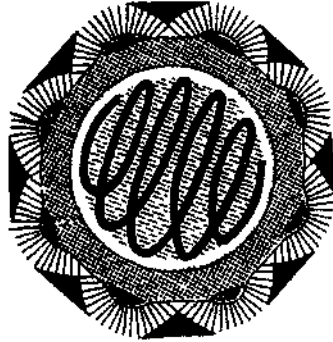




A Text Book of  
**Practical**  
**Botany 1**

**Bendre • Kumar**



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**Practical**  
**Botany -1**

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ALGAE, FUNGI, LICHENS, MICROBIOLOGY,  
PLANT PATHOLOGY, BRYOPHYTA, PTERIDOPHYTA,  
GYMNOSPERMS AND PALAEOBOTANY

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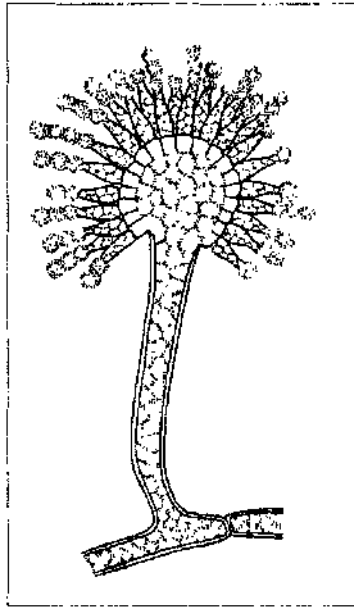
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# A Text Book of Practical Botany -1

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ALGAE, FUNGI, LICHENS, MICROBIOLOGY,  
PLANT PATHOLOGY, BRYOPHYTA, PTERIDOPHYTA,  
GYMNOSPERMS AND PALAEOBOTANY

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A Text Book of  
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Botany -1**

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# Introduction to Laboratory

# 1

Chapter

## Preamble

Science is a systematised study based on facts and observations. It involves curiosity, inquisitiveness and unbiased analysis. Most of the scientific work is done in a laboratory. It provides an opportunity to a person with scientific frame of mind to see and study various aspects of an object under observation. Hence, a biology student too is obliged to attend laboratory work-out with utmost sincerity, honesty and inquisitiveness.

## Laboratory Etiquette

The study of living things in laboratory requires that facilities provided are properly used.

One is expected to complete the assigned work within a specified time. This requires proper utilization and planning of time. One should, therefore, keep busy with own work and wherever necessary consult the teacher alone.

Laboratory provisions should be handled with utmost care. At the end of the laboratory period, working place should be left clean and in order.

Laboratory exercise to be performed should be read in advance and one is expected to arrive to the class theoretically prepared.

## Work Plan

1. Listen and understand the instructions and information given by teacher-in-charge.
2. Work out or observe the materials carefully.
3. Mount to prepare slides as per requirements.
4. Study the preparations or specimen carefully.
5. Draw suitable diagrams in a proper sequence and label them in your practical record.
6. Write down the observations sequentially and watch carefully if variations occur.
7. Get your work checked by teacher-in-charge and make necessary corrections.

## Necessary Instruments

The variety of instruments required depends upon the nature of work. It has, however, been found convenient to prepare a small kit in suitable containers such as a pencil box containing

1. a pair of forceps,
2. two fine, long handle, dissecting needles,
3. glass droppers,
4. good and sharp razor,
5. safety blade,
6. a fine hair brush,
7. a pair of sharpened pencils,
8. pencil eraser,
9. a clean and soft handkerchief and
10. practical record with cover file and spare pages, etc.

## Microscope

It is the most indispensable instrument in a biology laboratory, so much so that it comes to be called 'The primary instrument of the biologists'. It helps to increase the resolving power (property to distinguish objects lying very close as separate bodies) of human eye which fails to recognise objects lying closer between 0.01 to 0.25 mm.



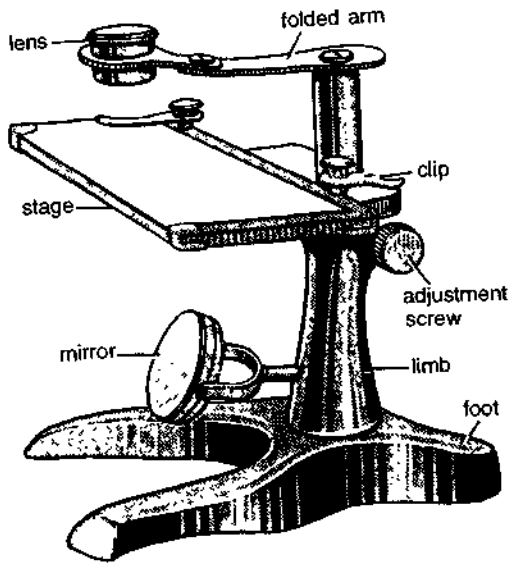


Fig. 1. A dissecting microscope.

Some common types of microscopes are listed below—

1. dissecting microscope,
2. compound microscope,
3. binocular microscope,
4. phase contrast microscope and
5. electron microscope.

Of these, dissecting microscope and compound microscopes are very commonly used by the students.

### [I] Dissecting microscope

It is used for dissection, specially during taxonomic studies, embryo separation, etc.

**Construction.** It consists of basal foot, a vertical limb, stage and a lens. The basal foot is a stand. The limb has an attached stage made of glass plate. A folded arm which can be moved vertically holds the lens. A mirror is attached at the base of the limb.

**Mechanical operation.** 1. Move the lens and adjust it over the object.

2. Illuminate the object suitably by adjusting the mirror.
3. Focus the object by using adjustment screw.

### [II] Compound microscope

It is one of the most commonly used and by far the most suitable microscope in the Botany Laboratory.

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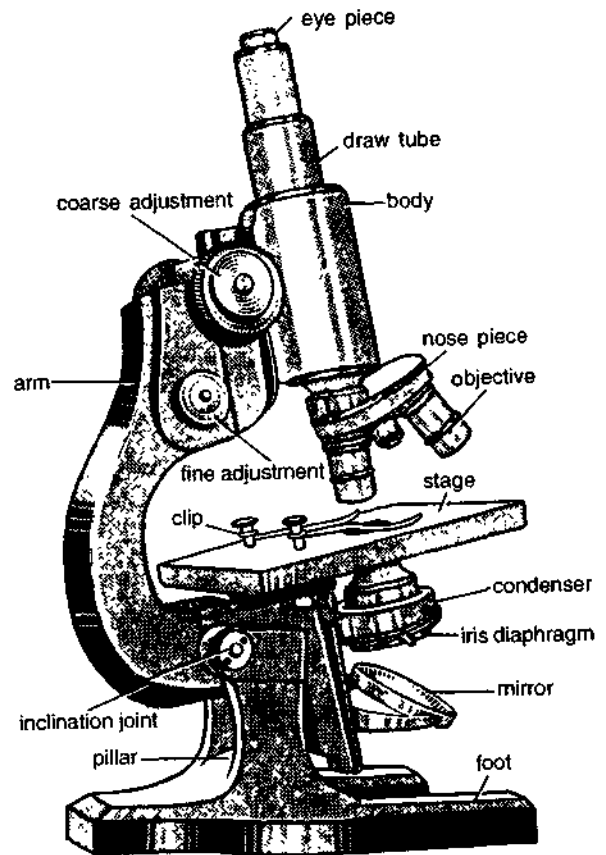


Fig. 2. Compound microscope.

At one time, it employs one ocular (eye piece) and one objective, in working position. As such, it is also known as monocular-mono-objective microscope.

**Construction.** The microscope is built around a strong basal foot and a vertical limb. The foot supports the vertical limb.

A round, rectangular or square stage is fixed to the limb. It is provided with spring clips to hold the slide in position.

A movable or fixed sub-stage is situated directly below the stage. It is provided with an iris diaphragm and condenser lens. Iris diaphragm is a wheel-shaped metal disc to regulate the aperture, through which light rays reach the condenser and are passed to an object. Condenser is a system of two or more lenses under the stage which receives parallel light rays from mirror and converge them at the level of stage.

A movable concave mirror is fixed at the lowermost part of the limb to focus a converging

cone of rays at the level of specimen. Whether day or artificial light is used as a source, concave mirror converges the light if there are no condensing lenses.

Body of the microscope is composed of a tube. At the upper end of the tube, is an ocular (eye piece) which can be changed for lower or higher values of magnifications. At the lower end of this tube is a revolving nose-piece with about three objectives viz. low power, high power and oil immersion. These magnifications range from 3.2x to 100x. The conventional low power objective is 10x.

Tube of the microscope is vertically movable with the help of coarse and fine adjustment screws on the limb, operated by a rack and pinion system. Coarse adjustment moves the tube rapidly, while fine adjustment screw does it gradually.

**Mechanical operation.** 1. Microscope is placed in maximum diffuse light. Direct sunlight is harmful for the eyes. The northern light is most suitable. If light source is artificial, filter (preferably blue coloured) is used.

2. Light is adjusted by turning the mirror towards the source of light and also by moving the sub-stage up and down, as well as with the help of iris diaphragm.
3. A prepared slide is placed on the stage. Object is adjusted just over the stage aperture.
4. The object is located and focussed with a low-power objective using coarse adjustment.
5. If higher magnification is desired, nose-piece is turned to next higher power. Fine adjustment can be used freely at this stage, while the use of coarse adjustment be avoided.
6. High power objective and subsequent higher powers are used only when object is properly mounted under coverslip.
7. The object should always be observed with both eyes open.

**Care.** 1. Before and after the use, all the lenses and metal parts including stage should be cleaned. The lenses are cleaned with tissue paper, muslin cloth or clean and soft handkerchief.

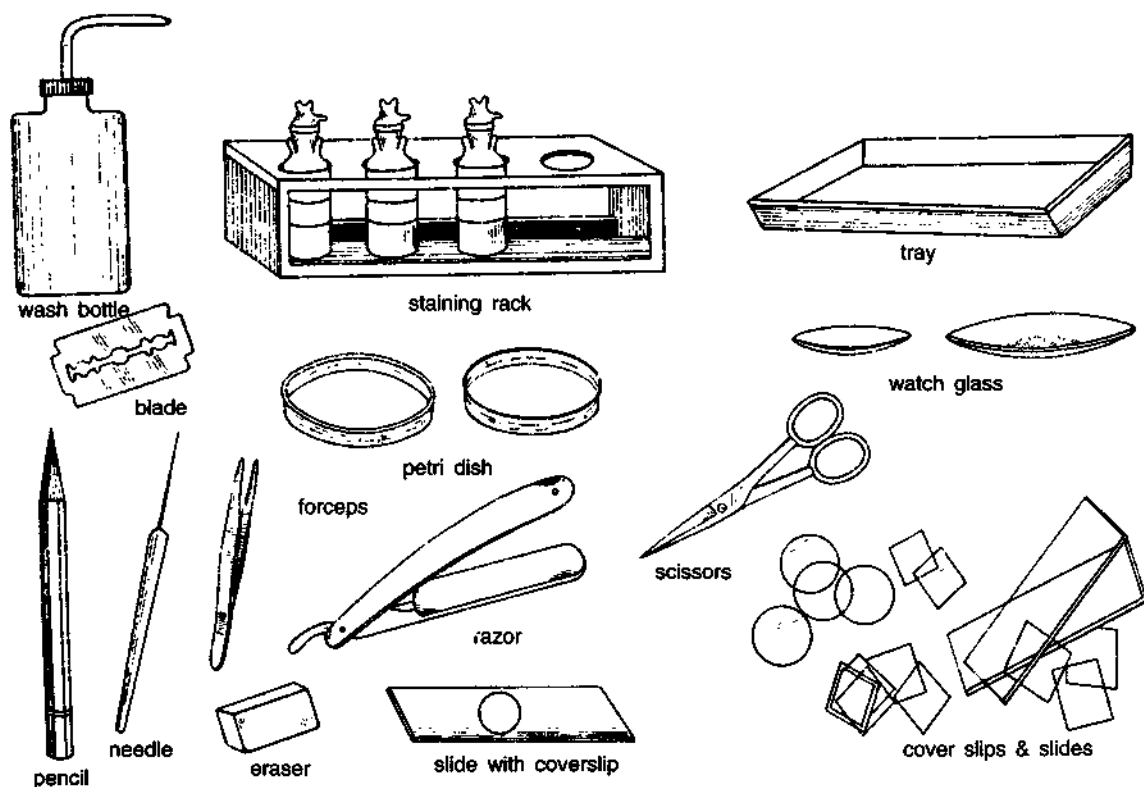


Fig. 3. Some laboratory provisions and necessary instruments.

2. Microscope is kept covered when not in use. Proper wooden box, plastic bags, bell jars or even a clean cloth can be used.
3. Objectives should not be ordinarily removed from the nose-piece.
4. Operating screws, condenser, iris diaphragm, mirror and stage or stage clips should always be handled carefully.

### Other Laboratory Provisions

Some other provisions available in the laboratory include staining rack, dropping bottles, slides, cover glasses, watch glasses, petri dishes, beakers, enamel trays, wash bottles, spirit lamp, hone, strop, dusters, etc. Some of these are described below—

1. **Staining rack.** It is mostly made of wood to hold the dropping bottles. The capacity of number of bottles per rack varies.

2. **Dropping bottle.** The stains, chemicals, mounting media, etc., are stored in these bottles. This glass bottle has a narrow mouth fitted with a slotted cock. Cock is provided with a beak that permits the liquid to flow out in drops.

3. **Slides.** The size of slides is mostly 3" × 1" (25 mm × 75 mm). It is about 1 mm thick. These are used to mount the material under study.

4. **Cover glasses.** The cover glasses are mounted on the object when the preparation is finally

ready. These may be either square or round shaped. The standard thickness of the coverslip is 0.17 mm.

### Fixing Agents and Preservatives

The plants or plant parts, collected fresh need to be immediately killed and subsequently preserved for a long time.

For this purpose, a few chemicals are used which do not cause any structural disturbance or distortion of the material. Carnoy's fluid, Formalin-aceto-alcohol, Formalin-propiono-alcohol, Randolph's modified Navashin fluid and Bouin's fluid are some of the common agents used.

Plants are generally fixed immediately after collection but these can also be fixed after bringing them to laboratory. The collected material must always be kept completely immersed in preservatives.

### Laboratory Techniques

#### [I] Section cutting

Sections of preserved material are cut in suitable planes for histological and ecological studies. Razor is suitable for cutting the sections in laboratory.

1. **Honing and stropping.** Razor should be sharp and free from nicks. Hence, it should be sharpened on a hone (fine-grit stone). Oblique,

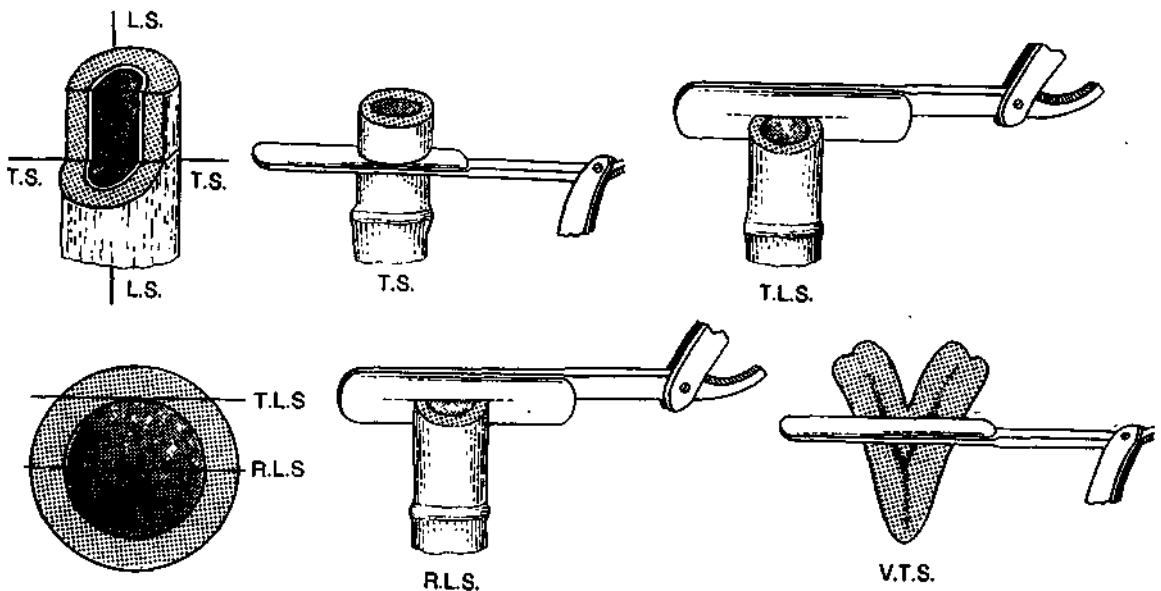


Fig. 4. Planes for section cutting.

uniform and slow strokes are carefully given to the razor with edge foremost on this stone.

After honing, uniform strokes are given on the strop (a smooth leather belt). The leather side of the belt is first slightly oiled and then razor is moved over. This should be done more frequently than honing, to maintain razor edge in good condition.

**2. Planes.** The following are a few commonly needed planes—

In case of cylindrical organs : (e.g., stems, roots, etc.).

**Transverse.** The section is cut by passing razor's edge at right angles to the longitudinal axis.

**Longitudinal.** The section is cut by passing razor's edge at right angles to the transverse axis. Two sections are possible in this plane.

(i) **Radial Longitudinal section (R.L.s.)** if it passes along one of the radii.

(ii) **Tangential Longitudinal section (T.L.s.)** if section is cut along one of its tangents.

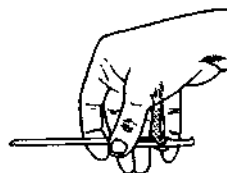
In case of dorsiventral organs (e.g. leaf, thallus of liverwort, etc.), transverse section is cut. It is known as vertical transverse section (being cut in vertical plane).

**3. Method.** Following steps would be useful for section cutting.

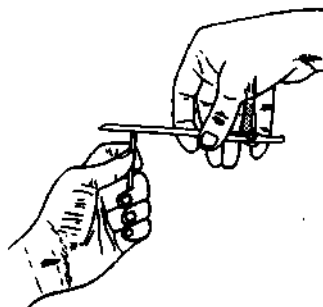
1. Soft, thin and small materials are placed in pith either by piercing a hole with a needle or by splitting it longitudinally with a blade. The pith used include carrot root and radish root, potato tubers, etc.
2. A razor must be held properly to cut the section. The handle and the blade of the razor should be at right angles to one another. The handle should remain free while the index finger is placed on the hooked end of the razor; 1st, 2nd and 3rd fingers pressed against the thick back edge of the razor and thumb against the milled surface of the thick shank of blade.
3. The material or the pith with embedded material is held between the thumb and the fingers of the left hand.
4. The material in the left hand and the razor's edge should form right angle.
5. The razor is now moved quickly over the material and the stroke is completed in one action only.



A



B



C

**Fig. 5. Method of section cutting.** A, holding the material, B, right way of holding the razor, C, holding the material and stroke of the razor.

6. More and more uniform strokes are used till desired quality and number of sections are obtained. Care is taken to keep the material and the razor flooded with water.
7. Sections float in water on the razor's edge. These are carefully lifted by a fine camel hair brush and then transferred to a watch glass containing water.
8. After the section cutting is over, the razor is tapped dry and cleaned without disturbing the edge. It is honed, stropped and encased.
9. The sections which float on water in the watch glass are considered to be thin.
10. These sections are lifted by a hair brush, placed on a slide in a drop of water and observed through microscope. A thin and uniform section is selected for staining.

## [II] Stains and staining

The selected sections need to be stained. The stains help to distinguish different tissues, cells or inclusions from one another by developing specific colours. Acetocarmine, Aniline blue, Crystal violet, Erythrosine, Hematoxylins, Fast green, Light green and Safranin are some of the commonly used stains.

**1. Specificity.** Most of the stains are specific in reaction and are purposely used so that definite structures or substances are stained. The following are some of the stains used for staining different structures.

<b>Achromatic figure</b>	<b>Cutinised cell wall</b>
Aniline blue	Crystal violet
Erythrosine	Erythrosine
Fast green	Safranin
Light green	<b>Callose</b>
<b>Cellulose cell wall</b>	Aniline blue
Aniline blue	<b>Chitin</b>
Delafield hematoxylin	Safranin
Fast green	<b>Proteins</b>
Light green	Safranin
<b>Lignified cell wall</b>	<b>Mitochondria</b>
Crystal violet	Crystal violet
Safranin	<b>Plastids</b>
<b>Suberised cell wall</b>	Crystal violet
Safranin	Iron hematoxylin
<b>Cytoplasm</b>	<b>Nucleus</b>
Aniline blue	Crystal violet
Erythrosine	Hematoxylin
Fast green	Safranin
Light green	<b>Chromosomes</b>
	Hematoxylin
	Safranin

**2. Single stains.** Safranin or fast green is used alone to stain filaments of algae, fungi, sections of bryophytes, spores of pteridophytes, pollen grains of gymnosperms, etc. Aniline blue or safranin is suitable for algae.

Following is the common method of staining.

1. The material is kept in a watch glass. A few drops of stain are added so that the material is immersed in the stain.
2. The material is allowed to remain so for a few minutes and allowed to take stain. The time required varies with materials.
3. After the stain is taken up, the excess of stain is washed off in water. The washing is repeated till stain stops coming out.
4. In some cases, excess stain is removed by acid water or acid alcohol if water alone fails to do so.

5. The stained material is ready for mounting. Fungi are stained in cotton blue as given below—

1. A drop of cotton blue (prepared in lactophenol) is placed on a slide.
2. Fungal hyphae is now placed in this drop.
3. The slide is run over the flame of the spirit lamp so that the stain is warmed up.
4. The preparation is now ready for mounting.

**3. Combinations.** Commonly two or more stains are employed wherever tissue differentiation is found. Combination of acidic and basic dyes of contrasting colours is of general use. This permits the distinction of woody tissue from non-woody tissue. The following few combinations are commonly recommended—

1. hematoxylin and safranin,
2. safranin and fast green,
3. safranin and aniline blue,
4. safranin and crystal violet and
5. crystal violet and erythrosine.

**4. Staining procedures.** There are two types of preparations—semi-permanent and permanent. The procedures differ in both the cases. These are given below.

*(a) For semi-permanent and temporary preparations.* Certain preparations are made for temporary use. The material is studied and the slide is then discarded. The method for staining them is given below.

1. The selected sections are transferred from watch glass containing water to another watch glass containing principal stain (e.g. hematoxylin, safranin or crystal violet).
2. The sections are allowed to remain in the stain for sometime (for about 4-5 minutes).
3. Excess amount of stain is removed by washing the sections repeatedly with water. (This can be seen under the microscope. The stain should be taken either by lignified or non-lignified tissues. Otherwise the section should be washed till the stain disappears from one type of tissue).
4. If destaining is not achieved, sections are washed with acid alcohol. In this case, further washing with water is necessary till traces of acid are removed.
5. This is followed by transfer of sections to a watch glass containing counter-stain

(e.g., safranin, fast green, erythrosine). This stain acts on the tissue more rapidly than the principal stain. Therefore, section is kept in this stain for short period (about a minute or two).

6. Excess of stain is removed by washing stained sections with glycerine (15-20%). The section should distinctly bring out demarcation between tissue system while preserving the colour of the stain.
7. The section is now ready for mounting.

(b) *For permanent preparations.* In certain cases preparations need to be stored permanently as a future record. The method of preparation followed is described below.

1. The section is first stained with principal stain (aqueous hematoxylin, safranin or crystal violet).
2. The section is then washed with water till no more stain dissolves and water remains colourless.
3. Section is passed through a graded series of alcohol for dehydration. A watch glass is filled with requisite amount of alcohol, (beginning with 30% alcohol) and the section is transferred to it. This watch glass should always be covered with another larger one. In order not to disturb the section, used alcohol is removed by glass dropper. All the 30% alcohol is replaced with 50% alcohol. This procedure is repeated till 70% of alcohol grade is reached.
4. At this stage, counterstain is employed (e.g. safranin, fast green or erythrosine prepared in 80% or 90% alcohol).
5. This stain acts quickly and as such section is washed immediately after the requisite time is over.
6. Destaining is done by washing sections with 90% or 100% alcohol.
7. The section is now transferred to absolute alcohol to complete the dehydration.
8. Clearing now begins with 25% of xylol (25 cc of xylol and 75 cc of absolute alcohol). The sections are gradually passed through xylol series of 25%, 50%, 70%, 90% and finally transferred to pure xylol. If dehydration is not complete, pure xylol turns white or turbid. At this stage section should be passed through reverse series.

9. Pure xylol is the last stage of clearing. Section is now ready for mounting.
10. Mounting is done in Canada balsam.

### Specific Schemes for Staining Combinations

(for temporary and semi-permanent preparations)

1. Hematoxylin & safranin	2. Safranin & fast green or aniline blue
Select a section ↓	Select a section ↓
Stain with hematoxylin ↓	Stain with safranin (for 4-5 minutes) ↓
Wash with water ↓	Wash with water ↓
Wash with ammonia water till stain turns blue (tap water is suitable if alkaline) ↓	Destain with acid alcohol if necessary ↓
Wash with water ↓	Wash repeatedly with water ↓
Stain with safranin ↓	Stain with fast green or aniline blue (for about a minute) ↓
Wash with glycerine ↓	Wash with glycerine ↓
Mount in glycerine	Mount in glycerine

### [III] Mounting an object

Mounting is necessary to properly position an object for clear view. Lactophenol, glycerine and glycerine jelly are used for temporary mounting while Canada balsam is used for permanent mounting.

**1. Mounting media.** Following are some of the common media.

(a) *Canada balsam.* It is a resin obtained from a conifer—*Abies balsamea*, most suitable for permanent slide preparation. The material to be mounted should come through alcohol (dehydration) and xylol (clearing) series.

(b) *Lactophenol.* It is a mixture of equal parts of phenol crystals, lactic acid, glycerine (sometimes two parts) and distilled water. Stains may be mixed with this medium (e.g. cotton blue in lactophenol used to stain fungi) or copper acetate is added to preserve green colour of the pigment.

### Specific Schemes for Staining Combinations (for permanent preparations)

1. Hematoxylin & safranin	2. Safranin & fast green 3. Crystal violet & erythrosine
Select a section (If necessary use mordant)	Aqueous safranin/ crystal violet
↓	↓
Stain in hematoxylin (If necessary destain with mordant)	Water change, until colourless
↓	↓
Wash in ammonia water or tap water	Dehydration with 30% alcohol
↓	↓
Dehydration with 30% alcohol	50% alcohol
↓	↓
50% alcohol	70% alcohol
↓	↓
70% alcohol	90% alcohol
↓	↓
Stain with safranin	Stain with fast green/ erythrosine
↓	↓
Destain with 70% alcohol	Destain with 90% alcohol
↓	↓
90% alcohol	Absolute alcohol
↓	↓
Absolute alcohol	Clearing or de-alcoholizing with 25% xylol
↓	↓
Clear with 25% xylol	50% xylol
↓	↓
50% xylol	70% xylol
↓	↓
70% xylol	90% xylol
↓	↓
90% xylol	Pure xylol
↓	↓
Pure xylol	Mount in Canada balsam
↓	
Mount in Canada balsam	

(c) **Glycerine.** Pure glycerine diluted to 15-25% is widely used. Semi-permanent and temporary preparations are mounted in glycerine.

(d) **Glycerine jelly.** Jelly is also used for mounting. It is made of gelatin 1 : glycerine 7 : water 6.

Warm the gelatin for two hours by adding water. Phenol (1%) is added later. Add crystals of safranin if desired. Allow the solution to cool and settle into jelly.

Many other mounting media like cedar oil, dammar, balsam, venetian turpentine and synthetic resins are also used.

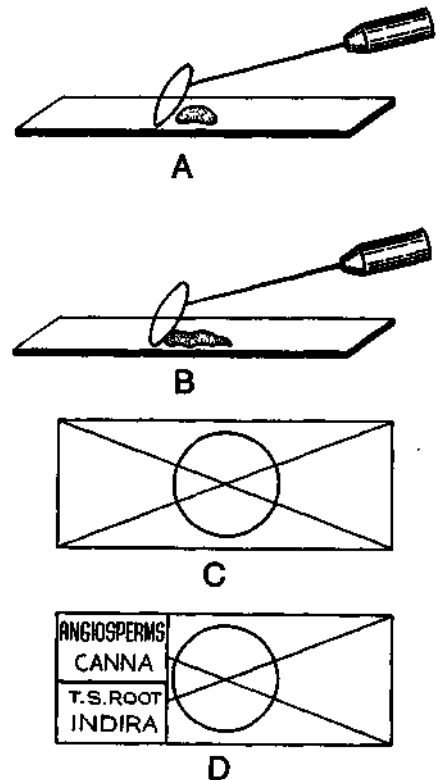


Fig. 6. Method of mounting coverslip.

2. **Care.** Following care should be taken during mounting—

- Object should be mounted in the centre of the slide. A simple method may prove suitable for this purpose. Take a piece of thick and white cardboard sheet larger than the size of the slide. Place the slide over it. Draw lines along all the four edges. Join all the four corner points diagonally by two lines. The point, where these two lines cross, gives the centre of the slide. While mounting an object, place the slide over this drawn sheet and an object on the central point.
- No air bubbles should enter the medium while mounting. This results in drying of medium and preparation is spoiled. To avoid air bubbles, touch one side of the coverslip to the drop of mounting medium on the slide. Support the coverslip by needle and lower it gradually before finally removing it.
- Use the necessary small quantity of mounting medium so that it does not flow on to the

slide. If so, use little lesser quantity for the next preparation. The extra amount can be soaked by touching a piece of blotting paper to the edge of the coverslip.

4. Preparation should be clean, hence the edges of slide and the coverslip alone should be held between the fingers.
5. Labels are pasted uniformly on one side of the prepared slide. It should carry the name of the division or generic and specific names, the part mounted and the section's plane. At the bottom be written, the name of the student who has prepared the slide.

**3. Sealing the coverslip.** Temporary preparations can be sealed with Canada balsam, gum, dammar, nail polish, etc. Such a preparation is called a semi-permanent preparation.

Sealing is done by simply painting the edges of the coverslip with sealing agent in such a way that the space between the slide and the coverslip gets filled with the agent. It will prevent the mounting medium from drying.

Similarly ringing table should be used for sealing the round coverslips. The use of Canada balsam for ringing is more convenient.

#### [IV] Maceration

This is a technique of separating individual cells from a group or tissue by dissolution of pectic middle lamella. There are three common methods.

**1. Jeffery's method.** The following are the steps—

1. Cut the fresh or dried material into small slices thinner than a tooth-pick.
2. Fill the test tube with material. Boil it in water till it settles down at the bottom indicating that it is free from air.
3. Replace water with the following macerating solution— (i) 10% Nitric acid  
(90 cc water + 10 cc nitric acid)  
(ii) 10% Chromic acid  
(90 cc water + 10 cc chromic acid)  
Mix both these acids in equal parts.
4. Heat the test tube filled with macerating fluid.
5. Stop heating as soon as the material becomes soft and pulpy.
6. Transfer the fluid to a watch glass.

7. Drain out all the macerating fluid. Wash the material repeatedly with water till all the traces of acids are removed.
8. The material is now stained with safranin and destained with water.
9. The pulp of the material is crushed with the glass rod and teased by a needle so that it is spread over the slide.
10. The material is mounted in glycerine or glycerine jelly.

**2. Harlow's method.** The following are the steps—

1. Sliced and boiled material is treated with chlorine water for two hours.
2. It is then washed with tap water.
3. The material is now boiled in sodium sulphate for about 15 minutes.
4. The liquid is transferred to a watch glass.
5. The material is now washed repeatedly with water.
6. It is teased with needle or crushed with glass rod.
7. The teased material is evenly spread on the slide, stained in safranin and then mounted in glycerine or glycerine jelly.

**3. Schultze's method.** The following are the steps—

1. Material is sliced and boiled in a test tube filled with water.
2. The tube is now filled with concentrated nitric acid, to which a few crystals of potassium chlorate are added.
3. The test tube is heated slowly and gradually till the material is bleached white.
4. The liquid is then transferred to watch glass and drained out leaving only the material.
5. The material is now washed with water.
6. Later it is teased or crushed, till individual cells appear isolated.

#### [V] Peelings

The removal of leaf epidermis, to study the number, arrangement, distribution and structure of stomata, is called peeling. The method consists of breaking the leaf irregularly with a force. This easily separates a little part of the lower epidermis which remains protruding on the lower surface of the leaf. It is



pulled out so that a long ribbon or strip of lower epidermis gets removed. If lower epidermis does not separate easily, a needle or forceps is inserted, and a small part is first slowly broken. This can now be held in hand and considerably large strip is pulled apart.

The stripped lower epidermis is stained in safranin and washed. It can be mounted in glycerine or glycerine jelly. If permanent preparation is desired, normal procedure of dehydration and clearing is followed before mounting it in mounting medium.

### [VI] Smearing

Smearing is used to study the chromosomes. The method consists of spreading the cells in a single layer. The cells are smeared at a stage when they are in the process of cell division. This permits the study of chromosome structure and various stages of cell division. Pre-requisite for such studies is the killing of dividing tissues at a proper stage of cell division and selection of material where cells are not firmly united with one another by middle lamellae. Microsporocytes of *Trillium* spp., *Lilium* spp. and *Oenothera* spp., as well as anthers of *Tradescantia* spp., *Triticum* spp. and *Nicotiana* spp. and root tips of onion, *Ficus*, etc. fixed at appropriate time are widely used for smear preparations.

**1. Technique.** The following are the steps—

1. Slides should be perfectly clean for preparation of smears. In order to do so these are immersed in sulphuric acid potassium bichromate mixture or concentrated nitric acid for a long time.
2. Slides are thoroughly washed with running water and finally dried with absolutely clean cloth, free from dust and lint.
3. Fresh anthers dissected out from the buds are placed in the centre of slide. The anthers on the slide are crushed with scalpel or another clean slide.
4. Slide is now inverted over a petri dish containing killing fluid (most suitable being Randolph modified Navashin fluid), in a way that smeared surface comes in contact with the fluid. It should be allowed in this position for about 10-15 minutes.

5. Slide is now inverted with smeared side upward. It is now ready for staining. It may also be stained immediately without immersing in killing fluid.

**2. Staining procedure.** The method described below is called Belling's iron acetocarmine method. The slides are stained in the following way.

1. A few drops of acetocarmine are placed on the smeared material or unsmeared anthers are kept on slide in a drop of acetocarmine. After a few minutes, stain is replaced with a fresh drop of stain.
2. At this stage, anthers are crushed and large pieces and debris are removed.
3. Slide is gently heated over a flame, cover glass is placed on the material and uniform pressure is applied on the material by placing blotting paper on the cover glass and then pressing it.
4. Slide is immediately sealed with melted wax.

Another simple method is followed where anthers are smeared on the cover glass. It is then inverted on the slide with a drop of acetocarmine. Cover glass is sealed with slide by melted wax.

### [VII] Squash

This technique is also useful in the study of cell division especially mitosis and the chromosome structure. Root tips give the best results. For this purpose allow the onion bulbs to grow in bottle filled with water. If the lower root portion of the bulb touches the water, it quickly sends forth large number of roots. Cut the root tips and fix them.

1. Place the fixed root tip in a drop of 45% acetic acid.
2. Place a cover glass over the tip and diffuse acetocarmine.
3. Tap and apply uniform pressure over the cover glass.
4. The squash preparation is ready.

### [VIII] Micrometry

(Measurement by means of microscope)

This is the procedure used to measure the size of microscopic objects like cell, spore, pollen grain, etc. The method consists of using a calibrated ocular micrometer (a glass disc with engraved scale). The calibration is done by comparing ocular with stage

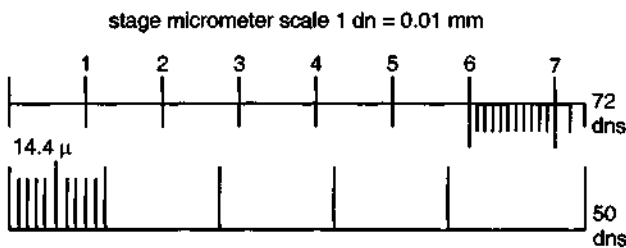


Fig. 7. Matching ocular micrometer with stage micrometer.

micrometer (a slide bearing an engraved scale of known values). The stage micrometer is usually ruled into tenths and hundredths of a millimeter (scales in hundredths of an inch are also obtainable). Each of the 100 parts of stage micrometer scale represents 0.01 mm or  $10\mu$  ( $1\text{ mm} = 1000\text{ microns or } \mu$ ).

**1. Calibration of ocular micrometer.** The calibration is done as follows.

1. Place the ocular micrometer inside the eye piece by unscrewing the upper lens.
2. The stage micrometer slide is now placed on the stage of the microscope and focussed to observe the scale.
3. The stage micrometer scale is moved in such a way that it lies by the side of the scale of ocular micrometer when focussed.
4. Now compare and count the divisions on both micrometers to find out the number of divisions where both scales are equally opposite.
5. For example when  $45\times$  objective and  $10\times$  eye piece are used, divisions of ocular micrometer are found equal to 72 divisions of stage micrometer.

6. Calibrate the ocular micrometer as given below.

Stage micrometer scale :

$100\text{ dns} = 1\text{ mm} (=100\mu\text{ or microns}^*)$

$1\text{ dn} = 0.01\text{ mm} (=10\mu\text{ or microns})$

If, 50 dns (ocular micrometer)

$= 72\text{ dns (stage micrometer)}$

then, 50 dns (ocular micrometer)

$= 0.72\text{ mm} (=720\mu\text{ or microns})$

therefore, 1dn (ocular micrometer)

$= 0.14\text{ mm} (=14.4\mu\text{ or microns})$

**2. Measurement of objects.** The following method is useful in actually determining the size of objects. An example is given below.

1. Thus as in above example (when objective  $45\times$  and eye piece  $10\times$  are used), each division of ocular (micrometer) would measure the distance of  $14.4\mu$  or microns.
2. Now remove the stage micrometer and place a slide with object to be measured.
3. Use oculometer (micrometer) to measure the width of a bacillus or diameter of a pollen grain or a fungal spore. For example a fungal spore measures 2 divisions.
4. The diameter of a fungal spore would be  $(2 \times 14.4\mu)$   $28.8\mu$ .

The length, breadth, diameter, etc. of different structures can be measured in this way.

## Record of Work

After the preparations are ready, these should be carefully observed, salient features noted and drawn on a practical record sheet. The following suggestions would prove useful.

1. Always use a sharp and pointed pencil for thin and uniform lines.
2. Punched holes should be on the left hand side of the drawing sheet.
3. Diagrams of the entire plant or its various aspects are drawn on the same page. The diagrams of unrelated specimens should in no case be drawn on the same page.
4. The sequence of the diagrams should always be—external features, anatomy and then reproduction.
5. For anatomical studies an outline diagram followed by a cellular sketch of its suitable sector are drawn one above the other on the same page.
6. All the parts of the diagram must be labelled. Capital letters are used for labelling. The labels are arranged one below the other in a row.
7. Labelling lines should never cross one another. Beautification and shading are not required until specific effects are to be produced.
8. Every diagram must have caption at its bottom (e.g. T.s. stem).
9. Date is written in the left hand corner of the page.
10. Classification and name of the plant are given in the right hand corner of the sheet.

\*One millimeter =  $1,000\mu$ .

$\mu$ , this Greek letter is an abbreviation for micron.

11. The description is written either on the reverse side of the drawing sheet or on a new facing page.
12. During description only technical terms are used. The points of identification are added in the end.
13. Anatomical studies are described as others. A section should be described starting from epidermis to the central region; give thickness of layer (how many cells deep), shape and size of the cells constituting it. Also give in details of the structure of stele and vascular bundle.

### Collection

Field work is one of the most essential part in the Botanical study. It permits to come across many types of plants, otherwise not seen and available in the laboratory. It is, therefore, advisable to go round many localities and explore their vegetation. Organised excursions or outings, led by experienced persons, add to the knowledge of common plants in nature.

While on a collection trip, local or outstation, following things are to be carried along.

**1. Containers.** For packing the collected material, preferably carry plastic unbreakable containers or polyethylene bags.

**2. Preservatives.** Formalin-Acetic-Alcohol (FAA) or Alcohol 70% or Alcohol 90%, and/or Formalin 6%-10%.

**3. Other requirements.** Scalpel, knife, blade, forceps, pencil, paper, a hand lens, a bag or vasculum for keeping plants or plant press with many newspapers or blotting papers.

After collecting the plant, it should be immediately killed and preserved or pressed to avoid its rotting and dehydration. Plants are either sprinkled or immersed with a little of the killing agent at the spot. On return to the laboratory collected material should be transferred to new and suitable containers with fresh preservative. The plants should be completely immersed in the preservative.

A few plants e.g. filamentous algae, fungi, reproductive parts of bryophytes, fertile parts of pteridophytes and different parts of gymnosperms, if collected in large quantities, are preserved in

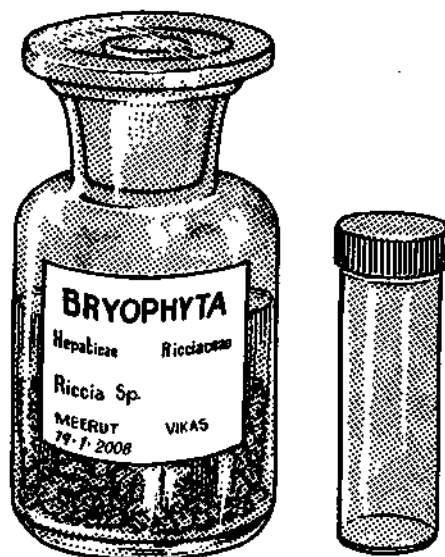


Fig. 8. Collection bottles.

containers. But if material (except a few algae and fungi) are collected in lesser quantities a herbarium sheet is prepared. Even if large quantity of such plants is available, one plant with fertile parts be preserved in the form of a herbarium sheet, while others should be packed in a container.

Every tube should be labelled. It is desired to write the name of the specimen, place and date of collection. The place of collection and date should also be written on a small piece of white card with a pencil, on the spot and inserted in the container. On return to laboratory, material is identified with the help of standard books. A label bearing name of the division and class to which the material belongs, the name of the material, date and place of collection and also the name of student is pasted on the container. All the containers should be of uniform size as far as possible.

### Herbarium

A collection of dried plant specimen, mounted on sheets is known as herbarium. Freshly-picked specimen are dried and pasted on mounting paper of regulation-sized herbarium sheets. The purpose of such a collection is to study the vegetation of a locality and maintain its record.

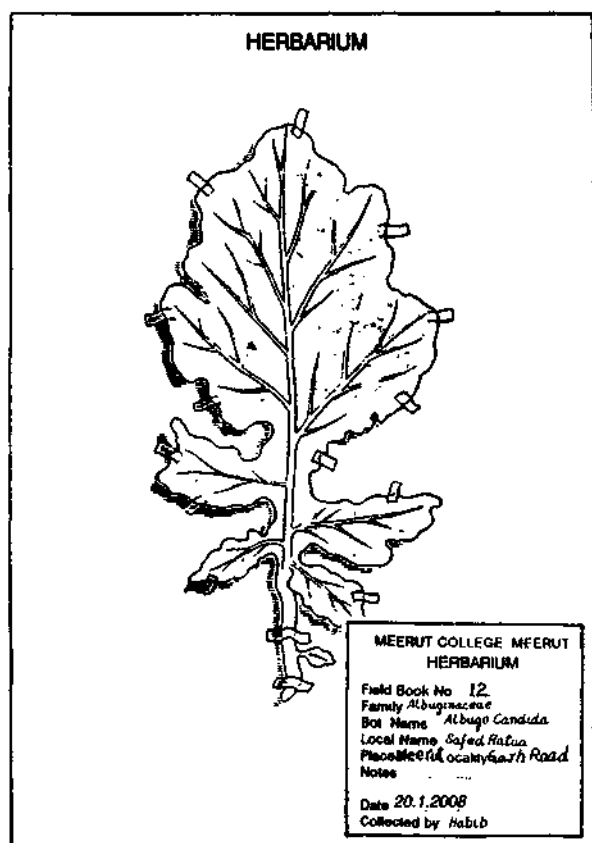


Fig. 9. A typical herbarium sheet.

### [I] Preparation of herbarium sheets

**1. Equipment.** On excursion, for the collection of plants, several items required to be carried include—

1. Trowel or pick,
2. Collecting can (vasculum) or field plant press,
3. Heavy laboratory plant press,
4. Blotting papers or newspapers,
5. Collecting sheets,
6. Mounting sheets,
7. Gum, gummed tape, labels, notebook, pen and pencil, etc.

Trowel or pick is used to dig out the plant as a whole, wherever possible. A light-weight field press is most practical. It is made by taking two pieces of plyboard or heavy binder's board of 12" × 17" size. These are held together by two pieces of heavy cord or straps tied or buckled together and press can be carried over the shoulders. A heavy plant press carries sheets of size at least 11½ × 17 inches. It is made of iron and tied and

tightened by iron chain and screws. This is used for pressing specimen after they are brought to the laboratory. Vasculum may be used in case only a small number of plants are to be brought back.

**2. Collection.** Collected plants are placed in the collecting sheets. The most practical size is 16½ × 23 inches; when folded 16½ × 11½ inches. Old newspapers serve this purpose to an appreciable extent and a large supply should always be included in the kit.

A specimen collected should represent root, stem, leaves and flowers. The plants are placed between the sheets or newspapers in such a way that relation between different organs is maintained. Herbaceous plants, 2 feet or less higher, may be collected entire. These can be bent to V or N shape whenever necessary. The most desirable is to collect a branch, about one foot high, containing leaves and flowers. In cases, where entire plant or branch cannot be folded to the size of herbarium sheet, only reproductive and fruiting parts and a stem with a few leaves are collected.

Delicate reproductive parts collapse even if pressed fresh. These can be pressed perfectly by applying bits of moist paper to the fresh reproductive structures and spreading them when plants are placed in the press. If parts of the herbaceous plant are thick and difficult to dry, split them before placing on the collecting sheet.

Water plants collapse if dried by usual method. These should be rolled up in wet paper when in the field and brought to the laboratory. On return to the laboratory, these plants are placed in water and floated out on sheets of white paper. The sheets are taken out of water carefully, so that the various parts do not cohere. The white sheets are placed in the blotting paper and then dried as usual.

After specimen has been collected and placed in collecting sheet, it is kept in plant press. This collecting sheet be placed in between blotting papers, one on either side.

While on collection it is important to note date, locality, habitat, height, method of branching, colour of reproductive parts, common name, etc. This should be noted separately in a field-book.

**3. Pressing.** The collecting sheets should be transferred to a heavy laboratory press. It must be remembered that specimen would acquire the same shape, as on collecting sheet, after pressing. The

press is securely tightened. It may also be equally useful if field press is kept under heavy weight. The press should be placed in a warm, well-aired place to dry.

After 24 hours, press is taken out and opened. The old newspapers and blotting sheets are replaced by new unused ones. At least such 3-4 changes are given at an interval of 2-3 days. An average specimen takes about a week for complete drying. Sometimes to hasten the process of drying, plant press may be placed near the source of heat.

**4. Mounting.** The specimen are ready for mounting once they are completely dry. The standard size of the sheet is  $16\frac{1}{2} \times 11\frac{1}{2}$  inches. However,  $16 \times 10$  inches size also has been used. The paper should be of good weight and not thin and flexible. The quality should be so, that it does not turn yellow even with a considerable lapse of time.

To mount, one of the following methods would be found convenient.

1. The gum is spread on a glass plate and specimen is laid on it. As soon as all the parts come in contact with gum, it is lifted and then placed in a position on a mounting sheet.
2. The specimen is inverted and painted with gum by a brush and then transferred to a mounting sheet.
3. The specimen is placed on a herbarium sheet and small strips of gummed tape or cellulose tape are pasted at suitable places, so that most of the part remains loose.

After mounting the specimen, a label is pasted in the right hand lower corner of the sheet. This carries information regarding botanical name of the plant, common name, date, collector's name, place of collection etc.

**5. Arrangement of sheets.** The sheets, are finally arranged in accordance with standard classification (preferably Bentham and Hooker's for Angiosperms or the most accepted ones for other groups of plants). The sheets are arranged into groups according to species, genera, families, classes, orders, series and sub-divisions, etc. Each group is placed in a separate envelope, slightly larger than the herbarium sheets (e.g.  $17 \times 12$  or  $17 \times 11$  inches). Each of such envelopes must be labelled and a proper index be written or pasted over it.

**6. Care of sheets.** Herbarium sheets are often attacked by museum pests, fungi, etc. To guard against them, specimen are fumed with carbon bisulphide, 3-4 times a year. Mounted specimen may also be treated with mercuric bichloride or copper sulphate. To prevent them from attack, powdered naphthalene balls or gamaxene powder be also spread from time to time. This ensures durability and long life of the herbarium sheet.

### [III] Some important herbaria

There are many institutions all over the world which house collections of herbarium sheets. The arrangement is mostly based on either Engler's, Bessey's or Bentham and Hooker's system of classification. A list of a few well-known herbaria of the world is given here.

**1. Herbarium Nationale' de Histoire Laboratories de Phanerogamie, Paris, France.** National institute, established in 1635, more than 5,000,000 specimens, mostly phanerogams and vascular cryptogams.

**2. Herbarium of Botanisches Institute de Universtate, Kiel, Germany.** Governed by Kiel University, established in 1875, more than 90,000 specimens.

**3. Royal Botanic Gardens, Herbarium, Kew, Great Britain.** Government institution of Royal Botanic Gardens, established in 1841, world-wide collection, more than 6,00,000 herbarium sheets.

**4. British Museum of Natural History, London, Great Britain.** A private body, established in 1753 more than 4,000,000 specimens, relating to all the plant groups.

**5. Gordon College Herbarium, Lahore, Pakistan.** A private mission, established in 1893, ferns of the Himalayas and flowering plants of Punjab, Kashmir, Afghanistan, Baluchistan, Pakistan and Nepal, numbering about 55,000.

**6. U.S. National Museum (Botanical Department), U.S. Smithsonian Institution, Washington, U.S.A.** Independent government agency, founded in 1868, world-wide collection of all groups, more than 2,700,000 specimens.

**7. U.S. National Arboretum, Herbarium, Washington, U.S.A.** Federal agency, established in

1934, 37,000 vascular plants of economic importance, cultivated and woody.

**8. Herbarium of the Department of Systematic and Plant Geography of the Botanical Institute of the Academy of Sciences of Leningrad, Russia.** Medical garden, established in 1714, herbarium added in 1823, state owned institution, representing flora of U.S.S.R., northern Asia and world-wide collections, more than 5,000,000 herbarium sheets.

**9. Indian Botanic Garden, Herbarium, Calcutta, India.** Established in 1787, government agency, more than 1,000,000 specimens, representing phanerogams and ferns of India and adjacent region.

**10. Herbarium Blatter, St. Xavier's College, Bombay, India.** Private body, representing more than

100,000 specimens from western India and collection of fungi established by Mundkur.

**11. National Botanic Research Institute, (formerly known as National Botanic Garden) Lucknow, India.** C.S.I.R. body, established in 1948, more than 40,000 specimens.

Besides these herbaria, many well-reputed collections exist; some of them being—Botanical Gardens Herbarium, Singapore; National Botanic Gardens Herbarium, Kirstenbosch, South Africa; Herbarium Bogoriensis, Bogor, Indonesia; Botanical Museum and Herbarium, of the State University of Utrecht, Netherlands; Forest Research Institute, Herbarium, Dehradun, India; Botanical Survey of India Herbarium, Pune, India; Indian Agricultural Research Institute, Botany Division Herbarium, New Delhi, India.

# 2

## Chapter

# Algae

### Preamble

Algae and Fungi were first included in the group cryptogamia, established by Linnaeus. Later, this group was divided into Thallophyta, Bryophyta and Pteridophyta. Of these, Thallophyta included Algae and Fungi. The group Thallophyta shows following characteristics.

- (1) Absence of differentiation into stem, root and leaves.
- (2) Sex organs generally unicellular, if multicellular, sterile envelope or jacket is absent.
- (3) Zygote does not develop into multicellular embryo while inside the female sex organ.

Thallophyta is one of the largest group of Plant Kingdom. The members of this group are found in almost all the types of habitats. Though algae and fungi are customarily placed under Thallophyta, they are very different from one another, in their morphological features, metabolism, reproduction and life histories.

Algae and fungi differ from one another mainly due to presence of chlorophyll in the former and its absence in the latter. This makes algae autotrophic and fungi remains heterotrophic. Reserve food in Algae is the form of starch while fungi store glycogen. Algal cell walls are made of cellulose and those of fungi are composed of chitin.

The present day delimitation of algae is due to A.L. de Jussieu (1789). Algae is mostly found in water though some are terrestrial and some are even parasitic (*Cephaleuros virescens*, a green alga causes Red Rust of Tea). They are as small as bacteria and as large as *Macrocystis* (a brown alga, 196 feet long). Algae show a great range of thallus structure—as simple as a single cell of *Chlamydomonas* and as complicated as an internally differentiated kelp (brown algae).

The algal cells are similar to those of higher plants. The characteristic colour of algae is due to specific pigments present in plastids.

Algae reproduce by vegetative, asexual and sexual methods. Sexual reproduction varies from simple isogamy to advanced oogamy. Few groups of algae also exhibit a distinct and well-defined isomorphic and heteromorphic alternation of generations.

Algae have been variously classified by numerous phycologists, their views always differing. The most simple and practical classification was proposed by British phycologist F.E. Fritsch in 1935.

### Distinguishing Characters of Taxa

#### SUB-DIVISION. ALGAE

- (1) Thallus simple
- (2) Chlorophyll present
- (3) Cell wall of cellulose

#### CLASS I. CHLOROPHYCEAE

- (1) Grass green plastids
- (2) Starch is reserve food
- (3) Flagella of reproductive structures equal in length.

#### Order 1. Volvocales

Thallus with motile flagellated cells.

##### Family 1. Chlamydomonadaceae

- (1) Thallus unicellular
- (2) Contractile vacuoles present

Example. *Chlamydomonas*

##### Family 2. Volvocaceae

- (1) Thallus colonial
- (2) Cells in a colony forming a flat plate

Examples. *Pandorina*, *Eudorina*, *Pleodorina*, *Volvox*

#### Order 2. Chlorococcales

- (1) Cells single and non-motile

- (2) Cells uninucleate
- (3) Reproduction by zoospores or autospores

Family 1. **Chlorellaceae**

- (1) Cells single, if united do not form a definite colony
- (2) Reproduction by autospores

Example. *Chlorella*

Family 2. **Hydrodictyaceae**

- (1) Cells united to form coenobe
- (2) Reproduction by zoospores and biflagellate gametes

Examples. *Pediastrum*, *Hydrodictyon*

Family 3. **Coelastraceae**

- (1) Reproduction by autospores
- (2) Autospores apposed to one another at the time of liberation

Example. *Scenedesmus*

Order 3. **Ulotrichales**

- (1) Thallus simple or a branched filament
- (2) Cells uni- or multinucleate
- (3) Single chloroplast with 1 or more pyrenoids

Family 1. **Ulotrichaceae**

- (1) Unbranched filaments
- (2) Cell walls not articulated
- (3) Cells uninucleate

Example. *Ulothrix*

Family 2. **Ulvaceae**

- (1) Thallus expanded, 1 or 2 cells thick
- (2) Cells uninucleate with laminate cup-shaped chloroplast

Examples. *Ulva*, *Enteromorpha*

Order 4. **Cladophorales**

- (1) Branched or unbranched filaments
- (2) Cells cylindrical and multinucleate

Family 1. **Cladophoraceae**

- (1) Cells more than eight times longer than broad
- (2) Chloroplasts do not form distinct transverse bands

Example. *Cladophora*

Order 5. **Chaetophorales**

- (1) Plant body heterotrichous
- (2) Hair or setae present

Family 1. **Chaetophoraceae**

- (1) Filaments branched, branches free from one another or pressed together forming pseudoparenchymatous thallus
- (2) Terminal cells modified into a long colourless setae (hair)

- (3) Cells uninucleate with a single laminate and parietal chloroplast.

Examples. *Draparnaldiopsis*, *Fritschiella*

Family 2. **Coleochaetaceae**

- (1) Vegetative cells with setae
- (2) Sexual reproduction oogamous

Example. *Coleochaete*

Order 6. **Oedogoniales**

- (1) Filaments branched or unbranched
- (2) Cell division resulting in 'cap' formation
- (3) Chloroplast reticulate
- (4) Zoospores and antherozoids multiflagellate

Family 1. **Oedogoniaceae**

Single family

Example. *Oedogonium*

Order 7. **Zygnematales**

- (1) Absence of flagellated reproductive cells
- (2) Sexual reproduction by conjugation

Family 1. **Zygnemataceae**

- (1) Filaments unbranched
- (2) Chloroplast parietal and ribbon-shaped or single or two axial chloroplasts.

Examples. *Spirogyra*, *Zygnema*

Family 2. **Desmidiaceae**

- (1) Cells composed of two semi-cells
- (2) Conjugating cells have chloroplast escaping from surrounding walls as they unite to form zygospores

Example. *Cosmarium*

Family 3. **Mesotaeniaceae**

- (1) Cells made of single piece and without pores
- (2) Conjugating cells do not transfer contents from one cell to another

Example. *Netrium*

Order 8. **Siphonales**

- (1) A single, multinucleate and tubular cell (coenocyte) represents the thallus
- (2) Chloroplasts many and discoid

Family 1. **Caulerpacae**

- (1) Thallus differentiated into rhizome, rhizoids and aerial folliar shoots, macroscopic
- (2) Internally shows the presence of trabeculae

Example. *Caulerpa*

Family 2. **Codiaceae**

- (1) Thallus freely branched and tubular
- (2) Sexual reproduction anisogamous and gametangia distinct

Example. *Codium*



**Order 9. Charales**

- (1) Thallus differentiated into nodes and internodes
- (2) Characteristic sex organs—globule and nuclei

**Family 1. Characeae**

Single family

Examples. *Chara*, *Nitella***CLASS II XANTHOPHYCEAE**

- (1) Chromatophores yellow-green
- (2) Photosynthetic reserves—oil droplets
- (3) Motile cells with unequal flagella

**Order 1. Heterosiphonales**

- (1) Thalli multinucleate, unicellular and siphonaceous

**Family 1. Botrydiaceae**

- (1) Thallus unicellular, multinucleate, vesicular
- (2) Zoospores biflagellate
- (3) Sexual reproduction isogamous

Example. *Botrydium***Family 2. Vaucheriaceae**

- (1) Thallus branched, coenocytic, tubular and filamentous
- (2) Zoospores multiflagellate
- (3) Sexual reproduction oogamous

Example. *Vaucheria***CLASS III. BACILLARIOPHYCEAE**

- (1) Chromatophores golden-brown or yellow with or without pyrenoids
- (2) Cell wall made of two silicified overlapping halves
- (3) Food reserve oil
- (4) Reproduction mostly by simple cell division

**Order 1. Centrales**

- (1) Valves circular, ornamentation radial or concentric
- (2) Statospores or microspores formed
- (3) Auxospores formed by oogamy

Example. *Melosira***Order 2. Pennales**

- (1) Valves bilaterally symmetrical, ornamentation bilateral
- (2) Valves always with raphe
- (3) Statospores or microspores never formed
- (4) Auxospores formed by oogamy

Example. *Pinnularia***CLASS IV. PHAEOPHYCEAE**

- (1) Yellowish-brown chromatophores
- (2) Laminarin and mannitol are reserve food products

(B-14)

- (3) Reproductive cells with two unequal, lateral flagella

**Order 1. Ectocarpales**

- (1) Thallus filamentous
- (2) Growth trichothallic
- (3) Reproductive organs—unilocular and plurilocular sporangia
- (4) Isomorphic alternation and generation

**Family 1. Ectocarpaceae**

- (1) Thallus monoaxial, branched; branches uniseriate
- (2) Uni- and plurilocular sporangia, terminal or intercalary

Example. *Ectocarpus***Order 2. Laminariales**

- (1) Sporophytes large, parenchymatous
- (2) Sporangia in sori, on stipe and blade
- (3) Gametophytes microscopic and dioecious

**Family 1. Laminariaceae**

- (1) Sporophytes differentiated into holdfast, stipe and blade
- (2) Blade simple or digitate
- (3) Paraphyses hyaline or with colourless appendages
- (4) Sporangia on both the surfaces of blade

Example. *Laminaria***Order 3. Fucales**

- (1) Plants parenchymatous with complex morphological and anatomical differentiation
- (2) Medulla filamentous
- (3) Asexual reproduction absent
- (4) Sex organs in conceptacles

**Family 1. Fucaceae**

- (1) Axes subterete to alate with midrib but not foliar
- (2) Vesicle when present intercalary
- (3) Oogonia with eight oospheres

Example. *Fucus***Family 2. Sargassaceae**

- (1) Axes terete, bearing distinct foliar organs
- (2) Vesicles usually present, lateral or immersed in the terminal branchlets
- (3) Branching of the thallus radial to the central axis

Example. *Sargassum***Order 4. Dictyotales**

- (1) Plants parenchymatous, less differentiated
- (2) Branching often dichotomous

- (3) Growth by apical cells
- (4) Asexual reproduction by tetraspores
- (5) Isomorphic alternation of generation

Family 1. **Dictyotaceae**

- (1) Gametophytes dioecious/monoecious
- (2) Oogonia and antheridia in definite sori
- (3) Sperms motile and eggs non-motile
- (4) Sporophyte with tetrasporangia

Example. *Dictyota*

CLASS V. **RHODOPHYCEAE**

- (1) Chromatophores pure red to dark purple
- (2) Photosynthetic reserve—Floridian starch or floridoside
- (3) Male gametes and female gametes non-motile and non-flagellated
- (4) Female reproductive organ with a receptive organ—trichogyne
- (5) Post fertilization product is called cystocarp

Sub-class 1. **Bangioideae**

- (1) Thallus simple, growth intercalary
- (2) Pit connections not well marked
- (3) Asexual reproduction by gonidia or monospores
- (4) Sexual reproduction mostly unknown

Sub-class 2. **Florideae**

- (1) Thallus basically filamentous
- (2) Pit connections distinct
- (3) Cells with more than one chromatophore
- (4) Carpogonium highly specialized

Order 1. **Nemalionales**

- (1) Plants filamentous, corticated, uni- or multi-axial
- (2) Cells uninucleate, chromatophores axial or lateral
- (3) Cystocarps superficial or embedded in the thallus
- (4) Life cycle without free living tetrasporophyte

Family 1. **Batrachospermaceae**

- (1) Fresh water members
- (2) Thallus uniaxial
- (3) Life cycle haplobiontic

Example. *Batrachospermum*

Order 2. **Gigartinales**

- (1) Plants filiform, fleshy-membranous or crustose; corticated and multi-axial
- (2) Tetrasporangia scattered on plants.
- (3) Spermatangia borne on surface at special points

- (4) Carpogonia embedded in the cortex

Family 1. **Gracilariaceae**

- (1) Branches terete and firm
- (2) Outer cells not radially serrate
- (3) Narrow, small celled assimilatory cortex bearing delicate, colourless hairs
- (4) Medulla parenchymatous
- (5) Tetrasporangia tetrapartite

Example. *Gracilaria*

Order 3. **Ceramiales**

- (1) Thalli uni- to multi-axial
- (2) Filaments corticated, polysiphonous
- (3) Spermatangia in clusters
- (4) Presence of trichoblasts

Family 1. **Ceramiaceae**

- (1) Thallus monosiphonous
- (2) Naked or corticated by secondary filaments developed at nodes
- (3) Procarp exterior to the thallus
- (4) Tetrasporangia usually tetrahedral and external

Example. *Ceramium*

Family 2. **Rhodomelaceae**

- (1) Axes polysiphonous
- (2) Axes naked, corticated or covered with branches
- (3) Main axis surrounded with pericentrals
- (4) Plants bushy, sparingly branched; branches delicate

Example. *Polysiphonia*

CLASS VI. **MYXOPHYCEAE**

- (1) Plastids not organised
- (2) Pigments blue-green in colour
- (3) Photosynthetic reserve—cyanophycean starch or glycogen
- (4) True nucleus absent, cells prokaryotic
- (5) Sexual reproduction not known

Order 1. **Chroococcales**

- (1) Plants unicellular or colonial
- (2) No differentiation into base and apex
- (3) Nannocytes often present

Family 1. **Chroococcaceae**

- (1) Cells unicellular or forming colonies
- (2) Filaments no known

Example. *Gloeocapsa*

Order 2. **Nostocales**

- (1) Thallus with trichomes
- (2) Trichomes unbranched or with false branching

- (3) Hormogones, heterocysts, exospores, endospores, etc. present
- Family 1. **Oscillatoriaceae**
- (1) Trichomes uniseriate, sometimes tapering at the ends
- (2) Heterocysts and spores absent
- (3) Sheath absent or diffluent
- Example. *Oscillatoria*
- Family 2. **Nostocaceae**
- (1) Trichomes simple, unbranched, uniseriate, undifferentiated
- (2) Heterocysts and akinetes present
- Example. *Nostoc*

## Classification of Algae

### Sub-division—ALGAE

Class	Sub-class	Order	Family	Example	
1. Chlorophyceae		1. Volvocales	1. Chlamydomonadaceae	<i>Chlamydomonas</i>	
			2. Volvocaceae	<i>Pandorina</i> <i>Eudorina</i> <i>Pleodorina</i> <i>Volvox</i>	
		2. Chlorococcales	1. Chlorellaceae	<i>Chlorella</i>	
			2. Hydrodictyaceae	<i>Pediastrum</i> <i>Hydrodictyon</i>	
		3. Ulotrichales	3. Coelastraceae	<i>Scenedesmus</i>	
			1. Ulotrichaceae	<i>Ulothrix</i>	
		4. Cladophorales	2. Ulvaceae	<i>Ulva</i> <i>Enteromorpha</i>	
			1. Cladophoraceae	<i>Cladophora</i>	
		5. Chaetophorales	1. Chaetophoraceae	<i>Draparnaldiopsis</i> <i>Frischiella</i> <i>Coleochaete</i>	
			2. Coleochaetaceae		
6. Oedogoniales	1. Oedogoniaceae	<i>Oedogonium</i>			
	7. Zygnematales	1. Zygnemataceae	<i>Spirogyra</i> <i>Zygnema</i> <i>Cosmarium</i>		
8. Siphonales		2. Desmidiaceae	<i>Netrium</i>		
	9. Charales	1. Caulerpaceae	<i>Caulerpa</i>		
		2. Codiaceae	<i>Codium</i>		
2. Xanthophyceae		1. Heterosiphonales	1. Characeae	<i>Chara</i> <i>Nitella</i>	
			2. Botrydiaceae	<i>Botrydium</i>	
3. Bacillariophyceae			2. Vaucheriaceae	<i>Vaucheria</i> <i>Melosira</i> <i>Pinnularia</i>	
			1. Centrales		
4. Phaeophyceae			2. Pennales		
			1. Ectocarpales	1. Ectocarpaceae	<i>Ectocarpus</i>
			2. Laminariales	1. Laminariaceae	<i>Laminaria</i>
			3. Fucales	1. Fucaceae	<i>Fucus</i> <i>Sargassum</i>
5. Rhodophyceae	1. Bangioidae 2. Florideae		2. Sargassaceae	<i>Diclyota</i>	
			4. Dictyotales	1. Dictyotaceae	
			1. Nemalionales	1. Batrachospermaceae	<i>Batrachospermum</i>
			2. Gigartinales	1. Gracilariaceae	<i>Gracilaria</i>
6. Myxophyceae			1. Ceramiaceae	<i>Ceramium</i>	
			3. Ceramiales	2. Rhodomelaceae	<i>Polysiphonia</i>
			1. Chroococcales	1. Chroococcaceae	<i>Gloeocapsa</i>
			2. Nostocales	1. Oscillatoriaceae	<i>Oscillatoria</i>
			2. Nostocaceae	<i>Nostoc</i>	
			3. Scytonemataceae	<i>Scytonema</i>	
			4. Rivulariaceae	<i>Rivularia</i> <i>Gloeotrichia</i>	

**Family 3. Scytonemataceae**

- (1) Trichomes uniseriate, without marked attenuation
- (2) Filaments free, with false branching
- (3) Sheath firm enclosing one or more trichomes

Example. *Scytonema*

**Family 4. Rivulariaceae**

- (1) Trichomes markedly attenuated from base to apex
- (2) Trichomes show false branching
- (3) Growth trichothallic with terminal hair

Examples. *Rivularia*, *Gloeotrichia*

**I. CHLOROPHYCEAE**

The members of chlorophyceae generally out number members of the other groups of algae. About 90% are freshwater forms while the rest are marine in habitat. Green algae also occurs in damp soils, rocks and cliffs, damp wood, bark and leaves of the tree, shells of the snails, snow or ice and sometimes even as internal parasites.

The green colour of these members is due to predominance of chlorophylls. Photosynthetic reserve is starch that is stored in pyrenoid. The cell walls are made of cellulose.

The range of thallus varies from unicellular, colonial, multicellular, filamentous to thalloid. Lower forms are generally unicellular or colonial. Higher members, however, are either filamentous or thalloid.

Thallus consists of cells which divide generally into two planes. Some members are remarkable in their absence of transverse walls thus nuclei are scattered throughout the thallus the coenocytes.

The reproductive structures are flagellated and, therefore, motile. The flagella are equal in length. There are generally two or four flagella but sometimes numerous flagella may also occur.

***Chlamydomonas*****Classification**

Sub-division	—	Algae
Class	—	Chlorophyceae
Order	—	Volvocales
Family	—	Chlamydomonadaceae
Genus	—	<i>Chlamydomonas</i>

**Exercise 1**

**Object : Study of Chlamydomonad cell.**

**Work procedure**

Study the slide showing unicellular thallus.

