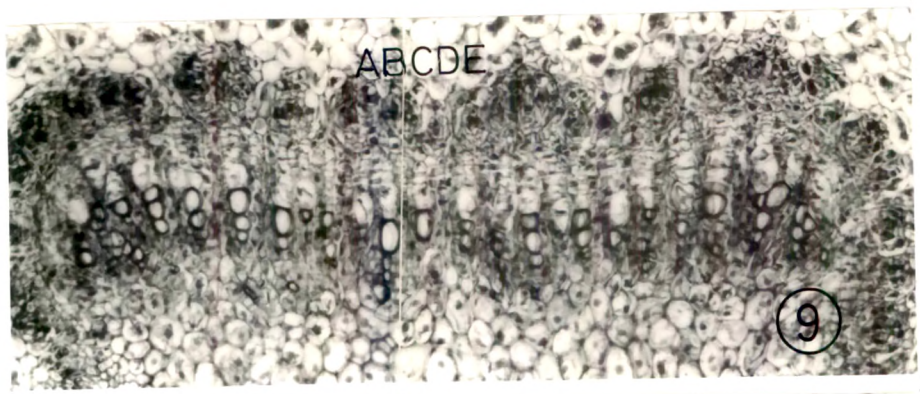

ABC
DE
8

Plate 3

Figs. 9 - 14.


ABCDE
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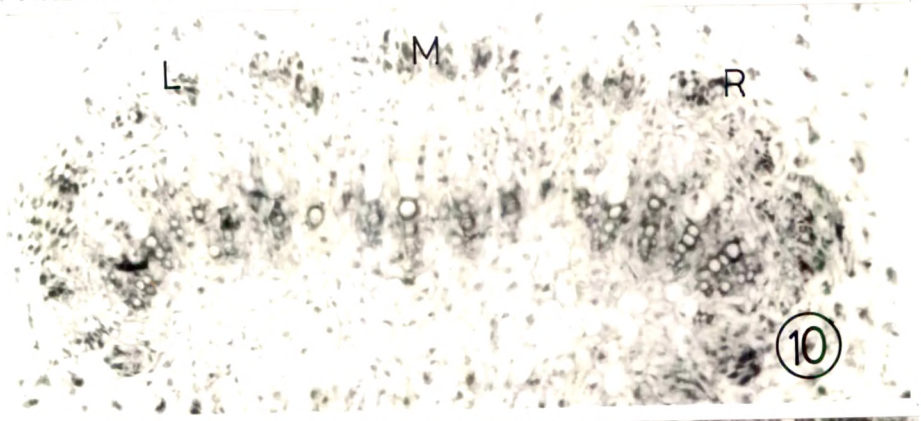
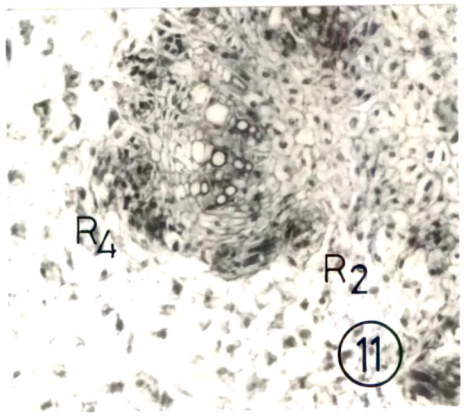
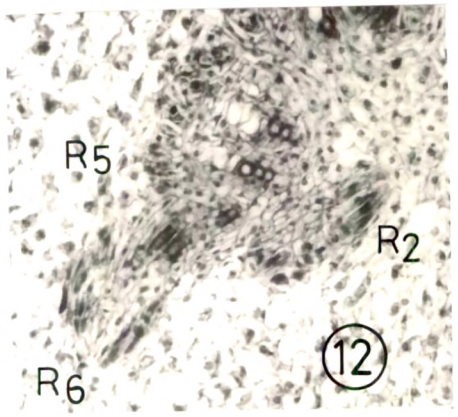
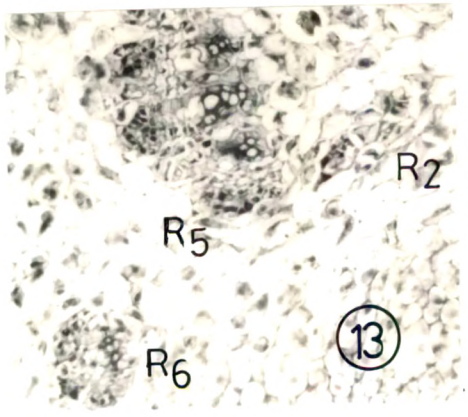

L
M
R
10


R4
R2
11


R5
R2
R6
12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R5
R6
13

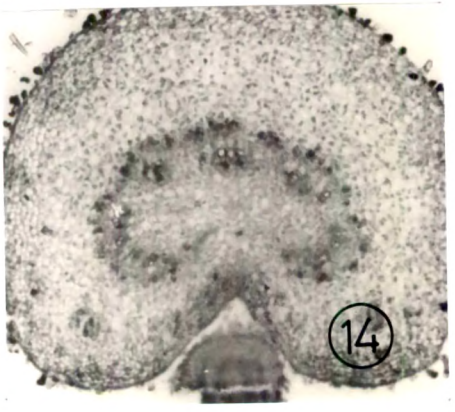
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Fig. 15.

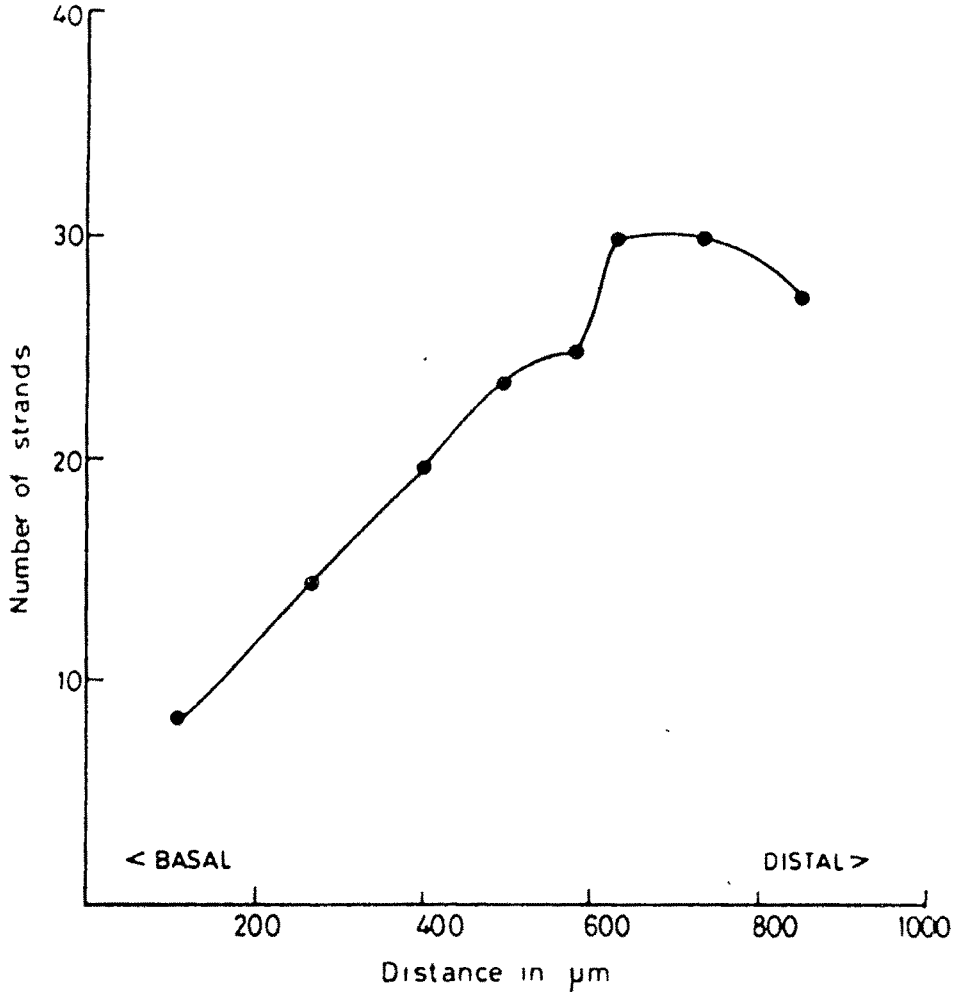

40
30
20
10
Number of strands
< BASAL
DISTAL >
200
400
600
800
1000
Distance in µm

Fig. 16.

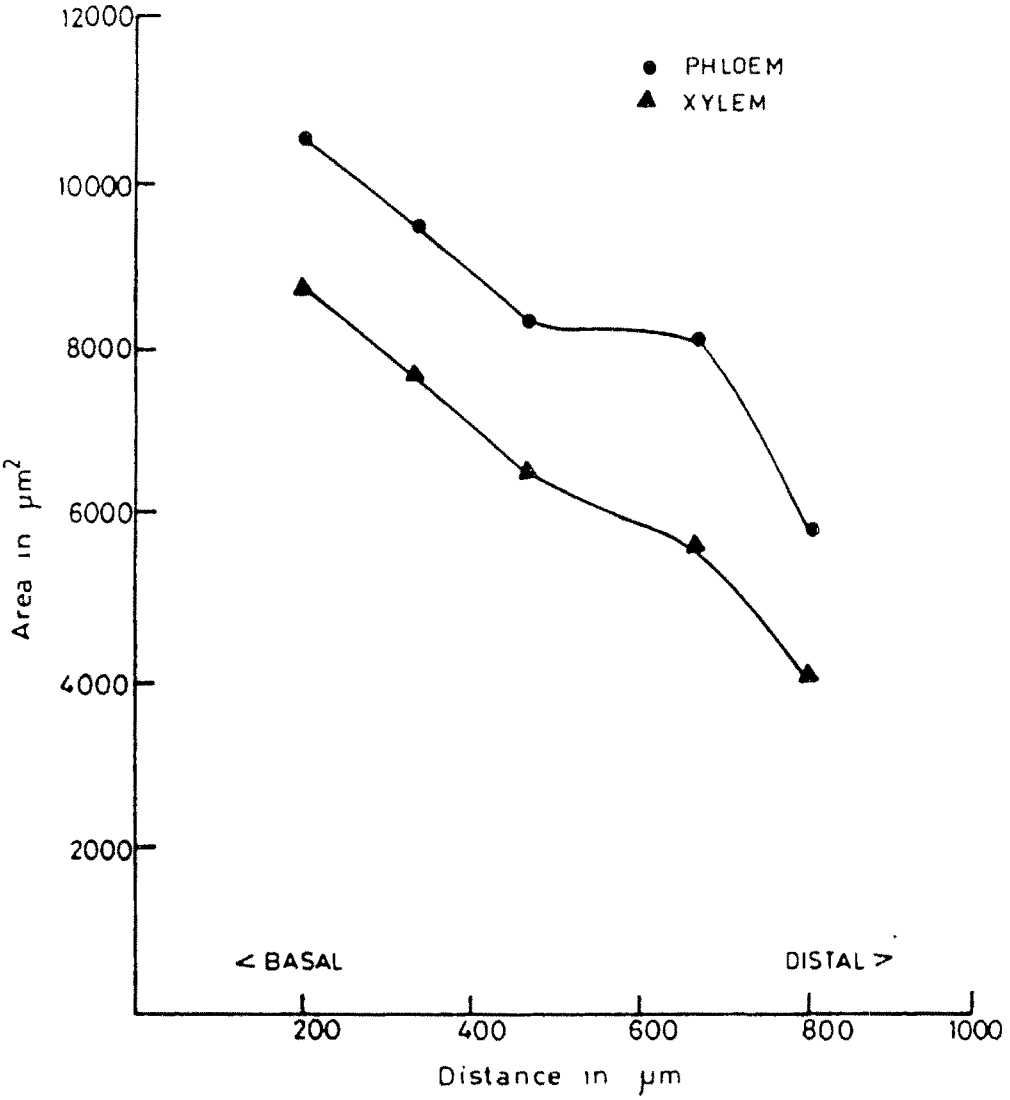
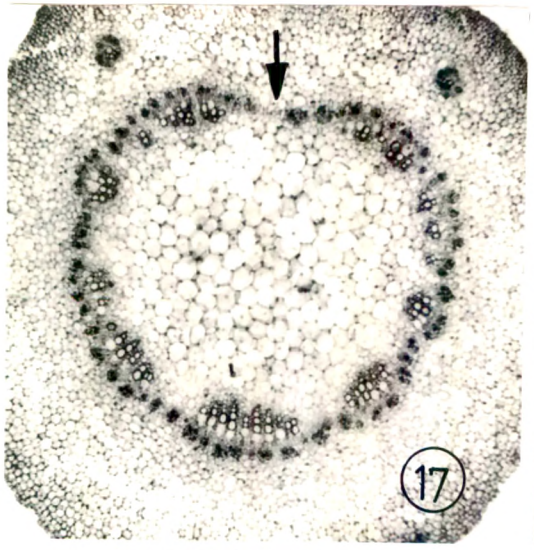

12000
10000
8000
6000
4000
2000
Area in μm^2
PHLOEM
XYLEM
< BASAL
DISTAL >
200
400
600
800
1000
Distance in μm

Plate 4

Figs. 17 - 24.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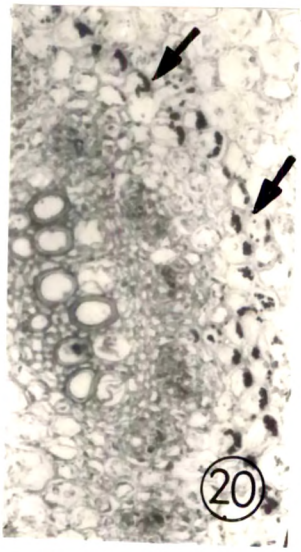
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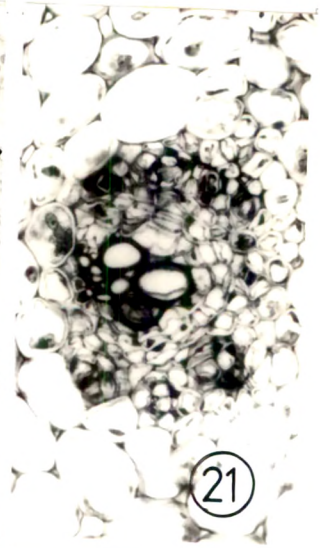
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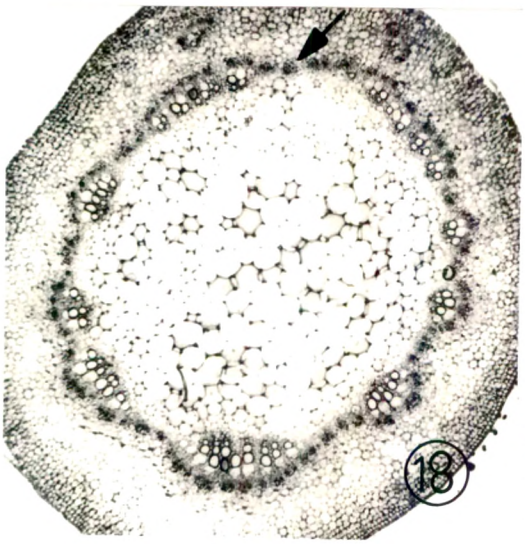
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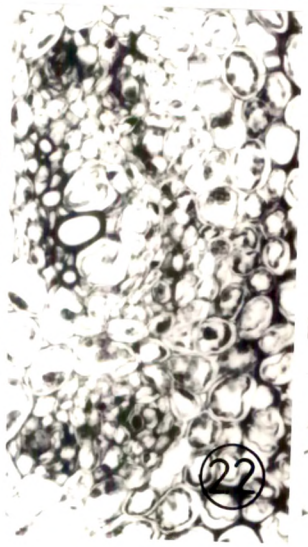
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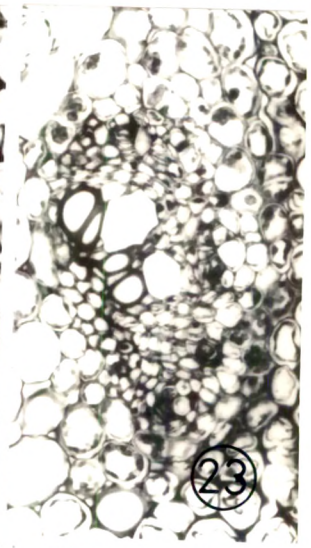
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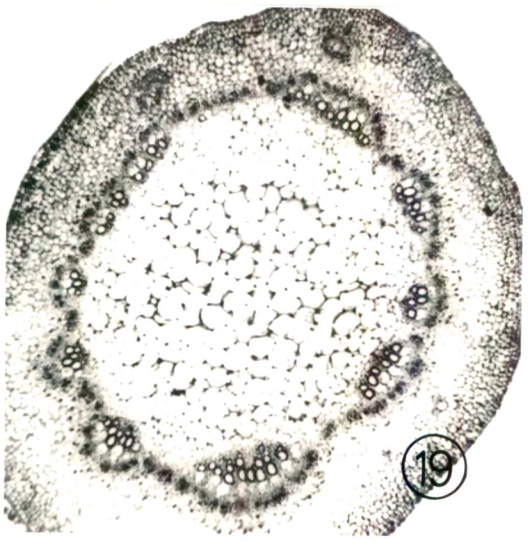
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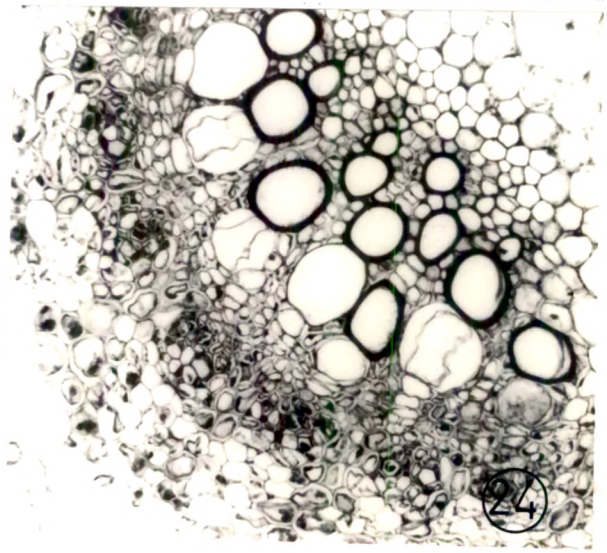
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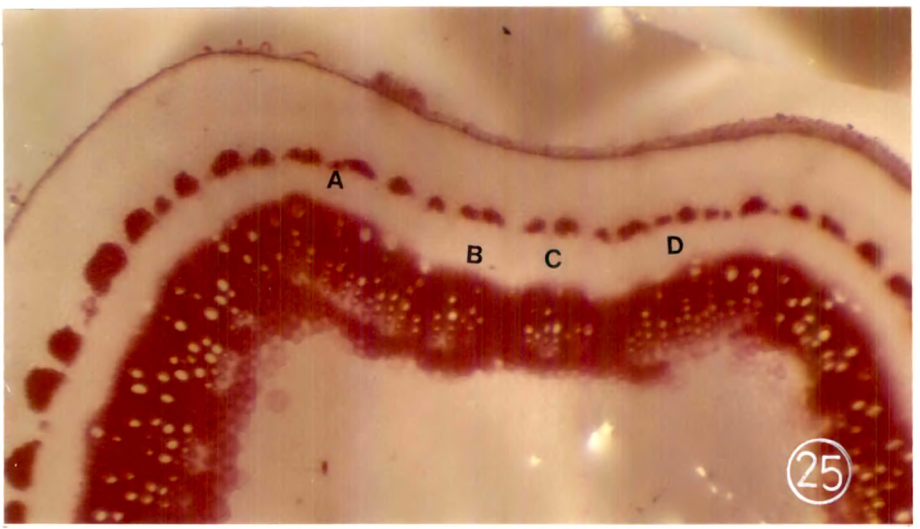
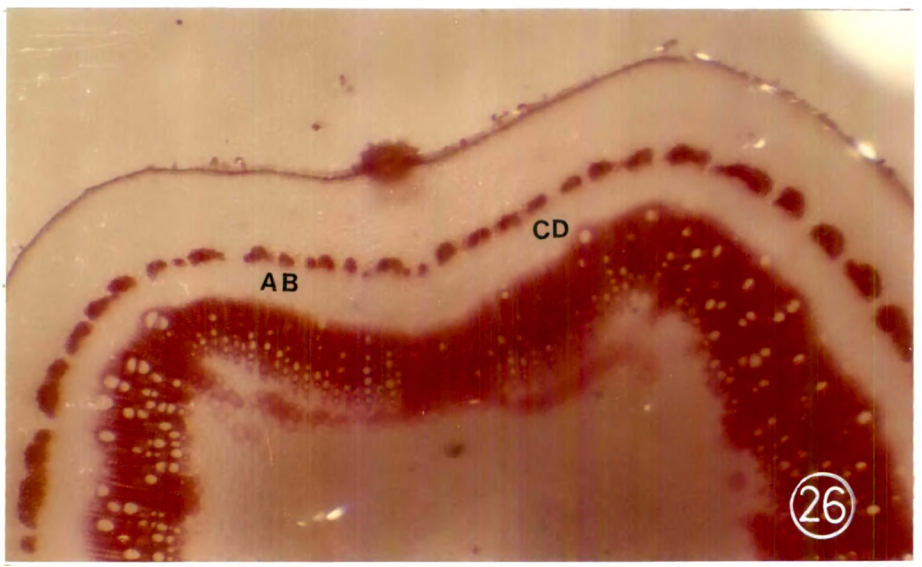
Plate 5

Figs. 25 - 27.


A
B
C
D
25


CD
AB
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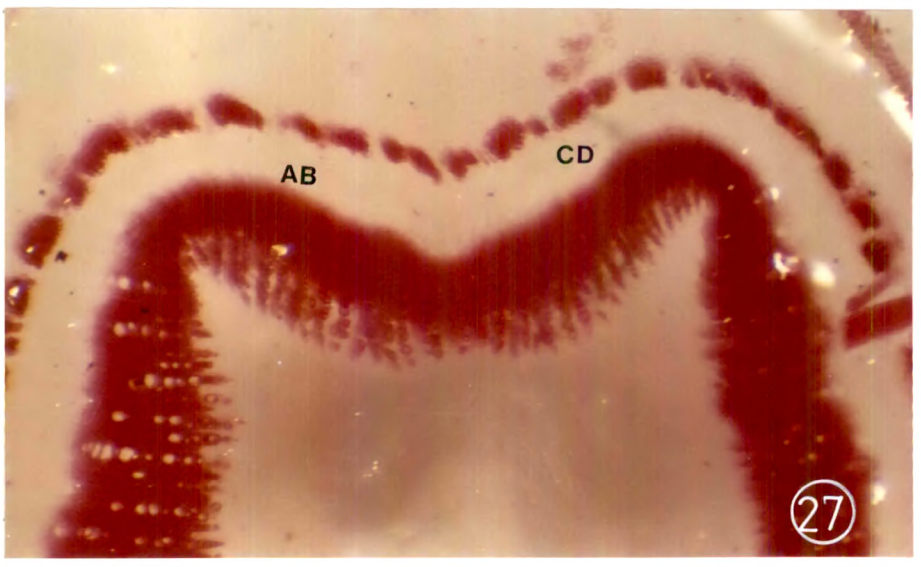
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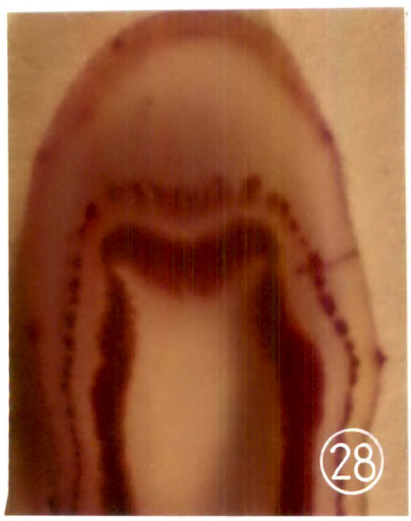
Plate 6

Figs. 28 - 33.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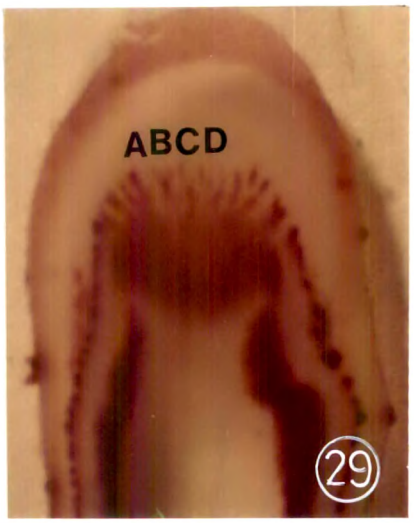
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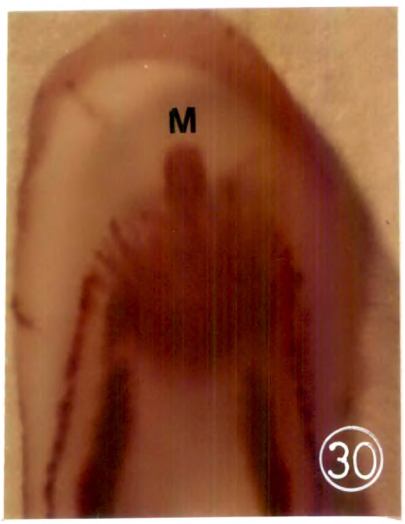
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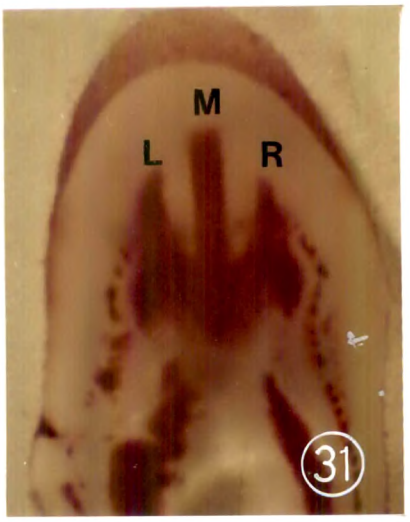
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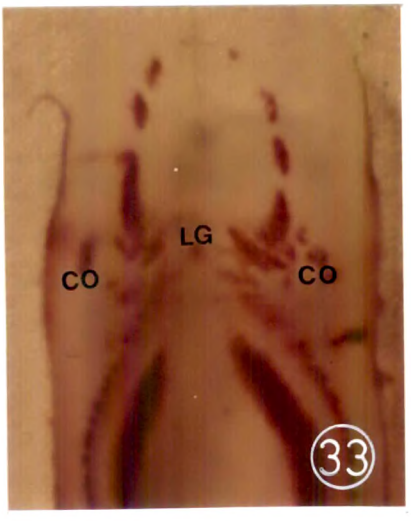
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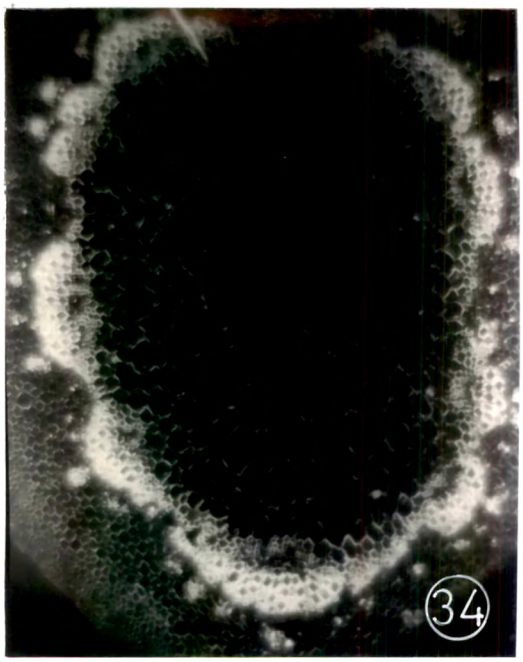
Plate 7

Figs. 34 - 44.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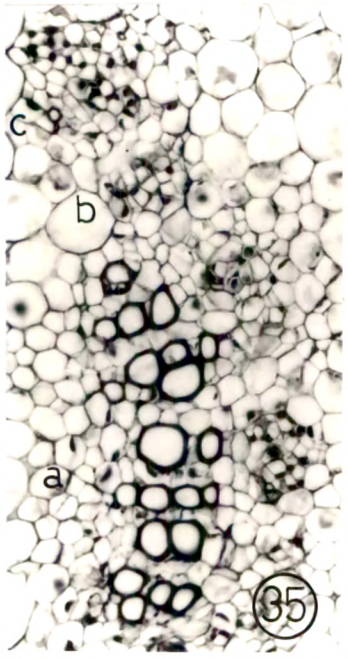

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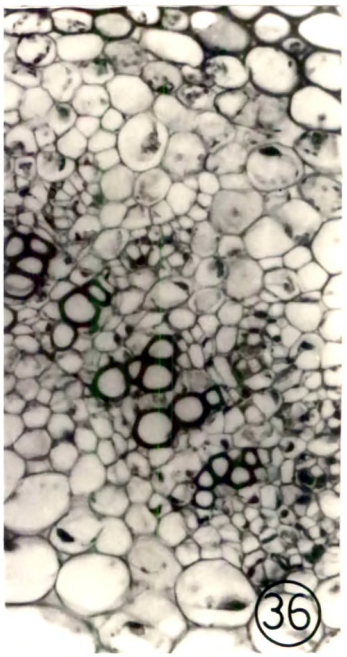
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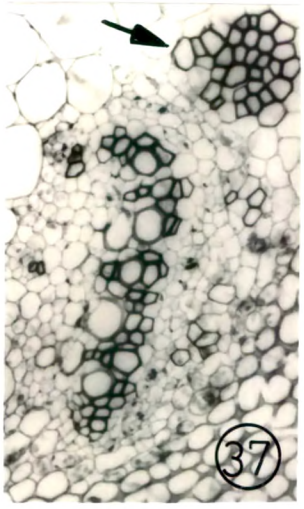
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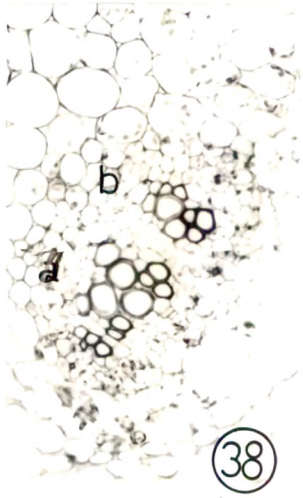

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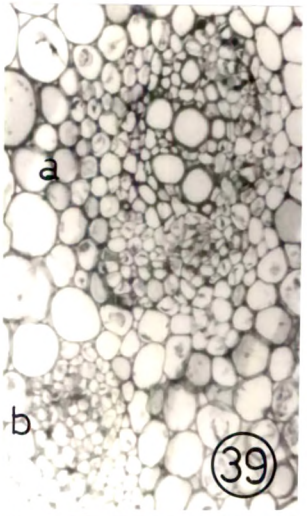

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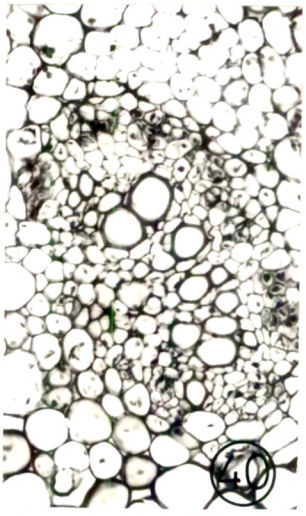
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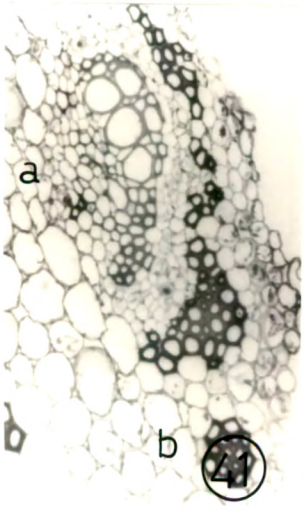

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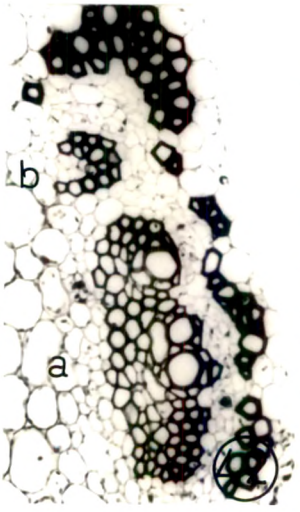

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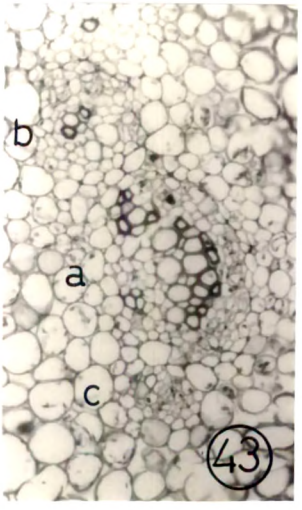

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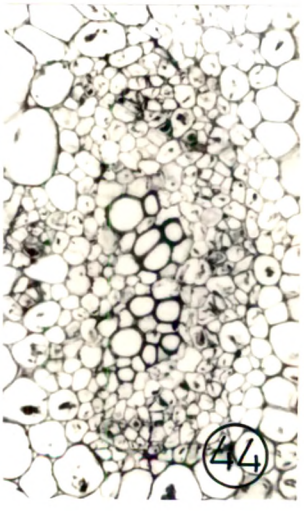
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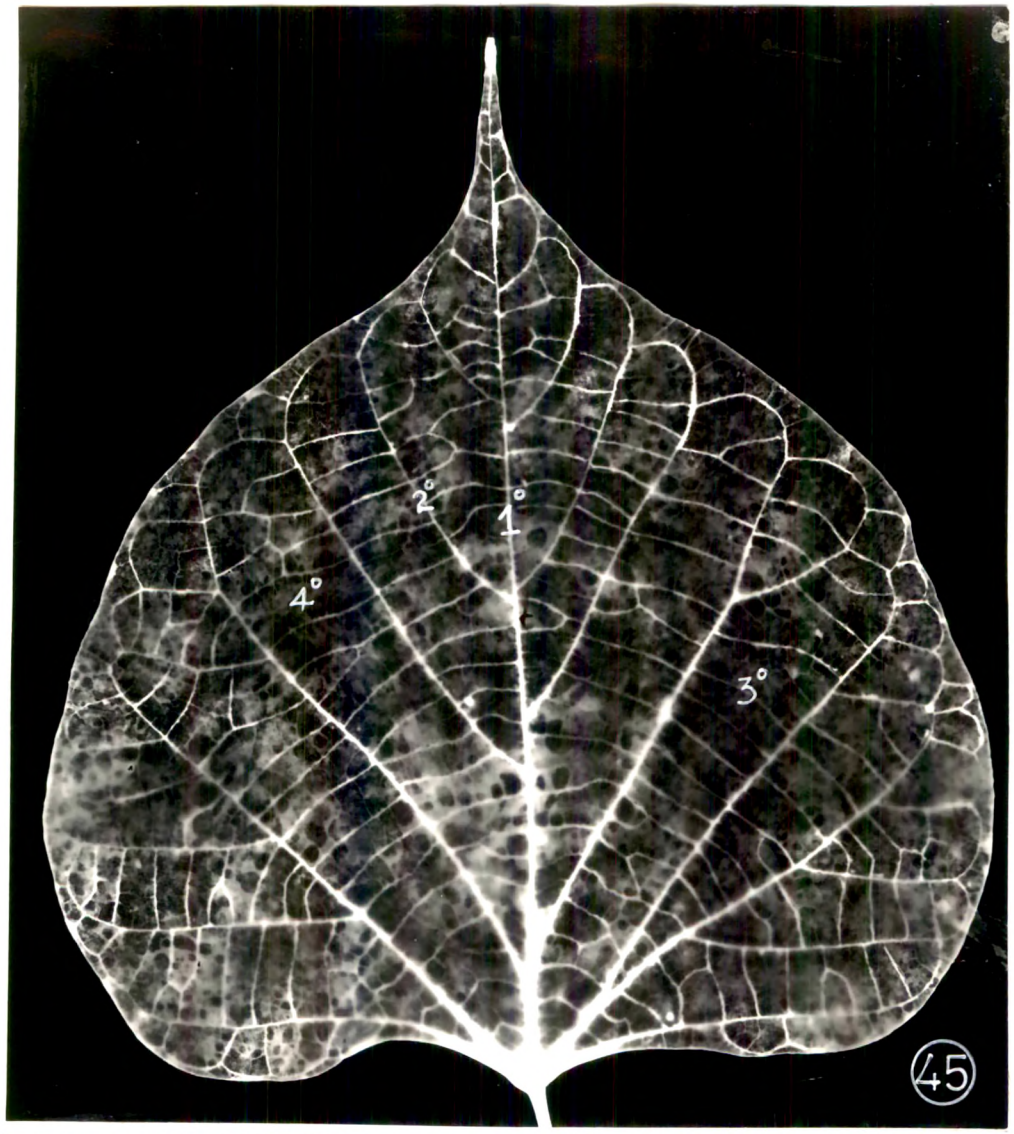
Plate 8

Figs. 45 - 48.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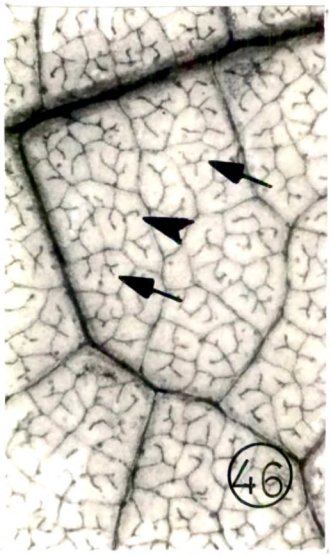
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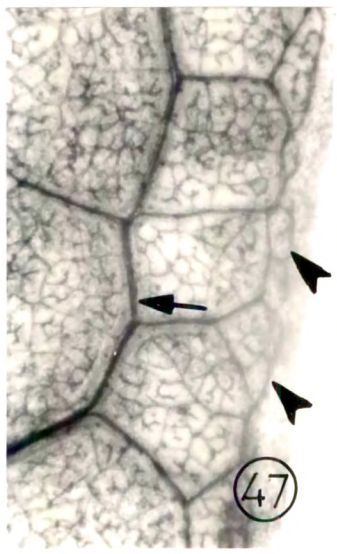
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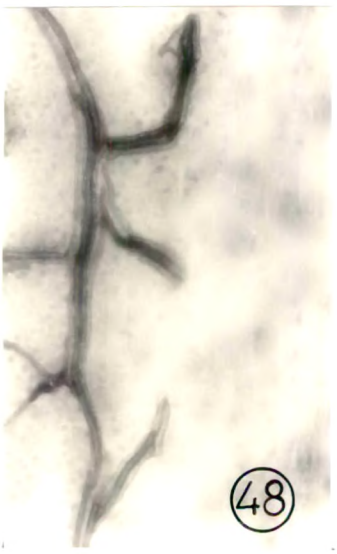
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Section II

3.1.5 Primary vascular differentiation in the petiole.

To understand the differentiation and development of the primary vascular system in the petiole both transverse and longitudinal course of development of the leaf procambial traces has been studied.

