M.Sc. BOTANY LAB MANUAL

2nd Semester

Prepared By

Biological Science Dept.

Botany

MIDNAPORE CITY COLLEGE

BOT 295
ANGIOSPERM TAXONOMY & BIOSYSTEMATICS
(Practical)

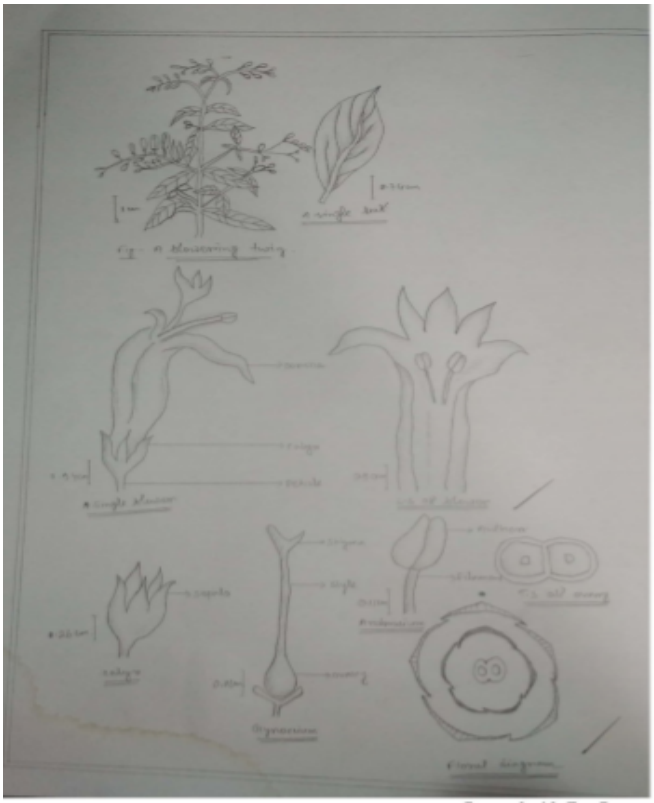
Drawing and Description of a specimen from locally available representative families, identification up to species :

Study of taxonomic specimen –A

Specimen – Supplied from the college laboratory .
Habitat –Terrestrial .
Habit –Herbaceous .
VEGETATIVE PARTS
Root – Not supplied .
Stem –Erect , nodes and internodes are conspicuous ,quadrangular ,deep green in colour with swollen nodes , minute hair present ,inter-nodal length -1.5 cm.
Leaf –Opposite decussate ,leaf margin entire ,leaf apex acute with equal leaf base ,lanceolate ,length of lamina 2.5 cm ,deep green in colour ,reticulate venation .

REPRODUCTIVE PARTS –·
Inflorescence – Recemose ,panicle .

- Flowers are pedicelled, biliped, white-purple or spotted purple and solitary. Pedicel is 2.5–10 mm in size, slender and glandular pubescent. Bracts are acicular and 2.5 mm long.
- Calyx - sepals 5,,gemosepalous ,valvate, lobes are subacute, 2.5-3.7 mm long and glandular. Corolla - petals 5, bilabiate , gamopetalous ,7.5-12.5 mm in size, tube about half as long as the corolla. Androecium - Stamens 4 ,epipetalous,didynamous ,filaments are attached nearly to the corolla tube . Filaments are hairy and anthers are two celled and exerted , purple beared at base.
- Gynoecium – carpels 2 ,syncarpous ,style slender ,stigma shortly two lobed ,ovary contains very minute hair ,two locules in ovary ,axile placentation ,ovules 2-6 in each locule.
- Fruit is a capsule, oblong, 18-20X4.5–5.0 mm, young ones sparsely glandular and hairy; when mature it is glabrous. Seeds are subquadrate, yellow to brownish in colour and rugose.



Identificatio-(1) stem with conspicuous nodes and internodes ,nodal region swollen

(2)leaves opposite decussate. lanceolate.

(3)Inflorescence – recemose panicle type .flowers bracts and bracteoles ,zygomorphic,bisexual and small .

(4) corolla bilabiate , stamen epipetalous ,didynamous ,anther bi-celled and exerted ,carpels -2, syncarpous, stigma shortly two lobed , style slender ,ovary with minute hair ,axile placentation ,seeds with retinacula .

……..hence .the plant belongs to the family Acanthaceae.

Erect or prostrate ,not climbing ,calyx distinct ,bracts are not sub-connate ,ovules more than two in each cell .

Seed supported on hard upward curving retinacula, ovules 2 ,not collateral or if more than two superposed in one row or else arranged alternately in each cell .

Corolla lobes, imbricate in bud .

Capsule compressed at right angles to the septum, seeds ovoid hardly compressed .

…….Genus – Andrographis .

Capsule nearly glabrous ,linears.

Leaves lanceolate ,glabrous.

Pedicels distinct.

……species – A. paniculata

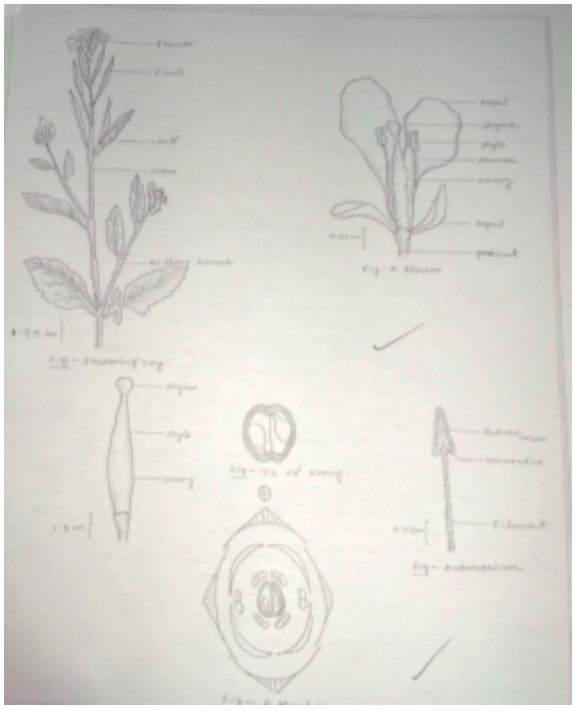
Hence the supplied plant specimen –**A** is *Andrographis paniculata* L.

Study of Taxonomic specimen –B

Specimen – Supplied from the college laboratory .
Habitat –Terrestria
Habit –Erect herb
VEGETATIVE PARTS
Root – Not supplied .
Stem –Solid, soft , terete, nodes and internodes are distinct ,alternate branching green in colour juice pungent.
Leaf –Exstipulate, sessile ,simple , alternate serrate ,leaf apex acute with equal leaf base ,glabrous ,reticulate venation, unicostate .
REPRODUCTIVE PARTS –·
Inflorescence – Recemose ,receme,axillary or terminal , ebracteate.

