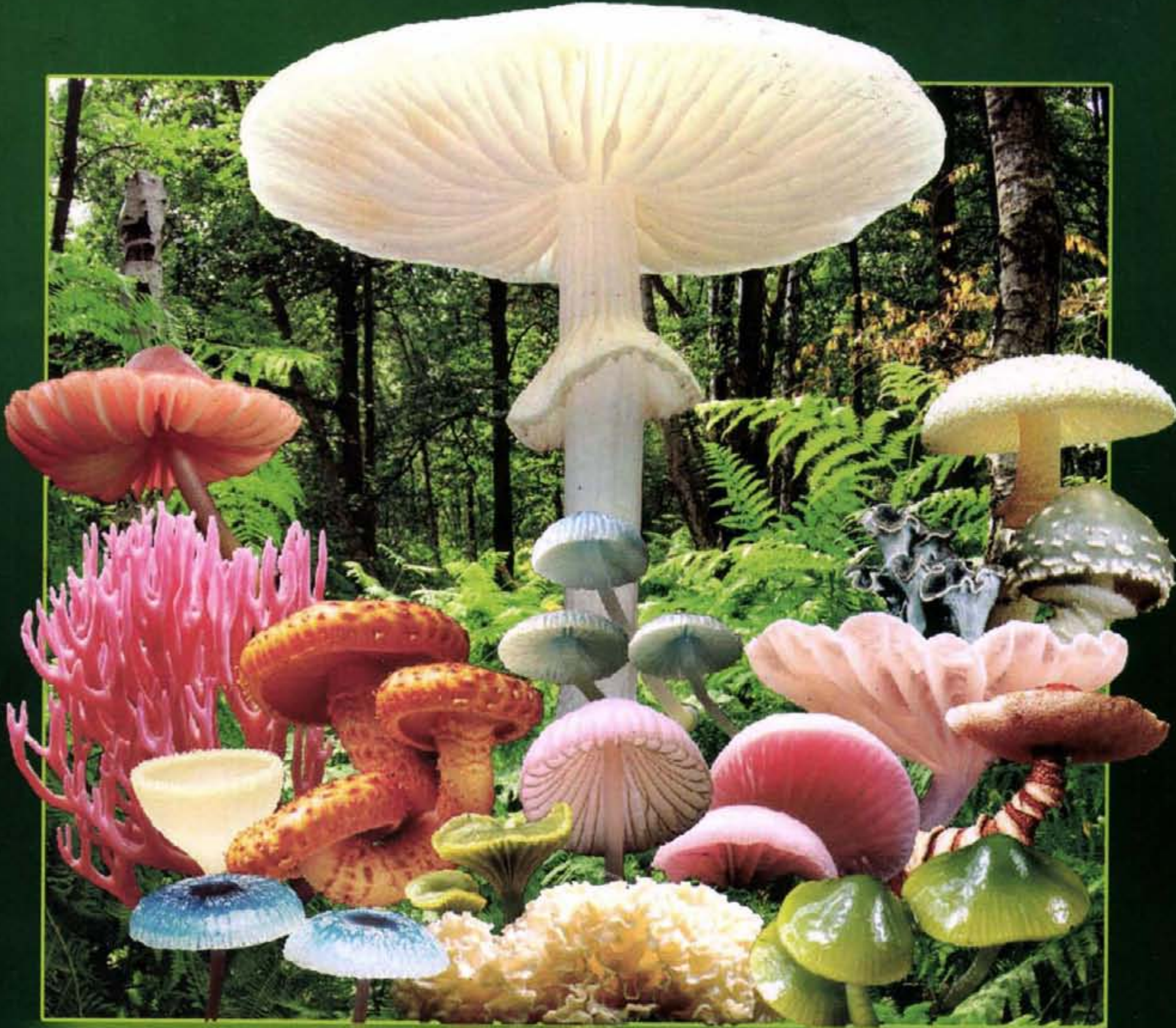

A Text Book of
Practical Botany 1

Bendre • Kumar

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Practical Botany -1

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

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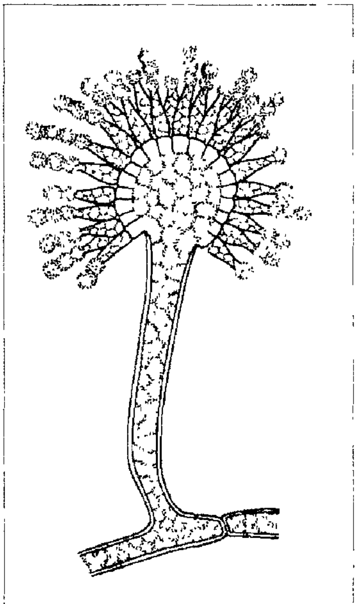

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Introduction to Laboratory

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.

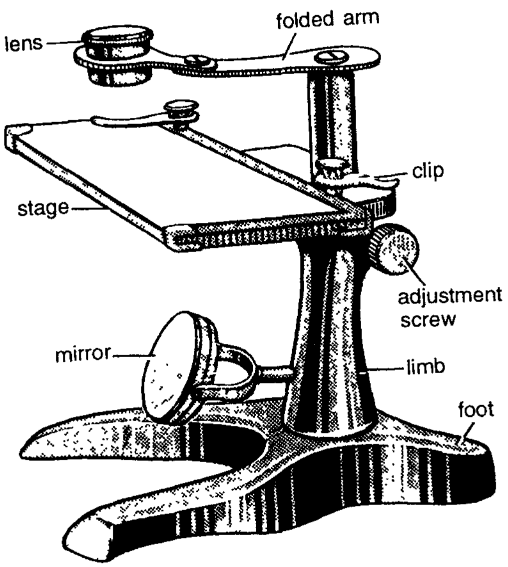


Fig. 1. A dissecting microscope.

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.

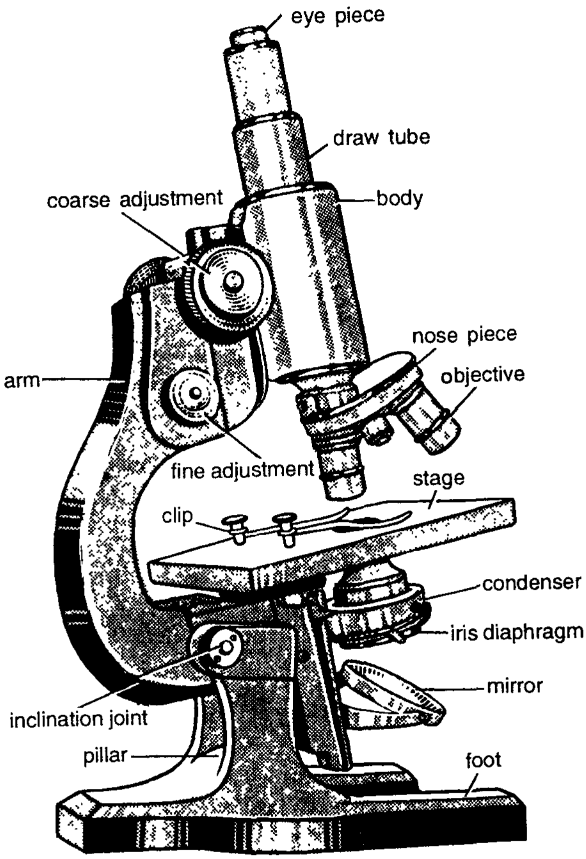


Fig. 2. Compound microscope.

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.

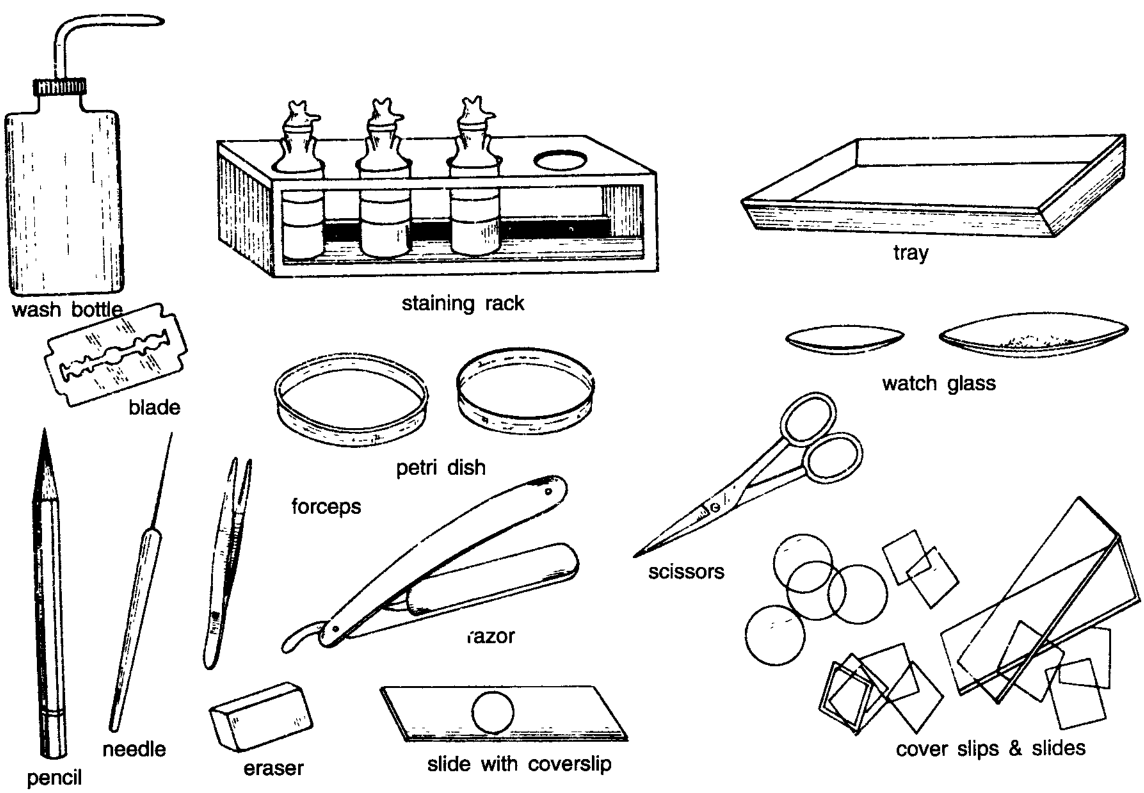


Fig. 3. Some laboratory provisions and necessary instruments.

2. Microscope is kept covered when not in use. Proper wodden 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,

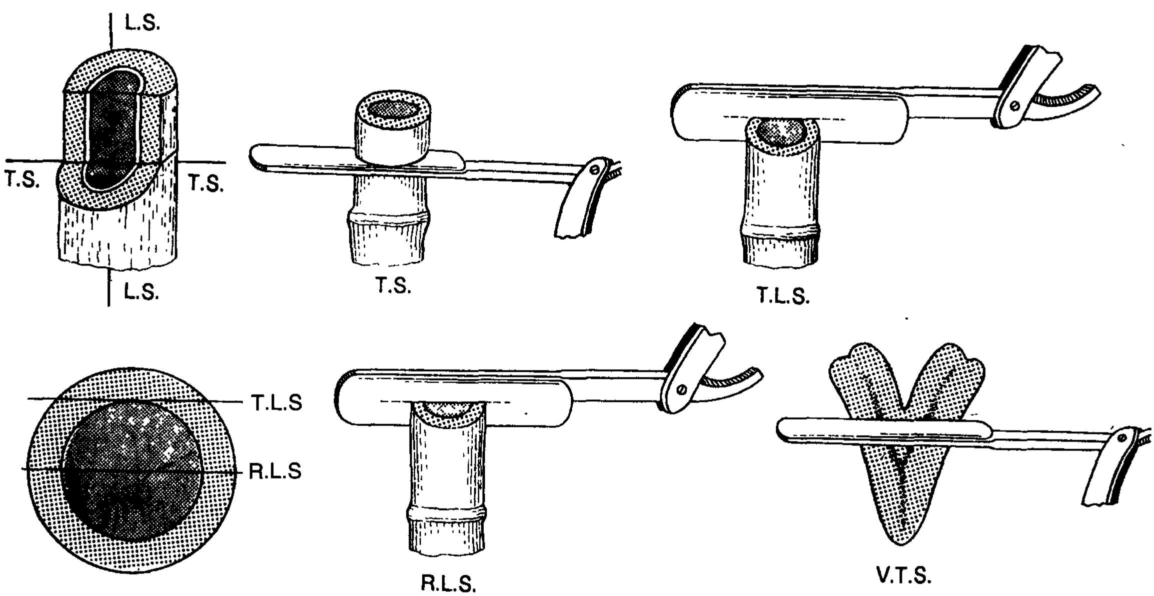


Fig. 4. Planes for section cutting.

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.

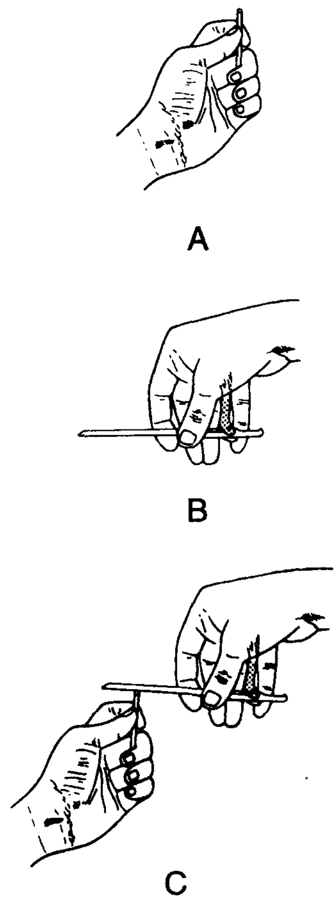


Fig. 5. Method of section cutting. A. holding the material, **B.** right way of holding the razor, **C.** holding the material and stroke of the razor.

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.

Achromatic figure
- Aniline blue
- Erythrosine
- Fast green
- Light green

Cellulose cell wall
- Aniline blue
- Delafield hematoxylin
- Fast green
- Light green

Lignified cell wall
- Crystal violet
- Safranin

Suberised cell wall
- Safranin

Cytoplasm
- Aniline blue
- Erythrosine
- Fast green
- Light green

Cutinised cell wall
- Crystal violet
- Erythrosine
- Safranin

Callose
- Aniline blue

Chitin
- Safranin

Proteins
- Safranin

Mitochondria
- Crystal violet

Plastids
- Crystal violet
- Iron hematoxylin

Nucleus
- Crystal violet
- Hematoxylin
- Safranin

Chromosomes
- 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 ↓ Stain with hematoxylin ↓ Wash with water ↓ Wash with ammonia water till stain turns blue (tap water is suitable if alkaline) ↓ Wash with water ↓ Stain with safranin ↓ Wash with glycerine ↓ Mount in glycerine	Select a section ↓ Stain with safranin (for 4-5 minutes) ↓ Wash with water ↓ Destain with acid alcohol if necessary ↓ Wash repeatedly with water ↓ Stain with fast green or aniline blue (for about a minute) ↓ Wash with 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 turpentines and synthetic resins are also used.

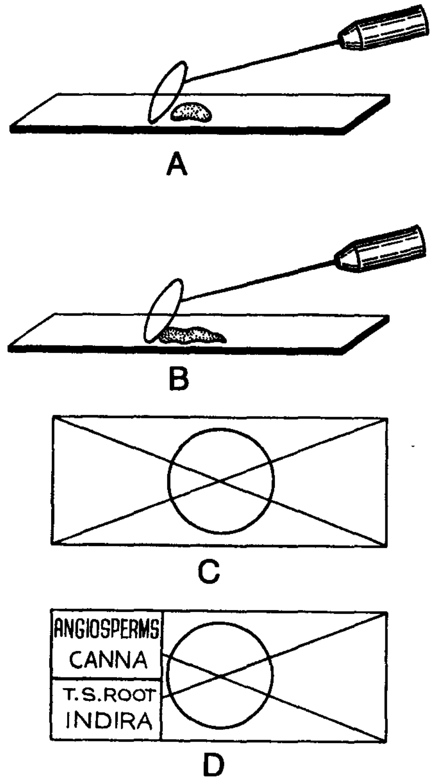


Fig. 6. Method of mounting coverslip.

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

1. 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.
2. 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.
3. 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

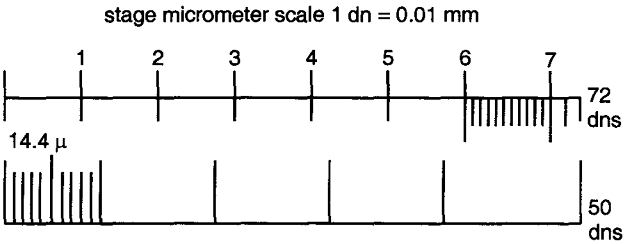


Fig. 7. Matching ocular micrometer with stage micrometer.

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 10μ (1 mm = 1000 microns or μ).

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 45x objective and 10x eye piece are used, divisions of ocular micrometer are found equal to 72 divisions of stage micrometer.
6. Callbrate the ocular micrometer as given below.
   Stage micrometer scale :
   100 dns = 1 mm (=100μ or microns*)
   1 dn = 0.01 mm (=10μ or microns)
   If, 50 dns (ocular micrometer)
   = 72 dns (stage micrometer)
   then, 50 dns (ocular micrometer)
   = 0.72 mm (=720μ or microns)
   therefore, 1dn (ocular micrometer)
   = 0.14 mm (=14.4μ 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 45x and eye piece 10x are used), each division of ocular (micrometer) would measure the distance of 14.4μ 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 × 14.4μ) 28.8μ.

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 milimeter = 1,000μ.

μ, 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 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.

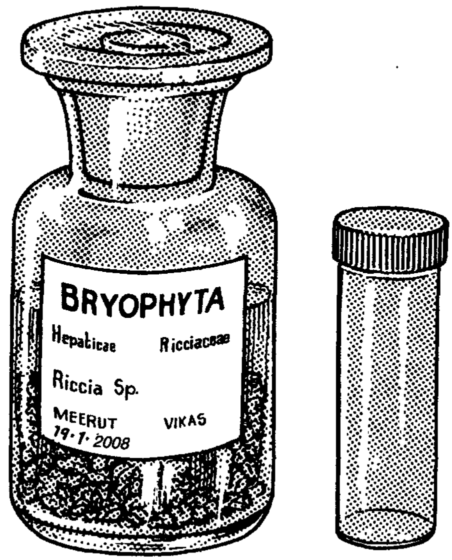


Fig. 8. Collection bottles.

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.

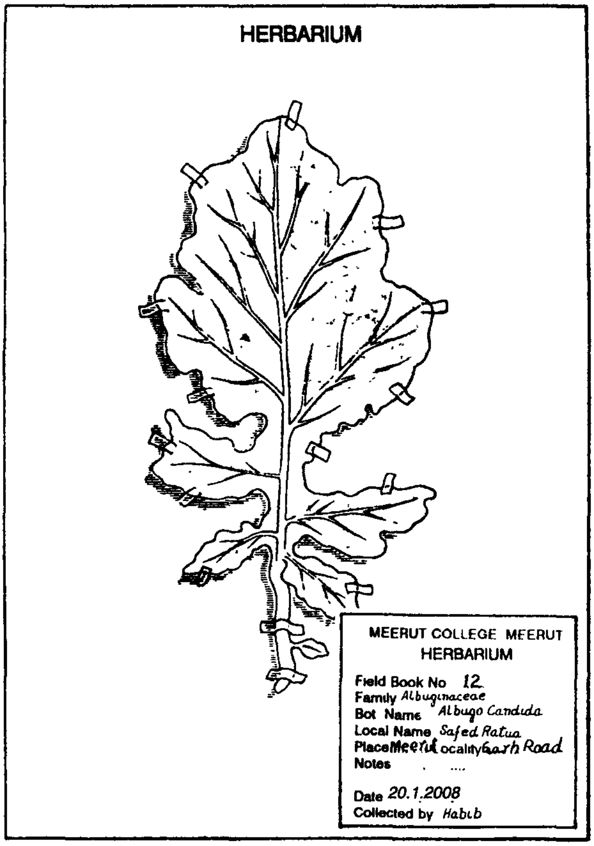


Fig. 9. A typical herbarium sheet.

[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 16½ × 11½ inches. However, 16 × 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 × 12 or 17 × 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.

[II] 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 virescence*, 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. **Caulerpaceae**
(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
(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 subterate to alate with midrib but not foliar
(2) Vesicle when present inercalary
(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 multiaxial
(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 multiaxial
(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 mutiaxial
(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	*Chlamydomonas*
			2. Volvocaceae	*Pandorina*
				Eudorina
				Pleodorina
				Volvox
		2. Chlorococcales	1. Chlorellaceae	*Chlorella*
			2. Hydrodictyaceae	*Pediastrum*
				Hydrodictyon
			3. Coelastraceae	*Scenedesmus*
		3. Ulotrichales	1. Ulotrichaceae	*Ulothrix*
			2. Ulvaceae	*Ulva*
				Enteromorpha
		4. Cladophorales	1. Cladophoraceae	*Cladophora*
		5. Chaetophorales	1. Chaetophoraceae	*Draparnaldiopsis*
				Frischiella
			2. Coleochaetaceae	*Coleochaete*
		6. Oedogoniales	1. Oedogoniaceae	*Oedogonium*
		7. Zygnematales	1. Zygnemataceae	*Spirogyra*
				Zygnema
			2. Desmidiaceae	*Cosmarium*
			3. Mesotaeniaceae	*Netrium*
		8. Siphonales	1. Caulerpaceae	*Caulerpa*
			2. Codiaceae	*Codium*
		9. Charales	1. Characeae	*Chara*
				Nitella
2. Xanthophyceae		1. Heterosiphonales	1. Botrydiaceae	*Botrydium*
			2. Vaucheriaceae	*Vaucheria*
3. Bacillariophyceae		1. Centrales		*Melosira*
		2. Pennales		*Pinnularia*
4. Phaeophyceae		1. Ectocarpales	1. Ectocarpaceae	*Ectocarpus*
		2. Laminariales	1. Laminariaceae	*Laminaria*
		3. Fucales	1. Fucaceae	*Fucus*
			2. Sargassaceae	*Sargassum*
		4. Dictyotales	1. Dictyotaceae	*Dictyota*
5. Rohodophyceae	1. Bangiodeae	—	—	—
	2. Florideae	1. Nemalionales	1. Batrachospermaceae	*Batrachospermum*
		2. Gigartinales	1. Gracilariaceae	*Gracilaria*
		3. Ceramiales	1. Ceramiaceae	*Ceramium*
			2. Rhodomelaceae	*Polysiphonia*
6. Myxophyceae		1. Chroococcales	1. Chroococcaceae	*Gloeocapsa*
		2. Nostocales	1. Oscillatoriaceae	*Oscillatoria*
			2. Nostocaceae	*Nostoc*
			3. Scytonemataceae	*Scytonema*
			4. Rivulariaceae	*Rivularia*
				Gloeotrichia

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	—	***Chlamydomonas***

Exercise 1

***Object* : Study of Chlamydomonad cell.**

Work procedure

Study the slide showing unicellular thallus.

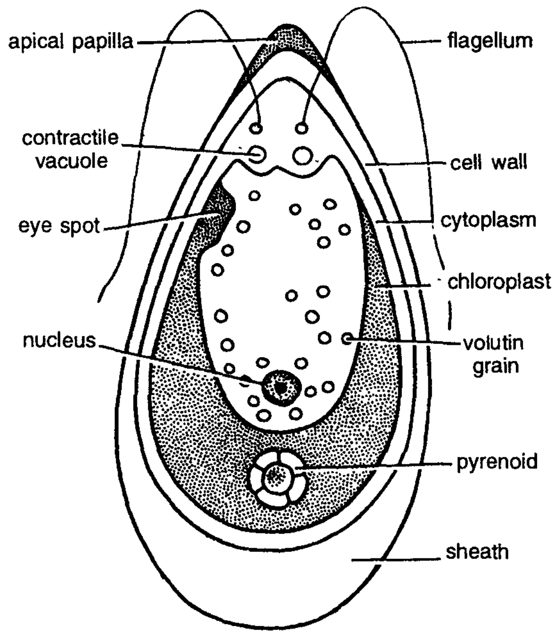


Fig. 1. ***Chlamydomonas.*** Structure of a single cell.

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).

11. The cavity of the cup-shaped chloroplast is completely filled with the cytoplasm in which lies a single nucleus.
12. 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

1. This is asexual reproductive body.
2. The groups of cells are embedded in a common mucilaginous envelope.
3. Each cell is chlamydomonad in structure. The flagella are absent.
4. It is a temporary phase—a structure of perennation.
5. 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.

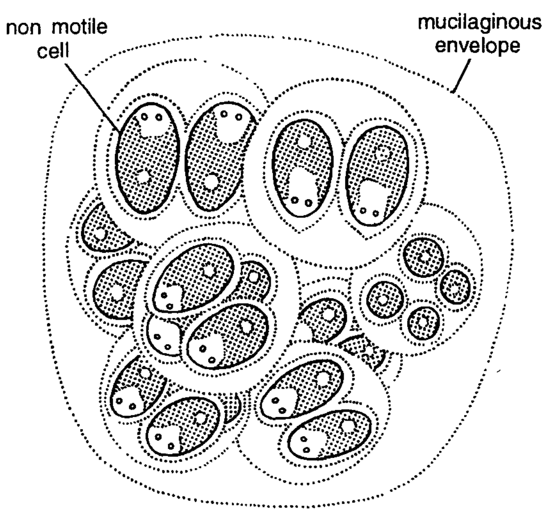


Fig. 2. ***Chlamydomonas***. Palmella stage.

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 vocuoles.
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

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

5. The number of cells in a colony varies from 500-6,500 according to the species.
6. Each cell of the colony is connected with a few of the neighbouring cells by thin and delicate cytoplasmic strands.
7. Each cell is enveloped by an individual gelatinous sheath.
8. All the cells of a colony are typically chlamydomonad in shape, size and structure.
9. 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 anterio-lateral position.

Exercise 2

***Object* : Study of asexual reproduction.**

Work procedure

Study the slide showing daughter colonies.

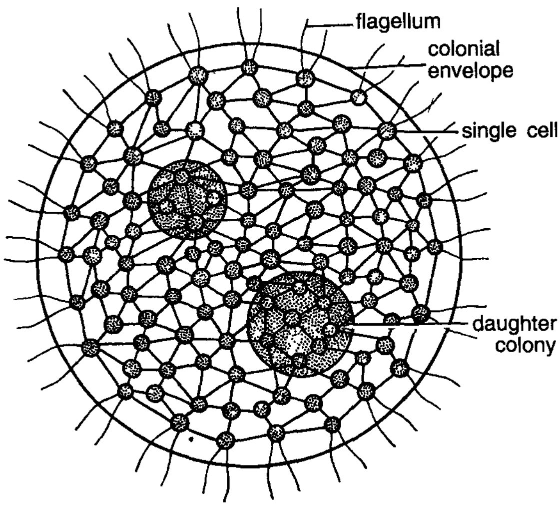


Fig. 1. ***Volvox.*** Parent colony with daughter colonies.

Comments

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

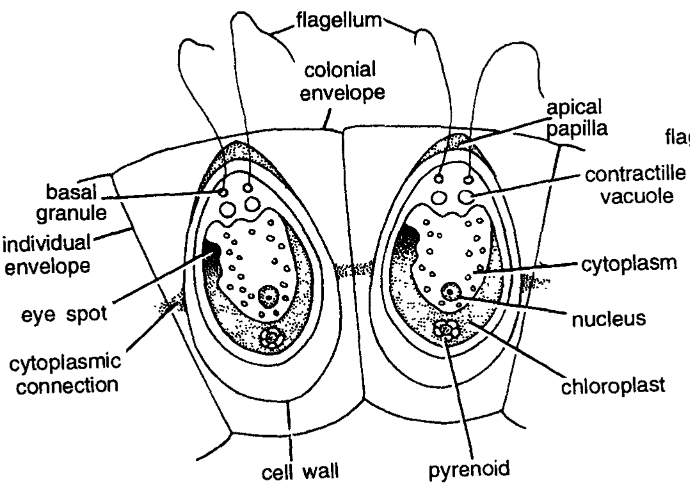


Fig. 2. ***Volvox.*** A part of the colony showing arrangement of cells.

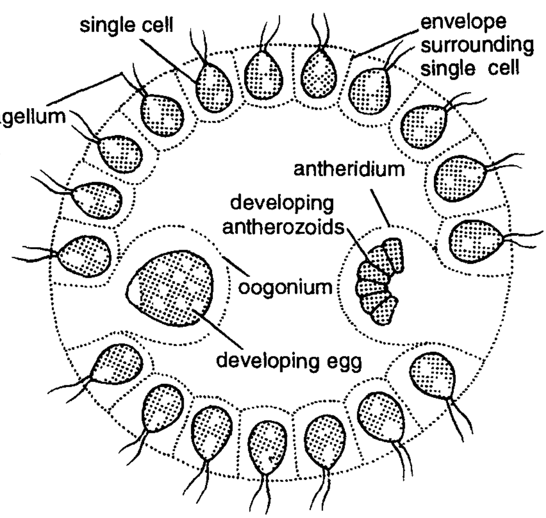


Fig. 3. ***Volvox.*** A colony with antheridium and oogonium.

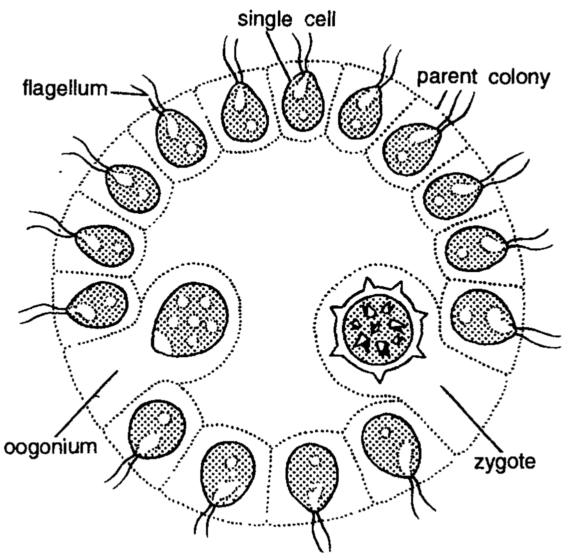


Fig. 4. *Volvox.* A colony with a zygote.

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.

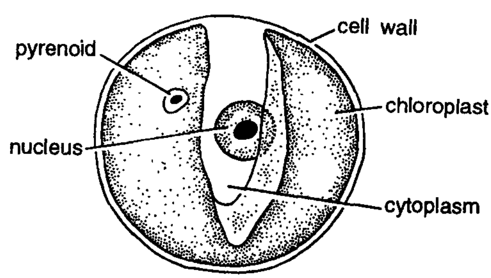


Fig. 1. ***Chlorella.*** A single cell representing a thallus.

2. The cell are found singly or sometimes in groups.
3. A cell is very small. It is spherical to ellipsoidal in shape.
4. A cell has a single cup-shaped or curved band of chloroplast, parietal in position.
5. A single pyrenoid is generally found (lacking at times).
6. Nucleus lies surrounded by the cytoplasm which fills up most of the cell.
7. 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*.

Hydrodictyon

Classification

Sub-division	—	**Algae**
Class	—	**Chlorophyceae**
Order	—	**Chlorococcales**
Family	—	**Hydrodictyaceae**
Genus	—	***Hydrodictyon***

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

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

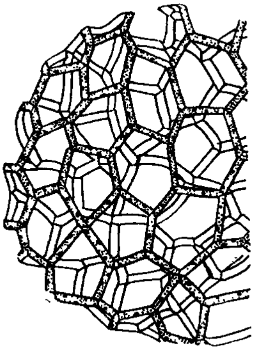


Fig. 1. ***Hydrodictyon.*** A part of saccate thallus.

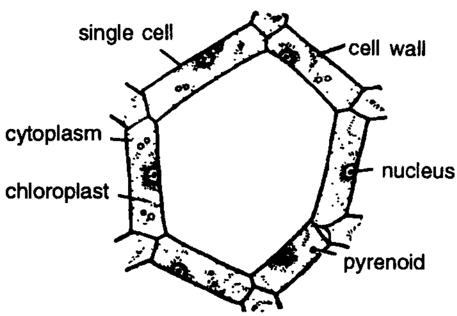


Fig. 2. ***Hydrodictyon.*** A part of reticulum.

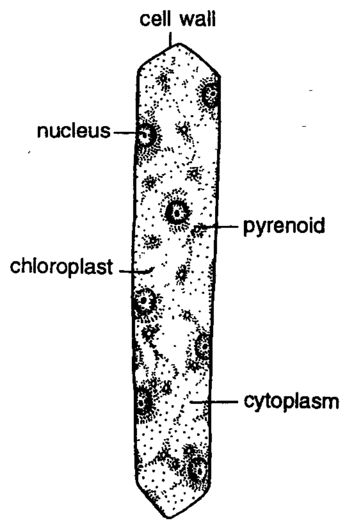


Fig. 3. ***Hydrodictyon.*** A single cell.

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).

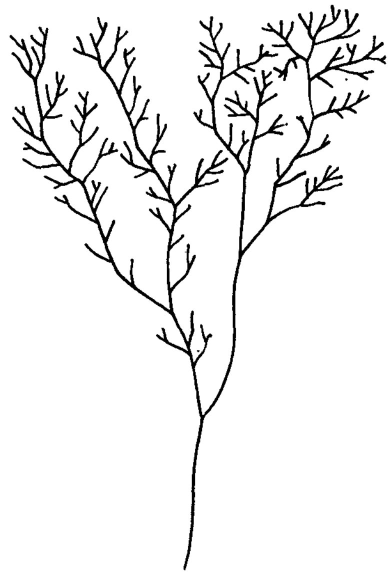


Fig. 1. *Cladophora*. A thallus.

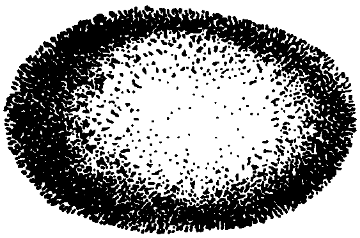


Fig. 2. *Cladophora.* Aegagropilous species.

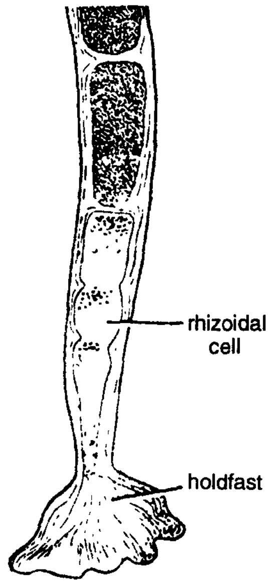


Fig. 3. *Cladophora.* Basal part of the filament with holdfast.

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

Exercise 2

***Object* : Study a cell.**

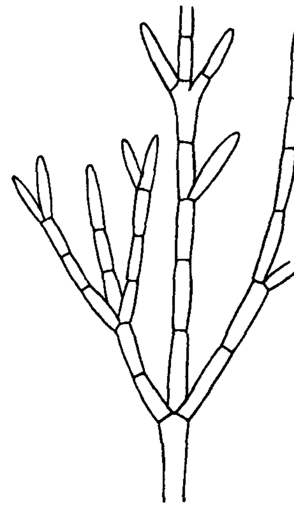


Fig. 4. *Cladophora.* A part of thallus showing mode of branching.

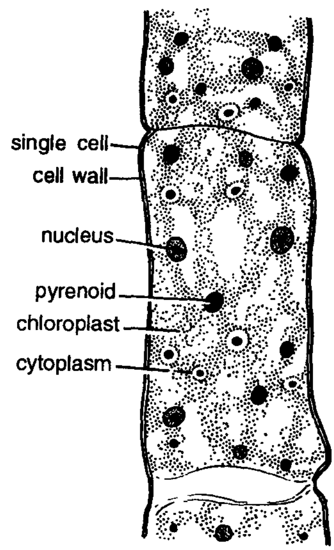


Fig. 5. *Cladophora.* A single cell.

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 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	—	***Fritschiella***

Exercise 1

***Object* : Study the Thallus.**

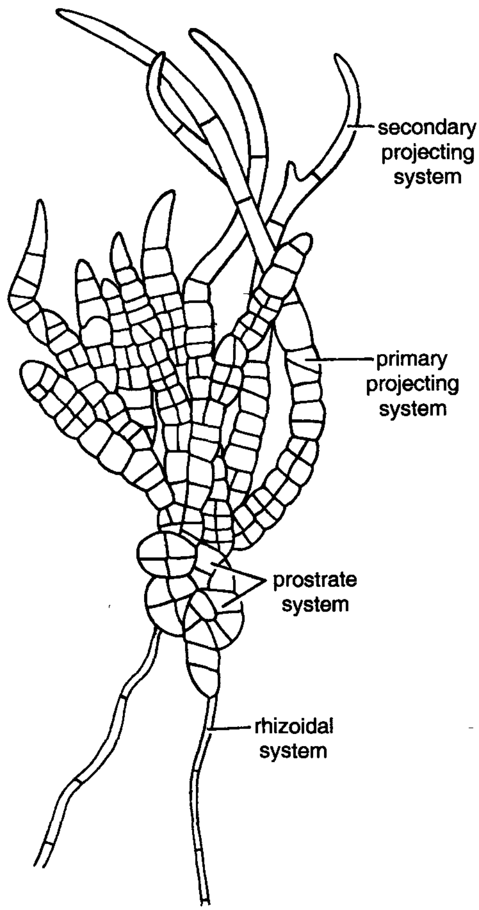


Fig. 1. ***Fritschiella.*** The thallus.

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.

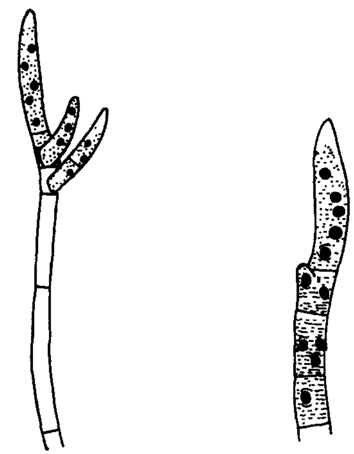


Fig. 2. ***Fritschiella.*** A single cell of the projecting system.

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 unbrahched. The cells of these filaments are small and rounded. They are green and photosynthetic.

6. 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.
7. 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—***Fristchiella***. (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	—	***Coleochaete***

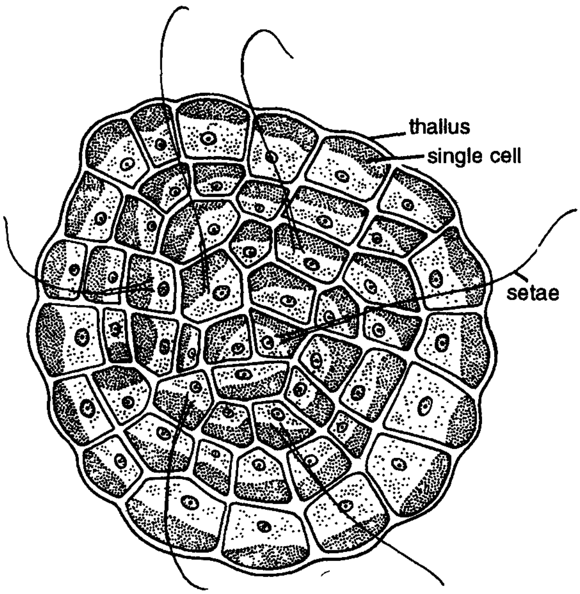


Fig. 1. *Coleochaete* sp. Discoid thallus.

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

1. Thallus is multicellular and heterotrichous.
2. 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.
3. It thallus is disc-like, the disc represents only the prostrate system while a few setae or hair, represent erect system.
4. Filamentous thallus exhibits typical heterotrichous habit with a branched prostrate system and a branched projecting (erect) system.
5. In both the cases a few cells possess a cytoplasmic outgrowth—setae. Setae are

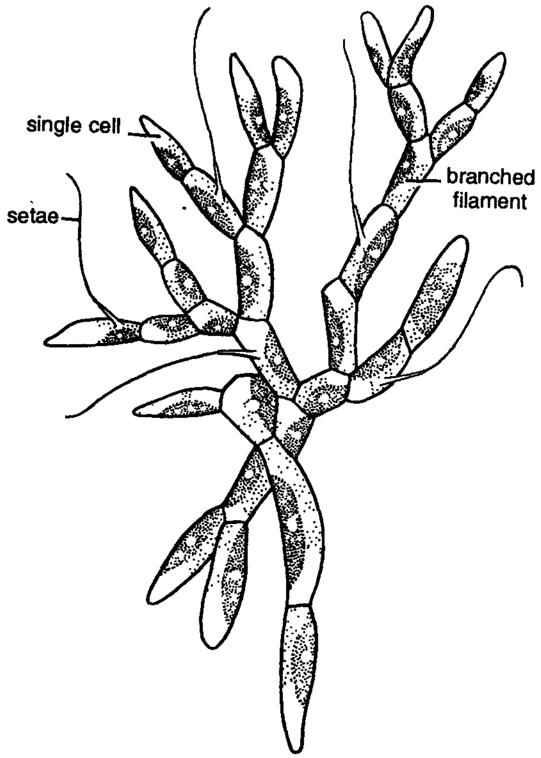


Fig. 2. *Coleochaete* sp. Cushionoid thallus.

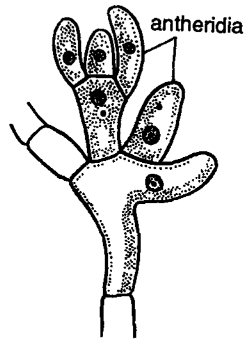


Fig. 3. *Coleochaete* sp. Thallus bearing antheridia.

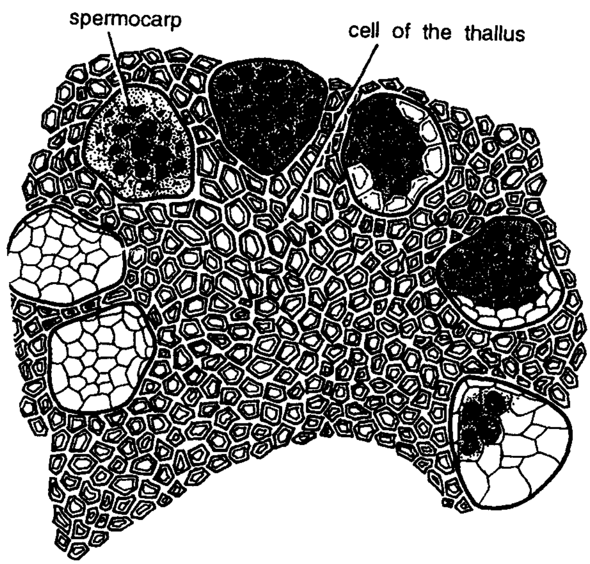


Fig. 4. *Coleochaete* sp. Thallus with spermocarps.

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.

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. Anteridia 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 spermocarp remains enveloped in a parenchymatous tissue formed by the development of neighbouring cells. It is conspicuously reddish-brown in colour.
9. Spermocarp 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 laminate chloroplast.

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

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.

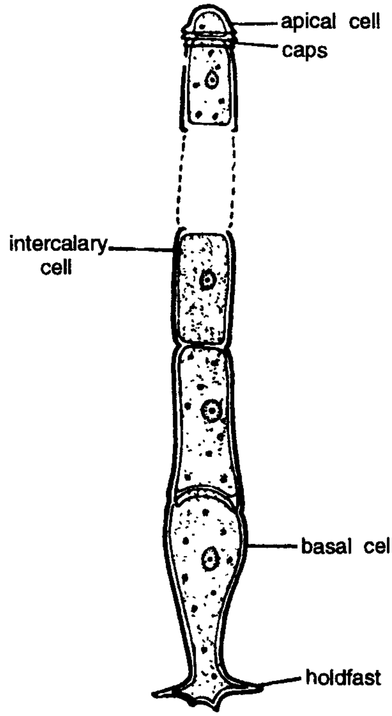


Fig. 1. ***Oedogonium.*** A filament.

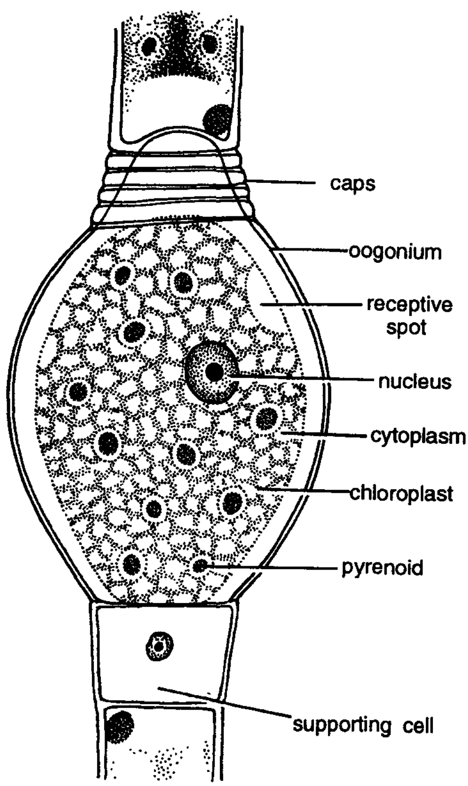


Fig. 2. ***Oedogonium.*** Filament showing a mature oogonium.

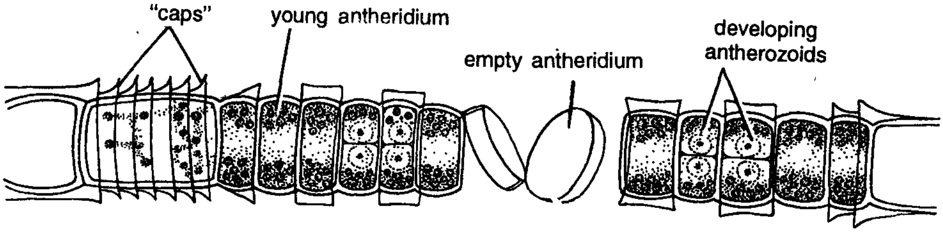


Fig. 3. ***Oedogonium.*** Part of a filament showing a chain of antheridia (Macrandrous species).

2. A filament is differentiated into three types of cells according to their position : (i) basal, (ii) intercalary and (iii) apical.
3. 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.
4. A cell at the tip of the filament is known as apical cell. It is rounded at its free surface.
5. The cells present between basal and the apical cells are intercalary cells. These show typical cell structure.
6. The typical cell is cylindrical.
7. Cell wall is thick and three layered.
8. 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.
9. The cell is uninucleate. The nucleus is situated near the cell wall and is held by thin and delicate cytoplasmic strands.
10. Mature and old cells show 'cap cells' at their upper end. These are characteristic of the members of Oedogoniales.

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

1. Oogonia are intercalary or terminal in position.
2. Oogonium may be solitary or occur in a row of 2-3 or even more.
3. Oogonium generally shows one or more cap cells at its upper end, indicating its development from a comparatively older cell.
4. It is mostly spherical or oval in shape and larger than a vegetative cell.
5. At the base of each oogonium lies a small and flat daughter cell, known as supporting or suffultory cell.
6. Oogonium encloses a single large ovum.
7. The wall of the oogonium has a small pore on one side, known as receptive pore.
8. Just opposite the receptive pore, protoplast of the oogonium has a hyaline area-receptive spot.
9. Uninucleate protoplast is rich in reserve food.

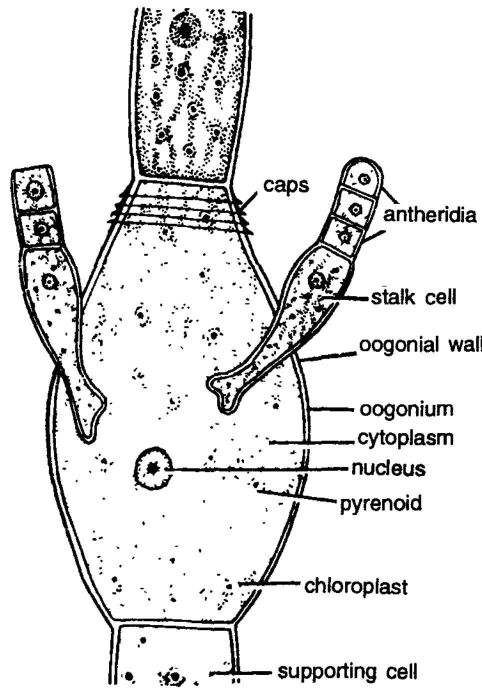


Fig. 4. ***Oedogonium.*** Oogonium with two dwarf males attached to it. (Nannandrous species).

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 anteridium 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 unbrahched, (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 1

***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 characteisitc 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.

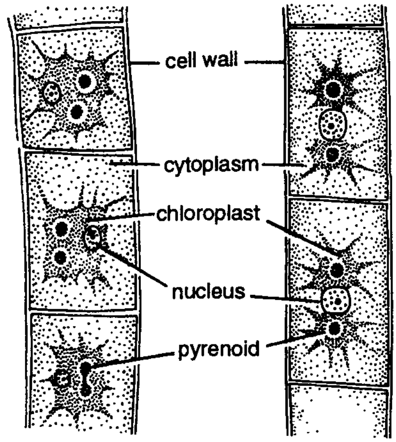


Fig. 1. ***Zygnema.*** Filaments showing internal structure of the cell.

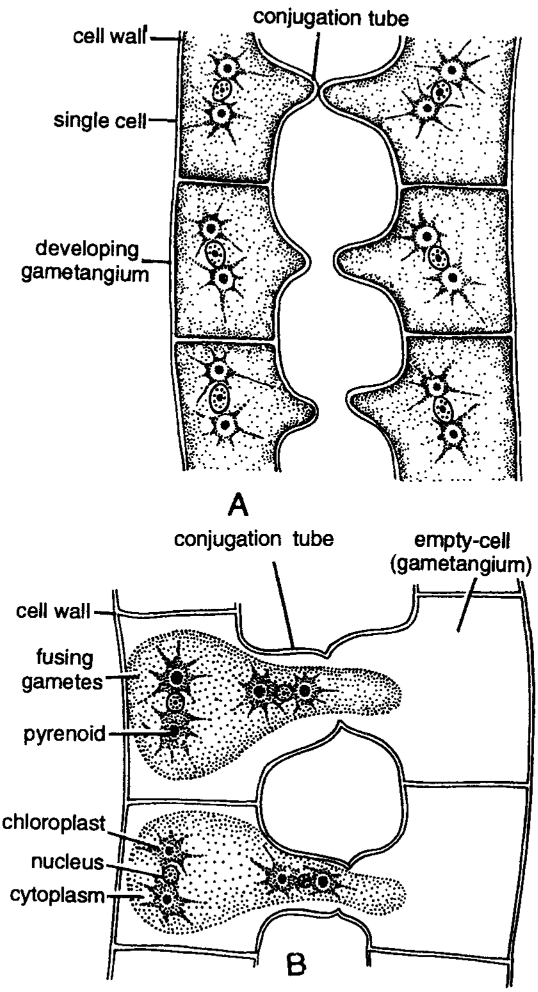


Fig. 2. ***Zygnema.*** **A-B.** Filaments showing various stages of scalariform conjugation.

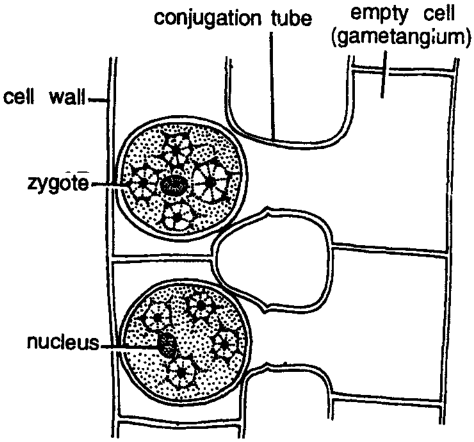


Fig. 3. ***Zygnema.*** Filaments showing zygospores after completion of scalariform conjugation.

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	—	***Chara***

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 macrocopic, 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 (ii) lateral branches of unlimited growth (long laterals).

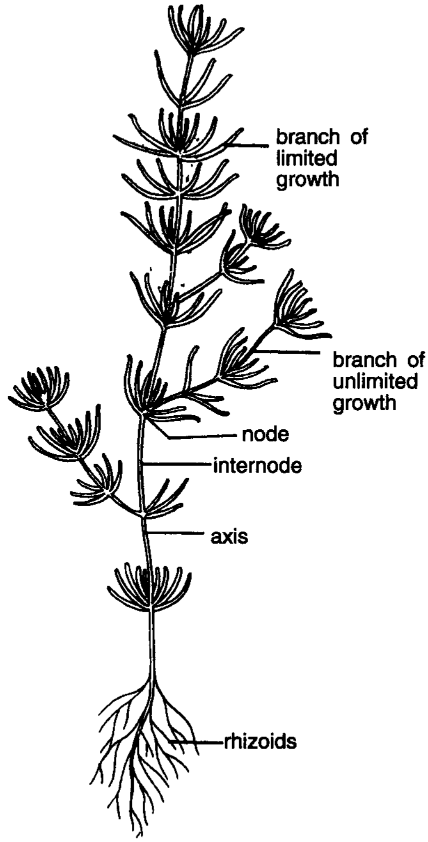


Fig. 1. ***Chara.*** A thallus to show habit.

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 characteistics 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.

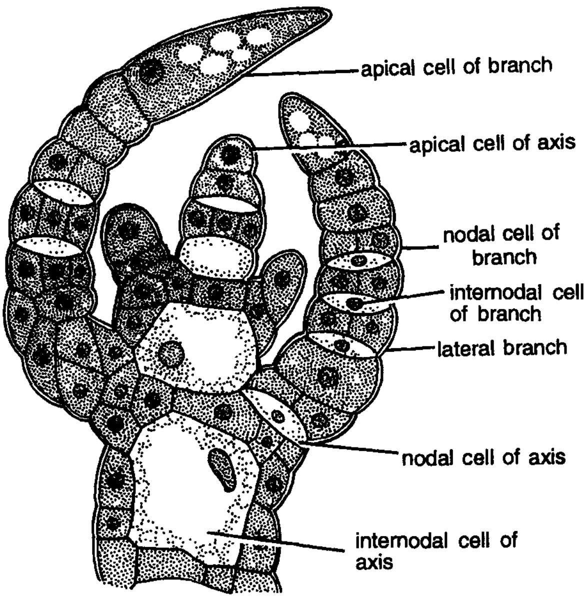


Fig. 2. ***Chara.*** L.s. apical region

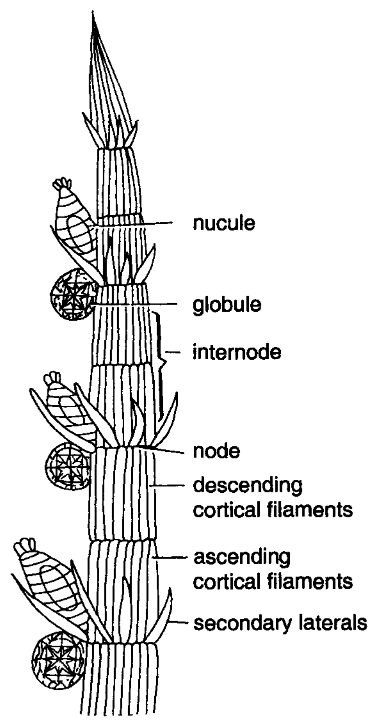


Fig. 3. ***Chara.*** Part of a fertile branch to show the position of sex organs.

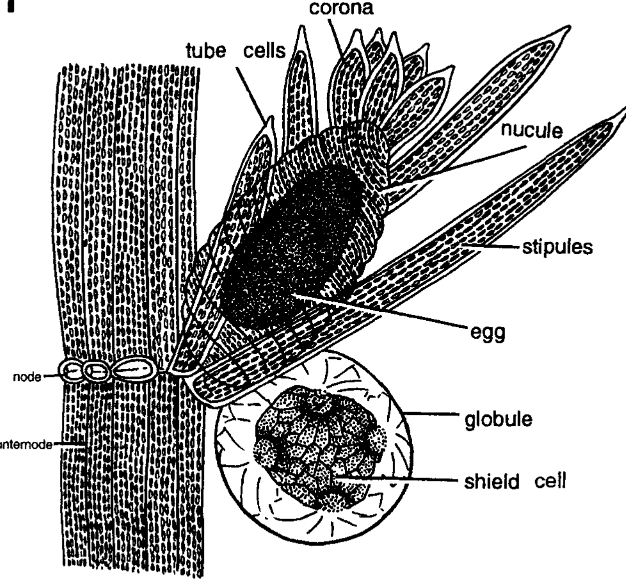


Fig. 4. *Chara.* Node bearing nucule and globule.

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.

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 amylum 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).

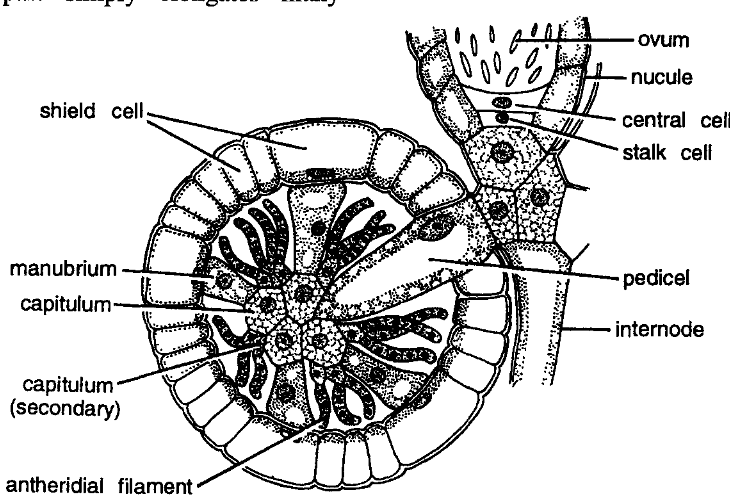


Fig. 5. *Chara.* L.s. globule.

2. The male reproductive organ is called a 'globule' while the female reproductive organ a 'nucule'.
3. Both the sex organs are borne at one point on the nodes of short laterals which bear stipules.
4. 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

1. A globule is a small, spherical and conspicuously red or yellow structure attached to the node by a long stalk cell.
2. Outermost wall of the globule is ornamented, composed of eight, large, curved and plate-like cells called shield cells.
3. Ornamentation of the shield cell is due to the foldings in the cell wall.
4. Each shield cell is attached to a long handle or a rod-shaped cell—manubrium.
5. 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.
6. Each secondary capitulum bears 2-4, long and unbranched antheridial filaments.
7. Each antheridial filament is made of 100-230 small cells.
8. 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

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

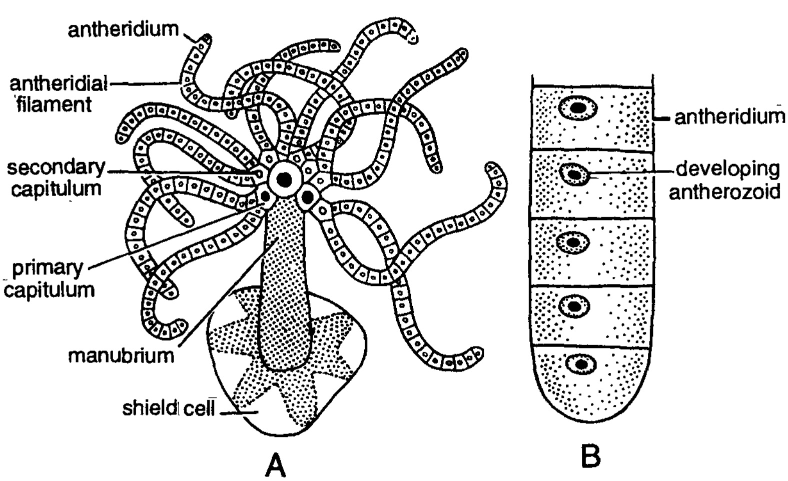


Fig. 6. ***Chara*** **A.** single shield cell with manubrium and antheridial filaments, **B.** A part of antheridial filament enlarged.

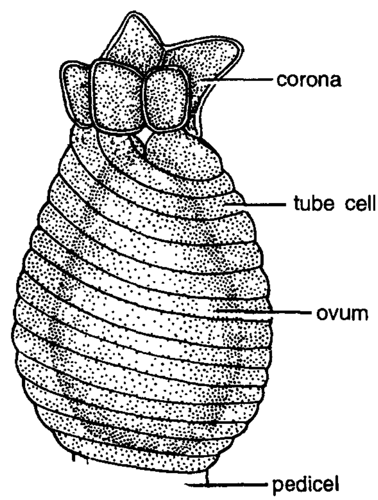


Fig. 7. ***Chara.*** A nucule (female sex organ).

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 (epipelic 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 β-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.

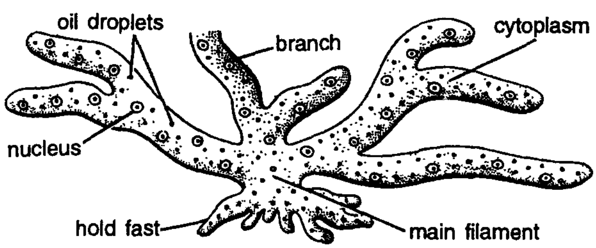


Fig. 1. ***Vaucheria.*** Thallus to show habit.

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.**

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.

Exercise 3

***Object* : Study of antheridia.**

Work procedure

Study a slide showing anteridia.

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.

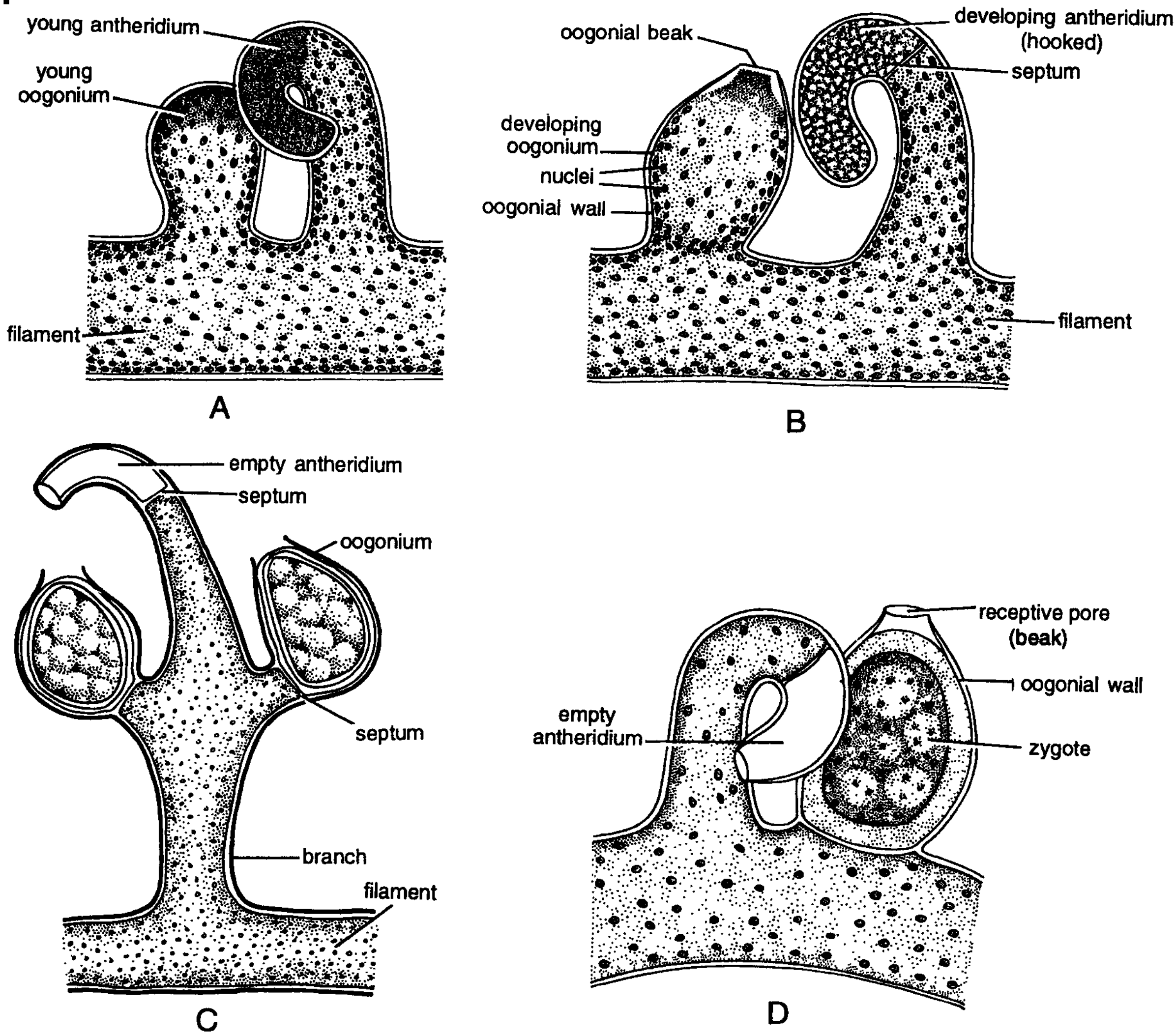


Fig. 2. ***Vaucheria.*** **A-D.** Various stages in sexual reproduction. **A.** Young oogonium with young coiled antheridium, **B.** Mature oogonium and antheridium, **C.** Stalked sex organs in *Vaucheria geminata*, **D.** Empty antheridium and zygote.

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, thubular 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^{0}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.

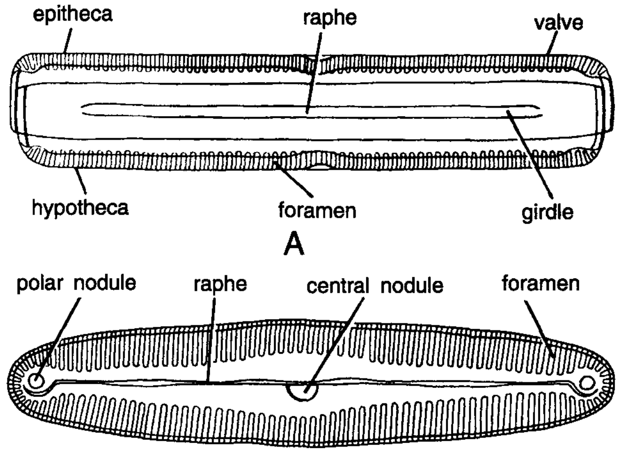


Fig. 1. Diatom, **A-B** ***Pinnularia***, **A.** Girdle view, **B.** Valve view of a frustule.

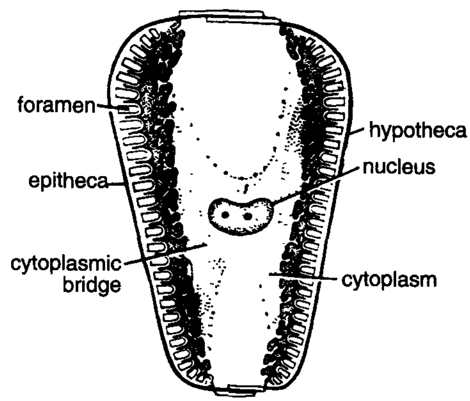


Fig. 2. ***Suriella***. Internal structure of a cell.

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 pennales, 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 pennales, 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.

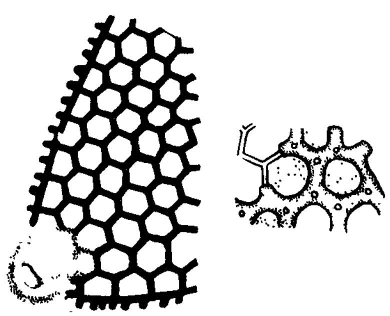


Fig. 3. *Triceratium.* Part of the wall magnified.

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 pennales.
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 swarmers, (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-raphe absent.

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

Exercise 1

***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 substratum.

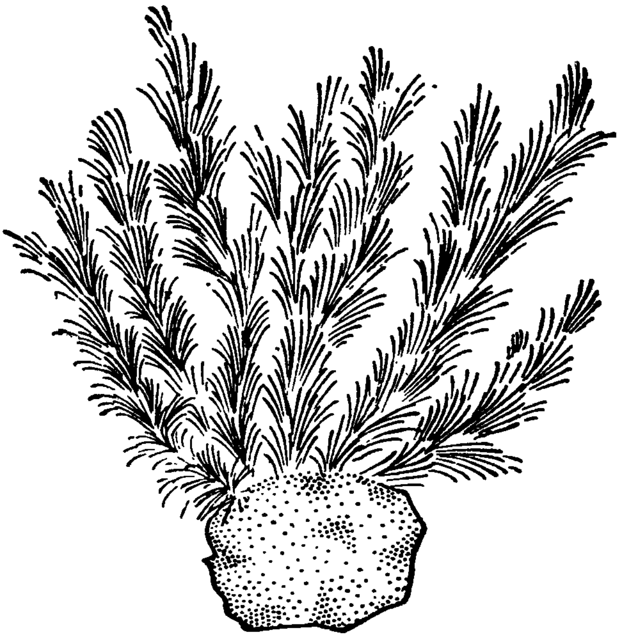


Fig. 1. ***Ectocarpus.*** Habit

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.

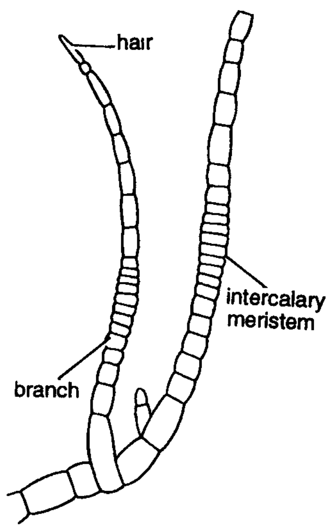


Fig. 2. ***Ectocarpus.*** A part of the filamentous thallus.

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.

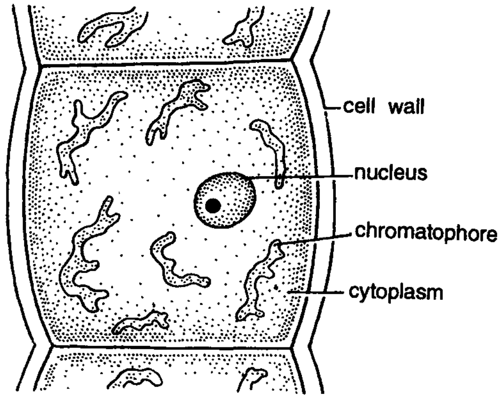


Fig. 3. ***Ectocarpus.*** A single cell.

Exercise 2

Object **: Study of unilocular sporangium.**

Work procedure

Search for fialments 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.

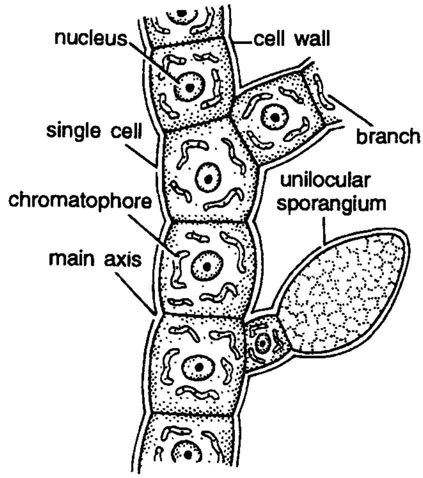


Fig. 4. ***Ectocarpus.*** A branch with unilocular sporangium.

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.

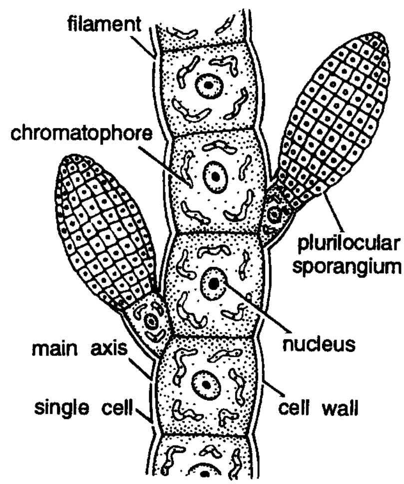


Fig. 5. ***Ectocarpus***. Filament with plurilocular sporangia.

2. The structure when borne by haploid plant serves as a gametangium whereas on diploid plant it functions as a sporangium.
3. Sporangia situated laterally may be sessile or stalked. These may be ovate to siliquose.
4. Plurilocular sporangium is divided into large number of cells.
5. A mature sporangium produces biflagellate swarmers, one each from every cell.
6. If plurilocular sporangium is borne on a haploid plant, the swarmers 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 swarmers 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

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

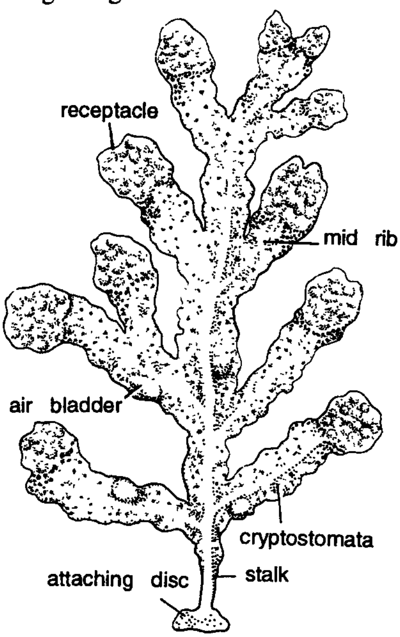


Fig. 1. ***Fucus.*** External features of thallus.

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 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).

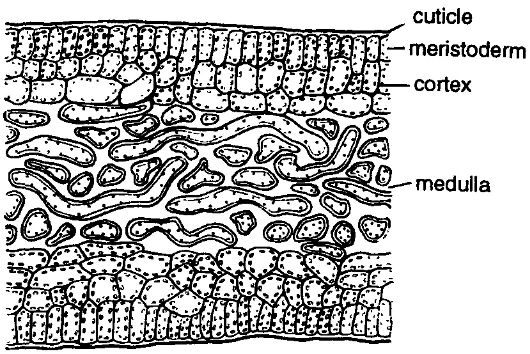


Fig. 2. ***Fucus.*** A part of T.s. through wing.

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.

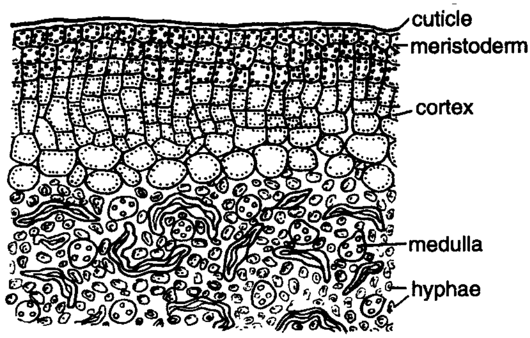


Fig. 3. ***Fucus.*** A part of T.s. through mid-rib.

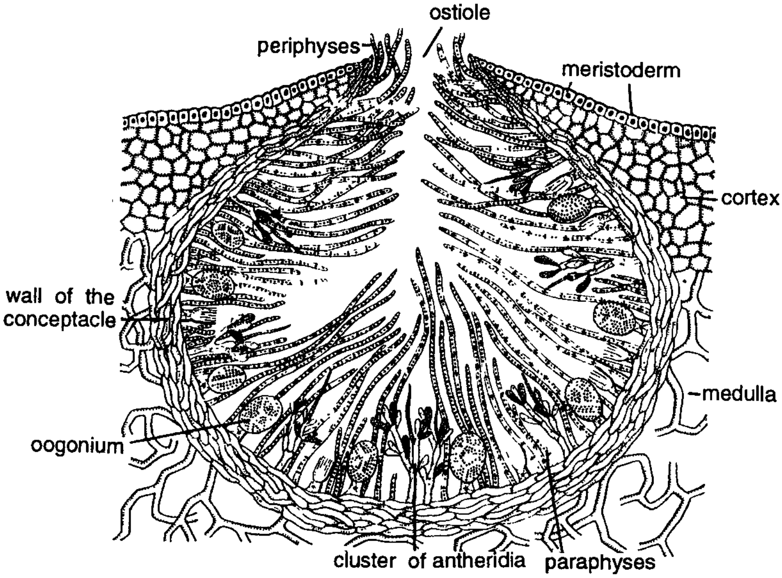


Fig. 4. ***Fucus.*** T.s. through conceptacle.

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.

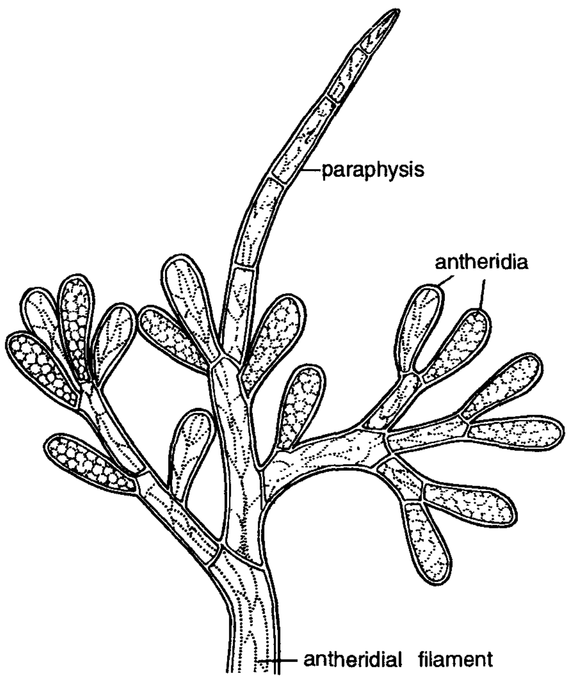


Fig. 5. ***Fucus.*** Antheridial filaments.

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 manitol, (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.

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.

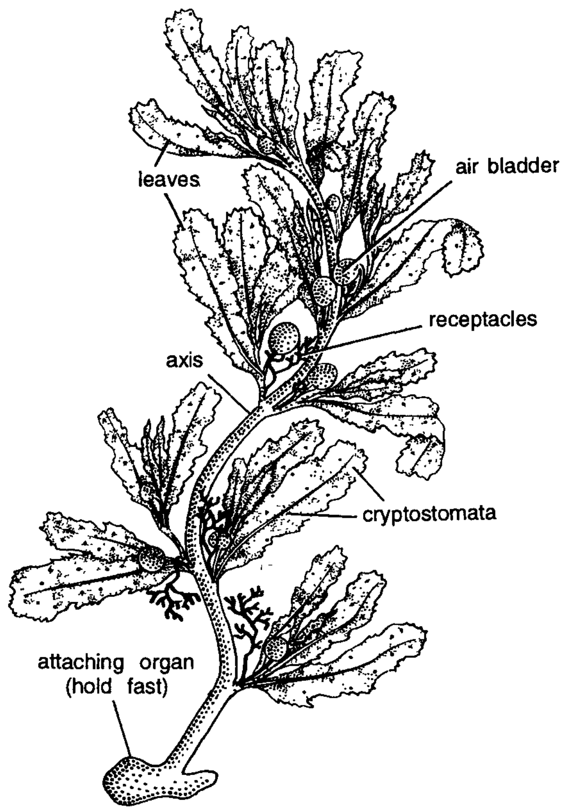


Fig. 1. ***Sargassum.*** Thallus to show external features.

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.

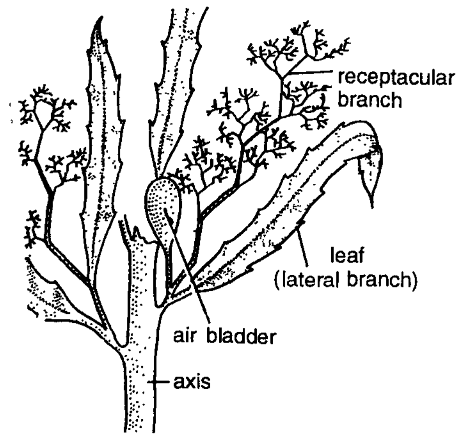


Fig. 2. ***Sargassum.*** A part of thallus to show details.

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 a 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.

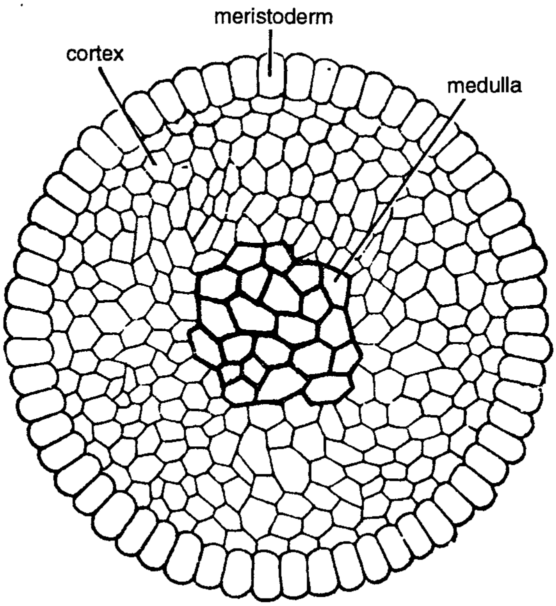


Fig. 3. ***Sargassum.*** T.s. axis.

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 cryptoblat opens to the exterior by an opening—ostiole (visible externally as black dots or pores).

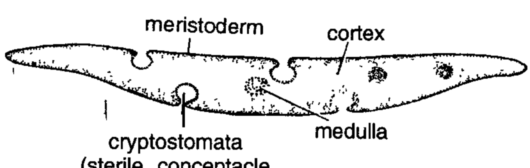


Fig. 4. ***Sargassum.*** T.s. leaf (diagrammatic).

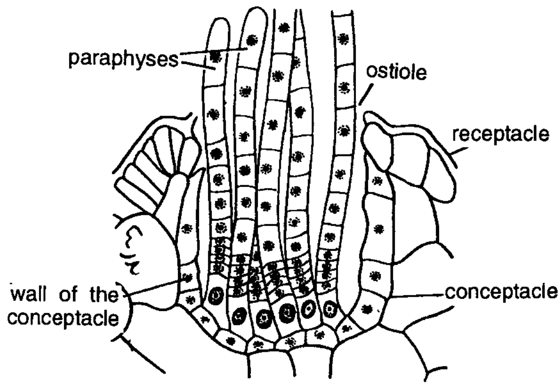


Fig. 5. ***Sargassum.*** L.s. through conceptacle.

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.

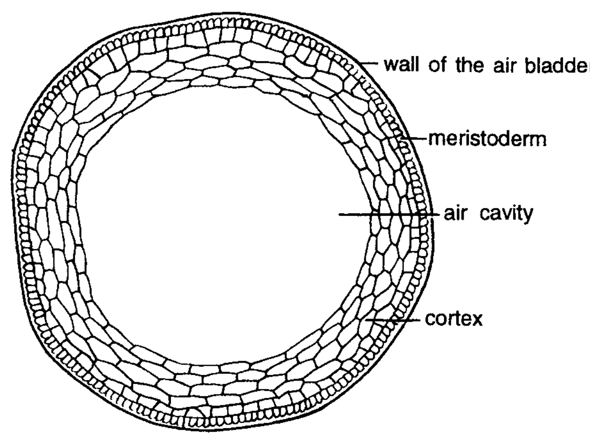


Fig. 6. ***Sargassum.*** T.s. through air bladder.

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 buyoancy.

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.
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 periphyses.
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.

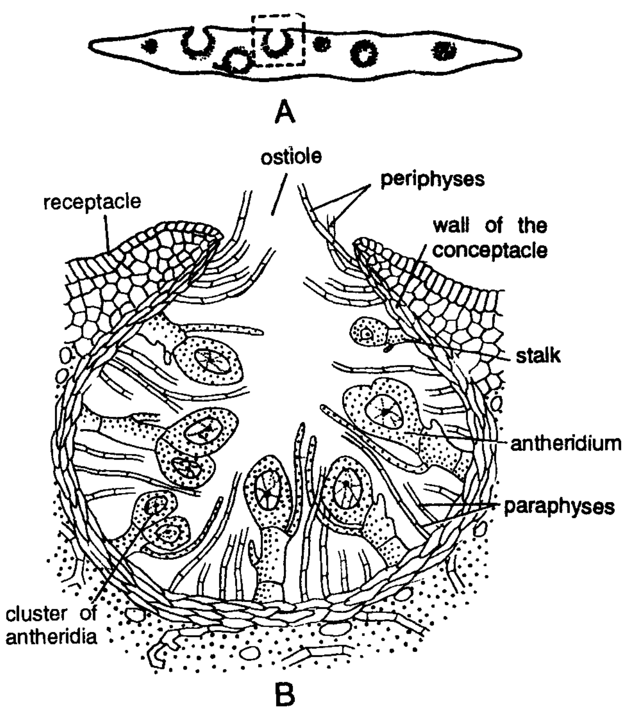


Fig. 7. ***Sargassum.*** **A-B. A.** T.s. through male receptacle, **B.** A male conceptacle as shown in fig. B enlarged.

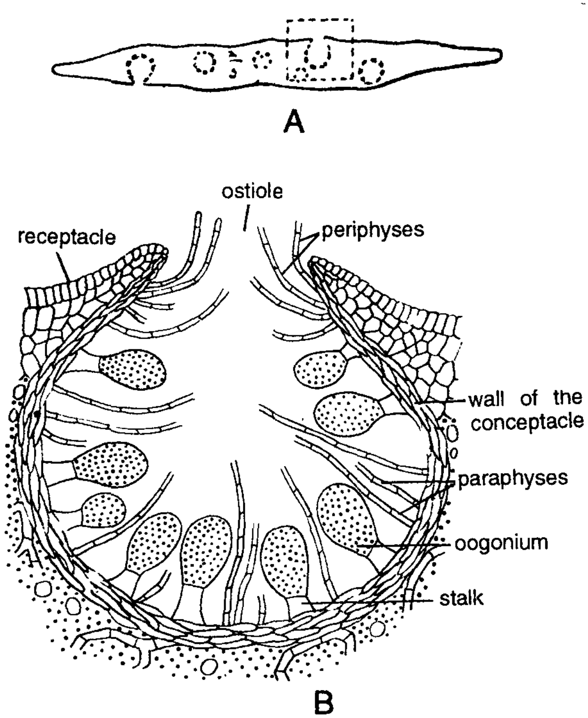


Fig. 8. ***Sargassum.*** **A-B. A.** T.s. through female conceptacle, **B.** A female conceptacle as shown in fig. B enlarged.

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-life 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-phycocyanin. 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, phamaceutical 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**
Calss	—	**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.

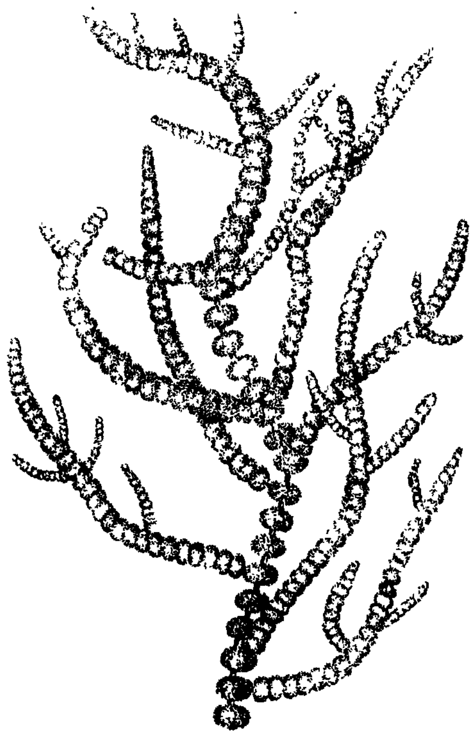


Fig. 1. ***Batrachospermum.*** Thallus to show external features.

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 carpospores.

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.

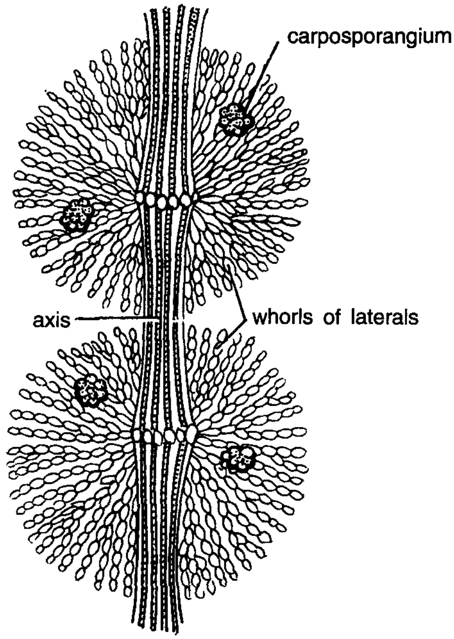


Fig. 2. ***Batrachospermum.*** A part of fertile branch with glomerule and carposporangia.

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-ferilization 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

Sub-division	—	**Algae**
Class	—	**Rhodophyceae**
Sub-class	—	**Florideae**
Order	—	**Ceramiales**
Family	—	**Ceramiaceae**
Genus	—	***Ceramium***

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.

