CERTAIN ASPECTS OF THE LIFE HISTORY OF HIBISCUS CANNABINUS LINN. WITH REFERENCE TO the development and properties of bast fibre

THESIS SUBMITTED IN PARTIAL FULFILMENT OF THE REQUIREMENTS FOR THE DEGREE OF DOCTOR OF PHILOSOPHY IN BOTANY

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## 1. INTRODUCTION

The most outstanding feature of the family Malvaceae is the presence of fibres. Two kinds of economically important fibres found in this family are the seed fibre and the bast fibre. By far the best known seed fibre is cotton. Bast fibres occur in genera Hibiscus, Sida, Abutilion, etc.

Malvaceae is known embryologically for quite sometime. Guignard (1904) and Stenar (1925) have observed the pollen tube branching in the region of nucellus in some members of Malvaceae. Lantis (1912) has studied the development of microsporangia and microspores of Abutilon theophrasti. Stenar (1925) has studied the number of pollen tubes from the pollen grains of Malva neglecta. On account of its economic importance, the genus Gossypium has received special attention. Gore (1932) has studied the development of female gametophyte and embryo in cotton. Ayyar and Ayyangar (1933) have studied the differentiation of hairs on the seed of cotton. Reeves (1936) has made a detailed study of the comparative anatomy of the seeds of cotton and other Malvaceous plants. Lang (1937) has investigated the morphology and cell inclusions and Iyengar (1938) has studied the pollen tubes of Gossypium. Cotton ovule, morphology and embryology have been studied by Joshi, Wadhwani and Johri (1967), Jensen (1968), Jensen and Fisher (1968a, 1968b) and Pundhir (1972).

The foregoing account indicates that development of gametophytes of the members of Malvaceae has been studied in detail but comparatively little has been studied of the embryogeny. The only complete account of embryogeny is that of Soueges (1922) in Malva rotundifolia.

Rao (1954, 1955) has dealt with sporogenesis, gametogenesis and development of endosperm and embryo in some species belonging to Malvaceae including Hibiscus solanora L., H. micranthus L., Hibiscus hirtus L. and Ablemoschus esculentus L. The gametogenesis of Hibiscus micranthus L. has also been studied by Ahuja (1964). A brief account on the embryology of $H$. cannabinus grown in Western Georgia has been reported by Medvedeva (1944).

Guttenberg (1947) has studied the development of apical regions of the roots of Malva alcea and Althea rosea. The root apices of Gossypium hirsutum and Althea rosea have been studied by Pillai and Girijamma (1965). Tolbert (1961) has stufed the vegetative shoot apex of Hibiscus syriacus. Tolbert and Johnson (1966) have made a survey of the vegetative shoot apices of 40 species of Malvaceae including a few species of Hibiscus. This work, however, does not include $H$. cannabinus.

Metcalfe and Chalk (1950) have reported leaf histology and petiole structure of Althea, Gossypium, Hibiscus cannabinus, Abutilon, Malva, Pavonia, Sida and Sidalcea. These authors (Metcalfe and Chalk, 1950) have
given an account of different trichomes present in Gossypium, Thespesia, Althea, Malva, Pavonia, Kokia and Lagunaria, including those of Hibiscus. Kundu and Rao (1955) have studied the origin and development of axillary buds in Hibiscus cannabinus.

Wilson (1937) in his study on the phylogeny of stamens has given an account on the floral vasculature of members of Malvaceae including that of Hibiscus tiliaceus. Floral vasculature of Abutilon indicum and Malvaviscus arboreus including two species of Hibiscus viz. H. solandra and $\underline{H}$. micranthus has been studied by Rao (1952). Floral anatomy of Hibiscus micranthus has been studied by Ahuja (1964).

In Malvaceae the seed fibres, commonly known as cotton fibres of Gossypium are the oldest and best known fibres. Much work has been done on cotton from eariy times and till date studies on cotton are continuing. Recent work on electron microscopic study of cotton fibre develop ment has been reported by Berlin and Ramsey (1970). Quite a few members of the family are known to yield bast fibres of economic importance. The bast fibres of Sida, Abutilon, Urena and species of Hibiscus have been reported by Matheas (1948). Iakushevskii (1959) has studied the commercial utilization of a new plant perennial, sida. A note on Hibiscus sabdariffa var altissima as an economically important plant has been published by Sambamurthy (1953). Kundu $(1941,1954)$ has studied the development of
bast fibres in the stems of di cotyledonous plants and in Hibiscus sabdariffa. The correlation of plant height, base diameter of the plants and fibre yield have been worked out in three varieties of Hibiscus sabdariffa (sanyal and Dutta, 1961). Maiti (1969) has given a full account on a new fibre crop, Hibiscus vitifolius.

Literature on Hibiscus cannabinus has been reviewed by Crane (1943) under botanical classification and description of varieties; polymorphism in leaves; pollination and cytogenetics; soil environment; cultural requirements; retting; diseases and insect pests and fibre uses. Ergle, Robinson and Dempsey (1945) have studied the growth conditions, yield and mechanical processing of bast fibres in Hibiscus cannabinus. Crane and Acuna (1945) have reported growth and development of kenaf (Hibiscus cannabinus L.) with special reference to fibre content of the stem and have suggested that $\underline{H}$. cannabinus should be harvested during the flowering stage in order to obtain good bast yield and separation of the fibre from the stem. Watkins (1946) has studied the growth and fibre production of kenaf Hibiscus cannabinus L. as affected by plant spacing in El Salvador. Das, Mitra anc Wareham (1950) have studied the composition of $\underline{H}$. cannabinus fibres in brief and compared with jute. Literature on Hibiscus cannabinus as a substitute for jute has been partially abstracted by McCann, Lewis (1952) under the following headings: history; distribution and use; current inportance; range of
adaptation; agronomic and cultural aspects; breeding and selection; uses and production possibilities in U.S.A. The experimental culture of Kenaf has been made by Seale, Joyner and Gangstad (1952) in South Florida for fibre and seeds to produce better yields than H. Sabdariffa. A comparative study on some chemical characteristics of jute and Hibiscus fibre has been done by MacMilian, sengupta and Roy (1956). Serbanescu-Jitariu and Mitroiu (1961) have investigated the differentiation of fibres in Althea cannabina, Hibiscus cannabinus and Abutilon theophrasti. Sakalo (1963) has observed that $\underline{H}$. cannabinus individuals with side branches have short fibres and low yield while those without side branches have long fibres and greater yield. Dutta and sen (1967) have studied the fibres of $\underline{H}$. sabdariffa and $\underline{H}$. cannabinus as a substitute fibre crop of India. Killinger (1969) has reported H. cannabinus as a multi-use crop. Preliminary results on the study of $\underline{H}$. cannabinus as a new material for the pulp and paper industry have been made by Claramello and Azzini (1971). Esau and Morrow (1974) have examined the spatial relation between $x y l e m$ and phloem in the stem of H. cannabinus L.

This review of literature indicates that investigations on different aspects of $H$. cannabinus have been fragmentary and theref ore the plant has been taken for investigation. The following aspects have been studied:

1. Development of gametophytes.
2. Fertilization, endosperm, seed coat and embryo development.
3. Root and shoot apical organization.
4. Seedling vasculature.
5. Phyllotaxy and primary vascular system.
6. Leaf histology, venation pattern and trichomes.
7. Floral vasculature.
8. Bark anatomy.
9. Origin and development of fibres from seedling upto post-fertilization stage and their properties.

Hibiscus cannabinus commonly known as 'kenaf' also 'Ambari', is a native of Africa and had been later introduced in other countries. It seems to grow well on all types of soils and upto an elevation of 1500 m . It requires four months from the time of sowing until harvesting.

Hibiscus cannabinus Linn. is an annual or perennial herb, 6-8 feet high, 1.5 cms in diameter, stem glabrous, sparsely prickly. Lower leaves unlobed and upper deeply palmately 3-5 lobed, ovate-cordate, mid-nerve glandular beneath, sparingly hairy on both surfaces, lobes are narrow lanceolate, serrate, acute; petioles long, lower petioles much longer, heiry, stipulate, stipules linear subulate, hairy. Flowers solitary, axillary or in terminal raceme
due to shortening of leaves, showy, yellow with crims on centre, pedicels very short; segments of epicalyx 7-10, free, linear, shorter than calyx, persistent; calyx 5, bristly, lanceolate, connate below the middle, lobes long, valvate in bud, persistent; petals free, large, obtuse, yellow with a crimson centre; staminal column long; filament short, deep red, arranged throughout the column, anthers reniform, open by vertical longitudinal slits, pollen many globular, spiny; ovary ovoid, densely covered with white silky hairs, 5 locular, ovules many in each chamber in two vertical rows on axile placenta, style connate, stigma 5 spreading,red; capsule globose, pointed, hairy, at maturity hairs become stiff, open by 5 loculicidal valves, seeds large, greyish black, reniform and 3 angled.

This description is based on author's observation and tallies closely with that given by Rakshit and Kundu (1970).

## MATERIALS AND METHODS

2.1 Materials
2.2 Fixatives
2.3 Processing
2.4 Staining
2.5 Clearing
2.6 Fibres
2.7 Chemical Analysis
2.8 Physical Analysis

## 2. MATERIALS AND METHODS

### 2.1 MATERIALS

Plants of Hibiscus cannabinus $L$. were raised from seeds in the Botanical Garden, Pilani, from which the material for the present study was collected.

For the study of floral vasculature and development of gametophytes and embryogeny, buds, flowers and seeds were collected at an interval of 24 hours. Young ovaries were dissected out and studied. Mature seeds were germinated in petri dishes to obtain seedlings and root apices. Seedlings were fixed at regular intervals of 4 th, 8 th, 12 th and 20 th days. Shoot apices were collected from the growing plants.

### 2.2 FIXATIVES

The fixatives used were Formalin-acetic acid alcohol and Allen-Bouin type fluid and and $4 \%$ formalin. The former two were employed for serial sectioning of the material and the latter for free hand sectioning. The second fixative was used for hard stems.

### 2.3 PROCESSING

The materials were dehydrated, infiltrated and blocked in paraffin through ethyl alcohol-xylene series
as well as ethyl alcohol-tertiary butyl alcohol series. sections were cut at 8 to 12 micron.

### 2.4 STAINING

Sections were stained in safranin-light green. For the seed coat study phloroglucinol test was used to determine the presence of lignin and was prepared by mixing a $2 \%$ solution of phloroglucinol in alcohol with an equal volume of concentrated hydrochloric acid. Tannin was determined by staining with a pinch of safranin powder in 1:1 alcohol xylol mixture.

### 2.5 CLEARING

Leaves fixed in F.A.A. and stored in $70 \%$ alcohol were cleared by bringing to aqueous medium by down grading. The material was kept in the clearing solution prepared by dissolving 1 gm of basic fuchsin in $100 \mathrm{c} . \mathrm{c}$. of boiling water and was allowed to cool. 5 gms of NaOH was added to this solution at $40^{\circ} \mathrm{C}$ for $24-48 \mathrm{hrs}$. The leaves were washed with water and kept in water for $24-48$ hours. The material was then dehydrated and permanent preparations were made.

### 2.6 FIBRES

For the study of fibres of $H$. cannabinus L. seeds were sown in bulk and in large well manured plots in the Botanical Garden. To study the course of fibre development
material was collected from the time when fibres were first seen in the stem upto post flowering stage with an interval of fifteen days. Study on development was largely on fresh materials cut free hand.

Permanent slides of bark were made by fixing stems at different stages in Bouin's fixative, dehydrated in Dioxan and blocked in paraffin. Sections were cut at 18 to $25 \mu$ thickness and stained in safranin-light green combination.

In very tender stems where a section does not reveal the fibre, maceration was carried out with $12 \%$ chromic acid and $12 \%$ nitric acid leaving the material in it for 24 hours.

Isolated fibres were taken for chemical and physical analysis. Fibres were isolated by the process of "Retting" as dealt in Chapter 11. An analysis of Hibiscus cannabinus fibres was done by the following methods:

### 2.7 CHEMICAL ANALYSIS

2.7.1 Moisture

Water content of the $f i b r e s$ was studied at three different stages: pre-flowering, flowering and postflowering. To estimate the moisture contents 2 gms of fibre was accurately weighed. Then three pre-weighed silica crucibles were taken and the weighed fibres were
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kept in them. These crucibles were heated at $110^{\circ} \mathrm{C}$ on the thermostat hot plate for 48 hours. Then the crucibles were placed in the dessicator for 5-10 minutes and weighed again. The difference of pre-heated weight of the fibres and the post-heated weight gave the total moisture content of the fibres. This experiment was repeated four times and a mean value of the four was taken.
2.7.2 Ash Content

To estimate the ash content in the fibres at three different stages as described above in 2.7.1, the same samples were taken in the crucibles and were kept in the furnace at $800^{\circ} \mathrm{C}$ for 30 minutes and the total ash content was calculated in percentage by taking the mean of four replicas.
2.7.3 Fat

To estimate fat percentage, 2 gms of powdered fibres of three different stages were taken in a preweighed filter paper which was given prior washing with solvent ether to remove any lipid. Then the filter paper with fibres was kept in the thimble of the soxlet apparatus and it was refluxed with petroleum ether for two days. Then it was dried in air. The defatted fibres were weighed. Difference in initial and final values gave the fat contents. This was repeated three times and a mean value was calculated.

This was estimated by the method as described in "Methods in Plant Physiology" by W.E. Loomis and C.A. Shull, 1937, pp. 292-293.

One gram of finely powdered fibre of the three different stages of flowering was taken and extracted with hot water. Each time fibres were kept in hot water at $100^{\circ} \mathrm{C}$ in an oven for 30 minutes and then water was filtered out on Whatman No. 1 filter paper. Five washings were given and the water extract was pooled in 1 litre flasks. The final volume was made to 300 ml and then 100 nil of 0.1 N NaOH was added in each of the flasks to hydrolyse methyl esters to $\mathrm{Na}^{+}$salts of pectic acid and hydrolysis was continued overnight at room temperature. Then 50 ml of 1 N acetic acid was added to each of the flasks to decompose the sodium salts and allowed to stay for 5 minutes. Next, 50 ml of molar $\mathrm{CaCl}_{2}$ fused or dehydrated was added to form calcium pectate. This solution was allowed to stand for 1 hr . and then the solution was boiled for $10-15$ minutes and the precipitate of calcium pectate was filtered on the Whatman No. 1 filter paper. The weight of filter paper was taken after drying it at $85^{\circ} \mathrm{C}$. The precipitate was washed repeatedly to remove the chloride with triple distilled water until the washings gave no ppt with silver nitrate solution. Then the filter paper was dried at $95^{\circ} \mathrm{C}$ to remove water and it was reweighed to
calculate the percentage. The whole experiment was repeated twice and the mean value was plotted.

### 2.7.5 Lignin

This was also estimated by the method as described in "Methods in Plant Physiology" by W.A. Loomis and C.A. Shull, pp. 298-299.

Before estimating the lignin 1 gm of fibre was weighed and partially hydrolized as given in page 290, Loomis and Shull's "Methods in Plant Physiology". Fibres were given washings with water and the extract was thrown. Then 100 ml of diluted $\mathrm{HCl}(1$ part of HCl and 19 parts of $\mathrm{H}_{2} \mathrm{O}$ ) was added in three flasks and autoclaved for 1 hr at 15 lbs pressure and the extract was filtered. To these partially hydrolyzed fibres was added 15 ml of prechilled $72 \% \mathrm{H}_{2} \mathrm{SO}_{4}(300 \mathrm{gms}$ acid + 100 ml of water). The temperature of this was kept below $12^{\circ} \mathrm{C}$ by keeping in the cold room at $4^{\circ} \mathrm{C}$ to prevent charring. The fibres were allowed to stand at the same temperature with occasional shaking for 48 hours. Then the gelatinous mass was transferred to a 1 litre beaker and diluted with water to 800 ml . All the flasks were covered with watch glass and boiled gently for 2 hours to hydrolyze the material completely. Hot water was added to the beaker to make up the loss due to evaporation. The lignin was allowed to settle and most of the solution was siphoned off and the lignin was washed repeatedly with water. The lignin precipitate was transferred to a
crucible dried at $100^{\circ} \mathrm{C}$, cooled and weighed. Then the samples were ashed and the weight of the ash was subtracted from the weight of residue of lignin. Three repetitions of the experiment were taken and a mean value was calculated in percentage.

### 2.7.6 Cellulose

This was determined by the method as described by H.F. Launer in the "Determination of cellulose in Methods in Carbohydrates Chemistry", Vol. III, edited by R.L. Whistler, 1963, pp. 29-31.

To estimate cellulose, 0.25 gm of fibres obtained at three different stages of flowering were weighed and taken in 500 ml flasks. Then 25.00 ml of 1.835 N potassium dichromate ( 90.00 gms AnalaR $\mathrm{K}_{2} \mathrm{Cr}_{2} \mathrm{O}_{7} \mathrm{dried}$ overnight at $100^{\circ} \mathrm{C}$ per litre) was pipetted in the beaker and the beaker was covered with a watch glass. Continuous rapid stirring was done and simultaneously 10 ml of conc. $\mathrm{H}_{2} \mathrm{SO}_{4}$ was added (AnalaR 1.84 d), after $15-30$ seconds 30 ml more of conc. $\mathrm{H}_{2} \mathrm{SO}_{4}$ was added and stirring was continued. After 3 minutes, 150 ml of water was added in each of the beakers. This solution was used for titration. The excess of $\mathrm{K}_{2} \mathrm{Cr}_{2} \mathrm{O}_{7}$ left was titrated with ferrous ammonium sulphate immediately $\left(0.5 \mathrm{~N} \mathrm{Fe}\left(\mathrm{NH}_{4}\right)_{2}\left(\mathrm{SO}_{4}\right)_{2}\right.$. $6 \mathrm{H}_{2} \mathrm{O}$ ). This was made by dissolving 195 gms of $\mathrm{Fe}\left(\mathrm{NH}_{4}\right)_{2}$ $\left(\mathrm{SO}_{4}\right)_{2} \cdot 6 \mathrm{H}_{2} \mathrm{O}$ and 10 ml conc. $\mathrm{H}_{2} \mathrm{SO}_{4} /$ litre. Solution is unstable, so every day solution was made anew. Then 5.00 ml of titrant was pipetted in a test tube and it was
titrated with $\mathrm{Fe}\left(\mathrm{NH}_{4}\right)_{2}\left(\mathrm{SO}_{4}\right)_{2} \cdot 6 \mathrm{H}_{2} \mathrm{O}$ solution which was added drop by drop and stirred until the indicator turned blue. The external indicator used was $\mathrm{K}_{3}\left[\mathrm{Fe}(\mathrm{CN})_{6}\right]$ potassium ferricyanide. Value obtained was denoted as
T. A reference titration was made by pipetting 5.00 ml of $\mathrm{K}_{2} \mathrm{Cr}_{2} \mathrm{O}_{7}$ solution and titrating it with $\mathrm{Fe}\left(\mathrm{NH}_{4}\right)_{2}\left(\mathrm{SO}_{4}\right)_{2}$. $6 \mathrm{H}_{2} \mathrm{O}$ solution to the end point. This reference value is Tr. Then cellulose was calculated by the formula given below:

$$
V=\frac{25.00-5.00\left(T / T_{r}\right)}{W}
$$

25.0 is the amount of $\mathrm{K}_{2} \mathrm{Cr}_{2} \mathrm{O}_{7}$ solution used in the oxidation of fibres and 5.00 is the titrant used to get the value $T$
$W$ is the weight of the fibre samples taken

$$
\% \text { cellulose }=1.24 \mathrm{~V}
$$

1.24 is a standard value derived from 0.01240 , the grams cellulose equivalent to 1 ml of 1.835 N dichromate times $100 \% . \mathrm{V}$ is the ml of dichromate consumed per gram of the test specimen.

The whole experiment was repeated five times and a mean value was obtained and percentage of cellulose was calculated and plotted.

### 2.8 PHY SICAL ANALYSIS

The following first five methods for physical analysis of fibres, at three different stages of flowering
viz., pre-flowering, flowering and post-flowering were carried out at the Technological Institute of Textiles, Bhiwani.
2.8.1 Tenacity

The tenacity of the fibres was determined at three different stages of flowering on a Stelometer. Fibres of 1.5 cms test length from the middle region of the reed were taken and fed into the stelometer. The reading that the pointer of the stelometer showed was noted in kilograms and the fibres of 1.5 cms test length were then weighed in milligrams. The unit for tenacity is 'tex' which was calculated as following:

$$
\text { tex }=\frac{\mathrm{Kg} \times 15}{\mathrm{mg}}
$$

where Kg is kilograms, mg is milligrams, and 15 is a standard value.

Five samples of each stage were taken and their mean was calculated.
2.8.2 Elongation at Break

This was also determined by the stelometer expressed in terms of percentage. Five samples of each of the three different stages of flowering were taken and their mean was calculated.
2.8.3 Fineness of Fibre

The fineness of 2 mm conbed fibre pieces was determined in terms of 'tex' at the three different stages of flowering where 'tex' is the unit defined as the weight in gms per km of fibre.
2.8.4 Youngs Modulus of Elasticity

Elasticity of fibres was determined by the formula $\frac{F L}{A \Delta L}$, where $F$ is the applied force in dynes, $L$ is the original length in cm of the fibre, A the area of crosssection in $\mathrm{cm}^{2}$ and $\Delta L$ the increase in length of fibre in cm . The unit of Youngs' modulus is dynes $/ \mathrm{cm}^{2}$. Elasticity of fibres was determined at the three different stages of flowering.

### 2.8.5 Density

The fibre density was calculated by dividing the weight of the fibres in grams by its volume in cubic centimetres. Readings at three different stages of flowering were taken.
2.8.6 Cross Sectional Area

The cork method of Viviani and Herzog was used ('Fiber Microscopy' by A.N.J. Heyn, 1954, pp. 145-146). A long sewing machine needle was threaded with a small thread through the centre of a small cork. A pin was placed through the protruding loop, the needle was
withdrawn, leaving the string threaded through the cork. A bundle of paralleled fibres, was placed in the loop and was pulled carefully into the cork. The surface of the cork was squared accurately. Sections moderately thin were cut from the cork with a sharp razor blade. The slice of cork containing the fibre was attached to the coverglass with ${ }^{2}$ drop of glycerin. The coverglass was then mounted on a slide supporting it by a ring of paraffin. The cork sections were cut accurately perpendicular to the axis of the fibre. About 50 images of cross sections were drawn with the camera lucida on graph paper. The graph squares were calibrated with the stage micrometer, and the value of the squares were converted in square microns. Then the number of squares were counted that were covered by the area of the fibre, the cross sectional area of the fibre was obtained. The average surface area of $50 \mathrm{fi} b r e s$ was taken and calculated in square centimetres: a $\times 900,000 \mathrm{~cm}$. where 'a' is area. The areas of 50 fibres at three different stages of flowering were taken and their average was calculated.
3.1 Microsporogenesis
3.2 Male Gametophyte
3.3 Ovule
3.4 Megasporogenesis
3.5 Female Gametophyte
3. DEVELOPMENT OF GAMETOPHYTES

### 3.1 MICROSPOROGENESIS

The hypodermal archesporium differentiates only at two places in the anther primordium (Fig. 1). Anthers are uniformly bisporangiate and the two loculi eventually fuse to form a single loculus, as anthers are monothecous. The archesporial cell divides periclinally to form an outer primary parietal cell and an inner primary sporogenous cell (Fig. 1 and Photomicro. 1). By further anticlinal and periclinal divisions, the parietal cell gives rise to three concentric layers (Figs.2,3 and Photomicro. 2). The epidermal layer stretches tangentially along with the growth of the anther. of the three wall layers, the outermost is the endothecium. The cells of the endothecium, enlarge, become radially elongated and develop fibrous thickenings by the time the pollen grains reach maturity. Inner to the endothecium is a single middle layer that gets crushed by the time microspore mother cells enter meiotic divisions. The innermost wall layer, the tapetum is of large cells with dense cytoplasm and prominent nuclei (Fig. 4). The tapetum is of the periplasmodial type. The cells enlarge and their nuclei undergo mitotic divisions when the microsporocytes are at the beginning of meiosis. The first nuclear division
is followed by further divisions and as a result multinucleate tapetal cells are formed (Fig. 5). The cytoplasm of tapetum leaves the walls of the cells and come to lie in the centre as the meiotic divisions in the microspore mother cells come to an end. The tapetal cells loose contact and lie separate in the anther loculus with their walls intact until the microspores attain a definite size and bear short soines and germ pores (Fig. 6). At this stage, the cell walls breakdown and the protoplasts protrude out and wander inside the loculus where they coalesce to form a continuous mass surrounding the growing mi crospores (Photomicro. 5). The periplasmodium becomes lesser and lesser as the pollen grains become more spinescent and is ultimately abs orbed (Fig. 7 and Photomicro. 6).