- Flowers – Actinomorphic .bisexual ,complete ,hypogynous ,tetramerous .
- Calyx-sepals 4, polysepalous, imbricate, deciduous, greenish.
  Corolla - petals 4, polypetalous ,cruciform ,distinctly clawed at the base, vavate ,yellow . Androecium - Stamens 6 , free, tetradynamous (2 –outer and 4-inner)filament long ,anther bi-celled ,basifixed dehiscing longitudinally .
- Gynoecium – carpels 2 ,syncarpous ,style short ,stigma simple ,capitate, ovary superior ,2 celled by a false partition - replum ,ovules many in parietal placentation .
- Fruit -Dry dehiscent ,elongated capsule (siliqua).
- Identificatio- (1) Plants herbaceous .
- (2)leaves simple, alternate ,reticulate ,unicostate ..
- (3) Inflorescence – recemose ebracteate, flower –bisexual ,complete ,regular ,sepals -4, free ,imbricate ,petals -4 ,cruciform ,clawed ,stamens -6 tetradynamous , carpels -4 ,syncarpous , superior , ovules in parietal placentation , fruit long dehising capsule (siliqua),seeds globose .
-
- ……..hence .the plant belongs to the family Cruciferae.
- Plant dehiscing ,narrow long ,seed-1 ,seriate,flower-yellow .
- …….Genus – *Brassica* .
- Leaves of the stem auriculate ,leaves without hairs ,less lobed.
- ……species – *B. napus* .
- Hence the supplied plant specimen – **B** is *Brassica napus* .
-

- ***N. B :- Drawing ,measuring and labeling are mandatory part of plant taxonomic description*** .
-
-



Study of taxonomic specimen –C

Specimen – Supplied from the college laboratory .
Habitat –Terrestrial .
Habit –Herbaceous

VEGETATIVE PARTS

Root – Not supplied .

Stem –Woody,erect ,cylindrical ,branched distinct nodes and internodes ,solid ,glabrous , length of internodes varies from 4-5 cm ,green in colour , minutely hairy .

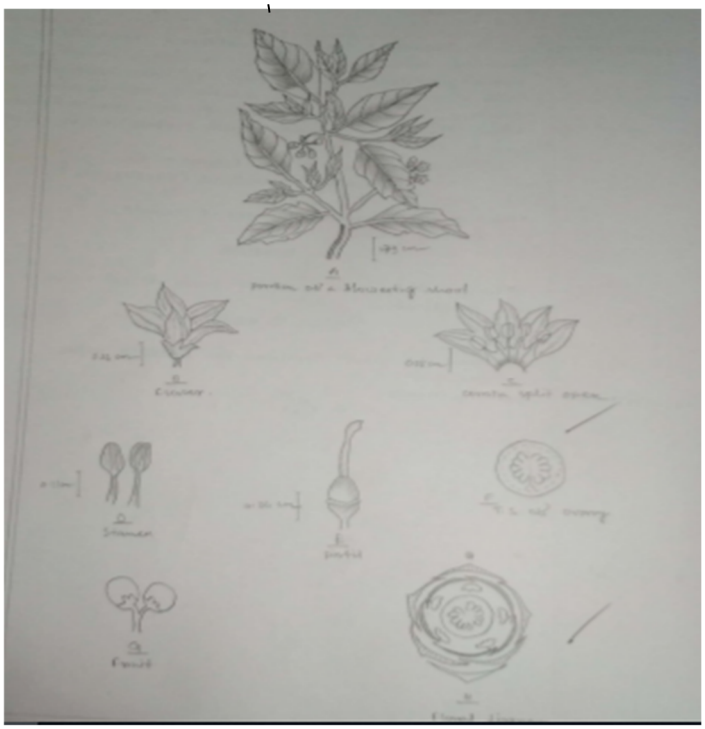
Leaf – ,simple , alternate petiolate, exstipulate ,ovate , serrated ,leaf apex acute ,membranous ,green in colour ,length of each leaf 3-4 with equal leaf base ,glabrous ,reticulate venation unicostate .

REPRODUCTIVE PARTS –·

Inflorescence – Flowers in dichotomous ,extra axilary ,cyme , ebracteate.

- Flowers – Actinomorphic .bisexual ,complete ,hypogynous ,pentamerous ,small, white in colour ,ebracteate ,pedicillate.

- Calyx - sepals 5,,gamosepalous ,imbricate, regular ,persistant ,small ,oblong ,acute ,hairy ,greenish. Corolla - petals 5 gamopetalous ,rotate , imbricate,small ,white in colour . Androecium - Stamens 5, epipetalous,alternate to petalous ,filament short ,anther 2- celled ,oblong ,basifixed ,dehiscent by apical pores .
- Gynoecium – carpels 2 ,syncarpous, ovary superior .style terminal,stigma simple and small , axile placentation ,slightly bifid..
- Fruit -Berry ,simple ,indehiscence with star shaped ,persistant calyx ,round ,purplish black when ripe and shiny .



Identification- .

1. Inflorescence – flowers in extra axillary cymes .
2. leaves simple, alternate ,exstipulate ..
3. Sepals united ,persistant.
4. Corolla campanulate ,rotate.
5. Stamen -5 epipetalous ,alternate with porous dehiscence.
6. Gynoecium bicarpellary ,ovary superior ,2- chambered with many ovule in each
7. Fruits are berry.

……..hence .the plant belongs to the family solanaceae

Fruit indehiscent berry ,corolla rotate ,leaves entire ,anther connivent in a cone ,dehiscent by apical poress,embryo curved or sub -spiral.

…….Genus – *Solanum*

Leaves ovate or oblong ,sinuately lobed .

……species – *Solanum nigram* .

Hence the supplied plant specimen – C is *Solanum nigram* .

Study of taxonomic specimen –D

Specimen – Supplied from the college laboratory .

Habit –Herb ,prostate .

VEGETATIVE PARTS

Stem –Aerial prostrate ,presence of hair ,green in colour . .

Root – Not supplied

Leaf –Simple ,exstipulate ,hairy ,leaf margin serrate, 1-2 cm long both the surface consist of hair ,deep green in colour ,reticulate venation .