3.1.5.1 Transverse course of differentiation

At the first node from the shoot apex, the petiole differentiation is not observed in the leaf primordium. My observations are confined to the basal region of the leaf primordium, which ultimately develops as a petiolar region. The procambial strands could not be detected in relation to the youngest primordium at the first node. At its extreme base an arc of actively dividing, densely stained small meristematic cells is present (fig. 1). This arc is termed here as the vascular meristem. It is a precursory stage of the petiole vasculature where procambial strands will differentiate. It is a template on which the future vascular system of the petiole develops. The vascular meristem cells are homogenously and densely stained and distinctly marked out from the surrounding vacuolated cells of the ground meristem (figs. 1 and 2). There are 2-3 loci in the vascular meristem (fig. 4) indicating meristematic activity leading to the establishment of a procambial strand. Figures 4 and 5 illustrate the appearance of the recognizable loci of

procambial differentiation at the basal region of the leaf primordium. The discreteness of the vascular meristem with its loci of procambial differentiation is indistinct in the distal region of the leaf primordium (fig. 3).

The further development of the vascular meristem is clearly evident in the basal region of the leaf primordium at the first node in figure 6. Here 3-4 localized areas show groups of small, actively and irregularly dividing polygonal or rectangular densely stained cells, which are the leaf trace procambial strands. The cells of the vascular meristem are lightly stained and generally radially elongated (fig. 6). In the distal region of this leaf primordium only an arc of vascular meristem with a single procambial strand in the centre is noticed (fig. 7).

In the basal region of the leaf at the second node where the differentiation of the petiole is noted there are five procambial strands (fig. 8). In the petiole further from the base the vascular meristem shows a median and two laterals on either side. These strands with the intervening vascular meristem cells are arranged in the form of an arc (fig. 9). Still further, the vascular meristematic arc with only three strands are present, i.e., a median 'M' and two laterals, 'L1' and 'L2' each on either side of the median (fig. 10). The intervening vascular meristem cells are elongated and lightly stained (fig. 10). The protoxylem and protophloem differentiation is also first observed in the median strand

at this node (fig. 11). The protophloem differentiation precedes that of the protoxylem and is in continuity with the older phloem below in the stem axis.

Subsequently at the third node vascular meristem of the petiole shows 13 strands at varying stages of their development (fig. 12). Between two procambial strands the interfascicular parenchyma is present. Its cells are less vacuolated and smaller than the surrounding parenchyma cells of the cortex and pith. At this stage the cortical procambial strand is observed (fig. 13) only at the basal region.

In the 4th node the petiole shows additional development of procambial strands. The cortical strands show both protoxylem and protophloem (fig. 14). In the basal region of the petiole there are 9 collateral and 8 phloic strands arranged in a crescent form.

3.1.5.2 Longitudinal course of differentiation

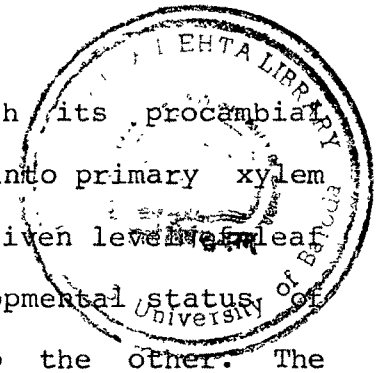
The shoot apex in Gmelina arborea is a high domed mound with a convex surface. It shows a tunica-corpus organization. The shoot apex has a single tunica layer overlying the randomly oriented cells of the corpus (fig. 15). All the cells of the tunica and part of the corpus are elongated and perpendicular to the curved surface of the apical dome. The shoot apex is distinguished into central, peripheral and pith meristems.

The leaf primordium at the first node shows the differentiation of the leaf trace procambial strand (fig. 16). It is in continuation of the procambium below in the axis. Its cells are elongated and narrower than lightly stained ground meristem cells (fig. 16). The procambium cells at the leaf base are densely stained and narrow, however these cells are densely stained distally.

A procambial strand extends from the base of the second leaf primordium to its apex (figs. 19). Its cells at the base are longer that those towards its distal region (fig. 17). In longitudinal section the cells of the vascular meristem show less affinity for staining (fig. 18), compared to the procambial cells. There were no other marked structural differences between them. The procambial cells when observed basipetally were in continuity with the existing procambial traces at lower levels in the shoot axis (fig. 19). This along with certain other evidences mentioned subsequently later in the chapter indicate that the differentiation of procambium is acropetal. These acropetally developing procambial strands extend into the petiole and lay the foundation of the petiole-leaf vascular system.

3.1.6 Procambium

Following the establishment of a leaf centre in the shoot apex and later its development into a recognizable leaf form, its vascular system is differentiated in stages, first



as a leaf trace, vascular meristem with its procambial strands and subsequently differentiating into primary xylem and primary phloem and parenchyma. At any given level of leaf development in Gmelina arborea, the developmental status of the leaf trace strands varies from one to the other. The petiole from its inception to the cessation of its elongation is considered to have primary growth and the meristem giving rise to the primary vascular tissue is termed as vascular meristem and procambium. There after the petiole shows transformative stages of development of procambium into the secondary vascular meristem, the transit secondary meristem cells.

Procambium is identified according to the criteria mentioned by Esau (1965a). The primary sieve elements were identified by their position in the vascular strand, i.e., on the outer periphery of the strand, their relative size, lack of cell contents and presence of callose. Protoxylem elements were identified by their small size, rounded appearance, birefringence of their walls in polarized light and thickening pattern. Metaxylem elements were distinguished from protoxylem by their late appearance, their large diameter, irregular shape in cross section, relative position with regard to protoxylem and cell wall thickening pattern.

The median strand in Gmelina arborea is the largest among the vascular strands and it extends throughout the entire petiole without much bifurcation, hence its

development is mainly considered and described here. The general development of the other strands is also studied. Critical observations on the other vascular strands, if any, have also been described at appropriate places.

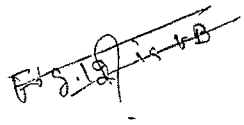
As mentioned earlier the procambium is distinguished in the form of a trace strand with the vascular meristem as the template. The leaf primordium at the second node shows a median strand and two lateral procambial strands. The procambial strands develop acropetally and are in continuity with the developmentally older procambial strands lower in the stem. Vascular elements in the median strand differentiate earlier than those in the laterals. The acropetal development of the median and other strands in the leaf primordium is indicated by the presence of more mature vascular elements at the base and developing elements at the distal end of the leaf. Also the size of the strand is largest at the base and it decreases towards the tip. The median strand is large and well developed at the base of the leaf, with a single tracheary element and one or two phloem elements (fig. 20). When its development is traced upwards, a differentiating tracheary element characterized by a nucleus and cytoplasm is observed. At the distal end, only protophloem is present (figs. 21). Still further up the median strand is procambial (fig. 22).

The procambial strand consists of apparently homogenous cells (fig. 22) irregularly oriented and dividing in varying

planes. At a distance of about 370 µm from the tip of the primordium a cell towards the inner side of the procambial strand divides periclinally producing the first radially aligned cells. Thus it is at this level of the primordium, the procambial strand shows a definite plane of cell divisions producing a radial seriation. Radially aligned cells of the procambium are mainly associated with xylem development after the establishment of the protoxylem pole, and procambial cells external to them are oriented irregularly and they divide in varying planes. They constitute the phloic procambium. It is only by this criterion that the xylary and phloic procambium could be somewhat distinguished.

The first vascular element to differentiate is a sieve tube element of the protophloem, observed at a distance of about 370 µm from the tip of the leaf primordia (fig. 21). The further details of phloem have been mentioned in the next section. The first xylem element is observed in the basal region of the leaf primordium. A single tracheary element is found towards the inner periphery of the procambial strand and adjacent to the pith (fig. 20). Of the two radially arranged cells in a row, the one towards the pith will differentiate into a tracheary element. Cortical bundles are absent in the petiole at this stage.

The basal end of the leaf at the third node from the shoot apex has 13 collateral and phloic strands arranged in a



crescent form (fig. 12). The strands are at different stages of development. The median strand increases in area radially and tangentially.

The periclinal divisions in the vascular strand increase in frequency and each vascular strand has 1-2 tiers of radially aligned procambial cells. The dividing procambial cells become increasingly vacuolated and are conspicuously set off from the xylem and phloem region. They occur as radially seriated tangentially aligned cells (fig. 23) or as isolated periclinally dividing cells (fig. 24). The derivatives of procambium towards the phloem are small and irregularly arranged and have no preferred directions of cell orientation. And those highly vacuolated and radially aligned are developing tracheary elements (fig. 23). The concomitant periclinal divisions within the protoxylem pole produce a radial file of cells. The cells in the interfascicular area keep pace with the radial increase in the area of the developing vascular strand by occasional periclinal divisions. Cortical procambial strands are present in the basal region of the petiole.

In a 3 mm long petiole there are 13 collateral and 10 phloic strands at the base and they extend to its tip. The structural features of the procambial cells resemble to those of the previous stage except that at few sites differentiation of the tracheary elements and isolated rectangular procambial cells are observed.

In a 6 mm long petiole there are 14 collateral and 13 phloic strands at the base. A marked difference noticed at this stage is the development of metaxylem and metaphloem elements. The vascular strands develop radially and tangentially. The xylem and phloem in the median strand are delimited by one or two rows of procambial cells, which have narrow radial diameter. Procambial cells at certain sites of xylem parenchyma differentiation are polygonal. Hence their structural homogeneity is not evident at this stage. It is difficult to delineate the last formed protoxylem and early formed metaxylem elements.

Cortical strands have 1-2 tiers of rectangular procambial cells in the basal region. Towards the middle and distal regions in the petiole polygonal and rectangular procambial cells are present.

In a 2.5 cm long petiole, there are discrete collateral and phloic strands interspersed either with interfascicular parenchyma or groups of procambial cells (fig. 25). Procambial cells are evident at the sites of anastomosing or fusion of the young strands. At this stage the procambial cells undergo frequent periclinal divisions forming a distinct radial seriation (fig. 25). The phloic strands also show 2-3 layers of procambial cells.

In transections of the petiole at its three different regions, the procambial region distinctly shows two types of

procambial cells (i) rectangular and (ii) polygonal. Their distribution varies in the vascular strands. In some vascular strands a tangentially continuous row of 2-3 radially aligned cells extend across the strand (fig. 26). In others the continuity is disrupted by the polygonal cells. The rectangular procambial cells are generally located in radial seriation with the developed and developing tracheary elements and the polygonal ones are located generally at sites where xylem parenchyma differentiation occurs (fig. 27). Such a histological distinction of procambial cells associated with the development of specific types of cells is not found towards the phloem region. As the repeated periclinal divisions result in radial seriation of procambial cells, the vessel elements and parenchyma cells in the primary xylem also appear in radial files. Procambial cells though mostly divide periclinally, anticlinal divisions are also present.

Generally it is believed that in the leaf all the vascular strands derived from the leaf trace strands develop acropetally (Esau, 1942). In the petiole of Gmelina arborea interfascicular parenchyma cells are present between two collateral strands and also between phloic strands. In the basal region of the petiole few procambial strands differentiate independently. The development of such a strand has been studied and described here. It is termed as subsidiary strands to distinguish them from the other vascular strands which have their origin from the leaf trace

strands. A strand positioned in the interfascicular region at the adaxial open end of the vascular system when traced either towards or away from the base shows precursors of the procambial cells (fig. 30), and when traced further in both the directions, only interfascicular parenchyma is observed at its site (figs. 28-33). The area of the strand also decreases in both the direction. So this strand shows both acropetal and basipetal development. The site at which the area is maximum is the locus of its origin (fig. 32). The development of this strand at different successive levels is illustrated in figs. 28-33.

The origin of such subsidiary strands is from the interfascicular parenchyma cells that dedifferentiate and redifferentiate as procambial cells and hence are considered to be of late in origin. The others derived from the leaf trace strand are of early origin.

In the young and elongating 7cm long petiole there is a rapid increase in the frequency of periclinal divisions in the procambial cells thus resulting into 3-5 layers of radial seriation delimiting xylem and phloem (fig. 35). As mentioned earlier two types of procambial cells are present. 3-4 layers of procambial cells delimit xylem and phloem of the cortical strand (fig. 34).

3.1.7 Transition of procambium to the secondary vascular meristem.

The transition of procambium has been studied in a 14.4 cm long petiole, which completed its full elongation and is considered as mature stage I. The petiole has discrete vascular strands with interfascicular parenchyma. The following morphological and micromorphological features distinguish this petiole from the petioles of the previous stages :-

(i) there is no further elongation.

(ii) a decrease in the length and radial diameter of the recently formed tracheary elements is noticed. They are shorter and narrower than the previously formed ones. The pattern of wall thickening in the earlier formed tracheary elements is annular or helical (figs. 36-39). Recently formed tracheary elements show closely helical or scalariform wall thickening (figs. 40, 41) in the three different regions of the petiole.

(iii) in the middle region of the petiole, the xylem parenchyma and interfascicular parenchyma cells are lignified and show birefringence at randomly distributed regions and not confined to any particular side of the petiole. The large cells outlying the periphery of the protophloem, described previously as the precursors developed as protophloem fibres.

All the vascular strands in the pulvinus base and the distal region of the petiole show thin walled protophloem

parenchyma cells capping the phloem complexes and when observed under polarized light xylem parenchyma and interfascicular parenchyma cells do not show any birefringence (fig. 42). In the middle region of the petiole the protophloem fibres capping the phloem complexes, xylem parenchyma and interfascicular parenchyma show birefringence (fig. 43) indicating lignification of their walls. Lignification is more pronounced in and between the vascular strands on the adaxial side of the petiole (fig. 45), than those of the median and other vascular strands on the abaxial side (fig. 46).

The median strand towards the pulvinus base is distinctly tangentially wide and the region between xylem and phloem apparently shows 2-3 layers of rectangular cells intervening with radially elongated cells or polygonal cells (fig. 47). Towards the basal region the intervening cells are less radially elongated than those in the pulvinus region (fig. 48). They do not divide frequently. In the middle and distal regions of the petiole the xylem and phloem region is set off by 1-2 layers of rectangular cells intervening with polygonal cells (fig. 45). The phloic strands also show 2-3 layers of similar cells.

In longisections they appear elongated with parallel side walls and the end walls are transverse or one end transverse and the other end oblique or fusiform. These cells divide mostly periclinally, but anticlinal divisions are also

present. The intervening cells between the rectangular cells divide, some of them differentiating as parenchyma cells and keep pace with the growth of the adjacent cells. In longisections they are shorter than the rectangular cells and have transverse end walls.

Generally the secondary growth in the petiole of a dicotyledonous leaf is accompanied by certain growth and structural features like cessation of elongation, differentiation of protophloem fibres, birefringence and lignification of xylem parenchyma and interfascicular parenchyma and the vascular elements of the same length as their precursors (see table II). In mature stage-I of the petiole of Gmelina arborea though the anatomical features associated with the beginning of the secondary growth are noticed, the recently formed vascular elements are more elongated than their precursors, the protophloem fibres show gradual stages of differentiation and the xylem parenchyma and interfascicular parenchyma are lignified only at few sites. Hence I conclude that this stage is an indication of transition of procambium to secondary vascular meristem, wherein some of the features associated with the secondary growth are observed. This stage of the petiole indicates that the beginning of secondary growth is discrete in the middle region.

3.1.8 Secondary vascular meristem:transit cells.

The next petiole examined was one which was collected

one month after the cessation of its elongation and designated as mature stage-II.

The vascular strands are discrete in the basal and distal region of the petiole. Xylem parenchyma within the strands and some fibres show birefringence. In its middle region due to the lignified xylem parenchyma and the interfascicular paranchyma,the vascular system do not show discrete strands but appear almost like a cylinder (fig.44). The protophloem fibres are well developed. In the basal and distal regions they mostly appear collenchymatous (fig.49).

In this petiole all the structural characteristics associated with the secondary growth are observed and hence the transition of procambium to the secondary vascular meristem has been recognised. The cambial system normally consists of two types of initials,the axially elongated fusiform initials being responsible for the development of the axial elements of the secondary xylem and phloem and the short ray initials being responsible for the development of the horizontal ray system. In the petiole of Gmelina arborea though features associated with secondary growth are noticeable, the secondary vascular meristem cells in transition do not resemble the fusiform or ray cells. No beaded thickening on the walls have been observed. A definite normal cambial structure is absent and hence these secondary vascular meristem cells are termed here as transit cambial cells. I have failed to observe any stage of transition where

initial cells could be identified.

The survival period of the leaf from its maturity to senescence is about 6 months during which no noticeable structural changes have been observed in these cells.In a senescent petiole the radial and tangential walls of the transit cells are thickened in the three regions of the petiole.

3.1.9 Morphological and dimensional changes during the transition of procambium.

Initially in a transverse view of the young petiole procambial cells appear structurally homogeneous. They are rectangular (figs. 23 and 24) with transverse or rounded end walls. Radial seriation of procambial cells is observed during the protoxylem development. In a longitudinal view these cells appear structurally homogeneous and elongated (fig. 50), with lateral walls parallel and transverse end walls. With the elongation of the petiole the procambial cells also elongate and increase in vacuolation.

With the onset of the metaxylem and metaphloem development micromorphologically two types of procambial cells are observed (1) rectangular and (2) polygonal. The rectangular cells as mentioned earlier are in radial seriation with the tracheary elements. They are tangentially wide, radially narrow and generally highly vacuolated (fig.27). The polygonal cells intervening between the

tracheary elements are densely stained and radially elongated (fig.27). In longisections there is no marked difference between these two types of cells except that polygonal cells are generally shorter and narrower and more densely stained than the rectangular ones (fig.51).

The structural heterogeneity of the procambium is the result of the following micromorphological development,

(i) - some procambial cells undergo frequent periclinal divisions followed by tangential widening resulting in the radially seriated rectangular cells,

(ii) the other procambial cells elongate vertically without much apparent periclinal divisions, mostly differentiating as vascular parenchyma cells, which are polygonal.

This heterogeneity is also a signal to the onset of metaxylem and metaphloem development. Some of the precursors of xylem elements are binucleate or even trinucleate and with one or two nucleoli. The end walls of the procambial cells are transverse or slightly oblique. In the mature stage I the procambial cells are in transition. The transit cells intervened by parenchyma give rise to xylem and phloem. The transit cells have parallel lateral walls with transverse, oblique or fusiform end walls (fig. 52). In the middle region of the petiole these cells are longer and narrower (fig. 53) than those in the basal and distal regions of the petiole (see table II). The layers of transit cambial cells are not continuous in the vascular strand itself. They are

separated by parenchyma cells which are radially elongated without any intercellular spaces. They divide anticlinally and periclinally keeping pace with the differentiation of adjacent secondary xylem and secondary phloem.

In mature stage-II the transit cells show transverse, oblique or fusiform end walls and parallel lateral walls (figs.54 and 55). Longitudinally the parenchyma cells between the transit cells are short with both end walls transverse. In the pulvinus base also they have transverse end walls. The dimensional features of the procambial cells through their transition in the young and mature petioles of MI and MII are shown in table II.

3.1.10 Ontogenetic relationship of transit cells and phloem elements.

Some of the vascular strands in the basal region of the mature petiole have their transit cells and phloic derivatives radially aligned in a complex which is clearly evident in transections. A portion of the median strand in the basal region of the petiole has been examined serially to understand the ontogenetic relationship of the transit cells and the phloem elements. Figure 58 illustrates the portion of the median strand examined at successive levels. The number and orientation of cell divisions occurring in a derivative cell leading to the differentiation and formation of a complex of phloem elements in the phloem complexes vary.

The assemblages within the tier were found containing various combinations of sieve elements with companion cells and parenchyma, and only sieve elements and companion cells. The term tier represents the radially arranged cells in a row.

Serial sections indicate three different patterns of cell divisions, (i) an anticlinal followed by a periclinal one in the mother cell results in a complex of 3 cells (fig.58A, assemblage 2). The first division is anticlinal giving rise to a large sieve tube mother cell and a parenchyma cell. The sieve tube mother cell divides periclinally and unequally giving rise to a large sieve tube element and small companion cell. (ii) the mother cell divides anticlinally giving rise to a large sieve element and a small companion cell. (fig.58B, assemblage 2). (iii) the mother cell undergoes one periclinal division and 2 or 3 anticlinal divisions resulting in an assemblage of 2 sieve tube elements and 3 companion cells (fig.58B, assemblage 1) or 2 sieve tube elements and 2 companion cells (fig.58A, assemblage 1) respectively. The two contiguous sieve tube elements have companion cells on opposite sides (fig. 58A) or on the same side also (fig. 58B, 1).

3.1.11 Cambium in the stem

The seasonal activity of cambium in the bark of Gmelina arborea has been reported (Dave and Rao, 1982). The vascular cambium is characterized by the presence of the two distinct

system of cells (1) fusiform initials and (ii) ray initials. The vascular cambium is non storied. The fusiform cells are elongated with tapering ends arranged in horizontal tiers with overlapping ends (fig.57). They are 242-436 µm long and 15-29 µm wide. Multiseriate rays are present. Ray cells are small and almost isodiametric (fig. 57). Rays are 146-556 µm high and 65-155 µm wide. Cambial cells are uninucleate and the walls show beaded appearance. Periclinal divisions are noticed in the fusiform cells indicated by the appearance of phragmoplast in tangential sections. In a transverse view 6-9 layers of cambial cells are present (fig.56). The fusiform cells are tangentially wide and radially narrow. A comparative account of the secondary vascular meristem in the petiole and the stem has been given in table III.

Table II. Dimensional relationship between the length and breadth of procambium cells, transit cells and secondary vessel elements in the different stages of the petiole at its three regions

Stages	Procambium/ Transit cells		Length in μm	Width in μm	Xylem Length in μm	Xylem Width in μm
0.3 cm	Procambium		26.69 - 35.38	4.71-6.28	-	-
0.6 cm	Procambium		32.97 - 53.38	6.28-7.85	-	-
	Rectangular Procambium	B	62.8 - 100.4	17.2 - 21.9	153.8 - 306.1	32.9 - 40.8
		M	47.1 - 94.2	15.7 - 20.4		
		D	61.2 - 102.5	15.7 - 20.4		
2.5 cm	Polygonal Procambium	B	31.4 - 91.06	10.9 - 15.7		
		M	28.2 - 70.6	10.9 - 14.13		
		D	32.9 - 81.6	10.9 - 14.1		
	Rectangular Procambium	B	85.0 - 105.0	11.9 - 25.5	196.2 - 282.6	40.8 - 48.6
		M	66.0 - 75.0	13.6 - 23.8		
		D	68.0 - 119.0	8.5 - 20.4		
7 cm						
	Polygonal Procambium	B	33.3 - 98.6	8.5 - 17.0		
		M	34.0 - 73.1	10.2 - 13.6		
		D	32.3 - 84.0	8.5 - 13.6		
M-I	Procambium - Cambium Transition	B	65.0 - 141.0	15.7 - 18.8	105.0 - 238.0	17.0 - 27.2
		M	94.2 - 160.1	10.9 - 14.1	90.1 - 297.0	18.7 - 34.0
		D	64.3 - 146.0	15.7 - 17.2	100.0 - 220.0	17.2 - 24.0
M-II	Transit Cells	B	78.5 - 163.2	15.7 - 20.4	78.5 - 157.0	15.7 - 29.8
		M	87.6 - 172.0	15.7 - 20.4	89.4 - 172.0	28.2 - 39.2
		D	69.0 - 161.7	17.2 - 20.4	70.6 - 160.1	23.5 - 37.6
M-III	Transit Cells	B	78.5 - 157.0	18.8 - 20.4	83.2 - 157.0	23.5 - 39.2
		M	81.6 - 172.7	17.2 - 20.4	78.5 - 160.0	29.8 - 34.5
		D	78.5 - 160.0	17.2 - 20.4	81.6 - 157.0	20.4 - 34.5

B - Basal region of the petiole

M - Middle region of the petiole

D - Distal region of the petiole

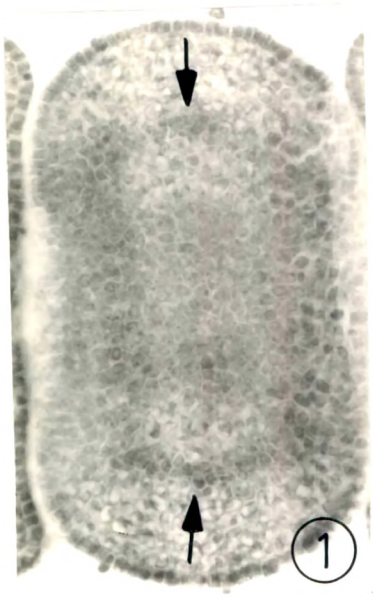
M-I, M-II and M-III = Mature stage - I, II and III

Table III. Comparative features of the secondary vascular meristem cells of the petiole and stem.

	Petiole	Stem
1) Cell types	Axial transit cells	Axial fusiform cells and vertical ray cells.
2) Arrangement	Non-storied	Non-storied
3) Number of layers	2-3	6-9
4) Fusiform cells/ transit cells		
a) Length µm	69-173	242-436
b) Width µm	15-20	15- 39
5) Rays	Absent	Present
a) Height µm	-	146-556
b) Width µm	-	65-155

Plate 9

Figs. 1 - 7.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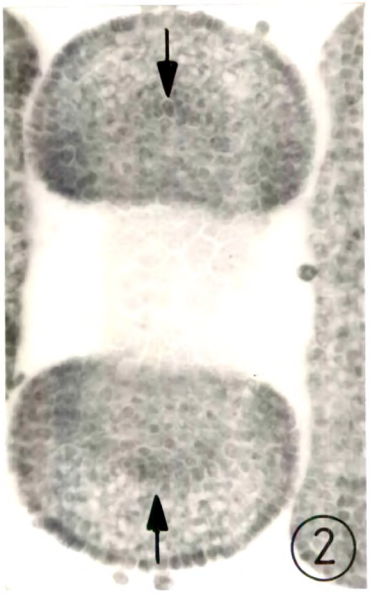
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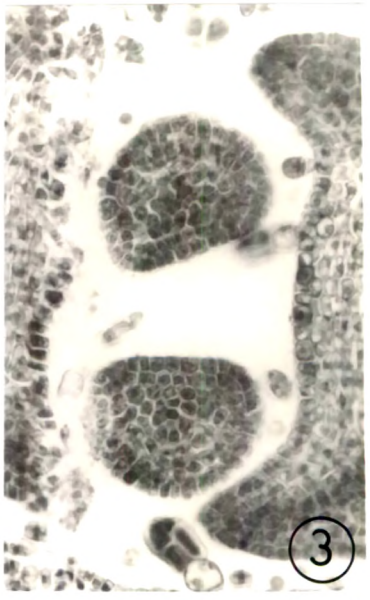
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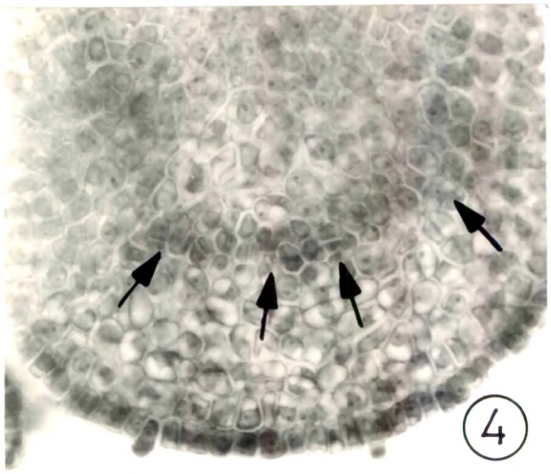
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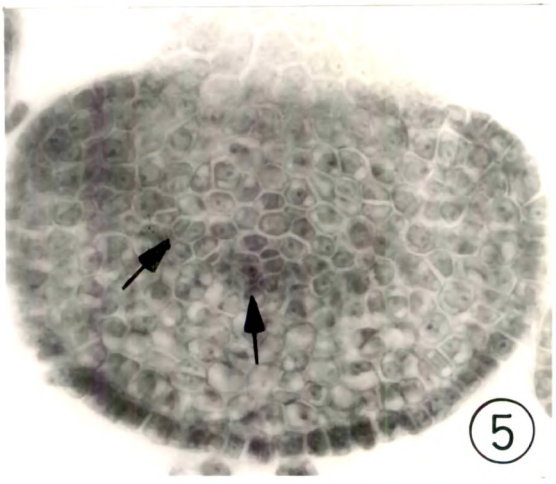
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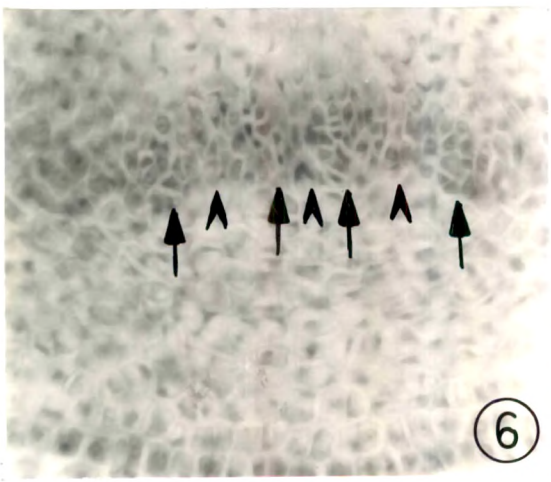
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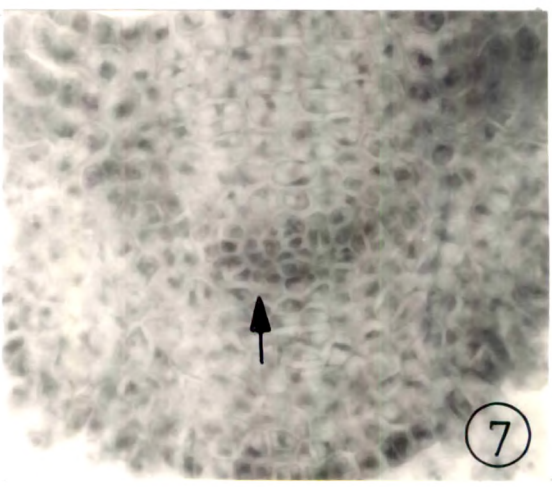
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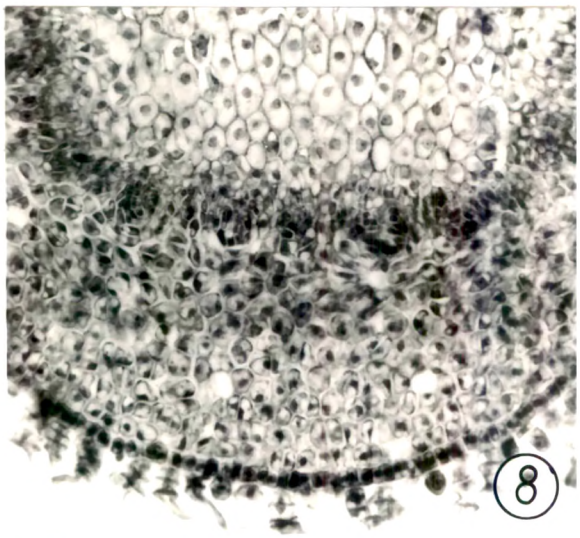
Plate 10