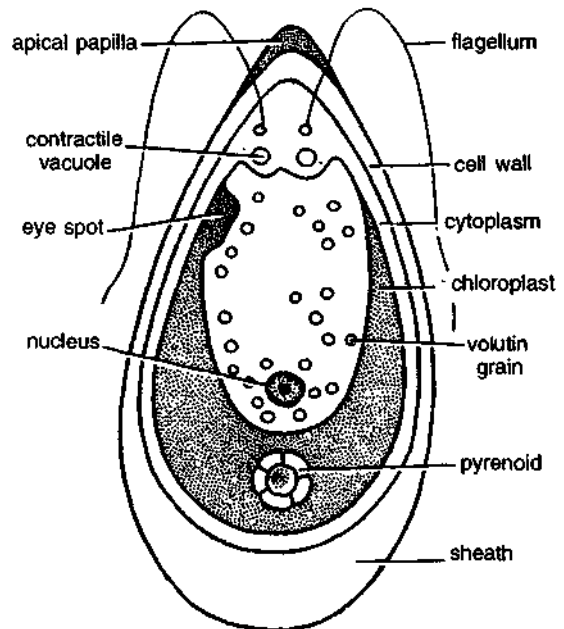


Fig. 1. *Chlamydomonas*. Structure of a single cell.

**Comments**

1. Thallus is unicellular and motile.
2. The cell is usually oval in shape. Sometimes spherical, oblong, pyriform or ellipsoidal.
3. The cell is surrounded by a cell wall. It is narrow at its anterior end and broad at the posterior end.
4. Anterior end bears two closely situated flagella (whiplash type).
5. At the base of each flagellum lies a blepharoplast or basal granule.
6. A small projection or papilla, known as apical papilla, is present in between the two anteriorly inserted flagella.
7. At the base of each flagellum one contractile vacuole is present.
8. Just near the cell wall, towards the anterio-lateral part of the cell, lies an orange or red coloured spot, called stigma or eye spot.
9. The broad posterior part has a large, massive and a single cup-shaped chloroplast. The thin sides of the chloroplast cup extend towards the anterior end.
10. The broad portion of the chloroplast has a single pyrenoid (sometimes two to many).

- The cavity of the cup-shaped chloroplast is completely filled with the cytoplasm in which lies a single nucleus.
- Many volutin grains, the main reserve food product, are irregularly distributed in the cytoplasm.

### Exercise 2

**Object : Study of Palmella stage.**

#### Work procedure

Study the slide showing Palmella stage. (It is known as Palmella stage because of its resemblance with another alga—Palmella of the order Tetrasporales).

#### Comments

- This is asexual reproductive body.
- The groups of cells are embedded in a common mucilaginous envelope.
- Each cell is chlamydomonad in structure. The flagella are absent.
- It is a temporary phase—a structure of perennation.
- This stage is formed under unfavourable conditions. On return of favourable conditions each cell of the Palmella stage develops flagella and becomes similar to the parent cell.

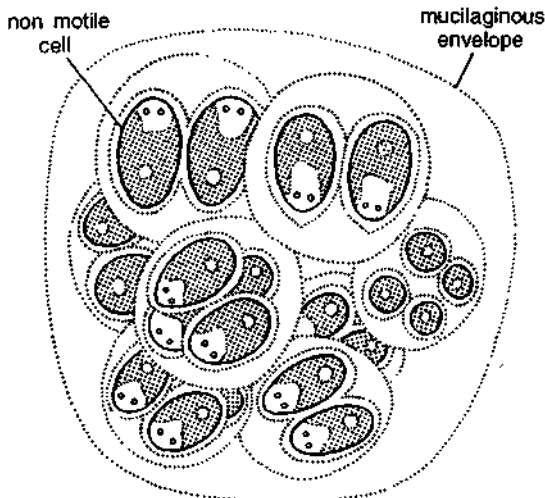


Fig. 2. *Chlamydomonas*. Palmella stage.

### Identification

**Sub-division—Algae.** (1) Presence of a simple thallus, (2) Chlorophyll present, (3) Cell wall made of cellulose.

**Class—Chlorophyceae.** (1) Presence of a definite nucleus, (2) Chloroplast present, grass green colour, (3) Presence of starch, (4) Reproductive structure motile and flagella equal in length.

**Order—Volvocales.** (1) Thallus motile, (2) Protoplast with contractile vacuoles.

**Family—Chlamydomonadaceae.**

**Genus—Chlamydomonas.** (1) Oval or pyriform shape of the thallus which is unicellular, (2) Cup-shaped chloroplast, (3) Presence of an eye spot, (4) Formation of Palmella stage.

### Hints for Collection

It is found free-swimming in freshwater, stagnant water and ditches. It also occurs on damp soil and mostly forms a green surface layer on the water.

## Volvox

### Classification

Sub-division	—	Algae
Class	—	Chlorophyceae
Order	—	Volvocales
Family	—	Volvocaceae
Genus	—	Volvox

### Exercise 1

**Object : Study of thallus.**

#### Work procedure

Stain a few colonies in safranin, wash in water and mount in glycerine to study the external features of the colony and structure of a cell.

#### Comments

- Thallus is multicellular, motile and a coenobial colony.
- Colonies are mostly spherical, rounded or oval in shape.
- It is hollow in the centre and cells are arranged in a single layer towards the periphery.
- Layer of cells is surrounded by a gelatinous mass which forms the outer and firm limiting layer.

- The number of cells in a colony varies from 500-6,500 according to the species.
- Each cell of the colony is connected with a few of the neighbouring cells by thin and delicate cytoplasmic strands.
- Each cell is enveloped by an individual gelatinous sheath.
- All the cells of a colony are typically chlamydomonad in shape, size and structure.
- Each vegetative cell is biflagellate, motile and ovoid. The two flagella are anteriorly inserted. A contractile vacuole is situated one each at the base of a flagellum. Cup-shaped chloroplast occupies much of the posterior part in which is situated a single pyrenoid. In the cavity formed by cup-shaped chloroplast lies a single nucleus, surrounded by cytoplasm. Cytoplasm is rich in volutin grains. Eyespot or stigma occupies antero-lateral position.

### Exercise 2

**Object : Study of asexual reproduction.**

### Work procedure

Study the slide showing daughter colonies.

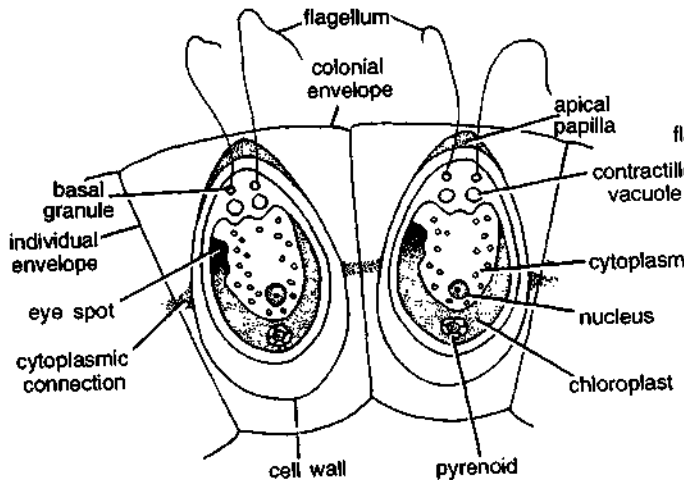


Fig. 2. *Volvox*. A part of the colony showing arrangement of cells.

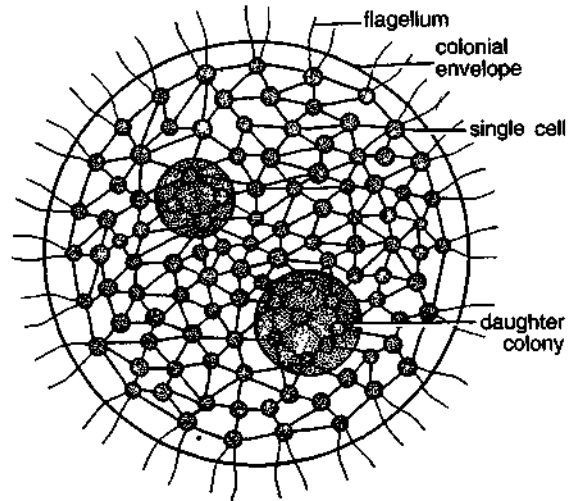


Fig. 1. *Volvox*. Parent colony with daughter colonies.

### Comments

- Asexual reproduction takes place by the formation of daughter colonies.
- Daughter colonies are formed mostly in the posterior part of the parent colony.
- Many small daughter colonies remain embedded in the parent colony. These appear similar to parent colony except its smaller size.
- Daughter colonies are liberated by the gelatinization of the wall of the parent colony.

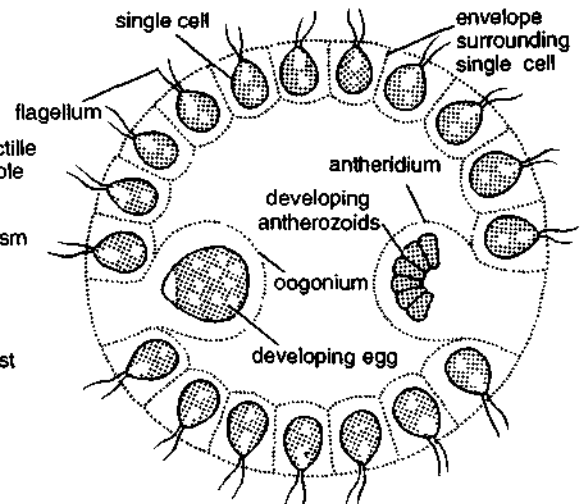


Fig. 3. *Volvox*. A colony with antheridium and oogonium.

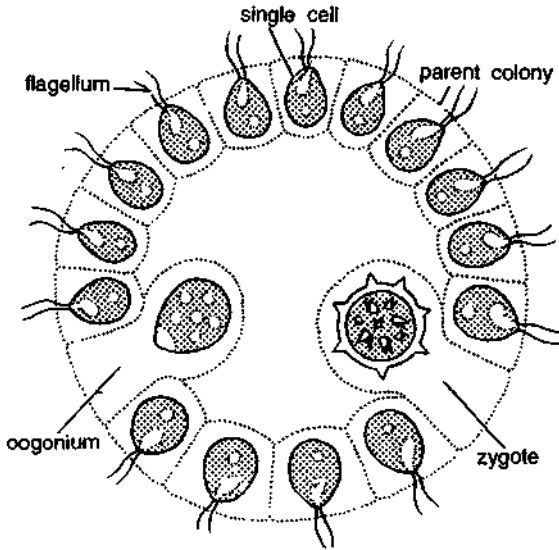


Fig. 4. *Volvox*. A colony with a zygote.

### Exercise 3

Object : Study of sexual reproduction.

#### Work procedure

Study the slide showing sex organs and the zygote.

#### Comments

1. Colonies may be monoecious or dioecious.
2. Sexual reproduction is oogamous.
3. Antheridia and oogonia are developed mostly in the posterior part of the colony.
4. Antheridium produces biflagellate antherozoids, while non-motile oogonium develops a single egg.
5. As a result of fertilization oospore or zygote is produced.
6. Zygote is orange-red in colour because of the presence of haematochrome.
7. Zygote is a thick-walled structure. The wall is made of two or three layers. Outermost layer is thick and may be smooth or ornamented.

#### Identification

**Sub-division—Algae.** (1) Thallus simple, (2) Presence of chlorophyll, (3) Cell wall of cellulose.

**Class—Chlorophyceae.** (1) Chloroplasts green, (2) Photosynthetic product starch, (3) Motile cells flagellated, (4) Flagella equal in length.

**Order—Volvocales.** (1) Vegetative cells flagellated, (2) Thallus motile.

**Family—Volvocaceae.** (1) Thallus colonial, (2) Division of cells in longitudinal plane.

**Genus—Volvox.** (1) Colony spherical or sub-spherical, (2) Number of cells at least 500.

### Hints for Collection

It can be collected from freshwater and permanent ponds and pools where it occurs as small green balls on the surface layer. The alga grows abundantly during spring and disappears during summer.

## Chlorella

### Classification

Sub-division	—	Algae
Class	—	Chlorophyceae
Order	—	Chlorococcales
Family	—	Chlorellaceae
Genus	—	Chlorella

### Exercise 1

Object : Study of single *Chlorella* cell.

#### Work procedure

Place a few cells on a slide, stain with safranin, wash in water and mount in glycerine. Study the cell structure.

#### Comments

1. Single cell represents the thallus. The cell is non-motile.

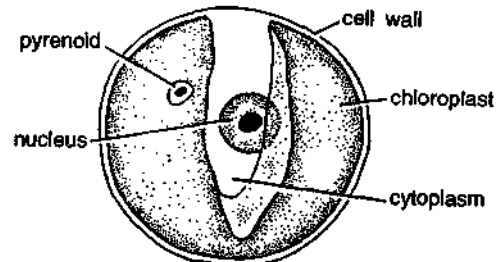


Fig. 1. *Chlorella*. A single cell representing a thallus.

- The cells are found singly or sometimes in groups.
- A cell is very small. It is spherical to ellipsoidal in shape.
- A cell has a single cup-shaped or curved band of chloroplast, parietal in position.
- A single pyrenoid is generally found (lacking at times).
- Nucleus lies surrounded by the cytoplasm which fills up most of the cell.
- The only method of reproduction is by autospores. Two to sixteen autospores are formed by the cell as a result of division.

### Identification

**Sub-division—Algae.** (1) Simple thallus, (2) Presence of chlorophyll, (3) Cell wall of cellulose.

**Class—Chlorophyceae.** (1) Chloroplasts grass-green, (2) Photosynthetic reserve—starch.

**Order—Chlorococcales.** (1) Cells single and non-motile, (2) Cells uninucleate, (3) Method of reproduction—zoospores or autospore formation.

**Family—Chlorellaceae.** (1) Cell single, if united do not form a definite colony, (2) Reproduction by autospores.

**Genus—Chlorella.** (1) Cells solitary and spherical with smooth walls, (2) Single chloroplast.

### Hints for Collection

It is free living and is found in stagnant and running waters. It also grows within the cells or tissues of invertebrates. *C. parasitica* (*Zoochlorella*) is found in *Ophrypodium spongilla*, etc. *C. conductrix* is known to occur in *Hydra*, *Paramecium* and *Stentor*.

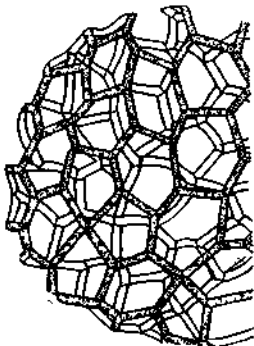


Fig. 1. *Hydrodictyon*. A part of saccate thallus.

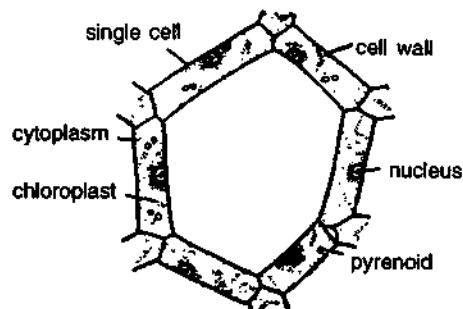


Fig. 2. *Hydrodictyon*. A part of reticulum.

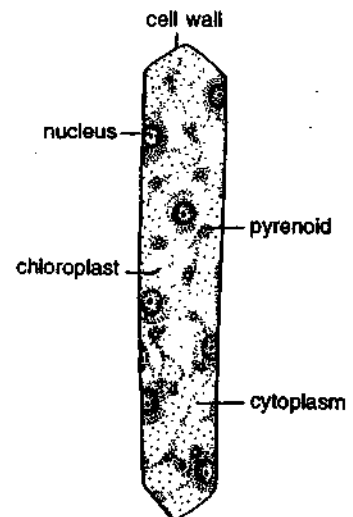


Fig. 3. *Hydrodictyon*. A single cell.

## Hydrodictyon

### Classification

Sub-division	—	Algae
Class	—	Chlorophyceae
Order	—	Chlorococcales
Family	—	Hydrodictyaceae
Genus	—	<i>Hydrodictyon</i>

### Exercise 1

**Object :** Study the thallus and a single cell.

### Work procedure

Take a small part of net-like thallus, stain in safranin, wash with water and mount in glycerine. Take care to spread the net-work in a way that cells do not get overlapped.

### Comments

- The thallus is a multicellular colony forming a net-like structure.
- Colony is a hollow and sac-like or saucer-like (saccate), cylindrical network, closed at both the ends.
- The spaces of the reticulum are bound by five or six cells (this number varies between 3-10).



cells) which form a pentagonal or hexagonal structure.

4. The cells are cylindrical. End walls are angular to facilitate the formation of a mesh.
5. A cell has a large central vacuole.
6. Cytoplasm lies towards the periphery.
7. Cells are multinucleate. The young cells are however, uninucleate.
8. Cell is called a coenocyte because of its multinucleate nature and presence of large central vacuole.
9. Young cells have zonate or entire chloroplast. Little mature cells possess reticulate chloroplast. However, during older stages chloroplast may assume discoid shape and diffuses throughout the cytoplasm.
10. Chloroplast contains large number of pyrenoids.

### Identification

**Sub-division—Algae.** (1) Thallus construction simple, (2) Presence of chlorophyll, (3) Cell wall of cellulose.

**Class—Chlorophyceae.** (1) Grass green chloroplasts, (2) Photosynthetic reserve starch, (3) Motile structures flagellated, (4) Flagella equal.

**Order—Chlorococcales.** (1) Cells mostly single; if united form non-filamentous colonies of definite shape and size, (2) Cells uni- or multinucleate, (3) Asexual reproduction by zoospores or autospores, (4) Sexual reproduction isogamous.

**Family—Hydrodictyaceae.** (1) Cells united to form coenobe, (2) Reproduction by zoospores and biflagellate gametes.

**Genus—Hydrodictyon.** (1) Coenobe a saccate reticulum, (2) Chloroplast reticulate or discoid, (3) Uni- or multinucleate coenocytic cells.

### Hints for Collection

This alga occurs in large quantities in permanent pools or other bodies of stagnant water. It forms an extensive net which covers the entire surface of water reservoir, and is as such called 'water net'.

## Cladophora

### Classification

Sub-division	—	Algae
Class	—	Chlorophyceae
Order	—	Cladophorales
Family	—	Cladophoraceae
Genus	—	Cladophora

### Exercise 1

**Object : Study the thallus.**

### Work procedure

Separate a few filaments, stain in safranin, wash in water and mount in glycerine. Study the characters of thallus.

### Comments

1. The thallus is multicellular, filamentous and branched.
2. In some cases (marine species of *Cladophora*) large number of filaments are held together by weaving of the branches. Such plants form dense masses and appear as compact cushions attached to some substratum or sometimes as loose hollow balls (frequently as big as human head). These species are known as aegagropilous and are found at the bottom of shallow lakes or in marine habitats.
3. Alga remains attached to the substratum by rhizoids. Rhizoidal outgrowths come out from the basal part of the thallus.
4. The filaments are profusely branched. Branching is lateral but appears dichotomous because of evection (process of pushing the main axis on one side during the development of lateral branches).

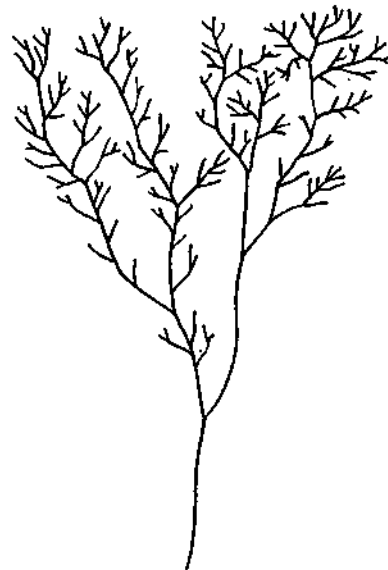


Fig. 1. *Cladophora*. A thallus.

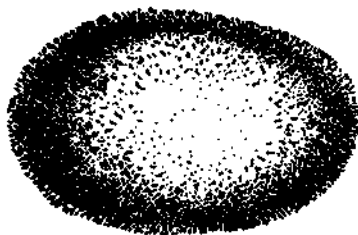


Fig. 2. *Cladophora*. Aegagropilous species.

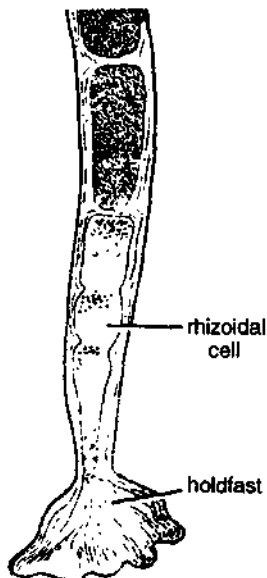


Fig. 3. *Cladophora*. Basal part of the filament with holdfast.

5. Branches arise just beneath the septum, from the upper end of the cell.

### Exercise 2

**Object :** Study a cell.

### Work procedure

Use the same slide prepared earlier and study the structure of a single cell.

### Comments

1. Cell walls are stratified being composed of three layers, out of which middle is conspicuously stratified.
2. A cell is coenocytic. Central region is occupied by a large vacuole, surrounded by outer cytoplasmic lining in which many nuclei are

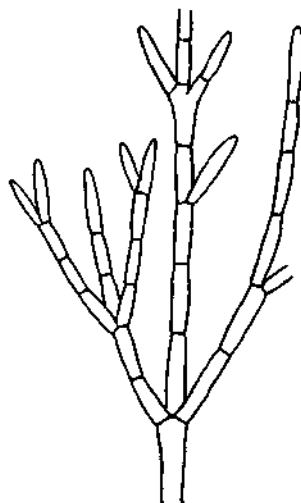


Fig. 4. *Cladophora*. A part of thallus showing mode of branching.

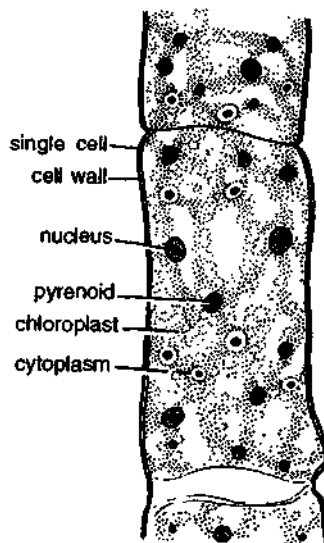


Fig. 5. *Cladophora*. A single cell.

present. Chloroplast is either reticulate or discoid. It is parietal in position and possesses many pyrenoids.

### Identification

*Sub-division*—Algae. (1) Thallus filamentous, (2) Presence of chlorophyll, (3) Cell wall of cellulose.

*Class*—Chlorophyceae. (1) Chloroplast grass-green, (2) Photosynthetic reserve—starch, (3) Motile structures flagellated, (4) Flagella equal in length.

- Order—Cladophorales.** (1) Cells multinucleate and cylindrical, (2) Cell united to form branched or unbranched filaments.
- Family—Cladophoraceae (Single family).** (1) Cells more than eight times longer than broad, (2) Chloroplasts do not form distinct transverse bands.
- Genus—Cladophora.** (1) Filaments freely branched, (2) Filaments without akinetes.

### Hints for Collection

It is the largest genus with world-wide distribution, found abundantly in fresh, brackish, salt and marine waters. It grows attached to stones, shells of snails, rocks, etc.

### *Fritschiella*

#### Classification

Sub-division	—	Algae
Class	—	Chlorophyceae
Order	—	Chaetophorales
Family	—	Chaetophoraceae
Genus	—	<i>Fritschiella</i>

#### Exercise 1

**Object :** Study the Thallus.

#### Work procedure

Place a small bunch of filaments on the slide, stain with safranin, wash in water and mount in glycerine. Study the structure of thallus and also a single cell.

#### Comments

1. Thallus is filamentous. The filaments show heterotrichous habit.
2. Thallus show typical heterotrichous nature with distinct rhizoidal system, the prostrate system, primary projecting system and the secondary projecting system.
3. The rhizoidal system consists of one or more rhizoid-like outgrowths. These arise from prostrate system.
4. The prostrate system is made of clusters of cells. These cells are either rounded or irregular in shape. The plants with well developed prostrate system consist of mature filaments.

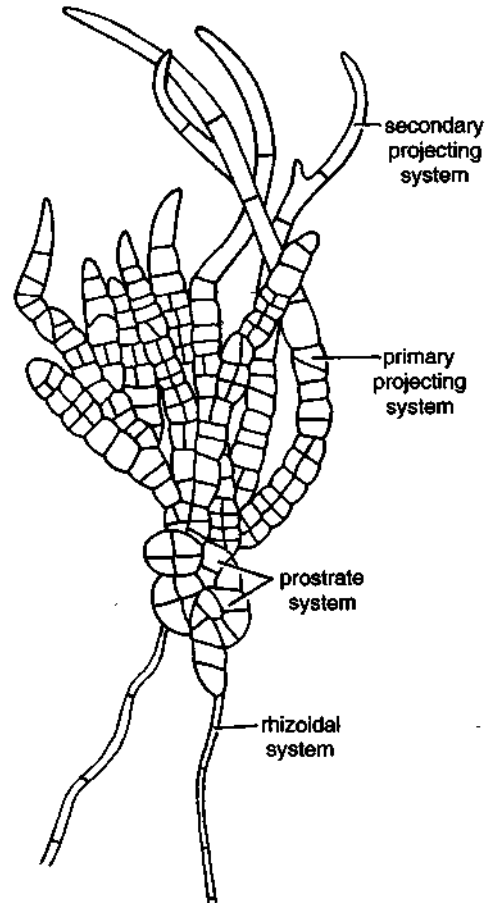


Fig. 1. *Fritschiella*. The thallus.

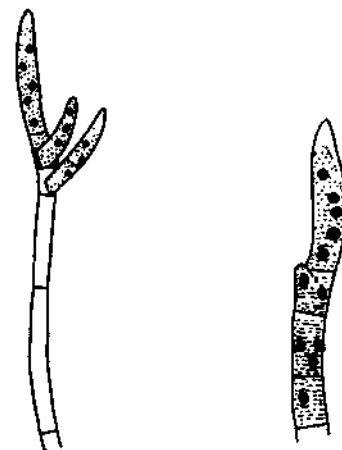


Fig. 2. *Fritschiella*. A single cell of the projecting system.

5. The primary projecting system arises from the prostrate system and is aerial in nature. The

filaments are uniseriate or biseriate which may be branched or unbranched. The cells of these filaments are small and rounded. They are green and photosynthetic.

- The secondary projecting system consists of freely branched uniseriate filaments. The cells are elongated. The end cells of the branches terminate into hair-like structures.
- The cells of the projecting system are thin walled. These are uninucleate with a curved plate-like chloroplast that has 2-8 pyrenoids. The cells of primary projecting system and the prostrate system have poorly developed chloroplasts. In rhizoidal system, chloroplasts are completely absent.

### Identification

**Sub-division—Algae.** (1) thallus construction simple, (2) Presence of chlorophyll, (3) Cell wall of cellulose.

**Class—Chlorophyceae.** (1) Chloroplasts green in colour, (2) Reserve food in the form of starch, (3) Motile structures flagellated, (4) Flagella equal in length.

**Order—Chaetophorales.** (1) Plant body heterotrichous, (2) Distinct prostrate and erect systems present.

**Family—Chaetophoraceae.** (1) Plants filamentous, (2) Terminal or lateral hairs present, (3) Cells with single chloroplast.

**Genus—Fritschiella.** (1) Plant body filamentous, (2) Chloroplast curved and plate-like, (3) Secondary projecting system composed of tuft of elongated branches with longer cells.

### Hints for Collection

*Fritschiella* grows abundantly on moist, alkaline soil and on silt forming lush green cushions. It is particularly common on moist soil, drying after the monsoon rains. This genus was recognised by Professor M.O.P. Iyengar and named to honour great British Algologist Professor F.E. Fritsch. The only Indian species is *Fritschiella tuberosa*.

## Coleochaete

### Classification

Sub-division	—	Algae
Class	—	Chlorophyceae
Order	—	Chaetophorales
Family	—	Coleochaetaceae
Genus	—	<i>Coleochaete</i>

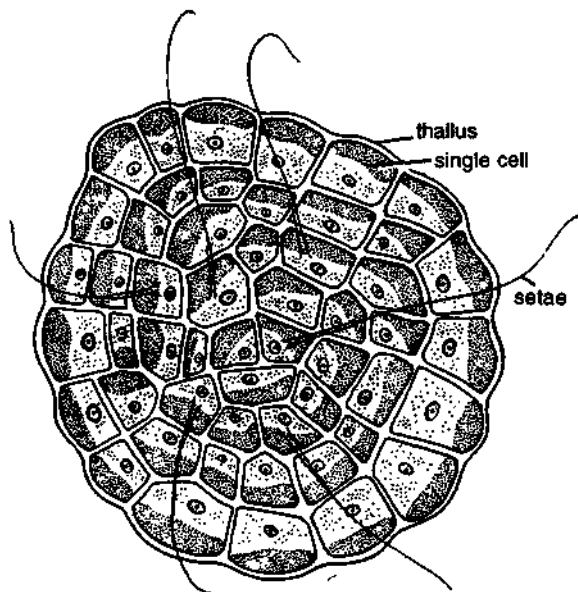


Fig. 1. *Coleochaete* sp. Discoid thallus.

### Exercise 1

**Object : Study the thallus.**

### Work procedure

Prepare a slide by first isolating a discoid or cushionoid epiphytic thallus from aquatic weed by scratching out the thallus with needle. Stain it with safranin, wash with water and then mount in glycerine. Study the structure of thallus and also a cell.

### Comments

- Thallus is multicellular and heterotrichous.
- It is either a disc-like structure in majority of the species (e.g. *C. scutata*) or cushionoid or filamentous (e.g. *C. pulvinata*) in others.
- If thallus is disc-like, the disc represents only the prostrate system while a few setae or hair, represent erect system.
- Filamentous thallus exhibits typical heterotrichous habit with a branched prostrate system and a branched projecting (erect) system.
- In both the cases a few cells possess a cytoplasmic outgrowth—setae. Setae are

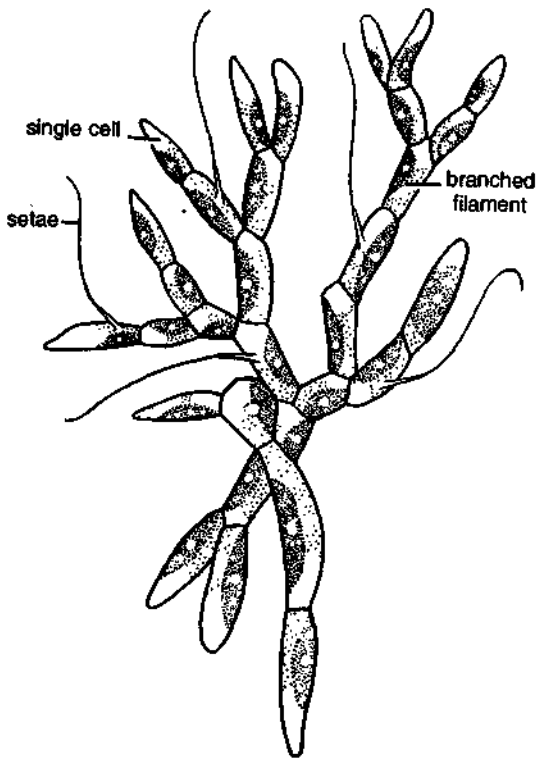


Fig. 2. *Coleochaete* sp. Cushionoid thallus.

surrounded partly or wholly by a gelatinous sheath at the base.

6. The thallus is distinctly enveloped by a gelatinous sheath or mucilage.
7. In discoid species cells of the thallus are joined end to end to form branches. These branches are laterally apposed to one another to form a pseudoparenchymatous disc.
8. Each cell is uninucleate. It has single, large, laminate and parietal chloroplast with a single pyrenoid. Rest of the cell is occupied by the cytoplasm.

### Exercise 2

**Object : Study the reproductive structures.**

#### Work procedure

Scrape out a few thalli from surface of aquatic weed. Observe under the microscope to see if spermocarps are present. Select such a thallus, stain in safranin, wash in water and mount in glycerine. Study the structure.

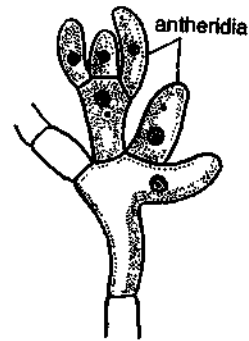


Fig. 3. *Coleochaete* sp. Thallus bearing antheridia.

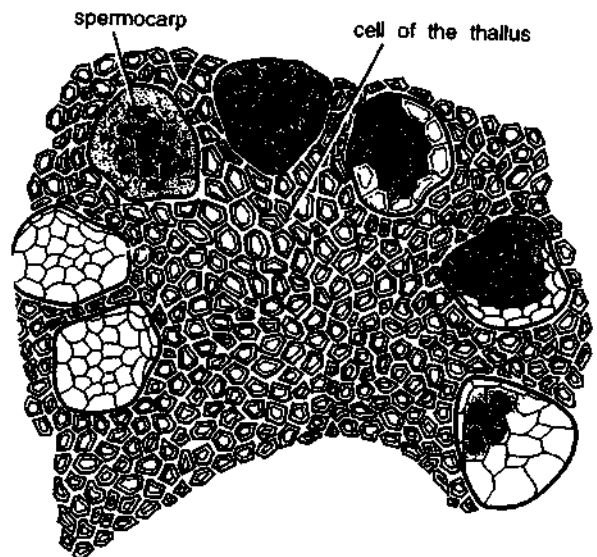


Fig. 4. *Coleochaete* sp. Thallus with spermocarps.

#### Comments

1. Thalli may be homothallic or heterothallic.
2. Sexual reproduction is oogamous.
3. Antheridia are generally borne at the tips in filamentous species and in the middle or peripheral region in the discoid species.
4. Antheridia appear as a group of small cells.
5. Oogonia are also borne terminally in filamentous species and towards periphery in the discoid species.
6. Oogonium is a flask-shaped structure with long tubular trichogyne.
7. The fertilization results in the formation of a zygote which remains embedded inside the wall of the oogonium. It is a thick walled structure.

8. Zygote known as spermatocarp remains enveloped in a parenchymatous tissue formed by the development of neighbouring cells. It is conspicuously reddish-brown in colour.
9. Spermatocarp remains dormant for a long period.

### Identification

**Sub-division—Algae.** (1) Thallus construction simple, (2) Presence of chlorophyll, (3) Cell wall of cellulose.

**Class—Chlorophyceae.** (1) Chloroplasts green in colour, (2) Reserve food in the form of starch, (3) Motile structures flagellated, (4) Flagella equal in length.

**Order—Chaetophorales.** (1) Plant body heterotrichous, (2) Presence of setae.

**Family—Coleochaetaceae.** (1) Vegetative cells with long cytoplasmic hair (setae), (2) Cells uninucleate, (3) Filaments branched, (4) Each cell with a single parietal and laminar chloroplast.

**Genus—Coleochaete.** (1) Plant body multicellular, (2) Thallus parenchymatous, (3) Presence of spermatocarp.

### Hints for Collection

It occurs as freshwater alga, mostly as an epiphyte on submerged water plants (e.g. *Ipomoea*, *Typha*, *Polygonum*, etc.) or on other algae. It also grows endophytically inside the cells of Charales.

## Oedogonium

### Classification

Sub-division	—	Algae
Class	—	Chlorophyceae
Order	—	Oedogoniales
Family	—	Oedogoniaceae
Genus	—	Oedogonium

### Exercise 1

**Object :** Study of thallus.

### Work procedure

Stain a few filaments in safranin, wash with water and mount in glycerine. Study the external features and structure of a cell.

### Comments

1. Thallus is multicellular, filamentous and unbranched.

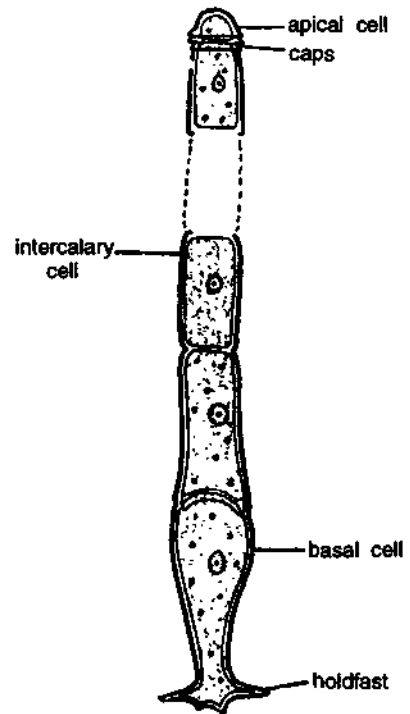


Fig. 1. *Oedogonium*. A filament.

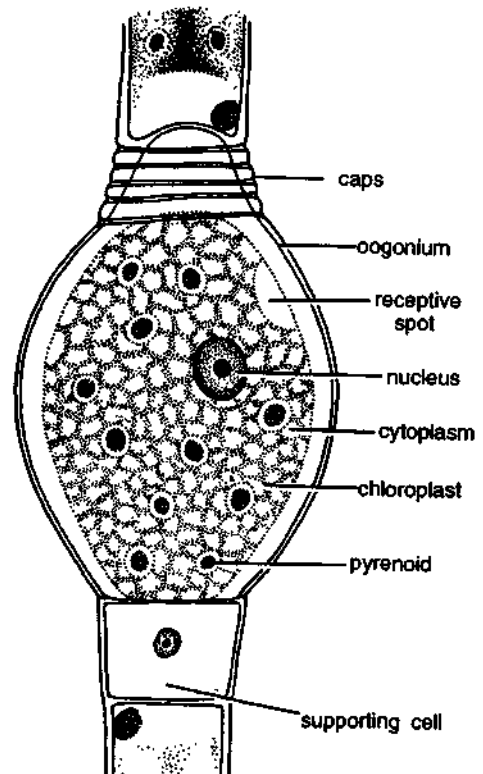


Fig. 2. *Oedogonium*. Filament showing a mature oogonium.

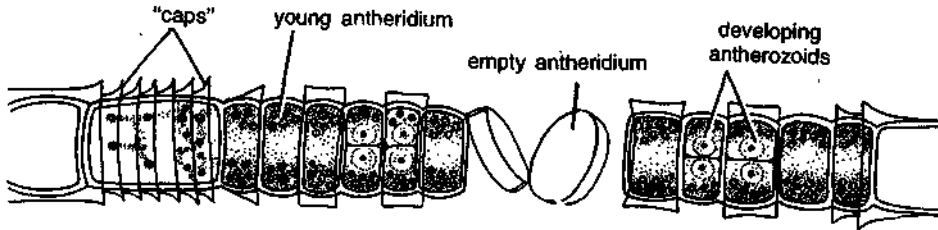


Fig. 3. *Oedogonium*. Part of a filament showing a chain of antheridia (Macrandrous species).

- A filament is differentiated into three types of cells according to their position : (i) basal, (ii) intercalary and (iii) apical.
- The basal cell of a filament functions as a holdfast. The lower part of the holdfast is either disc-like or finger-shaped. The upper part is mostly broad and rounded. The basal part of the cell generally lacks green pigment and, therefore, is non-green unlike other cells of the filament.
- A cell at the tip of the filament is known as apical cell. It is rounded at its free surface.
- The cells present between basal and the apical cells are intercalary cells. These show typical cell structure.
- The typical cell is cylindrical.
- Cell wall is thick and three layered.
- Inner to cell wall is a reticulate chloroplast that runs parallel to the long axis of the cell. Many pyrenoids are present in the chloroplast.
- The cell is uninucleate. The nucleus is situated near the cell wall and is held by thin and delicate cytoplasmic strands.
- Mature and old cells show 'cap cells' at their upper end. These are characteristic of the members of Oedogoniales.
- Oogonium generally shows one or more cap cells at its upper end, indicating its development from a comparatively older cell.
- It is mostly spherical or oval in shape and larger than a vegetative cell.
- At the base of each oogonium lies a small and flat daughter cell, known as supporting or suffultory cell.
- Oogonium encloses a single large ovum.
- The wall of the oogonium has a small pore on one side, known as receptive pore.
- Just opposite the receptive pore, protoplast of the oogonium has a hyaline area-receptive spot.
- Uninucleate protoplast is rich in reserve food.

### Exercise 2

**Object : Study of oogonium.**

#### Work procedure

Place a few filaments and observe a filament with oogonia. Stain such filaments with safranin, wash in water and mount in glycerine. Study the oogonia.

#### Comments

- Oogonia are intercalary or terminal in position.
- Oogonium may be solitary or occur in a row of 2-3 or even more.

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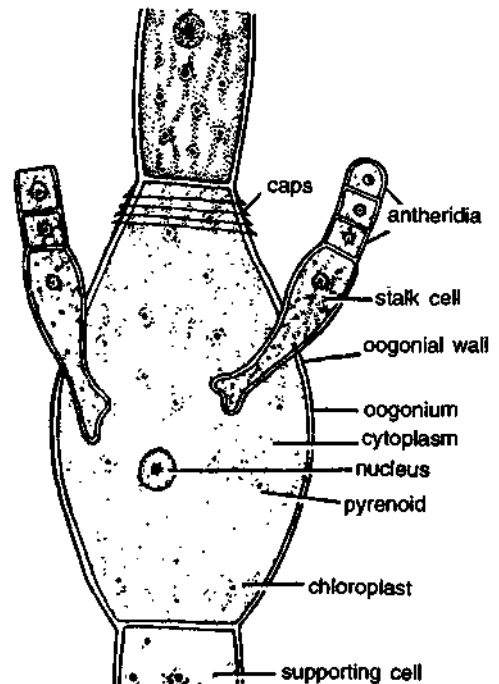


Fig. 4. *Oedogonium*. Oogonium with two dwarf males attached to it. (Nannandrous species).

**Exercise 3****Object : Study of antheridium.****Work procedure**

Place a few filaments on a slide in a drop of water, observe under the microscope for the presence of chain of antheridia. Isolate such filaments, stain in safranin, wash with water and mount in glycerine. Study the antheridia of macrandrous species.

**Comments**

1. Antheridia are mostly intercalary in position.
2. Numerous antheridia form a long chain being arranged in a series.
3. An antheridium is a small and flat cell.
4. Each antheridium has two nuclei lying side by side, surrounded by dense cytoplasmic contents. Each of such protoplasmic groups later on metamorphoses into a multiflagellate antherozoid.

**Exercise 4****Object : Study of dwarf male or nannandrium.****Work procedure**

Isolate few filaments bearing dwarf males and stain in safranin, wash in water, mount in glycerine and study. Draw the diagram.

**Comments**

1. The dwarf male (or nannandrium) is characteristic of nannandrous species.
2. The dwarf male is produced by the germination of androspore.
3. Androspores are formed inside the androsporangia.
4. Androsporangia form a long chain of small and flat cells in intercalary position in the filament.
5. Each androsporangium develops a single multiflagellate androspore (in contrast, per antheridium two antherozoids are produced).
6. Androspore germinates to produce a dwarf male or nannandrium which remains attached

either to the wall of the oogonium or to the suffultory cell.

7. A dwarf male is made of a stalk cell and a terminal row of 2-3 cells.
8. Stalk cell is at the base by which the dwarf male is attached to the filament. It has a disc-like or finger-like structure at its base.
9. The terminal row has 2-3 small, flat and narrow antheridia.
10. Each antheridium has two multiflagellate antherozoids.

**Exercise 5****Object : Study of zygote.****Work procedure**

Study a slide showing zygotes.

**Comments**

1. Zygote is thick-walled post-fertilization structure. The wall is generally three layered.
2. The layer outside the innermost may be smooth, ornamented or verrucose.
3. Zygote develops red colour due to the accumulation of reserve food in the form of reddish oil drops.

**Identification**

*Sub-division*—Algae. (1) Thallus filamentous, (2) Chlorophyll present, (3) Cell wall of cellulose.

*Class*—Chlorophyceae. (1) Chloroplasts grass-green, (2) Photosynthetic reserve—starch, (3) Motile structures flagellated, (4) Flagella equal in length.

*Order*—Oedogoniales. (1) Cells uninucleate, filaments branched or unbranched, (2) Cell division forming 'caps', (3) Chloroplast reticulate, (4) Zoospores and antherozoids bear a whorl of flagella, (5) Production of dwarf males.

*Family*—Oedogoniaceae (A single family).

*Genus*—Oedogonium. (1) Filaments unbranched, (2) Cells cylindrical, (3) Holdfast well developed.

**Hints for Collection**

It is mostly aquatic and occurs abundantly in small, permanent and semi-permanent bodies of water, such as pools and ponds. In fast-flowing stream, it is not generally found fruiting, except when the flow is slow. Filaments may form a free-floating mass or are attached to stones, etc. Species are also known

(B-14)



to occur widely as epiphytes, mostly on the water plants or on larger Oedogoniaceae or Cladophoraceae.

## Zygnema

### Classification

Sub-division	—	Algae
Class	—	Chlorophyceae
Order	—	Zygnematales (Conjugales)
Family	—	Zygnemataceae
Genus	—	Zygnema

### Exercise I

**Object : Study of thallus and a cell.**

#### Work procedure

Take a few filaments, stain with safranin, wash in water and mount in glycerine. Study the thallus and cell structure.

#### Comments

1. The thallus is multicellular and filamentous.
2. Filaments are uniseriate (composed of many cells in a row) and unbranched.
3. Each cell of a filament is cylindrical, length being not usually more than twice the breadth.
4. The cells are generally covered by a large amount of mucilage.
5. The transverse wall of the cell is thin while the lateral wall is thick (because of the thick pectose deposition).
6. The cells are filled with large amount of cytoplasm.
7. The most characteristic of the cell is the presence of two axile and stellate chloroplasts, arranged along the longitudinal axis of the cell.
8. Each chloroplast sends out thin or thick, laterally radiating strands which sometimes extend up to the cell wall.
9. A single pyrenoid is located centrally in each of the chloroplasts.
10. Cells are uninucleate. Nucleus is centrally located between the two stellate chloroplasts which lie on both of its sides. Nucleus is surrounded by a thick and broad strands of cytoplasm.

(B-14)

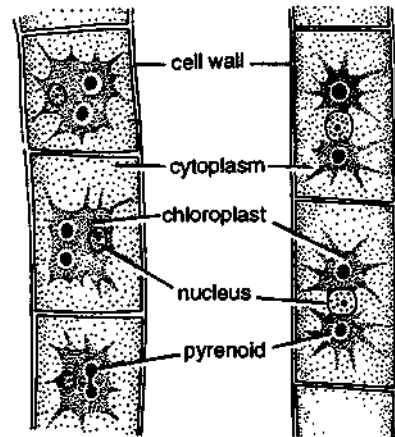


Fig. 1. *Zygnema*. Filaments showing internal structure of the cell.

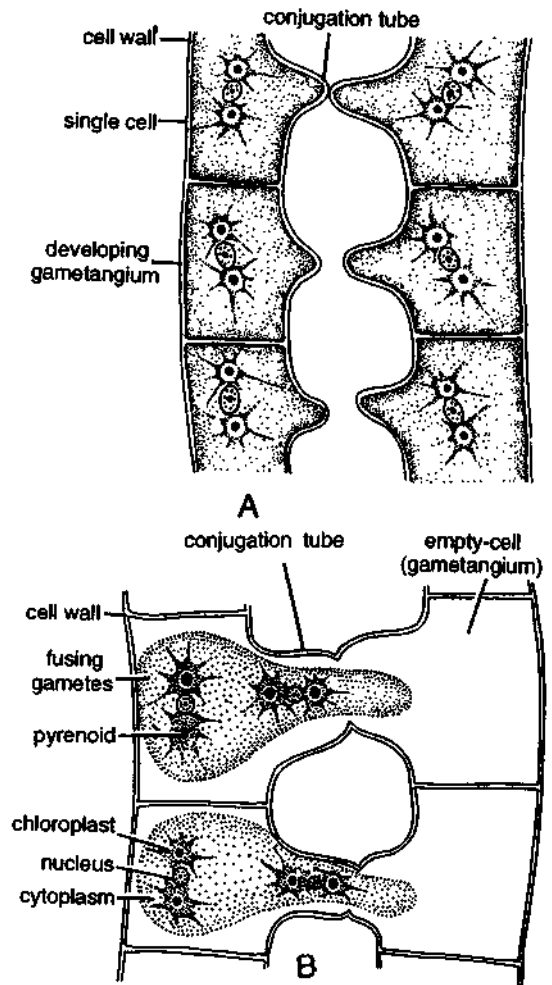


Fig. 2. *Zygnema*. A-B. Filaments showing various stages of scalariform conjugation.

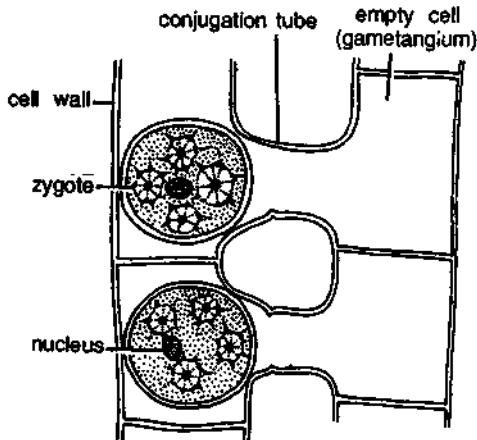


Fig. 3. *Zygnema*. Filaments showing zygospores after completion of scalariform conjugation.

### Exercise 2

**Object :** Study of scalariform conjugation.

#### Work procedure

Take a few filaments, see if stages of scalariform conjugation are present. Stain such filaments with safranin, wash in water and mount in glycerine. Study different stages and draw.

#### Comments

1. Sexual reproduction takes place by conjugation.
2. Each cell of the filament forms non-motile gamete.
3. Two filaments take part in this process. Thus the species showing scalariform conjugation are heterothallic.
4. Two filaments lie opposite one another throughout their whole length.
5. Each cell produces a conjugation tube towards the opposite cell of another filament.
6. Protoplasts are contracted, rounded or elliptical in shape, and are called gametes.
7. Gametes migrate from one gametangium to another during fusion. Thus cells of both threads are completely empty (because gametes generally fuse in conjugation tube). In other species zygospores occupy gametangium of one filament, leaving cells of the other filament empty.

8. A young zygospore has four stellate chloroplasts. Completely mature zygospore has thick, three-layered, ornamented and coloured (blue) wall.

### Exercise 3

**Object :** Study of lateral conjugation.

#### Work procedure

Study the slide showing various stages of lateral conjugation.

#### Comments

1. Lateral conjugation is comparatively rare.
2. Both male and female gametes are produced by the same filament. Hence, the species are called homothallic.
3. The cells or male and female gametangia produce a small conjugation tube each near the cross wall common to both these cells.
4. Both male and the female gametes creep into the conjugation tube where these fuse.
5. Both, male and female cells become empty, the zygospore being formed inside the conjugation tube.
6. Zygospore is ornamented or smooth. The wall is three layered and may also be coloured.

### Identification

*Sub-division—Algae.* (1) Filamentous thallus, (2) Presence of chlorophyll, (3) Cell wall of cellulose.

*Class—Chlorophyceae.* (1) Chloroplasts grass-green, (2) Photosynthetic reserve is starch.

*Order—Zygnematales* (Conjugales). (1) Absence of flagellated reproductive cells, (2) Sexual reproduction (conjugation) by amoeboid gametes.

*Family—Zygnemataceae.* (1) Unbranched and uniseriate filaments, (2) Chloroplasts either parietal and ribbon shaped or single/two, axial and stellate.

*Genus—Zygnema.* (1) Two axial chloroplasts per cell, (2) Zygospores in the conjugation tubes, (3) Conjugating cells not filled with gelatinous material.

### Hints for Collection.

It is widely distributed in stagnant/fresh water ponds and streams. It may form either floating masses or scum on the water surface. It is generally found in reproductive stages during spring.

***Chara***  
**(Stone-Wort)**

**Classification**

Sub-division	—	Algae
Class	—	Chlorophyceae
Order	—	Charales
Family	—	Characeae
Genus	—	<i>Chara</i>

**Exercise 1**

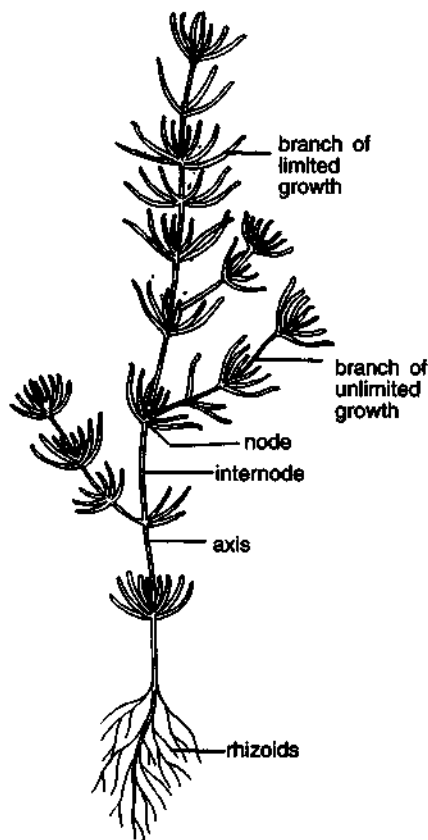
**Object : Study the external features of thallus.**

**Work procedure**

Study a slide showing mounted part of the thallus or study a preserved specimen. Magnifying lens or dissecting microscope would be very useful.

**Comments**

1. Thallus is macroscopic, branched and multicellular. Calcium carbonate is deposited all over.
2. It remains attached to the substratum by multicellular rhizoids which bear an erect and branched main axis above.
3. Multicellular rhizoids are branched.
4. These are borne by the lower nodes of the main axis.
5. Rhizoids possess oblique septa. The rhizoids are not differentiated into nodes and internodes.
6. The cytoplasm of a rhizoidal cell has a nucleus situated towards the upper side of the cell.
7. At the septum of a rhizoidal cell, the ends are protracted in opposite directions to form knotted part.
8. At this place, signifying a node, a plate of four cells or even more is formed which gives rise to rhizoidal branches. This part is known as rhizoidal plate.
9. Main axis is composed of long internodes alternating with small nodes.
10. Long internode is composed of a single cell enveloped by many corticating threads.
11. A node is a group of regularly arranged cells.
12. It bears two types of branches—(i) lateral branches of limited growth (short laterals) and



**Fig. 1. *Chara*. A thallus to show habit.**

- (ii) lateral branches of unlimited growth (long laterals).
13. Laterals of limited growth are borne in whorls around the nodes of the main axis.
14. Each short lateral is divided into nodes and internodes.
15. The internodes of short laterals are small as compared to those of the main axis.
16. Short laterals borne by the nodes of the main axis are also termed as primary laterals of limited growth.
17. From the nodes of the short laterals, secondary short laterals are produced which are usually small, unicellular and are variously termed as stipules or leaves.
18. Laterals of unlimited growth are borne by the nodes of the main axis only. These are situated in the axils of short laterals.
19. Long laterals possess the same characteristics as those of the main axis.

20. Long laterals are differentiated into long, corticated and unicellular internodes and small and multicellular nodes.
21. Nodes of the long laterals bear short laterals which in their turn give out stipules or leaves at their nodes.

### Exercise 2

**Object : Study of a cell.**

#### Work procedure

Study the slide showing T.s. of internodal cell.

#### Comments

1. In the centre is a large central, axial or internodal cell.
2. It is surrounded by corticating threads on all sides.
3. Internodal cell shows a typical cell structure.
4. Centre of the cell has a big vacuole surrounded by cytoplasm.
5. In the cytoplasm lies a single nucleus held by thin and delicate cytoplasmic strands.
6. Many discoid chloroplasts without pyrenoids are scattered in the peripheral cytoplasm.
7. The cell has an outermost, thick and firm cell wall.

### Exercise 3

**Object : Study L.s. of apex.**

#### Work procedure

Study the slide showing L.s. of apex.

#### Comments

1. The cell situated at the top is an apical cell. It contributes to the development of the main axis and lateral axes.
2. It cuts off a longitudinal series of cells below.
3. Upper biconcave cell of a series is a nodal cell and lower biconvex one is an internodal cell.

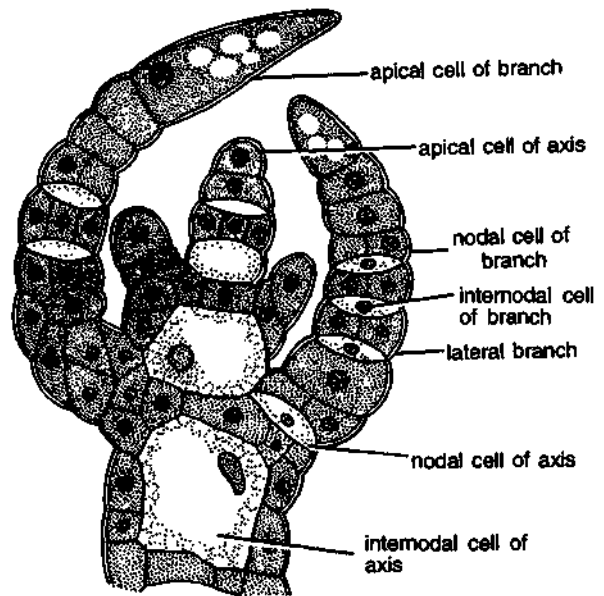


Fig. 2. *Chara*. L.s. apical region

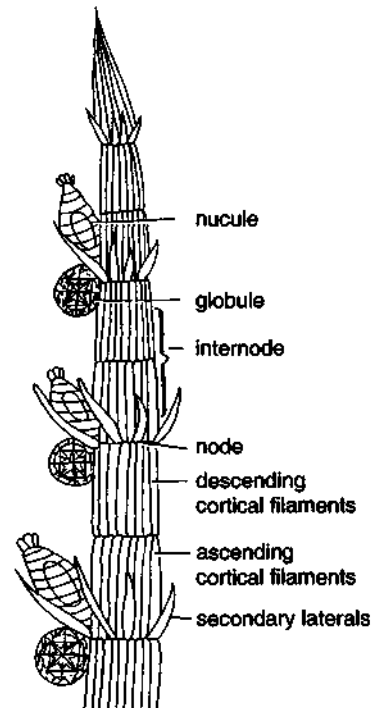


Fig. 3. *Chara*. Part of a fertile branch to show the position of sex organs.

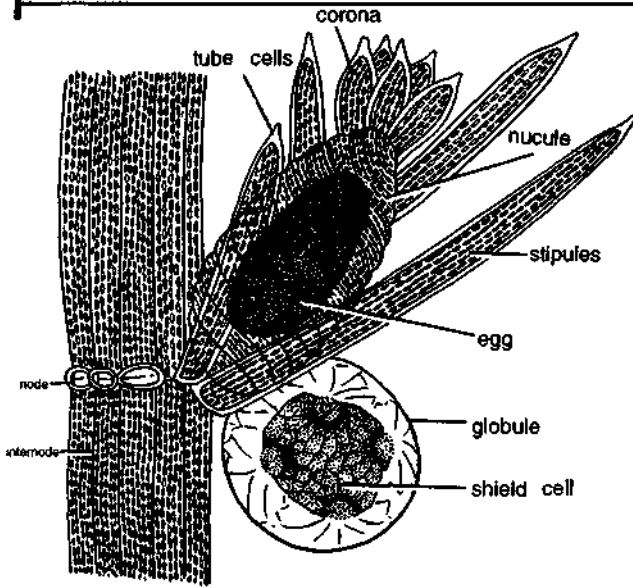


Fig. 4. *Chara*. Node bearing nucule and globule.

4. Biconcave nodal cell in the lower parts divides to produce a mass of peripheral cells which surrounds centrally located nodal cell.
5. Peripheral cells act as initials of the laterals of limited growth.
6. From peripheral cells of node, laterals are produced which show similar arrangement of nodal and internodal cells alternating with one another.
7. Biconvex internodal cell does not divide but in the lower part simply elongates many times.

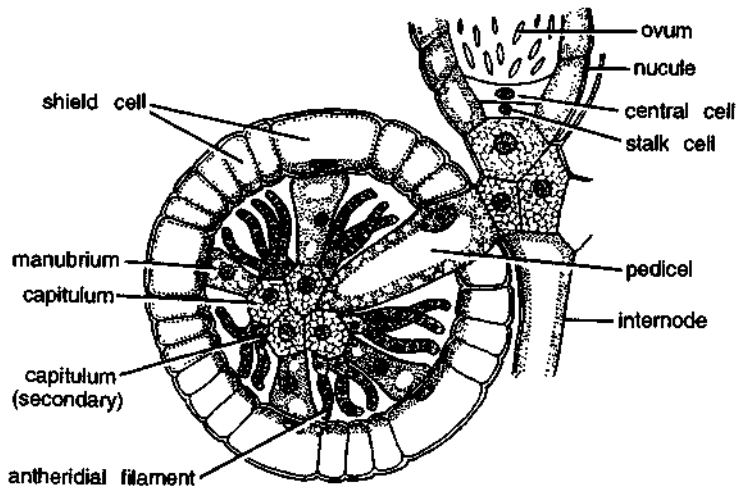


Fig. 5. *Chara*. L.s. globule.

#### Exercise 4

Object : Study of bulbils.

#### Work procedure

Study the slide of a thallus bearing bulbils.

#### Comments

1. Bulbils are present on the knotted part of the rhizoid or the basal nodes of the main axis.
2. These remain buried under the soil.
3. These are oval, tuber-like outgrowths.
4. Bulbils are rich in starch and hence also called amyllum stars.
5. These are the organs of vegetative propagation.

#### Exercise 5

Object : Study of sex organs.

#### Work procedure

Study the slide or a part of thallus bearing sex organs.

#### Comments

1. Most of the species are homothallic (monoecious) while a few are heterothallic (dioecious).

- The male reproductive organ is called a 'globule' while the female reproductive organ a 'nucule'.
- Both the sex organs are borne at one point on the nodes of short laterals which bear stipules.
- The characteristic feature of the genus *Chara* is the position of nuclei at the node above the globule.

### Exercise 6

**Object : Study of male sex organ.**

#### Work procedure

Study the external features of globule, then press it a little to observe internal structure. Also pierce through it by a needle to separate the shield cells and associated structures. Slide of L.s. of globule would also show almost the same structure.

#### Comments

- A globule is a small, spherical and conspicuously red or yellow structure attached to the node by a long stalk cell.
- Outermost wall of the globule is ornamented, composed of eight, large, curved and plate-like cells called shield cells.
- Ornamentation of the shield cell is due to the foldings in the cell wall.

- Each shield cell is attached to a long handle or a rod-shaped cell—manubrium.
- At the tip of each manubrium are two groups of six cells each. The group directly in contact with the manubrium is primary capitulum while the next is secondary capitulum.
- Each secondary capitulum bears 2-4, long and unbranched antheridial filaments.
- Each antheridial filament is made of 100-230 small cells.
- Each of these cells is an antheridium and produces a single biflagellate male gamete

### Exercise 7

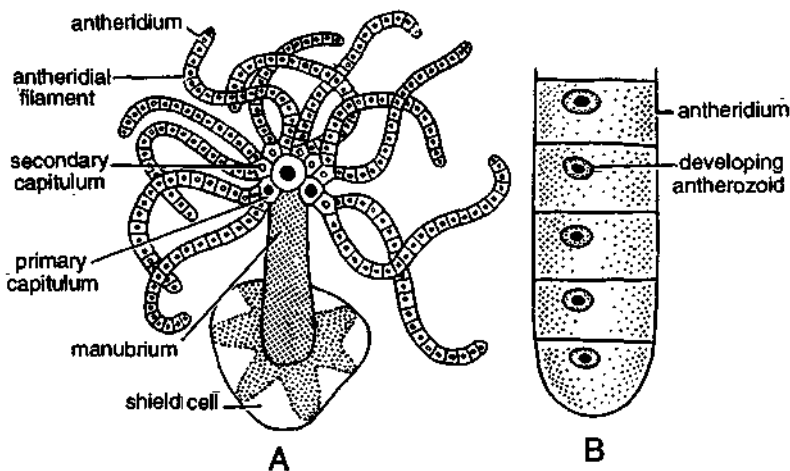
**Object : Study of female sex organ.**

#### Work procedure

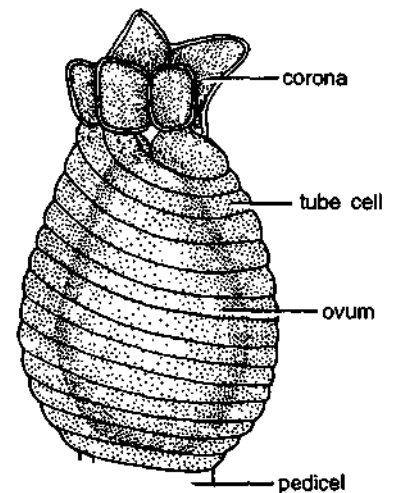
Study the whole mounts of nucule and a zygote.

#### Comments

- Nucule is oval in shape and is situated above the globule at the node.
- It is enveloped by spirally coiled (coiling clockwise) cells—tube cells.
- At the apex of the nucule is a corona of five small cells arranged in one tier and attached at one point.
- Oosphere is single celled where a nucleus lies surrounded by the cytoplasm.



**Fig. 6.** *Chara* A. single shield cell with manubrium and antheridial filaments, B. A part of antheridial filament enlarged.



**Fig. 7.** *Chara*. A nucule (female sex organ).

5. It is rich in food reserves which are in the form of starch and oil.
6. After fertilization the nucule gets modified into a zygote or oospore.
7. The coronal cells at the top of the oospore appear separated.
8. The oospore wall is thick and ornamented. It has a deposition of calcium.

### Identification

Sub-division—**Algae**. (1) simple construction of thallus, (2) Presence of chlorophyll, (3) Cell wall of cellulose.

Class—**Chlorophyceae** (1) Chloroplasts grass-green in colour, (2) Photosynthetic reserve-starch, (3) Motile reproductive bodies flagellated, (4) flagella equal in length.

Order—**Charales**. (1) Thallus differentiated into nodes and internodes, (2) characteristic sex organs—globule and nucule.

Family—**Characeae**. Single family

Genus—**Chara**. (1) Presence of corticating filaments around internodes, (2) Nucule lying above the globule, (3) Corona of nucule five celled.

### Hints for Collection

It is aquatic in habitat. It grows in fresh, clear and standing waters on a muddy or a sandy bottom (epipellic community). These form extensive sub-aquatic growth. *Chara* is found below the water level, growing to a considerable depth. It can be collected, almost from any standing or stagnant reservoir of water and occurs in fruiting state during early and late winters. Many species become encrusted with calcium carbonate and are rough and brittle. *Chara* owes its name 'Stone-worts' to calcium deposition.

## II. XANTHOPHYCEAE

The member of xanthophyceae are simple, only the most advanced being filamentous. Chromatophores are yellow-green due to the predominance of  $\beta$ -carotene. Pyrenoids are absent. Oil or leucosin is present instead of starch.

Cell wall is mostly composed of two equal or unequal halves, overlapping one another. Motile cells are flagellated. Flagella are borne anteriorly and are unequal in length (and, therefore, this group is also known as Heterokontae). A few members produce aplanospores instead of zoospores. Sexual reproduction is rare and, if present, is generally isogamous.

## Vaucheria

### Classification

Sub-division	—	Algae
Class	—	Xanthophyceae
Order	—	Heterosiphonales
Family	—	Vaucheriaceae
Genus	—	Vaucheria

### Exercise 1

**Object : Study of thallus.**

### Work procedure

Study the whole mount showing external features or stain the thallus in safranin and mount in glycerine. Observe the characters.

### Comments

1. Thallus is unicellular, multinucleate, filamentous and branched (coenocytic).
2. Filaments are profusely branched. The branching is lateral but appears dichotomous.
3. Filaments are without any septation (aseptate filaments).
4. If terrestrial in habitat, a few colourless rhizoidal branches are given out which penetrate soil.
5. Cell wall is two layered. Outer layer is composed of pectose while inner is that of cellulose.
6. In the centre lies a big vacuole, continuous throughout the length of the filament.

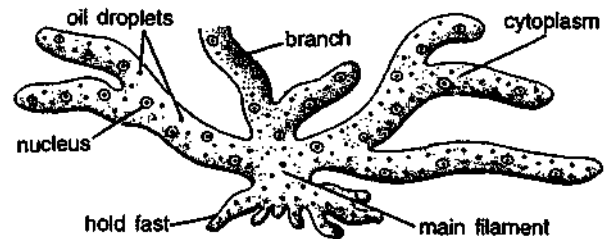


Fig. 1. *Vaucheria*. Thallus to show habit.

7. The cytoplasm lies between vacuole and the cell wall.
8. Many small nuclei are scattered in the cytoplasm near the vacuole.
9. Small chromatophores are also scattered in the cytoplasm. They are circular or elliptical in shape. Pyrenoids are absent.
10. The reserve food material is in the form of small oil droplets.

---

**Exercise 2**

**Object : Study of Gongrosira stage.**

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**Work procedure**

Study the slide showing Gongrosira stage.

**Comments**

1. This stage of asexual reproduction develops under extreme conditions of desiccation or low temperature.
2. It is called Gongrosira because it looks similar to another algal member—*Gongrosira*.
3. In this stage, filament is divided into many, short and thick-walled parts.
4. Each of these parts is called akinete or cyst.
5. Akinetes are thick walled and rich in oil contents.
6. Akinetes occur in long chains, alternating with a part of filament.

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**Exercise 3**

**Object : Study of antheridia.**

---

**Work procedure**

Study a slide showing antheridia.

**Comments**

1. Filaments are mostly monoecious but a few species are dioecious.
2. Sexual reproduction is oogamous.
3. Male reproductive bodies are antheridia and female reproductive bodies are oogonia.
4. The antheridia and oogonia are borne side by side on the same filament. Sex organs are

generally sessile but a stalk-like structure is present in a few species.

5. Antheridia are terminal. These are strongly curved, hook-like and cylindrical.
6. Antheridia are cut off from the main filament by a transverse septum at its base.
7. Protoplast accumulates towards the centre. It produces many biflagellate antherozoids.
8. Antherozoids are liberated through a small pore at the tip of antheridium.

---

**Exercise 4**

**Object : Study of Oogonia.**

---

**Work procedure**

Study a slide showing oogonia.

**Comments**

1. More than one oogonium are present at the tip of the stalks which once again branch at their tips.
2. Oogonia are oval or spherical and terminate into a short beak.
3. The entire protoplast forms a single oosphere.
4. In younger stages, oosphere is multinucleate but at maturity it is always uninucleate.
5. Near the beak, in the apical part, the protoplasm leaves a small colourless area, known as receptive spot.
6. Protoplast is rich in food reserve which is in the form of oil droplets.

---

**Exercise 5**

**Object : Study of zygote.**

---

**Work procedure**

Study a slide showing zygote.

**Comments**

1. Zygote is the result of fertilization.
2. It is present inside the oogonium.
3. It is a thick walled structure being made of 3-7 layers.
4. The protoplast of a zygote is very dense.