REPRODUCTIVE PARTS –·

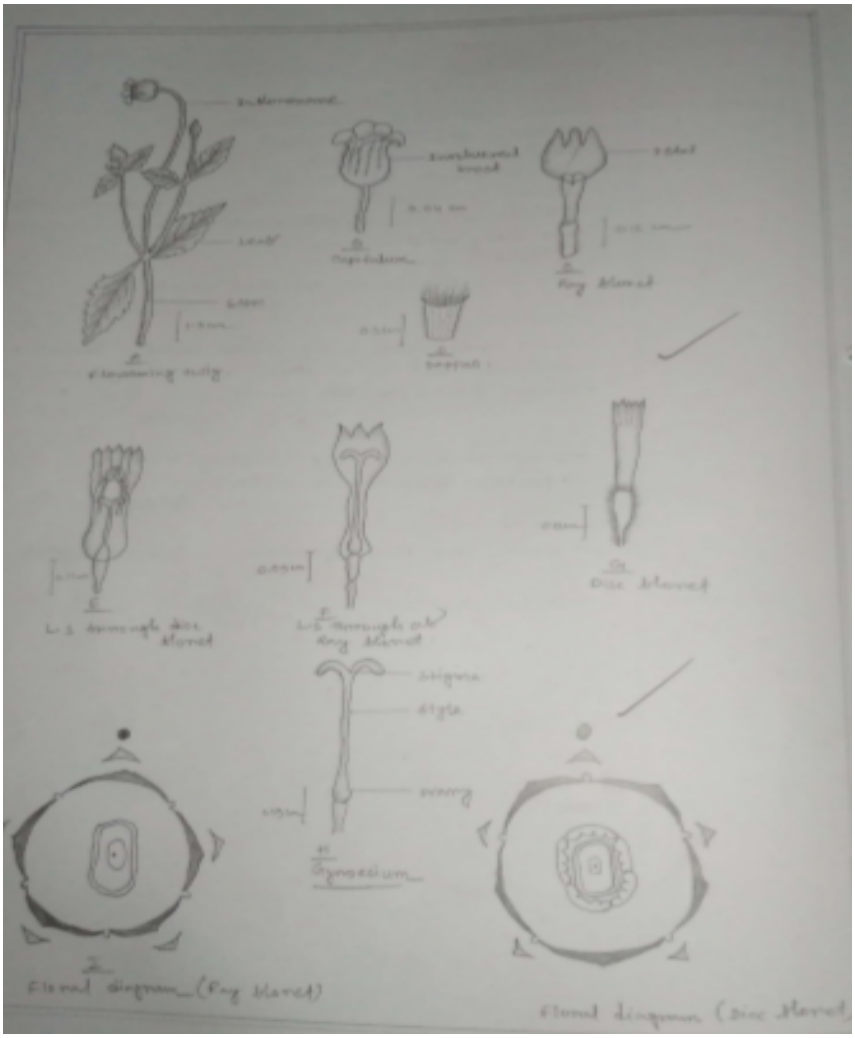
Inflorescence – Capitulum

Involucre –Many bracts and bracteoles basally fused together on the outer surface forming an involucres . It is green in colour and hairy .

- Flowers –Bracteate ,sessile ,two types of florets (1) Ray florets (2) Disc florets .
- Ray florets –Bracteate ,sessile ,unisexual ,dichlamydeous ,zygomorphic ,epigynous ,yellow in colour .
- Calyx - sepals ∞ ,free , highly reduced ,hairy and scale like structure called pappus ,superior . Corolla - petals 3, gamopetalous ligulate ,yellow ,superior ,

.

- Gynoecium – carpels 2 ,syncarpous , ovary inffferiorb,locule -1 ,one ovule in the basal placentation ,style single, terminal ,stigma bifid ,hairy ,coiled
- Disc florets – Bracteate ,sessile complte ,actinomorphic epigynous ,dichlamydeous ,pentamerous ,yellow.
- Calyx –Same as ray florets .
- Corolla – Petals -5 ,gamopetalous ,tubular ,superior ,yellow .
- Androecium –Stamen -5 ,epipetalous ,syngenesious ,bicelled superior .
- Gynoe cium –Carpels -2 ,syncarpous ,ovary inferior ,locule -1, basal placentation ,style single ,terminal ,stigma labid ,hairy ,coiled .



Identification –Ovary inferior ,stamen equal in number to corolla lobes ,alternate with lobes stamens attached to the corolla ,anther coronate ,syngenesious ,ovary -1 locular ,ovule -1 ,calyx reduced to pappus ,basal placentation ,flowers in heads surrounded by an involucres .

……..hence .the plant belongs to the family Asteraceae.

Corolla of all the flowers fused to near the mouth or if any expanded from a tubular base (ligulate)then only the marginal florets of the fiower –head (ray-florets) so, expanded .style long distinct or style sub entire then so only in the sterile flowers of heads with dissimilar .
Flowers dissimilar (heterogamous) disc.
Pappus of numerous scales ,head radiate .
Scales of pappus –feathery ,head medium .

…….Genus -*Tridex*
Only one species recorded in the key .
……species T- procumbens .
Hence the supplied plant specimen – **D** is *Tridex procumbens* .

Study of taxonomic specimen –E

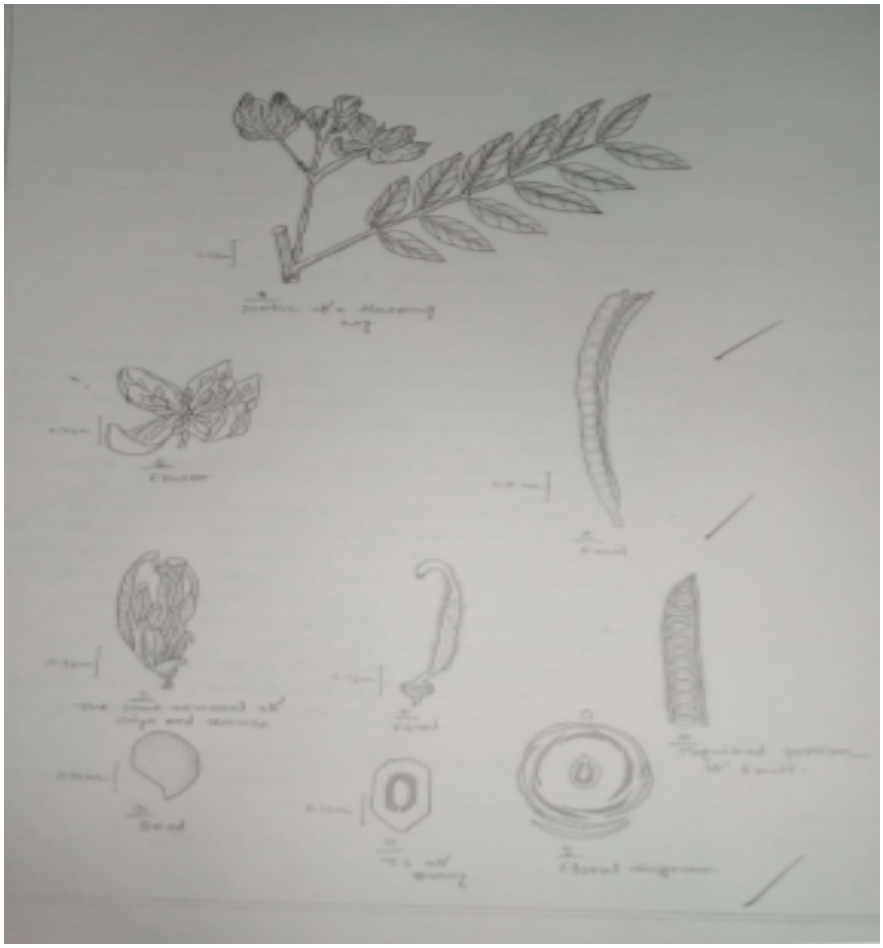
Specimen – Supplied from the college laboratory .
Habitat –Terrestrial .
Habit – Herbaceous .

VEGETATIVE PARTS

Root – Not supplied .
Stem –Solid, woody, erect, arial, branchedcyylindrical ,glabrous , nodes and internodes are distinct ,inter-nodal length 2.4-3.4 cm ,green in colour .
Leaf –Pinnately compound ,stipulate ,number of leaflet 6-12, reticulate venation unicostate entire ,ovate ,acute gland dotted ,lanceolate ,length of the leaf varies 10-12cm .
Leaflet – Ovate ,unicostate ,entire ,acute ,reticulate ,length of each leaflets varies from 3-5 cm .
REPRODUCTIVE PARTS –·
Inflorescence - Axillary racemes ,bracts and bracteole present ,corymb .

- Flowers – Bisexual regular ,pedicillate ,slightly zygomorphic ,pentamerous ,perigynous yellowish . .bisexual ,complete ,hypogynous ,tetramerous .