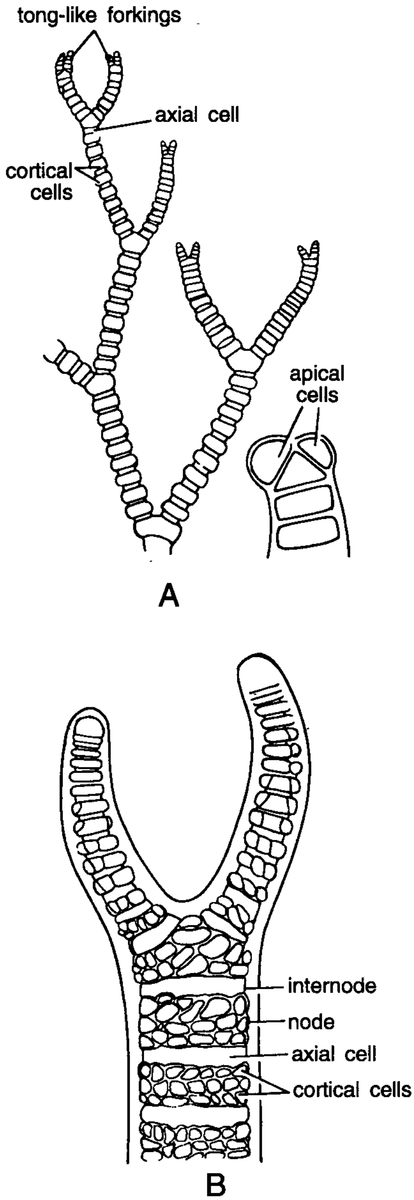


Fig. 1. ***Ceramium.*** Thallus. **A.** A part of thallus to show habit, **B.** Axis with nodes and internodes.

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.

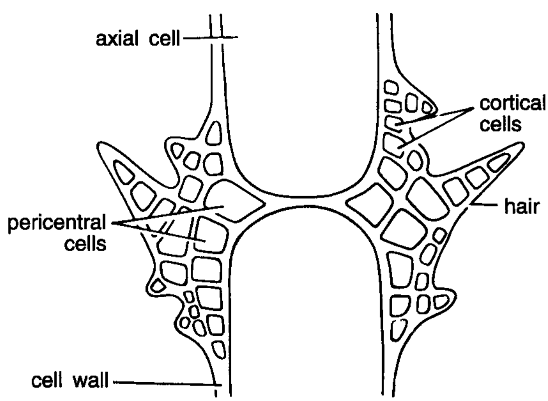


Fig. 2. ***Ceramium.*** L.s. through node.

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.

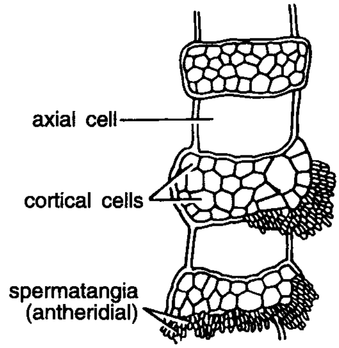


Fig. 3. ***Ceramium.*** An axis with antheridia or spermatangia.

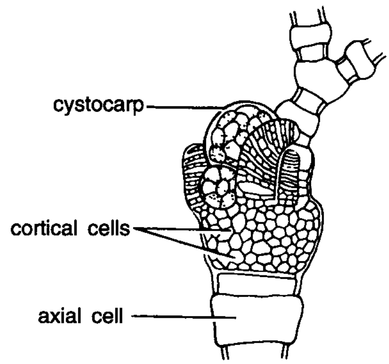


Fig. 4. ***Ceramium.*** An axis with carposporophyte showing cystocarp.

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.

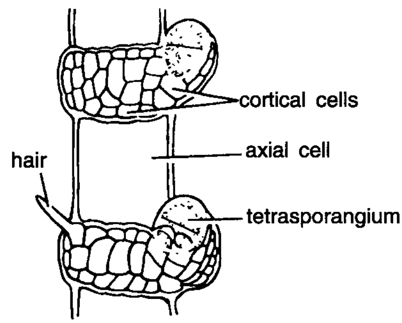


Fig. 5. ***Ceramium.*** An axis with tetrasporangia.

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—**Ceramiaceae**. (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.

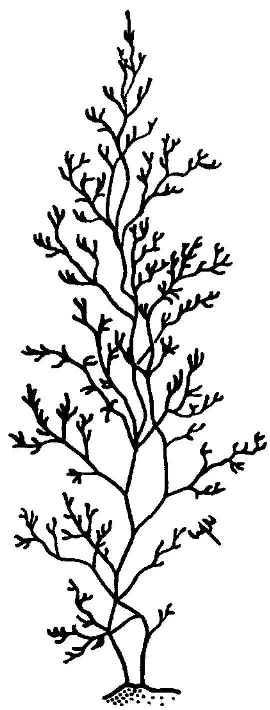


Fig. 1. ***Polysiphonia***. Thallus to show habit.

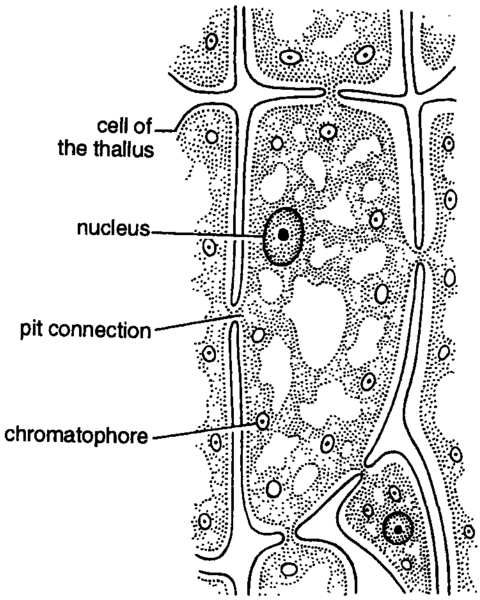


Fig. 2. ***Polysiphonia.*** A single cell.

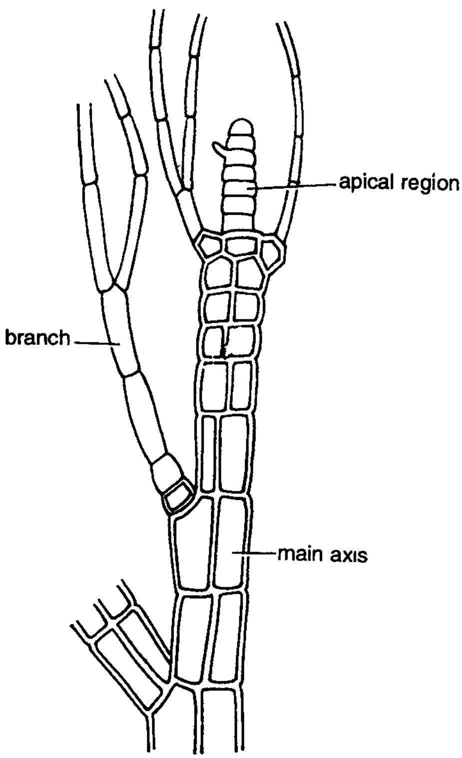


Fig. 3. ***Polysiphonia.*** A part of thallus with branches.

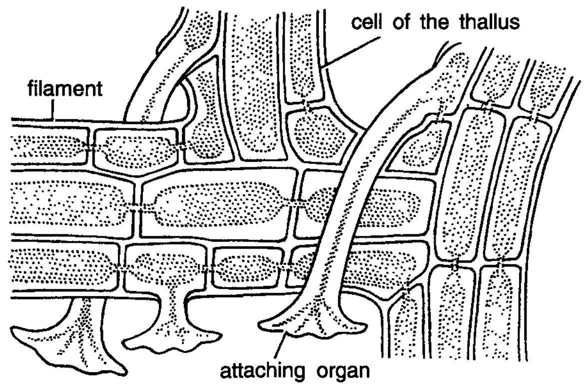


Fig. 4. ***Polysiphonia.*** A part of thallus showing rhizoids.

7. 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.
8. 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

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

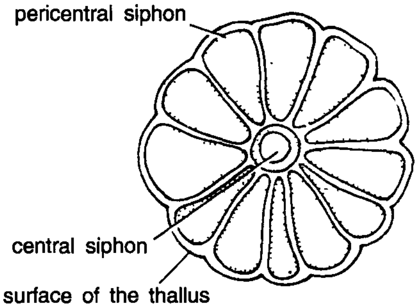


Fig. 5. ***Polysiphonia.*** T.s. thallus.

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.
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.

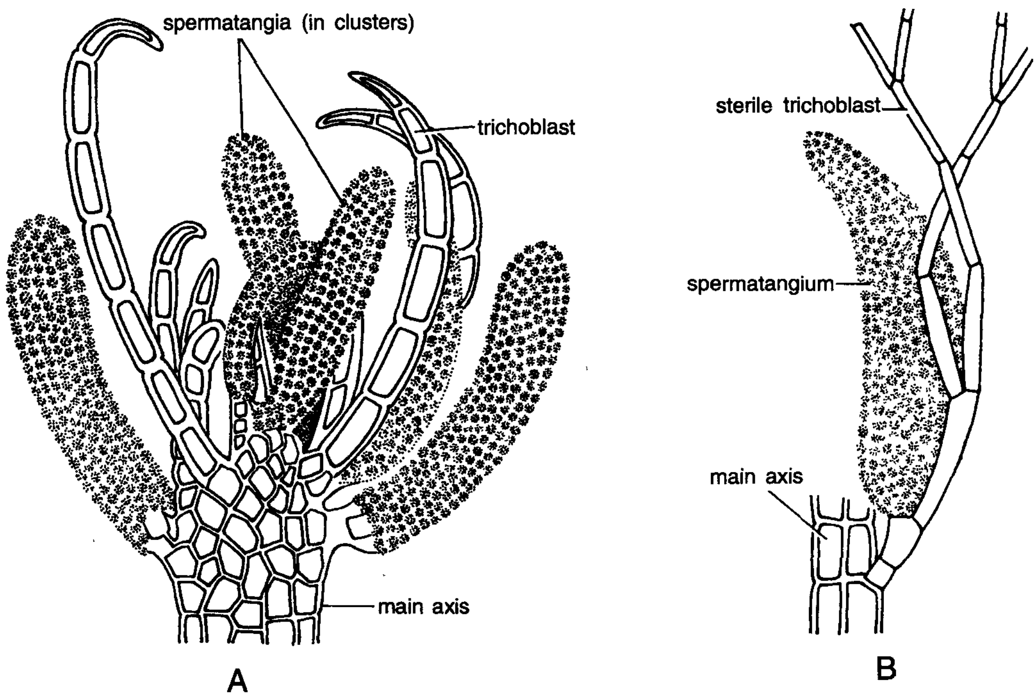
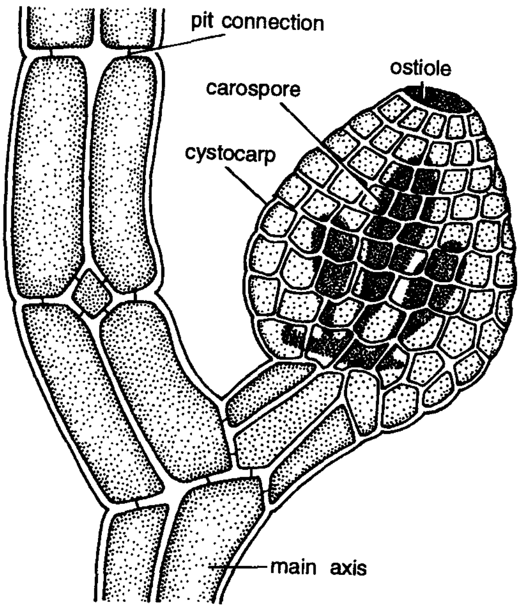


Fig. 6. ***Polysiphonia.*** **A-B A.** Cluster of spermatangia, **B.** A spermatangium.



Fi.g 7. *Polysiphonia.* A part of thallus with cystocarp.

5. Carpospores are produced from the base of the cystocarp. These are arranged in single spherical layer.
6. Each carpospore 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.

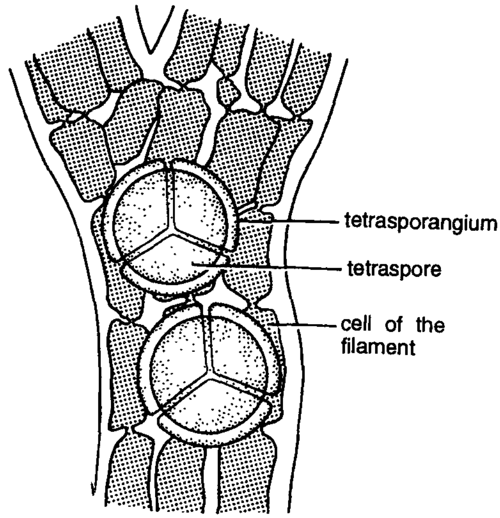


Fig. 8. ***Polysiphonia.*** A part of thallus with tetrasporangia.

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 centroplasm. 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 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	—	***Oscillatoria***

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 hormogone formation.
2. The hormogones (small pieces of trichome with one to many uniform cells) are formed as a result of the death of intecalary cell or by the formation of special biconcave separation discs.

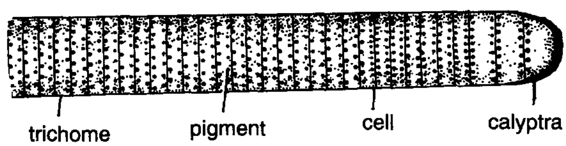


Fig. 1. ***Oscillatoria.*** Trichomes.

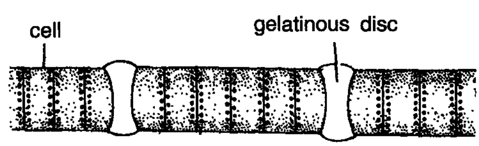


Fig. 2. ***Oscillatoria.*** A trichome showing separation disc.

(B–14)

3. Hormogones are liberated by the disintegration of discs.
4. Each hormogone 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) Hormogones, 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	—	***Nostoc***

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

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

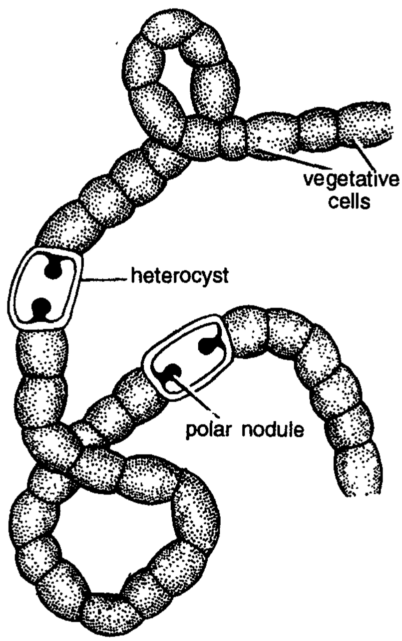


Fig. 1. *Nostoc.* A vegetative filament.

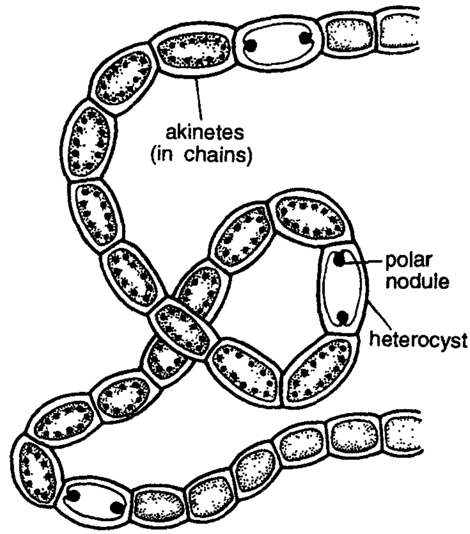


Fig. 2. *Nostoc.* Heterocyst with akinetes in chains.

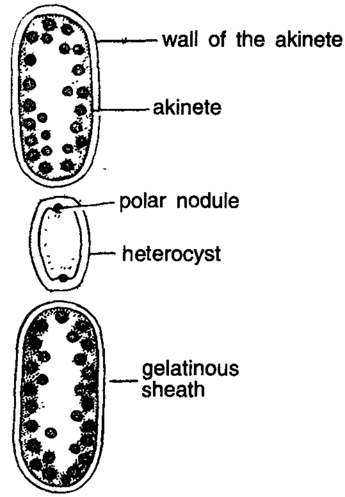


Fig. 3. *Nostoc.* Heterocyst with akinetes on both of its sides.

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.

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.

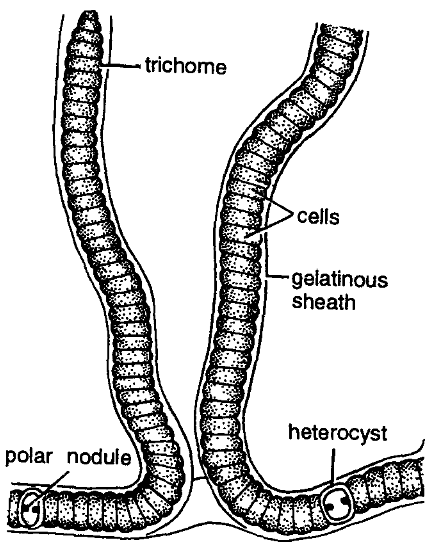


Fig. 1. ***Scytonema.*** A part of thallus to show false branching. *(B–14)*

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 centroplasm is enclosed by peripheral pigmented chromatoplasm. The centroplasm 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 fialmentous 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	—	***Rivularia***

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.

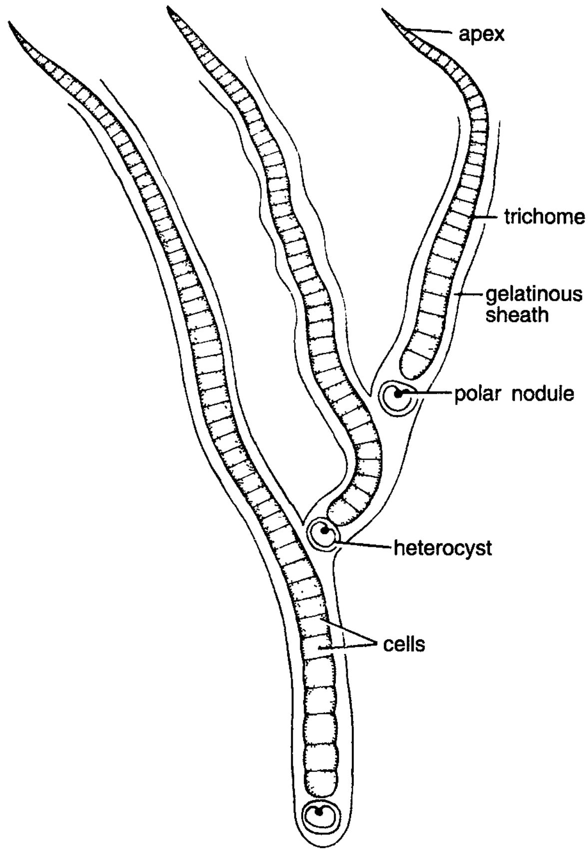


Fig. 1. ***Rivularia.*** A part of thallus to show false branching.

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 cyanophycean structure. There is a centrally located centroplasm or 'incipient nucleus'. Genetic material in the form of DNA is located in this region. Peripheral chromatoplasm shows dispersed pigments with shining cyanophycean granules.
9. Reproduction occurs by homogonia produced either singly or in series.
10. Hormogonia 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—cyanophycean 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) Trichomescommonly 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	—	***Gloeotrichia***

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.

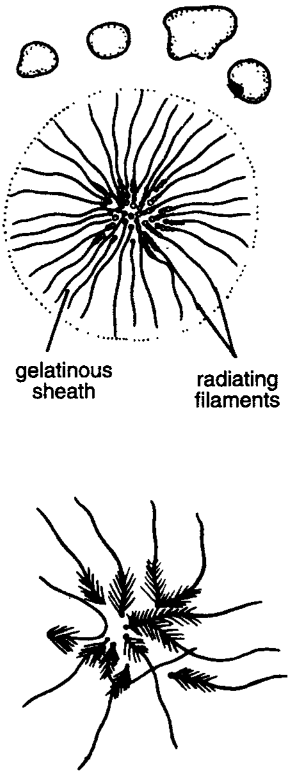


Fig. 1. ***Gloeotrichia.*** A few colonies.

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.

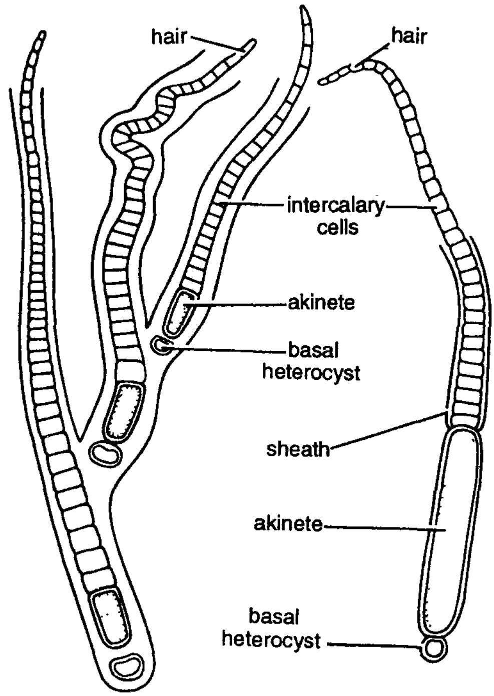


Fig. 2. ***Gloeotrichia.*** A filament to show heterocyst and akinete.

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 parasities 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 **PLASMODIOPHOROMYCETES**

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

(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 prosorus

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 cellulosic

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 perioplasm

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) Sporangiophores dichotomously branched, determinate
(2) Sporangia borne singly at the tips of branches

Examples. ***Sclerospora, Peronospora***

Family 3. **Albuginanceae**

(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 appearnce

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 chlamydospores 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**

Chlamydospores 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**

Thallospores 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	—	***Plasmodiophora***

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.

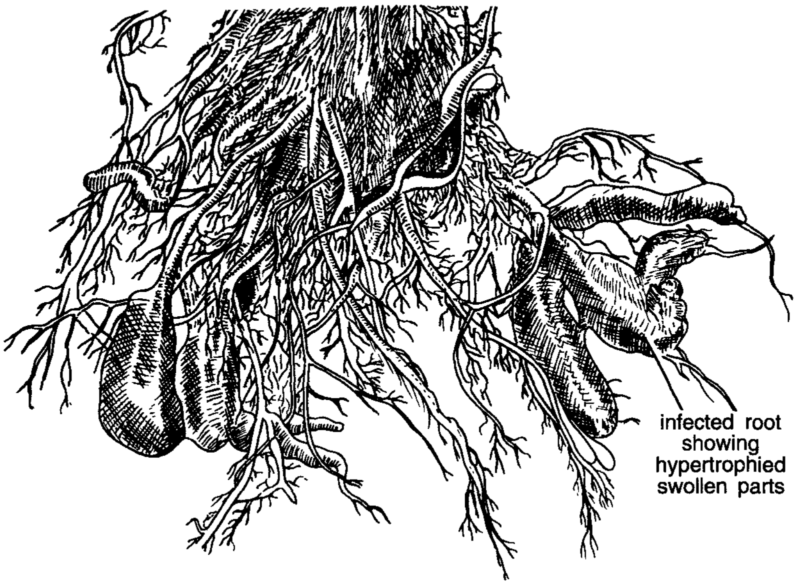


Fig. 1. ***Plasmodiophora***. Infected cabbage roots.

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.

Exercise 2

Object : **Study of thallus.**

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.

Exercise 2

Object : **Study of reproductive structures.**

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).

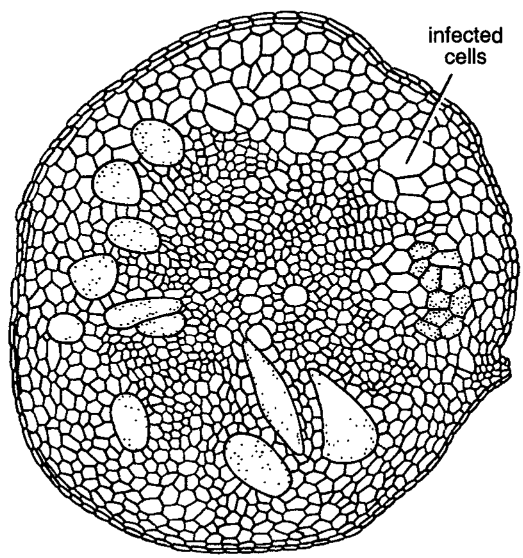


Fig. 2. ***Plasmodiophora.*** T.s. root of host showing fungus.

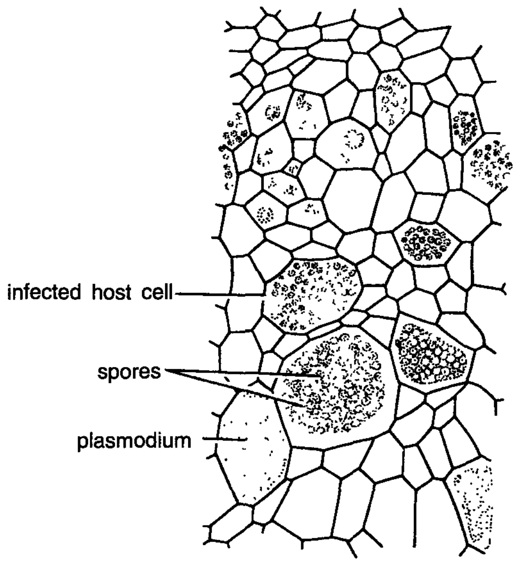


Fig. 3. ***Plasmodiophora.*** T.s. host root with spores.

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**. Endoparasite; vegetative development within living cells of the host.

Genus—***Plasmodiophora***. (1) Plant body plasmodium, (2) Hypertrophy results in club-shaped malformations.

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 seprophyte 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

Synchytrium

Classification

Kingdom	—	**Mycota**
Division	—	**Eumycota**
Sub-division	—	**Mastigomycotina**
Class	—	**Chytridiomycetes**
Order	—	**Chytridiales**
Family	—	**Synchytriaceae**
Genus	—	***Synchytrium***

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. Solauaceae) and 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.

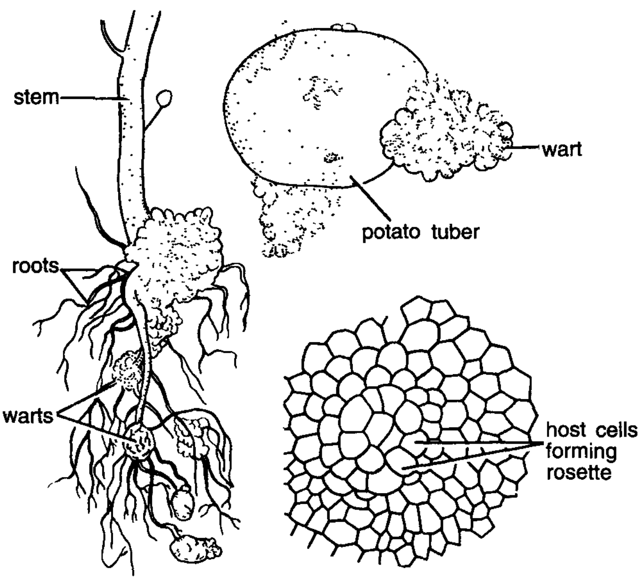


Fig. 1. *Synchytrium endobioticum*. Infected potato tuber causing black wart disease.

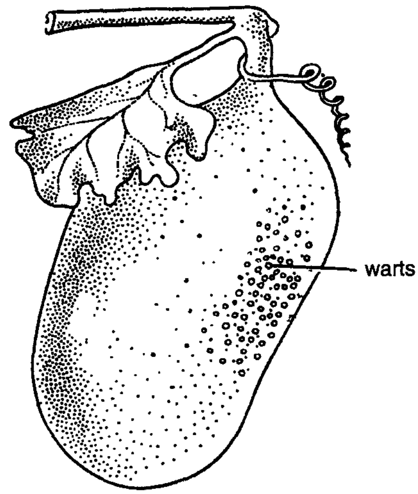


Fig. 2. *Synchytrium trichosanthoides*. Infected cucurbit fruit.

3. *S. trichosanthoides* and *S. lagenariae* attack cucurbits and cause warts.
4. *S. rhytzii* 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 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.

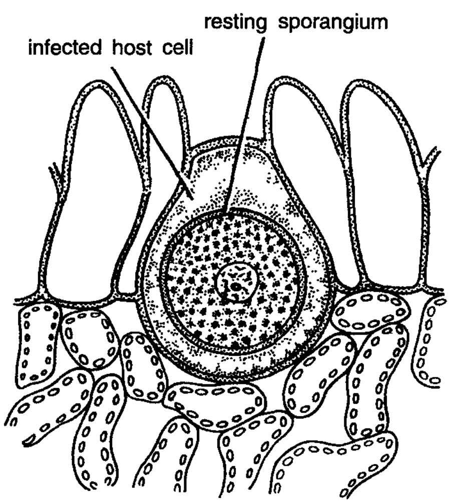


Fig. 3. ***Synchytrium.*** Resting sporangium in host cell.

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 may 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	—	***Saprolegnia***

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.

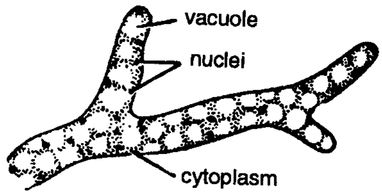


Fig. 2. ***Saprolegnia.*** A portion of mycelium to show vegetative structure.

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 of 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.

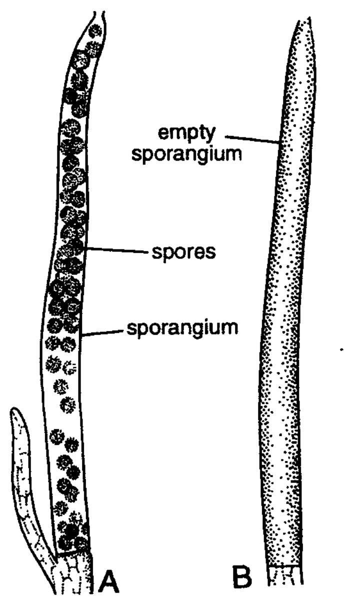


Fig. 3. ***Saprolegnia.*** A sporangium with **(A)** and without spores **(B)**.

2. 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.
3. A zoosporangium produces many pear shaped biflagellate (flagella apical) and uninucleate zoospores.
4. These primary zoospores produce secondary bean-shaped zoospores showing two laterally attached flagella.
5. The secondary zoospores encyst and later germinate into new hyphae.
6. The production of 2 types of zoospores is called diplanetic and phenomenon as diplanetism.
7. The new sporangia are formed by proliferation within the empty ones.
8. 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

1. The sexual reproduction is oogamous and most of the species are homothallic.
2. 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 monoclinous, diclinous or androgynous.
3. Oogonia are spherical and smooth walled.
4. Each oogonium has 1-20 uninucleate oospheres.
5. The oospheres are spherical and dark in colour owing to the presence of oil.
6. The antheridia are narrow, branched and multinucleate.
7. The antheridia grow closely apposed to the oogonia. Each sends a multinucleate ferilization tube which may be branched. It penetrates the oogonial wall and comes in contact with one or more eggs.

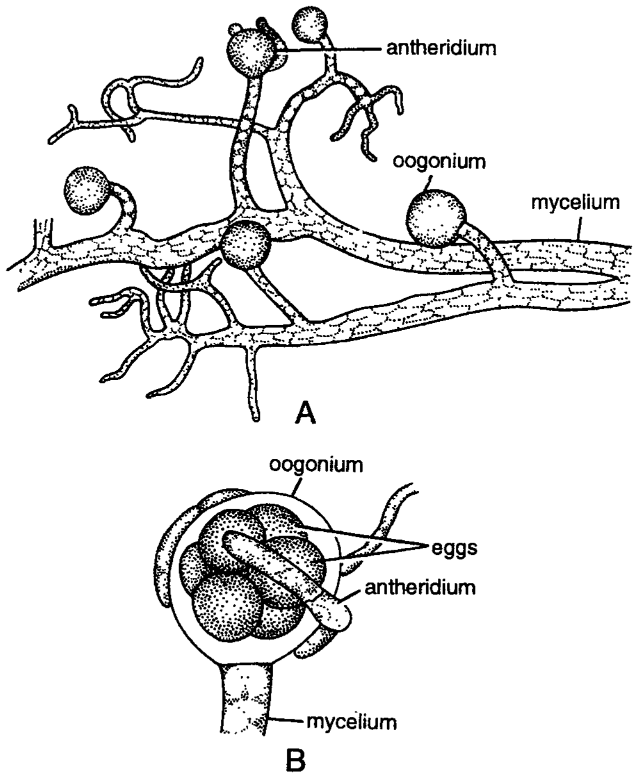


Fig. 4. ***Saprolegnia.*** A portion of mycelium showing **A.** Young and **B.** Mature sex organs.

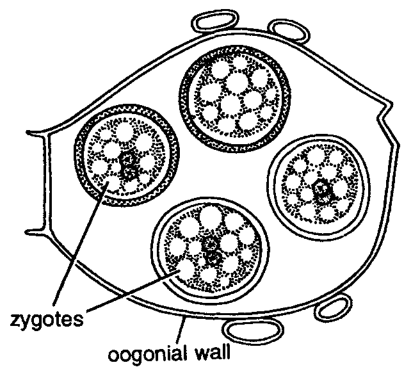


Fig. 5. ***Saprolegnia.*** Oogonium with zygotes.

8. The eggs get fertilized and one to many thick-walled oospores are formed within an oogonium.
9. 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	—	***Achlya***

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.

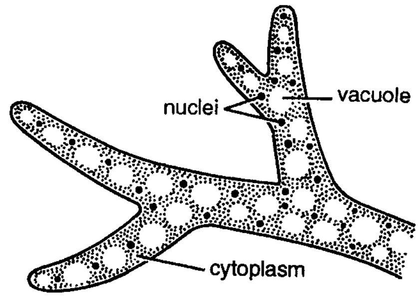


Fig. 1. ***Achlya.*** A portion of mycelium.

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.

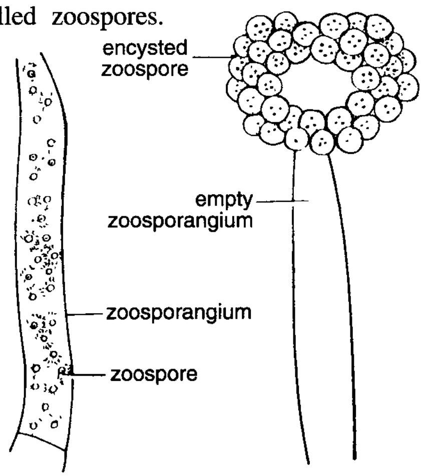


Fig. 2. ***Achlya.*** **A.** Mycelium showing zoospores inside zoosporangium, **B.** Empty zoosporangium with encysted zoospores at its tip.

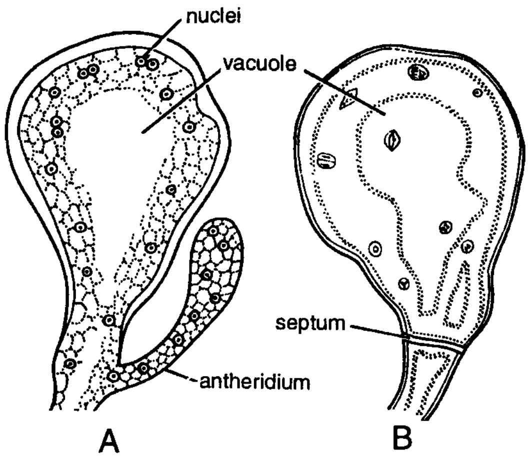


Fig. 3. ***Achlya.*** **A.** Oogonium with antheridium at its base, **B.** Oogonium with septum at its base.

2. 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.
3. 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.
4. 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

1. Sexual reproduction is oogamous. Most of the species are monoecious, but a few species are dioecious (e.g. *A. ambisexualis*, *A. bisexualis*).

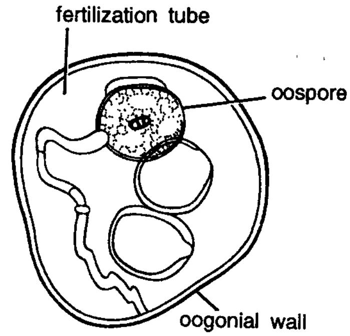


Fig. 4. ***Achlya.*** Oogonium with oospores.

2. 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).
3. 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*).
4. Each oogonium has 1-8 eggs (or oospheres) and rarely upto 50.
5. Each egg (oosphere) is surrounded by a thin delicate wall, contains dense cytoplasm and a single nucleus.
6. The antheridia are narrow, branched and multinucleate. Each is cut off from rest of the hyphae by a septum at its base.
7. Antheridia grow closely applied to oogonium, send fertilization tubes which penetrate the oogonial wall and fertilize the eggs.
8. 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	—	***Phytophthora***

Exercise 1

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.*

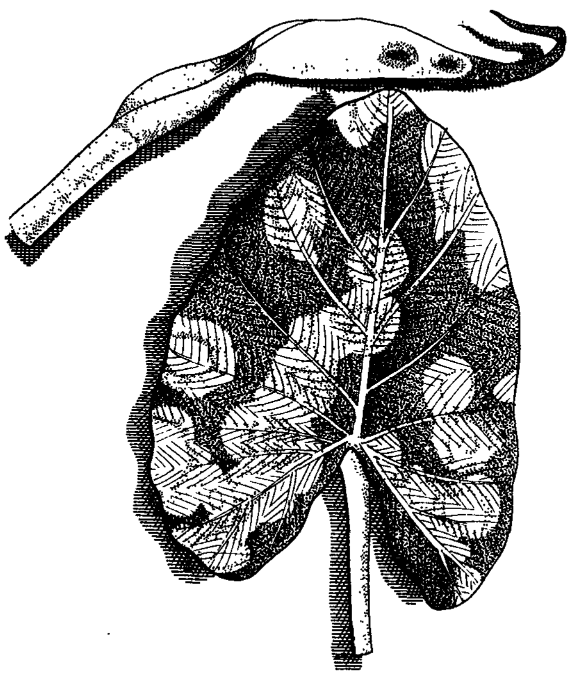


Fig. 2. ***Phytophthora.*** Inflorescence and the leaf of *Colocasia* infected by *P. colocasiae.*

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. *Phytòphthora* 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.

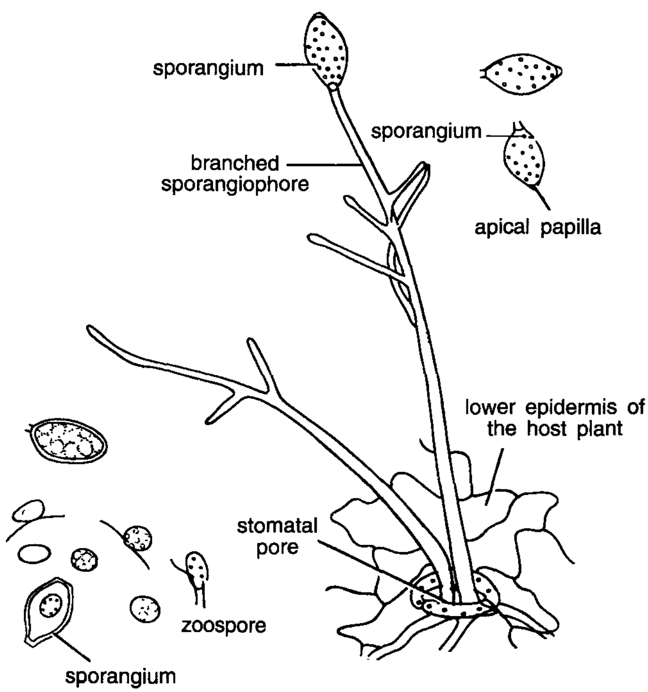


Fig. 3. ***Phytophthora.*** T. s. of infected leaf showing sporangiophores with sporangia.

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 (oospoes 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.

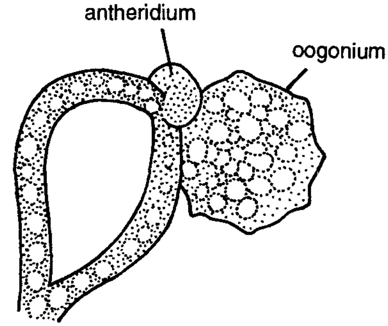


Fig. 4. ***Phytophthora.*** Sex organs.

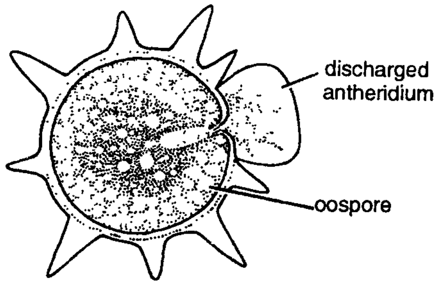


Fig. 5. ***Phytophthora.*** Oospore.

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**. Sporangiophores 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 *Phytophythora* 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	—	***Albugo***

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*; vern. phool gobhi), mustard (*Brassica campestris*; vern. *sarsaun*), radish (*Raphanus sativus*; vern. mooli) and toria (*Eruca sativa*; vern. tarra) etc.

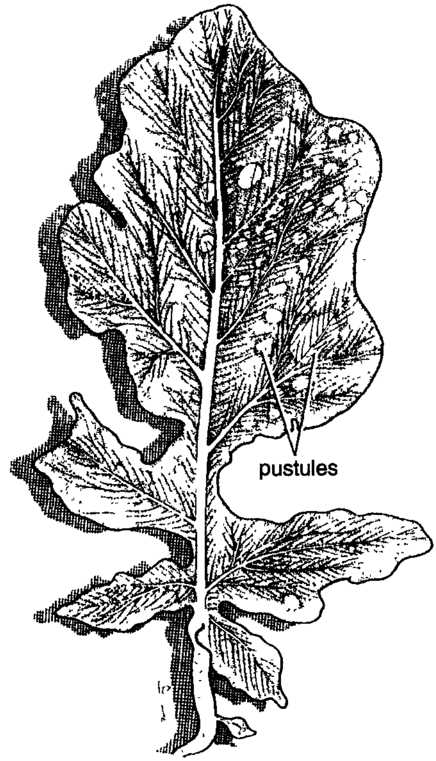


Fig. 1. ***Cystopus.*** Infected leaf of radish (*Raphanus sativus*) showing white pustules.

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.

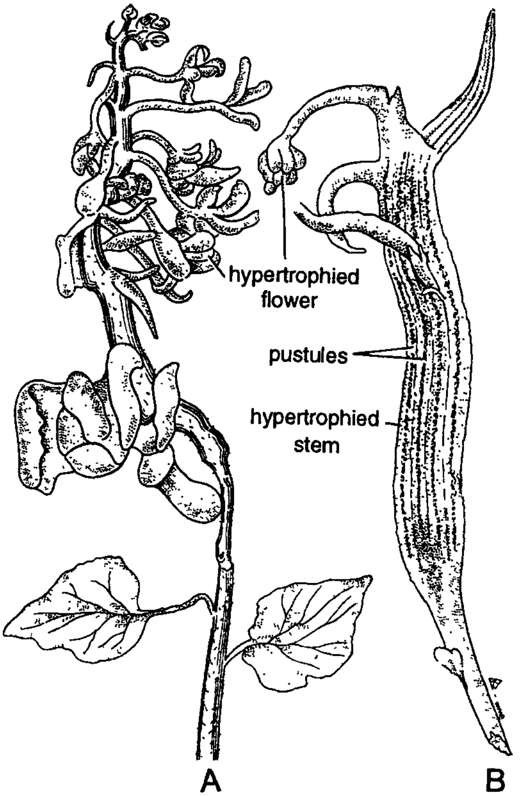


Fig. 2. ***Cystopus.*** Infection causing enormous hypertrophy in **A.** *Raphanus sativus* and **B.** *Eruca sativa.*

2. *A. ipomoeae-pandoranae* attacks *Ipomoea* species (fam. Convolvulanceae) and forms appreciable galls on the stem due to hypertrophy.
3. *A. bliti* is common on members of Amarantaceae.
4. *A. portulacae* infects leaves of *Portulaca* species.

Exercise 2

***Object* : Study the vegetative structure.**

Work procedure

Cut a section though 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 disjunctor.

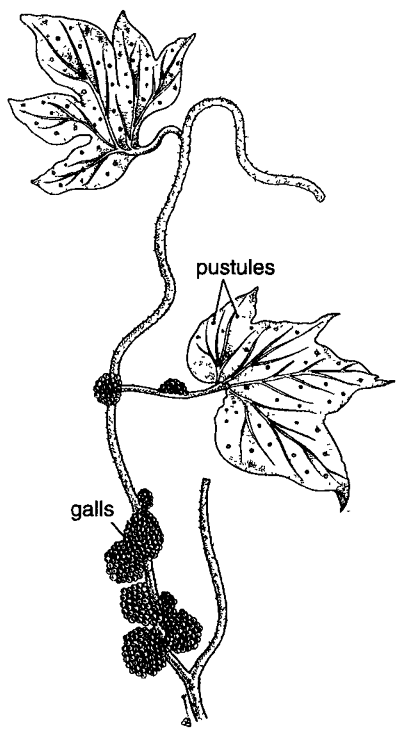


Fig. 3. ***Cystopus.*** *Ipomoea* sps. infected by *C. ipomoea pandoranae.*

5. Each conidium is multinucleate (5-8 nucleate), hyaline, smooth and spherical.
6. The conidiophores constantly cut off conidia forcing underlying epidermis to rupture.
7. 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).
8. 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.

Comments

1. Sexual reproduction is oogamous. The sex organs are generally formed in the stem towards the end of the growing season of the host.
2. 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).
3. The antheridium is paragynous in position and contains several nuclei.
4. The antheridia and oogonia are separated from remainder of the mycelium by a septum.
5. 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.
6. A single male nucleus enters and fuses with the nucleus in the oosphere thus effecting fertilization.
7. In some species (e.g. *A. bliti*), the oosphere or ooplasm is multinucleate and these nuclei get

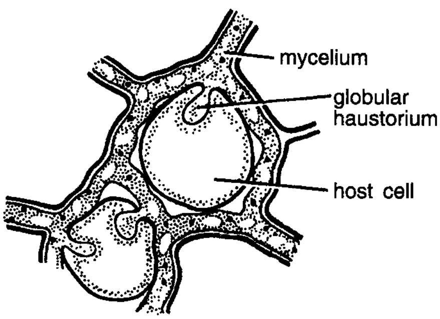


Fig. 4. ***Cystopus.*** A portion of intercellular mycelium.

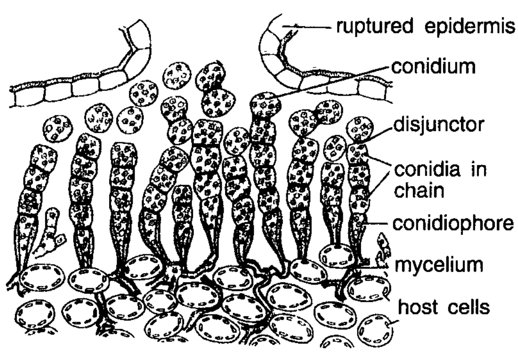


Fig. 5. ***Cystopus.*** T.s. of infected host leaf showing ruptured epidermis and conidia in chains.

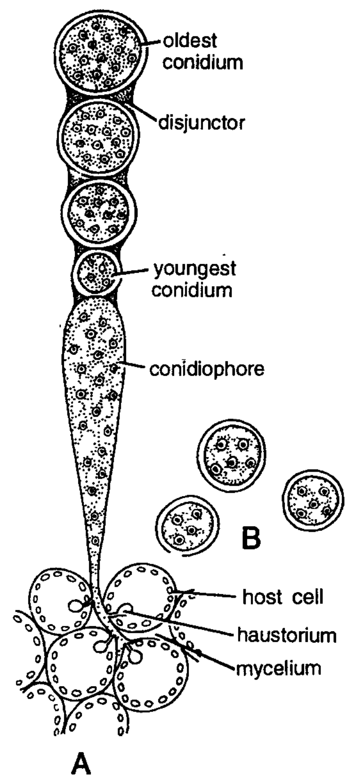


Fig. 6. ***Cystopus.*** **A.** Single conidiophore showing conidia in chain **B.** Conidia.

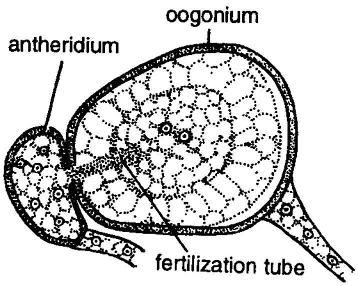


Fig. 7. ***Cystopus.*** Oogonium and antheridium.

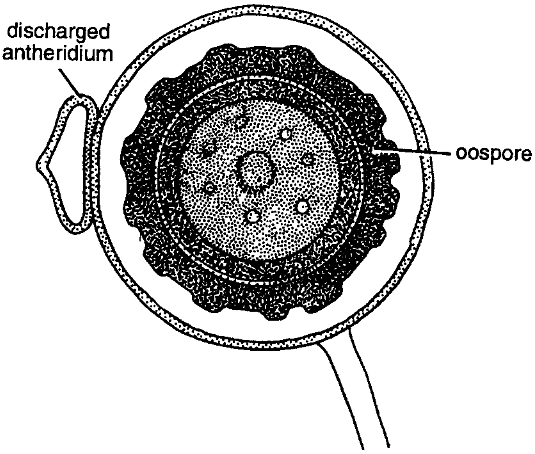


Fig. 8. ***Cystopus.*** Oospore.

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* sps. is a weed in the crop fields. *Cystopus* occurs on these hosts in January-February. *Cystopus* is also common on *Achyranthes* sps. (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	—	***Rhizopus***

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.

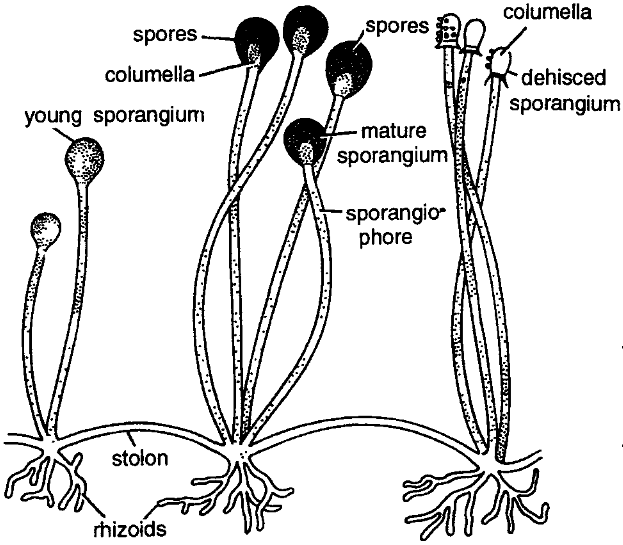


Fig. 1. *Rhizopus*. A portion of mycelium with sporangiophores.

Comments

Most of the species of *Rhizopus* are saprophytie 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. artocarpi* and *R. arrhizus* are weak parasites.

Only a few species of *Rhizopus* attack the plants.

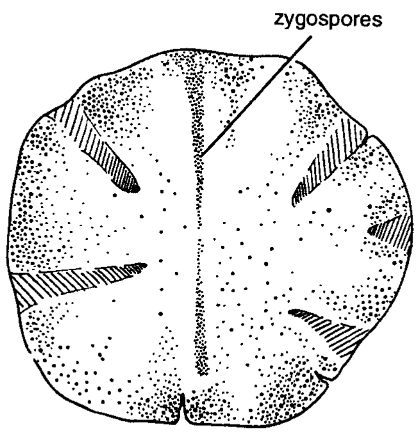


Fig. 2. *Rhizopus*. Potato culture plate showing heterothallism.

1. *R. artocarpi* 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. artocarpi* 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.

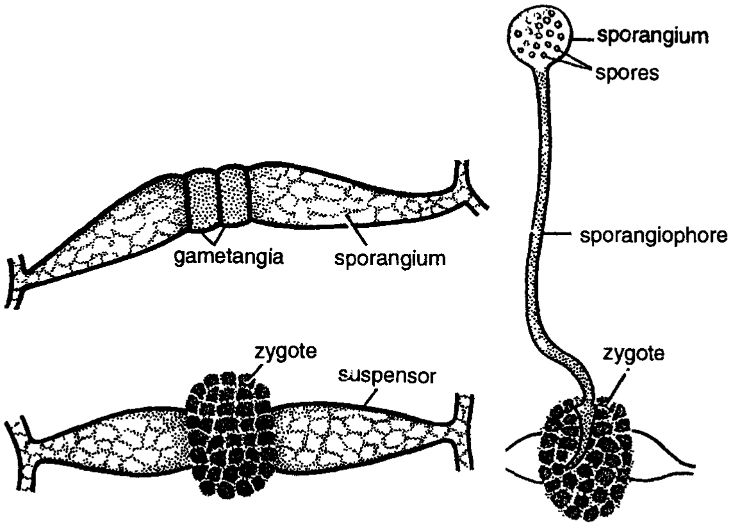


Fig. 3. ***Rhizopus.*** Various stages in sexual reproduction.

3. 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.
4. 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.
5. Spores are liberated by breaking of the sporangial wall.
6. Each spore germinates to form a 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

1. The genus is heterothallic and the sexual reproduction takes place only when mycelia of + (plus) and – (minus) strains meet.
2. 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.
3. 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.
4. The two isogametangia (also called coenogametangia) conjugate to form a zygospore (zygote) which has a thick wall and rough outer surface.
5. Zygospore divides meiotically after a period of rest. It germinates by producing a promycelium which develops a sporangium (often called zygosporangium) at its tip.
6. The sporangium produces many multinucleate spores. These germinate 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.

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***. Sporangiophores 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 chlamydospores.

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 sinesis*, *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

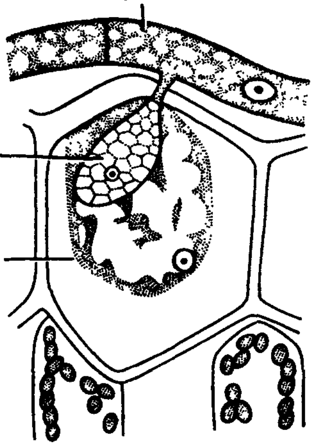


Fig. 1. ***Erysiphe.*** Haustorium of *E. cichoracearum* inside the host cell.

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.

2. *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.
3. *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.

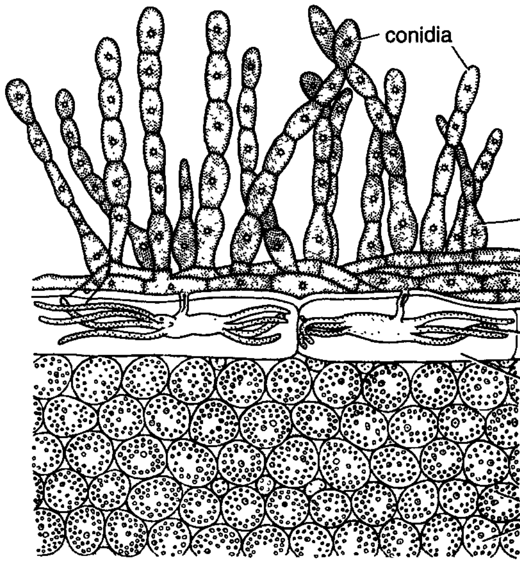


Fig. 2. ***Erysiphe.*** Section through host leaf showing haustoria, conidiophores and conidia.

Comments

1. Since the fungus is an ectoparasite, the mycelium forms a white, interwoven covering on the host surface.
2. The branched mycelium is septate and the cells are uninucleate.
3. Mycelium produces simple, globose, lobed haustoria which penetrate the epidermal cells of the host.
4. 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

1. The conidia are asexual reproductive bodies formed in chains at the tips of conidiophores.

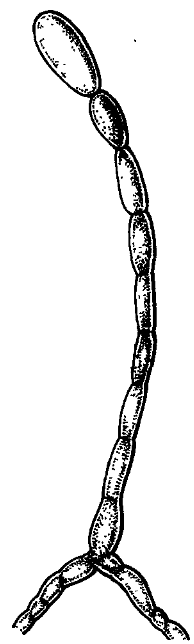


Fig. 3. *Erysiphe.* A conidiophore with conidia in chains.

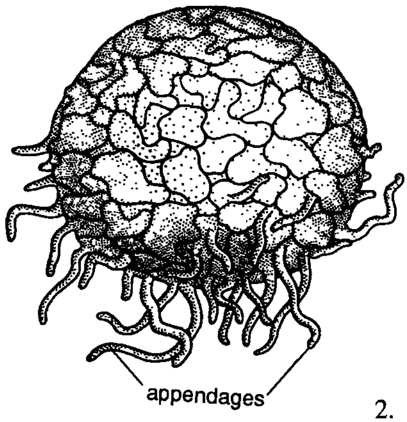


Fig. 4. *Erysiphe.* An ascocarp.

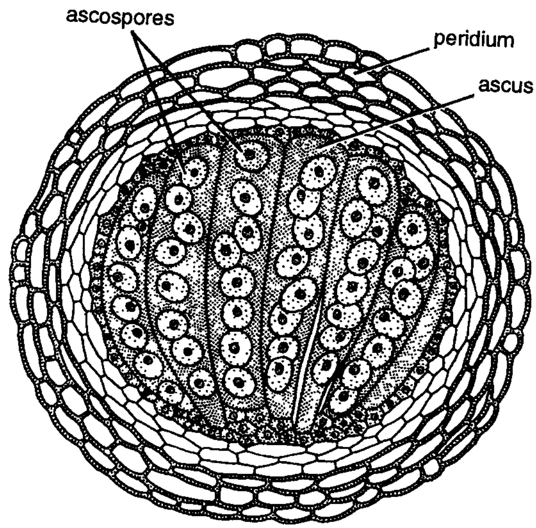


Fig. 5. *Erysiphe.* V.s. through an ascocarp.

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 supper 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-dividison	—	**Ascomycotina**
Class	—	**Plectomycetes**
Order	—	**Erysiphales**
Family	—	**Erysiphaceae**
Genus	—	***Sphaerotheca***

Exercise 1

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

Work procedure

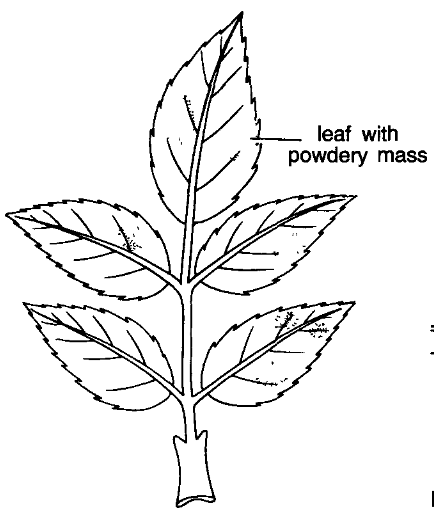


Fig. 1. *Sphaerotheca.* A rose leaf showing powdery mildew, caused by *S. pannosa.*

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.

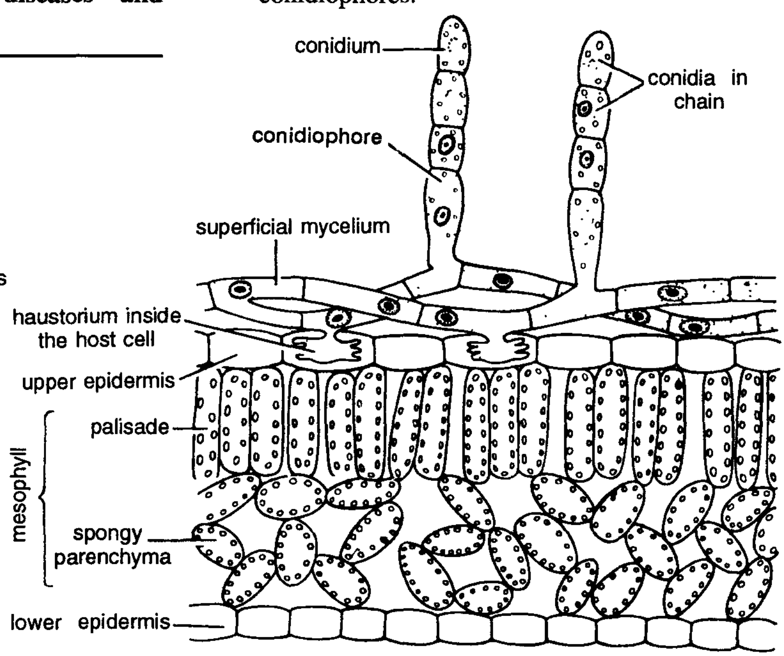


Fig. 2. *Sphaerotheca.* T.s. of infected host leaf showing mycelium and haustoria.

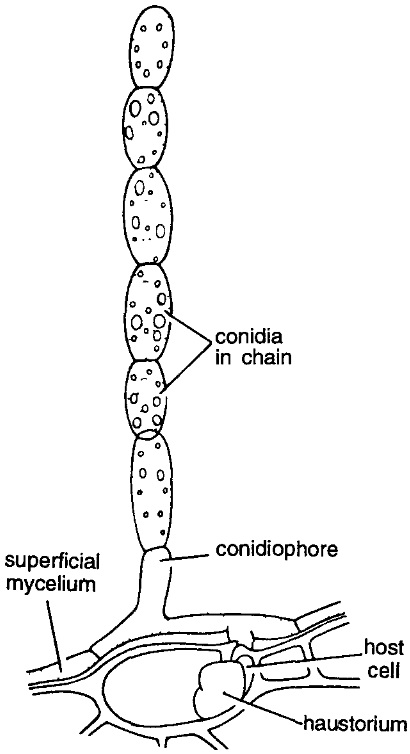


Fig. 3. *Sphaerheca.* Conidiophore with a chain of conidia.

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.

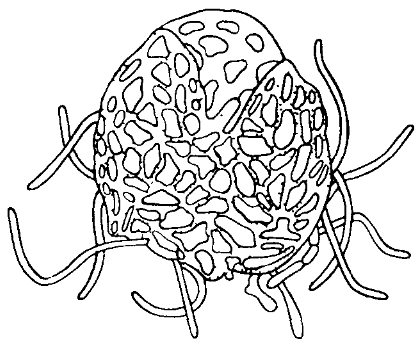


Fig. 4. *Sphaerotheca.* A single cleistothecium.

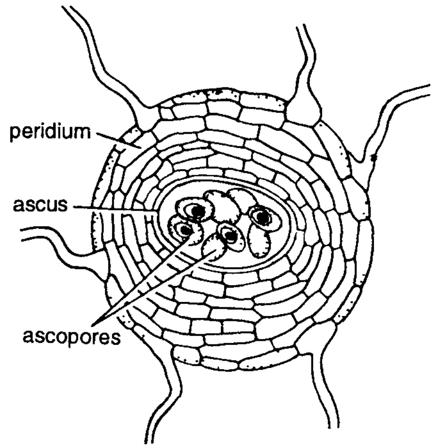


Fig. 5. *Sphaerotheca.* T.s. through cleistothecium.

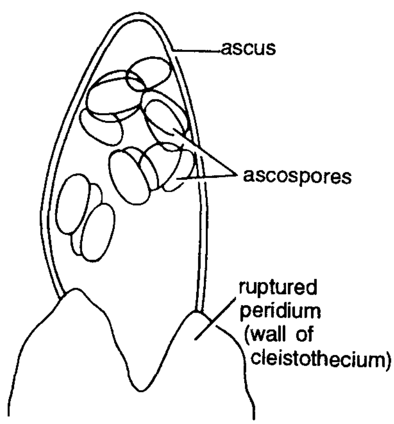


Fig. 6. *Sphaerotheca.* A ruptured cleistothecium showing single ascus.

2. Cleistothecium is globose and dark brown in colour. It bears simple myceloid appendages all over the surface.

3. The wall of the cleistothecium is thick and two layered. Each layer is several cells thick. The outer cells are darker.
4. 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 appearane.
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	—	***Phyllactinia***

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.

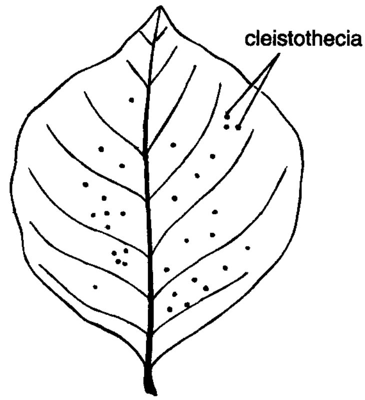


Fig. 1. *Phyllactinia*. Leaf of *Dalbergia sisso* showing cleistothecia.

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.

1. *P. guttata* (= *P. corylea*) is a very common species parasitizing *Corylus, Betula* (vern. bhojpatra; fam. Betulaceae), *Cassia fistula* (vern. amaltass; fam. Caesalpiniaceae), etc.
2. *P. dalbergiae* is very common in northern India and attacks *Dalbergia sissoo* (vern. shisham; fam. Papilionaceae).
3. *P. moricola* is found on leaves of *Morus alba* (vern. shahtoot; fam. Moraceae).
4. *P. acaciae* attacks some species of *Acacia* (fam. Mimosaceae).
5. *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.

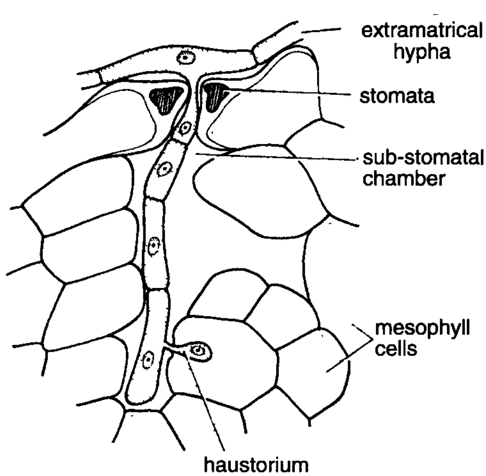


Fig. 2. ***Phyllactinia.*** Mycelium showing haustoria in the host cell.

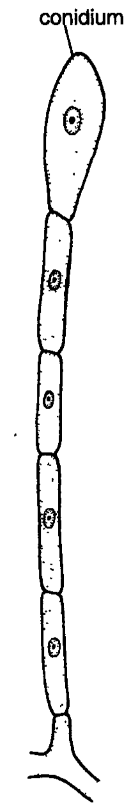


Fig. 3. ***Phyllactinia.*** A conidiophore with conidium.

4. Each conidium is clavate, thin-walled and uninucleate.

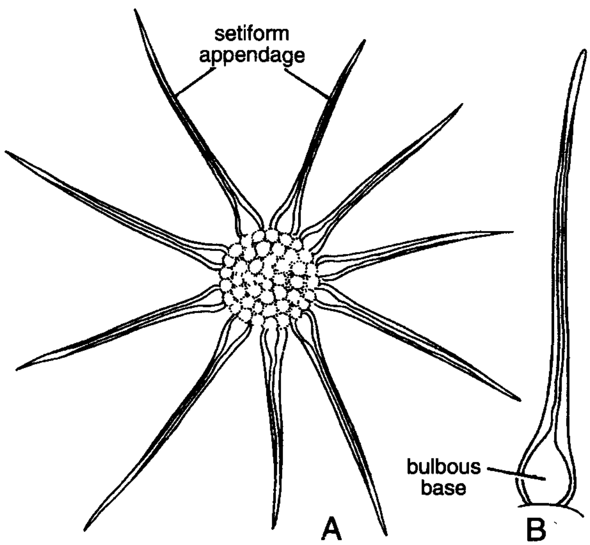


Fig. 4. ***Phyllactinia.*** **A.** Cleistothecium, **B.** Appendage.

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.

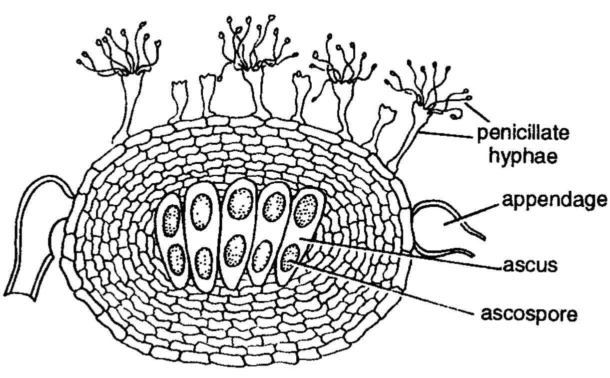


Fig. 5. *Phyllactinia.* V.s. cleistothecium.

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) Hemiendophyte, (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	—	***Aspergillus***

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

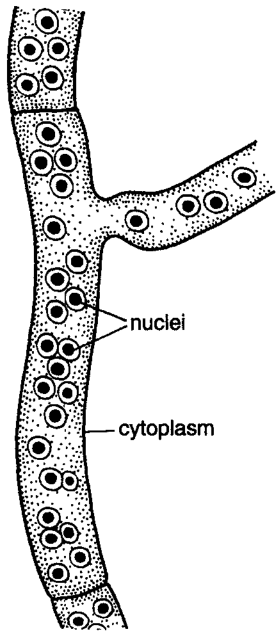


Fig. 1. ***Aspergillus.*** A portion of the mycelium.

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.

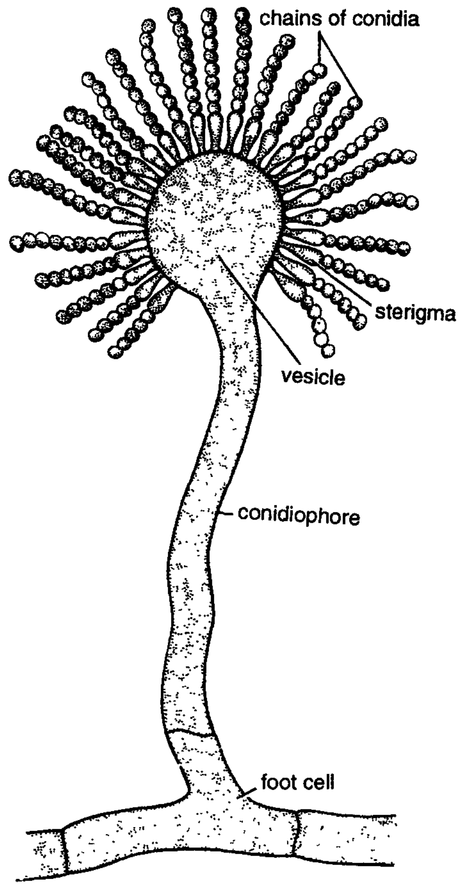


Fig. 2. ***Aspergillus.*** A mature conidiophore with chains of conidia.

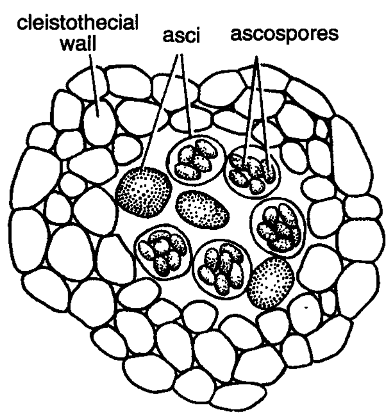


Fig. 3. ***Aspergillus.*** Cross section of a cleistothecium showing asci.

Comments

1. The mycelium is well developed, profusely branched and septate.

2. 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).
3. 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

1. Conidia are asexual reproductive units borne on conidiophores.
2. Each conidiophore arises from the foot cell of the mycelium and is long and erect hypha, terminating in a bulbous head—the vesicle.
3. The vesicle develops a number of bottle-shaped structures called the sterigmata (sing. sterigma) over its entire surface.
4. 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.
5. Each sterigmata cuts off a chain of basipetally arranged conidia.
6. The conidia are coloured and the colour of the conidia depends upon the species.
7. Each conidium is uninucleate with an outer finely spiny epispore and inner smooth endospore.
8. 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

1. The species of *Aspergillus* developing perfect stage i.e. cleistothecium are placed under the genus *Eurotium.*
2. The ascospores are produced after reduction division of ascospore mother cell in sac-like cell—the ascus.
3. The asci are developed inside a fruiting body–the cleistothecium.
4. Cleistothecium consists of wall called peridium formed by somatic hyphae, enclosing many asci.
5. Each ascus has eight uninucleate pulley wheel-like ascospores with an outer sculptured epispore and an inner smooth endospore.
6. 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	—	***Penicillium***

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).

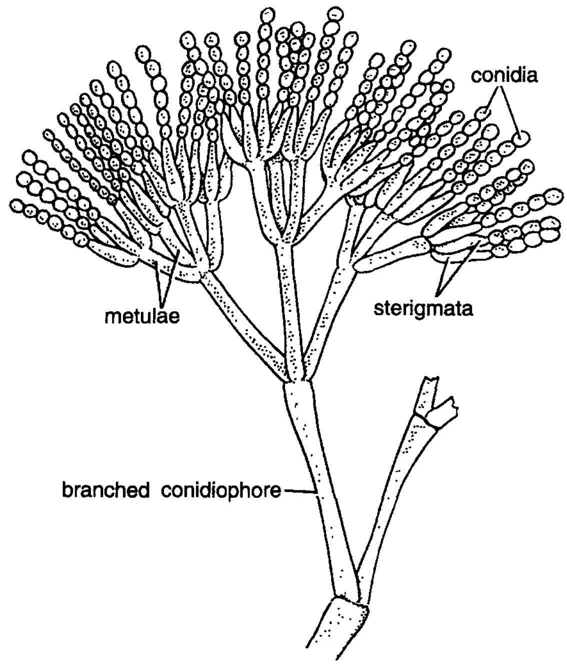


Fig. 1. ***Penicillium.*** Mycelium bearing conidiophore and chains of conidia.

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.

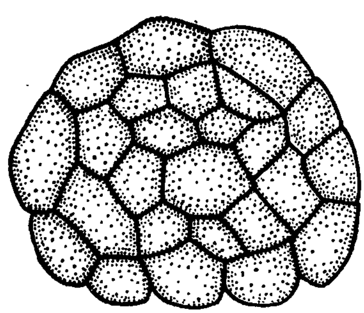


Fig. 2. ***Penicillium.*** A cleistothecium.

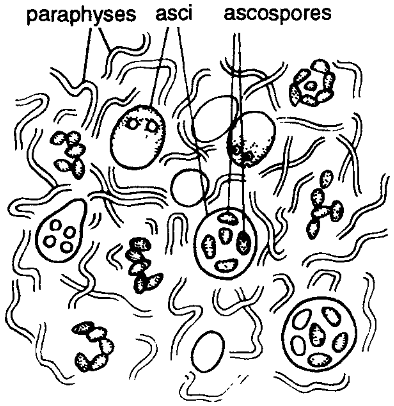


Fig. 3. ***Penicillium.*** A part of cleistothecium in cross section.

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, Teleromyces, 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

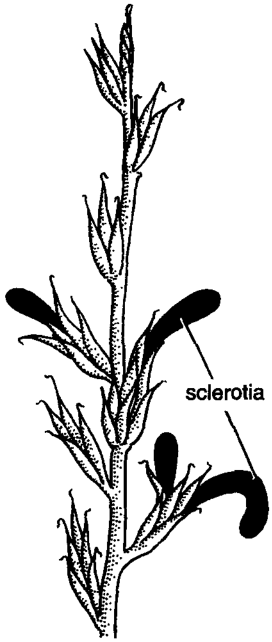


Fig. 1. *Claviceps.* An infected inflorescence of rye showing sclerotia.

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.

4. 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

1. Ascospores or conidiospores germinate on stigma, produce a germ tube that reaches the ovules and later develop mycelium in the grain.
2. The mycelium is white, branched, septate and the cells are multinucleate.

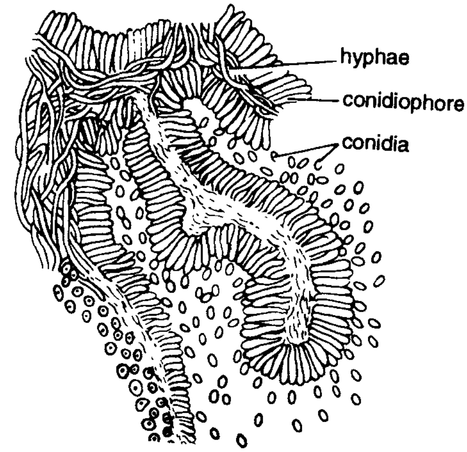


Fig. 2. *Claviceps.* Mycelium showing conidiophores.

Exercise 3

***Object* : Study of conidia.**

Work procedure

Crush the rye grain and locate conidiophores. Stain in cotton blue and mount in lactophenol.

Comments

1. The conidia are formed inside the grain at the tips of short conidiophores which form acervulus-like layers.
2. The conidia are minute, ovoid and uninucleate. These are budded off by each conidiophore.

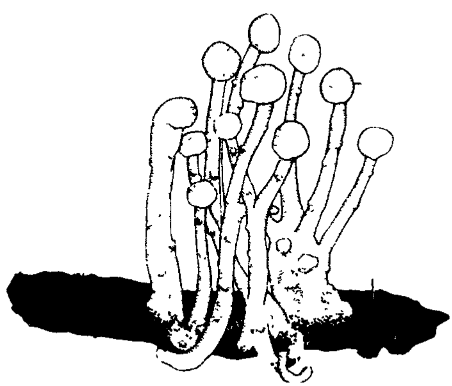


Fig. 3. *Claviceps.* Germinating sclerotium.

3. 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.
4. 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

1. The sclerotia are black and elongated structures which replace the grains in the inflorescence.
2. After winter, the black sclerotia germinate and form many long, stalked, mushroom-like and dark purple stromata with globose heads.
3. The stromata are nearly 1 cm in length.
4. Each stromatal head, just below its surface, has many minute flask-shaped cavities, known as perithecia, surrounded by the pseudoparenchymatous stromatic tissue.
5. Each mature perithecium is a flask-shaped cavity opening at the apex through a small pore—an ostiole.
6. Near the ostiole are present some hair-like structures known as paraphyses.

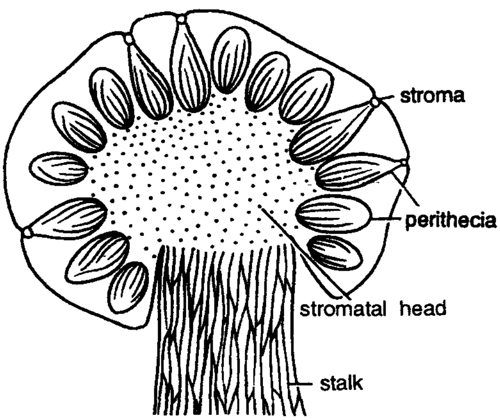


Fig. 4. ***Claviceps.*** V.s. stromata.

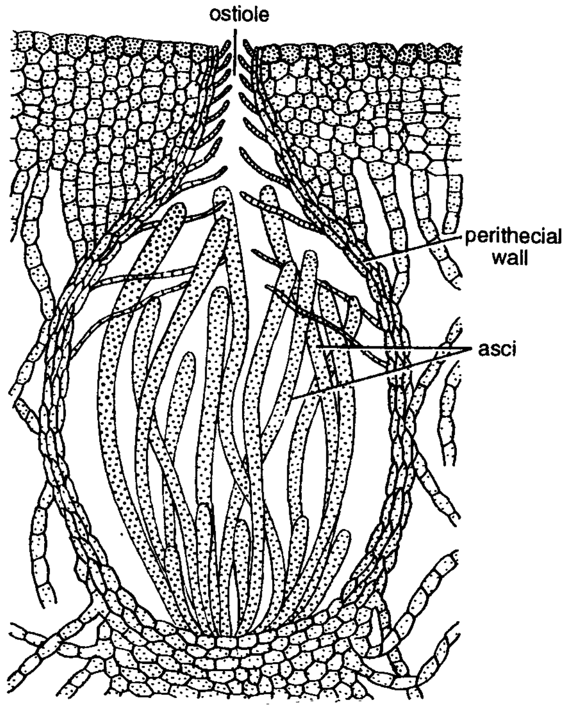


Fig. 5. ***Claviceps.*** V.s. ascocarp.

7. The asci are produced in tufts from the base of the perithecium.
8. An ascus is narrow, elongated and somewhat curved. It has a thick cap at apex through which the ascospores are discharged.
9. Each ascus produces eight haploid, uninucleate elongated and acicular ascospores that lie parallel to one another.

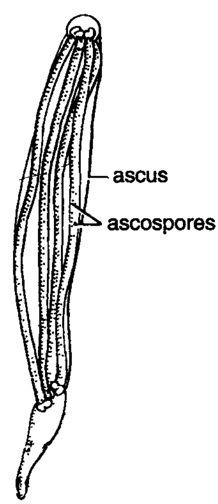


Fig. 6. ***Claviceps.*** Ascus showing ascospores.

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.

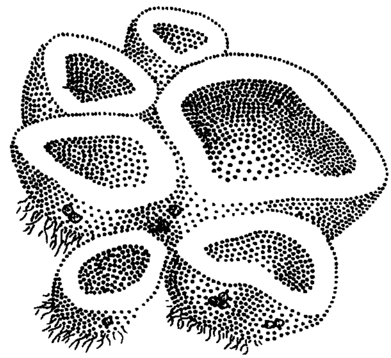


Fig. 1. ***Peziza.*** Ascocarps growing on decaying wood.

Exercise 2

Object **: Study the conidia/chlamydospores.**

Work procedure

Study the mycelium for the presence of conidia/ chlamydospores.

Comments

1. The conidia are the asexual reproductive bodies. These are produced rarely.

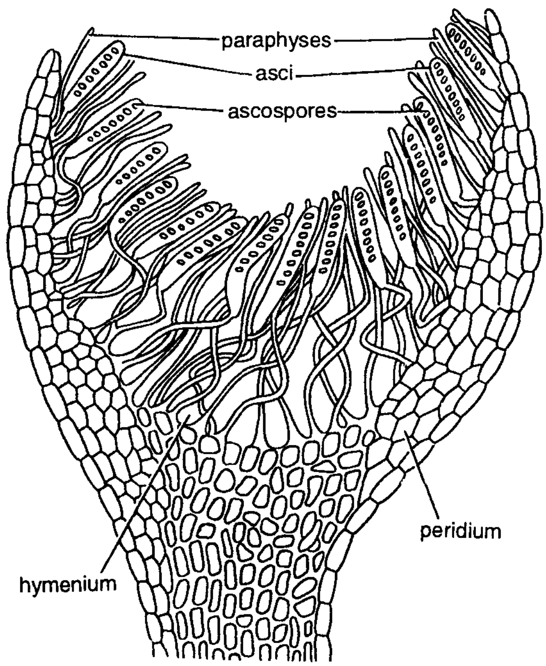


Fig. 2. ***Peziza*** V.s. ascocarp.

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 hypothectum 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.

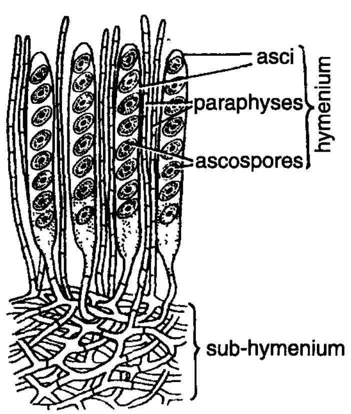


Fig. 3. *Peziza.* V.s ascocarp (portion magnified).

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	—	***Ascobolus***

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.

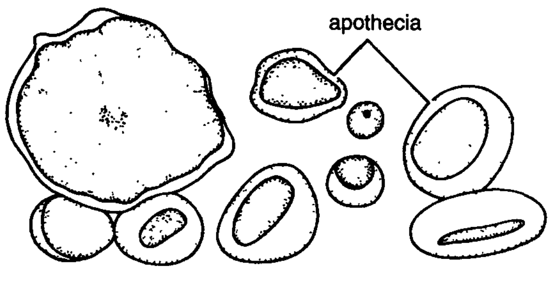


Fig. 1. ***Ascobolus.*** Ascocarps.

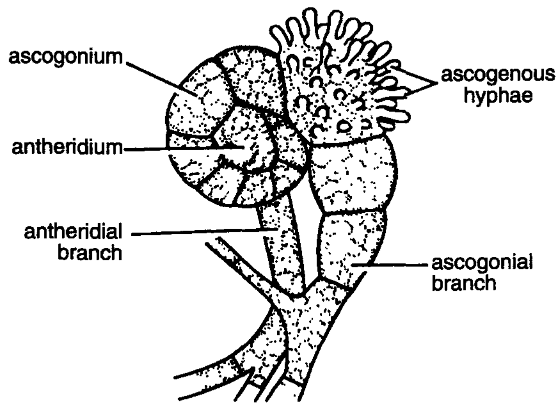


Fig. 2. ***Ascobolus.*** Sex organs.

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 moddle 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.

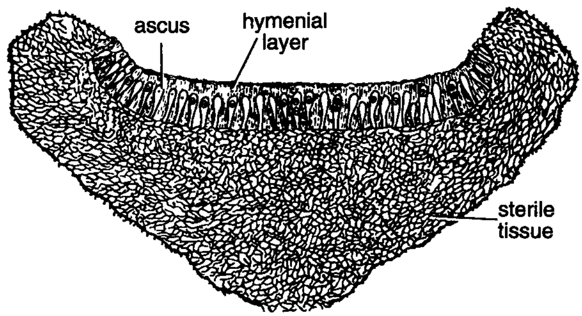


Fig. 3. ***Ascobolus.*** V.s. apothecium.

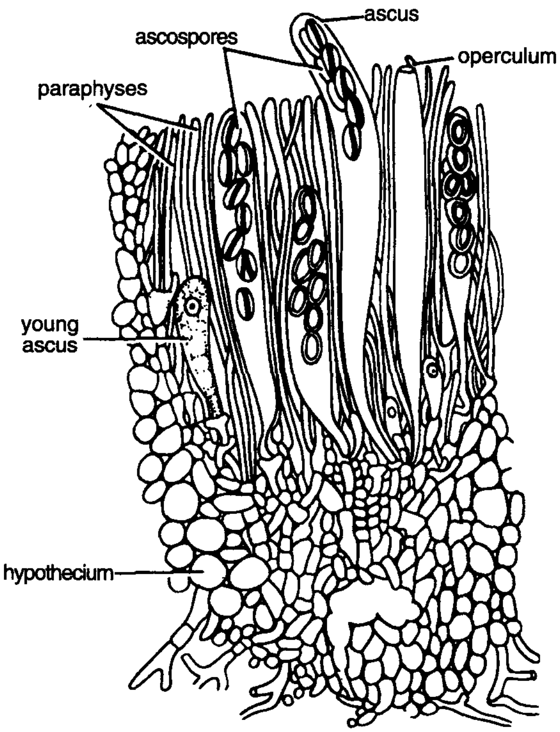


Fig. 4. ***Ascobolus.*** V.s. apothecium—a part enlarged.

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	—	***Morchella***

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.

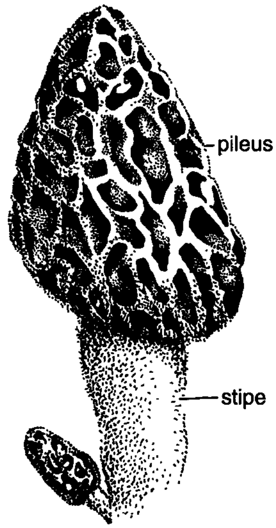


Fig. 2. ***Morchella.*** An ascocarp.

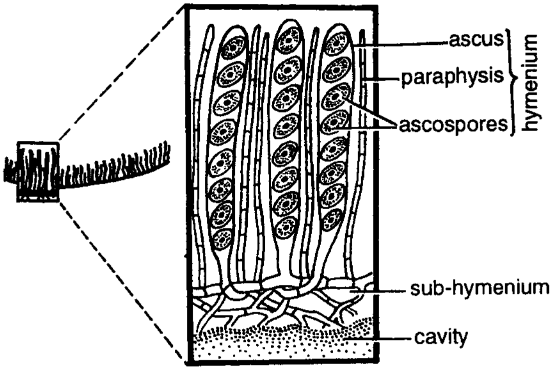


Fig. 3. ***Morchella.*** A portion of a section through depression of pileus.

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 lip.
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 the ascocarp. The stipe is cream coloured, thick, fleshly 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	—	***Ustilago***

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.

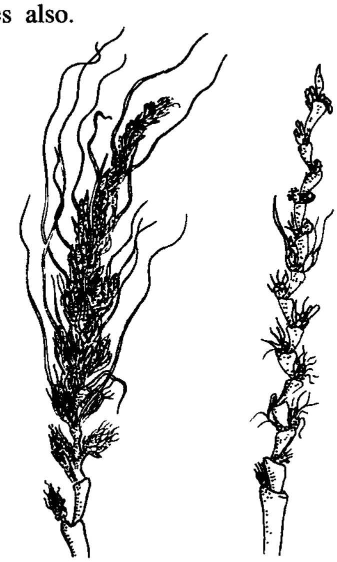


Fig. 1. *Ustilago*. Loose smut of wheat. Wheat ears infected by *U.tritici*.

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 resuls 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.

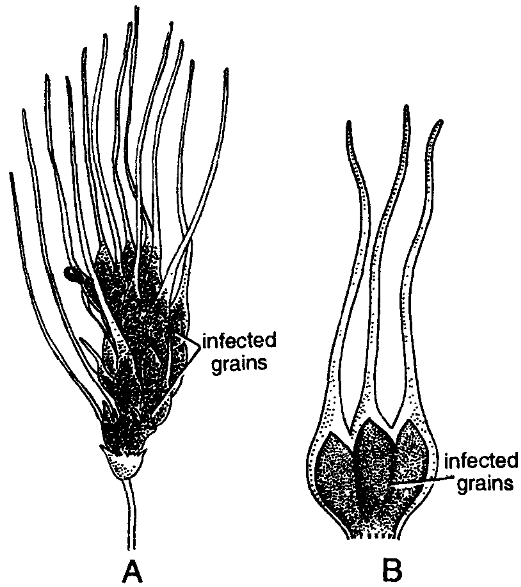


Fig. 2. ***Ustilago*. A.** Covered smut of barley. Spike of barley infected by *U. hordei*, **B.** Spikelet magnified.