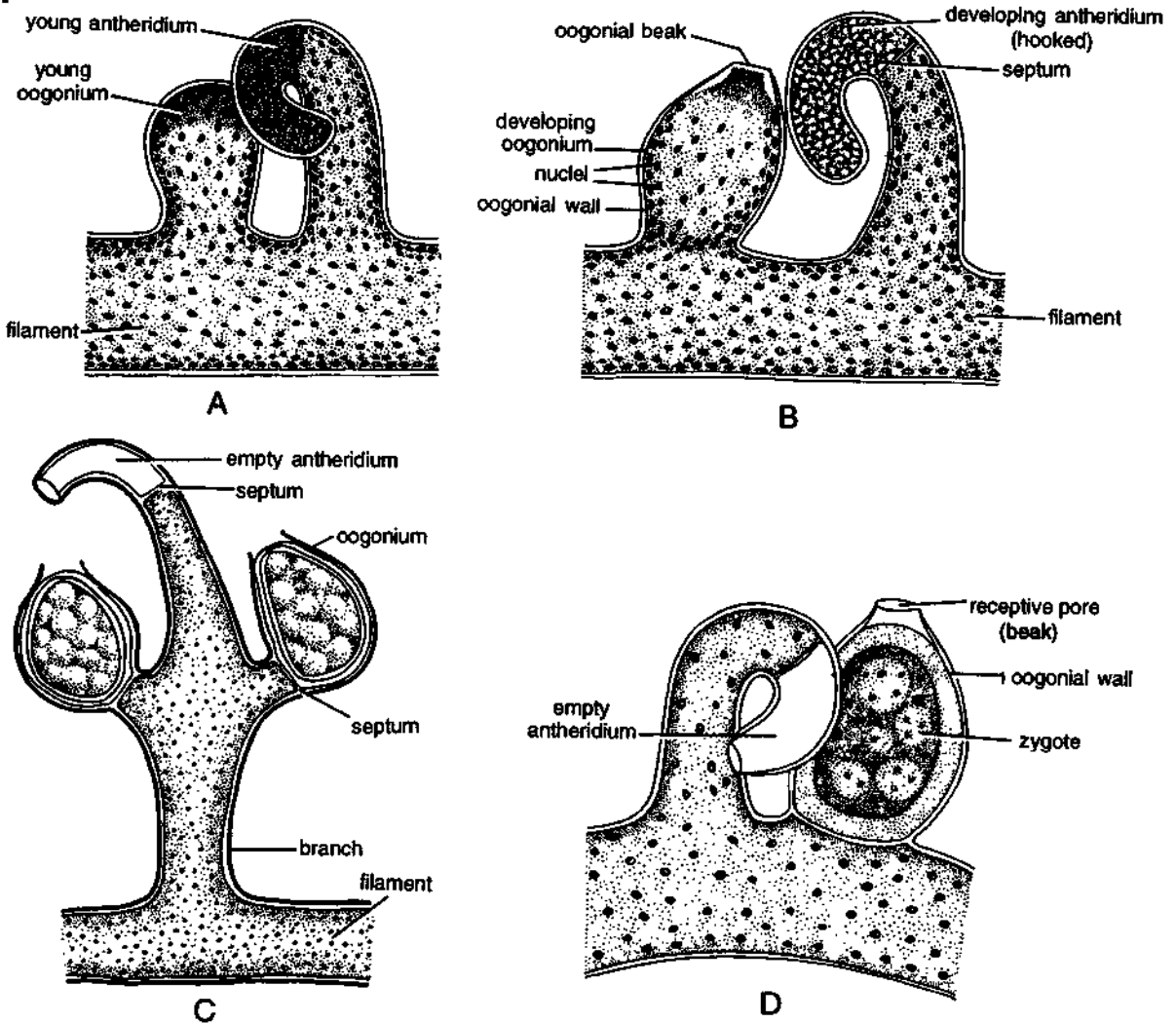


Fig. 2. *Vaucheria*. A-D. Various stages in sexual reproduction. A. Young oogonium with young coiled antheridium, B. Mature oogonium and antheridium, C. Stalked sex organs in *Vaucheria geminata*, D. Empty antheridium and zygote.

5. Numerous oil droplets are scattered throughout the protoplasm.
6. It is considered to be a diploid structure, as such there is said to be possibility of zygotic meiosis.
7. It is liberated through the oogonial beak.

### Identification

**Sub-division—Algae.** (1) Filamentous thallus, (2) Presence of chlorophyll, (3) Cell wall of cellulose.

**Class—Xanthophyceae.** (1) Chromatophores yellow-green, (2) Photosynthetic reserve-oil droplets, (3) Motile cells with unequal flagella.

**Order—Heterosiphonales.** (1) Thallus multinucleate, unicellular and siphonaceous.

**Family—Vaucheriaceae.** (1) Thallus branched, filamentous, tubular and coenocytic, (2) Zoospores multiflagellate, (3) Sexual reproduction oogamous.

**Genus—*Vaucheria*.** (1) Branching irregular or lateral, (2) Sex organs without constriction at the basal septum.

### Hints for Collection

Species of this genus grow in aquatic as well as in terrestrial habitats. Terrestrial species occur on damp soils of gardens, lake sides, ploughed fields and form an extensive green belt on the soil surface,

specially during early winters. The aquatic species also occur as a large green mat floating over the surface of water.

### III. BACILLARIOPHYCEAE

The members of this group are known as diatoms. Diatoms very commonly occur as planktons but are also found as epiphytes. The wall is made of two overlapping valves. Geometrical intricate sculpturing of the wall is due to deposition of silicon dioxide. The walls show either radial or bilateral symmetry. Each cell has one to many yellow or golden-brown chromatophores. The reserve food product is in the form of fats and volutin grains. The commonest method of multiplication is cell division. Under some circumstances, auxospores—the special structures of reproduction, are formed.

The cell walls of diatoms form a large and huge deposit, called 'diatomaceous earth' or 'siliceous earth'. This is commercially important being used in the preparation of metal polishes, tooth pastes, in sugar refineries, etc. It is an important source of food for the aquatic animals. Diatomaceous earth is used for lining the high temperature furnaces where the temperature reaches as high as 1500°C.

There are two major groups of diatoms—

1. **Centrales**—(i) Radially symmetrical,  
(ii) raphe absent,  
(iii) many chromatophores per cell,  
(iv) reproduction by auxospores.
2. **Pennales**—(i) Symmetrical or even asymmetrical,  
(ii) presence of raphe,  
(iii) one or two chromatophores per cell.  
(iv) reproduction by auxospores and statospores.

### Diatoms

(A general account has been given)

#### Classification

Sub-division	—	Algae
Class	—	Bacillariophyceae
Order-1	—	Centrales
Order-2	—	Pennales

#### Exercise 1

Object : Study of a cell.

#### Work procedure

Study a slide or a drop of plankton showing different types of diatoms.

#### Comments

1. Organisms are unicellular.

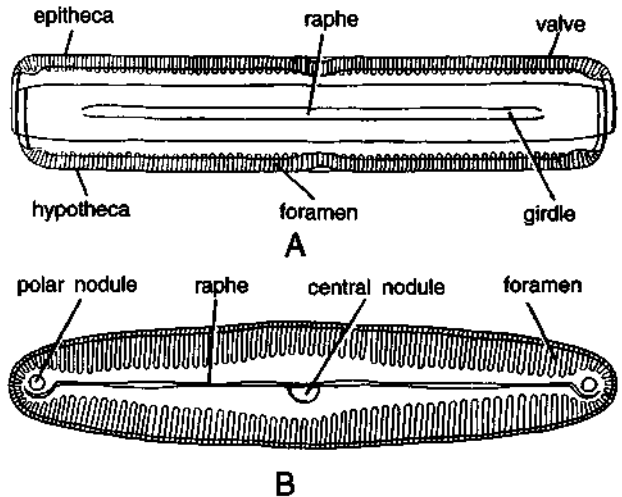


Fig. 1. Diatom, A-B *Pinnularia*, A. Girdle view, B. Valve view of a frustule.

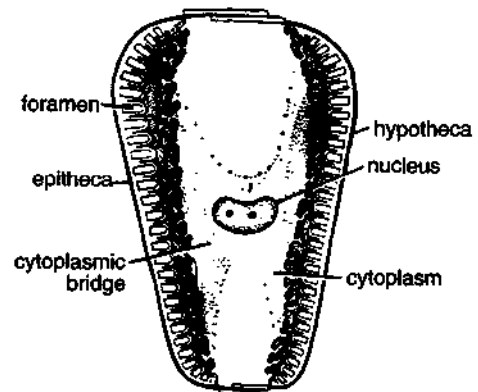


Fig. 2. *Suriella*. Internal structure of a cell.

2. The thallus mostly occurs singly or cells may be united in colonies.
3. A diatom cell is known as a frustule.
4. The wall is composed of two overlapping halves. The older part (valve) is known as epitheca which fits closely over the younger part (valve)—hypotheca.
5. The silicified parts of each valve are more or less flattened. Valves are joined with connecting bands (cingulum).
6. The two connecting bands together form a girdle.
7. The silicification of wall varies
  - (i) In centrales, walls possess areolae or striae arranged radially and symmetrically around a central point,

- (ii) In pennaes, walls are bilaterally symmetrical or asymmetrical with respect to an axial strip.
8. In some pennate diatoms, two systems of striae are separated from one another by a narrow, linear and smooth area occupying apical axis of the valve and is known as pseudoraphe.
  9. In some diatoms (e.g. *Pinnularia*, *Gomphonema*, etc.) the valves possess one central and two polar nodules which are internal thickenings of the wall. A longitudinal slit runs from one polar nodule to another. It is called raphe.
  10. Just inside and close to the cell wall is a cytoplasmic lining. Internal to this is situated a central vacuole. This vacuole is generally interrupted in some forms (e.g. pennate forms) by centrally located prominent band of cytoplasm, in which lies a nucleus. (In centrales this band is absent and nucleus remains near the valve).
  11. Nucleus is very prominent and is round or oval in shape.
  12. Chromatophores vary in shape. These are parietal in position. In pennaes, chromatophores are richly lobed and perforated and contain many naked pyrenoids.
  13. Reserve products are in the form of oils accumulated in large quantities.

### Exercise 1

**Object :** Study of reproductive structures.

#### Work procedure

Study a slide showing auxospores.

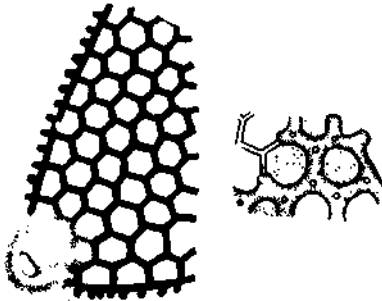


Fig. 3. *Triceratium*. Part of the wall magnified.

#### Comments

1. The common method of multiplication is cell division.
2. During division, two daughter cells of slightly unequal size are formed (comparing the size of the parent cell, one of the daughter cells is always smaller).
3. Continuous cell division results in progressive diminution of size. It is compensated by the formation of auxospores.
4. Auxospores are larger than the vegetative cells from which these are produced.
5. It is primarily the rejuvenation of the protoplast.
6. Sexual reproduction by conjugation is generally observed in pennaes.
7. The zygote formed as a result of sexual reproduction gives rise to a diploid auxospore, that produces a new set of vegetative cells.
8. In centrales, the sexual reproduction is oogamous where spermatozoid is uniflagellate and an egg cell is non-motile.

#### Identification

**Sub-division—Algae.** (1) Thallus simple, (2) Pigment present, (3) Cell walls of cellulose.

**Class—Bacillariophyceae.** (1) Chromatophores 1-2 or more, golden-brown or yellow, with or without pyrenoids, (2) Food reserve oil, (3) The cell wall of two overlapping halves, highly silicified, (4) Reproduction by flagellated swimmers, (5) Sexual reproduction by conjugation.

**Order-1—Centrales.** (1) Valves circular, polygonal or irregular in outline, (2) Ornamentation radial or concentric about a central point, (3) Raphe or pseudo-raphé absent.

**Order-2—Pennaes.** (1) Valves bilaterally symmetrical or asymmetrical in surface view, (2) Ornamentation always bilaterally symmetrical with respect to a line, (3) Raphe and pseudo-raphé present.

#### Hints for Collection

The diatoms are cosmopolitan in distribution being present in almost all the habitats. These constitute major part of the freshwater and marine planktons. In sea-water diatoms form bottom flora. Besides these habitats, diatoms may occur on soil, rock cliffs, bark of the trees, etc.

## IV. PHAEOPHYCEAE

The phaeophyceae or brown algae are distinctly marine, inhabiting littoral regions of Arctic and Antarctic seas. Marine brown algae form a clear vertical zonation on the rocks in the sea.

These algae are always multicellular and microscopic or macroscopic. A few members show external differentiation of the thallus into holdfast, stem-like axis and leaf-like blades. Internal differentiation, including tissue differentiation into sieve tube-like structures is also found.

Reproduction involves vegetative reproduction by fragmentation. Asexual reproduction takes place by biflagellate zoospores and non-flagellate tetraspores. Flagellated structures are typically pyriform in shape with two laterally inserted flagella.

Sexual reproduction ranges from isogamy to oogamy. In oogamy, one to eight female gametes are released from oogonium. The antherozoids are uni- or biflagellate. Isomorphic or heteromorphic alteration of generations is also seen in many of the members of the group.

### *Ectocarpus*

#### Classification

Sub-division	—	Algae
Class	—	Phaeophyceae
Order	—	Ectocarpales
Family	—	Ectocarpaceae
Genus	—	<i>Ectocarpus</i>

#### Exercise I

**Object :** Study of external features of thallus.

#### Work procedure

Mount a few filaments in glycerine after staining in safranin. Study the structure of thallus and also a single cell.

#### Comments

1. Thallus is multicellular, filamentous and branched.
2. Filaments are heterotrichous and differentiated into (i) prostrate portion and (ii) an erect portion.
3. In some species prostrate portion is irregularly and profusely branched or altogether absent. If present it remains attached to the

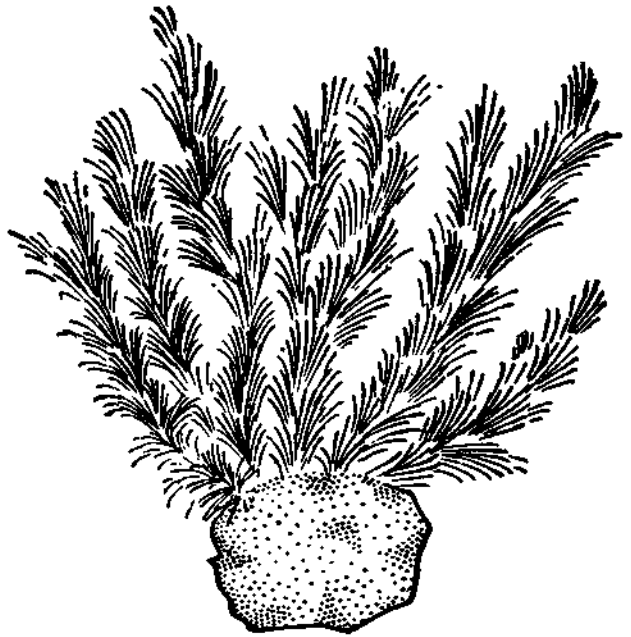


Fig. 1. *Ectocarpus*. Habit

- substratum.
4. Erect portion is a crowded tuft of branches. Main axis is broad from which lateral branches arise just beneath the septum and taper into a point. Ultimate branches give an appearance of a hair.

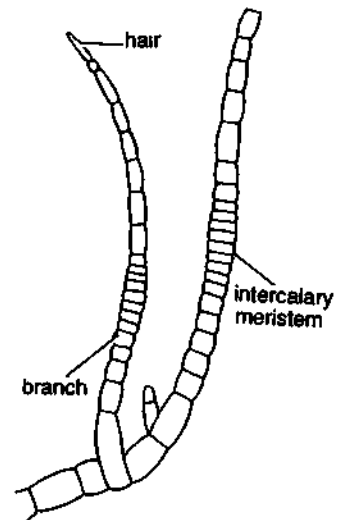


Fig. 2. *Ectocarpus*. A part of the filamentous thallus.

5. The branches and the main axis are uniseriate.
6. The erect branches have intercalary meristem just below the terminal hair. It results in trichothallic growth.
7. The cells are squarish to cylindrical and uninucleate.
8. The cell wall is double layered. Outer is gelatinous and inner is firm and cellulosic.
9. There may be one or many chromatophores varying from irregular to band-like to discoid. Pyrenoids are absent.
10. Reserve food products occur as shining fucosan granules.
11. The cell is filled with cytoplasm in which lies a single nucleus.

### Exercise 2

**Object :** Study of unilocular sporangium.

#### Work procedure

Search for filaments with unilocular sporangia, stain in safranin, mount in glycerine and study.

#### Comments

1. Unilocular sporangium is a structure of asexual reproduction, always present on diploid plants.
2. The sessile or stalked sporangium is situated terminally on lateral branches.
3. The shape varies from globose to ellipsoidal.
4. It is single celled and uninucleate when young but becomes multinucleate later.
5. Many biflagellate zoospores are produced when unilocular sporangium matures.

### Exercise 3

**Object :** Study of plurilocular sporangium.

#### Work procedure

Search for filaments with plurilocular sporangia, stain in safranin, mount in glycerine and study.

#### Comments

1. These occur on both haploid and diploid plants.

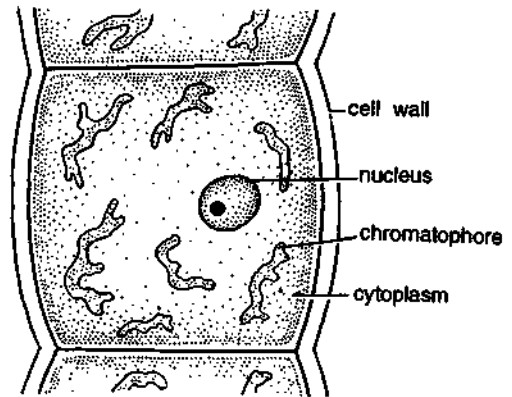


Fig. 3. *Ectocarpus*. A single cell.

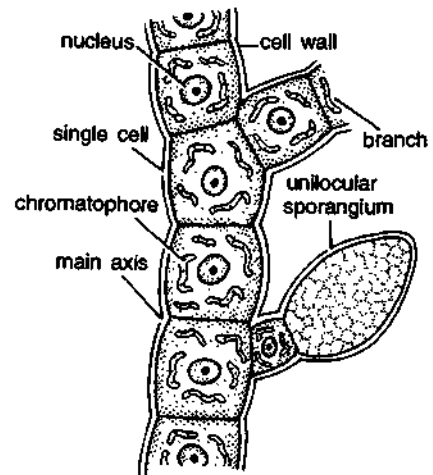


Fig. 4. *Ectocarpus*. A branch with unilocular sporangium.

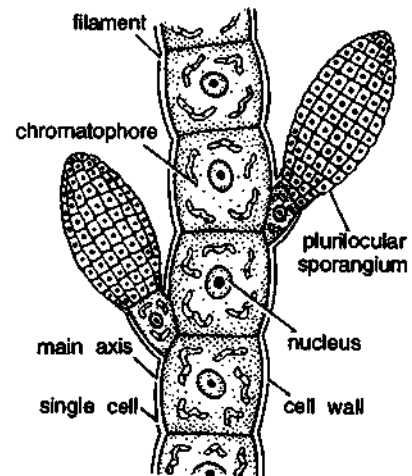


Fig. 5. *Ectocarpus*. Filament with plurilocular sporangia.

- The structure when borne by haploid plant serves as a gametangium whereas on diploid plant it functions as a sporangium.
- Sporangia situated laterally may be sessile or stalked. These may be ovate to siliquose.
- Plurilocular sporangium is divided into large number of cells.
- A mature sporangium produces biflagellate swimmers, one each from every cell.
- If plurilocular sporangium is borne on a haploid plant, the swimmers behave as gametes and if borne on diploid plant, these act as haploid zoospores.

(It is, therefore, suggested that instead of plurilocular sporangium it be called as neutral sporangium when zoospores are produced and spores as neutral spores and if it produces gametes on a haploid plant, it should be called gametangium and swimmers as gametes).

The life cycle exhibits typical alternation of generations (isomorphic).

### Identification

**Sub-division—Algae.** (1) Simple thallus, (2) Presence of chlorophyll, (3) Cell wall of cellulose.

**Class—Phaeophyceae.** (1) Yellowish-brown chromatophores, (2) Photosynthetic reserve—laminarin and mannitol, (3) Motile reproductive cell—pyriform and flagellated, (4) Flagella laterally inserted and unequal.

**Order—Ectocarpales.** (1) Thallus filamentous, (2) Growth trichothallic, (3) Reproductive organs-unilocular and plurilocular sporangia, (4) Isomorphic alternation of generations.

**Family—Ectocarpaceae.** (1) Thallus monoaxial, branched, branches uniseriate, (2) Growth trichothallic, (3) Sporophytes with uni- or plurilocular sporangia, terminal or intercalary.

**Genus—Ectocarpus.** (1) Chromatophores discoid or band-shaped, (2) Pyrenoids absent, (3) Reproductive parts terminal, stalked.

### Hints for Collection

It occurs in marine habitat the world over, mostly along the coasts of colder seas. It also grows along the Indian coasts and forms brown tufts attached to rocks and large kelps in littoral and sublittoral regions.

## Fucus

### Classification

Sub-division	—	Algae
Class	—	Phaeophyceae
Order	—	Fucales
Family	—	Fucaceae
Genus	—	Fucus

### Exercise 1

**Object :** Study external features of thallus.

### Work procedure

Study the external features of specimen provided.

### Comments

- Thallus is flat and dichotomously branched.
- It is attached by a rounded disc-shaped holdfast.
- A mid-rib stands erect from the holdfast. It is prominent in older parts of the thallus than in younger regions.

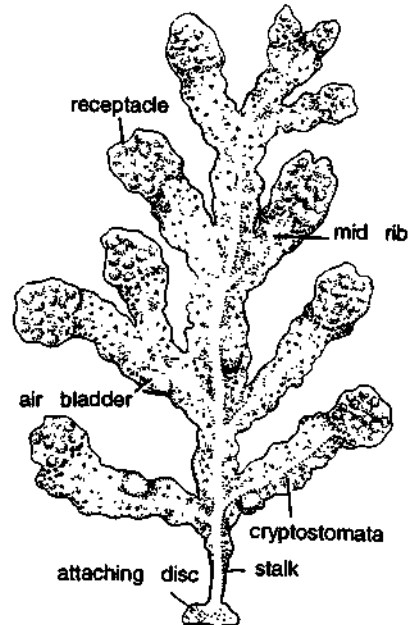


Fig. 1. *Fucus*. External features of thallus.

4. Thallus bears many flat strap-like, dichotomously branched blades or wings with smooth or entire margins.
5. Some species of *Fucus* bear air bladders within the thallus. These regions of the thalli appear inflated.
6. Wings show small openings of sterile conceptacles (also known as cryptostomata or cryptoblasts).
7. Fertile conceptacles are terminal. These swollen parts which lack midrib are called receptacles.

### Exercise 2

**Object : Study of internal structure of wing.**

#### Work procedure

Place a small piece of wing in pith, cut a T.s., stain in safranin and mount in glycerine.

#### Comments

1. Mature wing is internally differentiated into—(i) meristoderm, (ii) cortex and (iii) medulla.
2. The **meristoderm**. It is enveloped by thin cuticle consisting of small rectangular and closely compacted cells filled with dark brown plastids. This colour is due to a carotene—fucoxanthin (chlorophyll in these cells can be demonstrated by immersing a portion of the wing in freshwater. This dissolves fucoxanthin and chlorophyll is visible).
3. **Cortex** is composed of relatively large cells, size of which gradually increases toward the

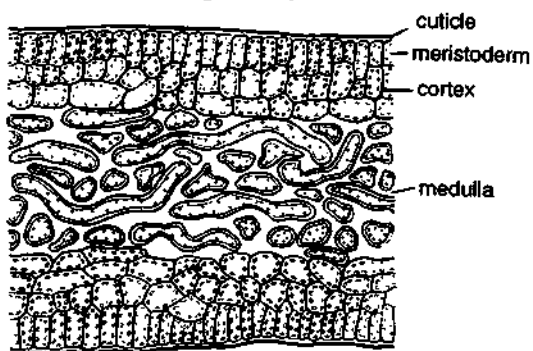


Fig. 2. *Fucus*. A part of T.s. through wing.

centre. The number of plastids in the cells gradually decreases towards medulla.

4. **Medulla** is located centrally. Cells are narrow, filamentous and loosely arranged. Spaces between the cells are filled with mucilage.
5. Along with medullary cells, running longitudinally are elongated cells—hyphae (which give mechanical support to the thallus).

### Exercise 3

**Object : Study of internal structure of mid rib.**

#### Work procedure

Place a small piece of blade in pith, cut a T.s., stain in safranin and mount in glycerine. Study only a part of midrib of the section.

#### Comments

1. Tissues are differentiated into—(i) outermost meristoderm, (ii) middle cortex and (iii) innermost medulla.
2. **Meristoderm** consists of small, rectangular and compacted cells arranged in a single layer. It is covered with a thin cuticle. The cells are rich in chromatophores.
3. **The cortex** forms a wide zone. The cells near the periphery are closely compacted but become loose and larger in size towards the centre.
4. **Medulla** shows irregular and longitudinal arrangement of hyphae, their narrow and thick nature making a compact and solid central tissue.

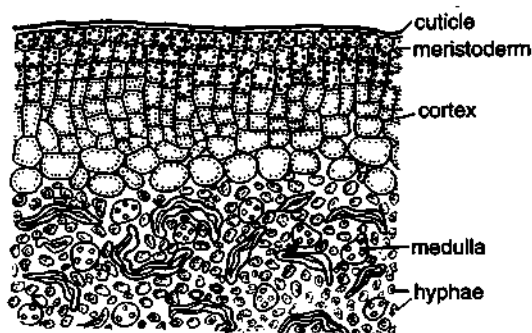


Fig. 3. *Fucus*. A part of T.s. through mid-rib.

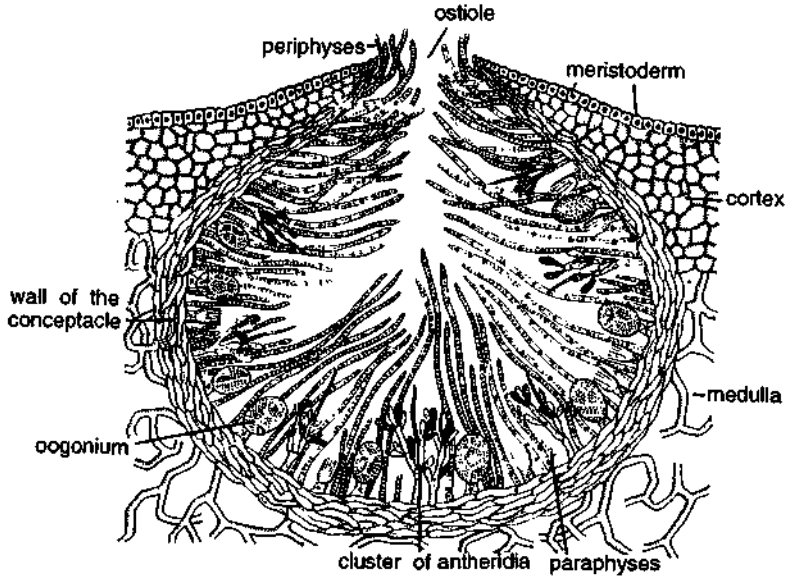


Fig. 4. *Fucus*. T.s. through conceptacle.

#### Exercise 4

**Object :** Study the structure of male conceptacle.

#### Work procedure

Study a slide showing male conceptacles and T.s. of male conceptacle.

#### Comments

1. Plants are either monoecious or dioecious.
2. The swollen tips of the branches are called receptacles.
3. Each receptacle has many cavities, each being known as conceptacle.
4. In monoecious species, antheridia and oogonia are present either in the same conceptacle or in two different conceptacles of the same plant.
5. In dioecious species antheridia and oogonia bearing conceptacles are found on male and female plants respectively.
6. T.s. of male conceptacle shows a flask-shaped cavity opening by a pore, called ostiole.
7. A few hair-like periphyses project outside from ostiole.
8. The wall of conceptacle is continuous with the external layer.
9. The floor of the cavity bears multicellular hair-like paraphyses and antheridia.

10. Antheridia are located on highly branched antheridial hairs or on short hairs arising from the wall of the conceptacle.
11. Antheridium is stalked, unicellular, oval with double layered wall.
12. Antheridium produces many pear-shaped, uninucleate and biflagellate antherozoids.

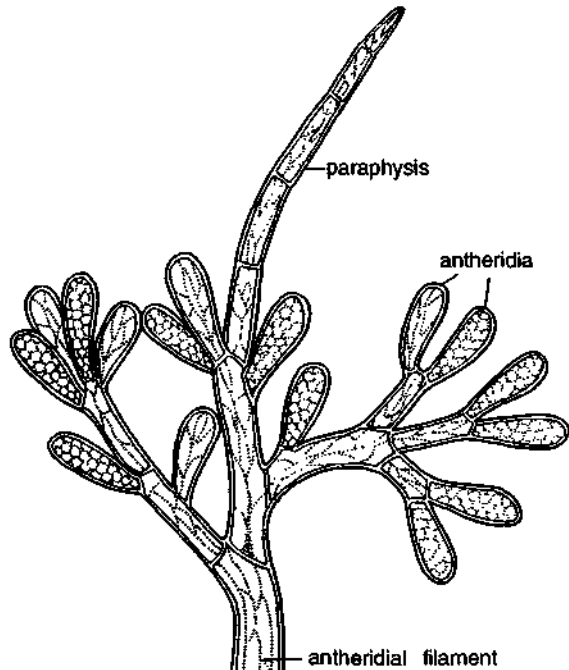


Fig. 5. *Fucus*. Antheridial filaments.



### Exercise 5

**Object :** Study of structure of female conceptacle.

#### Work procedure

Study a slide showing female conceptacles and T.s. of female conceptacle.

#### Comments

1. Plants are either monoecious or dioecious.
2. The swollen tips of the branches are called receptacles.
3. Each receptacle has many female conceptacles.
4. In monoecious species antheridia and oogonia are present either in the same conceptacle or in two different conceptacles on the same plant.
5. In dioecious species, antheridia and oogonia bearing conceptacles occur on male and female plants respectively.
6. T.s. of female conceptacle shows a flask shaped structure, the cavity of which opens by a pore called ostiole.
7. A few hair-like structures called periphyses project from near the ostiole.
8. The wall of the conceptacle is continuous with the external layer.
9. The floor of the cavity bears many multicellular and branched paraphyses and oogonia.
10. Oogonia arise singly, directly from the wall of the conceptacle.
11. Oogonium is shortly stalked, globose, swollen and has three layered thick wall.
12. Mature oogonium shows eight oospheres or eggs.

#### Identification

**Sub-division—Algae.** (1) Thallus simple, (2) Presence of chlorophyll, (3) Cell wall of cellulose.

**Class—Phaeophyceae.** (1) Yellowish-brown chromatophores, (2) Photosynthetic reserves—laminarin and mannitol, (3) Motile reproductive cells—pyriform and flagellated, (4) Flagella laterally inserted and unequal.

**Order—Fucales.** (1) Plants parenchymatous with complex morphological and anatomical differentiation, (2) Medulla filamentous, (3) Absence of asexual reproduction, (4) Sex organs in conceptacles.

**Family—Fucaceae.** (1) Axes subterete to alate with mid-rib, but not foliar, (2) Vesicles if present intercalary, (3) Oogonia with eight oospheres.

(B-14)

**Genus—Fucus.** (1) Plants erect, (2) Holdfast disciform or irregular, (3) Branching dichotomous or sub-pinnate, (4) Branches strap-shaped with a more or less distinct mid-rib.

#### Hints for Collection

Alga is exclusively marine. It is found attached to the rocks in the intertidal rocky coasts of the colder seas of the northern hemisphere. It is abundantly found along the coasts of British Isles, Northern European countries and Atlantic coast of America. During summers, rocks exposed to low tide remain exclusively covered with *Fucus*.

### Sargassum

#### Classification

Sub-division	—	Algae
Class	—	Phaeophyceae
Order	—	Fucales
Family	—	Sargassaceae
Genus	—	Sargassum

#### Exercise 1

**Object :** Study of external features of thallus.

#### Work procedure

Study the external structure of thallus.

#### Comments

1. Thallus is erect, thalloid and branched.
2. It remains attached to the substratum by a discoid holdfast.
3. Main axis stands out from the holdfast. It varies from a few to many centimeters in height.
4. Main axis bears large number of primary laterals forming a larger part of vegetative structure. Branches are radially symmetrical and spirally arranged.
5. Secondary branches are repeatedly branched.
6. Many branches are flattened along the plane of branching into leaf-like structures called 'leaves'.
7. Leaves are narrow and their margins are mostly serrate. A few species also show a clear mid-rib.

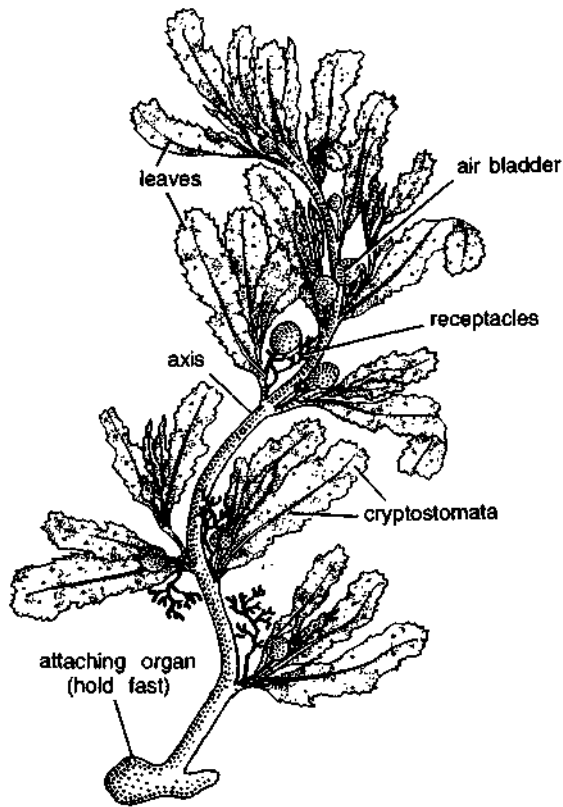


Fig. 1. *Sargassum*. Thallus to show external features.

8. In the lower parts, leaves are replaced by air bladders. However, leaf or its part is modified almost at any place into an air bladder.
9. Leaves show minute pores on both of the surfaces which are ostioles (or openings) of conceptacles (sterile) or cryptostomata or cryptoblasts.
10. In the axils of foliaceous branches (leaves) is situated a series of repeatedly branched receptacles which bear reproductive structures.

### Exercise 2

**Object :** Study the internal structure of axis.

#### Work procedure

Place a small piece of axis in the pith after removing the leaves. Cut a T.s., stain in safranin and mount in glycerine to study the internal structure.

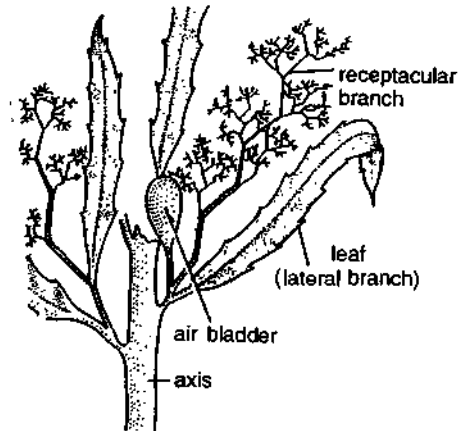


Fig. 2. *Sargassum*. A part of thallus to show details.

### Comments

1. The section is almost circular in outline.
2. It is differentiated into three regions :  
(i) meristoderm, (ii) cortex and (iii) medulla.
3. **Meristoderm** is the outermost single celled layer of meristematic cells. It consists of many, small and compactly placed cells covered by mucilage. Cells are rich in chromatophores and reserve food material.
4. Cells of the meristoderm are photosynthetically active and, therefore, constitute **assimilatory region**.
5. **Cortex** forms the major part of the axis. Cells are narrow and elongated with many intercellular spaces. Cells possess large amount of reserve food material. This region is also known as **storage region**.
6. **Medulla** occupies the central part of the axis. It consists of narrow, elongated and double-walled cells, inner wall being thin than the outer.
7. The medulla transports water and essential nutrients. Hence, it is also called as **conducting region**.

### Exercise 3

**Object :** Study of internal structure of leaf.

#### Work procedure

Place a leaf in the pith, cut a T.s., stain in safranin and mount in glycerine to study the internal structure.

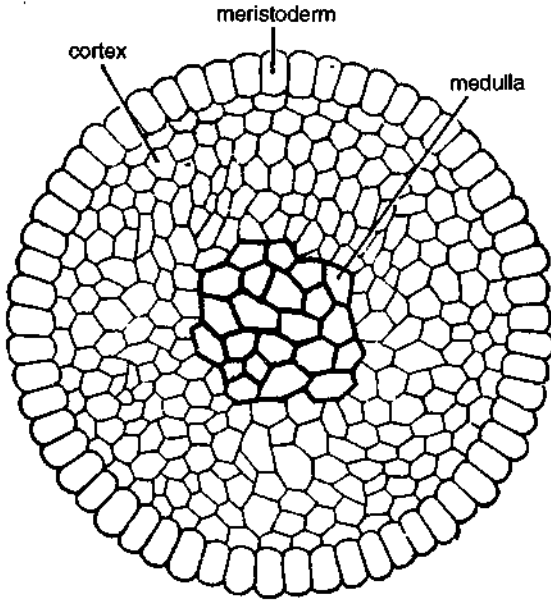


Fig. 3. *Sargassum*. T.s. axis.

### Comments

1. T.s. of leaf shows 3 regions similar to those found in axis. These are—meristoderm, cortex and medulla.
2. **Meristoderm** is the outermost layer. Cells are small, compactly arranged and rich in chromatophores and reserve food.
3. **Cortex** is a major part of the tissues. Cells are thin and contain large amount of reserve food material.
4. **Medulla** occurs only in central region of the leaf indicating mid-rib. It is absent from the wings.
5. In the leaf many sterile conceptacles are distributed on both of its surfaces (also known as cryptostomata or cryptoblasts).
6. Each cryptoblast opens to the exterior by an opening—ostiole (visible externally as black dots or pores).

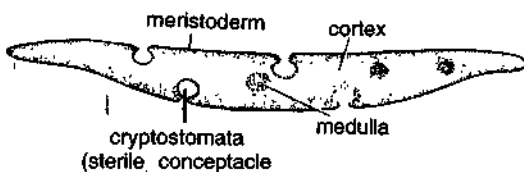


Fig. 4. *Sargassum*. T.s. leaf (diagrammatic).

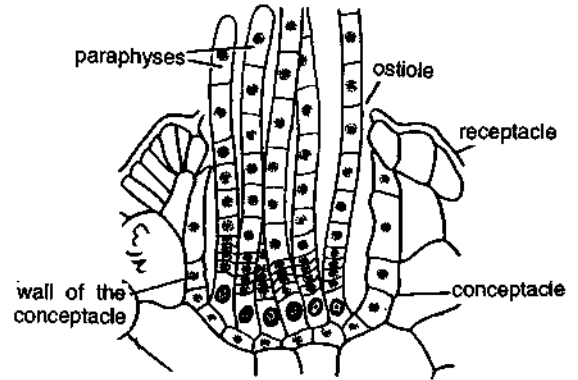


Fig. 5. *Sargassum*. L.s. through conceptacle.

7. Below an ostiole is situated a flask-shaped cavity—conceptacle. The wall of this cavity is lined by cells.
8. The floor of the wall bears many multicellular and unbranched hair called paraphyses. These protrude outside through an ostiole.
9. Thickness of the leaf is maximum in the mid-rib region and decreases toward the wings.

### Exercise 4

**Object :** Study of internal structure of air bladder.

### Work procedure

Place a leaf in the pith, cut a T.s. through swollen air bladder, stain the section in safranin, mount in glycerine and study the internal structure.

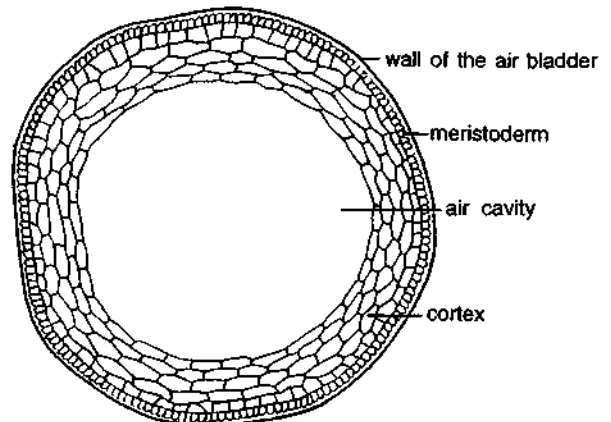


Fig. 6. *Sargassum*. T.s. through air bladder.

### Comments

1. Air bladder appears circular in outline.
2. It is enveloped by a distinct mucilage.
3. T.s. shows outermost meristoderm, followed by cortex and the central air cavity.
4. Meristoderm is made of radially elongated thin walled cells. It is followed by a large and thin-celled cortex.
5. In the centre is a large air cavity filled with gases.
6. Air bladders help in gaseous exchange and buoyancy.

### Exercise 5

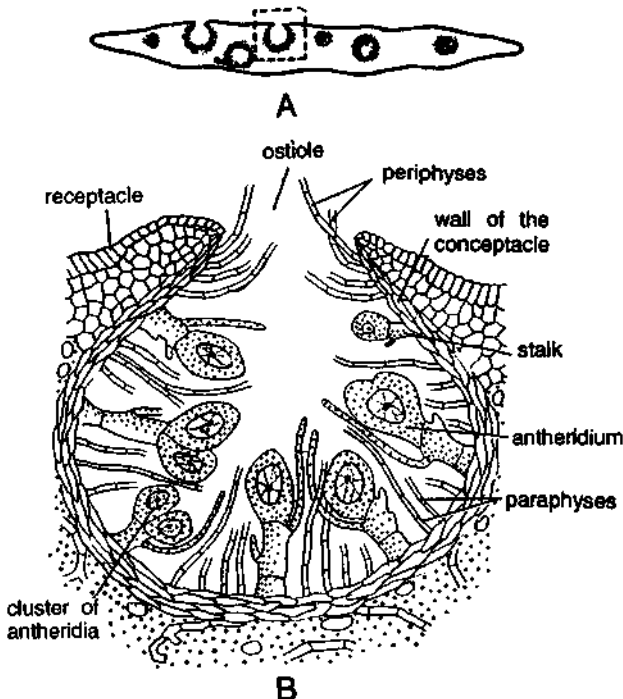
**Object :** Study of internal structure of male conceptacle.

### Work procedure

Study a slide showing T.s. of male conceptacle.

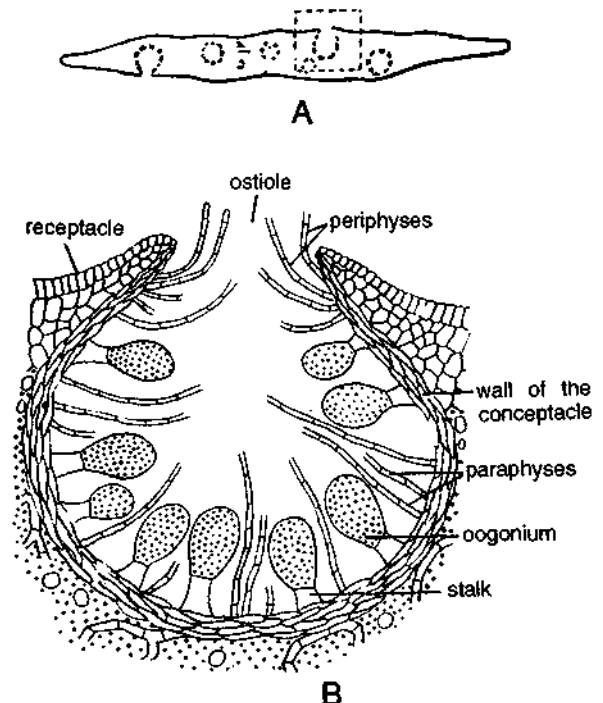
### Comments

1. Plants may be monoecious or dioecious.



**Fig. 7. *Sargassum*.** A-B. A. T.s. through male receptacle, B. A male conceptacle as shown in fig. B enlarged.

2. Antheridia are found in male conceptacles.
3. Conceptacles occur only in specialized branch system called receptacle or receptacular branch.
4. Male conceptacles are externally smooth.
5. Many conceptacles are found in a male receptacular branch.
6. Each conceptacle is a flask-shaped cavity opening by a pore called ostiole.
7. Wall of the conceptacle is made of small and flat cells rich in chromatophores.
8. Numerous multicellular hairs arising from near the ostiole project outside. These are called paraphyses.
9. Other types of multicellular hairs arising from the floor of the cavity are called paraphyses.
10. Some of paraphyses are branched and hold one or more antheridia at the tips of the branches.
11. Each antheridium has a thick wall made of two layers.
12. On maturity about 64 biflagellate antherozoids are produced.



**Fig. 8. *Sargassum*.** A-B. A. T.s. through female conceptacle, B. A female conceptacle as shown in fig. B enlarged.

### Exercise 6

**Object :** Study of internal structure of female conceptacle.

#### Work procedure

Study a slide showing T.s. of female conceptacle.

#### Comments

1. The plants may be monoecious or dioecious.
2. Oogonia are found in female conceptacles.
3. Conceptacles occur only in specialized branch system called receptacle or receptacular branch.
4. The female receptacular branch is spinous.
5. It bears many female conceptacles.
6. Conceptacle is a flask-shaped cavity opening by a pore called ostiole.
7. Many multicellular, unbranched hairs arising from near the ostiole called periphyses protrude outside.
8. The wall of the conceptacle is lined by small and flat cells, rich in chromatophores.
9. A few multicellular, unbranched hairs arise from the floor of the cavity and are called paraphyses.
10. Numerous oogonia arise directly from the wall of the conceptacle.
11. Oogonium is sessile or shortly stalked (most of the stalk cell being embedded in the wall).
12. Each oogonium is oval to sub-spherical with a three layered wall.
13. At maturity oogonium has a single, large and uninucleate egg.

#### Identification

**Sub-division—Algae.** (1) Simple thallus, (2) Chlorophyll present, (3) Cell wall of cellulose.

**Class—Phaeophyceae.** (1) Chromatophores yellowish-brown, (2) Photosynthetic reserves—laminarin and mannitol, (3) Motile reproductive cells—pyriform and flagellated, (4) Flagella laterally inserted and unequal.

**Order—Fucales.** (1) Plants parenchymatous, morphologically and anatomically differentiated, (2) Medulla filamentous, (3) Asexual reproduction absent, (4) Sex organs in conceptacles.

**Family—Sargassaceae.** (1) Axes terete, bearing distinct foliar organs, (2) Vesicles usually present, lateral or immersed in the terminal branchlets, (3) Branching of the thallus radial to the central axis.

**Genus—Sargassum.** (1) Foliar organs narrow, branched, leaf-like with a distinct mid-rib, (2) Vesicles generally lateral, (3) Fertile branches (receptacles) lateral, or terminal panicles.

#### Hints for Collection

It is marine in habitat and remains restricted to tropical seas, mainly of southern hemisphere. In India, species of this genus are found along the east coast, west coast, and Andman and Nicobar islands.

## V. RHODOPHYCEAE

Majority of red algae are marine. However, about a dozen or more genera with approximately 53 species occur in fresh water habitats. Marine species are distributed in almost all seas, including the Arctic and Antarctic, majority being found in polar seas. These form belts in the littoral region and also inhabit deeper waters. Freshwater members grow in well aerated waters of falls and rapidly flowing cold streams.

Colour of the thalli ranges from pure red to dark purple due to the presence of pigments r-phycoerythrin and r-phycoeyanin. Assimilatory product is either Floridean starch or Floridoside.

Thalli exhibit a great range of organization. It may be filamentous but most of the members are multicellular i.e. either monosiphonous or polysiphonous. Thalli are macroscopic and are either radially symmetrical or considerably compressed. Thallus consists of a system of filaments, sometimes simple, but mostly complex, resulting into a corticated structure built around a single central filament or a medulla of intertwining filaments. Members of florideae show pit connections between the adjacent cells. Chromatophores are mostly stellate but vary in shape. These are centrally located. Pyrenoid is a dense proteinaceous body without a surrounding sheath of starch grains, therefore, these pyrenoids are called naked.

Asexual reproduction takes place by many different types of spores (e.g. monospores, etc.). Sexual reproduction exhibits a high degree of oogamy and is complex. It is brought about by spermatangium (antheridium) and carpogonium (oogonium).

Male gametes termed as spermatia are non motile. These depend upon water current for their transportation to female gamete. Female reproductive organ is a one celled carpogonium. It possesses a tubular outgrowth—a receptive organ known as trichogyne, situated at its anterior end.

Post-fertilization changes are complex and result into a phase—carposporophyte. It lives parasitically on female gametophyte. Carposporophyte produces carpospores which on germination give rise to another diploid plant—a tetrasporophyte. Accordingly two types of life cycles are recognised.

(1) Two haploid phases—(i) Sexual individuals and (ii) carpospore bearing stages alternating with diploid phase (zygote nucleus). The life cycle is known as haplobiontic.

(2) Two diploid phases—(i) diploid carpospore bearing carposporophyte, and (ii) tetrasporic individual alternating with

single haploid phase (sexual individuals). This type of life cycle is known as diplobiontic.

Economically red algae are of importance. Agar—a colloid, useful as a microbiological culture medium, pharmaceutical emulsifying agent and necessary in baking and confectionery industries is obtained from a group of red algae popularly termed as agarophytes.

## Batrachospermum

### Classification

Sub-division	—	Algae
Class	—	Rhodophyceae
Sub-class	—	Florideae
Order	—	Nemalionales
Family	—	Batrachospermaceae
Genus	—	Batrachospermum

### Exercise 1

**Object :** Study the external features of thallus.

#### Work procedure

Take out a few filaments, stain in safranin and mount in glycerine. Study the external features.

#### Comments

1. Thallus is multicellular and filamentous. Filaments are branched.
2. Adult thallus appears as a chain of beads. It is mucilaginous and violet or blue in colour.
3. Thallus remains attached to the substratum by old shoots which form a prostrate system.
4. The main axis (primary axis) is corticated. It is differentiated into nodes and internodes. Branches are borne at the nodes.
5. Two types of branches occur—(i) branches of unlimited growth and (ii) branches of limited growth.
6. Primary axis and branches of unlimited growth arise from nodes. These show monopodial or pseudo-dichotomous branching.
7. Long branches or branches of unlimited growth are differentiated into small nodes and long internodes.

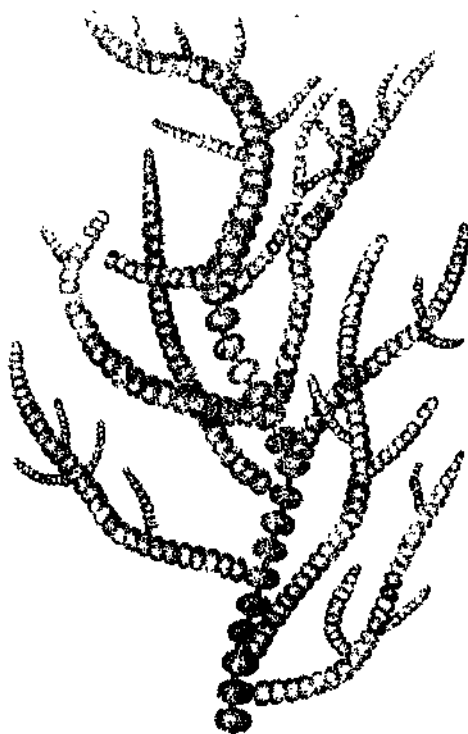


Fig. 1. *Batrachospermum*. Thallus to show external features.

8. Primary axes and branches of unlimited growth are enveloped by several layered cortex.
9. Dwarf branches or branches of limited growth arise laterally in whorls from the nodes of primary axes. (Clusters thus formed give beaded appearances to the thallus).
10. Each cluster formed at the node, is called a glomerule.
11. The laterals consist of small, ellipsoidal or moniliform and uninucleate cells.
12. Among these lateral branches are situated large clusters of carospores.

### Exercise 2

**Object :** Study of reproductive structures.

#### Work procedure

Mount a few filaments in a drop of water, search for reproductive structures. Stain filaments with reproductive organs with safranin, mount in glycerine and study.

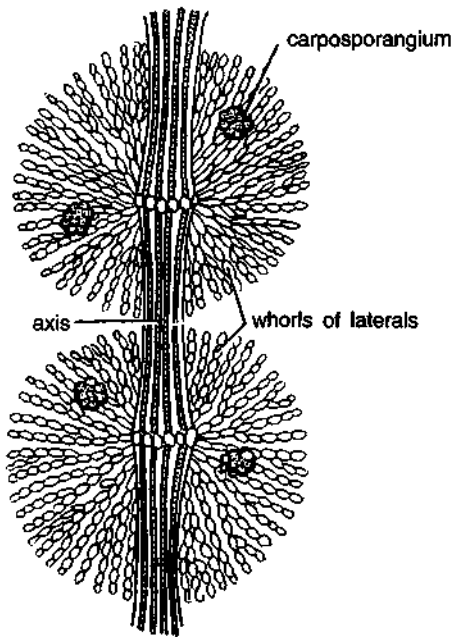


Fig. 2. *Batrachospermum*. A part of fertile branch with glomerule and carposporangia.

### Comments

1. The species are monoecious and male and female sex organs occur near the apex.
2. Male sex organs are antheridia. These are present in clusters on short branches of lateral filaments.
3. Antheridia are oblong or spherical and unicellular.
4. Each antheridium produces a single, spherical, colourless, naked, uninucleate and non-motile spermatium.
5. Female sex organs are carpogonia situated at the apex of 3-4 celled lateral carpogonial branch.
6. Carpogonium is made of a basal swollen portion with a terminal, elongated, tubular process called trichogyne.
7. As a result of fertilization cystocarp is formed. This appears as a cluster of carpospores in glomerules.
8. Cystocarp remains covered by sterile branches.
9. Inside the cystocarp lie many branched gonimoblast filaments.
10. The terminal swollen cells of these filaments are carposporangia. Each carposporangium produces a single carpospore.

### Identification

*Sub-division—Algae.* (1) Thallus simple, (2) Presence of chlorophyll, (3) Cell walls of cellulose.

*Class—Rhodophyceae.* (1) Chromatophores pure red to dark purple, (2) Photosynthetic reserve—floridean starch and floridoside, (3) Male gametes non-motile, (4) Female reproductive organ with trichogyne—a receptive structure, (5) Post-fertilization product a cystocarp.

*Sub-class—Florideae.* (1) Thallus basically filamentous, (2) Pit connections between sister cells, (3) Cells with more than one chromatophore, (4) Carpogonium highly specialised.

*Order—Nemalionales.* (1) Plants filamentous, corticated, uni- or multiaxial, (2) Cells uninucleate, chromatophores axial or lateral, (3) Cystocarps superficial or deeply embedded in the thallus, (4) Life cycle without free-living tetrasporophyte.

*Family—Batrachospermaceae.* (1) Inhabit freshwater, (2) Thallus uniaxial, (3) Life cycle haplobiontic.

*Genus—Batrachospermum.* (1) Main axis and branches free from one another, (2) Branching appears beaded, (3) Threads embedded in large amount of mucilage.

### Hints for Collection

It is commonly found in freshwaters. The filaments are attached to stones in slow moving waters of rivers and streams or on the margins of the lakes.

## Ceramium

### Classification

<i>Sub-division</i>	—	<b>Algae</b>
<i>Class</i>	—	<b>Rhodophyceae</b>
<i>Sub-class</i>	—	<b>Florideae</b>
<i>Order</i>	—	<b>Ceramiales</b>
<i>Family</i>	—	<b>Ceramiales</b>
<i>Genus</i>	—	<b>Ceramium</b>

### Exercise 1

**Object : Study the external features of thallus.**

### Work procedure

To study the external features of the thallus, stain a few filaments in safranin, mount in glycerine and observe the characters.

### Comments

1. The thallus is red or yellowish-green or red-brown in colour.

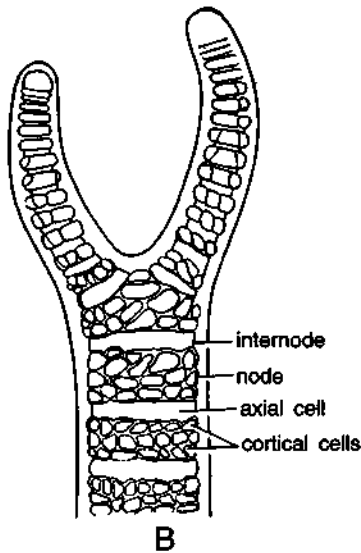
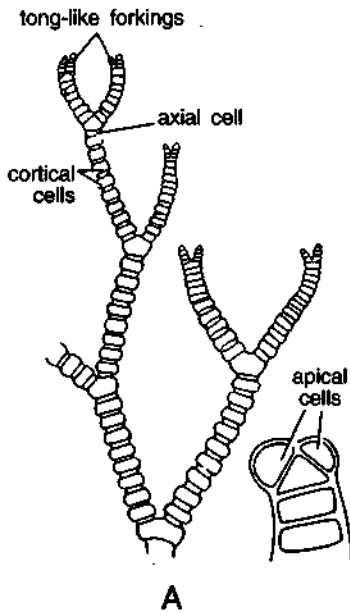


Fig. 1. *Ceramium*. Thallus. A. A part of thallus to show habit, B. Axis with nodes and internodes.

2. The plant body is filamentous, uniaxial, multicellular and richly branched showing distinct bands.
3. The filament is typically dichotomously branched. The apical region of the branches shows characteristic tong-like forkings.

4. The thallus is attached to the substratum by a cushion-like structure that produces rhizoids.
5. The plant body is made of a single row of large cylindrical or barrel shaped cells (the axial cells) arranged one over the other.
6. The banded appearance of the main filament is due to envelope of cortical cells (cortical filaments or branches) cut off by the axial cells.
7. The cortical cells or branches are produced discontinuously. This results in alternation of corticated and non-corticated regions.

### Exercise 2

**Object :** Study of spermatangia - the male sex organs.

### Work procedure

Stain a few filaments in safranin, mount in glycerine and study the structure.

### Comments

1. The spermatangia or antheridia are present in dense sori or clusters on the upper side of the lateral branchlets.
2. Each antheridium is oblong to spherical in shape.
3. It is unicellular and uninucleate structure.
4. The entire contents develop into a single, uninucleate and non-motile spermatium or male cell.

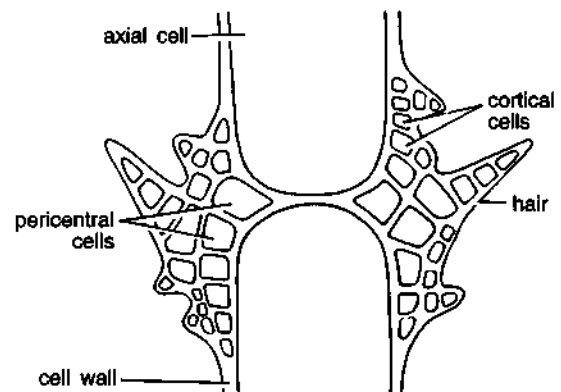


Fig. 2. *Ceramium*. L.s. through node.



### Exercise 3

**Object :** Study of carpogonium - the female sex organ.

#### Work procedure

Select a few filaments with carpogonia. Stain in safranin, mount in glycerine and study.

#### Comments

1. Carpogonium is borne terminally on short lateral branch called carpogonial branch.
2. The branch consists of a few cells, at the apex of which is a carpogonium. It is made of a basal swollen portion—the carpogonium, in which lies the female nucleus and the terminal long drawn out receptive organ called the trichogyne.
3. Post fertilization developments result in the formation of phase called carposporophyte. It is dependent on the female gametophytic plant.
4. The carposporophyte consists of gonimoblast filaments bearing carposporangia at the tips. Each carpogonium has only one carpospore.

### Exercise 4

**Object :** Study of tetrasporophyte, tetrasporangium and tetraspores.

#### Work procedure

Stain in safranin a few filaments of tetrasporophytic plant which can be identified by the presence of typical tetraspores seen on the surface. Mount in glycerine and study.

#### Comments

1. The external features of tetrasporophytic plant resemble that of the gametophyte. It is multicellular, uniaxial, filamentous, corticated and the apical portions are tong-forked.
2. The plant shows the presence of tetrasporangia.
3. Tetrasporangia are developed in clusters from the enlarged cells of the cortical bands.

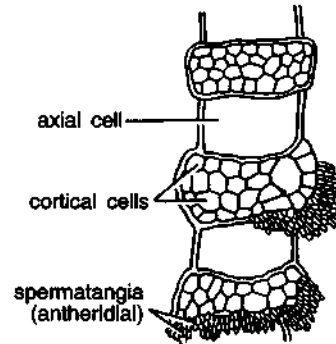


Fig. 3. *Ceramium*. An axis with antheridia or spermatangia.

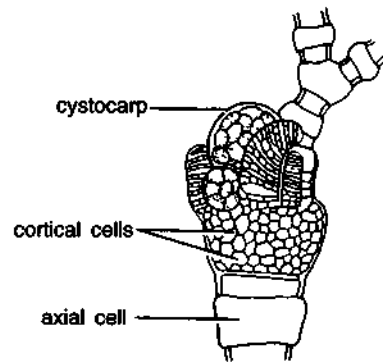


Fig. 4. *Ceramium*. An axis with carposporophyte showing cystocarp.

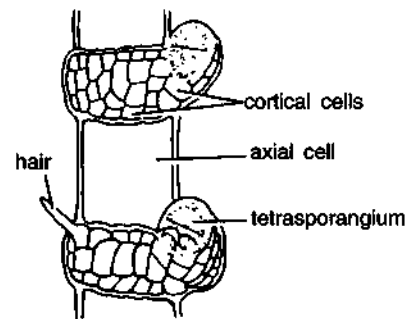


Fig. 5. *Ceramium*. An axis with tetrasporangia.

4. Tetrasporangia remain partly embedded in the cortical bands.
5. Each tetrasporangium has four tetraspores. Each tetraspore is uninucleate and haploid. It germinates to produce a new gametophytic generation.

## Identification

**Sub-division—Algae.** (1) Thallus simple, (2) Chlorophyll present, (3) Cell wall cellulosic.

**Class—Rhodophyceae.** (1) Chromatophores pure red to dark purple, (2) Photosynthetic reserve in floridoside, (3) Male gametes are non-motile, (4) Female reproductive organ with trichogyne, (5) Post fertilization product is cystocarp.

**Sub-class—Florideae.** (1) Thallus basically filamentous, (2) Pit connections between sister cells, (3) Cells with more than one chromatophore, (4) Carpogonium highly specialised.

**Order—Ceramiales.** (1) Thalli uniaxial or multiaxial, (2) Filaments corticated, polysiphonous, (3) Spermatangia in clusters, (4) Presence of trichoblasts.

**Family—Ceramiceae.** (1) Axes corticated, (2) Spermatangia developed on special determinate branchlets, (3) Cystocarps naked.

**Genus—Ceramium.** (1) Thallus is richly branched, banded due to discontinuous cortication, (2) Apical regions of the branches tong-forked, (3) Repeated dichotomous branching.

## Hints for Collection

*Ceramium* occurs very commonly between tide levels and also in deeper waters. The species of the genus are especially abundant in Mediterranean coast. The commonest Indian species include *Ceramium cruciatum*, *C. elegans*, *C. strictum*, *C. subdichotomum*, etc.

## Polysiphonia

### Classification

Sub-division	—	Algae
Class	—	Rhodophyceae
Sub-class	—	Florideae
Order	—	Ceramiales
Family	—	Rhodomelaceae
Genus	—	Polysiphonia

### Exercise 1

**Object :** The study of external features of thallus and a cell.

### Work procedure

Stain a few filaments in safranin, mount in glycerine and study the external features of thallus and a single cell.

## Comments

1. Plant body is filamentous. Filaments are multicellular, branched and polysiphonous.
2. Branching is dichotomous. Each branch terminates into a single celled apex, followed by a number of flat cells.
3. Thallus is polysiphonous i.e. made of series of parallel filaments.
4. Centre is occupied by a large barrel shaped cell (axial cell or central siphon). It is surrounded by 4-24 peripheral cells (pericentral siphons).
5. In the apical region, two or three cells below the apical cell, uniseriate, dichotomously divided, gradually tapering and multicellular filament is produced. It is known as trichoblast.
6. Many species remain attached to the substratum by thick walled, richly lobed and unicellular rhizoids (attaching organs) which arise from the peripheral cells of the creeping system.

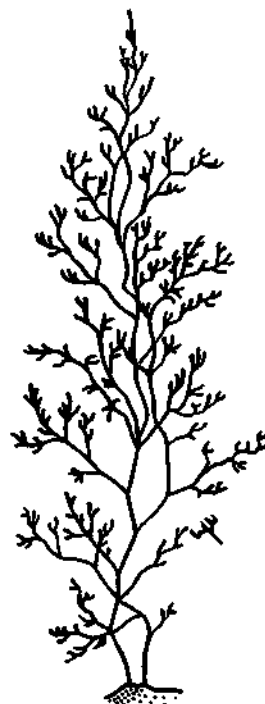


Fig. 1. *Polysiphonia*. Thallus to show habit.

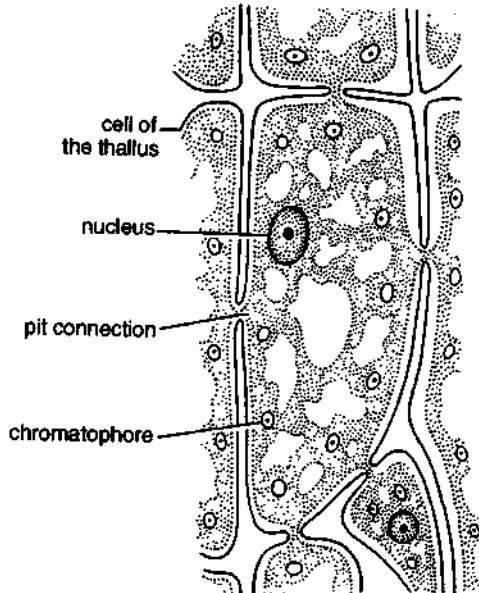


Fig. 2. *Polysiphonia*. A single cell.

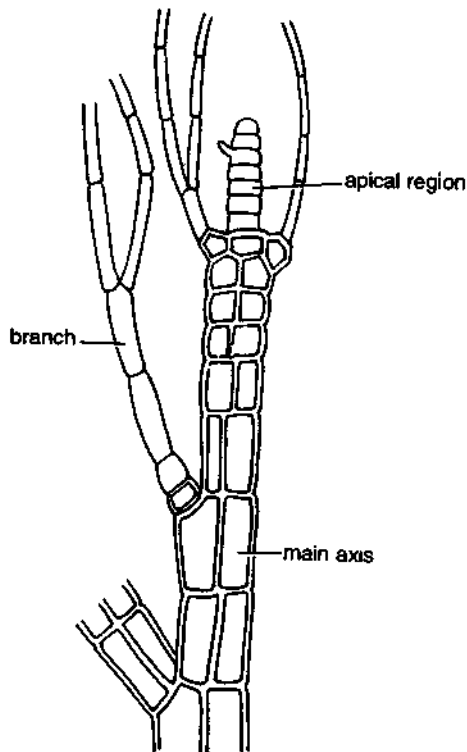


Fig. 3. *Polysiphonia*. A part of thallus with branches.

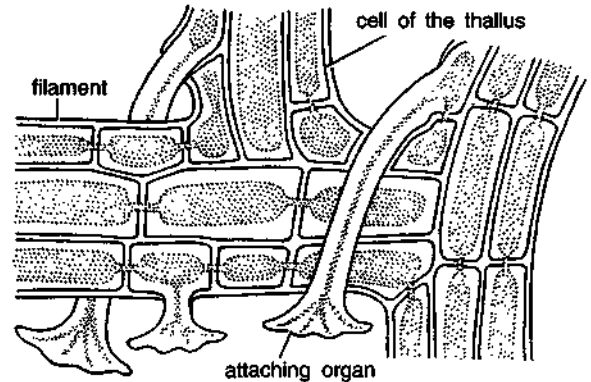


Fig. 4. *Polysiphonia*. A part of thallus showing rhizoids.

- The cell wall is thick. Cell is uninucleate. It has a large central vacuole. Chromatophores are small, discoid and many times without pyrenoids. Reserve food is in the form of starch grains—floridoside.
- The neighbouring cells are connected with one another by cytoplasmic lining known as pit connections.

### Exercise 2

**Object : Study of spermatangia.**

#### Work procedure

Select a portion of male plant bearing spermatangia, stain in safranin, mount in glycerine and study.

#### Comments

- The genus shows male plants bearing antheridia.
- Antheridia are produced in clusters by fertile trichoblasts situated near the apex.
- Antheridium is known as spermatangium. It is oval in shape, naked (without outer membrane) and contains many non-motile spermatia.
- Each spermatium is small, oval to spherical, uninucleate and non-motile.

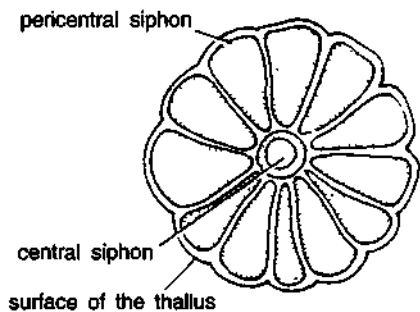


Fig. 5. *Polysiphonia*. T.s. thallus.