- Calyx - sepals 5,polysepalous ,imbricate, broad ,greenish .
  Corolla - petals 5 , polypetalous ,slightly irregular, imbricate,yellow .
  Androecium - Stamens 10 unequal ,filamentous, 6-fertile and 4- sterile,out of six , four small and two large ,anther 2-celled ,basifixed ,dehiscent longitudinally .
- Gynoecium – carpel- 1 ,apocarpous ,style short ,stigma terminal , ovary superior ,unilocular ,large curved slightly pubescent ,greenish , marginal placentation .
- Fruit -Woody ,dry dehiscent ,pod, lomentum .

- 

Identificatio- .

(1)leaves pinnatly compound .

(2) Flower bisexual ,irregular ,zygomorphic .

(3)Calyx of 5 sepals , free irregular

(4)Androecium of 10 stamens .

(5) Gynoecium monocarpellary, placentation marginal .

……..hence .the plant belongs to the family Caesalpiniaceae.

Leaves simple ,foliate or pinnate ,anther basifixed ,petals -5 ,leaves even pinnate .

…….Genus – *Cassia* .

Leaflets 3-5,ovate –oblong

Pods flattened ,impressed between the seeds .

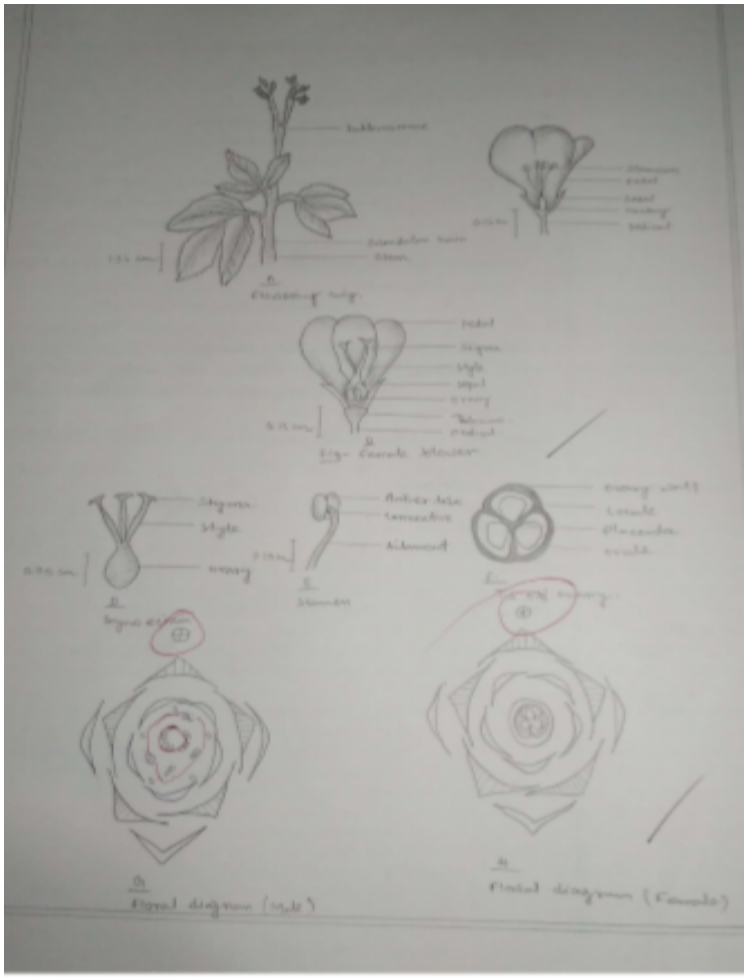
……species C- *occidentalis* .

Hence the supplied plant specimen – E is *Cassia occidentalis*

Study of taxonomic specimen –F
Specimen – Supplied from the college laboratory .
Habit –Under shrub, milky latex through the vegetative part .
VEGETATIVE PARTS

Stem –Aerial , erect ,solid ,reddish brown ,having gland differentiated into nodes and internodes , nodes swollen length of the internode -3 cm .
Root – Not supplied
Leaf –Simple ,palmate ,petiolate,reddish –green ,tri-penta foliate upper surface –brownish green ,shiny stipulate , stipule modified into glands ,gland are present on the upper surface .margin- entire, apex –acute ,base-cordate,venation –multicostate reticulate .texture of lamina –membranous ,glutinous dorsiventral alternate ,lower –glabrous ,upper-shiny .length of the the petiole 5-6.5 cm .
REPRODUCTIVE PARTS –·
Inflorescence –peniculate cyme .

- Flowers –Bracteate , unisexual ,actinomorphic ,hypogynous ,pentamerous ,dichlamydous ,red in colour ,length of flower -0.8 cm .
- Male flower –
- Calyx - sepals 5 ,gamosepalous, lanceolate ,imbricate ,glandular ,inferior ,green ,persistant .length of the calyx -0.3 cm .
  Corolla - petals 5,obovate ,imbricate-quencuncial,inferior ,reddish brown,length of the corolla -0.4 cm .
- Androecium – Stamens -8 in two bundle,3 in inner whorl ,3- in outer whorl,anther –reniform ,flattened ,dehiscence ,transversely .
- Female flower –
- Calyx – Sepals -5 gamosepalous ,lanceolate ,imbricate ,glandular ,inferior ,persistant .
- Corolla –Petals -5 obovate ,imbricate ,quincuncial ,inferior ,reddish brown .
- Gynoecium – Carpels -3 ,syncarpous ,triangular ,superior ovary ,ovule in each locule ,pendular ,axile placentation .
- Fruit –Regma ,seeds carrunculate .



Identification – Plants under shrubs ,leaves alternate ,stem with latex ,flower unisexual ,actinomorphic ,poly petalous ,,steams free mostly , carpel-3 ,united ,superior, ovule in axial placentation , fruit dehiscent –capsular type ,leaves simple,stipulate ,seeds with copious albumen

……..hence the plant belongs to the family Euphorbiaceae.

Flower monoecious ,stamen biserate ,stamens alternate with sepals .

Flowers in terminal cymes .

Leaves digitateiy lobed .

Central flower female .

Calyx regular 5- lobed .

Stamens many outer 5 in a series opposite the petals ,the rest in column in centre of flower .

…….Genus – *Jatropha* .

Leaves palmately 3 -5 lobed ,lobes widened in the middle ,glandular toothed ,bracts ,sepals and stipules glandular ,petiole glandular .

……species J -*gossipifolia*

Hence the supplied plant specimen – F is *Jatropha gossipifolia*

PALAEOBOTANY, PALYNOLOGY & PLANT REPRODUCTIVE BIOLOGY

Acetolysis of Pollen Grains

This method is used to study exine ornamentation in detail. Acetolysis removes all non-sporopollenin components of the pollen (the contents, intine and components present in and on the pollen wall) leaving only the exine intact.

Special Requirements A cetolysis mixture: acetic anhydride and concentrated sulphuric acid (9:1). Add the acid slowly to acetic anhydride in the fume hood; as it results in exothermic reaction, the container gets hot. Low-speed centrifuge. Glacial acetic acid.