Figs. 8 - 14.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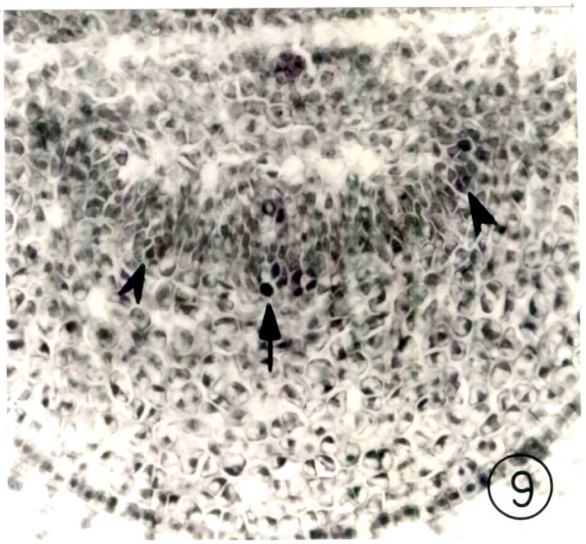
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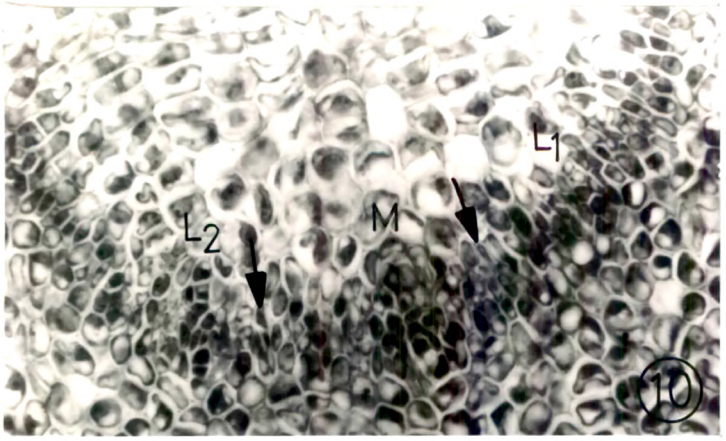

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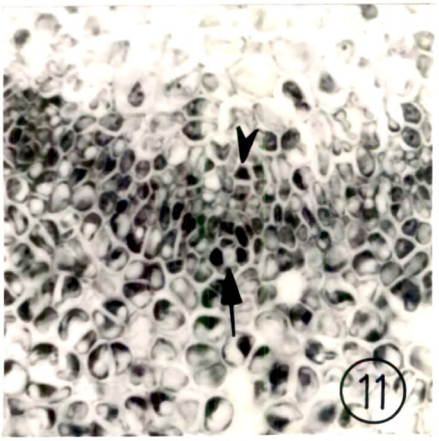
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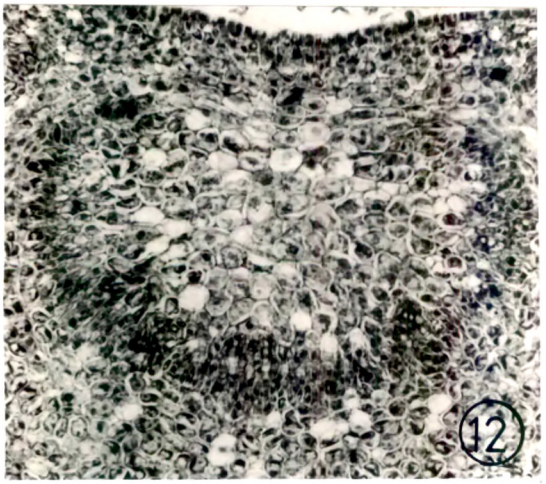
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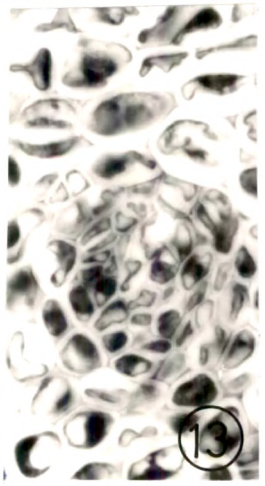
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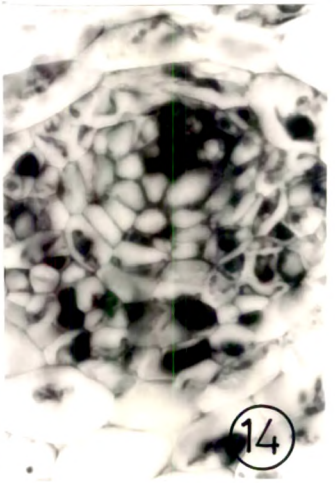
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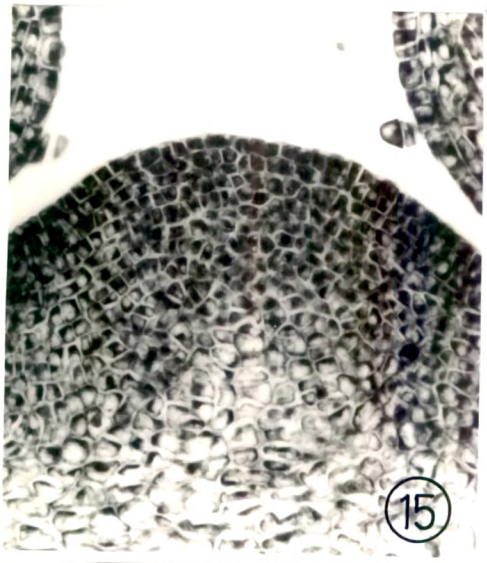
Plate 11

Figs. 15 - 19.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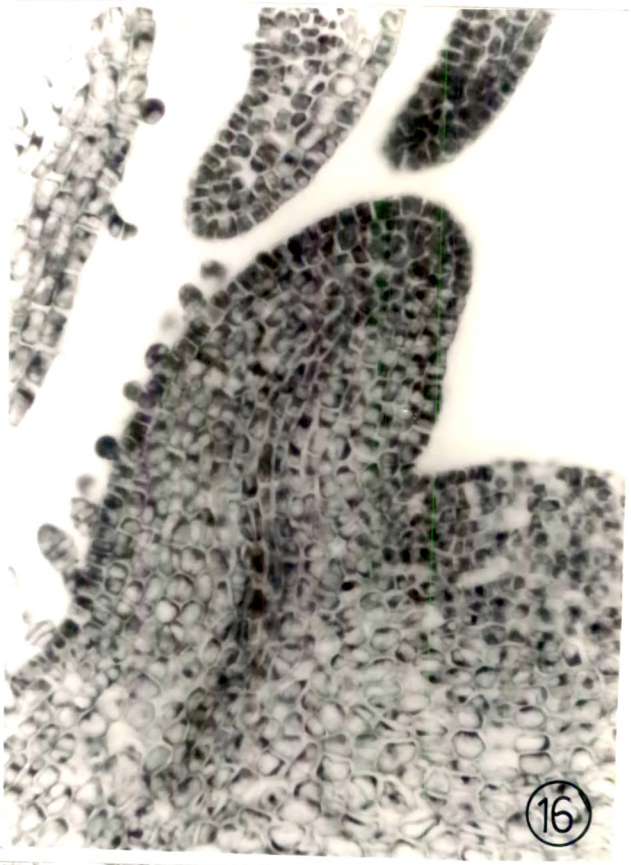
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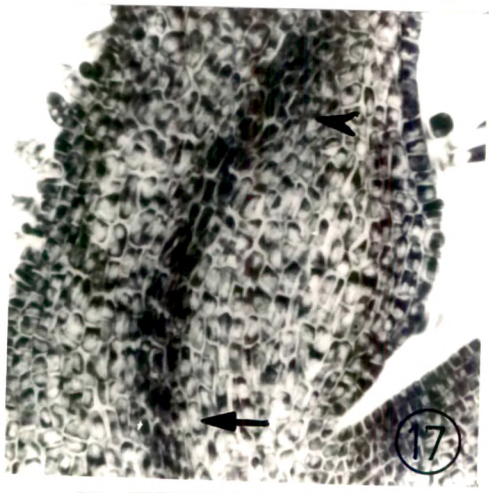
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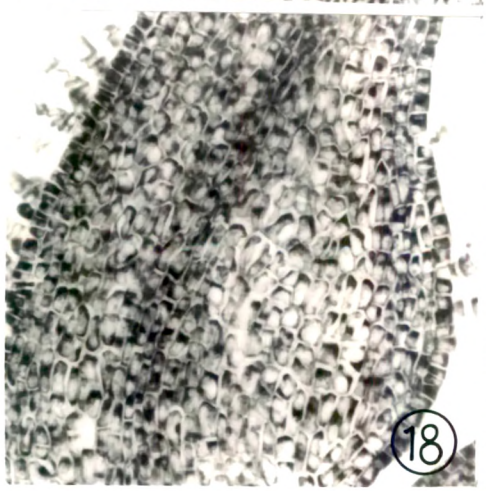
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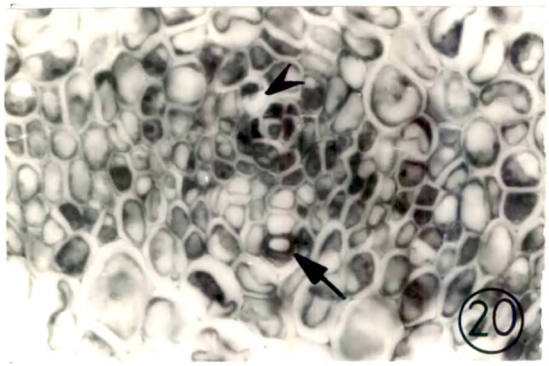
Plate 12

Figs. 20 - 27.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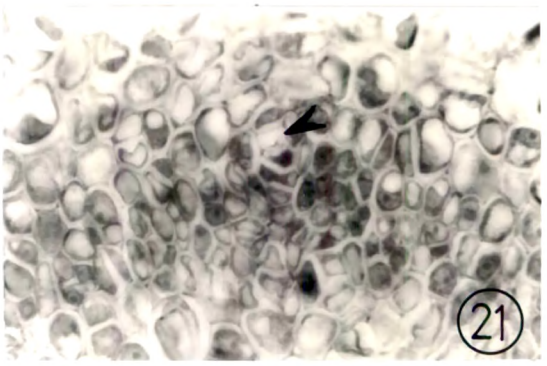
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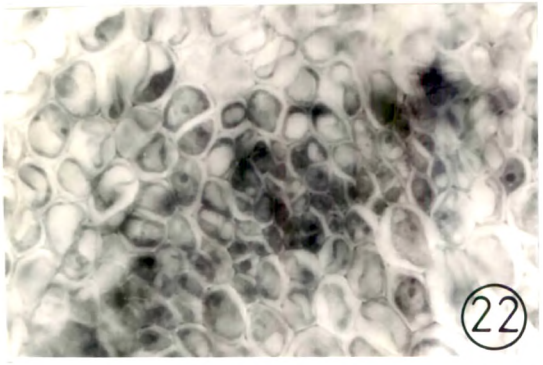
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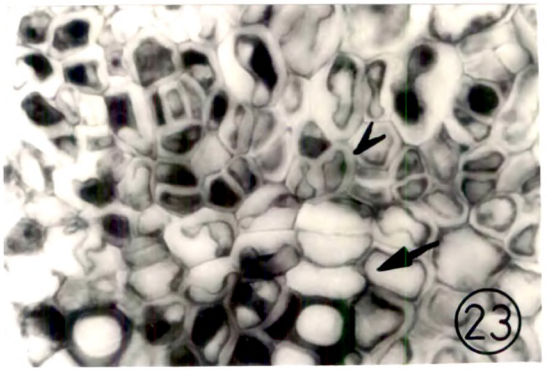
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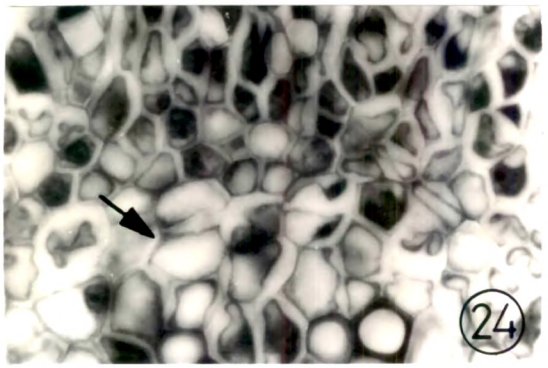
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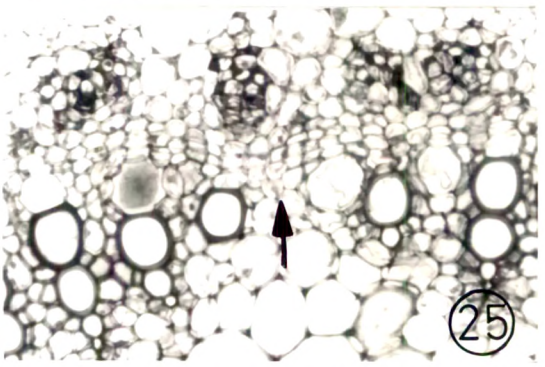
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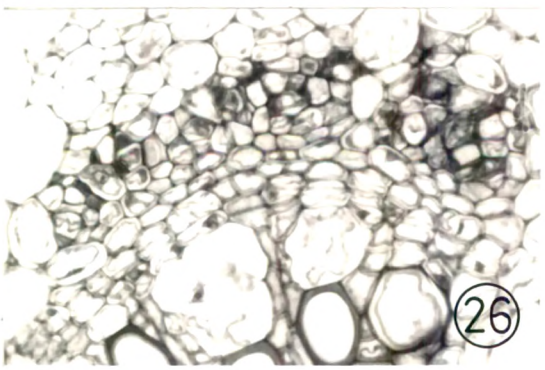
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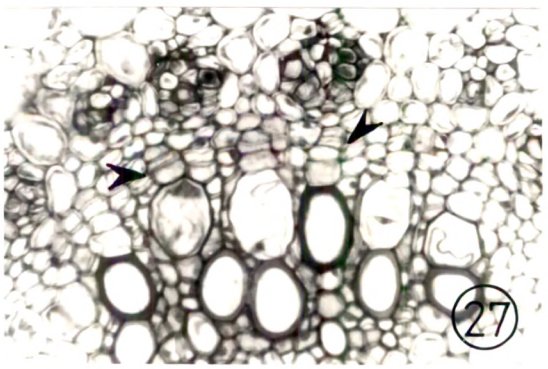
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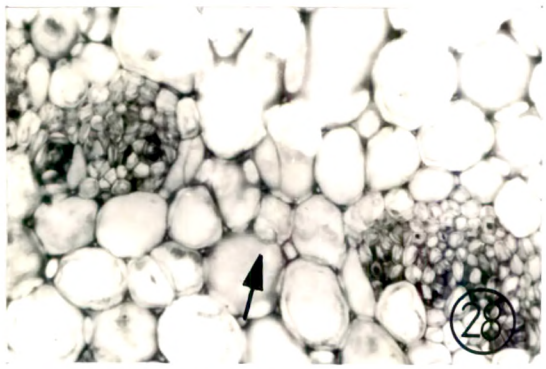
Plate 13

Figs. 28 - 35.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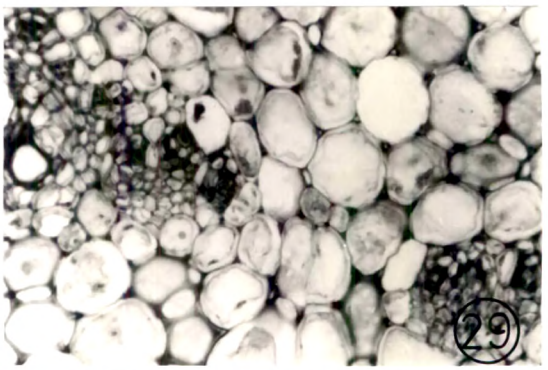
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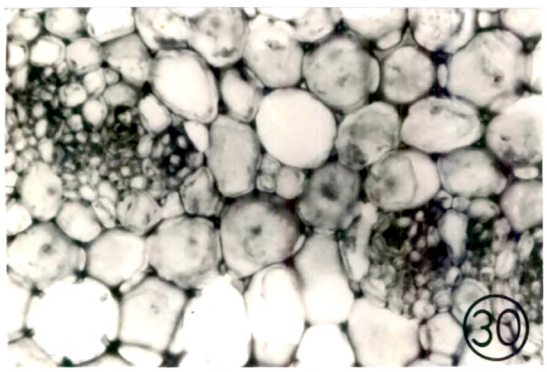
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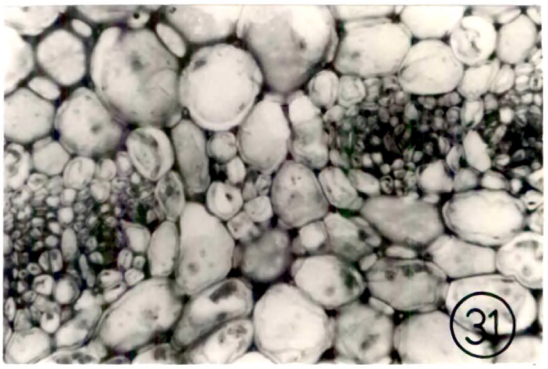
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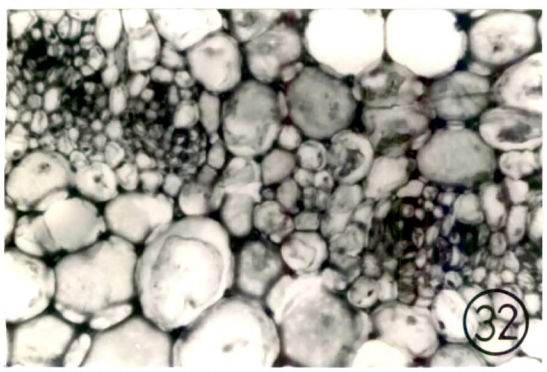
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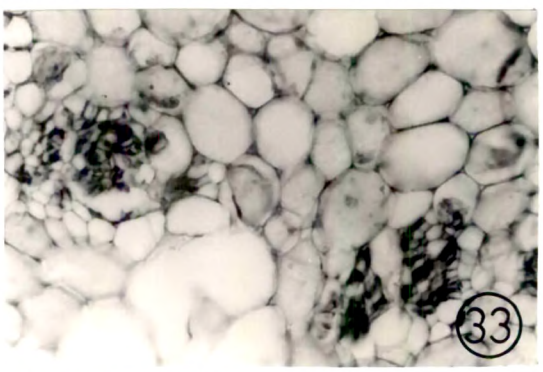
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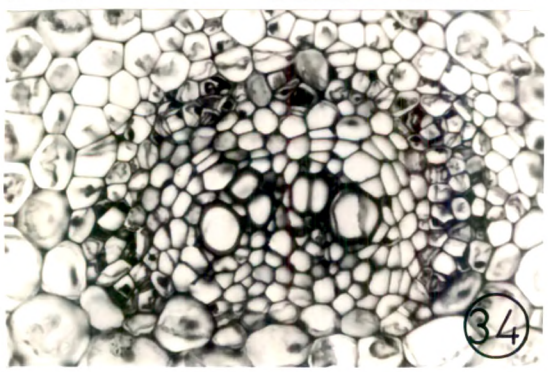
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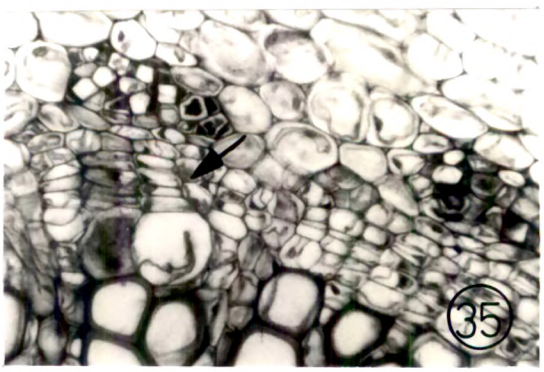
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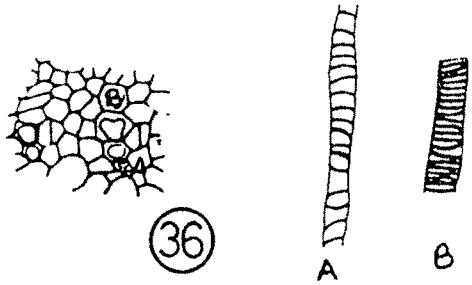
Figs. 36 - 41.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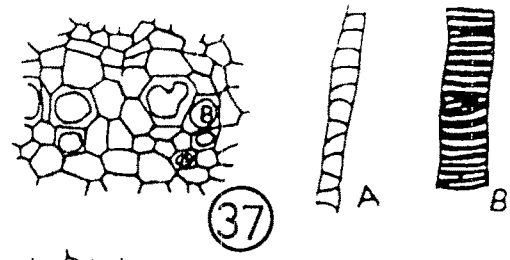
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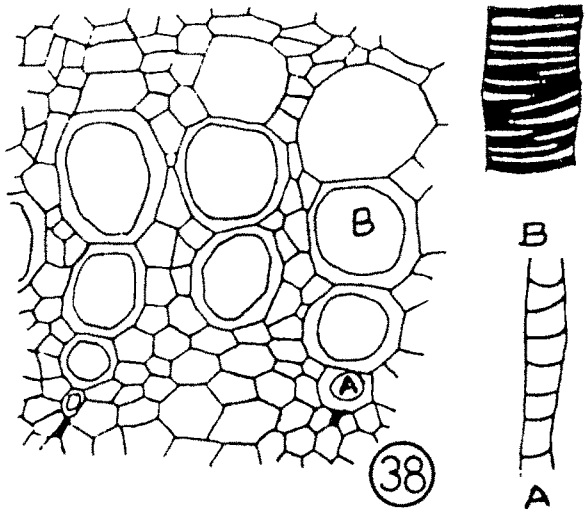
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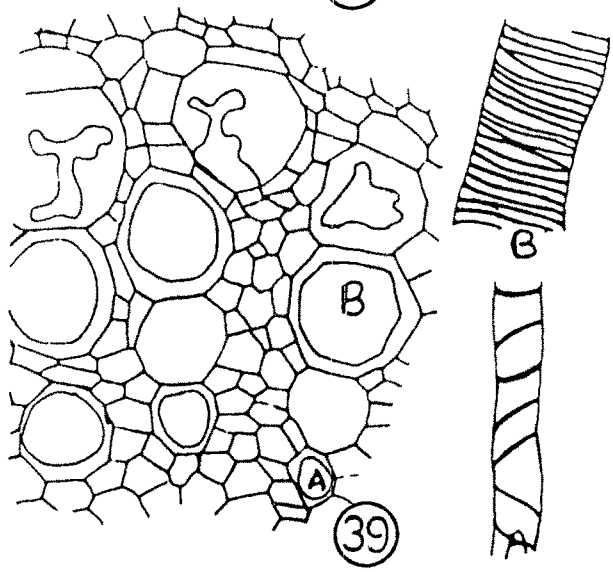
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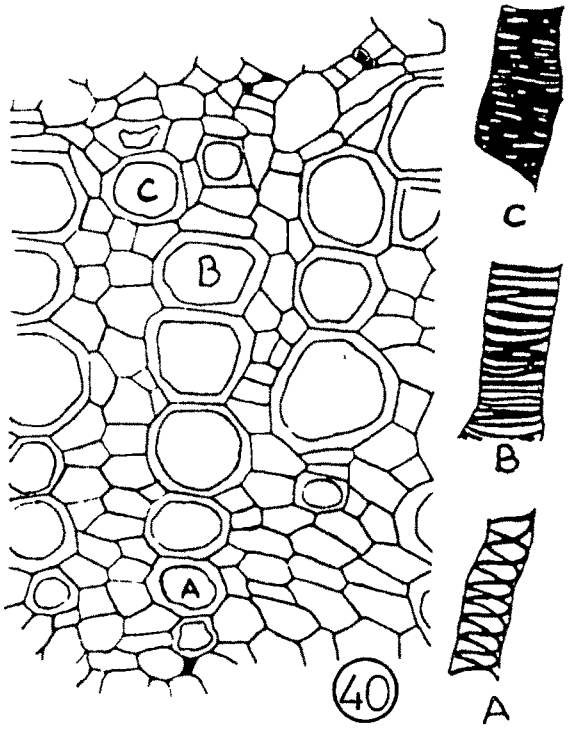
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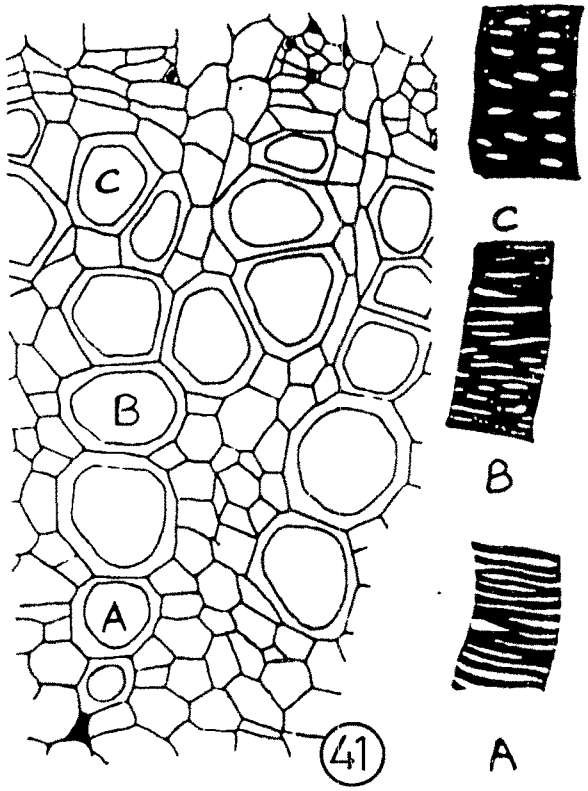
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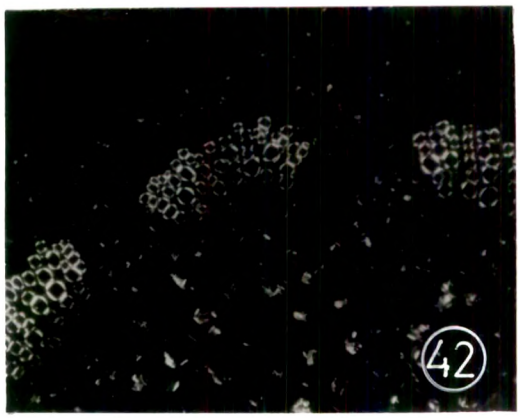
Plate 14

Figs. 42 - 49.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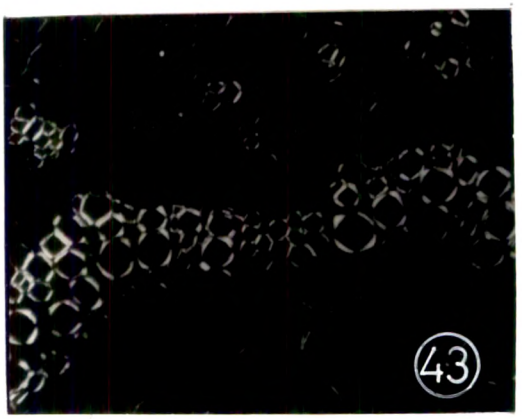
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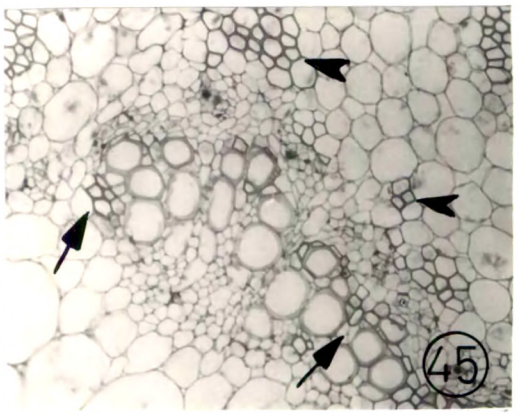
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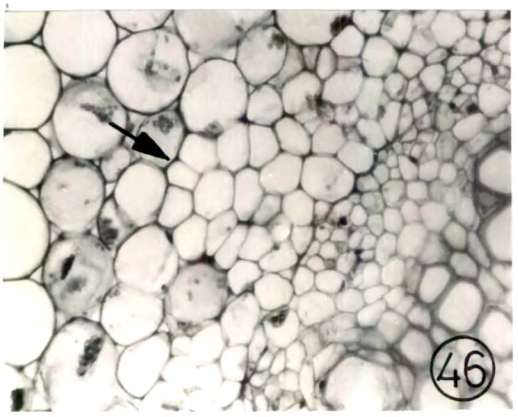
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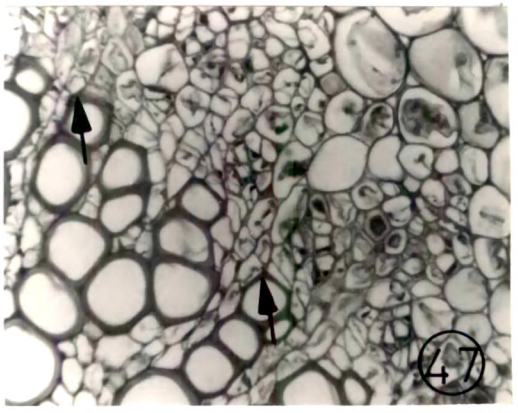
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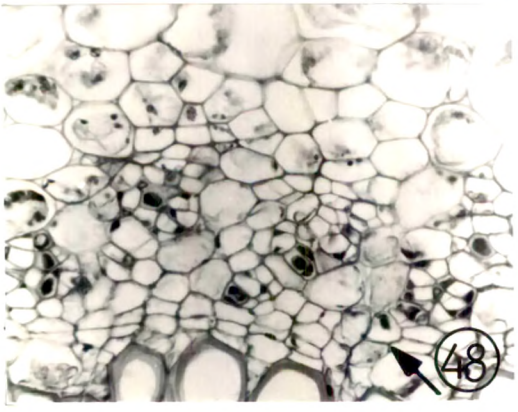
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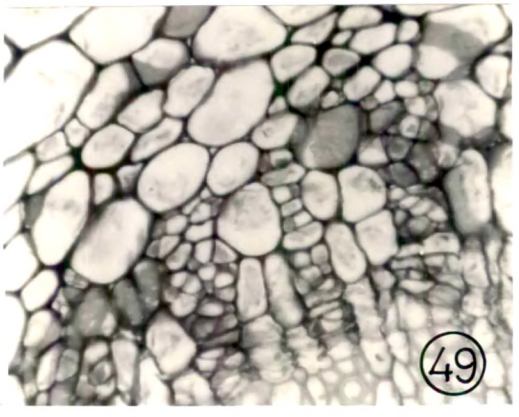
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Plate 15

Figs.50 - 57.