### Exercise 3

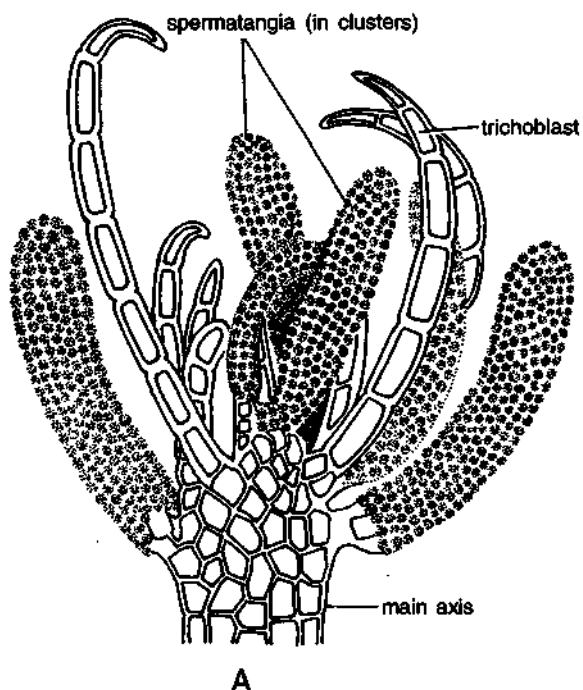
**Object :** Study the structure of procarp.

#### Work procedure

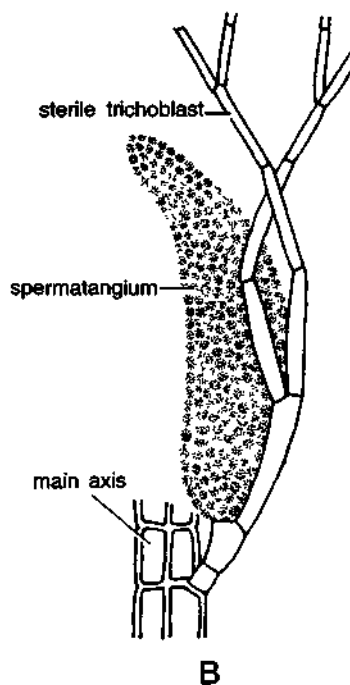
Select a part of female plant bearing procarps, stain in safranin, mount in glycerine and study.

#### Comments

1. The carpogonia are present on the female plants inside the procarp.



A



B

Fig. 6. *Polysiphonia*. A-B A. Cluster of spermatangia, B. A spermatangium.

2. Procarp is urn-shaped body. The wall is called pericarp that has an opening known as ostiole.
3. A long, tubular, receptive organ called trichogyne protrudes out of the ostiole.
4. At the base of trichogyne lies a swollen part, called carpogonium with a single female nucleus.

### Exercise 4

**Object :** Study of cystocarp.

#### Work procedure

Mount a few filaments with cystocarp, stain in safranin, mount in glycerine and study.

#### Comments

1. Cystocarp is a post-fertilization product. The thallus bearing this structure forms a phase called carposporophyte.
2. This oval or urn-shaped structure is attached to a lateral branch.
3. Cystocarp opens to the exterior by an opening called ostiole.
4. Wall of the cystocarp is called pericarp and is composed of a single layer of cells.

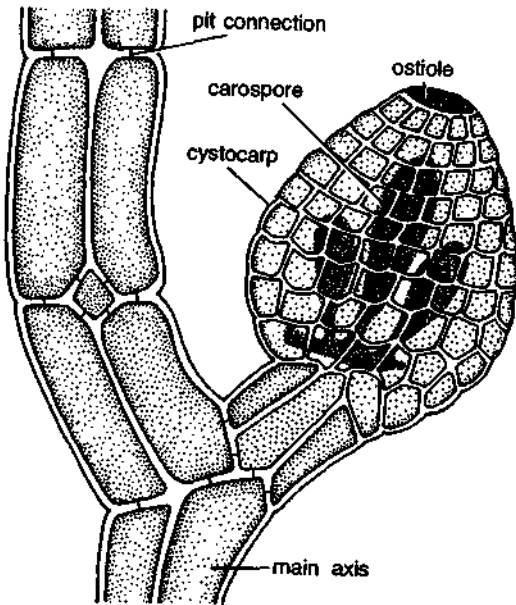


Fig. 7. *Polysiphonia*. A part of thallus with cystocarp.

5. Carospores are produced from the base of the cystocarp. These are arranged in single spherical layer.
6. Each carospore is oval, uninucleate and diploid.

#### Exercise 4

**Object :** Study the structure of tetrasporophyte and tetrasporangium.

#### Work procedure

Select a tetrasporophytic filament. It shows tetrahedrally arranged tetraspores in a tetrasporangium and can be easily detected. Stain in safranin, mount in glycerine and study.

#### Comments

1. Tetrasporophytes are morphologically similar to male and the female gametophytes.
2. The thallus is polysiphonous being made of a central siphon surrounded by pericentral siphons.
3. A cell shows a nucleus, discoid chromatophores and pit connections.
4. The plant is diploid and bears tetrasporangia in longitudinal series, produced mostly by pericentral cells.
5. Tetrasporangia are small and spherical bodies borne on short one-celled stalk.

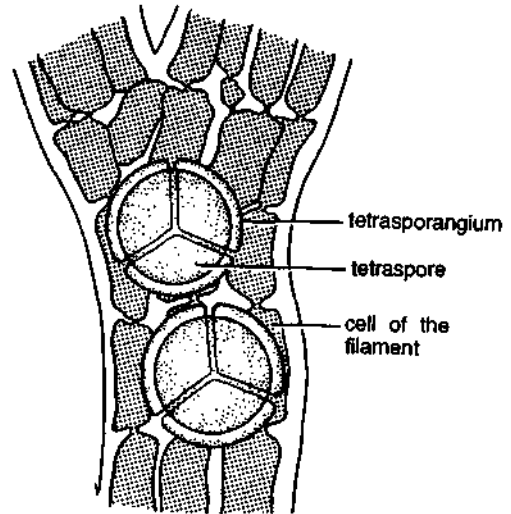


Fig. 8. *Polysiphonia*. A part of thallus with tetrasporangia.

6. Each tetrasporangium possesses four tetrahedrally arranged uninucleate and haploid tetraspores.

### Identification

**Sub-division—Algae.** (1) Thallus simple, (2) Chlorophyll present, (3) Cell walls of cellulose.

**Class—Rhodophyceae.** (1) Chromatophores pure red to dark-purple, (2) Photosynthetic reserve floridoside, (3) Male gametes non-motile, (4) Female reproductive organ with a receptive structure—trichogyne, (5) Post-fertilization product—cystocarp.

**Sub-class—Florideae.** (1) Thallus basically filamentous, (2) Pit connections between sister cells, (3) Cells with more than one chromatophore, (4) Carpogonium highly specialized.

**Order—Ceramiales.** (1) Thalli uni-multiaxial or filamentous, (2) Filaments corticated, polysiphonous, (3) Spermatangia in clusters, (4) Presence of trichoblasts.

**Family—Rhodomelaceae.** (1) Axes polysiphonous, (2) Axes naked, corticated or covered with branches, (3) Main axis surrounded with pericentrals, (4) Plants bushy, sparingly branched, branches delicate.

**Genus—Polysiphonia.** (1) Ultimate branches uncorticated, (2) Tetrasporangia borne singly.

### Hints for Collection

Species of *Polysiphonia* are exclusively marine. These are most commonly found along the Atlantic and Pacific coasts, in littoral and sub-littoral regions. A few species occur as epiphytes on mangroves or brown seaweeds. A small number of species are also found along the Indian coasts.

## VI. MYXOPHYCEAE

Popularly known as cyanophyceae, cyanobacteria or blue-green algae, the group is said to be highly successful since they inhabit almost every habitat. Majority of species are freshwater. The members also occur in marine as well as terrestrial habitats. Group, as a whole, forms pioneer communities of ecological interests. It also offers an example of symbiotic relations, forming lichens. Hot springs and snow are not left unoccupied. The ability to live in such diverse habitats points towards its great degree of adaptability and perhaps its occurrence since remote past.

Predominance of pigments, *c*-phycocyanin (a blue pigment) and *c*-phycoerythrin (a red pigment) imparts characteristic blue-green colour. The group is marked from all others, in its absence of definite nucleus and instead DNA strands are present (prokaryotic character) in the centropasm. Pigments are found diffused in the form of lamellae in the peripheral part of the protoplasm, called chromatoplasm. The plastids and pyrenoids are absent. The reserve food product is cyanophycean starch and glycogen.

Thallus is poorly developed. Plants are either unicellular, colonial or filamentous. Thalli of most of the members are surrounded by gelatinous envelope. Branching is either absent, or if present is false except a few which show true branching. Many members possess a unique structure—'Botanical engima'—heterocyst perhaps an active site of nitrogen fixation.

Sexual reproduction being absent, only methods of reproduction are by asexual means. Flagellated reproductive bodies are entirely lacking, and there is a total absence of gametic union. Cell division, endospores, hormogones, akinetes, etc. are a few of the methods by which reproduction takes place. A parasexual method of reproduction is now known.

A few members have been shown to possess the ability to fix the atmospheric nitrogen enriching the habitat in which they live. It is said that this character can be used to solve manuring problems of the rice fields. Other uses include reclamation of usar soils, in fisheries, etc.

### *Oscillatoria*

#### Classification

Sub-division	—	Algae
Class	—	Myxophyceae
Order	—	Nostocales
Family	—	Oscillatoriaceae
Genus	—	<i>Oscillatoria</i>

#### Exercise 1

**Object :** To study the structure of a filament and single cell.

#### Work procedure

Take a few filaments, stain in safranin, mount in glycerine and study the structure.

#### Comments

1. Filaments occur either singly or interwoven to form a flat or spongy, free swimming mat.
2. Filament consists of an inconspicuous and barely recognizable sheath enclosing unbranched trichome.
3. Trichome consists of a single row of cells.
4. The apical cell of the trichome may have calyptra—thick wall on its outer free face.
5. The cells show typical myxophycean cell structure. It has no definite nucleus, no chloroplasts or no membrane bound organelles (prokaryotic cell).
6. The cell shows many shining cyanophycean granules.
7. Floating species show numerous gas vacuoles.

#### Exercise 1

**Object :** Study of reproductive structures.

#### Work procedure

Place a few filaments in safranin, wash in water, mount in glycerine and study the reproductive structures—hormogones.

#### Comments

1. The only method of reproduction is hormone formation.
2. The hormogones (small pieces of trichome with one to many uniform cells) are formed as a result of the death of intercalary cell or by the formation of special biconcave separation discs.

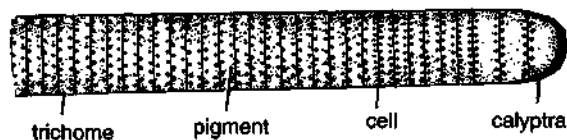


Fig. 1. *Oscillatoria*. Trichomes.

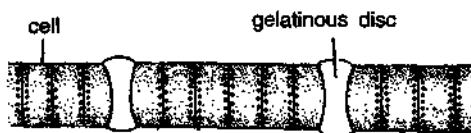


Fig. 2. *Oscillatoria*. A trichome showing separation disc.

(B-14)

- Hormones are liberated by the disintegration of discs.
- Each hormone develops into a new filament.

### Identification

**Sub-division—Algae.** (1) Thallus simple, (2) Presence of chlorophyll, (3) Cell wall of cellulose.

**Class—Myxophyceae.** (1) Chromatophore not organised, pigments diffused, blue-green, (2) Photosynthetic reserve cyanophycean starch, (3) True nucleus absent, (4) Sexual reproduction absent.

**Order—Nostocales.** (1) Thallus with trichomes, unbranched, or with false branching, (2) Hormones, heterocysts, exospores and endospores generally present.

**Family—Oscillatoriaceae.** (1) Trichomes uniseriate, sometimes tapering at the ends, (2) Heterocysts and spores absent, (3) Sheath absent or diffluent.

**Genus—Oscillatoria.** (1) Trichomes not in bundles, (2) Trichomes without a sheath, (3) Trichomes straight and cylindrical.

### Hints for Collection

One can hardly miss *Oscillatoria* when on a collection trip. It is almost cosmopolitan and occurs abundantly on moist rocks, cliffs, damp soil, muddy banks of streams, ponds, surface of the water reservoirs, sewage ponds, etc. A few species are marine and float on the surface.

## Nostoc

### Classification

Sub-division	—	Algae
Class	—	Myxophyceae
Order	—	Nostocales
Family	—	Nostocaceae
Genus	—	<i>Nostoc</i>

### Exercise 1

**Object :** Study of external features of thallus.

### Work procedure

Place a part of colony on the slide, press it little by another slide so that it spreads flat, stain in safranin and mount in glycerine.

### Comments

- Thallus is colonial. Young colonies are microscopic, spherical and solid.
- Mature colonies become irregular and hollow.
- Colonial envelope encloses many filaments. These are much twisted, curved and entangled with each other.
- A filament has diffluent gelatinous sheath.
- The trichomes are unbranched. Each trichome is made of cells of uniform size and shape except those called heterocyst which occur throughout.
- Structure of a cell is typically cyanophycean. It has a centrally located centropiasm, nucleus being altogether absent. Peripheral cytoplasm shows diffused pigments. A few shining cyanophycean granules are also present in this region.
- Heterocysts are intercalary. These are double walled, pale yellow coloured with two shining polar granules, one each near the neighbouring

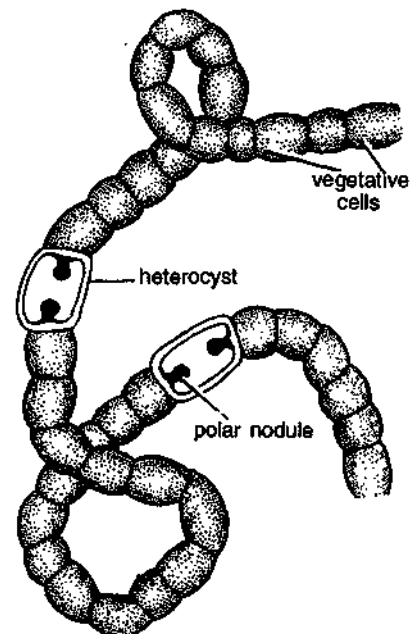


Fig. 1. *Nostoc*. A vegetative filament.

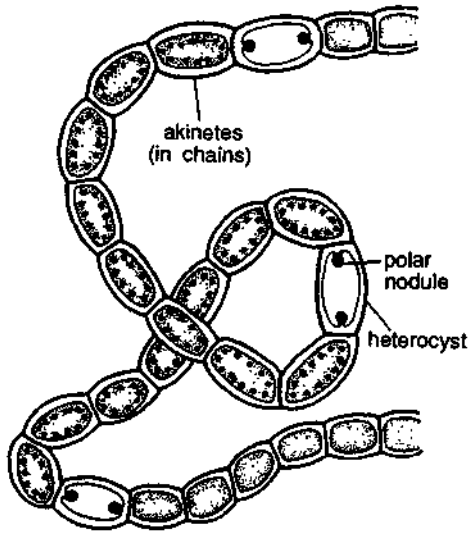


Fig. 2. *Nostoc*. Heterocyst with akinetes in chains.

cell on either side. Heterocysts are much of the same size of slightly bigger than the vegetative cells.

### Exercise 2

**Object : Study the akinetes.**

#### Work procedure

Press a piece of mature colony to spread the filaments. Stain in safranin, mount in glycerine and study.

#### Comments

1. Akinetes are developed only in a mature colony. These occur in large number, in series between two heterocysts. Usually all vegetative cells between two successive heterocysts develop into akinetes.
2. Akinetes are thick walled, sometimes ornamented, rich in food reserves and cyanophycean granules.
3. Akinetes are liberated due to decay of colonial sheath. These germinate to form a new thallus.

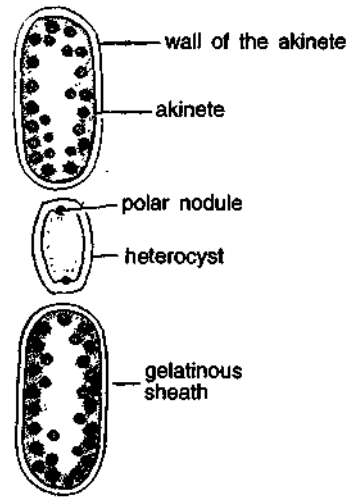


Fig. 3. *Nostoc*. Heterocyst with akinetes on both of its sides.

### Identification

*Sub-division—Algae.* (1) Thallus simple, (2) Presence of chlorophyll, (3) Cell wall of cellulose.

*Class—Myxophyceae.* (1) Chromatophore not organised, pigments diffused, blue-green, (2) Photosynthetic reserve of cyanophycean starch, (3) True nucleus absent.

*Order—Nostocales.* (1) Thallus with trichomes, unbranched or branching false, (2) Hormogones, heterocysts, exospores and endospores generally present.

*Family—Nostocaceae.* (1) Trichomes simple, unbranched, uniseriate and approximately of the same diameter throughout, (2) Heterocysts and akinetes present, (3) Trichomes not differentiated.

*Genus—Nostoc.* (1) Trichomes much twisted into a mass of definite form with a firm colonial envelope, (2) Heterocysts intercalary and single.

### Hints for Collection

It commonly occurs in abundance after the first few rains. It is terrestrial as well as aquatic. It is collected from water pools, paddy fields, waterlogged soil, moist rocks, stagnant water, etc. It is known to occur as epiphyte on aquatic weeds and endophytically inside *Cycas* coralloid roots, *Azolla*—a fern, *Blasia* and *Anthoceros*—bryophytes and form lichens in association with fungal members.



## Scytonema

### Classification

Sub-division	—	Algae
Class	—	Myxophyceae
Order	—	Nostocales
Family	—	Scytonemataceae
Genus	—	Scytonema

#### Exercise 1

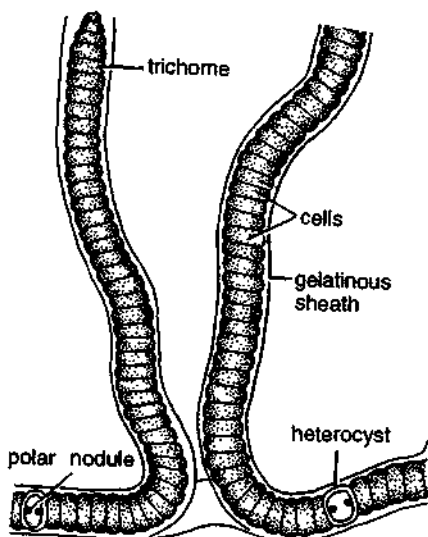
**Object :** Study the external features of thallus.

#### Work procedure

Take a few filaments, tease them so that these get separated from one another. Stain in safranin, mount in glycerine and study.

#### Comments

1. Filaments occur singly. The trichomes are enveloped in a sheath.
2. Filaments show false branching, branches being single or geminate (in pairs).
3. The trichomes are of the same diameter throughout its length.
4. The trichome is covered by an individual sheath which is firm and either hyaline or coloured. It may be homogeneous or lamellated.



**Fig. 1.** *Scytonema*. A part of thallus to show false branching. (B-14)

5. Heterocysts occupy intercalary position. It may be either single, in twos or threes. These are approximately of the same size as those of vegetative cells. Lateral branches are generally produced in between heterocysts.
6. Heterocysts show two shining granules, one each in contact with the cells on either side. It is pale yellow in colour and contents are homogenous.
7. Cell structure is typically cyanophycean. Central part known as centropiasm is enclosed by peripheral pigmented chromatoplasm. The centropiasm has genetic material in the form of DNA. The chromatoplasm has scattered pigments and cyanophycean granules.

#### Exercise 2

**Object :** To study hormogonia.

#### Work procedure

Examine part of the thallus, stained in safranin for terminally placed hormogonia.

#### Comments

1. Sexual reproduction is absent.
2. Akinetes are asexual reproductive bodies. These are of rare occurrence.
3. Hormogones are found very commonly. They are solitary and terminal.
4. Hormogone is a spherical structure that is covered by a thick mucilaginous envelope.

#### Identification

- Sub-division—Algae.** (1) Simple filamentous thallus, (2) Chlorophyll present, (3) Cell walls of cellulose.
- Class—Myxophyceae.** (1) Chromatophore not organised, (2) Pigments diffused, blue-green, (3) Photosynthetic reserve cyanophycean starch, (4) True nucleus absent, (5) Sexual reproduction absent.
- Order—Nostocales.** (1) Thallus with trichomes, (2) Trichomes unbranched or with false branching, (3) Hormogones, heterocysts, exospores or endospores generally present.
- Family—Scytonemataceae.** (1) Trichomes uniseriate, without marked attenuation, (2) Filaments always free, with false branching, (3) Sheath firm, enclosing one or more trichomes.
- Genus—Scytonema.** (1) Filaments with heterocysts, (2) One trichome within a sheath, (3) False branching present, (4) Branches arising in pairs.

## Hints for Collection

This genus is usually found in sub-aerial habitats such as damp walls, brick-work, bark of the trees, etc. Filaments are interwoven to form a felt-like mass of considerable extent. Few species grow best on damp soil and others on the dripping surfaces of rocky cliffs.

### *Rivularia*

#### Classification

Sub-division	—	Algae
Class	—	Myxophyceae
Order	—	Nostocales
Family	—	Rivulariaceae
Genus	—	<i>Rivularia</i>

#### Exercise 1

**Object :** To study the external structure of thallus.

#### Work procedure

Take a colony, spread it by pressing with another slide, allow it to dry a little, stain in safranin, mount in glycerine and study.

#### Comments

1. Thallus is colonial. Colony is enveloped by a mucilaginous sheath. The shape of the colony is spherical, hemispherical or irregular which attains macroscopic size when mature.
2. Trichomes are radially arranged in a colony. Basal part lies toward the centre, while tapering terminal end of the filament is nearer the periphery.
3. Trichome is partially or wholly surrounded by a confluent sheath.
4. Trichomes are unbranched, sometimes more or less irregularly false branched. These are broad at the base and gradually taper into a hair toward the apex.
5. The base has a single heterocyst with a single polar nodule (basal heterocyst).
6. Sheath is distinct in the basal part. It may be homogeneous or lamellated. Sheaths are more or less confluent at the distal ends.

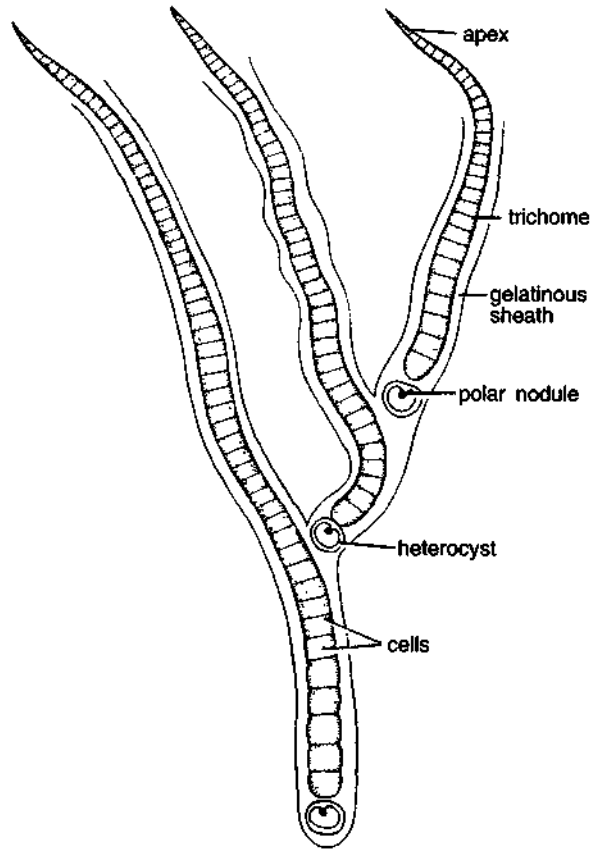


Fig. 1. *Rivularia*. A part of thallus to show false branching.

7. Radial arrangement of the trichomes in a colony is due to the repeated false branching in the basal portion of trichomes.
8. The cell shows typical cyanophyceean structure. There is a centrally located centropiasm or 'incipient nucleus'. Genetic material in the form of DNA is located in this region. Peripheral chromatoplasm shows dispersed pigments with shining cyanophyceean granules.
9. Reproduction occurs by homogonia produced either singly or in series.
10. Homogonia are special pieces of trichomes which on germination give rise to new thallus.

#### Identification

**Sub-division—Algae.** (1) Thallus simple, (2) Presence of chlorophyll, (3) Cell walls of cellulose.

**Class—Myxophyceae.** (1) Chromatophores not organised, pigments blue-green, (2) Photosynthetic reserve—cyanophyceean starch, (3) True nucleus absent, (4) Sexual reproduction absent.

**Order—Nostocales.** (1) Thallus with trichomes, unbranched or with false branching, (2) Hormogones, heterocysts, exospores and endospores generally present.

**Family—Rivulariaceae.** (1) Trichomes markedly attenuated from base to apex, or from middle to both extremities, (2) Growth trichothallic, terminal hair, (3) Trichomes commonly false branched.

**Genus—Rivularia.** (1) Trichomes without akinetes, (2) Each trichome singly enclosed by a sheath, (3) Heterocyst basal, (4) Filaments united into spherical or hemispherical thalli.

### Hints for Collection

It commonly occurs in aquatic habitats, as also on damp soil near river bed, between mosses on earth, moist rocks and in flowing waters. It is epiphytic, occurring on *Nitella* and other submerged stems and leaves of water plants.

## Gloeotrichia

### Classification

Sub-division	—	Algae
Class	—	Myxophyceae
Order	—	Nostocales
Family	—	Rivulariaceae
Genus	—	<i>Gloeotrichia</i>

### Exercise 1

**Object :** Study the external features of thallus and heterocyst.

### Work procedure

Take a colony, press it flat on the slide, stain in safranin and mount in glycerine.

### Comments

1. The thallus is colonial. It is surrounded by a large mucilage. The shape appears to be spherical or hemispherical. The old colony becomes inflated and hollow.
2. The colour of the colony ranges from dull brown to blackish green and is seldom blue green.

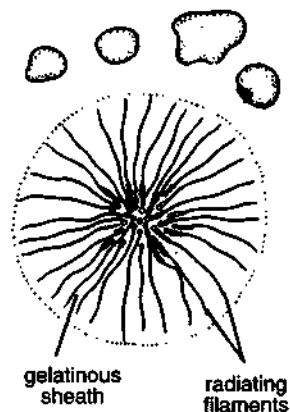


Fig. 1. *Gloeotrichia*. A few colonies.

3. The filaments in a colony are loosely arranged in radial fashion. These are more or less parallel with false branches.
4. Colourless sheath is present at the base where it is firm and gradually gelatinizes outwards.
5. Trichomes may be unbranched or more or less false branched. These are broad at the base and gradually taper into a hair toward the apex.
6. The trichomes show a typical and distinct trichothallic growth.
7. A single spherical heterocyst with a single polar granule is present at the base of the trichome.
8. Each cell shows a typical cyanophycean structure. The central region does not have a nucleus, the genetic material in the form of DNA being dispersed. The pigments occur scattered in the peripheral region. A few cyanophycean granules—the major reserve food material are also present in this region.

### Exercise 2

**Object :** To study the hormogonia and the spores.

#### Work procedure

Stain a colony with safranin, mount in glycerine and search for hormogonia and spores.

#### Comments

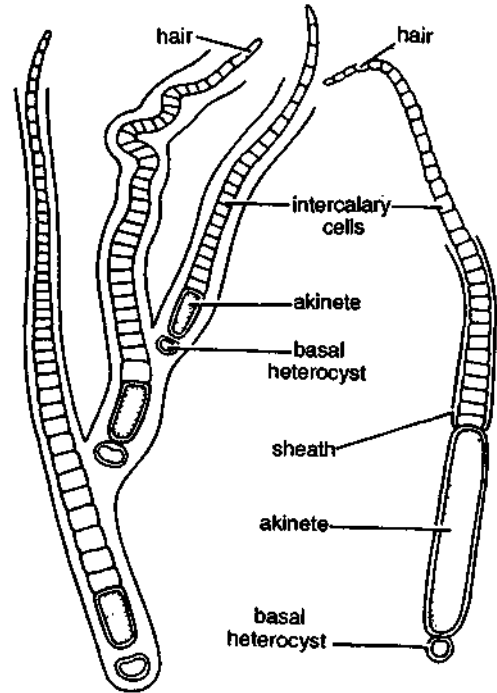
1. Reproduction takes place by hormogones and spores or akinetes.
2. Sexual reproduction is not known.
3. Hormogone formation is common. These occur either singly or in series.
4. Spores are also of common occurrence. These occur at the base of the trichome, generally singly, sometimes more.
5. Each spore is long, cylindrical, smooth or ornamented and thick walled.
6. It is rich in reserve food material in the form of cyanophycean granules.
7. Akinetes are the major organs of perennation.

### Identification

**Sub-division—Algae.** (1) Thallus simple, (2) Presence of chlorophyll, (3) Cellulose cell wall.

**Class—Myxophyceae.** (1) Chromatophores blue-green, (2) Cyanophycean starch as reserve food, (3) True nucleus absent.

**Order—Nostocales.** (1) Thallus with unbranched trichome or trichome with false branching, (2) Hormogones, heterocysts, etc. generally present.



**Fig. 2. *Gloeotrichia*.** A filament to show heterocyst and akinete.

**Family—Rivulariaceae.** (1) Trichomes gradually attenuated from base to apex, (2) Growth trichothallic, hair terminal, (3) Trichomes commonly false branched.

**Genus—*Gloeotrichia*.** (1) Spores large, formed singly, (2) Filaments in spherical thallus.

### Hints for Collection

*Gloeotrichia* occurs as mucilaginous balls or mass floating on the surface of the stagnant waters. Colonies are also found as epiphytes on aquatic weeds growing near the margins of ponds and pools.

# 3

## Chapter

# Fungi

### Preamble

The term 'Fungi' is used for those plants which lack chlorophyll and are, therefore, heterotrophic. Being the members of Thallophyta, their plant body is simple and is not differentiated into root, stem and leaves.

The branch that deals with fungi is known as 'Mycology'. It included the study of mushrooms only to begin with. The credit for laying the foundation of present day mycology goes to Italian Botanist Pier' Antonio Micheli who included his researches on fungi in a book—*Nova Plantarum Genera* published in 1729.

Fungi include 100,000 species, of which 40,000 are known to be valid species leaving 60,000 to be investigated and described in future. The earlier fungal taxonomists included bacteria also in this group, the practice which is now completely discarded. The members of this heterotrophic group exist either as parasites or saprophytes. Special absorbing organs called haustoria are developed for this purpose. Thallus is either coenocytic (aseptate) or septate forming compact structures like mushrooms, morels, etc. Hyphal system (filamentous) forms a thallus and is called mycelium. The cell wall is typically made of chitin while the reserve food occurs mainly in the form of glycogen.

Both asexual and sexual methods of reproduction are known in fungi. Asexual reproduction is through different types of spores like zoospores, conidiospores, basidiospores, chlamydospores, etc. Sexual reproduction in fungi is extremely reduced. In lower members distinct sexual reproduction is present but is gradually reduced in higher members and finally it is found to be absent in Fungi Imperfecti.

The diseases caused by this group have rendered it sufficient economic importance. The members of this group attack plants as well as animals including the human beings. Since the fungi are principal agents of decay, so through the decomposition of organic matter, they play an essential role in the nutrition of the green plants. Contrary to these harmful activities, fungi serve as food, used in preparation of medicines and antibiotics, and are employed in many industrial processes.

Fungi have been variously classified by numerous mycologists, time and again. Basically the group was divided into five classes—Myxomycetes, Phycomycetes, Ascomycetes, Basidiomycetes and Deuteromycetes. This classification was the most convenient. However, the trend now is to treat fungi as a kingdom rather than as a sub-group of Thallophyta equal in rank to algae. One of such classifications was proposed by G.C. Ainsworth (1973). It is being used in this chapter with certain modifications. Only those taxa have been described which are included in the subsequent text.

### Distinguishing Characters of Taxa

#### KINGDOM MYCOTA (FUNGI)

- (1) Chlorophyll absent
- (2) Reserve food glycogen
- (3) Cell wall of fungal cellulose

#### DIVISION I. MYXOMYCOTA

- (1) Thallus without cell walls
- (2) Thallus a naked mass of protoplasm

#### CLASS PLASMIDIOPHOROMYCETES

Plasmodium parasitic within the cells of host plants

#### Order **Plasmodiophorales**

Swarm cells anteriorly biflagellate

#### Family **Plasmodiophoraceae**

Vegetative development within the host cells

Example. *Plasmodiophora*

#### DIVISION II EUMYCOTA

Presence of definite cell wall throughout the vegetative phase

#### SUB-DIVISION I. MASTIGOMYCOTINA

- (1) Presence of motile spores or zoospores

## Classification of Fungi

### Kingdom MYCOTA

Division	Sub-division	Class	Order	Family	Example
I. MYXOMYCOTA		Plasmodiophoromycetes	Plasmodiophorales	Plasmodiophoraceae	<i>Plasmodiophora</i>
II. EUMYCOTA	1. Mastigomycotina	1. Chytridiomycetes 2. Oomycetes	Chytridiales 1. Saprolegniales 2. Peronosporales	Synchytriaceae Saprolegniaceae 1. Pythiaceae 2. Peronosporaceae	<i>Synchytrium</i> <i>Saprolegnia</i> <i>Achlya</i> <i>Pythium</i> <i>Phytophthora</i> <i>Sclerospora</i> <i>Peronospora</i>
	2. Zygomycotina	Zygomycetes	Mucorales	3. Albuginaceae 1. Mucoraceae 2. Pilobolaceae	<i>Albugo</i> <i>Rhizopus Mucor</i> <i>Pilobolus</i>
	3. Ascomycotina	1. Hemiascomycetes 2. Plectomycetes	Endomycetales 1. Erysiphales 2. Eurotiales	Saccharomycetaceae Erysiphaceae Eurotiaceae	<i>Saccharomyces</i> <i>Erysiphe</i> <i>Sphaerotheca</i> <i>Phyllactinia</i> <i>Aspergillus</i> <i>Penicilium</i>
		3. Pyrenomycetes	Sphaeriales	1. Sordariaceae 2. Xylariaceae 3. Clavicipitaceae	<i>Neurospora</i> <i>Xylaria</i> <i>Claviceps</i>
		4. Discomycetes	Pezizales	1. Pezizaceae 2. Helvellaceae	<i>Peziza</i> <i>Ascobolus</i> <i>Morchella</i>
	4. Basidiomycotina	1. Teliomycetes	1. Ustilaginales 2. Uredinales	Ustilaginaceae Pucciniaceae	<i>Ustilago</i> <i>Sphacelotheca</i> <i>Puccinia</i> <i>Uromyces</i>
		2. Hymenomycetes Sub-class : Holobasidiomycetidae	1. Exobasidiales 2. Agaricales 3. Aphyllophorales	Exobasidiaceae Agaricaceae Polyporaceae	<i>Exobasidium</i> <i>Agaricus</i> <i>Polyporus</i>
		3. Gasteromycetes	Lycoperdales	Lycoperdaceae	<i>Lycoperdon</i>
	5. Deuteromycotina	1. Hyphomycetes	Moniliales	1. Dematiaceae 2. Moniliaceae 3. Tuberculariaceae	<i>Alternaria</i> <i>Cercospora</i> <i>Helminthosporium</i> <i>Piricularia</i> <i>Fusarium</i>
		2. Coelomycetes	Melanconiales	Melanconiaceae	<i>Collectotrichum</i>

(2) Oospores produced as a result of sexual reproduction

### CLASS 1. CHYTRIDIOMYCETES

- (1) Thallus usually unicellular
- (2) Zoospores uniflagellate (flagella whiplash type)

### Order Chytridiales

Asexual reproduction by posteriorly uniflagellate zoospores

### Family Synchytriaceae

Thallus endobiotic, holocarpic, directly forms a sorus or prosoorus

Example. *Synchytrium*

**CLASS 2. OOMYCETES**

- (1) Thalli usually mycelial (mycelium aseptate)
- (2) Zoospores biflagellate (posterior flagellum whiplash type and anterior tinsel type)
- (3) Cell wall cellulose

**Order 1. Saprolegniales**

- (1) Mycelial thallus extensive and without a conspicuous hold fast
- (2) members aquatic, often called water molds
- (3) Sporangia cylindrical

**Family Saprolegniaceae**

Oogonium with many eggs and lacks periplasm

Examples. *Saprolegnia*, *Achlya*

**Order 2. Peronosporales**

- (1) Sexual reproduction aplanogametic and oogamous
- (2) Primarily terrestrial—in soil or parasitic on vascular plants
- (3) oogonium with a single egg surrounded by periplasm

**Family 1. Pythiaceae**

Sporangiophores similar to somatic hyphae or if different, indeterminate in growth

Examples. *Pythium*, *Phytophthora*

**Family 2. Peronosporaceae**

- (1) Sporangiohores dichotomously branched, determinate
- (2) Sporangia borne singly at the tips of branches

Examples. *Sclerospora*, *Peronospora*

**Family 3. Albuginaceae**

- (1) Mycelium intercellular and provided with knob-like haustoria
- (2) Conidia in basipetal chains on clavate, unbranched conidiophores

Example *Albugo*

**SUB-DIVISION 2. ZYGOMYCOTINA**

Asexual reproduction by non-motile spores, aplanospores, perfect stage spore—zygospore formed

**CLASS ZYGOMYCETES**

- (1) Gametangia morphologically similar
- (2) Sexual reproduction results in the formation of zygospores

**Order Mucorales**

- (1) Mostly saprophytic
- (2) Asexual reproduction by typical non-motile aplanospores

**Family 1. Mucoraceae**

- (1) Sporangia with many spores and well developed columella
- (2) Sporangial wall relatively thin and easily breakable or deliquescent
- (3) Suspensors rarely tong-like

Examples. *Rhizopus*, *Mucor*

**Family 2. Pilobolaceae**

- (1) Sporangia with many spores with moderate sized columella
- (2) Sporangium wall thickened above and not breaking up or deliquescent.
- (3) Sporangium violently discharged or passively discharged as a unit
- (4) Suspensor always tong-like

Example. *Pilobolus*

**SUB-DIVISION 3. ASCOMYCOTINA**

- (1) Mycelium septate, if not reproduces by budding
- (2) Ascospores borne endogenously in ascus
- (3) Ascospores in definite numbers, in multiples of two, usually eight.

**CLASS 1. HEMIASCOMYCETES**

- (1) Thallus yeast-like or unicellular
- (2) Asci naked and formed directly from the zygotes
- (3) Ascogenous hyphae and ascocarps not formed

**Order Endomycetales**

- (1) Mostly saprophytes
- (2) Zygote unicellular, forms ascus directly

**Family Saccharomycetaceae**

Cells multiply by budding

Example. *Saccharomyces*

**Class 2. Plectomycetes**

Ascocarp, a cleistothecium

**Order 1. Erysiphales**

Ectoparasites

**Family Erysiphaceae**

- (1) Aerial mycelium hyaline
- (2) Enormous production of conidia on host surface gives it a white powdery appearance

Examples. *Erysiphe*, *Sphaerotheca*, *Phyllactinia*

**Order 2. Eurotiales**

Ascocarp—a cleistothecium

**Family Eurotiaceae**

- (1) Asci scattered, 8-spored, thin walled, ascospores unicellular

- (2) Hymenium not formed
- (3) Peridium (outer wall of cleistothecium) of closely interwoven hyphae

Examples. *Aspergillus*, *Penicillium*

### Class 3. Pyrenomycetes

Ascocarp, a perithecium

#### Order Sphaeriales

Ascocarp borne singly or in stroma, dark, membranous or carbonous

##### Family 1. Sordariaceae

- (1) Perithecia dark coloured, not in stroma, free
- (2) Ostiole lined by periphyses

Example. *Neurospora*

##### Family 2. Xylariaceae

- (1) Perithecia embedded in stroma
- (2) Stroma consisting entirely of fungus tissue
- (3) Ascospores dark and inequilateral

Example. *Xylaria*

##### Family 3. Clavicipitaceae

- (1) Perithecia develop on fleshy stroma, ostiolate
- (2) Perforated cap at the apex of ascus
- (3) Ascospores long, narrow, often breaking up into short segments

Example. *Claviceps*

### Class 4. Discomycetes

Ascocarp, an apothecium

#### Order Pezizales

- (1) Apothecia fleshy or leathery
- (2) Apothecia usually not in stroma
- (3) Asci opening by a lid or operculum

##### Family 1. Pezizaceae

Apothecia not differentiated into stipe and pileus

Examples. *Pyronema*, *Peziza*, *Ascobolus*

##### Family 2. Helvellaceae

- (1) Apothecia not cup-shaped
- (2) Apothecia differentiated into stipe and pileus

Example. *Morchella*

### SUB-DIVISION 4. BASIDIOMYCOTINA

- (1) Mycelium septate
- (2) Characteristic reproductive body is basidium
- (3) Basidiospores usually four, produced exogenously

### CLASS 1. TELIOMYCETES

- (1) Basidiocarp lacking
- (2) Teliospores or chlamydo-spores in sori or scattered
- (3) parasitic on vascular plants

#### Order 1. Ustilaginales

- (1) Mycelial hyphae in the host inter—as well as intracellular
- (2) Telio- or teleutospores mostly intercalary, basidiospores not on sterigmata
- (3) Basidiospores indefinite in number

##### Family Ustilaginaceae

Chlamydo-spores formed in the host tissue from hyphal cells

Examples. *Ustilago*, *Sphacelotheca*

#### Order 2. Uredinales

- (1) Teleutospores formed terminally
- (2) Four basidiospores per basidium, Basidiospores on sterigmata
- (3) Infected plant rusty in colour

##### Family Pucciniaceae

- (1) Teleutospores stalked
- (2) Teleutospores free or united but never in the form of layer

Examples. *Puccinia*, *Uromyces*

### CLASS 2. HYMENOMYCETES

- (1) Basidiocarp usually well-developed
- (2) Mostly saprobic

#### Sub-class Holobasidiomycetidae

Basidia aseptate (holobasidia), club-shaped

#### Order 1. Exobasidiales

- (1) Basidiocarp present
- (2) Basidia covering the surface of host tissue

##### Family Exobasidiaceae

- (1) Basidiocarp septate prior to germination
- (2) Basidia club-shaped and formed externally

Example. *Exobasidium*

#### Order 2. Agaricales

- (1) Basidia borne on lamellae
- (2) Basidiocarp soft and putrescent

##### Family Agaricaceae

- (1) Basidiocarp fleshy
- (2) Gills narrow in section

Example. *Agaricus*

#### Order 3. Aphyllophorales

- (1) Basidiocarp developed gymnocarpously on the outer side
- (2) Texture of basidiocarp not soft and putrescent

##### Family Polyporaceae

- (1) Basidia line the inner surface of the pore or tube
- (2) Pores or tubes generally deep



Example *Polyporus*

### CLASS 3. GASTEROMYCETES

- (1) Basidiocarps permanently closed (angiocarpous)
- (2) Basidia do not become exposed until the spores are mature

#### Order Lycoperdales

- (1) Glebal cavities usually not separating from the peridium or from each other
- (2) Gleba powdery
- (3) Spores light coloured and small

#### Family Lycoperdaceae

- (1) Peridium distinguished into two layers
- (2) Presence of capillitium among spores

Example. *Lycoperdon*

### SUB-DIVISION 5. DEUTEROMYCOTINA

- (1) Mycelium septate
- (2) Perfect stages not known
- (3) Reproduction by asexual means only

### CLASS 1. HYPHOMYCETES

- (1) Mycelium sterile or bearing spores directly or on special branches
- (2) Not aggregated in pycnidia or acervuli

#### Order Moniliales

Conidia borne on free conidiophores

#### Family 1. Dematiaceae

The mycelium, conidiophores and usually the conidia are brown or black in colour

Examples. *Alternaria*, *Cercospora*, *Helminthosporium*

#### Family 2. Moniliaceae

Conidia are borne on free conidiophores, pycnidia or acervuli never formed

Example *Piricularia*

#### Family 3. Tuberculariaceae

Conidiophores usually rather short arising more or less radially from sporodochia

Example *Fusarium*

### CLASS 2. COELOMYCETES

Thallose spores or conidia borne in pycnidium or acervulus

#### Order Melanconiales

- (1) Conidia in acervuli, immersed in substratum
- (2) Conidia black or light coloured, accompanied by setae or not

#### Family Melanconiaceae

Single form-family

Example *Colletotrichum*

## I. MYXOMYCOTA

### 'Slime Molds'

Slime molds, as the members of this group are called, have perplexed the taxonomists due to their characteristics, some of which are plant-like and others like animals. The acellular creeping somatic phase is definitely animal-like in structure whereas the reproductive structures are certainly plant-like. There are about 450 species distributed throughout the world. The members of this group are terrestrial or inhabit decaying wood, bark, other fungi, etc.

The plant body is a naked mass of protoplasm called plasmodium. This thallus is made of either a single large multinucleate protoplasm or many small naked uninucleate protoplasts. Reproduction takes place by the formation of small uninucleate spores, each having a distinct wall. The plasmodia and fructifications of many species of 'Slime Molds' are coloured and beautiful, showing intricate designs.

### *Plasmodiophora*

#### Classification

Kingdom	—	Mycota
Division	—	Myxomycota
Order	—	Plasmodiophorales
Family	—	Plasmodiophoraceae
Genus	—	<i>Plasmodiophora</i>

#### Exercise 1

**Object :** Study the hosts, diseases and the symptoms.

#### Work procedure

Since *Plasmodiophora* is an obligate endoparasite of cabbage. The disease and symptoms are studied by observing a specimen infected by the fungus.

#### Comments

1. The fungus is an obligate endoparasite. It infects the roots of members of Cruciferae, particularly cabbage (*Brassica oleracea* var. *capitata*) cauliflower (*Brassica oleracea* var. *botrytis*) and turnip (*Brassica rapa*), grown in gardens particularly in acidic and poorly drained soil.
2. The disease caused by *Plasmodiophora brassicae* is known as 'club-root' or 'finger-and-toe' disease of crucifers.



Fig. 1. *Plasmodiophora*. Infected cabbage roots.

3. The typical symptoms are shown by the roots which become much swollen, lobed, club-shaped and branched. This is due to hypertrophy, the abnormal enlargement of cells.

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#### Exercise 2

Object : Study of thallus.

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#### Work procedure

A transverse section of the infected part of root is cut, stained in cotton blue and mounted in lactophenol to study the structure

#### Comments

1. The plant body is non-septate, naked and multinucleate mass of protoplasm.
2. The nuclei are free in the cytoplasm and not separated by walls hence it is a plasmodium.

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#### Exercise 2

Object : Study of reproductive structures.

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#### Work procedure

A section of infected part of the root is stained and observed if structures of reproduction could be seen.

#### Comments

1. The vegetative method consists of division of plasmodia into many daughter plasmodia.
2. Myxamoeba is present in the root hair cells.
3. It divides to produce many uninucleate amoebae, which in turn develop into multinucleate plasmodia (primary plasmodia).

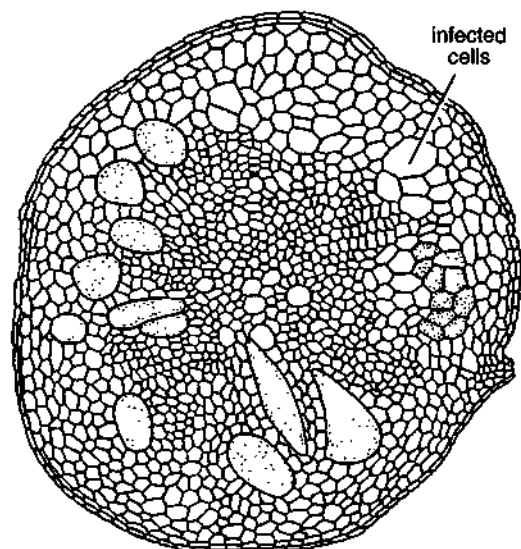


Fig. 2. *Plasmodiophora*. T.s. root of host showing fungus.

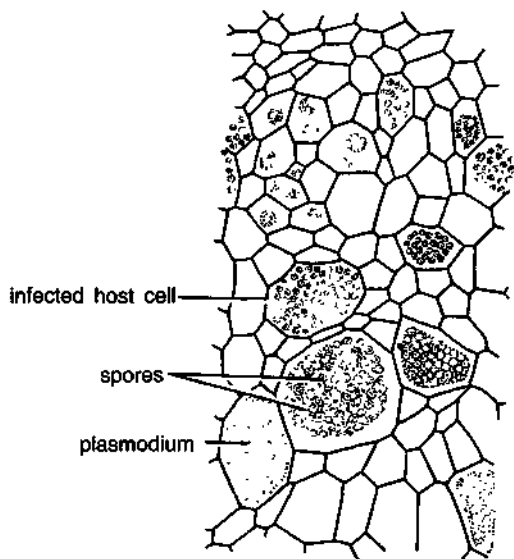


Fig. 3. *Plasmodiophora*. T.s. host root with spores.

## Hints for Collection

The hosts which are the members of Cruciferae are common in the fields. Many of the hosts are cultivated while some grow as weeds.

## II. EUMYCOTA

The group includes fungi with a definite cell wall throughout all stages of vegetative development, and with definite demonstrable nuclei. Most genera have branched filamentous thallus, known as mycelium, however, a few of the primitive forms are unicellular. A single filament or a branch of mycelium is termed as hypha. The mycelium may be coenocytic or septate and multicellular, made up of uni- or multinucleate cells.

Since there is no photosynthetic pigment, the fungus is either a saprophyte or a parasite. A few saprophytic species are aquatic but the great majority are terrestrial and grow either in the soil or upon the remains of plants and animals. When parasitic, the host may be plant or animal. Plant hosts range from the simplest algae to the most advanced angiosperm. The saprophytes (which derive their nutrition from dead and decaying organisms) or parasites (which derive their nutrition from the living cells of the host), are known as obligate saprophytes and obligate parasites respectively. There are other fungi which primarily grow as saprophytes in soil, but on getting a suitable host they turn to be parasites. Such fungi are known as facultative parasites. Similarly, those which live primarily as parasites but may grow as saprophytes after the death of the host, are known as facultative saprophytes.

Except for some simpler unicellular genera, where the entire thallus produces spores or gametes (holocarpic), the majority of genera have only a portion of the thallus becoming reproductive (eucarpic).

Asexual reproduction takes place by variety of spores generally formed within sporangia of distinctive shape. The ascospores and basidiospores are formed at a specific time in the life cycle, after meiosis. Sexual reproduction occurs in all true fungi except deuteromycetes. It takes place by the union of gametes or gametic nuclei. The gametes are formed in gametangia which may be similar in shape to sporangia or their shape may differ from them. The sexual reproduction ranges from isogamy to oogamy and involves two distinct phases—the plasmogamy, the union of cytoplasm, and the karyogamy, the union of two nuclei.

Division Eumycota is divided into 5 sub-divisions : (1) Mastigomycotina, (2) Zygomycotina, (3) Ascomycotina, (4) Basidiomycotina and (5) Deuteromycotina

### Sub-division

#### 1. MASTIGOMYCOTINA

This group includes those members of Phycomycetes which produce motile spores or zoospores. The structures produced as a result of sexual reproduction are oospores. Mastigomycotina is divided into three classes (1) Chytridiomycetes, (2) Hyphochytridiomycetes and (3) Oomycetes

4. Plasmodia form roughly spherical, thin walled zoosporangia which appear packed in the host cells.
5. A zoosporangium has 4-8 uninucleate, biflagellate (flagella unequal, whiplash) zoospores.
6. The zoospore forms haploid plasmodium inside another new host cell.
7. Sexual reproduction takes place by biflagellate isogametes produced in gametangia. The zygote is formed by fusion. The diploid zygote nucleus divides repeatedly to form many multinucleate plasmodia. The plasmodia are transformed into haploid resting spores.
8. Resting spores are released into the soil after root decay.
9. These plasmodia occur in the cortical cells which get hypertrophied.

### Identification

**Kingdom—Mycota.** (1) Chlorophyll absent, (2) Reserve food glycogen, (3) Cell wall of fungal cellulose.

**Division—Myxomycota.** Thallus naked mass of protoplasm—a plasmodium.

**Order—Plasmodiophorales.** Swarm cells (gametes) anteriorly biflagellate.

**Family—Plasmodiophoraceae.** Endoparasitic; vegetative development within living cells of the host.

**Genus—Plasmodiophora.** (1) Plant body plasmodium, (2) Hypertrophy results in club-shaped malformations.

## Synchytrium

### Classification

Kingdom	—	Mycota
Division	—	Euromycota
Sub-division	—	Mastigomycotina
Class	—	Chytridiomycetes
Order	—	Chytridiales
Family	—	Synchytriaceae
Genus	—	<i>Synchytrium</i>

### Exercise 1

**Object :** Study of hosts, diseases and symptoms.

#### Work procedure

Collect a few specimen of diseased plants including the potato tuber. Study the symptoms.

#### Comments

1. The fungus generally occurs as an obligate parasite in the epidermal cells of various angiosperms.
2. *S. endobioticum* attacks potato (*Solanum tuberosum*; vern. alu; fam. Solanaceae) and

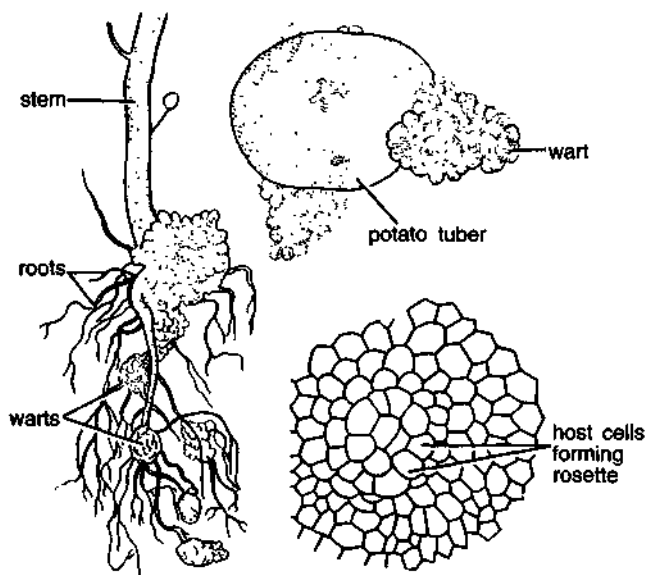


Fig. 1. *Synchytrium endobioticum*. Infected potato tuber causing black wart disease.

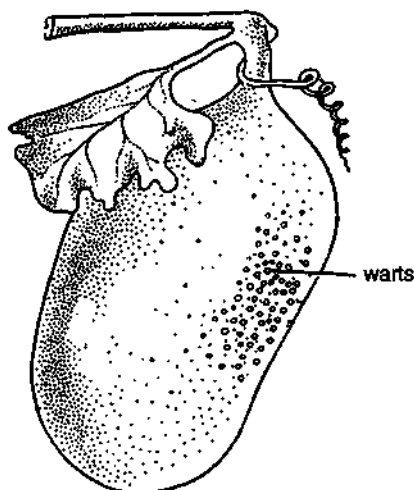


Fig. 2. *Synchytrium trichosanthis*. Infected cucurbit fruit.

causes serious disease called black wart disease or 'potato-wart disease', very common in Europe. In India, it is common in Darjeeling and West Bengal. Generally, the potato tubers are infected and show dark-brown, warty, cauliflower-like outgrowths. Galls may also be formed on aerial parts.

3. *S. trichosanthis* and *S. lagenariae* attack cucurbits and cause warts.
4. *S. rhytzi* attacks *Peristrophe bicalyculata*, (Acanthaceae) and members of Labiatae.
5. *S. sesami* attacks *Sesamum indicum* (vern. til; fam. Pedaliaceae).

### Exercise 2

**Object :** Structure of thallus.

#### Work procedure

Cut a transverse section of the diseased plant part (of cucurbits, *Peristrophe*, etc.) stain, in cotton blue and mount in lactophenol. Study the host cells and the endoparasitic fungus.

#### Comments

1. The thallus is unicellular and non-filamentous.
2. It is endobiotic (lying wholly within the host cell or cells) and holocarpic (whole of the vegetative thallus forms the reproductive unit).
3. The globose, one-celled thallus may either be surrounded by a thick or a thin wall.

### Exercise 3

**Object :** Study of asexual reproductive structures.

#### Work procedure

T.s. of diseased host shows various stages of reproduction.

#### Comments

1. Encysted zoospore infects the host by releasing the contents into the epidermal cell.
2. The fungal thallus enlarges and stimulates the host epidermal cell to enlarge.
3. Adjacent host cells also enlarge to form a rosette of hypertrophied cells surrounding the infected epidermal host cell.
4. The fungus after penetrating the host cell absorbs food material from surrounding protoplasm and grows in size. When it attains a certain size, a two layered wall is secreted, the outer being thick and golden yellow and the inner being thin and transparent. This is called as summer spore or prosorus.
5. Inner wall alongwith protoplast protrudes out in the form of a sac in the upper portion of the host cell, through a pore formed in the outer wall of the summer spore. The nucleus of the summer spore now divides mitotically till there

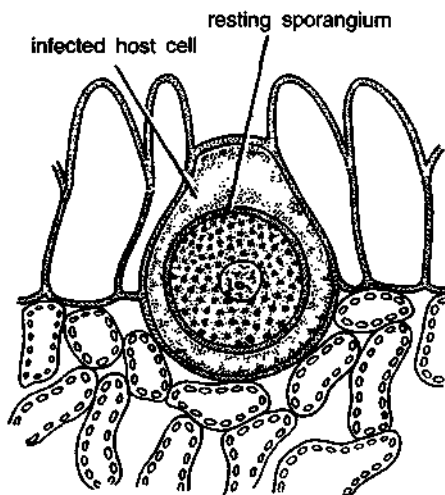


Fig. 3. *Synchytrium*. Resting sporangium in host cell.

are 32 nuclei. This multinucleate protoplast is termed as summer sporangium.

6. These form zoosporangia with many zoospores.
7. Each zoospore is uninucleate and has a single posterior flagellum.
8. The zoospore penetrates the fresh epidermal cells.

### Exercise 4

**Object :** Study of sexual reproductive structures.

#### Work procedure

Study T.s of infected part of the host plant and observe the characters of a resting sporangium.

#### Comments

1. Sexual reproduction takes place by gametes formed during unfavourable conditions.
2. The gametes are also formed in the same way as zoospores.
3. Fusion of isogametes forms a zygote which after swimming in water for a while, penetrates the host cell.
4. When zygote infects a host cell it undergoes hyperplasia (repeated cell division). As a result, zygote and its products get buried deep into the host tissue.
5. The zygote rests inside the host cell, secretes a two layer wall—outer thick and inner thin—around itself and is known as resting spore or winter sporangium.
6. On the return of spring, it divides meiotically into many uninucleate, haploid spores, which after liberation penetrate fresh epidermal host cells.

### Identification

**Kingdom—Mycota.** (1) Chlorophyll absent, (2) Reserve food glycogen, (3) Cell wall of fungal cellulose.

**Division—Eumycota.** A definite cell wall present.

**Sub-division—Mastigomycotina.** Presence of motile spores or zoospores.

**Class—Chytridiomycetes.** (1) Usually unicellular (2) Zoospores uniflagellate.

**Order—Chytridiales.** Asexual reproduction by posteriorly uniflagellate zoospores.

**Family—Synchytriaceae.** Thallus divided into a number of reproductive units and forms a sorus.

Genus—*Synchytrium*. (1) Galls are produced on host, (2) Resting spore is formed.

### Hints for Collection

The common hosts are potato, cucurbit fruits and *Peristrophe bicalyculata*. The former two are cultivated and latter grows wild in the rainy season.

## Saprolegnia (Water Mold)

### Classification

Kingdom	—	Mycota
Division	—	Eumycota
Sub-division	—	Mastigomycotina
Class	—	Oomycetes
Order	—	Saprolegniales
Family	—	Saprolegniaceae
Genus	—	<i>Saprolegnia</i>

#### Exercise 1

**Object :** Study of vegetative structure.

#### Work procedure

Place a few hyphae on the slide, stain in cotton blue and mount in lactophenol. Study the structure of hypha.

#### Comments

1. Most of the species of this genus are saprophytes on animals or vegetable matter, in water or soil. Rarely a few species may be parasitic e.g. *S. ferax* and *S. parasitica* (parasite on fish and their eggs).
2. The thallus is eucarpic, filamentous, coenocytic, multinucleate, much branched and without special organs of attachment.



Fig. 1. *Saprolegnia*. Growing on a dead fly.

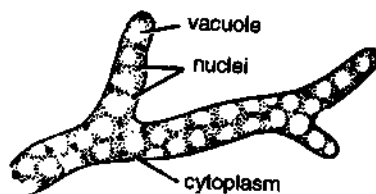


Fig. 2. *Saprolegnia*. A portion of mycelium to show vegetative structure.

3. When it grows as an aquatic saprophyte, some of the small and much branched hyphae penetrate the animal tissues whereas others, which are long, form a fringe around the animal.
4. These external hyphae are fairly stout, more or less straight, with pointed tips when vegetative and terminate in club-shaped zoosporangia.

#### Exercise 2

**Object :** Study of asexual reproductive structures.

#### Work procedure

Prepare a slide showing zoosporangia and study.

#### Comments

1. Asexual reproduction is by zoospores produced in zoosporangia.

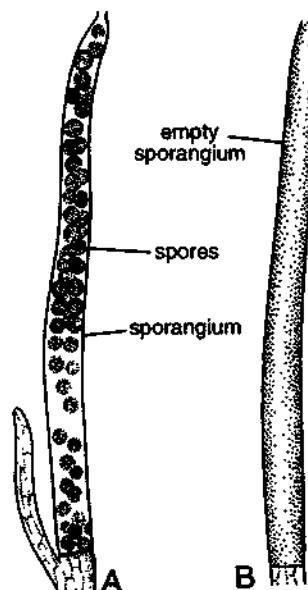


Fig. 3. *Saprolegnia*. A sporangium with (A) and without spores (B).  
(B-14)

- Each zoosporangium is an elongated structure with a pore at the apex and separated from the remainder of the thallus by a septum at its base.
- A zoosporangium produces many pear shaped biflagellate (flagella apical) and uninucleate zoospores.
- These primary zoospores produce secondary bean-shaped zoospores showing two laterally attached flagella.
- The secondary zoospores encyst and later germinate into new hyphae.
- The production of 2 types of zoospores is called diplanetic and phenomenon as diplanetism.
- The new sporangia are formed by proliferation within the empty ones.
- When a sporangium empties its contents, another sporangium is initiated inside the first by bulging out of the basal septum.

### Exercise 3

**Object :** Study of sexual reproductive structures.

#### Work procedure

Stain in cotton blue a part of mycelium showing antheridia and oogonia. Mount in lactophenol.

#### Comments

- The sexual reproduction is oogamous and most of the species are homothallic.
- Oogonia and antheridia are generally borne at the tips of branches or are occasionally intercalary. Arrangement of antheridia and oogonia varies in different species. It may be monoclinal, declinal or androgynous.
- Oogonia are spherical and smooth walled.
- Each oogonium has 1-20 uninucleate oospheres.
- The oospheres are spherical and dark in colour owing to the presence of oil.
- The antheridia are narrow, branched and multinucleate.
- The antheridia grow closely apposed to the oogonia. Each sends a multinucleate fertilization tube which may be branched. It penetrates the oogonial wall and comes in contact with one or more eggs.

(B-14)

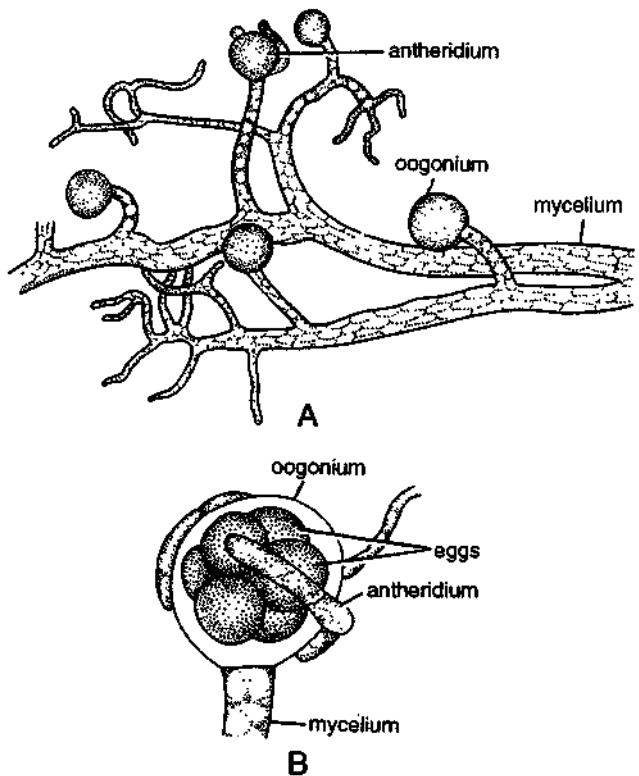


Fig. 4. *Saprolegnia*. A portion of mycelium showing A. Young and B. Mature sex organs.

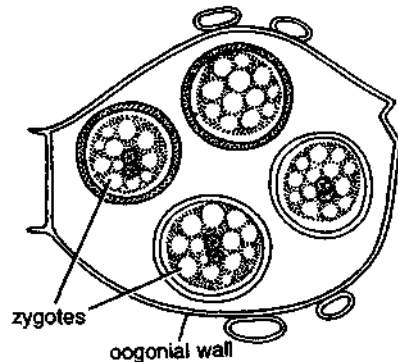


Fig. 5. *Saprolegnia*. Oogonium with zygotes.

- The eggs get fertilized and one to many thick-walled oospores are formed within an oogonium.
- Mature oospore contains membrane-bound vacuole-like body, the ooplast surrounded by cytoplasm containing organelles. The oospores germinate to form the new mycelia.

10. Sometimes the antheridia do not develop close to the oogonia. In such a case, eggs develop inside the oogonium parthenogenetically (i.e. without fertilization).

### Identification

**Kingdom—Mycota.** (1) Chlorophyll absent, (2) Reserve food glycogen, (3) Cell wall of fungal cellulose.

**Division—Eumycota.** A definite cell wall present.

**Sub-division—Mastigomycotina.** Presence of motile spores or zoospores.

**Class—Oomycetes.** (1) Usually mycelial (aseptate), (2) Zoospores biflagellate.

**Order—Saprolegniales.** (1) Mycelial thallus extensive and without a conspicuous holdfast portion, (2) Sporangia cylindrical.

**Family—Saprolegniaceae.** Oogonium with many eggs and lacks periplasm.

**Genus—Saprolegnia.** (1) Aquatic or semi-aquatic and generally a saprophyte, (2) Zoosporangia formed by proliferation.

### Hints for Collection

Boiled hemp (bhang) seeds can be floated on water from ponds, lakes, soil, mud, etc. Hyphae of *Saprolegnia* appears within 3-4 days. A dead fly can also be placed in muddy water and white, cottony mycelium appears in a few days.

## Achlya

### Classification

Kingdom	—	Mycota
Division	—	Eumycota
Sub-division	—	Mastigomycotina
Class	—	Oomycetes
Order	—	Saprolegniales
Family	—	Saprolegniaceae
Genus	—	<i>Achlya</i>

### Exercise 1

**Object :** The study of vegetative structure.

#### Work procedure

Pick up a few hyphae, stain in cotton blue, mount in lactophenol and study.

#### Comments

1. The species are aquatic and occur in a variety of water reservoirs.

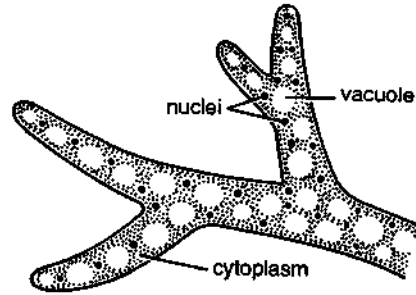


Fig. 1. *Achlya*. A portion of mycelium.

2. The thallus is eucarpic, filamentous, coenocytic, slender, white and much branched.
3. Hyphae are broader at the base and gradually taper towards the apex.
4. The basal cluster of hyphae forms holdfast. However, special organs of attachment are absent.

### Exercise 2

**Object :** Study of asexual reproductive structures—zoosporangia.

#### Work procedure

Select a part of mycetium, stain in cotton blue, mount in lactophenol and study zoosporangia and the spores.

#### Comments

1. Zoosporangia produce asexual reproductive cells called zoospores.

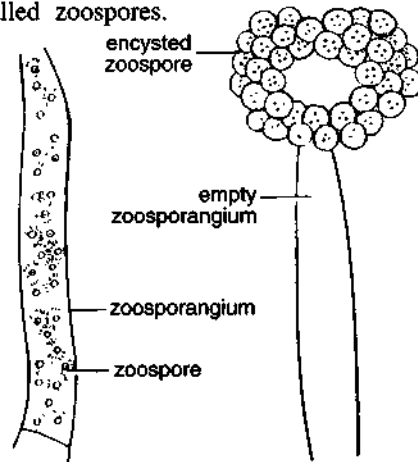


Fig. 2. *Achlya*. A. Mycelium showing zoospores inside zoosporangium, B. Empty zoosporangium with encysted zoospores at its tip. (B-14)



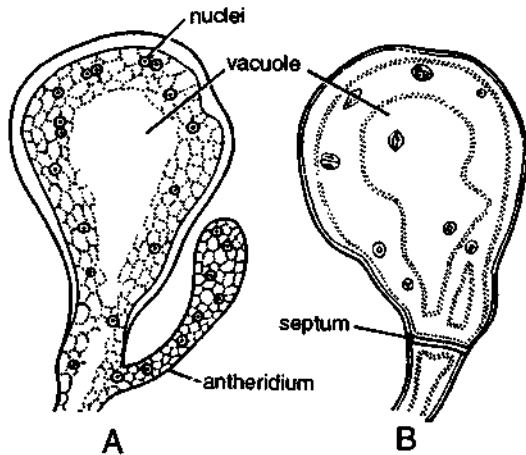


Fig. 3. *Achlya*. A. Oogonium with antheridium at its base, B. Oogonium with septum at its base.

- Each zoosporangium is an elongated, clavate or cylindrical structure. It is separated from rest of the hyphae by a septum at its base and opens by an apical pore.
- Zoospores are biflagellate and are released through apical pore. Zoospores aggregate around the pore and become encysted. (Cysts are liberated after a period of rest in the form of reniform biflagellated zoospores and are known as secondary zoospores). Zoospores germinate and give rise to new mycelium.
- In *Achlya* cymose branching of the hyphae occurs during the development of new zoosporangia, lateral zoosporangia growing out, below the primary and proliferation being absent.