Procedure:

1. Collect suffi cient amount of pollen grains (2–4 mg) and suspend them in about 10 ml of acetolysis mixture taken in a test tube. 2. H eat the suspension to boiling in a water bath for 1–2 min. Remove the tube and allow it to cool. 3. T ransfer pollen suspension into a centrifuge tube. Remove acetolysis mixture by low- speed centrifugation (about 2,000 rpm). 4. Add about 5 ml of glacial acetic acid to the centrifuge tube. Mix pollen pellet and remove acetic acid by centrifugation. 5. Add about 5 ml of water to centrifuge tube and mix pollen pellet in water. 6. Mount pollen grains on a slide in water or 10 % glycerol and observe under the microscope. They can also be mounted in glycerin jelly as semi-permanent preparations for later observations and also as reference slides.

Mounting of Pollen Grains in Glycerin Jelly

For any morphological study such as size, shape, apertures and exine ornamentation, pollen grains have to be mounted in a suitable medium. Although temporary preparations can be made in acetocarmine or safranin, the structure of the pollen grains may not be clear. Glycerin jelly, containing some stain such as basic fuchsin, has been used routinely as a mounting medium by pollen morphologists. Glycerin jelly becomes solid at room temperature, and it has to be melted for mounting. Both fresh and acetolysed pollen can be mounted in glycerin jelly

Special Requirements : Glycerin jelly , Spirit lamp/hot plate , Paraffin wax

Procedure :1. Take a small amount of pollen sample (to be mounted) on the middle of a clean microscope slide. 2 . Scoop a small piece of glycerin jelly by using forceps or a spatula and place the jelly in contact with the pollen sample so that pollen grains adhere to the jelly. 3 . Warm the slide gently on a spirit lamp/hot plate until the jelly melts. Do not allow it to boil. 4. Keep the slide under a stereomicroscope and spread the pollen in the melted jelly by gently stirring with a needle. Rewarm the jelly if it solidifi es during stirring. 5. Lower a cover glass carefully over the jelly. Allow it to spread into a circle of 4–5 mm

and rewarm the slide, if necessary. 6 . W hile the slide is still warm, place the slide inverted (cover slip facing down) with support on both the sides with some solid material so that the cover glass remains hanging without touching anything. This will allow pollen grains to settle near the cover glass so that pollen grains can be observed under higher magnifi cation of the microscope. 7. Observe the preparation or store for later observation. Glycerin jelly slides remain in good condition for many weeks or even months. They should be stored under dust-free atmosphere.

In Vitro Germination Test for Pollen Vigour

One of the simple methods to assess the vigour of pollen samples is to use pollen samples for controlled pollinations. One set of pistils is pollinated with fresh pollen which are expected to show maximum viability and vigour and another set with pollen sample to be tested for vigour. Some pistils from each set are fi xed at intervals and used to study pollen germination and pollen tube growth using aniline blue test and record the extent of tube growth in terms of mm/cm. n vitro germination assay can also be used to assess pollen vigour. In the protocol to assess pollen viability , per cent pollen germination is scored at one time without consideration of the time factor; the cultures are scored after maintaining them for a much longer period than that required for germination. To assess pollen vigour, however, germination and tube length in fresh pollen samples and test pollen samples are scored at regular intervals after culture and the responses are compared.

Special Requirements: Sucrose solution, Boric acid solution, Groove slide, Moisture chamber, microscope

Procedure : 1. Raise sitting drop cultures of fresh pollen and test pollen as described in Protocol 5.5.10 . As the same culture cannot be used for scoring at regular intervals, raise more number of cultures for each pollen sample depending on the scoring time intervals. Scoring intervals depend on the time taken for germination and the rate of pollen tube growth. Record the time of raising each culture. 2. Incubate the cultures in the humidity chamber. 3. After each time interval (for many species, 30-min intervals are satisfactory, and others

may require 1-h intervals), take two cultures from each pollen sample, add a drop of fi xative or acetocarmine, mix the pollen suspension and lower a cover glass. Keep the slide under humidity chamber to prevent drying if they are not scored immediately. 4. Repeat the procedure at selected time intervals. 5. Score all the cultures (2 from fresh pollen and 2 from test pollen, at each selected time interval) for pollen germination (following the procedure given under Protocol 5.5.10 and Table 5.1) . Measure pollen tube length from each fi eld used for scoring germination as follows: (a) Adjust the intensity of microscope illumination with the help of diaphragm so that the boundary of each grain and tube are clearly visible. From each microscope fi eld used for germination score, measure the length of 10 pollen tubes selected randomly (in ocular units) by using an ocular micrometre. Omit curved pollen tubes which cannot be measured accurately. If the number of pollen tubes in each microscope fi eld is <10, measure all the pollen tubes in the fi eld. If the number of tubes is >10, measure 10 randomly selected pollen tubes. (b) E nter both germination and tube length data in a tabular form (see the sample Table 5.1). Repeat the scoring for all the

cultures. Calculate mean pollen germination and pollen tube length for each pollen sample at each time interval and present the same with SE/SD. If necessary, ocular units can be converted into micrometres by calibrating the microscope with the use of stage micrometre 6. Present the results for germination and tube length in the form of a graph over time for each reading.

Pollen Germination and Pollen Tube Growth

In several studies, it becomes necessary to determine the rate of pollen germination and pollen tube growth. This is particularly important in selfincompatible species. In some self-i ncompatible species, pollen tubes may reach the base of the style in both self- and cross-pollinated pistils, but the growth of self-pollen tubes is much slower when compared with cross-pollen tubes; in such species even when pollinations are carried out with mixed pollen (self and cross), most of the seeds are sired by cross-pollen. Such studies are also needed in interspecifi c crosses. In this protocol, pollinated pistils are fi xed at various time intervals after pollination and the extent of pollen tube growth is determined.

Procedure : 1. Tag suffi cient number of fl ower buds before anthesis and anther dehiscence. Divide them into several sets and label each set as i, ii and so on. The number of sets depends on the f requency of time intervals needed; to some extent this depends on the time taken for the pollen tubes to reach the base of the style. The number of fl ower buds for each set should be at least six. 2. Emasculate all the fl ower buds and bag them. 3. On the day of anthesis or when the stigma becomes receptive, open the bags and carry out pollination with required pollen sample in all the sets of fl owers and rebag them. Note down the time of pollination. 4. Excise pollinated pistils at preselected intervals (starting with set i) and fi x them in a fi xative. Generally, interval of 2–4 h is suffi cient in short-styled fl owers; in long-styled fl owers such as tobacco, 6–12 h may be suitable. 5. Process each set of pistils separately to study pollen germination and pollen tube growth through aniline blue fl uorescence method following Protocol 8.5.1 . 6. Observe each pistil from each set and record the extent of pollen germination and the length of pollen tube growth in the pistil. One of the standard methods to measure the length of pollen tube growth in the pistil is to put a dot (on the mounted cover glass) with a marker at the region on the pistil up to which the pollen tubes have traversed, and then measure the length (mm/cm) from the tip of the stigma up to the dot. 7. After recording the extent of pollen germination and pollen tube growth in all the pistils of all the sets, calculate, for each set, average length up to which pollen tubes have grown in the pistil. Calculate the rate of pollen tube growth using the data from all the sets. Growth rate in the pistil is generally expressed as µm/h.