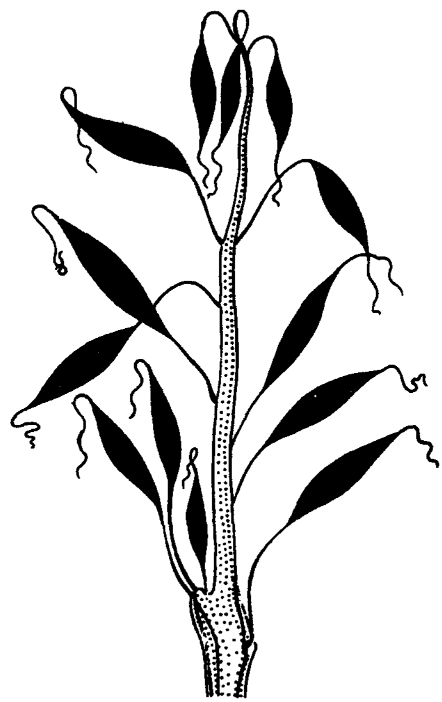


Fig. 3. ***Ustilago*.** Covered smut of oat. Oat inflorescence infected by *U. kolleri*.

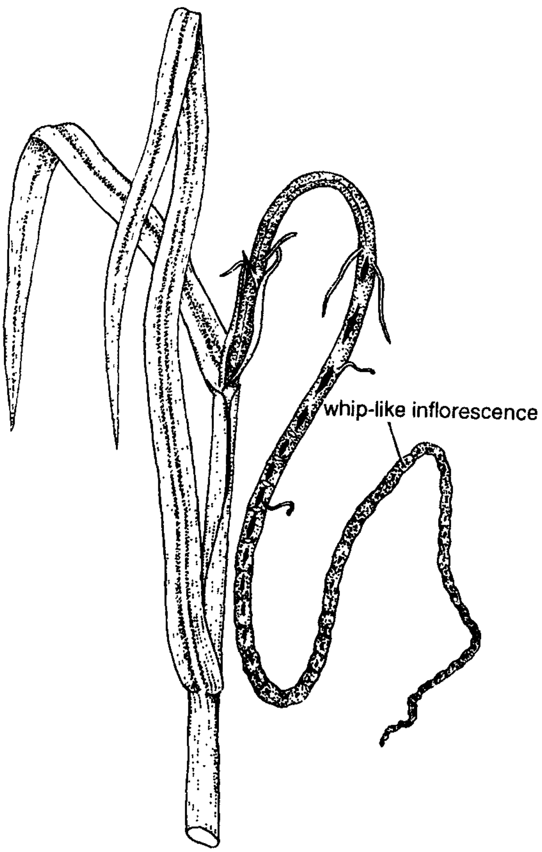


Fig. 4. ***Ustilago.*** Whip smut of sugarcane caused by *U. scitaminae.*

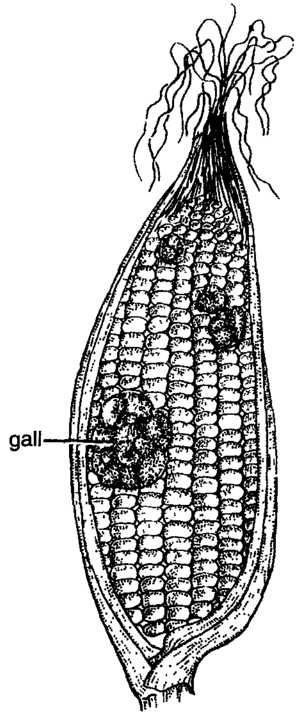


Fig. 5. ***Ustilago.*** Common smut of maize—Maize cob infected by *U. maydis.*

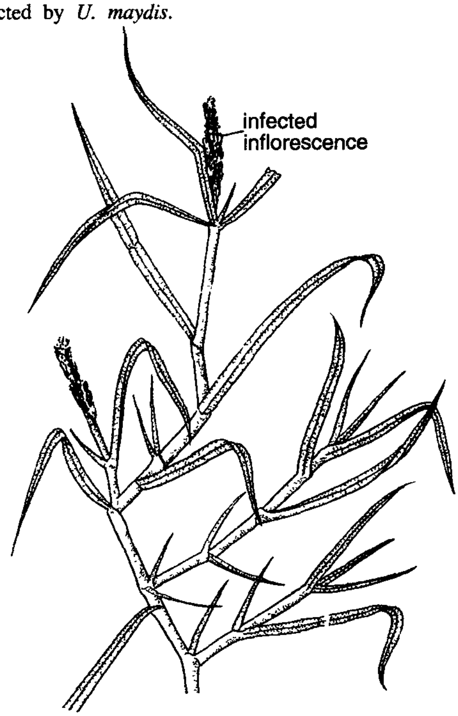


Fig. 6. ***Ustilago.*** Grass spike infected by *U. cynodontis.*

7. Other species of *Ustilago* infecting economically useful plants include *U. panicifrumentacei* and *U. paradoxa* on barnyard millet (*Echinocloa 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.

Exercise 2

Object : Study of vegetative structure.

Work procedure

Break open or crush the spikelet when young. Search for the mycelium, if seen, stain with cotton blue and mount in lactophenol.

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.

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) Characteistic 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.

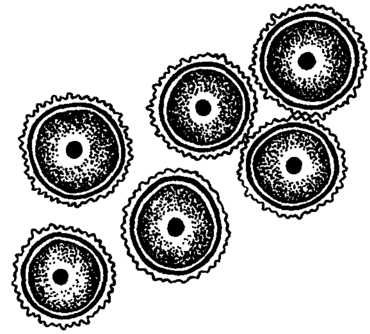


Fig. 7. ***Ustilago.*** Chlamydospores.

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	—	***Puccinia***

Exercise 1

***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 (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

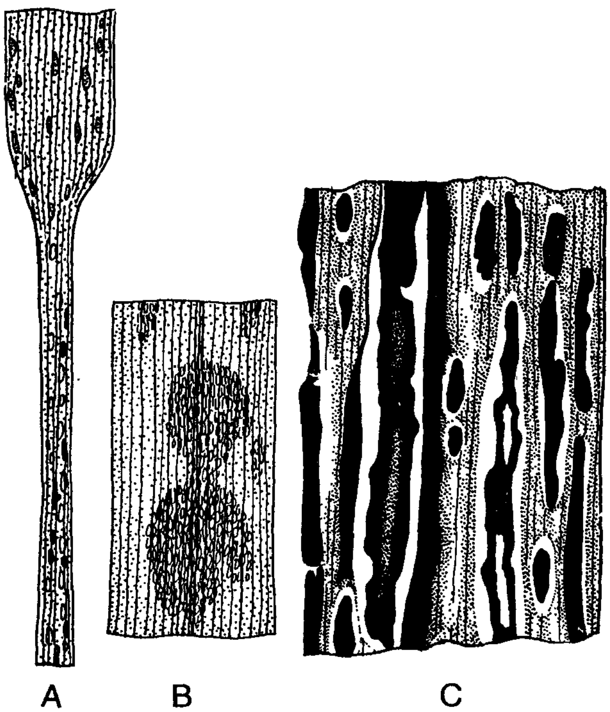


Fig. 1. *Puccinia graminis tritici*. Black or stem Rust. **A.** Leaf sheath showing pustules, **B.** A portion of leaf showing uredo-pustules, **C.** A portion of stem showing teleutopustules.

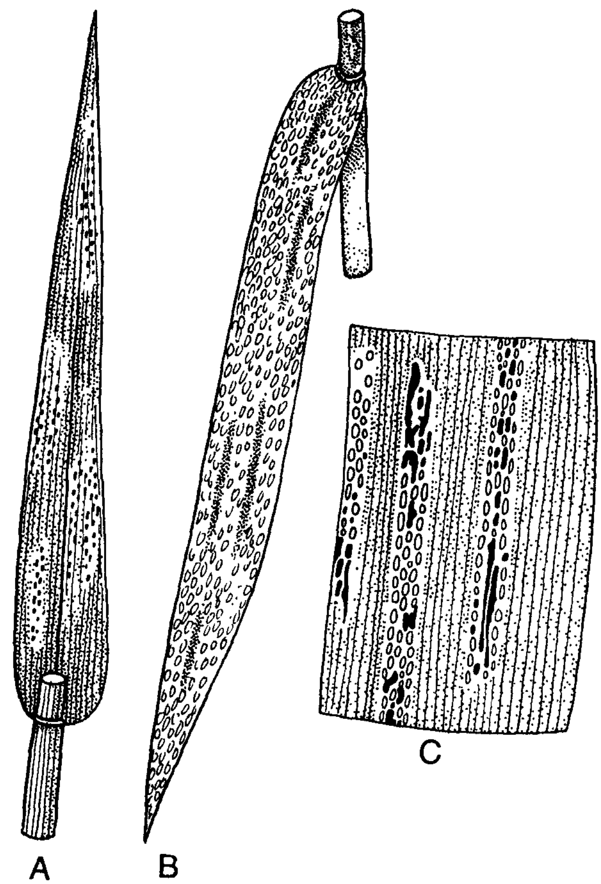


Fig. 2. *Puccinia striiformis*. Yellow or Stripe Rust. **A.** and **B.** Leaves showing pustules in the form of streaks **C.** A portion of leaf sheath showing uredo- and teleutopustules.

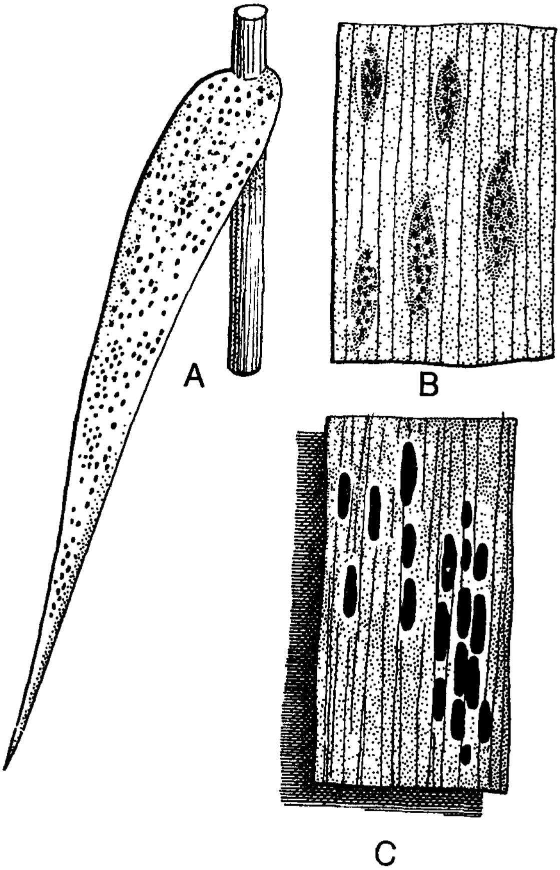


Fig. 3. ***Puccinia recondita.*** Orange Leaf Rust. **A.** Leaf showing pustules, **B.** A portion of leaf showing uredo-pustules, **C.** A portion of leaf showing teleutopustules.

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. triticina*) 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 host, *Thalictrum*, shows small brown specks in clusters on leaves.

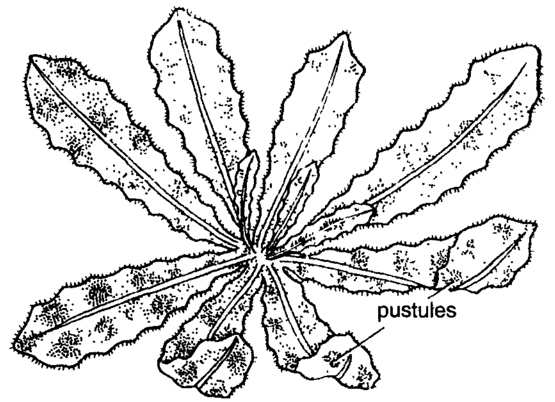


Fig. 4. ***Puccinia butleri.*** Infection on *Launea* sps.

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 diease 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. pennisetti* 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 telutosori 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.

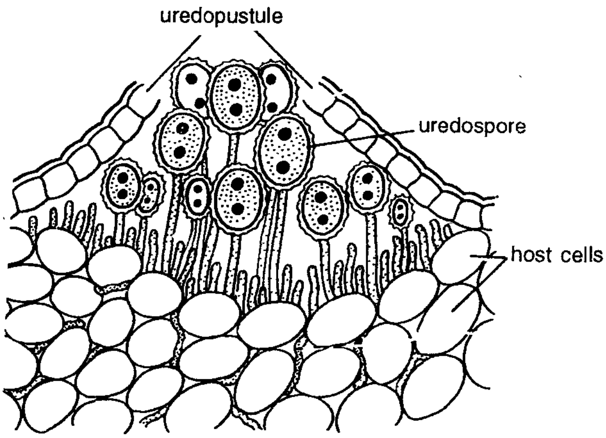


Fig. 5. ***Puccinia.*** T.s. of wheat leaf through uredopustule.

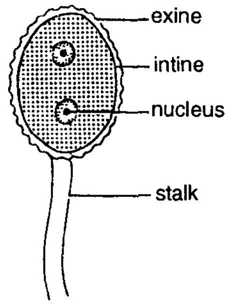


Fig. 6. ***Puccinia.*** Uredospore.

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 has 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 teleutosorus and teleutospores.**

Work procedure

These are found on wheat leaves. A T.s. of the leaf showing teleutosorus 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.

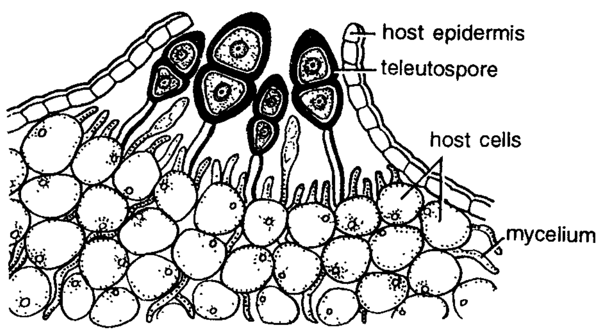


Fig. 7. ***Puccinia.*** T.s. of wheat leaf through teleutopustule.

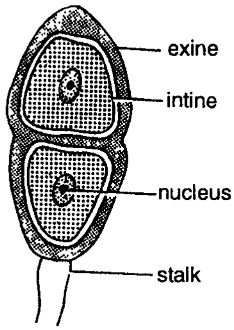


Fig. 8. ***Puccinia.*** Teleutospore.

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.

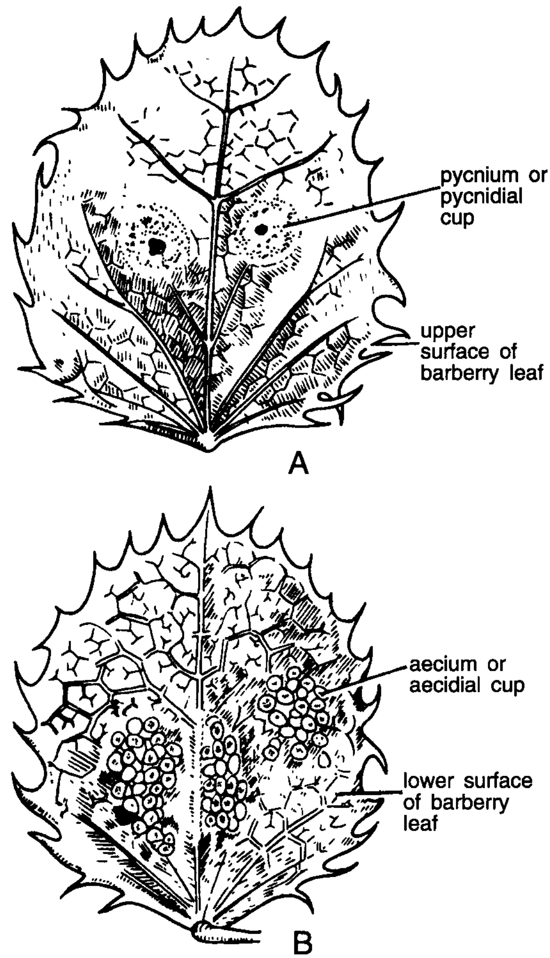


Fig. 9. ***Puccinia graminis.*** Leaves of barberry showing **A.** Pycnidial cups on upper surface **B.** Aecidial cups on lower surface.

Comments

1. Each basidiospore germinates on the leaf of alternate host producing the (monokaryotic) mycelium, that ultimately forms the pycnicial 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 periphysis and project through the ostiole.
5. Some of the periphyses are branched and thin walled. These are called receptive hyphae (or flexous hyphae). They project through the ostiole far beyond the periphyses.
6. The cavity of the pycnidium is lined by many elongated and uninucleate pycnidiophores or spermatophores.
7. The pycnidiophores 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 aescidium 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.

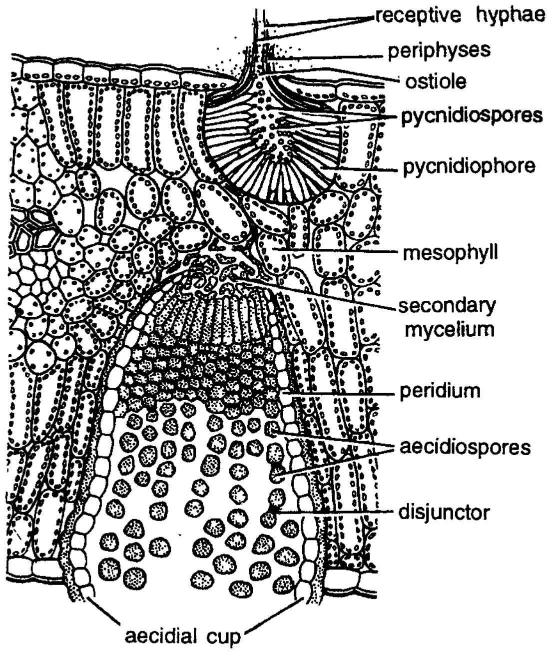


Fig. 10. ***Puccinia.*** T.s. of infected leaf of *Berberis* showing pycnidial and aecidial cups on upper and lower epidermis respectively.

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 disjunctor whereas the latter is the aecidiospore.
7. In younger conditions, aecidiospores are hexagonal and are held in chains by the disjunctor 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 or reinfecting 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	—	***Agaricus***

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.

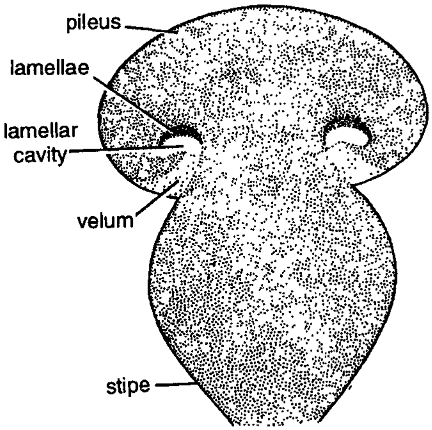


Fig. 1. ***Agaricus.*** L.s. button stage.

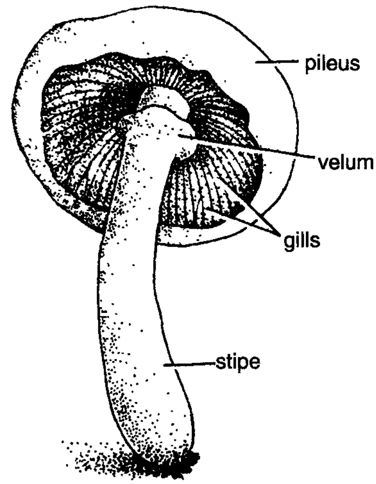


Fig. 2. ***Agaricus.*** A basidiocarp.

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.

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 mangification 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 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.

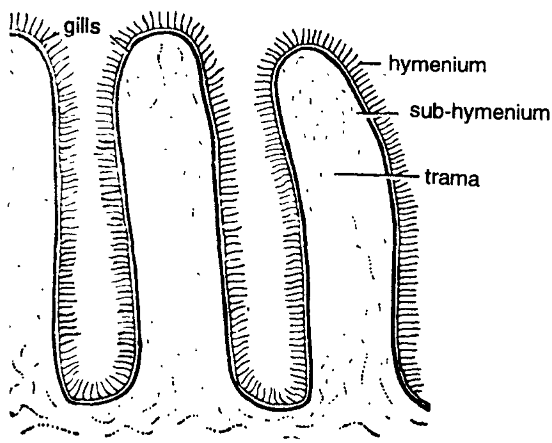


Fig. 3. ***Agaricus.*** Gills as seen in transverse section.

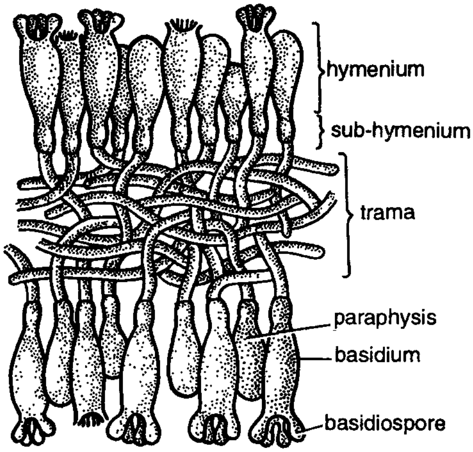


Fig. 4. ***Agaricus.*** Gills as seen in transverse section (magnified).

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.

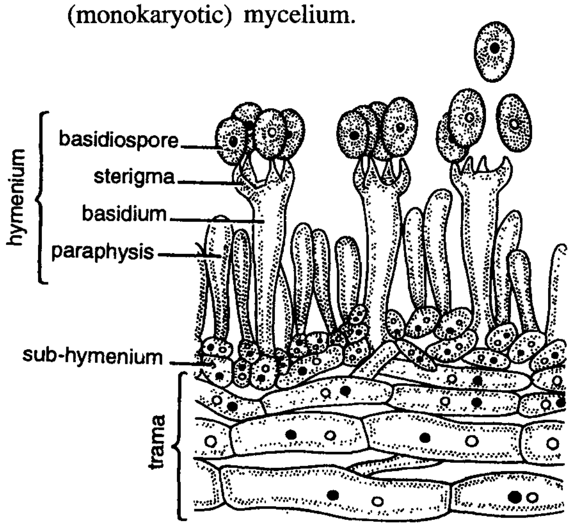


Fig. 5. ***Agaricus.*** A portion of gills in section showing basidia and basidiocarp.

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) Characteistic 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	—	**Myctoa**
Division	—	**Eumycota**
Sub-division	—	**Basidiomycotina**
Class	—	**Hymenomycetes**
Sub-class	—	**Holobasidiomycetidae**
Order	—	**Polyporales**
Family	—	**Polyporaceae**
Genus	—	***Polyporus***

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).

Exercise 2

***Object* : Study of vegetative structure.**

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.

Exercise 3

***Object* : Study of basidiocarp, basidia and basidiospores.**

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.

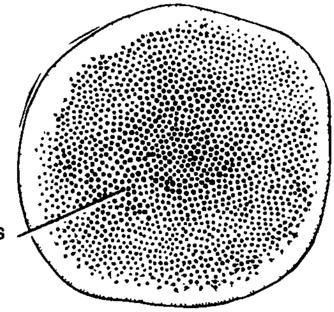


Fig. 2. *Polyporus*. Basidiocarp as seen from lower side.

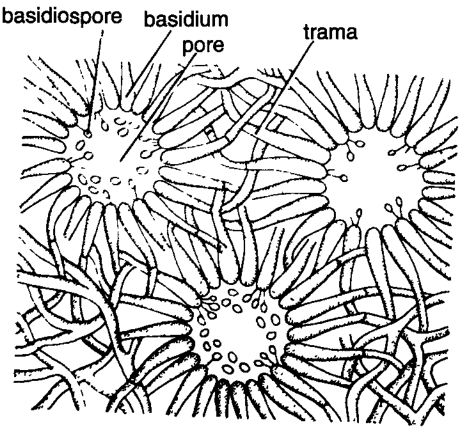


Fig. 3. *Polyporus*. Basidiocarp in section (a part only).

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 steile 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) Characteistic 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—**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 condiospores. The condiiospores 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).

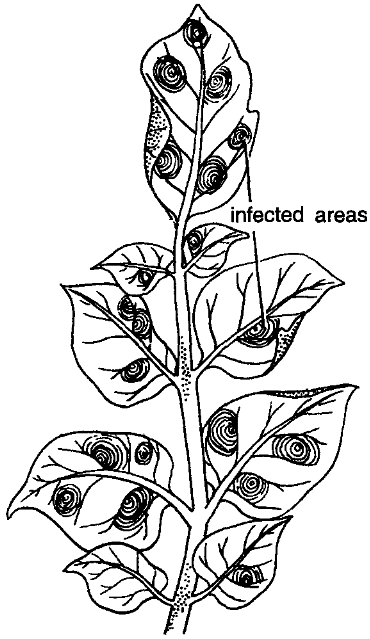


Fig. 1. ***Alternaria.*** Potato twig infected by *A. solani* causing Early Blight of Potato.

3. *A. burnsi* 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 'taget board' effect.
7. Other species which infect various hosts include
   (1) *A. lini*—Alternaria blight of linseed (*Linum usitatissimum*; vern. alsi),
   (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 conidiospores which are

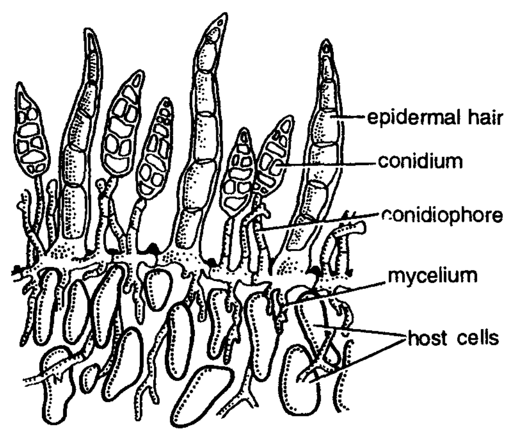


Fig. 2. ***Alternaria.*** T.s of infected host leaf showing conidiophores and conidia.

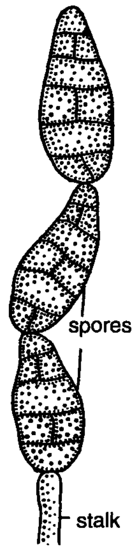


Fig. 3. ***Alternaria.*** A few conidia.

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 condidiospores.

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	—	***Cercospora***

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 parasits 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).

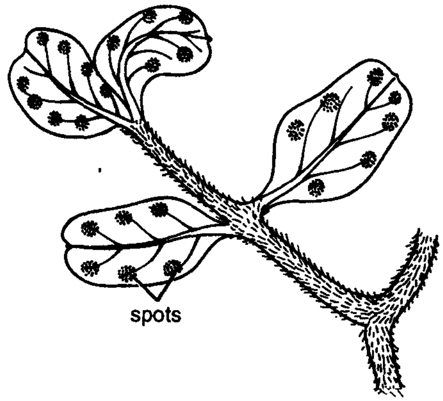


Fig. 1. ***Cercospora.*** Infected plant of *Arachis hypogaea* (Ground nut).

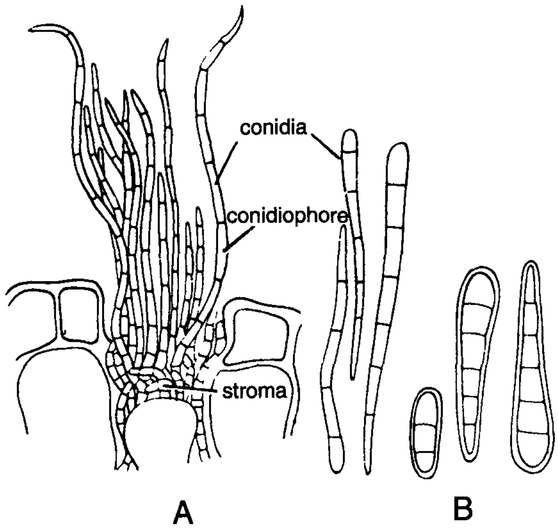


Fig. 2. ***Cercospora.*** **A.** T.s. host leaf, **B.** Conidia.

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 annuum* (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*

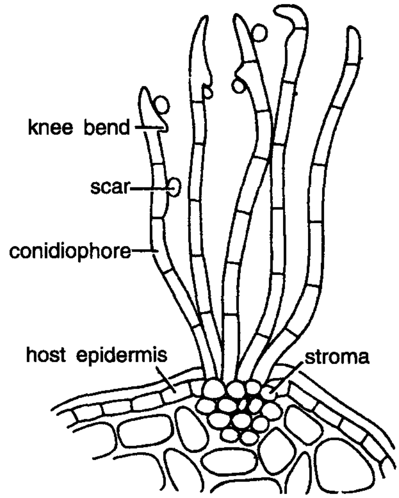


Fig. 3. ***Cercospora.*** Conidiophores with geniculate branching and the scars.

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 hyplhal 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	—	***Colletotrichum***

Exercise 1

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

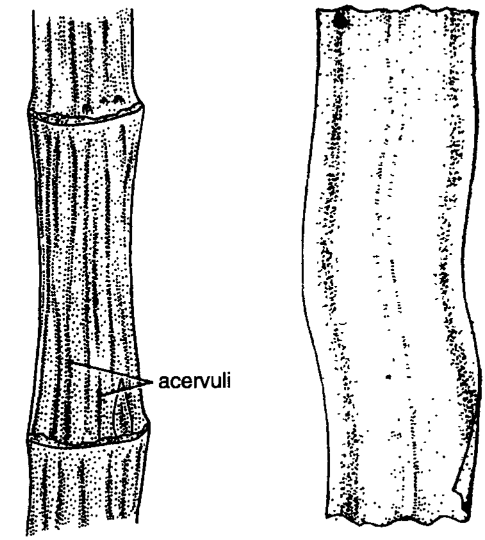
Work procedure

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.



A B

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. 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.

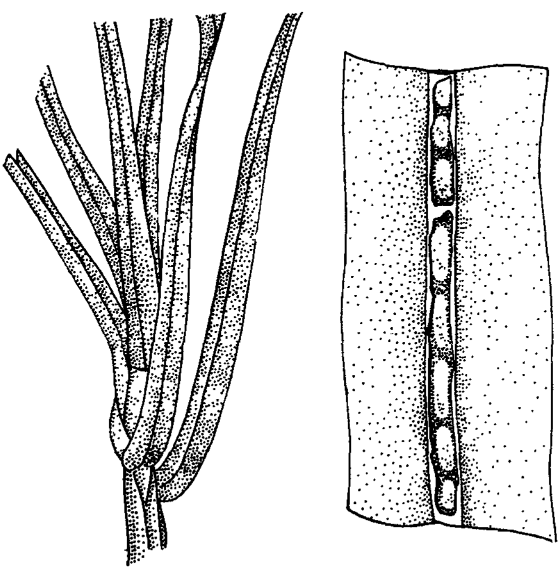


Fig. 2. ***Colletotrichum falcatum*** on sugarcane showing lesions on **A.** Stem and **B.** Leaf

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.

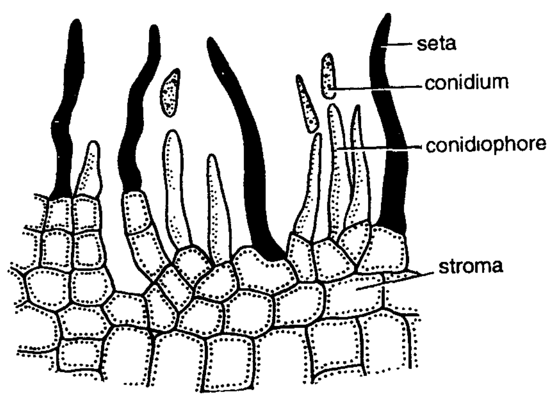


Fig. 3. ***Colletotrichum.*** Acervulus.

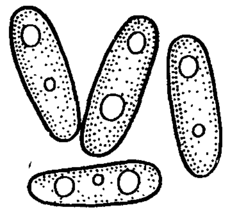


Fig. 4. ***Colletotrichum.*** Conidia.

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.

Lichens

4

Chapter

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 a 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—(i) Crustose or crustaceous, (2) Foliose of 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 a 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 and *Dhup* used in connection with regious 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.*

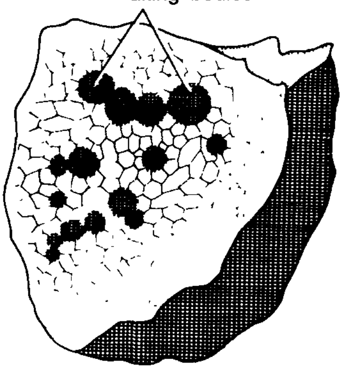


Fig. 1. ***Lecidia platycarpa.*** Crustose ascolichen growing on rocks : External features and fruiting bodies.

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.

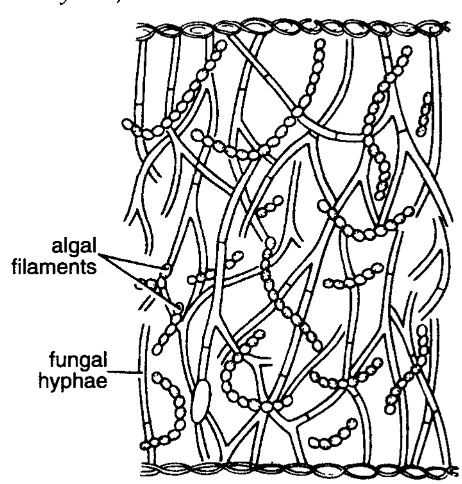


Fig. 3. ***Collema.*** Homoiomerous thallus showing internal structure.

Exercise 2

Object **: Study of internal structure.**

Work procedure

Study the slide showing internal structure.

Comments

1. The thallus is poorly differentiated.
2. The tissues forming the thallus are arranged more of 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 agal 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.

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.

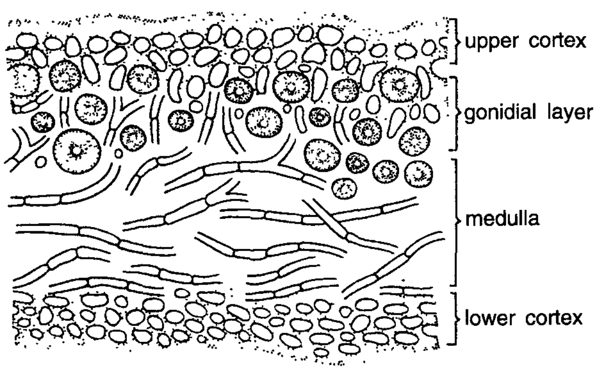


Fig. 4. *Physcia.* V.s. thallus to show heteromerous internal structure.

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 gaseious 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 pycinidial 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.

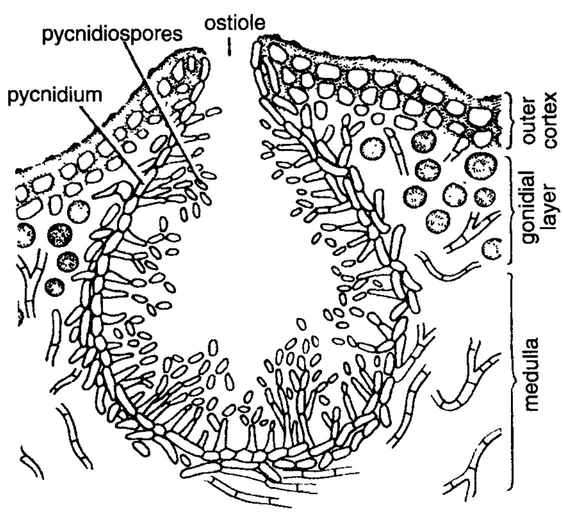


Fig. 5. *Physcia.* V.s. pycnidium to show pycniospores.

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.

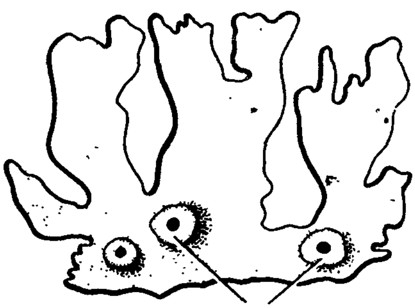


Fig. 6. ***Physcia.*** Thallus showing apothecia.

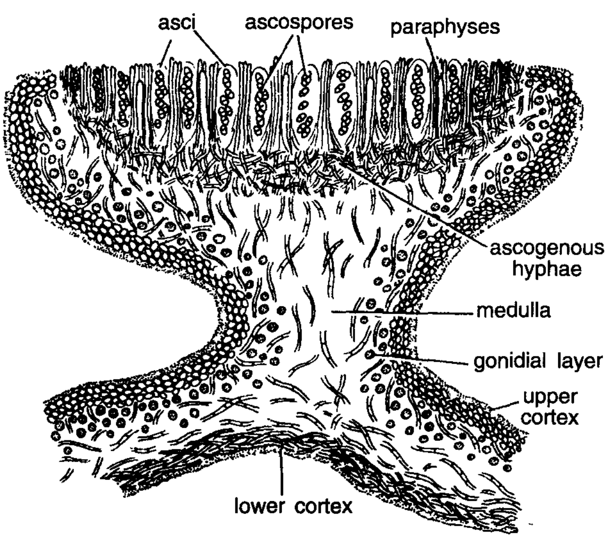


Fig. 7. ***Physcia.*** V.s apothecium for the study of ascospores.

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.

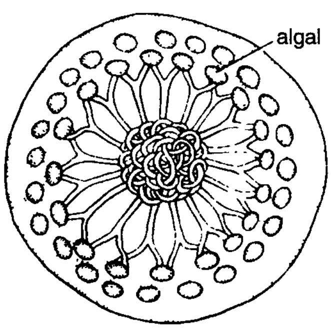


Fig. 10. ***Usnea*** sp. A fruticose thallus. Transverse section to show internal structure.

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.

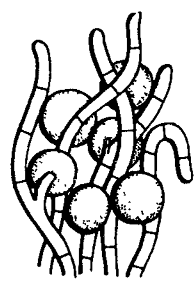


Fig. 11. ***Usnea.*** A soredium

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 superfically with 'bracket fungi'.

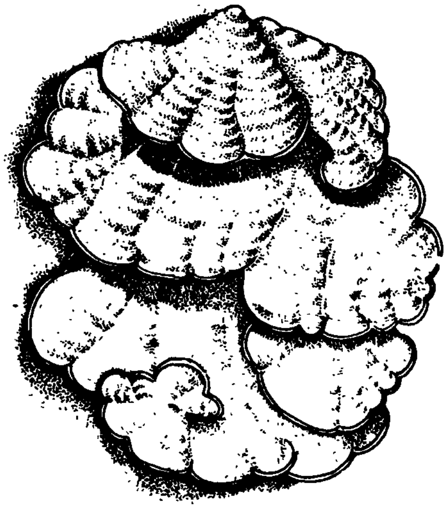


Fig. 12. *Cora pavonia.* External features.

Exercise 2

***Object* : Study of internal structrue 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.

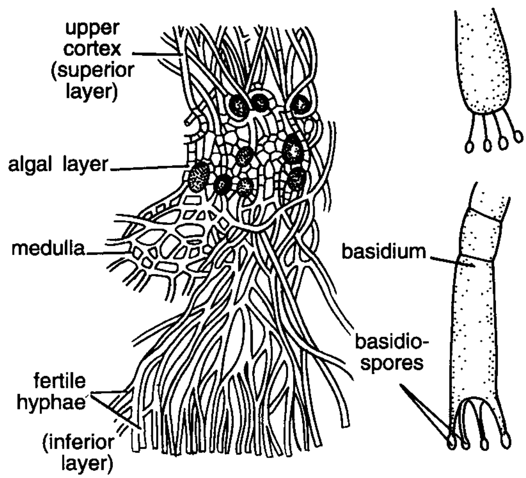


Fig. 13. *Cora pavonia.* V.s. thallus to show internal structure. **A.** Part of V.s. thallus, **B.** Basidia and Basidiospores.

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 instects 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 ($NaNO_3$), potassium hydrogen phosphate (K_2HPO_4), potassium chloride (KCl), hydrated magnesium sulphate ($MgSO_4$, $7H_2O$), ferrous sulphate ($FeSO_4$), sodium chloride (NaCl), sodium hydroxide (NaOH), sodium bicarbonate ($NaHCO_3$), 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 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.

[II] 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 .o 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, salvia, 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^{0}C. The cultures are ready for examination after about 2-3 days.

Exercise 2

***Object* : To isolate micro-organisms from mixed culture and grow a pure culture.**

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 (micro-organisms).**

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.

Stain with crystal violet for about 1 min.
↓
Wash with water
↓
Put a few drops of Gram's iodine on the film and allow it to stand for about 1 minute
↓
Rinse the slide in water
↓
Destain with 95% alcohol for 10-30 secs.
↓
Wash with water
↓
Counterstain with safranin for 30 sec.
↓
Wash with water and dry with blotting paper
↓
Put a drop of glycerine on the film and cover with a cover slip
↓
Now examine the slide under oil immersion objective of the microscope.

Observations

$Gram^+$ bacteria are stained violet or blue and $Gram^-$ 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 10μ (1 mm = 1000 microns or μ).

Ocular micrometer. It is a disc of glass with engraved scale. It is 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.

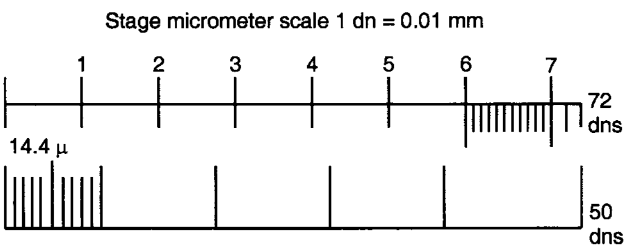


Fig. 1. Micrometer slide and oculometer being coincided.

4. Now coincide the divisions of the stage micrometer slide with those of ocular micrometer.
5. In this case, for example, 72 divisions of stage micrometer slide coincide with 50 divisions of ocular micrometer.

[II] Standardization of ocular micrometer scale

1. The set of lenses used is—objective 45x and eye piece 10x.
2. The value of stage micrometer is—one division of stage micrometer is equal to .01 mm or 10 microns or μ.
3. Therefore, 72 divisions = 0.72 mm or 720 microns or μ.
4. Since 72 divisions of stage micrometer = 50 divisions of ocular micrometer.
5. 0.72 mm or 720 microns or μ = 50 divisions of ocular micrometer.
6. Therefore, 1 division of ocular micrometer = 0.72 mm/50 or 720 μ/50 = 0.144 mm or 14.4 microns or μ.
7. The value of one division of ocular micrometer is, therefore, 0.144 mm or 14.4 microns or 14.4 μ.
8. 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

1. Remove the stage micrometer slide. The ocular micrometer should, however, be allowed to remain in the eye piece.
2. Place the bacteria or any other object under the objective lens. A slide with a cover glass should be used.
3. Move the eye piece or rotate, so that the ocular micrometer scale is adjusted over the object to be measured.
4. If the cell diameter measures 2 divisions of ocular micrometer, then the diameter would be 2 × 14.4 μ = 28.8 μ.
5. 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

1. Place a drop of milk on the slide and prepare a smear.
2. Let the smear dry.
3. Stain the smear with methylene blue.
4. Dry the smear again and place a drop of glycerine.
5. Mount a coverslip and study.

Observations

Numerous bacterial cells are seen scattered. These generally belong to the genera *Lactobacillus* or *Streptococcus*.

[I] *Lactobacillus*

1. These are rod-shaped bacteria. The rods are long and slender.
2. The rods generally occur in chains.
3. The bacterium shows peritrichous flagella but motility is rare.
4. These are Gram$^+$ bacteria but may become Gram$^-$ with increasing age and acidity.
5. Besides dairy products, the bacterium is also found in grains and meat products, water, sewage, beer, wine, fruits and fruit juices.
6. Metabolism is generally fermentative but some are strict anaerobes.

[II] *Streptococcus*

1. Cells spherical to ovoid, less than 2 μm in diameter.
2. The cells occur in pairs or chains.
3. Occasional motile strains are also found.
4. These are gram$^+$, 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
	Fungal Diseases	
1. Black wart disease (Wart disease of potato)	*Synchytrium endobioticum*	*Solanum tuberosum*, Potato, vern. aloo
2. Damping off	*Pythium* sp.	Tomato, tobacco, Ginger, Papaya
3. Late blight of potato	*Phytophthora infestans*	*Solanum tuberosum*, Potato, vern. aloo
4. Green ear disease of bajra	*Sclerospora graminicola*	*Pennisetum typhoideum*, Pearlmillet, vern. bajra
5. Downy mildew of pea	*Peronospora pisi*	*Pisum sativum*, Pea, vern mater
6. White rust of crucifers	*Albugo candida* (=*Cystopus candidus*)	*Raphanus sativus*, Radish, vern. mooli
7. Powdery mildew of barley	*Erysiphe graminis* var. *hordei*	*Hordeum vulgare*, vern. jaii

Contd...

8.	Loose smut of wheat	*Ustilago tritici*	*Triticum aesitivum*, vern. gehoon
9.	Covered smut of barley	*Ustilago hordei*	*Hordeum vulgare*, vern. jaii
10.	Whip smut of sugarcane	*Ustilago scitaminea*	*Saccharum officinarum*, vern. ganna
11.	Black stem rust of wheat	*Puccinia graminis tritici*	*Triticum aestivum*, vern. gehoon
12.	Brown/orange leaf rust of wheat	*Puccinia recondita*	*Triticum aestivum*, vern. gehoon
13.	Rust of linseed	*Melampsora lini*	*Linum usitatissimum*, vern. alsi
14.	Early blight of potato	*Alternaria solani*	*Solanum tuberosum*, vern. aloo
15.	Tikka disease of groundnut	*Cercospora personata, C. arachidicola*	*Arachis hypogaea*, vern. moongphali
16.	Wilt of cotton	*Fusarium oxysporum f.sp. vasinfectum*	*Gossypium* spp., vern. kapas
17.	Red rot of sugarcane	*Colletotrichum falcatum*	*Saccharum officinarum*, vern. ganna
		Bacterial Diseases	
18.	Bacterial blight of rice (paddy)	*Xanthomonas oryzae*	*Oryza sativa*, vern. dhan, chawal
19.	Citrus canker	*Phytomonas citri (=Xanthomonas citri)*	*Citrus aurantifolia*, vern. nimbu
20.	Tundu disease of wheat	*Phytomonas tritici*	*Triticum aestivum*, vern. gehoon
		Viral Diseases	
21.	Leaf curl of potato	Potato virus I or (*Solanum* virus 14)	*Solanum tuberosum*, vern. aloo
22.	Tobacco mosaic	Nicotiana virus 1	*Nicotiana tabacum*, vern. tambaku
23.	Leaf curl of tobacco	Potato virus x	*Nicotiana tabacum*, vern. tambaku
24.	Leaf curl of papaya	Tobacco virus 16, (*Nicotiana* virus 10)	*Carica papaya*, vern. papita
25.	Yellow vein mosaic of bhindi	*Hibiscus* Virus I	*Abelmoschus esculentus*, Lady's finger, okra
		Disease caused by Mycoplasma	
26.	Little leaf of brinjal	Mycoplasm (MLB)	*Solanum melongena*, vern. baingan
		Disease caused by Nematode	
27.	Root knot of vegetables	*Meloidgyne arneria, M. incognita, M. javanica*	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.

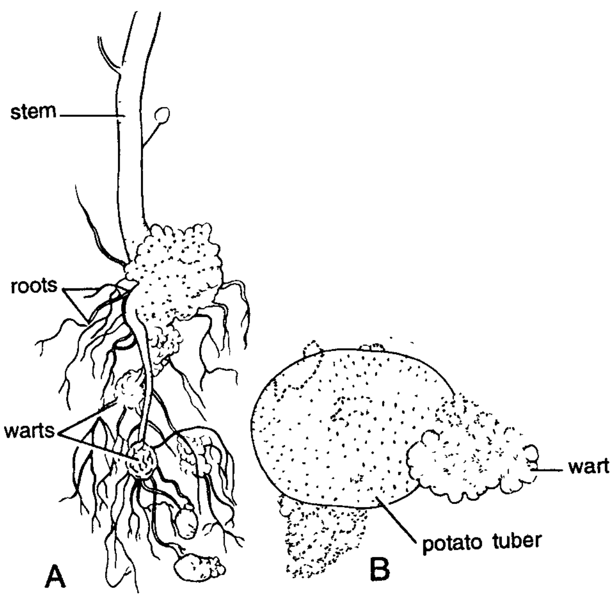


Fig. 1. Black wart disease of potato. A. Heavily warted plant. **B.** A potato tuber showing warts due to infection by *Synchytrium endobioticum.*

3. The diseased potato tubers show dark brown, warty, cauliflower like outgrowths.
4. The warts are produced due to stimulation of cells to divide in the presence of fungus.
5. The disease can be controlled by certain chemicals like $HgCl_2$, ammonium sulphocynate, 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

1. Potato wart disease is caused by a phycomycetous fungus *Synchytrium endobioticum.*
2. It is an obligate parasite.

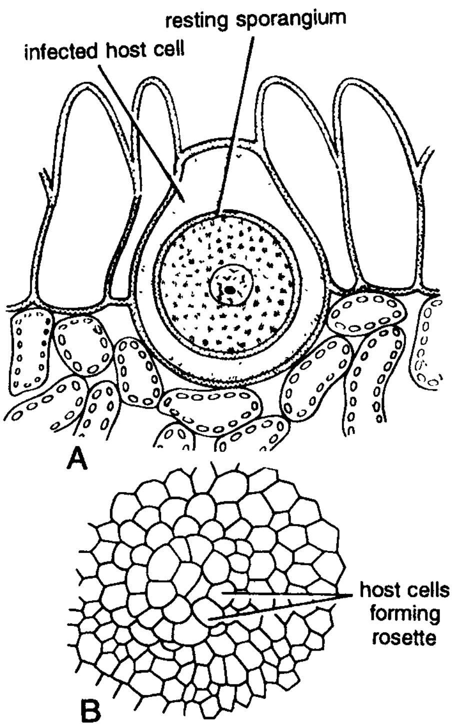


Fig. 2. *Synchytrium*. The causal organism producing black warts on potato. **A.** Summer sporangium in host cell. **B.** Rosette of host cells.

3. The thallus is a naked amoeboid protoplast and endobiotic.
4. Uniflagellate zoospores enter through the host epidermis and cause infection.
5. Thallus secretes golden-brown double layered chitinous wall to form prosorus.
6. 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

1. The disease is caused by a phycomycetous fungus *Pythium.* The common hosts include tomato, mustard, tobacco, ginger, papaya, wheat, etc.
2. The seedlings are affected. The disease also affects seeds, older plants of many vegetables, flowers and also the fruits.
3. Seeds infected in the soil fail to germinate. The seedlings become pale green.
4. A girdle of brown decaying cortex develops at the base of seedling. The seedlings finally collapse.
5. 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.

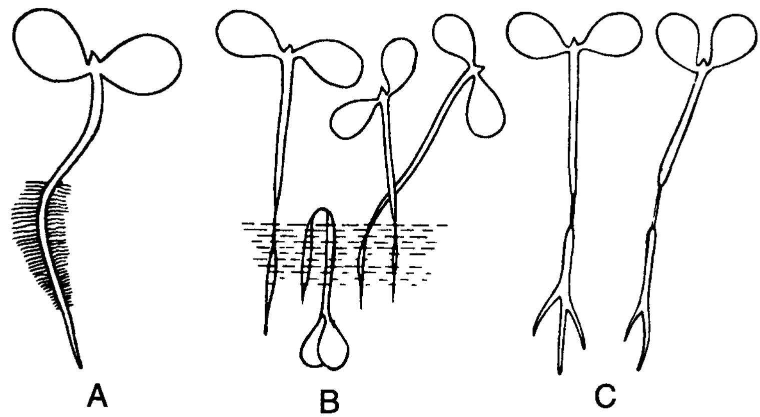


Fig. 1. Damping off. A. Disease free seedling **B.** Diseased seedling, **C.** Diseased seedlings in advanced stage.

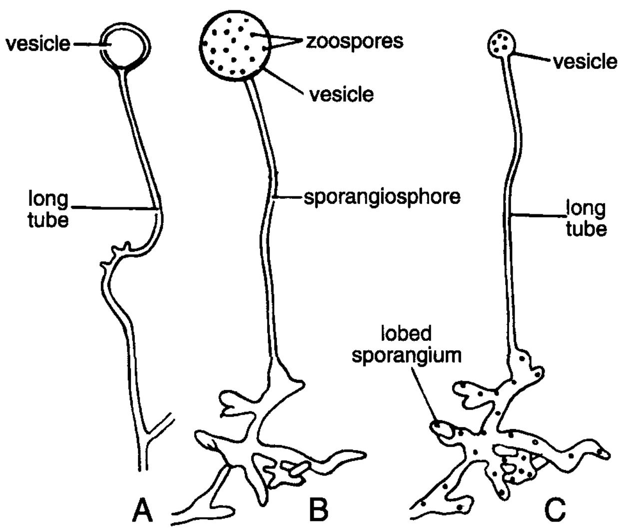


Fig. 2. *Pythium.* Causal organism of damping of **A, B, C.** different stages of sporangium.

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 phycomycetes.
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.

Fig. 1. **Late blight of potato.** Potato twig and tubers infected by *Phytophthora infestans*.

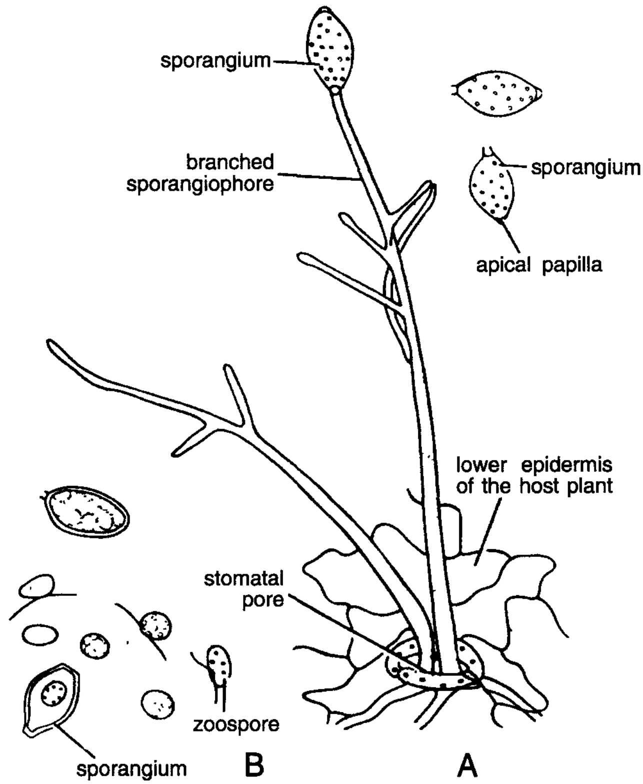


Fig. 2. ***Phytophthora.*** **A.** Sporangiophores coming out of stomata of potato leaf. **B.** Sporangia and the zoospores.

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

Tranverse 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.

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.

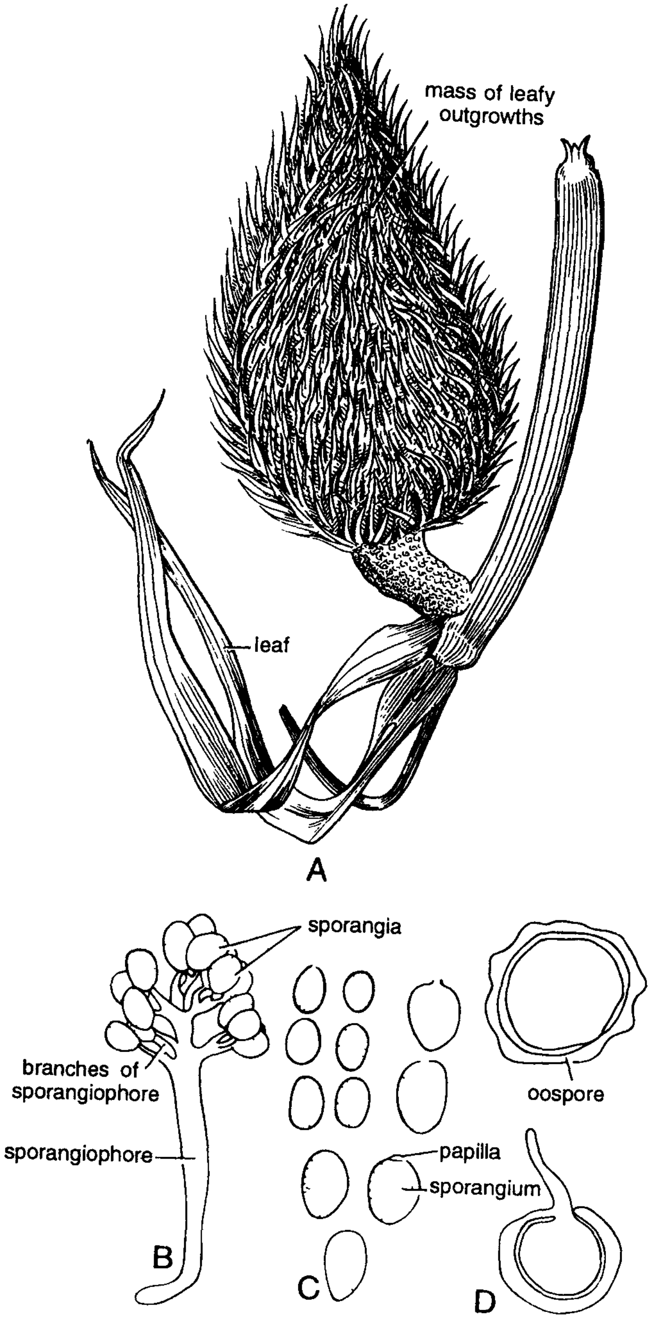


Fig 1. Green ear disease of bajra. A. Infected ear of bajra **B.** Sporangiophore with sporangia. **C.** A few sporangia. **D.** Oospore.

Comments

1. This common disease of bajra (*Pennisetum typhoideum*; Eng. pearlmillet; fam. Poaceae) is caused by a phycomycetous fungus, *Sclerospora graminicola.*
2. The disease is common to all bajra growing regions of the country.
3. Two types of symptoms are produced—downy mildew stage on the leaves and the green ear stage on the ear.
4. In the **downy mildew** stage, the leaves
   (i) become white in the beginning, later becoming brown.
   (ii) become whitish on the lower side due to sporangial growth, and
   (iii) finally become distorted, twisted and crinkled and tend to split.
5. In **green ear disease**
   (i) each flower is replaced by green leafy outgrowths thus turning the solid spicate ear, wholly or partly into a loose head,
   (ii) the bristles of spikelets become hypertrophied and variously contorted. The glumes become enlarged and green,
   (iii) number of florets in a spikelet also increase,
   (iv) 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

1. The causal organism is phycomycetous fungus—*Sclerospora graminicola.*
2. The thallus consists of freely branched coenocytic hyphae that produces haustoria inside the host cell.
3. 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.
4. The tips of the branches of sporangiophores are swollen and bear sporangia.
5. Each sporangium is hyaline, elliptic, smooth and bears apical papilla.

6. Oosporic stage is very common and is found in leafy structures.
7. 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

1. This is a common disease of pea *(Pisum sativum;* vern. matar; fam. Papilionaceae) and is caused by a phycomycetous fungus, *Peronospora pisi.*
2. The symptoms appear at an early stage when the plant is not more than 10-12 cms in height.
3. Yellow to brown spots or lesions appear on the upper surface of leaflets and stipules.
4. At the same time corresponding areas of lower surface of leaves become covered with a whitish downy mildew.
5. The symptoms on the pod appear at flat pod stage. The seeds near the infected tissues get considerably reduced in size and finally abort.
6. 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

1. The causal organism is *Peronospora* of phycomycetes.

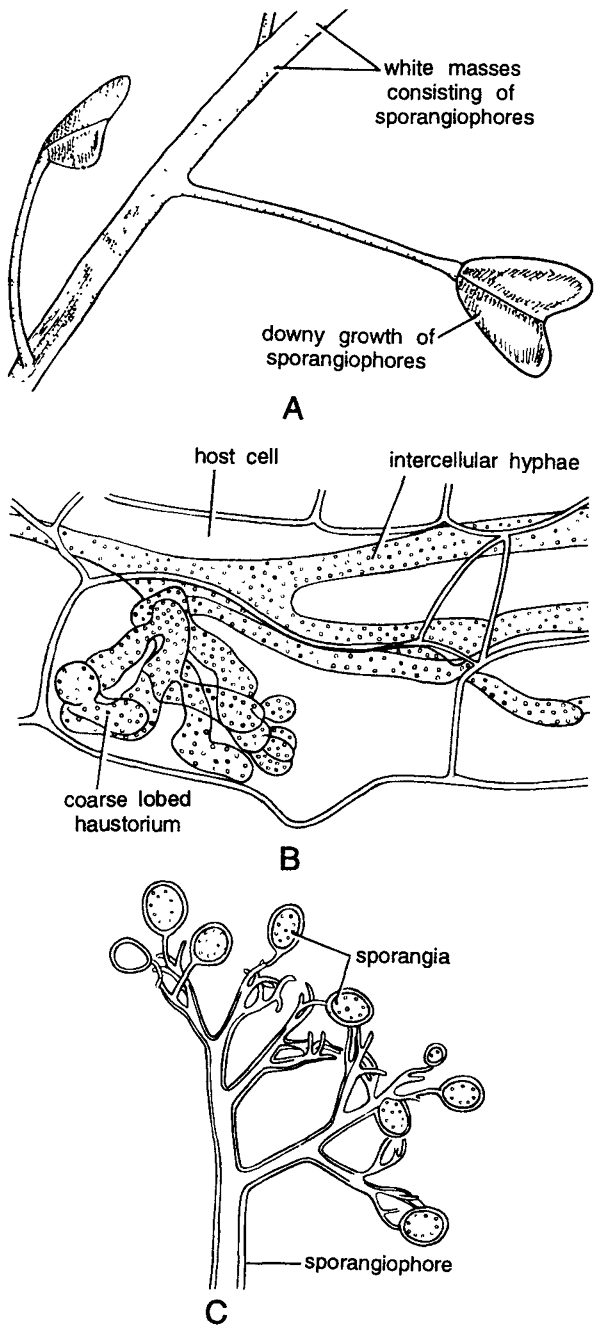


Fig. 1. Downy mildew of pea. A. Part of the plant infected with *Peronospora pisi.* **B.** Host cells with intercellular mycelium and haustoria. **C.** Mycelium with sporangiophore and sporangia.

2. 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. Sporagniophores 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 epispore.

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.

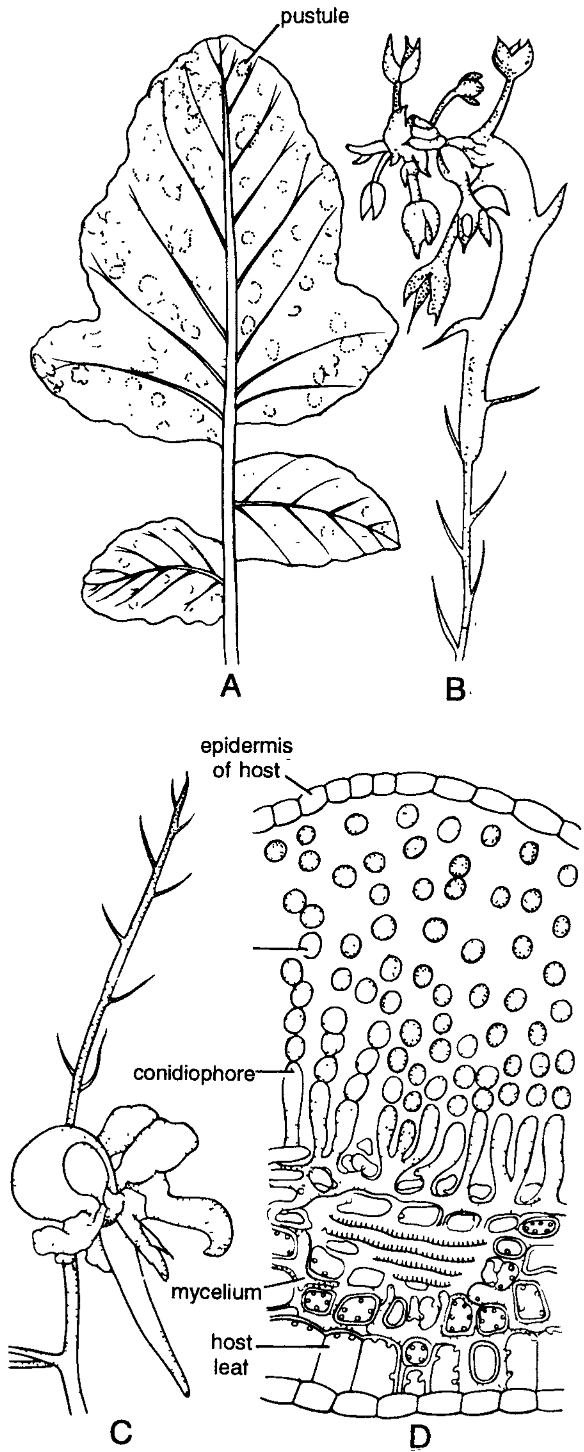


Fig. 1. White rust of crucifers. A. Leaf of *Brassica* (Mustard) infected with *Albugo candida*. **B.** Diseased inflorescence of *Brassica*. **C.** Diseased flower of *Brassica*. **D.** Section through pustules showing conidia.

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 zoopores.
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 epispore 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.

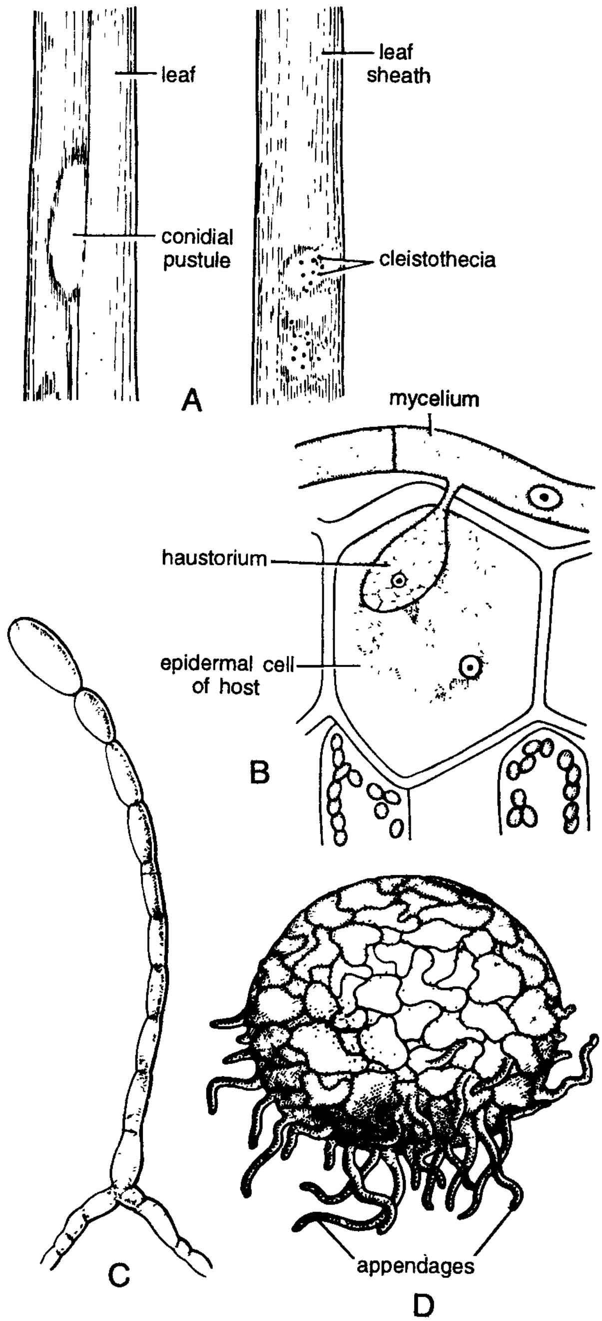


Fig. 1. Powdery mildew of barley. A. Conidial and cleistothecial stage on host leaves. **B.** Epidermal cell of the host showing haustoria. **C.** Conidiophore with a chain of conidia. **D.** Cleistothecium.

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.

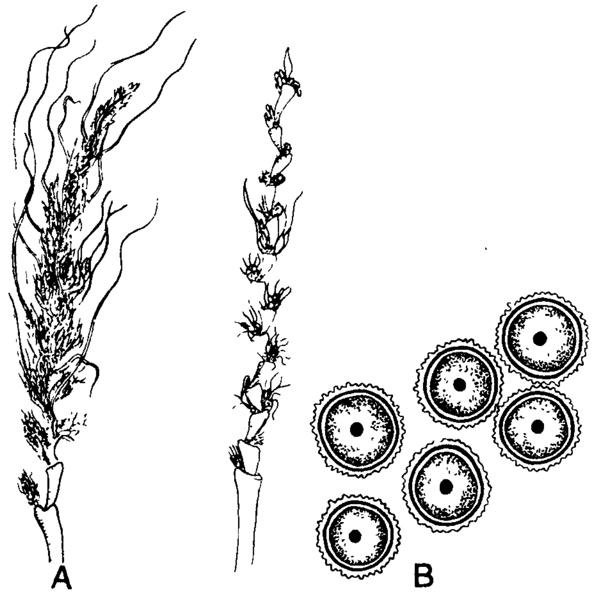


Fig. 1. Loose smut of wheat. A. Wheat ears infected by *Ustilago tritici.* **B.** Chlamydospores.

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.

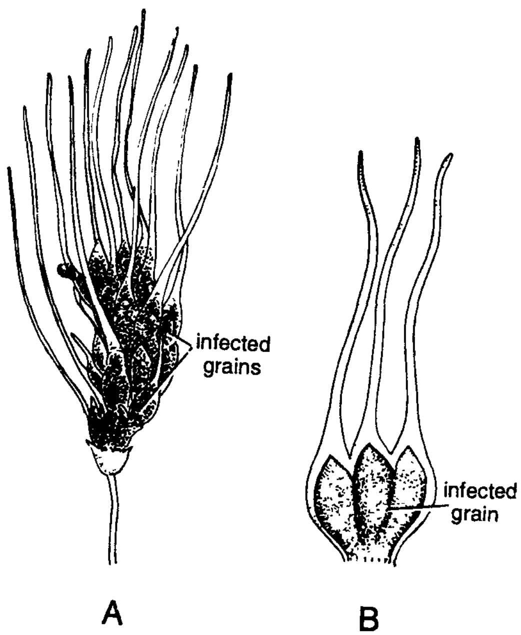


Fig. 1. Covered smut of barley. A. Ears infected by *Ustilago hordei.* **B.** Spikelet magnified.

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.

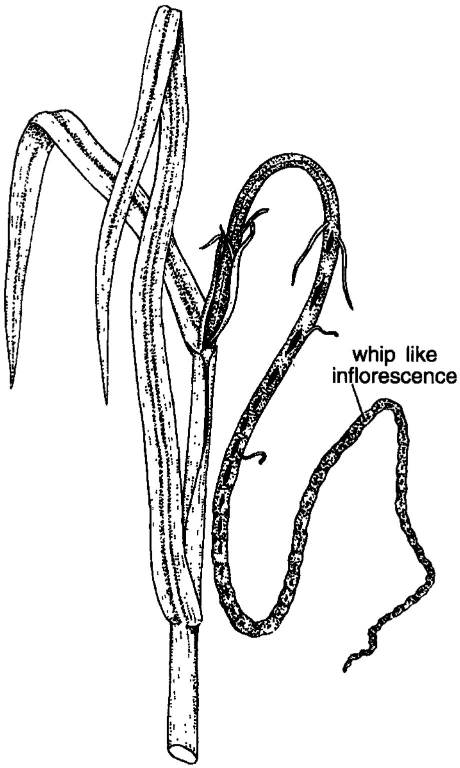


Fig. 1. Whip smut of sugarcane. Inflorescence infected by *Ustilago scitaminea.*

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.

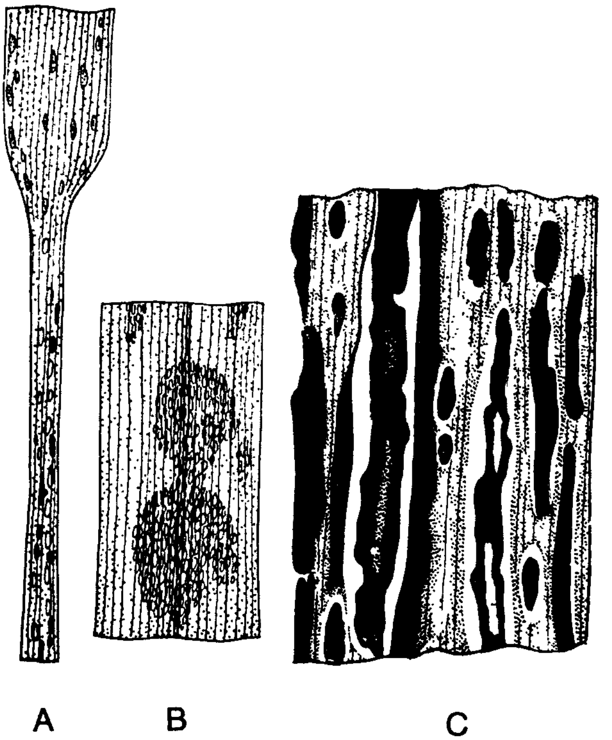


Fig. 1. Black stem rust of wheat. Infection caused by *Puccinia graminis tritici.* **A.** Leaf sheath showing pustules. **B.** A portion of leaf showing uredopustules. **C.** A portion of stem showing teleutopustules.

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

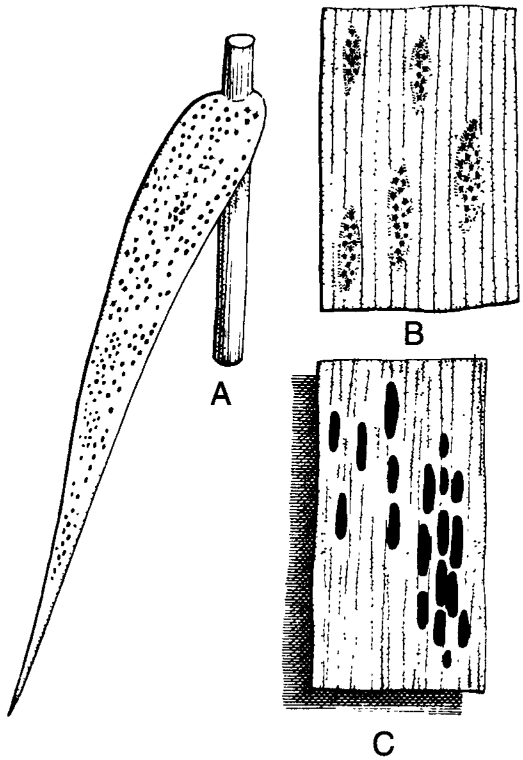


Fig. 1. Brown (orange) leaf rust caused by *Puccinia recondita.* **A.** Leaf showing pustules. **B.** A portion of leaf showing uredopustules. **C.** A portion of leaf showing telutopustules.

basidiomycetous fungus, *Puccinia recondita*, earlier known as *P. triticina*.
2. The rust is more common to northern and eastern parts of India than to Peninsular India.
3. Pycnial and aecial stages are present on *Thalictrum* spp. which is an alternate host for the rust.
4. Uredospores and teleutospores are found on the leaves of wheat plant.
5. Uredosori or uredopustules are very common on the leaves and are rare on leaf sheath and stalks.
6. These appear as bright orange coloured spots when burst open.
7. Uredosori may either be irregularly scattered or present in small clusters.
8. Uredospores are brown, spherical, with finely echinulate epispore and 3-4 germ pores scattered on the surface.
9. Teleutosori are rarely formed. These appear on the lower leaf surface as small, oval or linear, black groups covered by epidermis.
10. Paraphyses are abundantly present which divide the sorus into many chambers.
11. 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 ZnSO4, (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

1. The pathogen that causes Brown or orange leaf rust of wheat is *Puccinia recondita*.
2. Uredospores occur in uredosori or uredopustules. These are brown and spherical, and epispore is finely echinulate with 3-4 germ pores.
3. 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

1. The rust of linseed (*Linum usitatissimum*; vern. alsi; fam. Linaceae) is caused by a basidiomycetous fungus, *Melampsora lini*.
2. The affected plants become bright orange in colour due to the presence of large number of uredosori.
3. Uredosori occur on both the leaf surfaces and other aerial parts of the plant.
4. The leaves die prematurely.

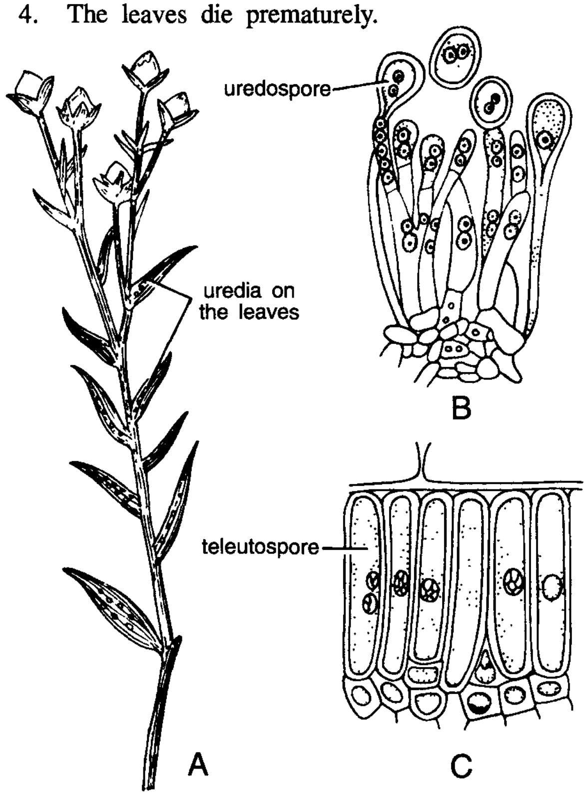


Fig. 1. Rust of linseed caused by *Melampsora lini*. **A.** Diseased linseed plant, **B.** Uredosorus with uredospores, **C.** Teleutosorus with teleutospores.

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,

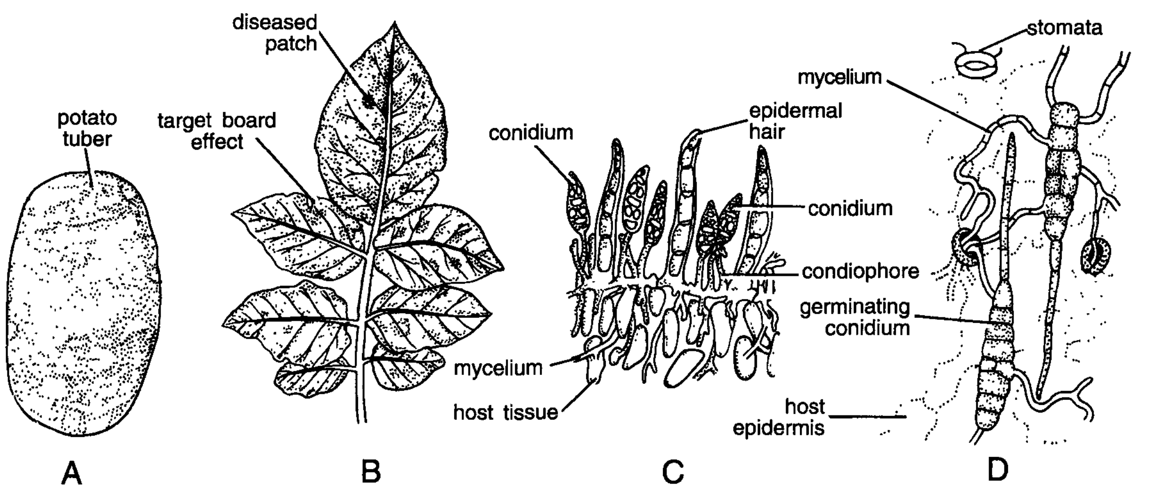


Fig. 1. Early blight of potato caused by *Alternaria solani.* **A.** Diseased potato tuber, **B.** Diseased leaf. **C.** T.s. Infected leaf showing conidiophores and conidia. **D.** Conidia and conidiophores.

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. Condiophores 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 $CuSO_4$, and fungicides like Bordeaux mixture, Dithane M-15, Benlate, etc.

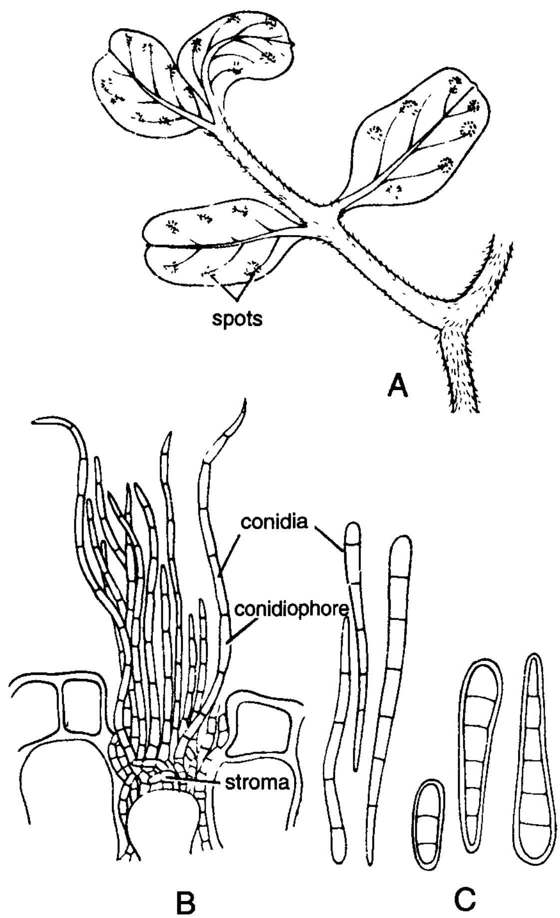


Fig. 1. Tikka disease of ground nut caused by *Cercospora*. **A.** Symptoms of disease on the leaf. **B.** T.s. of host leaf showing conidia and conidiophores. **C.** Conidia.

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. arachidiocola* 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.

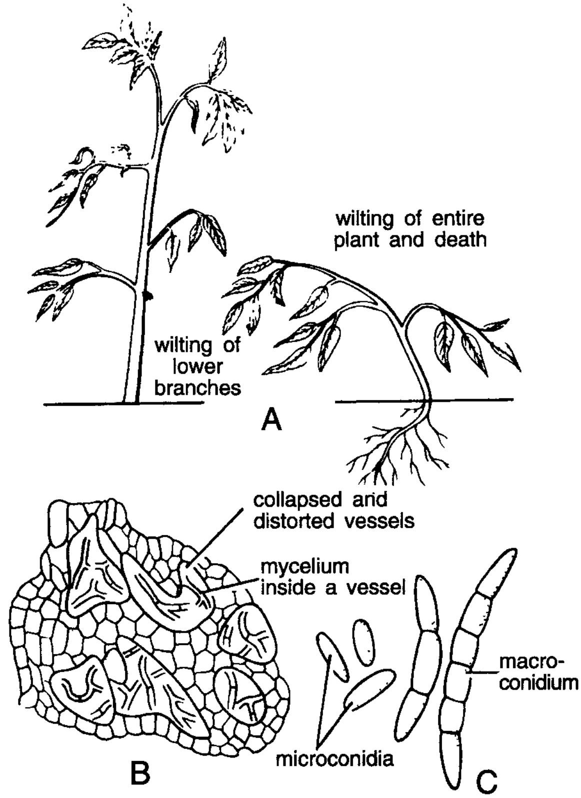


Fig. 1. Wilt of cotton caused by ***Fusarium***. **A.** Symptoms **B.** Mycelium in vessels. **C.** Macroconidia and microconidia.

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. Chlamydospores 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.

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, looses 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 logitudinally 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.

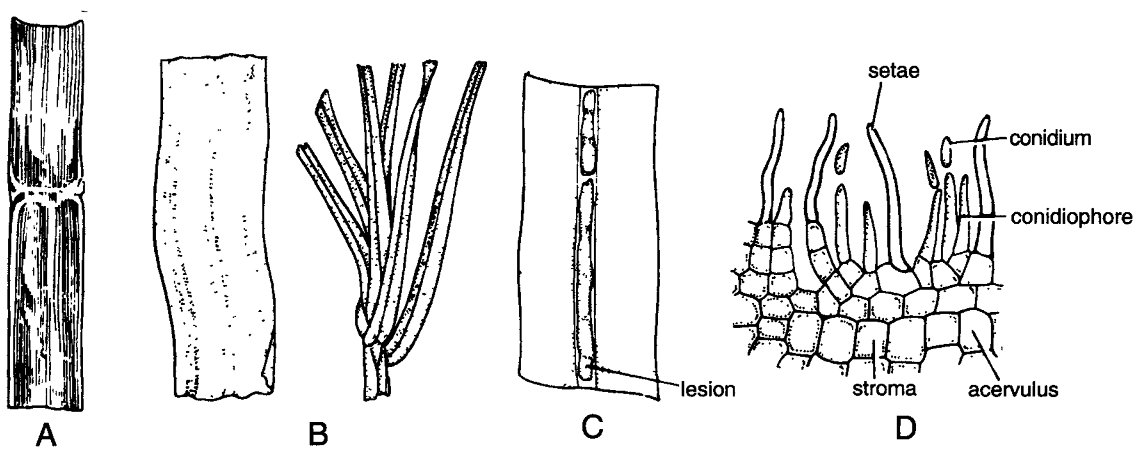


Fig. 1. Red rot of sugarcane caused by *Colletorichum falcatum.* **A.** Symptoms on stem, **B.** Leaves, **C.** Lesions on leaves. **D.** Acervulus with conidia.

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 condiophores 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 interpersed 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.

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 *Phytomonas 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.

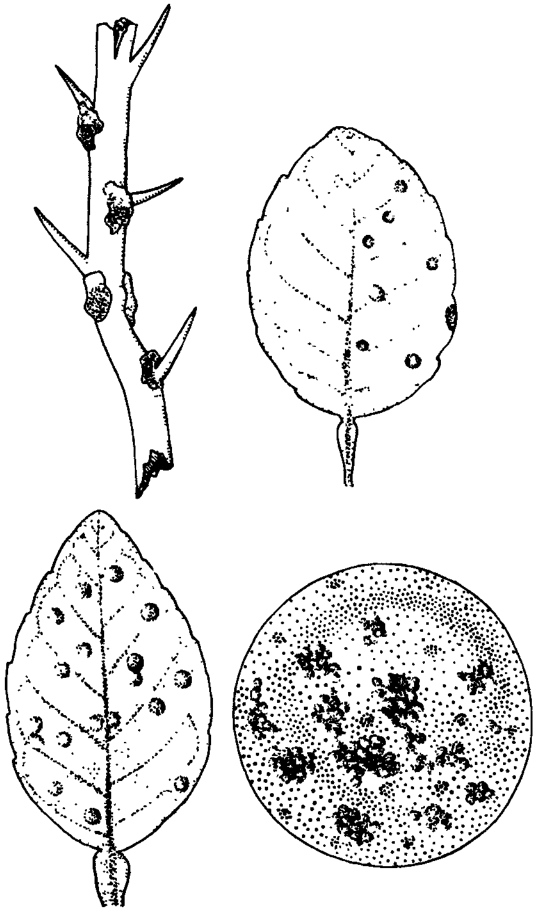


Fig. 1. Citrus canker. Twig, leaves and fruit, showing symptoms.

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.

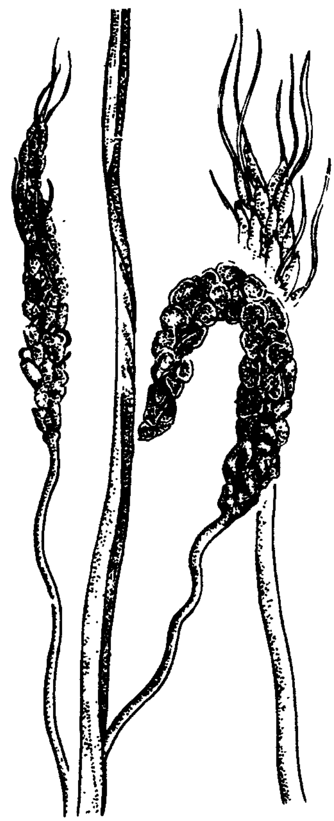


Fig. 1. Tundu disease of wheat. Wheat plant showing symptoms.

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.

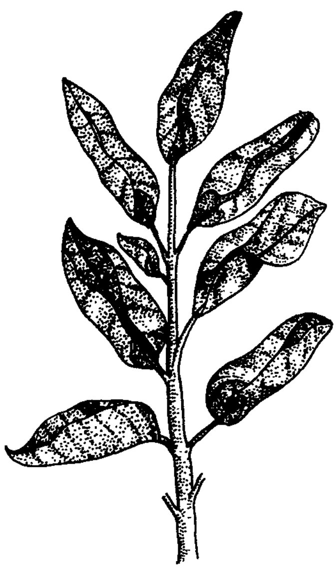


Fig. 1. Leaf curl of potato. Potato leaf showing symptoms.

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.

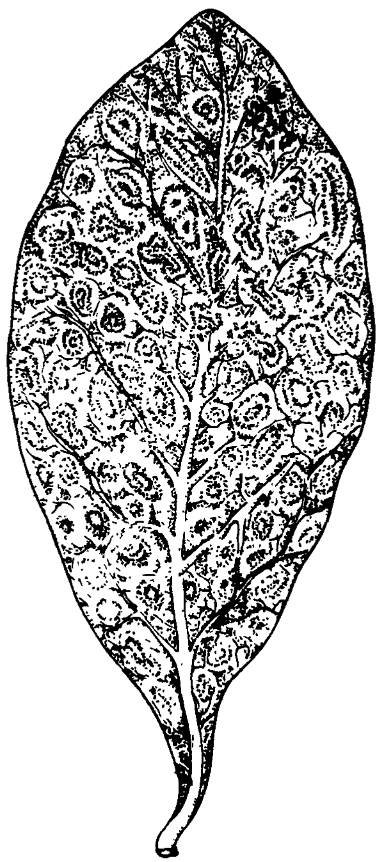


Fig. 1. Tobacco mosaic. Leaf of tobacco showing symptoms.