The primary sporogenous cells undergo a few mitotic divisions to increase the number of spore mother cells arranged in a single row in each anther lobe (Fig. 3 and Photomicro. 2). The nucleus of the microspore mother cell enlarges with the thickening of the cell wall. Meiosis occurs and the four daughter nuclei become organized (Photomicro. 3). Cytokinesis takes place by furrows and is simultaneous (Fig. 4). The microspores are arranged in tetrahedral and isobilateral fashion (Fig. 8A-8B and Photomicro. 4). Each microspore has dense cytoplasm and a prominent nucleus. It has a wavy and smooth outline to start with (Fig. 5) but soon takes
a rounded form. The pollen grains are quite large and range from 80-130 $\mu$ in size. Some degenerating pollen grains are also found associated with normal ones (Fig. 6). The pollen grains are spinescent and multiporate. The spines are long and tapering. The exine is thick ranging from $8-10 \%$. It has two layers, the inner thick and smooth layer and the outer thin layer. The spines and the pores are uniformly distributed on the exine (Fig. 7 and Photomicro. 5).

### 3.2 MALE GAMETOPHYTE

The microspore has dense cytoplasm and a centrally placed nucleus (Fig. 7). The nucleus undergoes division giving rise to the large vegetative and small generative nuclei (Fig. 9 and Photomicro. 6).

The vegetative nucleus has dense cytoplasmic sheath of its own. The generative nucleus migrates and comes to lie towards the wall (Figs. 10-11). At early stages, the generative cytoplasm is relatively hyaline in appearance. Division of the generative cell occurs in the pollen grain (Fig. 12 and Photomicro. 7). By the time the vegetative cell degenerates cytokinesis takes place resulting in bipartitioning of the generative cell. Two male gametes each with a nucleus and a sheath of cytoplasm are thus formed (Fig. 13). The pollen grains are shed after the formation of male gametes.

### 3.3 OVULE

The ovules are bitegmic, crassinucellate and campylotropous. Initially the ovule is anatropous but later curves to attain the campylotropous condition. The initials for both the integuments do not arise at the same time. The inner integumentary primordia differentiate first followed by those of the outer integument (Fig. 14 and Photomicro. 8). The outer integument grows faster than the inner and covers the nucellus by the time megaspore mother cell is fully grown. The outer integument is $2-3$ layers across and the inner is 4-8 layers across. The micropyle formed by both the integuments has a zigzag form. Uvules are variously inclined andthe micropyles point towards the axis of the ovary. The nucellus is markedly curved. The micropylar part of the nucellus is many layered thick, derived fromthe epidermal and parietal cells of the nucellus. These cells seem to be spreading out from the megaspore mother cell. The antipodal end of the emrbyo sac bears tanniferous cells.

### 3.4 MEGASPOROGENESIS

A single cell of the nucellus situated directly below the epidermis becomes more conspicuous than the rest of the cells because of its large size, dense cytoplasm and prominent nucleus. This is the hypodermal
archesporial cell (Fig. 14). As anticlinal divisions in the epidermis initiate the integuments, the archesporial cell undergoes periclinal divisions to give rise to a primary parietal cell and a primary sporogenous cell (Fig. 15 and Photomicro. 8). The parietal cell by further anticlinal and periclinal divisions contributes to the growth of the nucellus (Figs. 16,17 and Photomicro. 9). Occasionally two hypodermal cells function as archesporial cells (Fig. 18). Only one of the two functions beyond $L^{\text {The }}$ megaspore mother cell stage. The cells of the parietal tissue appear spreading out from the megaspore mother cell (Fig. 20A and Photomicro. 12). The primary sporogenous cell functions as the megaspore mother cell.

The megaspore mother cell is elongated and tapering (Fig. 17 and Photomicro. 10). It undergoes transverse division to form the dyad. The upper cell of the dyad is smaller than the lower (Fig. 19 and Photomicro. 11). The second division is also transverse, resulting in a linear tetrad of megaspore (Fig. 2OA). The chalazal megaspore functions further (Fig. 20 A and Photomicro. 12). Accessory sporogenous cells develop from one or more cells of parietal tissue. The accessory sporogenous cells become demarcated at the time of meiosis in the functional cell. These cells are seen till the functional cell has formed ints a tetrad. Leaving the functional chaluzal megaspore the upper three cells
of the linear tetrad and the accessory sporogenous cells degenerate (Figs. 20A and 20B). The cells in the central region of the nucellus meanwhile become elongated and those in the chalazal region divide in all planes to produce a mass of small cells of indefinite arrangement.

### 3.5 FFMALE GAMETOPHYTE

The functional megaspore beccmes vacuolated and its nucleus comes to lie in the centre (Fig. 20B). It divides to give rise to two nuclei each of which migrates to opposite poles(Fig. 21). Each of them divides to give rise to a four nucleate embryo sac (Fig. 22). Further divisions of the four nuclei give rise to the eight nucleate embryo sac. The synergids become slightly hooked. The polar nuclei meet at about the midale of the sac and travel together upwards and come to lie near the egg apparatus (Fig. 23). The three antipodals persist till the eight celled stage of embryo development.

# FERTILIZATION AND EMBRYO DEVELOPMENT 

### 4.1 Fertilization

4.2 Endosperm
4.3 seed coat
4.4 Embryo Development

### 4.1 FERTILIZATIUN

The pollen grains are large and polysiphonous. The entry of pollen tube is porogamous. Pollen tubes are formed in closed flowers. Thus self-pollination occurs. The pollen tubes reach nucellus through the michopyle In a zigzag manner. Inside the nucellus the pollen tubes become broad and send out branches. The diameter of the pollen tube ranges from 10-24 $\%$. Some shori branches end abruptly midway and others extend to the embryo sac. While the main tube progresses towards the egg destroying the cells in its path. When in contact with egg cell the pollen tube becomes broad, massive and branches profusely. The branches encircle the egg and the branch with one of the gametic nuclei sheds its nucleus into the egg cell (Figs. 33,34 and Photomicro. 13). The male gametic nuclei show a distinct nucleolus in the centre surrounded by a vacuolated region (Fig. 35 and Photomicro. 14) similar to that of egg nucleolus. This particular branch of pollen tube degenerates and the rest persist. The persistent tubes are recognizable till the heart shaped stage of embryo (Fig. 32). Its open end faces the base of the developing embryo. A cross section of the pollen tube shows a narrow lumen and a thick wall. The male nucleus remains embedded in the cytoplasm of the egg until the
endosperm attains $4-8$ nucleate stage, when the syngamy oc curs.

### 4.2 ENDOSPERM

The two polar nuclei remain adpressed until fertilization occurs (Fig. 22). At the time of fertilization the polar nuclei come to lie beneath the egg. of the two male gametes one fuses with the polar nuclei to form the triploid-3-nucleclate primary endosperm nucleus. Usually 10-16 endosperm nuclei are formed before the zygote diviaes (Fig. 24 and Photomicro. 15). The number of nucleoli in each nucleus varies from 1-4. Uptil early globular stage of thembryo the free nuclei of the endosperm aggregate in a compact manner encircling the growing proembryo (Fig. 25 and Photomicro. 16). The endosperm during cell formation first gets cut up into uninucleate protoplasts. Later cell walls are formed, starting from the micropylar end at the late globular stage of embryo. subsequently, it progresses towards the chalazal end. The endosperm can be distinguished in three zones: (a) micropylar cellular zone, (b) central partially cellular zone, and (c) chalazal free nuclear zone (Fig. 26 and Photomicros. 17-19). The nuclei at chalazal end are oval, spherical or elliptical. In chalazal portion of endosperm, nuclear fusion occurs resulting in large polyploid nuclei (Fig. 26). The chalazal endosperm becomes cellular about the time of the torpedo shaped embryo when the micropylar endosperm shows an advanced cellular condition. The endosperm is
largely consumed by the growing embryo so that only a part of it is left at the mature embryo stage.

The embryo sac lies in contact with the nucellus. The nucellus $f$ arms a storage tissue, the perisperm. At the globular stage of embryo the perisperm at the micropylar end is 12 tc 13 cells in thickness but as the embryo grows, the number of cells decrease so that at the mature embryo stage the nucellus is 2 to 3 cells in thickness.

### 4.3 SEEED COAT DEVELOAMENT

4.3.1 Structure of the Integument

The ovule is bitegmic. The outer integument, at early stages, overgrows the inner one. At the time of fertilization both the integuments form a 'zigzag' micra pyle. The outer and inner integuments are well marked at the fertilized egg stage (Fig. 27A and Photomicro. 20). The outer integument is composed of two definite layers except at the micropylar end where it forms a flared tip. The outer epidermis of the outer integument has uniform 1sodiametric cells. The cells of the inner epidermis of the outer integument are small with prominent nuclei. The flared tip of the outer integument is composed of parenchymatous cells (Figs. 27A, 27B).

The inner integument is 6 to 7 cells thick. The cells of its outer epidermis are isodiametric and richly protoplasmic at the fertillzed egg stage. Their nuclei

Ile at the same level giving it a characteristic appearance (Fig. 28 and Photomicro. 21). The inner epidermis of the inner integument comprises tangentially flattened cells. This layer is designated as the 'fringe' tissue. These cells contain tannin. Abutting the fringe tissue on the outside are 3 to 6 layers of parenchymatous thin walled cells. Next occurs, uniseriate layer of large tanniferous cells (Fig. 27B and Photomicro. 21) which, however, is multiseriate at micropylar and chalazal ends and scattered throughout the funicular region. This layer is termed as 'pigment layer'.
4.3.2 Changes in the Integuments

Outer integument: Both the integuments take part In the formation of seed coat. The outer epidermis of the outer integument, during the development of the seed, get tangentially flattened. Epidermal papillae develop when the embryo is at [he globular stage. The inner epidermal cells remain isodiametric, divide anticlinally to keep pace with the growing ov ule (Fig. 31).

Inner integument: The inner integument constitutes the major portion of the seed coat and forms the tegmen. The tegmen varies in thickness at different parts of the same seed. In the region of micropyle, it is thinner, and thicker at the chalaza and funiculum. At the globular stage of the embryo, the parenchymatous cells enlarge, the outer 2 to 3 rows of cells remain compact and the inner

4 to 5 layers become radially elongated at places. The outer epidermis enlarges further at this stage and their nuclei lie at the same level of each cell (Fig. 28 and Photomicro. 21). At the late globular stage the outer epidermal cells divide anticlinally. Concomitant with their divisions, the outer epidermal cell elongates radially and become columnar. The nuclei are still prominent but move upwards along with the cytoplasm, with the beginning of lignification at the lower region of the cells (Fig. 29). At the heart shaped embryo stage, the outer epidermal cells further undergo radial elongation, chalazal region upwards and become palisade like.

At the torpedo stage of embryo the inner walls of palisade cells, show fibrous thickenings. The fibrous thickening gradually covers more then half the region of the cell and nucleus by this time degenerates. A very littie portion at the upper end of the cell remains unlignified (Figs. 30A, 30B and Photomicro. 22).

At the mature stage of embryo the outer integument becomes membranous and has numerous papillae (Fig. 31). The palisade like cells of the tegmen are large and prominent covering more than half the thickness of the seed coat. Each cell becomes almost completely lignified. The pigment layer and the fringe tissue with a few crushed parenchymatous cells at the centre are left at this stage. (Fig. 31 and Photomicro. 23).
4.4 EMBRYO DEVELOPMENT
4.4.1 Proembryo

The first division of the zygote is transverse forming a terminal cell 'ca' and a basal cell 'cb' (Fig. 36 and Photomicro. 24). The terminal cell 'ca' divides longitudinally and the basal cell, 'cb' transversely into 'cí' and ' $\underline{\underline{\prime}}$ ' so that a $T$-shaped proembryo comprising of four cells disposed in three tiers is formed (Fig. 37). The terminal cell region is one-tiered after another vertical division in a plane perpendicular to that of the first divisions forming quadrants designated as ' $g$ '. In the next stage, the proembryo consists of eight cells placed in four-tiers. The original terminal cell has completely resolved into quadrants ' $g$ '. The tier ' $m$ ' divides longitudinally and 'ci-' divides transversely to give rise to two new tiers ' $\underline{n}$ ' and 'n' (Fig. 38 and Photomicro. 25). The $t$ erminal tier of quadrants ' $g$ ' divide in an oblique manner giving rise to four centrally placed cells designated as 'á' and four peripherally placed cells termed 'b' (Fig. 39).

Each of the eight cells become bipartitioned to give rise to a 16-celled proembryo disposed in five tiers. Tier ' $m$ ' now includes four circumaxial cells in one tier. ' $n$ ' divides vertically into two cells while region ' $n$ '' divides transversely into two tiers, the upper daughter cell termed as ' $o$ ', while 'he lower daughter cell as ' $\underline{\prime}$ ' the which ultimately gives rise to|suspensor. At this stage
transverse walls are laid in 'a' and 'b' regions of the terminal tier (Fig. 40). Thetier ' $m$ ' undergoes anticlinal divisions and ' $n$ ' also divides in the same manner as in 'm'. 'o' divides vertically forming two cells (Fig. 40). The demarcation of dermatogen in the terminal tier occurs first in the cells ' $b$ ' and then in the group of cells 'a' (Fig. 39). One of the two cells of 'a' of the tier ' $q$ ' divides transversely cutting off the dermatogen initials to the outside but the other divides in an inclined manner so as to form a cell that looks like an epiphysial cell (F1g. 41). In later stages it lobses its identity and appears like other cells. After the first radially vertical wall has been laid in ' $b$ ', the inner daughter cell divides again. The outermost cell is an epidermal initial. The inner daughter cell that has divided, gives rise to an outer periblem initial 'mc' and an inner plerome initial 'mv' (Fig. 41).

The quadrants of ' $m$ ' undergo periclinal divisions to demarcate the dermatogen initials to the outside and the common periblem-plerome initials to the inside (F1g. the 42). At the 16 -celled stage, ftier ' $n$ ' is composed of two cells both dividing longitudinally to give rise to four circumaxial cells (Fig. 41). Further by harizantal walls ' $n$ ' forms two distinct layers of 3 and 4 cells (Fig. 43). These cells form the root cortex. The tier ' 0 ' in the sixteen celled proembryo divides vertically to give two juxtaposed cells (Figs. 40-41). These cells divide horizontally to give rise to two groups of superposed
cells of unequal number (Fig. 43). They give rise to the root cap.

Thetier 'p' gives rise to the suspensor which is quite short and divides transversely into two cells (Figs. 42-43). The upper short stalk connecting the embryo to the larger vesicular lower cell.

The development of embryo, thus, conforms to the 'Urtica variation of the Asterad Type'. The schematic representation would be as follows:


### 4.4.2 Globular Embryo

The early globular embryo is embedded in the massive endosperm with its one celled suspensor intact, surrounded by perisperm. Nuclear divisions are randomly distributed, majority of the divisions are evident in the distal half, i.e., the part away from the suspensor, as a result the embryo becomes ovoid and broader than the suspensor end (Figs. 42-43 and Photomicro. 26). At the mid globular stage of the embryo the suspensor is still intact and becomes two celled. The cells of the region 1 mmediately beneath the suspensor, of tiers ' $O$ ' and ' $n$ ' are large and
narrower than the distal region due to lesser divisions (Fig. 43). Cell divisions are predominantly anticlinal although periclinal divisions may occur in some of the cells of the surface layer (Fig. 43).

By the late globular stage of embryo no remanant

Theof $h$ suspensor was $f$ ound. Due to frequent divisions the globular embryo flattens (Fig. 44 and Photomicro. 27), the axial symmetry is lost and cotyledon primordia appear making terminal region broader than the basal region. The surface layer continues to divide anticlinally, except in the middle region of the basal part where the root cap is to develop periclinal divisions are evident (Fig. 44). This is the first indication of the formation of root cap. By this stage transverse rows of large cells of ' $m$ ' become radially aligned because of root cap growth and form an elongated central core of cells (Fig. 44). The large, deeply stained distinct cell with a prominent nucleus could be recognized designated as 'central cell' (Photomicro. 27).
4.4.3 Heart Shaped Embryo

At the heart shaped stage of embryo the cotyledons are perceptible. In the cotyledmary regions the dermatogen at the tips shows anticlinal division. In the second and third layers periclinal divisions predominate, oblique divisions are also found (Fig. 45). Continued activity in the apices of the cotyledons results in their apical growth
so that they increase in length. The procambial central care extends into the cotyledons. The external layer of the embryo is continuous and keeps pace with the growing embryo dividing anticlinally. In the surface layer of the lower region of the embryo there is further occurrence of periclinal divisions which extend to the flanks forming periphery of the root cap (Fig. 45 and Photomicro. 28).
4.4.4 Torpedo-shaped Embryo

The torpedo-shaped embryo shows dorsiventrally flattened cotyledons. A slight dome shaped plumular apex shows single tunica layer and a corpus. The central cylinder, cortex andthe pith are clear. The protodermal cells at the radicular apex show kappa type of divisions giving rise to a few files of cells to form the peripheral part of the root cap (Photomicro. 29).

## CHAPTER 5

APICAL ORGANISATION
5.1 Radicular Apex
5.2 Apex of Seedling Root
5.3 Embryonic Shoot Apex
5.4 Mature Shoot Apex
5.5 Plastochronic Changes

## 5. APICAL ORGANIZATION

### 5.1 RADICULAR APEX

The radicular apex consists of two parts namely: the root-promeristem and the root cap. The root cap shows two distinct zones during embryogeny: a. the periphery of the cap, b. the columella.

The root cap is initiated at the hypophysial the region of the embryo from tier ' 0 ' during (late globular stage of the embryo. Development of the cap flanks is initiated by the characteristic 'Kappe' divisions in the cells of the surface layer at the late globular stage of the embryo. The cells of flanks abut the point of attachment of the suspensor to the embryo. This coincides with the appearance of the central cell (Fig. 46 and Photomicro. 27). Initially the kappe divisions are restricted only to the surface layer closer to the central cell.

At the heart shaped stage of the embryo kappe divisions extend to the sub-surface layers. The central cell is still prominent. The kappe divisions in the surface layer are slightly away from the central cell (Fig. 47 and Photomicro. 28). At the torpedo stage of the embryo additional kappe divisions occur in he surface layer more distal to the suspensor end. The daughter colle further divide similarly to add more layers to the
developing periphery of the cap. Due to subsequent divisions both in the surface and sub-surface layers a continuous zone of the cells is produced extending across the suspensor end of the embryo and results in the demarcation of the root cap (Fig. 48). Thus the flanks of the root cap or peripheryake histogenetically related to the surface layer. At the torpedo stage of embryo successive periclinal divisions occur resulting in 3-6 rows of cells which contribute to the lateral root cap lying between the differentiated epidermis and columella (Fig. 48), the development of which will be described later.

At the hypophysial region of the globular stage of embryo, belon the central cell four linear rows of cells are prominent. These cells are the precursors of columella (Fig. 46 and Photomicro. 27). At the heart shaped stage of embryo the primary columella consists of regular longitudinal rows of cells having their inftials at $t$ he proximal end. The divisions are transverse to start with (Fig. 47). All the layers of the root cap abut laterally on the columella. The cells of the columella are rectangular. At the torpedo stage of the embryo further linear divisions are added to the cells of the columella. The height of the columella varies from 6 to 8 cells and 4 to 5 cells in width. The cells maintain long vertical files arranged in linear rows (Fig. 48). By this stage dermatogen becomes clear and forms a boundary between the root body and root cap delimiting the columella from the lateral
portions of the cap (Fig. 48), and continues to divide periclinally to add cells to the periphery of the cap. During the torpedo stage of embryo there is no direct relationship between the cortex, central cylinder and root cap.

The ' $n$ ' and 'o' tiers of the globular embryo are responsible for further development of root body. At the late globular stage of the embryo there is a large, deeply stained distinct cell with a prominent nucleus. This cell is hypophysial in origin and is termed as the 'central cell'. The central cell is surrounded by a group of five prominent cells (Fig. 46 and Photomicro. 27). These cells divide transversely and constitute the promeristem. The central cell remains prominent upto the heart shaped stage of embryo (Fig. 47).

The structural configuration at the torpedo stage of embryo can be distinguished into four discrete histogens with a common initiating zone for dermatogen and calyptrogen and discrete central cylinder, cortical and columella initials. The common initial zone is concerned with the formation of both the dermatogen and the peripheral region of the cap. By further 'kappe' divisions the initials give rise to three cells, which appear like ' $T$ ' in the beginning and 'Y' later. Two of these behave as initials and repeat the pattern. The cell rows of the peripheral region of the cap curve inwards as they approach the columella. These curved rows merge all around the columella (Fig. 48). The columella initials continue to divide transversely like that
in a rib meristem (Fig. 48). The cortical initials are 5-6 cells wide. The cells of flanks exhibit 'korper' type of arrangement. These divisions widen the cortex towards the flanks. The 'korper' divisions are repeated a few times in the cortex after which the cells divide mostly anticlinally (Fig. 48 and Photomicro. 30-31). The central cylinder initials are $4-6$ cells wide at the torpedo stage of embryo. These cells generally divide by 'korper' divisions at the periphery. As a result the cells in the centre are broader than those at the periphery.

### 5.2 APEX OF SEEDLING ROOT

With further development of the root viz, at the seedling stage, the periphery of the root cap proliferates by more divisions and the root cap undergoes certain changes. Oblique divisions in the outer cortical initials contribute cells to the root cap so that continuity is established between the outer cortex and root cap. This changes the closed configuration of root apex to open type (Photomicros. 32-33). There is no further increase in the width of the primary columella. As the cortical cell layers traverse down towards the cap, they change their course abruptly and run parallel to the vertical files of columella resulting in 'L' shaped files of cells (Photomicro. 33). This formation of L-shaped files of cells is also known as 'knee' formation. As a result of these changes the convergence of layers at the apex is lost. The primary columella maintains its width of 4-5 cells.

The cell rows of the cap abutting on the columella undergo transverse, oblique and vertical divisions, forming linear rows around the columella. These rows thus formed can be regarded as 'secondary columella'. A few cells are added towards the periphery of the cap also. These cells are conspicuous. It appears as though the entire cortical rows arise from these cells. Such a row of cells has been termed as 'pericolumnar cells' (Schopf, 1943). They are known as pericolumnar initials because they are conspicuous and have dense protoplasm and it appears as though from each cell a row of cortex arises. The pericolumnar cells establish continuity with the stelar region above. The whole structure gives an appearance of an 'inverted cup'. The base of the cup is formed by the stele and the sides by the pericolumnar cells and packed with the columella cells (Photomicro. 33).

At this stage that is after the root apex opens the meristematic activity in the root body region shifts from actual site of initials to the cells immediately surrounding the initials which later continue to maintain minimal constructional centre.

### 5.3 BMBRYONIC SHOOT APEX

The terminal cell 'ca' of the two celled proembryo is responsible for the future shoot apex in Hibiscus cannabinus. Further divisions in the terminal cell forms a broader region at the chalazal end of the globular stage
of embryo than that produced by 'cb' at the suspensor end. At the globular stage, the embryo can be divided into three distinct regions based on shape and cell size. The narrow region at the suspensor end with large cells gives rise to the radicle. The middle two rows with large cells forms the future hypocotyl. The region below it which is broader, where the cells inside divide periclinally and of surface layer divide anticlinally, gives rise to the shoot apex and cotyledons. Periclinal divisions occur in the surface layer at levels where the future cotyledons are to be formed (Fig. 43). The cells of the terminal region have uniform staining capacity.

During further development of the embryo the cells of the terminal region divide repeatedly. Divisions are more pronounced on either sides of the terminal region. These are the sites of cotyledon initiation while the region between the two sites is the future epicotyl apex (Fig. 44 and Photomicro. 27).

At the heart shaped stage of the embryo, when the cotyledons have attained some height the epicotyl apex is concave (Fig. 45 and Photomicro. 28). A few cells of the surface and sub-surface layers have deep staining capacity. The cells of the sub-surface layer are larger, more active and divide periclinally (Fig. 49) to raise the epicotyl apex.