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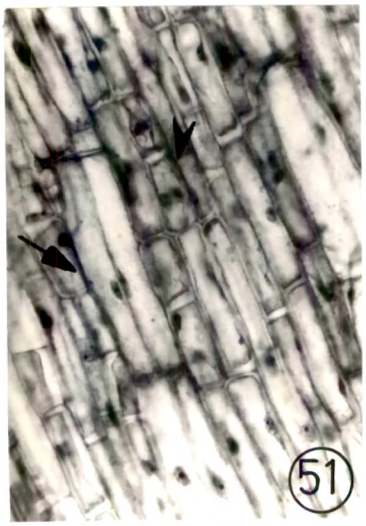
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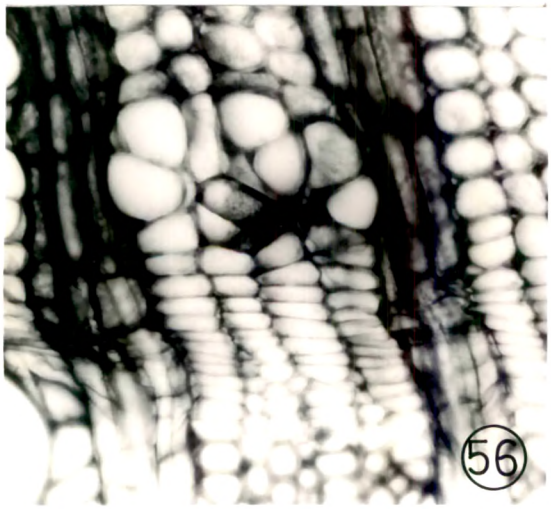
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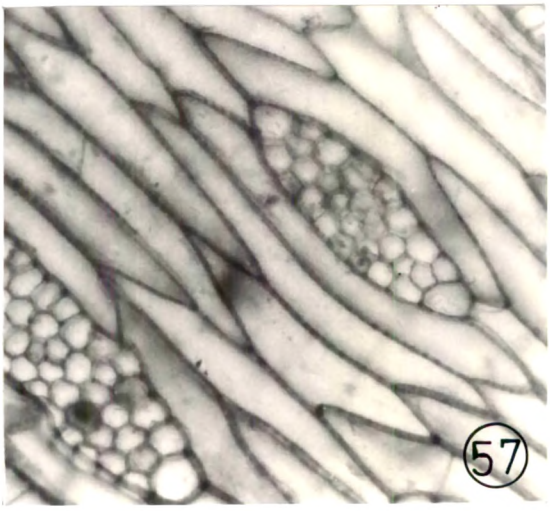
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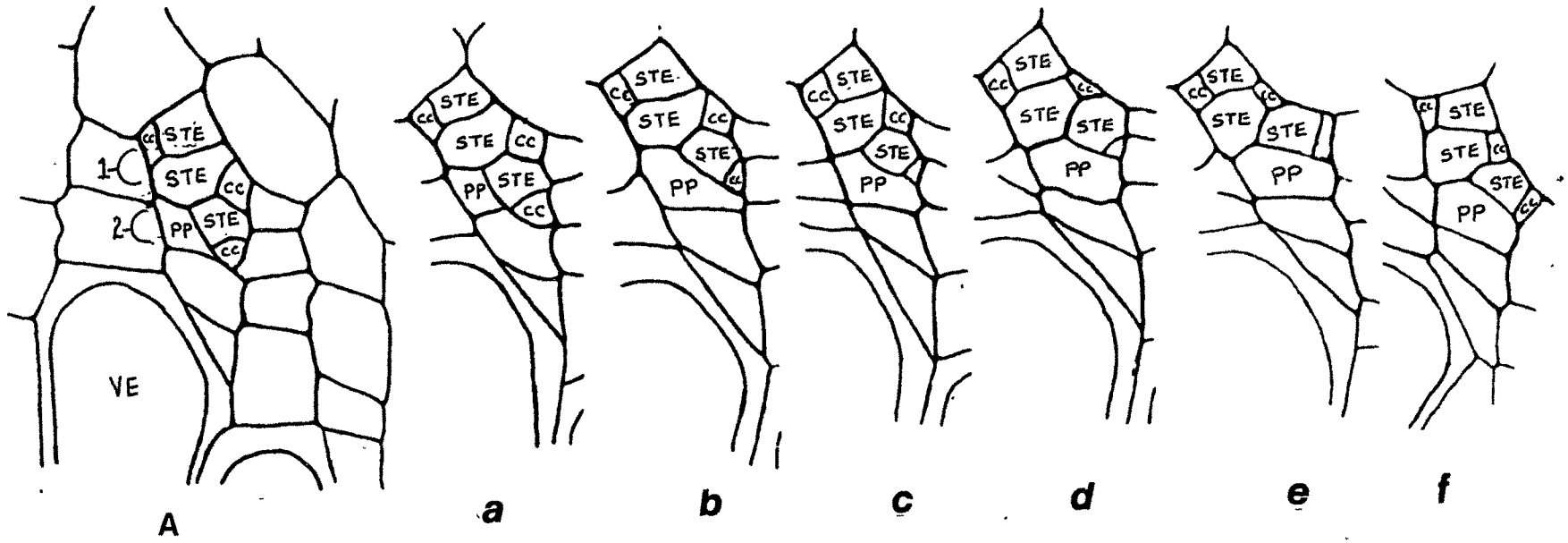
Fig. 58.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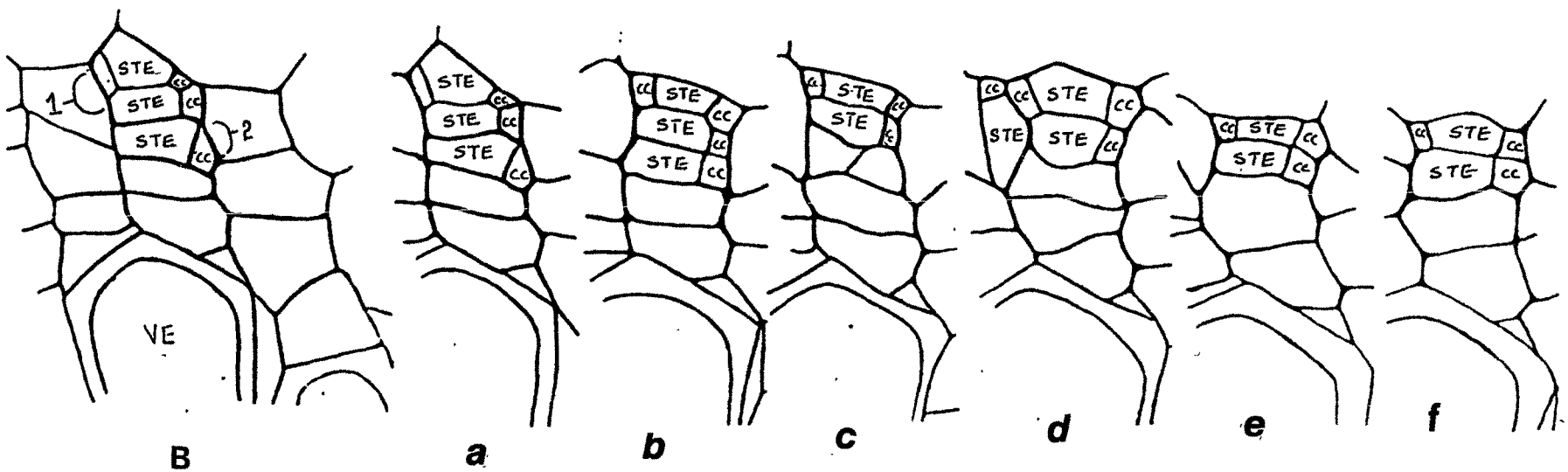
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Section III

3.1.12 Phloem

As already mentioned in the previous section, the vascular development of the petiole from its inception until the cessation of its elongation is considered primary, thereafter it is considered secondary with the production of secondary xylem and phloem. My observations are mainly confined to the median strand.

3.1.12.1 Development of phloem complexes

The establishment of procambium in a developing leaf is followed by the differentiation of certain of its cells into primary phloem and xylem elements. The first vascular element to mature within a procambial strand is the protophloem element and then the protoxylem element. The earliest stage of the development of phloem that is recognised is the differentiation of a sieve element in the second leaf primordium from the shoot apex (Section II, fig. 21) in a 0.8 mm long petiole.

In a 1.2 mm long petiole at the fourth node from the shoot apex, the sieve elements and their accompanying cells are organised in complexes interspersed with parenchyma (fig. 1). The complex includes the sieve tube elements, companion cells and contiguous and non-contiguous parenchyma cells. The number of complexes varies from one strand to the other and similarly varies in the same strand (the median

strand selected here) from the basal to the distal region in the same petiole (figs. 7-9 and 10-12). Figures 7-12 show the median strand in the basal, middle and distal regions of a 7 cm long young petiole and 14.4 cm long, one month old mature petiole. The median strand at the basal region of this mature petiole shows the secondary phloem elements also in complexes (fig. 10).

The primary and secondary phloem complexes are separated by one or two layers of parenchyma cells. In the middle and distal regions of the median strand, only certain sites show such a separation of complexes. In a senescent petiole the primary phloem sieve elements and companion cells are completely obliterated and the thickening observed at these sites indicate their previous positions.

The phloem has been categorized into primary phloem and secondary phloem and the former consists of protophloem and metaphloem.

3.1.12.2 Protophloem

3.1.12.2.1 Sieve tube element

In the median strand of the leaf primordium at the second node from the shoot apex, the protophloem development begins with the differentiation of one or two sieve tube elements followed by that of protoxylem tracheary elements. Additional sieve tube elements further differentiate

laterally and centripetally. The differentiation of protophloem is first noticed in the median strand and subsequently in the laterals (fig. 2). The clear lumen of the sieve tube element contrasts strikingly with the rather densely stained protoplasts of the companion and the phloem parenchyma cells. Sieve tube elements are generally associated with companion cells (fig. 3). The sieve tube element has transverse end walls (fig. 3) with a simple sieve plate and aggregate or scattered lateral sieve areas.

3.1.12.2.2 Companion cell

One to four companion cells are present. A single companion cell accompanying a sieve tube element extends from its one end to the other (fig. 3) or a longitudinal row of 2-4 companion cells completely aligned on any one of the lateral walls are present. The companion cell is uninucleate with dense cytoplasmic contents. The nucleus is round with a prominent nucleolus. Starch grains are absent.

3.1.12.2.3 Phloem parenchyma

This includes the parenchyma cells of the phloem complexes (fig. 1) and those of the interspersed areas. The contiguous and non-contiguous parenchyma cells may be as large as or larger than the sieve tube elements. One or two rows of broad and radially elongated interspersed parenchyma cells are present between the adjacent phloem complexes in the basal region (fig. 1). But towards the middle and distal

regions of the petiole few rows of small, almost rounded parenchyma cells separate the complexes. They differ from the contiguous parenchyma cells by their location, size and lighter staining.

3.1.12.2.3 Slime

The P-protein and other contents aggregate and accumulate at the sieve plate to form a plug. The mercuric bromophenol blue and Coomassie brilliant blue, stain the plugs, indicating a positive reaction for phloem specific proteins. The plug is generally observed only on any one side of the sieve plate.

3.1.12.3 Development of protophloem fibres

In a young petiole protophloem fibres differentiate from the parenchymatous precursor cells located outside the first formed sieve tube elements. They cap the phloem complexes (fig.1). In a 3 mm elongating young petiole these cells are small without intercellular spaces. In longitudinal view they appear elongated and narrow with transverse end walls. They become prominently large and elongated parallel with the longitudinal axis of the petiole. They also undergo some apical intrusive growth and show tapering end walls (fig. 6). They gradually develop wall thickening and lignification during the last phase of elongation of the petiole and develop as phloem fibres.

3.1.12.4 Developmental changes in protophloem

With the elongation of the petiole, radial seriation in the procambium of the young vascular strand becomes increasingly prominent and is clearly evident in the transection of a 2.5 cm long petiole (Section II, fig. 27). The protophloem sieve tube elements and their associated companion cells are physically stretched and become narrow in the elongating petiole. The sieve tube elements and companion cells close to the protophloem fibre precursors appear to have a short functional existence. Obliteration of protophloem sieve tube elements is first observed in a 2.5 cm long petiole. The other protophloem sieve tube elements and companion cells adaxial to the previously formed ones elongate and become narrow. Dimensional changes in the protophloem sieve tube elements during the elongation of the petiole are shown in table IV. Most of the protophloem sieve tube elements were obliterated in a 14.4 cm long one month old mature petiole in which mature protophloem fibres were present.

3.1.12.5 Metaphloem

In a 6 mm young elongating petiole the development of the earliest metaxylem was noticed. The transition from protophloem to metaphloem is not sudden. With the progressive development of metaphloem there is also a gradual increase in the diameter of its sieve elements. The radial seriation in the procambium of the vascular strand is

prominent as reported in a 2.5 cm long petiole (Section II, fig. 27).

There were no distinct criteria to delimit the last formed protophloem from first formed metaphloem sieve elements in the petiole of Gmelina arborea.

3.1.12.5.1 Sieve tube element

For dimensional details the last formed sieve tube elements, normally those which are present near the distinct metaxylem elements have been considered. The metaphloem sieve tube elements show rounded starch grains with a clear lumen mainly collected towards the sieve plate (fig. 4). They are absent in companion cells. When stained with I_2KI solution, they stained reddish brown and are easily distinguished from the blackish brown large starch grains of the cortical parenchyma (fig. 4). They are occasionally scattered in the lumen of the sieve tube element (fig. 5). Sieve tube elements show companion cells on one side or on either side of their lateral walls.

The sieve tube elements mostly have transverse or slightly oblique end walls with simple sieve plates. Metaphloem sieve tube elements are 61.2 - 187 µm long and 8-12 µm wide.

3.1.12.5.2 Companion cell

The number of companion cells, their size and arrangement show variation. Each sieve tube element is associated with 1-8 companion cells. They occur on one side (figs. 5 and 13) or on both the sides of the sieve tube element (figs. 15 - 17). The total number of companion cells for a sieve tube element can be a maximum of four on each lateral side (fig. 14). Longitudinally a row of companion cells may extend from one end of the sieve tube element to the other end (figs. 5, 13-16) or may not extend fully upto its end (fig. 17). When a vertical row of two companion cells are present, the length of one companion cell is almost half the length of the sieve tube element.

The companion cells are 22.1-127 μm long and 3.4-5.1 μm wide. It has a round nucleus and a nucleolus.

3.1.12.5.3 Slime

The contents of a sieve tube element are easily disturbed during processing of the material and hence exhibit varying configurations. They accumulate at the sieve plate to form a plug or else show varying densities of condensation at different levels away from the sieve plate.

The configurations of the aggregated slime may show funnel (fig. 18), or cylindrical (fig. 19) form. At times the dense mass of slime may be collected at different loci leaving spaces without any apparent contents

(figs. 19 and 20). The cylindrical and funnel form may also show one (figs. 21-23) or more cavities (fig. 24). Figure 25 shows the contents accumulated to form a dense cylindrical mass at the sieve plate.

3.1.12.6 Secondary phloem

The primary development of the phloem is normally completed in one month old petiole which ceases to elongate. As mentioned in Section II during the last phase of petiole elongation there is a transition from primary to secondary phloem development, during which few marked developmental changes take place. Although the cessation of petiole elongation can be correlated with these events, the changes take place gradually within the mature petiole and these changes do not necessarily coincide with one another.

A group of phloem cells in a complex, derived from an initial can be distinguished by its thick radial and tangential walls (fig. 26).

3.1.12.6.1 Sieve tube element

Sieve tube elements are short and they vary in length. Their range of length is similar to that of their precursors, the transit cells. The end walls are transverse or slightly oblique with a simple or a compound sieve plate, and they have aggregate scattered or solitary lateral sieve areas (figs. 28 and 29). Callose is present on the sieve areas.

Between the sieve tube element and companion cell lateral sieve areas are present only on the sieve element side. Companion cells are aligned on one or two lateral sides.

3.1.12.6.2 Companion cell

One to four companion cells are present for a sieve tube element, of which a maximum of two are present on either side.

Secondary growth becomes well established in the mature petiole by about two months and for dimensional relationship measurements have been taken from this petiole. Table V gives the dimensional relationship of the secondary phloem elements in the three different regions of the petiole. Sieve tube elements in the middle region are longer than those of the basal and distal regions.

3.1.12.6.3 Slime

P-protein and other contents of the sieve tube element are collected in the form of a plug at or near the sieve plate. Plastids mostly the starch grains also accumulate at the sieve plate or may be occasionally distributed in the lumen.

The configurations of the slime plug in the sectioned material vary. It may be funnel (fig. 32) or cylindrical form. Occasionally it coalesces to form a dense mass away from the sieve plate (fig. 33). The dense mass may be

collected at different loci leaving vacuole like spaces without any contents (fig. 34). Figure 30 shows contents in the peripheral position surrounding a clear lumen. Figure 35 also indicates such a lumen in longitudinal section. Figure 31 shows an artefact where more vacuole like cavities are observed. The funnel and cylindrical configuration of the slime plug may show varying densities of stained material (figs. 36-38, 39-43). Slime plugs with projections are also observed (figs. 44-46). Probably the projections represent a disturbed state of lumen with the contents accumulated at the end. Senescent petiole also shows sieve tube elements with cylindrical or funnel shaped configuration of slime plug. Sometimes the contents with massive callose deposition may almost completely fill up the entire lumen of the sieve tube element (figs. 47-49).

3.1.12.7 Obliteration

In the basal region of a mature petiole the primary and secondary phloem complexes are demarcated by a row of parenchyma cells. Hence the median strand in this region of the petiole has been selected to study obliteration.

The cortical strands do not show such a demarcation. The differentiation of vascular elements first noticed in the median strand in the basal region of the second leaf primordium from the shoot apex, begins with the development of few protophloem sieve elements followed by protoxylem elements. In a 8 mm long petiole at the third node from the

shoot apex the cortical strands are procambial and are present only in the basal region (Section II, fig. 13). In the petiole at the fourth node the cortical strands show both protoxylem and protophloem (Section II, fig. 14).

Protophloem differentiation continues till the radial seriation is apparent in the procambium between xylem and phloem. The first signs of obliteration in the median strand are noticed in a 2.5 cm long petiole. In a 7 cm long young elongating petiole the cortical strand shows signs of obliteration of the protophloem sieve tube elements and companion cells in the extreme basal region (fig. 50). No signs of obliteration are noticed in the other part of the basal region (fig. 51). The protophloem sieve tube elements mature during the early elongation of the petiole and due to physical stretching during the further elongation of the petiole they become stretched, functionless and ultimately crushed.

After the obliteration of the protophloem sieve elements, the parenchyma cells surrounding the sieve elements enlarge and align with parenchymatous precursors of phloem fibres/collenchymatous cells.

Within one month of growth the petiole ceases to elongate during which the primary phloem development is completed, after which the secondary phloem development begins. During the elongation of the petiole the protophloem

sieve elements obliterate and protophloem is mostly represented by protophloem fibres/collenchymatous cells and few parenchyma cells (fig. 52). When the sieve tube elements are appressed between the enlarging parenchyma cells, thickened places with callose spots temporarily indicate the former positions of obliterated sieve elements in the median and cortical strand.

Metaphloem remains relatively unchanged till one or two months after which obliteration begins (fig. 52). When the phloem is about 4-5 months old metaphloem is almost completely crushed.

The median strand in the four month old petiole shows the primary phloem completely crushed and only secondary phloem complex remains. As there is no demarcation of primary and secondary phloem complexes in the cortical strand, it could not be ascertained if metaphloem sieve elements were obliterated.

By the end of the growth period of the leaf i.e., after 6 months the sites of primary sieve elements is observed as thickened places between the secondary phloem complexes and the protophloem fibres/collenchyma (fig. 53).

Obliteration in protophloem and metaphloem is almost similar. Due to the elongation of the petiole the protophloem sieve tube elements are prone to stretching and hence the lumen of the sieve tube element is narrowed

(fig. 54). Cell wall of the sieve tube element appears crushed and disfigured (fig. 57) indicating excessive pressure exerted by the adjoining parenchyma cells. Later the empty lumen completely narrows, with the opposite lateral walls closely appressed and only the region of the sieve plate bulged out with heavy deposition of callose. Thickened regions sandwiched between the parenchyma cells (fig. 58) indicates the sites of completely obliterated sieve elements. In the metaphloem sieve tube elements other than physical stretching some of them become functionless by heavy deposition of callose at the lateral sieve areas (fig. 59) and the sieve plate (fig. 55). At times the lumen of the sieve tube element is almost completely masked with callose (fig. 56). When the petiole becomes senescent all the sieve elements are found to be blocked by callose. Figures 60-65 show phloic strands in the different stages of the petiole. Obliteration of sieve elements is first observed in a 7 cm long petiole (fig. 61).

3.1.12.8 Secondary phloem in the bark

The secondary phloem in the bark of Gmelina arborea consists of the axial and ray systems. The axial system consists of sieve tube elements, companion cells and phloem parenchyma cells almost arranged in a radial seriation (fig. 66). Comparison of sieve tube elements in the petiole and the bark has been given in table VI.

3.1.12.8.1 Sieve tube element

The sieve tube elements are conspicuously large with a clear lumen (fig. 66) with transverse or oblique end walls and simple sieve plate. They are 240-430 μm long and 15-39 μm wide.

3.1.12.8.2 Companion cell

The companion cells are relatively very small compared to the sieve tube elements and are densely stained. The number of companion cells varies from one to seven arranged in a longitudinal series on one side or on either of the lateral sides of the sieve tube element (figs. 67-71). Sometimes a single companion cell is found at one corner (fig. 70), or three very small companion cells somewhere in the middle region (fig. 69) of the sieve tube element. At times 7 small companion cells are radially aligned extending from one end of the sieve tube element to the other (fig. 68). They are 87-430 μm long and 8-12 μm wide.

3.1.12.8.3 Phloem parenchyma

Phloem parenchyma cells are intermediate in size between the sieve tube element and companion cell and are scattered more or less uniformly throughout the phloem (fig. 66). Rays are multiseriate.

3.1.12.8.4 Slime

The configuration of the slime plug in the sectioned material varies from scanty (fig. 71) to strand-form (fig. 72). The strand-form at times extends from one end of the sieve tube element to the other (fig. 72).

Table IV. Dimensions of protophloem and metaphloem sieve tube elements and companion cells during the elongation of the petiole. Dimensions are in μm.

Length of petiole in cm	Regions of the petiole	Protophloem				Metaphloem			
		Sieve tube element		Companion cell		Sieve tube element		Companion cell	
		Length	Width	Length	Width	Length	Width	Length	Width
0.3		43.0- 65.0	4-7	20.0- 64.0	1.7-3				
0.6		40.0- 68.3	4-8	13.6- 68.0	1.7-3				
2.5	Basal	153.8-282.0	5.2-6	34.4-103.0	2-3	86.0-140.0	9.4-10.9	22.1- 91.0	3.4-5.1
	Middle	122.0-272.0	5.1-6	25.5-119.0	3.4-4.2	90.0-187.0	8.5-10.2	22.1-127.0	3.4-5.1
	Distal	141.1-217.6	5.1-6.2	20.4- 59.5	3-4	61.2-153.0	8.0-10.2	34.0- 71.4	3.4-5.1
7.0	Basal	207.0-425.0	1.5-3	57.8-187.0	2-3	122.4-183.0	10.2-12	40.8-105.0	3.4-5.1
	Middle	173.5-286.0	4-5	44.2-102	2-3	112.0-180.2	10.2-11.9	32.3- 63.0	3.4-5.1
	Distal	153.0-235.0	4-5	42.5-105	2-3	69.7-184.0	10.2-11.9	25.5-61.2	3.4-5.1

TABLE V. Dimensional relationship of the secondary phloem sieve tube elements and companion cells in a two month old petiole. All the measurements are in μm

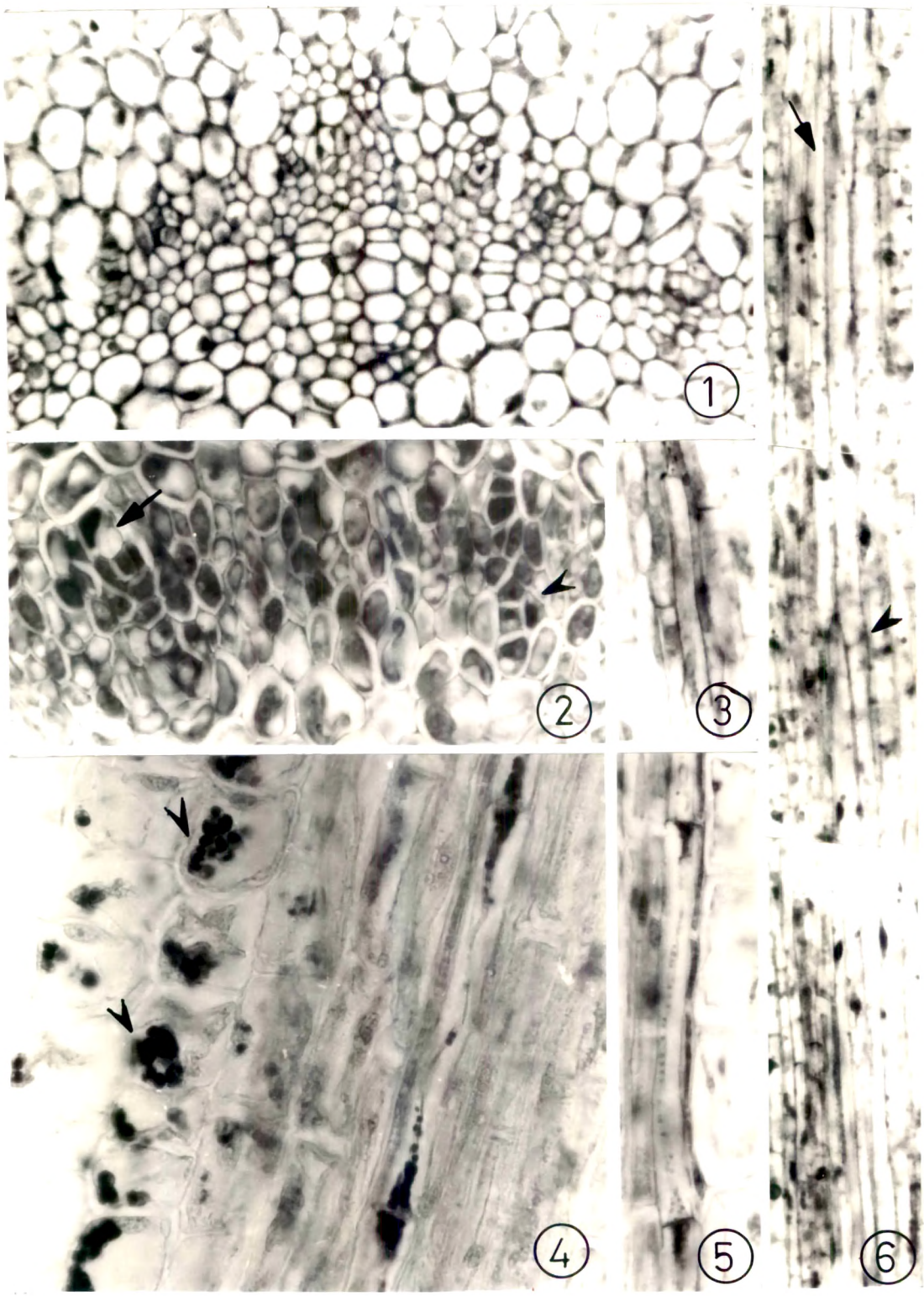
Regions	Sieve tube element		Companion cell	
	Length	Width	Length	Width
Basal	80-164	9-12	18-45	3-7.5
Middle	87-174	6-12	30-66	3-6
Distal	70-164	9-12	24-84	3-7.5

Table VI. Comparative structure of the secondary phloem sieve tube elements and companion cells of the petiole and stem

	Petiole	Stem
1) Tissue types	Axial system	Axial system and ray system
2) Components	Sieve tube elements, companion cells and axial parenchyma	Sieve tube elements, companion cells, axial parenahyma and ray parenachyma
3) Sieve tube element		
a) Length µm	70-174	240-430
b) Width µm	6-12	15-39
c) End walls	Transverse or oblique	Transverse or oblique
4) Companion cell		
a) Number	1-7	1-7
b) Length µm	18-84	87-430
c) Width µm	7-7.5	8-12

Plate 16

Figs. 1 - 6.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Figs. 7 - 12.

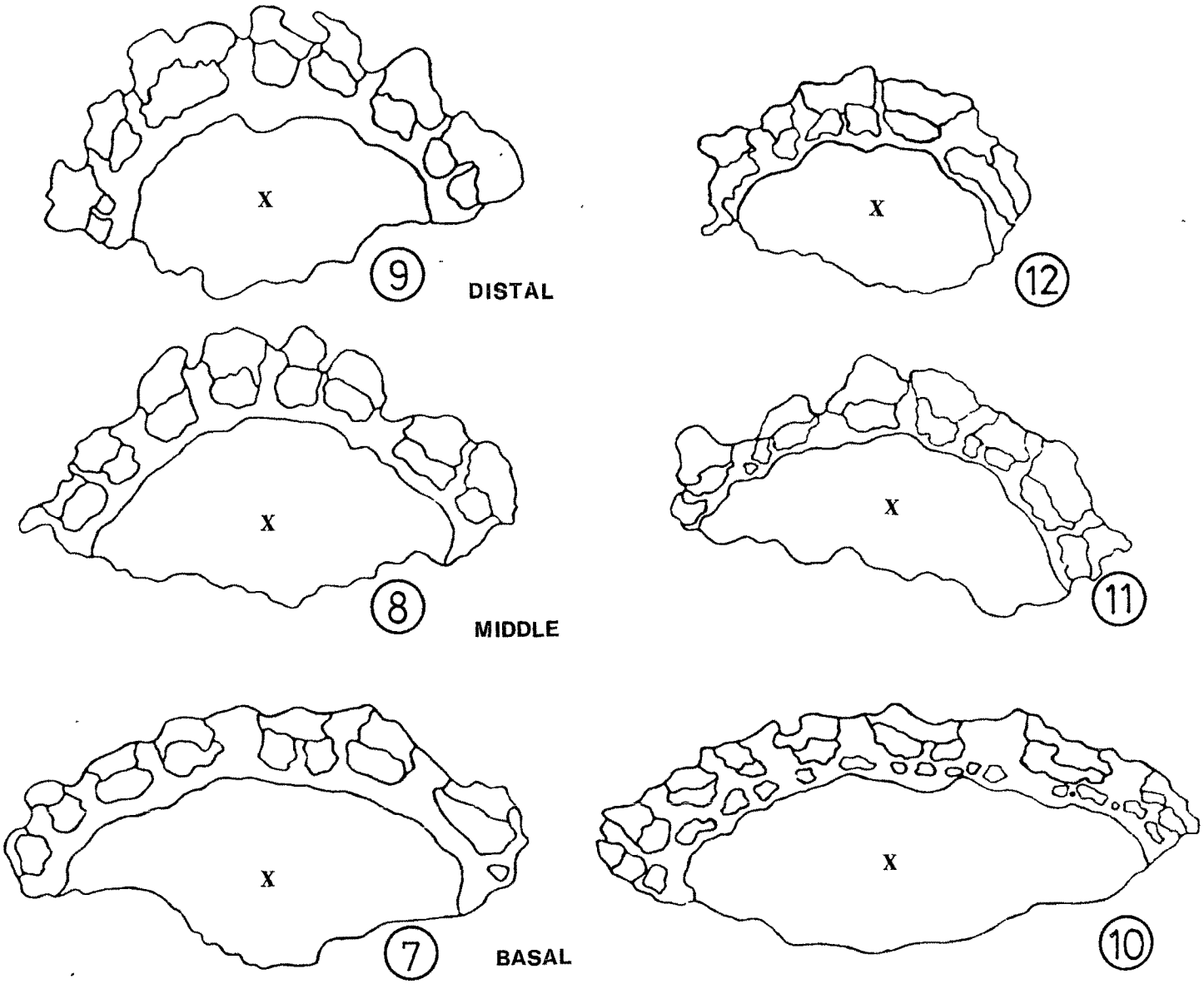
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Plate 17

Figs. 13 - 29.

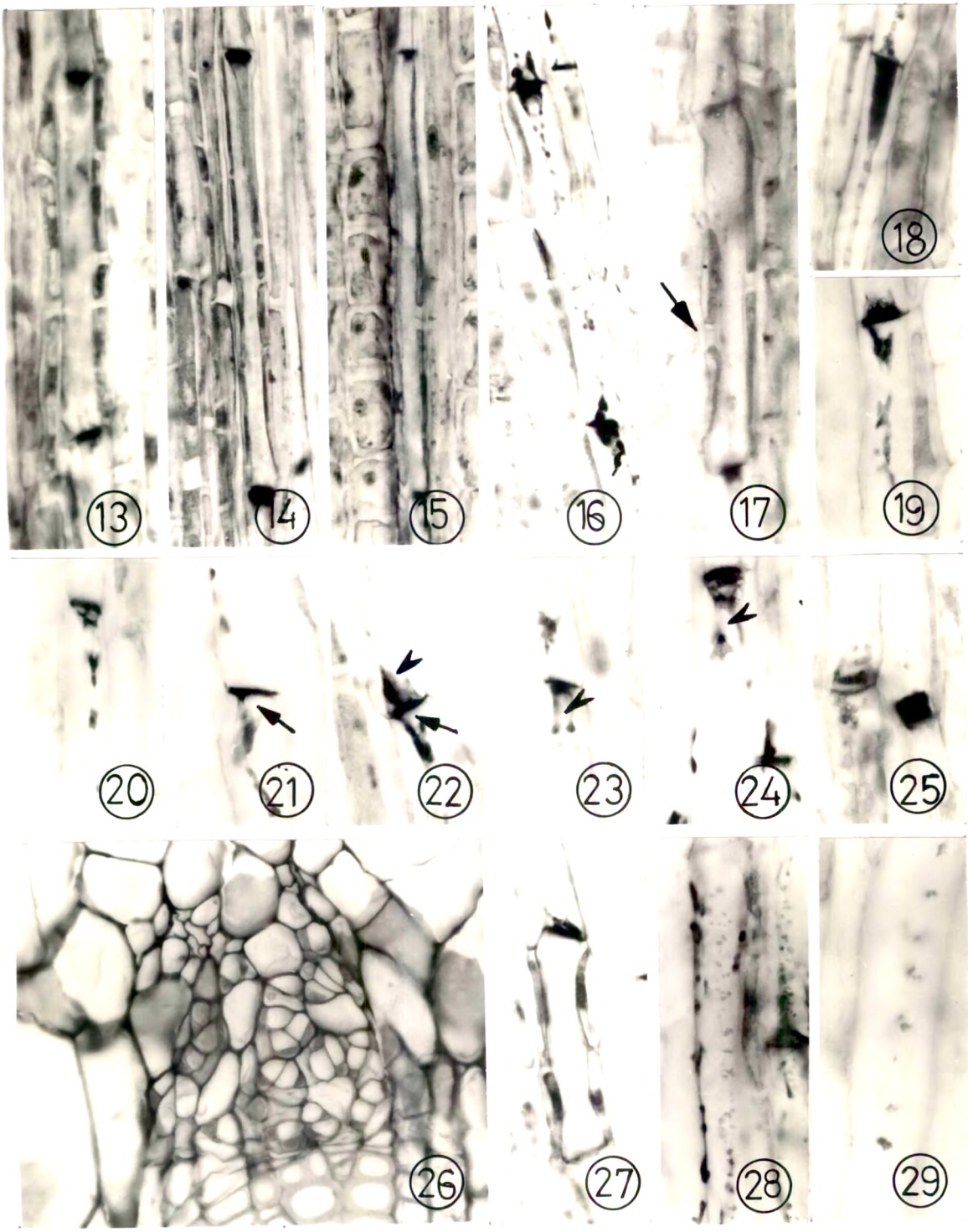
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Plate 18

Figs. 30 - 49.