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.

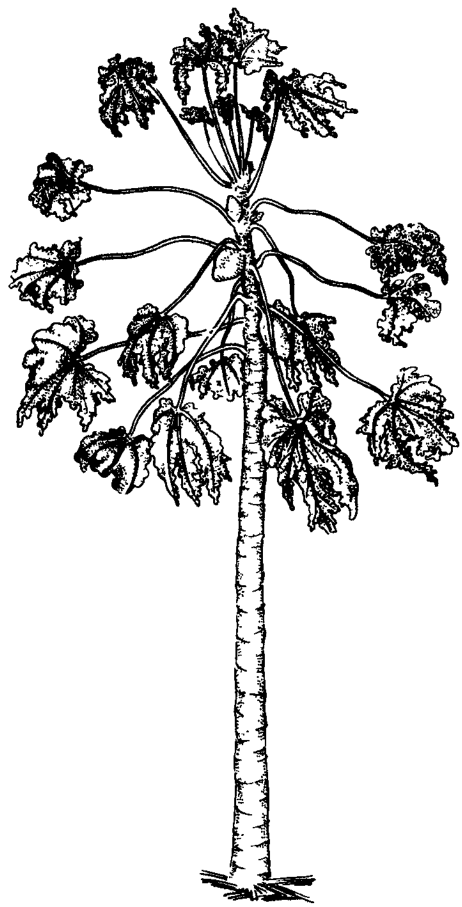


Fig. 1. Leaf curl of papaya. Plant of papaya showing symptoms.

Comments

1. *Carica papaya* acts as a host.
2. Leaves get rolled downward and inward, become deformed and leathery.
3. It is accompanied with vein clearing and reduction in the size of leaves.
4. The petioles are twisted in zigzag manner.
5. During severe infection, growth of the plants is checked, plants remain stunted and fruit bearing is reduced.
6. The causal virus is tobacco virus 16 or *Nicotiana* virus 10.
7. The most important agent of transmission is the white fly *Bemisia tabaci*.

Yellow Vein Mosaic of Bhindi

Exercise 1

Object : **Study the symptoms of yellow vein mosaic of bhindi.**

Work procedure

Study the symptoms shown by the diseased *bhindi* plant.

Comments

1. Yellow vein mosaic of bhindi (*Abelmoschus esculentus*) is a serious problem all over India wherever the crop is grown.

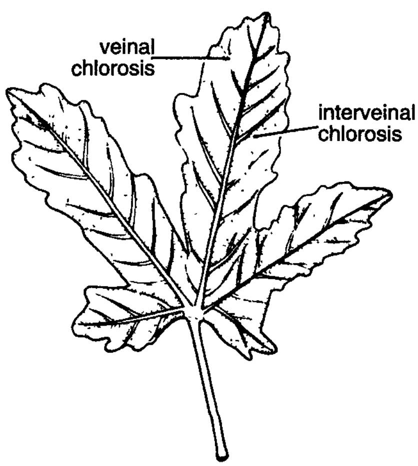


Fig. 1. Yellow vein mosaic of bhindi. A leaf showing symptoms.

2. The major symptoms include vein clearing followed by veinal chlorosis of the leaves.
3. A conspicuous yellow network of veins is formed. The veins and veinlets become thick.
4. In severe cases, chlorosis extends to interveinal areas and results in complete yellowing of leaves.
5. Fruits are dwarfed, malformed and are yellowish green in colour.
6. The causal virus is *Hibiscus* virus I.

Little Leaf of Brinjal

Exercise 1

Object : **Study the symptoms of little leaf of brinjal.**

Work procedure

Study the infected brinjal plant and note the symptoms.

Comments

1. This disease of brinjal (*Solanum melongena*; vern. baingan; fam. Solanaceae) is caused by MLB (mycoplasma-like bodies).
2. The infected plants show extremely reduced leaves and nodes. The plants thus become bushy.
3. The heavily infected plants flowering and fruit setting is negligible. Virescent and phylloid flowers are very common.
4. The disease is transmitted by *Cestius phycitis*.
5. The disease is graft transmissible.
6. Control methods include tetracycline therapy, eradication of weed hosts and use of insecticides for the control of insect vectors.

Root Knot of Vegetables

Exercise 1

Object : **Study the symptoms of root knot of vegetables.**

Work procedure

Study the infected plant parts and note down symptoms.

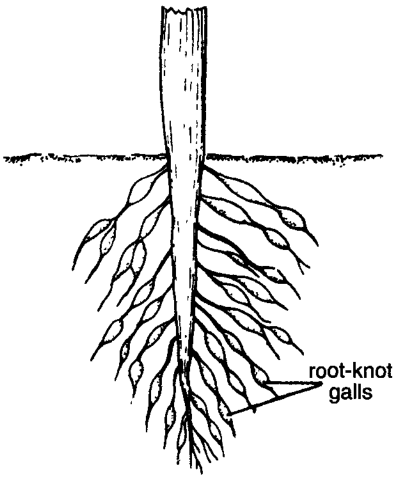


Fig. 1. Root knot of vegetables.

Comments

1. The disease is caused by root-knot nematodes *Meloidogyne*, the commonest species being *M. arneria*, *M. incognita* and *M. javanica*.
2. The common hosts include cucurbits, potato, tomato, brinjal, chillies, lady's finger, groundnut, carrot, radish, Colocasia, etc.
3. Since the symptoms are often not developed on the above ground parts, disease can not be easily recognised.
4. The diseased plants generally remain stunted, sickly and may dry suddenly and prematurely.
5. The roots show gall formation due to stimulation of excessive cell division.
6. Numerous female nematodes are present in the root galls.
7. Disease is controlled by fallowing or simple plowing, early or very late planting, application of DBCP, phorate, carbofuran, etc. and production of resistant varieties.

Exercise 2

Object : **Study of causal organism.**

Work procedure

Since the organism is present in the root galls, these are dissected and nematodes separated. The dissected galls are studied under the microscope.

Comments

1. The disease is caused by a nematode, *Meloidogyne*.
2. The adult males are worm-like while the adult females appear pear-shaped.
3. The female lays eggs in gelatinous substance. Egg develops into larva.
4. II stage larva emerges from the egg in the soil and enters the host root. These enter behind the root tip and become established.
5. It is only at the IV stage larva that male and female can be distinguished.

Important Plants and Their Bacterial Diseases

Host	Diseases	Causal organism	Symptoms
1. Alfala (*Medicago sativa*)	Bacterial disease	*Xanthomonas alfalfae*	Small, water soaked spots, surrounded by straw coloured spots on leaves, later turn brown.
2. Brinjal (*Solanum melongena*)	Bacterial wilt	*Pseudomonas solanacearum*	Lower leaves droop, before wilting.
3. Cabbage (*Brassica oleracea* var. *capitata*)	Bacterial leaf spot	*Xanthomonas campestris* var. *armoriaceae*	Water-soaked leaf spots of minute size, increase. with age, leaves dry up.
4. Chillies (*Capsicum annuum*)	Bacrterial leaf spot	*Xanthomonas vesicatoria*	Small, water soaked spots, surrounded by straw coloured spots on leaves, later turn brown.
5. Cotton (*Gossypium* spp.)	Black arm or angular leaf spot	*Xanthomonas malvacearum*	Angular lesions on leaf and boll, watery and necrotic at maturity.
6. Mango (*Mangifera indica*)	Bacterial leaf spot	*Pseudomanas mangiferae-indicae*	Small water soaked lesions by veins on leaf, lesions turn dark brown.

Contd...

7. Moong, Urd (*Phaseolus* spp.)	Bacterial bean blight	*Xanthomonas phaseoli indicus*	Large, irregular, dry, sunken areas, red or brown in colour.
8. Potato (*Solanum tuberosum*)	Blackleg and soft rot	*Erwinia carotovora*	Affected plants turn pale green or yellow, wilt and die. Affected haulms are jet black at soil level. Diseased tubers rot in storage.
9. Sugarcane (*Saccharum officinarum*)	Red stripe	*Pseudomonas rubrilinaeus*	Long, narrow red streaks on leaves and top shoots affected.
	Gummosis	*Xanthomonas vasculorum*	Pale yellow stripes on leaves, later turning brown, canes become thin, short internodes, vascular bundles red.
10. Tomato (*Lycopersicum esculentum*)	Bacterial wilt	*Pseudomonas solanacearum*	Lower leaves droop before wilting, vascular system becomes brown.

Important Plants and Their Viral Diseases

Host	Diseases	Symptoms
1. Apple (*Pyrus malus*)	Mosaic	Mottling of leaves, white and yellow spots, chlorosis.
2. Banana (*Musa* spp.)	Bunchy top	Dark green streaks on petioles and along veins, shortening of crown and stunted growth.
3. Brinjal (*Solanum melongena*)	Little leaf	Production of tiny yellow or pink discolouration.
4. Cardamom (*Elattaria cardamom*)	Chirke	Light and dark green patches on leaves.
5. Citrus (*Citrus* spp.)	Tristeza	Profuse flowering, heavy fruit setting, yellowing of leaves, defoliation and die back of twigs.
6. Coconut (*Cocos nucifera*)	Wilt	Wilting of leaves, necrosis of leaf tips, yellowing of leaves, crown smaller.
7. Cotton (*Gossypium* spp.)	Stenosis	Leaves crinkled, deformed, yellow or pink discolouration.
8. Cow pea (*Vigna sinensis*)	Mosaic	Yellow and green patches on leaves, mottling and stunting.
9. Ground nut (*Arachis hypogaea*)	Rosette disease	Dwarf shoot forms dense clump, yellow leaves.
10. Potato (*Solanum tuberosum*)	Leaf roll	Upwards rolling of leaves, tissue becomes rigid and leathery, Necrosis of phloem, retarded growth, tubers small.
	Mosaic	Symptoms differ with infection of virus X, virus Y, and virus A.
11. Sugarcance (*Saccharum officinarum*)	Grassy shoot	Profuse tillering, sprouting of lateral buds, poor cane formation.
12. Tobacco (*Nicotiana* spp.)	Mosaic	Mottling of leaves, dark and light green streaks.

Bryophyta

7
Chapter

Preamble

Bryophyta includes mosses, liverworts and hornworts — both living as well as fossils, and numbering about 24,000, distributed in approximately 900 genera. They are fundamentally terrestrial plants, growing in tufts and usually, occupying moist shady places such as grounds, walls, rocks and their crevices. Apart from this terrestrial habit, some plants have secondarily, acquired the aquatic habit (*Riella, Ricciocarpus natans, Riccia fluitans*) whereas the others show epiphytic habit (some mosses and Jungermanniales) and still others a saprophytic habit (as *Buxbaumia*-moss and *Cryptothallus mirabilis*-liverwort).

The, largest Bryophyte is probably an Australian genus *Dawsonia* which attains a length of about 40-70 cms, otherwise as a whole, bryophytes are small and many are indeed microscopic, e.g. *Zoopsis.* This group stands between Algae and Pteridophyta and shows a sharply defined heteromorphic alternation of generations. A noteworthy point about Bryophytes is, that the gametophyte bearing the sex organs is an independent plant and long lived, whereas the sporophyte, bearing the spores, is dependent on the gametophyte and is short lived. The gametophyte, though small, is highly developed and differentiated, and is nutritionally self-sufficient because of the presence of chloroplasts. The gametophyte is either thallose (not differentiated into stem and leaf) or foliose (differentiated into stem and leaf). The true roots are absent and their function has been taken up by rhizoids which are unicellular and unbranched (liverworts) or multicellular and branched (mosses).

Reproduction is oogamous and gametes are produced in multicellular sex organs provided with an outer jacket. The antheridium (male sex organ) consists of stalk and a club-shaped or spherical body, the latter produces the antherozoids. The archegonium (female sex organ) is also stalked and a flask-shaped organ, with a swollen basal venter and slender elongated neck. The former is occupied by a large basal egg cell and a small upper venter canal cell, whereas the latter contains many neck canal cells. Water is essential for fertilization and the embryo is retained within the archegonium whose basal portion enlarges to form calyptra, a protective covering. The simple sporophyte is not differentiated into stem, leaves and roots but usually consists of foot, seta and spore producing terminal capsule. The haploid spores are produced by a reduction division of spore mother cells which represent the last stage of sporophytic generation. The spores germinate to form a filamentous germ tube which later forms the thallus as in liverworts or they germinate into filamentous or thallose protonema from which many erect gametophores arise as in mosses.

Bryophytes have been variously classified many times. No single classification has yet been considered satisfactory, though the classification proposed by Campbell (1936) is generally useful. Classification given below is a modified version of Campbell's scheme. Only those taxa have been mentioned which are later described in the book.

The Distinguishing Characters of Taxa

DIVISION **BRYOPHYTA**
(1) True roots absent
(2) Presence of antheridia and archegonia
(3) True vascular tissues absent

CLASS **1. HEPATICOPSIDA**
(1) Rhizoids without septa
(2) Chloroplasts without pyrenoids
(3) Capsule lacks columella

Order **1. Marchantiales**
(1) Presence of scales
(2) Two types of rhizoids

Family **1. Ricciaceae**
(1) Air pores single
(2) Sex organs in mid-dorsal groove
(3) Sporophyte differentiated into capsule and foot; seta absent

Example. ***Riccia***

Family **2. Marchantiaceae**

(1) Sex organs borne on stalked receptacles
(2) Barrel-shaped air pores
(3) Elaters present in the capsule

Examples. ***Marchantia, Plagiochasma***

Order 2. **Jungermanniales**

(1) Scales absent
(2) Rhizoids smooth walled
(3) Antheridia and archegonia borne at apices
(4) Archegonial neck consists of 5 vertical rows of cells

Sub-order 1. **Metzgerineae**

(Jungermanniales Anacrogynae)

(1) Gametophyte usually thallus, rarely stem with leaves.
(2) Archegonia develop from segments of apical cell; apical cell not completely used in archegonia formation
(3) Capsule wall 2-5 layers thick

Family 1. **Pelliaceae**

(1) Sex organs scattered on the thallus surface
(2) The capsule has a basal elaterophore

Example. ***Pellia***

Sub-order 2. **Jungermannineae**

(Jungermanniales Acrogynae)

(1) Gametophyte with stem and two rows of dorsal and a third, ventral row of leaves
(2) Archegonia in terminal cluster and the last archegonium is formed by the apical cell

Family 1. **Porellaceae**

(1) Rhizoids less in number, form tufts at the bases of amphigastria
(2) Leaves incubously arranged
(3) Lobule (postical lobe) is distinct
(4) Perianth large, inflated with compressed mouth and bilabiate

Example. ***Porella***

Family 2. **Frullaniaceae**

(1) The stems usually pinnately branched
(2) The rhizoids form a tuft at the base or middle of the under leaf
(3) Lateral leaves complicate-bilobed
(4) Perianth compressed and obtusely triangular in cross section

Example. ***Frullania***

CLASS **II. ANTHOCEROTOPSIDA**

(1) Plant body thalloid
(2) Rhizoids without septa
(3) Each cell of the thallus has generally a single, large chloroplast with a pyrenoid

Order 1. **Anthocerotales**

(1) Thallus homogeneous
(2) Only smooth walled rhizoids present
(3) Scales and tuberculate rhizoids absent

Family **Anthocerotaceae**

(1) Sporophyte indeterminate in growth
(2) Presence of meristematic zone
(3) Capsule with central columella

Example. ***Anthoceros, Notothylas***

CLASS **III. BRYOPSIDA**

(1) Gametophores erect and leafy
(2) Rhizoids multicellular with oblique septa

Sub-class 1. **Sphagnidae**

(1) Protonema thallose
(2) Leaves without midrib, composed of two types of cells
(3) Antheridia lateral and archegonia terminal
(4) Archesporium formed from amphithecium
(5) Capsule elevated by elongation of apex of gametophore called pseudopodium
(6) Peristome absent

Order 1. **Sphagnales**

Characters same as sub-class

Family 1. **Sphagnales**

Single family, characters same as sub-class

Example. ***Sphagnum***

Sub-class 2. **Bryidae**

(1) Leaves with distinct midrib
(2) Seta long
(3) Spore sac usually separated from the capsule wall by air space

Order 1. **Funariales**

(1) Leaves ovate or spathulate
(2) Peristome usually double
(3) Calyptra usually distended

Family 1. **Funariaceae**

(1) Calyptra with a long beak
(2) Capsule pyriform and somewhat drooping

Example. ***Funaria***

Order 2. **Polytrichales**

(1) Gametophores tall and perennial
(2) Leaves narrow with lamellae on the upper surface of midrib
(3) Peristome teeth 32 or 64
(4) Calyptra cucullate; either smooth, spinulose or hairy

Family 1. **Polytrichaceae**

Single Family, Characters same as those of order

Examples. ***Polytrichum, Pogonatum***

DIVISION **BRYOPHYTA**

CLASS I **HEPTICOPSIDA** (Hepaticae) Liverworts	CLASS II **ANTHOCEROTOPSIDA** (Anthocerotae) Hornworts	CLASS III **BRYOPSIDA** (Musci) Mossess
Order 1. Marchantiales Family 1. Ricciaceae *Riccia* Family 2. Marchantiaceae *Marchantia* *Plagiochasma* Order 2. **Jungermanniales** Sub-Order 1. Metzgerineae (Jungermanniales Anacrogynae) Family Pelliaceae *Pellia* Sub-order 2. Jungermannineae (Jungermanniales Acrogynae) Family 1. Porellaceae *Porella* Family 2. Frullaniaceae *Frullania*	Order Anthocerotales Family Anthocerotaceae *Anthoceros* *Notothylas*	Sub-class 1. Sphagnidae Order **Sphagnales** Family Sphagnaceae *Sphagnum* Sub-class 2. Bryidae Order 1. Funariales Family Funariaceae *Funaria* Order 2. Polytrichales Family Polytrichaceae *Polytrichum* *Pogonatum*

Riccia

Classification

Division	—	**Bryophyta**
Class	—	**Hepaticopsida**
Order	—	**Marchantiales**
Family	—	**Ricciaceae**
Genus	—	***Riccia***

Exercise 1

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

Work procedure

Study the external features of the gametophyte, both from dorsal and ventral surfaces. Observe the two types of rhizoids and violet coloured scales.

Comments

1. The plant body is thalloid, dorsiventral, prostrate and ribbon-like.
2. A rosette is formed due to repeated dichotomies of the thalli.
3. The thallus is linear to wedge shaped with an apical notch at the apex and thickened midrib in the sagittal axis. On the dorsal side, the midrib is traversed by a mid-dorsal groove.
4. On the ventral side, scales and rhizoids are present. The scales are present at the margins. The rhizoids arise from the midrib region.
5. Each scale is violet coloured, multicellular and one celled thick.
6. Rhizoids are of two types—(i) smooth walled and (ii) tuberculate. The smooth walled rhizoids have inner smooth walls whereas tuberculate rhizoids produce tuber-like or peg-like ingrowths of their inner wall which project into the lumen of the rhizoids.
7. Sex organs are present in the mid-dorsal groove and are embedded in the thallus. The sporophytes, however, may be seen as black dots, when mature, under the dissecting microscope.

Exercise 2

***Object* : Study of anatomy of thallus.**

Work procedure

Place the thallus in pith. Cut T.s. and stain either in safranin or fast green. Mount in glycerine and study.

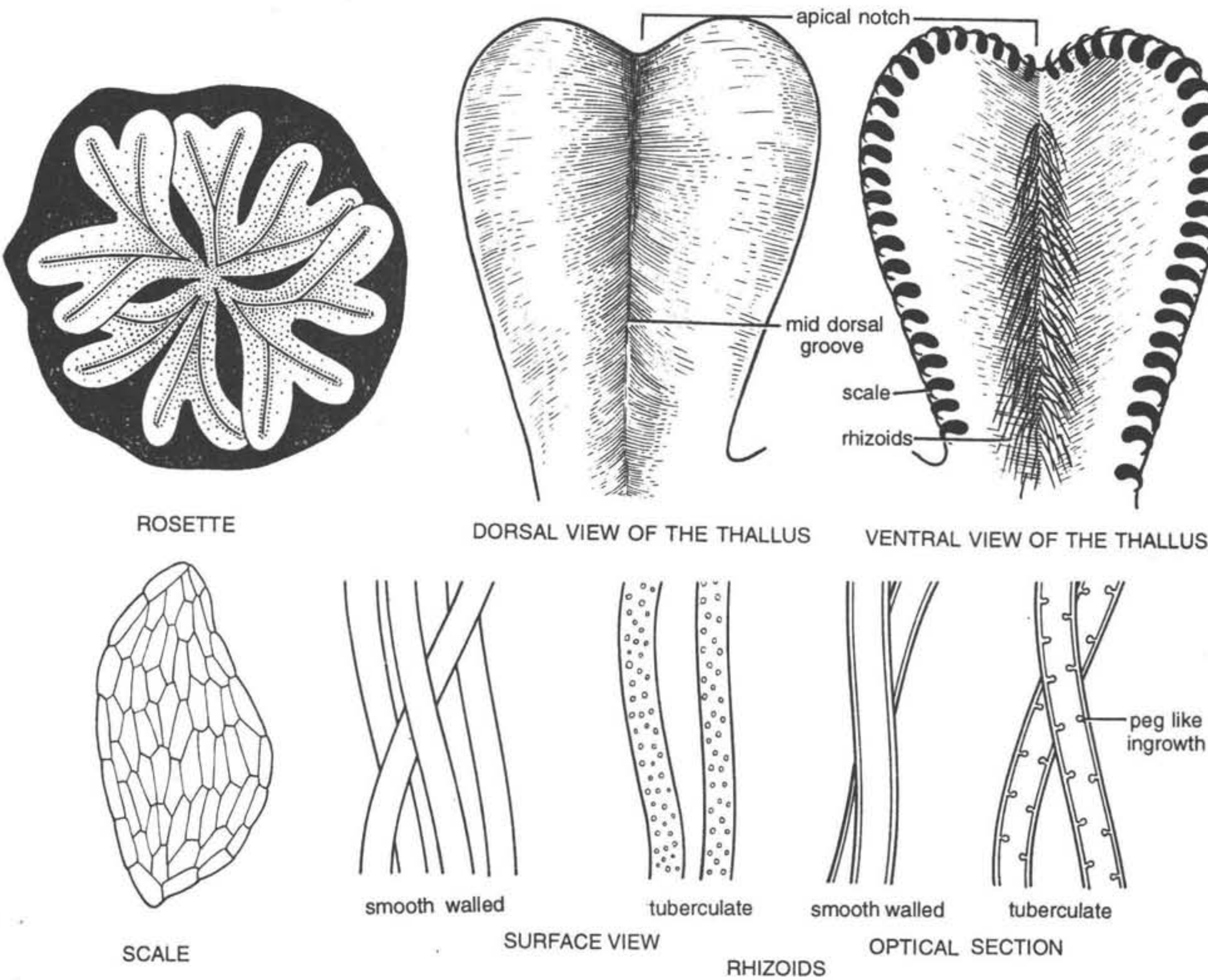


Fig. 1. ***Riccia.*** External features, scales and two types of rhizoids.

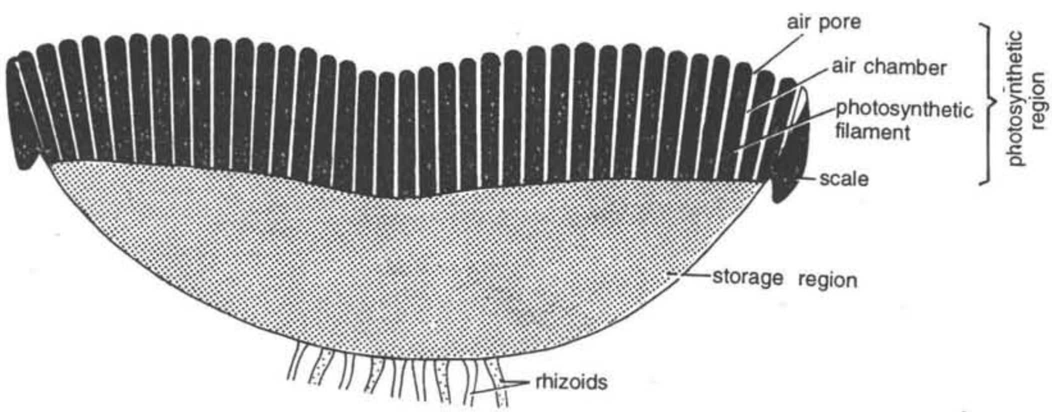


Fig. 2. ***Riccia.*** V.t.s. thallus (diagrammatic).

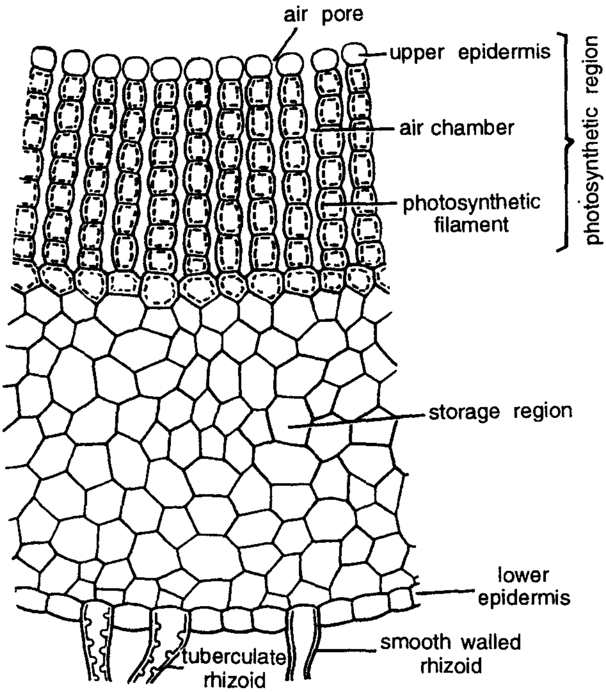


Fig. 3. ***Riccia.*** V.t.s. thallus (a part cellular).

Comments

1. The thallus is boat-shaped in a vertical transverse section.
2. It is thick in the midrib region and gradually thins out towards the margins.
3. The thallus is dorsiventrally differentiated into an upper green photosynthetic region and a lower colourless storage region.
4. The lower epidermis bounds the storage region on the lower side and bears the usual two types of rhizoids (smooth walled and tuberculate) in the centre.
5. The storage region consists of compactly arranged parenchyma. These cells contain starch.
6. The photosynthetic region consists of vertical rows of unbranched assimilatory filaments, separated by narrow air chambers. The cells of the filaments are barrel-shaped and each possesses numerous chloroplasts.
7. The air chambers open to the outside through simple air pores which are the intercellular spaces between the upper epidermal cells.
8. The uppermost cells of the assimilatory filaments are somewhat large. They lack chloroplasts and are thus colourless. These form an ill-defined upper epidermis.
9. On the two margins of the boat shaped section, violet coloured scales are present.

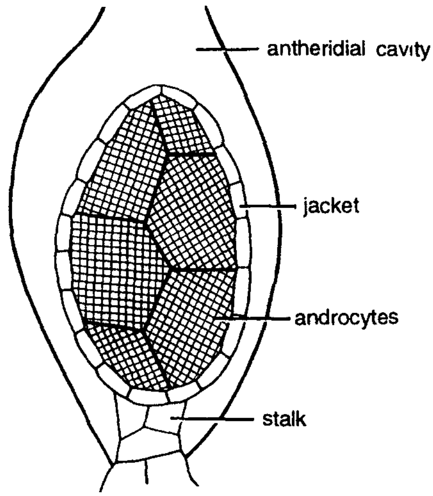


Fig. 4. ***Riccia.*** An antheridium.

Exercise 3

Object **: Study of antheridium.**

Work procedure

Cut L.s. of thallus through mid-dorsal groove. Stain in fast green, mount in glycerine and study the antheridia.

Comments

1. The thallus is monoecious, both the sex organs being situated in the mid-dorsal groove. (*R. bischoffi* and *R. curtisii* are dioecious).
2. The antheridium is present inside a cavity called antheridial chamber which opens outside by antheridial pore.
3. The antheridial chamber with antheridium, lies embedded partly in the tissue of the photosynthetic region and partly in the tissue of the storage region.
4. A mature antheridium consists of a small stalk and a globular or club-shaped body.
5. The stalk is short and few celled. The body is composed of a central mass of either androcytes or antherozoids, surrounded by a single layer

of sterile jacket. The cells of the jacket are tangentially elongated.

Exercise 4

***Object* : Study of archegonium.**

Work procedure

Cut L.s. of thallus through mid-dorsal groove, stain the section in fast green, mount in glycerine and study the archegonium.

Comments

1. The thallus is monoecious and both the sex organs are situated in the mid-dorsal grrove.
2. A nearly mature archegonium is flask-shaped.
3. Archegonium is shortly stalked and consists of a broad venter and a long neck.
4. Wall of the venter is one celled. The venter has one venter canal cell and an egg cell.
5. The neck consists of 6 vertical rows of cells and is 6-9 cells in height. It possesses 4 neck canal cells.
6. The neck is surmounted by four cover cells.
7. Before fertilization, all the axial cells except the egg cell degenerate and the cover cells spread open to facilitate the entry of antherozoids.

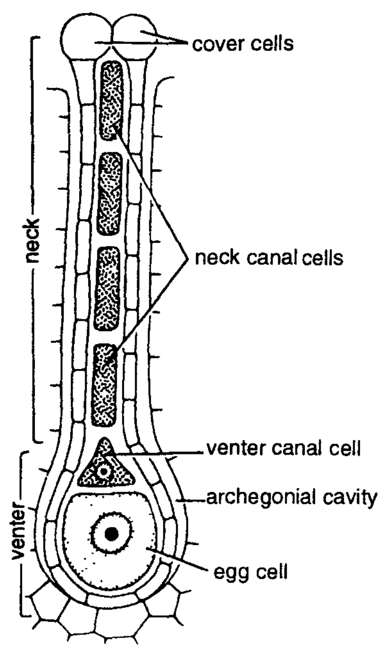


Fig. 5. ***Riccia.*** An archegonium.

Exercise 5

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

Work procedure

Cut L.s. of the thallus through mid-dorsal groove, stain in safranin or fast green, mount in glycerine and study the sporophyte.

Comments

1. The sporophyte is embedded in the tissue of the thallus. It is present in the venter of fertilized archegonium.
2. Sporophyte is represented only by the capsule, foot and seta being absent.
3. The young capsule has a jacket layer and a 2-layered calyptra, derived from venter.

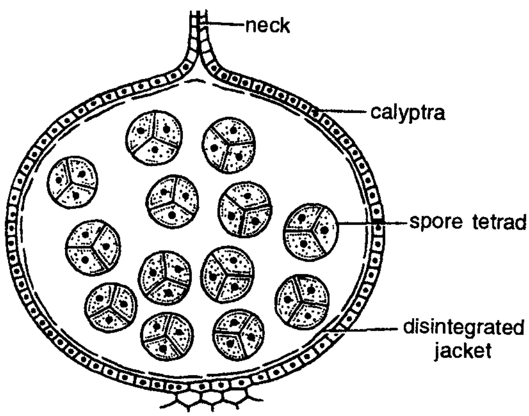


Fig. 6. ***Riccia.*** V.t.s. sporophyte.

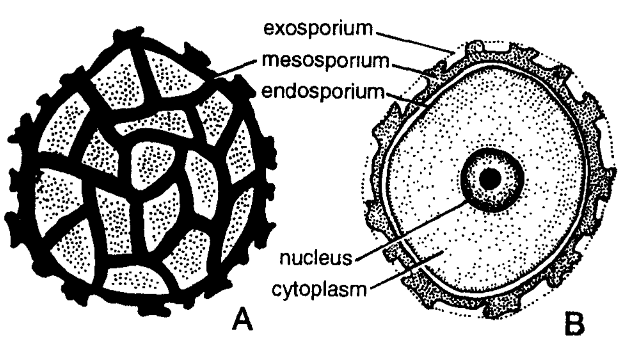


Fig. 7. ***Riccia.*** Spores **A.** Surface view. **B.** Optical view.

4. The mature sporophyte has spore tetrads arranged tetrahedrally (except *R. pearsonii*) or spores. These remain surrounded only by outer layer of calyptra, the inner layer of calyptra and the jacket disintegrates.
5. The spores are discharged only after the disintegration of the thallus.
6. Each spore ranges from 0.05 to 0.012 mm in diameter and consists of spore wall, enclosing within a rich cytoplasm and a nucleus.
7. The spore wall is three layered. The outermost layer is the exosporium which is thin and cutinized. The middle mesosporium is thick and the innermost endosporium is thin and homogenous. The entire spore wall is irregularly thickened and folded.

Identification

Division—**Bryophyta**. (1) True roots absent, (2) True vascular strands absent.

Class—**Hepaticopsida**. (1) Mostly thalloid, (2) Rhizoids without septa, (3) Chloroplasts without pyrenoids, (4) No columella in capsule.

Order—**Marchantiales**. (1) Scales present, (2) Two types of rhizoids present, (3) Air chambers and air pores present.

Family—**Ricciaceae**. (1) Air pores are simple, (2) Sex organs in the mid-dorsal groove, (3) Sporophyte composed only of capsule, foot and seta being absent.

Genus—***Riccia***. (1) Scales on the margins, (2) Assimilatory filaments are unbranched and vertical.

Hints for Collection

Riccia is very common in both hills as well as in plains. All the species grow on damp soil and rock. In plains *Riccia* can be seen growing amongst brick work or unused soil just after a few heavy showers.

Riccia fluitans

Comments

1. It is the only aquatic and free floating species of *Riccia*.
2. It is much dichotomously branched and thalloid.
3. The thallus is linear, elongated, ribbon-like, thin and lacks rhizoids and scales.
4. Vegetative reproduction takes place by adventitious branches.
5. It is sterile when free floating, but if water recedes, sex organs are formed. Therefore, fertile forms are terrestrial.

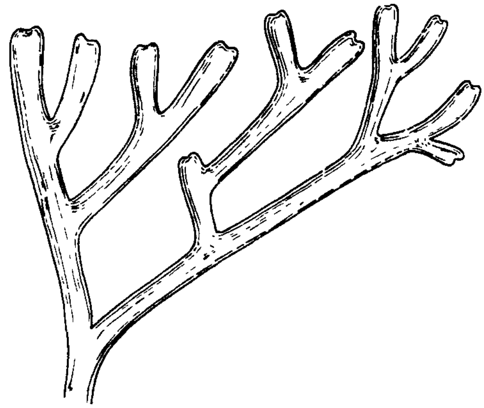


Fig. 1. ***Riccia fluitans***. External features.

Marchantia

Classification

Division	—	**Bryophyta**
Class	—	**Hepaticopsida**
Order	—	**Marchantiales**
Family	—	**Marchantiaceae**
Genus	—	***Marchantia***

Exercise 1

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

Work procedure

Study the external features of gametophyte. Observe the thallus from dorsal and ventral surfaces. Remove the rhizoids, stain in safranin and study. Also remove the scales from ventral side from the mid ventral region and margins. Mount in glycerine and study.

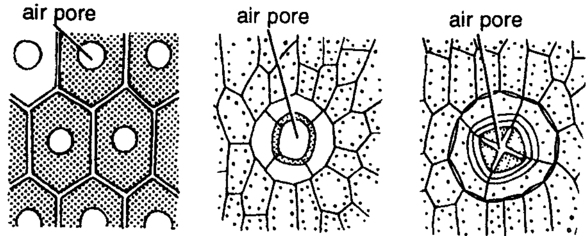


Fig. 2. ***Marchantia*****. A.** Polygonal area, **B.** and **C.** Air pores in surface view.

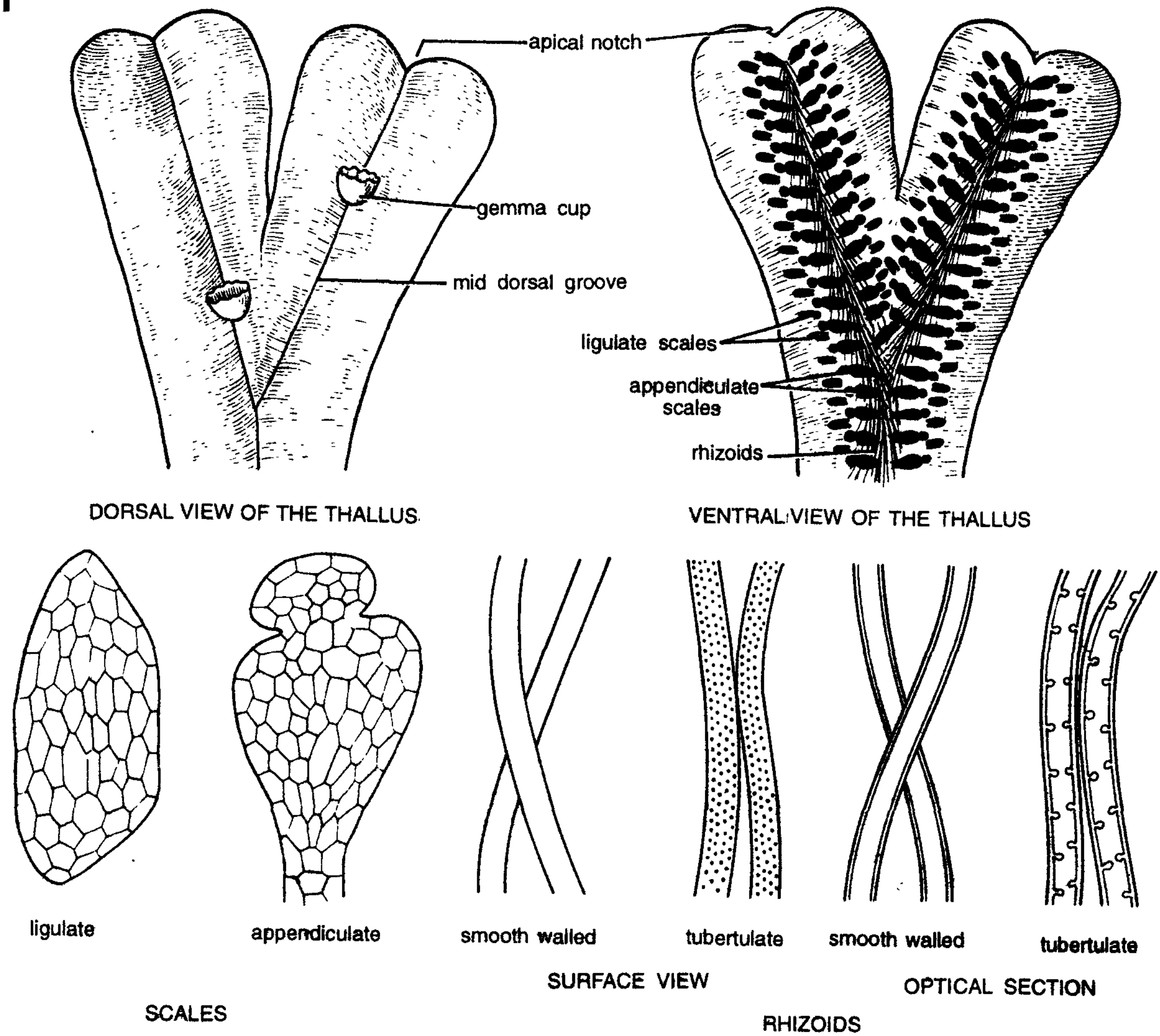


Fig. 1. ***Marchanita.*** External features; dorsal and ventral views, scales and rhizoids.

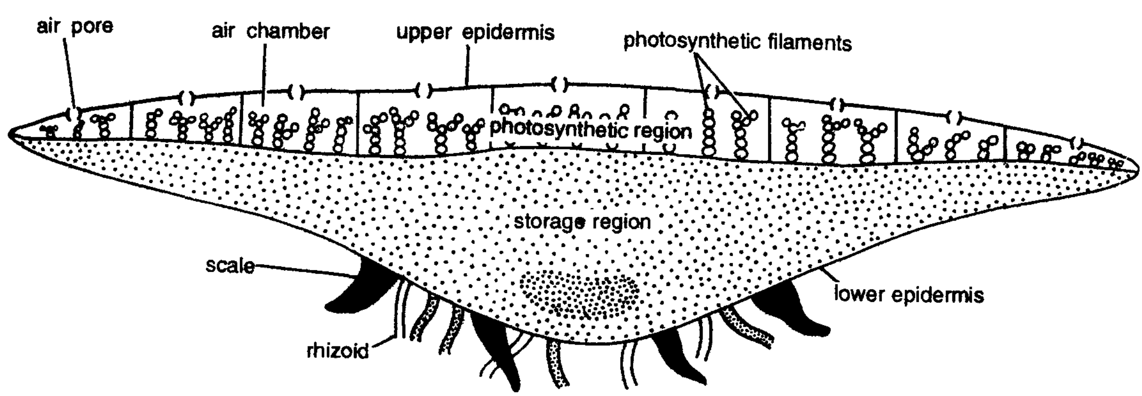


Fig. 2. ***Marchantia.*** V.t.s. thallus (diagrammatic).

Comments

1. Plants are thalloid, dorsi-ventral and prostrate.
2. Thallus is dichotomously branched and the apex of each branch is notched.
3. The dorsal side has a conspicuous midrib and many polygonal areas. These represent the underlying air chambers, each of which opens by a central air pore.
4. Each air pore is compound being made of 4-8 superimposed tiers of 3-4 cells each.
5. Certain cup-like structures present along the midrib are known as gemma cups. These contain gemmae, the vegetative reproductive bodies.
6. The ventral surface bears scales and rhizoids along the midrib.
7. Scales are arranged in two to four rows on either side of the midrib. Scales are of two types—(i) the simple or ligulate and (ii) the appendiculate. The appendiculate scales have a sub-rotund appendage at their tips.
8. The rhizoids are of two types—(i) smooth walled and (ii) tuberculate. The inner wall of the smooth walled rhizoid is smooth, while that of the tuberculate rhizoid has tuber-like or peg-like ingrowths. These appear just like circular dots in surface view.
9. The genus is dioecious, male and female thalli being different.
10. The sex organs are present on the stalked male and female receptacles. The male receptacle is known as antheridiophore and the female as archegoniophore. These structures arise from the growing apices of the thallus.

Exercise 2

***Object* : Study of anatomy of thallus.**

Work procedure

Cut V.T.s. of thallus by placing it in pith. Stain in safranin or fast green. Mount in glycerine and study.

Comments

1. Thallus is dorsiventrally differentiated into an upper photosynthetic or assimilatory region and a lower storage region.
2. Photosynthetic region is differentiated into upper epidemis and air chambers.
3. Upper epidermis is interrupted by compound, barrel-shaped air pores which open below into air chambers.
4. Air pore is made of 4-8 superimposed tiers of cells.
5. Each air chamber is filled with many branched assimilatory or photosynthetic filaments. The cell of these filaments and epidermis possess many chloroplasts.
6. Storage region is thick in the centre and gradually narrows towards margins.
7. Storage region consists of compactly arranged parenchymatous cells. A few cells are filled with oil bodies and mucilage.
8. The cells in the midrib or centre are slightly thickened to serve for conduction.
9. The lower surface of the thallus is bound by the lower epidermis, which bears scales (two types) and rhizoids (two types) in the middle region.

Exercise 3

***Object* : Study of vegetative reproductive structure : the gemma cup.**

Work procedure

Gemma cups are found on the upper side of thallus. Cut V.T.s. after placing the thallus in pith. Stain in safranin or fast green and mount in glycerine.

Comments

1. Outline is goblet-shaped with an outer wall and central cavity.
2. The outer wall shows outer photosynthetic region and inner storage region.
3. The internal structure of photosynthetic region and storage region is similar to that of thallus.
4. From the floor of the central cavity arise numerous discoid gemmae.
5. Intermingled with gemmae are many mucilage hairs or cells.
6. The gemma cup arises as a part of the thallus. It remains attached with the thallus by its base.

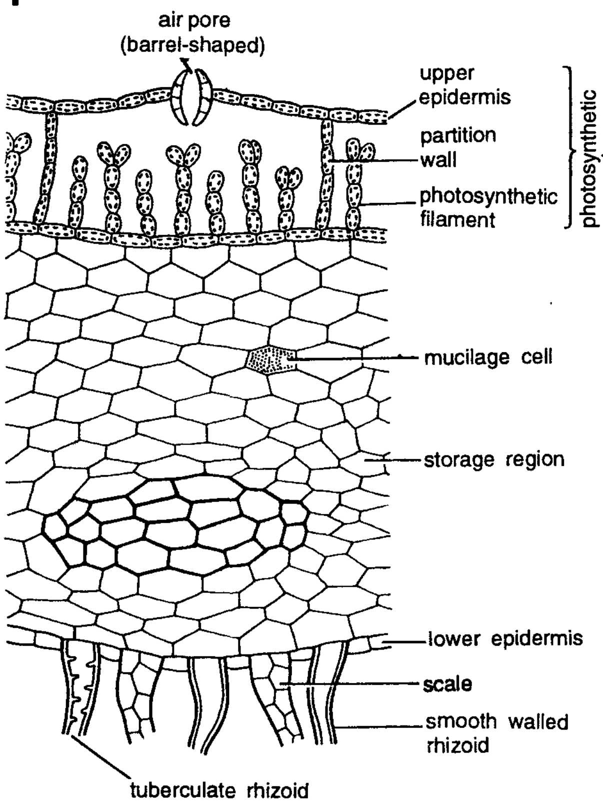


Fig. 4. ***Marchantia.*** V.t.s. thallus (a part cellular).

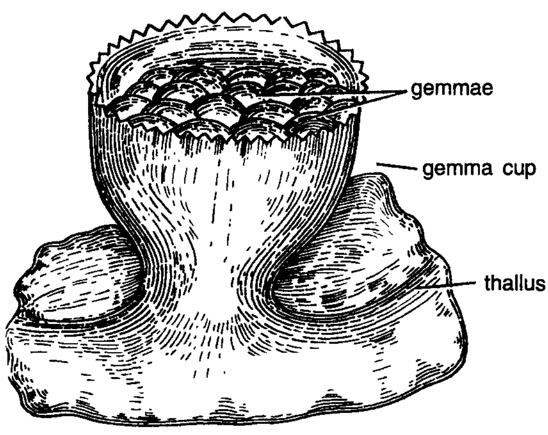


Fig. 5. ***Marchantia.*** A gemma cup.

7. Gemmae is one-celled, stalked structure. The stalk keeps gemma attached to the base of the gemma cup.
8. The disciform gemma has two shallow notches on both the lateral sides. Each notch possesses a row of apical cells.

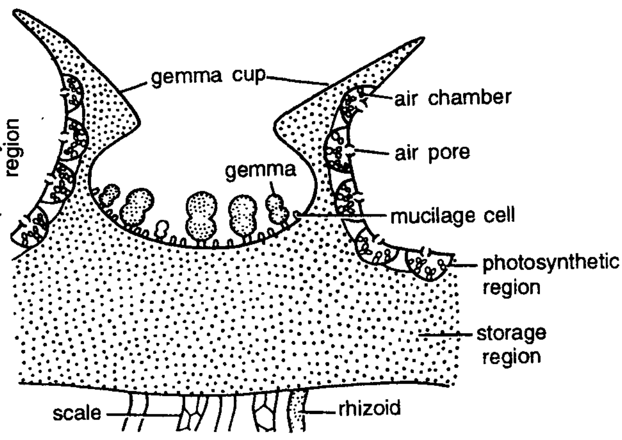


Fig. 6. ***Marchantia.*** V.t.s. of thallus through gemma cup.

9. Towards the periphery of the gemma colourless oil cells are present. Inner to them are the rhizoidal cells.
10. All the cells of gemma except the oil cells and rhizoidal cells contain chloroplast.

Exercise 4

Object **: Study of stalk of the receptacle.**

Work procedure

Sex organs are found on stalked receptacles known as antheridiophores (male) and archegoniophores (female). T.s. of stalks of any one of the receptacles is cut. It is stained in safranin or fast green and mounted in glycerine.

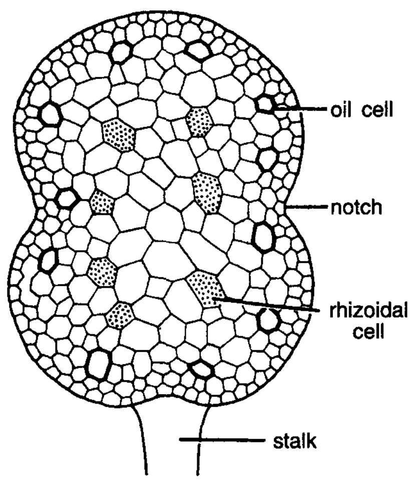


Fig. 7. ***Marchantia.*** A gemma.

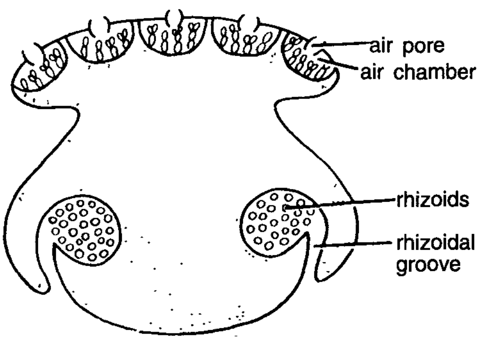


Fig. 8. ***Marchantia.*** T.s. of the stalk.

Comments

1. The stalk is dorsiventrally symmetrical.
2. It shows 2 rhizoidal grooves on the lower side, situated one on either side. It contains two types of rhizoids.
3. Upper side has photosynthetic region divided into many air chambers. It is similar to photosynthetic region of the thallus.
4. Stalks of both male and female receptacles are similar in structure.

Exercise 5

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

Work procedure

Cut a L.s. through antheridiophore, stain in safranin or fast green and mount in glycerine.

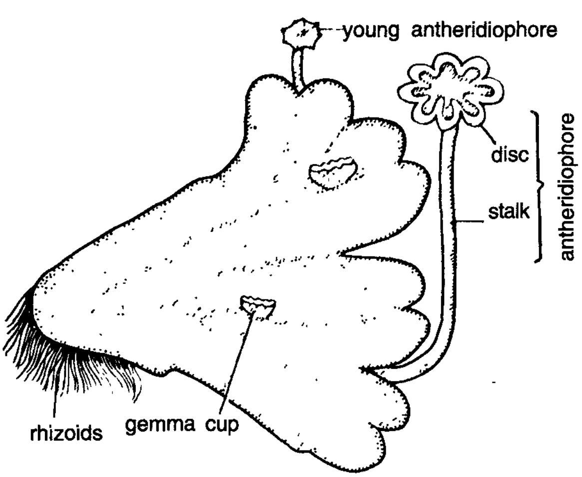


Fig. 9. ***Marchantia.*** Thallus bearing antheridiophore.

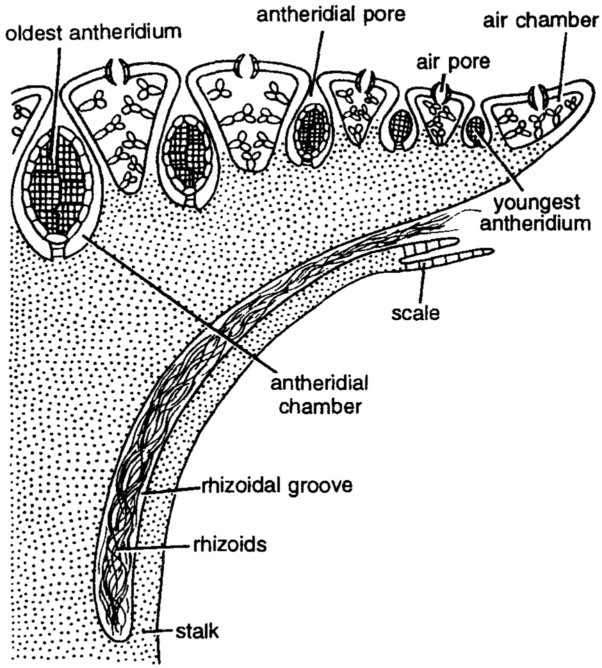


Fig. 10. ***Marchantia.*** A part of L.s. of antheridiophore.

Comments

1. The antheridiophore consists of 0.5 to 2.0 cms long stalk, bearing at its apex one eight lobed disc.
2. The peltate disc is slightly convex. The internal structure resembles with that of the thallus.
3. Epidermis is interrupted below by barrel-shaped air pores, each opening below, into an air chamber with branched assimilatory filaments.

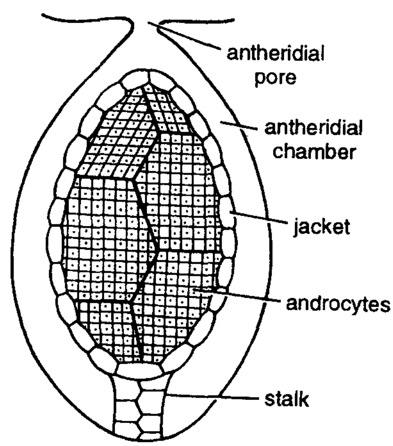


Fig. 11. ***Marchantia.*** An antheridium in anteridial cavity.

4. Alternating with air chambers, are antheridial cavities. Each antheridial cavity, that opens by an antheridial pore, has a single globular antheridium.
5. The antheridia are acropetally arranged i.e. oldest is nearest the centre and youngest nearest the margins.
6. It has a multicellular stalk attached to the base of the antheridial cavity.
7. The globular body has a single sterile jacket layer. Many androcytes or antherozoids occupy the space inside the jacket.

Exercise 6

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

Work procedure

Cut L.s. through archegoniophore, stain in safranin or fast green and mount in glycerine.

Comments

1. It is a stalked structure, (stalk 1 to 5 cms long) possessing a nine-rayed stellate disc at the apex. Groups of archegonia are found in between the rays. In each archegonial group, the archegonia are borne in radial rows.
2. After fertilization, sporophyte is formed in the same archegonium.
3. The peltate disc is convex. The internal structure is similar to that of thallus.

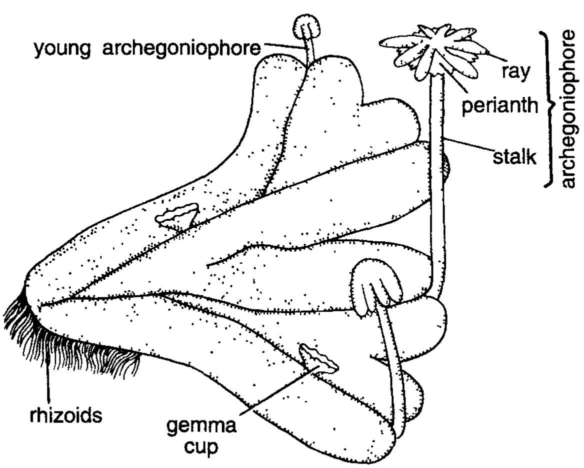


Fig. 12. ***Marchantia.*** Thallus with archegoniophores.

4. Outermost is the epidermis, interrupted by air pores. These open into air chambers with branched photosynthetic filaments.
5. In a young receptacle, archegonia are acropetally arranged on the upper side of the disc.
6. Due to the growth in the centre of the disc (which happens only after fertilization),

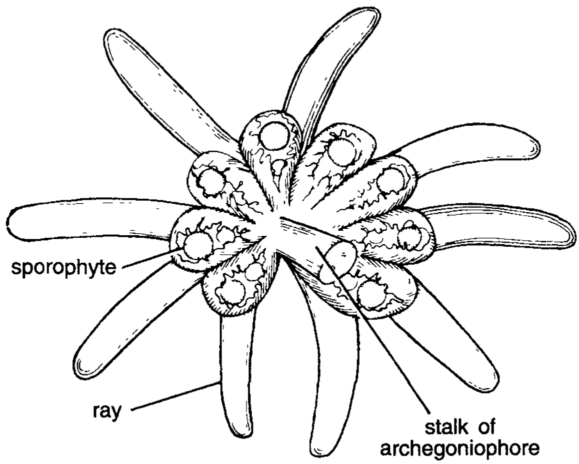


Fig. 13. ***Marchantia.*** Disc of archegoniophore as seen from lower side.

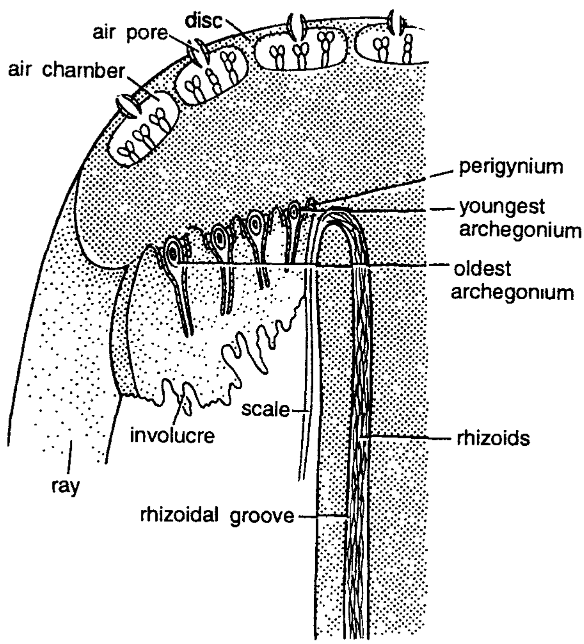


Fig. 14. ***Marchantia.*** A part of L.s. of archegoniophore.

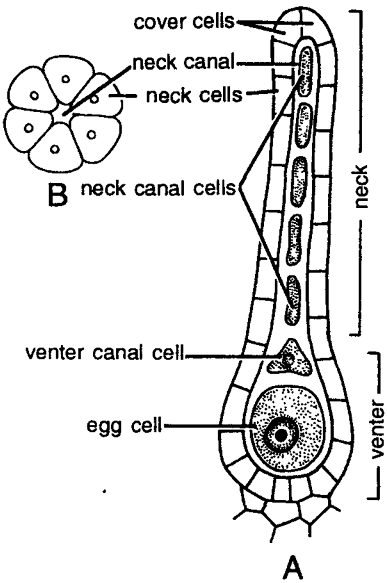


Fig. 15. ***Marchantia.*** **A.** An archegonium. **B.** T.s. through the neck.

archegonia finally become shifted towards the lower side.

7. The nearly mature archegonium has swollen venter and a long neck.
8. The venter encloses an egg cell and a venter canal cell, while the neck has 4-8 neck canal cells surrounded by six vertical rows of jacket cells.
9. The cover cells are not much distinct.
10. After fertilization perianth and involucre are developed.

Exercise 7

Object **: Study of sporophyte.**

Work procedure

Cut L.s. through disc of mature archegoniophore, stain in safranin and mount in glycerine.

Comments

1. Sporophyte develops in the same place as archegonium after its fertilization. Therefore, capsules are seen in a disc of mature archegoniophore, on the lower side. Only one sporophyte develops in one involucre.

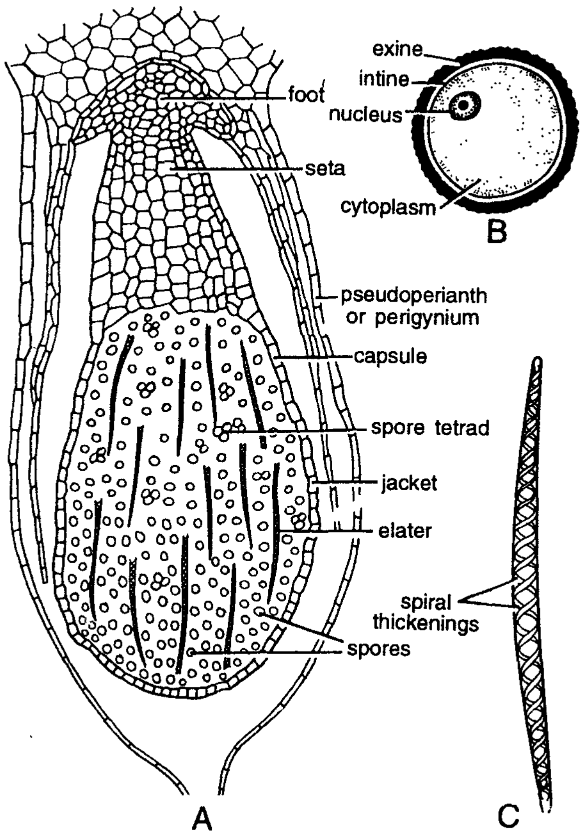


Fig. 16. ***Marchantia.*** **A.** L.s. of nearly mature sporophyte. **B.** Spore. **C.** An elater.

2. The sporophyte is enclosed by three coverings (i) calyptra, (ii) perigynium (perianth) and (iii) perichaetium (involucre). It is differentiated into a foot, seta and capsule.
3. Foot is basal and bulbous. Seta is middle and short and the capsule is spherical, occupying the distal end of the sporophyte.
4. Capsule has a single layered jacket, inside which lie many spores and elaters. Spores are arranged in tetrahedral tetrads.
5. A spore has an outer thick sculptured exine and a thin uniform intine. Every spore is uninucleate with rich cytoplasm.
6. The spores are very small in size.
7. Elaters are spindle-shaped and each possesses 2 spiral thickening bands. These are hygroscopic and help in the dispersal of spores.

Identification

Division—**Bryophyta**. (1) True roots absent and instead are present rhizoids, (2) No true vascular strand.
Class—**Hepaticopsida**. (1) Mostly thalloid, (2) Rhizoids without septa, (3) Chloroplasts without pyrenoids, (4) Columella absent from capsule.
Order—**Marchantiales**. (1) Scales present, (2) Two types of rhizoids.
Family—**Marchantiaceae**. (1) Sex organs borne on stalked receptacles, (2) Air pores barrel-shaped, (3) Elaters in capsule.
Genus—***Marchantia***. (1) Assimilatory filaments branched, (2) Scales ligulate and appendiculate both, (3) Gemma cup not crescent-shaped.

Hints for Collection

All the species of *Marchantia* found in India grow mainly in the Himalayas. Plants usually grow in moist places, either in wet open woodland or near the banks of streams. The best development is, however, seen in damp burnt soil.

Pellia

Classification

Division	—	**Bryophyta**
Class	—	**Hepaticopsida**
Order	—	**Jungermanniales**
Sub-order	—	**Metzgerineae**
Family	—	**Pelliaceae**
Genus	—	***Pellia***

Exercise 1

***Object* : Study of external morphology.**

Work procedure

Study the external features of gametophyte. Observe the thallus from dorsal and ventral sides. Remove the rhizoids and study.

Comments

1. The plants are thalloid, prostrate, drosiventral and dichotomously branched.
2. On the dorsal side there is an indistinct midrib and one celled thick lateral wings, with somewhat wavy margins.
3. At the apex is a notch in which growing point is situated. Club-shaped mucilaginous hairs are also present at the apex.
4. The shape of the thallus depends upon moisture conditions. If the thalli grow near water, they are narrow, ribbon-like, delicate and with distinct midrib. If the thalli happen to grow on dry soil, they become shorter, thick and bear an indistinct midrib.
5. Only smooth walled rhizoids are present on the ventral side towards the midrib portion. The tuberculated rhizoids and scales being altogether absent.

Exercise 2

***Object* : Study of anatomy of thallus.**

Work procedure

Cut V.T.s. of thallus, stain in safranin or fast green, mount in glycerine and study.

Comments

1. The section of the thallus shows homogeneous internal structure. Only the surface layers are little different while cells forming the rest of the tissues are all similar.
2. The section shows ventral projected midrib and wings. The midrib is 8-14 cells in thickness passing gradually on either sides into one celled wings.
3. The epidermal cells are smaller and contain more chloroplasts, than the other cells which have distinct starch grains and oil bodies.
4. In *P. epiphylla* and *P. neesiana* there are some thick-walled cells in the midrib portion which travel longitudinally from posterior to anterior end.
5. Rhizoids arise from the lower epidermis in the midrib region.
6. Vegetative reproduction takes place by adventitious shoots arising either from the midrib or from margins of the dorsal side. These branch on their separation from the parent plants and grow into new plants.

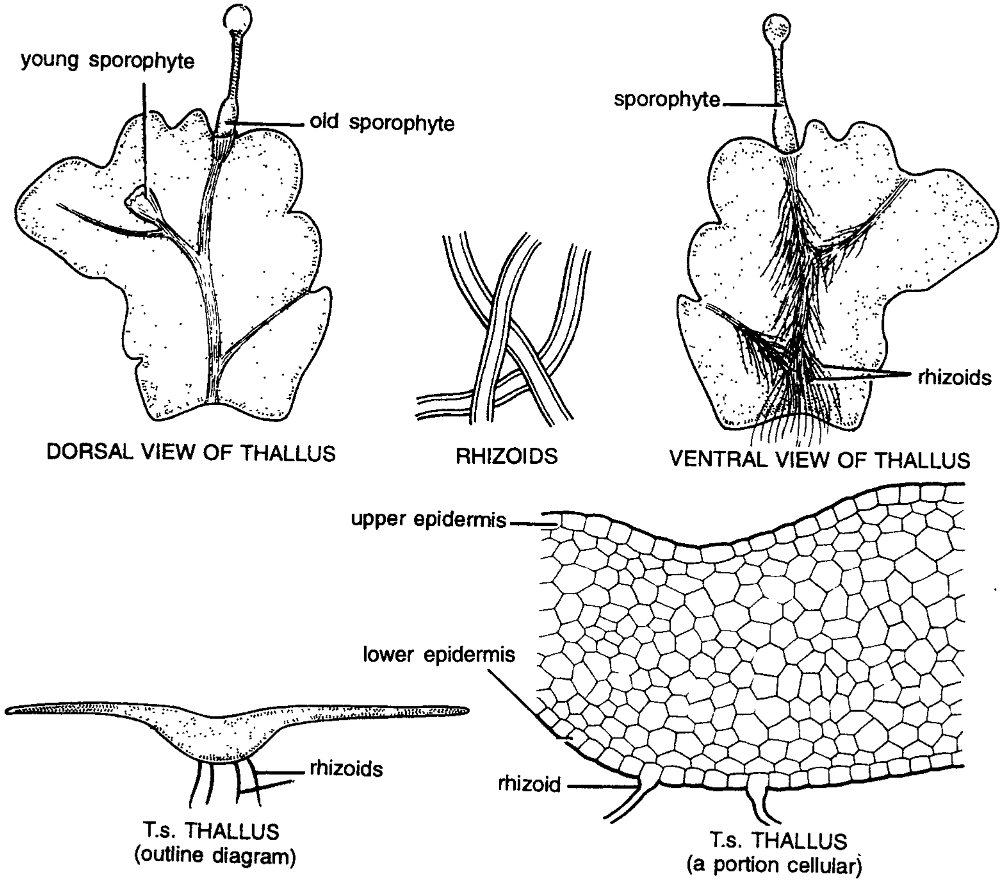


Fig. 1. *Pellia.* External features and anatomy of the gametophyte.

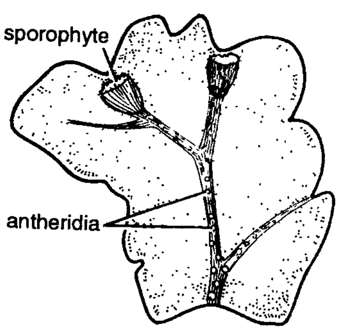


Fig. 2. *Pellia*. Fertile thalli of *P. calycina.*

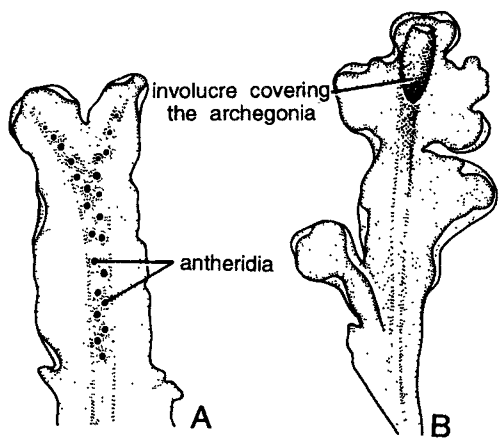


Fig. 3. *Pellia.* Fertile thallus of *P. epiphylla.*

Exercise 3

Object **: Study of male sex organs—the antheridia.**

Work procedure

Cut V.T.s. of thallus passing through midrib. Stain in safranin or fast green, mount in glycerine and study

Comments

1. All the species are dioecious except *P. epiphylla* which is monoecious and protandrous.
2. The antheridia are found just behind the growing points in two to three irregular rows in the midrib portion on the dorsal side.
3. Externally antheridia appear as wart-like projections.

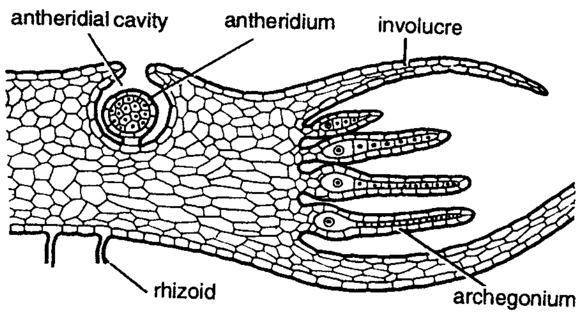


Fig. 4. ***Pellia.*** L.s. of thallus through sex organs.

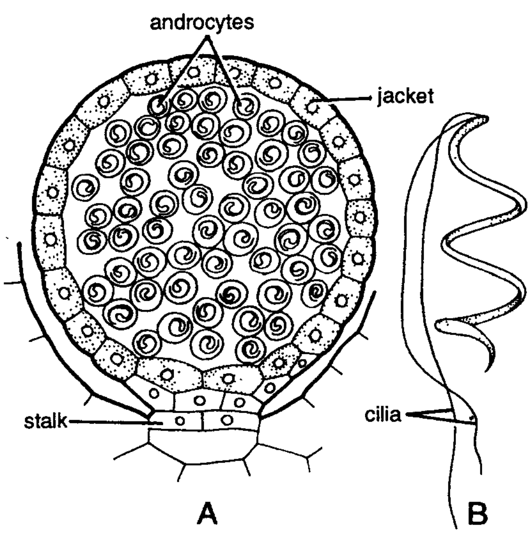


Fig. 5. ***Pellia.*** **A.** An antheridium. **B.** An antherozoid.

4. Each antheridium is present in the antheridial cavity that opens on the dorsal side by a narrow pore known as the antheridial pore.
5. Each antheridium consists of a short multicellular stalk and a nearly spherical body.
6. The body of the antheridium has a single-layered jacket enclosing many androcytes or antherozoids.
7. Each antherozoid is a spirally coiled, biciliate structure.

Exercise 4

Object **: Study of female sex organs—the archegonia.**

Work procedure

Cut L.s. of the thallus passing through growing point. Stain with safranin or fast green, mount in glycerine and study.

Comments

1. The archegonia are found in groups of 4 to 12, just near the apical cell.
2. Each archegonial group is surrounded by an involucre which may either be tubular, cylindrical or flap-like.

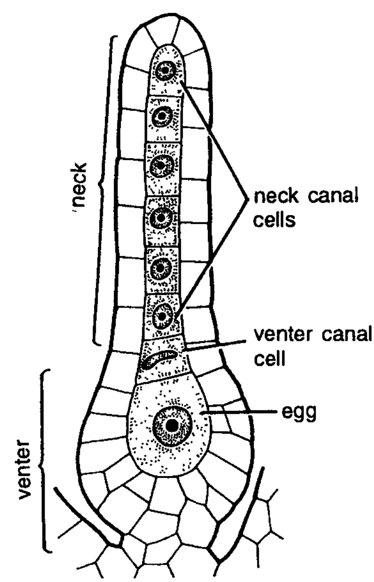


Fig. 6. ***Pellia.*** A nearly mature archegonium.

3. Archegonia are intermingled with each other without definite arrangement of older and younger archegonia.
4. A nearly mature archegonium has a short multicellular stalk, a broad venter and a long neck.
5. The jacket of a neck consists of five vertical rows of cells and encloses usually 6-8 neck canal cells.
6. The venter is two-layered thick and encloses a single venter canal cell and an egg cell.
7. The cover cells are 4 in number but are not very much distinct.

Exercise 5

Object : **Study of sporophyte.**

Work procedure

Study the external features and also cut L.s. Stain with safranin or fast green, mount in glycerine and study.

Comments

1. The mature sporophyte consists of foot, seta and capsule.
2. The foot is very prominent with its edges overlapping the basal portion of seta, thus assuming a collar-shape.
3. The seta is short when young but becomes very much elongated at maturity.
4. The capsule is covered by calyptra and involucre respectively.
5. The capsule is nearly spherical and consists of an outer jacket composed of 2 or 3 layers.
6. The jacket layers of capsule have radial thickenings except at 4 places at the top wherefrom the dehiscence takes place.
7. Inside the jacket at the base of the capsule is present a sterile tissue, known as elaterophore. On this elaterophore are attached some 20 to 100 elaters, radiating into the cavity of capsule.
8. In the remaining cavity of the capsule are present spores and elaters.
9. The spores are arranged in tetrahedral tetrads and are formed by lobing of spore mother cell.
10. Each spore is unicellular. The elater is a long, slender, spindle-shaped, structure with generally 2 but sometimes 3-4 spiral thickening bands.

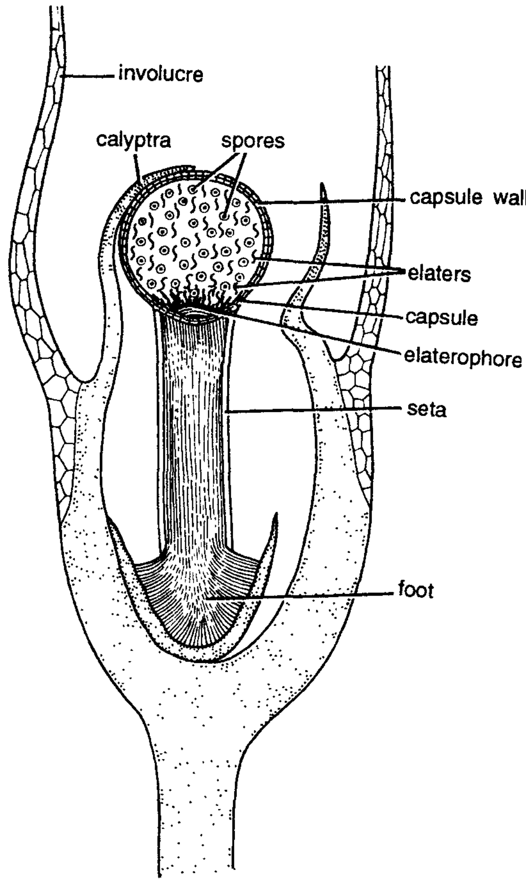


Fig. 7. ***Pellia.*** L.s. through the mature sporophyte.

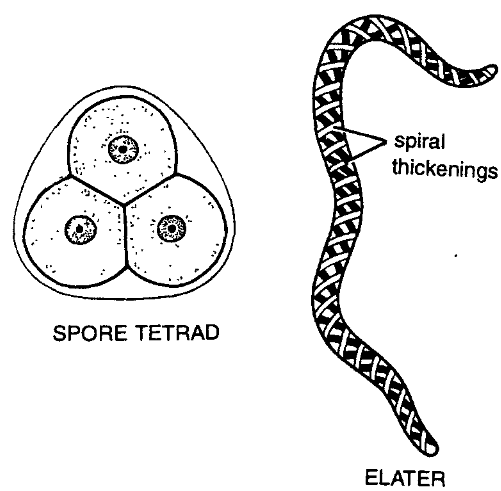


Fig. 8. ***Pellia.*** A spore tetrad and an elater.

Identification

Division—**Bryophyta**. (1) True roots absent and instead are present the rhizoids, (2) No true vascular strand.

Class—**Hepaticopsida**. (1) Rhizoids without septa, (2) Chloroplasts without pyrenoids, (3) Columella absent from capsule and there are stomata on capsule wall.

Order—**Jungermanniales**. (1) Scales absent, (2) Rhizoids smooth walled, (3) Antheridia and archegonia are borne at the apices, (4) Archegonial neck consists of 5 vertical rows of cells.

Sub-order—**Metzgerineae**. (Jungermanniales Anacrogynae) (1) Gametophyte usually a thallus, very rarely a stem with leaves, (2) Archegonia arise from the segments of apical cell. The apical cell is not consumed in the formation of archegonium, (3) Jacket of the capsule is 2 to 5 layers thick.

Family—**Pelliaceae**. (1) Sex organs are scattered on the dorsal surface of thallus, (2) The capsule has a basal elaterophore.

Genus—***Pellia***. (1) Thallus often lobed by irregular incisions, (2) Archegonia are present just behind the apical cell in groups of 4 to 12, (3) Capsule dehisces by 4 valves.

Hints for Collection

Two species of *Pellia* found in India are *P. calycina* and *P. epiphylla*. The former is chiefly found in western Himalayas and Kumaon, whereas the latter is common in eastern Himalayas and Skikkim. These very commonly grow in moist, shady places especially by the sides of ditches, streams and moist rocks, etc.

Porella (*Madotheca*)

Classification

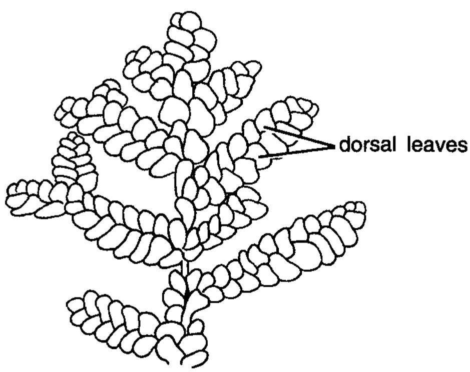
Division	—	**Bryophyta**
Class	—	**Hepaticopsida**
Order	—	**Jungermanniales**
Sub-order	—	**Jungermannineae**
Family	—	**Porellaceae**
Genus	—	***Porella***

Exercise 1

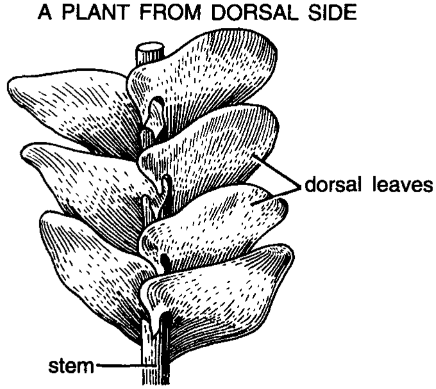
Object : **Study of external features.**

Work procedure

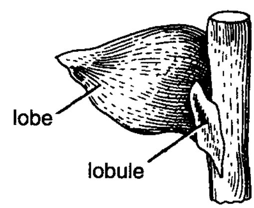
Study the gametophytic thallus and note down the characters.



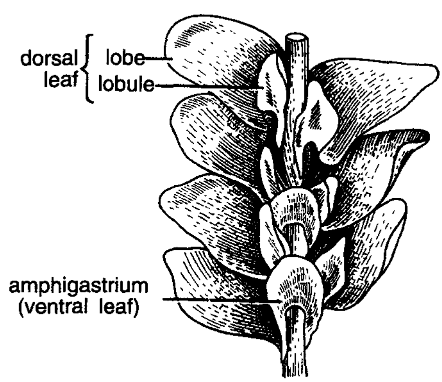
A PLANT FROM DORSAL SIDE



A PORTION OF BRANCH (dorsal view)



DORSAL LEAF



A PORTION OF BRANCH (ventral view)

Fig. 1. ***Porella.*** External features of gametophyte.

Comments

1. The plants are large, prostrate, dorsiventral and grow in compact greenish patches.
2. Thallus is differentiated into a branched stem and leaves, arranged in 3 rows.
3. Out of the three rows of leaves, the two are dorsal and one is ventral. These ventral smaller leaves are known as the amphigastria.
4. The dorsal leaves are incubously arranged (*the anterior margin of the lower leaf is covered by the posterior margin of the leaf next above it, when seen from above*) and each is divided into a larger (antical) lobe and a smaller (postical) lobule.
5. The lobe is usually ovate with rounded apex while the lobule is narrower with acute apex.
6. Many rhizoids are present on the lower side of the stem.

Exercise 2

***Object* : Study of anatomy of axis.**

Work procedure

Place the thallus in pith. Cut T.s. and stain either in safranin or fast green. Mount in glycerine and study.

Comments

1. The axis is differentiated into an epidermis, cortex and medulla.

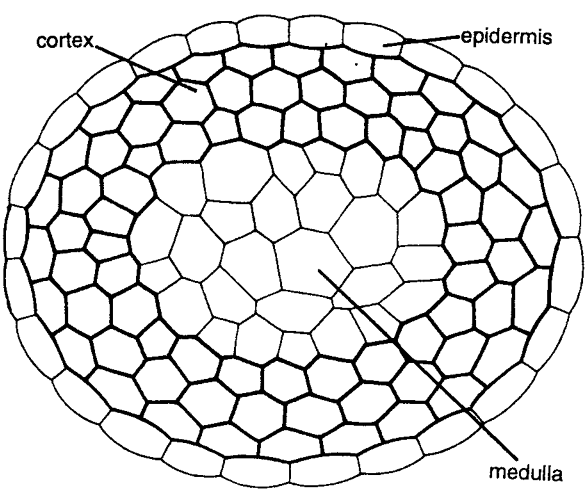


Fig. 2. ***Porella.*** T.s. axis (cellular).

2. The cortical cells are usually small and thick-walled, in comparison to the large, thin-walled medullary cells.

Exercise 3

***Object* : Study of antheridial branch and antheridium.**

Work procedure

Tease out the bracts and expose the antheridia, from antheridial branches. Stain in safranin, mount in glycerine and study.

Comments

1. The genus is dioecious and the male and female plants can be externally differentiated.
2. The male plants are usually smaller with the antheridial branches projecting nearly at right angles from the main axis.
3. The antheridial branch has many closely arranged bracts.
4. In the axil of each bract, antheridium is present.
5. Each antheridium consists of a long stalk, composed of two rows of cells subtending at its apex a globular body.
6. The body has a jacket layer composed of 2-3 layers in the basal part and one layer in the upper part.
7. Within the jacket are present many androcytes which ultimately go to form the biciliate antherozoids.

Exercise 4

***Object* : Study of archegonial branch and the archegonium.**

Work procedure

Tease the apex of archegonial branch. The archegonia appear between the leaves. Stain with safranin or fast green, mount in glycerine and study.

Comments

1. The genus is dioecious. The female plants are distinct from the male plants.
2. Female plants are usually larger though archegonial branches are shorter than the antheridial branches.

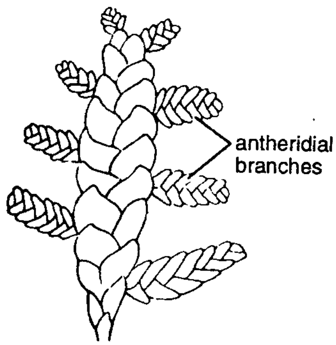


Fig. 3. *Porella.* A male plant with antheridial branches.

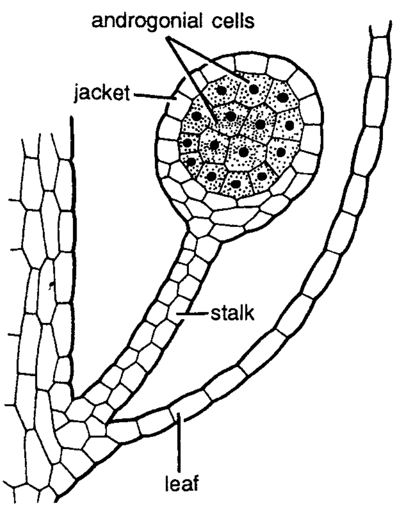


Fig. 4. *Porella.* A portion of an antheridial branch bearing antheridium in the axil of leaf.

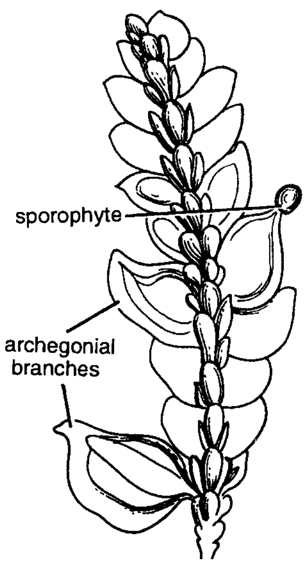


Fig. 5. *Porella.* A female plant with archegonial branches and a sporophyte.

3. The archegonia are found at the apex of archegonial branch.
4. The first few archegonia are arranged acropetally and the last archegonium is formed by the apical cell itself, thus checking the further growth of archegonial branch.
5. The archegonium consists of a broad neck and the venter.
6. The neck consists of five vertical rows of cells, typical of jungermanniales, and encloses 6-8 neck canal cells.
7. The venter wall is two layered. It encloses a venter canal cell and an egg cell.

Exercise 5

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

Work procedure

Cut a thin L.s. of the capsule. Stain in fast green, mount in glycerine and study.

Comments

1. The sporophyte consists of a small foot, a short seta and a globose capsule.

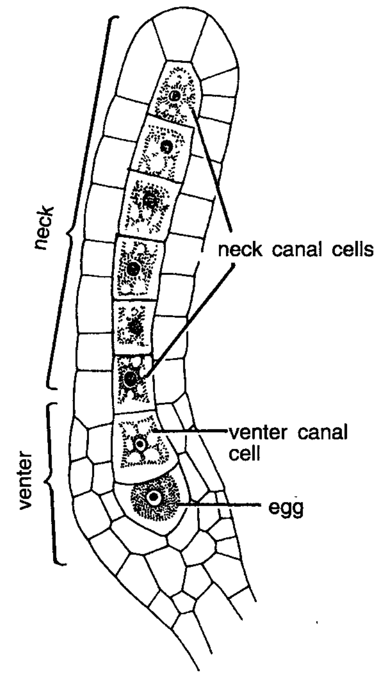


Fig. 6. *Porella.* A nearly mature archegonium.

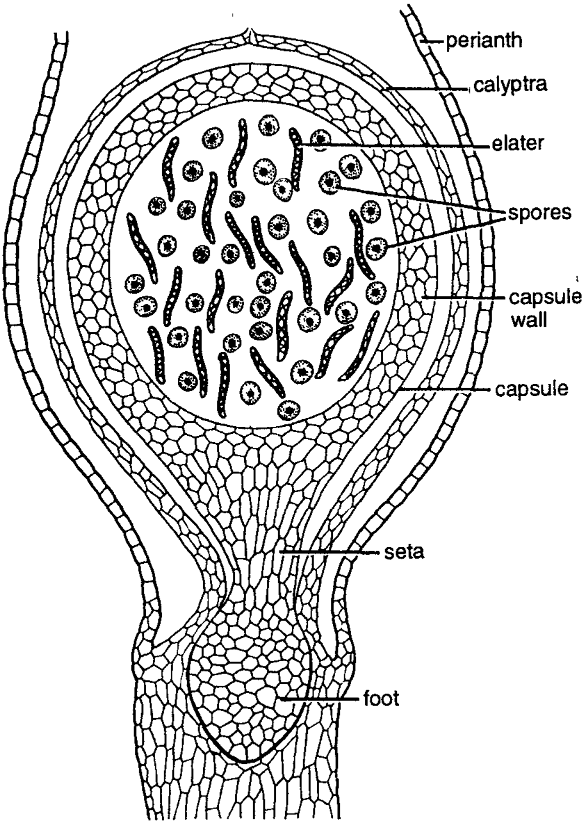


Fig. 7. ***Porella.*** L.s. mature sporophyte.

2. The young sporophyte is covered by calyptra and perianth.
3. The calyptra developed from venter of archegonium is more than one layered thick. It surrounds the sporophyte until its maturity.
4. The perianth is formed by the fusion of two uppermost perichaetial bracts and is more or less dorsiventrally compressed, in the anterior region.
5. The capsule is globose and consists of a jacket layer, two or six cells in thickness.
6. Enclosed within the jacket are many spores and elaters, the latter help in dispersal of spores.

Exercise 6

Object **: Study of a spore and an elater.**

Work procedure

Break the sporophyte open to release spores and elaters. Stain in safranin and mount in glycerine.

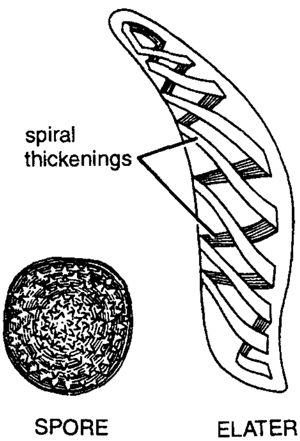


Fig. 8. ***Porella.*** A spore and elater.

Comments

1. A spore measures from 40 to 55 microns in diameter.
2. Each spore has an outer, smooth or papillose exospore and an inner, smooth endospore. Sometimes a third layer may also be present. It is known as outer exospore or perinuium.
3. Inside the wall layers, is a single nucleus embedded in rich cytoplasm.
4. An elater is a longitudinally stretched and coiled structure with 2-3 spiral thickening bands.

Identification

Division—**Bryophyta.** (1) True roots absent and instead are present the rhizoids, (2) No true vascular strand.

Class—**Hepaticopsida.** (1) Rhizoids without septa, (2) Chloroplasts without pyrenoids, (3) Columella absent from capsule and there are no stomata on capsule wall.

Order—**Jungermanniales.** (1) Scales absent, (2) Rhizoids smooth walled, (3) Antheridia and archegonia are borne at the apices, (4) Archegonial neck consists of 5 vertical rows of cells.