At late torpedo stage the epicotyl apex slightly bulges out due to pronounced periclinal divisions in the sub-surface layer and anticlinal divisions in the surface layer. The cells of the surface layer due to pronounced anticlinal divisions become narrow and radially elongated (Fig. 50). The apical bulge has a single covering layer the 'tunica' and the corpus beneath, the cells of which divide in all planes. The tunica and corpus have the same staining capacity and at $t$ his torpedo stage of embryo there is no cyto-histological zonation in the epicotyl apex (Fig. 50 and Photomicro. 34).

### 5.4 MATURE SHOOT APEX

The surface of the vegetative shoot apex is a low dome and shows a well marked tunica-corpus organisation. The tunica is composed of one discrete layer with a tendency for stratification of another layer. The cells of the corpus undergo divisions in all planes. The corpus is distinguishable into three zones, on the basis of its cell size and plane of division. Just below the tunica layer the cells are larger angular and show division in all planes. This group of cells constitutes the central mother cell zone. Below this is the pith rib meristem, consisting of cells that are in distinct files. The cells divide transversely and the pith has its origin in the pith rib meristem. The pith rib meristem has a surrounding flanking meristem. The flanking meristem is composed of fairly distinct files of small, dark staining very slightly
vacuolate cells. The cells undergo periclinal divisions first and then by anticlinal divisions. This flanking zone usually presents a definite stratified appearance due to predominance of anticlinal divisions. The cortex and the vascular tissue differentiate from the flanking meristem (FIg. 51 and Photomicro. 35).

### 5.5 PLASTOCHRONIC CHANGES

The shape of the shoot apex shows periodic changes during plastochronic cycles. The shoot apex at the maximal phase of the plastochron shows cytohistological zonation. Though the apical dome is low in Hibiscus cannabinus yet it is prominent. The single tunica layer has radially elongated cells. The flanks of tunica layer are more active than the apical region with the cells more deeply stained. These cells undergo anticlinal divisions. The cells of the central mother cell zone have larger nuclei than those of the surrounding cells. These cells undergo periclinal and oblique divisions (Fig. 51 and Photomicro. 35). The first periclinal division in the layer below the tunica layer, marks the formation of a leaf primordium. The initiation of leaf primordia also involves the deeper layers of flanking $z$ one. There is an increase in the anticlinal divisions in the tunica layer and with the appearance of the leaf primordia the shoot apex enters the minimal phase of the plastochron (Fig. 52 and Photomicro. 36).

The plastochronic cycle of the shoot apex can also be correlated with the size of the youngest leaf primordium. At the minimal phase of the plastochron the shoot apex is flat and has a width of $84.0 \mu$. This stage of the plastochron represents the maximum growth of the leaves before the initiation of the next primordium, attaining a height of 145-150 $\mu$. The tunica layer show more anticlinal divisions. The cytohistological zonation is less distinct (Fig. 52 and Photomicro. 36). The extent of the central mother cell zone and the flanking zone become reduced and become situated in close vicinity of the apex. As the apex passes from the minimal phase, the distal cells of the central mother cell zone becomes more active. Pith rib meristem and the corpus flanks become active. All these activities lead to the elevation of the apical dome. The cytohistological zonation becomes more distinct as the apex enters the mid plastochron. At this stage the width of the apical dome is 135-141 $\mu$ and height of the dome gradually increases to $20-24 \mu$ and the leaf primordia attains a height of $27 \mu$. The height of the dome varies as the apex enters into the maximal phase. At the maximal phase of the plastochron the shoot apex attains the maximum height of $36.0 \mu$ on the average and attains the maximum width of 147-153 $\mu$. The zonation of the apex becomes distinct and the apex shows tunica corpus organisation. The initiation of the leaf primordium takes place at the maximal phase.

CHAPTER 6
SEEDLING VASCULATURE
6.1 4th Day seedling
6.2 8th Day Seeding
6.3 12th Day seeding
6.4 20th Day seedling

## 6. SEEDLING VASCULATURE

### 6.1 4TH DAY SEEDLING

Serial transverse sections of the young seedling on the 4 th day shows a tetrarch stele with a solid core of xylem (Fig. 53). In the lower region of the hypocotyl the root shows four separate groups of xylem and eight strands of phloem (Fig. 54). Xylem groups 'a' and 'b' split into two each and gradually diverge on either sides still maintaining contact with the smallest peri pheral elements of xylem which later show signs of degeneration. simultaneously the phloem groups also split tangentially so that the number doubles in between the groups of $x y l e m$ (Fig. 55).

Each half of the split xylem groups ('a,a, and ' $b, b_{1}$ ') move apart, leaving behind a peripheral element of xylem 'x' (Figs. 56,57 and Photomicro. 37). A little below the cotyledonary node the xylem groups ' $c$ ' and ' $d$ ' also split in the same manner as that of 'a' and ' $b$ '. By this time the peripheral xylem is almost obliterated. While ' $a, a_{1}$ ' and ' $b, b_{1}$ ' move apart, ' $c, c_{1}$ ' and ' $d, d_{1}$ ' run close to each other (Fig. 57) and go together as traces to the cotyledons.

Immediately below the cotyledonary node the eight xylem groups become arranged into two collateral arcs ( $\mathrm{acc} \mathrm{c}_{1} \mathrm{~b}$ ) and ( $\mathrm{a}_{1} \mathrm{dd}_{1} \mathrm{~b}_{1}$ ) (Fig. 58 and Photomicro. 38). These
two arcs depart on either sides as traces to cotyledons. Here the xylem is endarch. The cotyledonary node is thus unilacunar with a single trace. The single partially differentiated $x y l e m$ ' $x$ ' between the cotyledonary traces extend into the epicotyl and goes as a trace to the first foliage leaf (Figs. 59,60 and Photomicro. 39).

The single trace that enters the cotyledon runs as such through the petiole upto the base of the lamina in the form of an arc. The cotyledonary strand divides into three secondary veins at the base of the lamina. The two on either side diverge to their respective sides into the lamina of the cotyledon and the central one runs upto the tip of the lamina. The secondaries divide profusely forming islets each having a number of veinlets ending freely in the mesophyll cells of the cotyledons.

### 6.2 8TH DAY SEEDLING

Just below the cotyledonary node, the strands lying on the cotyledonary plane go as cotyledonary traces, and those on the intercotyledonary plane go as traces to the first two leaves (Fig. 61 and Photomicro. 40). The first leaf is supplied by a single trace so that it is a unilacunar single trace node and the subsequent nodes show trilacunar condition (Fig. 62).

### 6.3 12TH DAY SESDLING

The number of vascular bundles at the cotyledonary node increases. The nature of cotyledonary supply is
similar to the 8 th day seedling. As the cotyledons separate from the axis the vasculature comes to lie in a ring. From this ring traces supply the leaves. First foliar node is unilacunar, the second and subsequent are trilacunar (Fig. 63).

### 6.4 20TH DAY SEEDLING

In older seedling the hypocotyl shows a ring of discrete endarch strands of different sizes at the base of the cotyledonary node (Fig. 64). The two large cotyledonary traces separate out from this ring from the opposite regions. Of the six strands left behind, two large strands $\left(M_{1} M_{2}\right)$ lie in the intercotyledonary plane and four small strands $L_{1} L_{2}$ and $L_{3} L_{4}$ lie separately in opposing groups of the cotyledonary plane (Fig. 65). The strands $M_{1}$ and $M_{2}$ go as medians to the first two leaves and $L_{1} L_{3}$ depart as laterals to one leaf and $L_{2} L_{4}$ as laterals to the other leaf. The foliar node is trilacunar (Figs. 66,67). The remaining partly differentiated vasculature extend higher up in the shoot and differentiate below the new leaves (Fig. 68). The first foliar node in the 20th day old seediling is also unilacunar and the subsequent nodes are triiacunar.

## CHAPTER 7

## PHYLLOTAXY AND NODAL VASCULATURE

## 7. 1 Phyllotaxy

7. 2 Primary Vasculature of the Axis
8. 3 Nodal Vasculature
9. PHYLLOTAXY AND NODAL VASCULATURE

### 7.1 PHYLLOTAXY

The arrangement of leaves on the shoot has been studied from a series of transverse sections of the shoot apex passing down through several nodes. Each node bears a single leaf. The leaves are stipulate.

The arrangement of leaves on the axis is spiral. The genetic spiral passes through the leaves in their numerical order. The spiral winds round the axis in an anticlockwise direction towards the shoot apex taking two complete rounds of the axis to connect two superimposed leaves. Thus five contact parastichies are observed connecting the leaves 0 and 5, 1 and 6,2 and 7,3 and 8 , 4 and 9 and so on. The phyllotaxis therefore, can be expressed as $2 / 5$. A diagrammatic representation of phyllotaxis is given in Fig. 70. Any two superimposed leaves are five plastochrons apart. The sixth leaf is five plastochrons older than the first leaf. Leaf seventh is five plastochrons older than leaf second and so on.

In addition to the contact parastichies which wind round in an anticlockwise direction, another set of contact parastichies is present connecting the leaves in the clockwise direction towards the apex. There are three contact parastichies in this set connecting the leaves $0-3-6-9$, 1-4-7-10, 2-5-8-11 respectively.

Thus in all there are two sets of contact parastichies, one winding around the axis clockwise and the other anticlockwise. The two sets thus can be represented as $3+5$ contact parastichies. The angular divergence between two successive leaves is approximately $137^{\circ}$.

### 7.2 PRIMARY VASCULATURE OF AXIS

The primary vasculature of the axis has been studied in serial transections through the shoot apex followed downwards through several nodes to determine the interrelationship of traces. There are five distinct sympodia which are recognisable at lower nodes where vascular differentiation is complete. They send off vascular traces at the nodes to supply the leaves. Both laterals and medians leave the sympodia at regular levels, so that each leaf is supplied by a median, an anadromic lateral and a katadromic lateral.

The median of any leaf taken on the axis, is just below or above the median of the sixth leaf. Similarly the medians of the subsequent four leaves in order are related to the next fifth leaf respectively. Each median is related to the laterals of the two leaves below. Of the two laterals one is two nodes below and is known as the katadromic and the other three nodes below the anadromic. Thus the vertical course of two laterals is different. The katadromic lateral of any leaf is related to the anadromic lateral from the preceding leaf.

The relationship of various traces from first to ninth leaf is represented in a reconstructed drawing (Fig. 69). The relationship of various traces is as follows:

| $\mathrm{M}_{12}$ | $\mathrm{M}_{11}$ | $\mathrm{M}_{10}$ | $\mathrm{M}_{9}$ | $\mathrm{M}_{8}$ |
| :--- | :--- | :--- | :--- | :--- |
| $\mathrm{KI}_{10}$ | $\mathrm{KI}_{9}$ | $\mathrm{Kl}_{8}$ | $\mathrm{KI}_{7}$ | $\mathrm{KI}_{6}$ |
| $\mathrm{Al}_{9}$ | $\mathrm{Al}_{8}$ | $\mathrm{Al}_{7}$ | $\mathrm{Al}_{6}$ | $\mathrm{Al}_{5}$ |
| $\mathrm{M}_{7}$ | $\mathrm{M}_{6}$ | $\mathrm{M}_{5}$ | $\mathrm{M}_{4}$ | $\mathrm{M}_{3}$ |
| $\mathrm{KI}_{5}$ | $\mathrm{KI}_{4}$ | $\mathrm{KI}_{3}$ | $\mathrm{KI}_{2}$ | $\mathrm{~K} I_{1}$ |
| $\mathrm{Al}_{4}$ | $\mathrm{Al}_{3}$ | $\mathrm{Al}_{2}$ | $\mathrm{Al} I_{1}$ |  |

From the above it is seen that each median is related to the laterals of the two leaves below, before its superimposed leaf comes into position. Of the two laterals, the first ${ }^{\text {is }}$ two nodes below the median and the second is related to the lateral of the next leaf. The former is katadromic and the latter is anadromic so that each median has its connection with second and third node below. As in Fig. 69, median $M_{9}$ is related to the katadromic lateral $K I_{7}$ and anadromic lateral $\mathrm{Al}_{6}$. Similarly, median $\mathrm{M}_{8}$ is related to the two laterals, the katadromic lateral $\mathrm{Kl}_{6}$ and anadromic lateral $\mathrm{Al}_{5}$ and so on.

## 7. 3 NODAL VASCULATURE

A distinct nodal vasculature in a mature shoot could be seen fully differentiated only after the third node. From the axis a single, prominent and large strand, the median, separates and moves out. Following it, move
out two smaller lateral strands on either side, leaving gaps in the procambial cylinder. The node is thus trilacunar (Figs. 71-72).

As the traces move out to the cortex, the two laterals divide and one branch on either side supply the respective stipules. The median strand of the leaf divides into three $\left(M_{1}, M_{2}\right.$ and $\left.M_{3}\right)$. Of these the middle one $M_{2}$ remains as such whereas $M_{1}$ and $M_{3}$ divide once to give $M_{1.1}$ and $M_{3.1}$ forming five strands (Figs. 73-75). By this stage the two lateral strands ( $L_{1}$ and $L_{2}$ ) undergo a series of changes. Each divide into two traces $\left(L_{1.1}\right.$ and $L_{1,2}$; $L_{2.1}$ and $\left.L_{2,2}\right)$. Those away from the median $\left(L_{1.1}\right.$ and $\left.L_{2,1}\right)$ go to the stipules on either side. The remaining two strands $L_{1.2}$ and $L_{2.2}$ move towards the median (Fig. 75). These strands undergo further divisions so that $L_{1.2}$ and $L_{2.2}$ gi ve a trace each towerds the adaxial side forming a complete ring. As they enter the leaf these traces change their position of protoxylem so that it becomes abaxial with reference to the stem and adaxial with reference to the petiole (Fig. 75).

Thus in all nine strands are formed to complete the circle that prolongs into the petiole. Both branching and fusion of branches of vascular bundles takes place within a very limited segment of the leaf base. At the distal end of the petiole nine isolated collateral bundles are seen in a circle (Fig. 76).

CHAPTER 8

## LEAF HI STOLOGY, VENATION PATTERN AND TRICHOMES

8. 1 Vasculature of Petiole
8.2 Leaf Histology
8.3 Venation Pattern
8.4 Trichomes
9. LEAF HI STOLOGY, VENATION PATTERN AND TRICHOMES

## 8. 1 VASCULATURE OF PETIOLE

The petiole is long and cylindrical. Numerous trichomes are present on the petiole. The transverse section through the distal end of the petiole exhibits isolated collateral bundles arranged in a circle. The central pith of parenchymatous cells is loosely arranged with druses present in many cells.

Gradual variati on in the number of bundles is observed while tracing from the axis towards the leaf blade. At the lower region of the petiole viz. at the point of attachment to the axis the petiole shows nine endarch bundles arranged in a circle. Bundles vary in size with more or less six large and three small bundles (Fig. 77 and Photomicro. 41). In the middle region of the petiole the number of bundles decreases to five as a result of fusion of the smaller bundles with the adjacent larger bundles (Fig. 78 and Photomicro. 42). Towards the upper region of the petiole the five bundles fuse to form a closed ring (Fig. 79). It runs upto the base of the leaf where it divides to give rise to $3-5$ secondary veins.

### 8.2 LEAF HISTOLOGY

Both the upper and lower epidermal layers bear stomata at the same level as that of the epidermal cells. Mucilage cells are fairly numerous in the epidermis. There
is a single layer of adaxial palisade on the upper surface of the leaf which continues in the midrib region leaving only a small strip which is occupied by collenchymatous cells. The spongy layer has 5 to 6 rows of loosely arranged cells (Fig. 80 and Photomicro. 45).

The mid vein is prominent and is more or less semicircular in transectional view. Smaller veins are embedded in the mesophy11. Stomata are ranunculaceous or anomocytic type. The guard cells are surrounded by 2 to 4 cells (Fig. 81 and Photomicro. 46). Occasionally druses occur in the lower epidermis and in the mesophyll tissue.

### 8.3 VENATION PATTERN

Venation pattern is craspedodromous type. Single primary vein runs through the petiole to the leaf lamina. The primary vein branches into three secondary veins which may later increase in number from 3 to 5 depending on the number of lobes present in the leaf. Of the three, the central vein runs to the median lobe and terminates at the tip. The lateral secondary veins divide at the leaf base Itself and run to the lateral lobes. Secondary veins give off strong tertiaries on either sides. The tertiaries with the quarternaries have a direct course to the marginal teeth (Photomicro. 47).

Quarternaries and their branches constitute the areoles. Areoles are polygonal and rectangular in shape. Three to six free vein ends in an areole are common. Free vein ends are simple, sometimes forked.

Some of the free vein ends appear to be jointed (Photomi cro. 48).

On the abaxial side of the midrib or the central vein of leaf an extra-floral nectary is situated. In a transverse section the extra floral nectary shows a group of secretory trichomes situated in a crypt surrounded by three to four rows of compressed cells. The glandular trichomes in the nectary are of two types. The uniseriate of 5-6 layered cells are restricted to the lower region near about the opening of the crypt. The biseriate trichomes occur along the rest of the concavity which has a stalk of 2 cells and biseriate of six cells. All the trichomes are of the same height. The crypt opens on the abaxial side of the leaf (Photomicros. 43-44).

### 8.4 TRI CHOMES

Two main groups of trichomes: (i) the glandular, and (ii) the nonglandular were observed on the vegetative and floral organs. On the basis of their structure eight types of trichomes have been recognized. Four types of trichomes are glandular and the other four nonglandular.

### 8.4.1 Glandular Trichomes

1. Uniseriate glandular trichame: The trichame initial in each type becomes recognizable because of its denser staining and large nucleus. The cell becomes papillose and vacuolated. It divides into a basal cell
and an apical cell by a periclinal wall. The basal cell does not divide further but occasionally an anticlinal division forms a two celled foot of the trichome. The apical cell divides into two. The lower stalk cell repeatedly divides periclinally to give rise to two to four celled stalk and the head cell alsc divides perfclinally to form a two to six celled uniseriate head. Anticlinal divisions in one or more cells form the head biseriate at places or as a whole (Figs. 98-105).
2. Fusiform glandular trichome: The trichome Initial divides periclinally to give rise to a basal cell that does not divide further, and an apical cell. The apical cell divides periclinally. The lower cell forms a stalk of two to three cells by periclinal divisions. The head cell results into a multicellular fusiform head by periclinal and anticlinal divisions (Figs, 110-113).
3. Capitate glandular trichome: The stalk cell divides periclinally to form two to many celled stalk. The head cell divides periclinally, anticlinally, rarely obliquely resulting into a multicellular capitate head. The basal cell does not divide further and forms a simple foot (Figs. 114,115).
4. Cylindrical glandular trichome: The trichome initial divides periclinally forming a basal cell which is the foot of the trichome. The apical cell divides to give a stalk cell below and head cell above. The stalk cell
divides periclinally to form two to three celled stalk. Repeated periclinal divisions in the head cell give rise to a multicellular head in a uniseriate cylindrical fashion (Figs. 105-109).

### 8.4.2 Nonglandular Trichomes

1. Simple unicellular trichome: This trichome originates from a papillose epidermal cell. The initial without dividing protrudes beyond the epidermal cells and undergoes considerable elongation till trichome is long and thin and tapers at the tip. The walls are generally thick. The cytoplasm and the nucleus degenerate during elongation (Figs. 82-84).
2. Simple bicellular trichane: The papillose trichome initial divides periclinally so as to form a basal cell and a pointed apical cell. The apical cell becomes much elongated axially, tapering above, pointed at the end and cytoplasmic contents absent at maturity. In some cases the basal cell undergoes longitudinal division to give rise to a bicelled foot (Figs. 85-87).
3. Capitate uniseriate filiform trichomes: The trichame initial divides periclinally into a basal cell and an apical cell. The latter divides further by a periclinal division into the lower and terminal cell. The lower cell divides transversely resulting into a foureight celled body. The terminal cell does not divide
having a sub-capitate appearance. These cells are broader in length (F1gs. 88-93).
4. Tufted trichomes: The epidermal cell enlarges, divides anticlinally resulting into two thick walled cells projecting above the epidermis. Each of these cells grow like a simple unicellular trichome. The trichomes diverge in various directions. Anticlinal divisions may exceed from two to many (Figs. 94-97).

All the eight types of trichomes are initiated from a single epidermal cell. When the trichome has been formed the neighbouring epidermal cells and the hypodermal cells lying just below the trichome become meristematic (Figs. 87, 97). This gives rise to the base of the trichome. In same cases the hypodermal cells may not take part in the formation of the base. By this meristematic activity the whole trichome is lifted up. Such a base has been noticed in the long tapering trichanes viz., 1. the simple unicellular trichome, 2. the bicellular trichome, and 3. the tufted trichome.

CHAPTER 9

FLORAL VASCULATURE

## 9. FLORAL VASCULATURE

Flowers are solitary, axillary, segments of epicalyx are 7-10 free, linear, shorter than calyx and persistent. Calyx are 5-lobed, adnate below and free above. Petals - 5 and free, staminal column present, anthers are arranged throughout the column, anthers monothecous. Ovary is pentacarpellary, ovules two to four in each chamber with axile placentation.

The pedicel has more or less a circular outline and contains a ring of 4 big and 4 small strands (Fig. 116 and Photomicro. 49). Three of the 4 large strands leave the ring (Fig. 117). As these three get tangentially stretched, the fourth large strand gives of $f$ a trace with the result the inner ring now comprises five small strands (Fig. 118 and Photomicro. 50). Now the four traces stretched tangentially join on either side to form a continuous ring of vascular tissue (Figs. 119, 120 and Photomicro. 51). For the convenience of description the outer vascular ring is termed as 'Bt Sm Ct' and the inner ring of five strands as 'SlPStCt' (Figs. 119, 120). From the 'BtsmCt' ring of vascular tissue the medians of the bracteoles 'Btm' which are the first to be given off move towards the bracteoles. Additional traces moving out come to lie on either side of the medians at a point where the bracteoles join. Each trace


#### Abstract

divides to form two strands, which enter the two adjacent bracteoles (Figs. 120, 121 and Photomicro. 52). Thus three strands traverse each bracteoles one as median and two as laterals 'Btl'. Occasionally five strands are seen in a bracteole because of its large size. Here the median has two laterals on either side (Fig. 122).


From the continuous ring of 'BtsmCt' vascular tissue, 5 traces are given off to the inner side which alternates the inner ring of five strands termed as 'SmCt'. The departure of these five traces is uneven. The first three strands formed (Fig. 119) take the position where the first three big strands were located after leaving the ring of eight strands as in Fig. 117. Later the other two are added so that in all five strands are formed (Fig. 120).

By this time the inner ring of five small strands 'SlPStCt' become lobed and 'U' shaped with the concavity on the inner side (Fig. 121 and Photomicro. 52). From the edges of the two inwardly projecting arms of the ' $U$ ' shaped, traces are pinched off so as to form ten traces at the centre (Figs. 121,122). At the same time the alternating five strands 'SmCt' give out traces that join the vascular cylinder in the centre formed by the 10 centrally placed traces so that it looks like a ring with five arms radiating (Figs. 122,123). The central ring of vascular tissue divides equally into five arcs
'CV' which are inversely oriented (Fig. 124 and Photomicro. 53). The outer 10 alternating strands 'smCt' and 'SlPStCt' now in a ring give out traces that join to form a hollow cylinder (Figs. 124, 125).

Higher up and at the base of the ovary the five dorsal carpellary traces 'CD' are given off on the petal radii from the hollow cylinder. This also gives off five prominent marginals 'Cul' on either side of the dorsals. The loculi are originally antipetalous in position but this alignment is disturbed higher up (Figs. 126,127). The centrally placed five arcs divide to form 10 ventrals 'CV'. Two ventrals go as traces to each carpel (Figs. 125,126).

After the organization of the carpellary vasculature, the 10 peripheral strands of the floral tube, alternately behave as 'Sm' and 'SlPSt' (Fig. 125). The 5 'Sm' strands directly go as medians of the sepals (Figs. 126,127). The alternating five strands 'SlPSt' give out traces on either sides of the sepal median as laterals 'Sl' that are confluent. Thus the calyx at the base is supplied by 10 traces (Figs. 127, 128 and Photomicro. 54). The laterals at higher level divide so that each sepal has a median and two laterals.