### Exercise 3

**Object :** Study of sexual reproductive structures—oogonia and antheridia.

### Work procedure

Mount a few hyphae in lactophenol after staining in cotton blue. Search for the sex organs and study them.

### Comments

- Sexual reproduction is oogamous. Most of the species are monoecious, but a few species are dioecious (e.g. *A. ambisexualis*, *A. bisexualis*).

(B-14)

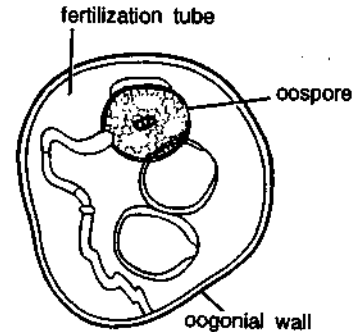


Fig. 4. *Achlya*. Oogonium with oospores.

- Oogonia are either terminal or intercalary. Antheridia arise either from the main hyphae, from secondary ones (diclinous), from the oogonial branches or from oogonial cell (androgynous).
- Oogonia are spherical with a septum at its base. The wall of the oogonium may be smooth (e.g. *A. racemosa*) or spinous (e.g. *A. colorata*).
- Each oogonium has 1-8 eggs (or oospheres) and rarely upto 50.
- Each egg (oospore) is surrounded by a thin delicate wall, contains dense cytoplasm and a single nucleus.
- The antheridia are narrow, branched and multinucleate. Each is cut off from rest of the hyphae by a septum at its base.
- Antheridia grow closely applied to oogonium, send fertilization tubes which penetrate the oogonial wall and fertilize the eggs.
- At this stage oogonium contains many thick walled oospores.

### Identification

**Kingdom—Mycota.** (1) Chlorophyll absent, (2) Reserve food glycogen, (3) Cell wall of fungal cellulose.

**Division—Eumycota.** A definite cell wall present.

**Sub-division—Mastigomycotina.** Presence of motile spores or zoospores.

**Class—Oomycetes.** (1) Usually mycelial (aseptate), (2) Zoospores biflagellate.

**Order—Saprolegniales.** (1) Mycelial thallus extensive and without conspicuous holdfast portion, (2) Sporangia cylindrical.

**Family—Saprolegniaceae.** Oogonium with many eggs and lack periplasm.

**Genus—Achlya.** (1) Some or all zoospores encysting at the mouth of zoosporangium, (2) Zoosporangia not proliferating.

## Hints for Collection

The species of *Achlya* are aquatic and can be collected from ponds, stagnant pools, sink holes and soil. It can be grown in the laboratory on hemp (*Cannabis sativa*), seeds and dead flies.

## Phytophthora

### Classification

Kingdom	—	Mycota
Division	—	Eumycota
Sub-division	—	Mastigomycotina
Class	—	Oomycetes
Order	—	Peronosporales
Family	—	Pythiaceae
Genus	—	<i>Phytophthora</i>

### Exercise I

**Object :** Study of hosts, diseases and symptoms.

### Work procedure

Collect as many hosts as possible with diseases from the following list, preserve them in F.A.A. or dry them for mounting on herbarium sheets.

About 27 species of this genus are found in India. Some are facultative parasites (which live in the soil as saprophytes but develop as parasites in the presence of suitable host), whereas others are parasites on higher plants. Though these species are parasites in their natural habitat, nearly all of them grow on artificial culture media. Thus, parasitism in this genus is less specific.

### Comments

Some of the diseases common in India are as follows—

1. Of all the species, *P. infestans* is economically the most important as it causes late or Irish blight of potato (*Solanum tuberosum*; vern. alu; fam. Solanaceae). The epidemics cause damage in hills and are not common to plains. Sometimes, besides the twigs, the mycelium may also invade the tubers and cause dry or wet rot.



Fig. 1. *Phytophthora*. Potato twig and tubes infected by *P. infestans*.

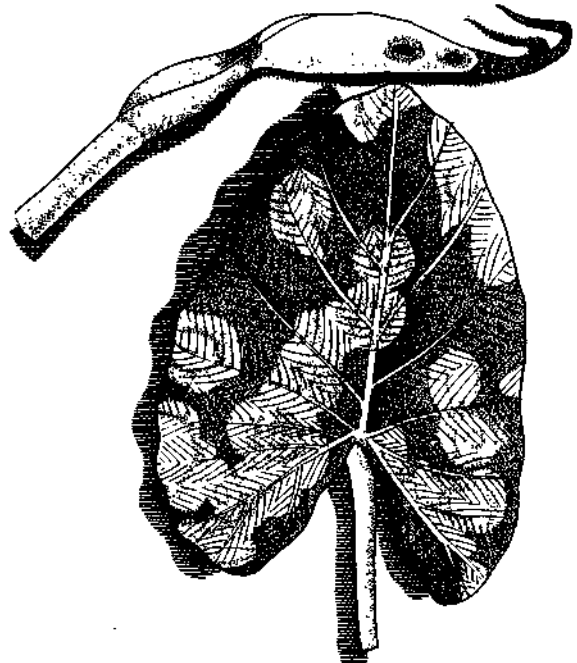


Fig. 2. *Phytophthora*. Inflorescence and the leaf of *Colocasia* infected by *P. colocasiae*.

The first symptoms appear on the leaves as small black patches which increase in size with time. Infection soon spreads to the stems also and the entire shoot falls off in a few days if environmental conditions are suitable. After the tops have been blighted, the underground tubers are also affected. In the earlier stages there is discolouration of skin. Later infection spreads inwards, so that the entire tuber turns brown and decays giving off pungent odour.

2. *P. colocasiae* causes the blight of *Colocasia* leaves and rot of succulent corms of *Colocasia antiquorum* (vern. arvi; fam. Araceae).

In the blight of *Colocasia*, small, dark, roundish specks are formed on leaves. These widen rapidly by centrifugal growth and become circular, oval or triangular. If the conditions are favourable, the petioles and inflorescences also get infected, and in severe cases the corms may rot completely.

3. *P. arecae* causes—**koleroga** of areca palms (*Areca catechu*; vern. supari; fam. Palmae), in western peninsular India. The preliminary symptoms appear on nuts as a water soaked area usually at the base. Ultimately the nuts loose their green colour and begin to fall. While the disease is predominantly on nuts, the tops of trees are occasionally attacked.
4. *P. palmivora* causes bud rot of toddy palms (*Borassus flabellifer*; fam. Palmae) and coconut palms (*Cocos nucifera*; vern. nariyal; fam. Palmae), fruit rot of bread fruit (*Artocarpus incisa*, *A. integrifolia*), black leg of tobacco (*Nicotiana*) and brown rot and gummosis of *Citrus* spp.
5. *P. parasitica* causes seedling blight of castor (*Ricinus communis*; vern. arandi; fam. Euphorbiaceae), leaf blight of sesame (*Sesamum indicum*; vern. til; fam. Pedaliaceae) and leaf and foot rot of betel (*Piper betel*; vern. paan; fam. Piperaceae). In seedling blight the cotyledons are blighted. Rotting of buds occurs in bud rot of various hosts.
6. *P. faberi* causes bud rot of coco (*Theobroma cacao*) and *Hevea*, etc.
7. *P. phaseoli* causes an abscission of the pods of *Phaseolus lanatus* (double bean; fam. Papilionaceae).

8. *Phytophthora* spp. causes damping off of brinjal (*Solanum melongena*), chillies (*Capsicum annuum*), tomato (*Lycopersicon esculentum*) and late blight of *Cinchona* spp.

### Exercise 2

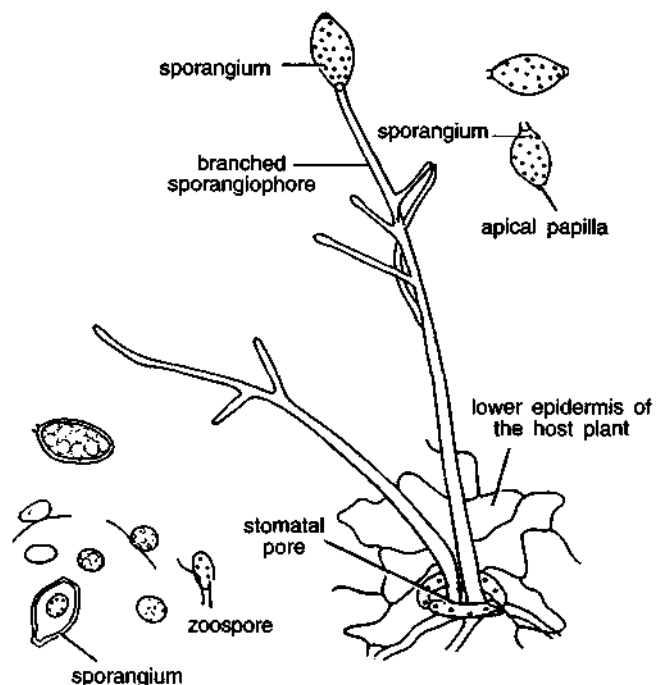
**Object :** Study of vegetative structure.

### Work procedure

Cut a section of infected part of plant. Stain in cotton blue, mount in lactophenol and study.

### Comments

1. *Phytophthora* is eucarpic, inter- or intracellular parasite.
2. The young mycelium is profusely branched and non-septate but old hyphae at the time of reproduction is septate.
3. Hyphae vary in diameter and develop finger-like haustoria which enter the cells of the host.



**Fig. 3.** *Phytophthora*. T. s. of infected leaf showing sporangiophores with sporangia.

### Exercise 3

**Object :** Study of the asexual reproductive structures—sporangia.

#### Work procedure

The spores or conidia appear on the lower side of potato leaf. Tease and stain them in cotton blue. Mount in lactophenol and study.

#### Comments

1. Asexual reproduction takes place by sporangia or conidia.
2. In a diseased patch, certain septate hyphae form the conidiophores which grow out from the stomata on the underside of the leaf.
3. The conidiophore produces ovoid or lemon-shaped, multinucleate, papillate (papilla lamellated) conidia which are at first terminal but become lateral later on, due to the sympodial branching of conidiophore.
4. Each conidium forms many biflagellate zoospores which emerge through papilla. Each zoospore germinates to form a new mycelium.

### Exercise 4

**Object :** Study of sexual reproductive structures—oogonia and antheridia

#### Work procedure

Cut a section of the infected part of tuber, stain in cotton blue, mount in lactophenol and study the sex organs (oospores in *P. infestans* rarely develop in nature but are very common in culture).

#### Comments

1. The sexual reproduction is of oogamous type.
2. The antheridium may either be paragynous i.e. at the side of the oogonium e.g. *P. cactorum* or amphigynous i.e. at the base of the oogonium e.g. *P. infestans*. The latter condition is more common.
3. The oogonium is spherical to pyriform, smooth and hyaline to yellowish. In the centre is an uninucleate oosphere.

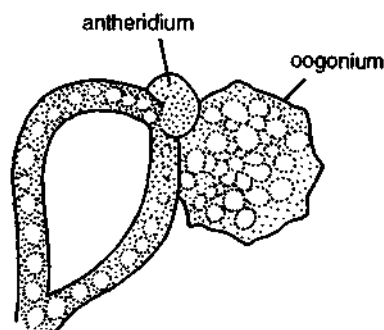


Fig. 4. *Phytophthora*. Sex organs.

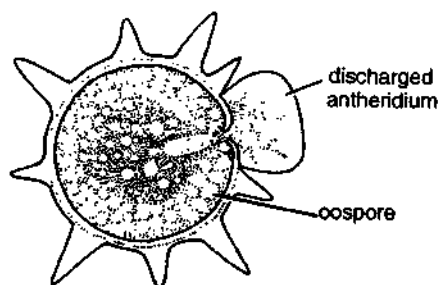


Fig. 5. *Phytophthora*. Oospore.

4. The antheridium has many male nuclei (multinucleate condition).
5. At maturity, the antheridium pierces the oogonium by a fertilization tube. One male nucleus passes into the ooplasm.
6. Male and female nuclei unite to produce oospore which develops a thick wall around itself.
7. Oospore undergoes a rest for a few weeks or months. It gives out a germ tube on germination. It either develops into new mycelium or may form sporangium at its tip.

### Identification

**Kingdom—Mycota.** (1) Chlorophyll absent, (2) Reserve food glycogen, (3) Cell wall of fungal cellulose.

**Division—Eumycota.** A definite cell wall present.

**Sub-division—Mastigomycotina.** Presence of motile spores or zoospores.

**Class—Oomycetes.** (1) Usually mycelial (aseptate), (2) Zoospores biflagellate.

**Order—Peronosporales.** (1) Sexual reproduction aplanogamic (gametes non-motile) and oogamous, (2) Oogonia contain single egg surrounded by periplasm.

**Family—Pythiaceae.** Sporangiohores similar to somatic hypha, or if different, of indeterminate growth.

Genus—*Phytophthora*. (1) Conidia formed sympodially.  
(2) Zoospores liberate individually and not in a vesicle.

### Hints for Collection

The common host of *Phytophthora* are potato, *Colocasia* (arvi), areca nut (supari), toddy palms, coconut palms, castor, coco, etc. All the hosts are cultivated for their economic value and can be collected from the fields. Toddy palms and coconut palms grow near the seacoasts.

### *Albugo* (= *Cystopus*) (White Rust)

#### Classification

Kingdom	—	Mycota
Division	—	Eumycota
Sub-division	—	Mastigomycotina
Class	—	Oomycetes
Order	—	Peronosporales
Family	—	Albuginaceae
Genus	—	<i>Albugo</i>

#### Exercise 1

**Object :** Study of hosts, diseases and symptoms.

#### Work procedure

Collect the diseased parts of hosts from the list given below. Preserve the specimen in F.A.A. or mount them dry on herbarium sheets.

#### Comments

All the twenty five species of *Albugo* are obligate parasites on flowering plants.

The fungus attacks many flowering plants especially the members of Cruciferae.

1. A common disease, the white rust of crucifers, is caused by *A. candida* (= *Cystopus candidus*) in crucifers. The fungus attacks wild as well as the cultivated plants of economic importance, the most important being cabbage (*Brassica oleracea* var. *capitata*; vern. patta gobhi), cauliflower (*Brassica oleracea* var. *botrytis*;

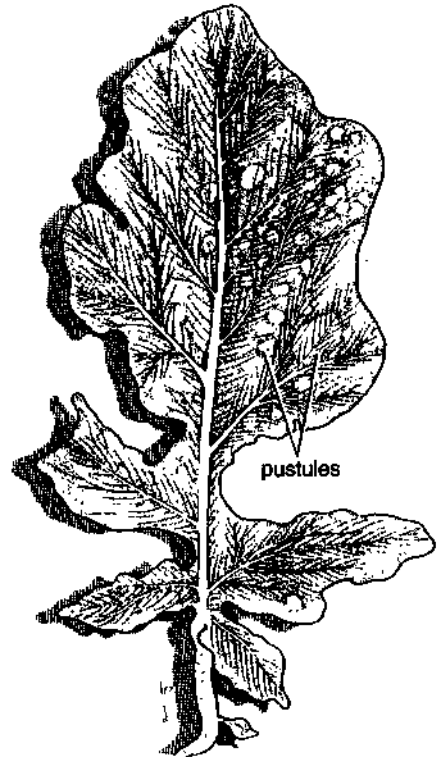


Fig. 1. *Cystopus*. Infected leaf of radish (*Raphanus sativus*) showing white pustules.

vern. phool gobhi), mustard (*Brassica campestris*; vern. sarsaun), radish (*Raphanus sativus*; vern. mooli) and toria (*Eruca sativa*; vern. tarra) etc.

The fungus may attack all parts of the plant with the exception of root. The preliminary symptoms of disease appear on the leaves in the form of white shining pustules which later on coalesce to form patches. When epidermis is ruptured by the pressure of underlying conidia, white powdery mass is seen on the underside of leaf, in the diseased white patch. The leaves in some cases become fleshy and thickened, inrolled and in case of severe infection reduced in size and the entire plant may remain dwarfed. The flowers and stems when infected, show various malformations and enormous hypertrophy (hypertrophy is the abnormal enlargement of the host tissue). The membranous petals become very thick and fleshy.

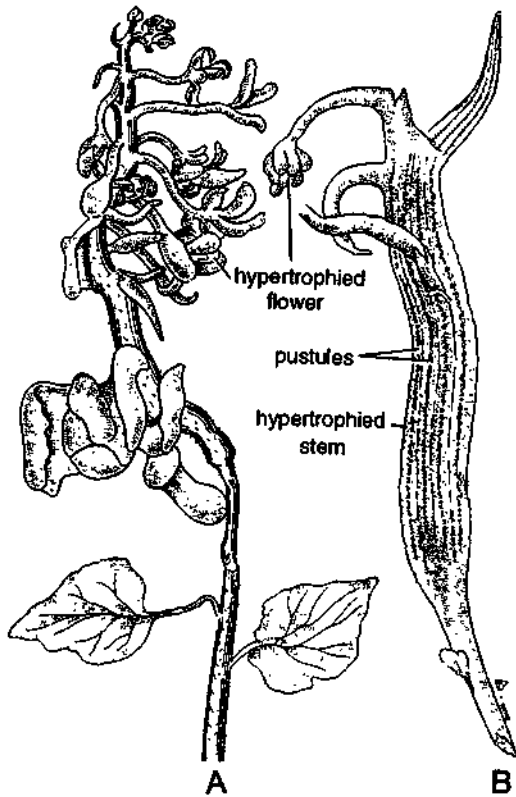


Fig. 2. *Cystopus*. Infection causing enormous hypertrophy in A. *Raphanus sativus* and B. *Eruca sativa*.

1. *A. ipomoeae-pandoranae* attacks *Ipomoea* species (fam. Convolvulaceae) and forms appreciable galls on the stem due to hypertrophy.
2. *A. bliti* is common on members of Amarantaceae.
3. *A. portulacae* infects leaves of *Portulaca* species.

### Exercise 2

Object : Study the vegetative structure.

#### Work procedure

Cut a section through a diseased patch on the leaf of radish, stain in cotton blue, mount in lactophenol and study.

#### Comments

1. The mycelium is branched, intercellular, unseptate and produce globular or knob-shaped haustoria.

2. Haustoria penetrate the host cells and absorb the food material.

### Exercise 3

Object : Study of the asexual reproductive structures — conidia.

#### Work procedure

Cut a T.s. of the infected leaf through a diseased patch. It can be stained in safranin and fast green combination and mounted in glycerine.

#### Comments

1. Asexual reproduction takes place by conidia.
2. The mass of intercellular hyphae beneath the host epidermis produces vertical palisade-like groups of conidiophores.
3. Each conidiophore bears at its tip a chain of conidia arranged basipetally i.e. the youngest at the base of the chain and oldest at the top.
4. In a chain, two conidia are joined with each other by a gelatinous pad called disjuncter.

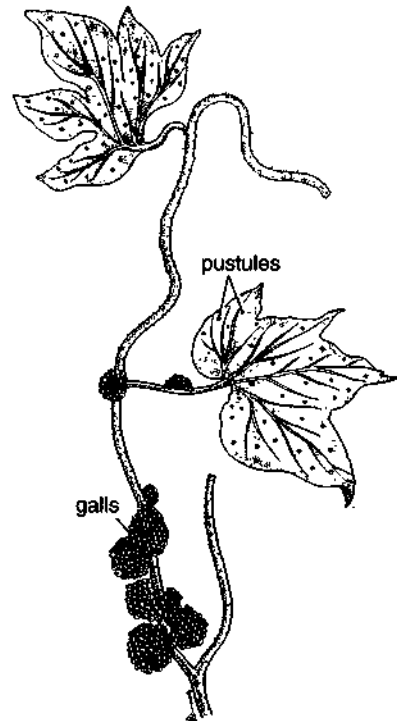


Fig. 3. *Cystopus*. *Ipomoea* sps. infected by *C. ipomoeae pandoranae*.

- Each conidium is multinucleate (5-8 nucleate), hyaline, smooth and spherical.
- The conidiophores constantly cut off conidia forcing underlying epidermis to rupture.
- The conidia are disseminated by wind and germinate either directly (when conditions are unfavourable) by producing a germ tube or may form zoospores (when conditions are favourable).
- The kidney-shaped biflagellate zoospores also germinate to form the new mycelia.

#### Exercise 4

**Object :** Study of sexual reproductive structures—**antheridia and oogonia**

#### Work procedure

Cut a transverse section of radish or toria. Observe the presence of sex organs. Stain with safranin and fast green combination. Mount in glycerine and study.

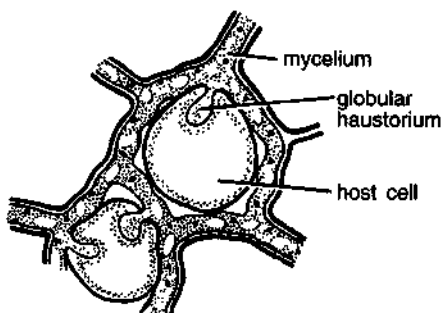


Fig. 4. *Cystopus*. A portion of intercellular mycelium.

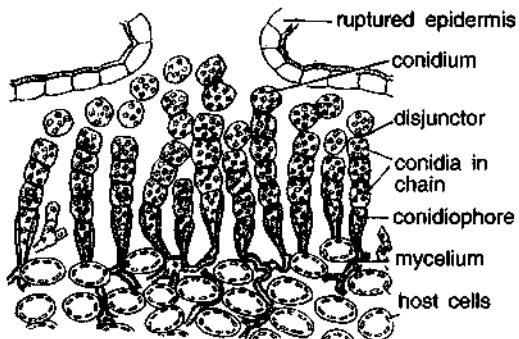


Fig. 5. *Cystopus*. T.s. of infected host leaf showing ruptured epidermis and conidia in chains.

#### Comments

- Sexual reproduction is oogamous. The sex organs are generally formed in the stem towards the end of the growing season of the host.
- The oogonium occurs in the intercellular spaces at the tips of mycelium. It is spherical with a central ooplasm and peripheral periplasm. In a *A. candida* ooplasm is uninucleate. (In *A. bliti* ooplasm is multinucleate).
- The antheridium is paragynous in position and contains several nuclei.
- The antheridia and oogonia are separated from remainder of the mycelium by a septum.
- A mature antheridium develops a slender fertilization tube which grows through the oogonial wall and the periplasm and penetrates deeply into the ooplasm or oosphere.
- A single male nucleus enters and fuses with the nucleus in the oosphere thus effecting fertilization.
- In some species (e.g. *A. bliti*), the oosphere or ooplasm is multinucleate and these nuclei get

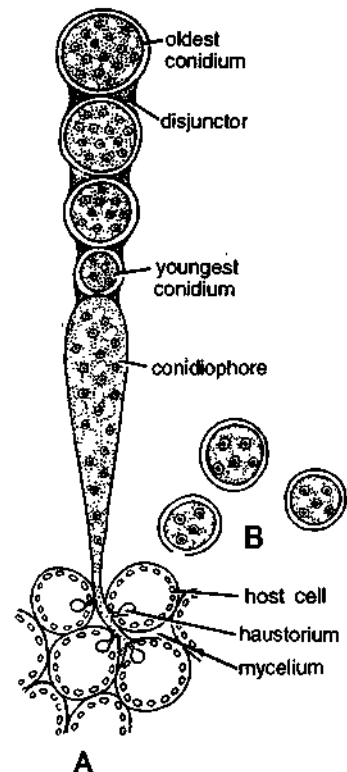


Fig. 6. *Cystopus*. A. Single conidiophore showing conidia in chain B. Conidia.

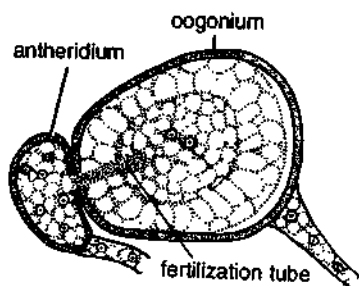


Fig. 7. *Cystopus*. Oogonium and antheridium.

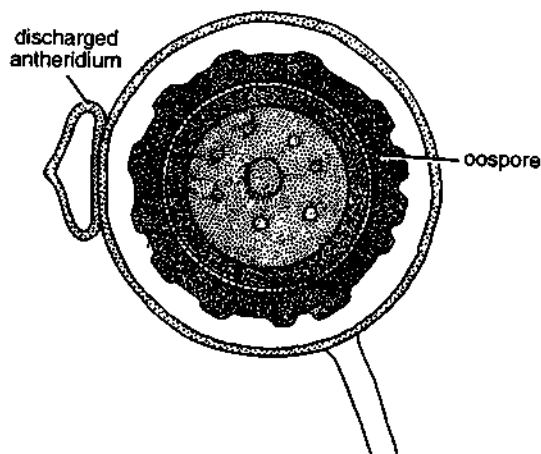


Fig. 8. *Cystopus*. Oospore.

fertilised by the entry of equal number of male nuclei. The resulting oospores or zygotes are thus multinucleate.

8. The oospore develops a thick, ornamented and three layered wall.
9. The nucleus of the oospore divides meiotically. A vesicle is produced in which 40-60 biflagellate zoospores are formed. Zoospore germinates into a new mycelium.

### Identification

**Kingdom—Mycota.** (1) Chlorophyll absent, (2) Reserve food glycogen, (3) Cell wall of fungal cellulose.

**Division—Eumycota.** A definite cell wall present.

**Sub-division—Mastigomycotina.** Presence of motile spores or zoospores.

**Class—Oomycetes.** (1) Usually mycelial (aseptate), (2) Zoospores biflagellate.

**Order—Peronosporales.** (1) Sexual reproduction aplanogametic (gametes non-motile) and oogamous, (2) Oogonia contain single egg surrounded by periplasm.

**Family—Albuginaceae.** (1) Mycelium intercellular, provided with haustoria, (2) Conidia in chains on clavate conidiophores.

**Genus—Albugo.** (1) White shining pustules on leaves, (2) Infected parts (stem and flower) exhibit hypertrophy, (3) Conidia arranged basipetally on conidiophores.

### Hints for Collection

The common hosts are cultivated crucifers, *Ipomoea*, members of *Amarantaceae* and *Portulaca*, etc. The cultivated crucifers and some members of *Amarantaceae* can be obtained from the fields. *Ipomoea* spp. is a weed in the crop fields. *Cystopus* occurs on these hosts in January-February. *Cystopus* is also common on *Achyranthes* spp. (*Amarantaceae*) which grows as a weed in dry soils. *Portulaca* is an ornamental plant common in gardens.

### Sub-division

## 2. ZYGOMYCOTINA

This group includes those members of fungi (Phycomycetes) which reproduce asexually by non-motile spores formed inside the sporangium. Thallus is usually mycelial and hyphae are aseptate. Sexual reproduction takes place by gametangial copulation. It is typically isogamous. Zygospores are formed as a result of sexual reproduction.

### *Rhizopus* (Bread Mold)

#### Classification

Kingdom	—	Mycota
Division	—	Eumycota
Sub-division	—	Zygomycotina
Class	—	Zygomycetes
Order	—	Mucorales
Family	—	Mucoraceae
Genus	—	<i>Rhizopus</i>

#### Exercise 1

**Object :** Study of hosts, diseases and the symptoms.

#### Work procedure

Collect diseased hosts, preserve them in formalin or alcohol. Also grow *Rhizopus* by keeping a slice of bread in moist chamber. This provides ready-made material for study.



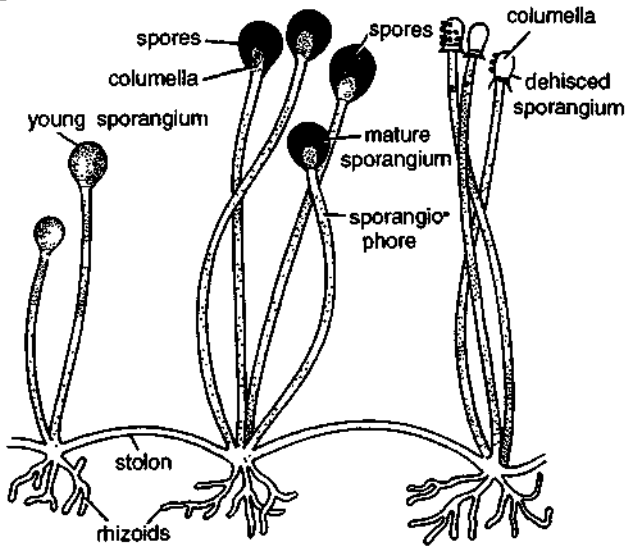


Fig. 1. *Rhizopus*. A portion of mycelium with sporangiophores.

### Comments

Most of the species of *Rhizopus* are saprophytic and grow on dead vegetables or animal matter. *R. stolonifer* grows so frequently on bread that it is often called the 'bread mold'. It is also called 'black mold' for its black coloured sporangia and 'Pin mold' for globose sporangia at the tips of branches look like pin heads. Only a very few species viz. *R. artocarp*i and *R. arrhizus* are weak parasites.

Only a few species of *Rhizopus* attack the plants.

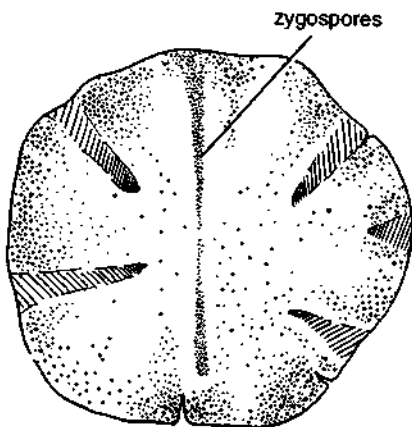


Fig. 2. *Rhizopus*. Potato culture plate showing heterothallism.

1. *R. artocarp*i causes fruit drop of jack fruit (*Artocarpus heterophyllus*; vern. kathal; fam. Moraceae).

In fruit drop of jack fruit, the premature fruits are shed due to the attack of the fungus. The attacked peduncles become black in colour.

2. *R. artocarp*i causes fruit rot of apples (*Pyrus malus*; vern. sev; fam. Rosaceae).

In fruit rot of apples, the apples rot away completely, in conditions of severe infection.

### Exercise 2

**Object : Study of vegetative structure.**

### Work procedure

Pick up a few hyphae growing on a slice of bread. Stain with cotton blue, mount in lactophenol and study.

### Comments

1. The mycelium shows abundant, white cottony growth.
2. The young mycelium is multinucleate, aseptate, with all the hyphae alike.
3. In the older mycelium three parts of hyphae can be distinguished (i) branched rhizoids that penetrate the substratum, (ii) stolon or runner growing horizontally above the substratum for some distance and then bending downward, producing another group of rhizoids and (iii) the sporangiophores which grow upward in tufts from the point where the stolons form rhizoids.

### Exercise 3

**Object : Study of structure —sporangia.**

### Work procedure

Pick up a black mass of mycelium, stain with cotton blue, mount in lactophenol and study.

### Comments

1. The asexual reproductive structures are sporangia borne by the sporangiophores.
2. Each sporangiophore is swollen at the tip and forms sporangium.

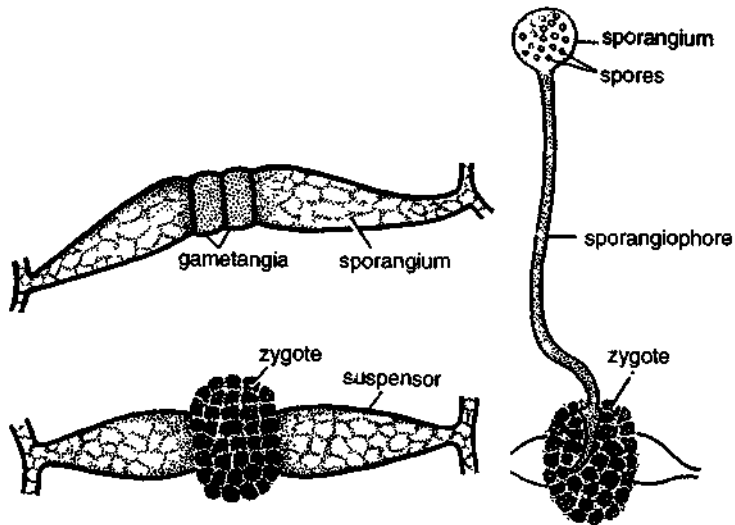


Fig. 3. *Rhizopus*. Various stages in sexual reproduction.

- The sporangium has a columella in the centre and the space between columella and wall of the sporangium is packed with aplanospores. This is known as the spore sac.
- The aplanospores are angular or rounded and multinucleate. The spores are colourless or coloured blue or brown with cuticularised or smooth wall showing longitudinal striations.
- Spores are liberated by breaking of the sporangial wall.
- Each spore germinates to form a new mycelium.
- The phenomenon can be demonstrated by a 'Potato culture plate' where, on the two opposite ends, mycelia of + and - strains were grown. These met in the centre to produce the zygospores. The zygospores appeared as a black streak in the centre of the potato chip.
- The hyphae of the opposite strains produce erect branches. A transverse division separates the terminal gametangium from a proximal suspensor cell. Suspensor is straight, large and swollen.

- The two isogametangia (also called coenogametangia) conjugate to form a zygospore (zygote) which has a thick wall and rough outer surface.
- Zygospore divides meiotically after a period of rest. It germinates by producing a promycelium which develops a sporangium (often called zygosporangium) at its tip.
- The sporangium produces many multinucleate spores. These germinate to form new mycelium.

#### Exercise 4

**Object :** Study of the sexual reproductive structures.

#### Work procedure

These are gametangia which are formed rarely. The spores are grown on potato chip by placing a few spores in sexual reproductive structures and then picking up zygospores from the centre. Zygospores are the fusion products of gametangia. Stain with cotton blue and mount in lactophenol.

#### Comments

- The genus is heterothallic and the sexual reproduction takes place only when mycelia of + (plus) and - (minus) strains meet.

#### Identification

**Kingdom—Mycota.** (1) Chlorophyll absent, (2) Reserve food glycogen, (3) Cell wall of fungal cellulose.

**Division—Eumycota.** A definite cell wall.

**Sub-Division—Zygomycotina.** Asexual reproduction by non-motile spores—aplanospores.

**Class—Zygomycetes.** (1) Gametangia morphologically similar, (2) Sexual reproduction forms a zygospore.

**Order—Mucorales.** (1) Mostly saprophytic, (2) Asexual reproduction by typical non-motile aplanospores.

**Family—Mucoraceae.** (1) Sporangia with many spores, and well developed columella, (2) Sporangial wall relatively thin and easily breakable or deliquescent.

**Genus—Rhizopus.** Sporangioophores arise at rooting nodes of the stolon.

### Hints for Collection

Since almost all the species of *Rhizopus* are saprophytes, the fungus can be grown on dead organic matter, viz. bread, butter, glycerine, etc., by keeping them in damp and dark conditions. Weak parasitic forms are common on jack fruit and apple which are grown in orchards.

## Sub-division 3. ASCOMYCOTINA (ASCOMYCETES)

The ascomycetes, well known as sac-fungi, comprise some 30,300 species, according to a conservative estimate. These are characterized by a reproductive body, known as ascus which has usually eight ascospores. Ascomycetes include both saprophytic and parasitic species; the saprophytes being generally terrestrial.

The hyphae, unlike those of phycomycetes, are septate and commonly form stromata, sclerotia and chlamydo-spores.

Asexual reproduction takes place by budding and conidia formed at the tips of conidiophores.

Sexual reproduction is isogamous in lower forms and heterogamous in higher forms where ascogonium (female sex organ) gets fertilized by antheridium (male sex organ) through a trichogyne which forms a receptive structure of the female cell. After fertilization, the ascogonium develops a number of ascogonous hyphae which ultimately form asci and paraphyses. The asci are surrounded by somatic hyphae to form typical fruiting bodies, known as ascocarps which are of 3 types viz.

1. **The cleistothecium.** It is a spherical body which opens by breakage of its outer wall, discharging the asci within.

2. **The perithecium.** It is a flask-shaped fruiting body with a narrow ostiole, through which the asci and ascospores are released.

3. **The apothecium.** It is an open saucer shaped fruiting body, lined on its inner side with asci arranged in parallel layer. The asci discharge the ascospores directly into the atmosphere.

The sub-division is further divided into (1) Hemiascomycetes, (2) Laculoascomycetes, (3) Plectomycetes, (4) Loboulbeniomycetes, (5) Pyrenomycetes and (6) Discomycetes.

## Erysiphe (Powdery Mildew)

### Classification

Kingdom	—	Mycota
Division	—	Eumycota
Sub-division	—	Ascomycotina
Class	—	Plectomycetes
Order	—	Erysiphales
Family	—	Erysiphaceae
Genus	—	Erysiphe

### Exercise 1

**Object :** Study of hosts, diseases and symptoms.

### Work procedure

*Erysiphe* infects large number of hosts given below. Collect as many specimen as possible and either preserve them in formalin or dry for mounting on herbarium sheets.

### Comments

All the species (approximately 22) of *Erysiphe* are obligate parasites which grow superficially on host (i.e. as ectoparasites). These generally parasitize angiosperms.

The following three species are most important.

1. *E. polygoni* infects pea (*Pisum sativum*; vern. matar; fam. Papilionaceae) and other economically important legumes viz. *Vigna sinensis*, *Lathyrus sativus*, *Phaseolus lunatus*, *P. vulgaris* and *Vicia faba*. It also parasitizes *Coriandrum sativum*, *Cuminum cyminum*, *Papaver somniferum*, etc.

This fungus causes a disease commonly known as powdery mildew of peas. The earliest symptoms appear on the upper surface of the older leaves as small, white, circular and powdery spots and enlarge rapidly and cover the entire leaf surface. In advanced stages the

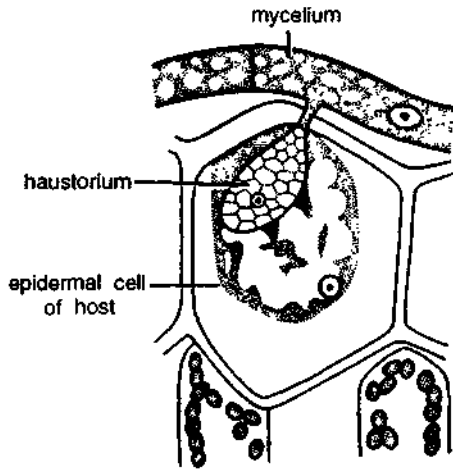


Fig. 1. *Erysiphe*. Haustorium of *E. cichoracearum* inside the host cell.

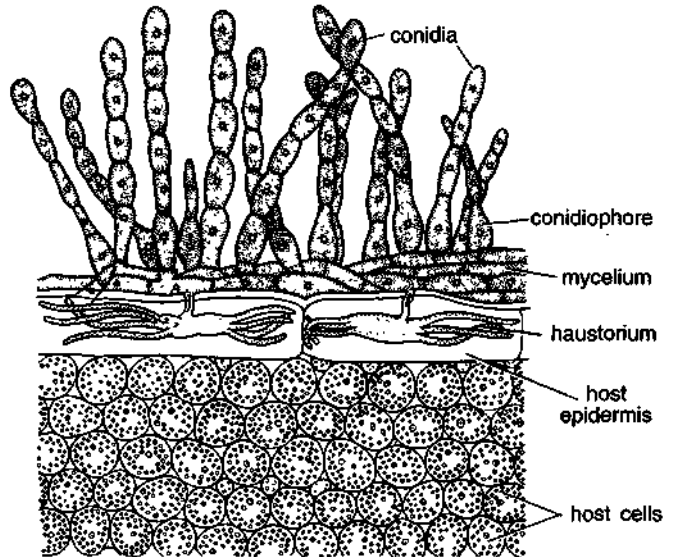


Fig. 2. *Erysiphe*. Section through host leaf showing haustoria, conidiophores and conidia.

leaves become covered with a white powder, get reduced in size and are finally shed. Immature pods are also attacked which become shrivelled and dried.

- E. graminis* f. sp. *hordei* parasitizes barley (*Hordeum vulgare*; vern. jaii; fam. Graminae) and causes the powdery mildew of barley. *E. graminis* f. sp. *tritici* infects wheat. It is first evidenced as numerous colonies of superficial, flocculent growth on the upper surface of the leaves. They are white to begin with turning grey or red later on with a powdery appearance and form a cushion-like growth. The infection increases the transpiration and the plants become stunted through reduction in size and number of leaves. The leaves that are not shed become wrinkled and deformed.
- E. cichoracearum* infects tobacco (*Nicotiana*), lady's finger or okra (*Abelmoschus esculentus*) and various cucurbits to cause powdery mildew.

#### Exercise 2

**Object :** Study of vegetative structure.

#### Work procedure

Collect hyphae from the leaf surface of pea, stain with cotton blue, mount in lactophenol and study.

#### Comments

- Since the fungus is an ectoparasite, the mycelium forms a white, interwoven covering on the host surface.
- The branched mycelium is septate and the cells are uninucleate.
- Mycelium produces simple, globose, lobed haustoria which penetrate the epidermal cells of the host.
- Generally haustoria are inflated but in others (*E. graminis*) these branch into finger-like projections.

#### Exercise 4

**Object :** Study of conidia.

#### Work procedure

Pick up mycelium from host surface. Stain in cotton blue, mount in lactophenol and study.

#### Comments

- The conidia are asexual reproductive bodies formed in chains at the tips of conidiophores.

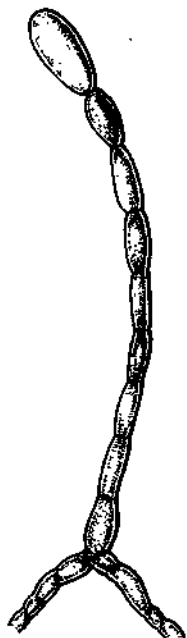


Fig. 3. *Erysiphe*. A conidiophore with conidia in chains.

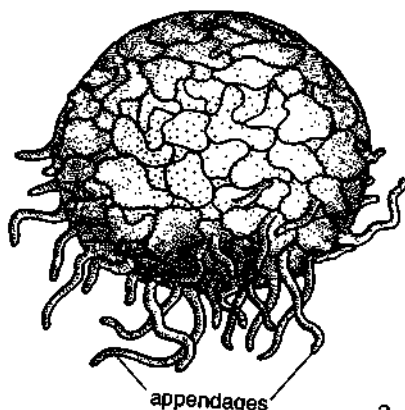


Fig. 4. *Erysiphe*. An ascocarp.

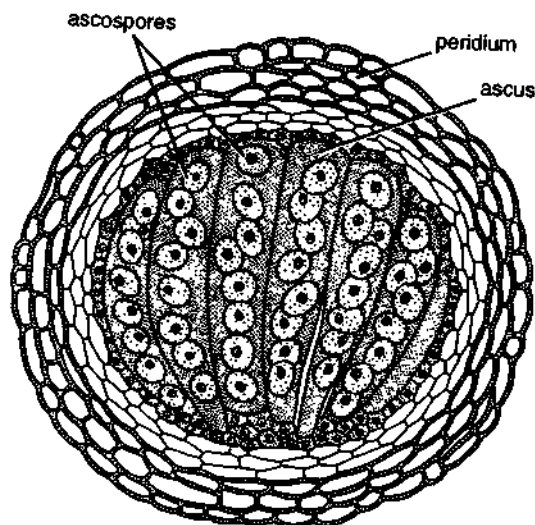


Fig. 5. *Erysiphe*. V.s. through an ascocarp.

2. A conidiophore arises vertically from the mycelium. It is unbranched and swollen at the base in a characteristic manner.
3. Each conidiophore bears at its tip many conidia arranged in a basipetal chain.
4. The mature conidia are elliptical, barrel-shaped or sometimes even cylindrical and measure about 30-45 microns in length and 10-19 microns in breadth.
5. The conidia are disseminated by wind and germinate by producing many germ tubes.

#### Exercise 5

**Object :** Study of ascocarp, asci and ascospores.

#### Work procedure

Pick up the mycelium at a stage when sexual reproduction is over. Stain in cotton blue, mount in lactophenol and study the ascocarp.

#### Comments

1. The ascocarp is a cleistothecium. It is formed after fertilization.

2. It is a globose structure surrounded by a protective covering—the peridium which at maturity becomes 6-10 cells in thickness.
3. Certain superficial cells of the peridium develop into characteristic elongated unbranched appendages with free ends.
4. Within the peridium are present many asci which are more or less stalked and ovate.
5. Each ascus generally contains eight ascospores (four to five in *E. polygoni*), two or rarely three in *E. cichoracearum*.
6. The ascospores are elliptical, one celled, uninucleate and hyaline.
7. The ascospores are set free either by an irregular cracking of the upper part of the peridium or the cleistothecium may split transversely from its upper part and may come off as a lid.
8. The ascospores after liberation, germinate by producing a germ tube, if they happen to fall on a suitable host.

#### Identification

**Kingdom—Mycota.** (1) Chlorophyll absent, (2) Reserve food glycogen, (3) Cell wall of fungal cellulose.

**Division—Eumycota.** A definite cell wall present.

**Sub-division—Ascomycotina.** (1) Mycelium septate, (2) Spores borne endogenously in the ascus, (3) Spores in definite numbers, in multiples of two, usually eight.

**Class—Plectomycetes.** Ascocarp, a cleistothecium.

**Order—Erysiphales.** Ectoparasites.

**Family—Erysiphaceae.** (1) Aerial mycelium hyaline, (2) Enormous production of conidia on host surface gives it a white powdery appearance.

**Genus—Erysiphe.** (1) Ascospores one celled, (2) Cleistothecia normally containing many asci, (3) Appendages or fruiting body hypha-like.

### Hints for Collection

The fungus is found as an ectoparasite on pea, wheat, cow pea, lathyrus, moong, urd, coriander, cumin, poppy, lady's finger, cucurbits and barley which are cultivated all over India. The fungus would be readily identified by the white powdery covering on the host leaves.

## Sphaerotheca

### Classification

Kingdom	—	Mycota
Division	—	Eumycota
Sub-division	—	Ascomycotina
Class	—	Plectomycetes
Order	—	Erysiphales
Family	—	Erysiphaceae
Genus	—	Sphaerotheca

### Exercise 1

**Object :** Study of hosts, diseases and symptoms.

#### Work procedure

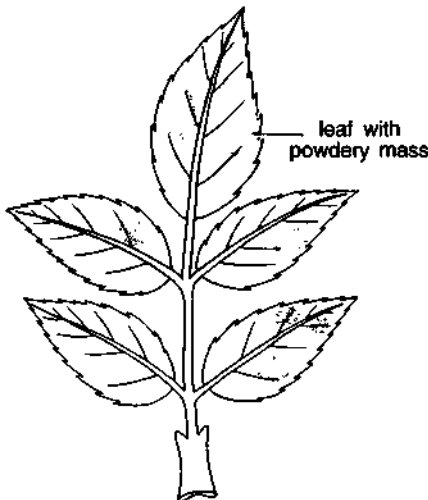


Fig. 1. *Sphaerotheca*. A rose leaf showing powdery mildew, caused by *S. pannosa*.

Collect as many infected hosts as possible out of the list given below. Note the symptoms.

#### Comments

1. *Sphaerotheca fuliginea* causes powdery mildew of cucurbits like *Lagenaria*, *Luffa* and *Cucurbita*.  
Tiny white superficial spots appear on leaves and stem. The spots become powdery on enlarging. Ultimately, there is premature defoliation.
2. *Sphaerotheca pannosa* is parasitic on roses.

### Exercise 2

**Object :** Study of vegetative structure.

#### Work procedure

A T.s. of the host leaf is very useful for studying the mycelium. Stain in cotton blue, mount in lactophenol and study.

#### Comments

1. The thallus is mycelial and consists of richly branched and septate hyphae. The cells are uninucleate.
2. The mycelium is superficial but develop simple, globose and lobed haustoria inside the host cells.
3. The superficial mycelium bears erect conidiophores.

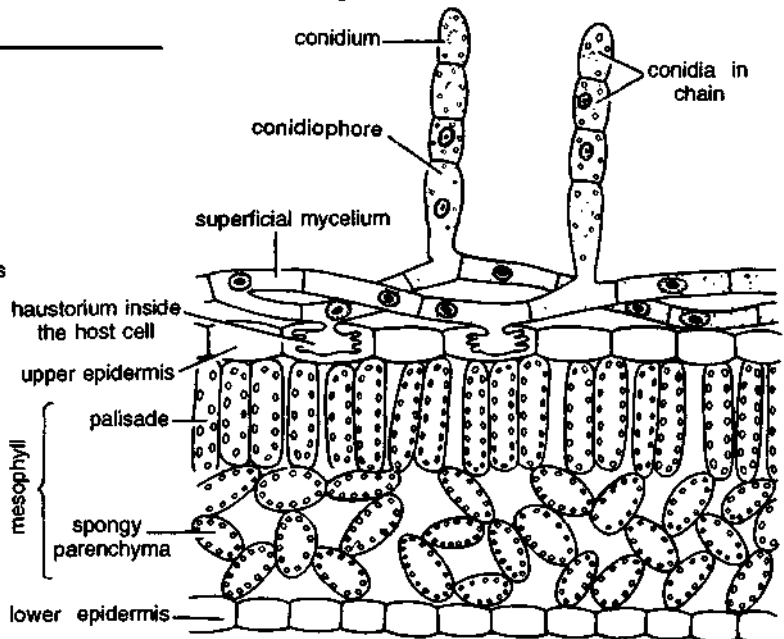


Fig. 2. *Sphaerotheca*. T.s. of infected host leaf showing mycelium and haustoria.

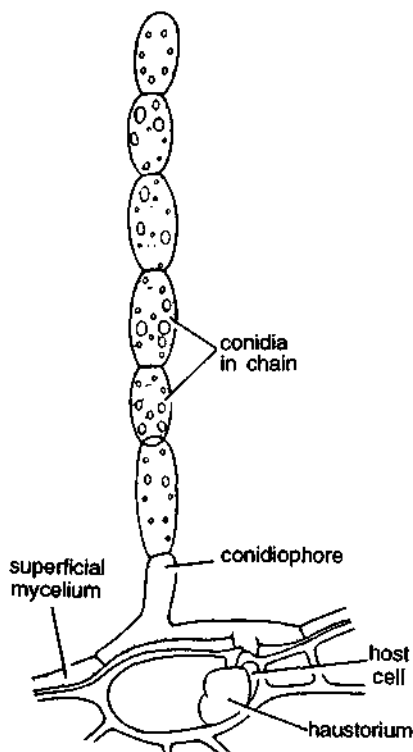


Fig. 3. *Sphaeroteca*. Conidiophore with a chain of conidia.

4. Conidia, the bodies of asexual reproduction, are present in chains in basipetal succession at the tip of conidiophore.
5. The conidia are barrel-shaped, hyaline and uninucleate. It germinates to produce a forked germ tube.

### Exercise 3

**Object :** Study of ascocarp, asci and ascospores.

#### Work procedure

Ascocarps are cleistothecia found mainly on twigs in thick mycelial felt. These are stained in cotton blue and studied. A little pressure is applied to the coverslip to break cleistothecia so that asci and ascospores are studied.

#### Comments

1. Ascocarps are closed ball-like structures called cleistothecia. These are the products of sexual reproduction.

(B-14)

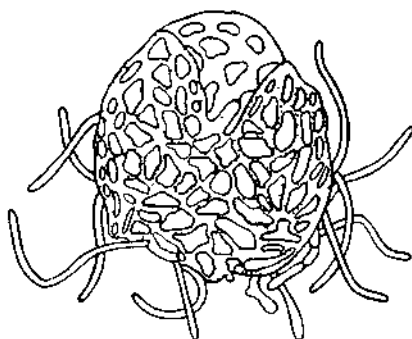


Fig. 4. *Sphaeroteca*. A single cleistothecium.

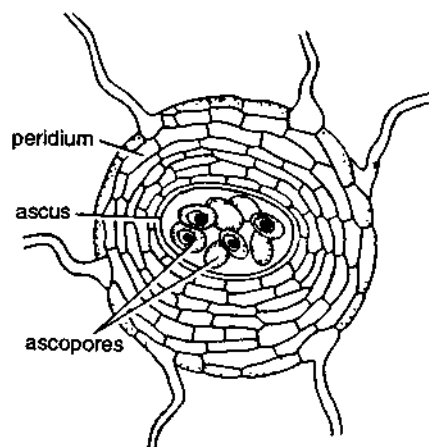


Fig. 5. *Sphaeroteca*. T.s. through cleistothecium.

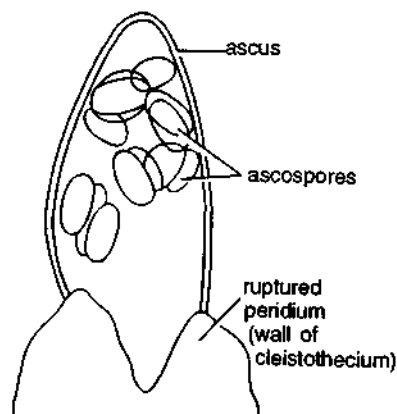


Fig. 6. *Sphaeroteca*. A ruptured cleistothecium showing single ascus.

2. Cleistothecium is globose and dark brown in colour. It bears simple myceloid appendages all over the surface.

- The wall of the cleistothecium is thick and two layered. Each layer is several cells thick. The outer cells are darker.
- Each cleistothecium has only one obovoid to elliptic ascus. Inside the ascus are present eight haploid ascospores.

### Identification

**Kingdom—Mycota.** (1) Chlorophyll absent, (2) Reserve food glycogen, (3) Cell wall of fungal cellulose.

**Division—Eumycota.** A definite cell wall present.

**Sub-division—Ascomycotina.** (1) Mycelium septate, (2) Spores borne endogenously in the ascus, (3) Spores in definite numbers, in multiples of two, usually eight.

**Class—Plectomycetes.** Ascocarp, a cleistothecium.

**Order—Erysiphales.** Ectoparasites.

**Family—Erysiphaceae.** (1) Aerial mycelium hyaline, (2) Enormous production of conidia on host surface gives it a white powdery appearance.

**Genus—Sphaerotheca.** Ascocarp with a single ascus.

### Hints for Collection

The fungus is an ectoparasite on cucurbits like *Lagenaria*, *Luffa* and *Cucurbita*. These plants are seasonal and are grown for their edible parts. The fungus can be easily collected from them during their growth season.

## Phyllactinia

### Classification

Kingdom	—	Mycota
Division	—	Eumycota
Sub-division	—	Ascomycotina
Class	—	Plectomycetes
Order	—	Erysiphales
Family	—	Erysiphaceae
Genus	—	<i>Phyllactinia</i>

#### Exercise 1

**Object :** Study of hosts, diseases and symptoms.

#### Work procedure

Collect specimen as mentioned below. Preserve in a container with formalin, F.A.A. or alcohol 90%. A dry specimen can also be mounted on a herbarium sheet.

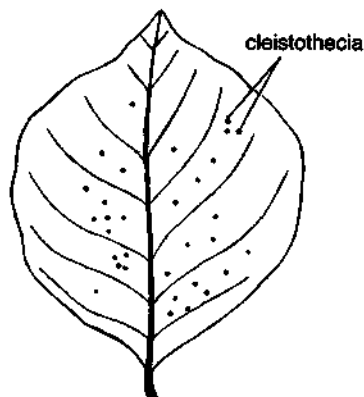


Fig. 1. *Phyllactinia*. Leaf of *Dalbergia sissoo* showing cleistothecia.

### Comments

The fungus is a hemiendophytic parasite and commonly causes powdery mildew.

*Phyllactinia* parasitizes leaves of over hundred species of plants, chiefly the deciduous trees. Some of the important species are listed below.

- P. guttata* (= *P. corylea*) is a very common species parasitizing *Corylus*, *Betula* (vern. bhojpatra; fam. Betulaceae), *Cassia fistula* (vern. amaltass; fam. Caesalpiniaceae), etc.
- P. dalbergiae* is very common in northern India and attacks *Dalbergia sissoo* (vern. shisham; fam. Papilionaceae).
- P. moricola* is found on leaves of *Morus alba* (vern. shahtoot; fam. Moraceae).
- P. acaciae* attacks some species of *Acacia* (fam. Mimosaceae).
- P. yarwoodii* is found on the leaves of *Dalbergia volubilis* and *D. lanceolaria*.

The disease caused by species of *Phyllactinia* is known as 'powdery mildew', owing to the presence of white powder on the host surface. It is the result of profuse extramatrical mycelium spreading on the host surface. The disease is air borne.

#### Exercise 2

**Object :** Study the vegetative structure.

#### Work procedure

Pick up the mycelium from the host leaf surface, stain with cotton blue, mount in lactophenol and study.



### Comments

1. The mycelium which spreads over the surface of the host possesses septate hyphae with uninucleate cells.
2. The saccate haustoria are formed in the mesophyll cells bordering the sub-stomatal chamber by 6-7 celled hyphal branches of limited growth. The mycelium is thus hemiendophytic.

### Exercise 3

**Object :** Study of asexual reproductive structures.

### Work procedure

Mount the mycelium from lower leaf surface, stain in cotton blue, mount in lactophenol, search for conidia and study.

### Comments

1. Conidia are the asexual reproductive structures formed on conidiophores; the latter develop from the superficial mycelium.
2. The conidiophores are abundant, first on both the surfaces of the leaves but later become restricted only to the lower surface.
3. Each conidiophore is 3-4 celled erect branch terminating into a single conidium.

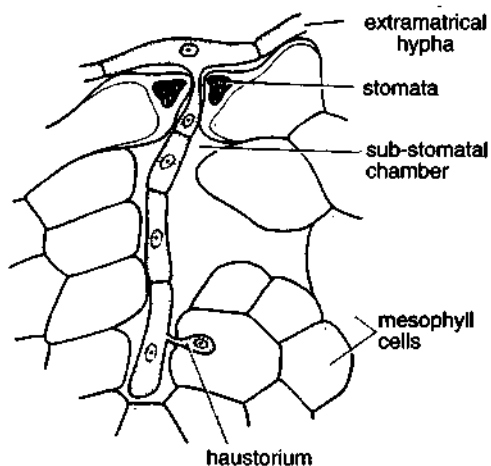


Fig. 2. *Phyllactinia*. Mycelium showing haustoria in the host cell.



Fig. 3. *Phyllactinia*. A conidiophore with conidium.

4. Each conidium is clavate, thin-walled and uninucleate.

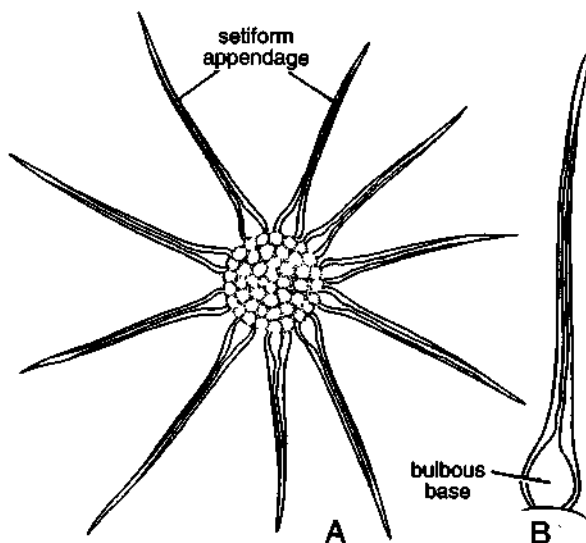


Fig. 4. *Phyllactinia*. A. Cleistothecium, B. Appendage.

5. The conidia are disseminated by wind. These germinate into a new mycelium under favourable conditions.

#### Exercise 4

**Object :** Study of ascocarp, asci and ascospores.

#### Work procedure

Prepare slide of mycelium with ascocarps. Press the cover-slip to break it open so that asci and ascospores could be studied.

#### Comments

1. The ascocarp or the fruiting body is cleistothecium provided with long, unbranched, setiform and rigid appendages.
2. The appendage has bulbous base which helps in release of ascospores after freeing the cleistothecium from hyphal mat.
3. Besides the appendages, there is an apical crown of penicillately branched hyphae over the cleistothecium. These hyphae give out a slimy substance which helps fruiting body to get attached to the host surface.
4. The asci get exposed and the ascospores are liberated, only after the rupture of the cleistothecial wall.
5. Asci are clavate, arranged more or less in parallel manner on the floor of the ascocarp.
6. Each ascus contains two ascospores. Each is ovate to elliptical and uninucleate.

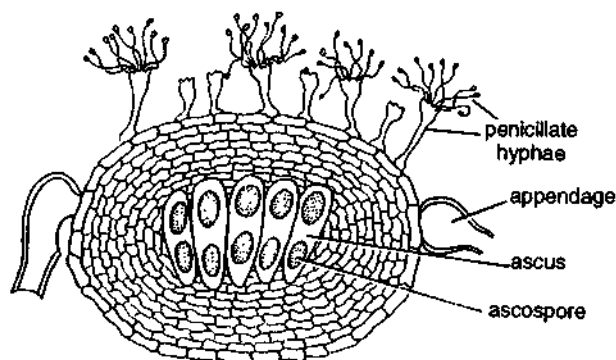


Fig. 5. *Phyllactinia*. V.s. cleistothecium.

### Identification

**Kingdom—Mycota.** (1) Chlorophyll absent, (2) Reserve food glycogen, (3) Cell wall of final cellulose.

**Division—Eumycota.** A definite cell wall present.

**Sub-division—Ascomycotina.** (1) Mycelium septate, (2) Spores borne endogenously in characteristic reproductive body—the ascus, (3) Spores in definite numbers, in multiples of two, usually eight.

**Class—Plectomycetes.** Ascocarps cleistothecium.

**Order—Erysiphales.** Ectoparasites.

**Family—Erysiphaceae.** (1) Aerial mycelium hyaline, (2) Enormous production of conidia on host surface gives it a white powdery appearance.

**Genus—Phyllactinia.** (1) Hemieudophyte, (2) Cleistothecial appendages with bulbous base, (3) Ascospores two per ascus.

### Hints for Collection

*Phyllactinia dalbergiae* is very common on the leaves of shisham and shahtoot. The leaves show white powdery mass on the lower surface.

## Aspergillus

### Classification

Kingdom	—	Mycota
Division	—	Eumycota
Sub-division	—	Ascomycotina
Class	—	Plectomycetes
Order	—	Eurotiales
Family	—	Eurotiaceae
Genus	—	<i>Aspergillus</i>

#### Exercise 1

**Object :** Study of hosts, diseases and symptoms.

#### Work procedure

A list of hosts is given below to identify and collect species of *Aspergillus*. Hosts should be preserved in F.A.A. or alcohol 90% or dried for mounting on herbarium sheets.

#### Comments

Most of the species of *Aspergillus* are saprophytes growing on decaying vegetables, butter, bread, rice, jams, leather, cloth, fabrics, etc. However, a few

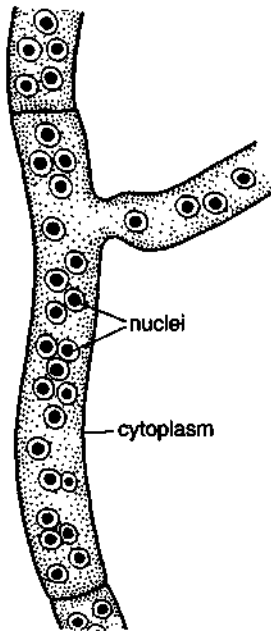


Fig. 1. *Aspergillus*. A portion of the mycelium.

species are parasites on plants and animals, including human beings.

1. *A. niger* causes rot disease of pomegranates (*Punica granatum*; vern. anar; fam. Punicaceae), dates (*Phoenix dactylifera*; vern. pind khazoor; fam. Palmae) and figs (*Ficus carica*; vern. anjir; fam. Moraceae).

In rot disease, the fruits decay, fungus enters the host through cuts and wounds in the fruits.

2. *A. fumigatus*, *A. flavus* and *A. niger* attack animals including human beings and cause a group of lung diseases collectively known as Aspergilloses.

The symptoms of Aspergilloses closely resemble to those of tuberculosis.

3. Some species of *Aspergillus* infect human ear and cause Otomycosis.

### Exercise 2

**Object :** Study the vegetative structure.

#### Work procedure

A few hyphae could be mounted in lactophenol after staining with cotton blue.

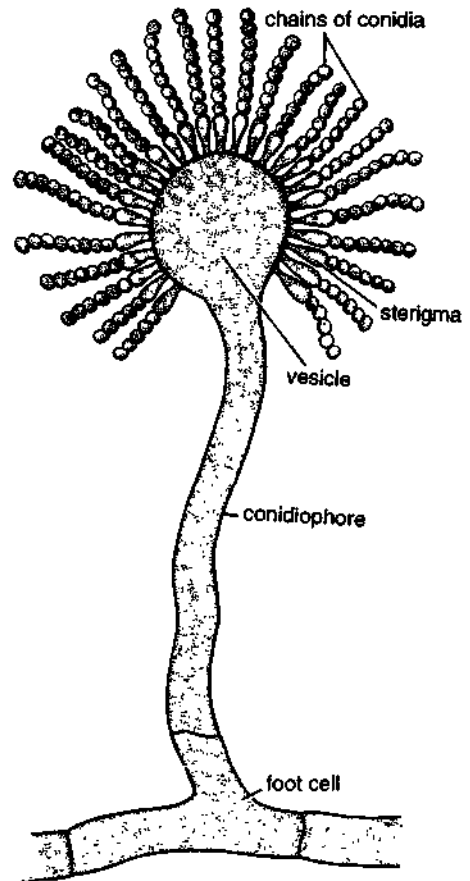


Fig. 2. *Aspergillus*. A mature conidiophore with chains of conidia.

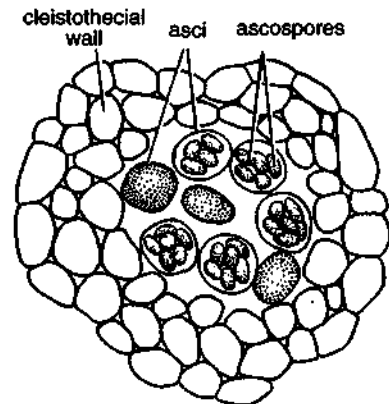


Fig. 3. *Aspergillus*. Cross section of a cleistothecium showing asci.

#### Comments

1. The mycelium is well developed, profusely branched and septate.

- The segments of the mycelium are uni- or multinucleate. The pigments in the cytoplasm give a characteristic colour to the mycelium of various species (similarly coloured conidiophores and conidia would be present in the same species).
- Some of the hyphae spread superficially over the substratum while others penetrate deep into the substratum. The latter absorb food for the mycelium.

### Exercise 3

**Object :** Study of the asexual reproductive structures.

#### Work procedure

Mount mycelium showing conidia in lactophenol after staining in cotton blue.

#### Comments

- Conidia are asexual reproductive units borne on conidiophores.
- Each conidiophore arises from the foot cell of the mycelium and is long and erect hypha, terminating in a bulbous head—the vesicle.
- The vesicle develops a number of bottle-shaped structures called the sterigmata (sing. sterigma) over its entire surface.
- In some species two layers of sterigmata one above the other, are formed. In such a case those of the lower layer are called primary sterigmata and those of the upper, secondary sterigmata. The conidia are borne only by secondary sterigmata.
- Each sterigmata cuts off a chain of basipetally arranged conidia.
- The conidia are coloured and the colour of the conidia depends upon the species.
- Each conidium is uninucleate with an outer finely spiny epispore and inner smooth endospore.
- A conidium germinates to form mycelium if it happens to fall on a suitable substratum.

### Exercise 4

**Object :** Study of ascocarp, asci and ascospores.

#### Work procedure

Mount mycelium having cleistothecia in lactophenol after staining in cotton blue.

#### Comments

- The species of *Aspergillus* developing perfect stage i.e. cleistothecium are placed under the genus *Eurotium*.
- The ascospores are produced after reduction division of ascospore mother cell in sac-like cell—the ascus.
- The asci are developed inside a fruiting body—the cleistothecium.
- Cleistothecium consists of wall called peridium formed by somatic hyphae, enclosing many asci.
- Each ascus has eight uninucleate pulley wheel-like ascospores with an outer sculptured epispore and an inner smooth endospore.
- On germination each ascospore gives rise to a haploid mycelium.

### Identification

**Kingdom—Mycota.** (1) Chlorophyll absent, (2) Reserve food glycogen, (3) Cell wall of fungal cellulose.

**Division—Eumycota.** A definite cell wall present.

**Sub-division—Ascomycotina.** (1) Mycelium septate, (2) Spores borne endogenously in the ascus, (3) Spores definite in numbers, in multiples of two, usually eight.

**Class—Plectomycetes.** Ascocarp a cleistothecium.

**Order—Eurotiales.** Cleistothecia sessile.

**Family—Eurotiaceae.** (1) Asci lie scattered, hymenium not formed, (2) Peridium made of closely interwoven hyphae.

**Genus—Aspergillus.** Conidiophore unbranched.

### Hints for Collection

*Aspergillus* can be grown on butter, bread, leather or any other similar substance in humid conditions. In rainy season, the fungus is commonly seen on shoes as black, yellow, brown or green dust.

It can also be cultivated in a laboratory from soil on suitable medium (Czapak, etc.).

***Penicillium***  
**(Blue Mold)**

**Classification**

Kingdom	—	Mycota
Division	—	Eumycota
Sub-division	—	Ascomycotina
Class	—	Plectomycetes
Order	—	Eurotiales
Family	—	Eurotiaceae
Genus	—	<i>Penicillium</i>

**Exercise 1**

**Object : Study of vegetative structure.**

**Work procedure**

Mycelium is stained with cotton blue and mounted in lactophenol.

**Comments**

1. The fungus is a saprophyte and is commonly found on citrus and other fruits, jellies and other foodstuffs.
2. The mycelium is freely branched, septate and each cell is uni- or multinucleate.
3. The mycelium may grow superficially on the surface of substratum or may penetrate deeply.
4. The hyphae are generally coloured due to pigments on the surface of hyphal walls.

**Exercise 2**

**Object : Study of conidia.**

**Work procedure**

Prepare a slide of material when it becomes green, yellow or blue coloured, for at this time it bears conidia.

**Comments**

1. The conidia are the asexual spores borne on long, erect and branched conidiophores.
2. The branched conidiophore, with its conidia looks like a small 'Penicillus' (a brush in Latin).