Sub-order—**Jungermannineae.** (Jungermanniales Acrogynae) (1) Gametophyte with stem and two rows of dorsal, and a third ventral row of leaves, (2) Archegonia in a terminal cluster and the last archegonium is formed by the apical cell.

Family—**Porellaceae.** (1) Rhizoids are scarce and form tufts at the bases of amphigastria, (2) Leaves incubously arranged, (3) Locule (postical lobe) is distinct, (4) Perianth large, inflated with compressed mouth and is bilabiate.

Genus—***Porella.*** Characters same as those of Porellaceae.

Hints for Collection

Out of the 34 species so far recorded from India, only a few are found in South India, others have been collected from the Himalayas. The genus usually grows on shaded, moist rocks, stones or on the bark of trees particularly at the base and rarely on the soil.

Frullania

Classification

Division	—	**Bryophyta**
Class	—	**Hepaticopsida**
Order	—	**Jungermanniales**
Sub-order	—	**Jungermannineae**
Family	—	**Frullaniaceae**
Genus	—	***Frullania***

Exercise 1

Object **: Study of external morphology.**

Work procedure

Study the external features of the gametophyte, observe the arrangement and structure of leaves.

Comments

1. The plants are large or medium sized but rarely small. These are reddish-brown or nearly black in colour.
2. The plant body is a gametophyte. It is differentiated into a branched, prostrate, central, stem-like axis and the leaves.
3. The thallus is attached to the substratum by unicellular, smooth walled rhizoids which arise in tufts from the bases or middle of the amphigastria.
4. The prostrate stem is pinnately or bipinnately branched.
5. The leaves on the stem are arranged in three rows—two rows of lateral leaves and a row of underleaves or amphigastria. Each lateral leaf is bipartite forming a large expanded antical lobe and a small postical lobe.
6. The antical lobe is obliquely ovate or sub-orbicular and almost always entire.
7. The postical lobe, also called lobule is cucullate, galeate or saccate. It may either form a water sac or may remain open.
8. The postical lobe usually bears a short subulate process, the stylus near its attachment between the postical lobe and the stem.
9. Underleaves or amphigastria are usually smaller. These are rounded, notched or deeply lobed.

Exercise 2

Object **: Study of anatomy of the stem.**

Work procedure

Cut a T.s. of the axis, stain in safranin, mount in glycerine and study.

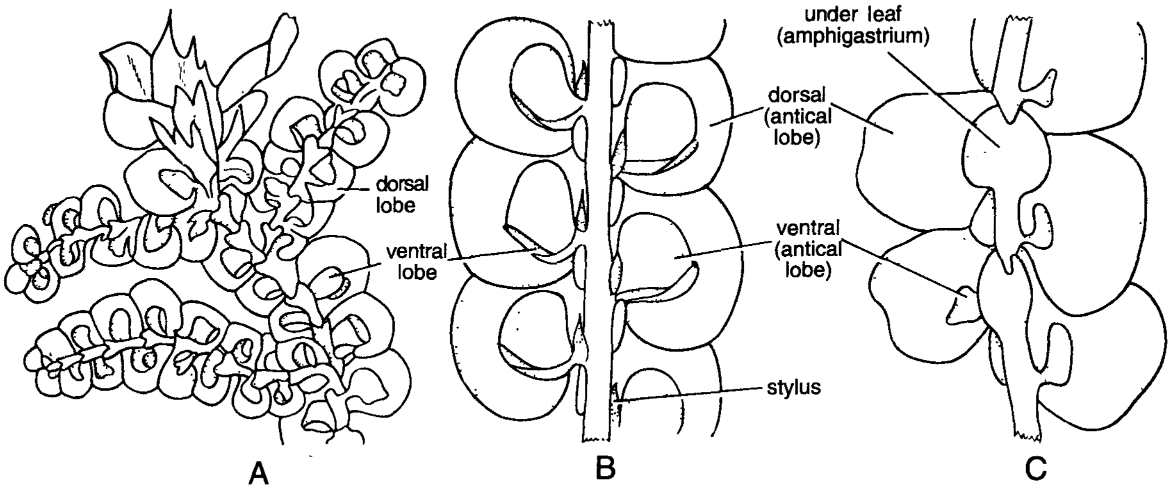


Fig. 1. ***Frullania.*** External features of the gametophyte.

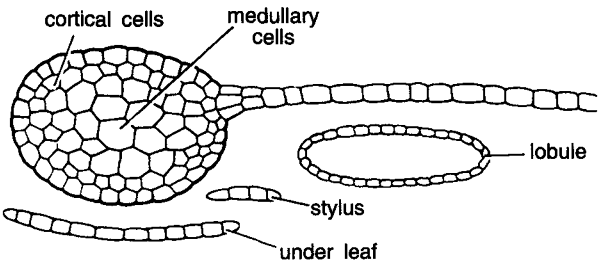


Fig. 2. ***Frullania.*** Anatomy of the stem.

Comments

1. T.s. of the stem is almost circular in outline.
2. Well defined epidermis is absent.
3. The section shows cortical and medullary regions.
4. The cortex is 2 to 3 cells deep. It consists of small cells with distinct thickened pigmented walls.
5. The central part is called medullary region. It is made of large and elongated cells with thin coloureless walls.

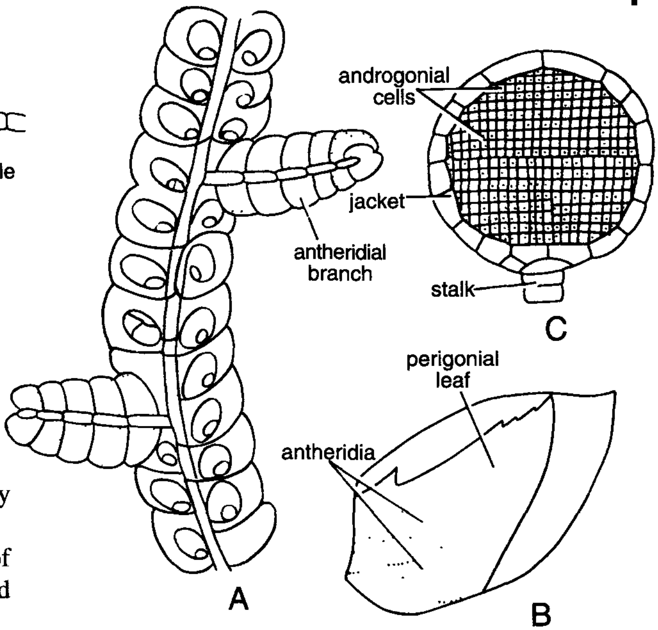


Fig. 3. ***Frullania.*** **A.** Antheridial branch, **B.** Perigonial leaf from ventral surface showing two antheridia. **C.** Single antheridium.

Exercise 3

Object **: Study of the antheridial branches and antheridia.**

Work procedure

Antheridia are found on short lateral branches. These could be seen by carefully removing the leaves, staining of antheridia with safranin and then by mounting in glycerine.

Comments

1. The species of this genus are dioecious as well as monoecious.
2. Monoecious species are autoicous i.e. antheridia and archegonia are borne by different branches.
3. The antheridia occur in the axils of bracts (perigonial bracts) borne on short laterals.
4. The perigonial bracts are densely imbricate and are divided into two almost equal lobes, large in size.
5. There are usually two antheridia in the axil of each bract.
6. A mature antheridium consists of a stalk and a body.
7. The stalk is slender and multicellular being composed of two rows of cells.
8. The body of the antheridium is globose. It consists of a central mass of androcyte mother cells and one-celled wall or jacket.
9. Androcyte mother cell divides, each forming two biflagellate antherozoids.

Exercise 4

Object **: Study of the archegonial branch and archegonium.**

Work procedure

Tease out the apices of short lateral branches of the female plant or female branch. Locate the groups of archegonia, stain with safranin, mount in glycerine and study.

Comments

1. The archegonia occur in groups at the apices of short lateral branches.
2. Each group has two to four archegonia.

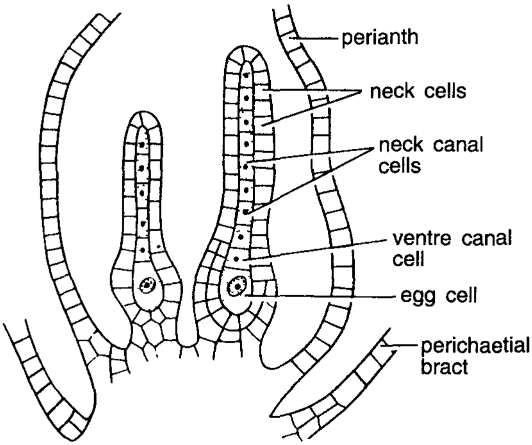


Fig. 4. ***Frullania.*** L.s. of archegonial branch showing two nearly mature archegonia.

3. A group of archegonia is surrounded by 2 to 5 pairs of perichaetial bracts which are dentate or lobed and larger than foliage leaves.
4. The inner pairs of bracts are connate with each other.
5. The uppermost bracts are laterally fused to form perianth.
6. The perianth is inversely heart shaped and much contracted to form a small, tubular mouth.
7. A mature archegonium is a flask shaped structure consisting of a basal ventre and the upper neck.
8. The venter has two layered wall that surrounds a small egg cell and a venter canal cell situated just above it.
9. The neck consists of five vertical rows of neck cells which surround an axial row of eight neck canal cells.

Exercise 5

Object : **Study of the sporophyte.**

Work procedure

Study the sporophyte externally. Also study the L.s. of the sporophyte.

Comments

1. A mature sporophyte consists of foot, seta and capsule.
2. The foot remains embedded in the tissue of the female branch.
3. The seta is very short and only 8-9 cells thick. It is not very distinct from the capsule because of the mass of thin walled tissue at the base of the capsule (resembling apophysis of moss sporogonium).
4. The capsule wall is made of two layers of cells.

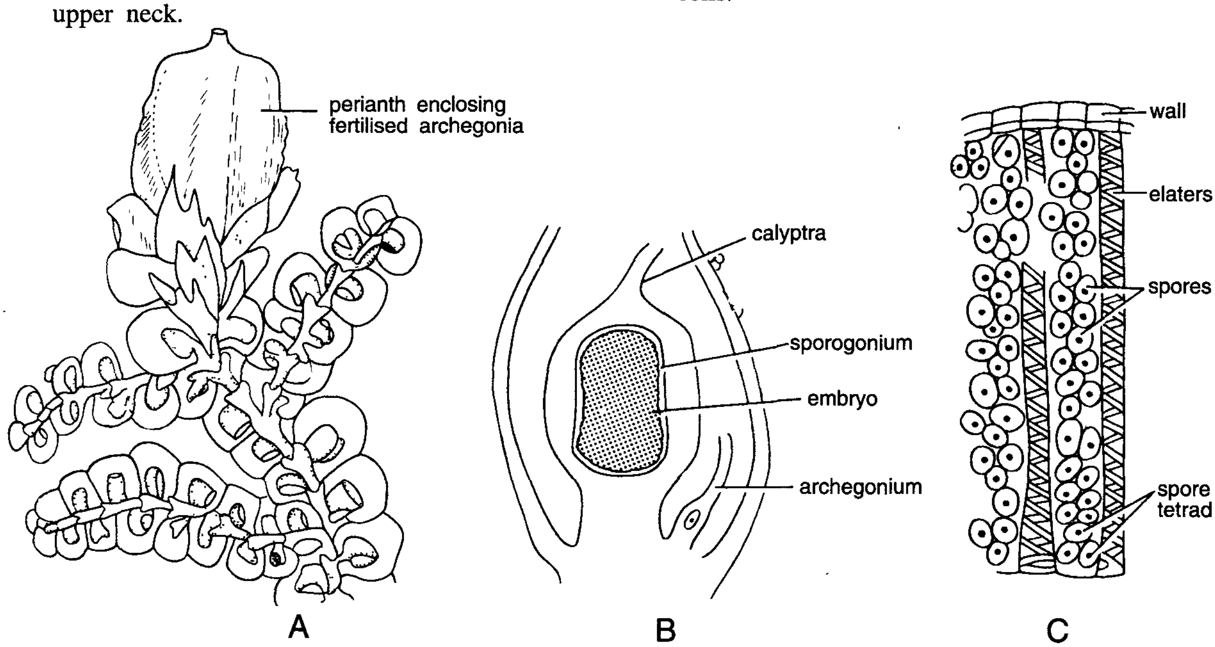


Fig. 5. ***Frullania.*** The sporophyte. **A.** External features, **B.** L.s. of young sporophyte, **C.** L.s. of nearly mature capsule.

5. The cells of the outer layer have thick rod-like fibres on their lateral walls especially at the corners.
6. The cells of the inner layers have walls with irregular network of thickening fibres.
7. Inside the wall there are about 80 elaters and large number of spores.
8. The elaters run from the roof to the floor of the capsule. These alternate with vertical rows of spores. Each elater is flattened. It has a single broad spiral band of thickening. The lower end of the elater is trumpet shaped.
9. The spores are large, oblong to roundish. The wall is two layered, the exospore and the endospore. The exospore is rough, tuberculate and verrucose. The cytoplasm has a nucleus and a chloroplast.

Identification

Division—**Bryophyta**. (1) The roots absent, but rhizoids present, (2) True vascular strand lacking.

Class—**Hepaticopsida**. (1) Rhizoids without septa, (2) Chloroplasts without pyrenoids, (3) Columella absent from capsule and there are no stomata on capsule wall.

Order—**Jungermanniales**. (Jungermanniales Acrogynae). (1) Gametophyte with stem and two rows of dorsal and a third ventral row of leaves, (2) Archegonia in terminal cluster and the last archegonium is formed by the apical cell.

Family—**Frullaniaceae**. (1) Stems usually pinnately branched, (2) Rhizoids form a tuft at the base or middle of the under leaf, (3) Lateral leaves complicate-bilobed, (4) Perianth compressed and obtrusely triangular in cross section.

Genus—***Frullania***. (1) Branch develops from the ventral half of a lateral segment replacing lobule of the leaf, (2) Presence of stylus.

Hints for Collection

Frullania is represented by about 700 species, of which about 39 are reported from India. The species can be collected from wet shady rocks or trunks of tree. It usually forms flat patches or extensive mats on the rocks or on tree trunks.

Anthoceros (Horn Wort)

Classification

Division	—	**Bryophyta**
Class	—	**Anthocerotopsida**
Order	—	**Anthocerotales**
Family	—	**Anthocerotaceae**
Genus	—	***Anthoceros***

Exercise 1

Object **: Study of external features.**

Work procedure

Study the external features of the gametophyte from dorsal and ventral sides. Note the absence of scales and presence of only smooth walled rhizoids.

Comments

1. Plant body is thalloid, somewhat lobed or radially dissected and generally suborbicular.
2. The thallus is less often dichotomously branched and lack a definite midrib.

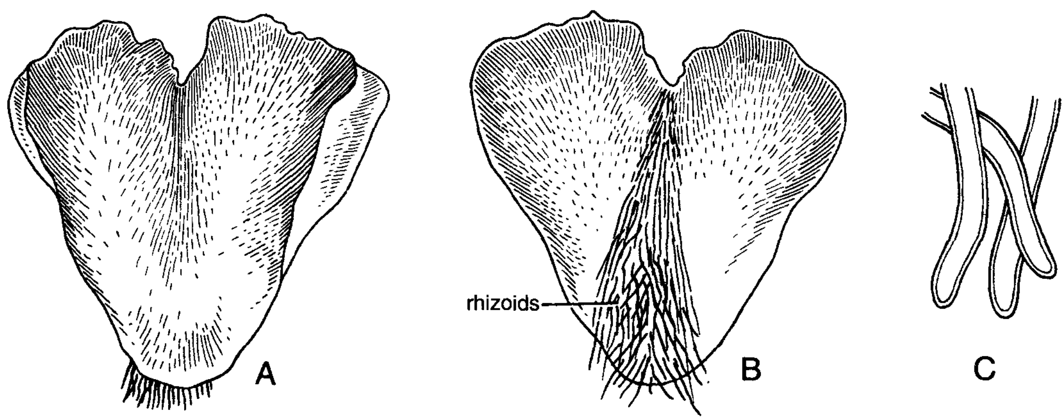


Fig. 1. *Anthoceros*. External features of gametophyte. **A.** Dorsal surface. **B.** Ventral surface. **C.** Rhizoids.

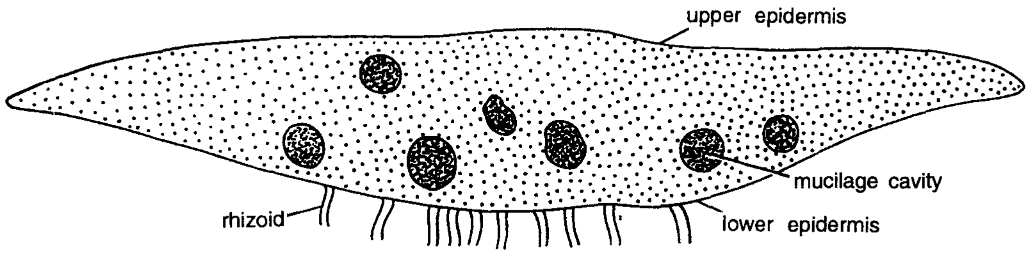


Fig. 2. ***Anthoceros.*** V.t.s. thallus (diagrammatic).

3. The dorsal surface of the thallus is generally smooth, velvety or rough.
4. The ventral surface bears smooth walled rhizoids only.
5. On the ventral side, a few bluish spots are seen indicating the presence of filaments of blue-green-alga (viz. *Nostoc* or *Anabaena*).
6. Sex organs are situated on the dorsal side and are embedded in the tissue of the thallus.
7. The sporophyte, however, is linear and elongated structure, arising from the dorsal side.

Exercise 2

Object : **Study of anatomy of thallus.**

Work procedure

Place the thallus in pith. Cut V.t.s. and stain in fast green. Mount in the glycerine and study.

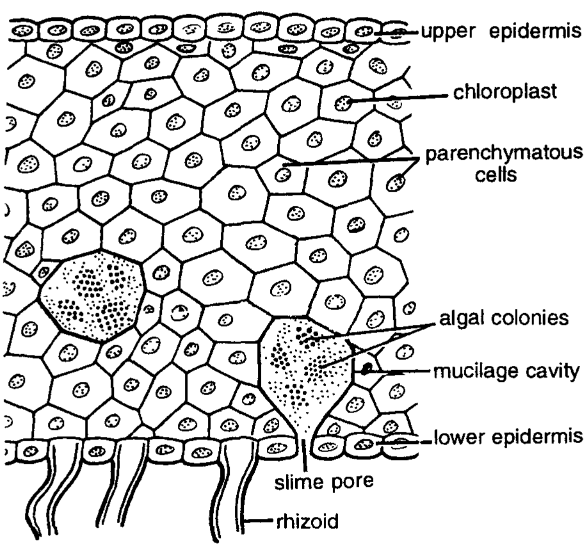


Fig. 3. ***Anthoceros.*** A part of V.t.s. of thallus (cellular).

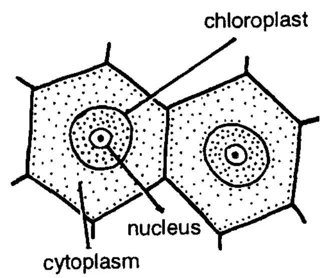


Fig. 4. ***Anthoceros.*** Cells of thallus with chloroplast.

Comments

1. Thallus is few cells in thickness in the middle and becomes thinner towards the margins.
2. Internal structure is homogeneous (i.e. all cells are alike). Air pores and air chambers are absent.
3. On upper side is present upper epidermis and lower epidermis on the lower side.
4. Parenchymatous tissue lies between these two layers.
5. Each parenchymatous cell has a distinct nucleus and a chloroplast.
6. Each of the cells has a large chloroplast with a single pyrenoid except the cells of lower epidermis producing rhizoids. There are two chloroplasts in the cells of *A. pearsonii* and four in *A. hallii.*
7. On the ventral side there are certain mucilage-filled cavities which open by slime pores, through the ventral epidermis.
8. The endophytic algae *Nostoc* or *Anabaena* present in he mucilage cavities, enter through these slime pores.
9. Rhizoids are smooth-walled and arise in the middle region of the thallus from the lower epidermis.

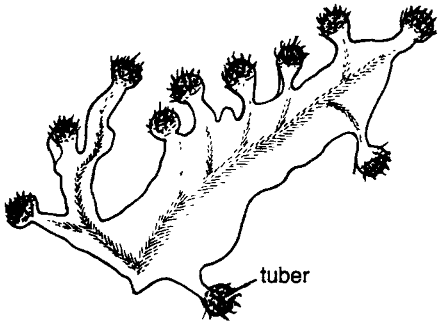


Fig. 5. ***Anthoceros.*** Thallus showing tubers.

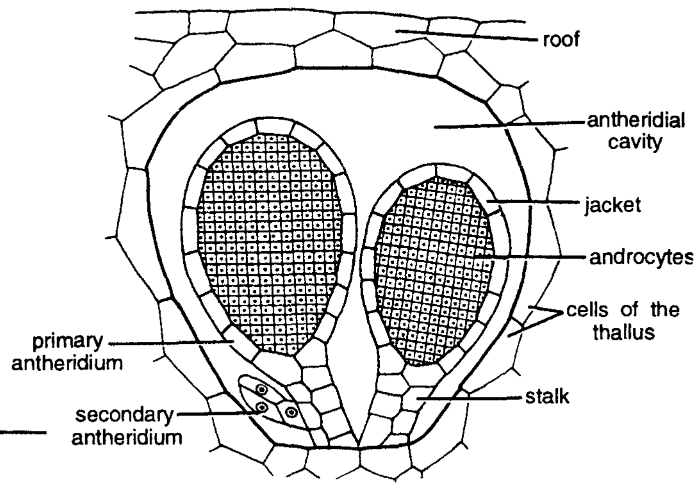


Fig. 6. ***Anthoceros.*** Antheridial cavity with primary and secondary antheridia.

Exercise 3

Object **: Study of tuber.**

Work procedure

Study a thallus with tubers. It can be stained with safranin and mounted in glycerine.

Comments

1. The tubers are the vegetative reproductive structures.
2. Tubers are formed under unfavourable conditions, on the dorsal side along the margins of the thallus.
3. A tuber in section shows outer 2-3 corky layers, protecting the inner tissue, containing the reserve food material.
4. On return of the favourable conditions, tubers develop into new thalli.

Exercise 4

Object **: Study of antheridium.**

Work procedure

Cut L.s. of thallus, select a section showing antheridia, stain in fast green, mount in glycerine and study the antheridia.

Comments

1. Both antheridia and archegonia remain embedded in the dorsal region of the thallus and are acropetally arranged.
2. Few species are monoecious (homothallic), but some are dioecious (heterothallic).
3. Monoecious species are frequently protandrous (antheridia maturing first).
4. The antheridia are present in the antheridial cavity or antheridial chamber with a sterile roof of 2-3 layers.
5. Each antheridial cavity contains about 1-4 or more primary antheridia. Secondary antheridia arise from the stalks of primary antheridia and ultimately there may be as many as 25 in each antheridial cavity.
6. The mature antheridium is a stalked, club-shaped structure with single layered jacket. Each cell possesses a prominent plastid.
7. Inside the jacket, there are large number of androcytes.

Exercise 5

Object **: Study of archegonium.**

Work procedure

Cut L.s. of thallus, select a section that shows archegonia, stain in fast green, mount in glycerine and study archegonia.

Comments

1. The thalli are generally monoecious and protandrous (antheridia maturing first).

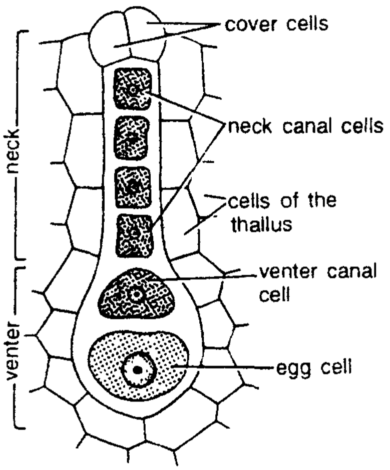


Fig. 7. ***Anthoceros.*** An archegonium.

2. The archegonia are embedded in the thallus and only the cover cells project beyond the general surface of the thallus.
3. They are in direct contact with the vegetative cells, lateral to them.
4. Archegonium consists of a neck and a swollen venter.
5. The nearly mature archegonium has 4 cover cells, 4-6 neck canal cells, one venter canal cell and one egg cell.

Exercise 6

Object **: Study of sporophyte.**

Work procedure

Study the external features of the sporophyte. Cut a L.s., stain in safranin and fast green, mount in glycerine and study the internal structure.

Comments

1. The sporophytes are linear, 2-3 cms long, elongated structure, arising from the dorsal side of the thallus.
2. The base of each sporophyte is enclosed by an involucre, made of gametophytic tissue.

   (The internal structure of the sporophyte can be understood by studying the transverse and longitudinal sections).

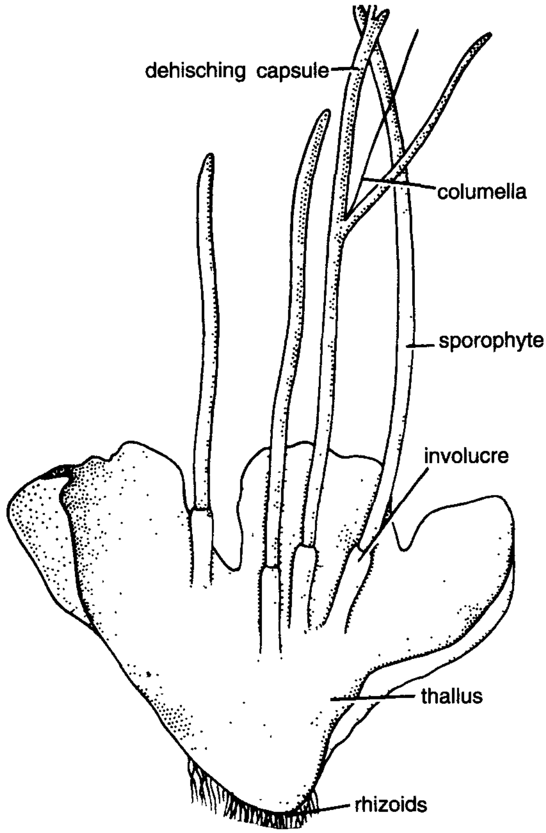
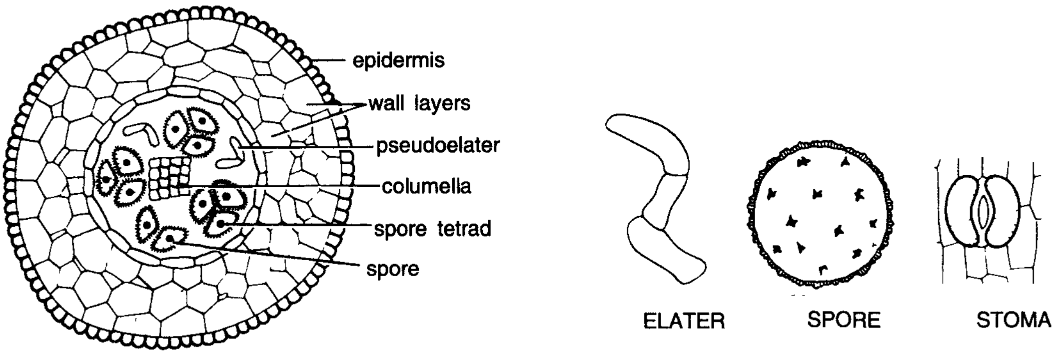


Fig. 8. ***Anthoceros.*** Gametophytic thallus bearing sporophytes.

L.s. of the sporophyte shows following characters:

3. The mature sporophyte is made of a lower foot, middle meristematic zone and upper capsule.
4. The foot is bulbous and is deeply rooted in the gametophytic tissue.
5. The seta is absent and instead is present the meristematic zone.
6. In the centre is columella, composed of 16 vertical rows of cells, extending from the base to the tip of the capsule.
7. Surrounding the columella is a cylinder of sporogenous tissue which extends from the base to the tip of the capsule where it over-arches the central columella. It reveals sporogenous series, from one layered archesporium at the base, to mature spores and pseudo-elaters at the tip.



T.S. CAPSULE

Fig. 9. ***Anthoceros.*** T.s. capsule, elater, spore and stoma on the capsule wall.

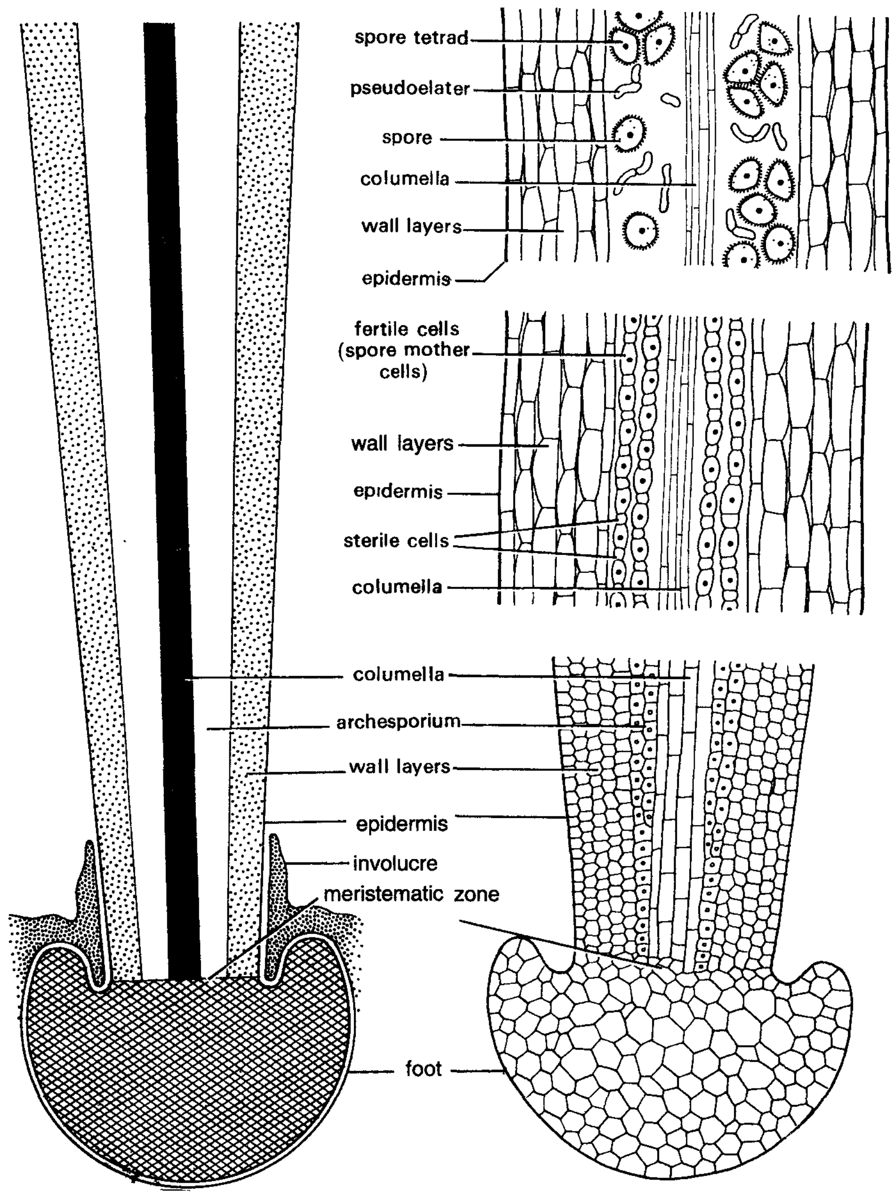


Fig. 10. ***Anthoceros.*** L.s. through sporophyte (diagrammatic and parts cellular).

8. At the base of the capsule is a single to double layered archesporium, while little higher up, archesporium is differentiated into alternately placed fertile cells (spore mother cells) and sterile cells (which form pseudo-elaters). A part of sporogenous tissue, upper to this region shows tetrahedral tetrads of spores and many pseudo-elaters. In the upper most region of the capsule are present the separated spores and pseudo-elaters.
9. The wall of the capsule is 4-6 layered.
10. The outermost layer forms a well defined epidermis, ventilated with stomata.
11. The cells of the layers beneath the epidermis possess generally two chloroplasts each and these chloroplasts make the sporophyte partially self-sufficient with regard to food.
12. Each spore is uninucleate and has a thick ornamented exospore and a thin endospore.
13. Pseudo-elaters consist of 2-3 cells, joined end to end, in a simple or branched structure. (These do not possess characteristic spiral thickenings of true elaters and, therefore, are called pseudo-elaters).

Identification

Division—**Bryophyta**. (1) True roots absent and instead are present the rhizoids, (2) No vascular strand.

Class—**Anthocerotopsida**. (1) Thalloid, (2) Rhizoids without septa, (3) Each cell of the thallus has generally a single large chloroplast, with pyrenoid.

Order—**Anthocerotales**. (1) thallus homogeneous, (2) Only smooth walled rhizoids present, scales and tuberculated rhizoids absent.

Family—**Anthocerotaceae**. (1) Sporophyte indeterminate in growth, (2) Presence of meristematic zone, (3) Capsule with central columella.

Genus—***Anthoceros***. (1) Capsule partly covered with involucre, only at the base, (2) Capsule wall ventilated, (3) *Nostoc* colonies inside the thallus.

Hints for Collection

Anthoceros is common in both hills and plains. Plants grow, as a rule, in moist, shady places on the sides of ditches or in moist hollows among rocks.

Sphagnum
(Peat Moss or Bog Moss)

Classification

Division	—	**Bryophyta**
Class	—	**Bryopsida**
Sub-class	—	**Sphagnidae**
Order	—	**Sphagnales**
Family	—	**Sphagnaceae**
Genus	—	***Sphagnum***

Exercise 1

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

Work procedure

Study the branching, the branches and arrangement of leaves on the axis.

Comments

1. The plants are aquatic, growing about the margins of small lakes and ponds or growing on dripping rocky banks.
2. The pH of water in which *Sphagnum* grows ranges from 3.7 to 4.9. Since this water accumulates year after year to form peat and hence the name peat moss.
3. The size of the plant varies from a few inches to a maximum of 7 inches.
4. The plant is erect, branched and differentiated into stem and the leaves. The colourless rhizoids are formed at the base but soon disappear. Hence, there are no rhizoids on mature gametophores.
5. At the apex of the gametophore there are a number of short branches densely crowded in a cluster, called coma.
6. In the posterior part of the stem, the branches arise in tufts in the axil of every fourth leaf and in each tuft there are 3-8 branches.
7. These branches are of two types—(i) diverging branches which are stout, short, growing

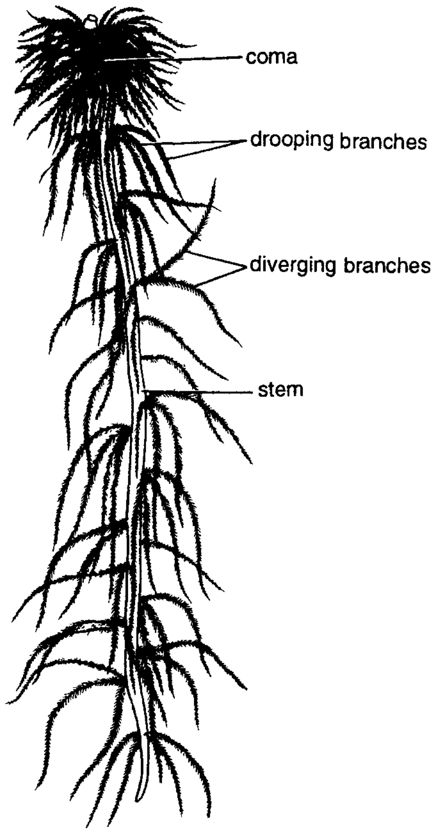


Fig. 1. ***Sphagnum.*** A part of gametophore.

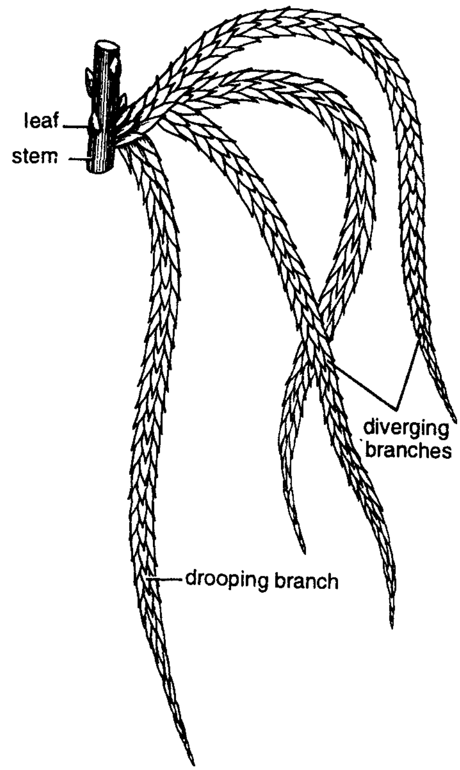


Fig. 2. ***Sphagnum.*** Portion of a plant showing tuft of branches.

outward and upward and (ii) drooping or flagelliform branches which hang downward, around and close to main stem. These are absent from submerged forms.

8. At intervals, one of the branches in the tuft grows and forms an apical cluster of branches like the main stem. This is called an innovation. It helps in vegetative propagation by separating from the main branch.
9. When first formed, the leaves are in three vertical rows or three ranked. Later the arrangement changes to 2/5.
10. The leaves lack a midrib an exception to mosses.

Exercise 2

Object **: Study of anatomy of axis.**

Work procedure

Cut a T.s. of the axis, stain in safranin and fast green, mount in glycerine and study.

Comments

1. The outermost layer is cortex, made of compactly arranged cells.
2. In some species, as *S. subsecundum,* it remains one cell thick throughout its life but in some others, it becomes 3-5 cells thick.
3. At maturity these cortical cells lack protoplast, become hyaline and dead.
4. In some species viz. *S. cymbifolium,* they develop spiral thickenings and develop even the pores. These cortical cells contain air and water.
5. In some species as *S. molluscum* and *S. tenellum,* some of the cortical cells become greatly enlarged and become flask shaped. The cells also accumulate water and are known as retort cells. Such type of cortex is also known as hyalodermis.
6. The cells inner to cortex are prosenchymatous. These give mechanical support to the stem. The region is also known as hadrome.
7. The innermost region is known as medulla. The cells are colourless, parenchymatous and somewhat vertically elongated.

Exercise 3

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

Work procedure

Take out a few leaves, stain in safranin, mount in glycerine and study.

Comments

1. The mature leaf is sessile, entire, acute and one-celled thick.
2. The leaf lacks a midrib and the surface view shows two kinds of cells (i) the narrow, chlorophyll containing assimilatory cells, and (ii) the large, dead, rhomboidal hyaline cells with spiral thickenings and pores.
3. The spiral thickenings provide mechanical support and keep the hyaline cells from collapsing when they are empty.
4. The pores help in rapid intake of water.
5. The green assimilatory cells of the leaf are joined together and form a network with sinuous walls.
6. At the base of very young leaf there are one or more glandular hairs but these disappear as the leaf matures.

Exercise 4

***Object* : Study of leaf anatomy.**

Work procedure

Cut a T.s. of the leaf stain in safranin, mount in glycerine and study.

Comments

1. In a cross section, the leaf appears like a beaded structure with hyaline and chlorophyllous cells alternating with each other.
2. The chlorophyllous cells are triangular and the base of triangle is towards the upper side e.g. *S. acutifolium*. In *S. tenellum* the condition is just reverse i.e. the base of the triangle is towards the lowerside. In *S. squarrosum* the chlorophyllous cells are not triangular but appear spindle-shaped and are hemmed in, above and below, by the hyaline cells.

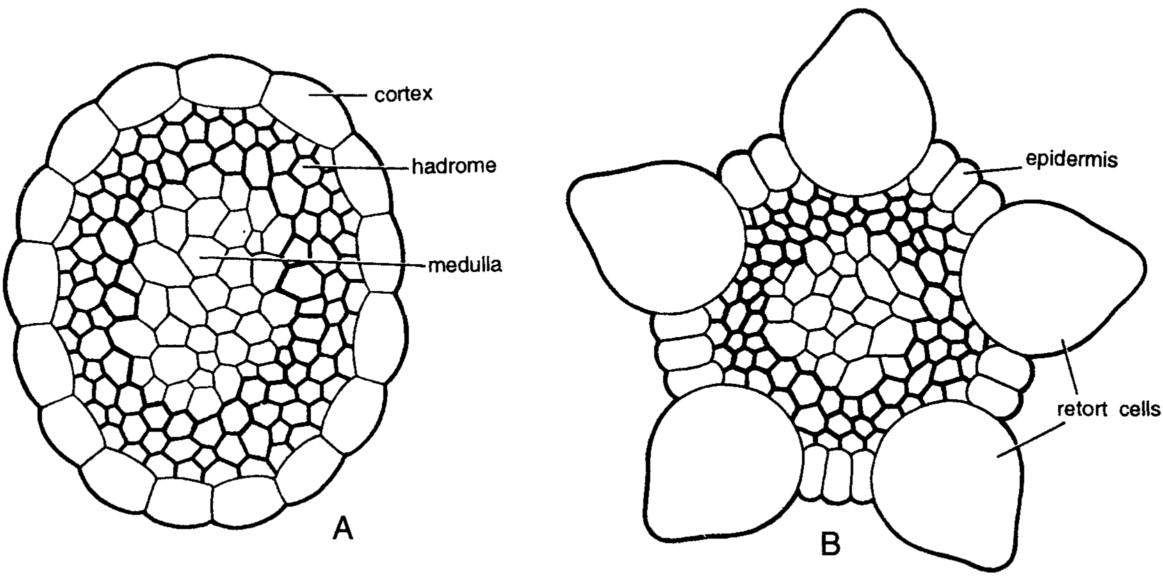


Fig. 3. ***Sphagnum*. A.** T.s. stem in most species. **B.** T.s. of stem of *S. molluscum*.

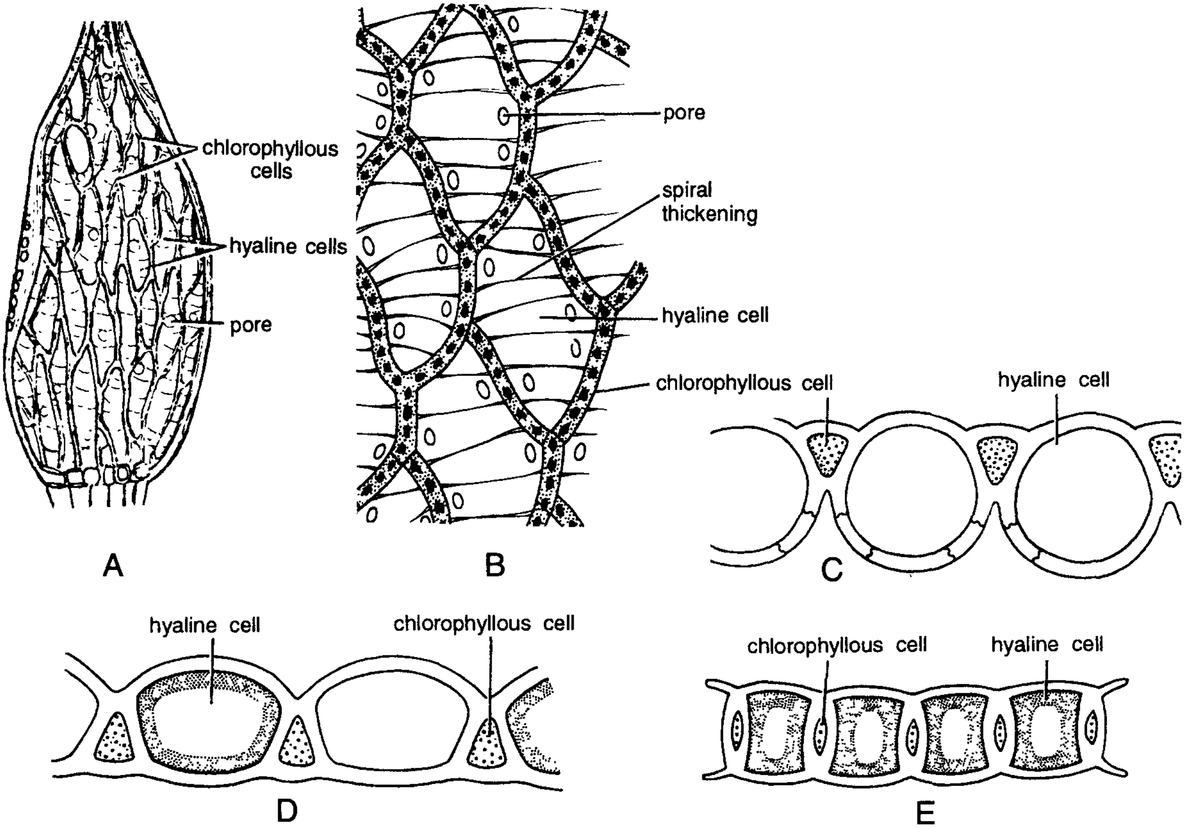


Fig. 4. ***Sphagnum.*** **A.** Leaf in surface view. **B.** A part of leaf surface magnified, **C,D,E.** Portions of transverse sections of leaves of **C.** *S. acutifolium*, **D.** *S. tenellum*, **E.** *S. squarrosum*.

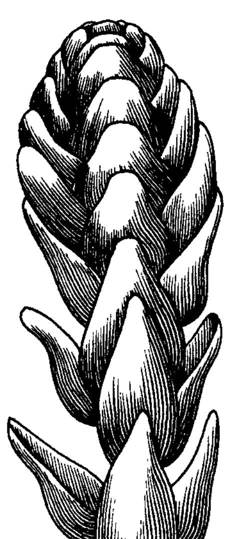


Fig. 5. ***Sphagnum.*** External features of antheridial branch.

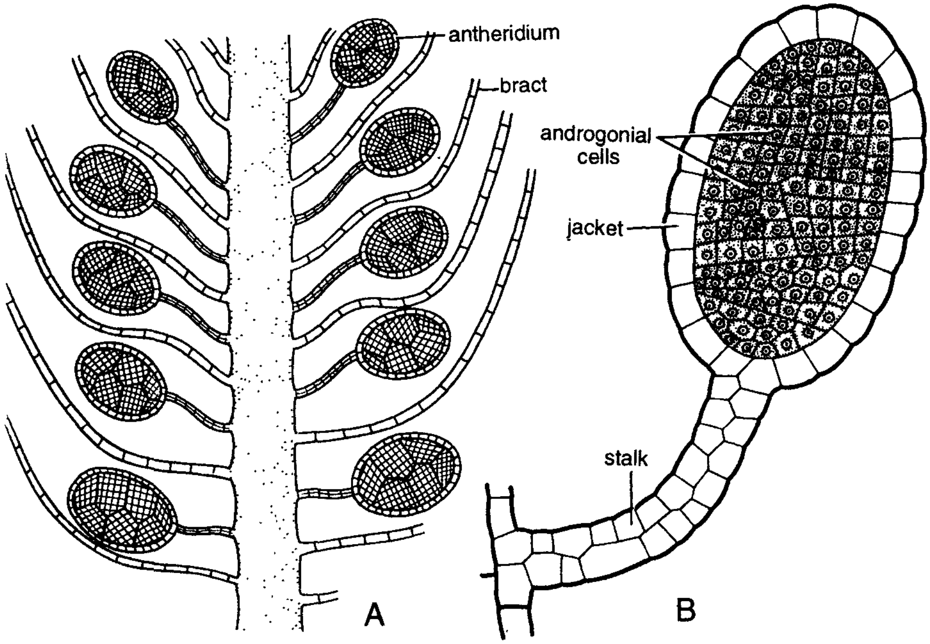


Fig. 6. ***Sphagnum.*** **A.** Portion of antheridial branch to show position of antheridia. **B.** Single antheridium.

Exercise 5

Object **: Study of antheridial branch and antheridium.**

Work procedure

Select antheridial branch by its catkin-like appearance at the tip of the branch. Tease out antheridia or else cut a longitudinal section, stain in safranin and mount in glycerine to study.

Comments

1. The plants are either monoecious or dioecious and both antheridial and archegonial branches occur at the apices of the gametophores.
2. The catkin-like antheridial branches are either red, brown or yellow in colour with club-shaped appearance.
3. The antheridia present in the axil of leaf are arranged acropetally.
4. The mature antheridium has a long stalk, 2 cells in breadth and a globular body.
5. The body of the antheridium consists of single-layered jacket surrounding number of androgonial cells.
6. The androgonial cells ultimately form the biciliate antherozoids.

Exercise 6

Object **: Study of archegonial branch and archegonium.**

Work procedure

Select archegonial head, tease out to observe archegonia or cut a longitudinal section, stain in fast green and mount in glycerine to study.

Comments

1. The archegonial branches are purple coloured. These bear large leaves known as perichaetial leaves.
2. Each archegonial branch has usually 3 archegonia (sometimes 1 or 5 also). Paraphyses are absent.
3. The position of the archegonium is acrogynous i.e. the first archegonium is formed by the apical cell and is known as the primary archegonium, whereas the other two archegonia are developed from the last two segments cut off by the apical cell and are known as secondary archegonia.
4. The structure of both types of archegonia is similar. A mature archegonium is a stalked structure with a broad venter and a long twisted neck.

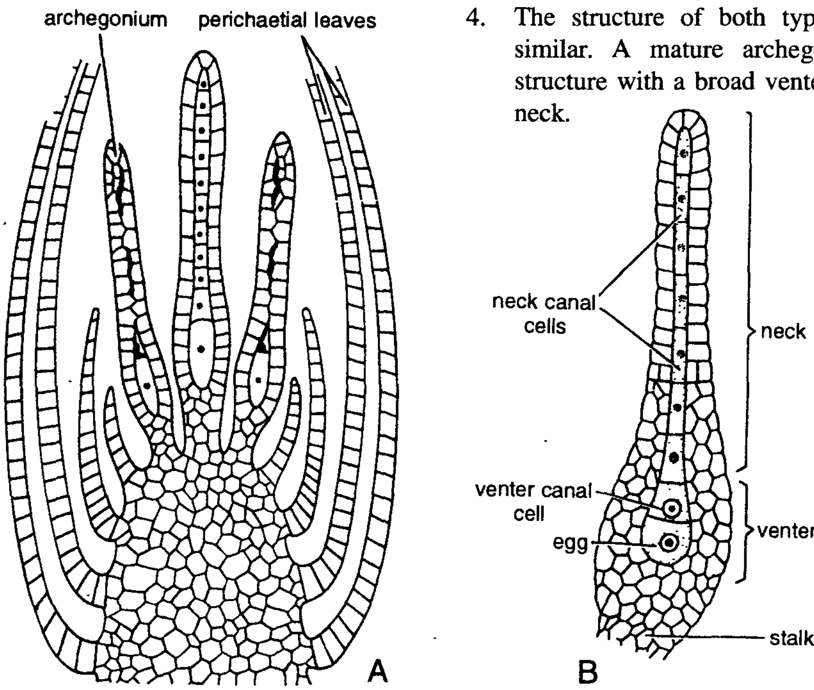


Fig. 7. *Sphagnum*. A. L.s. archegonial branch, **B.** A single archegonium.

5. The venter as well as lower portion of neck are 2-4 cells in thickness.
6. The cover cells are not distinct and in the axial row are found 8-9 neck canal cells, one venter canal cell and one egg cell.

Exercise 7

Object **: Study of sporophyte.**

Work procedure

Study the external features of the sporophyte. Cut longitudinal section, stain with safranin and fast green, mount in glycerine and study.

Comments

1. It consists of a large bulbous foot and a nearly spherical, black coloured capsule. Seta is absent and occurs as a small constriction between foot and capsule.
2. At maturity the apex of the archegonial branch elongates to form stalk-like portion called pseudopodium.
3. The calyptra covers the upper part of the mature capsule.
4. Longitudinal section of the sporophyte shows following characters.
5. Sporophyte consists of a bulbous foot and a spherical capsule connected by a neck-like suppressed seta.
6. The jacket of the capsule is 4-6 layered. The outermost layer is differentiated into an epidermis with rudimentary, non-functional stomata.
7. All cells of the capsule wall contain chloroplasts, hence the sporophyte is not completely dependent on the gametophyte.
8. At the top of th᾿ capsule there is a convex operculum separated from the remainder of the thallus by a thin-walled transverse ring of cells called the annulus.
9. Over the operculum is the calyptra which arises from the venter of the archegonium and completely covers the sporophyte.
10. The central region of the capsule is occupied by the columella. It is overarched by dome-shaped spore sac.

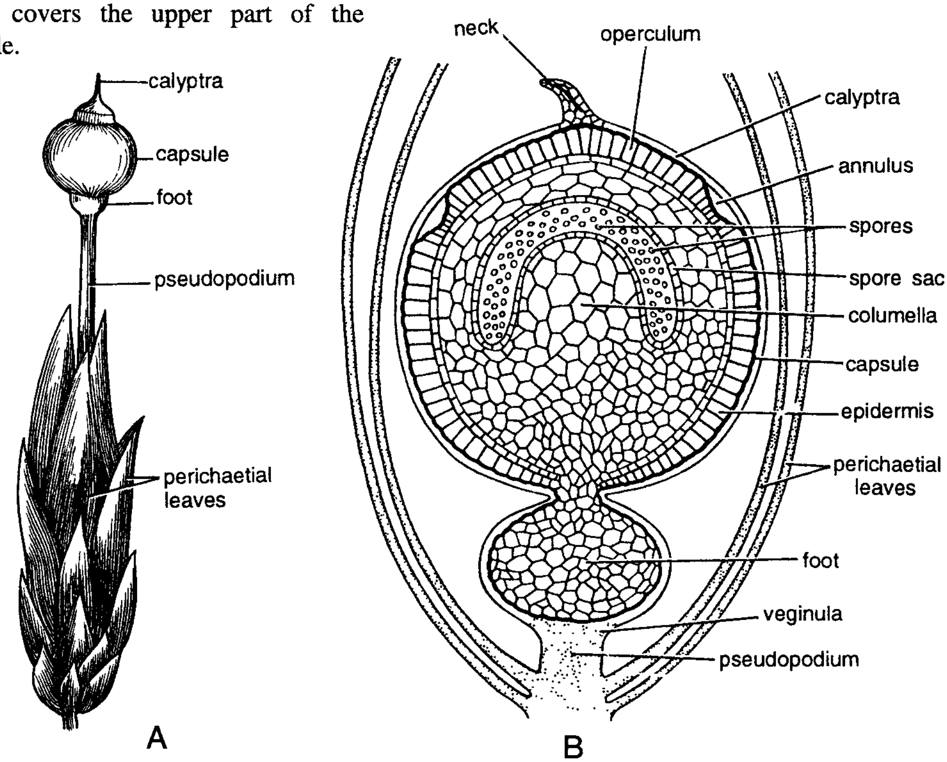


Fig. 8. ***Sphagnum.*** **A.** External features of sporophyte. **B.** L.s. sporophyte.

11. The spore sac is filled only with spores, elaters being altogether absent. Each spore has an outer exine and inner intine.
12. A mature saprophyte is situated at the top of an elongated archegonial branch, the pseudopodium.
13. The apex of the pseudopodium is enlarged which together with the basal portion of calyptra is known as veginula.
14. Veginula is a sac-like structure into which the foot of the sporophyte is embedded.
15. When the spores mature, the operculum breaks away at the place of annulus thus releasing the spores.

Exercise 8

Object **: Study of protonema.**

Work procedure

Collect the protonema from the soil or allow the spore to germinate into protonema. Stain in fast green and mount in glycerine.

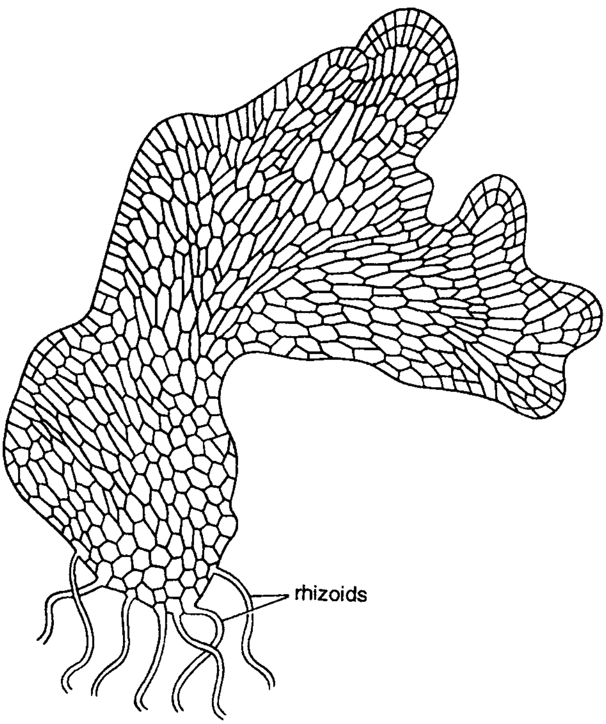


Fig. 9. ***Sphagnum.*** Thalloid lobed protonema.

Comments

1. The spore germinates to form a thalloid protonema.
2. At the posterior end of the thalloid protonema are borne the rhizoids which are multicellular with oblique septa.
3. Secondary protonema may arise from the primary protonema
4. Later, at the base of the protonema a single bud is differentiated with a 3-sided apical cell. This bud develops into the leafy gametophore. Thus, only one gametophore is developed from one protonema.

Identification

Division—**Bryophyta**. (1) True roots absent and instead are present the rhizoids, (2) No true vascular strand.

Class—**Bryopsida**. (1) Gametophore erect and leafy, (2) Rhizoids multicellular with oblique septa.

Sub-class—**Sphagnidae**. (1) Protonema thallose, (2) Archesporium from amphithecium, (3) Seta is very small but the capsule is elevated by the elongation of the gametophore apex known as pseudopodium, (4) Peristome teeth absent.

Order—**Sphagnales**. Character same as sub-class.

Family—**Sphagnaceae**. Characters same as those of Sphagnidae.

Genus—***Sphagnum***. Characters same as Sphagnidae.

Hints for Collection

All the 11 species of *Sphagnum* found in India are restricted to the Himalayas. The plants are aquatic or semi-aquatic forming dense cushions commonly seen in swamps, ponds and lake margins, moorelands and wet hill sides.

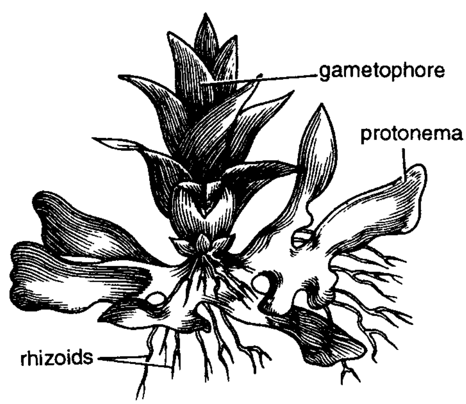


Fig 10. ***Sphagnum.*** Mature protonema producing a leafy gametophore.

Funaria (Cord Moss)

Classification

Division	—	**Bryophyta**
Class	—	**Bryopsida**
Sub-class	—	**Bryidae**
Order	—	**Funariales**
Family	—	**Funariaceae**
Genus	—	***Funaria***

Exercise 1

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

Work procedure

Observe the branching, arrangement of leaves, midrib of the leaf, branched and multicellular rhizoids. Pick up a few rhizoids, stain in safranin and observe oblique septa.

Comments

1. The gametophyte shows a prostrate underground protonema and an erect leafy gametophore.
2. The gametophore that arises from protonema is differentiated into (i) rhizoids, (ii) axis or 'stem' and (iii) leaves.
3. Many rhizoids are present at the base. These are slender, branched, and multicellular. The septa are oblique.
4. Young rhizoids are colourless while mature are coloured brown. They also develop chloroplast and become green if exposed to sunlight.
5. The axis is erect and branched. It is 1-3 cms high. The branches arise below a leaf and are thus extra-axillary.
6. The stem and branches are covered with small, simple, sessile and spirally arranged leaves with 3/8 phyllotaxy.
7. The leaves at the apex of the gametophore are crowded to form a bud-like head.
8. Each leaf is nearly ovate in shape and bears a clear midrib except when young.
9. Sex organs are borne at the apices of the axis.

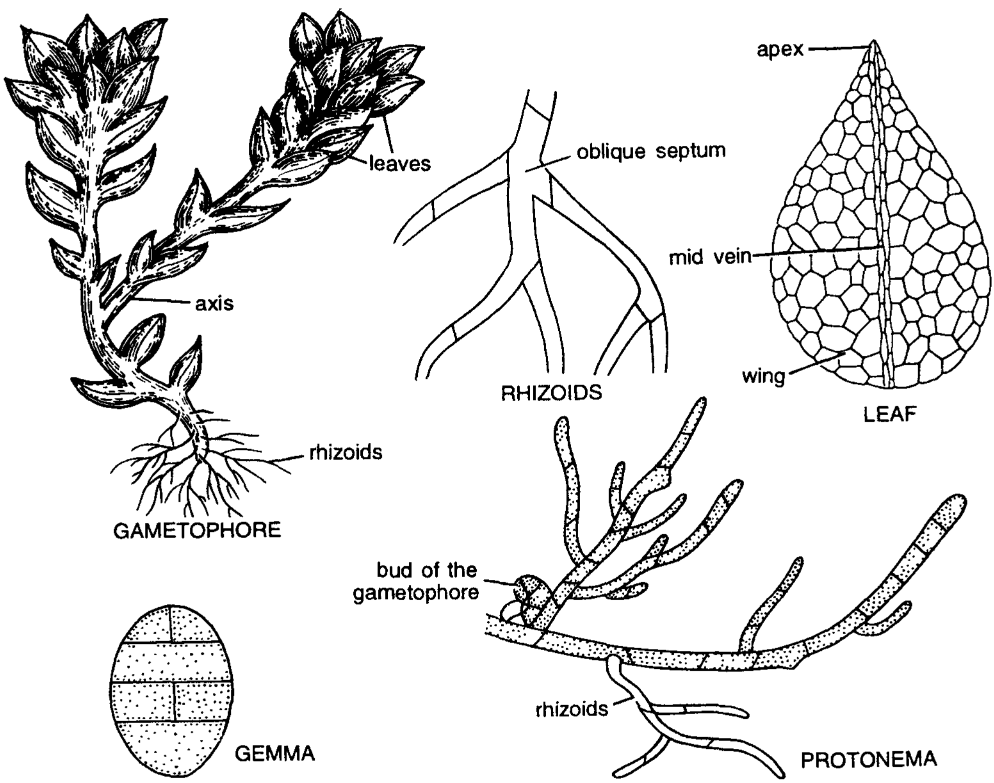


Fig. 1. ***Funaria.*** External features of the gametophyte : gametophore, rhiozoids, leaf, gemma and protonema.

Exercise 2

Object : **Study of anatomy of the axis.**

Work procedure

Place gametophore in the pith and cut T.s. with a sharp razor or blade. Stain in safranin or fast green, mount in glycerine and study.

Comments

1. The transverse section shows an almost circular outline.
2. It is differentiated into (i) an epidermis, (ii) cortex and (iii) the central cylinder.
3. The single-layered epidermis, with tangentially elongated cells has chloroplasts and bounds the underlying cortex.
4. The multilayered cortex surrounds central cylinder. Peripheral cells of the mature cortex are slightly thick walled than the rest.
5. Near the periphery of the cortex, small leaf traces with blind ends are present.
6. The outer cells of the cortex sometimes contain the chloroplasts.
7. The central cylinder is present in the centre. The cells are vertically elongated, smaller and the walls are slightly thickened. The cells are dead due to lack of protoplasm. The cylinder

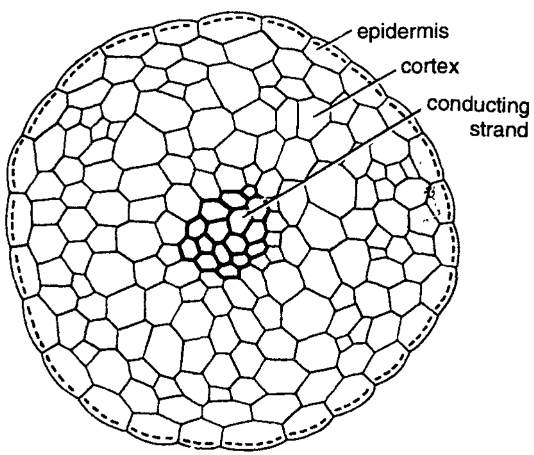


Fig. 2. ***Funaria.*** T.s. axis (cellular).

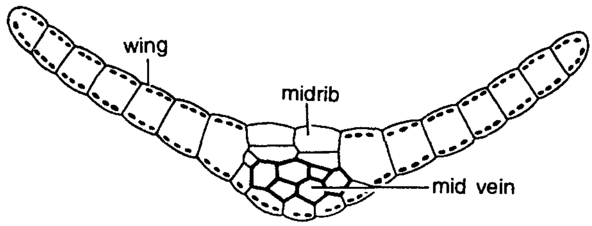


Fig. 3. ***Funaria.*** T.s. leaf (cellular).

takes part in the conduction of water and food materials.

Exercise 3

Object : **Study of anatomy of leaf.**

Work procedure

Cut a T.s. of leaf, stain in safranin or fast green, mount in glycerine and note down the observations.

Comments

1. The leaf consists of a single layer of cells containing chloroplasts except in the middle where it forms a distinct midrib.
2. The centre of the midrib is occupied by a small strand of narrow and slightly thick-walled cells.

Exercise 4

Object : **Study of gemmae.**

Work procedure

Shake leaves or brush off basal part of axis on the slide for collecting gemmae. Mount in glycerine and study.

Comments

1. These are vegetative reproductive structures.
2. Multicellular and green gemmae are produced on stem and leaves. On detachment, these germinate to give rise to new plants.
3. Gemmae, when grow on rhizoids, become brown in colour and are then known as bulbils.
4. Each gemma is composed of 8-12 cells. It is transversely and vertically septate.

Exercise 5

***Object* : Study of antheridial branch and antheridium.**

Work procedure

Tease a few tips of branches. Remove the cluster of leaves to bring out antheridia. Stain in safranin, mount in glycerine and study.

Comments

1. The sex organs are present at the apices of branches. These are enclosed by a group of leaves at the apex.
2. At the tip of the stem, is an antheridial branch or 'male flower'—a cluster of antheridia.
3. Intermingled with antheridia are multicellular capitate hairs, known as paraphyses.
4. Both antheridia and paraphyses are surrounded by large leaves, known as perichaetial leaves.
5. In the antheridial branch antheridia in various stages of development occur together.
6. The mature antheridium consists of massive stalk and a club-shaped body.
7. The body has a single layered outer jacket, the cells of which contain chloroplasts.
8. At the apex of the jacket is an operculum, which helps in liberation of antherozoids.
9. A dense central mass of androcytes lies within the jacket.

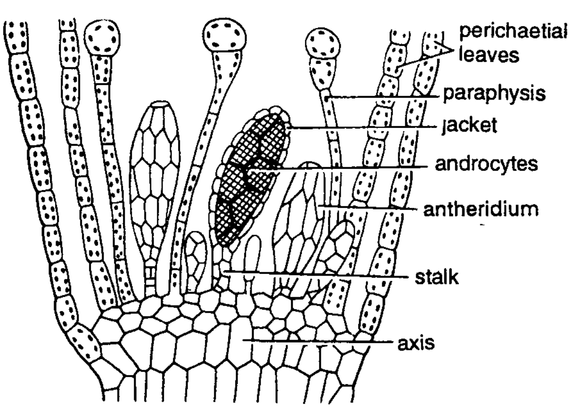


Fig. 4. ***Funaria.*** L.s. apex of the male branch showing antheridia.

Exercise 6

***Object* : Study of archegonial branch and archegonium.**

Work procedure

Tease the apex of the female branch to remove the leaves. Stain in safranin, mount in glycerine and study.

Comments

1. The sex organs are situated at the apices of branches inside the cluster of leaves.
2. The archegonia also arise in clusters at the apex of the archegonial branch.
3. Intermingled with archegonia are paraphyses.
4. The archegonia and paraphyses are surrounded by closely folding, unmodified leaves.
5. All the archegonia of this cluster are almost of the same age and developmental stage.
6. The nearly mature archegonium is a multicellular, stalked structure, with a broad venter and narrow twisted neck.
7. The wall of the venter is double layered. The neck consists of six longitudinal rows of cells surrounding a central canal.
8. In the neck there are six or more neck canal cells and the venter has one venter canal cell and one egg cell.

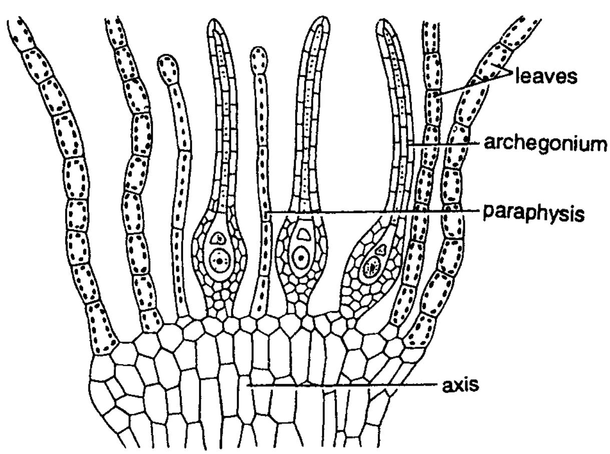


Fig. 5. ***Funaria.*** L.s. apex of the female branch showing archegonia.

Exercise 7

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

Work procedure

Select out a plant that has a sporophyte. Place it on the stage of a dissecting microscope and study.

Comments

1. A gametophyte shows a sporophyte attached to it.
2. The sporophyte is developed at the apex of the archegonial branch.
3. A mature sporophyte shows three parts (i) foot, (ii) seta and (iii) capsule.
4. Foot is poorly developed and is embedded in the apex of the archegonial branch.
5. Seta is long, slender and twisted. It bears a capsule at the top.
6. The capsule is slightly oblique and pear-shaped. Calyptra covers the apex of capsule.

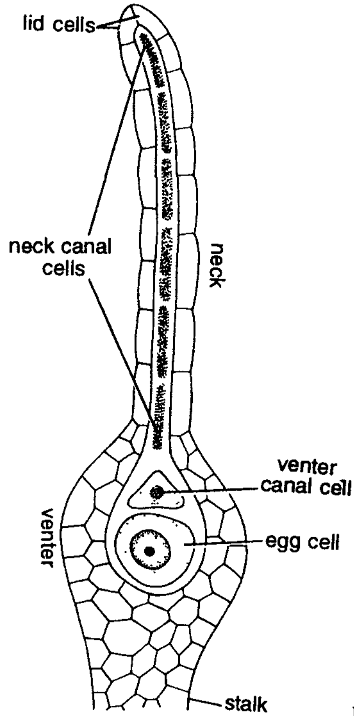


Fig. 6. ***Funaria.*** An archegonium.

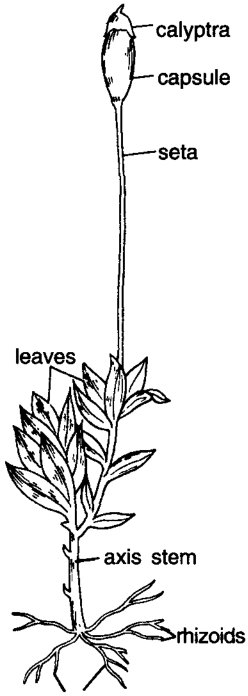


Fig. 7. ***Funaria.*** A gametophyte with sporophyte.

Exercise 8

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

Work procedure

Cut a T.s. of the capsule and stain in safranin and fast green; mount in glycerine and study.

Comments

1. L.s. of the capsule can be divided into three regions—(i) apophysis, (ii) theca proper and (iii) upper region.
2. Apophysis is the basal region. In its centre is a conducting stand in continuation with that of seta.
3. Around the conducting strand are few layers of cells with intercellular spaces and chloroplast. The epidermis in this region is ventilated (stomata present).
4. The theca proper is the fertile region. It has a central columella, the upper part of which is cone-shaped, projecting into the concavity of the operculum. On the basal end, it is

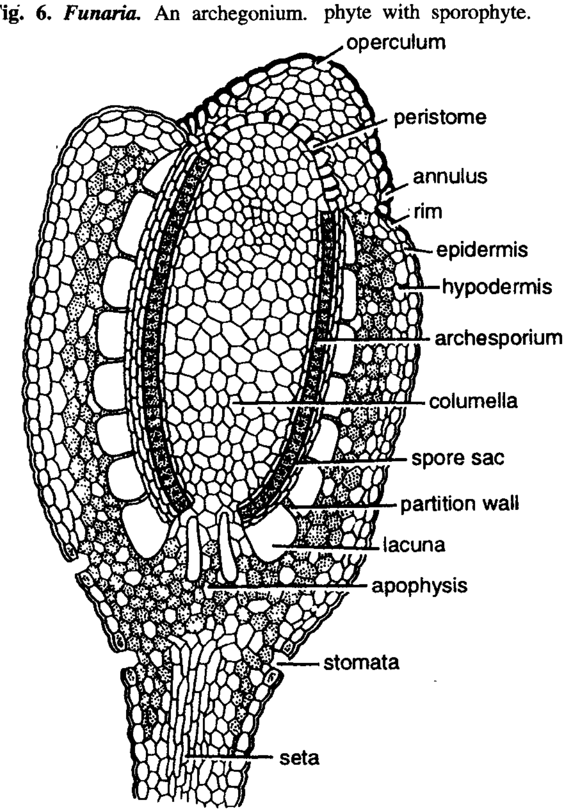


Fig. 8. ***Funaria.*** L.s. capsule.

connected with the central tissue of the apophysis.
5. Around the columella is a U-shaped spore sac, broken at the base, thus separating the two arms of U.
6. Spore sac has an outer wall of 3-4 layers of cells and an inner of one layer. Between these, only spores are present, elaters being absent.
7. Each spore has an inner hyaline endosporium and a coloured, almost smooth exosporium.
8. Inside the endosporium is the cytoplasm, with a nucleus, oil globules and chloroplasts.
9. Outside the spore sac, is an air space that is divided into many air cavities by green filaments which run from the external tissue of the wall to the outer wall of the spore sac.
10. The wall of the capsule is many layered. Two to three inner wall layers of the capsule in theca region are green and show intercellular spaces while outermost 2-3 layers just beneath the epidermis are compact parenchymatous and colourless.
11. The upper region consists of operculum and peristome. It is marked off by a conspicuous constriction, immediately below which is a rim and above the annulus.
12. Calyptra covers the capsule. The peristome teeth encircle the operculum.

Exercise 9

***Object* : Study of peristome.**

Work procedure

Keep a mature capsule on the stage of a dissecting microscope, remove the operculum by needles while viewing through the lens, yellow-black or yellow-brown coloured ring of peristomial teeth would be seen; mount in glycerine and study.

Comments

1. The peristome consists of 2 rows of curved triangular plate-like teeth. Each row has 16 teeth.
2. Outer peristomial teeth are ornamented with thick transverse bands and are spirally twisted to the left.

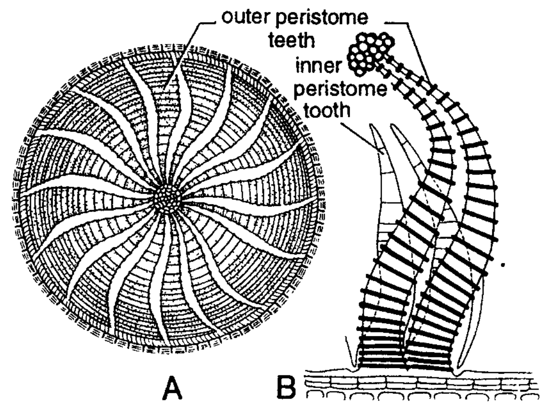


Fig. 9. *Funaria*. Outer and inner peristome.

3. Inner peristomial teeth are colourless, shorter and comparatively more delicate.
4. The bases of inner peristome teeth are directly covered by the teeth of the outer peristome, but as they move away from the base, they curve, thus narrowing the slits between outer peristome teeth.
5. Hygroscopic movements in the outer peristome teeth assist in liberation of spores from capsule.

Identification

Division—**Bryophyta**. (1) True roots absent, instead are present the rhizoids, (2) No vascular strand.

Class—**Bryopsida**. (1) Gametophore erect and leafy, (2) Rhizoids multicellular with oblique septa.

Sub-class—**Bryidae**. (1) Leaves with distinct midrib, (2) Seta long, (3) Spore sac usually separated from the capsule wall by air space.

Order—**Funariales**. (1) Leaves ovate or spathulate, (2) Peristome usually double, (3) Calyptra usually distended.

Family—**Funariaceae**. (1) Calyptra has a long beak, (2) Capsule pyriform and somewhat dropping.

Genus—***Funaria***. (1) Leaves arranged spirally and 3/8 phyllotaxy, (2) Stem internally distinguished into an epidermis, cortex and conducting strand, (3) Leaves crowded at the apex to form a bud-like head.

Hints for Collection

Funaria is very common in hills as well as in plains. It is generally found growing on moist walls and tree trunks, during rainy season.

Polytrichum
(Pigeon Wheat Moss)

Classification

Division	—	**Bryophyta**
Class	—	**Bryopsida**
Sub-class	—	**Bryidae**
Order	—	**Polytrichales**
Family	—	**Polytrichaceae**
Genus	—	***Polytrichum***

Exercise 1

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

Work procedure

Take a gametophore, observe the branching, arrangement of leaves and the rhizoids. Pick up a few rhizoids, stain in safranin and observe oblique septa.

Comments

1. The gametophyte is differentiated into an underground rhizome and aerial, erect, leafy stems 20 cms or more tall. In *P. commune*, transitional zone between rhizome and upper leafy shoot is present.
2. Rhizoids are produced by the rhizome. These are long and thick walled with oblique septa. The rhizoids coil round one another to form a rope like structure. These rhizoidal strands provide mechanical support also.
3. The leaves on the rhizome and middle transitional region occur in 3 vertical rows. These are either brown in colour or colurless. The leaves on aerial leafy shoot are green, large and spirally arranged.
4. Each leaf possesses a broad, colourless, membranous, one celled sheathing leaf base that narrows above into a lanceolate limb. The margins of the wings are coarsely toothed. The leaf has a dark green midrib.

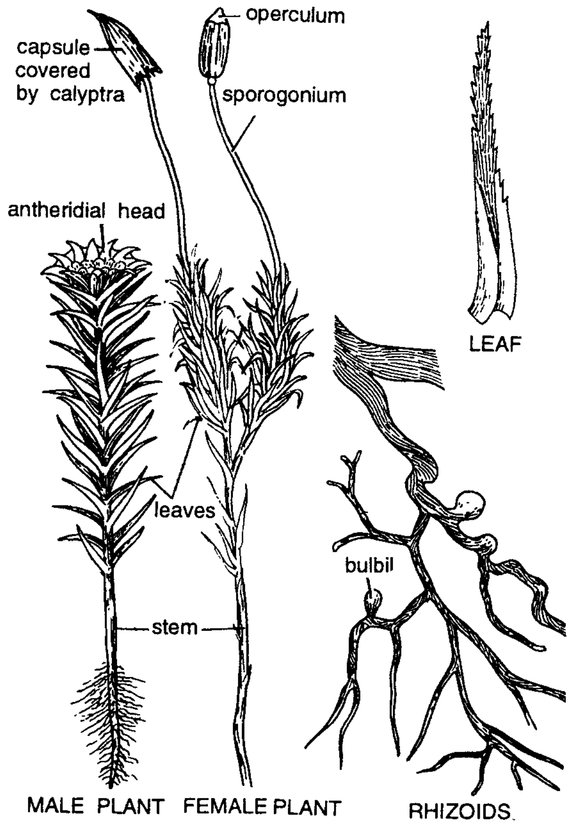


Fig. 1. *Polytrichum*. External features of gametophore : male and female plant, leaf and rhizoids.

Exercise 2

***Object* : Study of anatomy of rhizome.**

Work procedure

Place rhizome in the pith and cut T.s. with a sharp razor or blade. Stain in safranin or fast green, mount in glycerine and study.

Comments

1. The transverse section shows an almost circular outline.
2. It is differentiated into piliferous layer, cortex, endodermis and the central cylinder.

3. A piliferous layer is the outermost. It bears rhizoids.
4. This is followed by two or three layers of cortical parenchyma. It is interrupted by three hypodermal strands, the cells of which are distinctly prosenchymatous with pointed ends.
5. Passing radially inwards from the hypodermal strands are cells of greater diameter, which do not show a clear demarcation with the cells of hypodermal strands. These are called the radial strands.
6. The cortex is delimited from the conducting strand by large, radially elongated endodermal cells. The endodermis is discontinuous and consists of three arcs separated by radial strands.
7. The central cylinder is trilobed. The central mass consists mainly of very thick-walled, elongated living cells (with oblique end walls), known as stereids. The stereids are collectively called as stereom.
8. Scattered among the stereids are the hydroids which serve for the conduction. The hydroids are of about the same diameter as the stereids, or slightly bigger and as a whole being called as hydrome.
9. Surrounding the trilobed central strand is an interrupted pericycle composed of 2 or 3 layers.
10. The furrows between the lobes are occupied by 6-8 polygonal cells known as leptoids, collectively called as leptom. These cells appear similar to sieve tubes.
11. In between the leptom and hydrome is a layer of starchy parenchyma called as amylom.

Exercise 3

***Object* : Study of anatomy of axis.**

Work procedure

Place a piece of axis in the pith and cut T.s. with a sharp razor or blade. Stain in safranin or fast green mount in glycerine and study.

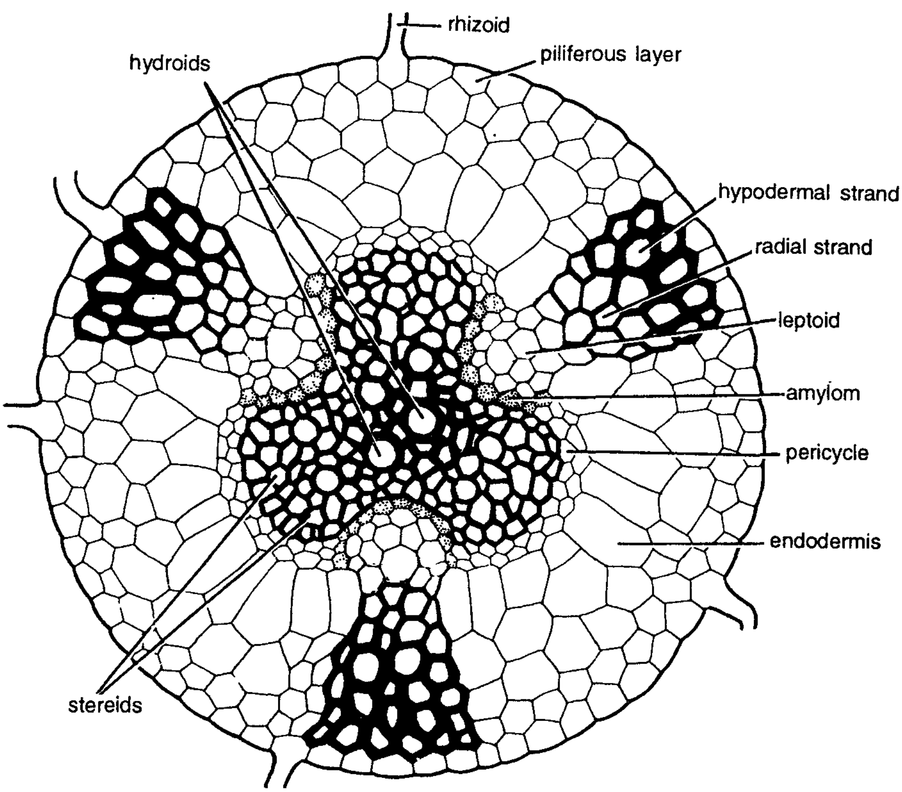


Fig. 2. ***Polytrichum.*** T.s. rhizome (cellular).

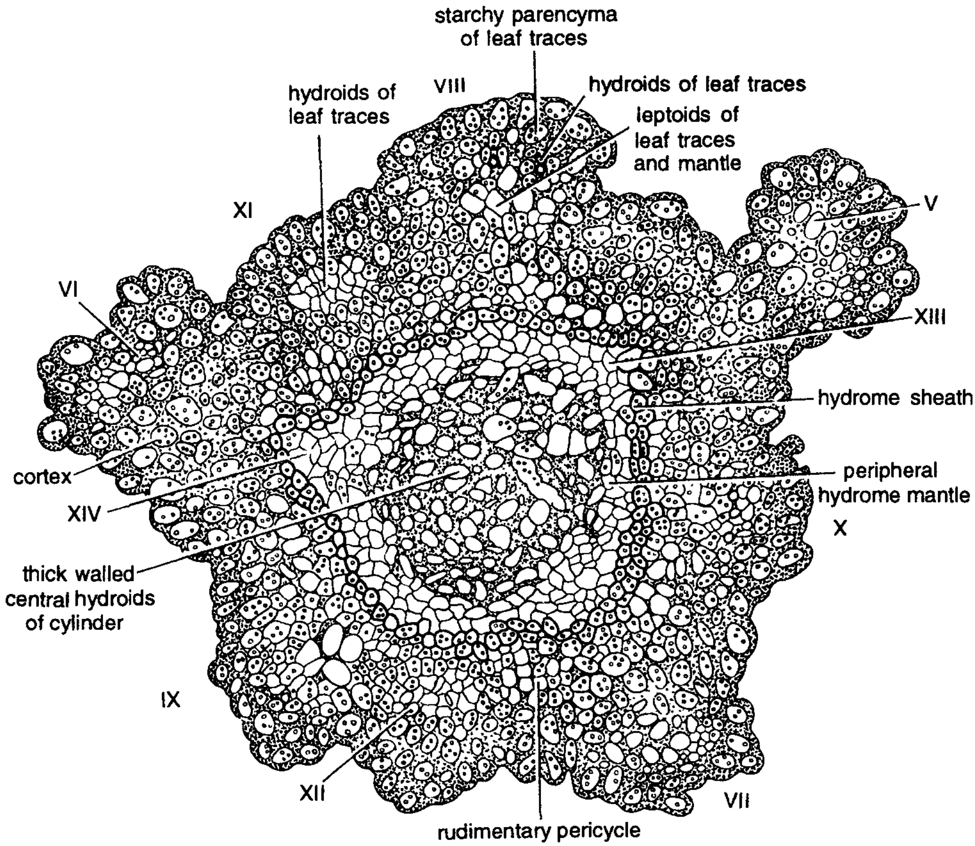


Fig. 3. ***Polytrichum.*** T.s. axis (cellular)

Comments

1. The outline of the section is irregular due to attachment of leaves.
2. The tissues show outermost superficial layer followed by cortex, pericycle, leptom mantle, hydrom sheath, hydrom mantle and the hydrom cylinder.
3. The superficial layer does not form clearly defined epidermis.
4. The cortex is divisible into outer and inner cortex. The outer cortex is made of compact, elongated prosenchymatous cells gradually merging into the inner cortex made of parenchymatous cells. Leaf traces are quite common in cortex.
5. Following the cortex is present the rudimentary pericycle which is not clearly differentiated.
6. Inner to pericycle is the leptom mantle, the cells of which are typical sieve tube-like. The leptom mantle is regarded as equivalent to the phloem of vascular plants.
7. Internal to the leptom mantle is the hydrom sheath (amylom layer) composed of one or two layers of cells with prominent starch.
8. Immediately following the hydrom sheath is the hydrom mantle which is composed of thin-walled cells without contents.
9. The centre of axis is occupied by the hydrom cylinder, made of thick walled cells.

Exercise 4

Object **: Study of anatomy of leaf.**

Work procedure

Place the leaf in pith, cut T.s. with a sharp blade or razor. Stain in safranin or fast green, mount in glycerine and study.

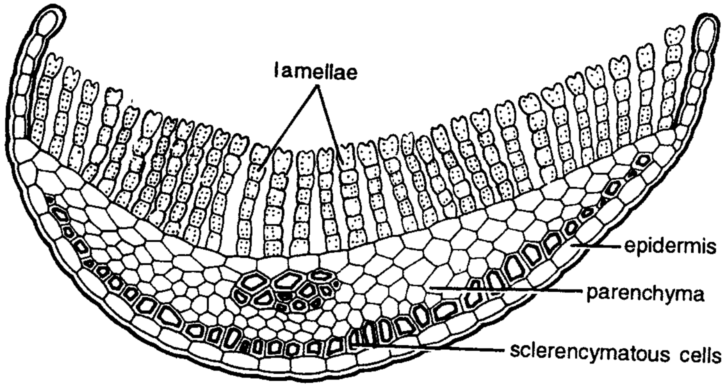


Fig. 4. *Polytrichum*. T.s. leaf (cellular).

Comments

1. The section shows several celled thick midrib that gradually merges into rudimentary wings which are made of hyaline cells.
2. On the lower side is the cuticularized epidermis. Just inside the epidermis are one or two layers of small, sclerenchymatous elongated cells.
3. The central tissue of the leaf is made up of large parenchymatous cells with some groups of sclerenchymatous cells scattered between them.
4. On the upper surface there is a layer of large cells from which arise many parallel plates known as lamellae.
5. Each lamella is uniseriate and is composed of 5-8 cells. Each cell contains chlorophyll. The terminal cell of each lamella is wider or papillose. Terminal cells of the adjacent lamellae almost touch each other.
6. These lamellae are the chief photosynthetic tissue of the leaf and compensate for the reduced wing.

Exercise 5

***Object* : Study of antheridial head and antheridium.**

Work procedure

Select a male plant, select antheridial head that is situated at the tip, tease out antheridia or cut L.s. of head, stain in safranin, mount in glycerine and study.