Modifi cations 1. Pollen tube growth can also be studied by pollinating excised fl owers/pistils maintained in a beaker or a Petri plate 2. To compare the rate of pollen tube growth in self- and cross-pollinated pistils, divide emasculated fl ower buds into two sets. Carry out self-pollinations on fl owers of one set and cross-pollinations on the other set. Divide each set into subsets for fi xing at regular intervals. And follow later steps as described above. 3. For studying the speed of pollen germination, fi x pollinated pistils at 15 min intervals and score percent pollen germination/number of pollen grains germinated on the stigma .

BOT 296

PLANT ANATOMY & PHARMACOGNOSY

Comparative study of various types of stomata

Stomata are tiny openings or pores which are commonly noticed being distributed on the epidermis of the leaves and also in young stems. Typically, stomata are found on the lower surface of a dicot leaf and in a monocot leaf, on both its surfaces. Stomata carry out the function of regulating gas exchange and water vapor between the leaves of the plant and the atmosphere.

Aim

To prepare a temporary mount of a leaf peel in order to show the stomata of a leaf

Principle/Theory

Plants are the primary producers. They carry out physiological processes such as photosynthesis and respiration which requires gas exchange between the tissues of plants and the atmosphere. This process is carried out through tiny openings located in leaves, known as stomata. Stomata are small elliptical openings on leaves that contain chloroplasts. They are girdled by two-kidney shaped cells known as guard cells on either side of the stomata. The guard cells possess a thick inner wall and a thin outer covering which control the closing and opening of the pores of stomata.

According to Metchalfe and Chal(1950) ,there are four types of stomata considering the position of subsidiary cells around the guard cell. They are (1) **Anomocytic or Ranunculus type -**In these stomata, accessory cells are absent. The guard cells are surrounded by ordinary epidermal cells, e.g., families Ranunculaceae, Cucurbitaceae, Papaveraceae and Malvaceae. (2) **Anisocytic or Cruciferous type -**In these stomata the guard cells are surrounded by three accessory cells. Of these two are larger whereas one is smaller in size.g., family Brassicaceae. (3)**Paracytic orRubiaceous type** - In these stomata the guard cells are also surrounded by two accessory cells, but their common walls are parallel to guard cells, e.g., families Rubiaceae, Fabaceae etc. .(4) **Diacytic or Caryophyllaceous type -**In these stomata the guard cells are surrounded by two accessory cells. Their common walls are at right angle to the walls of guard cells, families Caryophyllaceae, Acanthaceae.

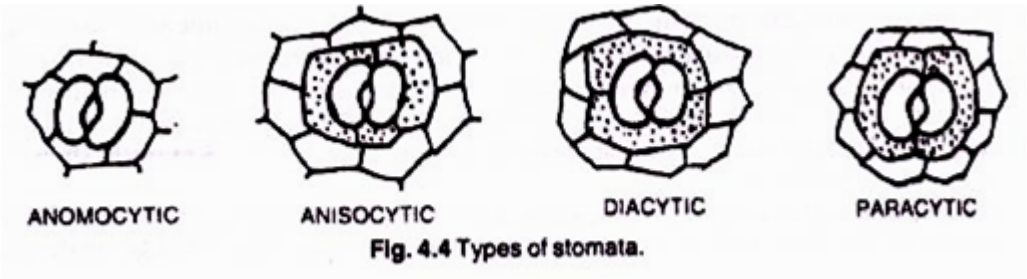


Fig. 4.4 Types of stomata.

Material Required

- Fresh leaves of *Hibiscus rosa-sinnensis* (A) ,*Brassica* sp.(B*) Oldenlandia corymbosa*(C) and *Adhatoda vasica* (D).
- Needles
- Forceps
- Watch glass
- Dropper
- Glass slides
- A brush
- Coverslips
- Blotting paper
- Safranin
- Compound microscope
- Glycerine
- Procedure: -
- Pick a healthy leaf from the supplied sample
- Fold the leaf to gently pull the peel apart to separate a peeled section from the lower surface of the leaf. Use the forceps to perform this step. Allow the peel to remain in a watch glass holding water for some time.
- In the watch glass, stain the sample by adding some drops of safranin through a dropper.
- Take the peel out after 2-3 minutes. Set it on a clear glass slide
- Add a drop of glycerin on the peel. Put a clear coverslip over it gently using a needle.
- Excess glycerin and stain can be removed using blotting paper
- Examine the slide first under a low-power and then under a high-power magnification of a compound microscope.

The following characters are observed

For sample –A , (1) Accessory cells are absent.

(2) The guard cells are surrounded by ordinary epidermal cells

Hence the supplied sample ‘ A ‘ is **Anomocytic type** of stomata

For sample-B , (1) the guard cells are surrounded by three accessory cells. Of these two are larger whereas one is smaller in size

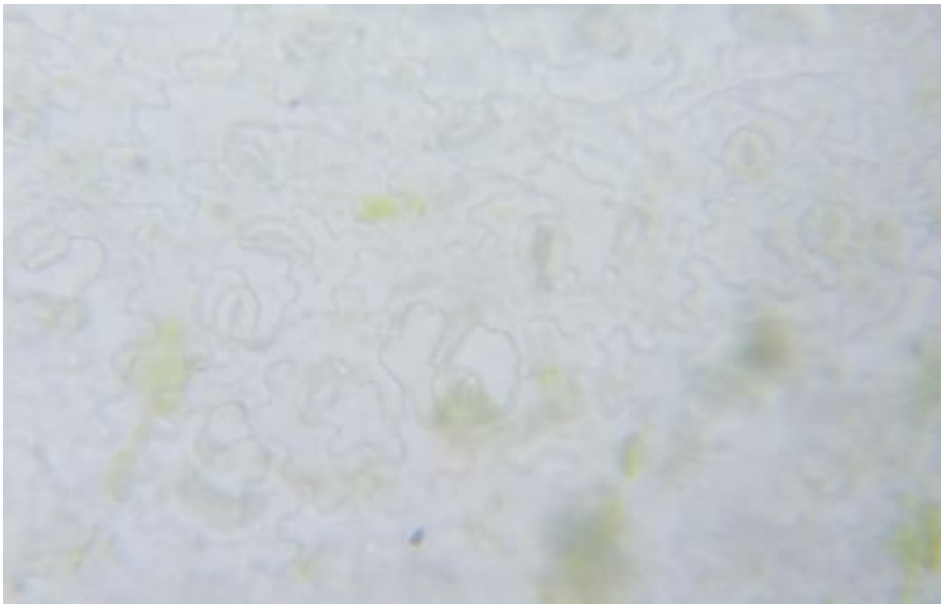
Hence the supplied sample ‘ B ‘ is **Anisocytic type** of stomata