The whorl of five traces 'PSt' after the separation of sepal laterals, supply as conjoint petalstamen traces. At first the dual nature of these traces
is not evident but while moving to the periphery of the receptacular cortex each trace gets tangentially stretched and becomes three lobed (Fig. 128). As the lobing becomes clearer the middle lobe separates and enters the petal 'Pt' and resolves into a median and laterals (Fig. 129 and Photomicros. 55,56).

The laterals, higher up, divide twice so that each petal is supplied with five strands (Fig. 130). Still higher up the strands of the petals divide several times to form a more or less complete band of vascular tissue (Fig. 131).

At the level of separation of petals from the staminal tube five pairs of traces are left behind inner to the bases of petals (Fig. 130 and Photomicro. 55). Higher up they get equally spaced. The ten staminal traces 'St' divide to form staminal strands at different levels. All the staminal traces get consumed in the formation of staminal strands (Fig. 131 and Photomicro.56).
10.1 External Morphology
10.2 Structure of $Y$ oung Twig
10.3 Structure of Mature Bark
10.4 Expansion Tissues
10.5 Lenticels
10.6 Periderm

## 10. BARK ANATOMY

## 10. 1 EXTERNAL MORPHOLOGY

The tender bark is green and smooth with short glandular and long non-glandular hairs and is mucilagenous. The stem has stomata on the epidermis. As development and differentiation of tissues proceed, the colour of the bark becomes brownish at the lower and thicker part of the stem and has numerous tiny black openings, the lenticels. Hairs are absent on the older bark.
10.2 STRUCTURE OF YOUNG TWIG

The pith is wide, parenchymatous and contains mucilage cells. The pith cells are larger in the centre and decrease in size towards the xylem. The phloem is delimited from the cortex by protophloic fibres which form blocks alternating with blocks of parenchyma. In the thickest internode of the young stem three to five tangential layers of secondary phloem fibres were present. The rays are parenchymatous. Next follows layers of parenchyma with numerous mucilage cells which get torn in sections and crystals in the form of druses are abundant in the parenchyma. Next to parenchyma follows two to three layers of collenchyma. The single layered epidermal cells are rectangular. Stomata are present in the epidermis. One to three layers of cortex beneath the epidermis contain chloroplasts (Photomicro. 57).

According to Esau (1965) all tissues outside the vascular cambium comprise bark. This concept is followed in the present study.

### 10.3 STRUCTURE OF MATURE BARK

### 10.3.1 Secondary Phloem

secondary phloem is characterised by the presence of phloem blocks which alternate with the phloem rays. The phloem blocks have strands of fibres arranged tangentially and radially forming a wedge shape. These phloem blocks become narrow towards the periphery of the bark due to ray expansion and phloem proliferation. One to five or six tangential layers of secondary phloem fibres were present in the thickest internode of the stem in each wedge. The fibres alternate radially with soft phloem regions containing sieve tubes, companion cells and parenchymatous cells. The fibres and the sof phloem constitute the axial system. In a transverse section fibre bands are variable in shape. A detailed study of fibres will be dealt in the next chapter.
10.3.2 Phloem Parenchyma

The phloem parenchyma occurs in narrow bands on both sides of the fibre bands. The cells are thin walled and angular in appearance and are compactly arranged. Many cells contain crystals in the form of druses (Fig. 144, and Photomicros. 60, 61).

### 10.3.3 Sieve Tubes

Sieve tubes in a transverse section are angular, small, embedded in the parenchyma singly or in an aggregate form. In longitudinal section they are long, tubular and arranged in storied fashion with their end walls inclined. Transverse end walls are also present. The inclined walls are perforated to form sieve plates. Sieve plates are compound with a row of sieve areas (Figs.158,159 and Photomicros. 58,59). Number of sieve areas is variable from plate to plate. Occasionally sieve plates are steeply oblique. The angle of inclination of the sieve plates to the horizontal axis of the sieve tubes varies from $20^{\circ}$ to $90^{\circ}$ with an average of $37^{\circ}$. The average length of the sieve tube is $410.34 \mu$ the range being 225 to $645 \mu$. Maximum number of sieve tubes have a range of 400 to $500 \mu$ in length. $2 \%$ has a length of 200 to $250 \mu$. The sieve tubes are 45 to 107.5 microns in width and their average being $71.62 \mu$. Percentage of sieve tubes in different frequency classes of length and width is shown in Fig. 135. Each sieve tube is associated with one or two, triangular or rectangular shaped compan\$ion cells in transverse section. However, the number of companion cells associated along the length is either one or three to five. If one companion cell is associated with the sieve tube it runs either the whole length or a part of it.
10.3.4 Rays

The phloem rays are of two kinds. 1. In a transverse section the rays are many cells wide and become still wider towards the periphery, 2. while in tangential longitudinal section they are uni-, bi- and multiseriate. The multiseriate are from tri- to many seriate. The ray cells are rectangular to hexagonal in shape and heterogenous.

The individual cells of the rays extend longitudinally along the radius of the axis. The height of the rays ranges from $700 \mu$ to $3900 \mu$, the average height being $1790.3 \mu$. The average ray width is $53.3 \mu$ and the range is $15 \mu$ to $150 \mu$. Distribution of rays into different classes of width and height is presented in Figs. 132, 133 respectively.

Uniseriate rays have narrow conical or dome shaped end cells (Fig. 146). Height of the rays ranges from $700 \mu$ to $1170 \mu$. The average height is $860 \mu$. Width of uniseriate rays ranges from $15 \mu$ to $45 \mu$, the average being $20.5 \mu$. Uniseriate rays are very few in number in comparison to bi- and multiseriate rays. Only $19.38 \%$ of rays belong to uniseriate type.

Biseriate rays usually have uniseriate equal ends. In a few, one is short while the other is long (Fig. 147). some of the rays show one end uniseriate and the other biseriate (Fig. 149) and some the ve alternate biseriate and
uniseriate portions (Fig. 148). Height ranges from 900 to $2745 \mu$ and width from 30 to 75 microns. Average height and width is 1781.10 and 51.15 microns respectively. They are the maximum in number, about $68 \%$ in all.

Multiseriate rays have equal uniseriate ends
(Fig. 150). Some of the multiseriate rays have either one or both the ends biseriate. In some rays uni- and biseriate portions are included. Height ranges from $1650 \mu$ to $3900 \mu$ and width 60 to 150 microns . Average height and width is 2730.75 and 84.40 microns respectively. $21.84 \%$ oif the rays belong to multiseriate type.

Number of rays per mm ranges 10 to 18 with an average of 13. Percentage reading in different frequency classes of ray number per mm are shown in Fig . 134.

### 10.4 EXPANSION TISSUES

As a result of secondary growth and production of phloem tissues, tangential stretching of the cortical tissues occur. More tissues are produced by the cortical parenchyma and rays.

During the early secondary growth in the stem expansion in the cortex takes place. The cells of the cortex are stretched tangentially followed by anticlinal divisions. As a result the circumference of cortex increases to some extent and keeps pace with the increase in diameter.

Almost all the rays of the phloem undergo expansion. The ray cells show tangential stretching which gradually increases towards the periphery. The tangential stretching is followed by anticlinal divisions in the ray cells. Moreover, in the expanding stem, the outer parts of the rays undergo dilation constituting tangential enlargement and radial divisions of cells so that wedges of ray tissue narrow near the cambium and wide near the cortex is formed (Fig. 142 and Photomicros. 64, 66).

### 10.5 LENTICELS

Lenticels are abundant in the older regions of the stem and are formed before the periderm is initiated at the early secondary growth. Their number varies from one to eight per square mm . The first formed lenticel arise beneath the stomata. Later ones arise after the periderm formation and are irregularly scattered over the entire surface of the bark (Fig. 145 and Photomicro. 66).

The development of lenticel begins with the irregular divisions of the parenchyma cells above the substomatal chamber. The division is restricted to the subhypodermal layers. Periclinal divisions occur in this layer and a lenticel phellogen is formed. By the repeated divisions, the filling cells of the lenticel increases in number, it ruptures the epidermis and protrudes above the surface (Fig. 145). The size of the lenticel is variable depending on the stage of development. The aperture may be as small as $90 \mu$ in diameter to about $375 \mu$.

The phellogen is superficial and the initial activity is in localized strips (Fig. 145). It is initiated by periclinal division in the epidermal cells resulting in the formation of a cork cell and a phellogen cell. The inner phellogen cell being meristematic, divides repeatedly increasing the number of cork cells (Fig. 145). The localized periderm results into lenticels. The divisions gradually become continuous around the stem circumference (Photomicro. 66). The cells of the cork are rectangular and arranged in regular radial seriations of 8 to 12 cells in height. However, later in older regions they get compressed and become tangentially flattened. The phelloderm is not very deep.

# CHAPTER 

BAST FIBRES
11.1 Origin and Development of Fibres
11.2 Development of Secondary Fibres
11.3 Cell Wall of Fibre
11.4 Fibre Characteristics
11.5 Fibre Measurements
11.6 Retting
11.7 Chemical Analysis
11.8 Physical Analysis

Bast fibres are derived from that portion of the dicotyledonous plants, lying between the outer bark or epidermis and the woody central cylinder. The bast fibres are also called "soft fibres" (Mathews, 1948).

### 11.1 ORIGIN AND DEVELOPMENT OF FIBRES

In an eight day old seedling fibres occur in the protophloem forming a ring (as seen in cross-section) which consists of several blocks of fibres that are tangentially flattened (Figs. 138, 139 and Photomicro. 57). These protophloic fibres originate from the procambial cells below the shoot apex in the elongating internode. They are discernible only after the sieve tubes and companion cells become obliterated. They become several times longer than the neighbouring cells from which they are distinguishable by the appearance of vacuoles in cytoplasm. This process of fibre differentiation advances inwards and the entire protophloem region is converted into a solid patch of fibres.

The fibre length results by elongation in the meristematic region at the stem tip. There is also a gradual increase in the diameter of the fibre during the Iife of the plant. This unusual elongation of fibres results in forcing the ends of the cells past each other
and between the fibres below and above, so that the apices elongate and intrude between the associated cells. As a result of this intrusive growth, the fibres increase in number in each fibre block and their ends form different shapes, viz., pointed, serrated, forked and rounded (Figs. 154-157, Photomicro. 62). The fibre cells are arranged in the form of bundles of varying number of ultimate fibres (Fig. 143). Such a development of the bast fibres forms a network, in which the individual strands have no identity. This is known as 'meshiness of fibres' which is loose nearer the apex and compact towards the basal region. Similarly it is compact near the cambiun and loose towards the periphery of the stem.
11.2 DEVELOPMENT OF SECONDARY FIBRES

In a cross section the cells of the cambial $z$ one are arranged in radial rows (Photomicro. 65). The cambial initials are basically of two types:
(a) Ray initials which give rise to the rays.
(b) Fusiform initials which by repeated divisions give rise to fibre blocks interposed by soft tissue on the same radius.

The fibre blocks alternate with phloem rays (Photomicros. 60,61 ). The nature of sieve tubes and phloem rays is discussed separately under the bark. In seedings or
younger regions of the stem, fibres occur initially in the form of a ring as seen in cross-section which consists of several bundles of fibres that are tangentially flattened (Fig. 138, Photomicro. 57). In older or basal regions of stem the number of layers increases and the phloem is delimited from the cortex by protophloic fibres. The rays undergo dilation growth combining tangential enlargement and anticlinal divisions of cells (Photomicro. 63). As a result of this growth, the phloem in transections assumes a pattern of alternating wedges of fibres and rays, the former being wide near the cambium and narrow near the cortex, and the latter narrow near the cambium and wide near the cortex (Fig. 142, Photomicros. 64, 65).

### 11.3 CELL WALL OF FIBRE

In Hibiscus cannabinus the walls of young fibre cells are thin and delicate. As the fibre elongetes the secondary wall formation commences. When fully developed, tertiary walls are laid down and the fibres become thick, thickness varying in different blocks (Fig. 141). In a transverse section, the outermost protophloic fibres show tertiary wall formation and nearer to the cambium the fibres either have the primary wall or a secondary wall Indicating that the maturation of fibres commences in the innermost fibre bundles (Figs. 140,141, Photomicro. 51). In some of the sections the secondary wall of the fibres
appear detached from the primary wall and it gets infolded. On treating the retted fibres with swelling reagent ( $1 \%$ iodine solution and by carefully introducing $50 \% \mathrm{H}_{2} \mathrm{SO}_{4}$ on the slide) the fibres did not swell but showed the thickness of fibre varying at different levels of its length. Young macerated fibres showed even thickness of wall throughout its length whereas mature fibres showed at their ends, undifferentiated portions of hyaline membranes and the middle regions showed thickened walls with a narrow Iumen (Figs. 151-153, Photomicro. 62).

Cross-section and lumen measurements of fibres were made at three different stages of flowering. Sampling was at random and the measurements are the average of 100 fibres. A graphical representation of the lumen and crosssection distribution is given in Figures 160,161 respectively and summarized readings are as below:

Table 1: Pre-flowering stage (75-90 days)

Cross Section:

| Average | $17.0 \mu$ |
| :--- | :--- |
| Maximum | $21.0 \mu$ |
| Minimum | $12.0 \mu$ |

Lumen:

| Average | $9.0 \mu$ |
| :--- | ---: |
| Maximum | $12.0 \mu$ |
| Minimum | $6.0 \mu$ |

Table 2: Flowering stage (90-105 days)

Cross Section:

| Average | $16.8 \mu$ |
| :--- | :--- |
| Maximum | $21.0 \mu$ |
| Minimum | $12.0 \mu$ |

Lumen:

| Average | $7.3 \mu$ |
| :--- | :--- |
| Maximum | $9.0 \mu$ |
| Minimum | $3.0 \mu$ |

Table 3: Post-flowering stage (105-120 days)

Cross Section:

| Average | $16.6 \mu$ |
| :--- | :--- |
| Maximum | $21.0 \mu$ |
| Minimum | $15.0 \mu$ |

Lumen:

| Average | $7.0 \mu$ |
| :--- | :--- |
| Maximum | $9.0 \mu$ |
| Minimum | $3.0 \mu$ |

The Tables 1, 2 and 3 show that the maximum crosssection of the fibre remains $21.0 \mu$ at the three stages of flowering and minimum is $12 \mu$ at pre-flowering and flowering stages and $15 \mu$ at post-flowering stage. The lumen of the fibre is wide at pre-flowering stage but small and similar at flowering and post-flowering stages.

### 11.4 FIBRE CHARACTERISTICS

Fibre characters differ at various stages of flowering. Accordingly they are described under the following three headings:

### 11.4.1 Pre-Flowering Stage

Transverse section at the middle region of the stem shows 2-3 and at the basal region 3-5 concentric rings of fibre blocks. The innermost ring of fibres nearer the cambium do not show discrete bundles. The number of fibres in a block varies in different layers and at different regions of the stem. Each fibre block has a number of ultimate fibre cells and at this stage the number ranges between 30 to 80 (Fig. 140, Photomicro. 60). Tangentially the fibres do not show discrete bundles nearer the cambium. There is, therefore, lot of variation in the tangential extent. The maximum radial extent of the fibre blocks interposed by soft tissue is $600 \mu$ and minimum is $300 \mu$ and the average is $414.90 \mu$. Maximum tangential extent is $300 \mu$ and minimum is $90 \mu$ and average is $189.0 \mu$.
11.4.2 Flowering Stage

As secondary growth is vigorous at this stage the fibres are in many layers and smaller wedges are formed with rays expanding on either sides. The maximum radial
extent of the fibre wedge is $795 \mu$ and minimum is $480 \mu$ and the average is $615.73 \mu(6-8$ blocks in number). The maximum tangential extent of fibre wedge is $550 \mu$ and minimum is $75 \mu$ and the average is $307.75 \mu(1-6$ blocks in number). Each fibre block has 16 to 50 fibre cells (Photomicro. 64).

### 11.4.3. Post-Flowering Stage

At this stage maximum fibre blocks are formed and these form wedges that are broader towards the cambium and narrow towards the periphery. The outermost layer of the wedge has one to two fibre blocks. The maximum tangential extent of the wedges is $810 \mu$ and minimum is $150 \mu$ and average is 475.15 ( 8 to 14 blocks in number). The maximum radial extent of fibre wedge is $990 \mu$ and minimum is $525 \mu$ with an average of $747.75 \mu$ ( 6 to 10 blocks in number). Total number of fibre blocks in each wedge is 20 to 50 and the ultimate fibre cells in a block is 20 to 60 (Fig. 142, Photomicro. 66).

The tangential and radial extent of the fibre wedges at different stages of flowering has been represented graphically in Figures 136; 137 respectively.

### 11.5 FIBRE MEASUREMENTS

Fibre measurements taken at a designated point (basal region) on the stem, at different intervals, during the plant's growth, showed gradual increase in fibre length
and diameter. A comparis on of fibres at the base and tip of the plant reveals similar differences. The length and diameter of fibres at 15 days interval were measured uptil post-fiowering stage. A graphical representation of the diameter and length of fibres has been shown in Figures 162-165, and the readings are summarized as below:

Table 4a: Length of fibres

|  | 15th day <br> (microns) | 30 th day <br> (microns) | 45 th day <br> (microns) | 60 th day <br> (microns) | 75 th day <br> (microns) |
| :--- | :---: | :---: | :---: | :---: | :---: |
| Average | 910.05 | 1184.2 | 2100.0 | 3018.2 | 5080.5 |
| Maximum | 1120.00 | 1800.0 | 2600.0 | 5000.0 | 7000.0 |
| Minimum | 400.00 | 500.0 | 1600.0 | 2300.0 | 3800.0 |

Table $4 a$ shows that there is a gradual increase in. the length of the fibre from 15 th to 75 th day of the plant.

Table 4b: Diameter of fibres

|  | 45 th day <br> (microns) | 60 th day <br> (microns) | 75 th day <br> (microns) |
| :--- | :---: | :---: | :---: |
| Average | 18.87 | 18.50 | 19.60 |
| Maximum | 30.00 | 30.00 | 37.50 |
| Minimum | 15.00 | 15.00 | 15.00 |

The diameter of the fibre from 45 th day to 75 th day does not show much increase and there is no change in diameter in comparison to length of fibre.
and diameter. A comparison of fibres at the base and tip of the plant reveals similar differences. The length and diameter of fibres at 15 days interval were measured uptil post-flowering stage. A graphical representation of the diameter and length of fibres has been shown in Figures 162-165, and the readings are summarized as below:

Table 4a: Length of fibres

|  | 15th day <br> (microns) | $30 t h$ day <br> (microns) | 45 th day <br> (microns) | $60 t h$ day <br> (microns) | 75 th day <br> (microns) |
| :--- | :---: | :---: | :---: | :---: | :---: |
| Average | 910.05 | 1184.2 | 2100.0 | 3018.2 | 5080.5 |
| Maximum | 1120.00 | 1800.0 | 2600.0 | 5000.0 | 7000.0 |
| Minimum | 400.00 | 500.0 | 1600.0 | 2300.0 | 3800.0 |

Table $4 a$ shows that there is a gradual increase in. the length of the fibre from 15 th to 75 th day of the plant.

Table 4b: Diameter of fibres

|  | 45 th day <br> (microns) | 60th day <br> (microns) | 75 th day <br> (microns) |
| :--- | :---: | :---: | :---: |
| Average | 18.87 | 18.50 | 19.60 |
| Maximum | 30.00 | 30.00 | 37.50 |
| Minimum | 15.00 | 15.00 | 15.00 |

The diameter of the fibre from 45 th day to 75 th day does not show much increase and there is no change in diameter in comparison to length of fibre.

Fibre diameter and length at three different stages of flowering is given below after summarizing the readings from Figures $163,165$.

> Table 5: Length and diameter of fibres at pre-flowering stage $(75-90$ days)

|  | Length <br> (microns) | Diameter <br> (microns) |
| :--- | :--- | :--- |
| Average | 7009.2 | 19.6 |
| Maximum | 9000.0 | 30.0 |
| Minimum | 4200.0 | 15.0 |

Table 5 shows that by pre-flowering stage (75-90 days) the length increases and is gradual (compare Tables $4 a$ and 5) whereas there is no increase in diameter (compare Tables 4 b and 5) indicating that at a certain stage (between 45 th to 90 th day) the fibre ceases to increase in diameter but grows lengthwise to a larger extent.

Table 6: Length and diameter of fibres at flowering stage (90-105 days)

|  | Length <br> (microns) | Diameter <br> (microns) |
| :--- | :---: | :---: |
| Average | 10548.2 | 18.99 |
| Maximum | 13600.0 | 30.00 |
| Minimum | 7200.0 | 15.00 |

At flowering stage as Table 6 shows there is a tremendous increase in the length of the fibre from the prefflowering stage (compare Table 5) but the diameter is just the same as at pre-flowering stage.

Table 7: Length and diameter of fibres at post-flowering stage (105-120 days)

|  | Length <br> (microns) | Diameter <br> (microns) |
| :---: | :---: | :---: |
| Average | 11834.3 | 18.8 |
| Maximum | 14060.0 | 30.0 |
| Minimum | 7800.0 | 15.0 |

Table 7 of post-flowering stage shows that there is a slight increase in the length of the fibre from 13600 to $14060 \mu$ from flowering stage (compare Table 6). The diameter remains the same showing no change.
11.6 RETTING

For the study of chemical and physical properties, the fibres were separated from other portions of the plant through 'Retting'. Retting is the decomposition of the middle lamella and a consequent separation of the individual fibres. The decomposition is brought about by bacterial action. The plants were pulled out and were tied into bundles and were placed in a horizantal position and submerged by weighting with stones in water. The
plants were allowed to remain there until the bark slipped readily from the central woody core. The period of retting varied, depending upon:
(1) the maturity of plants,
(2) the degree of lignification,
(3) the type of water,
(4) the thickness of periderm.

In Hibiscus cannabinus the plants were taken at three different stages of flowering:

1. The pre-flowering stage which is the sub-period that runs between the 15 th day after germination and the appearance of the first flower bud.
2. The flowering stage, a period during flowering.
3. The post-flowering stage which is the post-period when seeds are being formed.

The time period varied at the three stages. The fibres at pre-flowering stage could be retted at the shortest period of $7-8$ days in pond water whereas in tap water retting took a longer time. At flowering stage the time taken was $10-11$ days and post-flowering stage it took 12-14 days. The isolated fibres or 'reed' were washed, dried and cleaned. To determine the exact time period samples were retted separately at different time intervals in tap and pond water. Retted fibres were


#### Abstract

creamy to silvery white, lustrous, soft to feel and meshiness was high. The fibres at pre-flowering stage were highly lustrous and at post-flowering stage the lustre of the fibres was lesser. Similarly meshiness of the fibre is less at pre-flowering and flowering stages and high at post-flowering stage. The fibres obtained after the completion of the above process were taken for further observations.


### 11.7 CHEMICAL ANALY SIS

The chemical analysis of fibres was made through different phases. Fibres of the pre-flowering, flowering and post-flowering stages were analysed separately consisting of the elements listed in Tables 8,9 and 10.

Table 8: Pre-flowering stage (75-90 days)

|  | Chemical constituents | Percentage |
| :--- | :--- | :---: |
| 1. | Ash | 0.43 |
| 2. | Moisture | 7.19 |
| 3. | Cellulose | 81.80 |
| 4. | Fats | 0.93 |
| 5. | Lignin | 8.59 |
| 6. | Pectin | 1.00 |

Table 9 shows the chemical composition of fibres at flowering stage.

```
Table 9: Flowering stage (90-105 days)
```

|  | Chemical constituents | Percentage |
| :--- | :--- | :---: |
| 1. | Ash | 0.91 |
| 2. | Moisture | 6.28 |
| 3. | Cellulose | 80.36 |
| 4. | Fats | 0.96 |
| 5. | Lignin | 10.90 |
| 6. | Pectin | 1.34 |

Table 10 shows the chemical composition of fibres at post-flowering stage.

| Table 10: Post-flowering stage | $(105-120$ day $)$ |  |
| :---: | :---: | :---: |
| 1. | Chemical constituents | Percentage |
| 2. | Ash | 1.32 |
| 3. | Moisture | 5.28 |
| 4. | Fatlulose | 76.40 |
| 5. | Lignin | 0.98 |
| 6. | Pectin | 14.51 |

From the above observations, Hibiscus cannabinus in its chemical composition shows that it is mainly composed of cellulose and lignin. At the three different stages of flowering cellulose shows a gradual decrease as the plant matures and simultaneously the
percentage of lignin increases. There is a gradual increase in the ash, fat and pectin contents and decrease in moisture.
11.7.2 Colour Reactions

Sections cut serially of the fresh stem from tip to base indicated that lignification is not uniform along the length of the stem. At the shoot apex the young fibres which have no secondary and tertiary walls show positive results for cellulose and moving downwards the reactions for lignin were positive though the quantity differed, judged by the colour of fibres after treating with the reagent. The mature fibre blocks at the basal region of the stem are strongly lignified. In Hibiscus cannabinus lignification of the fibre cells was observed as early as in eight day old seedling.