Comments

1. Plants are usually dioecious and the antheridia and archegonia are present at the apices of the gametophores.
2. The antheridia are surrounded by specialized leaves known as perichaetial leaves which are usually short and may be pale pink or rose.

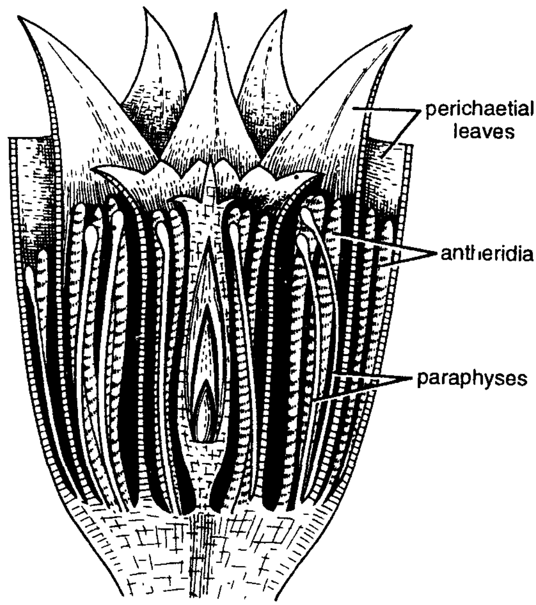


Fig. 5. *Polytrichum*. L.s. through antheridial head.

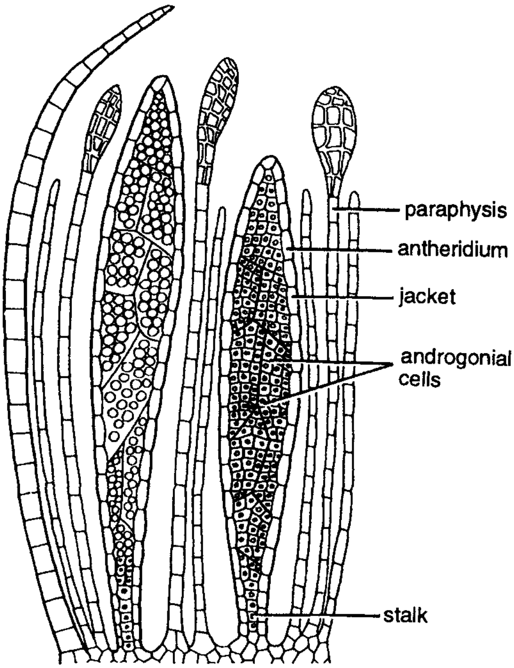


Fig. 6. ***Polytrichum.*** Antheridia and paraphyses.

They form a cluster or rosette, superficially resembling a small flower.

3. The antheridia are present in groups at the base of each perichaetial leaf in the position of lateral buds.
4. Intermingled with the antheridia are the paraphyses. Some of the paraphyses are filamentous, whereas the others are broadened at their tips.
5. A mature antheridium is usually stalked and somewhat club-shaped structure.
6. It consists of a jacket of cells surrounding a mass of androgonial cells.

(Since the apical cell is not consumed in the formation of the antheridia, the growth of the male shoot is not arrested by the development of antheridia. So after the antheridia have been matured, the vegetative axis may grow out in the following year through the antheridial cup and produce a new shoot. This new shoot may also behave likewise).

Exercise 6

Object **: Study of archegonial branch and archegonium.**

Work procedure

Select a female plant, select archegonial head present at the tip, tease out the perichaetial leaves to expose the archegonia or cut L.s. of head, stain in safranin or fast green, mount in glycerine and study.

Comments

1. The archegonia are surrounded by coloured perichaetial leaves. This gives the appearance of a small flower.
2. The archegonia are found in terminal groups at the apex of the gametophore, thus arresting the further growth of the axis.
3. In each group there are usually three archegonia.

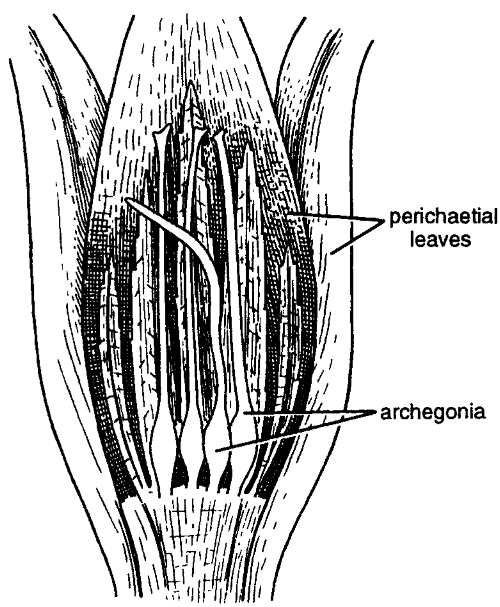


Fig. 7. ***Polytrichum.*** L.s. through archegonial head.

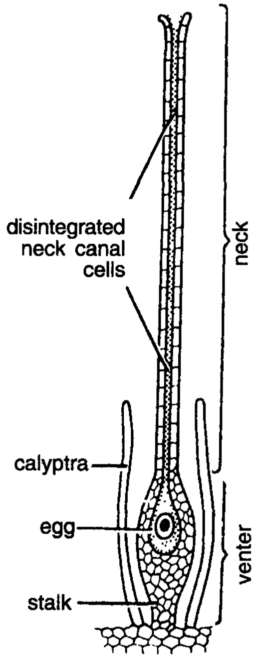


Fig. 8. ***Polytrichum.*** An archegonium.

4. Scattered among the archegonia are modified hair-like structures, the paraphyses.
5. The archegonia are also stalked and greatly elongated and consist of venter and a neck.
6. The venter is several cells thick and contains a venter canal cell and an egg.
7. The neck consists of six vertical rows of cells and contains a large number of neck canal cells which disintegrate as the archegonium matures.

Exercise 7

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

Work procedure

Study the specimen of gametophore that bears sporophyte.

Comments

1. The sporophyte is formed after fertilization and consists of foot, seta and capsule.
2. The foot is buried in the tissues of the leafy gametophore.

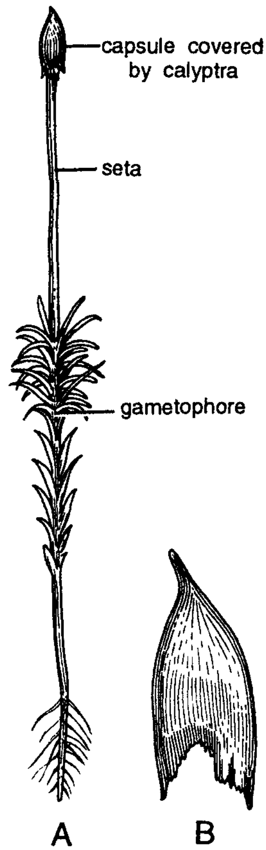


Fig. 9. ***Polytrichum.*** **A.** Sporophyte attached to the gametophore. **B.** Calyptra

3. Just above and in continuation of the foot is the long and slender seta which support the capsule at its apex.
4. With the growth of the sporophyte, the lower part of the archegonium enlarges following elongation, and is converted into a calyptra, covering the capsule.
5. The wall of the capsule is several layered and the outermost layer is differentiated into an epidermis with thick outer walls. All the cells of the wall layers contain chloroplast.
6. Inner to the wall there is an outer lacuna (air space), traversed radially by the chlorophyllous filaments. The filaments are connected internally with the outer wall of the spore sac.
7. The spore sac is internally bound by an inner lacuna made of filaments that connect the spore sac with the central columella.

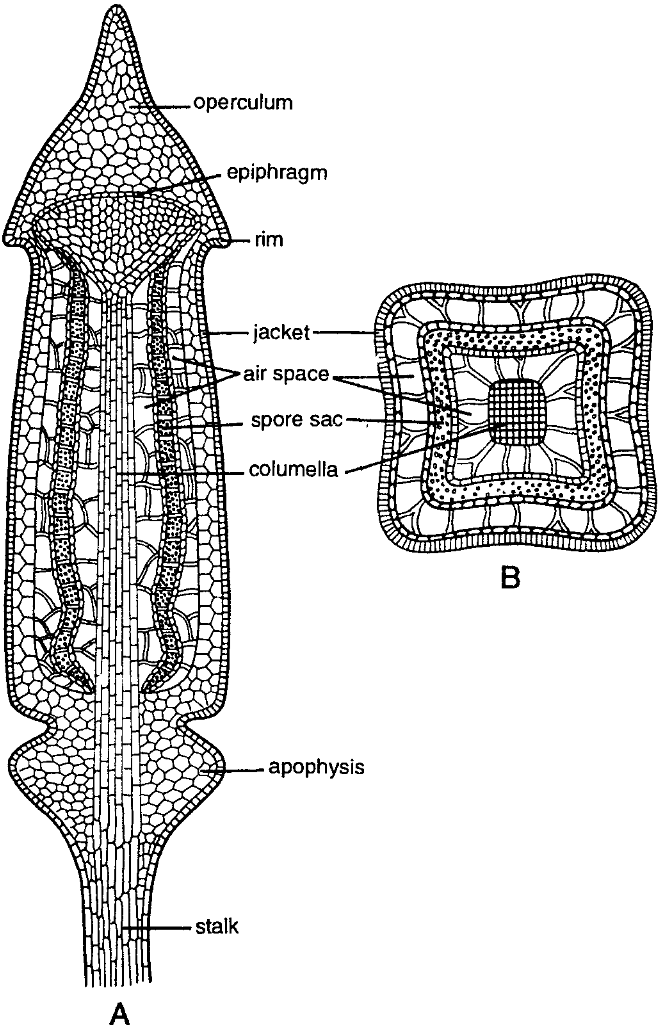


Fig. 10. ***Polytrichum.*** **A.** L.s. capsule. **B.** T.s. capsule.

8. The spore sac extends the entire length of the capsule. The archesporium (spore producing tissue) is 1 to 16 layered and all its cells develop into spore mother cells which after meiosis, give rise to spores.
9. At the top of the capsule is present a lid, the operculum.
10. Just below the operculum is present the epiphragm which stretches like a drum head over the opening of the capsule.
11. Just within the mouth of the capsule and under the epiphragm is a ring of peristome teeth.

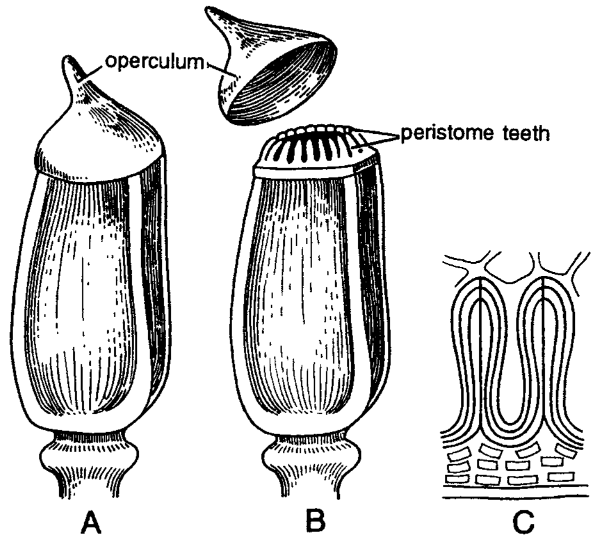


Fig. 11. ***Polytrichum.*** **A.** Ripe capsule after removing calyptra. **B.** Capsule after removing operculum. **C.** Two peristome teeth.

12. At maturity peristomial ring is composed of 32 or 64 short pyramidal teeth. These teeth are not hygroscopic but control the dispersal of spores.

Identification

Division—**Bryophyta**. (1) True roots absent and instead are present the rhizoids, (2) No vascular strand.

Class— **Bryopsida**. (1) Gametophore erect and leafy, (2) Rhizoids multicellular with oblique septa.

Sub-class—**Bryidae**. (1) Leaves with distinct midrib, (2) Seta long, (3) Spore sac usually separated from the capsule wall by air spaces.

Order—**Polytrichales**. (1) Gametophores tall and perennial, (2) Leaves narrow with lamellae on the upper surface of the midrib, (3) Peristome teeth 32 or 64 (4) Calyptra, cucullate, either smooth spinulose or hairy.

Family—**Polytrichaceae**. Characters same as those of Polytrichales.

Genus—***Polytrichum***. (1) Rhizoids rope-like, (2) Capsule angular.

Hints for Collection

Only three species of *Polytrichum* viz. *P. densifolium, P. xanthopiium* and *P. juniperinum* are found in India, mostly in hills. The species are found in a variety of habitats. Such as sandy ground, dry and stony places, peat, bog, marshy moores and damp soil, etc.

Pogonatum

Classification

Division	—	**Bryophyta**
Class	—	**Bryopsida**
Sub-class	—	**Bryidae**
Order	—	**Polytrichales**
Family	—	**Polytrichaceae**
Genus	—	***Pogonatum***

Exercise 1

Object **: Study of external morphology of the gametophyte.**

Work procedure

Observe the external features of the plant, the rhizome, rhizoids, axis and the leaves.

Comments

1. The gametophyte is differentiated into rhizoids, stem or axis and leaves.
2. The basal rhizomatous part of the erect stem is slightly stouter and stiffer. It bears rhizoids.
3. The rhizoids are thick walled and multicellular. The septa are oblique. Many rhizoids twist together in a rope-like manner forming strong cable-like strings.
4. The aerial part of the stem bears leaves. The leaves on the lower part are very small, scale-like and much paler in colour.
5. The upper leaves are crowded together spirally, spreading out from the stem and are rather stiff.
6. The leaves are sessile, pale in colour with sheathing broad base. The upper part of the leaf is deep green to brown or reddish brown in colour. The leaves gradually taper towards the apex and the margins are serrated.
7. A leaf has a distinct thick midrib in the centre with narrow wing-like lamina on either sides.
8. The upper surface of the midrib is completely covered by parallel longitudinal vertical plate-like structures called lamellae.

Exercise 2

Object **: To study anatomy of axis.**

Work procedure

Cut a thin transverse section of the axis. Stain in safranin, mount in glycerine and study.

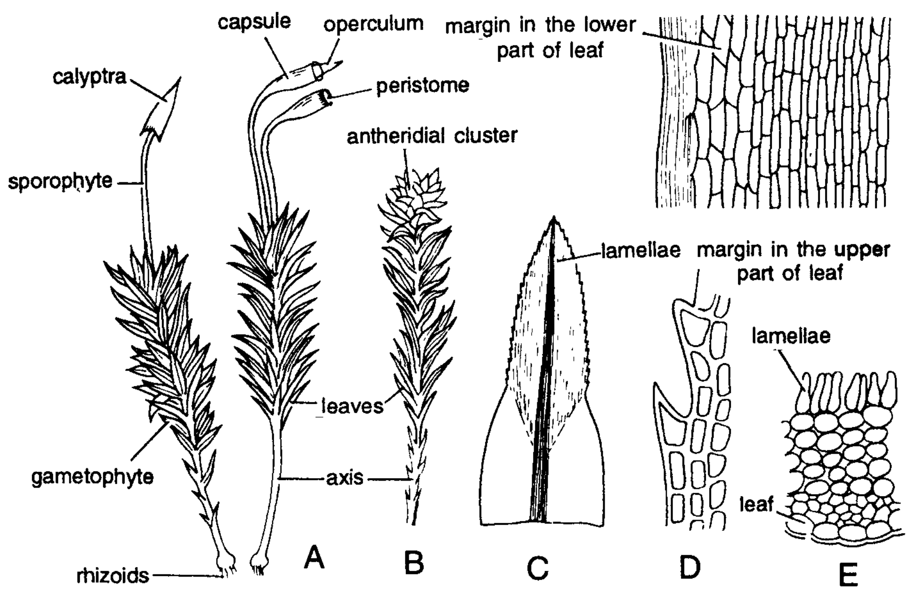


Fig. 1. ***Pogonatum.*** External features. **A.** Female plants. **B.** Male plant. **C.** Leaf showing lamina. **D.** Leaf cells at margin and base **E.** A part of T.s. of leaf showing lamellae.

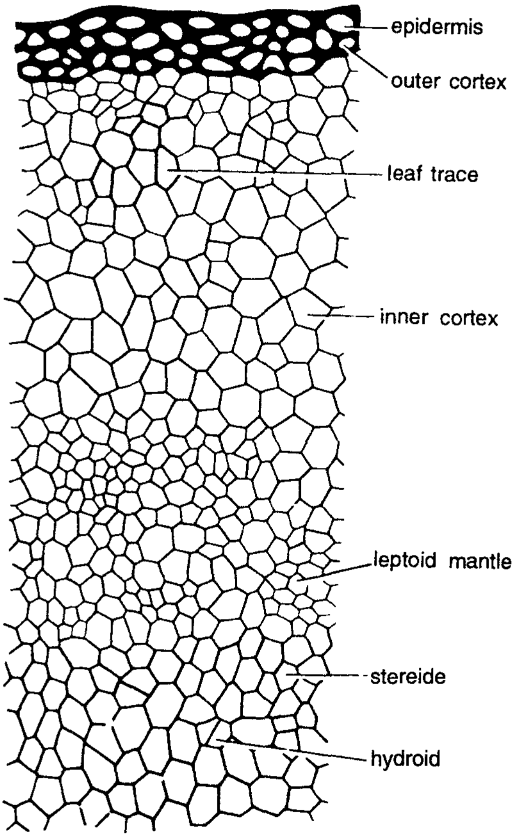


Fig. 2. ***Pogonatum.*** Anatomy of axis (a part cellular).

Comments

1. The transverse section shows epidermis, cortex, leptoid mantle, stereids and hydroids.
2. The outermost single layer of slightly thick walled cells is the epidermis. It is broken in the upper part of the stem due to presence of leaves.
3. A wide zone of cortex follows.
4. It shows outer and inner regions. The cells of the outer cortex are thick walled, elongated and deeply coloured.
5. The inner cortex is wider than the outer. The cells are thin walled, parenchymatous, lighter and compact.
6. A few thick walled leaf traces are also found in the cortex.
7. Inside the cortex is a zone of elongated cells with protoplasm but no starch. The cells are considered to be similar to sieve tubes and show oblique intervening cells like sieve plates. The cells are leptoids. This zone is called as leptoid mantle.
8. The centre of the stem shows hydrom cylinder which is made of two types of cells—the elongated, thick walled cells with living contents called stereids and similar cells devoid of contents called hydroids.
9. Hydroids are dispersed amongst stereids. They are considered to be helpful in water conduction.

Exercise 3

Object **: Study of the male plant and the antheridium.**

Work procedure

Study the position of antheridia on the male plant, tease or dissect the antheridial head, and study an antheridium.

Comments

1. Antheridia are borne at the apex of the branches of male plants.
2. Antheridia are surrounded by specialized leaves, the perichaetial leaves, at the apex. These are coloured red or orange and form a flower-like head.

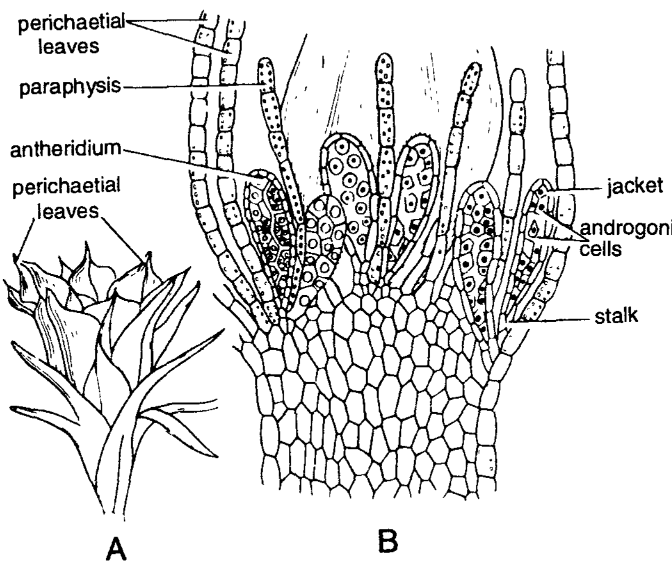


Fig. 3. ***Pogonatum.*** **A.** Male flower. **B.** L.s. of male head.

3. The group of these leaves enclose a cluster of antheridia and paraphyses.
4. The antheridia are borne in such a way that the center of the axis remains free for further growth.
5. Each antheridium is a club-shaped structure with a short stalk.
6. The stalk is multicellular and holds the body of the antheridium.
7. The body of antheridium is made of a single layered jacket that surrounds many androcytes.
8. Many paraphyses occur scattered amongst the anteridia. Paraphyses are long, multicellular, hair-like structures with rounded apical cell. Each cell has many chloroplasts.

Exercise 4

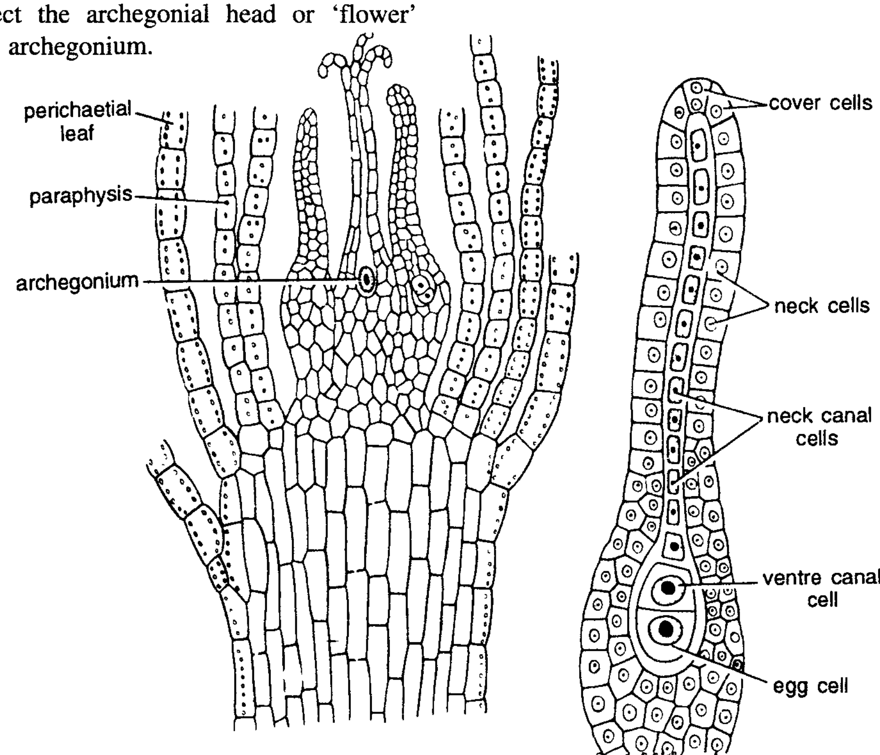
***Object* : To study the female plant and the archegonia.**

Work procedure

Study the position of archegonia on the female plant, tease or dissect the archegonial head or 'flower' and study the archegonium.

Comments

1. An archegonial cluster develops at the tip of the plant.
2. Each cluster consists of archegonia and paraphyses, surrounded by perichaetial leaves.
3. Perichaetial leaves are specially modified leaves located at the apex.
4. During the formation of archegonia, apical cell is used up and hence the axis stops any further growth.
5. The archegonium is attached to the stem apex by a short multicellular stalk.
6. The archegonium consists of a neck and a ventre. The neck consists of six vertical rows of cells, two cover cells and 8-12 (or more) neck canal cells. The ventre is made of venter canal cell and an egg cell. The wall of the ventre is made of 2 layers of cells.
7. The paraphyses occurring among the archegonia are multicellular, long, hair-like structures made of single row of cells. The cells are rich in chloroplasts.



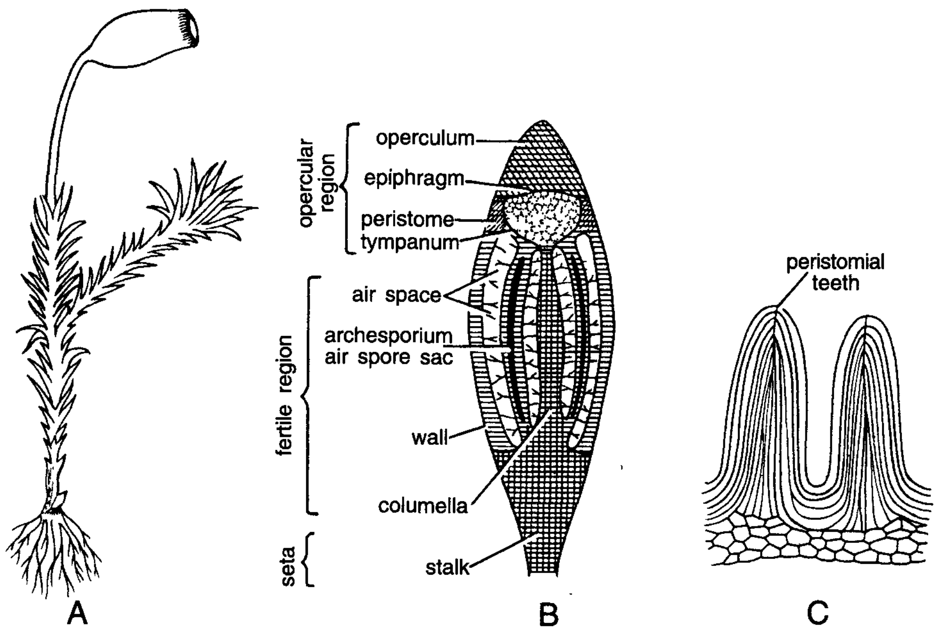


Fig. 5. ***Pogonatum.*** **A.** Sporophyte. **B.** L.s. sporophyte. **C.** Two peristome teeth.

Exercise 5

Object **: To study the sporophyte.**

Work procedure

Study the external features of the sporophyte and the internal structure by studying the slide of L.s. of the sporophyte.

Comments

1. The mature sporophyte consists of a foot, seta and a capsule covered by a calyptra.
2. L.s. of the capsule shows a lower stalk, middle fertile region and the upper operculum.
3. The stalk consists of a parenchymatous tissue that merges with columella. It is surrounded by chlorophyllous cells.
4. The capsule lacks a basal apophysis with stomata.
5. The fertile region of the capsule shows the following structure.
6. One layered epidermis with thick outer walls.
7. Several layered wall of chlorophyllous cells.
8. Large cylindrical air space or outer lacuna connected by string-like filaments of green cells with outer wall of the inner spore sac cylinder.
9. The spore sac has an outer one layered wall and inner two layered wall consisting of thin walled cells.
10. The spore sac is usually two layered thick. It shows archesporial tissue when young and spores when mature.
11. Inner spore wall is followed by another air space (inner lacuna).
12. The green filaments of inner air space are connected with centrally located parenchymatous columella. It is not cylindrical but produces four wing-like extensions.
13. Columella passes into opercular region and swells up to form drum-like roof of the capsule called epiphragm (tympanum).
14. Above the epiphragm is a conical and beaked operculum which is connected to the capsule by a ring-like diaphragm. Organised annulus is absent.
15. Above the diaphragm is a ring of 32 short peristomial teeth. These are formed of the

bundles of fibrous cells. The teeth are hygroscopic and control the dispersal of spores.

Identification

Division—**Bryophyta**. (1) True roots absent, (2) Presence of antheridia and archegonia, (3) True vascular tissues absent.

Class—**Bryopsida**. (1) Gametophore erect and leafy, (2) Rhizoids multicellular with oblique septa.

Sub-class—**Bryidae**. (1) Leaves with distinct midrib, (2) Seta long, (3) Spore sac usually separated from the capsule wall by air space.

Order—**Polytrichales**. (1) Gametophores tall and perennial, (2) Leaves narrow with lamellae on the upper surface of the midrib, (3) Peristome 32 or 64, (4) Calyptra cucullate, either smooth, spinulose or hairy.

Family—**Polytrichaceae**. (1) Single family, characters same as those of the order.

Genus—***Pogonatum***. (1) The leptoid and hydroid cells poorly demarcated, (2) Neck or apophysis absent from the capsule, (3) Peristomial teeth solid, 32 in number.

Hints for Collection

Pogonatum is the largest genus with about 199 species distributed all over the world. Of these, 34 are found in the temperate hills of India and Sri Lanka. It is commonly found in the hills of India, growing on the rock soil. The most common Indian species are *P. aloides*, *P. microstomum*, *P. junghunianum*, etc.

8 Chapter Pteridophyta

Preamble

The pteridophyta include those plants which are sometimes called "**Vascular cryptogams**", because of the presence of true xylem and hidden type of sexual reproduction. This group includes not only a large number of present day genera, but also a great many fossil types. Many of the genera which at the present day are quite small plants, have descended from ancient groups which in their days, formed large trees. Unlike bryophytes, the pteridophytes are found in great majority of habitats, ranging from aquatic to xerophytic. Generally they are land inhabiting but *Salvinia* and *Azolla* grow in aquatic habitat.

The members of the group vary greatly in form. These show two main evolutionary tendencies. One resulted in the production of large leaves and relatively small stems, and is known as the *megaphyllous* types. It is represented by ferns. The second tendency in which the leaves are small in relation to the stem and moreover the leaf trace leaves no gap in the stele, are known as the *microphyllous* types. This tendency is represented by club mosses and horse tails. The anatomy of these plants is to some extent dependent upon the type of the leaf they bear, but basically in all of them, the stem is divided into an outer cortex, and a central conducting system, the stele. A similar, though generallysimpler structure is found in roots. The leaves in megaphyllous types consist of a petiole and a leaf blade or lamina with many veins. Microphyllous leaves are much simpler, with no petiole and usually only one vein.

The reproductive organs are generally borne either on the leaves or in the axils between the leaves and the stem. They are made up of little capsules called sporangia, in which are developed the spores. All the spores may be of the same size, when the plant is said to be the homosporous, or they may be of the two different sizes and the plant is called as heterosporous. In heterosporous types, the smaller spores are termed as microspores, and are developed in microsporangia, while in larger spores, which are generally produced in smaller numbers, are termed as megaspores, and are found in megasporangia.

Since the plant body of the members belonging to this group, produces spores, they represent the sporophytes, being comparable to the sporogonium of bryophyta. The gametophytes are, however, small and insignificant and bear sex organs. The fern plant and the moss plant, thus cannot be compared, as they belong to different generations of life cycle.

Distinguishing Characters of Taxa

Various classifications of Pteridophytes have been proposed, time and again. Of these, a few classifications have been adopted by different workers. The classification followed in this book is a modified version of Riemers (1954). Below are given the characters of certain texa, described in the book.

DIVISION **PTERIDOPHYTA**

(1) True roots generally present
(2) Plant body differentiated into stem, roots and leaves
(3) True vascular strand present

SUB-DIVISION **1. PSILOPHYTOPSIDA**

(1) True roots absent
(2) Sporangia borne at the tips of erect branches either singly or in pairs
(3) Plants homosporous
(4) Plants found only as fossils

Order **Psilophytales**

(1) Sporophyte dichotomously branched
(2) Sporangia generally borne singly

Family **Rhyniaceae**

(1) Rhizoids unicellular, on rhizomes
(2) Aerial portion leafless

Classification of Pteridophyta

Division—PTERIDOPHYTA

Sub-division	Class	Order	Family	Examples
1. Psilophytopsida		Psilophytales	Rhyniaceae	*Rhynia**
				*Horneophyton**
2. Psilotopsida		Psilotales	Psilotaceae	*Psilotum*
3. Lycopsida		1. Lycopodiales	Lycopodiaceae	*Lycopodium*
		2. Lepidodendrales	Lepidodendraceae	*Lepidodendron**
				*Lepidocarpon**
		3. Selaginellales	Selaginellaceae	*Selaginella*
		4. Isoetales	Isoetaceae	*Isoetes*
4. Sphenopsida		1. Equisetales	Equisetaceae	*Equisetum*
		2. Calamitales	Calamitaceae	*Calamites**
5. Pteropsida	1. Eusporangiatae	1. Ophiglossales	Ophioglossaceae	*Ophioglossum*
		2. Marattiales	Angiopteridaceae	*Angiopteris*
	2. Leptosporangiatae	1. Filicales	1. Adiantaceae	*Adiantum*
			2. Polypodiaceae	*Dryopteris*
				Lastraea
				Nephrolepis
				Polypodium
				Pteridium
		2. Marsileales	Marsileaceae	*Marsilea*
		3. Salviniales	1. Salviniaceae	*Salvinia*
			2. Azollaceae	*Azolla*

* Fossils members

Examples ***Rhynia*, Horneophyton****

SUB-DIVISION **2. PSILOTOPSIDA**

(1) Sporangia are borne in the axils of scaly appendages or foliage leaves
(2) Plants living, true roots absent and homosporous

Order **Psilotales**

(1) Sporophyte dichotomously branched
(2) Sporangia generally borne singly
(3) Stele protostele to actinostele
(4) Eusporangiate and homosporous

Family **Psilotaceae**

(1) Axis branched
(2) Scale leaves small and minute

Example ***Psilotum***

SUB-DIVISION **3. LYCOPSIDA**

(1) Leaves microphyllous
(2) Sporangia borne singly on adaxial face of the sporophyll or in its axil
(3) Sporophytes homosporous

Family **Lycopodiaceae**

(1) Leaves without ligules
(2) Sporophylls and foliage leaves may be similar or dissimilar in shape

Example ***Lycopodium***

Order **2. Lepidodendrales**

(1) Plants tree-like
(2) Secondary tissues formed due to cambium
(3) Leaves microphyllous and ligulate
(4) Strobili heterosporous.

Family **Lepidodendraceae**

(1) Aerial portion freely branched
(2) Strobili at the tips of branches
(3) Trunk and branches with spirally arranged leaf scars

Examples ***Lepidodendron*, Lepidocarpon****

Order **3. Selaginellales**

(1) Each foliage leaf with a ligule at the base on adaxial side
(2) Sporophytes heterosporous

Family **Selaginellaceae**

(1) Stem herbaceous and dorsiventral or erect
(2) Gametophytes extremely reduced

Example ***Selaginella***

Order 4. **Isoetales**
(1) Herbaceous sporophytes with a massive rhizomorph at the base of the stem
(2) Leaves microphyllous and ligulate
(3) Sporophytes heterosporous
(4) Sporophylls may or may not be grouped in strobili
(5) Antherozoids multiflagellate

Family **Isoetaceae**
(1) Stem corm-like
(2) Sporophylls bearing sporangia on adaxial face, not grouped in strobili

Example ***Isoetes***

SUB-DIVISION 4. **SPHENOPSIDA**
(1) Stem branched, articulated, ridged and furrowed with distinct nodes and internodes
(2) Leaves microphyllous, small, scaly and arranged in whorls at the nodes

Order 1. **Equisetales**
(1) Stem branched, branches in transverse whorls
(2) Intenodes alternate with one another
(3) Vascular cylinder siphonostele, endarch

Family **Equisetaceae**
(1) No secondary growth
(2) Monosporous
(3) Sporagnia borne on sporangiophores which form a compact cone

Example ***Equisetum***

Order 2. **Calamitales**
(1) Tree-like sporophytes with considerable secondary thickening of stem and branches

Family **Calamitaceae**
(1) Stem branched, branches in whorls at nodes
(2) Stem shows endarch siphonostele

Example ***Calamites****

SUB-DIVISION 5. **PTEROPSIDA**
(1) Vascular cylinder siphonostelic, with leaf gaps
(2) Leaves megaphyllous, compound with rachis
(3) Leaves bear sporangia in sori
(4) Gametophytes small, green and free-living

CLASS 1. **EUSPORANGIATAE**
(1) Sporangium develops from a group of initials (eusporangiate development)
(2) Sporangial jacket more than one cell in thickness
(3) Large number of spores within sporangium
(4) Sporangia borne in spike or sori situated on the abaxial surface of the leaf.

Order 1. **Ophioglossales**
(1) Sporangia borne on a special structure called spike. It projects adaxially from a leaf and near the junction of blade and petiole.

Family **Ophioglossaceae**
(1) Single family, characters same as the order

Example ***Ophioglossum***

Order 2. **Marattiales**
(1) Young leaves with circinate vernation
(2) Leaves with fleshy stipules

Family **Angiopteridaceae**
(1) Sporangia almost free
(2) Sporangia linear in two rows on both the sides of the veins

Example ***Angiopteris***

CLASS 2. **LEPTOSPORANGIATAE**
(1) Sporangium develops from a single initial cell (leptosporangiatae development)
(2) Sporangial jacket one cell in thickness
(3) Definite number of spores

Order 1. **Filicales**
(1) Homosporous

Family 1. **Adiantaceae**
(1) Sori apparently marginal but superficial in origin
(2.) Indusia oblong or linear, usually many and distinct
(3) Leaflet margins bearing sori sharply reflexed

Example ***Adiantum***

Family 2. **Polypodiaceae**
(1) Annulus vertical
(2) Each sporangium with 32 to 64 spores

Examples ***Dryopteris, Lastraea, Nephrolepis, Polypodium, Pteridium***

Order 2. **Marsileales**
(1) Members heterosporous
(2) sporangia formed within sporocarps

Family **Marsileaceae**
(1) Members aquatic
(2) Sorus of gradate type, each producing both the types of sporangia
(3) Leaf circinately coiled in bud condition

Example ***Marsilea***

Order 3. **Salviniales**
(1) Members heterosporous

(2) Sporangia produced within sporocarps
(3) Sporocarp contains either a single megasporangium or numerous microsporangia
(4) The wall of the sporocarp is a modification of an idusium

Family **1. Salviniaceae**

(1) Sporocarps globose or ovoid, all of them of the same size

Example ***Salvinia***

Family **2. Azollaceae**

(1) Sporocarps of two types, one is larger and male-microsporocarp and the other smaller and female-megasporocarp.

Example ***Azolla***

Psilotum

Classification

Division	—	**Pteridophyta**
Sub-division	—	**Psilotopsida**
Order	—	**Psilotales**
Family	—	**Psilotaceae**
Genus	—	***Psilotum***

Exercise 1

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

Work procedure

Study the plant specimen.

Comments

1. The plant body may be pendent or erect and dwarfed (about 8 cm high) or may reach a height of 75-100 cm.
2. It is differentiated into (i) a basal rhizomatous system and (ii) aerial branches.
3. Basal rhizomatous system is subterranean, brown and rootless. The rhizome is repeatedly dichotomously branched and remains covered by small scales. The rhizome bears aerial branches.
4. Aerial shoots may be pendent (epiphytic species) or erect (terrestrial species). The slender and green aerial system is freely and dichotomously branched. The basal part of the shoot is cylindrical with longitudinal ribs. The distal green portion is radially cylindrical with three longitudinal ribs.

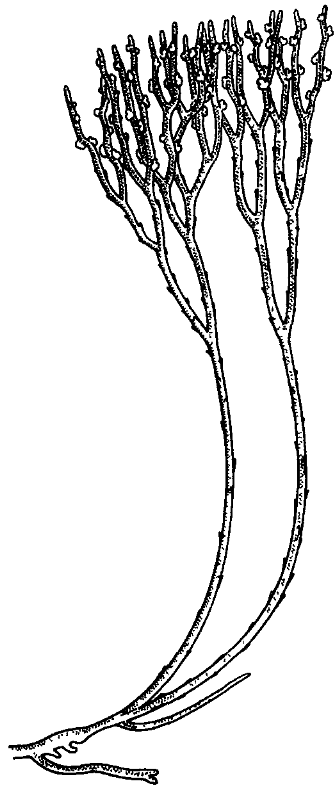


Fig. 1. *Psilotum.* External features.

5. Aerial shoots bear many, small and scale-like, irregularly distributed scale leaves.
6. Sporangia are borne in triads (synangium) on very short stalks in axil of leaves (bifid), mostly towards the tip of the aerial branches.

Exercise 2

***Object* : Study of anatomy of the rhizome.**

Work procedure

Cut a T.s. of the rhizome, stain in safranin and fast green combination, mount in glycerine and study.

Comments

1. The transverse section is almost circular in outline.

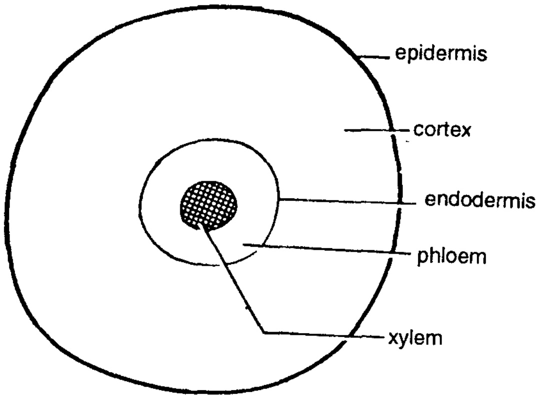


Fig. 2. ***Psilotum.*** T. s. rhizome (outlines)

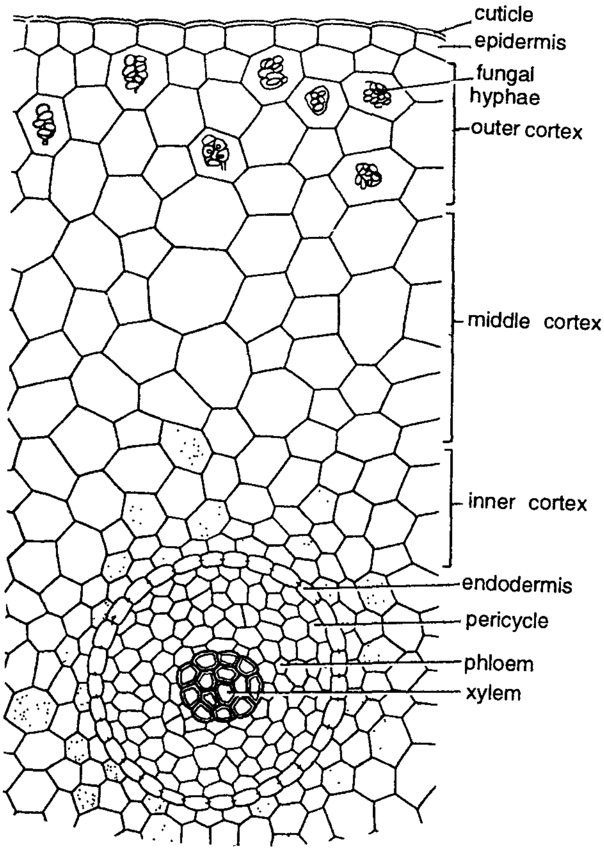


Fig. 3. ***Psilotum.*** T.s. rhizome (cellular).

2. It shows three distinct regions—epidermis, cortex and stele.
3. The cuticularised epidermis consists of rectangular or square cells, not much different from the underlying cells. The cells are slightly thick on their outer tangential faces.
4. The cortex is divisible into three regions—
(i) Outer: The cells are thin walled and parenchymatous containing the hyphae of endophytic mycorrhiza.
(ii) Middle : The cells are thin walled and parenchymatous with abundant starch grains.
(iii) Inner : The cells are small, thin walled and parenchymatous. These are coloured brown due to the presence of tannins.
5. Endodermis separates cortex from the stele. The cells are radially elongated and bear distinct casparian strips.
6. The centrally located stele is a protostele. It remains enclosed by a pericycle situated next to the endodermis. Pericycle is single layered and parenchymatous.
7. In the centre of the stele is a solid xylem strand, consisting of a few tracheids (the number and lobes of xylem vary with the age of rhizome. In young condition only 2-3 tracheids and a solid strand is present while in mature rhizome, number of tracheids increases and core becomes progressively lobed). The distinction between metaxylem and protoxylem elements is not clear.
8. The phloem extends from the xylem strand upto the pericycle thus completely surrounding the xylem strand.

Exercise 3

Object **: To study the anatomy of aerial shoot.**

Work procedure

Cut a T.s. of the aerial shoot, stain in safranin-fast green combination, mount in glycerine and study.

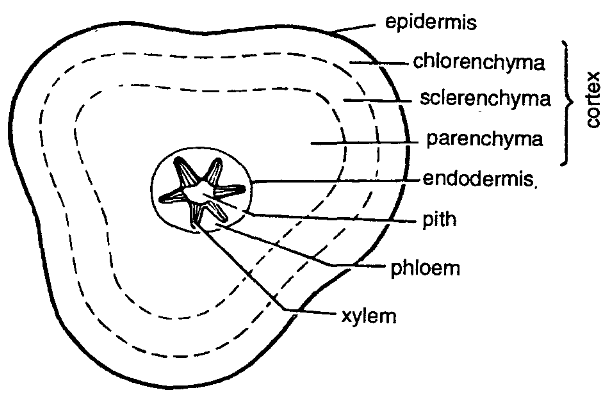


Fig. 4. ***Psilotum.*** T.s. aerial branch (outlines).

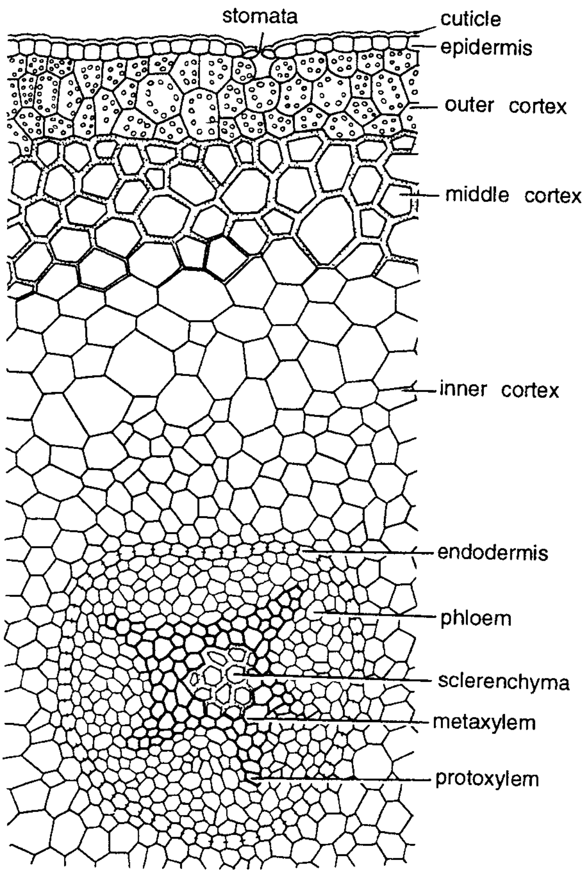


Fig. 5. ***Psilotum.*** T.s. aerial branch (a part cellular).

Comments

1. The transverse section appears slightly triangular in outline.
2. It is shows three distinct regions—epidermis, cortex and stele.
3. **The epidermis** consists of single layered, rectangular cells. The outer tangential walls are heavily cutinised and covered by a definite cuticle. Stomata are slightly sunken and are situated mainly in areas between the longitudinal ribs.
4. Internal to the epidermis is a broad cortex distinguishable into three zones—outer, middle and inner.
5. **Outer cortex**—the photosynthetic region, about 2-5 cells broad, is composed of vertically elongated cells and with small intercellular spaces. These cells contain numerous small chloroplasts.
6. **Middle cortex.** This zone is made of thick walled cells. The cells are sclerenchymatous and become progressively thinner towards inner zone.
7. **Inner cortex** is many celled broad, parenchymatous and contain numerous starch grains progressively towards centre.
8. **Endodermis** separates stele and cortex. The cells of the endodermis are tangentially elongated and exhibit distinct casparian strips on the radial end walls.
9. **The stele** is actinostelic, generally with six lobes. Each lobe has a few protoxylem elements at the tip while metaxylem is situated at its base.
10. The lobes of xylem are surrounded by phloem which extends upto the endodermis. (Typical sieve tubes are said to be absent).
11. Centre of the xylem is occupied by thick walled sclerenchymatous fibres with simple pits on their walls.

Exercise 4

Object **: To study the anatomy of leaf.**

Work procedure

Study a slide of T.s. of leaf.

Comments

1. The leaf is divisible into (i) epidermis, (ii) cortical tissue and (iii) a small leaf trace if present.
2. **The epidermis** consists of thin walled cells while the rest of the foliar appendage is filled by photosynthetic tissue.
3. **A leaf trace** ends into the base of foliar appendage (e.g. *P. flaccidium*), however, in *P. nudum* there is no vascular bundle.

Exercise 5

Object **: To study the spore producing organ–the synangium.**

Work procedure

Study the external features of synangium, slide of T.s. and also a single spore.

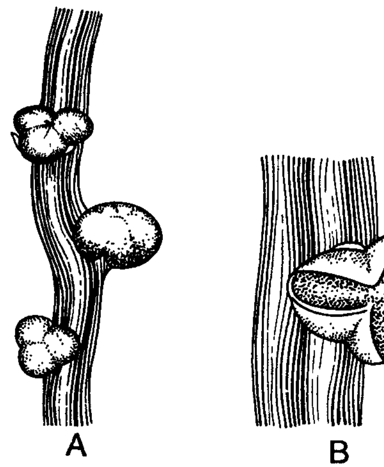


Fig. 6. ***Psilotum*. A.** Aerial branch with synangia, **B.** A single synangium.

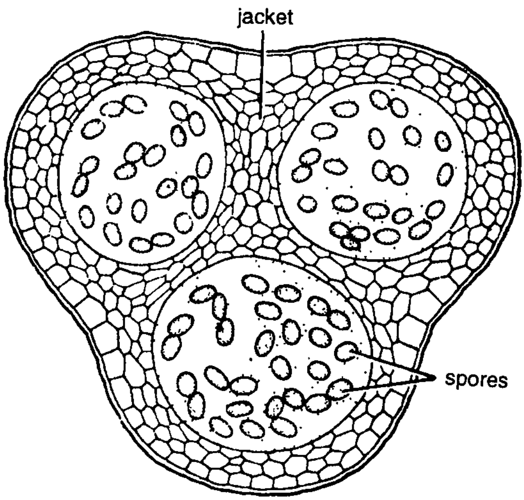


Fig. 7. ***Psilotum*.** T. s. synangium.

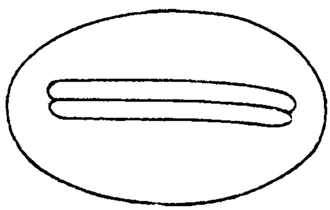


Fig. 8. ***Psilotum*.** A single spore.

Comments

1. Sporangia, the spore producing organs, are produced on the aerial branches.
2. These are borne in triads on minute appendages subtended by a bract. (Since the sporangia are fused with one another, the structure is called as synangium).
3. In a transverse section, synangium reveals 4-5 layered jacket, outer of which is made of thick walled cells. The loculi are filled with numerous spores. Interspersed among the spores are

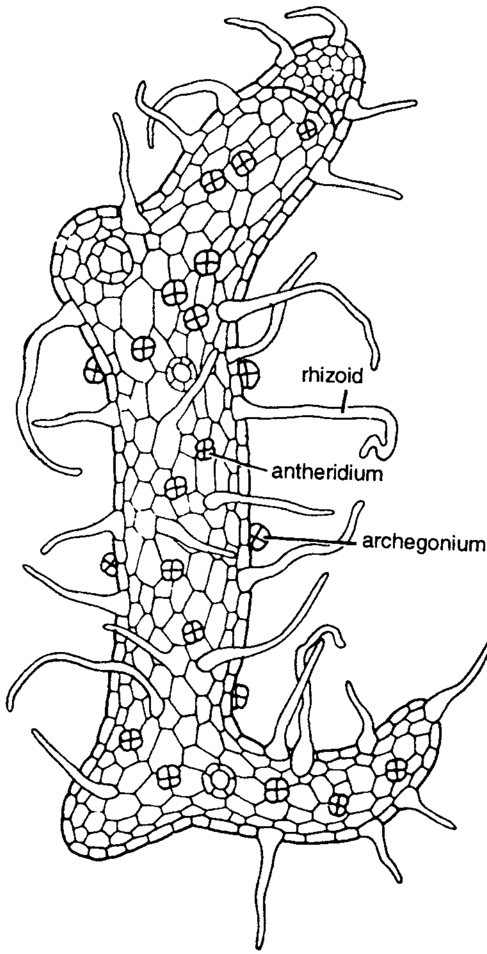


Fig. 9. ***Psilotum*.** A gametophyte.

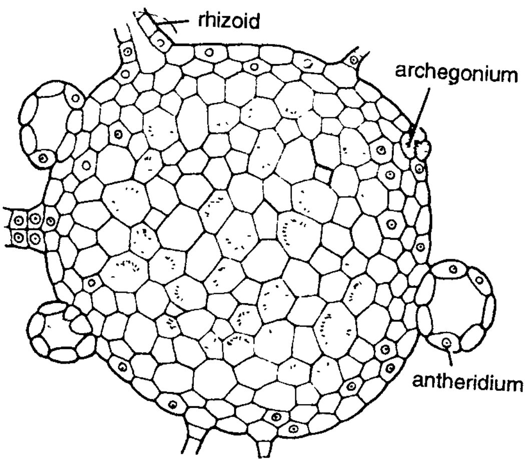


Fig. 10. ***Psilotum.*** T.s. gametophyte.

disintegrated sporocytes which serve as nutritional fluid.

4. Individual spores are bean-shaped or bilaterally symmetrical. The wall pattern is reticulate. There is a narrow slit with a median ridge.

Exercise 6

***Object* : Study of gametophyte.**

Work procedure

Study the slide of prothallus.

Comments

1. Gametophyte is subterranean and colourless. It is saprophytic and lives through the medium of symbiotic phycomycetous fungi.
2. Gametophytes are irregularly cylindrical and once or twice dichotomously forked. Rhizoids are also given out.
3. Gametophytes are homothallic. Sex organs are scattered over the entire surface. Archegonia are more in number than antheridia (or otherwise).

Identification

Division—**Pteridophyta**. (1) True roots generally present (except in Psilopsida), (2) True vascular strand present.

Sub-division—**Psilotopsida**. (1) True roots absent, (2) Shoot differentiated into subterranean rhizome and aerial portion, (3) Sporangia borne terminally.

Order—**Psilotales**. (1) Sporophyte dichotomously branched, (2) Sporangia generally borne singly, (3) Stele protostele, generally actinostele, (4) Eusporangiate and homosporous.

Family—**Psilotaceae**. (1) Axis branched, (2) Scale leaves small and minute

Genus—***Psilotum***. Sporangia borne in triads (synangium).

Hints for Collection

Only two species of *Psilotum* viz. *P. nudum* (=*P. triquetrum)* and *P. flaccidum* (=*P. complanatum*) are known. These are widespread in tropical and subtropical regions of both the hemispheres. *P. nudum* is of widespread occurrence and is known to occur at Pachmarhi in M. P. and Darjelling, West Bengal. It is primarily terrestrial and is collected from crevices of rocks and occasionally as epiphyte on tree ferns and palms.

Lycopodium (Club Moss)

Classification

Division	—	**Pteridophyta**
Sub-division	—	**Lycopsida**
Order	—	**Lycopodiales**
Family	—	**Lycopodiaceae**
Genus	—	***Lycopodium***

The genus *Lycopodium* is divided in to 2 sub-genera—Urostachya and the Rhopalostachya. The following are the differences between the two.

Urostachya	Rhopalostachya
1. Plant body erect or pendent.	1. Plant generally trailing or creeping.
2. Branching rare; if present always dichotomous.	2. Branching is dichotomous at the base but in the upper region only one of the branches grows more prominently.
3. The roots take their origin only from the basal part of the stem.	3. Adventitious roots may arise from any part of the stem.
4. Organized strobili are rarely found.	4. Strobili are always well organized and borne on long stalks.
5. Sporophylls are almost similar to the foliage leaves, the only difference is in their size.	5. Sporophylls are different from foliage leaves. They are pale yellowish and chaffy.
6. The margins of sporophylls are entire.	6. Margins of sporophylls are toothed.
7. The spores possess a pitted surface without any external outgrowth.	7. Spores possess reticulate surface

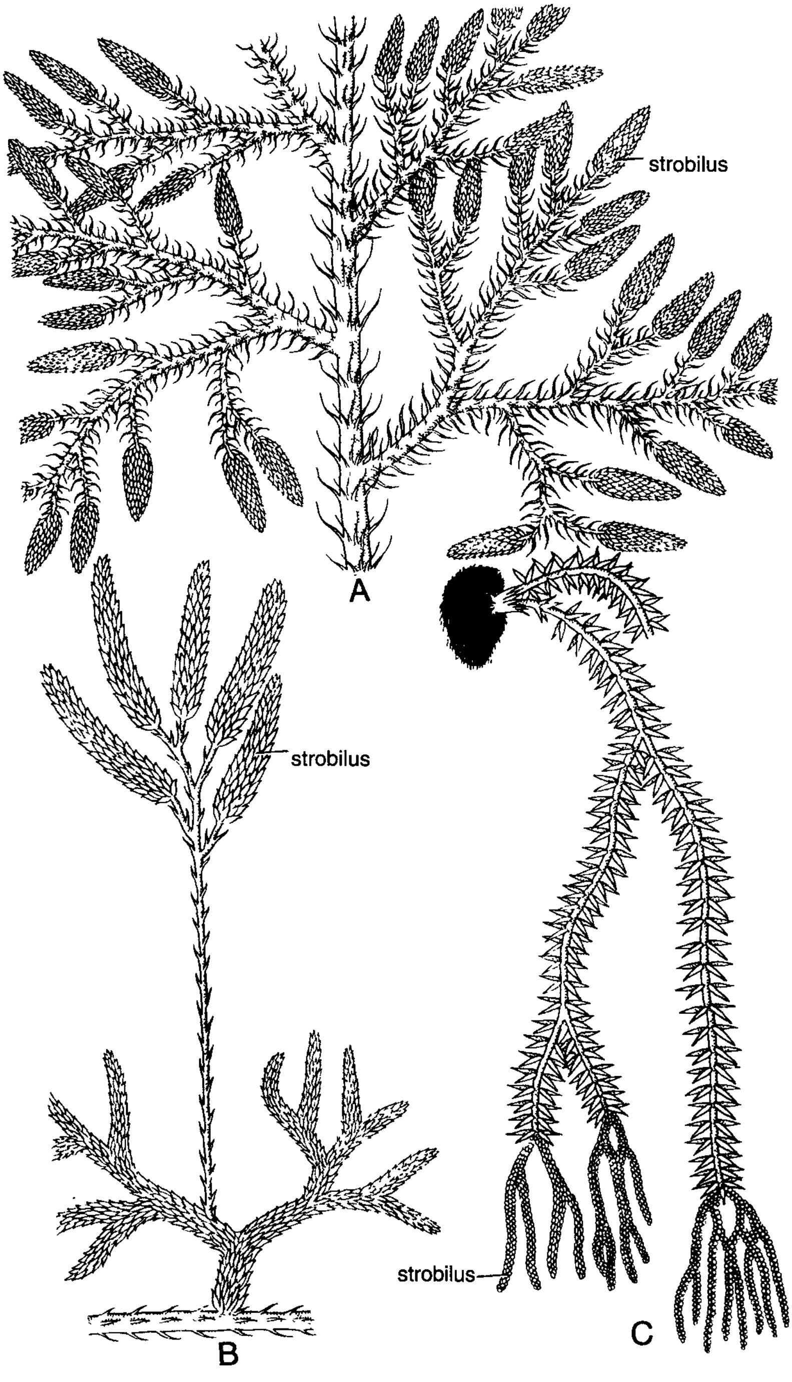


Fig. 1. ***Lycopodium.*** Sporophytes with strobili. **A.** *L. cernum* (terrestrial), **B.** *L. clavatum* (terrestrial), **C.** *L. phlegmaria* (epiphytic).

Exercise 1

***Object* : Study the external morphology.**

Work procedure

Study the specimen. Observe the differentiation of plant body into root, stem and leaves. Study the leaves and the stomata.

Comments

1. **The plant body** consists of creeping rhizome which gives off slender, elongated aerial branches from the upper side and adventitious roots from the lower. The aerial branches vary from 3-8 inches in length. *L. cernum* is exceptional in attaining a height of 2 feet or more.
2. **Habitat.** Most of the species are terrestrial and the sporophyte may either have an upright stem or a horizontally creeping stem. Some species grow as epiphytes on higher plants which show pendent habit e.g. *L. phlegmaria, L. squarrosum.*
3. **The branching** is mostly dichotomous but in some species it may be monopodial also.
4. **The stem** and its branches are densely covered with small leaves present in close spirals or whorls.
5. **The leaves** are entire, small and membranous, rarely, exceeding 1 cm in length. Each leaf is supplied by a single mid-vein which runs almost unbranched right upto the apex.
6. **Epidermis.** The walls of the epidermal cells of the leaf are sinuous.
7. **Stomata.** Stomata are more or less parallel to the midrib. These are equally distributed on both of the leaf surfaces.

Exercise 2

***Object* : Study of anatomy of root.**

Work procedure

Cut a T.s. of the root, stain in safranin and fast green combination, mount in glycerine and study.

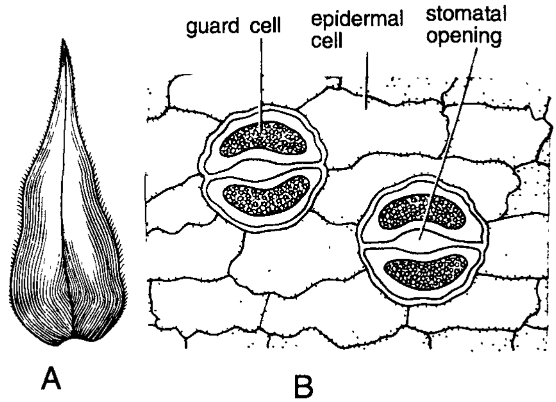


Fig. 2. ***Lycopodium.*** **A.** Entire leaf, **B.** A part of leaf epidermis showing stomata.

Comments

1. The root is differentiated into an epidermis, cortex and the stele.
2. **The epidermis** is single layered and gives rise to root hairs, the latter occur in pairs.
3. **The cortex** is several layered and in older roots a few of the outer layers become sclerified. The inner cells are thin walled and parenchymatous without any intercellular spaces.
4. **The stele** ranges from monarch to tetrarch but generally it is diarch with two protoxylem masses.

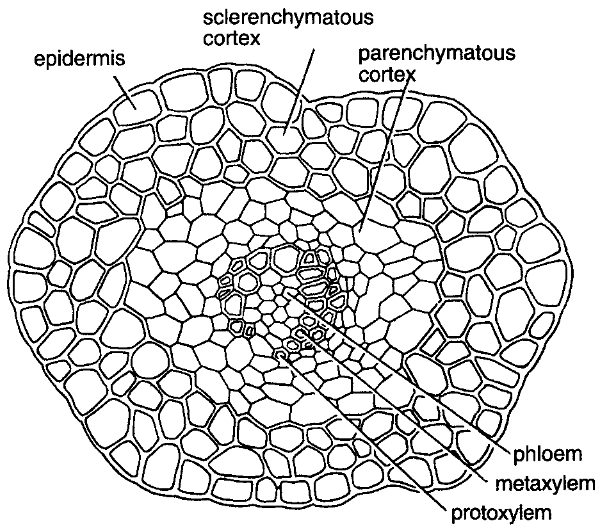


Fig. 3. ***Lycopodium.*** T.s. root.

5. The xylem is C or U shaped and is so oriented that the opening of C or U faces away from the stem.
6. The protoxylem is present at the tips of C or U and the intervening portion consists of metaxylem.
7. The phloem is present in between the arms of C or U.

Exercise 3

***Object* : Study of anatomy of stem.**

Work procedure

Cut T.s. of the stem, stain in safranin-fast green combination, mount in glycerine and study.

Comments

1. A transverse section of the stem shows an epidermis, a wide cortex and a stele.
2. **Epidermis** is single layered and is provided with stomata.
3. **The stem** has certain ridges and grooves. Chloroenchyma is present in the ridges.
4. **Cortex.** The structure varies from species to species. In some species, it is parenchymatous throughout, in others the inner and other portions are thick walled, and in still others the entire cortex is thick walled.
5. **Endodermis.** It is single layered and lies inner to cortex. In older stems, however, the endodermis may not be well defined.
6. **Pericycle.** The endodermis is follows this layer which is composed of 3 to 6 cells.
7. **Stele.** The centre is occupied by a protostele. Three different types of styles are found in species of *Lycopodium*.
   (i) In some species viz. *L. clavatum* and *L. complanatum*, it is definitely organised into a plectostele i.e. xylem and phloem occur in alternating bands that are more or less parallel.
   (ii) In other species viz. *L. seratum* and *L. phlegmaria*, it is star-shaped with 4 arms in which grooves are occupied by phloem. This is known as actinostele.
   (iii) In still other species, as exemplified by *L. cernum*, it is a haplostele in which the xylem strands lie scattered in phloem.

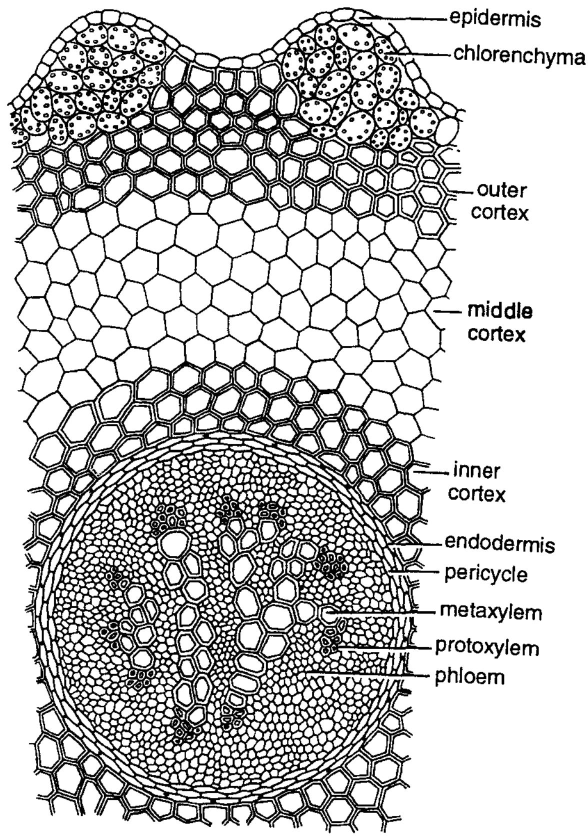


Fig. 4. ***Lycopodium.*** T.s. stem (a part cellular).

8. **The xylem** is exarch with protoxylem facing towards the periphery and metaxylem towards the centre.
9. **The tracheids** in metaxylem are scalariform while in protoxylem they are spiral or annular.
10. **The phloem** consists of unicellular sieve tubes, with numerous sieve plates, and phloem parenchyma. The companion cells are absent.
11. **Leaf traces** are seen to traverse the cortex.

Exercise 4

***Object* : Study the anatomy of leaf.**

Work procedure

Place the leaf in pith, cut T.s., stain in safranin-fast green combination, mount in glycerine and study.

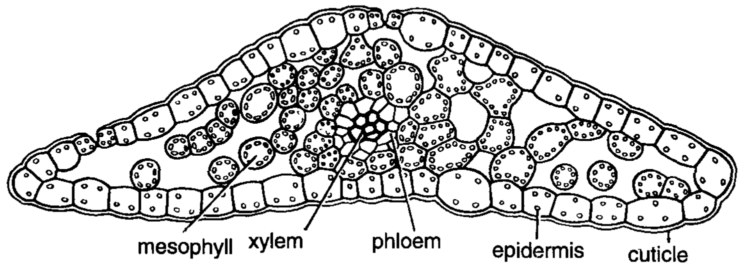


Fig. 5. ***Lycopodium.*** T.s. leaf.

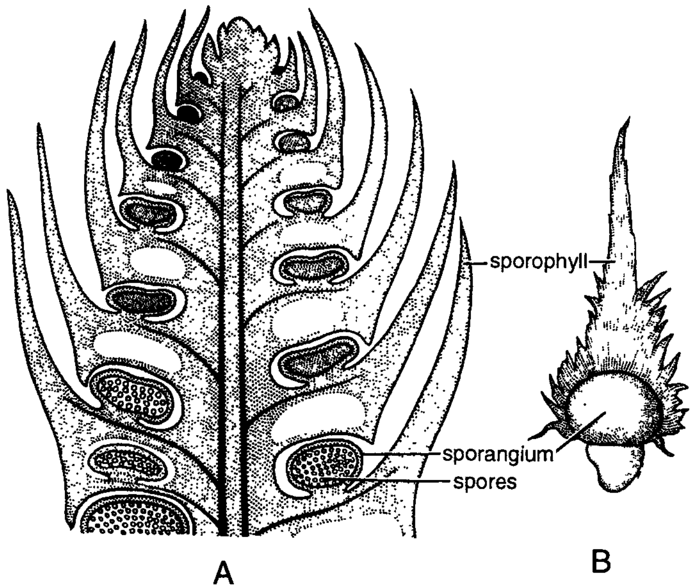


Fig. 6. ***Lycopodium.*** **A.** L.s. apical portion of strobilus. **B.** Dorsal view of sporophyll and a sporangium.

Comments

1. The epidermis is single layered. The stomata are equally distributed on both the sides.
2. The leaf has a single vascular strand which is concentric with xylem in the centre.
3. The cells between the epidermis and the vascular strand form spongy parenchyma.

Exercise 5

Object **: Study of spore producing organ.**

Work procedure

Study the strobili, L.s. of strobilus and the spores; by observing a prepared slide.

Comments

1. **External features of strobilus.** Sporangia are the spore producing organs. These are grouped to form strobili which are situated at the apices of branches. (In *L. selago* distinct strobilus is lacking and the vegetative and reproductive regions alternate each other.).
2. **L.s. of the strobilus** shows a central strobilar axis with spirally arranged sporophylls.
3. Each sporophyll bears a sporangium near its base on the adaxial side.
4. A sporangium is a black, kidney shaped structure, with a long or short massive stalk.
5. Sporangium consists of a wall and the cavity with spore.

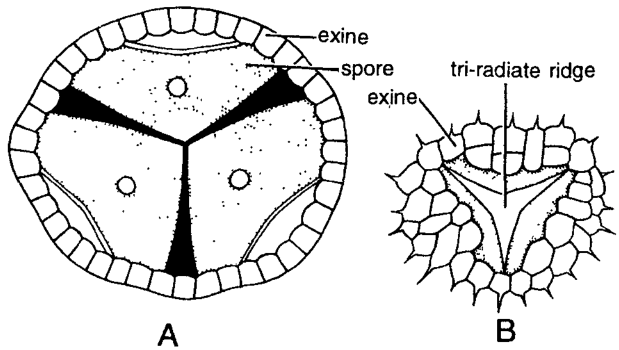


Fig. 7. *Lycopodium*. A. Spore tetrad, B. A spore.

6. The wall of sporangium is several layered thick. Tapetum forms the innermost layer.
7. The cavity has many spores, arranged in tetrahedral tetrads. Since all the spores are of one type, the plant is called homosporous.
8. Each spore is a minute structure with a triradiate ridge. It has a thick and spiny exine and a thin and membranous intine.
9. The spores germinate to form the prothallus.

Exercise 6

***Object* : To study the gametophyte.**

Work procedure

Study the slide showing gametophyte. Observe the structure of sex organs.

Comments

1. The gametophytes may be subterranean or sub-aerial.
2. The sub-aerial type is green, about 2 or 3 mm in length and bears the sex organs.
3. The subterranean type is non-green and is bigger as compared to the sub-aerial type. It generally consists of a tuberous body with a lobed crown that bears the sex organs.
4. The prothallus is monoecious and the sex organs lie almost wholly embedded in the tissue of the prothallus except the uppermost portion.
5. The antheridium is spherical with a single layer of jacket, containing within, a number of antherozoids or antherozoid mother cells. The antherozoids are fusiform and biciliate.

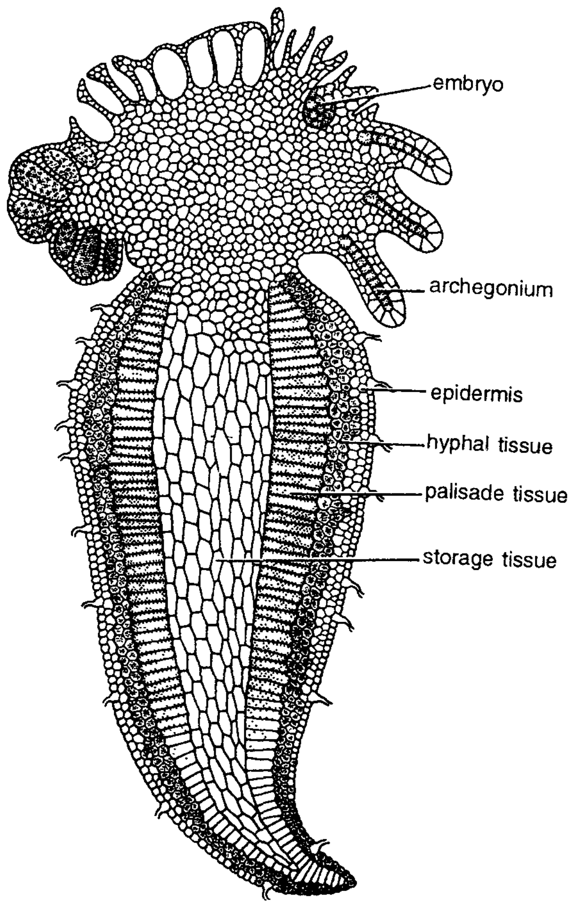


Fig. 8. *Lycopodium*. V.s. of mature gametophyte.

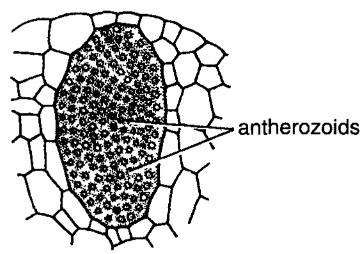


Fig. 9. *Lycopodium*. Mature antheridium.

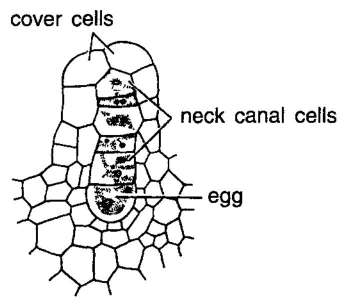


Fig. 10. *Lycopodium*. Nearly mature archegonium.

6. The archegonium is a narrow elongated structure. It has a narrow venter with an egg and venter canal cell and a long neck generally containing 4-6 neck canal cells.

Identification

Division—**Pteridophyta**. (1) Plant body differentiated into stem, roots and leaves, (2) A definite vascular strand present.
Sub-division—**Lycopsida**. (1) Laves microphyllous, (2) Sporangia borne singly on adaxial face of the sporophyll or in its axil, (3) Sporophylls borne in strobili.
Order—**Lycopodiales.** (1) Stem has protostele, (2) Sporophytes homosporous.
Family—**Lycopodiaceae**. (1) Leaves without ligules, (2) Sporophylls and foliage leaves may be similar or dissimilar in shape.
Genus—***Lycopodium***. (1) The sporophyte is long and always more than 2 inches, (2) Sporangia kidney-shaped, (3) Stele either a plectostele or actinostele or haplostele.

Hints for Collection

Out of the 100 species of *Lycopodium* about 18 occur in India. These are found in Himalayan and sub-Himalayan tracts, Garhwal, Assam, Bengal and also in Nilgiris.

Selaginella
(Small Club Moss)

Classification

Division	—	**Pteridophyta**
Sub-division	—	**Lycopsida**
Order	—	**Selaginellales**
Family	—	**Selaginellaceae**
Genus	—	***Selaginella***

Exercise 1

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

Work procedure

Study the plant, specimen observe the differentiation of plant body into root, stem and leaves. Study the two types of leaves, their arrangement and structure. Also observe the structure of a ligule. Note the presence of rhizophore.

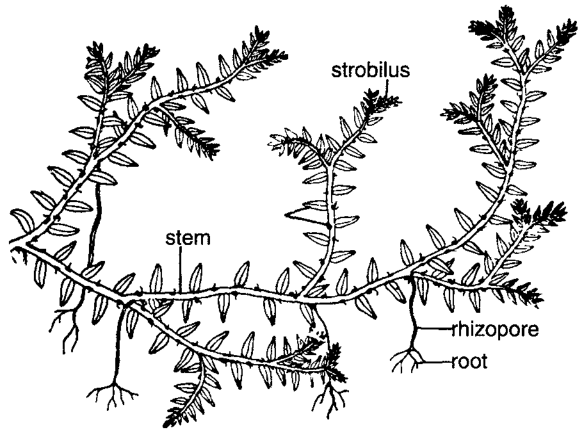


Fig. 1. ***Selaginella.*** External features.

Comments

1. Many species are prostrate, creeping on the ground e.g. *S. kraussiana*, others are sub-erect e.g. *S. trachyphylla* or erect e.g. *S. erythropus*. A few species climb with the help of rhizophores e.g. *S. alligans*.
2. The plant body is divided into root, stem and leaves.
3. The primary root is short lived and all other roots are adventitious.
4. On the basis of nature of stem and form of the leaves, the genus is sub-divided into two

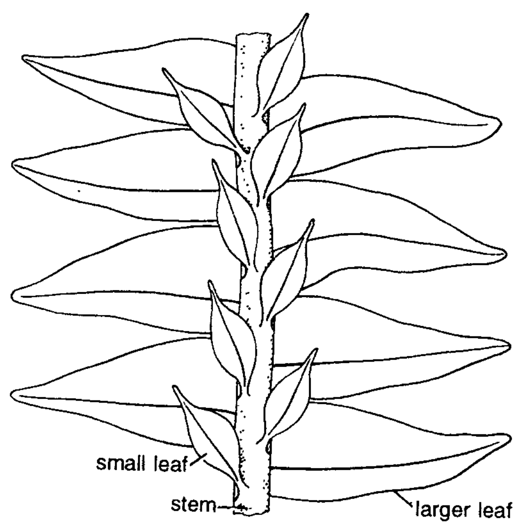


Fig. 2. ***Selaginella.*** A part of stem showing arrangement of leaves.

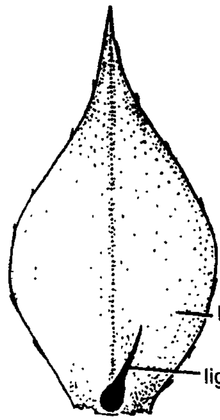


Fig. 3. ***Selaginella.*** Leaf (adaxial face showing ligule).

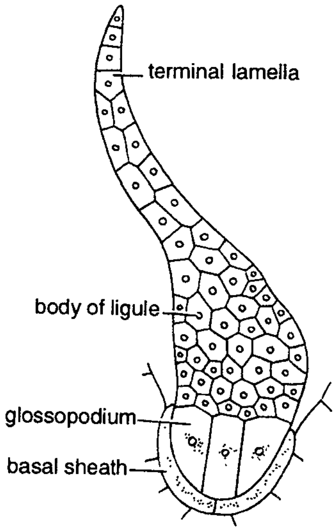


Fig. 4. ***Selaginella.*** A ligule.

sub-genera—the homoeophyllum and the heterophyllum.

5. In homoeophyllum species (*S. selaginoides*, *S. rupestris*, etc.) the stem is upright and all leaves are alike, while in heterophyllum species (majority of the species), the stem is prostrate and dorsiventral; and leaves are dimorphic (small and large).
6. In homoeophyllum, all the leaves are alike, spirally arranged, small and simple.
7. In heterophyllous species, they are dimorphic and are borne in pairs on dorsiventral stem. The two leaves are markedly different in size (one is larger and other smaller).
8. The smaller leaf of each pair is inserted on the dorsal side of the stem while the larger leaf is inserted on the ventral side.
9. The successive pairs of leaves are so arranged, that large leaf always alternates with the large leaf, and small leaf with the small leaf.
10. Each leaf is sessile, generally obovate with acute apex, and has a distinct midrib.
11. At the base of each young leaf, on the adaxial face, their is small tongue-like out growth, the ligule.
12. It is differentiated into basal sheath, glossopodium and the body of the ligule.
13. Whereas the cells of the sheath are tubular in shape and are dead, those of the glossopodium are vertically elongated.
14. The body of the ligule has parenchymatous cells with dense protoplasm.
15. From the point where stem branches, a cylindrical leafless organ is seen growing downward. This is known as rhizophore.
16. On reaching the ground, rhizophore terminates into roots (The morphological nature of the rhizophore is still open to question).
17. Certain vertical branches from the stem are reproductive in nature and bear strobili.

Features of special interest

1. Presence of rhizophore.
2. Dimorphic leaves (in heterophyllous species).
3. Presence of ligule.

Exercise 2

***Object* : To study anatomy of the root.**

Work procedure

Cut a T.s. of the root, stain in safranin-fast green combination, mount in glycerine and study.

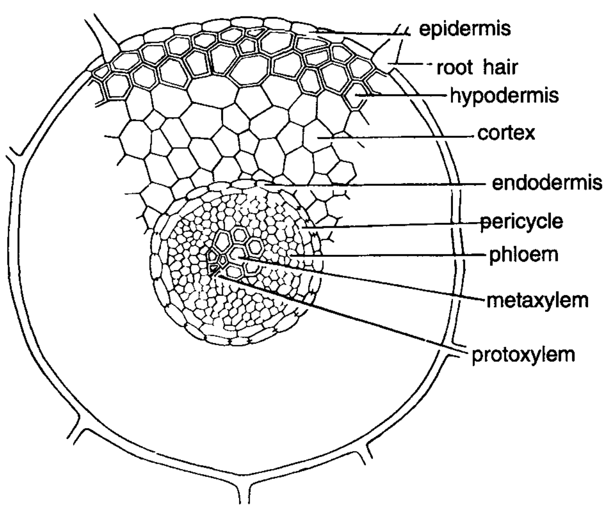


Fig. 5. ***Selaginella.*** T.s. root (a part cellular).

Comments

1. The section is almost circular in outline.
2. The tissues are differentiated into epidermis, cortex and stele.
3. **The epidermis** is single layered and its cells are tangentially elongated. Few of these cells give rise to the root hairs.
4. **Cortex** may either be made of parenchymatous (thin walled) cells or the outer layer of cells may form a sclerenchymatous (thick walled) hypodermis.
5. **The stele** lies in the centre. It is protostelic, monarch and exarch.
6. **Endodermis** is one layered and generally indistinct.
7. **Pericycle** is one to three layered.
8. **Xylem** forms only one group. Protoxylem is situated towards the periphery.
9. **Phloem** surrounds the centrally located xylem.

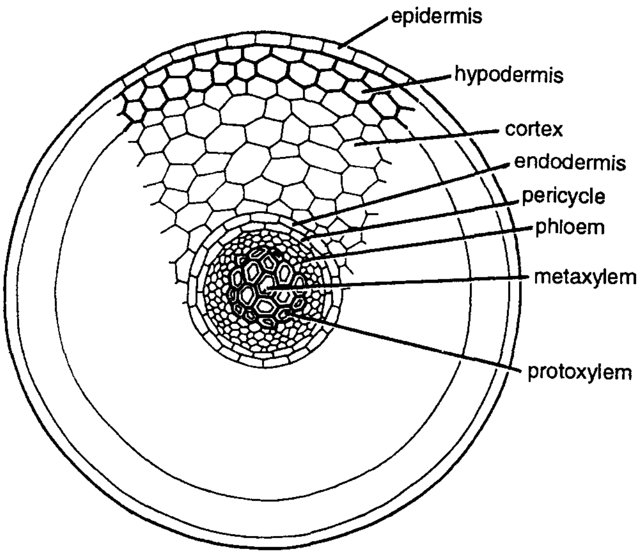


Fig. 6. ***Selaginella.*** T.s. rhizophore (a part cellular).

Exercise 3

***Object* : Study of anatomy of rhizophore.**

Work procedure

Cut a T.s. of the rhizophore, stain in safranin-fast green combination, mount in glycerine and study. Anatomically, the structure of the rhizophore is similar to that of root, with some minor differences, which occur on account of its environment.

Comments

1. The outline of the section is almost circular.
2. The section shows epidermis, hypodermis, cortex, endodermis and stele.
3. **The epidermis** is cuticularised.
4. **Hypodermis** that follows is 2-3 celled thick.
5. **Cortex** is few celled and parenchymatous. It occupies most of the part of section.
6. **Endodermis** is present between the stele and the cortex. It is followed by a single layered parenchymatous pericycle.
7. **The stele** is a protostele. It shows monarch and exarch condition. In some species (e.g. *S. atroviridis)* the metaxylem is lunar shaped and many protoxylem groups are situated on the concave adaxial side.

Exercise 4

***Object* : Study of anatomy of the stem.**

Work procedure

Cut a T.s. of the stem, stain in safranin-fast green combination, mount in glycerine and study.

Comments

1. **The outline** of the section appears slightly wavy.
2. **The section** shows epidermis, cortex and the stele.
3. **Epidermis** is the outermost layer. It is cuticularised and lack stomata.
4. **The cortex** consists of parenchymatous cells, without any intercellular spaces. All the cells of the cortex are thin walled.
5. **Hypodermis** occurs close to epidermis. It develops from cells of outer cortex which become thick walled. In xerophytic, species (e.g. *S. rupestris*, *S. lepidophylla*) hypodermis is more thickened.
6. **The stele** is generally a protostele.
7. **Endodermis** separates vascular tissue from the cortical region, by radially elongated endodermal cells, called as trabeculae, with

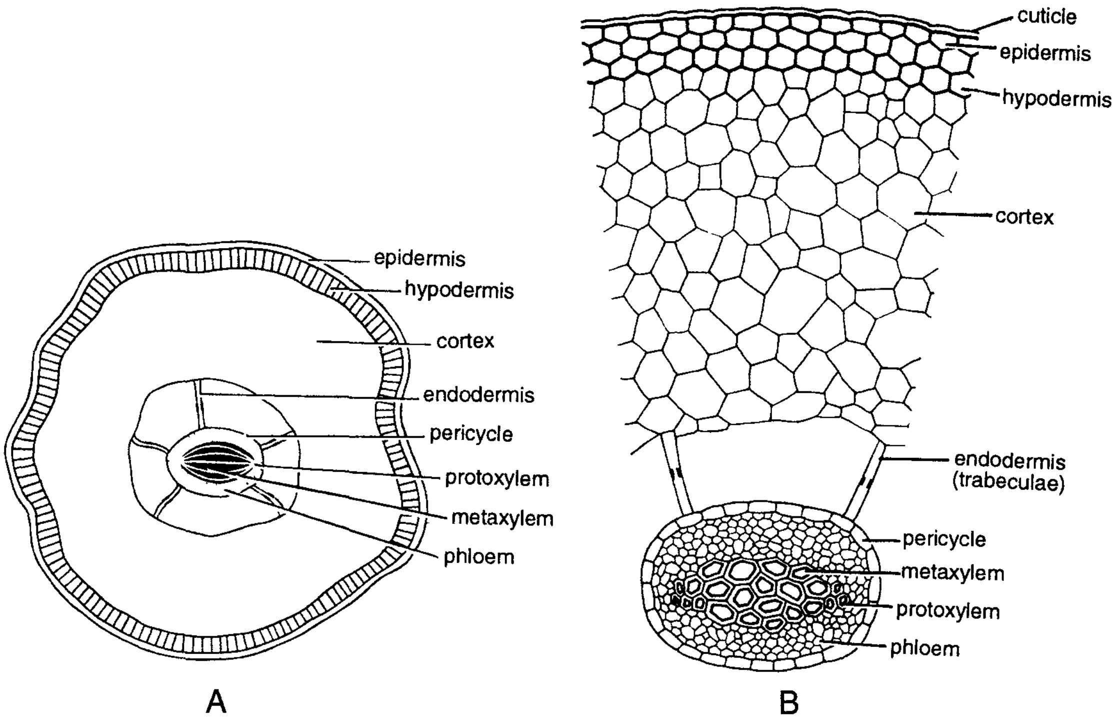


Fig. 7. ***Selaginella.*** T.s. stem **A.** Outline **B.** A part cellular.

conspicuous intercellular spaces between two trabeculae.

In spite of their great elongation, trabeculae still retain the transverse thickenings, the casparian strips, on their radial walls, characteristic of endodermal cells. Xerophytic species lack trabeculae (e.g. *S. lepidophylla* and *S. rupestris*).

8. **Pericycle** is a single layer surrounding the xylem and phloem and follows endodermis.
9. **Stele.** The number of steles in a stem varies from 1-16 thus exhibiting a polystelic condition.
10. Single stele, when present is generally diarch and exarch.
11. In *S. kraussiana,* the commonest species, there are two steles, each with a single exarch mass of protoxylem.
12. **The protoxylem** masses of the two steles point in opposite directions.
13. **The phloem** consists of smaller cells with dense protoplasm and completely surrounds the central core of xylem, in each stele.

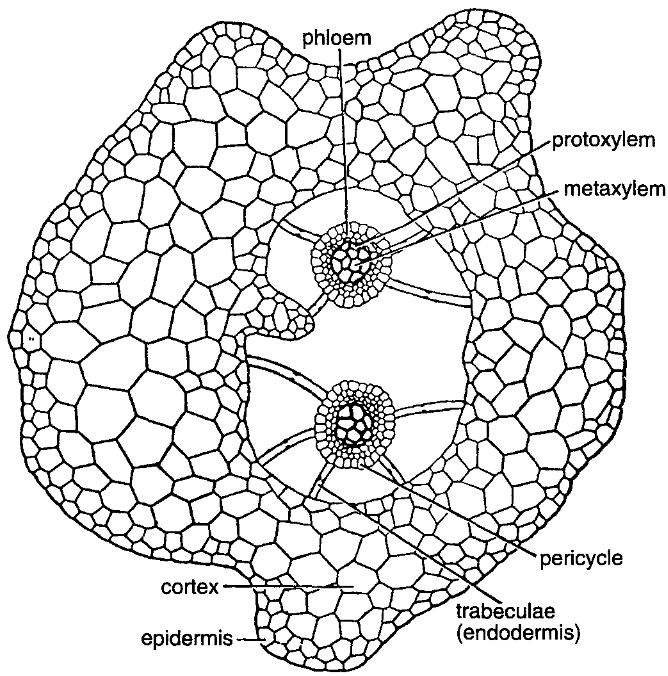


Fig. 8. ***Selaginella.*** T.s. stem—*S. kraussiana* (cellular).