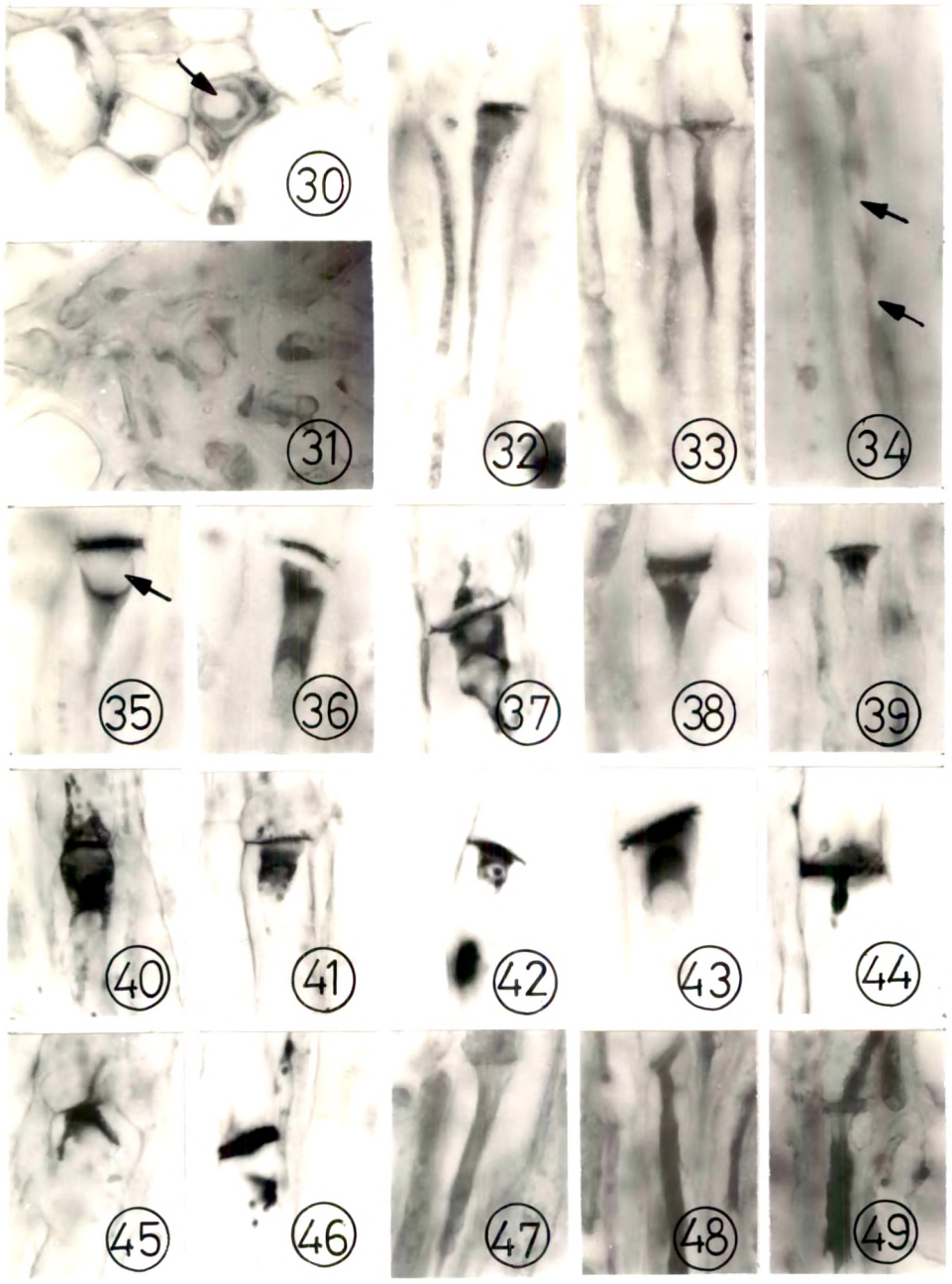
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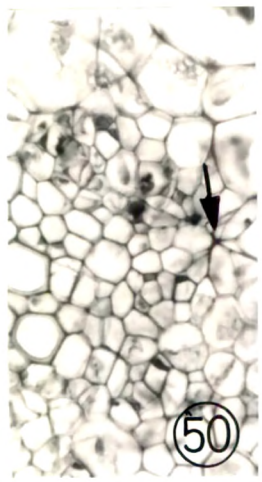
Plate 19

Figs. 50 - 59.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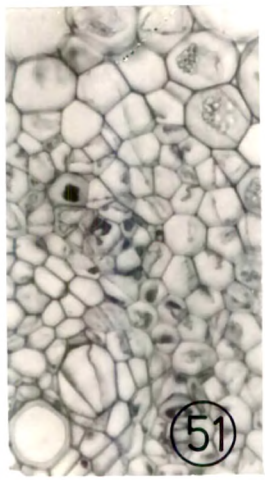
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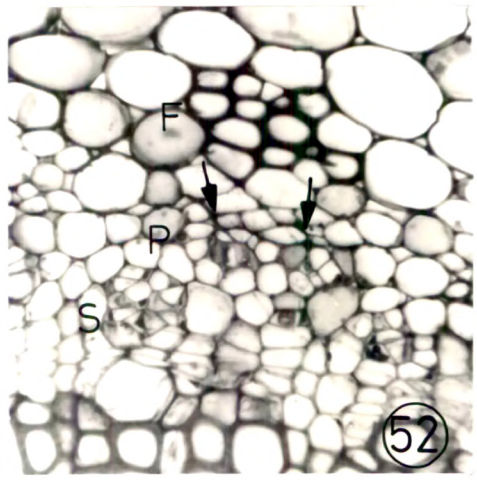
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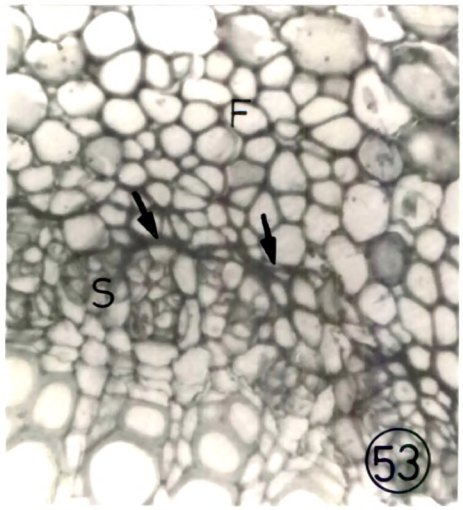
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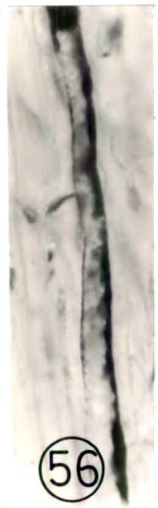
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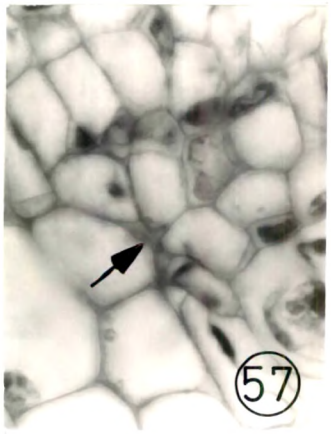
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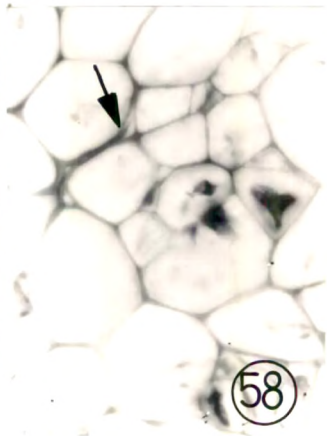
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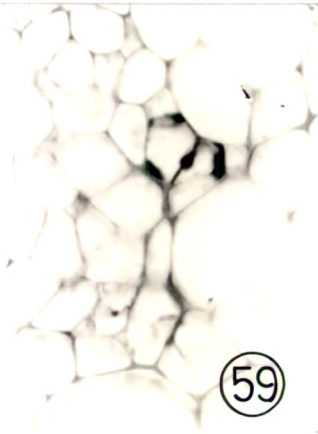
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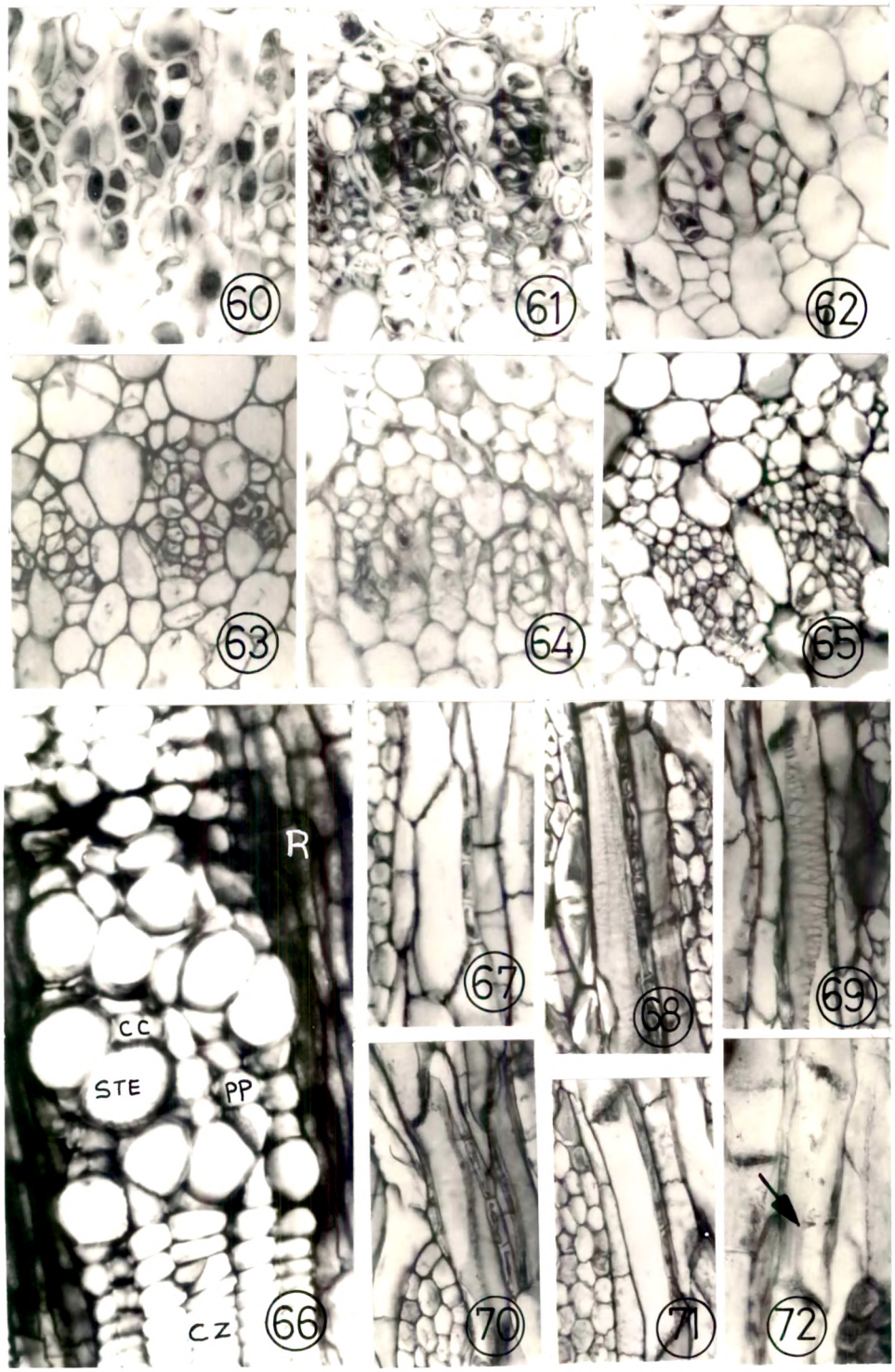
Plate 20

Figs. 60 - 72.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3.2 Tabebuia rosea DC. (Bignoniaceae)

Section IV

3.2.1 General morphology

Tabebuia rosea DC. commonly known as 'Pink Pui' or 'Trumpet tree', is an ornamental tree grown chiefly for their showy flowers. They are upright - evergreen trees with large digitately palmately compound leaves in opposite, decussate manner (figs. 1 and 2). Leaves are pentafoliolate with five leaflets, median and two pairs of laterals, rarely trifoliate with three leaflets, median and a pair of laterals. All the leaflets are petiolulate.

A mature petiole has a pulvinus base and to its dilated distal end petiolules of the five leaflets are articulated. The petiole is about 13-20 cm long. Leaflets are entire with their apex slightly acuminate. The petiolules of the median and the first laterals have a pulvinus base and a dilated distal end to which the lamina is attached.

The petiolules of the median and first lateral leaflets are about 2.5-4.5 cm and 1.2-4.0 cm long respectively. The petiolule of the second pair of lateral leaflet is about 0.3-0.5 cm long. Often the petiolule of the second lateral leaflet is very short and almost ill defined.

According to the classification of Longmann and Jenik (1974) Tabebuia rosea can be grouped as the periodic growth

evergreen type of tree wherein the leaf shedding occurs long after bud opening. The life span of the leaves varies from 7 - 8 months.

3.2.2 Sequential elongation of the petiole

The rates of elongation of the three regions of the petiole and of the petiolules were measured at a fixed time for about 16 days. Among the 20 petioles studied the longest petiole measured was 13.5 cm long and hence this has been considered a typical one. The length of the petiole when marked was 5 cm. The pattern of elongation is the same in the other petioles and petiolules.

The petiole ceases elongation within 15 days from the day of marking. Of the three different regions marked equally in the young petiole, the distal region elongates more and ceases its elongation after the simultaneous cessation of elongation of the basal and middle region of the petiole (fig.3). The basal and middle region of the petiole ceases its elongation within 3-4 days, with the middle region elongating more than the basal region. The distal region of the petiole still elongates further and ceases its elongation by 13-14 days from the day of marking.

Of the five leaflets, the petiolules of the median and first laterals elongate more than those of the second laterals (fig.4). Petiolules of the second laterals ceased elongation within a week followed by the petiolules of the

median and first laterals which ceased elongation by the eleventh or twelfth day from the day of marking.

3.2.3 Internode-node-petiole vascular continuum

The growing shoot of Tabebuia rosea produces petiolate leaves in an opposite and decussate manner, each pair separated at maturity by a clearly defined internode. The vascular system of the petiole at the third node from the shoot apex is discrete and hence the continuum has been studied in this node.

3.2.3.1 Nodal anatomy

The young stem is rectangular consisting of about 42 vascular strands including both collateral and phloic strands, peripherally arranged. The two opposite leaves depart from the main stem axis at the same levels. All the observations are described from the direction of internode-node-petiole continuum.

In the internode region at a distance of 540 um from the apical summit, the primary vascular system consists of 42 more or less distinct collateral and phloic strands. Five strands, each on the opposite sides of the internode are associated with the organization of the petiole vasculature. They include three collateral, i.e., a median and two laterals one on either side and adaxial to it and a pair of phloic strands one on each on the adaxial side of the

lateral (fig.5). The two phloic strands when traced basally towards the fifth node was found to be a divergence from the two lateral collateral strands. The group of five procambial strands, each lying adaxial to the phloic strands are associated with the formation of axillary bud vasculature (fig. 5, arrow).

3.2.3.2 Nodal vasculature

At a distance of about 80 µm from the internode, these strands 1,2,3,4 and 5 together arch out separating from the main stem vasculature and diverge towards the petiole base. The vascular system for the petiole and the axillary bud together with that of the internode form a clover shape. The incurve formed demarcates the two system (fig. 6, arrow). Further up 50 µm away, the axillary vascular system is completely separated from that of the petiole (fig. 7). Now the three vascular systems, i.e., of the petiole, axillary bud and stem axis are distinguished. At a distance of 40 µm further, the axillary bud alongwith the petiole separates from the main stem, and subsequently the two are separated.

Thus each opposite leaf is supplied by a median and two lateral collaterals and two phloic strands associated with a single leaf gap. Hence the leaf trace in Tabebuia rosea is unilacunar, multitrace.

3.2.3.3 Petiole vasculature

All the five trace strands enter the petiole base at

which level the median strand gives rise to a phloic strand on its either side. The lateral strands on either side of the median also anastomose (fig.15) thus forming 11 strands arranged in a crescent (fig.8). At a distance of 70 µm from the above the petiole completely separates from the axillary branch. Subsequently 2A and 4A lying immediately adjacent to the phloic strand at the end of the crescent bifurcate (fig.15) giving rise to two collateral strands 2D, 2C and 4D, 4C respectively, hence forming 13 vascular strands. The vascular strands are now organized in the form of a circle with an open end (fig. 9). The vascular strands are discrete and undivided till the distal end of the petiole.

3.2.3.3.1 Number and arrangement of the vascular strands

They are discrete, widely separated out, interspersed with interfascicular parenchyma and organized in the form of a crescent with incurved ends at the basal region of the petiole (fig. 8), the open side of which faces the adaxial surface of the petiole. The vascular strands include five collaterals and six phloic ones.

The arc of vascular strands in the middle region appears more rounded. The number of strands increases to seven collaterals and six phloic ones, because of the bifurcation of the 2A and 4B strands (fig. 9).

The arrangement of the vascular strands remains unchanged in the distal region (fig. 10). The two phloic

strands 1B, 1A and 5A, 5B lying at the open end unite (fig.5) and hence 11 strands, seven collaterals and four phloic ones are present in the distal region.

3.2.3.4 Petiolule vasculature

Towards the extreme distal end of the petiole the two phloic strands 1 and 5 lying at the adaxial open end of the vascular system diverge from the petiole vascular system (fig. 11). At a distance of about 50 μm from the divergence the collaterals 2D and 4D on the abaxial side near 2C and 4C respectively, traverse away from the petiolar vascular ring and fuse with strands 1 and 5 and traverse into the base of the petiolule of the second lateral leaflet.

Petiolule of the second laterals separate out first. At a distance of about 50 um from the separation of the second laterals petiolule strands 2C and 4C traverse and fuse with the phloic strands 3A and 3C respectively, lying on either side of the median strand (fig. 12). The two collaterals 2B and 4B give rise to phloic strands 2E, 2G and 4E, 4G on their either side (fig. 12). At a distance of about 120 μm from the above the median collateral strand with a phloic strand on its either side traverses into the median petiolule and the lateral collateral strands with phloic strand on their side traverse into the base of the petiolule of the first lateral leaflet (figs. 13 and 14). This condition remains till the distal end of the petiolule.

3.2.4 Vascular system of a mature petiole

The petiole vascular system of the young leaf is open and the vascular strands are well separated by interfascicular parenchyma. Distally in the petiole the vascular cylinder decrease in size as strands diverge to vascularize the petiolule of the lateral leaflet pair. However it retains its open structure. The petiole vascular system of a mature leaf in contrast exhibits a closed structure.

3.2.4.1 Nodal anatomy

In the nodal region the axial vascular system forms a closed cylinder with extensive secondary vascular development. The either side of the node responsible to give rise to the petiole vasculature shows an arc of vascular strands consisting of three collateral (fig. 16). They were identified on the basis of the number of primary xylem groups.

3.2.4.2 Nodal vasculature

The three collateral strands arranged in an arc, during the course of their traverse divide many times forming an arc of discrete vascular strands (figs. 19 and 20). They gradually arch out and separate from the stem axis and enter into the petiole forming a parenchymatous gap (fig. 21). Hence the condition in the mature node is also unilacunar, multitrace.

3.2.4.3 Petiole vasculature

The transection of a mature petiole is almost circular in outline. The epidermis shows a single layer of barrel shaped compact cells. Two-armed 'T' shaped unicellular epidermal hairs are present. A hypodermis of 8-10 layered collenchyma is present. The ground tissue is parenchymatous. The vascular system in the petiole delimits the cortex from the pith.

3.2.4.3.1 Number and arrangement of vascular strands

In the pulvinus base there are 23 collateral strands, compactly arranged in a ring (fig. 22) and interspersed by uniseriate or biseriate ray parenchyma. In the basal and middle region due to the lignification of the xylem parenchyma and ray parenchyma and development of xylem fibres, the vascular system forms a closed ring (figs. 23-25). In the proximal distal region the vascular system cylinder has a wavy outline (fig. 26). Towards the extreme distal region of the petiole the vascular strands are again discrete with elongated ray parenchyma. They further diverge and organize to form the vasculature of the five petiolules (fig. 27).

3.2.5 Leaf architecture

The basic axis of orientation in the leaf is towards the base (downward) and the curvature of the leaf elements is

generally convex. The lamina of the second laterals shows a concave curvature of the leaf margin (fig. 28). The leaf is palmately compound with the laminar subunits attached at the apex of the petiole which is distinctly swollen. Generally the lamina and the base are symmetrical in both the median and the first laterals. The lamina of the second lateral and its base is asymmetrical (fig.28). The lamina of the leaflets is elliptic with the axis of the greatest width perpendicular to the approximate mid point of the leaf axis. The apex of the lamina is acute and the base obtuse. The margin of the lamina is entire forming a smooth line without any noticeable projections, with a coriaceous texture, leathery, thick and stiff.

The petiole and petiolules are inflated with a thickened base, the pulvinus and a thickened distal end to which petiolules are attached, each with a lamina.

3.2.5.1 Venation

Venation is pinnate with a single primary vein (mid vein) serving as the origin for the higher order venation. The course of the mid vein is straight (fig. 28), lacking noticeable curvature or change in course and is unbranched lacking ramifications of primary rank.

3.2.5.1.1 Secondary veins

Secondary veins are camptodromous with the veins not terminating at the margin but joined together in a series

of prominent arches. Hence it is categorized as the brochidodromous type.

The angle of divergence measured between the branch and the continuation of the source vein (mid vein) above the point of branching is acute and wide. The course of the secondary veins is curved bending in an arc (fig. 28). It is uniform with the arc smooth and increasing in degree of curvature towards the margin of the leaf. The secondary veins join the superadjacent secondary vein at right angle and are also sometimes enclosed by secondary arches.

3.2.5.1.2 Tertiary veins

The tertiary veins may be ramified, branching into higher order (fig. 30), with or without rejoining the secondary veins. The branching is transverse, oriented across intercostal area. Some of the tertiary veins are percurrent joining with the tertiary veins of the opposite secondaries. The course of the tertiary veins is sinuous with a repeatedly changing direction of curvature (fig. 30). Its relationship to the mid vein is oblique tending in an obtuse angle to the mid vein.

3.2.5.1.3 Quarternary and quinternary veins

The quarternary veins are distinct and they have a relatively randomly oriented course. The quinternary veins are mainly orthogonal arising at right angle.

The areoles show mostly no veinlets (fig. 30). Some of them show simple veinlets, without branches and mostly curved or linear (fig. 30). Areoles are imperfect with meshes of irregular shape, more or less variable in size and pentagonal in shape. The arrangement of the areoles does not show a similar pattern of alignment within particular blocks or domain (fig. 30).

The marginal ultimate venation is with a fimbrial vein. The higher order veins are fused into a vein running just inside the margin (fig. 29).

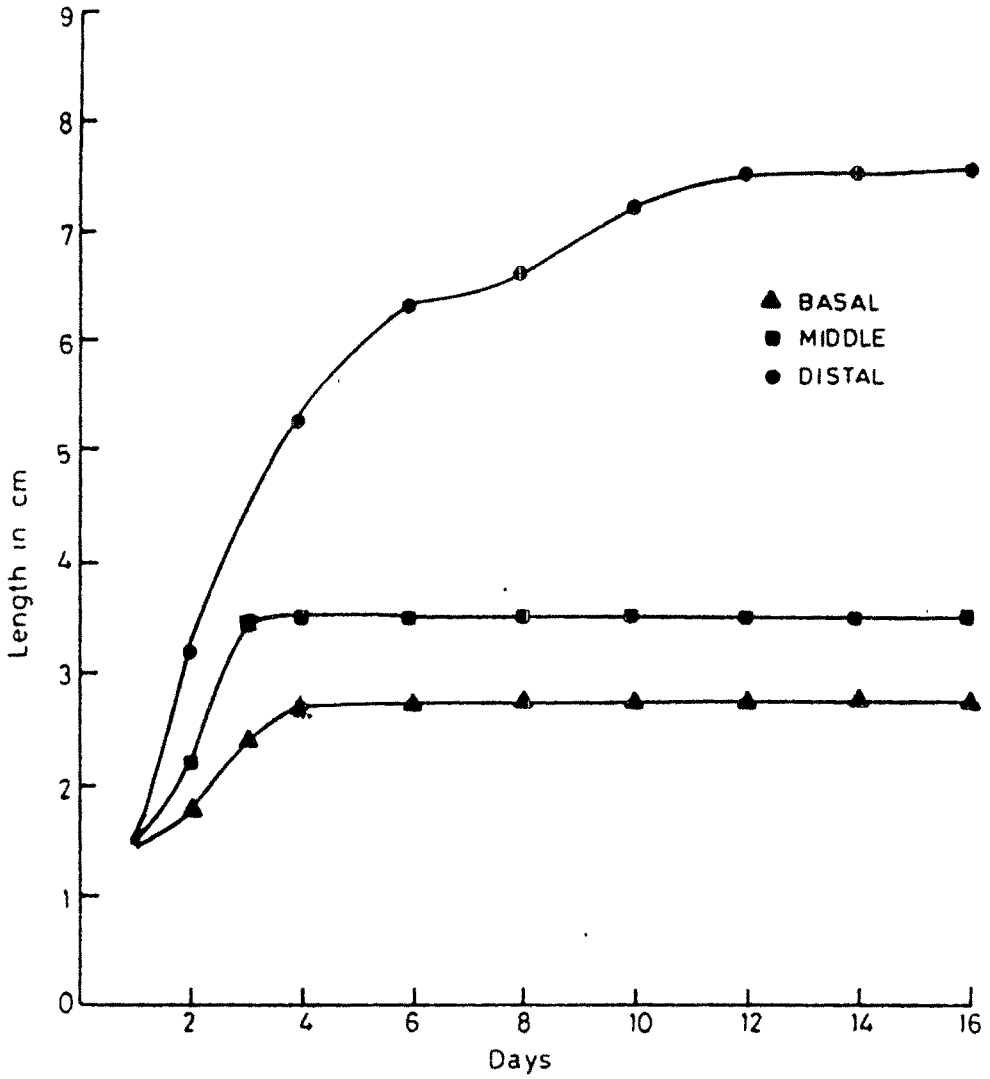
Plate 21

Figs. 1 and 2.


1


2

Fig. 3.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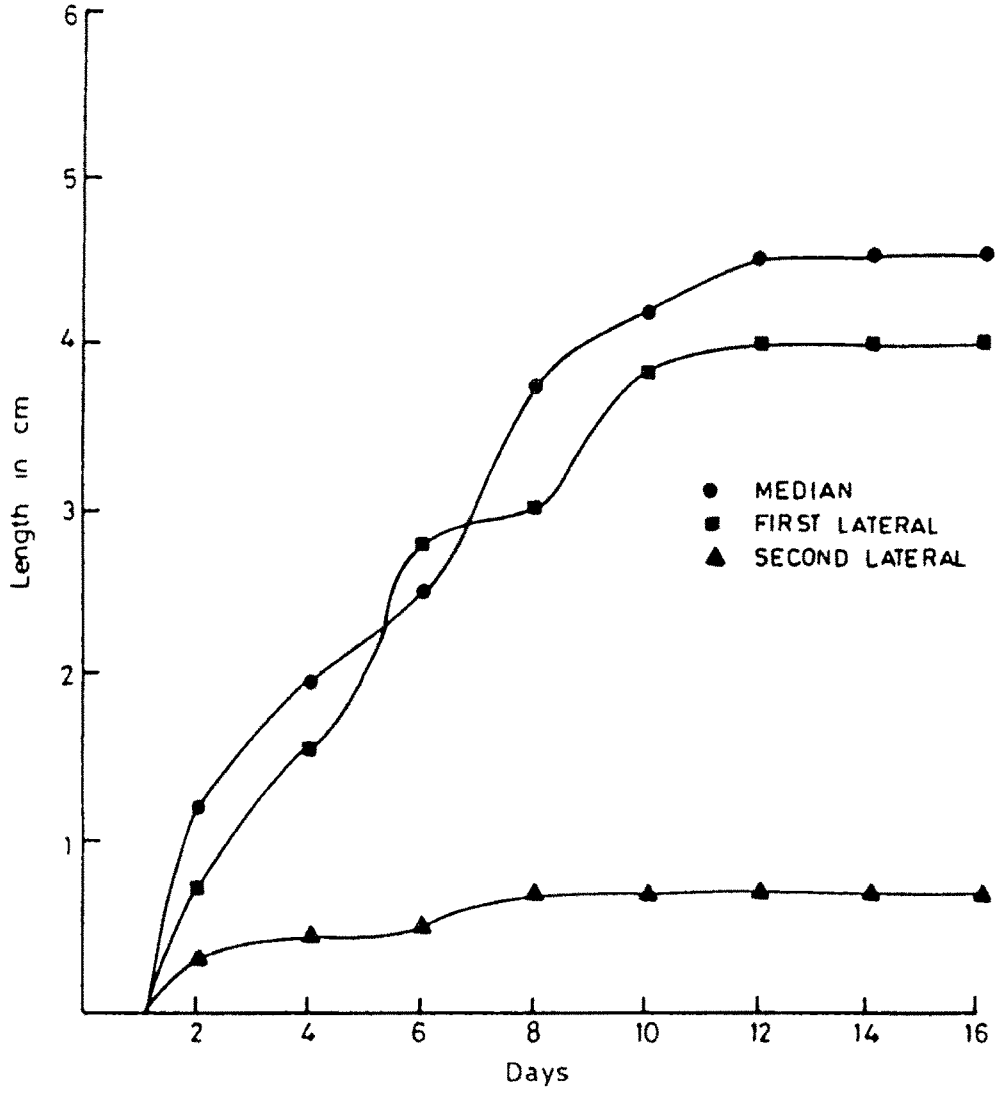
(3)

Fig. 4.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Length in cm
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(4)

Figs. 5 - 14.

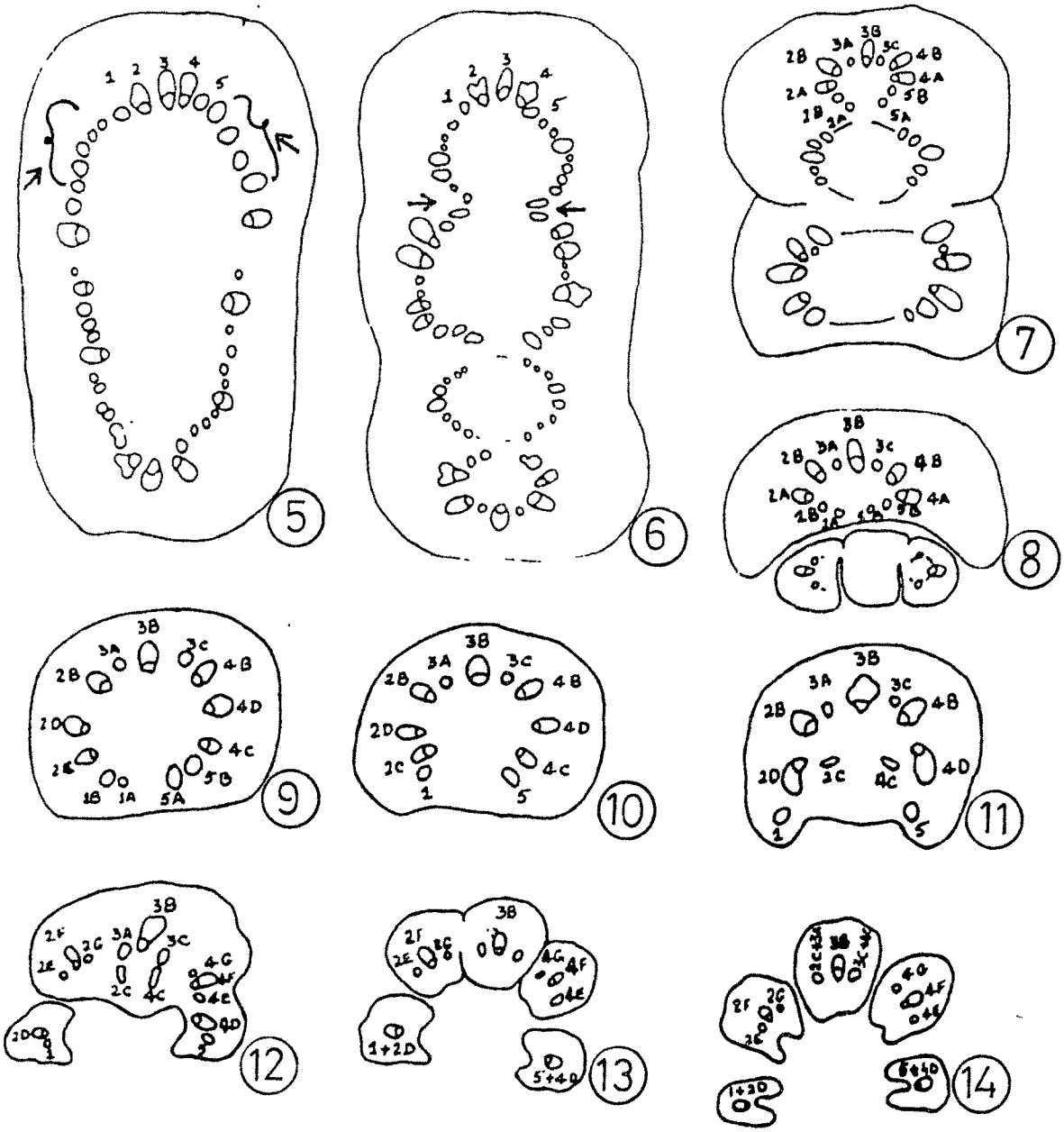
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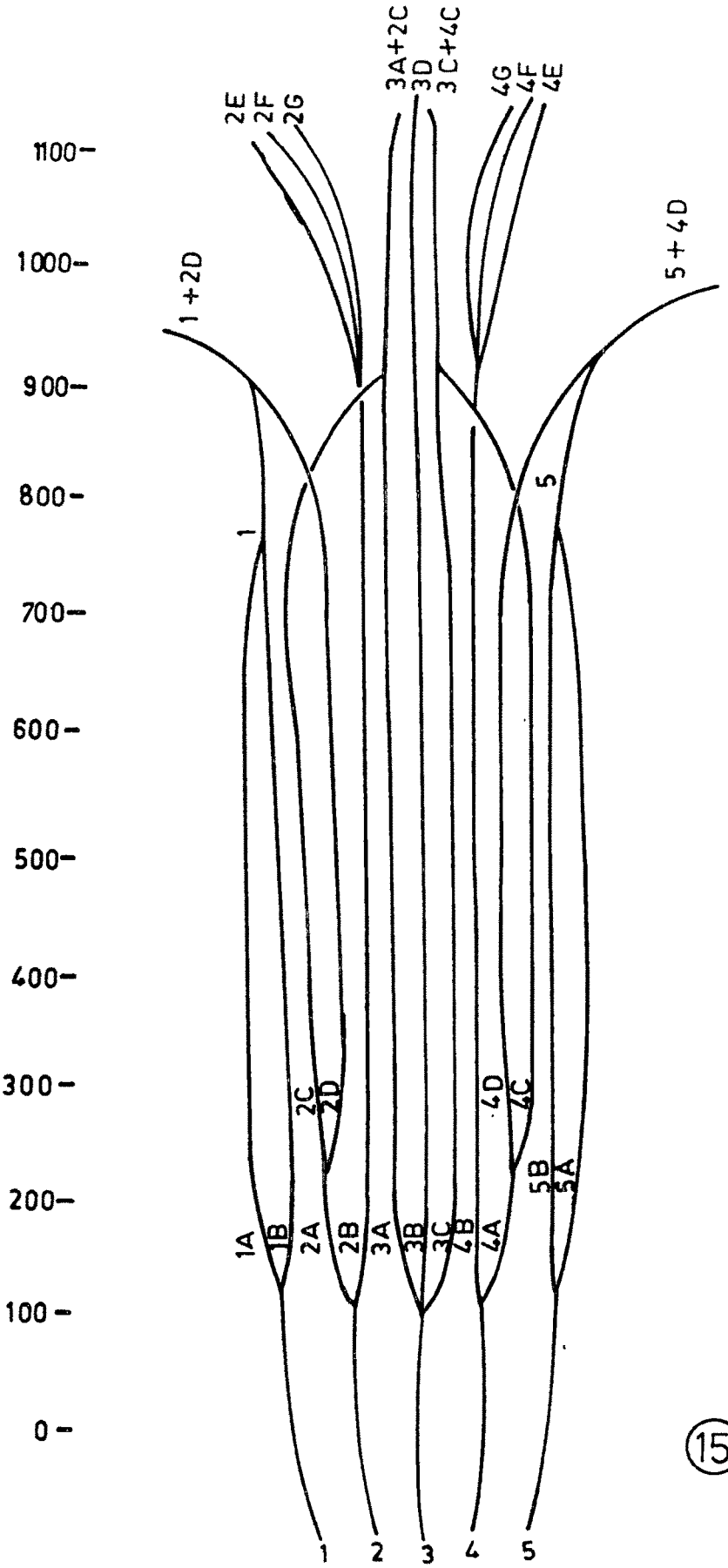
Fig. 15.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2D
4D
4C
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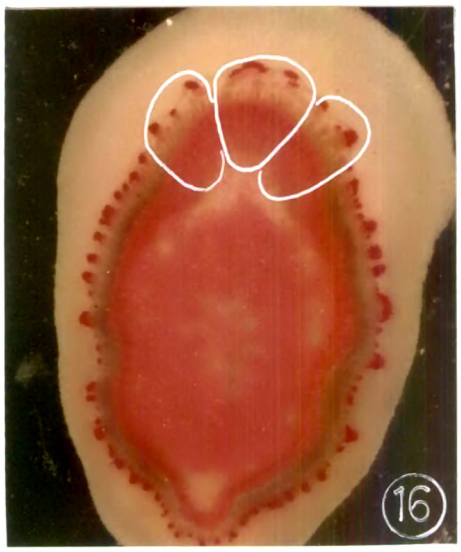
Plate 22