The tests were confirmed by the various reagents given below:

1. The fibres gave yellow colour when treated with iodine and sulphuric acid showing the presence of lignocellulose.
2. The fibres were steeped in $2 \%$ aniline hydrochloride in a watch-glass for a few minutes, ligno cellulose turned yellow.
3. With chloroiodide of zinc the fibres gave a yellow colour showing the presence of lignocellulose.
4. Sections of fresh material when treated with phloroglucinol and HCl gave deep magenta colour.

### 11.7.3 Dyeing

The fibres in $H$. cannabinus have an affinity to combine directly with dyes. When treated with the dyes the reactions were as following:

| Dyes | Colour <br> reactions |
| :--- | :--- |
| Phloroglucinol + HCl | Magenta |
| Chloroiodide + zinc | Lustrous yellow |
| Congo red | No colour |
| Orange G | Orange |
| Methylene Blue | Dark blue |
| Gentian violet | Violet |
| Aniline blue | Blue |

### 11.7.4 Bleaching

The fibres were bleached by scouring with alkali, and carefully treated with hypochlorite resulting into a permanent white colour.
11.8 PHYSICAL ANALYSIS

The average reed length of Hibiscus cannabinus at pre-flowering stage is $75-110 \mathrm{cms}$; at flowering stage

140-165 ms and at post-flowering stage 165-185 ms. The physical properties of the fibres at three different stages of flowering are given in Tables 11,12 and 13.

Table 11: Pre-flowering stage (75-90 days)

## Physical characters

1. Fibre bundle tenacity

$$
\begin{aligned}
& 24.72 \mathrm{gms} / \text { tex on } 1.5 \mathrm{~cm} \\
& \text { test length }
\end{aligned}
$$

2. Breaking elongation
3. $2 \%$
4. Fineness of 2 mm fibre pieces (combed fibre)
1.8 tex
5. Young's modulus of elasticity of fibre
$1.02 \times 10^{11}$ dynes $/ \mathrm{cm}^{2}$
6. Area in cross-section
7. Density
$141 \times 10^{-5} \mathrm{~cm}^{2}$
$1.25 \mathrm{gm} / \mathrm{cc}$

Table 12: Flowering stage (90-105 days)
Physical characters

1. Fibre bundle tenacity
$37.5 \mathrm{gms} /$ tex on 1.5 cm
test length
2. Breaking elongation
$5.2 \%$
3. Fineness of 2 mm fibre pieces (combed fibre)
2.4 tex
4. Young's modulus of elasticity of fibre
$1.25 \times 10^{11}$ dynes $/ \mathrm{cm}^{2}$
5. Area in cross-section
$156 \times 10^{-5} \mathrm{~cm}^{2}$
6. Density
$1.32 \mathrm{gm} / \mathrm{cc}$

Table 13: Post-flowering stage (105-120 days)

## Physical characters

1. Fibre bundle tenacity
2. Breaking elongation
3. Fineness of 2 mm fibre pieces (combed fibre)
4. Young's modulus of elasticity of fibre
5. Area in cross-section
6. Density

> 37.7 gms $/$ tex on 1.5 cm test length
$4.9 \%$
2.5 tex
$1.15 \times 10^{11}$ dynes $/ \mathrm{cm}^{2}$
$168 \times 10^{-5} \mathrm{~cm}^{2}$
$1.38 \mathrm{gm} / \mathrm{cc}$

A comparative study of the physical analysis at three different stages of flowering, shows the tenacity of fibre is very low at pre-flowering stage which is $24.72 \mathrm{gms} /$ tex and at flowering and post-flowering stage the difference is of $0.2 \mathrm{gm} / \mathrm{tex}$. The breaking elongation is highest at flowering stage viz. $5.2 \%$ and very low at pre-flowering stage 1.2\%. Fineness of the fibre is maximum at post-flowering 2,5 tex and minimum at preflowering 1.8 tex. Area of cross-section is maximum at post-flowering stage $168 \times 10^{-5} \mathrm{~cm}^{2}$ and minimum at preflowering stage $141 \times 10^{-5} \mathrm{~cm}^{2}$. The elasticity of the fibre is maximum at flowering stage $1.25 \times 10^{11}$ dynes $/ \mathrm{cm}^{2}$ and minimum viz. $1.02 \times 10^{11}$ dynes $/ \mathrm{cm}^{2}$ at pre-flowering stage. Density is maximum $1.38 \mathrm{gm} / \mathrm{cc}$ at post-flowering stage and minimum $1.25 \mathrm{gm} / \mathrm{cc}$ at pre-flowering stage.
12.1 Mi crosporogenesis
12.2 Ovule
12.3 Megasporogenesis
12.4 Fertilization
12.5 Endosperm
12.6 Seed Coat
12.7 Embryo Development
12.8 Root Apical Organization
12.9 Shoot Apical Organization
12.10 Plastochronic Changes
12.11 Seedling Vasculature
12. 12 Nodal Vasculature
12.13 Phyllotaxy and Trace Relatioms
12.14 Petiole
12.15 Leaf Histology
12.16 Leaf Venation
12.17 Trichomes
12.18 Floral Vasculature
12.19 Morphology of Bark
12.20 sieve Tubes
12.21 Phloem Rays
12.22 Expansion Tissue
12.23 Periderm
12.24 Origin and Development of Fibres
12.25 Secondary Fibre Development
12.26 Cell Wall of Fibre
12.27 Fibre Characteristics
12.28 Retting
12.29 Chemical Properties
12.30 Physical Properties
12. DISCUSSION

## 12. 1 MICROSPOROGENESIS

The archesporium in Hibiscus cannabinus differentiates at two places in the anther primordium as the anthers are monothecous. Same was also reported by Rao (1954) in other members of Malvaceae. The number of microspore mother cells in a row is 13-15. Rao (1954) observed 5 cells in Sida cordifolia, 10 cells in Abutilon indicum and 15 cells in pavonia zeylanica in a single row.

The epidermal cells become tangentially stretched and get deeply stained which Rao (1954) observed in other members of Malvaceae and called the deeply stained contents as tannin. The endothecium gets radially elongated and develops fibrous thickenings. The single middle layer gets crushed. The innermost layer, the tapetum is one layered. Rao (1954) observed some cells of the tapetum dividing periclinally and becoming two-layered at places in Althaea rosea.

In Hibiscus cannabinus the tapetum is of the plasmodial type, a feature shared by all the members of Malvaceae investigated by Schnarf (1931) and Rao (1954). The tapetal cells enlarge and their nuclei divide mitotically, and as a result $2-7$ nuclei are formed in each cell. A similar situation has been reported in

Hibiscus solandra and Malvaviscus arboreus (Rao, 1954) where each tapetal cell has 3-20 nuclei. The protoplasts of the tapetal cells gradually get absorbed and disappear at the 2-nucleate stage of microspore. Rao (1954) reported in Urena lobata the tapetal protoplasts disappear before microspore nucleus has divided and in sida carpinifolia and Pavonia zeylanica the remnants persist till the microspores have become 2-nucleate.

In Hibiscus cannabinus primary sporogenous cells function directly as microspore mother cells as also reported by Rao (1954) in Althaea rosea, Pavonia zeylanica and Sida cordifolia. In Abutilon indicum (Rao, 1954), however, primary sporogenous cells undergo divisions to increase the number of sporogenous cells.

In Hibiscus cannabinus microspore tetrads are tetrahedral and bilateral. Rao (1954) observed these two types of tetrads in other members of Malvaceae. In H1 biscus solandra (Rao, 1954) in addition to the above two types of tetrads linear tetrads were also noticed.

In Hibiscus cannabinus the microspore nucleus divides in the centre of pollen grain. The larger vegetative nucleus remains in the centre surrounded by its own cytoplasm and the generative nucleus moves towards the periphery. The vegetative nucleus degenerates by the time the generative nucleus divides to form two spherical male gametes inside the microspores before they are shed. The
pollen grains are shed at the two male gamete stage, however, in Hibiscus hirtus and Malachra capitata (Rao, 1954) the pollen grains are shed at the two nucleate nucleus
stage where the generativekhas not yet divided. Guignard (1904) noticed the division of generative nucleus in the pollen tube in Althaea, Hibiscus and Lavatera. In Kydia calycina (Rao, 1954) the pollen grains are shed at the two male gamete stage.

The pollen grains in Hibiscus cannabinus are 80-130 $\mu$ in size. Rao (1954) reported the largest pollen grain of $120 \mu$ in Hibiscus solandra and of $60 \mu$ in Abutilon Indicum.

The pollen grailns are large, spinescent and multiporate, a characteristic feature in all the members of Malvaceae. The exine is thick ranging from 8-12 $\mu$. In Sida and Malachra capitata (Rao, 1954) the exine is $5 \mu$ and 10-12 $\mu$ in thickness respectively.

### 12.2 OVULE

The ovule in Hibiscus cannabinus is bitegmic, crassinucellate and campylotropous. Rao (1954) has reported bitegmic, crassinucellate and campylotropous ovules in the members of Malvaceae except in Kydia calycina and Thespesia populnea where the ovules are anatropous. In Gossypium species (Gore, 1932) also the ovules are anatropous.

The initials for the inner integument differenttiate first but the outer integument grows faster than the inner and covers the nucellus by the time the megaspore tetrads are formed. A similar situation has been recorded in other members of Malvaceae (Rao, 1954). The micropyle in Hibiscus cannabinus is formed by both the integuments and has a zigzag course as observed in Side veronicaefolia (Roo, 1954). The micropyle in Hibiscus cannabinus points towards the axis of the ovary. Similar observation has been recorded for Hibiscus micranthus, Hibiscus hirtus and Abutilon indicum (Rap, 1954). In Pavonia zeylanica, Melachra capitata, Urena Lobata, Malvaviscus arboreus and Althea rosea (Rap, 1954) it faces the base of the ovary.

The nucellus in Hibiscus cannabinus is markedly curved. Rio (1954) observed such a markedly curved nucellus in side species, Abutilon indicum and Hibiscus solandra and straight nucellus in Kydia calycina and Thespesia populnea.

### 12.3 MEGASPOROGENESI S

In Hibiscus cannabinus the archesporium consists of a single hypodermal cell. Occasionally two cells function till megaspore mother cell stage. Rad (1954) observed the archesporium consisting of a single hypodermal cell in sida species, Abutilon indicum, Hibiscus hirtus and Hibiscus solandra and a group of hypodermal cells as
in Althaea rosea, Urena lobata and Malvaviscus arboreus. In Malachra capitata (Rao, 1954) two cells may function till megaspore mother cell stage but in Urena lobata and Kydia calycina the cells continue to function till embryo sac stage.

In Hibiscus cannabinus at the time of meiosis in the megaspore mother cell, accessory sporogenous cells develop from one or more parietal cells, a feature to which Stenar (1925) had drawn attention and was observed in Sida species, Altheea rosea, Abutilon indicum, Urena and Malvaviscus arboreus (Rao, 1954). The megaspore mother cells are elongated and tapering in Hibiscus solandra and Hybiscus micranthus (Rao, 1954). This feature is sirailar to that in Hibiscus cannabinus. In dyads in the second division the cell wall is laid in a horizontal manner so that the megaspore tetrads are linear which is common in many members of Malvaceae (Rao, 1954). The chalazal megaspore functions further. The accessory sporogenous cells in Hibiscus cannabinus degenerate before undergoing meiotic divisions, whereas in other members of Malvaceae the accessory sporogenous cells further undergo meiotic divisions (Rao, 1954).

The eight nucleate embryo sac is derived in Hibiscus cannabinus by three successive free nuclear divisions, a common feature in Malvaceae. The two polar nuclei maet in the middle of the embryo sac and remain adpressed. They move together upwards and lie near the egg till fertilization occurs. Sinilar situation has been observed by

Rao (1954) in members of Malvaceae except in Althaea rosea and Malvaviscus arboreus where the two polar nuclei fuse bef ore fertilization. Antipodals are three in number and degenerate when the embryo is at the 8 -celled stage. This is in contrast to the earlier report of Medvedeva (1944) who observed that the antipodal cells degenerate usually before fertilization in $H$. cannabinus. In sida veronicaefolia, Kydia calycina, Malvaviscus arboreus and Hibiscus solandra the antipodals are three in number whereas in Althaea rosea, Abutilon indicum, Malachra capitata and Hibiscus micranthus either the antipodal nuclei divide in free nuclear manner or a cell wall is formed after the division so that binucleate and supernumerary anti podals are formed (Rao, 1954).

### 12.4 FERTILIZATION

The pollen grains in Hibiscus cannabinus are large and polysiphonous as has been already noticed by stenar (1925), Lang (1937) and Rao (1954). The pollen tubes become broad and send out branches of different sizes crushing the nucellar tissue. A similar situation has been observed in Hibiscus micranthus (RaO, 1954). The pollen tubes are 10-24 $\mu$ wide and persist till the heart shaped stage of the embryo. Ras (1954) observed 15-20 $\mu$ wide and persistent pollen tubes in Hibiscus solandra and Abutilon indicum.

The male nuclei show nucleoli surrounded by a vacuolated region in Hibiscus cannabinus. The male gametic
nuclei do not show nucleoli in Malva rotundifolia and Lavatera thuringiaca (Stenar, 1925) but Rao (1954) observed nucleoli in male nuclei of Abutilon indicum.

### 12.5 ENDOSPERM

The two polar nuclei remain adpressed and do not fuse before fertilization. After fertilization and fusion of the male gamete with the polar nuclei the so formed triploid endosperm nucleus canes to lie just below the egg cell. In Hibiscus cannabinus the primary endosperm nucleus divides to form 10-16 nuclei before the zygote undergoes division. In Hibiscus solandra (Rao, 1955) 4-8 endosperm nuclei are formed when the fertilized egg undergoes the first division. In Hibiscus cannabinus uptil the early globular stages of embryo the free nuclei of endosperm aggregate in a compact manner, encircling the proembryo. At the late globular stage of embryo, cell wall commences at the micropylar end and proceeds towards the chalazal end, as was also reported in other members of Malvaceae by Rao (1955). Endosperm is completely cellular at the heart shaped stage of embryo. According to Rao (1938) a correlation exists between the occurrence of nuclear endosperm and embryo differentiation confirming the easy availability of nourishment from free nuclear endosperm and in early differentiation of the embryo. However, Kapil and Bhatnagar (1972) do not agree with this view, as in Euphorbiaceae the cell wall formation begins around the embryo at the early globular stage and becomes completely
cellular by the time embryo becomes heart shaped. This situation is similar to that in Hibiscus cannabinus. In Hibiscus cannabinus endosperm nuclei in the middle and chalazal region of the embryo as was also seen in other members of Malvaceae (RaO, 1955) where polyploid nuclei in endosperm are restricted to the antipodal part.

In the mature seeds a little endosperm could be seen in Hibiscus cannabinus as in other members like Sida, Pavonia zeylanica and Abutilon indicum (Rao, 1955) while in Hibiscus micranthus (Rao, 1955) mature seeds lack endosperm.

Perisperm of two to three layers was observed in the mature seeds of Hibiscus cannabinus. Reeves (1936) observed perisperm of one cell in thickness in members of Malvaceae. According to Rao (1955) no perisperm was found in the seeds of the members of Malvaceae studied by him.
12.6 SEED COAT

Broadly the structure of seed coat of the presently investigated species Hibiscus cannabinus resembles with the earlier records of Sida cordifolia, Pavonia zeylanica, Malachra capitata, Abutilon indicum and Hibiscus species by Reeves (1936) and Rao (1955).

The sequence of changes during development of the seed coat at different stages of embryo and the structure, reveals that the seed coat is formed by both the inner and
outer integument. The outer integument is very thin consisting of two layers of cells which become shrivelled in the mature seed. Hairs were not observed in the early stages but in mature seeds numerous papillae were observed in Hibiscus cannabinus. Rao (1955) reported in Hibiscus solandra an d Pavonia zeylanica the cells oi the testa developing into papillae whereas in Hibiscus micranthus and Hibiscus hirtus the testa developed hairs. In Hibiscus cannabinus the inner integument is very thick in early stages but as the seed matures it gets reduced in thickness as the parenchymatous cells between pigment and fringe tissue get crushed. A similar observation was recorded by Reeves (1936) and Rao (1955) in other members of Malvaceae.

The outer epidermis of the inner integument also known as palisade layer differentiates in the region nearest the hilum and progresses towards the rest of the sides of ovule. The palisade cells begin to elongate radially. Lignification of the palisade cell commences from the lower region of the cells and gradually advances upwards. By the time the seed matures the entire cell is lignified and thickened. The cells have fibrous thickening. The protoplast of the palisade cells degenerate completely. Rao (1955), in other Hibiscus species, reported remnants of the protoplast in lumen of palisade cells confined to the outer half and has not mentioned the fibrous thickenings. The palisade layer of the mature seed occuples more than half the thickness of the seed coat.

In Hibiscus cannabinus immediately below the palisade layer is a single pigmented layer on all sides except at the micropyle and funicular region where it is many layered. These pigmented cells are not confined to the inner integument only but are also found in the thickened tip of the micropyle and the funicular region of the outer integument. Rao (1955) reported such cells only in the inner integument in other members of Malvaceae.

The fringe tissue in Hibiscus cannabinus is lignified as was also observed by Reeves (1936) and Rao (1955) in other members of Malvaceae.

### 12.7 EMBRYO DEVELOPMENT

The first division of the zygote is transverse resulting in a two celled proembryo in H. cannabinus. The apical cell 'ca' undergoes a vertical division Eiving rise to two juxtaposed cells while the basal cell ' cb ' undergoes a transverse division forming a middle cell ' $m$ ' and an inner cell 'ci'. The two cells of the terminal tier divide longitudinally forming quadrants termed ' $q$ '. ' $M$ ' divides vertically and 'ci' transversely forming two superposed cells ' $n$ ' and ' $n$ '. The quadrants of tier ' $q$ ' divides obliquely to give four centrally placed cells 'a' and four peripherally placed cells ' $b^{\prime}$. ' $N$ ' and ' $m$ ' divide vertically and ' $n$ ', by a transverse division giving rise to two cells ' 0 ' and ' $p$ '. The embryo at this stage is 16-celled and 5-tiered. Thus embryo development in Hibiscus
cannabinus confirms the Urtica variation of the Asterad type (Johansen, 1950). Similar development of the embryo occurs in all the members of Malvaceae studied by Rao (1955).

The demarcation of dermatogen in the terminal tier occurs in the cells of 'a' and then in the group of cells 'b'. Similar situation was observed in Sida cordifolia and reverse in Sida veronicaefolia (Rao, 1955). In Hibiscus cannabinus one of the two cells of 'a' of the tier ' $q$ ' divides transversely cutting of $f$ the dermatogen initial to the outside but the other divides in an inclined manner so as to form a cell that looks like an epiphysial cell. In later stages it loses its identity and appears like other cells. Such a variation was observed by Soueges (1922) in Malva rotundifolia and Rao (1955) in Hibiscus solandra.

The two groups of superposed cells of tier 'o' give rise to the root ceap in Hibiscus cannabinus. In Abutilon indicum the root cap initiating layers are variable being one or two and in Hibiscus species the root cap initiating layers are variable but more commonly it is two (Rao, 1955).

The tier ' $p$ ' in Hibiscus cannabinus divides transversely once to form two celled suspensor. The upper short stalk connecting the embryo to the larger vesicular lower cell. The suspensor in Hibiscus hirtus, H. micranthus and $\underline{H}$. solandra becomes large, boot shaped and many

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celled (Rao, 1955).
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12.8 ROOT APICAL ORGANIZATION

In Hibiscus cannabinus at the late globular stage of embryo the promeristem with a prominent large 'central cell' has been observed. Guttenberg, Butrmeister and Arosel. (1955) have reported a large central cell during the late embryogeny of Helianthus which is transitory and gives place to a plate composed of a large number of cells to add to the promeristem. Clowes (1961) has also observed one large cell in embryos of same species but criticized the central cell theory stating that the central cell does not have any ontogenetic role. In Hibiscus cannabinus the central cell is surrounded by five prominent cells at the late globular stage of embryo. These cells divide transversely and constitute the promeristem. The central cell loses its identity by torpedo stage of the embryo and does not seem to play any further ontogenetic role. In Prosopis fuliflora (Trivedi, 1969) the central cell remains during all stage of embryo development as well as in mature root apex but does not play any ontogenetic role.

At the torpedo stage of embryo in Hibiscus cannabinus discrete groups of initials have been observed for central cylinder, cortex and columella. Byrne and Heimsch (1970) have reported discrete initials for the central cylinder, cortex and a layer of secondary columella in the fully developed embryo of Malva sylvestris. Perhaps
these authors have not traced the columella initials from the early stages of embryo and so it is difficult to accept the position of the secondary columella. Initials described by them. In Hibiscus cannabinus the primary columella initials could be distinguished as early as globular stage of embryo. If the plate of initials surrounding the cortical initials are the secondary colunella as in Malva sylvestris (Byrne and Heimsch, 1970) then there will be distinct patches of columella cut off by primary and secondary columella initials. Such a situation is not noticed in root apex of Hibiscus cannabinus. But Trivedi (1969) could distinguish the columella initials at the globular stage in Prosopis juliflora and distinguished primary and secondary columella.

The next aspect that requires attention is the nature of root cap of Hibiscus cannabinus. Through stages of development the cap shows two regions:
(a) periphery of the cap,
(b) central column or columella of the cap.

These two regions are ontogenetically independent. The columella has a separate set of initials. The cells divide in a rib meristem fashion, thereby the rows of columella maintain the same width as that of its initials. Consequently cells of columella neither add nor become a part of the peripheral region of cap. In contrast the periphery of the cap is derived by proliferation of surface
layer by Kappe type of division which shows oblique division figures so that there is a tapering of peripheral rows of the cap. As the peripheral rows are sloughed off during growth new layers are added in the same fashion. These ontogenetic differences have been earlier pointed out by Deshpande and Bhatnagar (1961) for Ephedra foliata and Deshpande and Joneja (1962) for Leptadenia pyrotechnica, Pillai and Sukumaran (1969) for Cyamopsis tetragonoloba.

Guttenberg et al. (1955) reported the transition from a closed pattern to an open pattern of organization during root elongation in Helianthus. Guttenberg (1960) introduced the term 'closed' to describe the embryonic condition where the cell rows converge at the apex and the term 'open' to describe the ontogenetic change in the pattern where the convergence is lost. Seago and Heimsch (1969) and Byrne and Heimsch (1970) have reported the ontogenetic opening of the closed embryonic pattern of organization in some members of Convolvulaceae and Malva sylvestris respectively. In contrast Heimsch (1960) and Byrne and Heimsch (1968) have reported that the closed embryonic pattern is maintained during growth in tomato and flax. In Hibiscus cannabinus the embryonic root apex show closed type of organization. But as the growth proceeds the initials of central cylinder becone continuous with the secondary columella so that the root apex becones open. The cells between the columeila and
differentiating epidermis contribute to the lateral root cap by successive periclinal divisions. This has been observed in the root cap of dicotyledons (Clowes, 1959; Esau, 1965; and Byrne and Heimsch, 1970).

Many authors have attempted to interpret root apical organization in terms of certain concepts. According to Hanstein's histogen concept (1868) as it appears in the literature any apical meristem should have at least three superimposed tiers of initials of which the cells of the outermost should divide only in anticlinal plane. Although initially rigid and put forward for explaining shoot apex organization, this concept with suitable modifications and in combination with 'Korper-Kappe' has proved its utility for explaining root apical meristem rather than shoot apical meristem which now and for the past many decades are explained in terms of tunica-corpus organization. Trying this concept for root apex of Hibiscus cannabinus it should be restated that apex here shows three superimposed tiers of initials viz. columella initials, cortical initials and stelar initials. These three tiers of superimposed initials do satisfy the first requirement of the concept. The second requirement of the concept is that the outermost layer should divide only in anticlinal plane. In the iiterature on histogen concept, outermost layer is regarded as dermatogen. In Hibiscus the dermatogen (protoderm or epiblema) does divide anticlinally towards
root body side but toward root cap the layer undergoes divisions in a proliferating manner so as to form the periphery of the root cap. Hanstein perhaps did not envisage the possibility of this concept being applied to root apices and so the formation of cap was not considered. This was later done by Janczewski (1874) who took into account the $r$ elationship of root body with the root cap and designated cap histogen as 'calyptrogen'. The situation obtaining in H. cannabinus has been described by Janczewski as 'Dermacalyptrogen' which is quite common among dicotyledons.