Features of special interest

1. Presence of modified endodermis in the form of trabeculae.
2. Presence of more than one stele i.e. polystelic condition.

Exercise 5

***Object* : Study of anatomy of leaf.**

Work procedure

Cut a T.s. of leaf, stain in safranin-fast green combination, mount in glycerine and study.

Comments

1. The section shows a slightly bulged midrib in the centre and the wings.
2. It shows definite upper and lower epidermis, usually undifferentiated mesophyll and a central vascular bundle.
3. **The epidermis** is one layered. The stomata are generally present on the abaxial surface (lower) but may also be present on the adaxial surface (upper), or on both the surfaces.
4. **The mesophyll** is usually not differentiated into palisade and spongy parenchyma. It shows many conspicuous intercellular spaces.
5. The cells of mesophyll contain chloroplasts, each of which has several pyrenoid-like bodies.
6. **The vascular bundle** is concentric with xylem surrounded by phloem and is bounded by a bundle sheath.

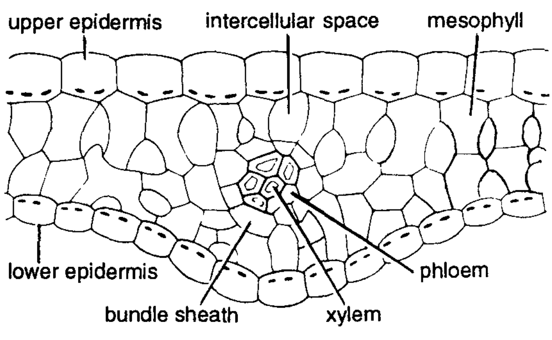


Fig. 9. *Selaginella.* T.s. leaf (a part cellular).

Exercise 6

***Object* : Study of spore producing organs.**

Work procedure

Study the external features of the strobilus. Cut L.s. of the strobilus, stain in safranin-fast green combination, mount in glycerine and study. (Alternatively study the slide of L.s. of the strobilus).

Comments

1. The spore producing organs are sporangia, aggregated in strobili which are generally present at the apices.
2. In some cases (as exemplified by *S. patula*) the axis may grow beyond the strobilus, terminating into a vegetative shoot or even in a second strobilus.
3. L.s. of the strobilus shows a strobilar axis, around which sporophylls are spirally arranged. Each sporophyll is ligulate and similar to a foliage leaf.
4. The sporangia are of two types, borne in the axils of the sporophylls, attached either strictly to the axils or to the axis just above.
5. *Selaginella* is heterosporous, with megaspores (large) and microspores (small), borne in their respective sporangia, known as megasporangia and microsporangia.

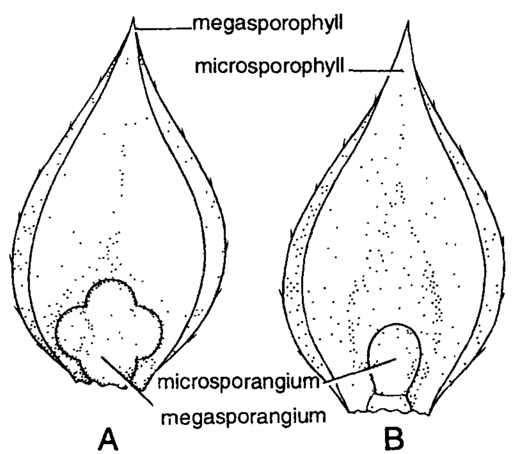


Fig. 10. *Selaginella.* Adaxial views of sporophylls showing sporangia; **A.** Megasporophyll, **B.** Microsporophyll.

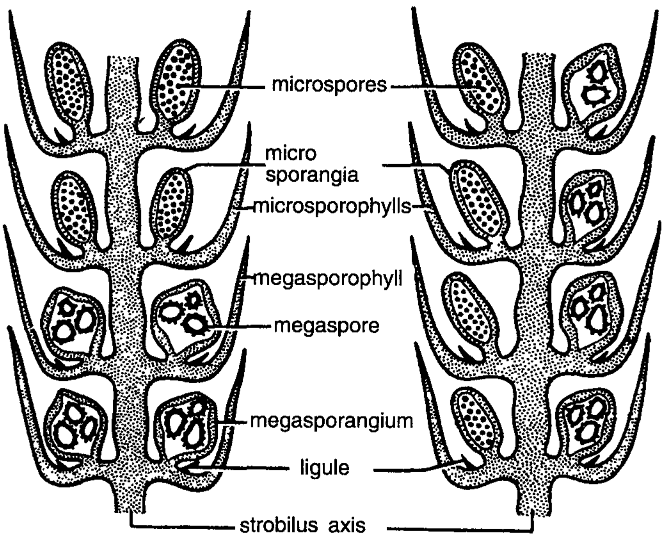


Fig. 11. ***Selaginella.*** L.s. strobilus showing different positions in which megasporangia and microsporangia occur.

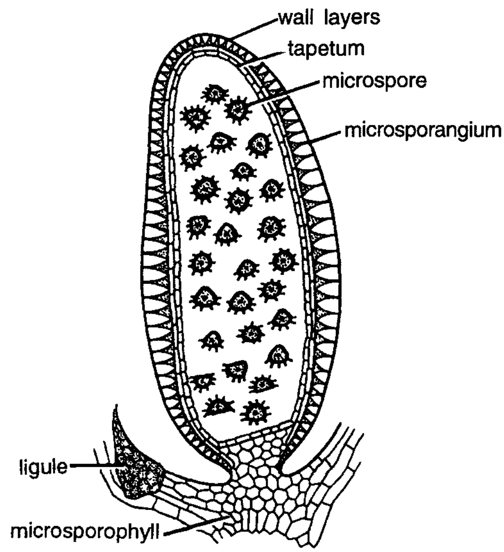


Fig. 13. ***Selaginella.*** L.s. microsporangium.

6. If a microsporangium is borne in the axil of the sporophyll, it is known as a microsporophyll but if it is a megasporangium, the sporophyll is termed as a megasporophyll.
7. Generally strobilus bears both types of sporangia but in *S. gracilis*, there are only one type of sporangia (either mega-or micro sporangia).
8. When both kinds of sporangia occur in one and the same strobilus, their arrangement differs from species to species:
   (i) In some species (e.g. *S. oregana*) there are only megasporangia on one side and only microsporangia on the other.

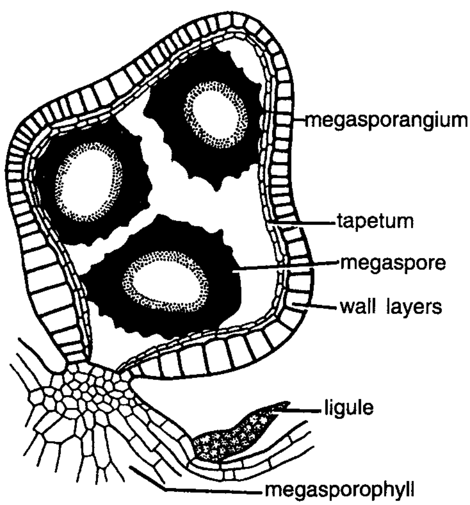


Fig. 12. ***Selaginella.*** L.s. megasporangium.

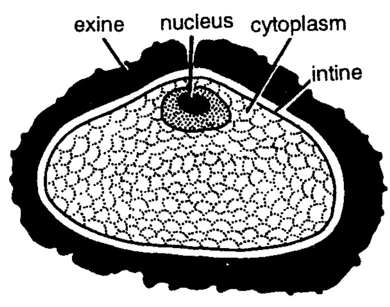


Fig. 14. ***Selaginella.*** A megaspore

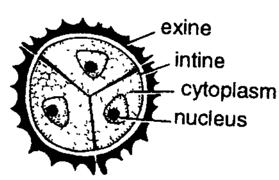


Fig. 15. ***Selaginella.*** Microspore tetrad.

(ii) In most of the species (e.g. *S. kraussiana*) there are only one or two megasporangia at the base and the rest are microsporangia.

9. Both types of sporangia are stalked and have two layered jackets. The outer layer of the jacket is chlorophyllous and has columnar cells, whereas the inner layer has tangentially elongated cells. It may form tapetum.
10. The cells of the outer jacket are thickened, except at the apex.
11. The two types of sporangia when ripe, differ in their size, form, structure and colour.
12. The megasporangium is much larger, four lobed, pale green or orange in colour and has only four megaspores.
13. The microsporangium is smaller with uniform outline. It is dark brown or red in colour and has many spores.
14. The megaspores are large in size and posses a triadiate ridge at its apex. It has thick sculptured exine and thin uniform intine.
15. The microspores are pyramidal in shape, and have thick, ornamented exine and a thin, uniform intine.
16. Both types of spores have a nucleus suspended in a rich cytoplasm.

Identification

Division—**Pteridophyta**. (1) Plant body differentiated into stem, roots and leaves, (2) A definite vascular strand present.
Sub-division—**Lycopsida**. (1) Leaves microphyllous, (2) Sporangia borne singly on the adaxial face of the sporophyll or in its axil, (3) Sporophylls borne in strobili.
Order—**Selaginellales**. (1) Each foliage leaf with a ligule at the base on adaxial side, (2) Sporophytes heterosporous.
Family—**Selaginellaceae**. (1) Stem herbaceous and dorsiventral or erect, (2) Gametophytes extremely reduced.
Genus—***Selaginella***. (1) Roots arise from rhizophore, (2) Trabeculae present, (3) Stele generally a protostele, sometimes siphonostele.

Hints for Collection

About 58 species of *Selaginella* have so far been reported from India. Many Indian species are found growing in western and eastern Himalayas and the hills of South India on damp shady sides. A few species are xerophytic. *S. oregana* is epiphyte on trunks and branches of moss covered trees in the forests.

Equisetum (Horse Tails)

Classification

Division	—	**Pteridophyta**
Sub-division	—	**Sphenopsida**
Order	—	**Equisetales**
Family	—	**Equisetaceae**
Genus	—	***Equisetum***

Exercise 1

Object : **Study of external morphology.**

Work procedure

Study the external features of the plant. Observe the differentiation of plant body into roots, rhizome, aerial branches and leaves. Note the ribbed nature of the stem, its branching and the scaly leaves. Also see the difference between sterile and fertile branches.

Comments

1. The plants are erect and bushy.
2. The plant is differentiated into roots, rhizome, aerial branches and leaves.
3. **The underground rhizome** has distinct nodes and internodes. The nodes bear aerial branches and roots.
4. **The roots** are produced on the lower side of the node. These are slender and fibrous.
5. **The aerial stems** are less than a metre in height with characteristic joints. Stem is rough due to the deposition of silica.
6. **The aerial branches** fall into two general categories—(i) typical sterile branches which are green and branched, and (ii) typical fertile branches which are non-green, unbranched and terminate in a cone. Such branches die after the spores are shed.
7. Some species have green, branched fertile shoots, with a cone at the apex of each lateral branch. Such branches do not die after the spores are shed.
8. Organization of the rhizome and aerial branch is the same, but is best seen in aerial branches.

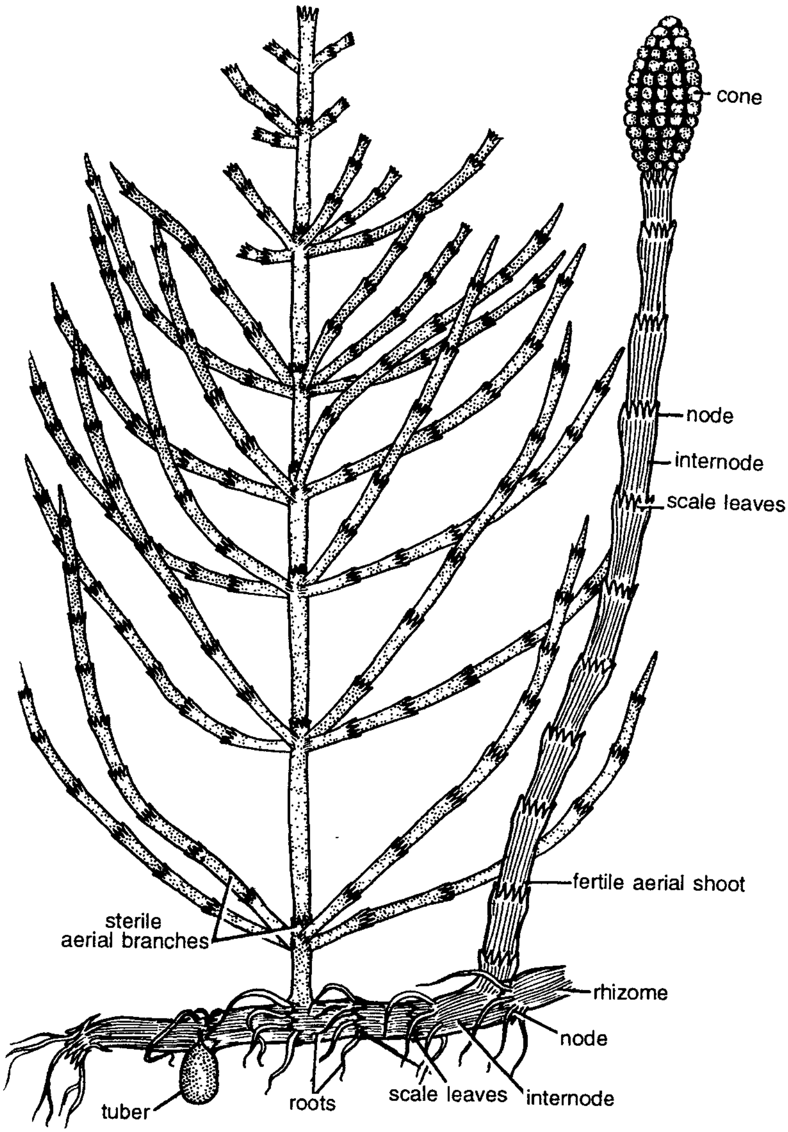


Fig. 1. ***Equisetum.*** External features.

9. **Each internode** of an aerial branch is longitudinally ribbed. The number of ridges is same as the number of leaves, and each leaf stands directly above a ridge present in the internodes below.
10. The ridges on the stem of successive internodes alternate, as also the leaves of the successive nodes.
11. **Leaves** are simple, small, scaly, whorled and fused laterally and possess longer or shorter free tips.
12. Leaves are present at nodes in whorls. Each whorl forms a sheath closely appressed to the node. The number of leaves in a whorl varies with the species and the size of the stem.
13. The leaves are non-chlorophyllous and scaly. These alternate at the successive nodes.
14. **The branches** develop at the node in between each two leaves. Therefore, the branches are equal in number to the leaves and appear to arise in a whorl.

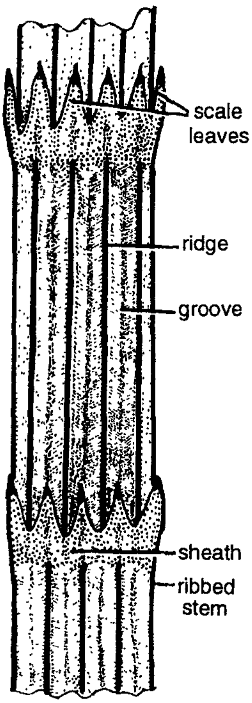


Fig. 2. ***Equisetum.*** A part of stem showing alternation of ridges and grooves and scale leaves.

Exercise 2

Object : **Study of anatomy of root.**

Work procedure

Cut a T.s. of the root, stain in safranin-fast green combination, mount in glycerine and study.

Comments

1. The section appears almost circular in outline.
2. **Epidermis** is single layered and possesses a few root hairs.
3. **The cortex** is often divided into an outer cortex and an inner cortex.
4. **The outer cortex** is a few layered deep. It is made of thick walled cells.
5. **The inner cortex** is also a few layered deep. The cells are large sized and parenchymatous with intercellular spaces.
6. **Endodermis** separates from the vascular tissues. It is two layered—outer and inner endodermis. The pericycle is absent.

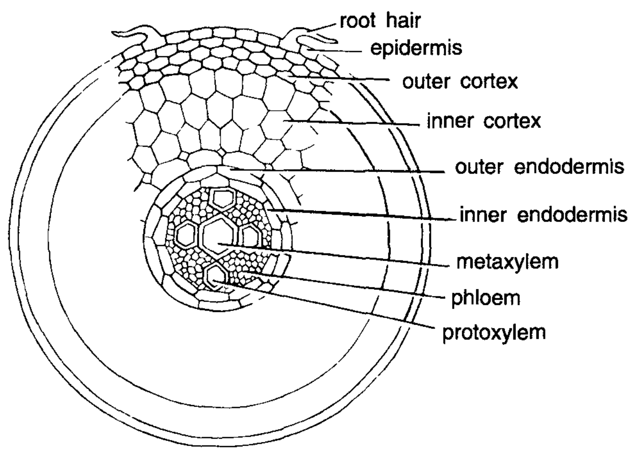


Fig. 3. ***Equisetum.*** T.s. root (a part cellular).

7. **The vascular bundle** shows a single, large metaxylem element in the centre with 3 to 4 protoxylem, triarch to tetrarch elements surrounding it. The number of protoxylem groups increases with increase in the diameter of the root.
8. The angles between the protoxylem are occupied by phloem.

Exercise 3

Object : **Study of anatomy of internode of aerial shoot.**

Work procedure

Cut a T.s. of the aerial shoot passing through the internode, stain in safranin-fast green combination, mount in glycerine and study.

Comments

1. The outline is wavy with ridges and grooves.
2. The tissues are organised into epidermis, cortex, stele and a pith cavity.
3. **The epidermis** is cuticularized with tangentially elongated and silicified cells.
4. **The stomata** are mostly found in the grooves. The guard cells are surrounded by two subsidiary cells, one on either side.
5. **Cortex** follows the epidermis and is highly differentiated. It is divided into outer and inner cortex.

6. **Outer cortex**, below the ridges has a group of sclerenchyma. Small patches of sclerenchyma may also occur, below the grooves.
7. Beneath the ridges radially elongated chlorenchymatous cells (palisade tissue) are present. The amount of palisade beneath the grooves is lesser.
8. **The inner cortex** is composed of large and thin-walled, parenchymatous cells.
9. **Vallecular canals** are present in the cortex. These are situated below the grooves.
10. **The stele** is an ectophloic siphonostele that consists of ring of vascular bundles.
11. **Endodermis** occurs at different positions in different species.
    (i) Most commonly, the endodermis forms a simple sheath, outside the ring of bundles.
    (ii) In some cases, in addition, there is also an internal endodermis and outer endodermis dips in between the bundles.
    (iii) In third condition, each bundle is surrounded by an individual endodermis.
12. **Pericycle** lies below the endodermis.
13. **The vascular bundles** are collateral and endarch, arranged in a ring and each bundle lies below each ridge.
14. Each bundle has one inner strand of protoxylem and two outer of metaxylem.

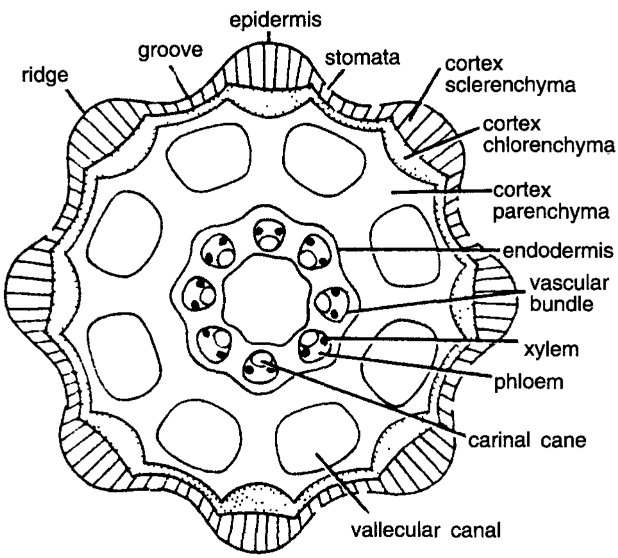


Fig. 4. ***Equisetum.*** T.s. aerial shoot : internode (diagrammatic).

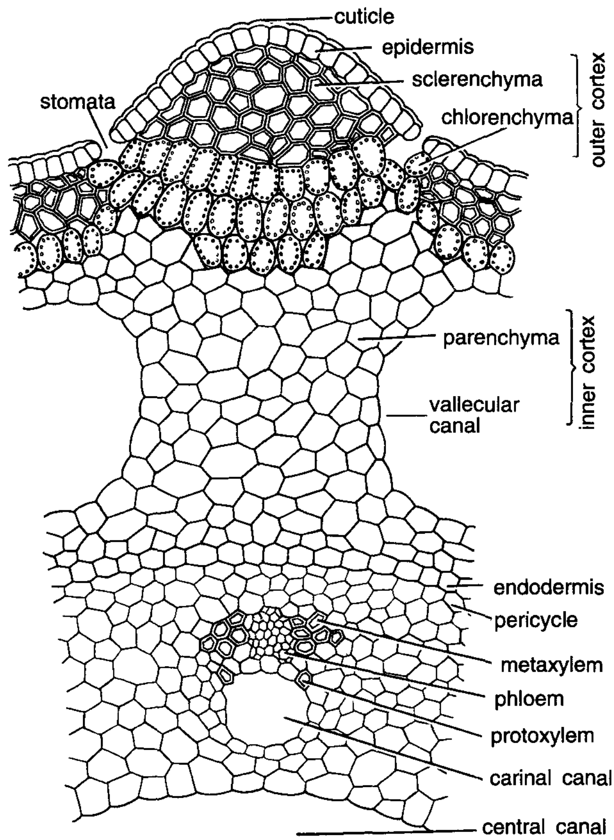


Fig. 5. ***Equisetum.*** T.s. aerial shoot : internode (a part cellular).

15. **The protoxylem** elements lie on the sides of a protoxylem lacuna, the carinal canal, formed by the disintegration of protoxylem elements.
16. **The two metaxylem** groups lie on two lateral sides of carinal canal (i.e. on the shoulders of the bundle).
17. The rest of the tissue of the vascular strands is parenchymatous.
18. Pith cavity known as central canal lies in centre.

Features of special interest

Anatomy shows both xerophytic as well as hydrophytic characters.

Xerophytic characters

(1) Presence of ridges and grooves.
(2) Position of stomata in grooves.
(3) Thick cuticle over epidermis.
(4) Well developed sclerenchyma below the ridges.
(5) Presence of palisade.

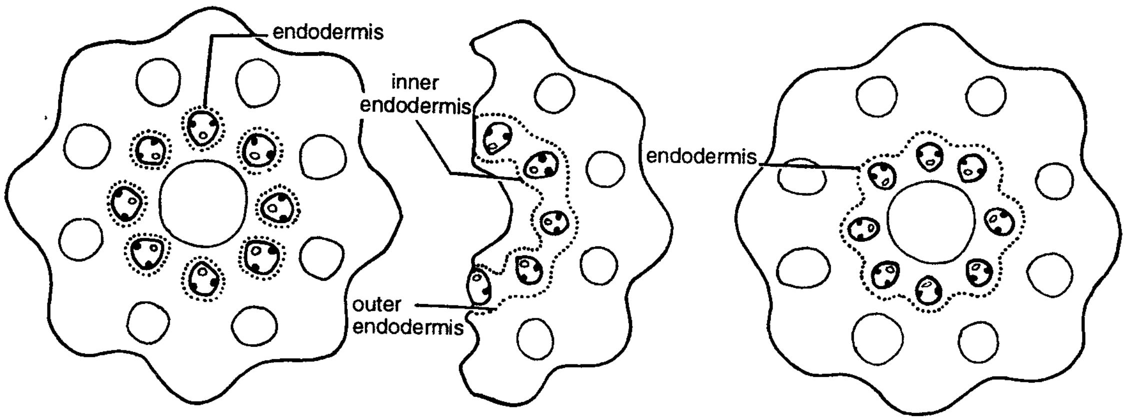


Fig. 6. ***Equisetum.*** T.s. aerial shoot : internode showing different conditions of endodermis.

Hydrophytic characters

(1) Presence of vallecular, carinal and central canals.

Exercise 4

***Object* : Study the anatomy of node of aerial shoot.**

Work procedure

Cut a T.s. of the aerial shoot passing through the node, stain in safranin-fast green combination, mount in glycerine and study.

Comments

1. The section shows distinct ridges and grooves.
2. The anatomy is almost similar to that of internode except for a few differences.
3. The section shows epidermis, cortex, stele and nodal diaphragm instead of pith cavity in internode.
4. **The epidermis** is the outermost thickly cuticularised layer.
5. **The cortex** is divisible into outer, middle and inner cortex.
6. **The outer cortex** is sclerenchymatous. It is followed by middle cortex made of palisade (chlorenchyma) tissues.
7. **The inner cortex** is parenchymatous and occupies most part of the section.
8. **Vallecular canals** are absent. Many leaf traces and branch traces are found scattered all over the inner cortex.
9. **Vascular bundles.** Instead of ring there is a complete vascular cylinder with outer ring of phloem enclosing a ring of xylem.
10. **Leaf and branch traces** are given off from the vascular cylinder. Leaf traces arise beneath the ridges and do not produce leaf gaps in the vascular cylinder. Branch traces arise beneath the grooves.
11. **Nodal diaphragm**. In the centre there is parenchymatous or sclerenchymatous tissue. It is known as nodal diaphragm. In L.s. it appears like an arc. The internodes easily break and separate at these places.

Exercise 5

***Object* : Study of anatomy of rhizome.**

Work procedure

Cut a T.s. of rhizome, stain in safranin-fast green combination, mount in glycerine and study.

Comments

1. The outline is wavy with ridges and grooves.
2. **Epidermis.** This is the outermost thickly cuticularised layer. Stomata are absent.
3. **The cortex** consists of a few layers of sclerenchyma just below the epidermis and a large zone of parenchyma spread upto the ring of vascular bundles.
4. **Large vallecular canals** are present in the parenchymatous cortex below the grooves.

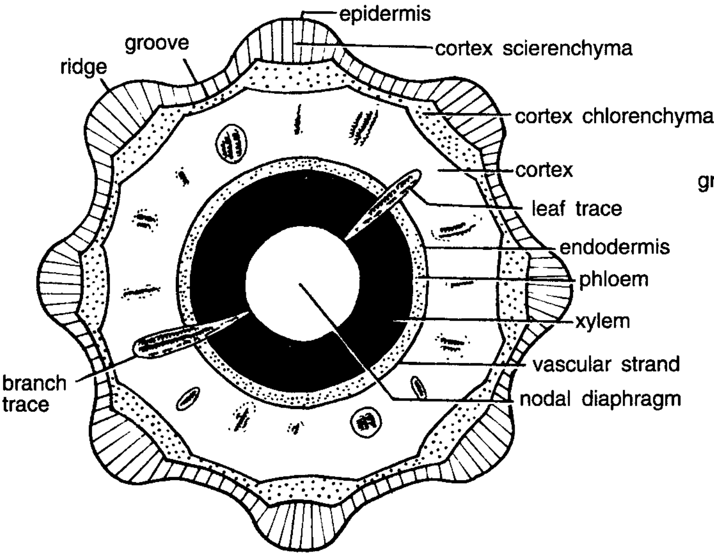


Fig. 7. ***Equisetum.*** T.s. aerial shoot : node (diagrammatic).

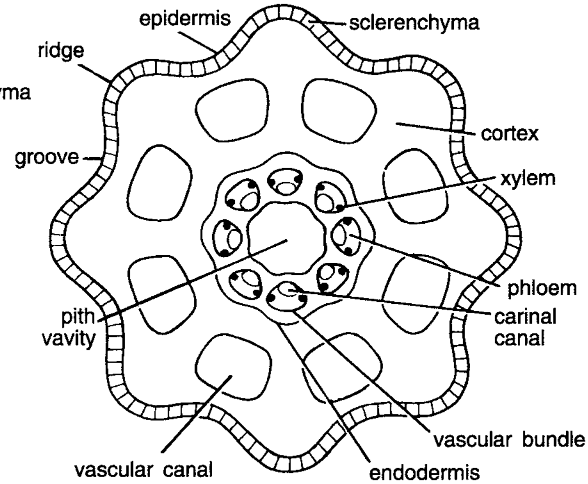


Fig. 8. ***Equisetum.*** T.s. rhizome : internode (diagrammatic).

5. **Endodermis** is single layered and encloses a ring of vascular bundles.
6. **Each bundle** is located below the ridge.
7. The bundle is conjoint, collateral and endarch.
8. The bundle has a large protoxylem lacuna, carinal canal.
9. **Pith cavity**. The centre has a large cavity, called pith cavity.

Exercise 6

Object **: Study of spore producing organs : L.s. cone.**

Work procedure

The spore producing organs are sporangia borne in cones, generally terminating the main axis and sometimes the lateral branches. The structure is best studied by observing L.s. of cone, single sporangiophore and spores. Study the features shown by respective slides.

Comments

1. L.s. of the cone shows cone axis and attached sporangiophores.
2. Cone axis is centrally located.
3. It bears sporangiophores in whorls which are mostly alternate though not regularly.

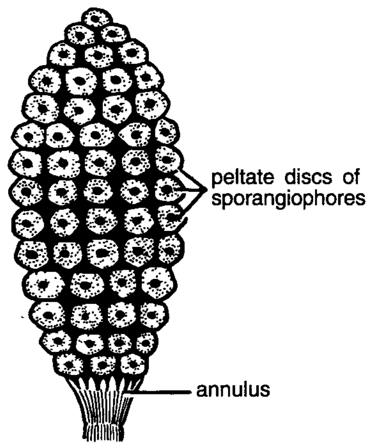


Fig. 9. ***Equisetum.*** A cone.

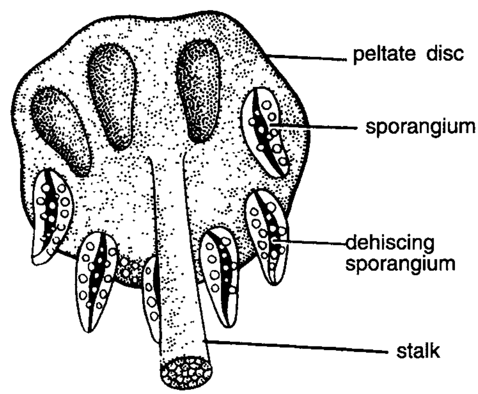


Fig. 10. ***Equisetum.*** Sporangiophore from ventral side.

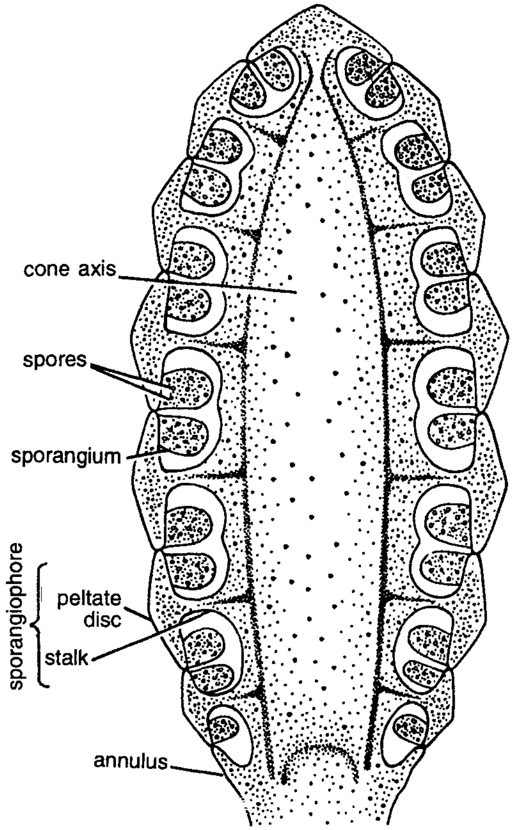


Fig. 11. ***Equisetum.*** L.s. cone

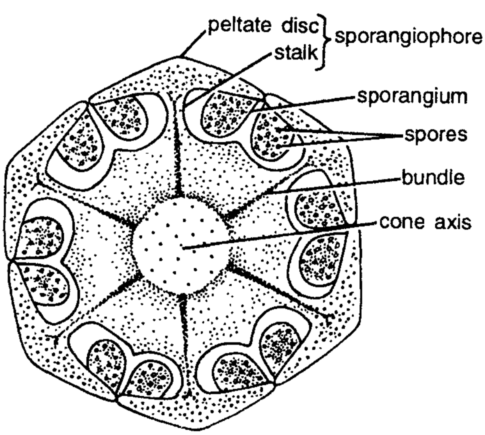


Fig. 12. ***Equisetum.*** T.s. cone.

4. At the base of the cone is a calyx-like whorl, the annulus (which most probably represents a modified leaf whorl).
5. The sporangiophores are attached to the cone axis at right angles with its stalk.
6. The stalk holds a polypogonal peltate disc at right angles to it. The peltate discs of sporangiophores fit closely to form a protective cover for the sporangia below.
7. Sporangia appear attached on the lower side of the disc.
8. Each sporangium is elongated and sac-like. It has one-layered jacket that encloses numerous spores.

Exercise 7

Object **: Study of spore producing organs : T.s. cone.**

Work procedure

Study the characters of the cone by observing various features as shown by the slide of T.s. cone.

Comments

1. T.s. of cone shows a cone axis and sporangiophores attached to it.
2. Centrally located part is called cone axis.
3. Sporangiophores are attached in a whorl.
4. Each sporangiophore consists of a stalk and a disc.
5. Stalk keeps the disc attached to cone axis.
6. The peltate disc bears sporangia on the underside, with one layered jacket which enclose the spores.
7. Each sporangium appears elongated and cylindrical.
8. Sporangiophore is one of the units, of which cone is made of.
9. These are attached to the central cone axis in successive whorls.
10. Each sporangiophore consists of a stalk and a polygonal peltate disc.
11. The stalk is attached to the cone axis on one side and to the peltate disc on the other.
12. About 5-10 cylindrical sporangia are arranged in a ring near the margins on the lower side of the disc.
13. Sporangium has a one layered jacket with helical thickenings.
14. Numerous spores, all similar (homosporous condition) are present in the sporangial cavity.
15. A longitudinal line of dehiscence is also clearly seen.

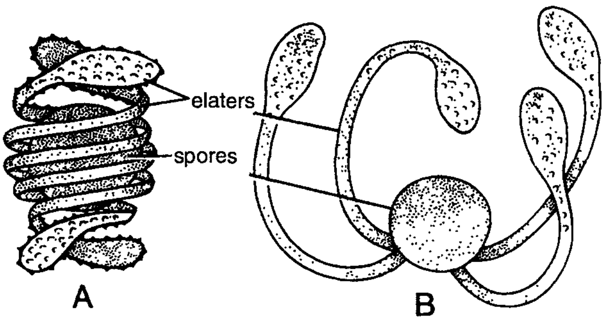


Fig. 13. ***Equisetum.*** Spores. **A.** Spore with elaters coiled, **B.** Spore with elaters uncoiled.

Exercise 8

***Object* : Study of spores.**

Work procedure

Tease a sporangium with needle. Mount spores in water. These can also be stained with safranin and then studied.

Comments

1. The spores consist of a four layered wall.
2. Surrounding the two usual wall layers, there is a third cuticular layer known as the middle layer and a fourth, thick, outermost layer known as perispore.
3. The perispore of each spore is differentiated into four narrow spirally wound bands, with flat-spoon tips, all attached at the common point.
4. These projecting bands are called as "elaters", but are very different from the elaters of bryophyta.
5. The elaters are hygroscopic and with the changes in the atmospheric humidity, they coil and uncoil. (this can be observed under the microscope by allowing the wet spores to dry on a slide).
6. Each spore in a section shows a single nucleus with rich cytoplasm and all the four wall layers.

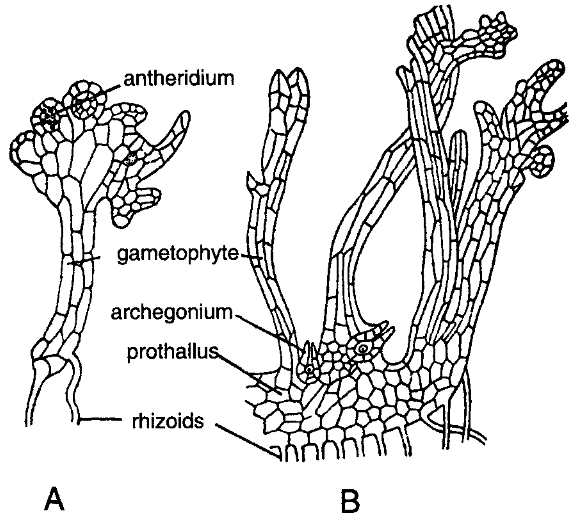


Fig. 14. ***Equisetum.*** Prothalli. **A.** Prothallus with antheridia, **B.** Prothallus with archegonia.

Exercise 9

***Object* : Study of prothallus.**

Work procedure

Study a prepared slide of prothallus.

Comments

1. Both male (antheridia) and female (archegonia) sex organs are borne on the same prothallus. Thus it is monoecious.
2. In younger stages only antheridia are developed. Therefore, small and younger prothalli show antheridia only. In older prothalli, however, archegonia are found. Hence it is protandrous.
3. The multicellular central part gives out many flat branches. These branches further get irregularly dissected in uniseriate filaments.
4. From the central region, long, brown and unbranched rhizoids are also given off.
5. Archegonia remain embedded in the tissue of the prothallus, at the place where branches are given out.

6. Prothallus that bears antheridia, is less branched and smaller. Antheridia may arise at the base of the branch or on the branch itself.

Identification

Division—**Pteridophyta**. (1) Plant body differentiated into stem, roots and leaves, (2) A definite vascular strand present.
Sub-division—**Sphenopsida**. (1) Stem branched, articulated, ridged and furrowed with distinct nodes and internodes, (2) Leaves microphyllous, small, scaly and in whorls at nodes.
Order—**Equisetales**. (1) Stem branched. Branches borne in transverse whorls, (2) Internodes alternate with one another, (3) Vascular cylinder endarch, siphonostele.
Family—**Equisetaceae**. (1) Homosporous. (2) Sporangia borne on sporangiophores which form a compact cone, (3) No secondary growth.
Genus—***Equisetum***. (1) Leaves scaly and colourless, (2) Sunken stomata in grooves, (3) Presence of palisade in the stem, (4) Presence of valecular, carinal and central canals.

Hints for Collection

Equisetum debile which is common in India grows abundantly along the banks of rivers, in sandy soil and on the woods along the river. Another common species, *E. arvense,* grows in grasslands.

Adiantum (Maiden Hair Fern)

Classification

Division	—	**Pteridophyta**
Sub-division	—	**Pteropsida**
Class	—	**Leptosporangiatae**
Order	—	**Filicales**
Family	—	**Adiantaceae**
Genus	—	***Adiantum***

Exercise 1

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

Work procedure

Study a fresh plant or a preserved specimen for external morphology. Observe the differentiation of plant body into roots, rhizome and leaves. Note the circinate vernation of young leaf, compound nature of mature leaf and reflexed margins of the leaflets.

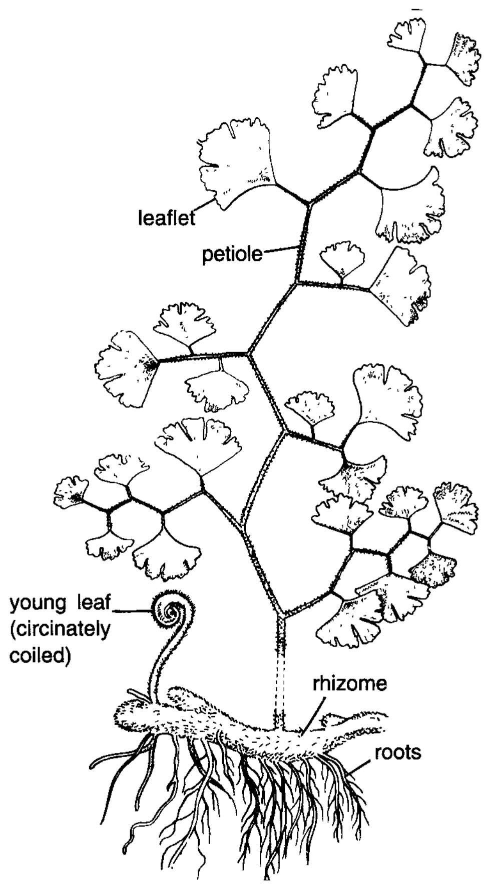


Fig. 1. ***Adiantum.*** External features.

Comments

1. **Plant body** is a sporophyte. It is differentiated into roots, rhizome and leaves.
2. **Roots** are produced on the lower side of the rhizome. Primary root is short-lived. Secondary roots are adventitious and branched.
3. **Rhizome** may either be creeping or erect. It is scaly, covered with hairs and bears adventitious roots and leaves.
4. **Leaves** with a long petiole, are spirally or alternately arranged on the rhizome. Young leaves are circinately coiled. Young rhizome, petiole, rachis and circinately coiled leaves are covered with hairs known as ramenta.
5. The leaves are compound and are borne on shining black and brittle petiole.

6. Leaves are often dichotomously branched into many leaflets.
7. The blade of the leaflets may be entire, and either simply or repeatedly branched. The leaflets are deltoid in shape. When fertile, the leaflet margin remains folded toward the lower side forming a false indusium which encloses many sori.
8. **Leaflet** is traversed by dichotomously branched veins which generally do not unite to form a reticulum. The venation is, therefore, of open dichotomous type.

Exercise 2

***Object* : Study of anatomy of root.**

Work procedure

Cut a T.s. of the root, stain with safranin-fast green combination, mount in glycerine and study.

Comments

1. The root is almost circular in outline.
2. It is differentiated into epidermis, cortex and stele.
3. **The epidermis** is single layered, cells are thin walled and tangentially elongated. It bears a few unicellular root hairs.
4. **Cortex** is inner to the epidermis. It is multilayered and parenchymatous.

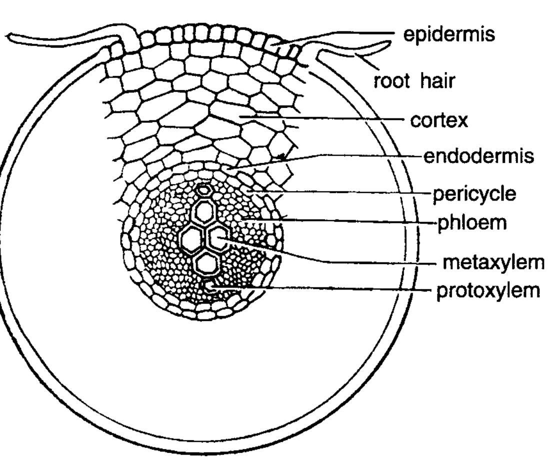


Fig. 2. *Adiantum.* T.s. root (a part cellular).

5. **Endodermis** is single layered and is followed by a single layered pericycle.
6. **A protostele** is present in the centre. It is diarch and exarch.
7. **Xylem** elements occur in the centre. Of these, metaxylem elements are present in the centre. Protoxylem groups are situated on the two opposite sides of the metaxylem (Thus xylem groups are exarch and diarch).
8. **Phloem** surrounds the xylem on all the sides.

Exercise 3

***Object* : Study of anatomy of rhizome.**

Work procedure

Cut a T.s. of rhizome, stain in safranin-fast green combination, mount in glycerine and study.

Comments

1. Rhizome appears almost circular or gutter-shaped in a transection.
2. It shows differentiation into epidermis, hypodermis, ground tissue and stele.
3. **Epidermis** is single layered and bears numerous multicellular hairs.
4. **Hypodermis** that follows epidermis is a 2-3 layered deep and sclerenchymatous.
5. **Ground tissue** occupies major part of rhizome. It is parenchymatous and many layered deep.

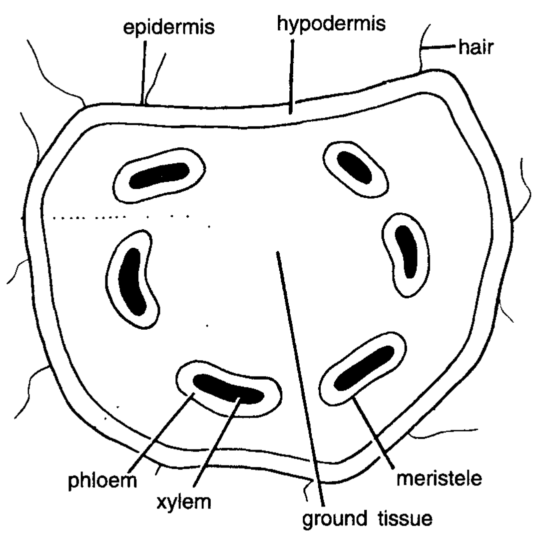


Fig. 3. *Adiantum.* T.s. rhizome (outlines : diagrammatic).

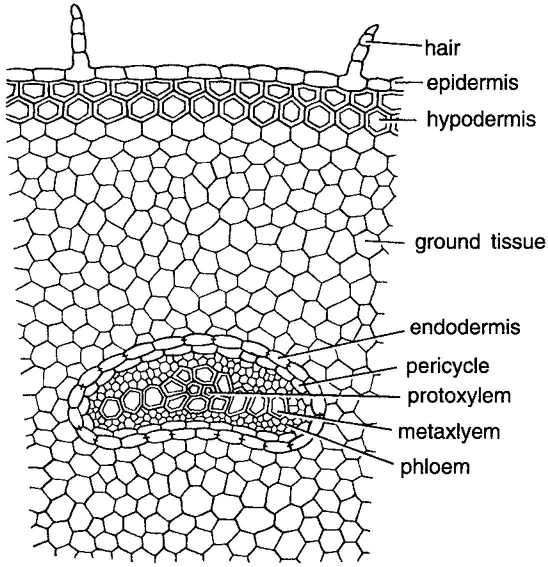


Fig. 4. ***Adiantum.*** T.s. rhizome (part shown by dotted lines in Fig. 3 : cellular).

6. **Stele** is variable in nature, differing from one region to another. Species with elongated rhizome (*A. pedatum and A. hispidulum*) show actual solenostele (amphiphloic siphonostele). But commonly the stele is gutter shaped and appears as several meristeles arranged in a ring, due to numerous leaf gaps (dictyostele).
7. **Meristeles** varying in number, but more often 5-7, lie arranged in ground tissue in a gutter-shaped ring, thus exhibiting dictyostelic condition. The spaces between neighbouring meristeles are leaf gaps.
8. Each of the meristeles is surrounded by a distinct, single-layered endodermis subsequently followed by a single layered pericycle.
9. **Xylem** elements occupy the central part. Metaxylem and protoxylem are arranged in a way to form mostly mesarch condition.
10. **Phloem** surrounds xylem.

Exercise 4

Object : **Study of anatomy of rachis.**

Work procedure

Cut a T.s. of rachis, stain with safranin-fast green combination, mount in glycerine and study.

Comments

1. Tissues of the rachis are differentiated into epidermis, hypodermis, cortex and stele.
2. **Epidermis** is a single layer of cells, covered by a cuticle.
3. **Hypodermis** follows the epidermis. It is few layered deep. The cells are sclerenchymatous.
4. **Cortex** forms larger part of the rachis. It is made of many layers of parenchyma.

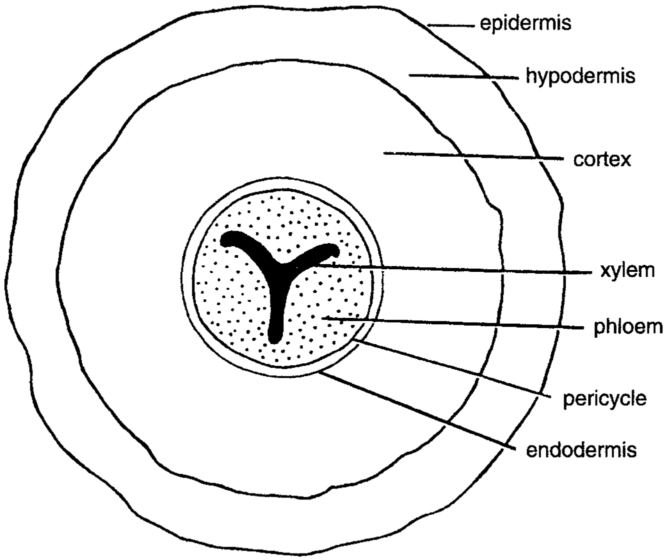


Fig. 5. ***Adiantum.*** T.s. rachis (outlines : diagrammatic).

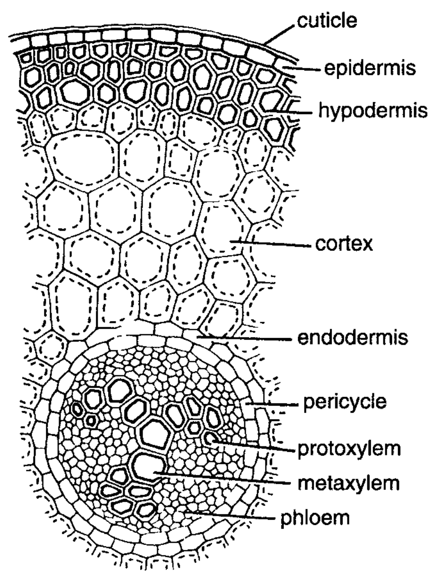


Fig. 6. ***Adiantum.*** T.s. rachis (a part cellular).

5. **Stele** is a protostele.
6. **Endodermis** is single layered and is followed by a pericycle.
7. **The xylem** group is almost Y-shaped. Protoxylem elements are situated at the tips of three free ends; while the rest of the part is occupied by metaxylem.
8. **Phloem** surrounds the xylem on all the sides.

Exercise 5

Object : **Study of anatomy of leaflet.**

Work procedure

Cut a T.s. of leaflet by keeping a leaf in pith. Stain in safranin-fast green combination, mount in glycerine and study.

Comments

1. Leaflet is differentiated into upper and lower epidermis, mesophyll, sclerenchyma and vascular bundle.
2. **Epidermis.** The cells of the upper and lower epidermis possess chloroplast. The lower epidermis is frequently interrupted by stomata.
3. **Chloroenchyma.** Just above the lower epidermis lies a single layer of compactly arranged cells containing numerous chloroplasts.
4. **Mesophyll.** Following this compact layer, mesophyll tissue extends up to the upper epidermis. It is undifferentiated into palisade and spongy parenchyma but is composed of loosely arranged spongy parenchyma only.
5. **Vascular bundle.** Each vascular bundle is surrounded by a thick sclerenchymatous sheath.

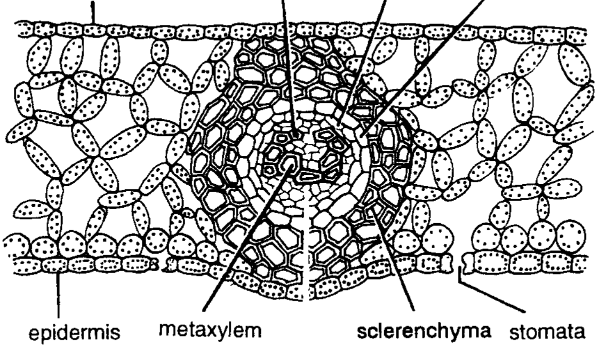


Fig. 7. ***Adiantum.*** T.s. leaflet (part cellular).

6. **Endodermis.** Surrounding the vascular bundle is endodermis followed by a pericycle.
7. **Xylem.** Centrally located xylem has protoxylem groups, facing towards the adaxial surface of the leaf.
8. **Phloem** surrounds xylem.

Exercise 6

Object : **Study of structure of sorus.**

Work procedure

Since the sori are present on the lower reflexed side of the leaflet, sections of this part are cut to study the arrangement and structure of sorus. The sections are stained in safranin-fast green combination, mounted in glycerine and studied.

Comments

1. The spore producing organs are the sporangia grouped in sori. Each sorus is mixed in nature and shows sporangia of different ages.
2. Sori are present, along both the sides of the veins, on the dorsal side of the marginal reflexed lobe. This part remains folded towards the lower side and acts as a false indusium which is membranous and brown coloured.
3. Lower part of each leaflet shows many such reflexed lobes along its margins. The reflexed part is traversed by veins in continuation with those in the unfolded part.

To study the relation of the folded part with the unfolded, cut a L.s. of leaflet in folded condition.

1. It shows the upper part of the leaflet possessing vascular bundles cut longitudinally and the reflexed or folded margin on its lower side.
2. This reflexed part is a portion of the leaflet and bears sporangia in sori, thus forming a false indusium.
3. Many sporangia are seen arranged in groups.
4. Sporangia are attached to the indusium by their long slender stalks.

To study the relation between veins, sori and sporangia with indusium, unfold the reflexed lobe and cut its T.s.

1. Reflexed lobe of leaflet covers sori and is called false indusium.

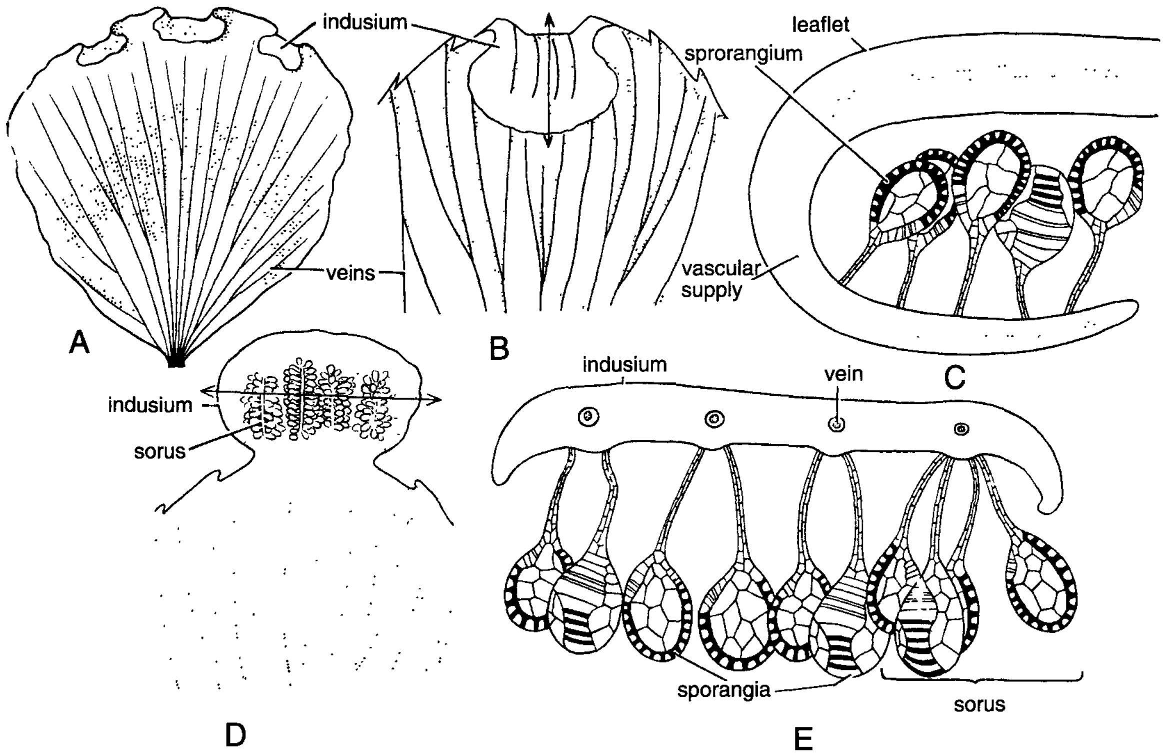


Fig. 8. ***Adiantum.*** **A.** Surface view of leaflet showing reflexed margin (indusium) and venation. **B.** Magnified portion of leaflet showing single reflexed lobe (indusium). **C.** L.s. leaflet through indusium (plane shown by arrow in Fig. B) **D.** Unfolded reflexed lobe (indusium) showing sori on both the sides of vein. **E.** T.s. through indusium (plane shown by arrow in Fig. D).

2. Veins traverse the indusium but do not form reticulum (open dichotomous venation).
3. Along both the sides of each vein are many sporangia, attached by their long and slender stalks. This group of sporangia is called a sorus and many such sori are situated along each of the veins.

Exercise 7

Object **: Study of structure of a sporangium and a spore.**

Work procedure

Study the preparation of section of leaflet showing sori or unfold the reflexed lobe of a leaflet, tease out a few sporangia, also crush open a few of these sporangia to release spores. Stain with safranin, mount in glycerine and study.

Comments

1. Each sporangium is attached to the indusium by a slender, long and multicellular stalk.
2. The sporangium (capsule) is oblong in shape and borne at the tip of the stalk.
3. The wall of the sporangium is made of thin-walled cells.
4. Some cells of this wall lying in a vertical row are characteristically thickened on their radial and inner tangential walls. These together are called as annulus.
5. On one side of the annulus are a few (2-3) thin walled cells forming a stomium wherefrom dehiscence of the sporangium takes place.
6. The wall encloses many spores inside.
7. Each spore is a double walled structure. The outer layer is exine which is thick and ornamented. The inner layer, called intine, is thin and smooth.

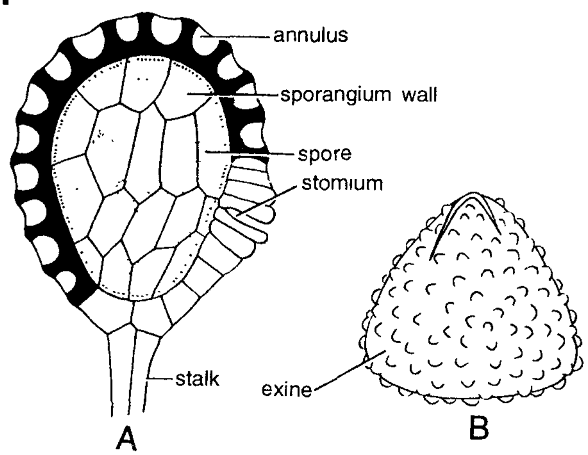


Fig. 9. *Adiantum.* A. A sporangium. B. A single spore.

8. It has a single big nucleus, surrounded by cytoplasm.

Exercise 8

Object : **Study of structure of prothallus.**

Work procedure

Mount young prothallus, stain in fast green, mount in glycerine and study.

Comments

1. It is formed after the germination of a spore and is thus a gametophytic structure.
2. It is leafy and heart-shaped.
3. It consists of a single layer of cells, one cell in thickness, except in the central region where apical notch is situated.
4. Many unicellular rhizoids are given out from the ventral surface.
5. Antheridia are located in the posterior part of prothallus away from the apical notch.
6. Archegonia lie near the apical notch, on the thickened, central apical cushion.
7. Parts of antheridia and necks of archegonia protrude outside the general surface of the prothallus.
8. All the cells of the prothallus are thin walled and bear many discoid chloroplasts.

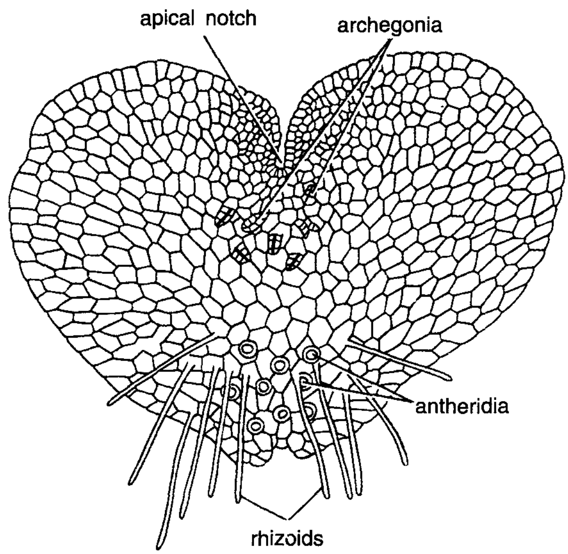


Fig. 10. *Adiantum.* Prothallus bearing young leafy sporophyte.

Exercise 9

Object : **Study of old prothallus with sporophyte.**

Work procedure

Mount an old prothallus that bears a sporophyte stain in fast green and mount in glycerine.

Comments

1. Sporophyte is formed as result of fertilization.
2. Gametophyte (prothallus) still persists.
3. Young sporophyte is differentiated into young leaves, primary root and secondary roots.
4. Leaves stand erect and appear near the apical notch. They are simpler than the mature leaves. They may also be circinately coiled.
5. Primary root grows on the lower side and gives out secondary roots.

Identification

Division—**Pteridophyta**. (1) Plant body differentiated into stem, roots and leaves, (2) A definite vascular strand present.
Sub-division—**Pteropsida**. (1) Vascular cylinder siphonostelic, with leaf gaps, (2) Plants macrophyllous, leaves compound, with rachis, (3) Leaves bear sporangia in sori, (4) Gametophytes small, green and free living.

Class—**Leptosporangiatae**. (1) Sporangium with a jacket layer one cell in thickness, (2) Definite number of spores.
Order—**Filicales**. (1) Sori are mixed, (2) Homosporous.
Family—**Adiantaceae**. (1) Sori marginal, (2) Indusium oblong or linear, formed of the more of less changed and reflexed margin of the frond. opening inwardly.
Genus—***Adiantum***. (1) Sori apparently marginal, but superficial in origin, (2) Indusia globose to linear, usually many and distinct, (3) Leaflet margins bearing sori are sharply reflexed, (4) Open dichotomous venation of the leaflet.

Hints for Collection

Adiantum is very common in hills, lower slopes of the hills and in the plains. In hiils it is commonly seen in dense evergreen forests and also on limestone rocks. In plains, it can easily be collected from moist places such as banks of rivers, etc. It is also seen growing on the inner walls of the wells.

Nephrolepis (Sword Fern)

Classification

Division	—	**Pteridophyta**
Sub-division	—	**Pteropsida**
Class	—	**Leptosporangiatae**
Order	—	**Filicales**
Family	—	**Polypodiaceae**
Genus	—	***Nephrolepis***

Exercise 1

***Object :* Study of external morphology.**

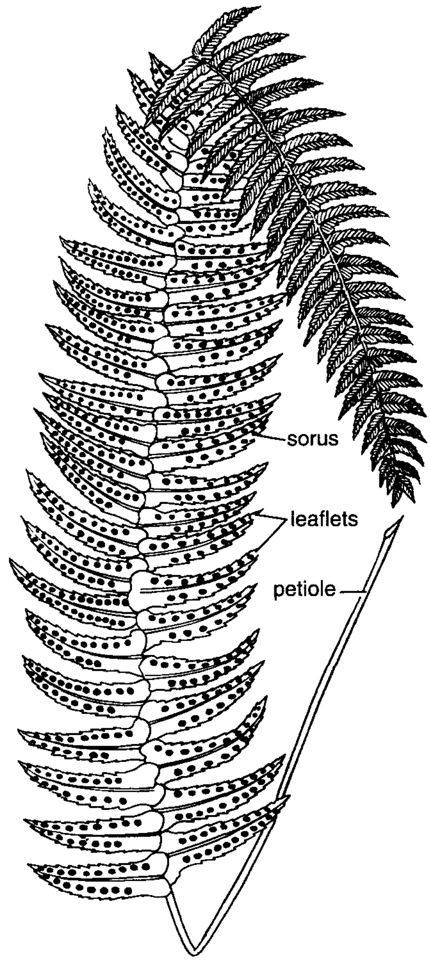


Fig. 1. ***Nephrolepis.*** External morphology.

Work procedure

Study the characters of roots, rhizome and leaves of a potted plant or a museum specimen.

Comments

1. The plant body is a sporophyte. It is differentiated into roots, rhizome and leaves.
2. The rhizome gives out adventitious roots from its underside. These adventitious roots are small and branched.
3. The stem is modified to rhizome. It is subterranean, short and erect. The rhizome produces elongated slender stolons. Peltate scales cover the rhizome.
4. In *N. tuberosa*, rhizome bears tubers. These are reservoirs of carbohydrates and water.
5. The leaves are long, narrow, sub-coriaceous and unipinnate.
6. The pinnae are sessile or shortly petioled. They have a usually rounded or cordate base. Each pinna has articulation with a pouch-like structure at the base.
7. The veins are prominent and the veinlets are branched with open ends. The tips of veinlets are gland dotted and they extend up to the margins.

Exercise 2

***Object* : Study of anatomy of the root.**

Work procedure

Cut a thin transverse section of the root. Stain with safranin-fast green combination. Mount in glycerine and study.

Comments

1. **The outline** of the section is almost circular.
2. The tissues are differentiated into—epiblema, cortex and the vascular cylinder.
3. **Epiblema** is the outermost single layer of cells. The cells are unicellular and thin walled. A few root hairs are produced by this layer.
4. **Cortex** forms the major part of the section. It shows outer parenchymatous region and the inner small sclerenchymatous region.
5. **Endodermis** follows the cortex and separates it from the vascular tissues. The cells of endodermis show casparian strips.
6. **Pericycle.** Endodermis is followed by 1 or 2 layered parenchymatous pericycle.
7. **Vascular cylinder** is represented by a radial, diarch and exarch vascular bundle.

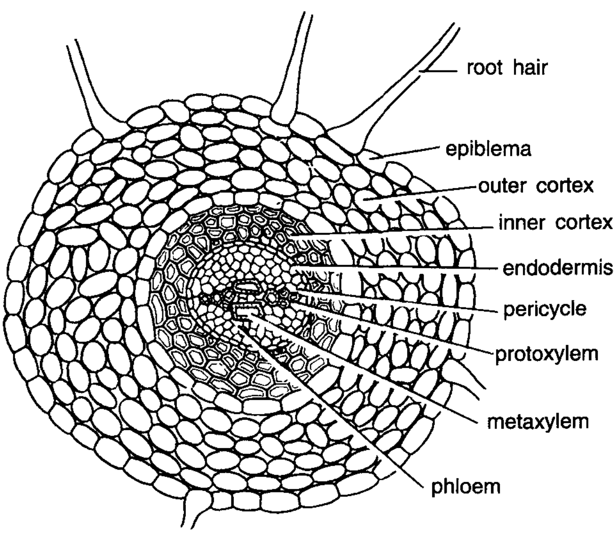


Fig. 2. ***Nephrolepis.*** T.s. root (diagrammatic).

Exercise 3

***Object* : Study of anatomy of rhizome.**

Work procedure

Cut a thin transverse section of rhizome, stain in safranin-fast green combination, mount in glycerine and study.

Comments

1. **The outline** of the section is almost biconvex.
2. The section can be divided into epidermis, hypodermis, ground tissue and the stele.
3. **Epidermis** is the outermost single layer of thickly cuticularised cells.
4. **Hypodermis** that follows epidermis is made of a few sclerenchymatous layers.
5. **Ground tissue**. Rest of the tissue is called ground tissue. It is parenchymatous with numerous starch grains in the cells.
6. **Stele**. The structure of the stele varies with the age of the rhizome.
   (i) In youngest part of rhizome, it is protostele.
   (ii) In a few weeks old plant with a few leaves, the rhizome shows ectophloic siphonostele.
   (iii) The old part of rhizome shows a dictyostele.
7. **Dictyostele** is made of two rings of meristeles, separated by two sclerenchymatous bands.
8. **Meristele** has its own endodermis and pericycle. The centre is occupied with xylem which is completely surrounded by phloem on all its sides.

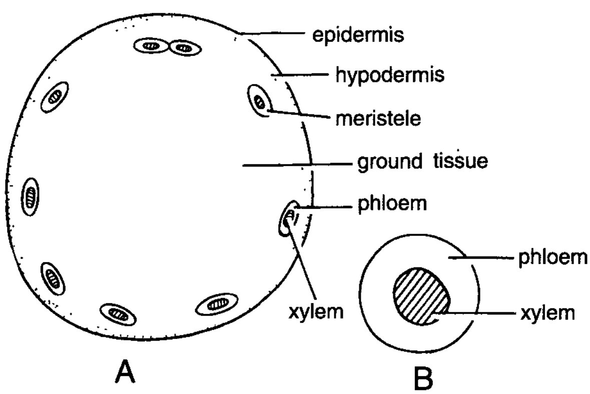


Fig. 3. ***Nephrolepis.*** T.s. rhizome (diagrammatic).

Exercise 4

***Object* : Study of anatomy of rachis.**

Work procedure

Cut a thin and uniform transverse section of the rachis, stain with safranin-fast green combination, mount in glycerine and study.

Comments

1. The section appears horse-shoe shaped.
2. It shows epidermis, hypodermis, ground tissue and the stele.
3. **Epidermis** is made of single layer of thickly cuticularised cells.
4. **Hypodermis** lies below the epidermis. The cells are sclerenchymatous.
5. **Ground tissue**. The rest of the parenchymatous region extending throughout the section is called ground tissue.
6. **Stele**. In the ground tissue is situated U-shaped or horse-shoe shaped stele.
7. **Endodermis**. The stele is surrounded by a single layered endodermis followed by a few layered pericycle.
8. **Xylem and phloem**. Centrally located xylem is surrounded by phloem on all sides.
9. **The structure of the stele** differs at various levels of rachis—
   (i) In younger parts, there is a single U-shaped stele.
   (ii) Little above the base, the U-breaks at the bottom, thereby producing two steles.
   (iii) In mature parts, dissection of the stele results in many meristeles.

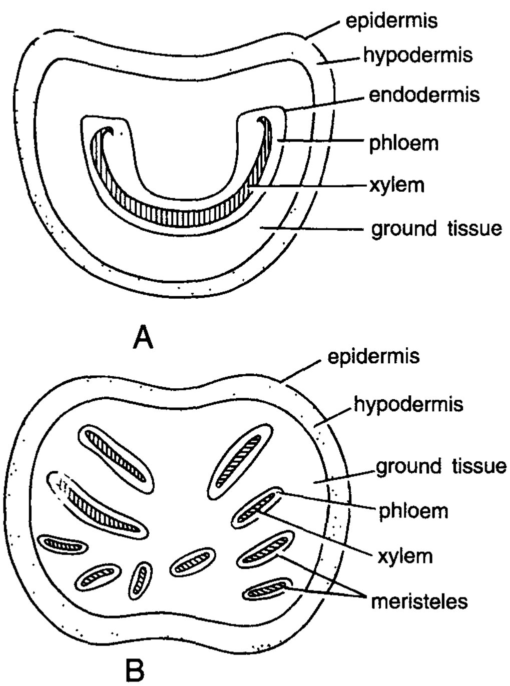


Fig. 4. ***Nephrolepis.*** T.s. rachis (diagrammatic).

Exercise 5

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

Work procedure

Observe the underside of the sporophyll, cut a transverse section of pinnae passing through a sorus. Stain in safranin-fast green combination, mount in glycerine and study.

Comments

1. The leaf bearing sori is called sporophyll.
2. The sporangia are present on the lower side of

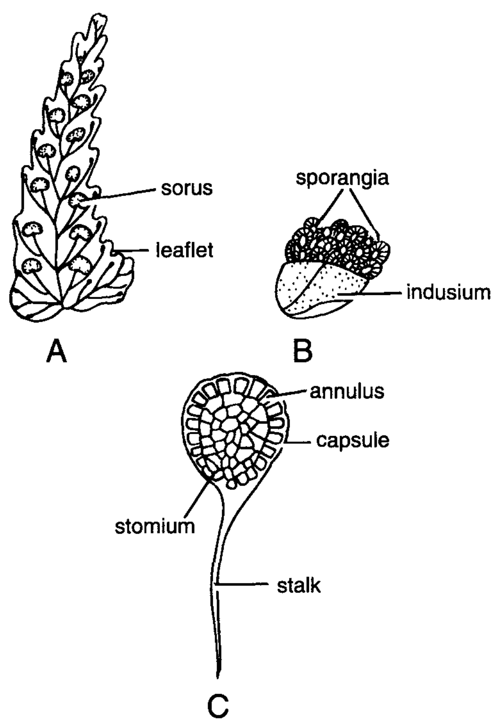


Fig. 5. ***Nephrolepis.*** **A.** Sporophyll. **B.** Sorus. **C.** A sporangium.

mature pinnae. These occur in groups called sori.
3. Sori are superficial and form definite rows, one on either side of the vein.
4. The sorus appears semi-rounded, and arises at the tip of the veinlet.
5. The sori are indusiate. The indusium is reniform (kidney-shaped), roundish or sub-orbicular.
6. Each sporangium has a stalk and a capsule.
7. The stalk is long, slender and multicellular.
8. The wall of sporangial capsule is one celled thick. A ring of thick walled cells called annulus is present. A few thin walled cells forming stomium are situated in the ring.
9. The capsule wall encloses 32 or 64 spores. All the spores are similar, the fern being homosporous.

Identification

Division—**Pteridophyta**. (1) Plant body differentiated into stem, root and leaves, (2) A definite vascular strand present.
Sub-division—**Pteropsida**. (1) Vascular cylinder siphonostele/dictyostele, (2) Plants macrophyllous with large leaf gaps, (3) Leaves bear sporangia in sori, (4) Gametophytes small, green and free-living.
Class—**Leptosporangiatae**. (1) Sporangial wall one-celled thick, (2) Number of spores per sporangium definite.
Order—**Filicales**. Mixed sori.
Family—**Polypodiaceae**. (1) Annulus of sporangium vertical, (2) Each sporangium with 32-64 spores.
Genus—***Nephrolepis***. (1) Leaves unipinnate with articulate or pouch like base, (2) Sori distinct and enclosed by individual indusium, (3) Indusium true.

Hints for Collection

It is commonly found in tropics, but a few species like *Nephrolepis acuta, N. tuberosa,* etc. are also grown as ornamentals.

Pteridium (Bracken Fern)

Classification

Division	—	**Pteridophyta**
Sub-division	—	**Pteropsida**
Class	—	**Leptosporangiatae**
Order	—	**Filicales**
Family	—	**Polypodiaceae**
Genus	—	***Pteridium***

Exercise 1

Object : **Study of external morphology of the plant.**

Work procedure

Study a fresh plant or a preserved specimen, observe the differentiation of plant body into roots, rhizome and leaves.

Comments

1. **The plant body** is a sporophyte. It is differentiated into roots, rhizome and leaves.
2. **Stem** is modified to rhizome. It is subterranean. The rhizome is long, slender and dichotomously

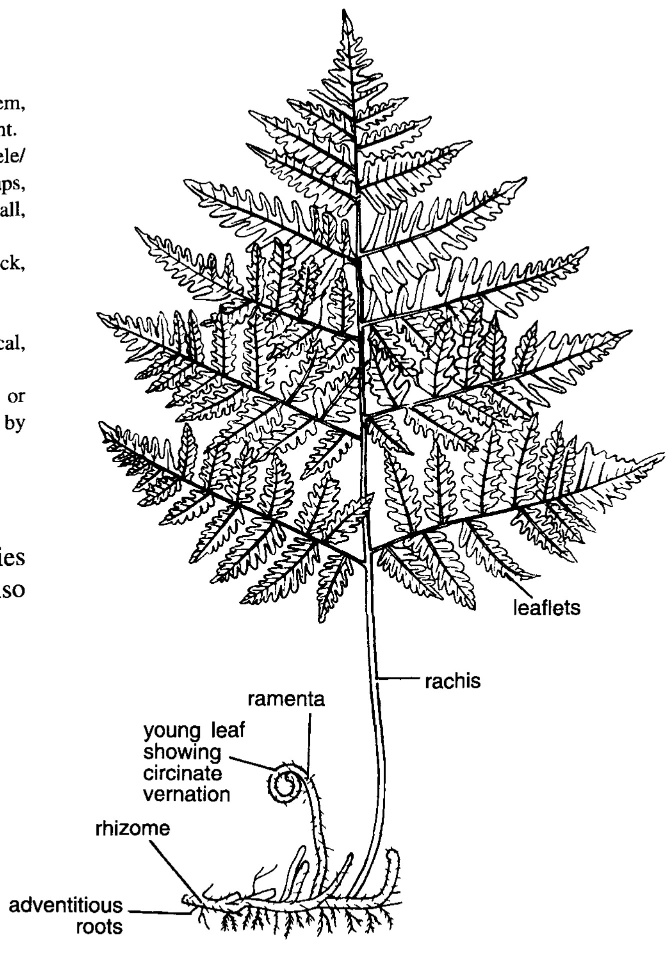


Fig. 1. *Pteridium.* External morphology : Plant showing young and mature leaves, rhizome and roots.

branched. It is covered with brown and multicellular hairs called ramenta.
3. **Roots**. The rhizome gives out adventitious roots on its underside. These are small and branched.
4. **The leaves** are borne alternately on the upper side of the rhizome at the nodes.
5. **The young leaves** are circinately coiled. The rachis is covered with ramenta.
6. Each leaf is tripinnately compound. Each pinna is sessile. It has a distinct midrib that gives out lateral branches.

Exercise 2

***Object* : Study of anatomy of the root.**

Work procedure

Cut a thin transverse section of the root. Stain with safranin-fast green combination. Mount in glycerine and study.

Comments

1. **The outline** of the section is almost circular.
2. It shows three regions—epiblema, cortex and the vascular cylinder.
3. **Epiblema** is the outermost single layer of cells. The cells are thin walled and produce unicellular root hairs.

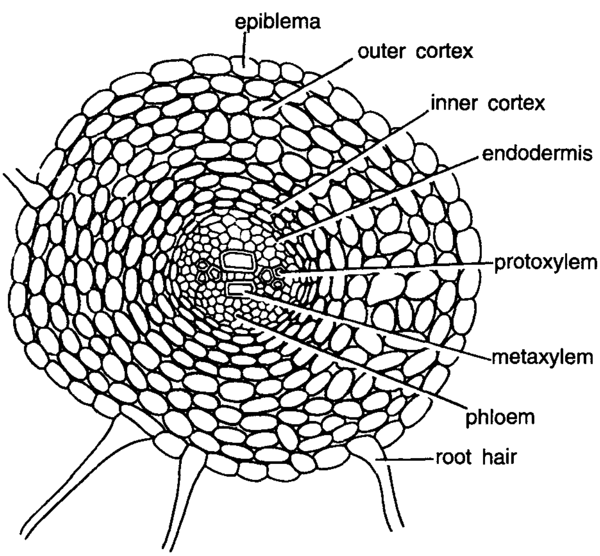


Fig. 2. *Pteridium.* T.s. of root (cellular).

4. **Cortex** occupies most part of the section It is differentiated into outer and inner regions.
5. The outer region is parenchymatous while the inner few layers are sclerenchymatous.
6. **Endodermis** follows the cortex. The radial walls of endodermal cells are characterised by casparian thickenings.
7. **Pericycle** is situated inner endodermis. It is 1 or 2 layered and parenchymatous.
8. **Vascular cylinder** shows radial, diarch and exarch conditions.
9. **The xylem** consists of two central metaxylem tracheids with groups of small protoxylem elements on their both sides.
10. **Phloem** is present on both the sides of xylem plate.

Exercise 3

***Object* : Study of anatomy of rhizome.**

Work procedure

Cut a transverse section of rhizome, stain in safranin-fast green combination, mount in glycerine and study.

Comments

1. **The outline** of the section appears almost like a biconvex lens.
2. The tissues are differentiated into epidermis, hypodermis, ground tissue and the stele.
3. **Epidermis** is the outermost single layer of cells. The cells are thickly cuticularised.
4. **Hypodermis** lies below the epidermis. The cells are sclerenchymatous which often show pitted walls. It is generally interrupted on the lateral sides by parenchyma.
5. **Ground tissue** follows the hypodermis. It is parenchymatous and is spread up to the centre of the section. The cells are filled with starch grains.
6. **Stele**. The structure of the stele varies with the age of the rhizome.
   (i) In just formed rhizome, condition is protostelic.
   (ii) In a few weeks old plant with 2-3 leaves, the rhizome shows ectophloic siphonostele.

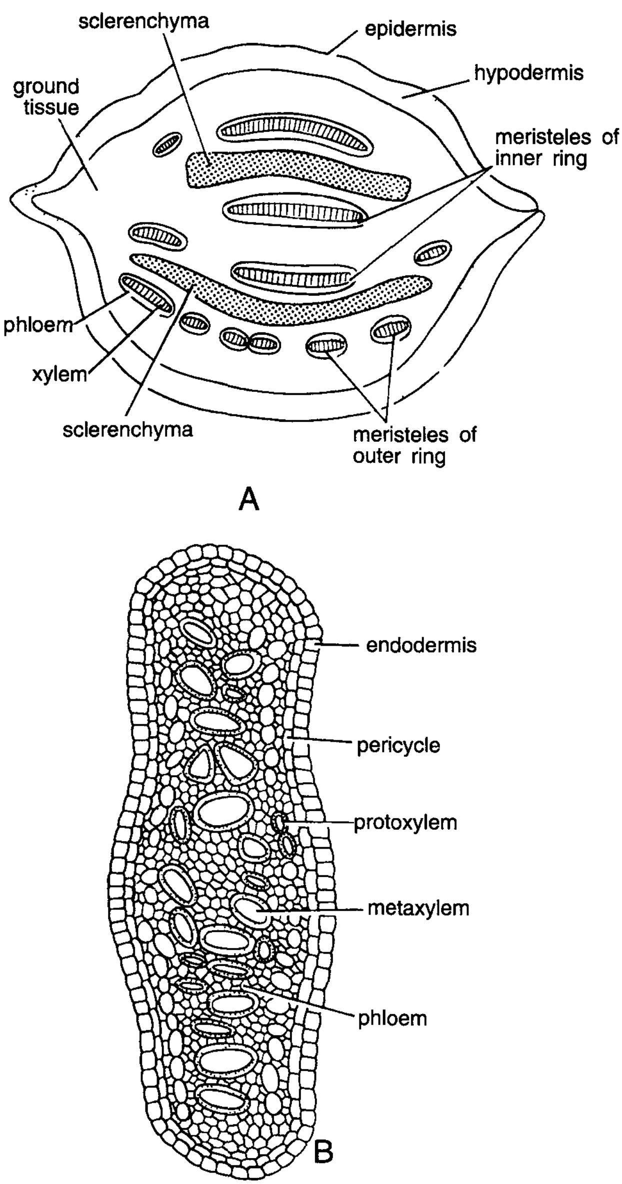


Fig. 3. ***Pteridium.*** T.s. rhizome; **A.** Diagrammatic, **B.** A meristele enlarged.

(iii) In mature plant, the old part of rhizome shows a dictyostele.

7. **Dictyostele** is made of meristeles arranged in two rings, separated by two sclerenchymatous bands.
8. **Meristele** is surrounded by its own endodermis, which is followed by one or two layers of parenchymatous pericycle.

9. The centre of the meristele is occupied by xylem which is completely surrounded by phloem on all sides.

Exercise 4

Object : **To study the anatomy of rachis.**

Work procedure

Rachis is thin and wiry, hence a sharp blade or razor would be required to cut a section. Cut transverse section, stain in safranin-fast green combination , mount in glycerine and study.

Comments

1. **The outline** of the section appears horse-shoe shaped or hemispherical.
2. The tissues of the section are differentiated into epidermis, hypodermis, ground tissue and the stele.
3. **Epidermis** which is the outermost single layer of cells is thickly cuticularised.
4. **Hypodermis** is present below the epidermis. It is 2 to 3 layered thick. The cells are sclerenchymatous.
5. **Ground tissue**. Following the hypodermis is a large region of parenchyma called ground tissue.
6. **Stele**. In the ground tissue is situated U-shaped or horse-shoe shaped stele.
7. **Endodermis and pericycle**. Stele is surrounded by a single layered endodermis followed by a few layered parenchymatous pericycle.
8. **Xylem**. The centre of the stele is occupied by massive xylem. Metaxylem is present in the centre with protoxylem located at two of its ends.
9. **Phloem**. The region between xylem and the pericycle is filled by phloem.
10. The nature of the stele varies with the maturity of the rachis.
    (i) In younger parts stele is U-shaped.
    (ii) Little above the base, it gets dissected into two large meristeles.
    (iii) In mature parts, many meristeles are present as a result of further dissection of the original stele.

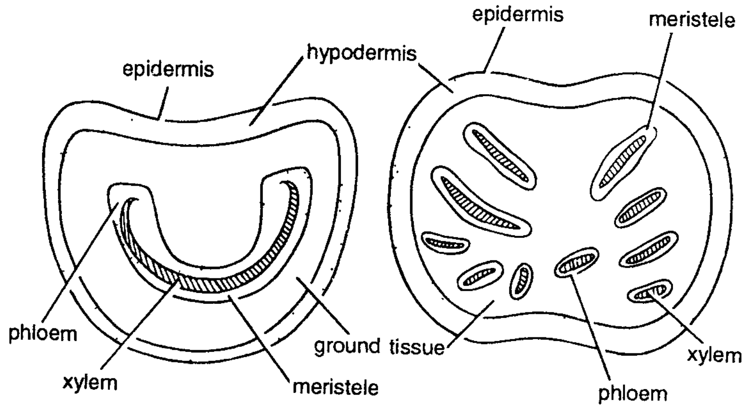


Fig. 4. ***Pteridium.*** Anatomy of rachis; **A.** T.s. of young rachis, **B.** T.s. of old rachis (both diagrammatic).

Exercise 5

Object **: Study of anatomy of pinnule.**

Work procedure

Cut vertical transverse section of the pinnule. Stain with safranin-fast green combination, mount in glycerine and study.

Comments

1. The section shows the 'midrib' region and the wings.
2. **The midrib region** consists of compact parenchyma in which a single concentric vascular bundle is situated. It shows centrally located xylem surrounded by phloem. A distinct parenchymatous bundle sheath surrounds the bundle.
3. **The upper and the lower epidermis** are single layered. The stomata are present only on the lower surface.
4. **Mesophyll** that lies between the two epidermal layers is differentiated into palisade and spongy parenchyma.
5. **The spongy tissue** is situated close to the lower epidermis. The cells are loosely arranged and contain many chloroplasts. The intercellular spaces open into stomata.

Exercise 6

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

Work procedure

Cut a vertical transverse section of the pinnule that has sori on the lower side. Stain in safranin and mount in glycerine. Study the characters of sorus and the sporangium.

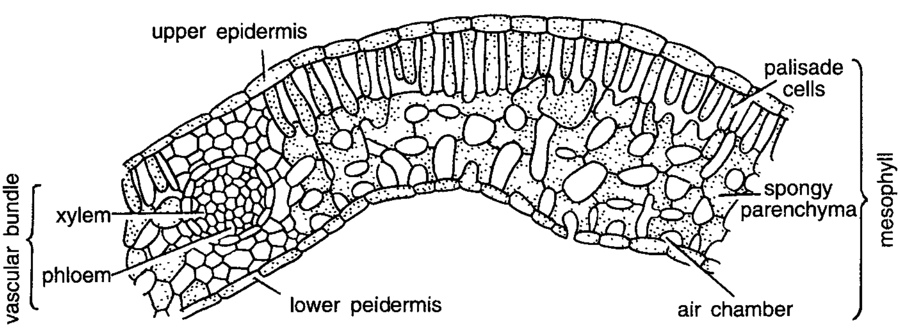


Fig. 5. ***Pteridium.*** T.s. pinnule.

Comments

1. The leaf bearing sori is called sporophyll.
2. The sporangia occur in groups called sori on the lower or abaxial side of pinnules. The sporangia form a continuous linear sorus along the margins. Such a confluent sorus is called coenosorus. The identity of sorus is thus lost and only one long sorus appears along the two lateral margins of the fertile pinnules.
3. The sorus is protected by indusium. It is made of upper indusial flap formed by the incurved margins of the pinnule and the lower true indusial flap that is poorly developed.
4. The sporangia in the sorus occur mixed. The development is leptosporangiate.
5. Each mature sporangium is differentiated into a stalk and a capsule.
6. The stalk of the capsule is made of three rows of cells. It is long and slender.
7. The capsule is ovate or biconvex. The sporangial jacket is single layered thick. A ring of thick walled cells forms the annulus. A few thin walled cells of the ring form the stomium. The capsule wall encloses 32 or 64 spores.
8. All the spores being similar, the fern is homosporous. Spores are haploid and uninucleate. The wall is two layered. The outer thick layer is called exine and the inner thin layer is called intine.

Exercise 7

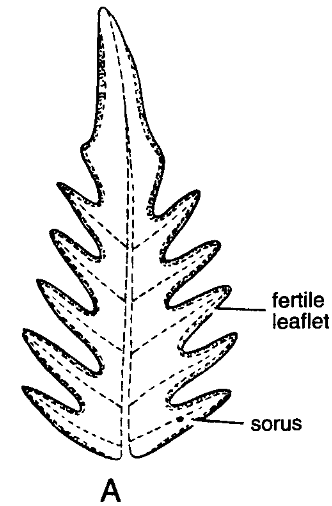
***Object* : Study of structure of prothallus.**

Work procedure

Study a slide of prothallus showing sex organs. Note the positions of sex organs and also the young sporophyte.