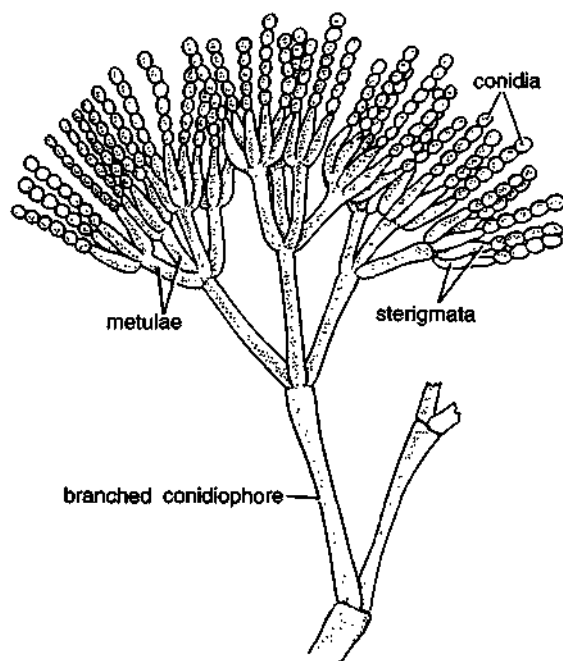


Fig. 1. *Penicillium*. Mycelium bearing conidiophore and chains of conidia.

3. Each conidiophore grows vertically from the mycelium and branches at its upper end. The ultimate branches are known as metulae.
4. Each branch of conidiophore ends in bottle-shaped sterigmata bearing a group of conidia arranged basipetally.
5. The conidia are generally blue, sometimes green or yellow and give characteristic colour to the colony.
6. The conidia are globose to ovoid in shape and appear as glass beads under the microscope.

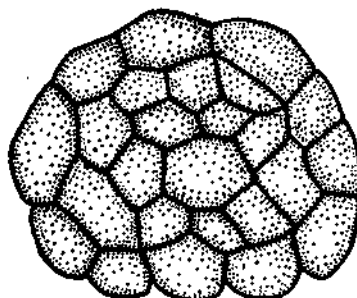


Fig. 2. *Penicillium*. A cleistothecium.

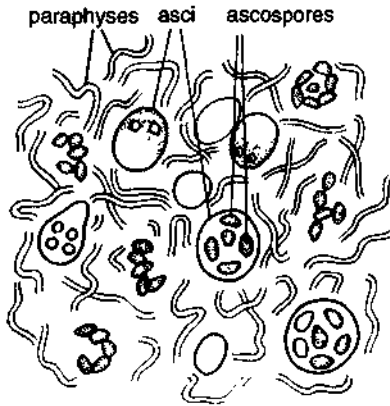


Fig. 3. *Penicillium*. A part of cleistothecium in cross section.

### Exercise 3

**Object :** Study of ascocarp, asci and ascospores.

#### Work procedure

Prepare a slide as usual and search for cleistothecia. Apply little pressure over the coverslip to break them, so that asci and ascospores come out.

#### Comments

1. The fruiting body or ascocarp is called cleistothecium. It has a wall—peridium made of sterile hyphae which encloses many asci and paraphyses.
2. The globose or pear-shaped asci lie scattered inside the cleistothecium.
3. Each ascus has eight uninucleate and wheel-shaped ascospores.
4. The ascospores are ultimately released by the rupture of cleistothecium and after falling on a suitable medium germinate into new mycelia.
5. The perfect stage i.e. cleistothecium is known as *Eupenicillium*, *Teleroomyces*, *Carpenteles*.

### Identification

**Kingdom—Mycota.** (1) Chlorophyll absent, (2) Reserve food glycogen, (3) Cell wall of fungal cellulose.

**Division —Eumycota.** A definite cell wall present.

**Sub-division—Ascomycotina.** (1) Mycelium septate, (2) Spores borne endogenously in the ascus, (3) Spores in definite numbers, in multiples of two, usually eight.

**Class—Plectomycetes.** Ascocarp, a cleistothecium.

**Order—Eurotiales.** Cleistothecia sessile.

**Family—Eurotiaceae.** (1) Asci lie scattered and hymenium not formed, (2) Peridium (outer wall of cleistothecium) of closely interwoven hyphae.

**Genus—Penicillium.** Branched, brush-like conidiophore.

### Hints for Collection

The fungus can easily be found or grown on citrus and other fruits and on foodstuff. It generally grows in association with *Aspergillus*, but since it is a weak saprophyte it is dominated by *Aspergillus*.

### Claviceps

#### Classification

Kingdom	—	Mycota
Division	—	Eumycota
Sub-division	—	Ascomycotina
Class	—	Pyrenomycetes
Order	—	Sphaeriales
Family	—	Clavicipitaceae
Genus	—	Claviceps

### Exercise 1

**Object :** Study of hosts, diseases and symptoms.

#### Work procedure

Collect as many specimen as possible out of the list given below. Preserve in alcohol 90% or dry to mount on herbarium sheets.

#### Comments

1. The fungus is an obligate parasite. *C. purpurea* causes ergot disease of rye (*Secale cereale* vern. rye; fam. Poaceae).
2. It also infects other cereals such as wheat to cause 'Ergot disease'.
3. *C. microcephala* parasitizes bulrush millet (*Pennisetum typhoidium*; vern. bajra; fam. Poaceae) and causes ergot disease. The disease is air borne.

As a result of disease the infected grain becomes

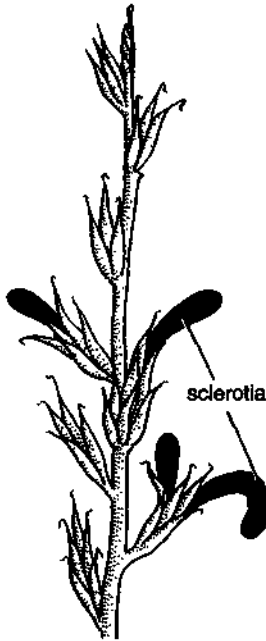


Fig. 1. *Claviceps*. An infected inflorescence of rye showing sclerotia.

very much elongated and the ovary is replaced by black compact mass of fungal tissue, the sclerotium. This sclerotium is known as Ergot in commerce.

- Whereas the mycelium causes disease in plants, the sclerotia, if eaten by animals including human beings, cause a disease known as Ergotism. It causes poisoning and ultimately the death.

### Exercise 2

**Object :** Study of vegetative structure.

#### Work procedure

Crush the rye grain and study the mycelium.

#### Comments

- Ascospores or conidiospores germinate on stigma, produce a germ tube that reaches the ovules and later develop mycelium in the grain.
- The mycelium is white, branched, septate and the cells are multinucleate.

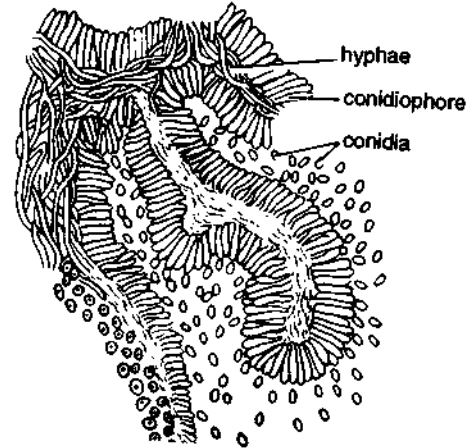


Fig. 2. *Claviceps*. Mycelium showing conidiophores.

### Exercise 3

**Object :** Study of conidia.

#### Work procedure

Crush the rye grain and locate conidiophores. Stain in cotton blue and mount in lactophenol.

#### Comments

- The conidia are formed inside the grain at the tips of short conidiophores which form acervulus-like layers.
- The conidia are minute, ovoid and uninucleate. These are budded off by each conidiophore.

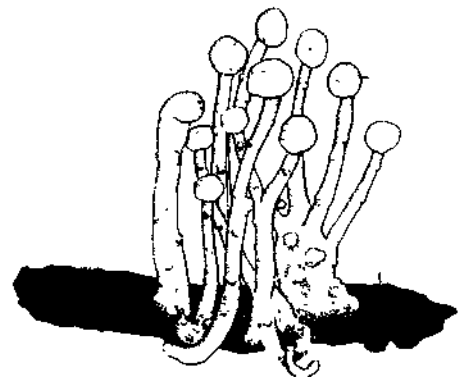


Fig. 3. *Claviceps*. Germinating sclerotium.

- The conidia are mixed with a sticky, sweet, nectar-like secretion, known as honey dew. Insects get attached by honeydew and serve to spread the disease.
- Conidial stage is also known as *Sphacelia* stage being earlier described under the species *Sphacelia segetum*.

#### Exercise 4

**Object :** Study of sclerotia, stromata, asci and ascospores.

#### Work procedure

Study the sclerotia, the V.s. of stromata and an ascocarp.

#### Comments

- The sclerotia are black and elongated structures which replace the grains in the inflorescence.
- After winter, the black sclerotia germinate and form many long, stalked, mushroom-like and dark purple stromata with globose heads.
- The stromata are nearly 1 cm in length.
- Each stromatal head, just below its surface, has many minute flask-shaped cavities, known as perithecia, surrounded by the pseudoparenchymatous stromatic tissue.
- Each mature perithecium is a flask-shaped cavity opening at the apex through a small pore—an ostiole.
- Near the ostiole are present some hair-like structures known as paraphyses.

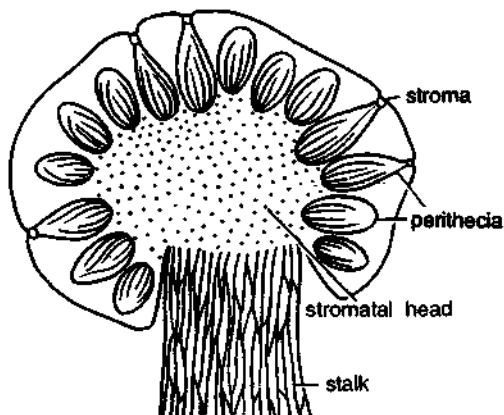


Fig. 4. *Claviceps*. V.s. stromata.

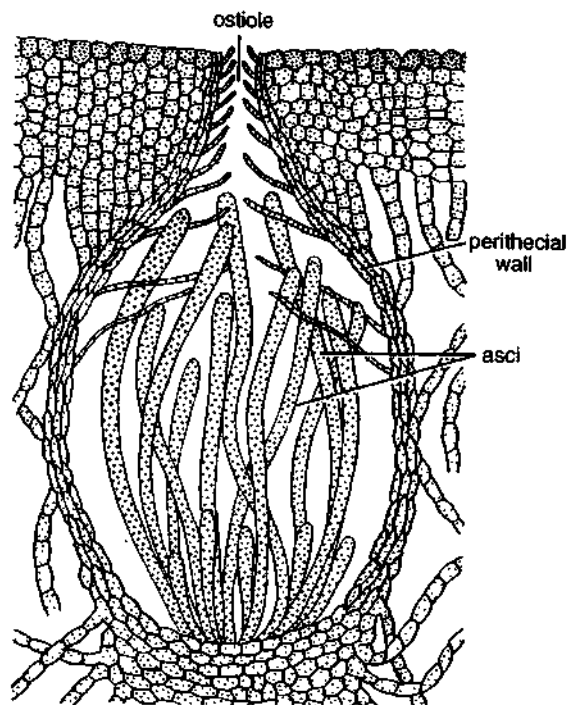


Fig. 5. *Claviceps*. V.s. ascocarp.

- The asci are produced in tufts from the base of the perithecium.
- An ascus is narrow, elongated and somewhat curved. It has a thick cap at apex through which the ascospores are discharged.
- Each ascus produces eight haploid, uninucleate elongated and acicular ascospores that lie parallel to one another.

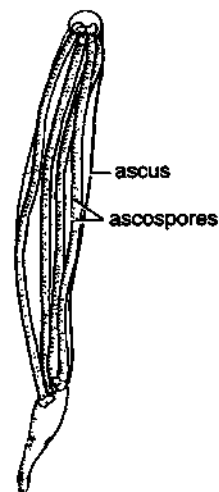


Fig. 6. *Claviceps*. Ascus showing ascospores.



## Identification

**Kingdom—Mycota.** (1) Chlorophyll absent, (2) Reserve food glycogen, (3) Cell wall of fungal cellulose.

**Division—Eumycota.** A definite cell wall present.

**Sub-division—Ascomycotina.** (1) Mycelium septate, (2) Spores borne endogenously in ascus, (3) Spores in definite numbers, in multiples of two, usually eight.

**Class—Pyrenomycetes.** Ascocarp, a perithecium.

**Order—Sphaeriales.** (1) Ascospores thread-like, (2) Perithecia within a well-developed stroma.

**Family—Clavicipitaceae.** Perforated cap at the apex of the ascus.

**Genus—Claviceps.** (1) Sclerotia black & well marked, (2) Ascospores lie parallel in ascus.

## Hints for Collection

The fungus can be collected from the cereals (especially rye), growing in the crop fields cultivated for their edible grains.

### Peziza

## Classification

Kingdom	—	Mycota
Division	—	Eumycota
Sub-division	—	Ascomycotina
Class	—	Discomycetes
Order	—	Pezizales
Family	—	Pezizaceae
Genus	—	Peziza

### Exercise 1

**Object :** Study of vegetative structure.

#### Work procedure

Take apothecium, tear off a part to study mycelium.

#### Comments

1. It is a common saprophyte growing on rich humus soils and decaying woods. Sometimes it becomes coprophilous (i.e. grows on dung).
2. The mycelium is a complex system that penetrates the substratum.
3. The mycelium is profusely branched, septate and the cells are multinucleate.
4. The mycelium becomes visible only in the form of apothecial cups above the ground surface.

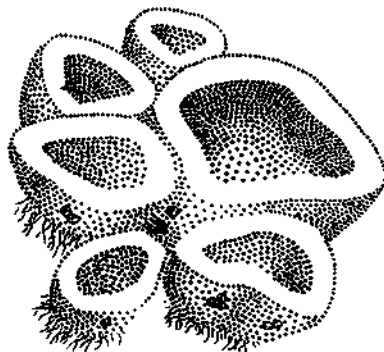


Fig. 1. *Peziza*. Ascocarps growing on decaying wood.

### Exercise 2

**Object :** Study the conidia/chlamydo spores.

#### Work procedure

Study the mycelium for the presence of conidia/chlamydo spores.

#### Comments

1. The conidia are the asexual reproductive bodies. These are produced rarely.

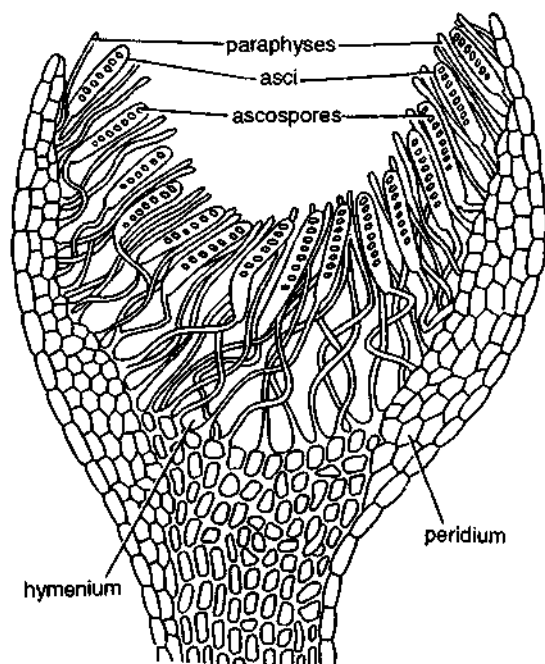


Fig. 2. *Peziza* V.s. ascocarp.

2. Conidia are hyaline, to lightly coloured and elliptical.
3. In some species, thick walled and intercalary chlamydospores are produced singly or in series on mycelium.
4. On germination chlamydospore produces a new mycelium.

### Exercise 3

**Object :** Study of ascocarp, asci and ascospores.

#### Work procedure

Cut a V.s. of apothecial cup, stain with cotton blue, mount in lactophenol and study.

#### Comments

1. The ascocarp is an apothecium. It is fleshy, shortly stalked, about 5 cm in diameter with a bright red or bright grey lining.
2. A vertical section of ascocarp shows a cup-shaped structure made up of mycelium. It shows 3 regions—hymenium, hypothecium and excipulum.
3. Hymenium consists of asci and paraphyses arranged vertically in organe-red-coloured palisade-like layer.
4. The hypothecium consists of thin and lightly coloured hyphae that runs parallel to hymenium.
5. Excipulum forms a basal large part of loosely interwoven hyphae of apothecium.

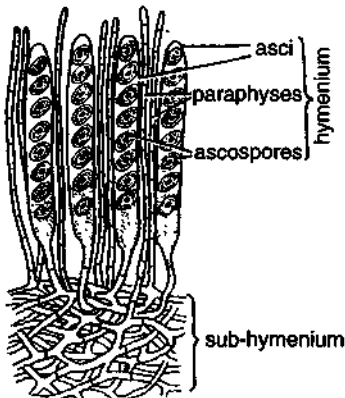


Fig. 3. *Peziza*. V.s ascocarp (portion magnified).

6. The hymenium is encircled by densely interwoven hyphae forming the wall of the apothecium—the peridium.
7. Ascus is elongated with a single row of eight ascospores, arranged obliquely.
8. Each ascospore is uninucleate, hyaline or faintly coloured, elliptical, surface smooth or coarsely reticulate and ellipsoidal. It germinates to form new mycelium.

### Identification

**Kingdom—Mycota.** (1) Chlorophyll absent, (2) Reserve food glycogen, (3) Cell wall of fungal cellulose.

**Division—Eumycota.** A definite cell wall present.

**Sub-division—Ascomycotina.** (1) Mycelium septate, (2) Spores borne endogenously in ascus, (3) Spores in definite numbers, in multiples of two, usually eight.

**Class—Discomycetes.** Ascocarp, an apothecium.

**Order—Pezizales.** (1) Apothecia fleshy or leathery, (2) Apothecia usually not in stroma.

**Family—Pezizaceae.** (1) Apothecia cup-shaped or discoid, (2) apothecia not differentiated into stipe and pileus.

**Genus—Peziza** (1) Apothecium 1 to 5 cm in diameter and conspicuous, (2) Vegetative mycelium penetrating the substratum, (3) Ascospores not apiculate.

### Hints for Collection

Apothecia are seen growing on dung and on decaying wood, in damp places in rainy season.

### *Ascobolus*

#### Classification

Kingdom	—	Mycota
Division	—	Eumycota
Sub-division	—	Ascomycotina
Class	—	Discomycetes
Order	—	Pezizales
Family	—	Pezizaceae
Genus	—	<i>Ascobolus</i>

### Exercise 1

**Object :** Study of the vegetative structure.

#### Work procedure

Pinch a small part of the ascocarp or cut a section of the sterile part of the ascocarp. Stain with cotton blue, mount in lactophenol and study.

### Comments

1. The fungus mostly grows on the dung of herbivores and is called coprophilous. A few species (*A. carbonarius*) grow on burnt soils.
2. The thallus is made of richly branched mycelium that forms a complex structure and finally a cup shaped structure.
3. The hyphal masses penetrate the substratum. These act as organs of absorption for the aerial branches.
4. The hyphae are branched and septate. Each cell is multinucleate.

### Exercise 2

**Object :** Study the sexual reproductive structures.

### Work procedure

The mycelium shows sex organs just before apothecia appear fully formed. The mycelium at this stage be stained with cotton blue and mounted in lactophenol and sex organs are searched.

### Comments

1. The male reproductive organs are antheridia and the female reproductive organs are called ascogonia.
2. Antheridia and ascogonia are borne at the tips of separate branches.
3. Antheridium is borne at the tip of antheridial branch. It is cylindrical or clavate in shape.
4. Each antheridium is multinucleate.
5. Ascogonium is also present at the tip of the ascogonial branch. It is sub-globose in shape and is multinucleate.

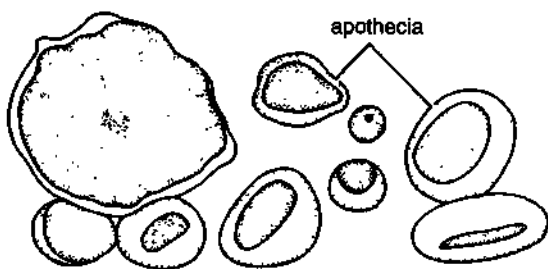


Fig. 1. *Ascobolus*. Ascocarps.

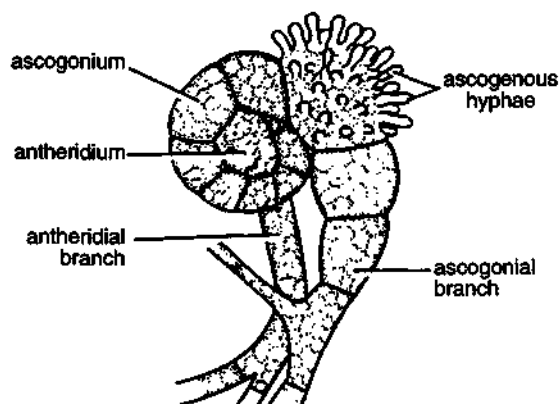


Fig. 2. *Ascobolus*. Sex organs.

6. The tip of ascogonium is drawn into a long and terminal organ called trichogyne. It reaches the tip of an antheridium.

### Exercise 3

**Object :** Study the ascocarps, asci and ascospores.

### Work procedure

Cut a V.s. of apothecium, stain in cotton blue, mount in lactophenol and study.

### Comments

1. The ascocarps are apothecia and develop as a result of fertilization.
2. These are yellowish and saucer shaped.
3. Apothecium is a cup-shaped structure that is made of mycelium.
4. The section shows three zones—the outermost called hymenium, the middle called sub-hymenium and the lowermost called hypothecium.
5. The lowermost hypothecium, is made of sterile hyphae, loosely packed to form pseudoparenchymatous region called trama.
6. The middle zone consist of a few erect hyphae. This later merges with hymenial layer which is called sub-hymenium.
7. The hymenium consists of asci intermingled with paraphyses.

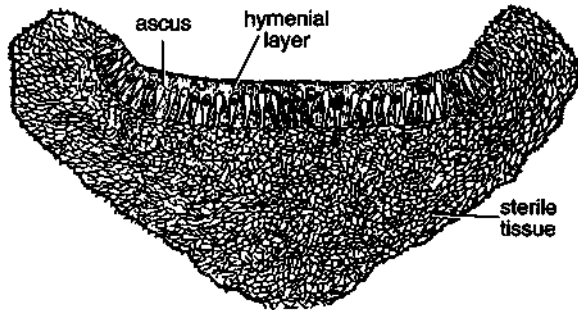


Fig. 3. *Ascobolus*. V.s. apothecium.

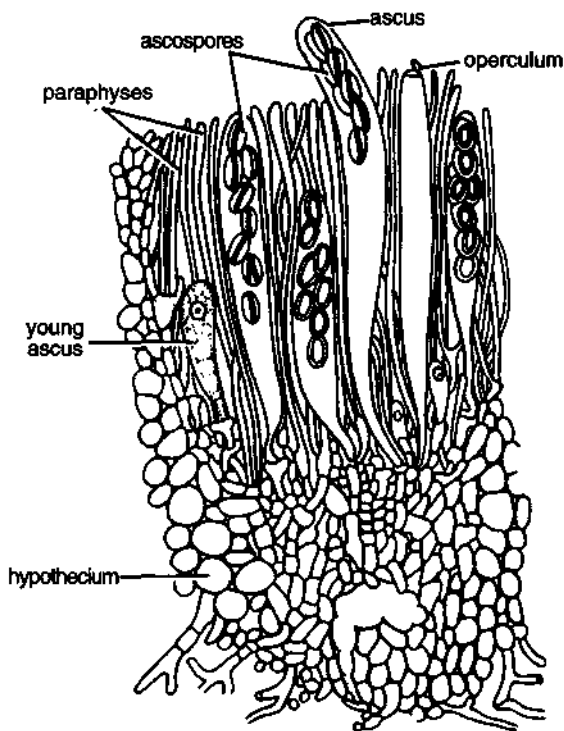


Fig. 4. *Ascobolus*. V.s. apothecium—a part enlarged.

8. The asci elongate on maturation to protrude above the hymenial surface. These are found mixed with paraphyses. Ascus is a long and cylindrical structure. It opens by a terminal pore called operculum.
9. Each ascus contains eight ascospores. Ascospore is one celled, large in diameter,

purple or dark brown and the spore wall shows longitudinal colourless striations.

### Identification

- Kingdom—Mycota.** (1) Chlorophyll absent, (2) Reserve food glycogen, (3) Cell wall of fungal cellulose.  
**Division—Eumycota.** A definite cell wall present.  
**Sub-division—Ascomycotina.** (1) Mycelium septate, (2) Spores borne endogenously in the ascus, (3) spores in definite numbers, in multiples of two, usually eight.  
**Class—Discomycetes.** Ascocarp an apothecium.  
**Order—Pezizales.** (1) Apothecia fleshy or leathery, (2) Apothecia usually not in stroma.  
**Family—Pezizaceae.** (1) Apothecia cup-shaped or discoid, (2) Apothecia not differentiated into stipe and pileus.  
**Genus—*Ascobolus*.** (1) Apothecia upto 5 mm in diameter, (2) Apothecia saucer-shaped and growing on dung.

### Hints for Collection

Apothecia could be collected from the decaying dung of herbivores. These are also common on burnt soils.

## *Morchella* (Morel)

### Classification

Kingdom	—	Mycota
Division	—	Eumycota
Sub-division	—	Ascomycotina
Class	—	Discomycetes
Order	—	Pezizales
Family	—	Helvellaceae
Genus	—	<i>Morchella</i>

### Exercise 1

**Object :** Study the vegetative structure.

### Work procedure

Pinch off a small piece of fungus or cut a section. Stain in cotton blue, mount in lactophenol and study.

### Comments

1. All the species are saprophytes and grow in deciduous forests on decaying wood or in humus soil. These are commonly known as morels.

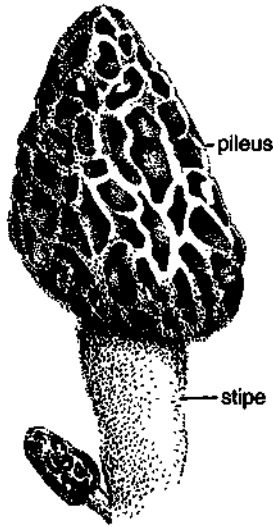


Fig. 2. *Morchella*. An ascocarp.

2. The mycelium is underground, growing a few inches deep in the soil.
3. Mycelium is freely branched and the hyphae are septate. Each cell has many nuclei.
4. The mycelium is woven to form a stalk and a conical pileus at the tip.
5. The pileus is ridged and grooved. The grooves develop ascocarps.

### Exercise 2

**Object :** Study of ascocarp, asci and ascospores.

### Work procedure

Cut a L.s. through a groove that represents an apothecium, stain with cotton blue, mount in lactophenol and study.

### Comments

1. The ascocarp is an apothecium, varying from 1 to 5 inches in height.
2. The colour of the apothecium varies from greyish white to dark brown depending on the species and age.
3. The mature ascocarp consists of a stalk, known as stipe, surmounted by a hollow conical cap called the pileus which is the fertile portion of

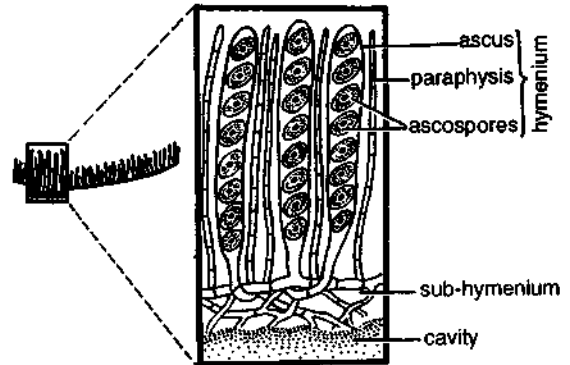


Fig. 3. *Morchella*. A portion of a section through depression of pileus.

- the ascocarp. The stipe is cream coloured, thick, fleshy and hollow.
4. The surface of the pileus is thrown into many strong longitudinal and transverse folds, so that it becomes coarsely pitted (i.e. with ridges and grooves).
5. The ridges are the sterile areas whereas depressions are the fertile areas.
6. A section of the pileus through the depression shows the hymenium with asci and paraphyses, the latter being sterile structures.
7. Below the hymenium, the mycelium forms a close interwoven structure known as sub-hymenium.
8. The asci and paraphyses in the hymenium are arranged perpendicular to the surface of depression and form a palisade-like layer.
9. Each ascus is a long and cylindrical structure containing eight ascospores arranged obliquely and uniseriately.
10. The ascospores are large, hyaline, oval and arranged in a single row.

### Identification

**Kingdom—Mycota.** (1) Chlorophyll absent, (2) Reserve food glycogen, (3) Cell wall of fungal cellulose.

**Division—Eumycota.** A definite cell wall present.

**Sub-division—Ascomycotina.** (1) Mycelium septate, (2) Spores borne endogenously in ascus, (3) Spores in definite numbers, in multiples of two, usually eight.

**Class—Discomycetes.** Ascocarp, an apothecium.

**Order—Pezizales.** (1) Apothecia fleshy or leathery, (2) Apothecia usually not in stroma.

**Family—*Helvellaceae*.** (1) Apothecia not cup shaped, (2) Apothecia differentiated into stipe and pileus.

**Genus—*Morchella*.** (1) Pileus costate, (2) Pileus thrown into many ridges and grooves, (3) Sterile stipe and fertile pileus distinct.

### Hints for Collection

The fungal ascocarps can commonly be found in deciduous forests either in humus soil or on decaying wood, etc. In India, it is abundantly found in Kashmir and Kumaon hills. This is an edible fungus and is grown commercially.

#### Sub-division 4.

### BASIDIOMYCOTINA (BASIDIOMYCETES)

This large class comprises about 25,000 species of great variety of form and structure. The unifying character, of this class, is their possession of a special reproductive structure—the basidium which bears at its tip usually four exogenously formed basidiospores. The basidiomycetes consists of forms called as mushrooms, toadstools, puffballs, stinkhorns, earthstars, bird's nest fungi, rust, smuts, jelly fungi and shelf or bracket fungi, etc.

The heterotrophism in basidiomycetes varies from obligate saprophytism to obligate parasitism with intermediate facultatism. The fleshy fungi are mainly saprophytes, the rusts are obligate parasites and the bunts and smuts, facultative saprophytes.

The mycelium is freely branched, typically septate and frequently perennial in the substratum. In higher basidiomycetes, the mycelium living in the soil sometimes form fairy rings, sclerotia or rhizomorphs. The mycelium of mushrooms, puffballs shelf fungi, etc., forms appreciable fruiting bodies—the basidiocarps. The mycelium (dikaryotic mycelium) of higher basidiomycetes is characterized by clamp connections.

Asexual reproduction is common by conidia (uredospores) and oidia.

Sexual reproduction is very much reduced. The basidia are formed on dikaryotic mycelium. Some of these are vertically or transversely septate, or have deeply incised apex, and are known as phragmobasidia and the basidia that are more or less cylindrical, have a rounded apex and are without septa, are known as holobasidia. The two nuclei of opposite strains fuse in young basidium forming a diploid nucleus. The basidium generally develops four outgrowths at its apex. Meanwhile, the diploid nucleus in basidium undergoes reduction division and forms four haploid nuclei which ultimately migrate into the outgrowths, now known as basidiospores. The basidiospore is uninucleate and on germination gives rise to monokaryotic mycelium. Basidiomycotina is further divided into 3 classes—(1) Teliomycetes, (2) Hymenomycetes and (3) Gasteromycetes.

### *Ustilago* (Smuts)

#### Classification

Kingdom	—	Mycota
Division	—	Eumycota
Sub-division	—	Basidiomycotina
Class	—	Teliomycetes
Order	—	Ustilaginales
Family	—	Ustilaginaceae
Genus	—	<i>Ustilago</i>

#### Exercise 1

**Object :** Study of hosts, diseases and the symptoms

#### Work procedure

Collect the infected plants. Preserve them in F.A.A. or dry the specimen and mount them on herbarium sheets.

#### Comments

Various species of *Ustilago* are parasitic in their natural habitat but many of them grow as saprophytes also.

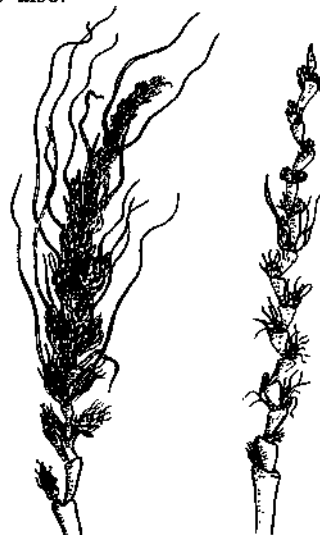


Fig. 1. *Ustilago*. Loose smut of wheat. Wheat ears infected by *U. tritici*.

Most of the species infect members of the grass family (Poaceae) and cause enormous loss. The cereal smuts are classified into loose smut and covered smut. Both are seed borne types.

- (i) In loose smut, the spores occur in the grains which are exposed only on maturity of the ear and are easily blown away by wind (infection air borne).
- (ii) In the covered smut, spore masses remain covered by the wall of grain and glumes (not exposed). These are liberated when wall is ruptured under pressure (infection seed borne).

Only the inflorescences are attacked in all the cases. The disease, in general, caused by species of *Ustilago*, on various hosts, is known as smut disease because of production of black dusty mass of spores. The following are common hosts and diseases.

1. *U. tritici* infects wheat (*Triticum vulgare*; vern. gehun) and causes loose smut of wheat.
2. *U. nuda* and *U. hordei* infect barley (*Hordeum vulgare*; vern. jau) and cause loose smut and covered smut of barley, respectively.
3. *U. avenae* and *U. kolleri* infect oat (*Avena sativa*; vern. jaii) and cause loose smut and covered smut respectively.

In covered and loose smuts of various cereals, the symptoms are seen only at the time of flowering and not before. The grains in the inflorescence of the hosts are replaced by black and sooty mass of teliospores. The infected plants produce deformed spikelets. As the grain is directly affected, the yield of the crop is considerably reduced.

4. *U. scitaminea* attacks sugarcane (*Saccharum officinarum*; vern. ganna) causing whip smut of sugarcane.

The whip smut of sugarcane causes the floral axis of infected plant to become long and whip-like, covered by black sooty spores.

5. *U. maydis* attacks maize (*Zea mays*; vern. makka) and results into a disease called, common smut of maize.

In common smut of maize, large galls are produced on the cob which are sometimes as large as man's fist. These galls partly consist of host tissue and partly of fungus tissue.

6. *U. cynodontis* infects grass (*Cynodon dactylon*; vern. doob ghas) and causes loose smut of grass.

(B-14)

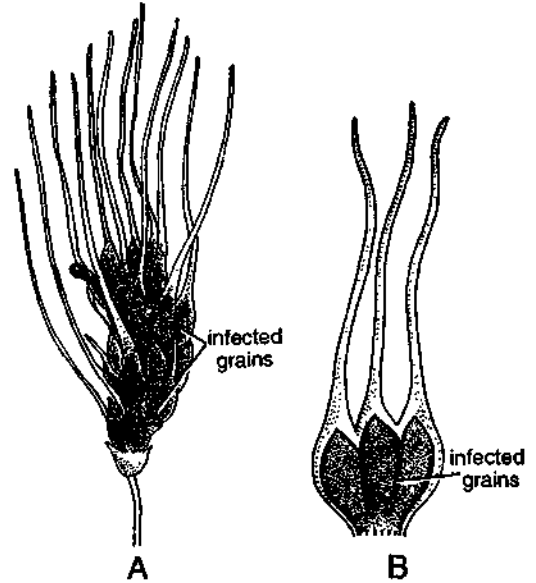


Fig. 2. *Ustilago*. A. Covered smut of barley. Spike of barley infected by *U. hordei*, B. Spikelet magnified.

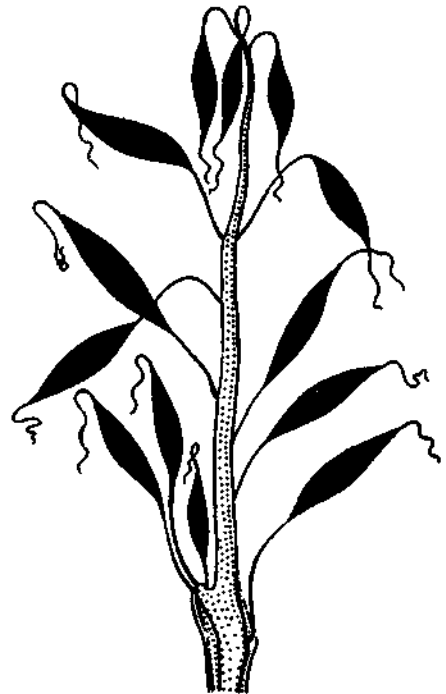


Fig. 3. *Ustilago*. Covered smut of oat. Oat inflorescence infected by *U. kolleri*.

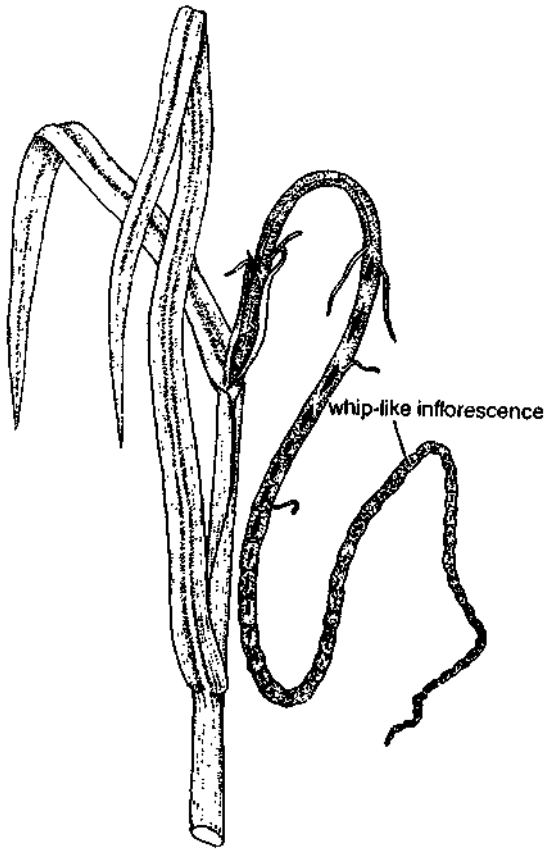


Fig. 4. *Ustilago*. Whip smut of sugarcane caused by *U. scitaminae*.

7. Other species of *Ustilago* infecting economically useful plants include *U. panicifrutentacei* and *U. paradoxa* on barnyard millet (*Echinochloa frumentacea*), *U. crameri* on Italian millet (*Setaria italica*), and *U. coicis* and *U. lacrymaejobi* on job's tear millet (*Coix lacryma-jobi*).
8. *U. violacea* causes another smut of Caryophyllaceae.

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### Exercise 2

Object : Study of vegetative structure.

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#### Work procedure

Break open or crush the spikelet when young. Search for the mycelium, if seen, stain with cotton blue and mount in lactophenol.

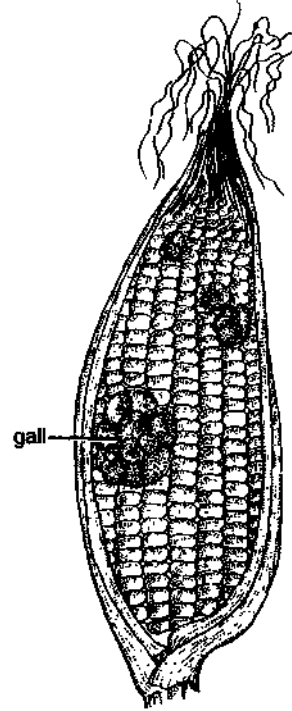


Fig. 5. *Ustilago*. Common smut of maize—Maize cob infected by *U. maydis*.

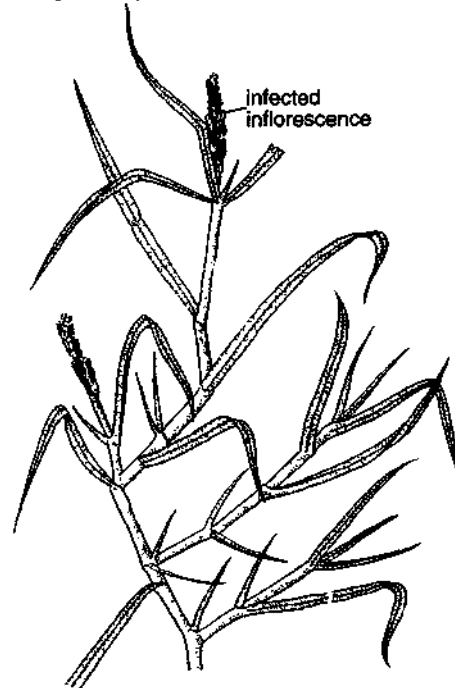


Fig. 6. *Ustilago*. Grass spike infected by *U. cynodontis*.



### Comments

1. The mycelium is well developed. It is generally intercellular without haustoria but sometimes, it is intracellular also.
2. The mycelium is branched and septate and can be seen only in younger portions.
3. The mycelium can be monokaryotic when it has only one nucleus in each cell. It also becomes dikaryotic when it possesses two nuclei of different strains in each cell.

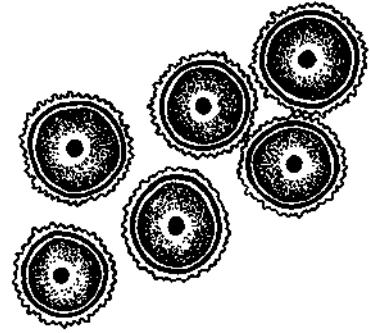


Fig. 7. *Ustilago*. Chlamydospores.

### Exercise 3

**Object :** Study of chlamydospores or teleutospores.

### Work procedure

Tap the infected inflorescence or seed on the slide. The spores would be shed. These could be mounted in glycerine without staining.

### Comments

1. The chlamydospores are formed in the grains of the host by repeated partition of the mycelium.
2. The mature chlamydospores are black soot-like in colour.
3. Each chlamydospore (teliospores) at maturity is unicellular, uninucleate, diploid and globose. The wall is thick with exospore and endospore. Exospore is thick and spiny while endospore is thin and smooth.
4. It germinates to form four basidiospores which in their turn produce the monokaryotic (primary) mycelium.

### Identification

**Kingdom—Mycota.** (1) Chlorophyll absent, (2) Reserve food glycogen, (3) Cell wall of fungal cellulose.

**Division—Eumycota.** A definite cell wall present.

**Sub-division—Basidiomycotina.** (1) Mycelium septate, (2) Characteristic reproductive body, basidium, (3) Basidiospores usually four, produced exogenously.

**Class—Teliomycetes.** (1) Basidiocarp lacking, (2) Teliospores or chlamydospores in sori or scattered, (3) Parasitic on vascular plants.

**Order—Ustilaginales.** (1) Hyphae inter- as well as intracellular. (2) Teleutospores (teliospores) mostly intercalary, basidiospores not on sterigmata.

**Family—Ustilaginaceae.** Chlamydospores formed in the host tissue from the hyphal cells.

**Genus—Ustilago.** (1) Chlamydospores separate and not adhering in pairs, (2) Sori dusty at maturity.

### Hints for Collection

The fungus parasitizes mostly the cultivated grasses as wheat, barley, oat, sugarcane, etc. grown in crop fields. *Cynodon dactylon*, another host, occurs wild in shades, also as a weed on boundaries of fields and is very common grass in lawns.

### Puccinia (Rusts)

Species of *Puccinia* are known as Rusts, because the infected parts look like rusted iron. Rusts are known from very ancient times, because of the enormous loss caused by them to the crop. In ancient Rome, cereal rust diseases were thought to be caused by two Gods, Robigus and Robigo. To please these Gods, the ancient Romans used to annually celebrate a festival, Robigalia.

### Classification

Kingdom	—	Mycota
Division	—	Eumycota
Sub-division	—	Basidiomycotina
Class	—	Teliomycetes
Order	—	Uredinales
Family	—	Pucciniaceae
Genus	—	<i>Puccinia</i>

### Exercise I

**Object :** Study of hosts, diseases and the symptoms.

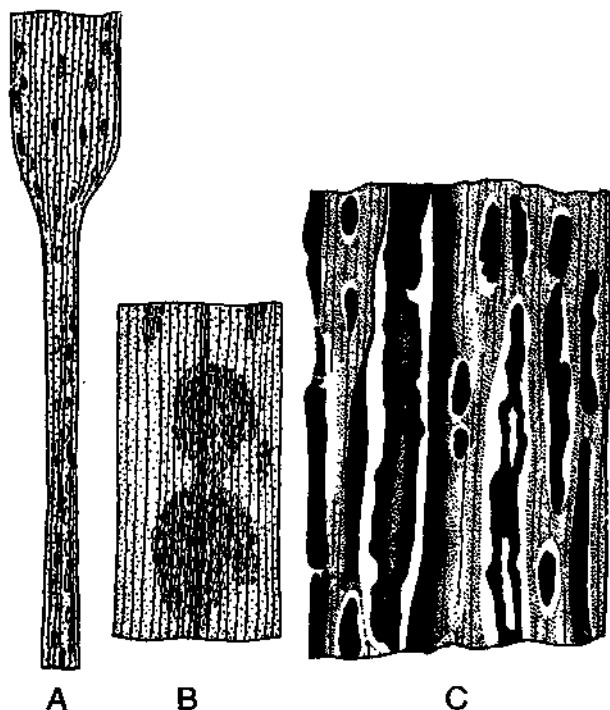
#### Work procedure

Numerous species of *Puccinia* occur on different hosts. A list is given below so that specimen could be collected and preserved in alcohol or as dried herbarium sheets.

#### Comments

All the species of *Puccinia* are obligate parasites on some of the important cereals (fam. Graminae or Poaceae) viz. wheat, maize and oat, on millets as bajra and jowar, and on other plants as *Berberis* and *Thalictrum*, etc.

All the species of *Puccinia* are polymorphic. Some species such as *P. graminis* are heteroecious (i.e. they complete their life cycle on two different hosts), while others such as *P. Butleri* are autoecious

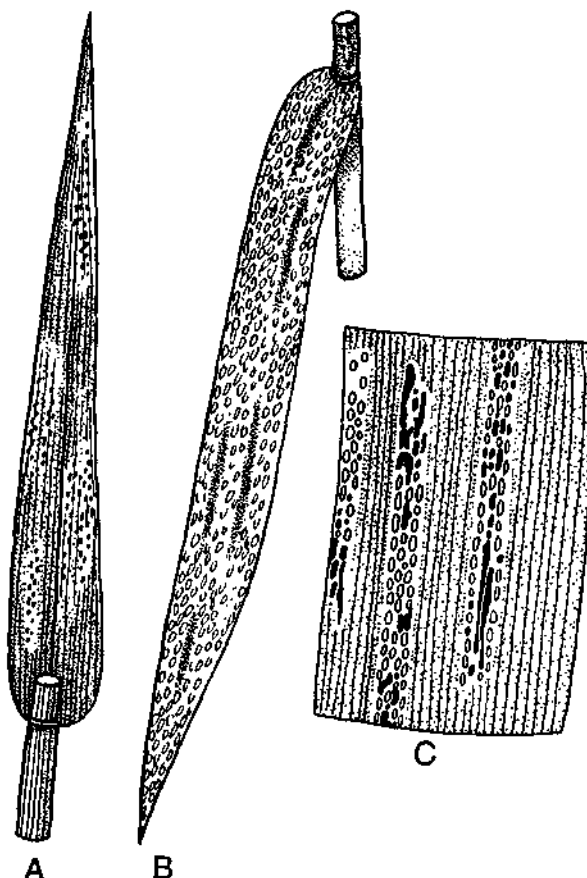


**Fig. 1.** *Puccinia graminis tritici*. Black or stem Rust. A. Leaf sheath showing pustules, B. A portion of leaf showing uredo-pustules, C. A portion of stem showing teleutopustules.

(i.e. those which complete their life cycle on one host only). In almost all the heteroecious rusts, uredo- and teleutostages are found on primary host while pycnidial and aecidial stages are found on alternate host. Some of the hosts, their causal organisms and diseases are as follow:

1. *P. graminis tritici* and *P. striiformis* (= *P. glumarum*) infect wheat (*Triticum* sp., vern. gehun), the primary host and barberry (*Berberis* sp.; fam. Berberidaceae), the alternate host. They cause Black rust or Stem rust and Yellow or Stripe rust respectively on wheat.

The symptoms of the disease are seen on leaves, leaf sheaths and sometimes on stem also. The floral organs are generally not affected (cf. *Ustilago*). In Black or Stem rust, (*P. graminis tritici*) dark brown or black, oblong to linear



**Fig. 2.** *Puccinia striiformis*. Yellow or Stripe Rust. A. and B. Leaves showing pustules in the form of streaks C. A portion of leaf sheath showing uredo- and teleutopustules.

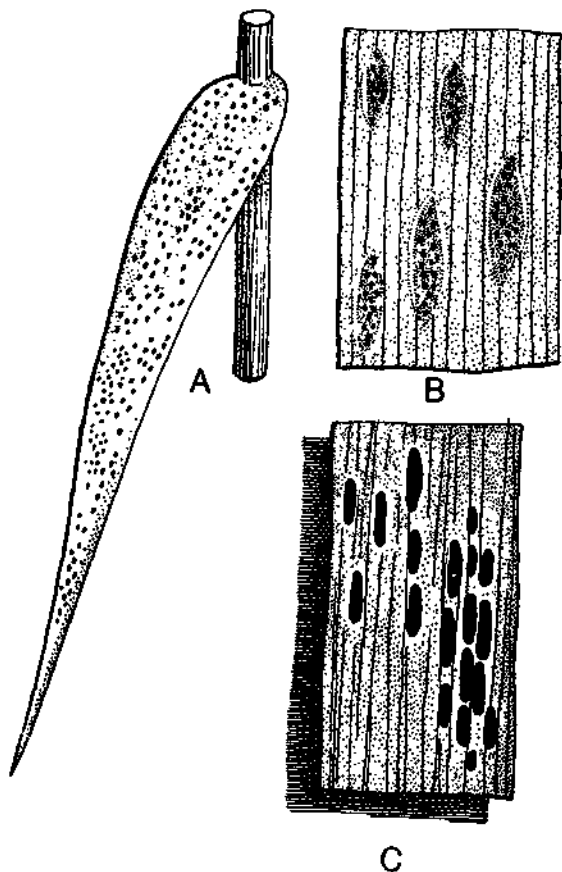


Fig. 3. *Puccinia recondita*. Orange Leaf Rust. A. Leaf showing pustules, B. A portion of leaf showing uredopustules, C. A portion of leaf showing telutopustules.

lesions are produced on leaves, leaf sheaths and stems which in case of severe infection, coalesce to form large patches.

Yellow or stripe rust (*P. striiformis* = *P. glumarum*) is chiefly confined to leaves but if the attack is severe it may also spread to leaf sheaths and stalks. As such the green colour of leaves fades, producing long streaks on which small oval and lemon yellow lesions are found.

2. *P. recondita* (= *P. trititina*) infects wheat, the primary host and *Thalictrum* sp. (fam. Ranunculaceae), the alternate host and causes Orange leaf rust of wheat.

It shows round to slightly oblong, orange coloured irregularly scattered pustules or lesions or form clusters on the leaf blades. They are never found in rows or stripes. The alternate

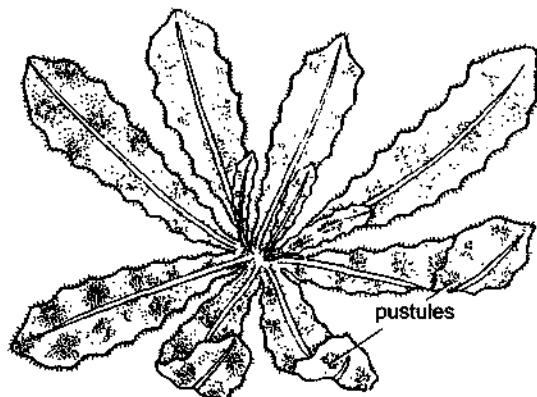


Fig. 4. *Puccinia butleri*. Infection on *Launea* sp.

host, *Thalictrum*, shows small brown specks in clusters on leaves.

3. *P. purpurea* parasitizes *Sorghum* (vern. jowar; fam. Graminae or Poaceae), the primary host and *Oxalis* (vern. khatti buti; fam. Oxalidaceae), the alternate host. The disease is known as Leaf rust.
4. *P. sacchari* causes Leaf rust on sugarcane (*Saccharum officinarum*; vern. ganna; fam. Graminae or Poaceae).
5. *P. butleri*, an autoecious rust, attacks *Launea* sp. (fam. Compositae or Asteraceae) and causes Leaf rust. In case of other Leaf rusts orange to black pustules are seen on leaves and sometimes also on leaf sheaths.
6. *P. penniseti* infects *Pennisetum typhoideum* (vern. bajra; fam. Graminae), the primary host and *Solanum melongena* (vern. baingan; fam. Solanaceae), the alternate host.
7. *P. coronata* parasitizes oat (*Avena sativa*; vern. jaii; fam. Graminae or Poaceae), the primary host, causing Crown rust and *Rhamnus* (fam. Rhamnaceae), the alternate host.

### Exercise 2

**Object :** Study of vegetative structure.

### Work procedure

To study the mycelial structure, section of a very young wheat leaf is cut. If it shows inter- or intracellular hyphae, it is stained in cotton blue, mounted in lactophenol and studied.

### Comments

1. The mycelium is well developed, branched and septate. It is generally intercellular and sometimes shows globular haustoria also.
2. The mycelium is called dikaryotic because it possesses two nuclei of different stains in each cell.  
(The description mostly applies to heteroecious rusts. The uredosori and teliosori are developed on primary hosts).

### Exercise 3

**Object : Study of uredosorus and uredospores.**

### Work procedure

These are found on wheat leaf. A section is cut through the pustule, stained in cotton blue and mounted in lactophenol or stained in safranin-fast green combination for permanent mounting.

### Comments

1. The uredosori or uredopustules appear as red, oval or lemon shaped lesions on the leaves and leaf sheaths.
2. The uredosorus in section reveals the ruptured host epidermis due to the pressure of underlying uredospores.
3. The (dikaryotic) intercellular and branched mycelium is aggregated beneath the epidermis.

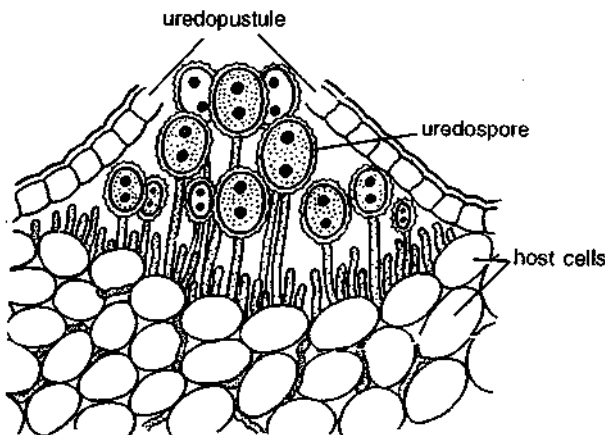


Fig. 5. *Puccinia*. T.s. of wheat leaf through uretopustule.

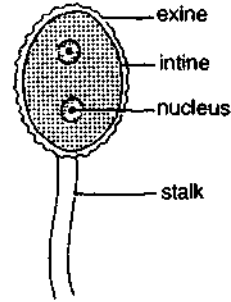


Fig. 6. *Puccinia*. Uredospore.

4. The uredospores are produced in massive groups from this mycelium.
5. Each uredospore is binucleate, stalked and rounded or oblong in shape.
6. It has an outer exine which is finely verrucose or echinulate and an inner smooth intine.
7. Each uredospore has four equatorial germ pores.
8. The uredospores get disseminated by wind and infect the fresh wheat plants.

### Exercise 4

**Object : Study of teliosorus and teliospores.**

### Work procedure

These are found on wheat leaves. A T.s. of the leaf showing teliosorus could be stained in cotton blue and mounted in lactophenol for temporary preparation. Safranin-fast green combination is used if permanent preparation is to be made.

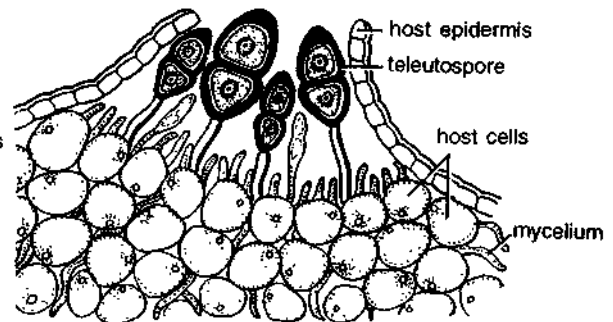


Fig. 7. *Puccinia*. T.s. of wheat leaf through teliosorus.

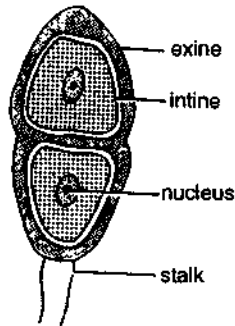


Fig. 8. *Puccinia*. Teleutospore.

### Comments

1. The teleutosori or teleutopustules appear on leaves, leaf sheaths and stem as black, oval pustules that fuse to form patches in case of severe infection.
2. A teleutosorus in a section reveals the (dikaryotic) intercellular, branched mycelium, a bunch of teleutospores and the ruptured host epidermis.
3. The host epidermis is ruptured due to the pressure of underlying teleutospores.
4. The teleutospores are formed by the same mycelium which earlier produced uredospores.
5. Each teleutospore is borne terminally by the mycelium. It is stalked, elongated and bicelled structure.
6. The apex of the teleutospore may be rounded or pointed as in *P. graminis* or it may be nearly flat as in *P. recondita* and *P. striiformis*.
7. The teleutospore has a very thick but smooth exine and delicate thin intine. The exine turns black at maturity.
8. At first each of the two cells of the teleutospore is binucleate but later on, the nuclei fuse making each of them uninucleate.
9. Each cell of the bicelled teleutospore has a single germ pore.
10. The teleutospores are incapable of infecting the primary host (wheat plant). They germinate to form the basidiospores which infect the barberry plant or *Thalictrum*, etc., the alternate host.  
(The pycnidial and aecidial cups are formed only on alternate hosts).

### Exercise 5

**Object :** Study of pycnidial cup and pycnidiospores.

### Work procedure

These are found on the upper leaf surface of barberry. A T.s. of leaf is cut and stained with cotton blue or safranin and fast green combination for temporary or permanent preparations respectively.

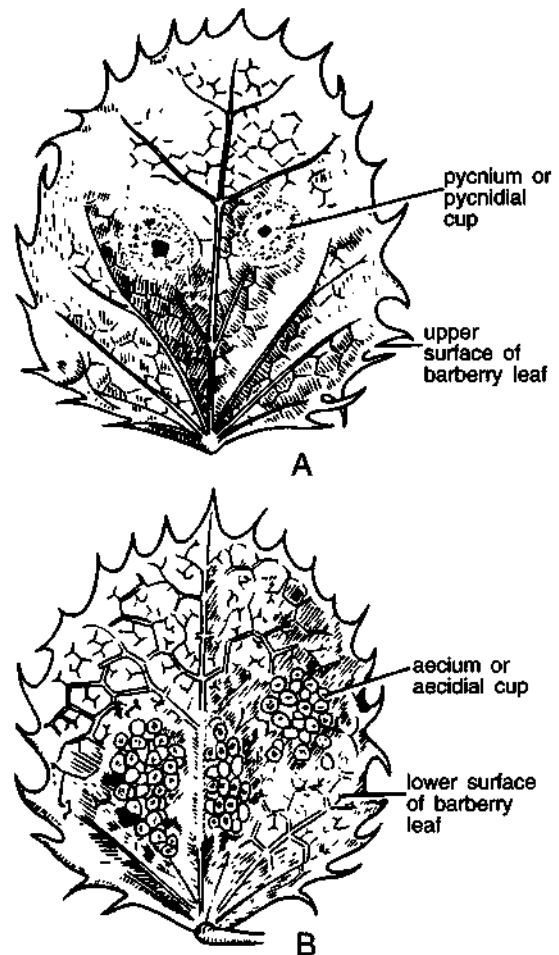


Fig. 9. *Puccinia graminis*. Leaves of barberry showing A. Pycnidial cups on upper surface B. Aecidial cups on lower surface.

### Comments

1. Each basidiospore germinates on the leaf of alternate host producing the (monokaryotic) mycelium, that ultimately forms the pycnial cup or pycnidium.
2. The pycnidia are generally present on the upper surface of the leaf and may be best studied in a transverse section of the host leaf.
3. A mature pycnidium is flask-shaped with a pore known as ostiole at its apex.
4. The hyphae near the ostiole are unbranched, pointed and orange coloured. These are called periphyses and project through the ostiole.
5. Some of the periphyses are branched and thin walled. These are called receptive hyphae (or flexuous hyphae). They project through the ostiole far beyond the periphyses.
6. The cavity of the pycnidium is lined by many elongated and uninucleate pycnidiohores or spermatophores.
7. The pycnidiohores are arranged in a palisade-like layer and each cuts off a chain of pycnidiospores or spermatia.
8. The pycnidiospores or spermatia are discharged through the ostiole and help in producing the dikaryotic mycelium.

### Exercise 6

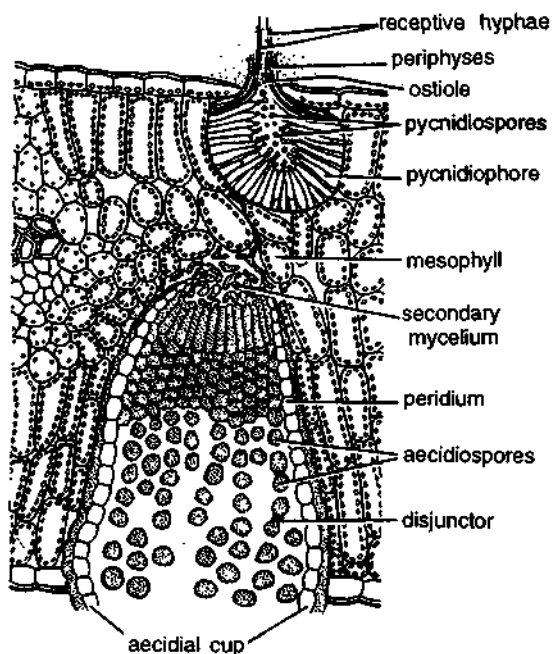
**Object :** Study of aecidial cup and aecidiospores.

### Work procedure

A transverse section of barberry leaf is stained with cotton blue and mounted in lactophenol. If permanent preparation is to be made safranin-fast green combination is useful.

### Comments

1. The aecidial cup or aecidium can only be formed by a dikaryotic mycelium.
2. The aecidia are generally present on the lower surface of leaf and thus both pycnidia and aecidia can be seen in the same section of the host leaf.



**Fig. 10.** *Puccinia*. T.s. of infected leaf of *Berberis* showing pycnidial and aecidial cups on upper and lower epidermis respectively.

3. Each aecidium is cup-like structure with an outer protective layer called peridium.
4. The developing aecidium elongates and is pushed through the host epidermis.
5. At the base of aecidium there are many elongated cells known as sporophores, arranged in a palisade-like manner.
6. Each sporophore cuts off alternately, a small and a large cell. The small cell is a disjunctors whereas the latter is the aecidiospore.
7. In younger conditions, aecidiospores are hexagonal and are held in chains by the disjunctors cells. The spores round off as soon as they get separated.
8. Each aecidiospore is a binucleate structure with a thick and smooth wall.
9. The aecidiospores are blown away by wind and infect wheat plant (primary host). They are not capable of re-infecting barberry (the alternate host).

**Exercise 7****Object : Study of *Puccinia butleri*.****Work procedure**

Collect *Launea* that grows wild. The pustules are prominently seen on leaves. Cut T.s. of the leaf or scrap the spores from the surface and study.

**Comments**

It is an autoecious rust, hence all the stages are found on a single host i.e. *Launea* sp., member of family Compositae. In this case there is no alternate host.

**Identification**

**Kingdom—Mycota.** (1) Chlorophyll absent, (2) Reserve food glycogen, (3) Cell wall of fungal cellulose.

**Division—Eumycota.** A definite cell wall present.

**Sub-division—Basidiomycotina.** (1) Mycelium septate, (2) Basidium is reproductive body, (3) Basidiospores usually four, produced exogenously.

**Class—Teliomycetes.** (1) Basidiocarp lacking, (2) Teliospores or chlamydospores in sori or scattered, (3) Parasitic on vascular plants.

**Order—Uredinales.** (1) Teleutospores formed terminally, (2) Basidiospores on sterigmata, (3) Infected plants rusty in colour.

**Family—Pucciniaceae.** (1) Teleutospores stalked, (2) Teleutospores free or united but never in the form of a layer.

**Genus—Puccinia.** (1) Teleutospores bicelled, (2) Aecia cupulate.

**Hints for Collection**

The fungus is an obligate parasite. Some of the hosts as wheat, bajra, etc., are cultivated for their grains. The fungus can be collected from the wheat fields in February-March in Northern India. *Berberis* and *Thalictrum* are common weeds on hills. The former is a bush, whereas the latter is a large herb. *Launea* is very common as a weed in the fields and lawns. The fungus on *Launea* can be collected in March-April.

**Agaricus  
(Mushroom)****Classification**

Kingdom	—	Mycota
Division	—	Eumycota
Sub-division	—	Basidiomycotina
Class	—	Hymenomycetes
Sub-class	—	Holobasidiomycetidae
Order	—	Agaricales
Family	—	Agaricaceae
Genus	—	<i>Agaricus</i>

**Exercise 1****Object : Study the vegetative structure.****Work procedure**

Collect white cotton growth on decaying matter, stain in cotton blue, mount in lactophenol and study.

**Comments**

1. The mycelium is underground and consists of much branched hyphae, anastomosing at their points of contact, forming a network in the substratum.
2. Hyphae are septate, dikaryotic with granular protoplasm and prominent oil globules.
3. The aerial portion constitutes the fruiting body. (Fruiting body is formed only by the dikaryotic mycelium).

**Exercise 2****Object : Study of button stage.****Work procedure**

Study a small and young mushroom. Cut L.s., stain with safranin or fast green and mount in glycerine to study.

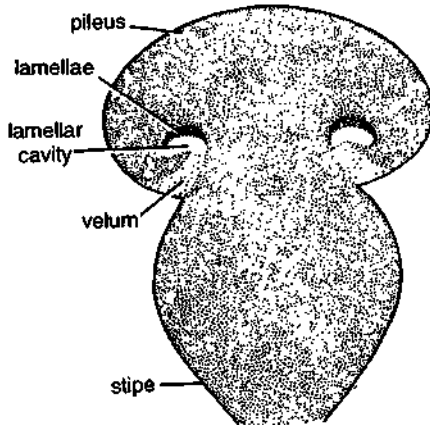


Fig. 1. *Agaricus*. L.s. button stage.

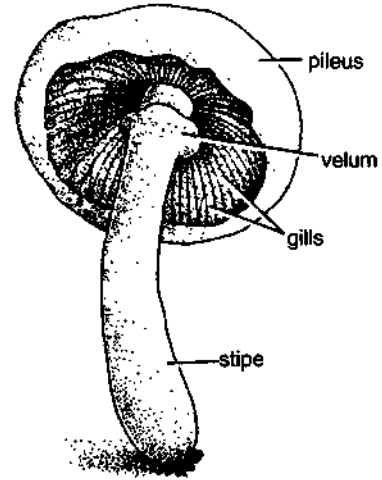


Fig. 2. *Agaricus*. A basidiocarp.

### Comments

1. This is a developmental stage of the basidiocarp.
2. It is formed above the ground in the form of a small, globose body.
3. In a longitudinal cut, it reveals a small stipe surmounted by the pileus.
4. In between pileus and stipe, there is a constriction.
5. At the level of this constriction are seen two lamellar cavities or chambers, one on either side.
6. The lamellar cavities or chambers have small lamellae or gills.
7. The edge of the pileus is connected to the stipe by a thin sheet of tissue—the veil or velum.

remains in the form of a ring (annulus) on the stipe, just below the pileus.

3. The upper surface of pileus is flesh coloured and tough.
4. The pileus, on the underside, bears many lamellae or gills which hang down vertically and extend almost radially from stipe to the margin of the pileus.
5. The gill in transverse section exhibits a trama, a sub-hymenium and a hymenium.
6. The trama forms a central core of elongated sterile hyphae.
7. The hyphal cells of trama curve outwards on either side of the gill forming a more or less compact tissue of cells, the sub-hymenium.

### Exercise 3

**Object :** Study of basidiocarp, gills, basidia and basidiospores.

### Work procedure

Pluck the pileus of the mushroom after studying it externally. Observe it from the lower side under a high magnification lens. Also study a slide showing T.s. of gills.

### Comments

1. The mature basidiocarp consists of a stalk or the stipe, having an expanded pileus at its top.
2. With the growth of the basidiocarp, the veil or velum ruptures and in mature basidiocarp it

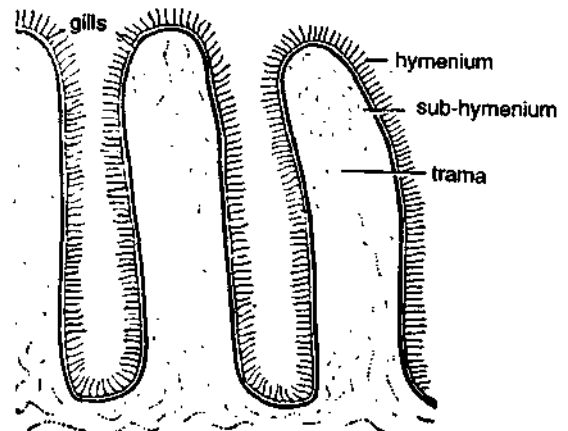


Fig. 3. *Agaricus*. Gills as seen in transverse section.



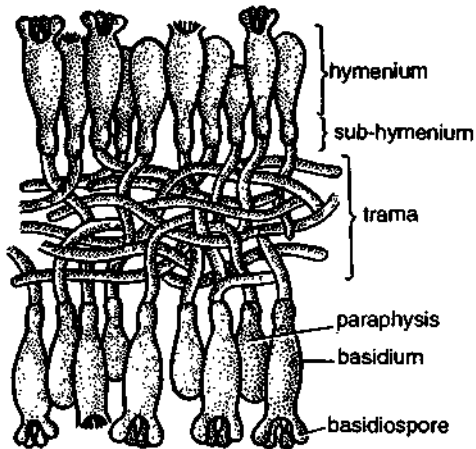


Fig. 4. *Agaricus*. Gills as seen in transverse section (magnified).