Figs.16 - 21.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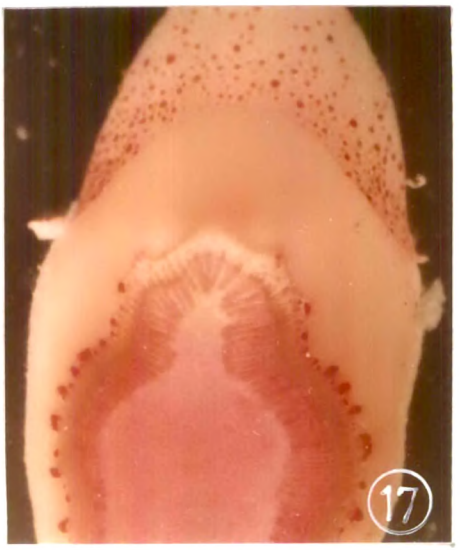
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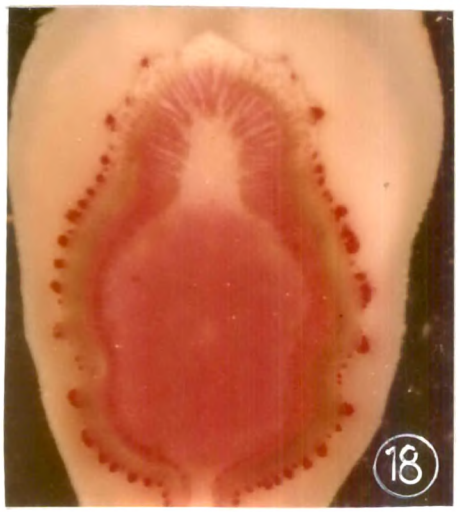
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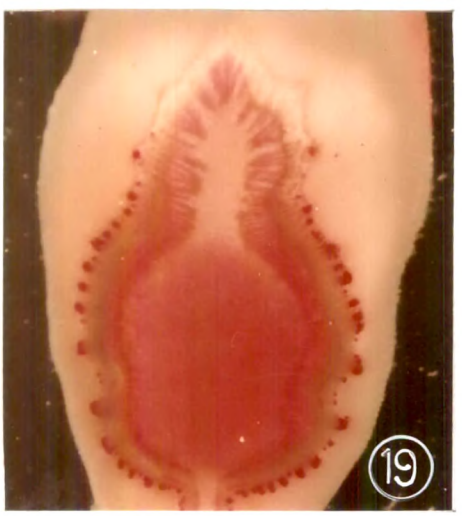
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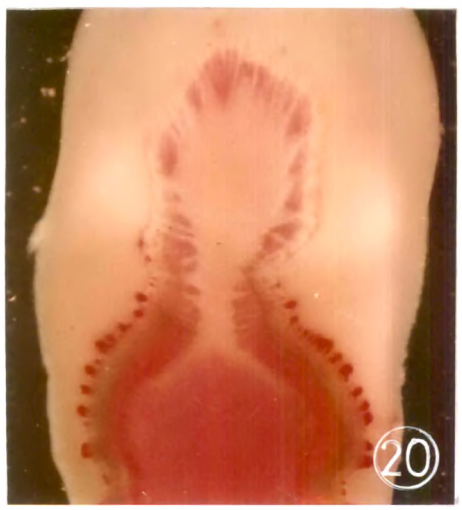
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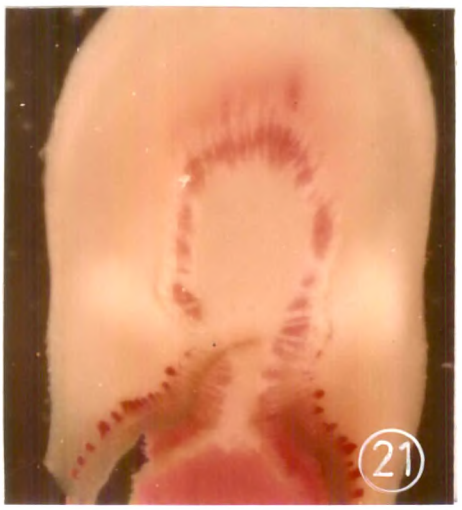
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Plate 23

Figs. 22 - 27.


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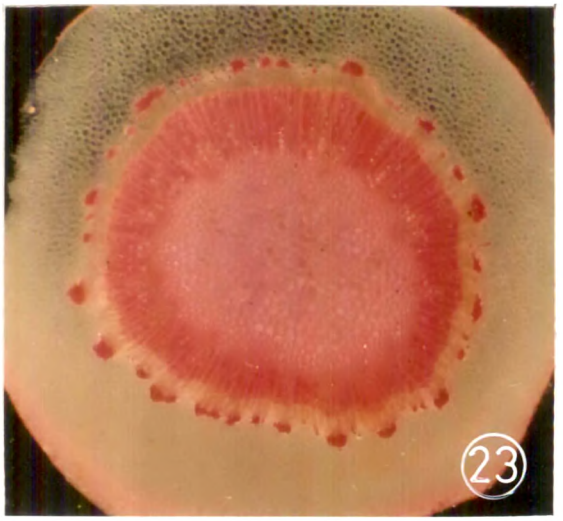
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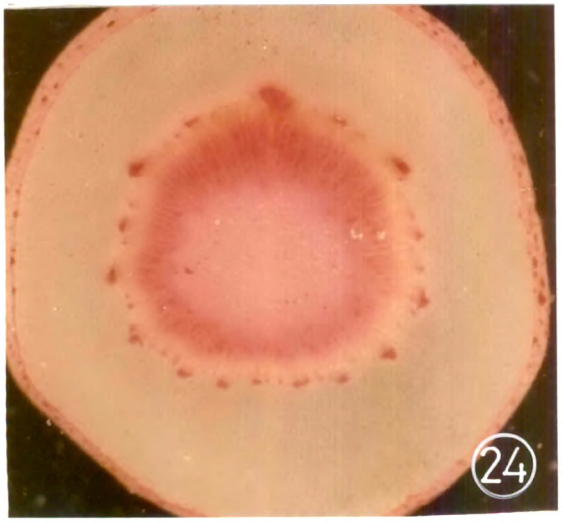
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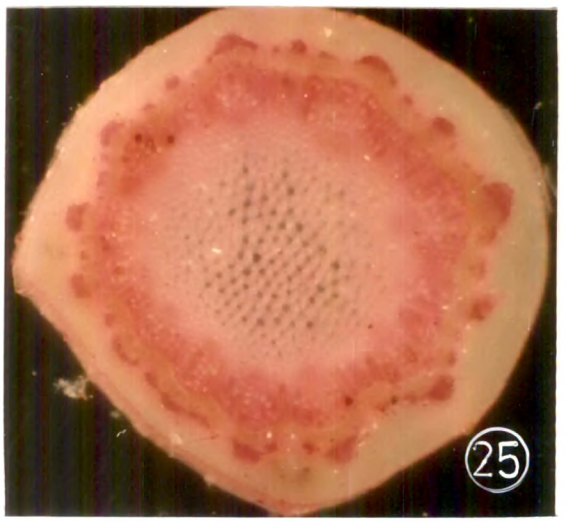
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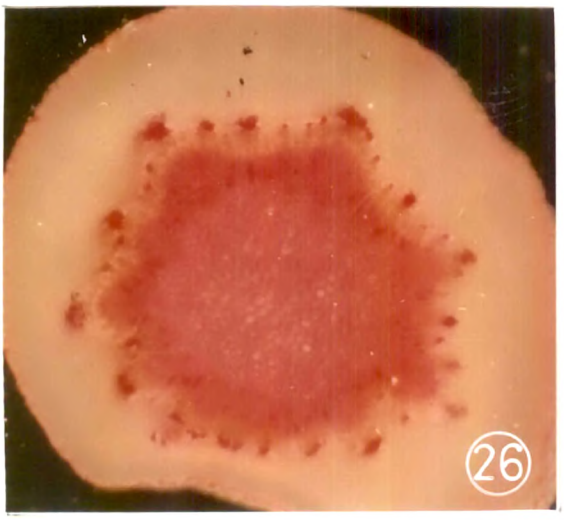
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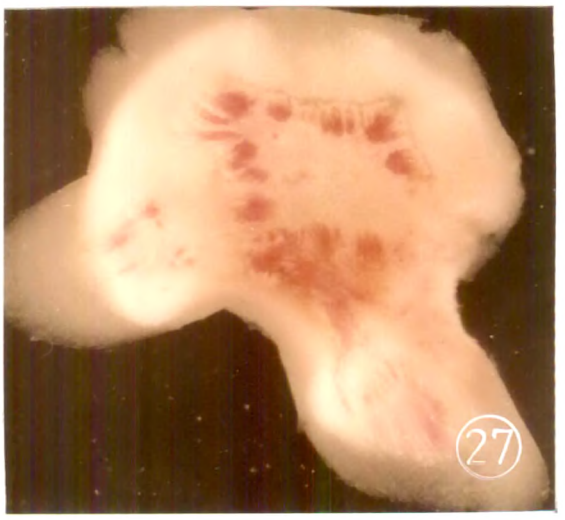
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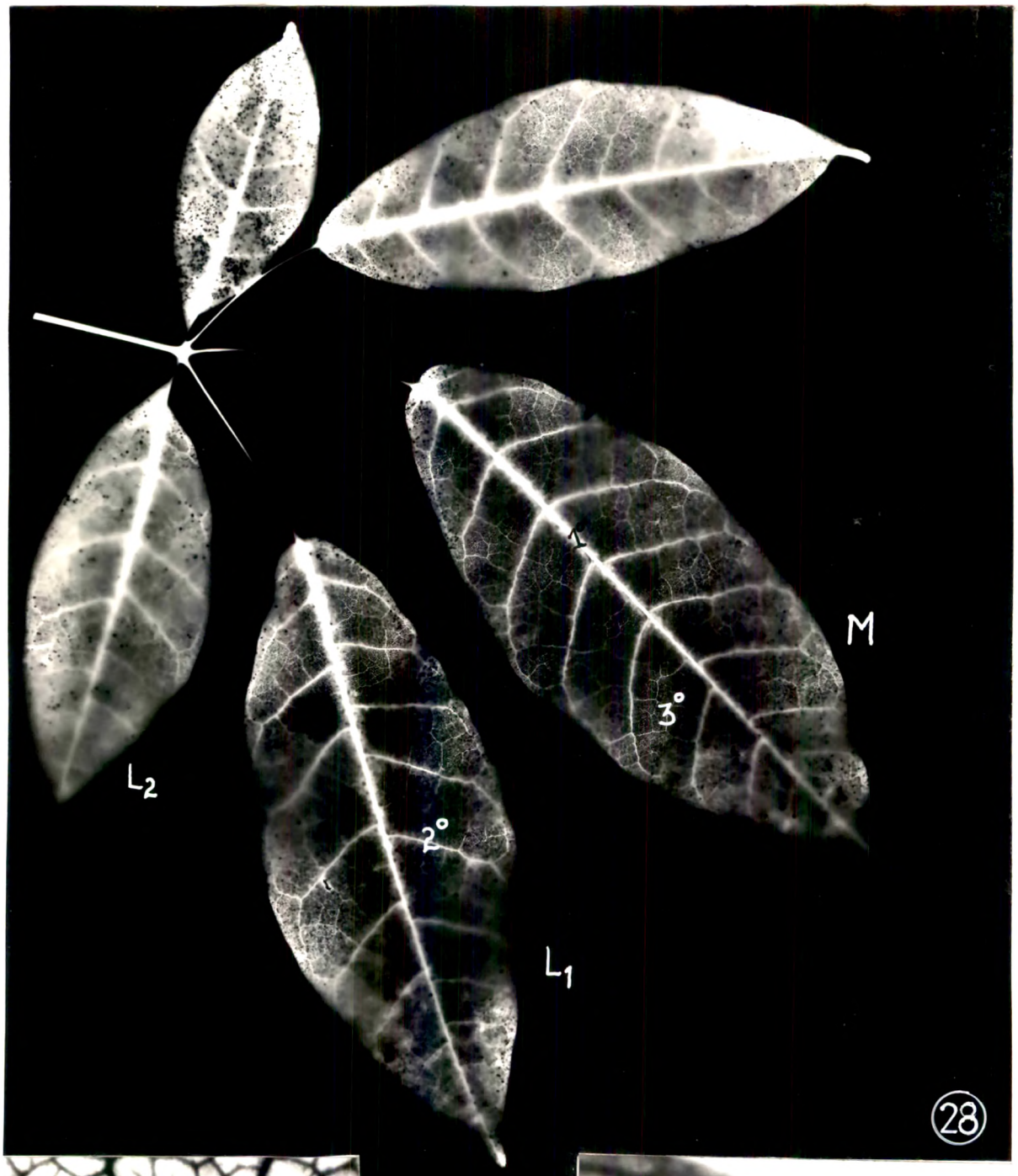
Plate 24

Figs. 28 - 30.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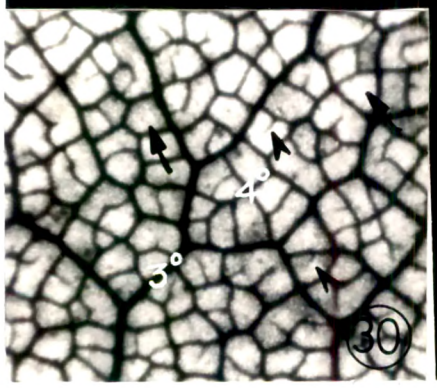
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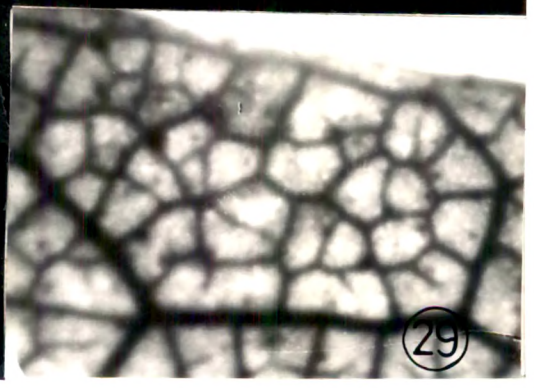
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Section V

3.2.6 Primary vascular differentiation in the petiole

Both transverse and longitudinal sections were examined to study the primary vascular differentiation in the petiole. The leaves are opposite and decussate and diverge from the main stem axis almost at the same level.

3.2.6.1 Transverse course of differentiation

Transverse sections of the shoot apex were studied at successive levels to trace the course of procambium. The position of the successively appearing leaves is referred to the respective nodes in the shoot apex.

A recognizable developing petiole and five developing petiolules are present only at the third node. At the nodes where the petiole and petiolules are not differentiated, my observations are mainly confined to the basal region of the leaf primordium, as it develops into the petiole. Four loci of actively dividing cells extend through the node towards the base of the first leaf primordium (fig 1). In the extreme basal region of the leaf primordium at the first node, a densely stained vascular meristematic arc is observed. It shows three loci of actively dividing cells, a median and two laterals (fig. 2). The three loci appear to be the precursors of procambial strands. At the level of the separation of the primordium from the shoot axis, only the median one traverses

the base of the primordium (figs. 3 and 4). This locus of actively dividing cells may be considered as the forefront of a developing procambial trace. It extends upto the distal region of the leaf primordium (fig. 5).

The extreme basal region of the leaf primordium at the second node shows three discrete collateral strands, a median and two laterals (fig. 6). The laterals further bifurcate giving rise to five strands (figs. 7 and 8). On separation of the leaf primordium from the axillary bud, the median strand alone traverses the base of the leaf primordium (figs. 9 and 10). Thus there is a single trace strand for the leaf primordium at this stage. The rest of the strands converge forming the vascular supply of the axillary branch. The median strand shows both protoxylem and protophloem (fig. 11) and extends upto the distal region of the leaf primordium (fig. 12).

The petiole at the third node is served by five strands at different stages of development. The origin of these five strands has been traced and described earlier. They include a median collateral and one collateral and a phloic strand on either side of the median (Section IV). These strands form the primary vascular system of the petiole. They bifurcate before traversing the basal region of the petiole giving rise to 13 strands organized in the form of an arc with incurved ends (Section IV).

3.2.6.2 Longitudinal course of differentiation

The shoot apex in Tabebuia rosea has a tunica corpus organization with a single layered tunica overlying the corpus (fig. 13). The shoot apex is a high dome shaped mound and leaf primordia are initiated on the sloping flanks.

The leaf primordium at the first node of the shoot apex shows longitudinal files of centrally located meristematic cells which are densely stained than the surrounding cells (fig. 14). This constitutes the forefront of a developing procambial strand.

The leaf primordium at the second node shows an advanced stage of procambial development. A procambial strand is distinguished from the surrounding cells of the prospective pith and cortex by the frequency of longitudinal divisions, deep staining of their protoplast, large round nuclei and densely packed long cells (fig. 15). Development of axillary bud is noticed in the axil of the leaf primordium.

3.2.7 Procambium

The early developmental stages of the petiole are categorized on the basis of their length into four stages Y_1, Y_2, Y_3 and Y_4, their lengths being 1.2, 2, 5 and 8.5 cm respectively. Procambium and the primary phloem elements are identified on the basis of the criteria described in Gmelina arborea (Section II).

As mentioned earlier in this chapter the petiole and petiolules are first recognized at the third node.

3.2.7.1 Procambial development

The young procambial strands consist of structurally homogeneous meristematic cells. Although the divisions in procambial strands appear to be random they none the less follow a pattern in the sense that a well organized procambial system is established.

Concurrent with the foregoing process, some isolated procambial cells on the inner side divide periclinally later form a continuous tangential layer (fig. 16) and as a result they show radial seriation. The radially aligned cells resulting from the first periclinal divisions have been termed as the 'initiating layer' by Larson (1976). The subsequent development of a definite periclinal orientation of cells is a marked change in the procambial differentiation. Procambial strands show the radially aligned cells in 2-3 layers (fig. 16). They are not equidistantly placed between the two poles of the strand, rather they appear inside, towards the pith. The outer region of relatively small cells forms the phloic procambium while the inner region constitutes the xylary procambium.

Protophloem differentiates before the protoxylem. In contrast to the protophloem that differentiates within the well developed phloic procambium external to the periclinally

dividing and radially seriated cells in the procambial strand, protoxylem pole originates at a site internal to it (fig. 17). The xylary procambium within which the protoxylem develops consists of highly vacuolated cells. Once initiated the protoxylem develops peripherally and centrifugally. The procambial cells show active periclinal divisions and extend across the vascular strand (fig. 18).

In the basal region of a 1.2 cm long petiole there are 20 vascular strands of varying developmental stages. Differentiation of metaxylem is first observed at this stage (fig.19). The procambial zone at this stage consists of narrow radially flattened cells interrupted by the radially elongated cells (fig. 20). Phloic strands also show similar 2-3 layers of procambium. The earliest metaxylem elements may be initiated from the randomly and periclinally divided procambial cells (fig. 19).

3.2.8 Metacambium

In a 2 cm long petiole vascular strands show a zone of cambiform (cambium like) cells internal to the primary phloem. It consists of a seriated band of 2-3 layers of cells characterized by almost regular periclinal divisions (fig.21). A similar zone is reported in the young vascular strands in the stem of Populus deltoides by Larson (1976). He referred it as metacambium which is an intermediate stage between the procambium and cambium. It is an advanced stage of procambium in which the procambial cells undergo frequent

periclinal divisions forming a distinct radial seriation of cell layers extending across the entire strand to form a tangentially continuous zone. I have borrowed this term to designate this intermediate stage in the petiole of Tabebuia rosea. The metacambial cells are more radially flattened than the procambial cells. However, there are some radial rows of cells, having radial diameter being greater than their tangential diameter (fig. 21). These are the cells which may later differentiate to form radial rows of parenchyma cells resembling the xylem ray. Some of the parenchyma cells in the interfascicular region also divide periclinally.

In a 5 cm long petiole at its extreme base, the radial seriation of metacambial cells become more conspicuous and they form a tangentially continuous band even between the vascular strands (fig. 22), thus developing a tangentially continuous metacambial cylinder of radially aligned cells. I have concluded that the periclinally dividing interfascicular parenchyma cells initially form the interfascicular metacambium. Observations regarding its development follow subsequently in this section. The metacambial cells are radially flattened (fig. 22) and they form a radial seriation of 3-4 cells. The increased tempo of cell division activity resulting in cells arranged in a more definitive radial and tangential alignment helps in distinguishing the metacambium from the other surrounding cells of the cortex and pith.

In the basal region of a 5 cm long petiole, parenchyma cells lying below the interfascicular metacambium, xylem parenchyma and protophloem fibers show lignifications of their walls (fig. 23).

New xylem elements develop from the interfascicular metacambium cells in between the main vascular strands (figs. 24 and 25). These elements can be considered as metaxylem because they appear simultaneously with the metaxylem elements formed from the metacambium in the vascular strand. These elements can also be termed as the late metaxylem elements because they appear after the differentiation of protoxylem and early metaxylem in the vascular strands.

In a 8.5 cm long petiole a complete cylinder of xylem is present. Cell wall thickening becomes more apparent in the protophloem fibers and also in the interfascicular parenchyma cells. A layer of tangentially elongated metacambial cells is interrupted by radially elongated cells (fig. 26). Although metacambial in micromorphology, these cells assume characteristics more resembling those of the vascular cambium. None the less neither the vascular meristem nor its derivatives assume characteristics attributed to the cambium until the petiole elongation has ceased. Lignification of protophloem fiber precursors and xylem parenchyma cells is pronounced. Development of tracheary elements with pitted wall sculpturing and differentiation of xylem fibers generally

associated with the secondary xylem are observed at this stage. Although differentiating secondary elements are observed earlier, they do not mature until the petiole elongation has ceased. The elongated cells resembling the fusiform cells of the cambium do not show a beaded wall structure which is a general characteristic associated with these cells. Secondly, the tracheary elements originating from the cambial cells generally have the same length as their precursors, but in the 8.5 cm long petiole studied they are elongated than their precursors. Apart from these observations, another additional aspect which led me to conclude that the transformed vascular meristem has not yet micromorphologically developed into a typical vascular cambium is that the petiole is still in its elongating phase.

In view of the above mentioned observations I am inclined to consider that this stage is a transition between procambium and cambium eventhough the vascular cylinder shows both the general appearance and many of the structural characteristics attributed to the cambium and its activity.

3.2.9 Micromorphological and dimensional changes during the transition of procambium-metacambium-cambium

The procambium is a structurally homogeneous tissue in its early developmental stage (figs. 27 and 28). The procambial cells divide anticlinally and periclinally. They

are closely packed. They have conspicuously stained cytoplasm and round and large nuclei. They show a more or less storied arrangement in longitudinal view (figs. 27 and 28), which distinguish them from surrounding cells of the pith and cortex. The end walls are rounded or transverse in radial view (figs. 16 and 17). The first protoxylem elements are with annular wall sculpturing which are physically stretched with the petiole elongation.

With the elongation of the petiole the procambial cells also elongate. They show transverse or oblique end walls, with a less dense cytoplasm (fig. 29). The nucleus becomes elongated and is spindle or oval shaped.

During further elongation, in a 2 cm long petiole the radially flattened cells of the metacambium are organized (fig. 21). These cells are more radially flattened than those in the preceding stage. In longitudinal view the metacambium exhibits two systems, one composed of axial files of relatively short cells with transverse end walls and the other with transverse or oblique end walls (figs. 30 and 31). The axial file of short cells later differentiate as cambial ray cells.

The differentiation of the two cell systems in the metacambium results apparently from the uneven distribution of transverse divisions in some of the procambial cells and the axial elongation of some of the cells without transverse divisions during the elongation of the petiole.

Metaxylem elements at this stage of the petiole show helical or closely helical wall sculpturing. Young xylem fibers have tapering end walls.

In a 5 cm long petiole apical intrusive growth of the long cells of the transition stage of metacambium becomes prominent (fig. 32) and in a 8.5 cm long petiole they gradually attain fusiform nature (figs. 34 and 35). Periclinal division in the metacambial cells is evident by the presence of phragmoplasts (fig. 33).

Developmental features accompanying the secondary growth are gradually evident beginning from a 5 cm long young elongating petiole. Both apical intrusive growth of the elongated metacambium-cambium transit cells and the blocking out of the prospective ray tissue occur in a 5 cm long petiole and when the petiole ceases elongation it assumes the true characteristics attributed to cambium, i.e., the development of fusiform and ray cells.

In the 5 cm long petiole, xylem parenchyma cells close to the metaxylem elements show wall lignification and it gradually extends to the cells of the interfascicular region. Lignification of the phloem fibers begin simultaneously with that of the xylem parenchyma cells. Metaxylem elements at this stage show closely helical or scalariform wall sculpturing. Along with the lignified xylem parenchyma and fiber cells the vascular strands form a complete cylinder

where the full identity of the strands becomes less evident. This is clearly evident in a 8.5 cm long petiole. In this petiole the tracheary elements show scalariform and pitted wall sculpturing but they are more elongated than their precursors (mother cell). Both fusiform and ray cells are present but do not show beaded appearance. Hence this stage is considered as a precursors stage of the vascular cambium.

With the cessation of petiole elongation the differentiation of protophloem and metaphloem is completed. In a 13.5 cm long petiole, which is ceased to elongate, show tracheary elements with pitted wall sculpturing and they are as long as their precursors. The long fusiform cambial cells show beaded appearance. Xylem parenchyma, xylem fibers and protophloem fibers show birefringent walls. Hence I conclude that the secondary growth is organized in this petiole. The dimensional changes observed in the procambium, metacambium and cambium cells are mentioned in table VII.

3.2.10 Vascular cambium

In a mature petiole at the pulvinus base the vascular strands are discrete and when viewed under polarized light tracheary elements and protophloem fibers show birefringence. In the middle region also these cells show birefringence (fig. 36). Secondary growth in the petiole of Tabebuia rosea is not confined only to the vascular strands but it extends in the interfascicular region. In a transverse view the vascular cambium is a cylinder of actively dividing cells in

radial files of 3-4 cells (fig. 37). It consists of fusiform and ray cell system. I have preferred to use the term fusiform and ray cells and not initials as I could not distinguish between fusiform initials and their immediate derivatives.

3.2.10.1 Fusiform and ray cells

The vascular cambium in the petiole of Tabebuia rosea consists of two system of cells, the axially elongated fusiform cells being responsible for the production of the axial elements of the secondary phloem and secondary xylem and the shorter ray cells being responsible for the production of horizontal ray system (figs. 58 and 59). Fusiform cells are long, generally uninucleate with tapering end walls or fusiform end walls and they show beaded appearance and frequent transverse divisions.

Rays are uniseriate (fig. 59) or biseriate (fig. 51) composed of short cells with rounded or transverse end walls.

3.2.10.2 Development of ray cells

Vascular rays on the basis of their origin can be categorized into the primordial ray cells and secondary ray cells. Primordial rays are the rays occurring during the transitional stage in the transitional tissues during the elongation of the petiole. The long uniseriate and biseriate rays arise generally through the uneven septation of some of

the long metacambial cells to give rise to short cells in axial files which later form the ray cells (figs. 31 and 32). Philipson et al. (1971) have termed these rays as the primary rays.

The secondary rays originate from the cambium ray cells during the development of the secondary xylem after the petiole ceases elongation. The fusiform cells also give rise to ray cells. Ray cells may be formed by a single cell cut off from the fusiform end by pseudotransverse divisions (fig. 61) or a part of the fusiform cell may be segmented by transverse divisions to form a tier of ray cells (fig. 58). The height of the long rays may be at times reduced by the dividing intruding fusiform cell (fig. 60).

3.2.11 Development of the interfascicular cambium

As mentioned earlier the vascular strands in the petiole at the third node are discrete with interfascicular parenchyma. They differ from those of the pith and the cortex by their small size and absence of intercellular spaces. Longitudinally they show transverse end walls and are with a prominent nucleus (fig. 41). The cells are more or less structurally homogeneous.

In a 1.2 cm long petiole at some loci the interfascicular parenchyma cells show periclinal divisions. Longitudinally these cells are elongated and narrower than those of the previous stage (figs. 42 and 43). In a 2 cm long

petiole the periclinal divisions are more evident and tangentially continuous with the 2-3 layered metacambium of the vascular strand (fig. 38). Still at certain loci interfascicular parenchyma persists. So the tangential advancement of periclinal divisions occurs from the fascicular region to the interfascicular region. In a longitudinal view at this stage like the meta cambium within the strand, the interfascicular cells also show two systems of cells, long cells and short cells. Some of the long cells show oblique end walls. As these cells appear similar to metacambium within the strand and as they are present between the strands, I have used the term interfascicular metacambium to designate the periclinally dividing cells in the interfascicular region.

In a 5 cm long petiole the interfascicular region shows 3-4 layered cells (fig. 39) in radial seriation and in continuity with the band of metacambial cells in the strand to form a complete metacambial cylinder. As mentioned earlier in this chapter, it is at this stage of the petiole that new xylem was observed to develop in the interfascicular region. The tracheary elements produced were of metaxylem type and hence this was another additional criterion which prompted me to coin the term 'interfascicular metacambium'. These cells are radially flattened and appear similar to those of the metacambium within the strand. The two system of cells in the metacambium are distinctly seen in the longitudinal view (fig. 44). The long cells show tapering or transverse end walls.

In a 8.5 cm long petiole, which is the transitional stage between metacambium and cambium, the interfascicular metacambial cells in radial view show radially flattened cells and radially elongated cells. The radially elongated cells later differentiate as the ray cambium cells. Longitudinally the long cells appear with fusiform or oblique end walls (figs. 45-47). Long axial file of short cells with more or less rounded end walls (fig. 45) later develop as the ray cells. The long cells like those of the late metacambial cells in the vascular strands do not show a beaded appearance.

In a mature petiole 2-3 layers of radially flattened cells are interrupted by the elongated ray cells (fig. 40). The interfascicular cambium thus arises from periclinal divisions in the interfascicular parenchyma by its dedifferentiation and redifferentiation into metacambium which during its further course of development assumes and follows the same pattern of development as the fascicular cambium.

3.2.12 Ontogenetic relationship between secondary phloem elements and cambium

The secondary phloem elements appear in radial seriation with cambial cells (fig. 48). In order to examine the ontogenetic relationship of secondary phloem elements and cambial cells, serial transections were examined and the

sequence and plane of divisions were studied in the phloic cambial cells.

Derivatives of a fusiform cell divide periclinally and anticlinally to form a complex of sieve tube elements, companion cells and phloem parenchyma. The number and orientation of cell divisions occurring in a derivative cell leading to the formation of the phloem elements to form a complex may vary. The complex generally contains two sieve tube elements each with a companion cell and a phloem parenchyma cell. Cell 'A' (fig. 48, A) is a derivative of the fusiform cell. When examined serially it is observed that a fusiform derivative cell gives rise to a complex of two sieve tube elements associated with the companion cells and a parenchyma cell. The complex is formed probably by the mother cell undergoing two unequal periclinal divisions giving rise to 3 cells radially aligned. Out of them two of the contiguous cells undergo anticlinal divisions giving rise to a large cell and a small cell. The large cell differentiates as a sieve tube element and the small one as a companion cell. The third differentiates as a phloem parenchyma cell (fig. 48, C). Sometimes the elements of the complex are so obliquely placed that it is difficult to determine the plane of cell divisions.

3.2.13 Vascular cambium in the stem

Vascular cambium in the stem of Tabebuia rosea consists

of fusiform cells and ray cells. The vascular cambium is storied with fusiform cells arranged in vertical rows with overlapping ends (fig. 63). The fusiform cells are generally uninucleate but multinucleate condition is also present. The nucleus is spindle shaped or oval shaped with nucleoli. The walls of the fusiform cells show beaded appearance. Average length of the fusiform cell varies from 271.7 - 347.6 μm in length and 18-25 μm in width.

The rays are uniseriate (fig. 63) or multiseriate. They are 44.2-56.2 μm in width and 208.5 - 265 μm in height. Transverse sections of the bark show 8-9 layers of radially aligned cambial cells (fig. 64).

Comparative features of the vascular cambium cells of the petiole and stem are mentioned in table VIII.

Table VII. Dimensional changes of the procambium, metacambium and cambium cells during the development of the petiole in Tabebuia rosea. Dimensions are in μm.