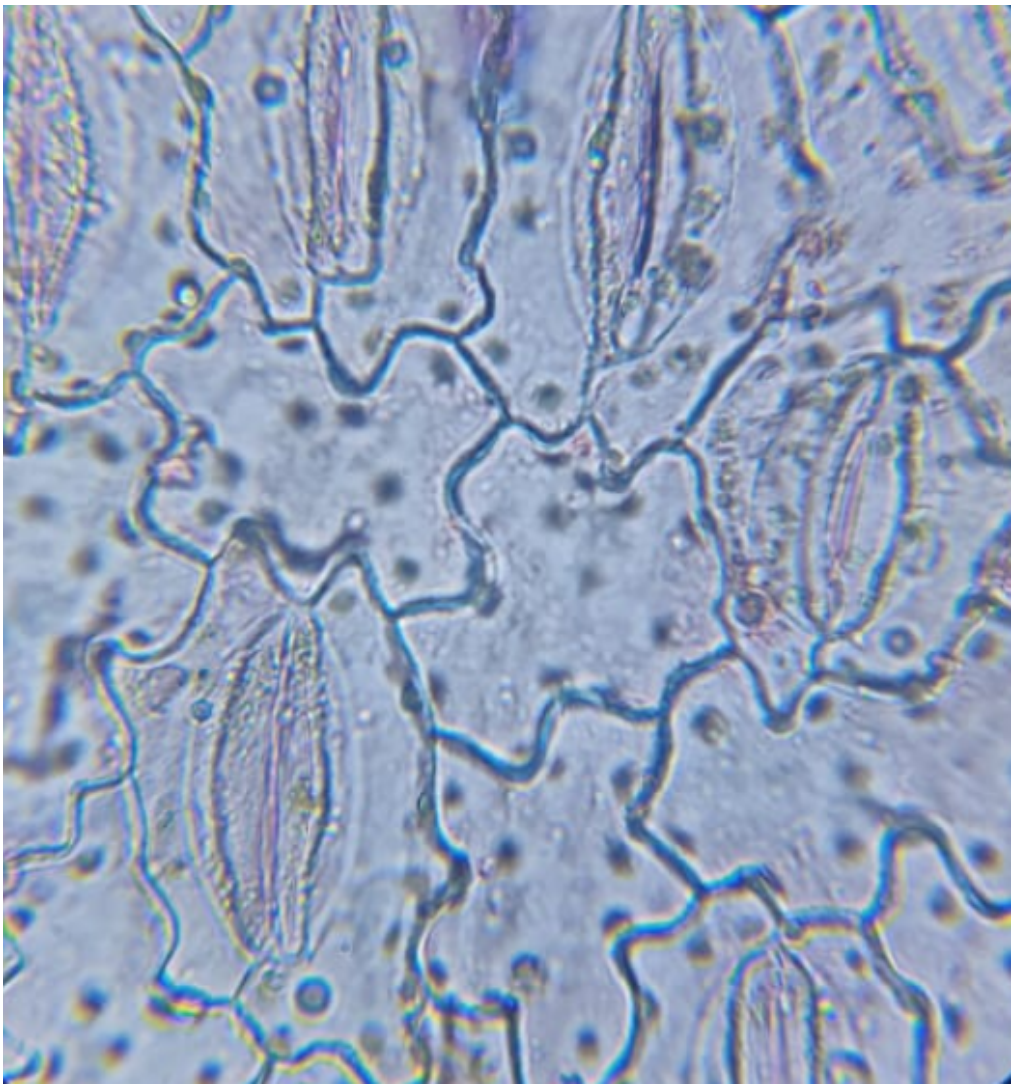
For sample-C , (1) The guard cells are surrounded by two accessory cells, but their common walls are parallel to guard cells,

Hence the supplied sample ‘ C ‘ is **Paracytic type** of stomata

For sample –D, (1) The guard cells are surrounded by two accessory cells. Their common walls are at right angle to the walls of guard cells.

Hence the supplied sample ‘ D ‘ is **Diacytic type** of stomata





Study of different types of crystals

Introduction :

Raphides are sharp needle-shaped crystals of calcium oxalate found in various tissues including leaves, roots, shoots, fruits, etc., of wide varieties of plant species, and are typically kept in highly specialized cell called idioblast . Raphides are commonly found in monocots families such as Araceae, Agavaceae, Orchidaceae, Smilacaceae, Discoreaceae, Bromeliaceae, Arecaceae, Commelinaceae, Musaceae and also in some dicot families such as Rubiaceae, Solanaceae, Actinidiaceae, Vitaceae. Study of crystals in plant body is an important part of plant taxonomy for plant identification .

Material and method –(1)Fresh leaves of *Ficus* sp. ,*Nerium* sp. ,*Solanum* sp. and colocasia sp.

(2) compound microscope

(3) watch glass

(4) slide and cover slip

(5) Safranin stain.

(6) needle

Procedure – At first a leaf of any supplied sample has to be sectioned by T.S process . Then a thin section has to be kept on the slide .By putting a drop of glycerin, a cover slip has to be placed upon it carefully . Then the slide has to be placed under the microscope to observe the crystals of various shape and structure. If it is needed to add safranin staining during the preparation of slide. In this way all sample have to be examined thoroughly .

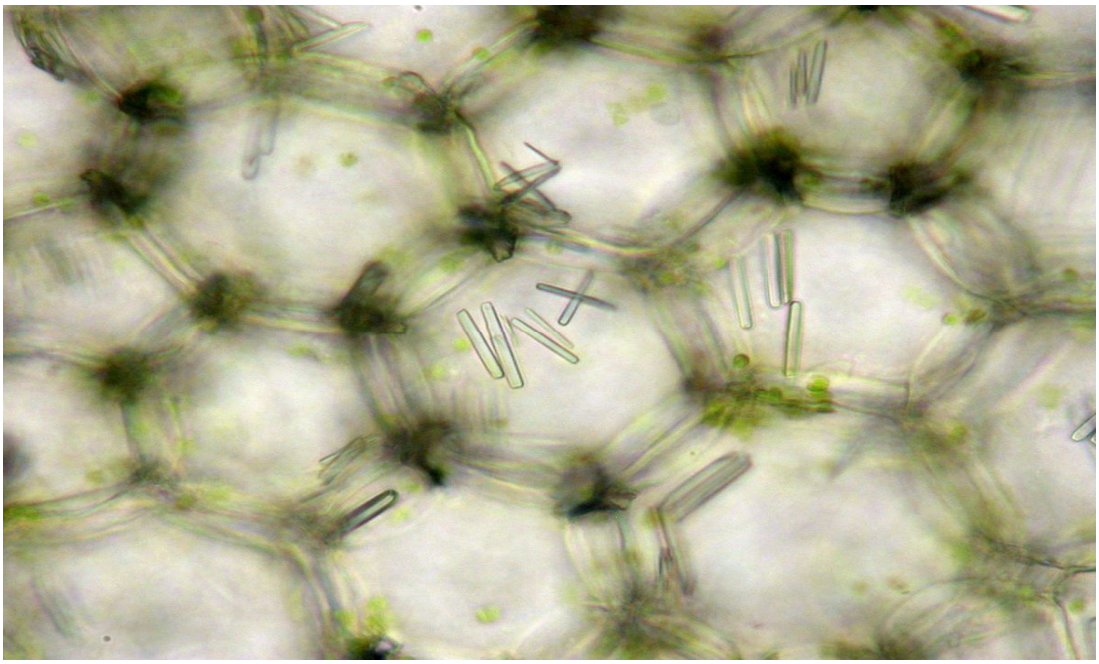
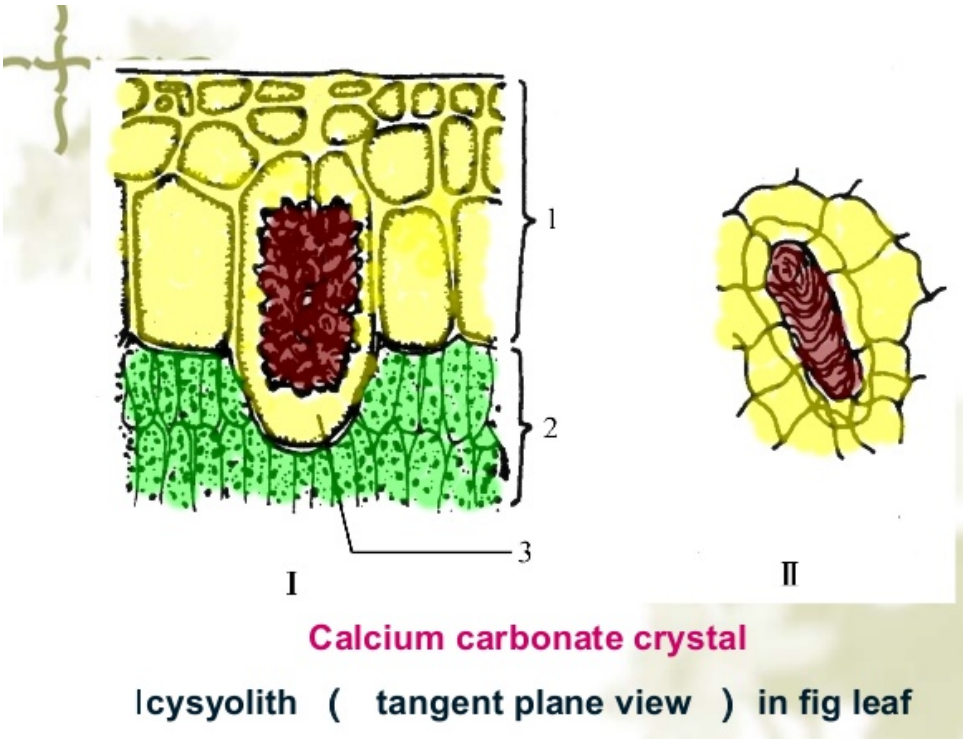