8. Finally the hyphae terminate in elongated, club-shaped cells, forming a superficial layer of the gill, known as the hymenium.
9. The hymenium at maturity, consists of the fertile cells, the basidia, intermingled with the sterile cells, the paraphyses. (The paraphyses are undeveloped basidia).
10. Each basidium is a club-shaped structure, bearing at its top generally four but sometimes two basidiospores, on short slender stalks known as sterigmata.
11. Each basidiospore is oval in shape and uninucleate.
12. On germination, it produces the new (monokaryotic) mycelium.

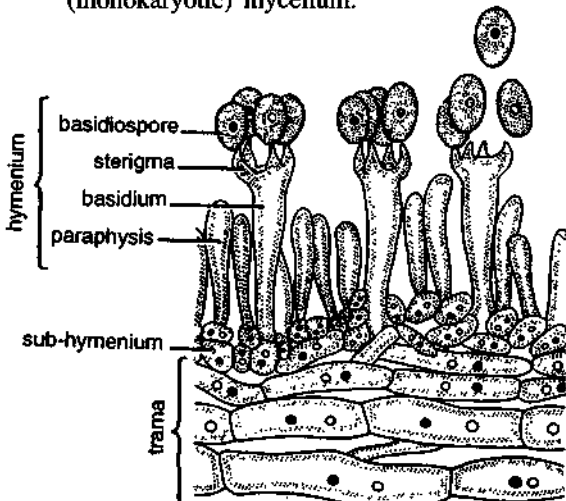


Fig. 5. *Agaricus*. A portion of gills in section showing basidia and basidiocarp.

## Identification

**Kingdom—Mycota.** (1) Chlorophyll absent, (2) Reserve food glycogen, (3) Cell wall of fungal cellulose.

**Division—Eumycota.** A definite cell wall present.

**Sub-division—Basidiomycotina.** (1) Mycelium septate, (2) Characteristic reproductive body is basidium, (3) Basidiospores, usually four and exogenous.

**Class—Hymenomycetes.** (1) Basidiocarps usually well developed, (2) Mostly saprobic.

**Sub-class—Holobasidiomycetidae.** Basidia club-shaped and non septate.

**Order—Agaricales.** (1) Basidia borne on lamellae, (2) Basidiocarp soft and putrescent.

**Family—Agaricaceae.** (1) Basidiocarp fleshy, (2) Gills narrow in section.

**Genus—*Agaricus*.** (1) Pileus centrally stipitate, (2) Annulus typically present, (3) Gills free, stipe readily separating from pileus.

## Hints for Collection

The mushroom (vern. kukarmutta, saap ki chatri) is very common in humus soil, dung, rotten logs of wood and other similar decaying organic substances during the rainy season.

**Precaution.** Stay away from beautifully coloured mushrooms for they are often poisonous.

## *Polyporus* (Bracket Fungus)

## Classification

Kingdom	—	Mycota
Division	—	Eumycota
Sub-division	—	Basidiomycotina
Class	—	Hymenomycetes
Sub-class	—	Holobasidiomycetidae
Order	—	Polyporales
Family	—	Polyporaceae
Genus	—	<i>Polyporus</i>

## Exercise 1

**Object :** Study of hosts and diseases.

## Work procedure

A list of various hosts is given below. Collect this bracket fungus in polythene bags.

### Comments

1. Many species of the genus are destructive parasites and cause disease in forest and other shade trees. Many others grow on lumber and destroy it.
2. *P. sulphureus* (sulphur mushroom) causes Wood rot of oaks (*Quercus* sp.; fam. Fagaceae).
3. *P. squamosus* causes a serious Heart rot in elms (*Ulmus* sp.; fam. Ulmaceae) and other trees.
4. *P. versicolour* grows on various woods and is known as Wood rotter.
5. *P. betulinus* is very common on birch (*Betula* sp.; fam. Betulaceae).

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### Exercise 2

**Object :** Study of vegetative structure.

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### Work procedure

Pick up a part of bracket with forceps or cut a thin section, stain in cotton blue and mount in lactophenol.

### Comments

1. The mycelium is well developed, branched and septate.
2. Generally the mycelium grows within and below the bark but in case of severe attacks, it completely invests the central wood cylinder.

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### Exercise 3

**Object :** Study of basidiocarp, basidia and basidiospores.

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### Work procedure

Study the external features of a fungal organization, cut a T.s., stain in cotton blue, mount in lactophenol and study.

### Comments

1. The mycelium (dikaryotic) forms a more or less flat fruting body, the basidiocarp.
2. The basidiocarp is characteristically shelf-like, shortly stalked and arises from the tree trunks.
3. It is leathery, corky or woody, whitish or slightly greyish or brownish in colour.



Fig. 1. *Polyporus*. Basidiocarp growing on wood.

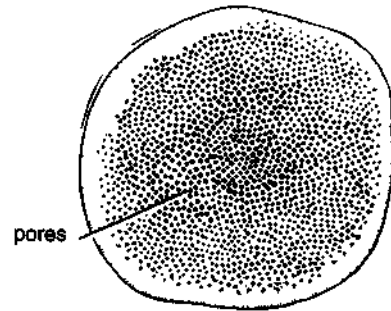


Fig. 2. *Polyporus*. Basidiocarp as seen from lower side.

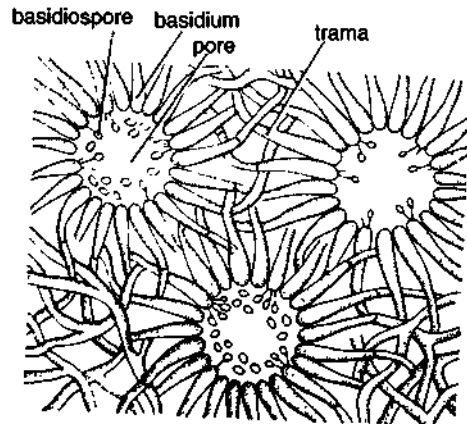


Fig. 3. *Polyporus*. Basidiocarp in section (a part only).

4. The upper surface is generally smooth, sometimes rough, often undulating, while the lower surface is porous. In some species the upper surface is distinctly striated.
5. The section of basidiocarp shows an outer context, trama, pores and hymenium.
6. The context is the outer fibrous part made up of thick walled hyphae.
7. The trama is a loose mass of much branched, septate and anastomosing hyphae.

8. The pores or tubes extend from below the context to the lower surface.
9. The hymenium is made up of a distinct layer of basidia, lining each pore or tube.
10. The basidia are club-shaped, somewhat larger than the sterile cells of the hymenium and project slightly into the cavity of the pore.
11. Each basidium has four short sterigmata at its free end, terminating into a basidiospore each.
12. The large number of basidiospores are discharged in the pore.
13. Each basidiospore is small, oval and is uninucleate.
14. The basidiospore on germination gives rise to the mycelium (monokaryotic).

### Identification

**Kingdom—Mycota.** (1) Chlorophyll absent. (2) Reserve food glycogen, (3) Cell wall of fungal cellulose.

**Division—Eumycota.** A definite cell wall present.

**Sub-division—Basidiomycotina.** (1) Mycelium septate, (2) Characteristic reproductive body is basidium, (3) Basidiospores, usually four and exogenous.

**Class—Hymenomyces.** (1) Basidiocarps usually well developed, (2) Mostly saprobic.

**Sub-class—Holobasidiomycetidae.** Basidia club-shaped and non-septate.

**Order—Polyporales.** Texture of basidiocarp not soft and putrescent.

**Family—Polyporaceae.** (1) Basidia line the inner surface of the pore or tube, (2) Tubes or pores, generally deep.

**Genus—Polyporus.** (1) Bracket or shelf-like shape, (2) Basidiocarp grow from wood, (3) Spores round, somewhat radially elongated, (4) Context always white or light coloured.

### Hints for Collection

Bracket fungi are common on tree trunks or on wood in damp forests.

### Sub-division 5.

## DEUTEROMYCOTINA (THE FUNGI IMPERFECTI)

The deuteromycotina is a more or less artificial and heterogenous group of some 15,000 to 20,000 species, phylogenetically unrelated to each other. The forms of fungi, in which the 'perfect stage' (sexual reproduction) was unknown and which, therefore, could not be placed in the already existing classes, were kept in this form class. The members, reproduce only asexually (imperfect stages).

The parasitism is mainly of the facultative type, enabling the fungi to cover summer under adverse conditions. Many species are, however, obligate saprophytes.

The mycelium is profusely branched and septate and may either be hyaline or coloured. The mycelium frequently produces rhizomorphs and sclerotia.

The reproduction is chiefly by coloured or hyaline conidia borne on conidiospores. The conidiospores are either free or organized into groups or clusters as sori, pycnidia, acervuli, sporodochia, cinnemata, etc. However, chlamydospores are also produced by some forms.

The sub-division is divided into three classes—(1) Blastomycetes, (2) Hyphomycetes and (3) Coelomycetes.

## Alternaria

### Classification

Kingdom	—	Mycota
Division	—	Eumycota
Sub-division	—	Deuteromycotina
Class	—	Hyphomycetes
Order	—	Moniliales
Family	—	Dematiaceae
Genus	—	Alternaria

### Exercise 1

**Object :** Study of host, diseases and symptoms.

### Work procedure

A list of common hosts is given below for facilitating the collection of various diseased hosts. These may be preserved in F.A.A. or dried for mounting on herbarium sheets.

### Comments

Majority of the species are weak parasites but some are saprophytes also.

Species of *Alternaria* parasitize plants as well as animals.

1. Of all the species, *A. solani* is economically very important, as it attacks potato plant (*Solanum tuberosum*; vern. alu; fam. Solanaceae) and causes early blight of potato. In Indian plains, early blight is more destructive than late blight.
2. *A. brassicae* and *A. brassicicola* cause grey and dark leaf spots of *Brassica* spp. (fam. Cruciferae).

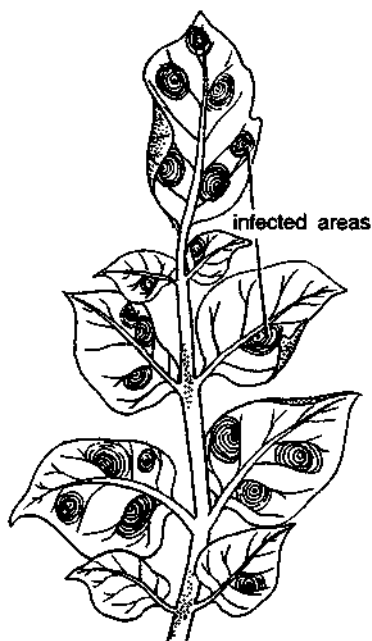


Fig. 1. *Alternaria*. Potato twig infected by *A. solani* causing Early Blight of Potato.

3. *A. burnsii* parasitizes cumin (*Cuminum cyminum*; vern. jeera; fam. Umbelliferae).
4. *A. triticina* causes leaf blight of wheat (*Triticum* spp.; vern. gehun; fam. Graminae).
5. *A. alterata* (= *A. tenuis*) infects sunflower (*Helianthus annuus*; fam. Compositae or Asteraceae) and large number of other hosts causing Leaf spot disease.
6. Conidia of *A. tenuis* also cause 'alternariasis' in guinea pig.

Generally the blight develops at the margins of the leaves but the whole leaf can get infected. The preliminary symptoms appear in the form of small, isolated and pale brown spots. As the spots grow, they become black, irregularly circular and show a series of concentric rings which produce a 'target board' effect.

7. Other species which infect various hosts include
  - (1) *A. lini*—Alternaria blight of linseed (*Linum usitatissimum*; vern. alsii),
  - (2) *A. carthami*—Alternaria disease of safflower (*Carthamus tinctorius*; vern. kusum),
  - (3) *A. palandui*—blight of garlic (*Allium sativa*; vern. pyazi).

### Exercise 2

**Object :** Study of vegetative structure.

#### Work procedure

Cut a section of host. Stain the section in cotton blue and mount in lactophenol for study of mycelium.

#### Comments

1. Mycelium is intercellular or sometimes intracellular and yellowish brown in colour.
2. Hyphae are multicellular and branched.
3. Hyphae are also septate. Each cell is uninucleate.

### Exercise 3

**Object :** Study of conidia.

#### Work procedure

Stain the T.s. of infected host leaf in safranin-fast green combination.

#### Comments

1. The conidia also are usually yellowish brown in colour.
2. The conidia may either be single or in chains. These are borne on conidiophores which are

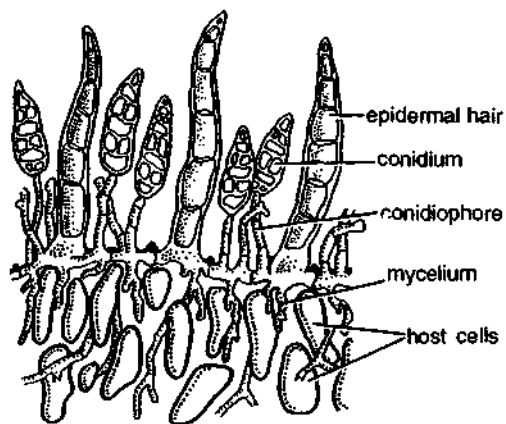


Fig. 2. *Alternaria*. T.s of infected host leaf showing conidiophores and conidia.



Fig. 3. *Alternaria*. A few conidia.

not much distinguishable from the vegetative hyphae emerging through stomata.

3. They are long, dark coloured, muriform (beaked), multicellular and dictyosporous i.e. spindle shaped or ovoid with both transverse and longitudinal septa.
4. The perfect stage of this form genus wherever known belongs of Loculoascomycetes-genus *Pleospora*.

### Identification

**Kingdom—Mycota.** (1) Chlorophyll absent, (2) Reserve food glycogen, (3) Cell wall of fungal cellulose.

**Division—Eumycota.** A definite cell wall present.

**Sub-division—Deuteromycotina.** Perfect stage absent, reproduction by asexual means only.

**Class—Hyphomycetes.** (1) Mycelium sterile or bearing spores directly or on special branches, (2) Not aggregated in pycnidia or acervuli.

**Order—Moniliales.** Conidia borne on free conidiospores.

**Family—Dematiaceae.** The mycelium, conidiospore and usually the conidia are brown or black in colour.

**Genus—*Alternaria*.** (1) Conidiospores distinct, mostly erect, (2) Conidia dictyosporous.

### Hints for Collection

Various species of *Alternaria* can be collected on hosts mentioned above. *A. solani* can be gathered from potato fields, three or four weeks after the crop is sown.

## Cercospora

### Classification

Kingdom	—	Mycota
Division	—	Eumycota
Sub-division	—	Deuteromycotina
Class	—	Hyphomycetes
Order	—	Moniliales
Family	—	Dematiaceae
Genus	—	<i>Cercospora</i>

### Exercise 1

**Object :** Study of hosts, diseases and the symptoms.

### Work procedure

Collect as many diseased hosts as possible. Preserve them in containers with F.A.A. or dry the specimen for mounting them on herbarium sheets.

### Comments

Majority of the species are facultative parasites. Most of these often turn out as destructive parasites commonly producing Leaf spot diseases.

It causes 'Leaf spot' diseases in the following plants.

1. **Cereals.** Narrow brown leaf spot *C. oryzae* on *Oryza sativa*, (rice, vern. chawal), *C. sorghii* on *Zea mays* (maize; vern. makka).
2. **Pulses.** *C. canescens* and *C. cruenta* on *Vigna sinensis* (cow pea), *C. cruenta* and *C. dolichi* on *Phaseolus* (vern. moong), *C. indica* on *Cajanus cajan* (pigeon pea; vern. arhar), *C. sesbaniae* on *Sesbania*, *C. sojina* on *Glycine max* (soya bean), *C. dolichi* on *Dolichos lablab* (vern. sem).
3. **Oil seeds.** *C. ricinella* on *Ricinus communis* (castor; vern. arandi); tikka disease is caused by *C. personata* and *C. arachidicola* on *Arachis hypogaea* (ground nut; vern. moongphali), *C. carthami* on *Carthamus tinctorius* (safflower; vern. kusum), *C. sesami* on *Sesamum orientale* (sesamum; vern. til).
4. **Fibre crops.** *C. gossypina* on *Gossypium* spp. (cotton, vern. kapas).

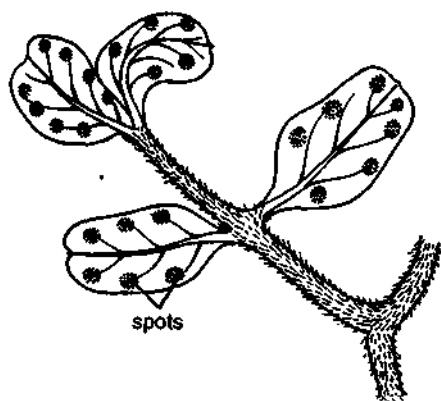


Fig. 1. *Cercospora*. Infected plant of *Arachis hypogaea* (Ground nut).

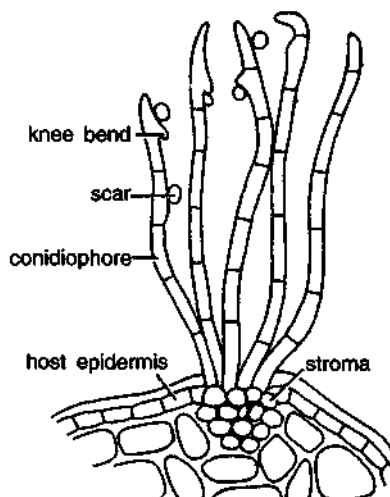


Fig. 3. *Cercospora*. Conidiophores with geniculate branching and the scars.

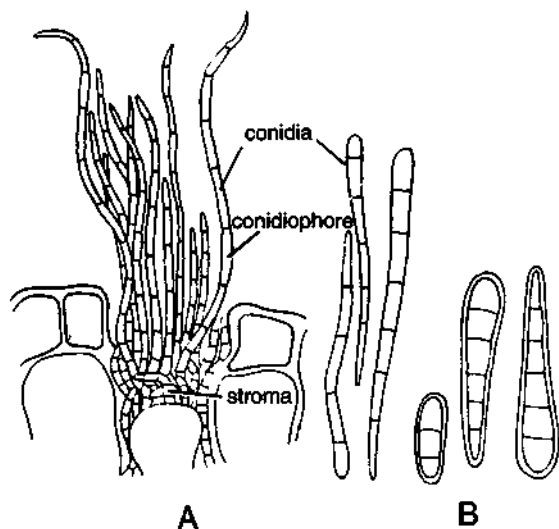


Fig. 2. *Cercospora*. A. T.s. host leaf, B. Conidia.

5. **Fodder crops.** *C. medicaginis* on *Medicago sativa* (Alfalfa or Lucerne), *C. traversiana* on *Trigonella foenum-graceum* (vern. methi).
6. **Vegetables.** *C. canavaliae* on *Canavalia ensiformis* (sword bean), *C. beticola* on *Beta vulgaris* (beet root; vern. chukander), *C. melongenae* on *Solanum melongena* (brinjal; vern. baingan), *C. concors* on *Solanum tuberosum* (potato; vern. alu), *C. capsici* on *Capsicum annum* (chillies; vern. Mirch), *C. batatae* and *C. bataticola* on *Ipomoea batatas* (sweet potato; ven. shakarkandi).
7. **Fruit crops.** Sigatoka disease caused by *C. musae* on *Musa* spp. (plantain; vern. kela).
8. **Plantation crops.** *C. coffeicola* on *Coffea*

*arabica* (coffee), *C. longipes* (causing brown leaf spot) and *C. kopkei* (causing yellow leaf spot) on *Saccharum officinarum* (sugar cane; vern. ganna).

9. **Narcotics.** *C. nicotianae* (causing frog eye leaf spot) on *Nicotiana* spp. (vern. tambaku).
10. **Condiment.** *C. apii* on *Apium graveolens* (celery; vern. ajmud).

During leaf spot diseases, fungus produces pale green spots on the upper surface of the leaf. These gradually turn yellow and ultimately become brown. The foliage finally dries up and is destroyed when disease is severe and destructive. Also, either the fruits are not formed or remain smaller.

### Exercise 2

**Object :** Study of vegetative structure.

### Work procedure

T.s. of the host leaf is stained with safranin-fast green combination.

### Comments

1. Mycelium consists of multicellular, septate and branched hyphae.
2. Parasitic hyphae is slender and intracellular. Inside the host, it forms lobed haustoria which penetrate the cells.

### Exercise 3

**Object :** To study the conidia.

#### Work procedure

T.s. of the host leaf is stained with cotton blue—lactophenol or safranin-fast green combination for permanent preparation.

#### Comments

1. The hyphal mass is aggregated beneath the epidermis as pseudoparenchymatous stroma.
2. A tuft of short, septate, geniculate, (knee-like) thin walled and unbranched conidiophores emerge through the epidermis. Mature conidiophores are dark coloured, and somewhat thicker than the rest of the hyphae.
3. Conidium is produced at the tip of conidiophore. This conidium is pushed to a side and the tip of conidiophore resumes its growth. Later, a new conidium is produced at its apex.
4. At the places of attachment, conidia leave a scar after falling off.
5. Conidium is inversely clavate (rounded at base and tapering towards apex) and straight or slightly curved. It is generally 4-5 septate (at times 12-15 septate).
6. The colour of the conidium ranges from hyaline to ash-gray to light brown.
7. The perfect stage of this form genus is *Mycosphaerella*.

#### Identification

**Kingdom—Mycota.** (1) Chlorophyll absent, (2) Reserve food glycogen, (3) Cell wall consists of fungal cellulose.

**Division—Eumycota.** Definite cell wall.

**Sub-division—Deuteromycotina.** (1) Perfect stage absent, (2) Reproduction by asexual means only.

**Class—Hyphomycetes.** (1) Mycelium sterile or bearing spores directly or on special branches, (2) Not aggregated in pycnidia or acervuli.

**Order—Moniliales.** Conidia borne on free conidiophores; pycnidia and acervuli never formed.

**Family—Dematiaceae.** The mycelium, conidiophore and usually the conidia are brown and black in colour.

**Genus—Cercospora.** (1) Conidiophores geniculate, (2) Conidia usually clavate.

### Hints for Collection

The common hosts are sugarbeet, tomato, potato, tobacco, ground nut and many others, listed above. Infection can be observed when these plants are well grown in the fields.

## Colletotrichum

#### Classification

Kingdom	—	Mycota
Division	—	Eumycota
Sub-division	—	Deuteromycotina
Class	—	Coelomycetes
Order	—	Melanconiales
Family	—	Melanconiaceae
Genus	—	<i>Colletotrichum</i>

#### Exercise 1

**Object :** Study of hosts, diseases and the symptoms.

#### Work procedure

Most of the species of *Colletotrichum* are parasitic on higher plants, such as sugarcane, cotton, beans, onion, etc. while others are saprophytes.

#### Comments

Species parasitic on important cultivated plants and causing diseases include—

1. *C. falcatum* infects sugarcane (*Saccharum officinarum*; vern. ganna; fam. Graminae) and causes red rot.  
In red rot, the fungus generally infects the stem and midrib of the leaves. The stem gets rotten within, the rind becomes dull in appearance and shrinks at the nodes. The upper leaves turn paler, droop slightly and split open, they show a red colour in the internodes. On the midribs, the infection is seen in the form of dark-reddish areas, which elongate rapidly, forming blood red lesions with dark margins.
2. *C. gossypii* and *C. lindemuthianum* parasitize cotton (*Gossypium* sp.; vern. kapas; fam.

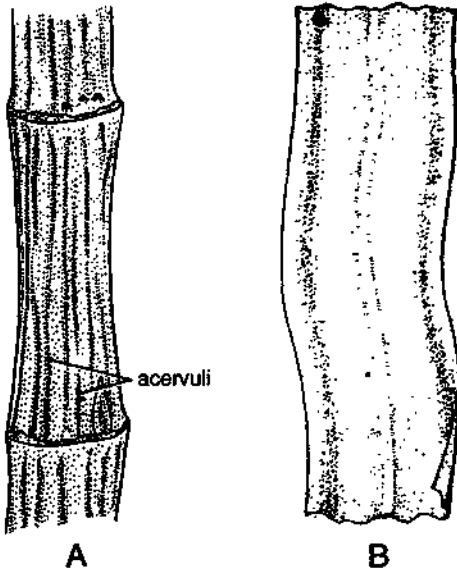


Fig. 1. *Colletotrichum falcatum* on sugarcane showing lesions on A. Stem and B. Leaf.

Malvaceae) and beans respectively and cause a disease called anthracnose.

3. *C. circinans* causes smudge in onion (*Allium cepa*; vern. pyaj; fam. Liliaceae).
4. Other species infecting various economically important plants include : *C. capsici* causing leaf spot on turmeric (*Curcuma longa*; vern.

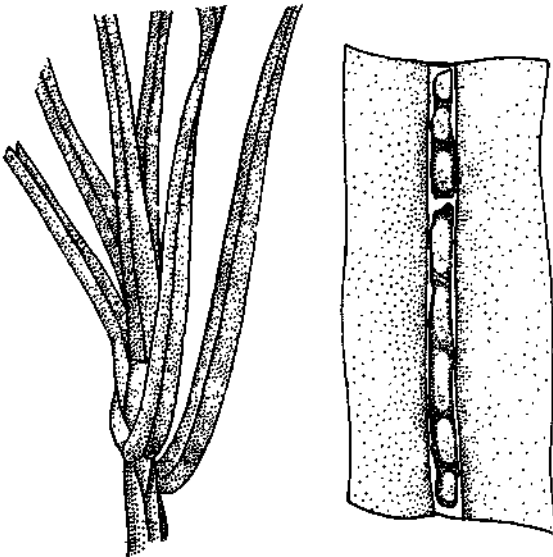


Fig. 2. *Colletotrichum falcatum* on sugarcane showing lesions on A. Stem and B. Leaf

haldi); die back on cow pea (*Vigna sinensis*), and anthracnose and ripe rot on chillies (*Capsicum annuum*; vern. mirch); *C. graminicola* causing red leaf spot of sudan grass (*Sorghum sudanense*); *C. gloeosporioides* causing anthracnose of mango (*Mangifera indica*; vern. aam), and black pepper (*Piper nigrum*; vern. kali mirch); *C. coffeanum* causing die back of coffee (*Coffea arabica*) and *C. camelliae* infecting tea (*Camellia sinensis*) to cause brown blight.

### Exercise 2

**Object :** Study of vegetative structure.

#### Work procedure

Study the section of stem of sugarcane for mycelium.

#### Comments

1. The mycelium is inter- and intracellular.
2. Hyphae are freely branched, septate, colourless and contain characteristic oil droplets.

### Exercise 3

**Object :** Study of acervulus and conidia.

#### Work procedure

Cut a section of superficial acervulus or pick up acervulus by a needle, stain in cotton blue, mount in lactophenol and study.

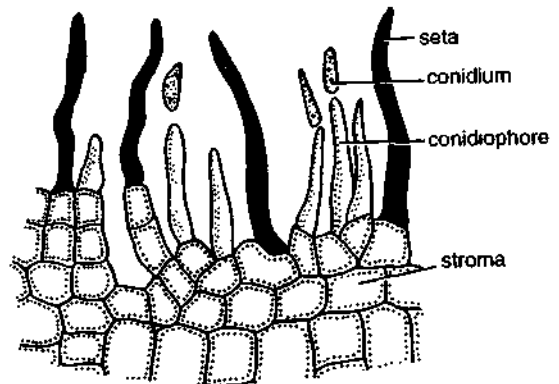


Fig. 3. *Colletotrichum*. Acervulus.



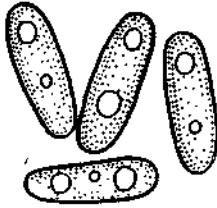


Fig. 4. *Colletotrichum*. Conidia.

### Comments

1. The conidia are always formed in acervuli.
2. The acervulus is formed on the surface of the rind as minute black clusters, just above or below the nodes. These develop from the stromatic mass of hyphae just beneath the epidermis.
3. Each saucer-shaped acervulus has a layer of unseptate conidiophores arranged in a palisade-like manner.
4. Intermixed with conidiophores are black, long, rigid, bristle-like and septate setae. Sometimes, setae form a fringe around the acervulus.

5. The conidia are borne on conidiophores.
6. Each conidium is one celled, falcate and is typically elongated with rounded ends. It is hyaline and densely granular.
7. The perfect stage is *Glomerella singulata*.

### Identification

*Kingdom*—Mycota. (1) Chlorophyll absent, (2) Reserve food glycogen, (3) Cell wall of fungal cellulose.

*Division*—Eumycota. A definite cell wall present.

*Sub-division*—Deuteromycotina. Perfect stage absent, reproduction by asexual means only.

*Class*—Coelomycetes. Thallospores or conidia borne in pycnidium or acervulus.

*Order*—Melanconiales. Conidia are borne in acervuli.

*Family*—Melanconiaceae. Single family.

*Genus*—*Colletotrichum*. (1) Acervulus dark coloured, (2) Setae present in or around acervulus.

### Hints for Collection

All the hosts are cultivated for their valuable edible products. Diseased plants can be collected from crop fields. Sugarcane is an important crop in U.P., Bihar, Punjab and parts of South India.

### Preamble

Lichens are a group of organisms, composite or dual in nature. The thallus of an alga and a fungus. The association is so intimate that it often gives an appearance of a single plant. The fungal partner is called a mycobiont and the algal partner as phycobiont. The phycobiont generally belongs to cyanophyceae or sometimes to chlorophyceae. The alga is unicellular. The Phycobiont is generally an ascomycete but in rare cases it is a basidiomycete.

The lichens were first discovered by Tulasne in 1892. The relationship between the two partners is a matter of controversy, some hold it to be a typical case of symbiosis whereas others consider it to be parasitism. However, it is now considered to be a case of helotism, a type of symbiotic association where the fungus has an upper hand.

The lichens grow on a variety of habitats, and are common on rocks, bark of trees, etc. Many of them grow under extreme conditions of cold, humidity and drought. *Cladonia rangifera*, the reindeer moss, for instance grows in arctic tundra which is a very cold region.

On the basis of their general growth, form and nature of attachment to the substratum, lichens are classified into following three categories—(1) Crustose or crustaceous, (2) Foliose or foliaceous and (3) Fruticose or filamentous.

Depending upon the fungal component in the lichen, the group is divided into (1) ascolichen, if the fungal component is an ascomycete and (2) basidiolichen, if the fungal component is a basidiomycete.

Lichens have been used as food by man e.g. *Cetraria islandica* (Iceland Moss) is eaten by man in Iceland and Scandinavia. Lichens like *Cladonia*, *Stereocaulon* are fodder lichens being eaten by reindeers. Lichens are also useful in brewing and distillation, cosmetics and perfumes, tanning, dyeing, etc. In fact the *Hawan Samagri* and *Dhup* used in connection with religious ceremonies, mainly consists of lichens.

## ASCOLICHENS

According to the external shape and habitat, ascolichens are divided into three principal groups, crustose lichens, foliose lichens and fruticose lichens.

### Crustose Lichens

#### Exercise 1

**Object :** Study of external features.

Study the specimen provided.

### Comments

1. The crustose lichens are hard, granular crusts on rocks and bark of the trees.
2. These adhere very closely and firmly to the substratum.
3. The thalli are partially or completely embedded in the substratum.
4. These lichens are generally ash-coloured. However, the colours may vary.
5. The surface of the thallus is generally divided into polygonal areas called areolae.
6. Some of the commonest examples include *Lecidea*, *Graphis*, *Lecanora*.

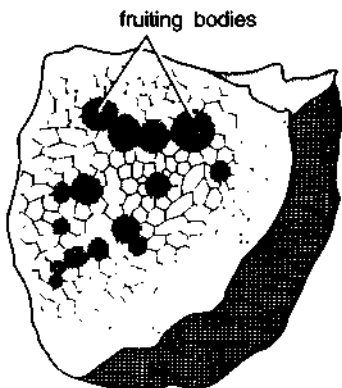


Fig. 1. *Lecidia platycarpa*. Crustose ascolichen growing on rocks : External features and fruiting bodies.

### Exercise 2

**Object :** Study of internal structure.

#### Work procedure

Study the slide showing internal structure.

#### Comments

1. The thallus is poorly differentiated.
2. The tissues forming the thallus are arranged more or less in strata, one above the other.
3. The upper cortex is made of rudimentary or sometimes highly developed hyphal layer.
4. Algal layer lies just beneath this layer. The layer shows algal filaments and the fungal hyphae in close association.
5. Much below the algal layer lies the medulla composed of loose tissue of branching hyphae.
6. The lower cortex lies next to medulla. It may be well developed or entirely absent.

### Foliose Lichens

### Exercise 1

**Object :** Study of external features.

#### Work procedure

Study the specimen provided.

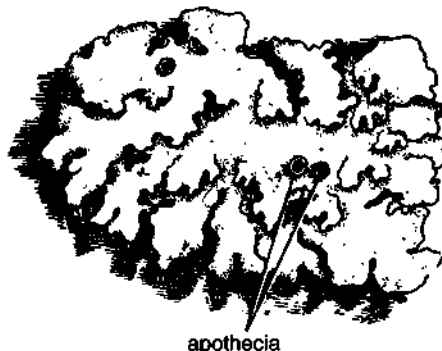


Fig. 2. *Parmelia flavicens*. Foliose ascolichen. A thallus showing external features and apothecia.

#### Comments

1. The foliose lichens have a flat, leaf-like, lobed or deeply incised thallus.
2. It is attached to the substratum only at certain points by rhizines.
3. Rhizines are rhizoid-like outgrowths which arise from the under surface.
4. The thallus may be attached to the substratum either by a single rhizine or by several rhizines.
5. The thallus is generally greyish or brownish in colour.
6. Certain small, hard, dark and gall-like outgrowths called cephalodia may also be present. These help in retaining moisture.
7. The common examples include *Parmelia*, *Physcia*, etc.

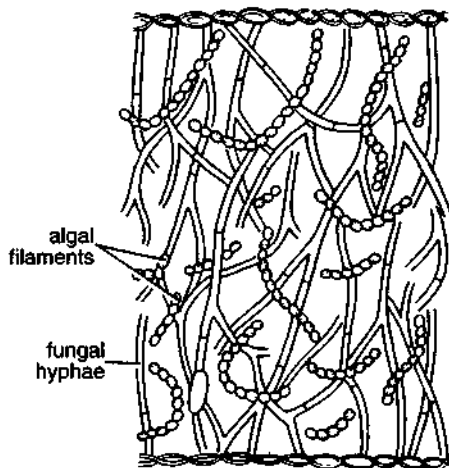


Fig. 3. *Collema*. Homoiomerous thallus showing internal structure.

**Exercise 2**

**Object :** Study of internal structure of homoiomerous thallus.

**Work procedure**

Study the slide showing homoiomerous structure e.g. *Collema*, *Leptogium*, etc.

**Comments**

1. These types of thalli are not very common.
2. Internal structure is not much differentiated.
3. The algal cells are irregularly scattered throughout the fungal hyphae.
4. Both algal cells and fungal hyphae are enveloped in a gelatinous matrix or the ground substance.

**Exercise 3**

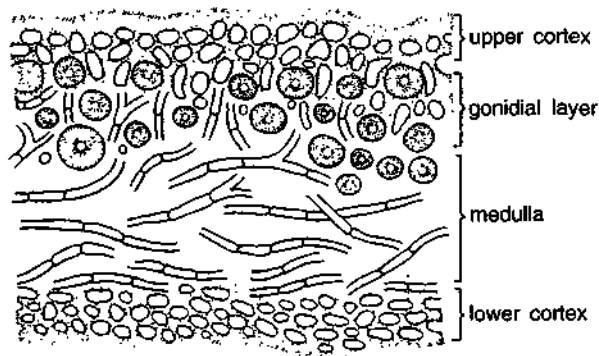
**Object :** Study of internal structure of heteromerous thallus.

**Work procedure**

Study the section of heteromerous thallus of a lichen e.g. *Parmelia*, *Physcia*, etc.

**Comments**

1. Internal structure of the thallus shows four distinct regions or zones. These are upper cortex, gonidial layer or algal layer, medulla and the lower cortex.



**Fig. 4.** *Physcia*. V.s. thallus to show heteromerous internal structure.

2. Upper cortex may or may not be bounded by an epidermis-like layer or hyphae.
3. Certain breathing pores may also be present in the epidermis. These help in gaseous exchange.
4. The upper cortex itself consists of vertical hyphae without intercellular spaces or with such spaces filled with gelatinous materials.
5. Beneath the upper cortex is algal or gonidial layer. It consists of numerous algal cells held together in the network of fungal hyphae.
6. Below the gonidial layer is the medulla made of very loosely interwoven hyphae.
7. On the lower side is the lower cortex, consisting of compact cells, lying either parallel or perpendicular to the lower surface.
8. Cells of the lower cortex produce rhizines.

**Exercise 4**

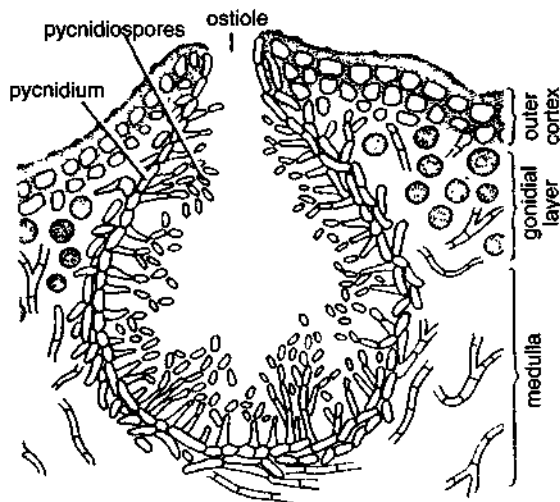
**Object :** Study of pycniospores.

**Work procedure**

Study a section passing through pycnidial cup or pycnidium.

**Comments**

1. Pycnidia or pycnidial cups are produced on the upper surface of the thallus.
2. Pycnidium is a flask-shaped cavity with a small opening at its apex, an ostiole.



**Fig. 5.** *Physcia*. V.s. pycnidium to show pycniospores.

3. The pycnidium is lined by the hyphae throughout its entire inner cavity.
4. Pycniospores are produced at the tip of these hyphae.
5. Pycniospores are released through an ostiole and germinate to produce a lichen if it comes in contact with an alga.

### Exercise 5

**Object :** Study of ascospores.

#### Work procedure.

Study a section passing through an apothecium. Study the asci and the ascospores.

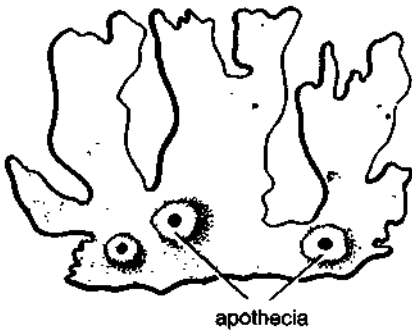


Fig. 6. *Physcia*. Thallus showing apothecia.

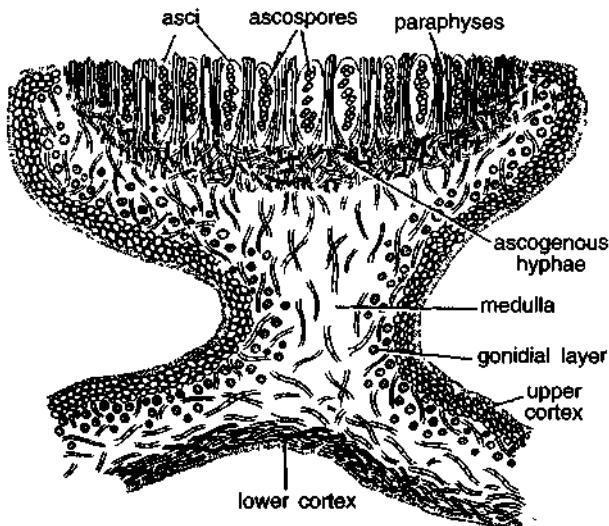


Fig. 7. *Physcia*. V.s apothecium for the study of ascospores.

#### Comments

1. Apothecia are saucer-shaped fruiting bodies (e.g. *Usnea*, *Physcia*, etc.)
2. In some cases, perithecia, flask-shaped fruiting bodies are also formed.
3. Apothecium (also perithecium) is lined with palisade-like layer of cells called hymenium.
4. The hymenium consists of a series of elongated cells—the asci, intermixed with sterile hyphae—the paraphyses.
5. Each ascus usually contains eight ascospores but the number may vary from one to eight.
6. Ascospores, when liberated, if come in contact with the suitable alga, produce the lichen thallus.

## Fruticose Lichens

### Exercise 1

**Object :** Study of external features of thallus.

#### Work procedure

Comment upon the specimen provided.

#### Comments

1. These appear shrubby with cylindrical, flat or ribbon-like body.
2. It is upright, generally branched and pendulous.
3. It remains attached to the substratum by rhizoid-like structures forming a disc.

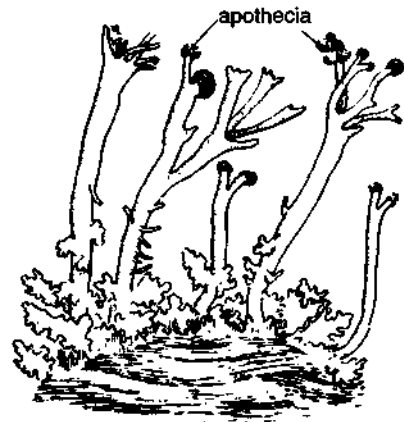


Fig. 8. *Cladonia flabelliformis*. A fruticose ascolichen showing external features.

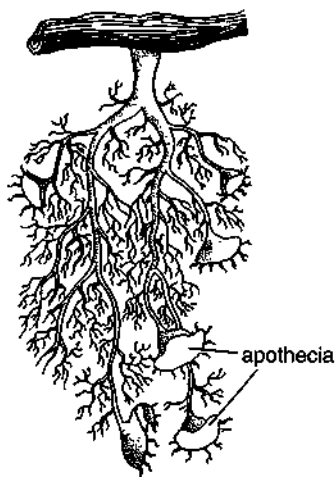


Fig. 9. *Usnea* sp. A fruticose ascolichen to show external features.

4. The common examples include *Usnea*, *Alectonia*, *Cladonia*, etc.

### Exercise 2

**Object : Study of internal structure of thallus.**

#### Work procedure

Study the T.s. of thallus of a common fruticose lichen, e.g. *Usnea*.

#### Comments

1. The transverse section appears almost circular.
2. The internal structure shows four distinct regions, outermost cortex, the algal zone, medulla and the centrally located chondroid axis.
3. Thallus structure is radially symmetrical.

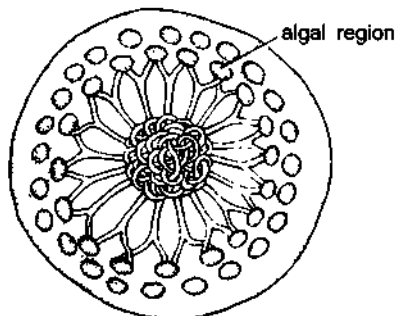


Fig. 10. *Usnea* sp. A fruticose thallus. Transverse section to show internal structure.

4. The cortex is made of closely packed and interwoven fungal hyphae. Intercellular spaces are absent.
5. The algal zone consists of cell of unicellular green alga, *Protococcus*.
6. Medulla follows the algal zone. It consists of algal cells loosely mixed with fungal hyphae scattered in different directions.
7. The central chondroid axis is made of longitudinally arranged, compact, thick walled and closely grouped fungal hyphae.

### Exercise 3

**Object : Study of vegetative structures.**

#### Work procedure

Study various structures which develop at different times on the thalli.

#### Comments

1. Vegetative propagation takes place by fragmentation, isidia and soredia.
2. Fragmentation is the commonest of all the methods.
3. Isidia develop as outgrowths of thallus. These develop into new lichen thalli after separating from the parent thallus.
4. Soredia are bud-like outgrowths developing either from the entire surface or in localised patches called soredia.
5. Soredia are developed in the gonidial layer.
6. A soredium consists of a few algal cells surrounded by hyphae.
7. Soredia get detached from the thallus and are carried away by the wind. Germination occurs on suitable substratum.

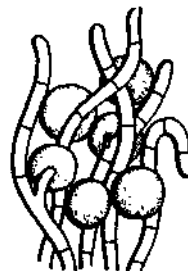


Fig. 11. *Usnea*. A soredium

## BASIDIOLICHENS

### Exercise 1

**Object :** Study of external features of the thallus.

#### Work procedure

Study the characters of a specimen provided.

#### Comments

1. The basidiolichens are tropical in distribution and are found growing upon bare soil, rocks and trees.
2. There are only three genera of basidiolichens and out of these *Cora pavonia* is the best known.
3. The thallus is much lobed, internally attached to the substratum by rhizines and resembles superficially with 'bracket fungi'.

### Exercise 2

**Object :** Study of internal structure of thallus.

#### Work procedure

Study the section of the thallus as seen in slide.

#### Comments

1. As seen in a vertical cut, the thallus is differentiated into three layers—the superior layer, the algal or gonidial layer and the inferior layer.
2. The superior layer is uppermost and consists of loose felt of more or less perpendicular hyphae.
3. The algal layer is the middle one. It is made of algal cells (*Chroococcus* species) intermixed with loose hyphae.
4. The inferior layer is the lowermost which is a dense felt of hyphae running in all directions.
5. The lowest surface bears concentric outgrowths of more or less perpendicular hyphae.
6. Each outgrowth is known as sub-hymenium and its lower face bears a palisade-like layer of basidia.

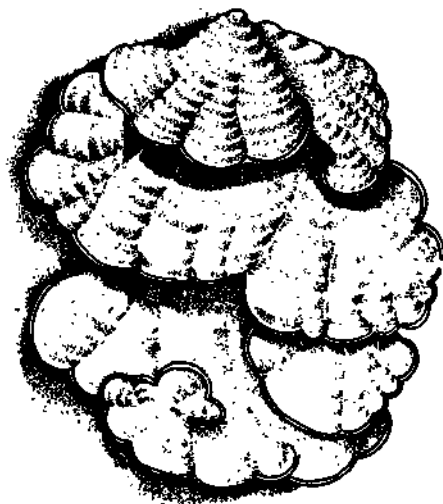


Fig. 12. *Cora pavonia*. External features.

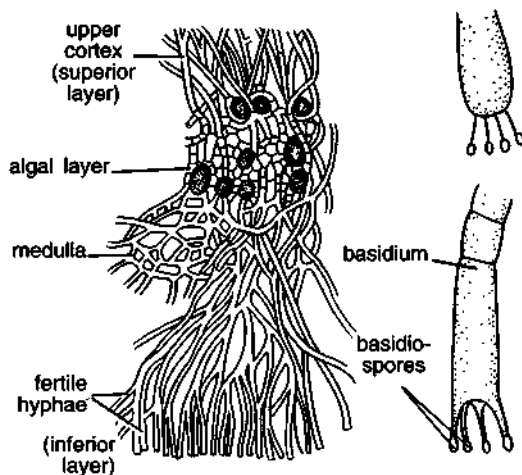


Fig. 13. *Cora pavonia*. V.s. thallus to show internal structure. A. Part of V.s. thallus, B. Basidia and Basidiospores.

7. Each basidium bears four terminal sterigmata with basidiospores.
8. Basidiospores are thus shed from the underside of the lichen. These on contact with alga form a lichen.

### Hints for Collection

The lichens are very common in hills. They are specially found on tree trunks and rocks.

# Microbiology

# 5

## Chapter

### Preamble

Microbiology is the study of micro-organisms. It is mainly concerned with the morphology, reproduction, physiology and identification of micro-organisms. It also includes the study of their distribution in environment, their interrelationships, their association with other living beings their beneficial and harmful effects on man, and the changes they bring to their environment. The micro-organisms include algae found most commonly in aquatic environments or in damp soil; bacteria and fungi found in practically all natural environments; single celled protozoans which cause diseases in man and other animals; rickettsiae obligate intercellular parasites found in many insects which transmit them to man and other animals; and the fascinating viruses—the well known pathogens of plants, animals and bacteria. Because of the small size of the micro-organisms, they are studied with the aid of microscope. Viruses are so small that they can be visualized only by electron microscope.

With this fundamental knowledge, one can find micro-organisms wherever they exist, know about their transmission and evolve methods for prevention of their sporadic growth. Thus the information emanated from this discipline does well to help identify micro-organisms, in the preservation of food, in the industry, in space laboratories, to prevent their spread and then the protection against various diseases.

### Exercise 1

**Object :** To culture or cultivate bacteria (and micro-organisms).

### Requirements

Potato, dextrose, sucrose, sodium nitrate ( $\text{NaNO}_3$ ), potassium hydrogen phosphate ( $\text{K}_2\text{HPO}_4$ ), potassium chloride (KCl), hydrated magnesium sulphate ( $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$ ), ferrous sulphate ( $\text{FeSO}_4$ ), sodium chloride (NaCl), sodium hydroxide (NaOH), sodium bicarbonate ( $\text{NaHCO}_3$ ), hydrochloric acid (HCl), alcohol, agar agar, water, beef, etc.

Conical flasks (250 ml, 500 ml, 1 l) culture tubes, petri dishes, beakers (500 ml), 100 ml pipette, 500 ml graduated measuring cylinder, etc.

Cotton, potato peeler, Bunsen burner/spirit lamp, wire test tube basket, wax pencil, forceps, inoculating needles, towel, etc.

Autoclave/pressure cooker, inoculation chamber, staining rack, balance, weights, etc.

### Work procedure

Cultivation of bacteria and their storage in laboratory is a process that involves sterile conditions and subsequent care. Therefore, precautions must be taken to avoid contamination by other micro-organisms. The major steps are— (1) Preparation of culture medium, (2) Sterilization (3) Filling test tubes, and petri dishes, (4) Inoculating or transfer of organisms to culture medium and (5) Storage.

### [I] Preparation of culture medium

A few common culture media and their methods of preparation are given below.

#### (a) Meat broth or meat extract broth

Beef extract	3 g
Peptone	10 g
NaCl	5 g
Distilled water	1000 ml
Agar agar	15 g

Meat infusions or beef extracts are prepared by soaking meat or beef in water for many hours.



Add beef extract, peptone and sodium chloride together and heat to 65°C, stirring until the materials are completely dissolved. Filter and adjust the pH to 7.2 to 7.6 by adding a pinch of sodium bicarbonate. Add agar to the broth. Heat slowly until agar is dissolved. Pour the medium in large conical flasks to be used as stock.

(b) *Potato dextrose agar*

Potato (peeled)	200 g
Dextrose	20 g
Distilled water	1000 ml
Agar agar	15 g

Weigh about 200 g of peeled potato. Wash it thoroughly. The pieces are boiled in flask for about 30 minutes. Allow to cool down and decant the supernatant potato extract. Add agar to the extract. Heat the extract slowly till agar is completely dissolved. Dextrose is now added and the whole mixture is agitated till it becomes homogeneous. pH of the medium is adjusted to around 6.4 by using 1/10 N HCl or 1/10 N NaOH. The medium is now poured into flasks as per the requirement. This is the stock to be used in future.

### [III] Sterilization

Conditions must be sterile or completely aseptic when bacteria are cultured. There should be no chances of contamination. There are many methods to achieve this. However, following methods would be more practical.

1. **Dry sterilization.** Glassware including petri dishes, test tubes, flasks, etc. may be sterilized by putting them in an oven at a temperature of 160° to 190°C for at least one hour.

2. **Steam under pressure (Autoclave).** Sterilize all the glassware, cotton, culture media, inoculating needles, etc. in an autoclave set at 15 pounds pressure for 15 minutes. Test tubes, petri dishes, etc. be kept in wire baskets. All the other glassware that could be useful in further procedure be also be placed in the autoclave. The flasks containing stock medium be plugged with cotton. For sterilization of small quantities of medium and glassware, pressure cooker is more practical.

### [III] Filling the test tubes

If stock medium is prepared and stored in large conical flasks, these flasks must be sterilized before

use. The cotton plug of the flask is opened under complete sterile conditions and close to the flame of the burner, to avoid any contamination.

The culture medium is now poured in the test tubes in such a way that it does not stick to the sides of the tubes and when the medium contains agar, it must be poured quickly so that agar does not have time to solidify. Fill the tubes 1/2 or 2/3 and plug them with sterile cotton. Medium may similarly be poured in the petri dishes from the large conical flasks with stock medium.

Finally sterilize all the test tubes and petri dishes by placing them in wire baskets and putting them in an autoclave for 20 minutes at 30 pounds pressure. Remove the test tubes from the autoclave and tilt the half full test tubes and set the medium at an angle so that the medium when set forms a slant (angle).

### [IV] Transfer of microorganisms or bacteria to the medium

The pathogen or bacteria are collected from the diseased host. More often, bacteria are found growing along with other micro-organisms. Hence, a culture prepared from this natural source would have a mixed population of different micro-organisms. Later, if pure cultures are desired, a colony that belongs to a desired bacterium is transferred to a fresh medium. This is generally known as sub-culturing and would permit the growth of one type of bacterium (or a micro-organism) alone (pure culture).

In order to do this, the first step is to transfer the organism directly from the natural source. Large number of bacteria removed from the tissues or substances like milk, curd, saliva, urine, etc. are diluted with distilled water. This mixture is kept in a petri dish.

The table top is cleaned with spirit or with 90% alcohol. Bunsen burner or a spirit lamp is lighted. A test tube containing nutrient agar is placed in hot water bath for melting the agar. It is now taken out and cotton plug is removed bringing the mouth of the test tube across the flame to eliminate contaminants around the mouth of the tube.

The mixture of bacteria or inoculum is now transferred to the agar slants or to the petri dish either by simply pouring a drop of distilled water or by using a dropper or pipette.

To inoculate or transfer to a fresh culture medium, following procedure is generally used. Take inoculating needle and heat it over the flame of the spirit lamp. Let the needle cool before being used once again. Remove the cotton plug from the sterile culture tube or flask and hold it in hand. The mouth of the tube be placed close to the flame of spirit lamp. The sterile and cooled inoculating needle is now inserted into the old culture growing in the tube. A small part of the colony is removed by the tip of the needle. This needle is now inserted into another sterile test tube with agar slant but without bacterial or any other culture. The needle is streaked along the entire length of sterile agar surface. The needle is flamed once again before putting it aside. Also flame the mouth of the test tubes before placing the cotton plugs.

When petri dishes are streaked, the cover of the petri dish is slightly raised and the needle is streaked over the surface in three equally distant parallel lines. The cover of the petri dish is replaced quickly. The dish is sealed with adhesive tape, its contents are labelled and the dish is stored.

#### [V] Storage

A special room is generally marked for the storage of cultures. The temperature of this room is maintained at 27°C. The cultures are ready for examination after about 2-3 days.

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#### Exercise 2

**Object :** To isolate micro-organisms from mixed culture and grow a pure culture.

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#### Requirements

Stock solution of meat extract medium/beef extract medium/PDA (Potato Dextrose—Agar)/any other suitable nutrient medium; conical flasks, culture tubes, petri dishes, cotton, Bunsen burner/spirit lamp, wire test tube basket, wax pencil, inoculating needles; autoclave/pressure cooker, inoculating chamber, etc.

#### Work procedure

The initial or original inoculum from the natural source has generally a mixed population of micro-organisms, hence when cultured it grows into a

mixed or contaminated culture. If an individual organism is to be studied, it must be isolated from the mixed culture and then grown again into a pure culture (a culture containing individuals or one species only). Many methods are available, however, the following is the simplest that can be adopted.

1. Observe different kinds of colonies growing in mixed culture.
2. Note different characteristics of colonies like size, form, elevation, margin, contour, surface, texture, colour, radial growth, etc. to identify an organism wherever possible.
3. The transfer of the desired colony is now done under the sterile conditions. The working table is wiped clean by cotton dipped in 90% alcohol.
4. The inoculating needle is heated red hot on a flame. It is then allowed to cool. The needle is generally not placed on the table.
5. The test tube with mixed culture is held in left hand and the sterilized needle in the right. The cotton plug of the tube is pulled out in between the fingers of the right hand. The open mouth of the culture tube is passed over the flame of the spirit lamp. The needle is now inserted into the tube and the colony or a part of it is lifted by the loop of the needle. The mouth of the tube is once again passed over the flame and the cotton plug is quickly replaced.
6. A fresh sterilised test tube with nutrient medium is taken out of wire test tube basket. The cotton plug is now pulled out between the fingers of the right hand. The open mouth is passed over the flame. The loop of the inoculating needle carrying colony (or a part of it) is now inserted into the tube and streaked along the surface of the agar slant. The mouth of the tube is passed over the flame once again and then plugged quickly.
7. Freshly inoculated tube (sub-culture) is now stored under suitable temperature.
8. The colonies appear within two to three days. (Blue-green algae take about 20-30 days to appear).
9. The fully grown up colonies can once again be taken out by using the same procedure as described in item no. 5.
10. The colonies are suitably stained and the organisms studied under the microscope.

### Exercise 3

**Object :** To stain and study bacteria (microorganisms).

#### Requirements

Crystal violet (aq. 0.5%), Iodine, alcohol 95%, Safranin (aq. 1%), microscope, needles, blotting papers, slides, glass dropper, etc.

#### Work procedure

##### [I] Preparation of bacterial film

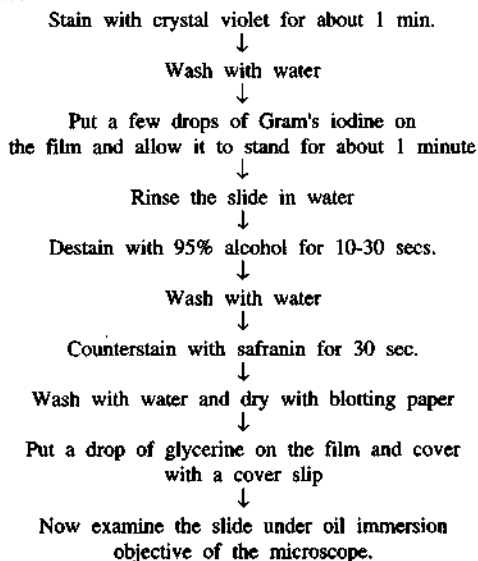
1. Take a clean slide and gently warm it over the spirit lamp. Let the slide cool.
2. Take drop of broth culture or pick up a part of colony by a needle loop.
3. Place it over the slide and spread evenly to form a film. Let the film dry.

##### [II] Fixing the bacterial film

1. Pass the film quickly 3-4 times over the flame of the spirit lamp.
2. Let the slide cool so that the film sticks firmly to the slide.

##### [III] Staining the bacterial film

Gram's stain is generally used to stain bacteria. The method is described separately. A summary is given below.



#### Observations

Gram<sup>+</sup> bacteria are stained violet or blue and Gram<sup>-</sup> bacteria are stained pink to reddish. Also observe the shape of the bacterial cells, the flagella if present, the capsule, etc.

### Exercise 4

**Object :** To measure the bacterial/other cells.

#### Requirements

Ocular micrometer (also known as oculometer), stage micrometer, slide of bacteria or any other structure to be measured.

#### Work procedure

##### [I] Coinciding the scales

**Stage micrometer.** It is a slide with engraved scale. It is generally ruled into tenths and hundredths of a millimeter. Thus each of the hundredth part or one division of stage micrometer represents 0.01 mm or 10 $\mu$  (1 mm = 1000 microns or  $\mu$ ).

**Ocular micrometer.** It is a disc of glass with engraved scale. It does not have any standard value and hence needs to be standardized with each microscope and the lenses (objective and eye piece) of different powers.

1. Place a stage micrometer slide on microscope stage. Focus the scale.
2. Place the ocular micrometer correctly on the metal diaphragm inside the eye piece. The lines of the micrometer should be sharp and clearly visible.
3. Move stage micrometer slide while looking through eye piece in a way so that both ocular and stage micrometer scales appear side by side.

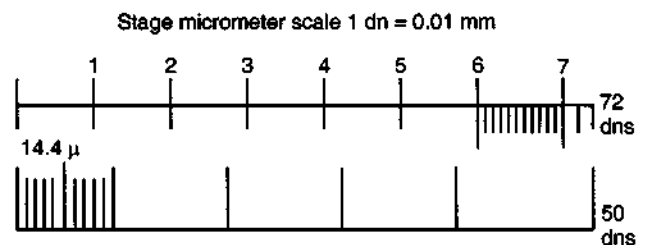


Fig. 1. Micrometer slide and oculometer being coincided.

- Now coincide the divisions of the stage micrometer slide with those of ocular micrometer.
- In this case, for example, 72 divisions of stage micrometer slide coincide with 50 divisions of ocular micrometer.

### [II] Standardization of ocular micrometer scale

- The set of lenses used is—objective 45x and eye piece 10x.
- The value of stage micrometer is—one division of stage micrometer is equal to .01 mm or 10 microns or  $\mu$ .
- Therefore, 72 divisions = 0.72 mm or 720 microns or  $\mu$ .
- Since 72 divisions of stage micrometer = 50 divisions of ocular micrometer.
- 0.72 mm or 720 microns or  $\mu$  = 50 divisions of ocular micrometer.
- Therefore, 1 division of ocular micrometer =  $0.72 \text{ mm}/50$  or  $720 \mu/50 = 0.144 \text{ mm}$  or 14.4 microns or  $\mu$ .
- The value of one division of ocular micrometer is, therefore, 0.144 mm or 14.4 microns or 14.4  $\mu$ .
- The following formula can be used.

1 Division of ocular micrometer

$$= \frac{10 \times \text{divisions of stage micrometer}}{\text{divisions of ocular micrometer}}$$

After substituting the values as given above

$$= \frac{10 \times 72}{50} = \frac{720}{50} = 14.4 \text{ microns } (\mu).$$

### [III] Measuring an object

- Remove the stage micrometer slide. The ocular micrometer should, however, be allowed to remain in the eye piece.
- Place the bacteria or any other object under the objective lens. A slide with a cover glass should be used.
- Move the eye piece or rotate, so that the ocular micrometer scale is adjusted over the object to be measured.
- If the cell diameter measures 2 divisions of ocular micrometer, then the diameter would be  $2 \times 14.4 \mu = 28.8 \mu$ .

- In this way different dimensions of different objects can be measured.

### Exercise 5

**Object : Microscopic examination of curd.**

#### Requirements

Milk, dropper, methylene blue, glycerine, slides, coverslips, etc.

#### Work procedure

- Place a drop of milk on the slide and prepare a smear.
- Let the smear dry.
- Stain the smear with methylene blue.
- Dry the smear again and place a drop of glycerine.
- Mount a coverslip and study.

#### Observations

Numerous bacterial cells are seen scattered. These generally belong to the genera *Lactobacillus* or *Streptococcus*.

#### [I] *Lactobacillus*

- These are rod-shaped bacteria. The rods are long and slender.
- The rods generally occur in chains.
- The bacterium shows peritrichous flagella but motility is rare.
- These are Gram<sup>+</sup> bacteria but may become Gram<sup>-</sup> with increasing age and acidity.
- Besides dairy products, the bacterium is also found in grains and meat products, water, sewage, beer, wine, fruits and fruit juices.
- Metabolism is generally fermentative but some are strict anaerobes.

#### [II] *Streptococcus*

- Cells spherical to ovoid, less than 2  $\mu\text{m}$  in diameter.
- The cells occur in pairs or chains.
- Occasional motile strains are also found.
- These are gram<sup>+</sup>, chemo-organotrophs, facultative anaerobes.

# 6

## Chapter

# Plant Pathology

### Preamble

Plant pathology or phytopathology is a branch of Botany that deals with the study of nature, development and control of plant diseases.

Disease is, in a sense any deviation from normal physiological processes or structural uniformity of an organism. Plant diseases have brought tremendous sufferings to the human race. The famous Irish famine of 1845 was due to Late Blight of Potato caused by *Phytophthora infestans*. It caused destruction of almost entire potato crop of the country. The famine resulted in the death of about a million people and about the same number of people were forced to migrate to other countries.

A similar historical catastrophe occurred in our country in 1942 which is known as Bengal famine. It was due to destruction of rice crop by a fungus *Helminthosporium oryzae*. An estimated 2 million people died of starvation.

This branch of Botany seems to be older than the origin of human civilization since the diseases seem to have originated with the origin of plants. The first book in which plant diseases have been discussed is *Vraksha Ayurveda* written by Surapal in India (1500 B.C.). Theophrastus was perhaps the first botanist to study and write about the diseases of trees, cereals and legumes.

Disease is caused by several factors, one being an organism that generally parasitizes the host. Such an organism is called pathogen. The potential capacity of pathogen is called pathogenicity. When pathogen enters the host, it causes infection which results in a disease causing numerous harmful effects on the host. Some of the significant effects include membrane permeability, photosynthesis, respiration, etc. Plant diseases are caused by fungi, bacteria, viruses, mycoplasma, nematodes, etc.

Infection most often produces viable symptoms like blights, damping off, hypertrophy, hyperplasia, galls, necrosis, chlorosis, etc. Sometimes the symptoms are so specific that disease could be easily detected and causal organism becomes known.

### List of Diseases Described

Disease	Causal organism	Host/s
<b>Fungal Diseases</b>		
1. Black wart disease (Wart disease of potato)	<i>Synchytrium endobioticum</i>	<i>Solanum tuberosum</i> , Potato, vern. aloo
2. Damping off	<i>Pythium</i> sp.	Tomato, tobacco, Ginger, Papaya
3. Late blight of potato	<i>Phytophthora infestans</i>	<i>Solanum tuberosum</i> , Potato, vern. aloo
4. Green ear disease of bajra	<i>Sclerospora graminicola</i>	<i>Pennisetum typhoideum</i> , Pearl millet, vern. bajra
5. Downy mildew of pea	<i>Peronospora pisi</i>	<i>Pisum sativum</i> , Pea, vern. mater
6. White rust of crucifers	<i>Albugo candida</i> (= <i>Cystopus candidus</i> )	<i>Raphanus sativus</i> , Radish, vern. mooli
7. Powdery mildew of barley	<i>Erysiphe graminis</i> var. <i>hordei</i>	<i>Hordeum vulgare</i> , vern. jaii

Contd...

8. Loose smut of wheat	<i>Ustilago tritici</i>	<i>Triticum aestivum</i> , vern. gehoon
9. Covered smut of barley	<i>Ustilago hordei</i>	<i>Hordeum vulgare</i> , vern. jaii
10. Whip smut of sugarcane	<i>Ustilago scitaminea</i>	<i>Saccharum officinarum</i> , vern. ganna
11. Black stem rust of wheat	<i>Puccinia graminis tritici</i>	<i>Triticum aestivum</i> , vern. gehoon
12. Brown/orange leaf rust of wheat	<i>Puccinia recondita</i>	<i>Triticum aestivum</i> , vern. gehoon
13. Rust of linseed	<i>Melampsora lini</i>	<i>Linum usitatissimum</i> , vern. alsii
14. Early blight of potato	<i>Alternaria solani</i>	<i>Solanum tuberosum</i> , vern. aloo
15. Tikka disease of groundnut	<i>Cercospora personata</i> , <i>C. arachidicola</i>	<i>Arachis hypogaea</i> , vern. moongphali
16. Wilt of cotton	<i>Fusarium oxysporum</i> f.sp. <i>vasinfectum</i>	<i>Gossypium</i> spp., vern. kapas
17. Red rot of sugarcane	<i>Colletotrichum falcatum</i>	<i>Saccharum officinarum</i> , vern. ganna
<b>Bacterial Diseases</b>		
18. Bacterial blight of rice (paddy)	<i>Xanthomonas oryzae</i>	<i>Oryza sativa</i> , vern. dhan, chawal
19. Citrus canker	<i>Phytoplasma citri</i> (= <i>Xanthomonas citri</i> )	<i>Citrus aurantifolia</i> , vern. nimbu
20. Tundu disease of wheat	<i>Phytoplasma tritici</i>	<i>Triticum aestivum</i> , vern. gehoon
<b>Viral Diseases</b>		
21. Leaf curl of potato	Potato virus I or ( <i>Solanum</i> virus 14)	<i>Solanum tuberosum</i> , vern. aloo
22. Tobacco mosaic	<i>Nicotiana</i> virus I	<i>Nicotiana tabacum</i> , vern. tambaku
23. Leaf curl of tobacco	Potato virus x	<i>Nicotiana tabacum</i> , vern. tambaku
24. Leaf curl of papaya	Tobacco virus 16, ( <i>Nicotiana</i> virus 10)	<i>Carica papaya</i> , vern. papita
25. Yellow vein mosaic of bhindi	<i>Hibiscus</i> Virus I	<i>Abelmoschus esculentus</i> , Lady's finger, okra
<b>Disease caused by Mycoplasma</b>		
26. Little leaf of brinjal	Mycoplasma (MLB)	<i>Solanum melongena</i> , vern. baingan
<b>Disease caused by Nematode</b>		
27. Root knot of vegetables	<i>Meloidogyne arneria</i> , <i>M. incognita</i> , <i>M. javanica</i>	Cucurbits, potato, tomato, brinjal, chillies, lady's finger, carrot, groundnut, radish etc.

## Black Wart Disease or Wart Disease of Potato

### Exercise 1

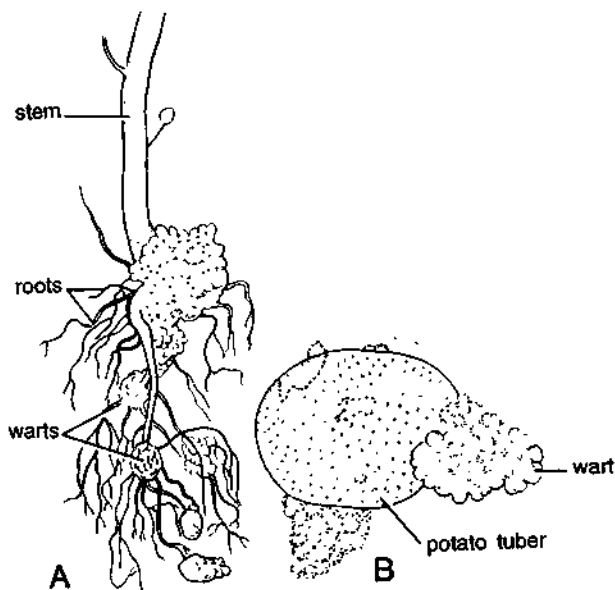
**Object :** To study the symptoms of wart disease of potato.