Length of petiole in cm	Procambium		Metacambium		Cambium			
					Fusiform		Ray	
	Length	Width	Length	Width	Length	Width	Length	Width
0.07	31.4-50.2	4.7-6.2						
1.2	47.0-84.7	4.7-7.8						
2			54.9-141.3	3.1-4.7				
5			54.9-116.1	3.0-4.7				
8.5			145.3-347.0	12.0-18.9				
13.5					111.4-243	9-12	233.8-1137.6	12.6-18.9

Table VIII. Comparative features of the vascular cambium of petiole and stem

	Petiole	Stem
1) Cell types	Axial-fusiform cells Vertical-ray cells	Axial-fusiform cells Vertical-ray cells
2) Arrangement	Non-storied	Storied
3) Number of layers	3 - 4	8 - 9
4) Fusiform cells/ transit cells		
a) Length μm	111.4-243	271.7-347.6
b) Width μm	9-12	18-25
Number of nucleus	Uninucleate	Uninucleate or multinucleate
5) Rays	Uniseriate or biseriate	Uniseriate or multiseriate
a) Height μm	233.8-1137.6	208.5-265
b) Width μm	12.6-18.9	44.2-56.2

Plate 25

Figs. 1 - 6.

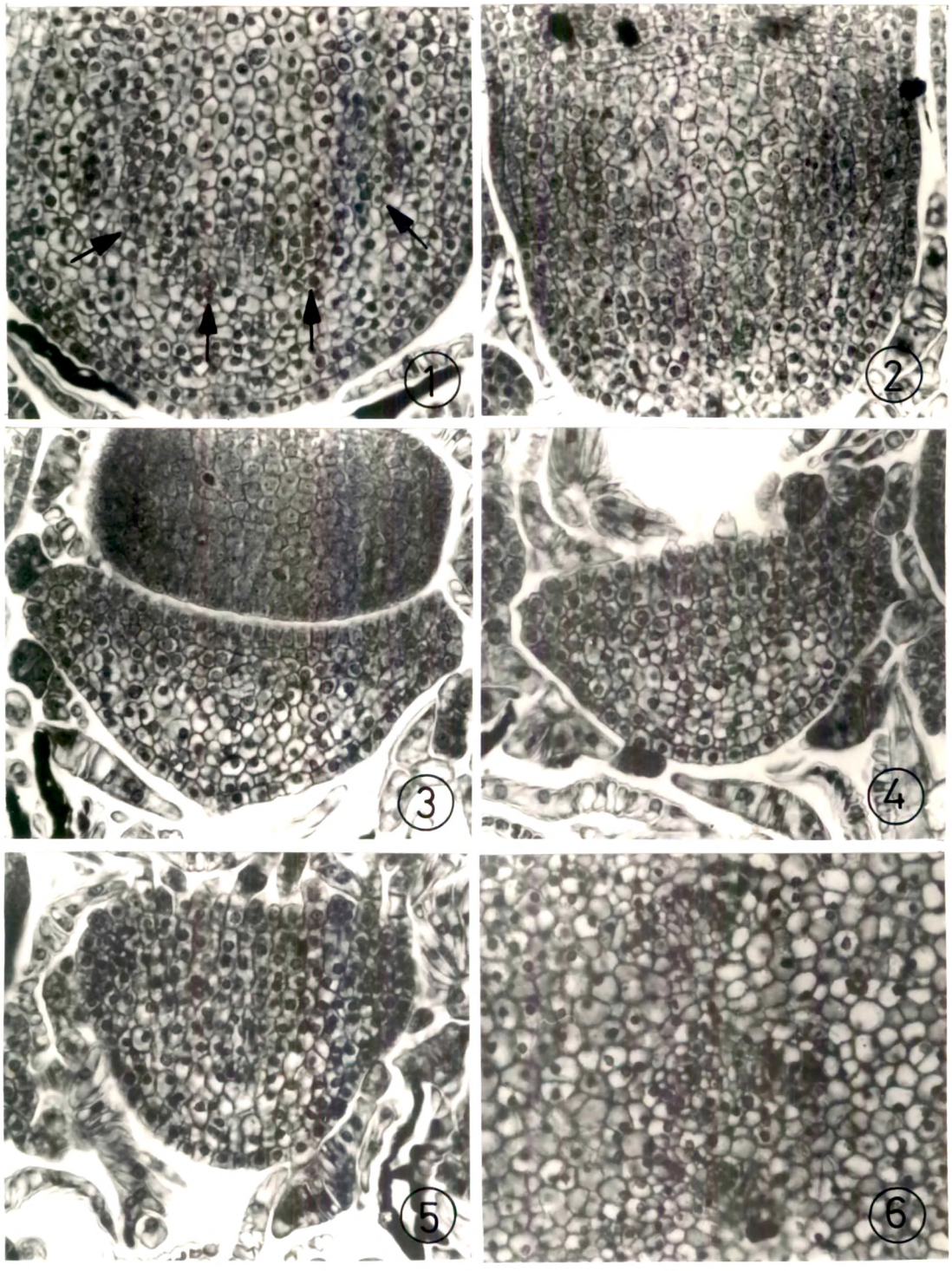
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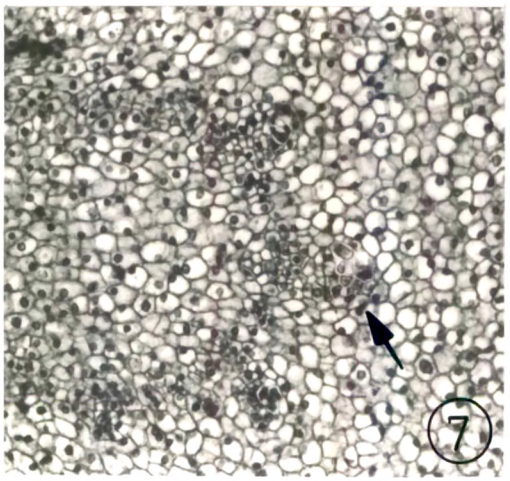
Plate 26

Figs. 7 - 12.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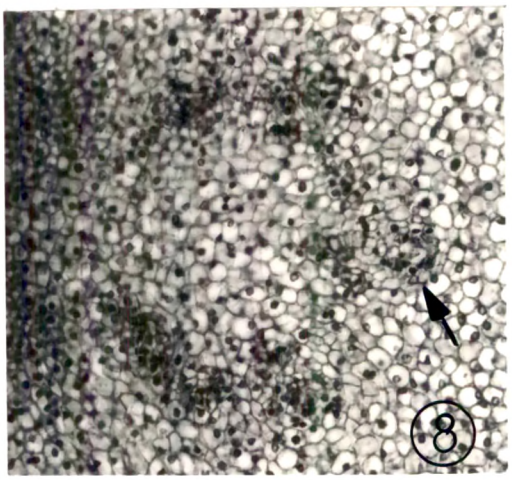
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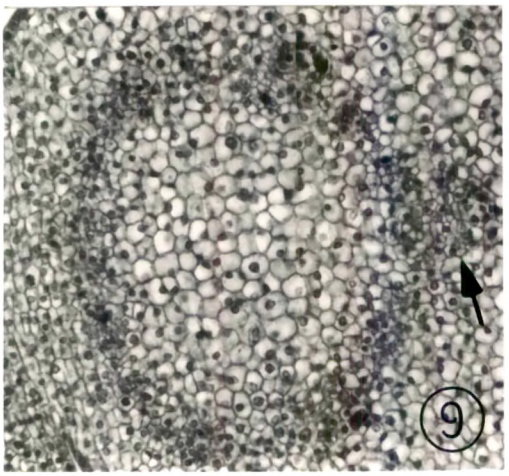
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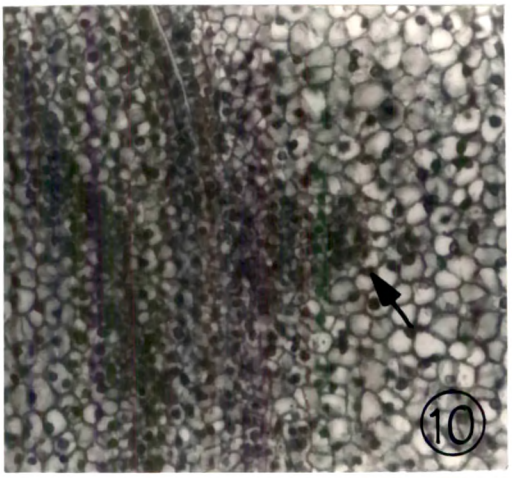
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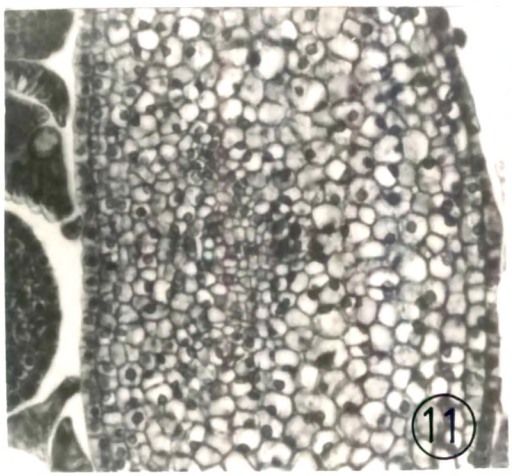
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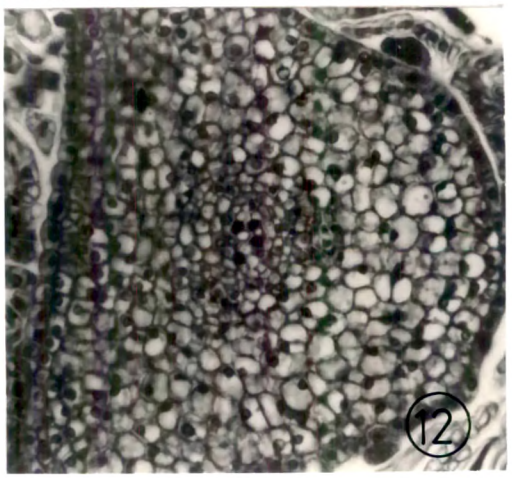
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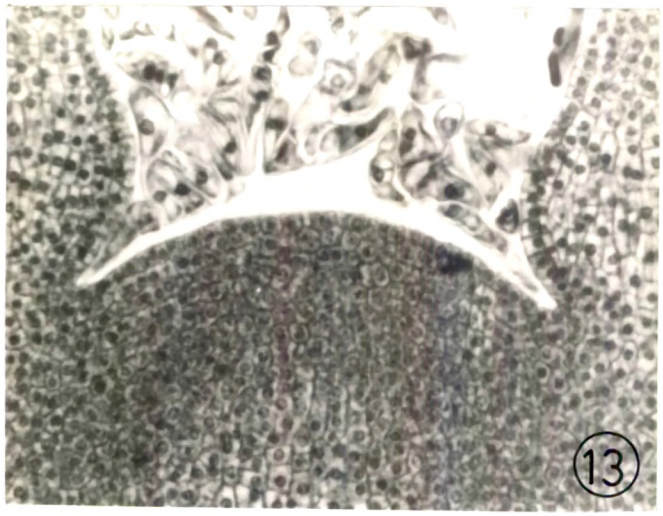
Plate 27

Figs. 13 - 20.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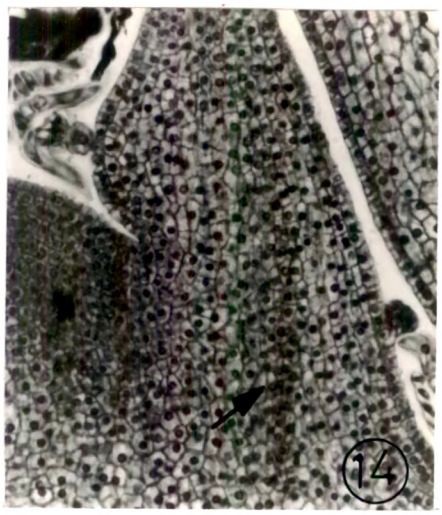
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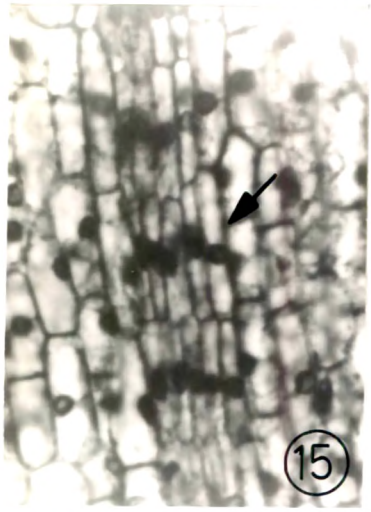
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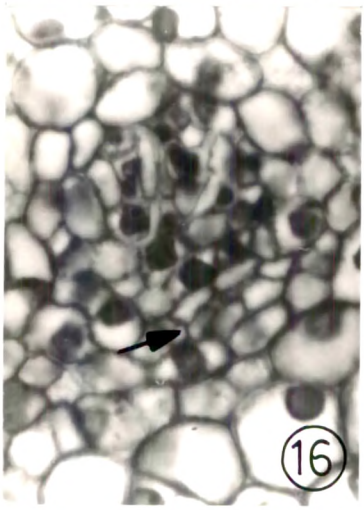
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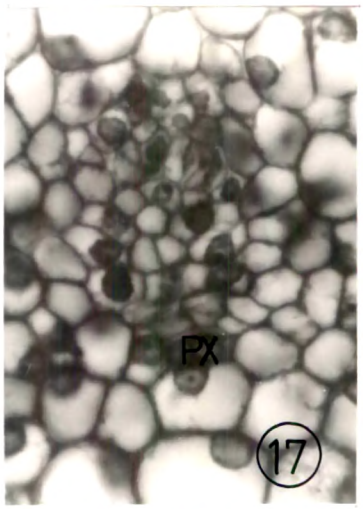

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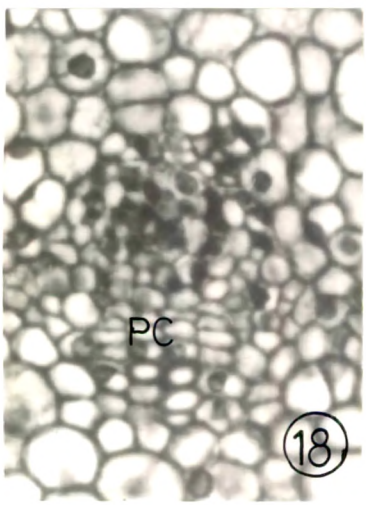

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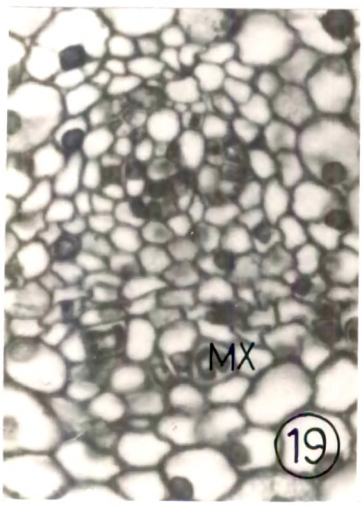

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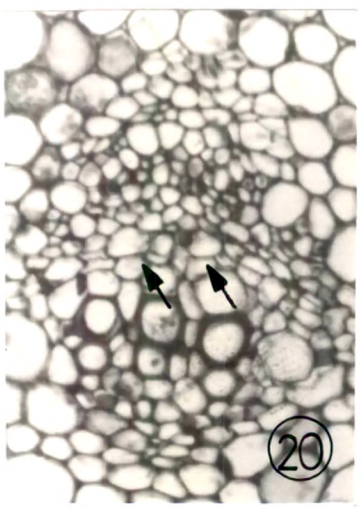
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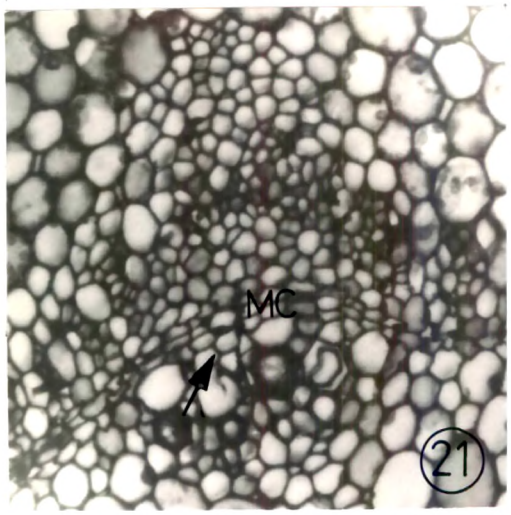
Plate 28

Figs. 21 - 26.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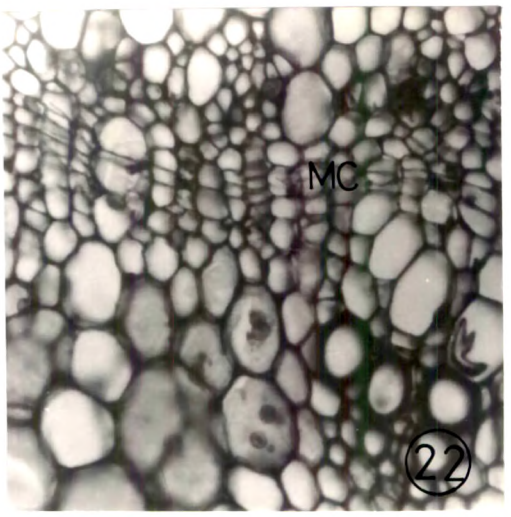
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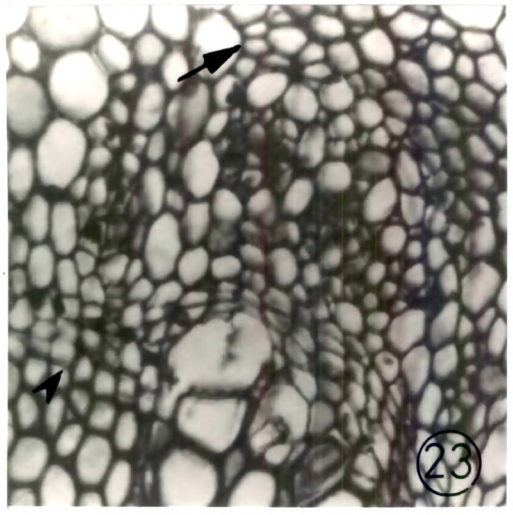
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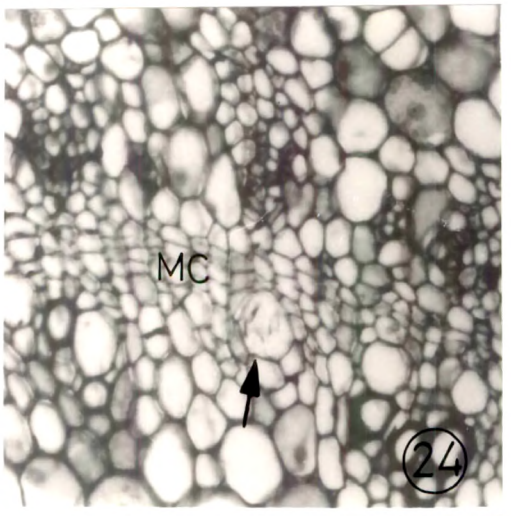
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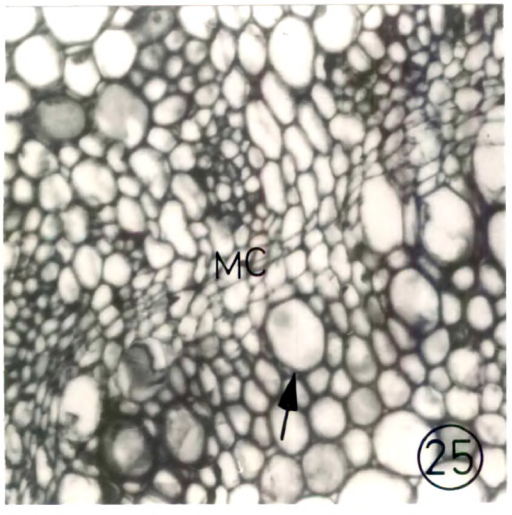
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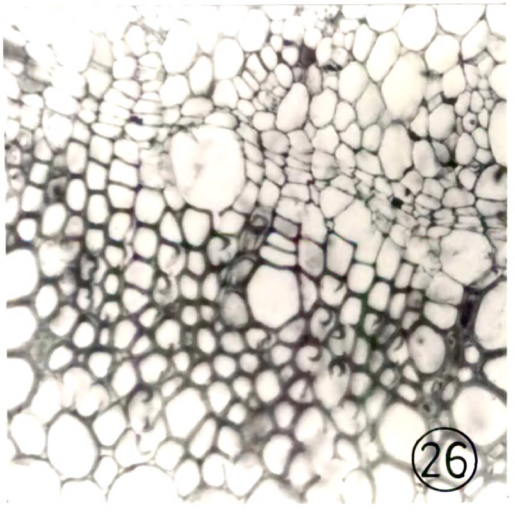
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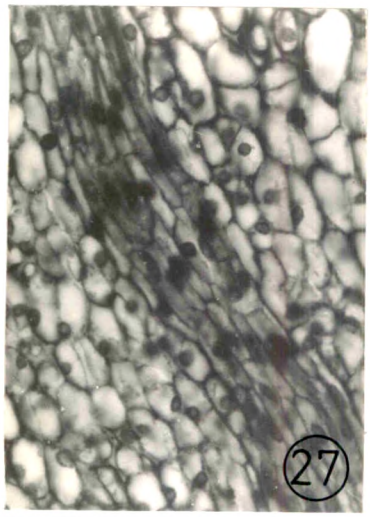
Plate 29

Figs. 27 - 35.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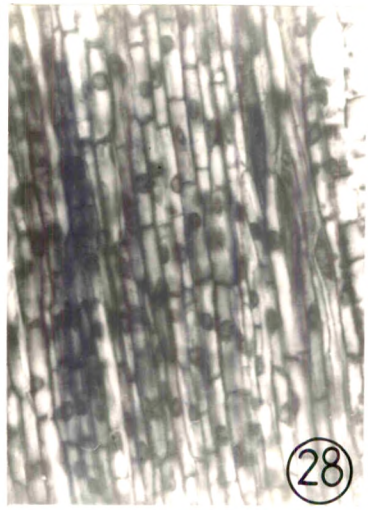
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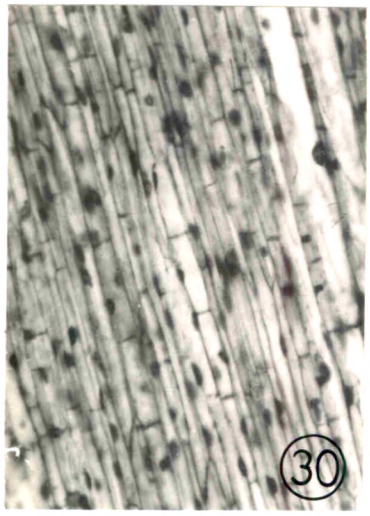
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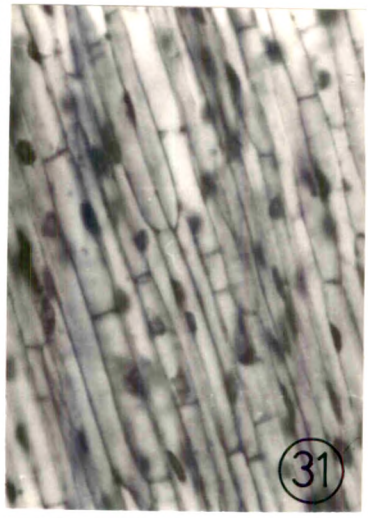
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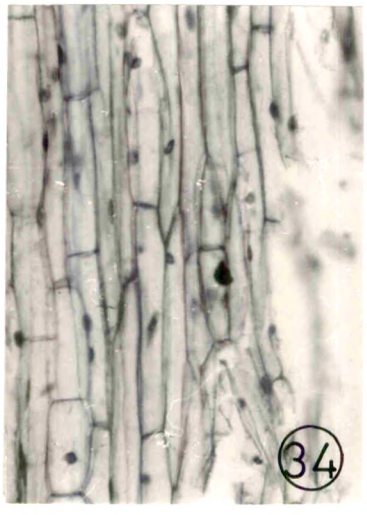
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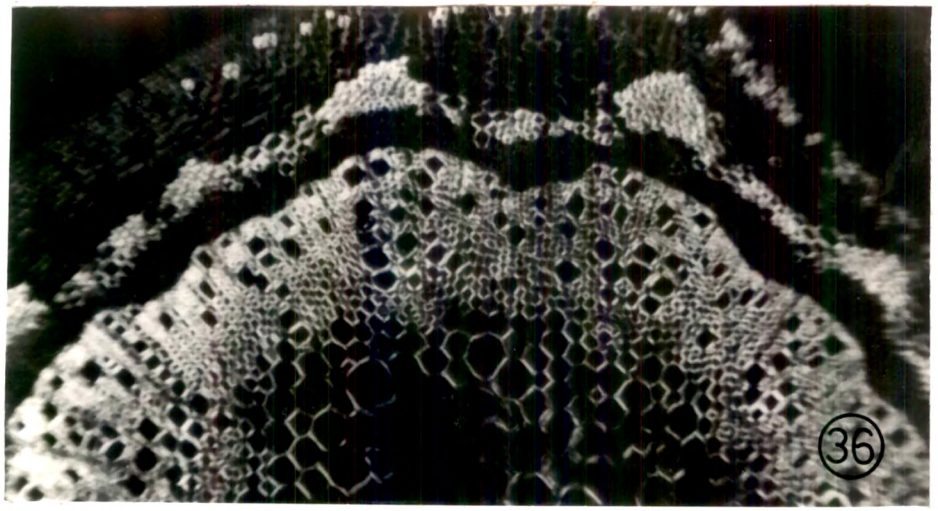
Plate 30

Figs. 36 - 40.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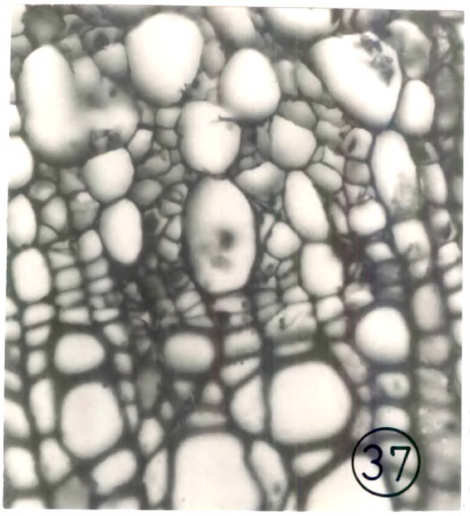
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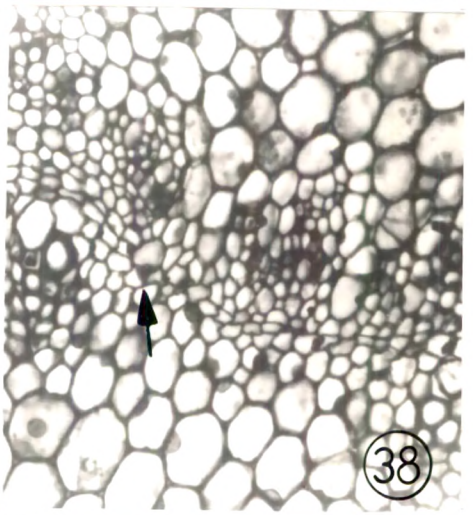
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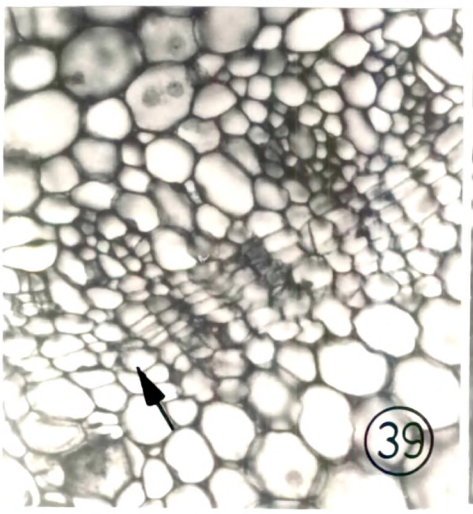
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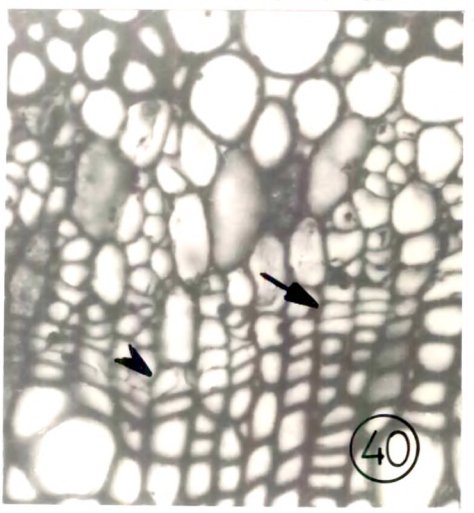
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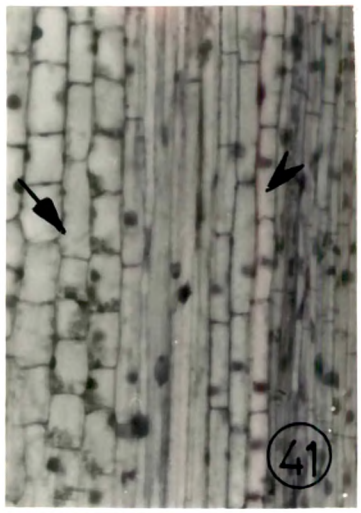
Plate 31

Figs. 41 - 47.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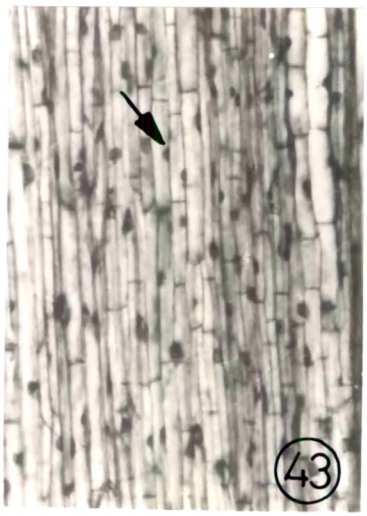
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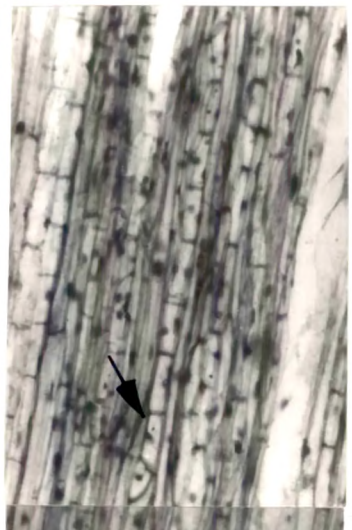


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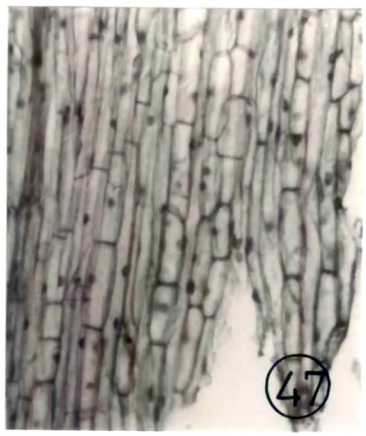
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Figs. 48 - 57.