The configuration with tiers at the apex continues until the seedling stage of the plant. During germination at varying days the root apical organization undergoes certain changes. The first change is that some of the rows formed by proliferation of surface and subsurface layers continue to contribute to the peripheral region of the cap, but as the columella rows become longer the peripheral rows change their orientation abruptly forming 'L' shaped files of cells. These 'L' shaped files surround primary columella and have similar orientation as that of the rows of primary columella. On this ground the author would like to designate these files as 'secondary columella'. Concomitantly the cells around the tiers of cortical and stelar initials become conspicuous and assume the cytogenerative function. This brings in the consideration of Clowes $(1953,1954)$ concept of root promeristem as being composed of 'minimal constructional
centre' and 'cytogenerative centre'. According to this concept minimal constructional centre is composed of cells which had a meristematic role sometime in early phases of ontogeny but later, as the root grows, meristematic activity shifts to the cells surrounding the initial tiers. These cells are termed as cytogenerative centre. The minimal constructional centre is said to take no part in histogenesis but can provide for 'repair' of the cytogenerative centre (or promeristem). The so-called initials of earlier stages now maintain an apical configuration only.

The next change that takes place in the root apex of Hibiscus cannabinus as it grows is that some of the cells of secondary columella on which abut the cortical rows contributing to the periphery of the cap become conspicuous and appear meristematic looking like initials. These are termed as 'pericolumnar initials' following Schopf (1943) who used this term for Larix lariciana. Although Larix belongs to a different group of plants, situation as far as these cells are concerned, is similar to Hibiscus irrespective of apical configuration. In Hibiscus the pericolumnar initials along with cytogenerative centre $4 s$ described earlier give an appearance of an inverted cup or deep saucer shaped promeristem. The sides of the cup on cap side are composed of pericolumnar initials, on root body side by cortical cells of cytogenerative centre and base by stelar cells of cytogenerative centre. The inside of the cup consists of primary
columella rows, and the entire minimal constructional centre which includes the earlier tiers of cortical initials and stelar initials which are now quiescent. The foregoing account is a direct pointer to the inverted cup concept and quiescent centre concept of Clowes (1950, 1956a,b). Pillai (1963) has reported the 'inverted cup' concept for the root apices of Cycads and Ginkgo biloba.

Thus, it would appear that 'Histogen concept' with some modification is applicable to root apical meristem at embryo stage of Hibiscus cannabinus and promeristem of cytogenerative centre and minimal constructional centre, inverted cup shaped concept and quiescent centre concept are all applicable as the root grows during seedling stages. The fact that all these concepts are applicable at one stage or the other of the same root apex only indicates that earlier authors considered only a particular stage and put forward a concept instead of tracing the development of root apical meristem.

All the concepts mentioned in the preceding pages are concerned with the meristematic activity at the apex. These concepts do not consider the cell lineages away from the apex and in the root cap. To explain these we have to consider the 'Korper Kappe' concept of Schuepp (1917). According to this concept the histological pattern just behind the promeristem shows $T$ shaped divisions where head of T is pointed towards the promeristem region. This according to Schuepp is 'Korper' type of division.

Contrasting this is 'Kappe' division where head of $T$ is directed towards root body. 'Korper' type predominates root body and 'Kappe' type is exclusive to root cap. The greatest merit of this concept is that it works in combination with any concept that has so far been put forward to explain root promeristem. This concept was used by Wagner (1939) in analysing the root cap in several plants. Clowes (1950) used this concept to explain root apex of Fagus sylvatica in combination with histogen concept.

In the present study histogen concept with 'Korper Kappe' is applicable to the root promeristem at embryo stage, inverted cup and quiescent centre concepts in combination with 'Korper Kappe' are applicable to the root apical meristem of seedlings.
12.9 SHOOT APICAL ORGANIZATION

There is a controversy in the literature as to whether shoot pole precedes root pole in differentiation during histogenesis of the embryo. In Hibiscus cannabinus the peripheral part of the root cap becomes distinct at the late globular stage by the Kappe divisions of the protodermal cells. This situation is similar to that described by Miller and Wetmore (1946), Reeve (1948) and Guttenberg et al. (1954a,b, 1955, 1960) in Angiosperms where the root pole is first to become distinct.

In contrast, Meyer (1958) and Mahlberg (1960) have put forward the view that the shoot pole is the first to become distinct in the embryo of McIntosh apple and Nerium respectively. In Avicenia and Epithema (Padmanabhan, 1964, 1967) and Albizzia (Pillai et al., 1974) similar observations have been recorded.

The shoot apex in Hibiscus cannabinus is a low dome and shows marked cytohistological zonation namely a single tunica layer, central mother cells zone, pith rib meristem and flanking zone. Hence, the shoot apex is the normal angiosperm type (Popham, 1951). The single tunica layer undergoes only anticlinal divisions. The stratification of the upper corpus forms a continuous layer below the single tunica layer. The cells of this layer divide periclinally during the initiation of leaves. Tolbert and Johnson (1966) reported in their study on 40 species of Malvaceae including Hibiscus, that majority of the species have single layered tunica and the stratification of the upper corpus is common. The apices of many species of Malvaceae are domed and some are flat and do not extend above the insertion of the youngest leaf primordium, nevertheless all of them show more or less marked cytohistological zonation.

Johnson and Tolbert (1960) introduced the concept of 'metra meristem' to designate the central part of the shoot apex, which include the tunica and the corpus initials, which later Tolbert and Johnson (1966) used in
describing the apical structure and activity of the apices in Malvaceae members. The the ory of the French School (authors of French School) can be explained as conceiving the apical zone including the tunica corpus together as 'meristeme $d$ ' attente' the flank meristem as the 'anneau initial' and the pith meristem as the 'meristeme medullaire'. Newman (1961, 1965) on the other hand recognized the 'monoplex' (with a single apical cell at the shoot apex), the 'simplex' (with a single zone of initial which is superficial at the shoot apex) and the 'duplex' (having two discrete zones of inftials, one zone superimposed on the other) types of shoot apical organizations on the basis of a continuing meristematic residue'.

## 12. 10 PLASTOCHRONIC CHANGES

Schmidt (1924) has recognized two phases of plastochron on the basis of the size of the shoot apex; the 'maximal phase' - a stage of maximal apical area with the beginning of leaf initiation; and the 'minimal phase' - a stage of reduced apical area after the appearance of a leaf primordium. One plastochron stage gradually enters the other showing structural changes so that it has been further sub-divided into many phases by various workers. Abbe, Phinney and Baer (1951) divided shoot apex of maize into 3 plastochrons viz., early, mid and late substages. Early substage: when the uppermost leaf primordium was barely visible and leaf primordium is
identifiable externally by the slightly perceptible bulge. Late substage: where the shoot apex is asso ciated with a well developed leaf primordium and mid substage: where all stages intermediate between the early and late substages. Paolillo and Gifford (1961) divided Ephedra apex into 5 stages viz., (1) minimal, (2) early post minimal, (3) late post-minimal, (4) pre-maximal, and (5) maximal. Cohen (1965) described 4 phases of plastochron in the inflorescence apex of Arceuthobium viz., (1) phase of maximal area, (2) phase of minimal area, (3) phase of early post-minimal area, and (4) phase of late post-minimal area. Trivedi (1969) has recognized three phases viz., maximal, minimal and mid-plastochron in Prosopis juliflora.

The shoot apex of Hibiscus cannabinus at the maximal stage of the plastochron is a low dome with an average height of $36.0 \mu$, and width of 147-153 $\mu$. The apex is almost flat during the minimal phase of the plastochron with an average width of $84.0 \mu$ and the leaf primordium at this phase attains the maximum height of 145-150 $\mu$ before the initiation of the next primordium. There is gradual increase in width and height of the apical dome as it enters from the minimal to the midplastochron stage along with the decrease in height of leaf so that at mid-plastochron the height of the dome is 20-24 $\mu$ and width is $135-141 \mu$ and the leaf primordia attains a height of $27 \mu$. At the maximal phase the leaf initiation just begins.

## 12. 11 SEEDLING VASCULATURE

Transition in Hibiscus cannabinus does not take place by splitting, rotation and fusion as put forward by Van Tieghem (1891) and the four different types recognized by Eames and MacDaniels (1947). According to Bonnier ( $1900 \mathrm{a}, \mathrm{b}$ ) there is a gradual shift in the pole of differentiation of xylem. He explains that in very young roots, rows of undifferentiated cells connect the first formed xylem vessels with the first formed phloem elements. Along these undifferentiated cells, differentiation proceeds simultaneously from phloem and xylem poles, in opposite direction. This displacement of the pole of xylem differentiation, along with approximation of phloem strands is the reason for varied arrangement of vascular tissue in the seedling axis.

Chauveaud (1911, 1923) has put forward still another view based on the study of the spermatophytic hypocotyl and recognition of three arrangements of vascular elements in the seedling - the alternate, the intermediate and the superimposed. In the first 'disposition alterni', the vessels differentiate radially and centripetally alternating with groups of sieve tubes round the circumference of the stele - the root structure. In the third, the 'disposition superposes', the vessels differentiate centrifugally and lie on the same radius as the sieve tubes - the stem structure. The 'disposition intermediaire' is exhibited by tangentially arranged
vessels and sieve tubes and is intermediate between the two arrangements. According to him these arrangements in the axis represent successive stages of vascular evolution.

In H. cannabinus the peripheral xylem gradually degenerate few centrifugal xylem develops from the inner part of procambial strands. Obliteration of peripheral xylem and formation of centrifugal xylem indicate a shift in the pole of differentiation of xylem. This observation goes in support of Bonnier's view. Sebastian (1971) finds a shift in the pole of xylem differentiation and obliteration of centrifugal xylem in Amaranthus leucocarpus. Pillai et al. (1974) also recorded degeneration of centripetal xylem in Albizzia lebbeck and the development of lateral and centrifugal xylem is independent of the centripetal xylem. Murray (1933) has given a physiological interpretation for the obliteration of centripetal xylem. Gradual alteration in position of the food conduction results in the change of focus of lignification of xylem vessels resulting in gradual degeneration of the original xylem.

### 12.12 NODAL VASCULATURE

The cotyledonary node and the first foliar node in Hibiscus cannabinus are unilacunar single trace and the subsequent nodes are trilacuriar. The trilacunar nodal condition was considered to be the besic type and that unilacunar and multilacunar derived from it (Sinnot, 1914).

Marsden and Bailey (1955), Bailey (1956) and Canright (1955) concluded on the basis of comparative, morphological and ontogenetic data that the unilacunar double trace condition is the most primitive and not the trilacunar node. Fahn (1967) reported that the unilacunar node of certain genera of Ranales is primitive and in certain dicotyledonous genera the unilacunar node has been derived from trilacunar node, and in still other dicotyledonous groups the tri- and multilacunar conditions have arisen from unilacunar node. According to Benzing (1967) the anatomy of cotyledonary node does not necessarily reflect ancestral conditions in the mature stem. The unique seedling mar phology and decussate insertion of the cotyledons make this unlikely. Benzing (1967) taking evidence from his studies on Ranales, further adds that primitive node in Angiosperms and possibly in all seed plants is one trace unilacunar or trilacunar. Two trace unilacunar condition may have evolved in conjunction with decussate phyllotaxy. While according to Bailey considerable number of plants have unilacunar cotyledonary node.

In Hibiscus cannabinus the cotyledonary node and the first foliar nodes are unilacunar single trace and the following foliar nodes are trilacunar indicating that unilacunar single trace is the basic type and by addition of laterals could give rise to trilacunar foliar nodes.

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Hibiscus cannabinus bears free lateral stipules. According to Sinnott (1914) and Dormer (1944) all the plants bearing stipules show trilacunar condition, the lateral traces branching for stipular supply, while those showing unilacunar are exstipulate. However, Borreria hispida (personal observation) and other members of Rubiaceae stipules are present but the nodal condition is unilacunar. It is interesting to note that in Hibiscus cannabinus the first follar node is unilacuriar and stipules are not differentiated, while subsequent foliar nodes show stipules, and the foliar node becomes trilacunar. Is the developmental difference in nodal pattern related to the development of stipules in Hibiscus cannabinus? Singh (1972) has noted that in species of Euphorbiaceae with trilacunar nodes, the lateral traces may supply the stipules before entering the leaf base or are entirely consumed in supplying to the stipules or may disappear soon after differentiation.

### 12.13 PhYLLOTAXY and trace relations

The arrangenent of leaves on the axis in Hibiscus cannabinus shows a $2 / 5$ phyllotactic pattern according to the genetic spiral and the angular divergence of the leaves. Here, two parastichies winding in opposite directions are recognized. The parastichies increase along with the increase in growth. The spiral arrangement is constant from the first leaf onwards, whereas the cotyledons are placed opposite. According to Esau (1965) the leaf production starts along two parastichies beginning
with cotyledons and the number of helices increases as the plant grows. The leaf arrangement in seedlings as well as old plants of Hibiscus cannabinus maintain regular pattern. This is in support of O'Niell's (1961) view that a tangential spread of leaf traces at higher levels of the shoot would make possible an increase in diameter of the vascular cylinder without a change in phyllotaxis.

In Hibiscus cannabinus the median trace of any leaf is related to two lateral traces, an anadromic lateral which is three plastochrons apart and katadromic lateral which is two plastochrons apart. Such a relation is of general occurrence in species with $2 / 5$ phyllotaxy and is also reported by Trivedi (1969) in Prosopis juliflora having $2 / 5$ leaf arrangement.

The arrangement of leaf traces varies in different groups of plants and is related to phyllotaxis (Ezelarab and Dormer, 1963; Dormer, 1954). Dormer (1954) has reported two types of vascular systems in shoots, the open type where the leaf traces form sympodia that are independent of one another and the closed type where the leaf traces interconnect the sympodia. In Hibiscus cannabinus the vascular system is of the open type, with an early development af secondary growth from the vascular cambium. Dormer (1945) suggests that the tangential continuity in open systems is accomplished in plants in which secondary growth from the vascular cambium comances early.

### 12.14 PETIOLE

The size and number of strands change at different levels in the petiole of Hibiscus cannabinus. At the base it is nine, in the middle five and towards the upper region it forms a closed ring. According to Metcalfe and Chalk (1950) the number of strands wherever discrete varies in different species but there are of ten about six strands. While in other members such as Abutilon, Althasa, Malva, Pavonia, Sida and Sidalcea in the distal end of the petiole, isolated collateral strands arranged in a circle were $f$ ound but the $x y l e m$ forms a closed ring in Anoda, Hibiscus, Lavatera and Sphaeralcea (Metcalfe and Chalk, 1950). The vascular system in transverse section through the middle region of the petiole of Gossypium (Metcalfe and Chalk, 1950) is in the form of closed ring, an arc or a circle of isolated strands.

### 12.15 LEAF HISTOLOGY

A transverse section of leaf in Hibiscus cannabinus shows upper and lower epidermis and stomata scattered on both the surfaces. Ranunculaceous stomata is a common feature in Malvaceae. The mesophyll can be distinguished into palisade and spongy layers in H1biscus cannabinus. Palisade tissue is single layered while $5-6$ layers of spongy tissue have been observed in this plant. Hibiscus cannabinus from Java has palisade
on both sides, but palisade tissue was observed only towards the upper side of leaves of the same species from Persia (Metcalfe and Chalk, 1950).

## 12. 16 LEAF VENATION

The venation of Hibiscus cannabinus belongs to the 'Craspedodromous type' of Ettinghausen (1861) wherein the secondaries follow a direct course to the margin terminating at lobes or in the marginal teeth. Pray (1954) divided venation pattern of dicotyledons into two categories: major and minor venation. Major venation comprises primary, secondary, tertiary and intermediate veins. In Hibiscus cannabinus a single primary vein runs through the lamina. The primary vein branches into three to five secondaries depending on the number of lobes in the leaf. According to Pray (1954) tertiaries are of two types: (1) Those connecting adjacent secondaries more or less distinctly, and (2) those producing a series of loops along the margin. Both types of tertiaries were observed in Hibiscus cannabinus. The minor venation includes quaternary and their branches. In Hibiscus cannabinus quaternaries constitute the areoles. Minor venation forms polygonal and rectangular areoles. According to Pray (1959) minor venation with polygonal areoles is a basic type from which other types could be derived.

Hibiscus cannabinus possesses one large nectary at the base of the midrib of leaf. These are present on the lower side of the midrib or three principal veins in certain species of Hibiscus, Ingenhouzia, Fulostylis, Kydia, Gossypium, Thespesia, Urena, etc. (Metcalfe and Chalk, 1950). The nectary in Hibiscus cannabinus is ifned with secretory trichomes situated in the crypt. The crypt opens on the abaxial side of the leaf. Metcalfe and Chalk (1950) have reported groups of secretory trichomes, situated either on the surface or in concavities or depressions. The nectaries are situated close to the vascular tissues. Frei (1955) has examined 160 species of dicotyledons and found the presence of phloem elements in nectaries of some plants, phloem and xylem elements in some species and absence of vascular elements in the rest.

### 12.17 TRICHOMES

In Hibiscus cannabinus eight different types of trichones have been observed viz., simple unicellular, simple bicellular, capitate uniseriate glandular, cylindrical glandular, fusiform glandular and capitate glandular trichomes. According to Metcalfe and Chalk (1950) eight types of trichomes cccur in Malvaceae viz., stellate, tufted, peltate, bristles, multicellular, capitate glandular, smaller glandular and x-shaped hairs. In genus Hibiscus (Metcalfe and Chalk, 1950) four types of trichomes viz., peltate, multicellular glandular,
capitate glandular and smaller glandular have been recorded. In Hibiscus cannabinus all the above types except the peltate have been recorded.

All trichomes develop from a single epiermal initial. This agrees with the conclusion drawn by Carlquist (1959a, b), Ramayya (1972) and Jain and Singh (1973). Ontogenetically four different modes of development of trichomes were observed:

1. The epidermal initial dividing periclinally a common mode found in glandular and non-glandular trichomes.
2. The initial undergoes anticlinal division. This occurs in non-glandular trichomes only.

The (1) and (2) modes of development were reported earlier by Netolitzky (1932) and Uphof (1962).
(3) First two or more divisions are anticlinal. This occurs in non-glandular trichomes. Similar develop ment has been reported by Ramayya (1969) in the trichomes of Compositae.
(4) The unicellular non-glandular trichome that develops from an epidermal initial without undergoing any division. Jain and Singh (1973) have reported such trichomes in Dombeya natalensis.

The next division during development in all the trichames occurs in the apical cell. The foot cell does not divide further except in bicellular trichome where
occasionally anticlinal divisions were observed. Ramayya (1972) has reported anticlinal divisions in the foot cell of the trichome of Boerhaavia diffuse.

The initials of non-glandular and glandular trichome were distinguished by their pointed and rounded papilla formed by the first division of the epidermal cell respectively (Uphof, 1962, and Sahasrabudhe and Stacte, 1974). But in Hibiscus cannabinus the non-glandular trichomes also have rounded papilla so that there exists no such distinction. A similar situation has been recorded in trichome of Compositae (Ramayya, 1969) and Dombeya natalensis (Jain and Singh, 1973).

The single epidermal initial in Hibiscus cannabinus gives rise to the head, stalk and foot of the glandular and non-glandular trichomes. In the nonglandular trichome viz., the bicellular, unicellular and tufted trichomes, the hypoderma cells and neighbouring epidermal cells divide meristemazically forming a cushite on below to lift the trichome. Similar observation has been recorded by Sahasrabudhe and Stace (1974).
12.18 FLORAL VASCULATURE

Flower of hibiscus cannabinus is typically tetracyclic. The vasculature of the flower is complicated because of cohesion of stamens and adanation of corolla and staminal tube. The pedicel has eight strands which become organized into two rings. The outer ring is
composed of continuous vascular tissue and the inner ring comes to possess five discrete strands. The outer ring comprises the medians and laterals of the bracteoles, sepal medians and the carpellary traces. The strands of the inner ring go as sepal laterals, petal traces, androecium and the carpellary traces. Thus there are two sets of compound traces "Bt Sm Ct" and "Sl P st Ct".

Hibiscus cannabinus does not agree in many floral characters, anatomically and mor phologically, with the other members of Malvaceae investigated. In flowers of Malvaviscus arboreus, Hibiscus solandra, Hibiscus micranthus and Abutilon (Rao, 1952), the pattern of vascular supply to the floral parts is in contrast to the vasculature of Hibiscus cannabinus. In these plants the traces for all the parts of the flower originate independently from the central stele except for the conjoint petal-stamen traces. Wilson (1937), studied the vascular supply of the floral parts in Hibiscus tiliaceus. In this plant the origin of bracteole and sepal median traces are completely independent whereas the traces that go as sepal laterals, petal traces and the stamens are conjoint. No mention has been made of carpellary traces.

The medians and laterals of the bracteoles are given off independently from the outer ring of vascular tissue "Bt $\mathrm{sm} C t$ " in Hibiscus cannabinus whereas Rao (1952) observed in Malvaviscus arboreus, Hibiscus micranthus and Hibiscus solandra that a single trace divides to form a
a median and the two laterals of each bracteole. In Hibiscus cannabinus after the traces for bracteoles are given of $f$, the carpellary traces are organized from the two compound traces 'Bt Sm Ct' and 'Sl P St Ct'. Only after the organization of all carpellary traces viz., the dorsals, marginals and ventrals, depart, the traces for sepals at higher levels. Further the compound traces 'Bt $S m C t$ ' and 'Sl $P$ St $C t$ ' separate in their component parts.

The sepal medians arise conjointly with carpellary traces. Once the carpellary traces separate, the remaining strands directly enter as medians of sepals. The five laterals of sepals arise conjointly with petal-stamen traces. Each lateral separates and comes to lie in between the two medians so that in all there are ten strands. In view of their position and behaviour the laterals of adjacent sepals remain confluent until the extent of fusion of sepals. As the sepals become free the confluent laterals divide into two each and run as separate laterals for each sepal thus each sepal has a median and two laterals. This clearly indicates that the sepals are fused below and free above. In Hibiscus solandra and Hibiscus micranthus (Rao, 1952) the sepal traces are next to be given off after the bracteoles. Five traces enter the sepals and each divides into a median and two laterals indicating the free condition from below. In Hibiscus tiliaceus (Wilsan, 1937) 5 sepal medians come from the central stele and the 5 laterals from the conjoint petal
stamen trace so that the sepals have 10 traces in all but the separation of laterals has not been mentioned indicating complete fusion of sepals.

Rao (1952) reported origin of the conjoint petalstamen traces to be characteristic feature and remarkably constant in the whole of Malvales. The same has been noticed in Hibiscus cannabinus. The petal traces are given off after the laterals of sepals depart leaving behind the staminal traces. The ten staminal traces left behind after the separation of the petal traces become equally spaced and get consumed in the formation of staminal strands. The repeated chorosis of the staminal traces results in the great congestion of the stamens with the result on the staminal tube anthers emerge at different levels. Wilson (1937) and Rao (1952) have noticed this in other species of Hibiscus.

The gynoecium is pentacarpellary, pentalocular with axile placentation. The vascular supply comprises 15 strands - five dorsals, five marginals and five inverted ventrals. Each of the ventrals divide into two to supply the either half of the placenta. Dorsals are not recognizable as distinct as other carpel wall bundles. The dorsals fade of $f$ and the marginals extend to the style. This indicates that the dorsals are derived from the marginals. The marginals continue their course in the style. The inverse orientation of ventrals is characteristic of axile placentation (Puri, 1952). Such
orientation of placental bundles has been observed in Hibiscus cannabinus. However, Rao (1952) has not mentioned about the inverse orientation of the ventrals in the members of Malvaceae that he studied.