Fig : Crystals of calcium carbonate



Study of trichomes

Introduction to Trichomes:

Some of the epidermal cells of most plants grow out in the form of hairs or trichomes. They may be found singly or less frequently in groups. They may be unicellular or multicellular and occur in various forms. They vary from small protuberances of the epidermal cells to complex branched or stellate multicellular structures.

The cells of the hairs may be dead or living. Very frequently the hairs lose their protoplasm in their cells.

Types of Trichomes: Trichomes may be classified into different morphological categories. One common type is referred to as hair.

The hairs may be subdivided into:

(i) Unicellular; and

(ii) Multicellular.

The unicellular hairs may be un-branched or branched. Multicellular hairs may consist of a single row of cells or several layers. Some multicellular hairs are branched in dendroid (tree-like) manner; others have branches oriented largely in one plane (stellate hairs.)

Stinging Hairs:

They are one of the most interesting types of the trichomes. It contains a poisonous liquid and consists of a basal bulb like portion from which a stiff, slender and tapering structure is given out. This tapering structure ends in a small knob like or a sharp point.

Glandular Hairs: Many plants possess glandular hairs. These hairs may secrete oil, resin or mucilage. A typical glandular hair possesses a stalk and an enlarged terminal portion, which may be referred to as gland. The glandular hairs may be uni- or multicellular.

Active secretory cells of glandular trichomes have dense protoplasts and elaborate various substances, such as volatile oils, resins and mucilage's, and gums. These substances are excreted and accumulate between the walls and cuticle. Their final removal from the hair occurs by rupture of the cuticle.

Scale or Peltate Hair:

A common type of trichome is the scale, also called peltate hair (from the latin peltatus, target-shaped or shield like, and attached by its lower surface). A scale consists of a discoid plate of cells, often borne on a stalk or attached directly to the foot.

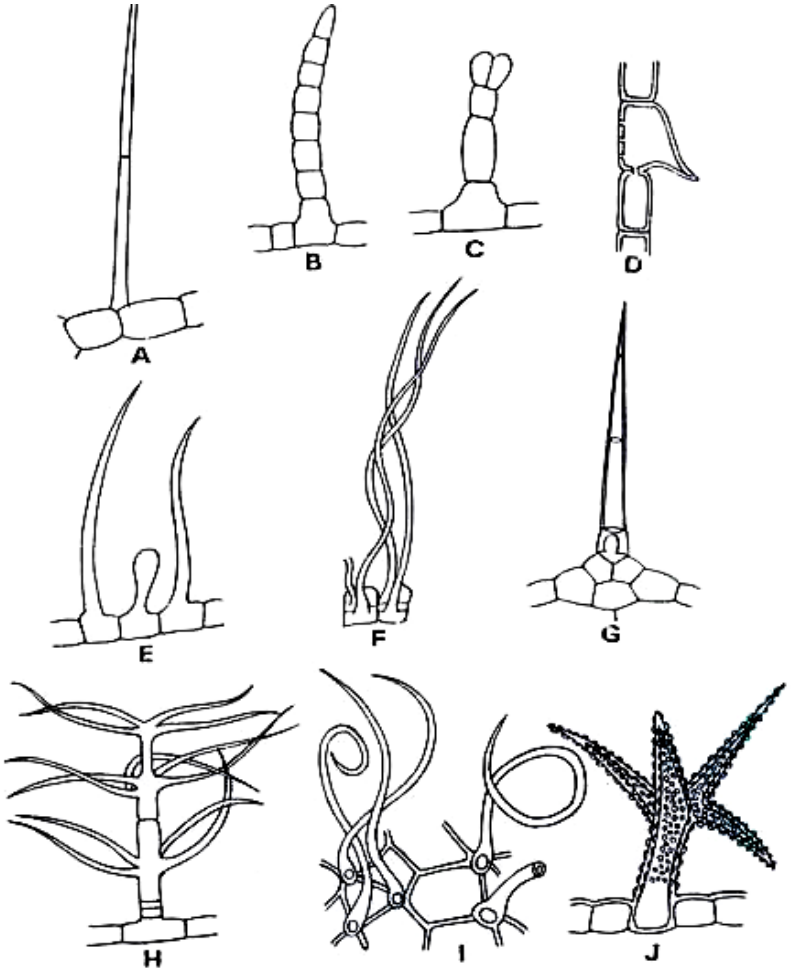


Fig. 37.21. Various types of trichomes (hairs). A, from petal of *Epigaea*; B, from leaf of *Coreopsis*; C, rom petal of *Phryma*; D, from leaf of *Avena*; E, from sepal of *Heliotropium*; F, from stem of *Onopordum*; G, from *Cucumis* leaf; H, from *Platanus* leaf; I, from *Rubus* fruit; J, from *Aubrietia* stem. (After E. & M)

(only visible pictures have to be drawn on practical copy)

Material and method –(1)Fresh and young leaves of Nerium sp.,Datura sp.,lantana camara, Adhatoda sp.and Solanum lycopersicum .

(2) Bright field microscope

(3) watch glass.

(4) slide and cover slip

(5) glycerine

Method –Upper epidermal layer of any sample leaf is peeled off and then placed it on the slide .If it is needed to take help of sharp blade ,by it we can cut the thin layer of upper epidermal cell .Then a drop of glycerine is fell on the epidermal cells which was kept on the slide previously . After that a cover slip is placed on it and observed under the bright field microscope .Under the microscope , different types of trichomes will be observed in different leaf samples which is very important taxonomic character for plant identification and also for pharmacognosy study.