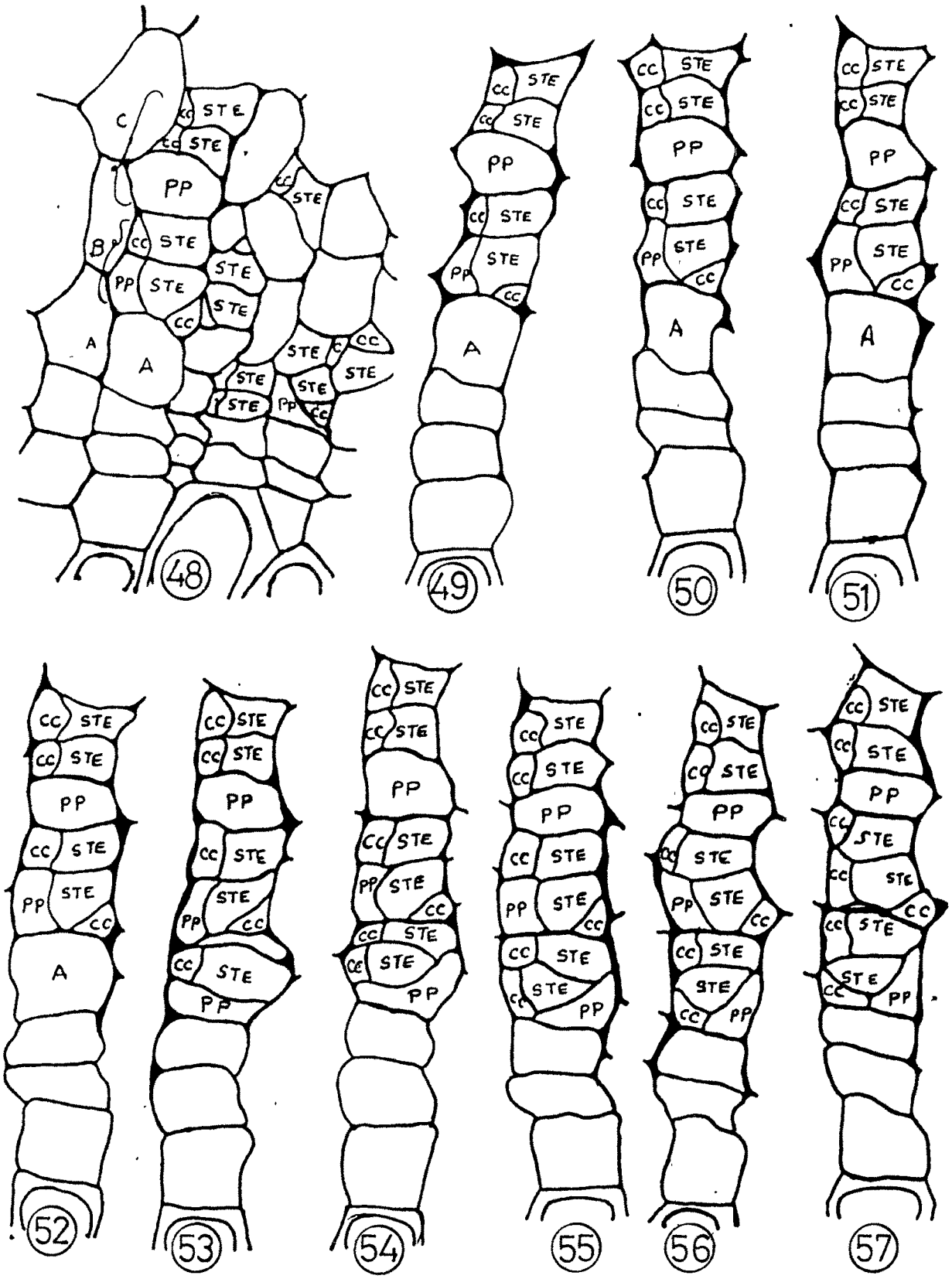
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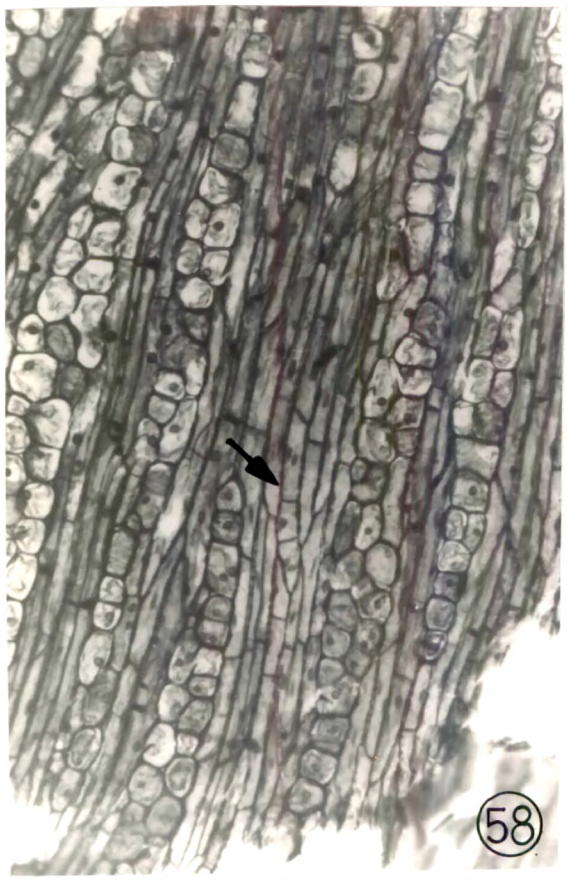
Plate 32

Figs. 58 - 64.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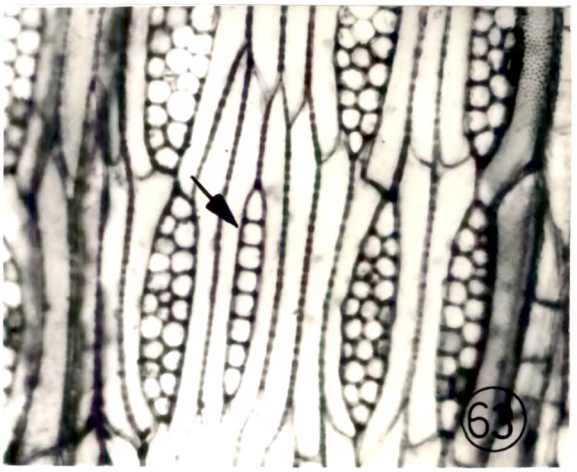
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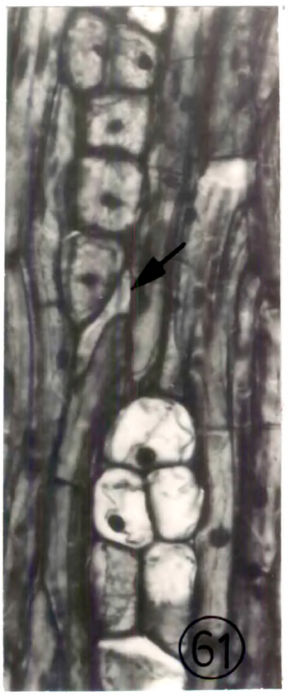
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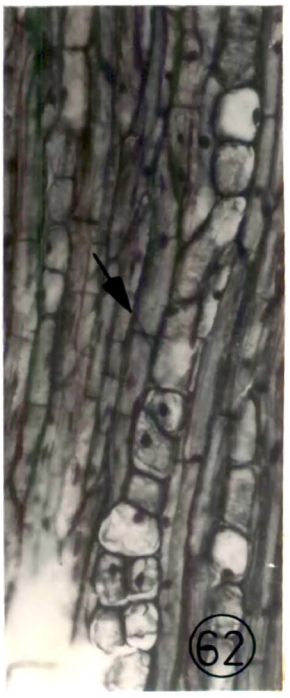
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Section VI

3.2.14 Phloem

As reported earlier certain developmental features apparently noticed in the vascular system are related with the elongation, cessation and maturation of the petiole. The developmental stages of phloem in Tabebuia rosea have been categorized into protophloem, metaphloem and the secondary phloem.

3.2.14.1 Protophloem

Young vascular strands at the abaxial region which traverse more or less without much anastomoses from the basal to the distal region of the petiole were selected at random for the study of phloem differentiation and structure. The observations have been compared with those of the other vascular strands and the critical observations, if any, have been mentioned at the appropriate places.

In the petiole at the third node the vascular strands are at varying stages of development. All of them do not develop simultaneously and hence the state of their procambial differentiation is variable.

3.2.14.1.1 Sieve tube element

The young procambial strand shows a group of more or less structurally appearing homogeneous cells (fig. 1) within which the protophloem first differentiates. Its

differentiation begins with one or two sieve tube elements (figs. 2 and 3), along the outer periphery of the procambial strand. These sieve tube elements differentiate inner to a layer of small parenchyma cells outlying the procambial strand (fig.3). Differentiation of protoxylem is observed after the protophloem (fig. 4).

After the differentiation of a few sieve tube elements radial seriation is evident in the procambium (figs.4 and 5). The first formed sieve tube elements appear solitary or in pairs, rather randomly distributed amongst the phloem parenchyma cells (figs. 5 and 6). They are with or without companion cells, with transverse (fig. 7) or oblique (fig. 9) end walls, with simple sieve plate and scattered, aggregate lateral sieve areas.

3.2.14.1.2 Companion cell

Each sieve tube element is normally associated with a single companion cell aligned on any one of its lateral walls (figs. 7-9). The companion cell is as long as the sieve tube element. They are uninucleate with dense cytoplasmic contents and without starch grains. The nucleus is round (fig. 10) or elongated (fig. 9) with a single nucleolus.

3.2.14.1.3 Slime

The P-protein with other contents accumulate at the sieve plate in the form of a slime plug usually formed on one

side of the sieve plate. It generally attains a funnel form of configuration which may be elongated (fig. 11) extending almost half of the sieve tube element or a short one (figs. 12 and 13). At times elongated dense mass is observed away from the sieve plate (fig. 14).

3.2.14.1.4 Phloem parenchyma

The contiguous and non-contiguous parenchyma cells are generally larger than the sieve tube elements. They are also more or less densely stained.

3.2.14.2 Metaphloem

As noted earlier, metacambium is observed in a 2 cm long petiole. Once the metacambium has attained a certain level of distinct micromorphological identity within a strand then some of its derivatives differentiate into metaxylem and metaphloem. Metaxylem elements show a radial pattern of arrangement, whereas the metaphloem does not show any specific pattern of orientation. For taking dimensional details metaphloem sieve elements were identified by the same criteria used in Gmelina arborea.

3.2.14.2.1 Sieve tube element

Metaphloem contains groups of sieve tube elements, companion cells and phloem parenchyma cells (fig. 15). Each sieve tube element is associated with a companion cell. The thick walls of the sieve tube element and their clear lumen

strikingly contrasts from the other cells of the phloem i.e., companion cells, phloem parenchyma cells (figs. 17 and 18). The early formed metaphloem sieve tube elements are short (fig. 19) but during the late stages of metacambium, long sieve tube elements are observed (figs. 20 and 21).

Sieve tube elements have transverse (fig. 19) or oblique (fig. 20) end walls and the lateral sieve areas are aggregate and scattered (fig. 22).

3.2.14.2.2 Companion cell

A single companion cell is aligned on any one of the sieve tube element wall (figs. 19-21), and is as long as its associated sieve tube element.

3.2.14.2.3 Slime

The metaphloem sieve tube elements also show the P-protein and other contents accumulated in the form of dense mass away from the sieve plate (fig. 23) or at the sieve plate forming a funnel (fig. 24) configuration.

3.2.14.2.4 Phloem parenchyma

The contiguous and non-contiguous parenchyma cells vary in width. Some are as large as or larger than the sieve tube elements. The parenchyma cells interspersed in between the groups of metaphloem are generally larger than the other phloem parenchyma cells (fig. 15).

3.2.14.3 Development of protophloem fibers

In a young petiole at the third node the parenchyma cells outlying the first formed sieve tube elements are small without intercellular spaces (fig. 2). Along with the development of the primary phloem they widen conspicuously (fig.25) and elongate. This is evident in the vascular strand in a 2 cm long petiole. They show apical intrusive growth. The phloic strands also show these widened parenchyma cells (fig. 26). The obliteration of sieve elements is noticed when a petiole is 1.2 cm long, where the parenchyma cells surrounding them also widen. When the petiole is about 5 cm the fiber precursors show wall thickening (fig. 26) and they differentiate into fibers in a mature petiole (fig. 28). In contrast to the middle region and proximal pulvinal region of the petiole, the distal region of the petiole do not show well developed protophloem fibers (fig. 27).

3.2.14.4 Secondary phloem

The structural characteristics accompanying the transition of procambium-metacambium-cambium are already mentioned in section V. They generally signal or/and accompany the development of secondary tissues. They are evident in a 5 cm long petiole, but as mentioned earlier the vascular cambium is observed only after the petiole ceases to elongate. The secondary phloem is derived from the vascular cambium which is composed of two kinds of cells, the elongated fusiform cells oriented with their long axis

parallel with the longitudinal course of tissue, and short ray cells. The phloem also hence reflects the structure of cambium and is composed of two systems, the axial and ray system. Secondary phloem elements are observed in radially arranged complexes along with the cambium (figs. 27 and 28). Derivative of a fusiform cambium cell divides and arranges to form a component of the complex of phloem cells which include sieve tube element, companion cells and phloem parenchyma. Adjacent complexes are distinguished by thick radial walls. The ontogenetic relationship between the cambial cell and phloem elements is already explained in section V.

3.2.14.4.1 Sieve tube element

The sieve tube elements vary in length. Ontogenetically they appear short because of the transverse division in the fusiform cambial cell. A sieve tube element is generally associated with a single companion cell (fig. 29) as in the primary phloem. The end walls are mainly transverse and the lateral sieve areas are aggregate (fig. 31).

3.2.14.4.2 Companion cell

Each sieve tube element is associated with a single companion cell as long as the associated sieve tube element (fig. 29).

3.2.14.4.3 Slime

P-protein and other contents accumulate at the sieve plate and have a funnel form of configuration (fig. 30).

3.2.14.4.4 Phloem parenchyma

The phloem parenchyma cells vary in width and may be as large as or larger than the sieve tube element. It includes both the axial and ray parenchyma (fig. 27). Axial parenchyma cells are elongated and with mainly transverse or oblique end walls. These cells are uninucleate and have large prominent round nuclei.

The phloem rays are continuous with the xylem rays. They are one or two cells wide (fig. 29) and many cells high. They have more or less rounded or transverse end walls and are uninucleate with round nuclei.

3.2.14.5 Developmental changes in phloem

Protophloem sieve tube elements especially the first formed ones appear singly or in pairs rather uniformly distributed among parenchyma cells (figs. 1 and 3). They have a very short existence. They mature during the early elongation of the petiole and within a short time become stretched and crushed. Obliteration is first noticed when a petiole is 1.2 cm long (fig. 32). With the elongation of the petiole radial seriation in the procambium young of the vascular strand becomes prominent. The protophloem elements are physically stretched with the elongation of the petiole

and obliterated (fig. 33). With the appearance of metacambium the radial seriation becomes increasingly prominent. There is a gradual increase in the width of the sieve tube element. Parenchyma cells in radial rows differentiate (fig. 15) with the development of the two distinct cell systems in the metacambium.

With the cessation of the petiole elongation the protophloem undergoes structural changes due to obliteration and only fibers and parenchyma cells are evident in the mature petiole. The thickened sites adjacent to the protophloem fibers represent the obliterated sites of the sieve tube elements and companion cells (fig. 27).

As there is no clear demarcation between the last formed protophloem and first formed metaphloem it was difficult to say when exactly obliteration began in the metaphloem elements. But when a petiole is 5-6 months old metaphloem was almost completely obliterated (fig. 28). The dimensional features of the primary and secondary sieve tube elements and companion cells in the petiole are shown in table IX.

3.2.14.6 Secondary phloem in the bark

In transection the phloem cells are in radial rows (fig.34) along with the cambium. The secondary phloem consists of the axial and ray system cells. The axial system of cells consists of sieve tube elements, companion cells and phloem parenchyma cells arranged in radial seriation. Groups

of sclereids separate the soft tissue of the phloem in compartments (fig. 27). A comparative structure of the secondary sieve tube elements and companion cells of the petiole and bark is mentioned in table X.

3.2.14.6.1 Sieve tube element

The sieve tube elements are conspicuously large with a clear lumen (fig. 35). They have transverse or oblique end walls and simple sieve plate.

3.2.14.6.2 Companion cell

Each sieve tube element is associated with a single narrow companion cell as long as or shorter than its associated sieve tube element.

3.2.14.6.3 Phloem parenchyma

Phloem parenchyma cells are intermediate in width between sieve tube element and companion cell and are more or less uniformly distributed throughout the phloem (fig. 34).

3.2.14.6.4 Slime

The contents and P-protein accumulate at any one end of the sieve plate in the form of a dense mass (fig. 35).

3.2.14.6.5 Rays

Rays are uniseriate, biseriate or multiseriate. They are 1-3 cells wide.

Table IX. Dimensions of protophloem, metaphloem and secondary phloem sieve tube elements and companion cells during the elongation of the petiole. Dimensons are in μm. Measurements are confined to the middle region of the petiole.

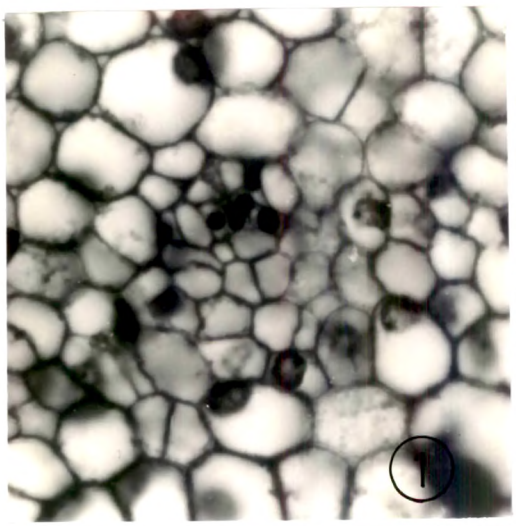
	Protophloem				Metaphloem				Secondary phloem			
Length of petiole in cm	Sieve tube element		Companion cell		Sieve tube element		Companion cell		Sieve tube element		Companion cell	
	Length	Width	Length	Width	Length	Width	Length	Width	Length	Width	Length	Width
0.07	40.3-61.1	2.0-3.0	40.0-60.0	1.5								
1.2	67.5-86.3	3.9-4.7	67.0-86.3	1.5-2.0								
2					73.8-165.4	5.4-6.2	74-165	1.5-2.0				
5					62.8-136.5	5.8-6.2	61-136.5	2.0-3.0				
8.5					160.4-280.0	6.0-6.5	160.0-278.5	2.5-3.0				
13.5									112.0-242.6	9.0-13.0	112.0-242.6	3-4

Table X. Comparative structure of the secondary phloem sieve tube elements and companion cells of the petiole and stem

	Petiole	Stem
1) Tissue types	Axial and ray system	Axial and ray system
2) Components	Sieve tube elements, companion cells, axial parenchyma and ray parenchyma	Sieve tube elements, companion cells, axial parenahyma and ray parenachyma
3) Sieve tube element		
a) Length µm	112-242.6	270-346
b) Width µm	9-13	18-25
c) End walls	Transverse	Transverse or oblique
4) Companion cell		
a) Number	one	one
b) Length µm	112-242.6	270-346
c) Width µm	3-4	9-13

Plate 33

Figs. 1 - 6.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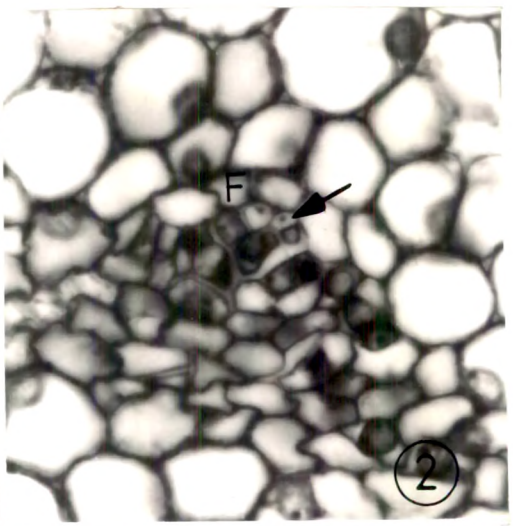
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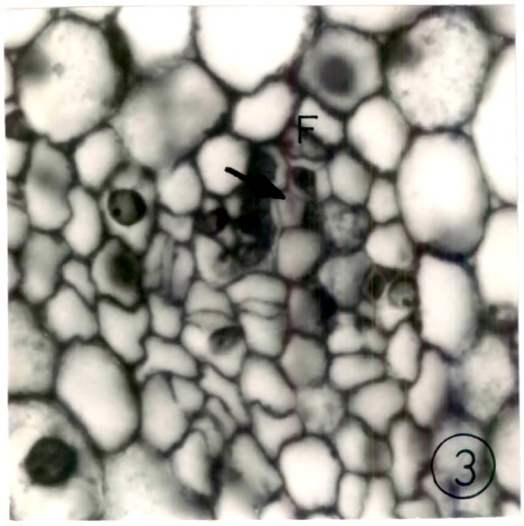
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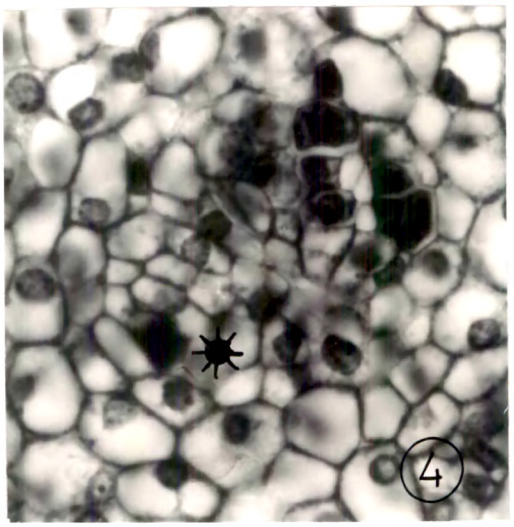
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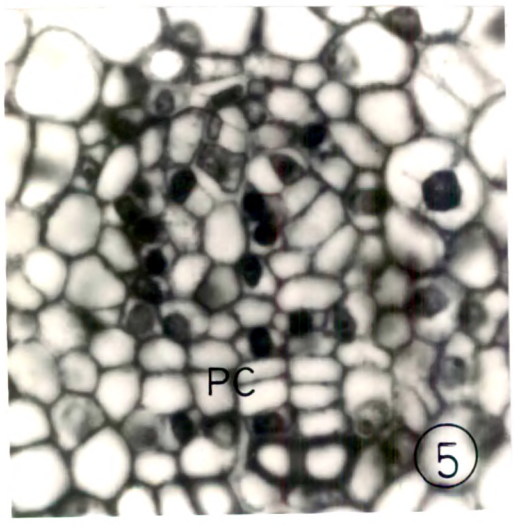
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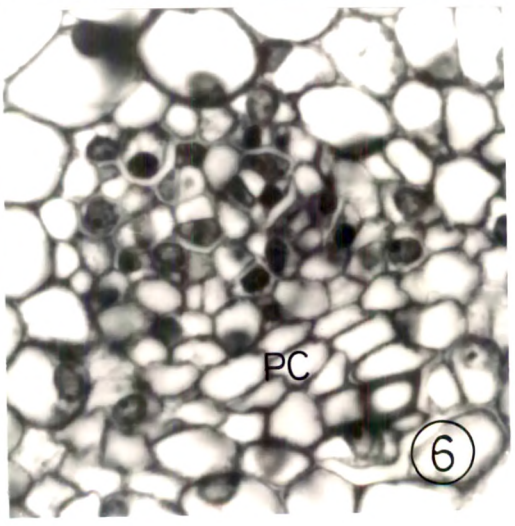
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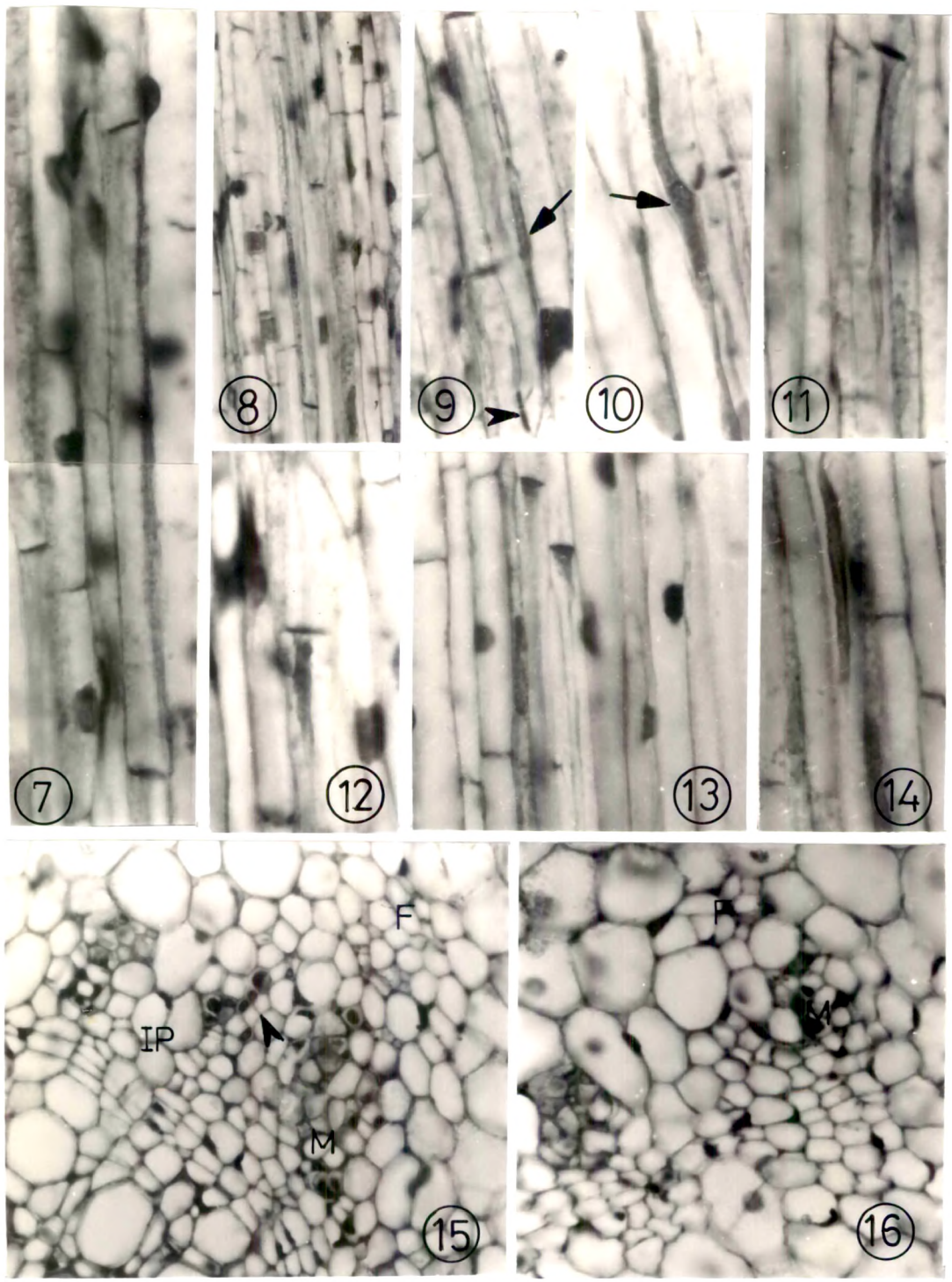
Plate 34

Figs. 7 - 16.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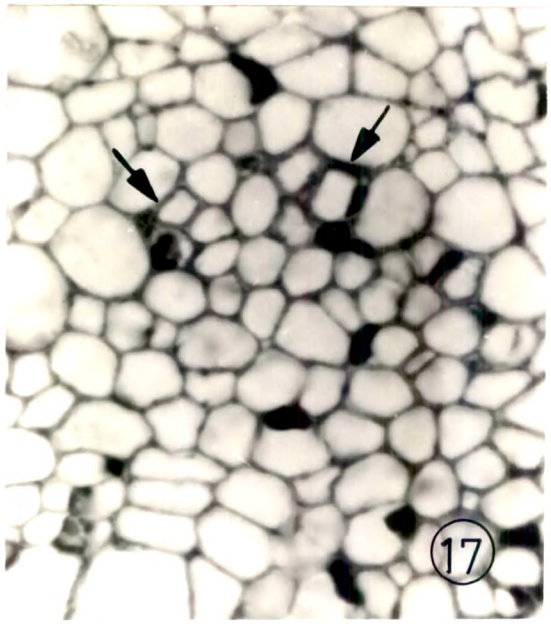
Figs. 17 - 26.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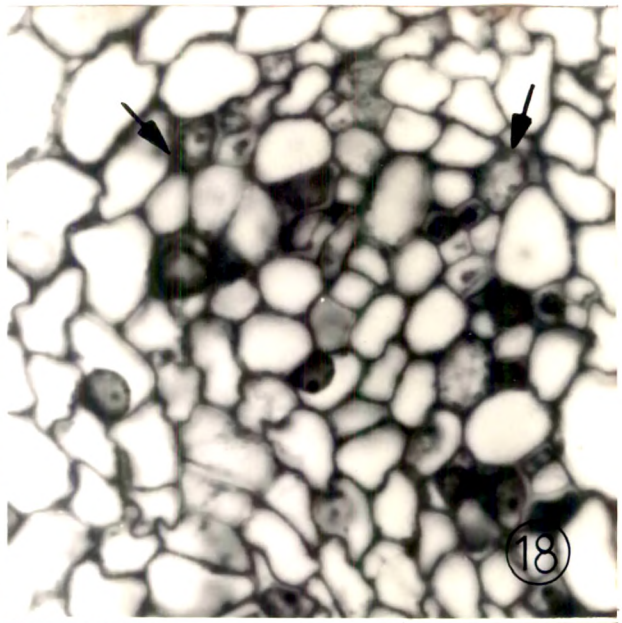
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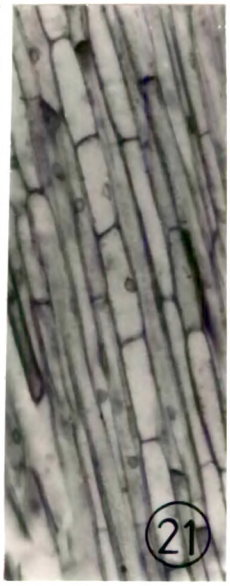
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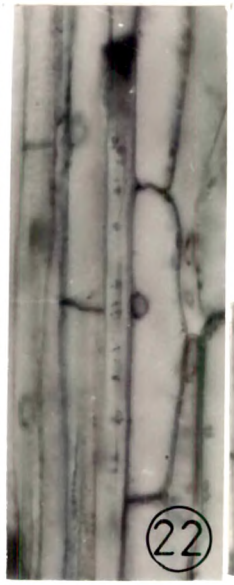
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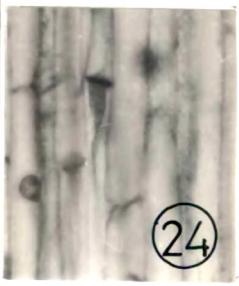
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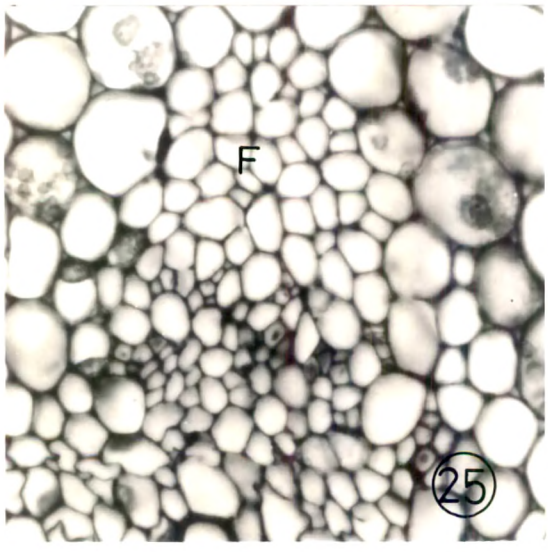
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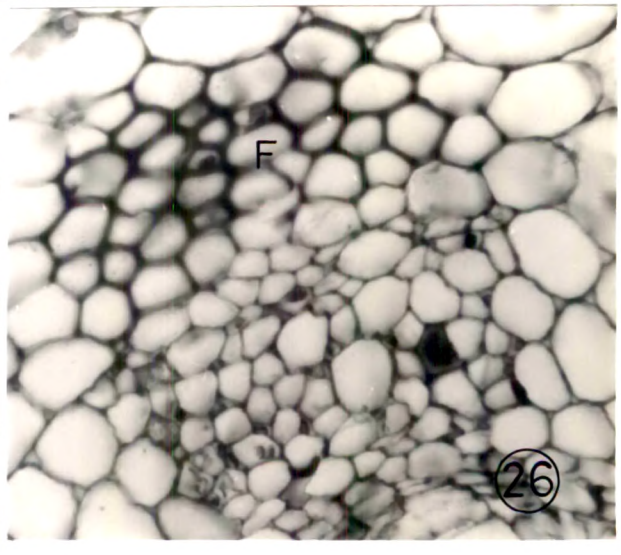
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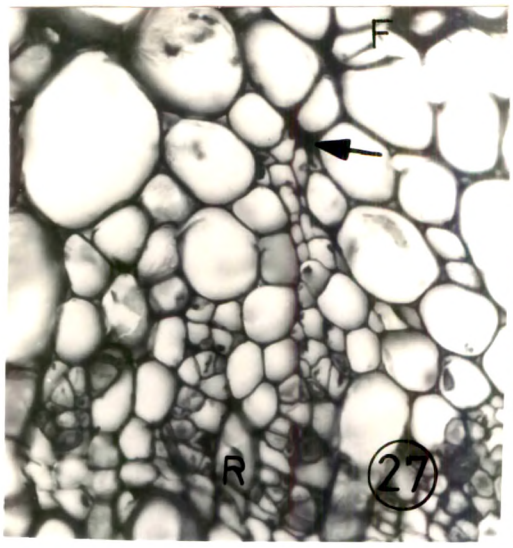
Plate 36

Figs. 27 - 35.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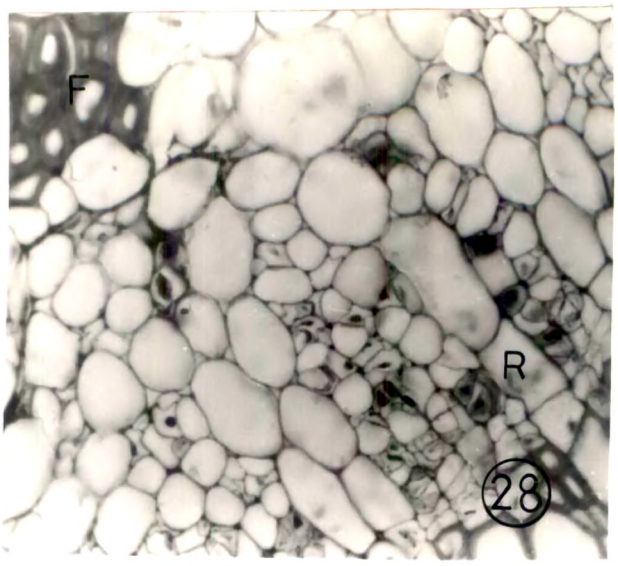
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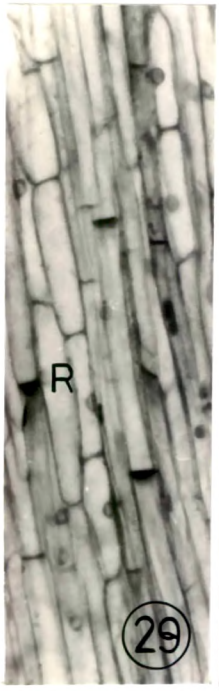
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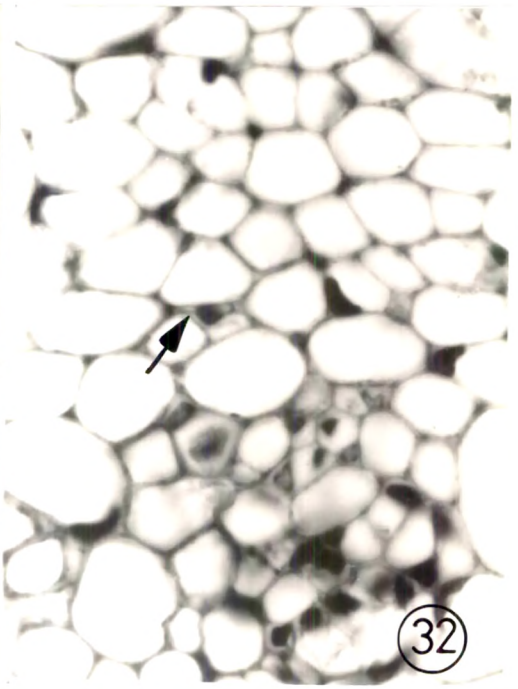
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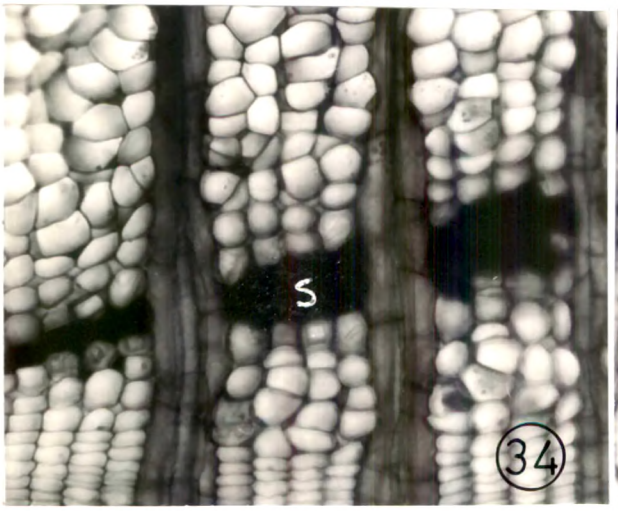
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