The level of separation is correlated with the degree of hypogyny. Since the bases of the sepals, petals and stamens are adnate to the ovary wall to some length, the ovary is to some degree inferior. Anatomically too, it indicates partly hypogynous nature due to the separation of carpellary traces before sepal-petal and stamen traces. Based on this point, the author suggests that ovary here is not hypogynous. However, extensive investigation of the carpellary traces in Malvaceae would be necessary for a decisive conclusion.
12.19 MORPHOLOGY OF BARK

The tender bark of Hibiscus cannabinus is smooth with hairs and contains, only the primary phloem tissues. As growth proceeds the phloem is delimited from the cortex by protophloic fibres which form strands alternating with parenchyma. Initiation of cambial activity results in the formation of fair amount of secondary phloem tissues. Flbres also differentiate in the secondary phloem.

The older bark has black openings and becomes rough due to development of lenticels. Superficial
periderm is formed . Layers of secondary phloem fibres are formed. The fibre bands alternate radially with soft phloem regions containing sieve tubes, companion cells and parenchyma cells. The fibres and the soft phloem form the axial system. As the stem expands circumferentially the ray cells undergo dilatation growth by both radial divisions and tangential enlargement so that it resmlts in alternating wedges of axial phloem, wide near the cambium and narrow near the cortex and wedges of ray are narrow near the cambium and wide near the cortex. Esau (1964) finds that the secondary phloem may lack fibres in some species, in others it is present and distributed irregularly or in regular tangential bands, while some have scleretds alone or fibres and sclereids. Presence or absence of fibres and the pattern of distribution when it is present, however, impart special characteristic to the secondary phloem (Zahur, 1959).
12.20 SIEVE TUBES

In Hibiscus cannabinus the sieve tubes have end walls that are transverse to very inclined. The average angle of inclination of the end walls is $37^{\circ}$. According to Cheadle and Whitford (1941) and Cheadle (1948) the transverse to slightly oblique end walls are highly specialized; very oblique end walls primitive. Ashok Kumar (1969) in his studies on the bark anatomy of

Leguminous species has considered in Acacias where the end walls are at an angle of 33.5 to 45.4 degrees as very inclined. In $\underline{H}$. cannabinus the end walls are very inclined.

In Hibiscus cannabinus the transverse to inclined end walls show regular as well as irregular arrangements of the sieve tubes and so that there is no correlation between the transverse and very oblique end walls and the arrangement of sieve tubes. Cheadle and Whitford (1941) and Cheadle (1948) have considered the sieve tube member with transverse to slightly oblique end walls to be associated with regular arrangement than are the oblique end walls. Very oblique end walls are nearly with an irregular arrangement of the sieve tube elements. Regular arrangement of sieve tubes is highly specialized and advanced and irregular the primitive.

Chalk's (1978) classification of vessels based on tangential diameter was used by Ashok Kumar (1969) for sieve tube classification. The classification is as follows:

1. Small

$$
\begin{array}{lr}
\text { 1.1 Extremely small } & \text { upto } 25 \mu \\
\text { 1.2 Very small } & 25-50 \mu \\
\text { 1.3 Moderately small } & 50-100 \mu
\end{array}
$$

2. Medium

100-200 $\mu$

| 3. Large | 3.1 Moderately large | $200-300 \mu$ |
| :--- | :--- | :--- |
|  | 3.2 Very large | $300-400 \mu$ |
|  | 3.3 Extremely large | over $400 \mu$ |

The sieve tubes in $H_{\text {. }}$ cannabinus are small; either very small (8.2\%) or moderately small (91.8\%).

Adopting Metcalfe and Chalk's (1950) classification of vessel length:

| 1. Short | upto $350 \mu$ |
| :--- | ---: |
| 2. Medium | $350-800 \mu$ |
| 3. Long | above $800 \mu$ |

The sieve tubes are either short or medium in Hibiscus cannabinus.

### 12.21 PHLOFM RAY S

The vascular rays in the secondary phloem follow a pattern similar to that of the wood. Kribs (1935) and Metcalfe and Chalk (1950) have classified the wood rays broadly into two types which is classified further into sub-divisions based on dominant type of rays:

## e

1. Homogenfous (composed of procumbent cells only)

Type I: Entirely uniseriate
Type II: Rays all not uniseriate
2. Heterogenfous (composed of procumbent and upright cells)

Type I: Rays entirely uniseriate; procumbent and upright cells which are taller than broad, more or less alternating with each other by irregularly arranged.

Type IIA: Upright cells taller then broad, confined to the margins and the uniseriate portions.

Type IIB: Uniseriate rays composed of alterrating upright and procumbent cells; multiseriate rays with upright cells confined to the terminal uniseriate portions.

Type III: Uniseriate of two sorts: composed of either upright or procumbent cells. Multiseriate rays composed of square upright cells.

Hibiscus cannabinus shows uni-, bi- and multiseriate phloem rays belonging to type IIA since it consists both procumbent and upright cells and so they are hetero genfous. The biseriate rays are the dominant ones. In biseriate rays the procumbent cells are relatively few and the uniseriate cells are composed entirely of upright cells. The multiseriate rays are $3-5$ cells wide with more of procumbent cells and the upright cells at the terminals and in between the procumbent cells. Chalk (1938) suggested a classification of rays on the basis of ray width which has been accepted by the International Associa. lion of Anatomists (1939). According to this, the different categories are:

| 1. Extremely fine | $15 \mu$ |
| :--- | ---: |
| 2. Very fine | $15-25 \mu$ |
| 3. Moderately fine | $25-50 \mu$ |
| 4. Medium | $50-100 \mu$ |
| 5. Moderately broad | $100-200 \mu$ |
| 6. Very broad | $200-400 \mu$ |
| 7. Extremely broad | above $400 \mu$ |

According to this classification and taking into consideration width of the phloem rays in Hibiscus cannabinus can be classified as the uniseriate rays are very fine or moderately fine. The biseriate rays belong to moderately fine or medium category and the multiseriate rays are medium or moderately broad. Metcalfe and Chalk (1950) have mentioned that the frequency of rays is a useful character. In Hibiscus cannabinus the average number of rays per mm is $10-18$.
12.22 EXPAN SION TISSUE

The stress and strains put on the bark as a result of secondary growth is partly accommodated by the develop ment of phellogen. The tissue outside the vascular cambium shows secondary growth which is termed as 'diffuse secondary growth' by Tomlinson (1961) and as 'dilation growth' by Schneider (1955), Chattaway (1955) and Esau (1965), keep pace with growth in diameter. On the other hand Whitmore (1962) has differentiated dilation growth in Dipterocarpaceae into (1) phloem proliferation, (2) phloem expansion. Ashok Kumar (1969) differentiated dilation growth into cortical expansion, pericycle expansion, ray expansion and phloem proliferation. Dilation growth in Hibiscus cannabinus is effected by cortical and ray expansion. Tangential stretching and anticlinal divisions take place in most of the cells of the cortex. The expansion of rays is brought about by the division of the body cells expanding into wedges.

Periderm consists typically of three parts: the phellogen which is the initiating layer, the tissues producing inwards the phelloderm and outside the phellem. The phellogen is the secondary meristematic tissue (Fahn, 1967). The phellogen may be initiated at different depths outside the vascular camblum (Esau, 1948; Metcalfe and Chalk, 1950, and Whitmore, 1962). In the epidermis (Nerium oleander, Quercus Suber, Solanum dúlcemara); in subepidermal layer (Populus, Juglans, Ulmus); in the second or third cortical layers (Aristolochia, Robinia) and near the phloem or from within the phloem parenchyma itself (Arbutus, Camellia, Punica) and development from the subepidermal layer is most common (Eames and McDaniels, 1947; Esau, 1965; Fahn, 1967).

In Hibiscus cannabinus the phellogen arises in the epidermal layer which forms the 8 to 10 layered cork cells. In all the members of Malvaceae the phellogen arises in the epidermal layers that form: the cork (Metcalfe and Chalk, 1950). According to Lier (1955) the meristematic activity initiating the periderm begins either in localized regions or around the entire circumference. In Hibiscus cannabinus initially the periderm is locallzed forming lenticels and later the divisions are continuous around the stem circumference forming files of cork cells and with scattered lenticels. The first division results in the formation of a cork cell and a phellogen cell which by successive
periclinal divisions gives rise to the radial rows of cork cells. A similar situation has been observed in Pyrus (Esau, 1965).
12.24 ORIGIN AND DEVELOPMENT OF FIBRES

Bast fibres have received considerable attention, for their interpretation involves the problematic concept of pericycle (Blyth, 1958; Esau, 1939, 1943a, 1950). Morot (1885) has frequently found fibres in the secondary phloem but that the primary phioem has ordinarily a sof $t$ tissue. Later Leger ( $1895,1897 \mathrm{a}, \mathrm{b})$ distinguished the first sieve elements in shoot and followed their obliteration and the development of the remaining cells into fibres. But he applied the term pericycle to species in Caryophyllaceae, Cucurbitaceae, Liliaceae and others in which the fibres were separated from the outermost phloem and appeared to arise independently.

The phloic origin of the so-called pericycle was suggested by many authors. Baranetzky (1900) has observed fibre strands on the periphery of the vascular region in Nerium oleander containirig phloem cells, which later got crushed. Gaucher (1902), reported in Euphorbiaceae that the fibres of pericycle originated in the phloem and the pericycle was a useful term to designate a particular location in the stem. In Linum perenne, Vitis vinifera, and Tilia sp. (Esau, 1943b, 1948, 1965) developmental studies on the primary phloem showed
that the cells differentiating in association with first sieve elements elongated considerably, developed secondary walls and matured as fibres after the sieve tubes and their companion cells were destroyed. Similar relation was reported by Kundu $(1941,1942,1954$ ) in Crotolaria, Hibiscus, Cannabis, Corchorus and Linum, and Kundu and Sen (1961) in Boehmeria nivea, the plants which serve as sources of commercial fibres. Serbanescu-Jitariu and Mitroiu (1961) misinterpreted protophloem fibres as pericyclic fibres in Althea cannabina, Hibiscus cannabinus and Abutilon theophrasti and consequently did not recognize the differences between the primary and secondary phloem fibres. Metcalfe and Chalk (1950) have acknowledged the concept of pericycle in distribution of phloem fibres as unsolved but found it convenient to cantinue the use of the term.

In Hibiscus cannabinus the fibres are common in the primary phloem, specifically in the protophloem. These outermost fibres develop from the elements of the procambium in the protophloem after the sieve tubes and companion cells are obliterated by the expansion and elongation of the surrounding cells. They become several times longer than the neighbouring cells. Thus the first formed fibres are protophloic.

The fibre primordia elongate without undergoing divisions and attain a considerable length before they
mature (Aldaba, 1927). In addition, they commonly undergo intrusive growth (Kundu, 1942, 1954; Kundu and Sen, 1961). In Hibiscus cannabinus also the fibre cells elongate considerably and undergo intrusive growth due to which the fibre ends are sometimes forked and differently shaped depending upon their spatial arrangement associated with the cells as was also suggested by Esau (1965).

### 12.25 SECONDARY FIBRE DEVELOPMENT

In some species protophloic fibres are the anly fibres produced as in Linum. In others, however, fibres appear in later formed tissue, usually the secandary phloem. Metaphloem is typically devoid of fibres (Esau, 1969). Kundu (1954) has stated that in the case of bast fibres like Corchorus, Cannabis, $H$. cannabinus, and $H$. sabdariffa, the secondary fibres are formed by the activity of the cambium. Kundu and Sen (1961) have also reported in Boehmeria nivea that the secondary fibre cells originate in the secondary phloem tissue and are shorter than the primary fibres. According to Kundu (1954) as a result of cambial activity and formation of secondary rays the fibre cylinder towards the base of the stem, becomes more compact and meshy. This kind of meshiness reduces the quality of the fibre and weakens the fibres as they are split apart by combing operations during spinning.

In Hibiscus cannabinus the secondary fibres are formed by the cambial activity in the phloem region. The fibre bundles form a network. Meshiness is more at postflowering stage and less at pre-flowering and flowering stages.
12.26 CELL WALL OF FIBRE

The secondary wall formation in relation to cell elongation wes studied in detail in primary phloeni fibres by Aldaba (1927) and Anderson (1927). These cells undergo intrusive growth at their tips while secondary thickening is deposited in their middle regions (Esau, 1965). The secondary wall does not appear to be cemented to the primary, for in sections the wall gets detached from the primary (Aldaba, 1927; Anderson, 1927; Kundu, 1942). In Hibiscus cannabinus the fibre cell elongates to some extent when it remains delicate and thin and later secondary and tertiary wall formation begins which is limited to the middle regions and the fibre ends are hyaline. The secondary walls get detached from the primary and get infolded and wrinkled in some of the fibre sections.

The sec ondary wall varies in thickness. In Linum usitatissimum the secondary thickening occupies $90 \%$ of the area of the cell in cross section (Esau, 1969). Jute has a maximum cross section of $18.6 \mu$ and minimum of $12.3 \mu$ and a lumen with maximum $4.58 \mu$ and minimum of $2.05 \mu$ (Mathews, 1948). In Hibiscus cannabinus the cross section
at three different stages shows that the maximum ( $21 \boldsymbol{\mu}$ ) and minimum ( 12 to $15 \mu^{2}$ ) values are more than jute. The average cross-section in pre-flowering, flowering and post-flowering stages has a difference of $0.2 \mu$ and is highest at pre-flowering stage ( $17.0 \mu$ ) (Ref. Tables 1, 2, 3).

The lumen of the fibre cell in Hibiscus cannabinus at pre-flowering stage is maximum $12 \mu$ and minimum $6 \mu$. At flowering and post-flowering stages maximum is $9 \mu$ and minimum is 3 which is two times greater than in Jute. There is a decrease in the size of lumen in Hibiscus cannabinus from re-flowering to post-flowering stage indicating that the wall gradually thickens as the fibre matures with the result the lumen becomes smaller.

### 12.27 FIBRE CHARACTERS

According to Kundu (1954) the characters like greater bundle size, larger number of ultimate fibre cells in a fibre bundle, and greater length of ultimate fibres improve the quality of the fibre. In $\underline{H}$. cannabinus and H. sabdariffa it has been found that greater the number of ultimate fibres in the fibre strand, higher is the quality. The number of ultimate fibres in Corchorus, Cannabis, $\underline{H}$. sabdariffa and $\underline{H}$. cannabinus may vary from 6 to 60 depending on the variety, seas on and cultural conditions (Kundu, 1954). In Hibiscus vitifolius the number of fibre cells in a bundle varies greatly ranging
from 4 to 40 (Maiti, 1969). In Hibiscus cannabinus the study at three different stages shows that the number of fibre cells increases and is maximum at post-flowering stage 20-60. At pre-flowering stage the number of cells is apparently more because of the blocks that are fused tangentially (blocks are not discrete). In this respect Hibiscus cannabinus is better than Hibiscus vitifolius.

The number of bundles at the basal portion of fibre wedges is 8 to 20 in Hibiscus vitifolius and 8 to 24 in jute (Maiti, 1969) whereas in Hibiscus cannabinus it is 8 to 14 and is highest at post-flowering stage. The distance between two adjacent layers is greater in H. cannabinus as compared with jute and $\underline{H}$. vitifollus as shown by Maiti (1969). Varieties having bigger fibre blocks with larger number of ultimate fibres and arranged in a compact manner are known to yield better quality fibre in flax and in jute (Kundu, 1954). As compared to jute, Hibiscus cannabinus shows bigger blocks and larger number of ultimate fibres which are compact.

Fibres are usually long cells. Length of fibres in some species like Linum usitatissimum is $9000-70000 \pi$, Cannabis sativa 5000-55000 $\mu$, Corchorus capsularis 800-6000 $\mu$ and Boehmeria nivea 50000-250000 $\mu$ (Esau, 1965). According to Mathews (1948) jute fibres are 1270-4826 $\mu$ in length and 20 to $25 \mu$ in diameter. Dutta and sen (1736) studied the fibres of H . cannabinus and $\underline{H}$. sabdariffa by
cutting plants in three equal zones on top, middle and base and kept in petri dishes containing pond water for retting. The average maximum lengths of $257.62 \mu$ and 278.75 were observed in the middle regions of the stem of $\underline{H}$. cannabinus and $\underline{H}$. sabdariffa respectively. According to Mathews (1948) Hibiscus cannabinus has fibre length ranging from 1981.2 to $6096 \mu$ and the diameter from 14 to $33 \mu$. The above readings are shown in Table 14 for comparison.

Table 14: Comparative study of length of different fibres

| Plant | Length <br> (microns) | Author |
| :--- | :---: | :--- |
| Linum usitatissimum | $9000-70000$ | Esau (1965) |
| Cannabis sativa | $5000-55000$ | Esau (1965) |
| Corchorus capsularis | $800-6000$ | Esau (1965) |
| Boehmeria nivea | $50000-250000$ | Esau (1965) |
| H1biscus cannabinus | 257.62 | Dutta and Sen |
| Hibiscus sabdariffa | 278.75 | Dutta and sen |
| (1967) |  |  |
| H. cannabinus | $1981.2-6096$ | Mathews (1948) |
| Corchorus capsularis | $1270-4826$ | Mathews (1948) |

The following table (Table 15) shows a comparative account of the length and diameter of Hibiscus cannabinus and Hibiscus vitifolius (Maiti, 1969).

Table 15: Dimensions of ultimate fibre
$\frac{H}{\text { (present }} \frac{\text { cannabinus }}{\text { investigation) }}$$\quad \stackrel{\text { Hitifolius }}{\text { (Maiti, 1969) }}$

Length:

| Minimum | $7800 \mu$ | $645.00 \mu$ |
| :--- | ---: | ---: |
| Maximum | $14060 \mu$ | $6665.00 \mu$ |

Diameter:

| Minimum | $15 \mu$ | $6.86 \mu$ |
| :--- | :--- | ---: |
| Maximum | $30 \mu$ | $20.52 \mu$ |

The above observations show that the length of the fibres of $H$. cannabinus is almost two times greater than H. Vitifolius and the diameter is also greater than that of $H$. Vitifolius. The length of the $f i b r e s$ of $H$. cannabinus as reported by Mathews (1948) and Dutta and $\operatorname{Sen}(1967$ ) is many times smaller than the present investigation. It is, however, not known at what stage the fibre length was studied by the above authors.
12.28 RETTING

Retting in Hibiscus cannabinus at three different stages of flowering typically signifies the pace at which the process takes place. The fibre retting at preflowering stage took 7-8 days, at flowering stage 10-11 days and at post-flowering stage 12-14 days. This is because percentage of lignin is less at pre-flowering
stage and there is no formation of periderm, so that the decomposition by bacterial action is quicker. During flowering stage there is a gradual increase in the percentage of lignin and interrupted periderm and lenticels are formed due to which the retting takes 2 to 3 days more than the pre-flowering stage. At the post-flowering stage when the plants attain the maximum height there is maximum percentage of lignin with the fibrous layer encapped by a thick periderm of 6-10 layers of cork cells and secondary cortex. This periderm is frequently interrupted by lenticels. These need longer time to ret with the result retting period is more by $3-4$ days. Retting in tap water took a longer duration and in pond water the reaction was quicker having a difference of two days. Retting at all the three stages was carried out without any difficulty. Aldaba (1927) has suggested that degree of lignification influences the retting to a marked extent. Heavily lignified fibres do nct undergo aecomposition as easily as unlignified fibres.

According to Crane and Acuna (1945) kenaf (H. cannabinus) should be harvested during the flowering stage in order to obtain best yield and the bast separation of the fibre from the stem. Retting of the fibre proved to be more rapid when the ribbons were stripped of $f$ with a wooden mallet and retted in slowly moving water in $\underline{H}$. cannabinus (Watkins, 1946). According to Kundu (1954) compact fibres have strength and quality, and also
facilitate retting and extraction. In Hibiscus cannabinus the fibre bundles are bigger and more compact than they are in jute and flax. Maiti (1969) has considered the flowering stage as the proper period for retting in Hibiscus vitifolius and the period of retting was 4 to 7 days depending on the temperature of the water. The time taken for retting $\underline{H}$. vitifolius in pond water was 6 days and in tap water 7 d\&ys. In Hibiscus cannabinus retting was possible at all stages and the time taken in retting in pond water was $2-3$ days lesser than in tap water.
12.29 CHEMICAL PROPERTIES
12.29.1 Chemical Composition

The chemical composition of the fibres at three different stages of flowering in Hibiscus cannabinus (Ref. Tables 8, 9, 10) gave an idea of the increase and decrease in the percentage of different chemical constituents. of the six chemical constituents detected, percentage of cellulose was maximum at pre-flowering stage viz. $81.80 \%$, at flowering stage $80.36 \%$ and at post-flowering stage $76.40 \%$ showing a gradual decrease in percentage. Lignin is next highest in percentage about $8.59 \%$ at pre-flowering, $10.90 \%$ at flowering and $14.51 \%$ at post-flowering stages. Thus there is a gradual increase in lignin quantity. The cellulose here is known as lignocellulose, which is a compound of cellulose with lignin. Ash content shows a total increase of about $0.40 \%$ from pre- to post-flowering
stage, whereas moisture shows a total decrease of about 1.0\% from pre- to post-flowering stages. Fats and pectin shows a total increase of about $0.03 \%$ and $0.24 \%$ respectively from pre- to post-flowering stage.

The above observations show that at all stages cellulose and lignin are maximum making the bulk of the fibres.

Chemical composition of some of the bast fibres also known as "soft fibres" as reported by Mathews (1948) is given below.

Table 16: Comparative study of chemical properties

| Chemical <br> constituents | Flax | Jute | Hemp | Ramie | Sida | $\frac{\text { Hibiscus }}{\text { cannabinus }}$ <br> (present <br> investigation) |
| :--- | :---: | :---: | :---: | :---: | :---: | :---: |
| Ash | 1.32 | 0.68 | 0.82 | 5.63 | 0.91 | 0.91 |
| Moisture | 10.70 | 9.93 | 8.88 | 10.15 | 9.55 | 6.28 |
| Aqueous <br> extract | 6.02 | 1.03 | 3.48 | 10.34 |  |  |
| Fat | 2.37 | 0.39 | 0.56 | 0.59 | 0.85 | 0.96 |
| Cellulose | 71.50 | 64.24 | 77.77 | 66.22 | 70.24 | 80.36 |
| Lignin and | 9.41 | 24.41 | 9.31 | 12.70 | 1.00 | 10.90 |
| Pectin |  |  |  |  | 14.79 | 1.34 |

Mathews (1948) has not reported the stage at which the values presented above in Table 16 for flax, jute, hemp,
ramie and sida have been studied. The values shown for Hibiscus cannabinus are at the flowering stage.

Except for flax all others show lesser percentage of fat than $H$. cannabinus which is $0.96 \%$ at flowering stage. H. cannabinus shows a high percentage of cellulose i.e. $80.36 \%$ and $10.90 \%$ of lignin. According to Das, Mitra and Wareham (1950) fibres of Hibiscus cannabinus and Hibiscus sabdariffa are the stiffest and contain highest amount of fat and lowest amount of lignin. According to these authors high hemicellulase is responsible for the stiffness of these fibres. Maiti (1969) has reported that the fibres of Hibiscus vitifolius are more lignified than that of jute. In the present investigation hemicellulose content was not estimated. However, looking at the fat and lignin percentage the fibres of $\underline{H}$. cannabinus appear to be soft.
12.29.2 Colour Reactions

Lignification in $f l a x$ begins after flowering and is not general in all fibres in a cross-section of stem but occurs in isolated groups or individuals (Aldaba, 1927). According to Harris (1954) in Linum, cannabis and Boehmeria the secondary wall contains 75 to 90 per cent cellulose and little or no lignin. In H. cannabinus lignification was observed from the early stages of fibre development that is in an eight day old seedling. The presence of lignin was gradual from the tip to the base
of the stem. Similarly the first formed fibres in transection were lignified and the inner later formed fibres showed gradual decrease in lignification.

The reactions to lignin and cellulose show that it behaves similar to that of jute (Mathews, 1948) indicating the presence of lignocellulose. Maiti (1969) found that Hibiscus vitifolius on reaction with aniline sulphate and phloroglucinol show that, like jute, the fibre contains a considerable amount of lignin and cellulose.
12.29.3 Dyeing

Mathews (1948) has recorded that the chief difference between jute and the pure cellulose fibres is in the ability of jute to combine with basic dyes. H. vitifolius gave similar reaction to dyes (Maiti, 1969). Similarly Hibiscus cannabinus has a great affinity for basic dyes.
12.29.4 Bleaching

Hibiscus cannabinus has a natural lustre which is greater at the preflowering and flowering stages and so does not require longer period for bleaching. Jute on the other hand is more sensitive to the action of chemicals and being more yellow cannot be bleached, as treatment with alkalies weakens the fibre (Mathews, 1948).