#### Work procedure

Observe the specimen provided. Note the symptoms. Recall control measures.

#### Comments

1. Wart disease of potato (*Solanum tuberosum*, vern. aloo; fam. Solanaceae) is caused by phycomycetous fungus *Synchytrium endobioticum*.
2. The disease is very commonly found in the hills particularly in Nilgiris, Shimla, Sikim and Darjeeling.



**Fig. 1.** Black wart disease of potato. A. Heavily warted plant. B. A potato tuber showing warts due to infection by *Synchytrium endobioticum*.

- The diseased potato tubers show dark brown, warty, cauliflower like outgrowths.
- The warts are produced due to stimulation of cells to divide in the presence of fungus.
- The disease can be controlled by certain chemicals like  $HgCl_2$ , ammonium sulphocyanate, copper sulphate, formalin, etc. and also by using resistant varieties.

### Exercise 2

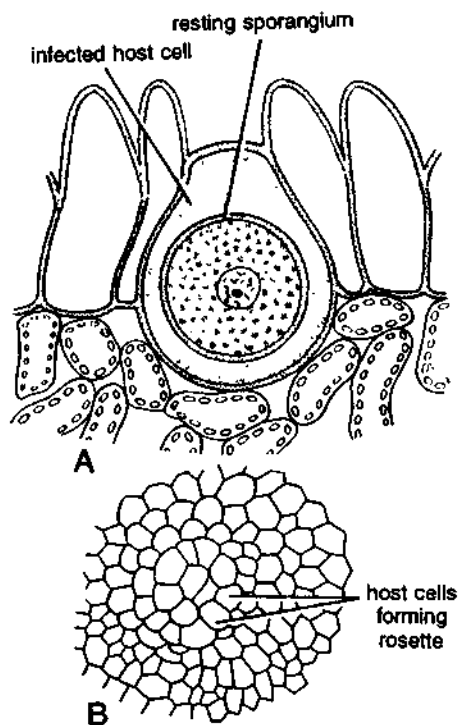
**Object :** Study of causal organism.

#### Work procedure

Cut a thin section of diseased part of potato tuber, stain in cotton blue and mount in lactophenol.

#### Comments

- Potato wart disease is caused by a phycomycetous fungus *Synchytrium endobioticum*.
- It is an obligate parasite.



**Fig. 2.** *Synchytrium*. The causal organism producing black warts on potato. A. Summer sporangium in host cell. B. Rosette of host cells.

- The thallus is a naked amoeboid protoplast and endobiotic.
- Uniflagellate zoospores enter through the host epidermis and cause infection.
- Thallus secretes golden-brown double layered chitinous wall to form prosorus.
- Zygote is formed as a result of sexual reproduction. It enlarges and secrets a two layered thick wall. The outer thick layer is folded and dark brown in colour. The inner layer is thin and hyaline.

## Damping Off

### Exercise 1

**Object :** Study of the symptoms of damping off.

#### Work procedure

Collect the specimen of diseased mustard or tomato. Study the symptoms. Recall the control measures.

#### Comments

- The disease is caused by a phycomycetous fungus *Pythium*. The common hosts include tomato, mustard, tobacco, ginger, papaya, wheat, etc.
- The seedlings are affected. The disease also affects seeds, older plants of many vegetables, flowers and also the fruits.
- Seeds infected in the soil fail to germinate. The seedlings become pale green.
- A girdle of brown decaying cortex develops at the base of seedling. The seedlings finally collapse.
- The disease is controlled by chemicals like—formalin, thiran, captan, etc. and also by various cultural practices like good drainage, aeration, etc.

### Exercise 2

**Object :** Study of causal organism.

#### Work procedure

A transverse section of the host is stained in cotton blue and mounted in lactophenol.

(B-14)

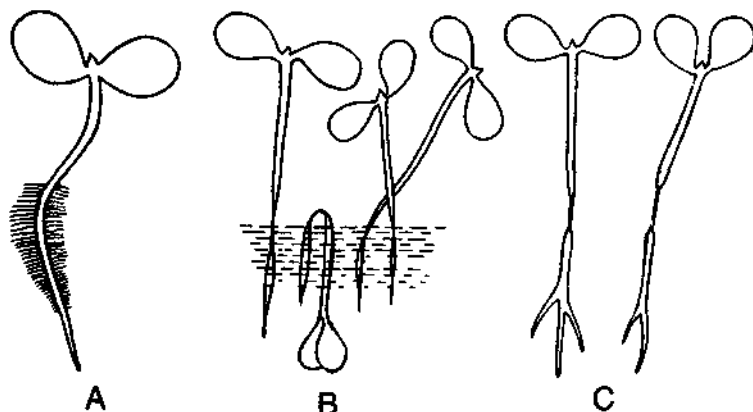


Fig. 1. Damping off. A. Disease free seedling B. Diseased seedling, C. Diseased seedlings in advanced stage.

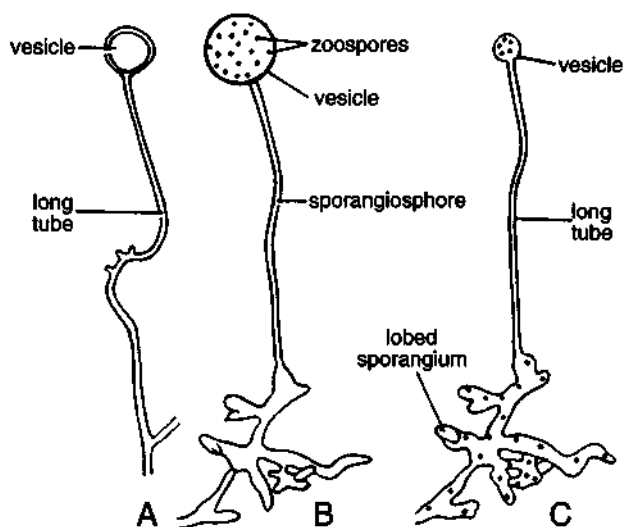


Fig. 2. *Pythium*. Causal organism of damping of A, B, C. different stages of sporangium.

### Comments

1. The causal organism is *Pythium*. The common pathogenic species are *P. aphanidermatum*, *P. debaryanum* and *P. ultimum*.
2. It is a saprophyte but can also grow as facultative parasite.
3. The thallus consists of freely branched coenocytic hyphae.
4. The fungus reproduces asexually by zoospores produced at the tips of hyphae. The zoosporangia are typified by the presence of an apical pailla.
5. Sexual reproduction is oogamous. The species are homothallic.

## Late Blight of Potato

### Exercise 1

**Object :** Study of the symptoms of late blight of potato.

### Work procedure

Collect a potato tuber showing disease. Note the symptoms and make a mention of control measures.

### Comments

1. This disease of potato (*Solanum tuberosum*; vern. aloo; fam. Solanaceae) is caused by *Phytophthora infestans* of phycomyces.
2. The disease infects potato twig and the tubers. The famous Ireland famine between 1845 and 1847 was due to this disease of potato.
3. The disease appears after the blossoming period.
4. The first symptoms are the brown spots or necrotic areas on the leaves.
5. These areas later become larger brownish-black lesions.
6. The lesions first appear at the tips of margins of the leaves. These later spread downward and inward and increase in size.
7. Generally the lower leaves are attacked first.
8. The underside of the leaves show whitish or greyish fungal growth—the downward hanging of sporangiophores with sporangia.
9. Tubers get infected while they are still attached to the plant.

(B-14)





Fig. 1. Late blight of potato. Potato twig and tubers infected by *Phytophthora infestans*.

10. The first symptoms of the tuber are brown purple discolouration of the tuber skin followed by brownish dry rot.
11. Late blight of potato is controlled by spraying of Bordeaux mixture, Blitox-50, Diathane Z-78, etc., destruction of haulms, seed selection, storage, production of disease resistant varieties, etc.

### Exercise 2

**Object :** Study of causal organism.

#### Work procedure

Transverse section of potato tuber from infected part would show characteristic features. Leaf may also be used for studying the characters of sporangia.

#### Comments

1. Causal organism is *Phytophthora infestans* of phycomycetes.

(B-14)

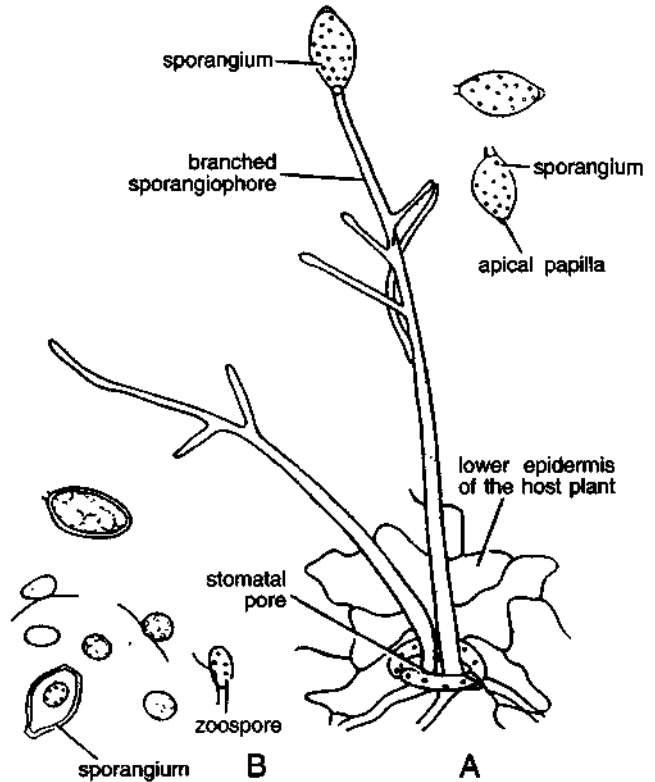


Fig. 2. *Phytophthora*. A. Sporangiophores coming out of stomata of potato leaf. B. Sporangia and the zoospores.

2. The thallus consists of hyaline, branched, coenocytic, intercellular mycelium with haustoria which enter the host cell.
3. Sporangiophores come out of the stomata of leaves and lenticels of tubers. They are simple or poorly branched, bearing a sporangium at the apex.
5. The sporangia are multinucleate, thin walled, hyaline, oval or pear-shaped with an apical papilla.

## Green Ear Disease of Bajra

### Exercise 1

**Object :** Study of symptoms of green ear disease of bajra.

#### Work procedure

Study the symptoms of the disease on leaves and the inflorescence.

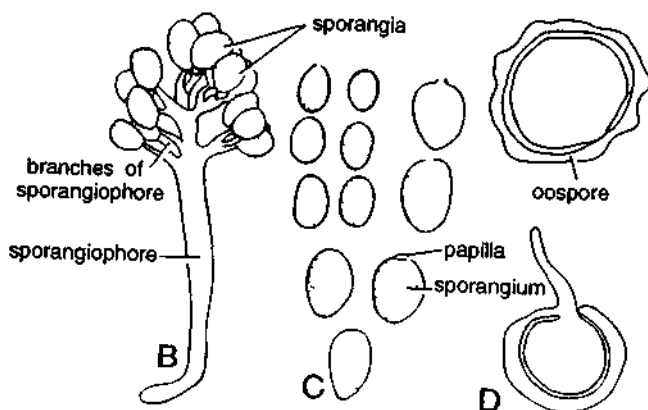
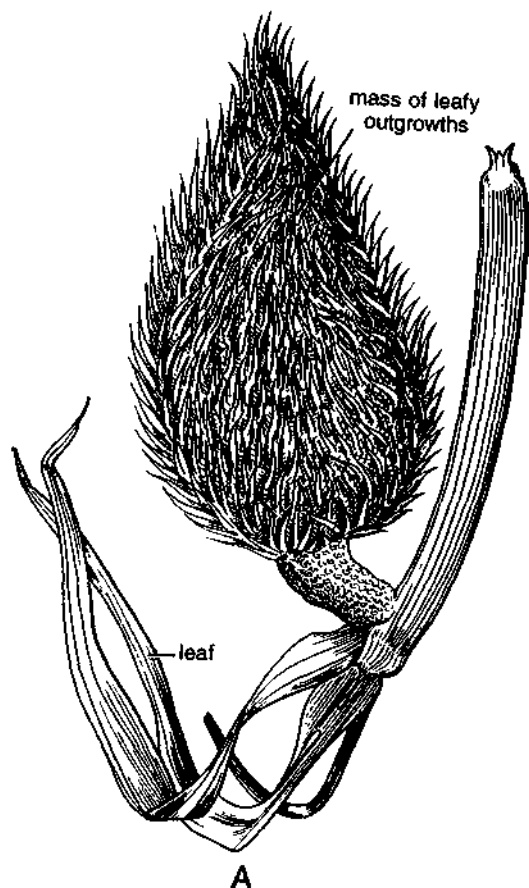


Fig 1. Green ear disease of bajra. A. Infected ear of bajra B. Sporangiophore with sporangia. C. A few sporangia. D. Oospore.

### Comments

1. This common disease of bajra (*Pennisetum typhoides*; Eng. pearl millet; fam. Poaceae) is

caused by a phycomycetous fungus, *Sclerospora graminicola*.

- The disease is common to all bajra growing regions of the country.
- Two types of symptoms are produced—downy mildew stage on the leaves and the green ear stage on the ear.
- In the **downy mildew** stage, the leaves
  - become white in the beginning, later becoming brown.
  - become whitish on the lower side due to sporangial growth, and
  - finally become distorted, twisted and crinkled and tend to split.
- In **green ear disease**
  - each flower is replaced by green leafy outgrowths thus turning the solid spicate ear, wholly or partly into a loose head,
  - the bristles of spikelets become hypertrophied and variously contorted. The glumes become enlarged and green,
  - number of florets in a spikelet also increase,
  - carpels are also replaced by leafy shoots or horny outgrowths.

### Exercise 2

**Object :** Study of causal organism.

### Work procedure

Study the lower leaf surface, T.s. of the leaf and also the infected grain of the inflorescence.

### Comments

- The causal organism is phycomycetous fungus—*Sclerospora graminicola*.
- The thallus consists of freely branched coenocytic hyphae that produces haustoria inside the host cell.
- Sporangiophores come out of the stomata of the leaf. Each sporangiophore is broad and unbranched at the base but may be dichotomously branched near the apex.
- The tips of the branches of sporangiophores are swollen and bear sporangia.
- Each sporangium is hyaline, elliptic, smooth and bears apical papilla.

- Oosporic stage is very common and is found in leafy structures.
- The oospores develop at the end of growing season of the host and are confined to brown coloured areas.

### Downy Mildew of Pea

#### Exercise 1

**Object :** Study of symptoms of downy mildew of pea.

#### Work procedure

Study the specimen of diseased parts of pea plant like leaflets, stipules and the pod.

#### Comments

- This is a common disease of pea (*Pisum sativum*; vern. matar; fam. Papilionaceae) and is caused by a phycomycetous fungus, *Peronospora pisi*.
- The symptoms appear at an early stage when the plant is not more than 10-12 cms in height.
- Yellow to brown spots or lesions appear on the upper surface of leaflets and stipules.
- At the same time corresponding areas of lower surface of leaves become covered with a whitish downy mildew.
- The symptoms on the pod appear at flat pod stage. The seeds near the infected tissues get considerably reduced in size and finally abort.
- Since the primary inoculum is present in the soil, sanitation and crop rotation help reduce the infection.

#### Exercise 2

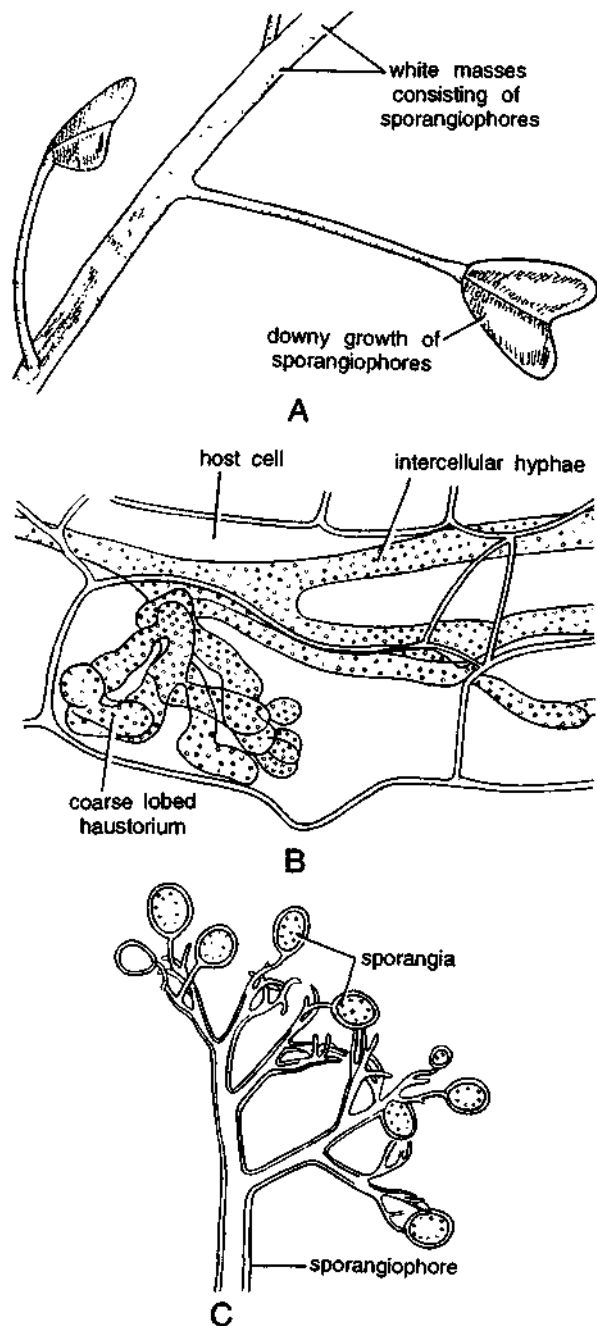
**Object :** Study of causal organism.

#### Work procedure

Pick up the downy coating from the lower leaf surface of pea. Also cut T.s. of the leaf, stain in cotton blue and mount in lactophenol.

#### Comments

- The causal organism is *Peronospora* of phycomycetes.



**Fig. 1.** Downy mildew of pea. A. Part of the plant infected with *Peronospora pisi*. B. Host cells with intercellular mycelium and haustoria. C. Mycelium with sporangiophore and sporangia.

- The thallus consists of mycelium that is freely branched, septate or coenocytic with knob-like haustoria inside the host cell.

3. Asexual reproduction takes place by sporangia, borne at the tips of sporangiophores.
4. Sporangiophores are unbranched at the base but are dichotomously branched near the apex. The tips of the branches are reflexed and bear the sporangia.
5. Oospores are formed as a result of sexual reproduction. These occur embedded in the old tissues of the host. They are spherical brown-yellow with thick and ornamented episporium.

### White Rust of Crucifers

#### Exercise 1

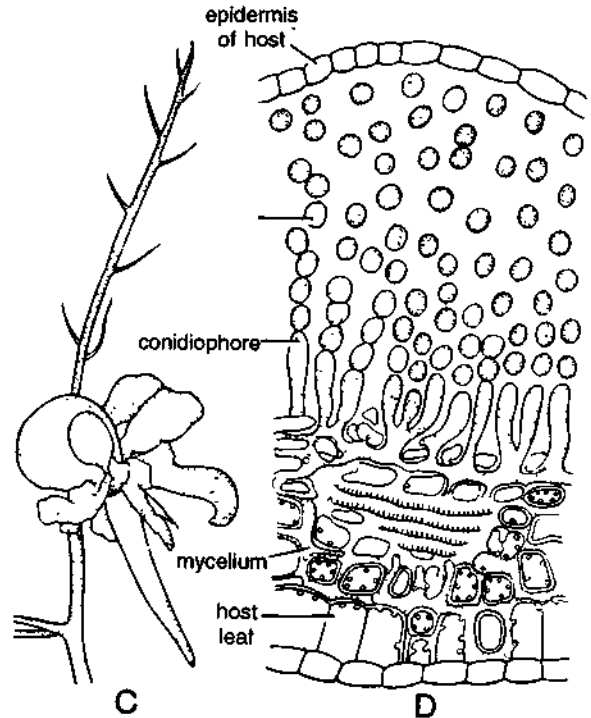
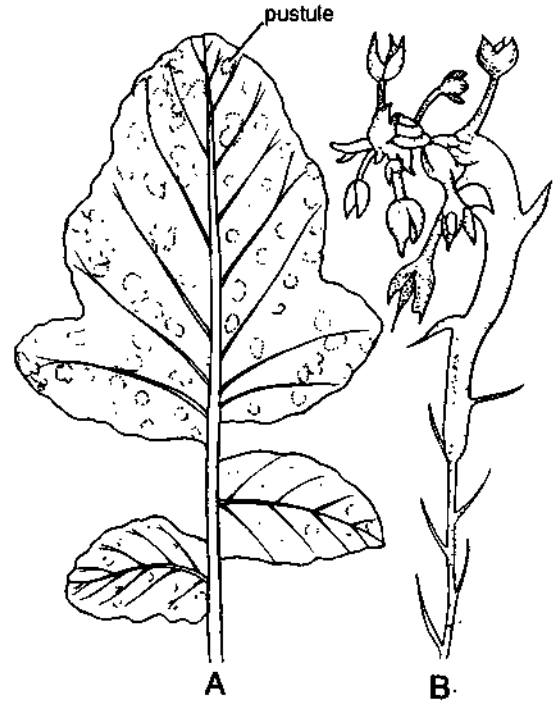
**Object :** Study of symptoms of white rust of crucifers.

#### Work procedure

Study the symptoms appearing on the leaf and stem of radish.

#### Comments

1. The disease is caused by *Albugo candida* (= *Cystopus candidus*) which is a phycomycetous fungus. *Raphanus sativus* (radish; vern. mooli; fam. Cruciferae) serves as host.
2. Other hosts include cabbage, cauliflower, mustard, toria, etc.
3. All the plant parts except the root are infected.
4. The first symptoms appear on the leaves as white shining pustules. These later coalesce to form large patches. The lower epidermis gets ruptured.
5. In case of severe infection leaves become fleshy, thickened and inrolled. The entire plant remains stunted.
6. Sometimes stems and flowers may also get infected. These show structural distortions due to hypertrophy.
7. The membranous floral parts like petals become very thick and fleshy.
8. The disease can be controlled by sanitation through destruction of weeds and infected plant debris, also by crop rotation and the use of Bordeaux mixture.



**Fig. 1.** White rust of crucifers. A. Leaf of *Brassica* (Mustard) infected with *Albugo candida*. B. Diseased inflorescence of *Brassica*. C. Diseased flower of *Brassica*. D. Section through pustules showing conidia.

**Exercise 2****Object : Study of causal organism.****Work procedure**

Study T.s. of infected host leaf.

**Comments**

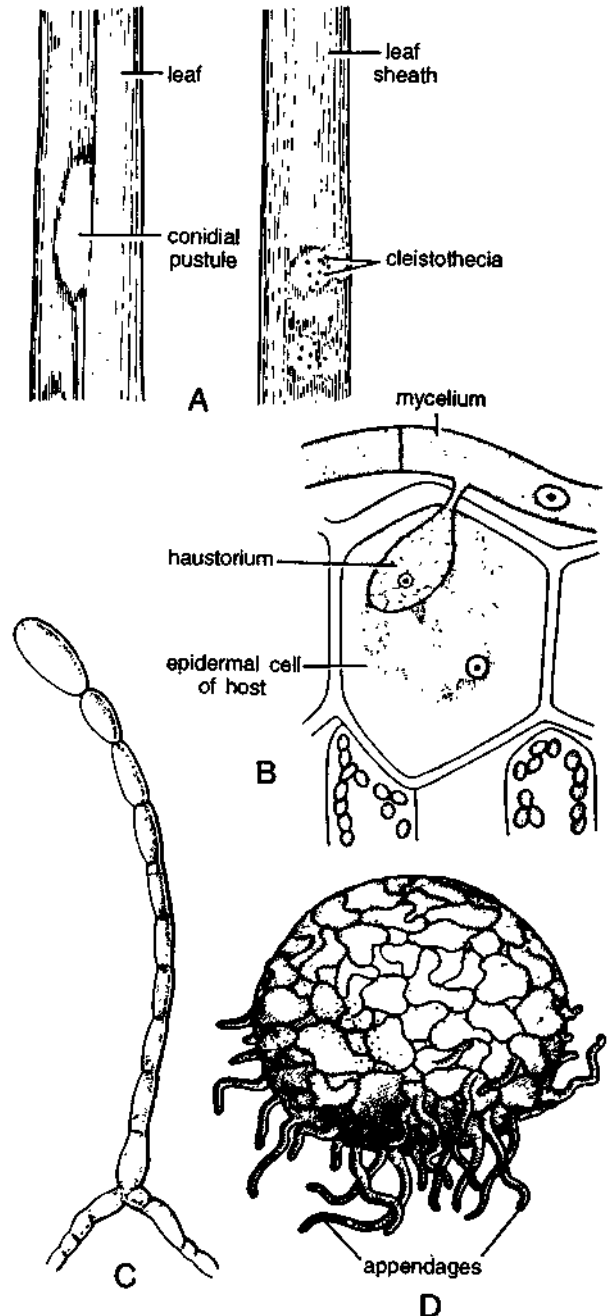
1. *Albugo candida* is the causal organism of White Rust of Crucifers.
2. The thallus consists of branched, intercellular and unseptate mycelium that penetrates the host cell by globular or knob-like haustoria.
3. Asexual reproduction takes place by conidia borne basipetally on conidiophores which are present in palisade-like manner just below the lower epidermis.
4. Conidia are hyaline and nearly spherical. Each germinates to produce four to eight biflagellate zoospores.
5. Sexual reproduction is oogamous and takes place at the end of growing season of the host.
6. Oogonia and antheridia are present in the intercellular spaces of the host tissue.
7. Globular, thick walled oospores with brown episporae are also found in intercellular spaces.

**Powdery Mildew of Barley****Exercise 1****Object : Study of symptoms of powdery mildew of barley.****Work procedure**

Study the leaves, stem and inflorescence for symptoms.

**Comments**

1. Powdery mildew of barley (*Hordeum vulgare*; vern. jaii; fam. Poaceae) is caused by ascomycetous fungus *Erysiphe graminis* var. *hordei*.
2. Infection occurs on leaves, stem and even inflorescence.



**Fig. 1. Powdery mildew of barley.** A. Conidial and cleistothecial stage on host leaves. B. Epidermal cell of the host showing haustoria. C. Conidiophore with a chain of conidia. D. Cleistothecium.

3. Numerous superficial colonies are formed on the upper surface of the leaves. The colonies are white in the beginning during conidial stage

but later turn grey or red due to cleistothecia formation.

4. The plants get stunted and number and leaf size also gets reduced.
5. The leaves still remaining attached to the plant are weak, twisted and deformed.
6. Field sanitation, foliar spray with Karathane and Calixin and use of resistant varieties are the major methods of disease control.

### Exercise 2

**Object :** Study of causal organism.

#### Work procedure

Pick up various stages in the form of mycelium covering the host surface, stain in cotton blue and mount in lactophenol. Study as many stages as possible.

#### Comments

1. Causal organism is an ascomycetous fungus, *Erysiphe graminis* var. *hordei*.
2. The thallus consists of branched and septate mycelium. The cells are uninucleate. The haustoria are elliptic with long finger-shaped appendages.
3. Conidiophores are short and bear chains of ellipsoid conidia.
4. The ascocarp is a cleistothecium. The peridium bears elongate and unbranched appendages.
5. There are eight ascospores inside the ascus. These are elliptic and sub-hyaline to pale brown.

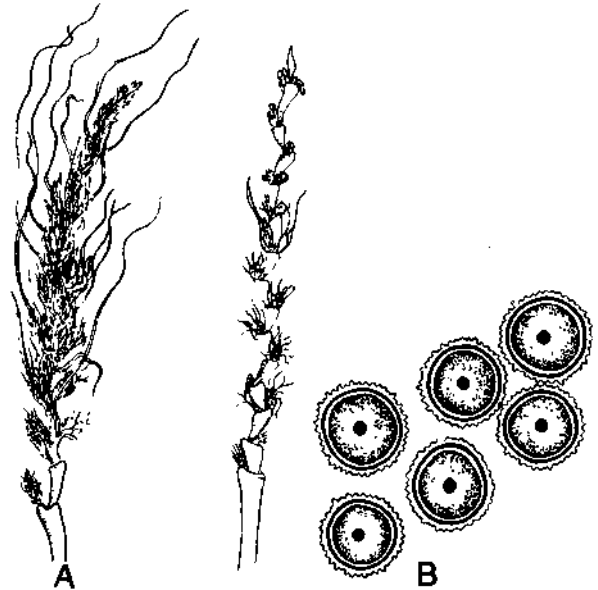
## Loose Smut of Wheat

### Exercise 1

**Object :** Study of symptoms of loose smut of wheat.

#### Work procedure

Observe the symptoms shown by a specimen of diseased wheat inflorescence.



**Fig. 1.** Loose smut of wheat. A. Wheat ears infected by *Ustilago tritici*. B. Chlamydo spores.

#### Comments

1. The loose smut of wheat (*Triticum aestivum*; vern. gehoon; fam. Poaceae) is caused by basidiomycetous fungus, *Ustilago tritici*.
2. The disease appears when the plant is in inflorescence (ear) stage.
3. Diseased ears come out of the bracts little earlier than the normal ones.
4. The flowers become brittle and get filled by powdery mass of spores.
5. The diseased ears bear deformed spikelets filled with black, dry, powdery mass of spores which replace all the floral parts and also the glumes.
6. Since the disease is seed-borne, some of the following control measures would be useful—hot water treatment, solar energy treatment, use of carboxin, benlate, etc. and breeding of resistant varieties.

### Exercise 2

**Object :** Study of causal organism.

#### Work procedure

Tap the infected ear on the slide to release spores. Mount in lactophenol and study.

**Comments**

1. *Ustilago tritici* causes the loose smut of wheat.
2. The spores, commonly called as chlamydospores are produced in large quantities in the inflorescence.
3. Chlamydospores or teliospores are pale olive with minute echinulate walls.

**Covered Smut of Barley****Exercise 1**

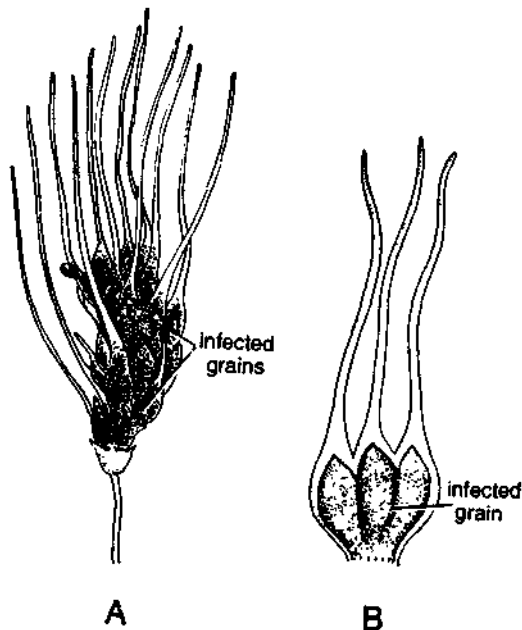
**Object :** Study of symptoms of covered smut of barley.

**Work procedure**

Study the diseased inflorescence (ear) of barley.

**Comments**

1. The covered smut of barley (*Hordeum vulgare*, vern. jau; fam. Poaceae) is caused by a basidiomycetous fungus, *Ustilago hordei*.
2. The disease is seen only when the blackened ears come out of leaf sheaths.



**Fig. 1.** Covered smut of barley. A. Ears infected by *Ustilago hordei*. B. Spikelet magnified.

3. All the ears on the plant and all the grains in all the ears are generally infected.
4. The ovaries are full of spore masses and grains are not formed.
5. The black spore masses are firmly held inside the persistent membranes of the grains and the basal part of the glumes.
6. Since the disease is externally seed-borne, seed treatment is effective. Use of ceresan and agrosan and use of resistant varieties help in controlling the disease.

**Exercise 2**

**Object :** Study of causal organism.

**Work procedure**

Mount the spores from diseased grains in lactophenol and study.

**Comments**

1. Causal organism is *Ustilago hordei* belonging to basidiomycetes.
2. The spores are round to elliptical and brown-black. The epispore is smooth.
3. The fungus is found inside the seed.

**Whip Smut of Sugarcane****Exercise 1**

**Object :** Study of symptoms of whip smut of sugarcane.

**Work procedure**

Study the specimen of the diseased sugarcane plant.

**Comments**

1. The whip smut of sugarcane (*Saccharum officinarum*; vern. ganna; fam. Poaceae) is caused by a basidiomycetous fungus, *Ustilago scitaminea*.
2. Floral axis of the affected plant becomes long, whip-like, dusty black shoot, often several feet long.
3. Floral shoot is curved on itself.

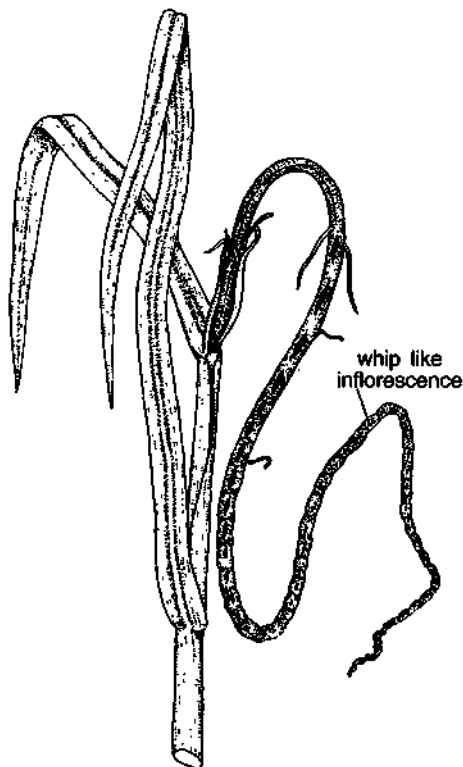


Fig. 1. Whip smut of sugarcane. Inflorescence infected by *Ustilago scitaminea*.

4. Whip-like floral shoot is covered with silvery thin membrane which soon flakes away.
5. The disease can be controlled by (i) removal of smutted whips, (ii) discouraging ratooning, (iii) disinfection of sets by chemicals such as mercuric chloride, formalin and fungicides like vitavax, benlate, etc., (iv) use of resistant varieties and (v) avoiding cultivation of susceptible varieties.

### Exercise 2

**Object :** Study of causal organism.

#### Work procedure

Dust away a few spores from the whip-like floral axis. Mount in glycerine or lactophenol and study.

#### Comments

1. The causal organism is *Ustilago scitaminea* of basidiomycetes.

2. Infection is due to intercellular mycelium found in the tissues of the cane below the whip smut.
3. Hyphae forms dense masses towards the surface of spore bearing shoot where spores are formed.
4. The spores are globose to sub-globose, reddish-brown and typically smooth or punctate.

## Black Stem Rust of Wheat

### Exercise 1

**Object :** Study of symptoms of black stem rust of wheat.

#### Work procedure

Study the diseased stems, leaf sheaths, leaves and ears of the infected wheat plants.

#### Comments

1. This disease of wheat (*Triticum aestivum*; vern. gehoon; fam. Poaceae) is caused by basidiomycetous fungus, *Puccinia graminis tritici*.
2. First to appear are uredosori which are found on culms, leaf sheaths and leaves.
3. Uredosori are large, elongated, coalescing and dehiscing early and result in breaking large pieces of epidermis.
4. Teleutosori appear later in the season. These are black in colour and burst early in the season.
5. Teleutosori are found on all green parts of the plant but least on leaf blades.
6. Control measures include eradication of infected plants, sanitation, mixed cropping, use of various chemicals like (i) sulphur dust, (ii) Dithane M-15, (iii) Nabam and  $ZnSO_4$ , (iv) antibiotics, (v) fungicides like vitavax, plantvax, etc. and (vi) use of resistant varieties.

### Exercise 2

**Object :** Study of causal organism.

#### Work procedure

Study the sections of the stem passing through uredosorus and teleutosorus.



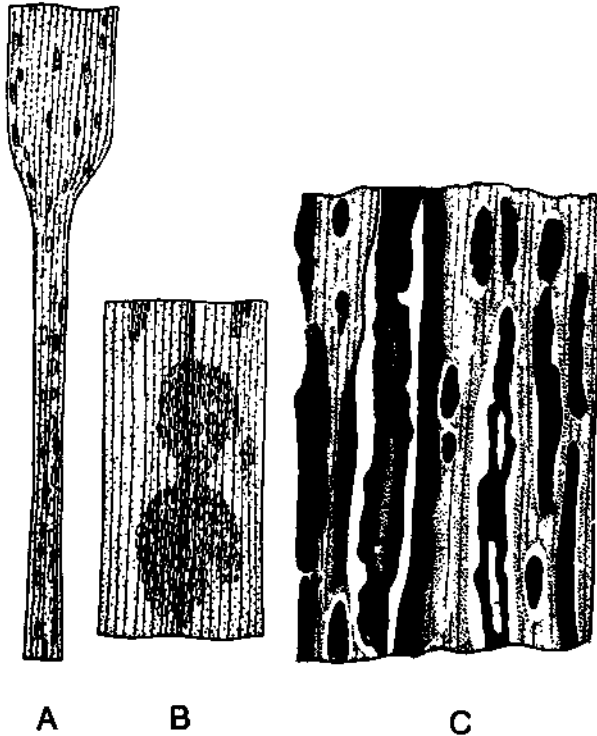


Fig. 1. Black stem rust of wheat. Infection caused by *Puccinia graminis tritici*. A. Leaf sheath showing pustules. B. A portion of leaf showing uredopustules. C. A portion of stem showing teleutopustules.

### Comments

1. The causal organism is *Puccinia graminis tritici*.
2. It is a heteroecious rust. The pycnidial and aecidial stages are found on *Berberis vulgaris*, an alternate host for the rust.
3. The pycnidial cups are flask-shaped. The floor of the flask bears pycniospores. Receptive hyphae is produced from near the ostiole of the flask.
4. Aecidial cups are found on the lower leaf surface. These are surrounded by periderm. Aeciospores which lie within the cup are echinulate and show six germ pores. Aeciospores are in chains arising from the base of the cup.
5. Uredosori and teleutosori are produced on wheat plant.
6. Uredosori are groups of brown and oval uredospores. Each spore has a single thick wall with small spines and four equatorially placed germ pores.

7. Teleutosori are groups of brown and two-celled teleutospores. Each teleutospore has a thick and smooth wall with rounded or sometimes pointed apex. Each cell has a germ pore.

## Brown (Orange) Rust of Wheat

### Exercise 1

**Object :** Study of symptoms of brown rust of wheat.

### Work procedure

Study the leaves of diseased plant.

### Comments

1. Brown rust of wheat (*Triticum aestivum*; vern. gehoon; fam. Poaceae) is caused by

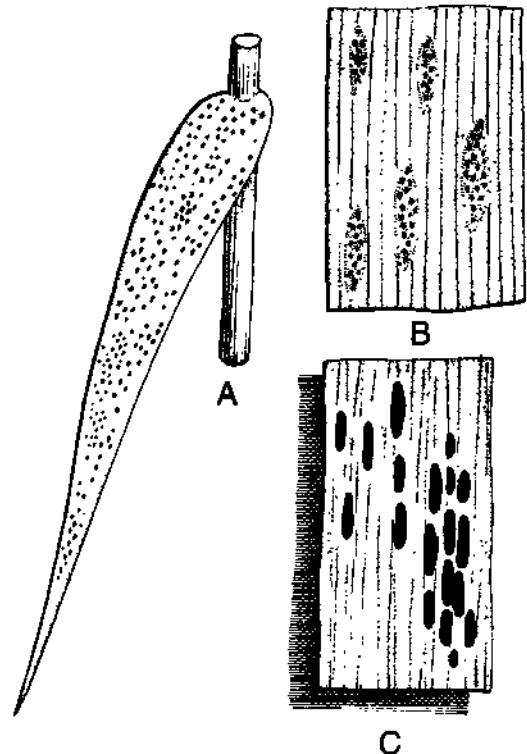


Fig. 1. Brown (orange) leaf rust caused by *Puccinia recondita*. A. Leaf showing pustules. B. A portion of leaf showing uredopustules. C. A portion of leaf showing telutopustules.

basidiomycetous fungus, *Puccinia recondita*, earlier known as *P. trititica*.

- The rust is more common to northern and eastern parts of India than to Peninsular India.
- Pycnial and aecial stages are present on *Thalictrum* spp. which is an alternate host for the rust.
- Uredospores and teleutospores are found on the leaves of wheat plant.
- Uredosori or uredopustules are very common on the leaves and are rare on leaf sheath and stalks.
- These appear as bright orange coloured spots when burst open.
- Uredosori may either be irregularly scattered or present in small clusters.
- Uredospores are brown, spherical, with finely echinulate epispore and 3-4 germ pores scattered on the surface.
- Teleutosori are rarely formed. These appear on the lower leaf surface as small, oval or linear, black groups covered by epidermis.
- Paraphyses are abundantly present which divide the sorus into many chambers.
- Control measures include eradication of infected plants, sanitation, mixed cropping, use of various chemicals like (i) sulphur dust, (ii) Dithane M-15, (iii) Nabam and ZnSO<sub>4</sub>, (iv) antibiotics, (v) fungicides like vitavax, plantvax, etc. and use of resistant varieties.

### Exercise 2

**Object : Study of causal organism.**

#### Work procedure

Study the T.S. of leaf through uredosori and teleutosori

#### Comments

- The pathogen that causes Brown or orange leaf rust of wheat is *Puccinia recondita*.
- Uredospores occur in uredosori or uredopustules. These are brown and spherical, and epispore is finely echinulate with 3-4 germ pores.
- Teleutospores are oblong to cuneiform and slightly constricted at the septum. The apex is rounded with prominent thickenings.

## Rust of Linseed

### Exercise 1

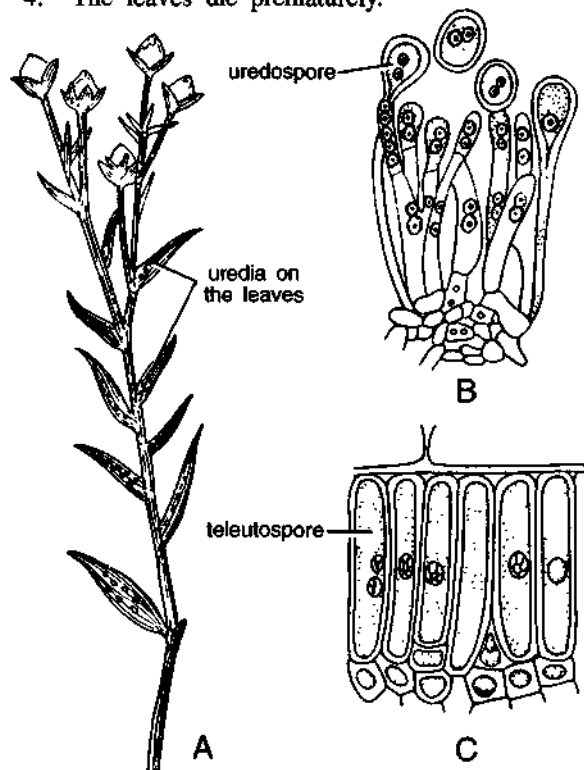
**Object : Study of symptoms of rust of linseed.**

#### Work procedure

Study the diseased plant specimen of linseed.

#### Comments

- The rust of linseed (*Linum usitatissimum*; vern. als; fam. Linaceae) is caused by a basidiomycetous fungus, *Melampsora lini*.
- The affected plants become bright orange in colour due to the presence of large number of uredosori.
- Uredosori occur on both the leaf surfaces and other aerial parts of the plant.
- The leaves die prematurely.



**Fig. 1.** Rust of linseed caused by *Melampsora lini*. A. Diseased linseed plant, B. Uredosorus with uredospores, C. Teleutosorus with teleutospores.

5. Teleutosori appear later and are formed on the stem. These are brown to black crusts covered by epidermis.
6. Control measures include sanitation, use of resistant varieties, seed treatment with chemicals to inactivate teleutospores, etc.

### Exercise 2

**Object : Study of causal organism.**

#### Work procedure

Study T.s. of leaf passing through uredosori and teleutosori.

#### Comments

1. The pathogen causing rust of linseed is *Melampsora lini*, of basidiomycetes.
2. It is an autoecious rust.
3. About 5-10 amphigenous pycnia are grouped together.
4. Aecia lack peridium, are epiphyllous and surround pycnia to form ring.
5. Uredia are amphigenous and irregularly scattered. Paraphyses occur mixed with uredospores which are ellipsoidal or obovoidal. The wall is hyaline and finely echinulate.
6. Telia are amphigenous, circular or elongated along the stems, often fused and covering large areas, subepidermal and black.

## Early Blight of Potato

### Exercise 1

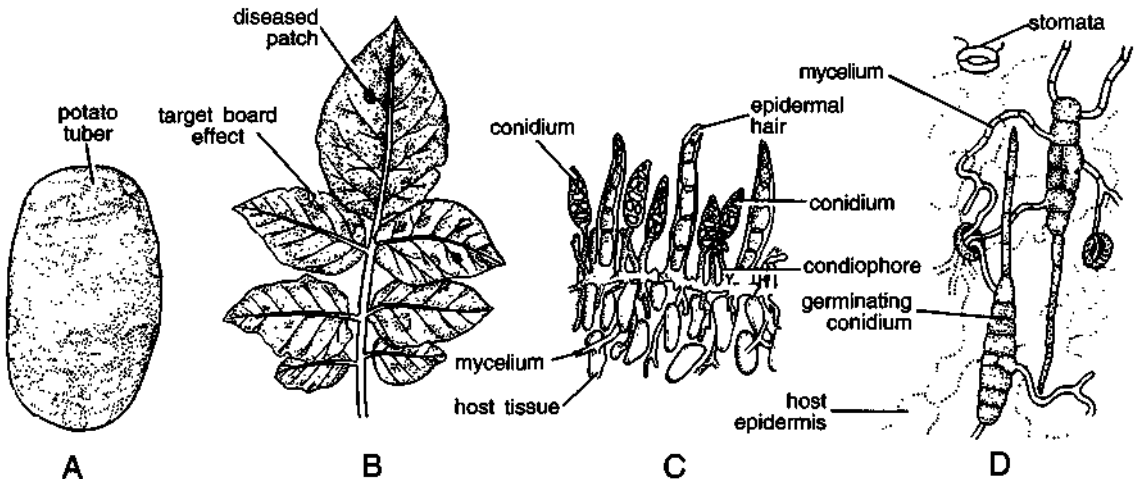
**Object : Study the symptoms of early blight of potato.**

#### Work procedure

Study the symptoms appearing on leaves, stem and also tubers.

#### Comments

1. Early blight of potato (*Solanum tuberosum*; vern. aloo; fam. Solanaceae) is caused by deuteromycetous fungus, *Alternaria solani*.
2. This common disease of potato causes serious damage to the crop.
3. The disease appears earlier than the late blight of potato in almost all climatic conditions.
4. Certain other plants like tomato, chillies, egg plant, etc. are also infected by the same pathogen.
5. The disease appears first on lower leaves as small, isolated, scattered, pale brown spots on leaflets.
6. Later, the spots become covered with deep greenish blue growth of fungus.
7. In necrotic spots, concentric rings appear on older leaves and darkened areas on the stem,



**Fig. 1.** Early blight of potato caused by *Alternaria solani*. A. Diseased potato tuber, B. Diseased leaf. C. T.s. Infected leaf showing conidiophores and conidia. D. Conidia and conidiophores.

- giving target board-like effect. A narrow chlorotic zone surrounds the spots.
8. In severe attacks, leaves shrivel and fall down. All the aerial portions of the plant appear blighted.
  9. Tubers may also get infected. These develop brown to black necrotic lesions on the skin.
  10. Disease is soil borne and can be controlled by sanitation, crop rotation and use of fungicides like dithane, M-15, Blitox, Zineb, etc.

### Exercise 2

**Object :** Study causal organism.

### Work procedure

Study T.s. of infected leaf of potato.

### Comments

1. *Alternaria solani*, a deuteromycetous fungus causes early blight of potato.
2. Mycelium consists of septate, branched, light brown hyphae becoming darker with age.
3. The hyphae is intercellular but also penetrates the host tissue.
4. Conidiophores emerge out of stomata from the dead centre of spots.
5. Conidia are formed in chains. These are beaked, muriform and dark coloured.
6. Mature conidia show both transverse and longitudinal septa.

## Tikka Disease of Groundnuts

### Exercise 1

**Object :** Study of symptoms of tikka disease.

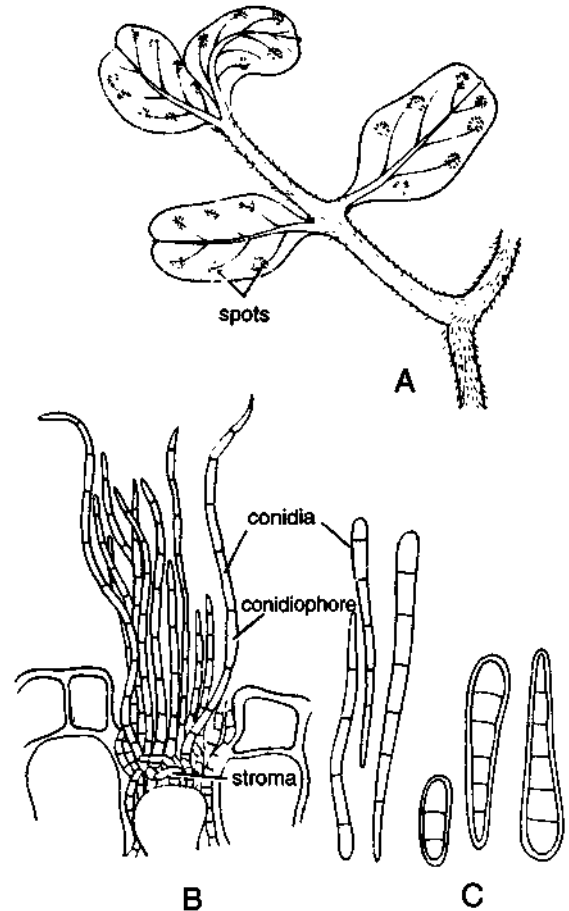
### Work procedure

Study the aerial parts of the plant and note down symptoms.

### Comments

1. Tikka disease of groundnut (*Arachis hypogaea*; vern. 'moongphali'; fam. Papilionaceae) is caused by *Cercospora personata* and *C. arachidicola*.

2. Symptoms first appear in one or two month old plants.
3. The leaves show excessive spotting causing defoliation. This results in fewer and smaller nuts.
4. The first symptoms appear in the form of pale areas on the upper surface of older leaves.
5. Circular or irregular reddish brown to brown lesions are formed. These are larger in size on the lower leaf than on the upper.
6. The spots are surrounded by a narrow yellow halo. These are indistinct on the lower surface than on the upper.
7. Disease is soil borne and can be controlled by crop rotation, seed treatment with agrosan GN and  $\text{CuSO}_4$ , and fungicides like Bordeaux mixture, Dithane M-15, Benlate, etc.



**Fig. 1.** Tikka disease of ground nut caused by *Cercospora*. A. Symptoms of disease on the leaf. B. T.s. of host leaf showing conidia and conidiophores. C. Conidia.

**Exercise 2****Object : Study of causal organism.****Work procedure**

Study T.s. of the leaf through spots or tease the spots to take out conidia.

**Comments**

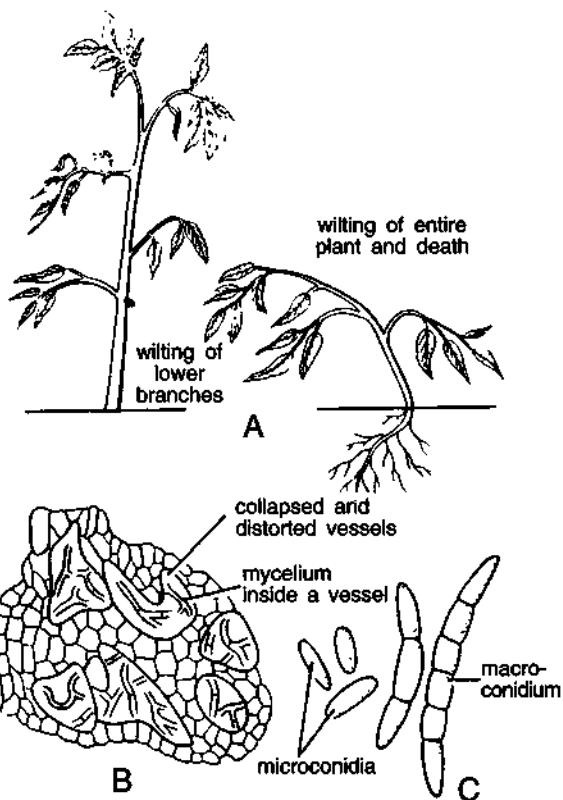
1. *Cercospora personata* and *C. arachidiicola* are the causal organisms of Tikka disease of groundnut.
2. The mycelium is septate and branched. Initially it is intercellular but becomes intracellular later after the host cells have been killed.
3. The mycelium enters the host when it is about 2-4 weeks old. Of the two pathogens, *C. personata* is more damaging because it produces more spots, spreads faster and causes earlier defoliation.
4. Conidiophores are septate and geniculate. Conidia leave definite scars on the conidiophores as they fall off.
5. Conidia are hyaline or pale yellow, obclavate and 1-12 septate.

**Wilt of Cotton****Exercise 1****Object : Study of symptoms of wilt of cotton.****Work procedure**

Study the entire plant for symptoms of the disease.

**Comments**

1. Wilt of cotton (*Gossypium* sp.; vern. kapas; fam. Malvaceae) is caused by a deuteromycetous fungus, *Fusarium oxysporum* f. sp. *vasinfectum*.
2. This is a very common disease of cotton in almost all the black cotton soil regions of the country.
3. The plants are infected during all the stages of plant growth.
4. The first symptoms appear on young seedlings. These are vein clearings of the leaves followed by interveinal tissue necrosis.



**Fig. 1. Wilt of cotton caused by *Fusarium*. A. Symptoms B. Mycelium in vessels. C. Macroconidia and microconidia.**

5. Cotyledons show yellowing and browning.
6. A brown ring is formed on the petiole.
7. The leaves become flaccid resulting into dropping and wilting starting from the oldest leaves at the base and proceeding upwards, finally involving branches and the whole plant. Defoliation is very common.
8. The stem is discoloured. The basal portion becomes black. In adult plants discolouration may be partial.
9. The control measures include field sanitation, crop rotation, mixed cropping and use of disease resistant varieties.

**Exercise 2****Object : Study of causal organism.****Work procedure**

Observe the mycelium that grows on surface of the stem. Also cut T.s. of stem near the base, stain in

cotton blue and mount in lactophenol. Study the mycelium and conidia.

### Comments

1. Causal organism is a deutermycetous fungus, *Fusarium oxysporum* f. sp. *vasinfectum*.
2. The aerial mycelium is white to greyish forming a mat on the collar region of the stem near the ground level.
3. Mycelium is both inter- and intracellular, profusely branched and septate. The stroma is brownish-white to violet and plectenchymatous.
4. Two types of conidia are produced—macroconidia and microconidia.
5. Macroconidia are present on cushion-like sporodochia. These are long crescent-shaped and multiseptate.
6. Microconidia are very small, unicellular or 1 or 2 septate, spherical, elongated or crescent-shaped.
7. Chlamydo spores are also formed either terminally or in intercalary position.

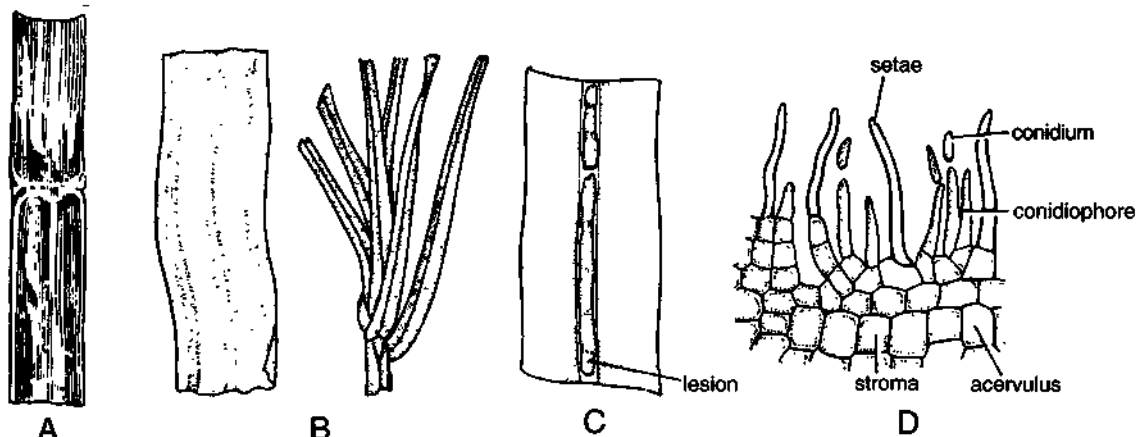
## Red Rot of Sugarcane

### Exercise 1

**Object :** Study the symptoms of red rot of sugarcane.

### Work procedure

Study all the aerial parts of the plant for symptoms.



**Fig. 1.** Red rot of sugarcane caused by *Colletotrichum falcatum*. A. Symptoms on stem, B. Leaves, C. Lesions on leaves, D. Acervulus with conidia.

### Comments

1. Red rot of sugarcane (*Saccharum officinarum*; vern. ganna; fam. Poaceae) is caused by deuteromycetous fungus, *Colletotrichum falcatum*.
2. It is a serious disease of sugarcane in subtropical conditions. Serious epiphytotics have occurred in U.P. and Bihar during 1939-1940 and 1946-1947 respectively.
3. The symptoms of the disease occur on all aerial parts being more prominent on stem and midrib areas of leaves.
4. The early symptoms show drooping of leaves and loss of colour.
5. Later the cane becomes completely rotted within, loses its natural bright colour, becomes dull and shrinks at node.
6. The leaves now begin to wither completely and droop.
7. The split stems show longitudinally reddened internodal tissue, generally at base. The characteristic symptom of the disease is formation of cross bars in reddened areas.
8. Juice often gives bad odour due to conversion of sucrose into glucose and alcohol.
9. On the leaves, infection appears in the midrib as dark reddish area that elongates rapidly forming blood red lesions whose margins become darker. In old lesions, the centre becomes straw coloured.
10. The lesions get covered with powdery mass at the time of reproduction of pathogen.

11. The control measures include field sanitation, use of healthy sets, discouraging ratooning, crop rotation, use of resistant varieties, etc.

### Exercise 2

**Object :** Study of causal organism.

#### Work procedure

Cut T.s. of the host leaf or tease the conidia from the infected spots.

#### Comments

1. The pathogen responsible for red rot is *Glomerella tucumanensis* (= *Physalospora tucumanensis*) a deuteromycetous fungus, that shows perfect stages.
2. The conidial stage of pathogen is, however, very common and is formed by *Colletotrichum falcatum*.
3. The fungus produces conidia from conidiophores arranged in acervuli. These are minute, black, dot-like bodies found on the host surface.
4. Acervuli are rounded or elongated. These consist of setae and conidiophores bearing conidia.
5. Setae are 1-4 septate and swollen at the base with tapered to rounded tip. These are interspersed with conidiophores.
6. Conidiophores are septate and bear conidia at their tips.
7. Each conidium is one celled with large oil globule in the centre.

## Bacterial Blight of Rice (Paddy)

### Exercise 1

**Object :** Study of symptoms of bacterial blight of rice.

#### Work procedure

Study the symptoms of the disease appearing on leaves.

(B-14)

#### Comments

1. Bacterial blight of rice (*Oryza sativa*; vern. chawal, dhaan; fam. Poaceae) is caused by a bacterium, *Xanthomonas oryzae*.
2. The pathogen is a rod-shaped, monotrichous, gram negative and non-spore forming bacterium.
3. The disease appears as water soaked spots on the margins of the fully developed lower leaves.
4. The lesions are large and yellow with wavy margins.
5. In advanced stage lesions cover the entire leaf blade.
6. Other symptoms include drying of the leaf tip and inward rolling and twisting of leaf blade, yellowish stripes on the leaf and marginal necrosis.
7. The control measures include seed treatment with Argimycin (antibiotic) and Ceresan and use of resistant varieties.

## Citrus Canker

### Exercise 1

**Object :** Study the symptoms of citrus canker.

#### Work procedure

Study the diseased parts of the plant such as leaves, twigs, fruits.

#### Comments

1. Citrus canker is produced by a bacterium *Phytoplasma citri* (= *Xanthomonas citri*) on citrus (*Citrus aurantifolia*; vern. kagzi nimboo; fam. Rutaceae).
2. Bacterium causing this disease is a short bacillus (rod-shaped), monotrichous and strictly aerobic.
3. It produces corky outgrowth (cankers) on any aerial part of the plant. Mostly leaves, twigs, young branches and fruits are infected.
4. Canker on the leaves appears first on the underside. In the beginning it appears as convex protuberance of dark green colour but later, spots become white-greyish and finally rupture.

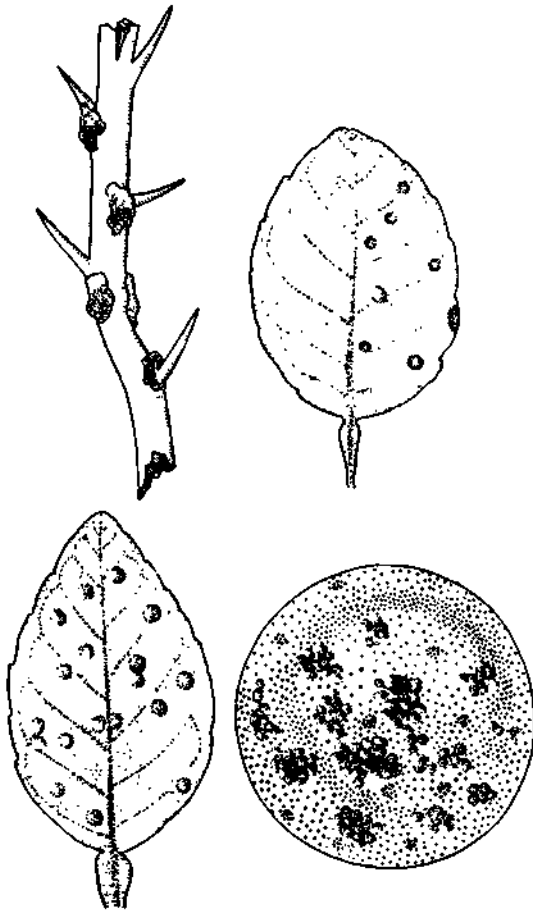


Fig. 1. Citrus canker. Twig, leaves and fruit, showing symptoms.

5. The spongy and rough formations remain scattered irregularly or several coalesce to form irregular scabby mass.

## Tundu Disease of Wheat

### Exercise 1

**Object :** To study the symptoms of Tundu disease of wheat.

#### Work procedure

Study the infected parts of the plant.

#### Comments

1. This disease is caused by *Phytomonas tritici*. Wheat (*Triticum* sp.; vern. gehun; fam. Graminae) serves as a host.
2. The disease causing organism is a gram positive and monotrichous bacillus.
3. Symptoms show twisting of stem, distortion of ear heads and rotting of spikelets.
4. Some yellow liquid oozes out from the affected parts and, therefore, disease is also known as 'yellow ear rot'.
5. The liquid collects over the ears and checks the growth of the plant. For this reason diseased plants remain stunted.
6. Heads are also abnormally small and slender, and flowers do not develop.

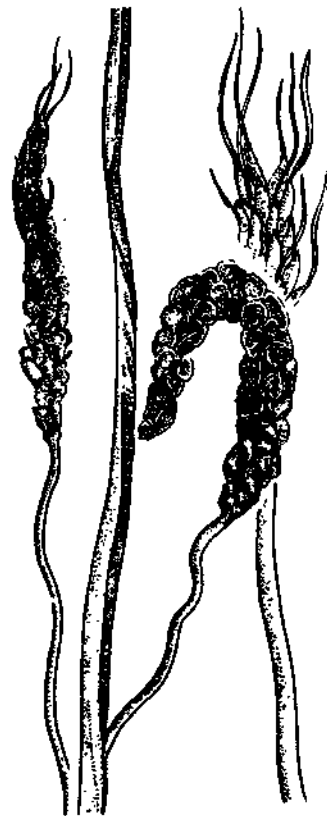


Fig. 1. Tundu disease of wheat. Wheat plant showing symptoms.



## Leaf Curl of Potato (Potato Leaf Roll)

### Exercise 1

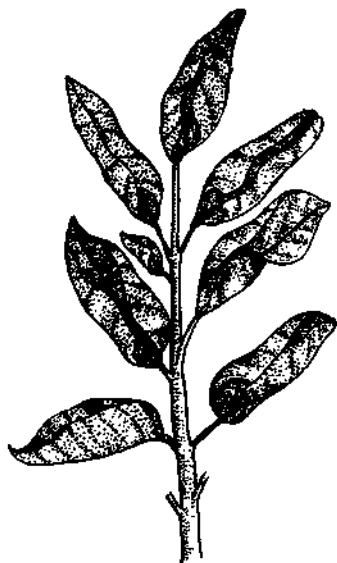
**Object :** Study the symptoms of leaf curl of potato.

#### Work procedure

Study the symptoms of disease appearing on the leaves.

#### Comments

1. Leaf curl virus or potato virus I or *Solanum virus 14*, causes serious disease of potato.
2. The most common symptom is the characteristic rolling of leaves. Leaves curl from the margins towards the mid-rib and in case of severe infection become almost tubular. The texture of the leaves also changes.
3. Colour of the foliage changes from dark green to yellowish.
4. The plants get affected in their growth. It may assume either spreading or abnormally erect V-shaped form.



**Fig. 1. Leaf curl of potato.** Potato leaf showing symptoms.  
(B-14)

5. Reduction in the size of tubers takes place. Net necrosis of tubers is also known. (The tubers in this case show a network of brown strands, particularly near the stem end of the tuber).
6. The virus is transmitted by an aphid, *Myzus persicae*.

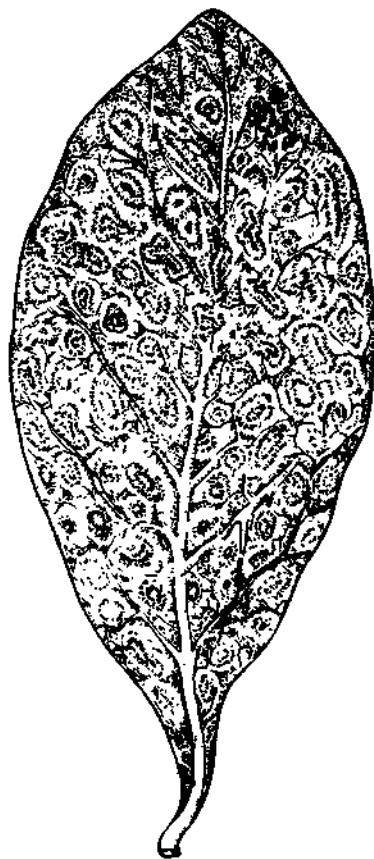
## Tobacco Mosaic

### Exercise 1

**Object :** Study the symptoms of tobacco mosaic.

#### Work procedure

Study the specimen showing symptoms of the disease.



**Fig. 1. Tobacco mosaic.** Leaf of tobacco showing symptoms.

**Comments**

1. About 200 species of plants belonging to about 36 botanical families are attacked. Solanaceae represents one-third of the recorded hosts. All species of tobacco (*Nicotiana*) are susceptible.
2. The disease is caused by *Nicotiana virus I*.
3. Symptoms exhibited are, leaf mottling of light and dark green patches, distortion of leaves, unnatural and irregular leaf shape, stunting of the whole plant, variegation, mosaic patterns, necrosis, etc.
4. The first symptoms to appear are clearing of veins, forming mosaic patterns, characteristic mottling and ultimately distortion.
5. The virus is sap transmissible and enters the host through wounds.

**Leaf Curl of Tobacco**

**Exercise 1**

**Object :** Study of symptoms of leaf curl of tobacco.

**Work procedure**

Study the symptoms of disease shown by tobacco plant.

**Comments**

1. Tobacco (*Nicotiana tabacum*) acts as a host. The causal organism is potato virus X.



**Fig. 1.** Leaf curl of tobacco. Tobacco plant showing symptoms.

2. Tobacco plants of the tropics are severely attacked.
3. Leaf curl occurs through the agency of white-fly *Bemisia gossypiperda* which acts as vector.
4. The symptoms display badly curled, twisted and distorted leaves.

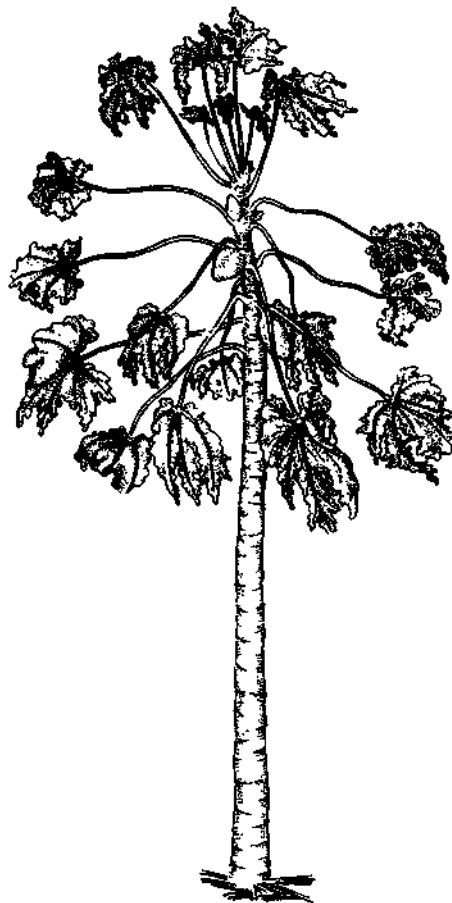
**Leaf Curl of Papaya**

**Exercise 1**

**Object :** Study the symptoms of leaf curl of papaya.

**Work procedure**

Study the symptoms of the disease shown by papaya plant.



**Fig. 1.** Leaf curl of papaya. Plant of papaya showing symptoms.