Table 17: Comparative table indicating the qualities and utilization of fibres at three stages of flowering

12.30 PHY SICAL PROPERTIES

According to Kundu (1956) quality of fibre is assessed on the basis of length, strength, colour, lustre, proportion of faults and defects, general qualities as fineness, stiffness and hardness.

Maiti (1969) has reported in Hibiscus vitifolius that the most desirable features of fibres for spinning good yarn are: 1. lower cross-sectional area, 2. great elasticity, 3. cleanliness, 4. strength, 5. fineness, 6. softness, 7. absence of specks. Considering the above features the fibres of Hibiscus cannabinus at pre-flowering stage are cleanest, soft, show lower cross-sectional area, and fineness. At flowering stage fibres show greater elasticity, clean, soft, absence of specks and fineness. Therefore, the fibre quality is at its best at the flowering stage for spinning (Table 17).

It is betterttian jute and almost similar to that of Hibiscus vitifolius according to the Table 18 given below:

Table 18: Comparative study of some physical characters

Characters Haiti (1969) \begin{tabular}{c}
Jute <br>
Haiti (1969)

$\underset{$

vitifolius <br>
$\text { Tigation })$

$}{$

H. cannabinus
\end{tabular}$}$

Density
$1.43 \mathrm{gm} / \mathrm{cc}$
$1.39 \mathrm{gm} / \mathrm{cc}$
$1.38 \mathrm{gm} / \mathrm{cc}$
Cross-
sectional area

Young's
Modulus of elasticity
$153 \times 10^{-5} \mathrm{~cm}^{2}$
$168 \times 10^{-5} \mathrm{~cm}^{2}$
$1.25 \times 10^{11}$

1. $25 \times 10^{11}$
dynes/ cm ${ }^{2}$

A comparative study of the fibres of Hibiscus cannabinus at three different stages of flowering in Table 17 shows that the fibre is of very good quality and can be utilized for spinning and easy bleaching at the pre-flowering stage. The main drawbacks of this stage are the yield, reed length and ultimate fibre length are poor compared to other stages and hence uneconomical.

At post-flowering stage though the yield, fibre length and reed length are the best but other qualities except for tenacity show that the fibres cannot be used for spinning but can be used for cordages.

The fibres et the flowering stage possess all the good qualities for spinning. Thus it can be concluded that the best period for harvesting is the flowering stage.

The development of male and female gametophytes is studied. The anther wall is four layered. The subepidermal layer of the anther develops into the fibrous endothecium and the tapetum is of the plasmodial type. The microspore tetrads are tetrahedral and bilateral. Cytokinensis is by furrowing. Pollen grains are spinescent and multiporate and are shed at 2 -male gamete stage. The ovule is bitegmic, crassinucellate and campylotropous. The archesporium is one celled, occasionally two celled archesporium is observed which functions uptil megaspore mother cell stage. Only one functions to form the linear megaspore tetrad and the other degenerates. Embryo sac develops according to Normal-type.

Pollen grains produce pollen tubes in a polysiphonous manner. The endosperm is nuclear to begin with and becomes cellular at later stages. Cell farmation commences at the micropylar end and gradually proceeds to the chalazal end. Seed coat is formed by the inner and outer integuments. Outer integument forms a membranous testa. The outer epidermis of the inner integument forms the palisade layer. Protoplast of these cells degenerate completely. In a mature seed the entire cell is lignified and has fibrous thickening.

Embryo development occurs according to the urtica variation of the Asterad type. The terminal cell of the two celled proembryo forms the cotyledons and stem tip and the basal cell gives rise to the hypocotyl root tip, root cap and suspensor.

The radicular apex is established by late globular stage of embryo. The embryonic root apex shows closed type of organization with four discrete groups of initials for central cylinder, cortex, columella, and a comman initial zone 'dermocalyptrogen' which is concerned with the formation of both the dermatogen and peripherall region of the cap. The root apex of seedlings becomes open as the inftials of central cylinder become continuous with the secondary columella. The shoot apex has single layered tunica, a central mother cell zone, pith rib meristem and flank meristem. The apex is a low dome at maximal stage of plastochron and almost flat at minimal stage.

The root is tetrarch. Along the hypocotyl the four xylem strands divide to form eight strands. The eight strands become arranged in two opposing groups. They enter as traces to the cotyledons. The cotyledonary node is unilacunar, single trace. The first follar node is unilacunar single trace and the subsequent nodes are trilacunar.

Phyllotaxy is spiral 2/5. Each median is related to the laterals of two different levels - katadromic lateral and anadromic lateral. The two laterals of the trilacunar node supply to the stipules.

Gradual variation in the number of vascular bundles is observed in the petiole at different levels. In leaf there is a layer of palisade tissue on the adaxial side and 3-5 layers of spongy tissue below. Venation pattern is 'craspedodromous' type. A single nectary is observed beneath the mid vein. Eight types of trichomes are recognized of which four are glandular and four are nanglandular. Their mode of development is studed.

The pedicel of the flower has eight strands which become organized into two rings. The outer ring of vascular tissue comprises the medians and laterals of the bracteoles, sepal medians and carpellary traces. The strands of the inner ring go as sepal laterals, petal traces, androecium and the carpellary traces. Due to the separation of carpellary traces before sepal, petal and stamen traces, the ovary is to some degree inferior.

The young bark is hairy. As growth advances lenticels develop that form black spots on the bark. The sieve tubes have very inclined end walls. Their diameter is very small or moderately small. They are of short or medium length. The biseriate phloic rays are dominant. Uniseriate and multiseriate rays also occur. Rays belong
to heterogenous type IIA. Dilation growth is effected by ray and cortical expansion. Periderm is superficial. The phellogen arises in the epidermal layer which forms 8-10 layered cork cells.

The origin and development of fibres is studied. The first formed fibres are protophloic. Secondary fibres are formed by the cambial activity. The fibre blocks thus formed alternate with the soft tissue radially and form the phloem wedge i.e. narrow near the periphery and broad near the camblum. The young fibres are thin walled and as the fibre matures secondary and tertiary walls are formed. The fibre cell attains a considerable length and diameter. Maximum length of fibre is at post-flowering stage. Fibre diameter is same at pre-flowering, flowering and post-flowering stages. Retting of fibres is carried out at the three different stages of flowering in pand water and thereafter their chemical and physical properties are studied. Chemically the fibre is mainly composed of lignocellulose. Lignin is detected at an early stage i.e. eight day old seedling. There is a gradual increase in the amount of lignin with the growth of the plant. Fibres have a great affinity for dyes and are easy to bleach. Fibres are soft, clean, have good lustre, greater elasticity and lower cross-sectional area. Fibres at pre-flowering stage have all the best qualities for spinning but the yield, ultimate fibre length and reed length is poor and therefore is uneconomical. At
flowering stage the fibres have all good qualities for spinning. At post-flowering stage they have all poor qualities for spinning but can be used for cordage.

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Figs. 1-13. Developmental stages of microsporogenesis and male gametophyte.

Fig. 1. T.S. of young anther showing outer parietal cell and inner sporogenous cell. Fig. 2. T.S. of anther showing divisions in parietal cells. Fig. 3. L. S. of anther lobe. Fig. 4. T.S. portion of anther, showing meiosis in microspore mother cells and tapetum and other outer layers. Fig. 5. T.S. portion of anther, showing microspores being liberated and mitotic divisions in tapetal cells. Fig.6. Mature pollen grains with abhortive pollen grain. Fig. 7. A single pollen grain with remnants of tapetal protoplast. Figs. 8a-8b. Bilateral and tetrahedral tetrads. Figs.9-12. Pollen grain nucleus showing divisions. Fig. 13. Pollen grain at two male gamete stage.

Figs.14-23. Stages in the development of megasporogenesis and female gametophyte.

Fig. 14. Ovule showing hypodermal archesporal cell. Fig. 15. Formation of primary parietal cell and primary sporogenous cell. Fig. 16. Anticlinal divisions in primary parietal cell. Fig. 17. Ovule at megaspore mother cell stage. Fig. 18: Ovule with two megaspore mother cells. Fig. 19. Megaspore mother cell resulting in the formation of dyad. Fig. 20a. Tetrad of megaspores with degenerating accessory sporogenous cells. Fig. 20b. A tetrad of megaspores with chalazal cell functioning. Figs. 21-22. Two and four nucleate embryo sacs. Fig. 23. Mature embryo sac at eight nucleate stage.

E - epidermis; ML - middle layer; TC - tapetal cell; MiMC - microspore mother cell; M - micro. spore; $T$ - tetrad; $D M$ - degenerating microspore; MMC - megaspore mother cell; MF - functional megaspore; GN - generative nucleus; VN - vegetative nucleus.



Fig. 24. Upper part of the embryo sac showing fertilized egg and free nuclear endosperm.

Fig. 25. Free nuclear endosperm lying compact at the micropylar end at early-globular stage of embryo.

Fig. 26. Endosperm at late-globular stage of embryo showing wall formation at micropylar end and proceeds towards the chalazal end.

Figs. 27A-30B. Structure and development of seed coat at different stages of embryo development.

Fig. 31. T.S. of mature seed showing papillae and fibrous thickening in the palisade-like layer.

Fig. 32. L.S. of upper part of the ovule showing persistent pollen tube at late-globular stage of embryo.

PT - pollen tube; EN - endosperm; PPT persistent pollen tube; $N$ - nucellus; E - embryo; CE - cellular endosperm; FNE - free nuclear endosperm; $M$ - micropyle; OI - outer integument; II - inner integument; $O E$ - outer epidermis; IE inner epidermis; $P$ - palisade-like layer; PTT - pigment tissue; FT - fringe tissue;


Fig. 33. Micropylar part of the ovule showing branching pollen tube and the egg cell.

Fig. 34. Male nucleus lying near the egg nucleus within the egg cell.

Fig. 35. Male and egg nuclei in advanced stage of fusion.

Figs. 36-45. Various stages in the development of embryo.

Fig. 44. Embryo just before heart-shaped stage with histogenic layers.

Fig. 45. Heart-shaped embryo with prominent cotyledonary primordia. Surface and sub-surface layers at the basal end show 'Kappe' divisions.

E - egg cell; EN - egg nucleus;
MN - male nucleus; PT - pollen tube;
N - nucellus; SAI - subapical initial;
D - dermatogen; PER - periblem;
PL - plerome; $C$ - central cell; CI columella, FE -free nuclear endosperm.


Figs. 46-48. Development of root apical meristem. Fig. 46. L.S. late globular stage of
embryo with prominent central
cell and columella initials.

Fig. 47. L.S. heart-shaped stage of embryo with central cell and 'Kappe' divisions in the surface and sub-surface layers.

Fig. 48. L. S. of radicle of late torpedo stage embryo showing cap flanks with 'Kappe' divisions, the linear rows of primary columella and the cortical and central cylinder initials giving rise to the cortex and central cylinder respectively by 'Korper' divisions.

Figs. 49-52. Development of shoot apical meristem.
Fig. 49. L.S. epicotyl apex at the heart-shaped stage of embryo showing concave apex.

Fig. 50. L. S. epicotyl apex at the torpedo stage of embryo showing a slight bulge in the apex.

Fig. 51. L. S. of mature shoot apex at maximal phase of plastochron showing clear cytohistological zones.

Fig. 52. The same at minimal plastochr on phase showing flat apex.
$C$ - central cell; PLI - plerome initial; PBI - periblem initial; CL - columella; DC - derma calyptrogen; EA - epicotyl apex; CMC - central mother cells; T - tunica; FZ - flanking zone;


FIGS. 46:47, 49,50 $\underbrace{50 \mu}$ FIGS. 48,51,52.
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Figs. 53-60. Transection showing the different stages of transition of 4 day old seedling.

Figs. 53-57. Sections from root to the higher level of hypocotyl.

Fig. 58. Just below the cotyledonary node.
Fig. 59. Section of the cotyledonary node.
Fig. 60. Section above the cotyledonary node with the first leaf.

Figs. 61-62. Transections of 8 day old seedling.
Fig. 63. Transection of 12 day old seedling above the cotyledonary node.

Figs. 64-68. Transections of 20 day old seeding from just below to above the cotyledonary node.

$$
\begin{aligned}
& L-\text { Leaf, Cot - Cotyledon, } \\
& M_{1}, L_{1}, L_{3}-\begin{array}{l}
\text { Median and two laterals of } \\
\text { one leaf }
\end{array} \\
& M_{2}, L_{2}, L_{4}-\begin{array}{c}
\text { Median and two laterals of } \\
\text { another leaf. }
\end{array}
\end{aligned}
$$



Fig. 69. Diagrammatic representation of trace relationship.

Fig. 70. Diagrammatic representation of phyllotaxis.

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Figs. 71-76. Transections showing the nature of nodal pattern.

Figs. 71-72. The median and two
lateral traces moving out.
Fig. 73. The traces show division.
Figs. 74-75. The laterals divide to supply the stipules.

Figs. 75-76. Formation of vascular crescent at the leaf base composed of adaxial and abaxial elements.


Figs. 77-79. Serial transection of the petiole from lower region to upper region.

Fig. 80. T.S. of leaf.
Fig. 81. Epidermal cells with anomocytic stomata.
Figs. 82-97. Stages of development of nonglandular trichomes.

Fig. 82. Epidermal initial for simple unicellular trichome.

Figs. 82-84. Development of simple unicellular trichome.

Figs. 85-87. Stages in the development of simple bicellular trichome.

Figs. 88-93. Stages in the development of simple uniseriate filiform trichome.

Figs. 94-96. Stages in the development of tufted trichomes.

Fig. 97. A tufted trichome with a multicellular base and trichomes sending in all directions.

Figs. 98-115. Stages of development of glandular trichomes.

Fig. 98. Epidermal initial of glandular trich ome.

Figs.99-105. Developmental stages of uniseriate glandular trichomes.

Figs.100-101. Two and three celled stages of glandular trichome.

Figs. 106-109. Stages of development of cylindrical glandular trichome.

Figs. 110-113. Stages of development of fusiform glandular trichome.

Figs. 114-115. Divisions in the head cell of capitate glandular trichome.


Figs. 116-123. Serial transections of the flower from pedicel and upwards.

BtSmCt - bracteole-sepal median carpellary strand; SlPStCt - sepal lateral-petal-stamen-carpellary strand; Btm - bracteole median; SmCt - sepal median-carpellary strand; Btl - bracteole laterals; Ct - carpellary strand.


Figs. 124-131. Continuation of serial transection of the flower.

Sm - sepal median; CV - carpellary ventral; CM - carpellary marginal; CD - carpellary dorsal; St - stamen strand; Sl - sepal lateral; Pt petal strand; SIPSt - sepal lateral-petal-stamen strand; PST - petalstamen strand.


Fig. 132. Percentage of uni-, bi- and multiseriate rays in different frequency classes of width.

Fig. 133. Percentage af uni-, bi- andmultiseriate rays in different classes of height.

Fig. 134. Percentage readings in different frequency classes of number of rays per mm.

Fig. 135. Percentage of sieve tubes in different frequency classes of length and width.

Fig. 136. Percentage of fibre wedges in different frequency class of tangential extent.

Fig. 137. Percentage of fibre wedges in different frequency class of radial extent.


FIG. 132



FIG. 134


FIG. 136

fig. 137

Fig. 138. Transverse section of Bth day seedling showing epideris with trichome and cortex with first formed fiore bunds.
Fig. 137. Portion of phloen block showing protophlate. fibres alternating with soft tissue and an the outer side is the pericycle and endodermis.

F1g. 140. Transverse section of a stem at pre-inowering stage showling inner sucondary fibre bands.

Fig. 141. Portion of phloers bloci showing fibre bands alternating with sieve tubes, companion cells and parenchyma and fibre walis of different thicicness.

Fig. 142. Transverse section of mature bark showing ray expansion, fibre wedges and periderm.

Fig. 143. A single fibre band with thick wall and lumen.
Fig. 144. Phloem tissue showing companion cells, sieve tubes and parenchyma cells with druses.

Fig. 145. Periderm showing cork cells and lenticel.
Fig. 146. Uniseriate ray.
F1gs. 147-147. 3iseriato rays.
Fig. 150. Multiseriate ray.
Figs. 151-153. Macerated fiore cells.
Fies. 154-157. Fibres showing different fiore ends.
Fig. 158. Sieve tubes with compound sieve plates in tangential 1 cngitudinal section.

F1g. 159. Sleve tubes with sieve plates, companion cells and rays in a tangentiaily longitudinal section.

F - endodermis; P-pericycle; F3 - fibre bundie; S - soft tissue; R-ray; F\% - Itbre wall; LU - lumen; Ty - tertiary wall: $3 T N$ beginning of tertiary wall; 3w-secondary wall: $9 T$ - sieve tube; CC - companion cell; RAEX - ray expansion; L-ienticel; C - cork;


Fig. 160. Percentage of fi bre lumen in different frequency classes at three stages of flowering.

Fig. 161. Percentage of fibre cross section in different frequency classes at three stages of flowering.

Fig. 162. Percentage of fibre diameter in different frequency classes at 15 days interval from 45 days to 75 days of germination.

Fig. 163. Percentage of fibre diameter in different frequency classes at three stages of flowering.

Figs. 164-165. Percentage of fibres in different frequency classes of length from 15 th day to $120 t h$ day of germination.


Photomicrographs 1-4. Developmental stages of microsporogenesis.

Photomicrograph 1. T.S. of young anther showing outer parietal and inner sporogenous cell

Photomi crograph 2. L.S. of anther lobe.

Photomicrograph 3. L.S. of anther showing meiosis in microspore mother cell.

Photomi crograph 4. L.S. of anther showing tetrads and tapetum.

SP - sporogenous cell; M1MC microspore mother cells; T - tetrads.




Photamicrographs 5-7. Developmental stages of male gametophyte.

Photomicrograph 5. L.S. of anther showing pollen grains surrounded by periplasmodium.

Photomi crograph 6́. Two nucleate pollen grain with a larger vegetative nucleus and smaller generative nucleus.

Photomicrograph 7. Pollen grain at two male gamete stage.

PG - pollen grain; TC - proto plasmoftapetal celis; GN generative nucleus; VN - vegetative nucleus; MG - male gametes.


Photomicrographs 8-11. Developmental stages of megasporogenesis.

Photomicrograph 8. L.S. of ovule showing an outer parietal and an inner sporogenous cell and initiation of inner integument.

Photomi crograph 9. L.S. of ovule showing anticlinal division in the parietal cell.

Photomicrograph 10. L.S. of ovule showing the elongated megaspore mother cell.

Photomicrograph 11. Megaspore mother cell resulting in the formation of dyad.

P - parietal cell; SP - primary sporogenous cell; II - inner integument; MMC - megaspore mother cell; D - dyad.


MMC

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Photomicrograph 12. L.S. of ovule showing tetrad of megas pores.

Photomicrograph 13. Part of ovule showing male nucleus lying near the egg nucleus within the egg cell surrounded by pollen tubes.

Photomicrograph 14. Part of ovule showing male and female nuclei in a stage of fusion.

Photomicrograph 15. L.S. of ov ule showing free nuclear endosperm before the fertilized egg undergoes first division.

II - inner integument; MF - functional megaspore;
OI - outer integument; PT - pollen tube; EN - egg nucleus; $M N$ - male nucleus; E - endos perm.



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Photomicrograph 16. L.S. of ovule showing the compact free nuclear endosperm at micropylar end.

Photomicrographs 17-19. Endosperm at different regions.

E - embryo; FNE - free nuclear endosperm; CE - cellular endosperm.


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CE



FNE


Photomicrograph 20. L.S. of ovule showing inner and outer integuments.

Photomicrographs 21-23. L. S. of ovule showing development of seed coat at different stages of embryo.

OI - outer integument;
P - palisade-like layer;
PI - pigment layer;
FT - fringe tissue.



Photomicrograph 24. L.S. two-celled proembryo.

## Photomi crograph 25. L.S. eight-celled proembryo also showing free nuclear endos perm.

Photomicrograph 26. L. S. globular embryo showing distinct surface layer.

PE - proembryo.


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Photomicrograph 27. L.S. late globular embryo showing prominent central cell and columella initials.

Photomicrograph 28. L.S. heart-shaped embryo showing prominent cotyledonary primordia and the central cell.

Photomicrograph 29. L.S. tor pedo-shaped embryo showing vasculature.

C - central cell;
Col - columella.


Photomicrograph 30. L.S. radicle of torpedo stage of embryo showing Kappe divisions in surface and sub-surface layers

Photomicrograph 31. L.S. radicle of torpedo stage of embryo showing distinct initials for central cylinder, cortex and columella.

Photomicrograph 32. L. S. median of root apex of seedling.

Photomicrograph 33. L.S. median of root apex of seedling showing knee formation and secondary columella formation.

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CCI - central cylinder initial;
COI - cortical initial; COLI -
columella initial; PL - plerome;
PER - periblem; COL - columella;
K - knee; SC - secondary columella;
PC - primary columella; dotted
line shows the inverted cup formation.
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Photomicrograph 34. L.S. tor pedo stage of embryo showing slightly bulging out epicotyl apex.

Photomicrograph 35. L.S. mature shoot apex showing maximal phase of plastochron.

Photomicrograph 36. L. S. mature shoot apex showing minimal phase of plastochron.

EA - Epicotyl apex.


Photomicrograph 37. T.S. of 4 day old seeding at higher level of hypocotyl.

Photomicrograph 38. T.S. of 4 day old seedling just below the cotyledonary node.

Photomicrograph 39. T.S. of 4 day old seedling above the cotyledonary node where traces have departed to the cotyledons and the first leaf has formed.

Photomicrograph 40.
T.S. of 8 day old seedling at the cotyledonary node.

Cot - Cotyledon
X - Xylem strand to the first
( $a c c, b$ ) and $(a, d d, b$,$) are two$ traces to each of the cotyledons.



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# Photomicrograph 41. T.S. of petiole at basal region. 

Photomicrograph 42. T.S. of petiole at middle region.

Photomicrograph 43. T.S. of leaf through midrib showing the nectary.

Photomicrograph 44. T.S. of leaf showing the opening of nectary.

NG - nectary glandular trichomes


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Photomicrograph 45. T.S. of leaf.

Photomicrograph 46. Surface view of the epidermis showing stomata.

Photomicrograph 47. Marginal dentation showing veins terminating.

Photomicrograph 48. Partion of cleared leaf showing the median, secondary and tertiary veins.


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Photomicrograph 49. T.S. of pedicel showing four large and four small strands. Photomicrographs 49-52. Serial transections of the flower from pedicel and upwards. BtSmCt - bracteole-sepal mediancarpellary strand SIPStCt sepal lateral-petal-stamencarpellary strands Btm - bractole median; SmCt - sepal mediancarpellary strands.



Photomicrophs 53-56. Continuation of serial transection of the flower

Pt - petal strand
Sl - sepal lateral
CV - carpellary ventral
CM - carpellary marginal
St - stamen strand


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Photomicrograph 57. T.S. 8th day seedling showing first formed fibres.

Photomicrograph 58. T.L.S. secondary phloem showing rays and sieve tubes.

Photomicrograph 59. T.L.S. secondary phloem showing sieve tubes with sieve plate.

EP - epidermis; FB - fibre bundle;
ST - sieve tube; SP - sieve plate;
CC - companion cell; $R$ - ray.


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Photomicrograph 60. T.S. of stem nearer to the shoot apex at pre-flowering stage showing a layer of fibre bands.

Photomicrograph 61. T.S. of stem showing fibre bands and phloem tissue and rays in between the fibre bands.

Photomicrograph 62. Fibres showing different types of fibre ends.

Photomicrograph 63. Fibre with tertiary thickening.

EP - epidermis; PT - parenchyma tissue; $P$ - phloem; FB - fibre band; FE - fibre end.


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# Photomicrograph 64. T.S. of stem at flowering stage showing the formation of fibre wedges and ray expansion. 

## Photomicrograph 65. T.S. of stem showing the cambium and the formation of secondary fibres.

Photomicrograph 66. T.S. of stem at post-flowering stage showing fibre wedges alternating with ray expansion, the cork cells and lenticels.

EP - epidermis; FB - fibre band; CA - cambialy; R - ray; CC companion cell; ST - sieve tube; L - lenticel; C - cork cells; RE - ray expansion; FW - fibre wedge.