Comments

1. The prothallus is a gametophyte formed as a result of spore germination.
2. It is dark green, heart-shaped and single layered sheet of cells. The midrib region becomes a



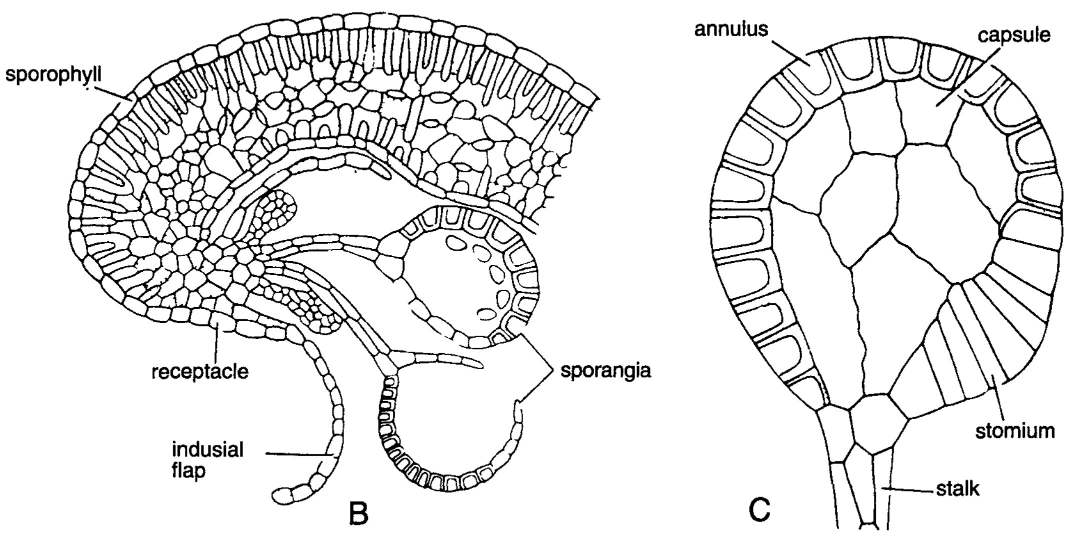


Fig. 5. ***Pteridium.*** **A.** sporophyll, **B.** Vertical section of a leaflet, **C.** A sporangium.

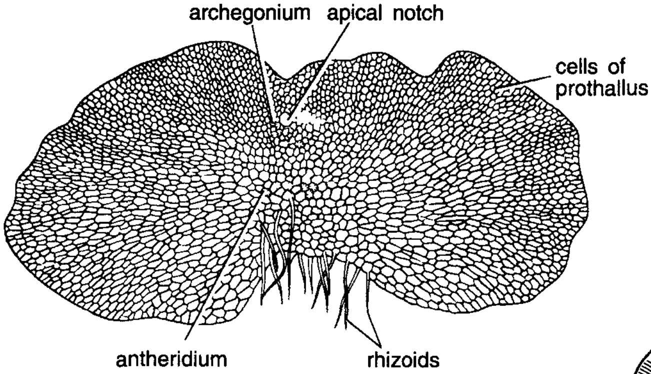


Fig. 7. ***Pteridium.*** Young prothallus with sex organs.

cushion of several cells. It remains attached to the substratum by rhizoids produced on the lower side in the central region.

3. The antheridium is surrounded by the cells of the prothallus. Each antheridium consists of wall of three rings of cells. It encloses 30-40 multiflagellate antherozoids at maturity.
4. Archegonia develop near the apical notch. Each is made of neck and ventre. The neck is 5-7 celled high with a single binucleate neck canal cell. The ventre has a small ventre canal cell and a large egg.

Exercise 8

***Object* : Study of prothallus with young a sporophyte.**

Work procedure

Study a slide of old prothallus. If prothallus is collected from the pot or natural habitat, stain in fast green, mount in glycerine and study.

Comments

1. Sporophyte is formed as a result of fertilization. The zygote grows into a sporophyte that still remains attached to the prothallus.
2. Young sporophyte is differentiated into young leaves, primary and secondary roots.
3. The leaves are petiolate and erect. These emerge through the apical notch. The leaves are simpler than the mature leaves. Sometimes these even show circinate vernation.

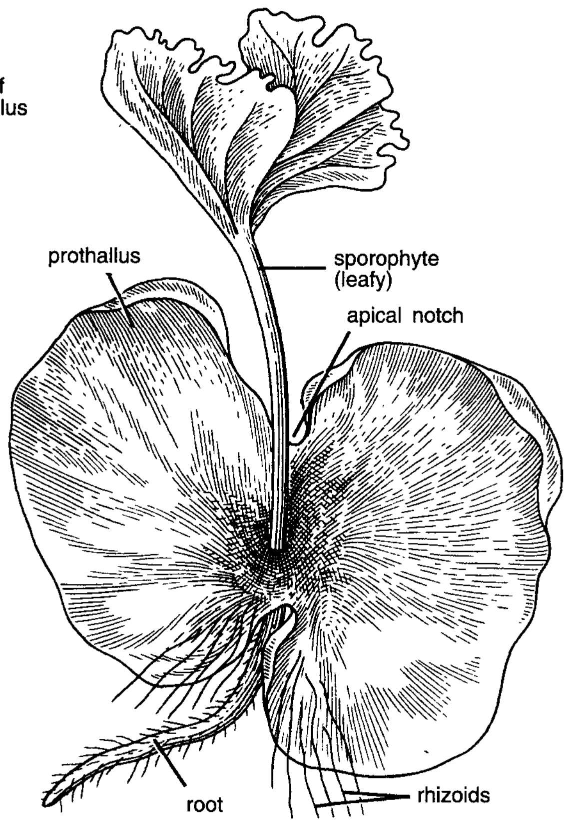


Fig. 8. ***Pteridium.*** Prothallus with a young sporophyte.

4. Primary root grows on the lower side and gives out secondary roots.
5. The sporophyte is dependent on the gametophyte till first leaf is formed. It absorbs its food through the foot of the young embryo.

Identification

Division—**Pteridophyta**. (1) Plant body differentiated into stem, root and leaves, (2) A definite vascular strand present.

Sub-division—**Pteropsida**. (1) Vascular cylinder siphonostele/ dictyostele, (2) Plants macrophyllous with large leaf gaps, (3) Leaves bear sporangia in sori, (4) Gametophytes small, green and free-living.

Class—**Leptosporangiatae**. (1) Sporangial wall one-celled thick, (2) Number of spores per sporangium is definite.

Order—**Filicales**. Mixed sori.

Family—**Polypodiaceae**. (1) Annulus of sporangium vertical, (2) Each sporangium with 32-64 spores.

Genus—***Pieridium***. (1) Leaves tripinnately divided, (2) Presence of coenosorus, (3) Sorus enclosed between indusial flaps.

Hints for Collection

Pteridium is cosmopolitan. It is widely distributed along the entire Himalayan tract. It grows particularly well at altitudes between 1,000 to 3,000 meters. *P. aquilinum* is found on forest floors, mountain slopes, open grasslands, etc.

Marsilea

Classification

Division	—	**Pteridophyta**
Sub-division	—	**Pteropsida**
Class	—	**Leptosporangiatae**
Order	—	**Marsileales**
Family	—	**Marsileaceae**
Genus	—	***Marsilea***

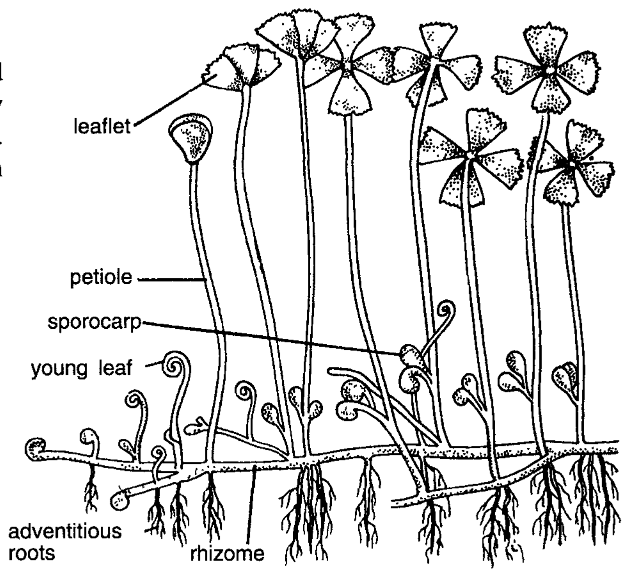


Fig. 1. *Marsilea*. External features.

Exercise 1

Object **: Study of external morphology.**

Work procedure

Study external characters of the plant. Observe various features of root, rhizome and leaves. Note the circinate vernation of young leaves and the characteristic leaf venation.

Comments

1. **The plant body** is differentiated into a rhizome, roots and leaves.
2. **The rhizome** is slender, creeping branched. It may either grow in water or attached by roots in the damp soil.
3. It bears nodes and internodes. The leaves and roots occur in acropetal succession (youngest towards the apex of rhizome) on the nodes. The adventitious roots grow downwards and the leaves grow upwards. Young leaves are circinately coiled, a characteristic of most ferns.
4. **The leaves** present at the nodes occur in two rows, (two ranked) one on either side of the mid-line of the rhizome.
5. Each leaf consists of a long petiole, bearing at its top generally four leaflets or pinnae, apparently arising from one common point. In M. *quadrifolia*, a common Indian species, six leaflets are found. (Puri and Garg, 1953, call the leaflets as pinnules).
6. The division of lamina into four pinnae is the result of three dichotomies, close to each other. Therefore, out of the four leaflets, two form a distal pair while the lower two are alternate. The leaflets, thus give a false impression of arising from one common point.
7. **Each leaflet** is obovate. The venation is dichotomous with several cross connections. The free veinlets at the apex of the leaflet are tied up with marginal loops.
8. Leaflets fold up in the night or early morning, thus showing sleeping movements.
9. The plant when grows in water, has long, flexible petioles and the leaflets float on the surface of the water but when it grows on

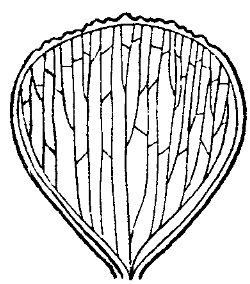


Fig. 2. *Marsilea*. Leaflet showing dichotomous venation.

mud or damp soil, the petioles become short and rigid.

(It is interesting to note that, when in a pond in which *Marsilea* is growing, water level rises, the petioles are also seen to increase in length. Contrary to it when level goes down, the petioles are found to coil, as such in both the conditions, the leaflets float on water surface).

10. The spore bearing structures known as sporocarps are commonly borne laterally near the base, on the petiole, but sometimes higher up. The two common Indian species, *M. minuta* and *M. quadrifolia* show variation in the number of sporocarps from one to four.

Exercise 2

Object **: Study of anatomy of the root.**

Work procedure

Cut a T.s. of the root, stain in safranin-fast green combination, mount in glycerine and study.

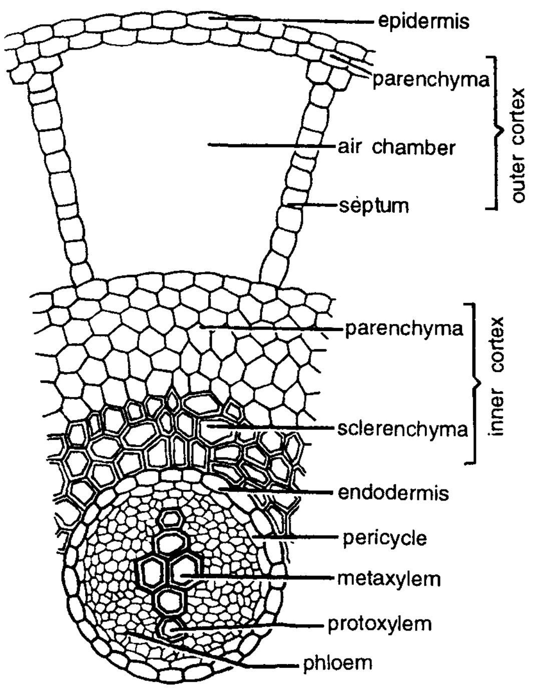


Fig. 3. ***Marsilea.*** T.s. root (a part cellular).

Comments

1. **The outline** of the section appears almost circular.
2. **The epidermis** is single layered with tangentially elongated cells.
3. **The cortex** is differentiated into an outer and an inner cortex.
4. **The outer cortex** has many air chambers separated by radial septa.
5. **The inner cortex** has either all the parenchymatous cells or some of the cells towards the inner side may become thick walled and sclerenchymatous.
6. **Endodermis** is single layered. It is followed by one layered pericycle. These surround vascular bundle.
7. **Xylem** is diarch and exarch xylem. It is situated in the centre. The protoxylem elements are situated opposite one another.
8. **The phloem** has smaller cells and forms two bands, one on either side of the xylem mass.

Features of special interest

The root shows aerenchyma in the outer cortex (hydrophytic character).

Exercise 3

Object **: Study of anatomy of rhizome.**

Work procedure

Cut a T.s. of rhizome, stain in safranin-fast green combination, mount in glycerine and study.

Comments

1. **The outline** of the section appears almost circular.
2. **The section** shows three regions—epidermis, cortex and stele.
3. **The epidermis** is single layered without stomata. The epidermis of aquatic plants lacks cuticle but that of terrestrial individuals has a distinct cuticle.
4. **The cortex** is differentiated into three regions—the outer, the middle and the inner.
5. **The outer cortex** has well-developed air

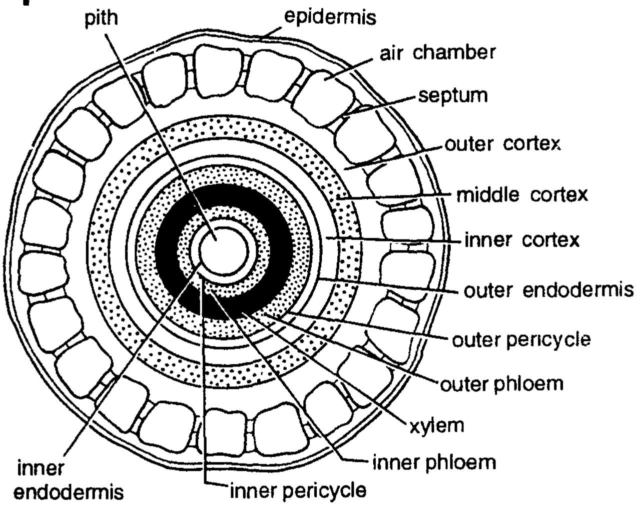


Fig. 4. ***Marsilea.*** T.s. rhizome (diagrammatic).

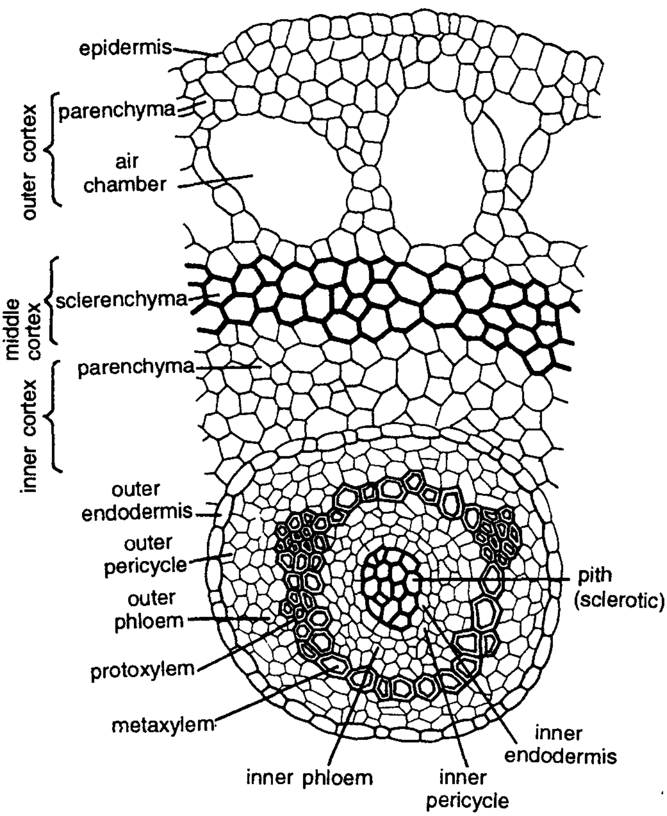


Fig. 5. ***Marsilea.*** T.s. rhizome (a part cellular).

spaces, separated by radially arranged parenchymatous cells (aerenchyma). The outermost cells of the cortex contain chloroplasts.

6. **The middle cortex** is thick walled, made up of sclerenchymatous cells and is only a few celled thick.
7. **The inner cortex** is composed of thin-walled parenchymatous cells containing starch.
8. **The stele** is an amphiphloic solenostele.
9. **Stele** shows a central xylem ring. On its outer side is outer phloem ring, outer pericycle and outer endodermis. On the inner side i.e. towards the pith are present inner phloem ring, inner pericycle and inner endodermis.
10. **Protoxylem** groups may or may not be distinct. They are generally exarch, but in some cases mesarch too.
11. **Pith** lies in the center. In aquatic plants it is parenchymatous and in terrestrial plants it is sclerotic.

Features of special interest

1. It shows hydrophytic character viz. presence of aerenchyma in the cortex, as well as some xerophytic characters viz.
   (i) thick walled middle cortex and
   (ii) sclerotic pith
2. Presence of amphiphloic solenostele.

Exercise 4

Object : **Study of anatomy of petiole.**

Work procedure

Cut a T.s. of petiole, stain in safranin-fast green combination, mount in glycerine and study.

Comments

1. **The outline** of the section is circular.
2. **Epidermis** is the outermost layer with rectangular cells.
3. **Hypodermis** is sometimes present below the epidermis. It is one or two layered.
4. **The cortex** is differentiated into an outer and an inner zone.
5. **The outer cortex** has many air chambers, separated by narrow radially arranged parenchymatous cells (aerenchyma).
6. **The inner cortex** has parenchymatous cells containing starch. A few cells contain tannins also.

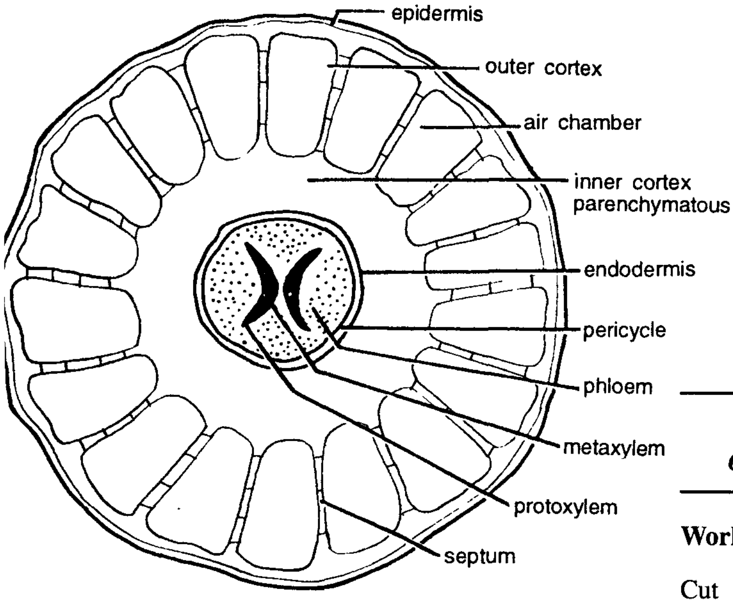


Fig. 6. ***Marsilea.*** T.s. petiole (outlines).

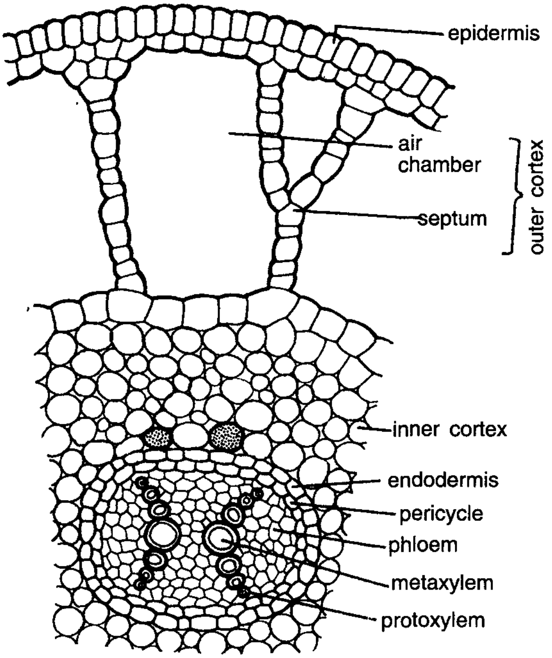


Fig. 7. ***Marsilea.*** T.s. petiole (a part cellular).

7. **The stele** is a protostele.
8. **Endodermis** is single layered. It is followed by a single layer of pericycle.
9. **The xylem** is 'V shaped' with exarch protoxylem. The two arms of 'V' are slightly curved and separate. Each arm has generally one or two large tracheids in the middle and smaller tracheids towards both the ends. The open end of 'V' always points towards the adaxial side of the petiole (towards the axis).
10. **Phloem** surrounds the xylem.

Features of special interest

1. Shows hydrophytic character viz. presence of aerenchyma in the outer cortex.
2. Presence of V-shaped xylem.

Exercise 5

***Object* : Study of anatomy of leaflet.**

Work procedure

Cut a T.s. of the leaflet, stain with safranin-fast green combination, mount in glycerine and study.

Comments

1. The section shows an upper and lower epidermis, mesophyll and a vascular bundle.
2. **The stomata** are found on both upper and lower epidermis if the plant is terrestrial but they are found only on upper epidermis if leaves float on water surface.
3. **Mesophyll** is differentiated into palisade and spongy parenchyma.
4. **Palisade** is arranged in one layer near the upper epidermis. Spongy parenchyma is located near

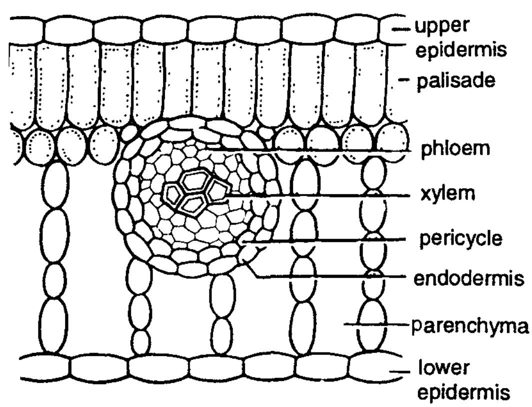


Fig. 8. ***Marsilea.*** T.s. leaflet (a part cellular).

the lower epidermis. It is loosely arranged to form large air spaces and is called aerenchyma.
5. There are many vascular bundles. Each bundle is concentric with centrally located xylem surrounded by phloem.
6. The distinct endodermis is present just outside the vascular bundle.

Exercise 6

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

Work procedure

Study the external characters of a sporocarp.

Comments

1. The spore-bearing organs are the sporocarps which are borne laterally on the adaxial side of the petiole. Their number and positions vary from species to species.
2. Sporocarp is stalked, bean-shaped or ovoid structure.
3. The place of attachment of the body of the sporocarp to the peduncle (stalk) is known as raphe.
4. Beyond the raphe, there are two projections known as teeth or tuberceles, one tooth being lower than the other.
5. The lower tooth is usually stouter and more prominent while the upper tooth, which lies a short distance above is usually more slender and delicate.
6. The side on which the raphe is present is the basal side and the side opposite to it is the apical side. The side on which the tubercles are present is the dorsal side and the side opposite to it is ventral side.

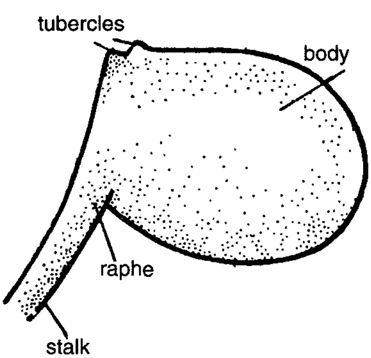


Fig. 9. ***Marsilea.*** A single sporocarp.

Exercise 7

***Object* : Study of V.T.s. of sporocarp.**

Work procedure

Cut a section of the sporocarp in a plane almost parallel to the stalk as shown in reference diagram. Stain the section in safranin, mount in glycerine and study.

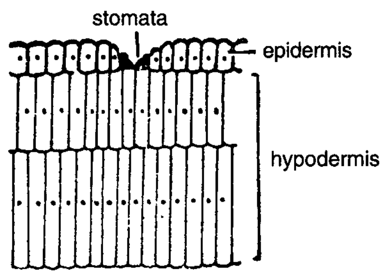


Fig. 10. ***Marsilea.*** A part of the sporocarp wall (cellular).

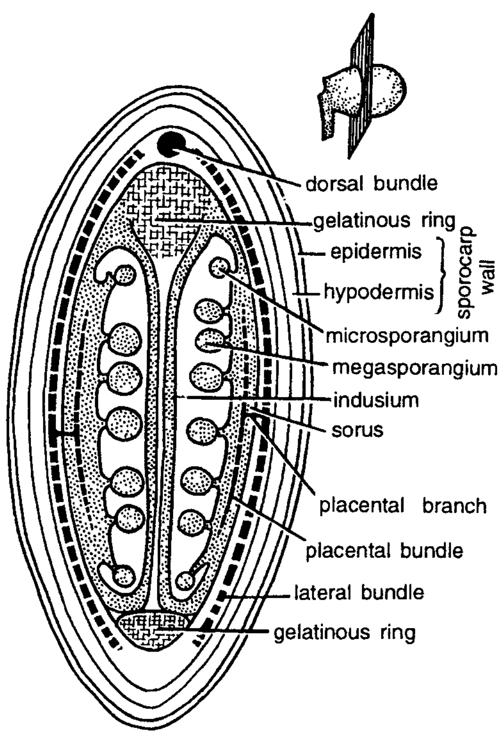


Fig. 11. ***Marsilea.*** V.T.s. sporocarp.

Comments

1. **The section** shows wall of the sporocarp which encloses sori.
2. **The wall** is made of outer epidermis followed by hypodermis.
3. **Epidermis** consists of thick walled cells. Numerous stomata are present in the epidermis.
4. **Hypodermis** consists of two layers of radially elongated cells. The cells of the inner layer are double in length as compared to the cells of the outer layer.
5. All the cells of both the layers have their nuclei arranged in one row.
6. **Receptacles** are cut longitudinally. Only two sori are seen, each of which is covered by its own 2 layered indusium. The receptacle of the sorus bears microsporangia at the corners and megasporangia all along the receptacular ridge.
7. On the upper and lower sides of the receptacles, two masses of gelatinous ring, cut transversely, are present. The upper one is bigger in size than the lower.
8. The dorsal bundle, lateral bundles, placental branches and placental bundles are seen.

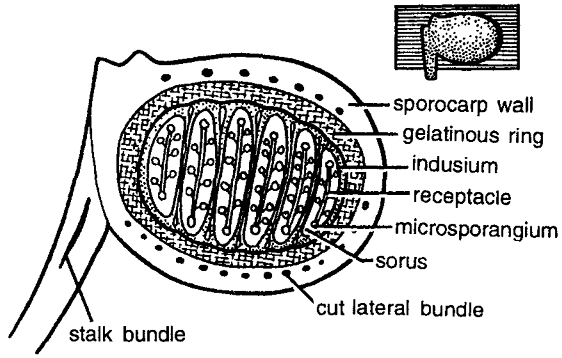


Fig. 12. ***Marsileal.*** V.L.s. sporocarp showing microsporangia.

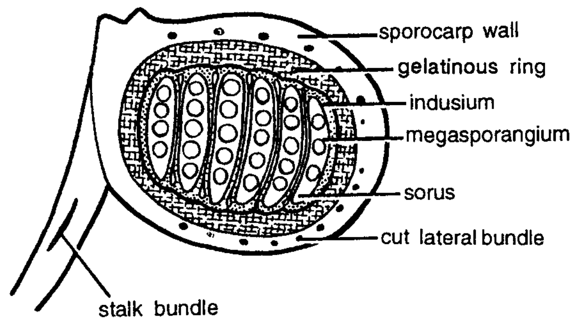


Fig. 13. ***Marsilea.*** V.L.s. sporocarp showing megasporangia.

Exercise 8

Object : Study of V.L.s. of the sporocarp.

Work procedure

Hold the sporocarp with tubercles pointing upwards. Split the sporocarp by a sharp blade in two halves. Study the section under dissecting microscope, section being thick.

Comments

1. The section shows wall of the sporocarp enclosing sori embedded in a gelatinous wall.
2. The outermost is the sporocarp wall. It is made of an epidermis with stomata and two layered hypodermis.
3. Below the sporocarp wall is a gelatinous ring which surrounds sori. It is relatively more prominent on the dorsal side than on the ventral.
4. The sori are cut longitudinally and appear in a row.
5. Each sorus is surrounded by its own indusium.
6. If the section passes through the centre, then megasporangia are seen in all the sori. Since the megasporangia are present at the apex of the receptacle, no receptacle is seen.
7. If the section is not perfectly median, then microsporangia are seen attached on either sides of the receptacle in each sorus.
8. In this section the stalk bundle and cut lateral bundles are seen.

Exercise 9

Object : Study of H.L.s. of the sporocarp.

Work procedure

Hold the stalk between the thumb and the index finger. Cut a section by passing a blade at right angles to the stalk axis (see reference diagram). Stain in safranin, mount in glycerine and study.

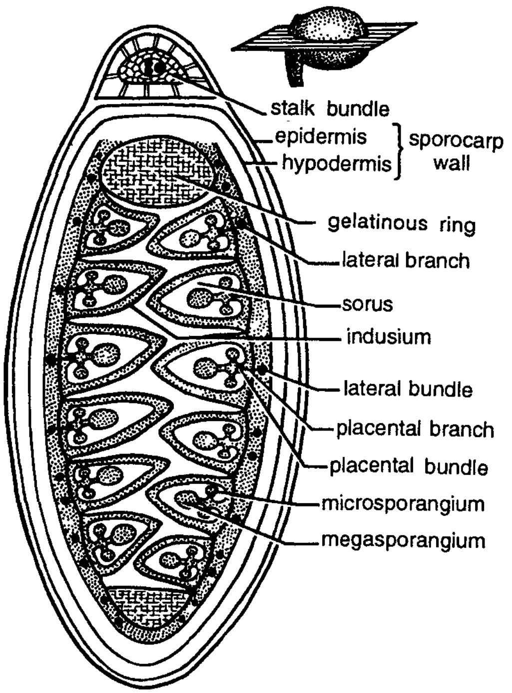


Fig. 14. ***Marsilea.*** H.L.s. sporocarp.

Comments

1. The section shows transversely cut stalk, wall of the sporocarp and two rows of sori.
2. Transversely cut stalk appears on one side. It shows the stalk bundle.
3. The wall of the sporocarp is made of epidermis with stomata and two layered hypodermis.
4. Gelatinous ring shows two patches, heavier on the dorsal side than on the ventral.
5. There are two rows of sori, one row alternating with the other.
6. Each sorus is covered by its indusium.
7. A sorus consists of a receptacle. Megasporangium is present at the apex of receptace while microsporangia are present on the sides.
8. The lateral bundles are cut transversely and each is seen to supply its own receptacle by a receptacular or placental branch.
9. Thus, in this section dorsal bundle, many lateral bundles and receptacular branches (placental branches) are seen.

Exercise 10

Object **: Study of dispersal of spores.**

Work procedure

Cut open the body of the sporocarp on ventral side by a sharp blade or scalpel. Place in water for some time. Gelatinous ring with sporangia attached to it comes out.

Comments

1. The sporocarp is hard and resistant to unfavourable conditions.
2. It opens through its ventral margins.
3. It imbibes water and the gelatinous ring inside swells up.
4. This ring ultimately comes out of the sporocarp wall.
5. Gelatinuous ring bears two rows of sporangia, one on each side, alternating with one another.

Identification

Division—**Pteridophyta.** (1) Plant body differentiated into stem, roots and leaves, (2) A definite vascular strand present.
Sub-division—**Pteropsida.** (1) Plants are always megaphyllous

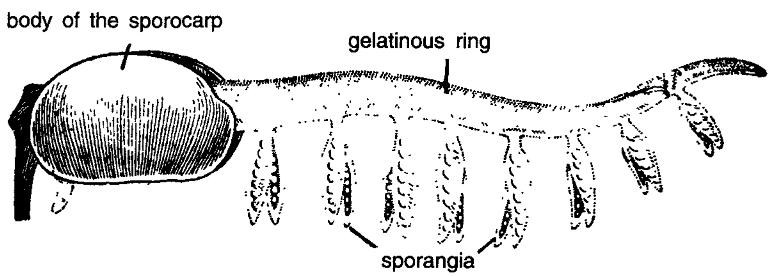


Fig. 15. ***Marsilea.*** Sporocarp showing extension of gelatinous ring during germination.

excluding a few exceptions, (2) Leaves differentiated into a petiole and dissected blade, (3) The sporangia are borne on abaxial surface of leaves.

Class—**Leptosporangiatae**. (1) Development of sporangium is of leptosporangiate type, (2) Jacket one cells in thickness, (3) Spores definite within a sporangium.

Order—**Marsileales**. (1) Members heterosporous, (2) Sporangia formed within sporocarps.

Family—**Marsileaceae**. (1) Members aquatic, (2) Sorus gradate type and each produces both types of sporangia, (3) Leaf circinately coiled in bud condition.

Genus—***Marsilea***. (1) Leaflet with dichotomous venation and cross connections, (2) Presence of aerenchyma in vegetative organs of the sporophyte, (3) Presence of amphiphloic solenostele in the rhizome, (4) Presence of V-shaped xylem in petiole.

Hints for Collection

The two Indian species, *Marsilea minuta* and *M. quadrifolia*, are commonly found growing either in shallow water or on moist banks of ponds and ditches. They grow either completely submerged or partially or entirely out of water in damp and wet places.

Azolla

Classification

Division	—	**Pteridophyta**
Sub-division	—	**Pteropsida**
Class	—	**Leptosporangiatae**
Order	—	**Salviniales**
Family	—	**Salviniaceae**
Genus	—	***Azolla***

Exercise 1

Object **: Study of external morphology of the sporophyte.**

Work procedure

Collect a fresh specimen or study a preserved plant.

Comments

1. The plant is a sporophyte. It grows free floating in ditches and ponds.

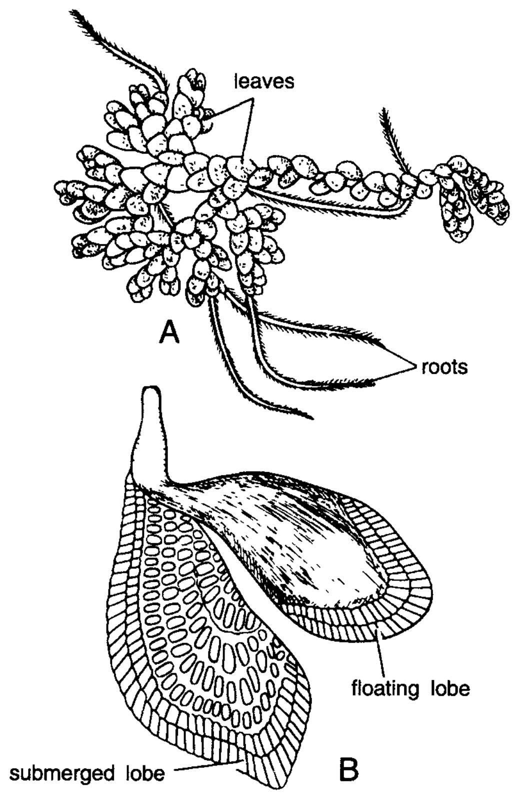


Fig. 1. ***Azolla***. External features of *A. filiculoides*; **A.** Plant showing roots and leaves, **B.** A single leaf.

2. **The plant body** is differentiated into stem, roots and leaves.
3. **The stem** is pinnately branched. It is horizontally floating. The branches are extra-axillary.
4. **Roots** are produced from the lower side of the stem. These remain submerged in water.
5. **Leaves** cover stem and its branches. These are present in two alternate and overlapping rows.
6. **Each leaf** is divided into two lobes of approximately equal size.
7. The upper or aerial lobe is thick and green. It is somewhat obliquely placed and only one of the edges touches the water.
8. The thin lower or submerged lobe is nearly colourless. The absorption of water is believed to take place through this lobe.

Exercise 2

Object : Study of anatomy of root

Work procedure

Cut a T.s. of the root, stain in safranin-fast green combination, mount in glycerine and study.

Comments

1. **The outline** of the section is almost circular.
2. It shows epidermis, cortex and the stele.
3. **Epidermis** is the outermost single layer of cells.
4. **Cortex** consists of 2-8 layers of parenchymatous cells.
5. **Endodermis** lies inner to cortex. It is made of a single layer consisting of 6 cells.
6. **Pericycle** that follows is also made of a single layer consisting of 6 cells.
7. **Xylem** lies in the centre. It is represented by two centrally placed metaxylem tracheids. These are surrounded by four small outer groups of protoxylem elements.
8. **Phloem** consists of only a few elements. These are placed on either sides of the metaxylem elements.

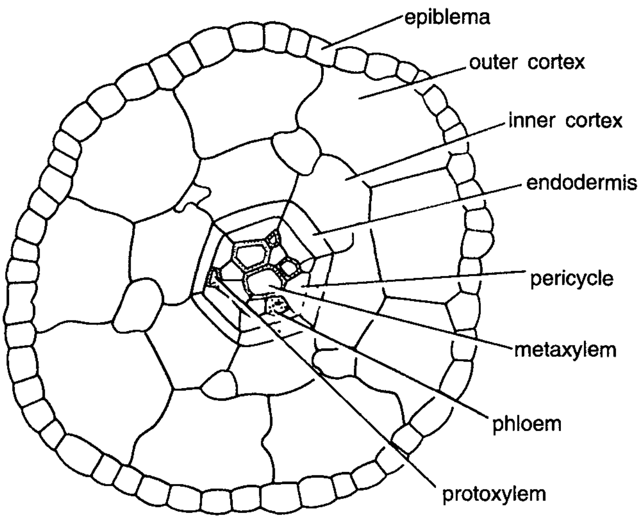


Fig. 2. *Azolla.* T.s. root (cellular).

Exercise 3

Object : Study of anatomy of stem.

Work procedure

Cut a transverse section of the stem, stain with safranin-fast green combination, mount in glycerine and study.

Comments

1. **Outline**. Transverse section of the stem is almost circular in outline.
2. It shows epidermis, cortex and stele.
3. **Epidermis** is the outermost single layer of cells.
4. **Cortex** is five to eight cells in thickness. The cells are thin walled and parenchymatous without intercellular spaces.
5. **Stele** is centrally located. It is surrounded by single layer of endodermis followed by a single layer of parenchymatous pericycle.
6. The central cylinder is protostelic. The vasculature of the stem is greatly reduced in response to aquatic habitat.
7. **Vascular tissues** are represented by about six xylem elements and twice as many phloem elements, in a stele.

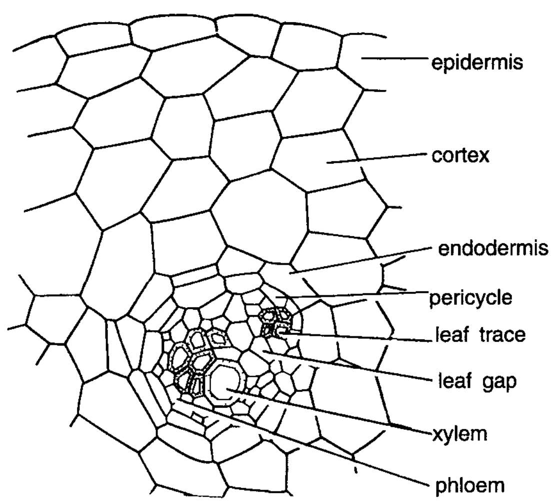


Fig. 3. *Azolla.* T.s. stem (cellular).

Exercise 4

Object **: Study of anatomy of leaf.**

Work procedure

Cut a vertical transverse section of the upper lobe by keeping it in suitable sized pith. Stain in safranin-fast green combination, mount in glycerine and study.

Comments

1. The upper lobe of leaf is bound on both the sides by upper and lower epidermal cells.
2. Both the layers possess stomata.
3. The upper epidermis has many unicellular or bicelled hairs.
4. Major portion of the leaf between both epidermal layers is made of palisade-like photosynthetic cells. Large intercellular spaces are present between them.
5. The upper lobe has a large cavity at its base. It opens to the outside through a circular pore.
6. The cavity is filled with the filaments of blue green alga—*Anabaena azollae.* The alga has a symbiotic relationship with the fern. It fixes atmospheric nitrogen.
7. The pore is later closed by ougrowths of the tissue of the margin. It becomes filled with mucilage.

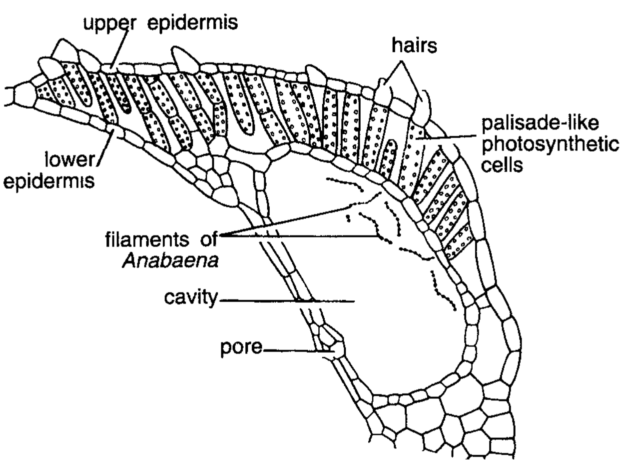


Fig. 4. ***Azolla.*** T.s. through dorsal (floating) lobe of leaf.

Exercise 5

Object **: Study of structure of sporocarp.**

Work procedure

Look for the sporocarp on the lower side of the plant. Identify microsporocarp and megasporocarp. Tease them. Stain with safranin, and study the internal structure.

Comments

1. Sporocarps are borne only on the lowermost leaf of a lateral branch at the end of annual season.
2. Submerged lobe of the leaf bears 2-4 sporocarps.
3. The upper lobe of the fertile leaf forms a hood-like covering around the sporocarp.
4. The sporocarps are dimorphic i.e. these are of two types: microsporocarps and megasporocarps.
5. Larger sized is a microsporocarp and the smaller sized is a megasporocarps.
6. Each sporocarp is a sorus covered by indusium.
7. Microsporocarp shows a central raised cushion on which sporangia develop basipetally. Each microsporangium has one layered jacket. It is followed by tapetum. The cavity encloses 64 microspores.
8. Microsporangium has a multinucleate periplasmodium formed as a result of breakdown of tapetum. Periplasmodium forms four or more quadrately arranged massulae in which spores remain embedded at periphery.
9. The surface of massulae has many anchor-shaped barbed hairs called glochidia which help the attachment of massulae to the microspore.
10. Megasporocarp shows a single large megasporangium. It is surrounded by a flask-shaped indusium. It envelops the sporangium completely except for a narrow slit at the apex.
11. Megasporangium is covered by a single layered wall. It encloses a single megaspore.
12. Megaspore is surrounded by a hardened vacuolate layer—the perispore. The megaspore wall is hard and ornamented. It is called epispore.

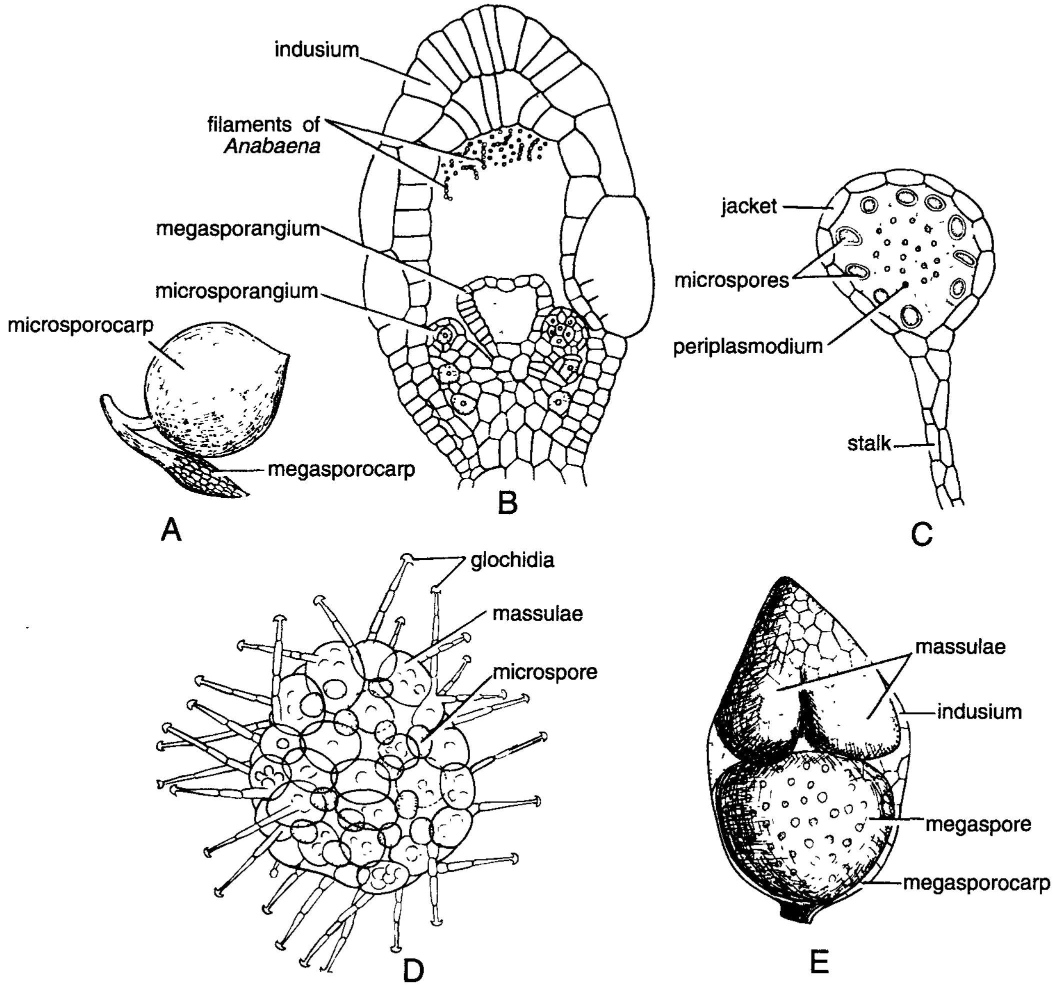


Fig. 5. ***Azolla.*** **A.** The fertile submerged lobe with one large microsporocarp and one small megasporocarp. **B.** L.s. of nearly mature microsporocarp. **C.** Nearly mature microsporangium. **D.** Massulae inside microsporangium. **E.** Massulae inside megasporangium.

13. At the distal end of the megaspore, four quadrately arranged massulae are present. These are formed by the remaining aborted spores and the tapetal cells.

Identification

Division—**Pteridophyta**. (1) Plant body differentiated into stem, root and leaves, (2) A definite vascular strand present.

Sub-division—**Pteropsida**. (1) Vascular cylinder siphonostele/ dictyostele, (2) Plants macrophyllous with large leaf gaps, (3) Leaves bear sporangia in sori, (4) Gametophytes small, green and free living.

Class—**Leptosporangiatae**. (1) Sporangial wall one celled thick, (2) Number of spores per sporangium is definite.

Order—**Salviniales**. (1) Sporocarp is a single sorus enclosing either megasporangia or microsporangia, (2) Sporocarp walls formed by the indusia.

Family—**Salviniaceae**. Single family.

Genus—***Azolla***. (1) Presence of endophytic blue green algae *Anabaena* in the leaf, (2) Each leaf divided into two lobes, (2) Megasporocarp with only one megasporangium.

Hints for Collection

Azolla forms red coloured bloom in ditches and ponds. It is found floating freely on the surface of water. Common Indian species is *A. pinnata*. Another species *A. filiculoides* is also known to occur frequently while the third species *A. imbricata* is found mostly in Eastern Himalayas.

Gymnosperms

9
Chapter

Preamble

Gymnosperms form a large group of evergreen, slow growing plants. Though true seeds are formed, the group differs from other group of seed-bearing plants the angiosperms, firstly in possessing naked ovules; secondly, in the lodging of pollen grains directly on the micropyle and thirdly, in the absence of true vessels and companion cells. This group is more ancient than angiosperms, claiming fossils as well as living members and form a bridge between the pteridophyta on one hand and the angiosperms on the other.

The gymnosperms vary in size from small plants to very large gigantic plants. *Sequoia sempervirens* grows up to a height of about 150 meters (California) and *Taxodium maxicanum* has a trunk with the enormous diameter of about 17 meters. (Contrary to this, *Zamia pygmia* is the smallest gymnosperm with and underground tuberous stem. In gymnosperms, there are two main structural types, the leaves. Most of the members of this group grow in relatively dry and poor soils, the plants thus exhibit thermographic features.

The fructifications (cones) are made up of an aggregation of sporophylls bearing sporangia, in which the spores are produced. The cones are generally unisexual. The male and female cones differ in shape and size. Whereas the male cones are usually smaller and short lived, the female cones are quite larger and long lived. Considerably the gametophytic generation is even more reduced than it is in any of the pteridophyta.

The gymnosperms are also important from economic point of view. Some conifers as *Cedrus deodara* (vern.deodar) form valuable timber. Canada balsam, a chief familiar mounting medium used in biological laboratories, is the resin of *Abies balsamea.* Turpentine oil which is chiefly used as medicine is also extracted from a conifer tree. Last, but not the least gymnosperms have also proved themselves for their food value viz. sago palm (*Cycas revoluta*) yield sago or sabudana (of course the chief commercial supply now comes from *Metroxylon rumphii*- an angiosperm) and the very familiar fruit of chilgoza is the seed of *Pinus gerardiana.*

Classification of Gymnosperms

Division. GYMNOSPERMS

Class	Order	Family	Examples
Cycadopsida	Pteridospemales	Lyginopteridaceae	*Heterangium**, *Lyginopteris**
		Glossopteridaceae	*Glossopteris**
	Bennettitales	Williamsoniaceae	*Williamsonia**
		Cycadeoidaceae	*Cycadeoidea**
	Cycadales	Cycadaceae	*Cycas*
Coniferopsida	Coniferales	Pinaceae	*Pinus*
	Taxales	Taxaceae	*Taxus*
Gnetopsida	Gnetales	Gnetaeceae	*Gnetum*
		Ephedraceae	*Ephedra*

*Fossil members

Distinguishing Characters of Taxa

DIVISION. **GYMNOSPERMS**
(1) Ovules naked
(2) Seeds attached to a scale
(3) Scales forming a strobilus

CLASS **1. CYCADOPSIDA**
(1) Wood manoxlic
(2) Large frond-like leaves
(3) Seeds with radial symmetry

Order **1. Pteridospermales**
(1) Leaves large, frond-like, pinnately compound
(2) Large leaf traces with one or more strands
(3) Spores formed in sporangia, aggregated in synangia

Family **1. Lyginopteridaceae**
(1) Stem monostelic
(2) Petioles with a strong midrib
(3) Seeds small

Examples. ***Heterangium*****, ***Lyginopteris*****

Family **2. Glossopteridaceae**
(1) Leaves with a strong midrib
(2) Stelar structure unusual, showing several plates of vascular tissues
(3) Reproductive structure cupulate and bisexual

Example. ***Glossopteris*****

Order **2. Bennettitales**
(1) Tree trunk covered by a mantle of persistent leaf-bases
(2) Microsporophylls in groups at the tip of frond-like leaves
(3) Megasporophylls in cone-like organization

Family **1. Williamsoniaceae**
(1) Stem delicate, branched
(2) Inflorescence stalked or sessile, not sunk in the scales of persistent leaf bases

Example. ***Williamsonia*****

Family **2. Cycadeoidaceae**
(1) Trunk columnar
(2) Trunk covered by a mantle of leaf bases
(3) Flowers' sunk in the distal part of the trunk

Example. ***Cycadeoidea*****

Order **3. Cycadales**
(1) Plants woody, stem unbranched
(2) Wood manoxylic
(3) Presence of mucilage canals
(4) Leaf trace diploxylic
(5) Dioecious plants
(6) Ovules orthotropous
(7) Sperm with spiral band of flagella

Example. ***Cycas***

CLASS **2. CONIFEROPSIDA**
(1) Wood pycnoxylic
(2) Leaves needle-shaped, or fan-shaped
(3) Seeds with bilateral symmetry

Order **1. Coniferales**
(1) Plants branched, leaves needle shaped
(2) Resin canals present
(3) Male and female cones compact
(4) Male gametes non-flagelate

Family 1. **Pinaceae**
(1) Wood resinous
(2) Plants monoecious
(3) Sporophylls spirally arranged
(4) Microsporophylls with two microsporangia
(5) Pollen grains winged
(6) Female cone woody
(7) Polyembryony present
(8) Seeds dry and winged

Example. ***Pinus***

Order **2. Taxales**
(1) Profusely branched trees or shrubs
(2) Leaves simple, solitary, flat and spirally arranged
(3) Wood pycnoxlic wihtou parenchyma
(4) Plants mostly dioecious
(5) Female strobilus represented by a single terminal ovule, enclosed in aril

Family. **Taxaceae**
(1) Typical of order

Example. ***Taxus***

CLASS **3. GNETOPSIDA**
(1) Wood with vessels,
(2) Flowers in compound strobili or inflorescence, unisexual, usually dioecious,
(3) Ovule surrounded by several envelopes.

Order **1. Gentales**
(1) Plants woody trees, shrubs or lianas
(2) Leaves simple, arrangement opposite or whorled
(3) Male flowers with perianth

Family **1. Gnetaceae**
(1) Ovules cauline
(2) Leaves - scaly and foliage; foliage leaves oblong-lanceolate

* Fossil members

(3) Plants dioecious
(4) Male and female cones in panicles
(5) Cones with 'cupules' or collars'
(6) Seeds with protective inner envelope

Example. ***Gnetum***

Family **2. Ephedraceae**

(1) Plants shrubs or woody climbers
(2) Leaves- scaly and foliage
(3) Presence of nodal diaphragm
(4) Stamens enclosed by bract
(5) Seeds covered with fleshy bracts

Example. ***Ephedra***

Cycas

Classification

Division	–	**Gymnosperm**
Class	–	**Cycadopsida**
Order	–	**Cycadales**
Family	–	**Cycadaceae**
Genus	–	***Cycas***

Exercise 1

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

Work procedure

Study the external features of the plant. Observe the armour of leaf bases on the stem, absence of branching, crown of leaves, two types of roots, etc.

Comments

1. **Plant body** is differentiated into an underground root system, that is distinguished into an erect stem and a crown of leaves.
2. **Roots** are of two types : (i) primary or normal root and (ii) secondary of coralloid root.
3. **Normal root** is a tap root, growing deep into the soil (positively geotropic). It is sparsely branched and sometimes grows as thick as aerial stem.
4. **Secondary roots** are negatively geotropic projecting above the soil surface, repeatedly dichotomously branched and appear as coralloid clusters.

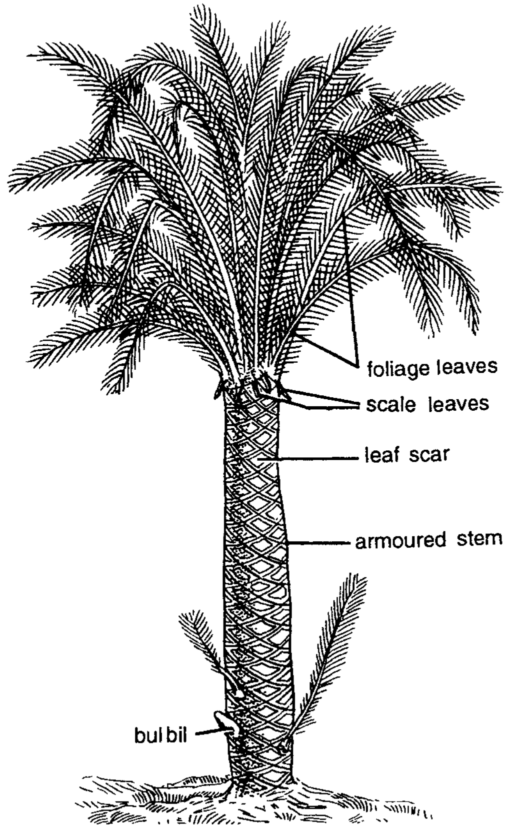


Fig. 1. ***Cycas.*** External features.

5. **The young stem** is almost tuberous but when grows old, it becomes thick, colummar and unbranched (Branching is rare and is caused due to injury, etc.). The trunk is covered by persistent leaf bases.
6. **Leaves.** The stem bears a terminal group of leaves which are dimorphic (i.e. of two types) (i) foliage leaves (green assimilatory fronds) and (ii) scale leaves (brown and hairy). These leaves alternate with one another.
7. Young foliage leaves are circinately coiled and are covered with ramenta (hairs).
8. **Mature leaves** are spirally arranged and pinnately compound. Each leaf has about 80-100 pairs of pinnae that are closely arranged, opposite one another on the rachis with a decurreht base. Each pinna is tough, leathery and entire with a definite midrib but no lateral veins.

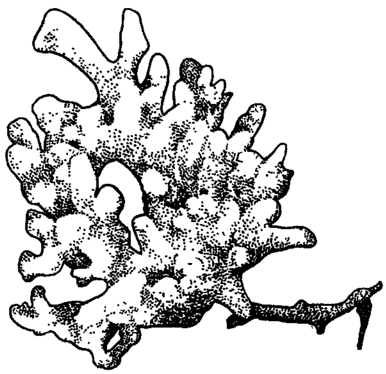


Fig. 2. *Cycas.* Coralliod root-external features.

9. **Scale leaves** are small, simple , brown with aborted lamina and covered with hairs. These leaves cover the apex and young developing foliage leaves. Scales are also persistent, like leaf bases.
10. **Reproductive organs.** *Cycas* is dioecious and, as such, bears terminally, either male cone or female reproductive structures.
11. **The male cone is** borne terminally at the apex of the stem and the further growth of the stem continues by axillary bud (developed at the base of the cone) which pushes the male cone on one side. The branching in *Cycas* stem is thus referred to as sympodial.
12. **The female reproductive structures** are the sporophylls developing in place of foliage leaves. The vegetative apex continues to grow as usual.
13. **The sporophylls** are smaller than the foliage leaves. They are brown or light brown in colour and are densely covered with wooly hairs.

Exercise 2

***Object* : Study of anatomy of normal young root.**

Work procedure

Cut a T.s. of the young part of primary root, stain in safranin-fast green combination, mount in glycerine and study.

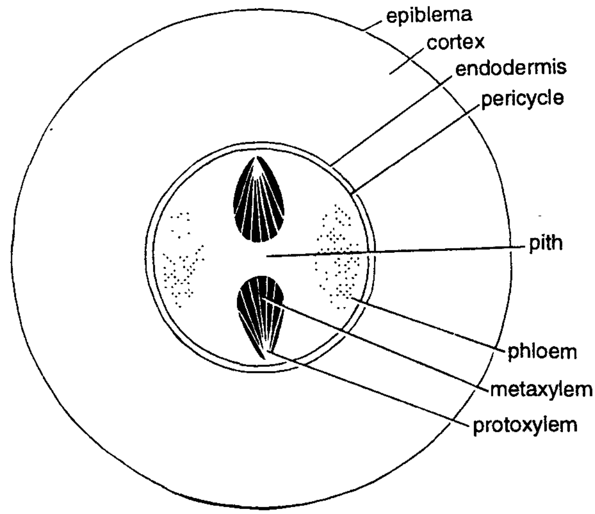


Fig. 3. *Cycas.* T. s. of normal young root (diagrammatic).

Comments

1. **The section** is circular in outline. It shows an outer layer or epiblema, cortex and centrally located stele.
2. **Epiblema** is made of single layer of thin walled cells. Some of these cells bear unicellular root hairs.
3. **Cortex** is multilayered with starch filled parenchymatous cells. A few tannin filed cells are also scattered in this region.
4. **Endodermis** is single layered and indistinguishable. Many-layered pericycle separates the cortex from vascular tissues.
5. **The central stele** is made of radial and exarch vascular bundles. There are two protoxylem groups and thus condition is diarch.

Exercise 3

***Object* : Study of anatomy of older part of normal root.**

Work procedure

Cut a T.s. of the older part of normal or primary root, stain in safranin-fast green combination, mount in glycerine and study.

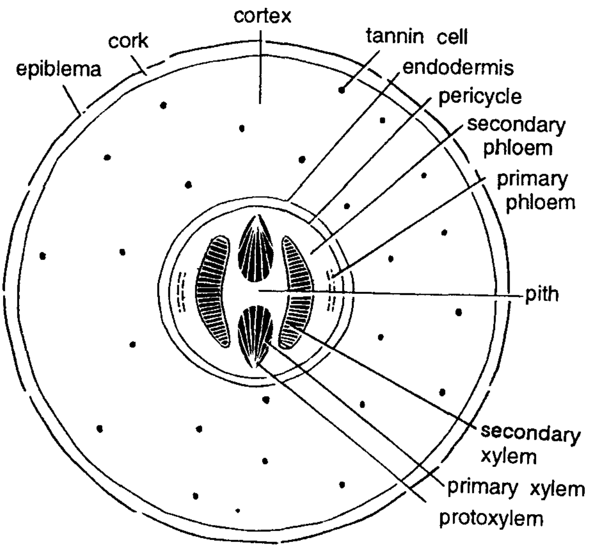


Fig. 4. ***Cycas.*** T. s. of normal old root (diagrammatic).

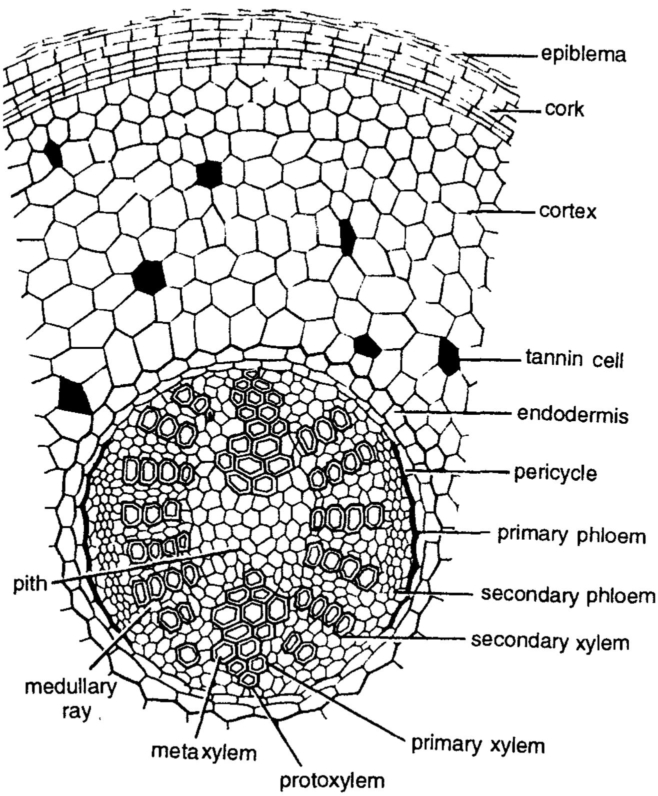


Fig. 5. ***Cycas.*** T. s. of old root (a part cellular).

Comments

1. It shows secondary growth, rest of the structures being similar to that of a young root.
2. **The epiblema** is ruptured due to the thick walled cork cells formed below it. Cork cells are a few layered deep and are arranged in brick-like fashion.
3. **Cortex** is large, parenchymatous and multilayered. It is present below the cork. A few tannin filled cells occur scattered in the cortex.
4. **Endodermis** is single layered. It is followed by many layered pericycle.
5. **Primary** phloem is the outermost (near the pericycle) and is crushed during secondary growth. Secondary phloem follows this layer, the cells of which are intact.
6. **Cambium** arcs are formed along the inner edges of phloem in the vascular region.
7. **Secondary xylem** is situated towards pith. The primary xylem is situated in the same region as it was before the secondary growth.
8. **Medullary rays** are formed.
9. **In the centre** is a small parenchymatous pith.

Exercise 4

Object : Study of anatomy of coralloid root.

Work procedure

Cut a T.s. of the root, stain in safranin- fast green combination mount in glycerine and study.

Comments

1. The structure is almost similar to that of a normal root. It consists of epiblema, differentiated cortex and vascular tissues.
2. **Epiblema is** outermost and single layered.
3. **The cortex** is divisible into three regions -outer, middle and inner. These are similar in size. Cortex parenchymatous.
4. **The middle** cortex is also called algal zone. The cells are radially elongated. A blue-green alga *Anabaena cycadae* occurs endophytically in these cells. It is believed to be symbiotic and helps in nitrogen fixation.
5. **Endodermis** separates cortex and vascular tissues . It is single layered and followed by many layered pericycle.
6. **Vascular bundles** are radial and xylem is triarch and exarch.

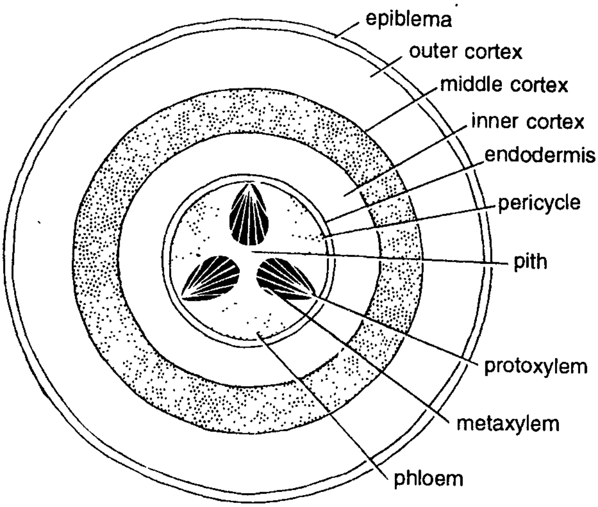


Fig. 6. *Cycas.* T.s. of coralloid root (diagrammatic).

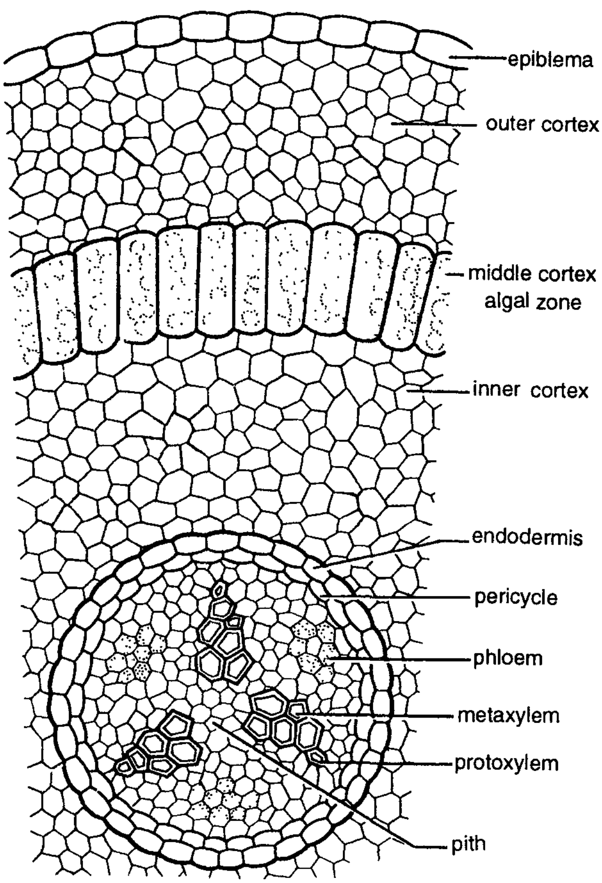


Fig. 7. *Cycas.* T.s. of coralloid root (a part cellular).

7. **Secondary** growth is generally absent; if present, it is verry less.

Exercise 5

Object : **Study the anatomy of young part of stem.**

Work procedure

Since the stem is very thick, unbranched and very slow growing, sections are not cut, instead a slice of stem cut transversely can be preserved as specimen. It shows some important anatomical characters.

Comments

1. **Outline** of the section is irregular due to the presence of numerous persistent leaf bases.
2. **The structure** is divisible into cortex, vascular tissue and pith.
3. **Cortex.** Greater part of the stem is made of starch filled parenchymatous cortex. It is traversed by many cut, girdle-shaped leaf traces, supplying the leaves. Many mucilage ducts are irregularly scattered in this region. (In *Cycas*, a leaf is supplied by two large girdle traces, two direct traces and numerous smaller radial traces. The two girdle traces arise from the side of the stele, opposite the leaf. These unite, bifurcate and take a circular route through the cortex before entering the leaf. The radial traces arise from other points of vascular ring but contrary to girdle traces, they adopt a straight radial course in the cortex. They bifurcate producing anastomosing branches which get attached to the girdle traces. In a trancverse section large number of girding leaf traces are cut. This is one of the most conspicuous features of the stem anatomy).
4. **Stele** is an ectophloic siphonostele.
5. **Endoermis** surrounds the stele. It is single layered while underlying pericycle is few celled thick
6. **Vascular cylinder** is composed of many vascular bundles arranged in a ring. Ring of vascular bundles lies near the centre and is very small in comparison to the massive cortex.
7. **The vascular bundles** are conjoint, collateral, endarch and open.

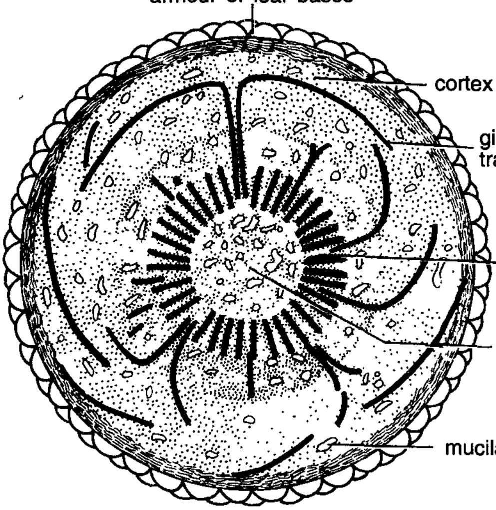


Fig. 8. ***Cycas.*** T.s. of young stem (diagrammatic).

8. **Xylem** is made of tracheids only and xylem parenchyma. Vessels are absent.
9. **Phloem** consists of sieve tubes, phloem parenchyma and phloem fibres.
10. The young stem is **monoxylic** (i.e. with one ring of vascular bundles only).
11. **Pith.** There is parenchymatous pith in the centre, with scattered mucilage canals.

Exercise 6

***Object* : Study of anatomy of the old stem.**

Work procedure

A thck slice of an old stem is generally preserved as specimen. Only prominent features could be observed.

Comments

1. It shows almost the same structures as those in young stem, except those formed after secondary growth.
2. **A periderm** is present on the outer side. It is composed of thick walled cells.
3. **Cortex** is large and parenchymatous. It forms most part of the section. A few mucilage canals are also present.

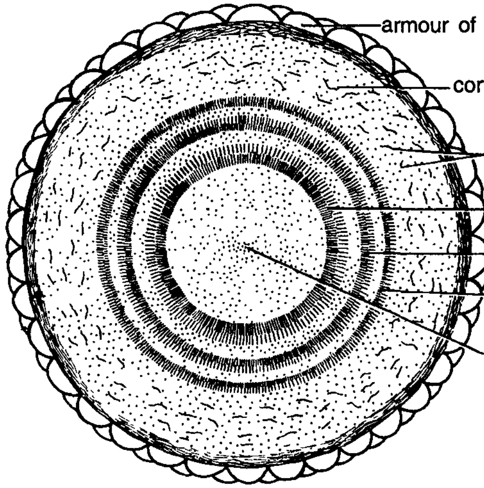


Fig. 9. ***Cycas.*** T.s. of old stem (diagrammatic).

4. **Vascular bundles** are formed, due to successive development of cambium rings. Thus, the old stem is **polyxylic** (with more than one ring of vascular bundles). The number of vascular bundles as well as the thickness of the successive vascular rings, thus formed, is lesser than the first formed ring.
5. **Secondary vascular tissues.** The successive rings of secondary vascular tissue are separated by parenchymatous zone. This loose, soft and scanty wood is called **manoxylic.**
6. **Medulllary rays** are present.
7. **Pith.** A large pith lies in the centre. Cells are parenchymatous and starch filled. Many mucilage canals are also present.

Exercise 7

***Object* : Study of anatomy of rachis.**

Work procedure

Cut a T.s. of rachis from its middle region, stain in sarfanin-fast green combination, mount in glycerine and study.

Comments

1. **Outline.** It is cylindrical. It shows insertion of pinnae on the adaxial side (upper side).

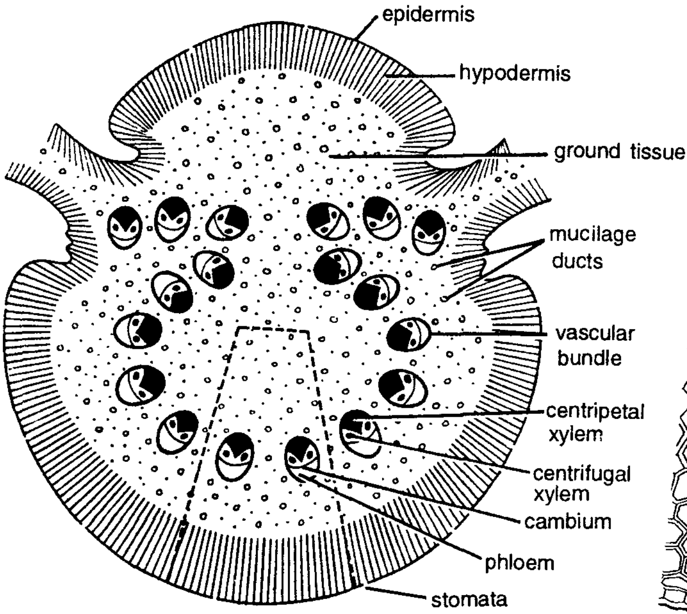


Fig. 10. *Cycas.* T.s. of rachis (diagrammatic).

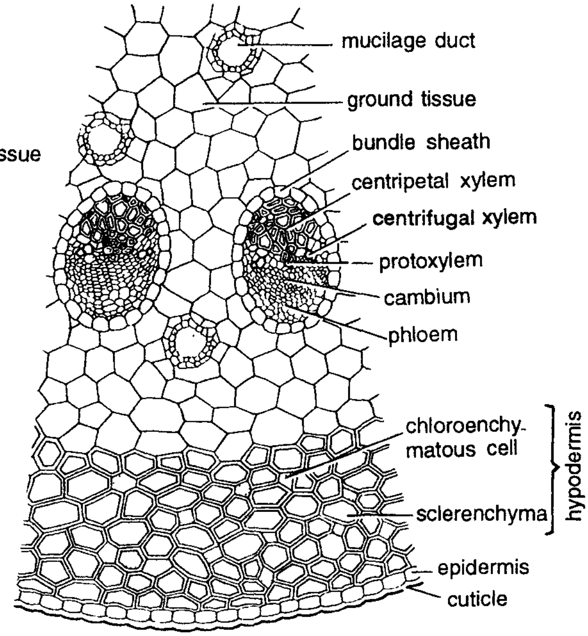


Fig. 11. *Cycas.* T.s. of rachis (a part shown by dotted lines in Fig. 11 in details).

2. **The rachis** is differentiated into epidermis, hypodermis, ground tissue and a ring of vascular bundles.
3. **Epidermis** is single layered, thickly cuticularized and is interrupted by stomata throughout its surface. The condition is known as amphistomatic.
4. **Hypodermis** is mainly composed of thick-walled cells (sclerenchyma). Intermixed with these cells are a few cells having chloroplasts-chlorenchyma.
5. This sclerenchymatous hypodermis is 2-3 layered toward adaxial side and many layered toward abaxial side.
6. **Ground tissue.** The rest of the tissue that forms most part of the section is called ground tissue. It is parenchymatous.
7. **Mucilage ducts** are scattered throughout the ground tissue. Mucilage ducts are double layered, the inner layer being composed of epithelial cells and the outer of tangentially elongated sclerenchymatous cells.
8. **The vascular bundles** are arranged in an inverted omega (Ω) shaped arc. Each vascular bundle is surrounded by a thick walled, single-layered bundle sheath. It is conjoint, collateral and open.
9. **The arrangement of xylem and phloem** differs in vascular bundles at the base, middle and upper region of the rachis.
   (i) Higher up and for most part of the rachis, bundles are **diploxylic** i.e. two types of xylem elements are present - centripetal and centrifugal xylem. The centrifugal xylem occurs in two small groups, present on both the sides of large triangular and centrally located centripetal xylem. The phloem is situated on the abaxial side of the rachis.
   (ii) At the very base of the rachis, vascular bundles show only centrifugal xylem which is endarch. Phloem occupies the abaxial side of the rachis.
   (iii) Little higher up the base of rachis, vascular bundles show centrifugal xylem on abaxial side and centripetal xylem on adaxial side. In the centre of these two xylem groups, lies the protoxylem. This conditionis is said to be mesarch.

Features of special interest

1. Presence of chlorenchyma, dispersed among the thick walled sclerenchymatous hypodermis.

2. Presence of sunken stomata all over the surface. (xerophytic characters).
3. Vascular bundles arranged in inverted omega (Ω) shaped arc.
4. Diploxylic nature of the vascular bundles.
5. Mucilage ducts scattered throughout.

Exercise 8

***Object* : Study of anatomy of leaflet (pinna).**

Work procedure

Cut a T.s. of leatlet, stain in safaranin-fast green combination, mount in glycerine and study.

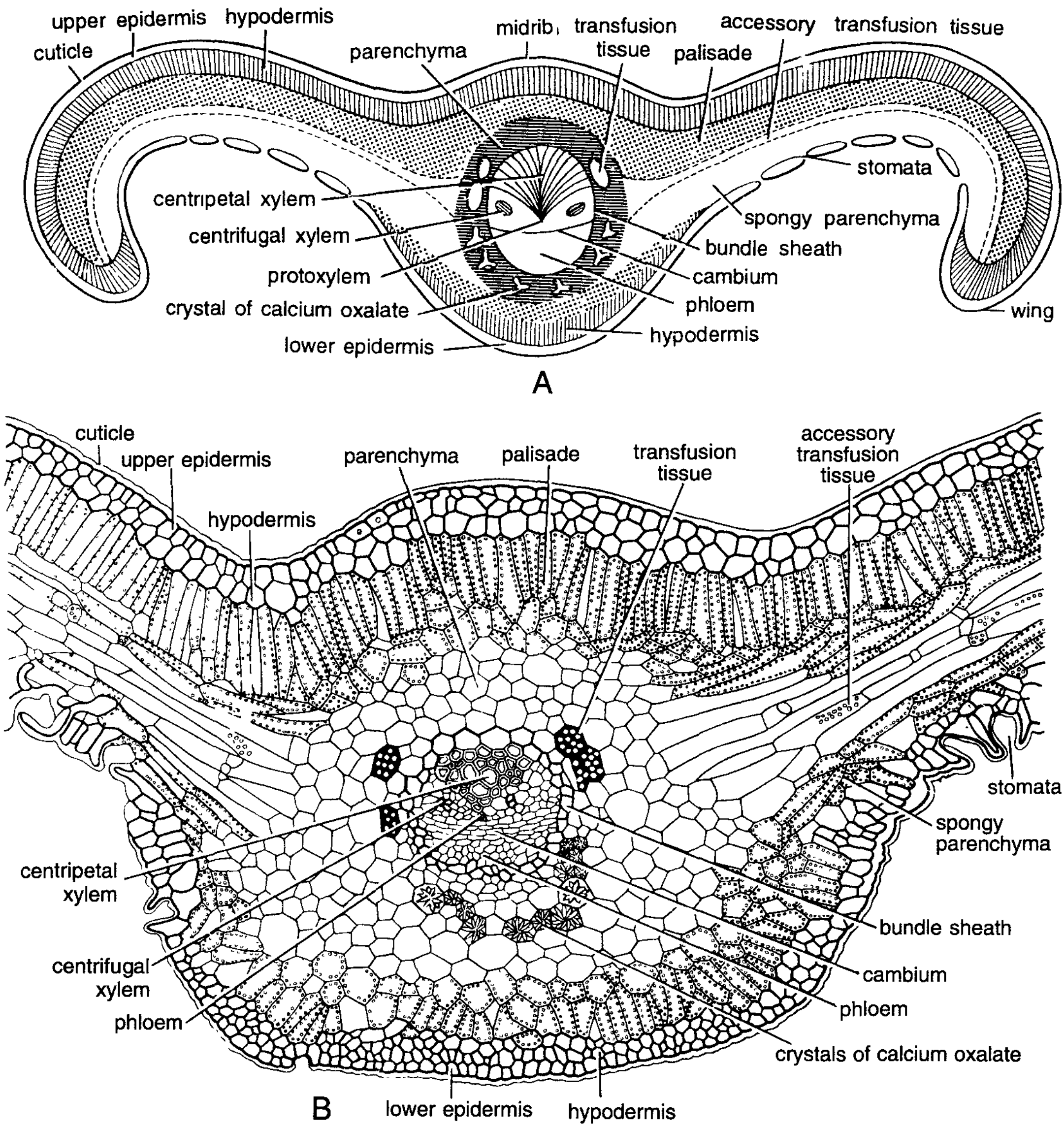


Fig. 12. ***Cycas.*** **A.** and **B.** T.s. of leaflet.

Comments

1. **The leaflet shows** a distinct midrib and the wings.
2. **The midrib** is swollen, while wings on the lateral sides are narrower and flattened.
   (i) In *C. revoluta* midrib is less projected than in *C. circinalis,* where it is much projected on the upper side.
   (ii) Margins of wings are revolute in *C. revoluta,* and *C. beddomei* while they are straight in *C. circinalis, C. rumphii, C. pectinata* and *C. siamensis.*
3. **Upper epidermis** is present on the upper side. It is thickly cuticularized and single-layered.
4. **Hypodermis** is present below the epidermis. It is sclerenchymatous.
   (i) In *C. revoluta*, hypodermis is present in the midrib (near both upper and lower epidermis) and wings (below the upper epidermis).
   (ii) In *C. circinalis*, hypodermis in the midrib region is present on both the sides (upper and lower) while in the wings, it occupies only the corners, being absent from rest of wings.
5. **Mesophyll** lies below the hypodermis and is well developed. It is differentiated into upper palisade layers and lower of spongy parenchyma.
   (i) In *C. revoluta*, palisade is present beneath the hypodermis, both in the midrib and the wings.
   (ii) In *C. circinalis* palisade is absent from the midrib region
6. **Spongy parenchyma** with many intercellular spaces lies immediately above the lower epidermis.
7. **Transfusion tissue.** On either side of the centripetal metaxylem of mid rib bundle and somewhat connected with it, are present two tracheid-like cells-transfusion tissue.
8. **Accessory transfusion tissue.** Between the palisade and spongy parenchyma cells, there are 3 or 4 layers of tracheid-like, long colourless cells which run transversely from the midrib to near the margin of the lamina. This is known as accessory transfusion tissue. It is connected with the xylem of the vascular bundle of midrib through the transfusion tissue.
9. **Lower epidermis** bounds the leaflet from lower side. It is thickly cuticularized and single layered. Sunken stomata are found in the lower epidermis in the midrib region.
10. **Stomata** are very much sunken in the lower epidermis in *C. revoluta*, while they are not so much sunken in *C. circinalis.*
11. **Midrib bundle.** In middle of the swollen portion representing the midrib lies a single vascular bundle surrounded by parenchymatous tissue (with calcium oxalate crystals). Vascular bundle has a definite and thickened, parenchymatous bundle sheath.
12. **The vascular bundle** is similar in all respects to that found in the upper region of the rachis. It is conjoint, collateral, open and diploxylic.
13. **Phloem** lies towards the abaxial (lower) side. In between xylem and phloem, cambium is present.
14. **Xylem.** It shows a large, triangular patch of centripetal xylem and two small groups of centripetal protoxylem.

Features of special interest

1. Lateral veins are absent.
2. Thickly cuticularized upper and lower epidermis.
3. Sunken stomata in the lower epidermis.
4. Presence of transfusion tissue.
5. Diploxylic nature of vascular bundle.

Exercise 9

Object : **Study of bulbil.**

Work procedure

Study the position on the lower part of the stem and observe external characters.

Comments

1. Bulbils are produced adventitiously, on basal part of the plant, in the crevices between the persistent leaf bases.
2. The decurrent base of the bulbil remains covered with scale leaves.
3. A few foliage leaves are given out from the central part.

4. It germinates under favourable conditions and produces new plant or else develops into a branch (rarely), giving an appearance of dichotomous branching.
5. This is the commonest method of reproduction in *C. revoluta* in north india, male plants being rare in this region.

Exercise 10

***Object* : Study of external features of male cone.**

Work procedure

Study the male cone attached to the plant if possible.

Comments

1. The male cone is terminal, shortly stalked, compact, large and oval or conical in shape and consists of a central cone axis around which numerous microsporophylls are spirally arranged. (Since the male cone terminates the growth of the apex of male plant, a lateral bud later grows and takes over the continuation of growth of apex. The male plant thus shows sympodial growth).
2. The outer covering of the male cone is formed by closely set sterile ends of the microsporophylls usually possessing upcurved apices, apophysis.

Exercise 11

***Object* : Study of L.s. of the male cone.**

Work procedure

Since the male cone is about 60-80 cms in height, it can be split into two and specimen is studied.

Comments

1. The L.s. shows stalk and the cone.
2. Male cone is attached at the apex of the plant by a stout and broad stalk.
3. The cone itself consists of a central cone axis with many microsporophylls.
4. Each microsporophyll is attached to the cone axis. The part of microsporophylls away from the axis is upcurved and is called apophysis.

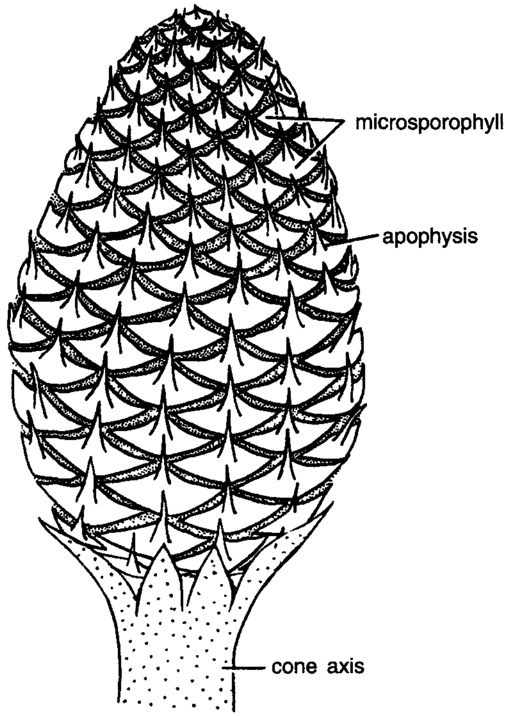


Fig. 13. ***Cycas.*** A male cone (external features).

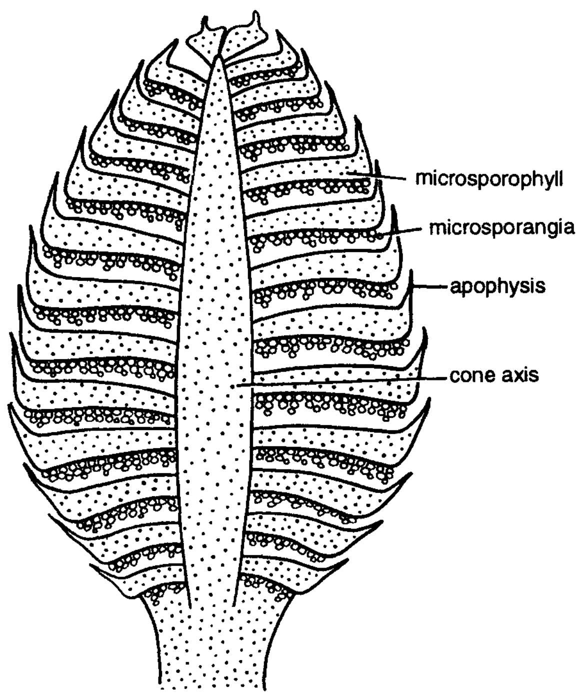


Fig. 14. ***Cycas.*** L.s. of male cone.

5. The upper surface of the microsporophyll is sterile.
6. The lower surface of the microsporophyll is fertile and bears many microsporangia in groups (sori).
7. Microsporophylls in the middle part of the cone are largest and get gradually smaller towards the base and the apex.

Exercise 12

Object **: Study of microsporophyll and microsporangia.**

Work procedure

Take out a microsporophyll from the male cone. Study both- upper and lower surfaces. Observe the sporangia on the lower surface with a magnifying lens.

Comments

1. A single microsporophyll is woody, more or less horizontally flattened and triangular structure.
2. It is differentiated into a fertile and sterile parts. Fertile part is wedge-shaped and is expanded distally from a narrow point of attachment. Sterile part is the distal part of the microsporophyll which tapers into an upcurved apophysis.
3. Lower (abaxial) surface of the fertile part of the microsporophyll bears microsporangia in groups of 3-4, forming definite sori.
4. Microsporangia are arranged in sori around central papilla. Sporangia show radial lines of dehiscence. Many hairs are distributed on this surface mixed with sporangia.

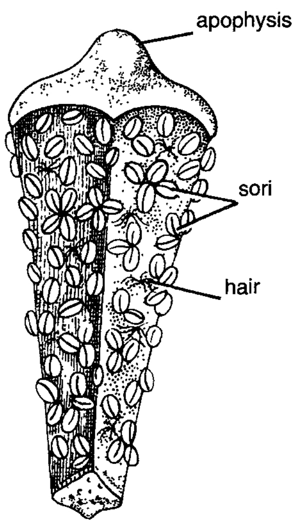


Fig. 15. ***Cycas.*** A microsporophyll from lower side.

Exercise 13

Object **: Study of T.s. of microsporophyll.**

Work procedure

Study the characters observed in sliok of T.s. of microporophyll.

Comments

1. The section shows microsporangia attached to the abaxial (lower) surface by their short stalks.
2. A mature microsporangium has three layered wall. The outermost layer is thick and cutinized, termed as exothecium. The remaining inner layers are thin and are collectively known as endothecium and enclose a tapetum.
3. Numerous microspores remain enclosed inside the wall of the microsporangium.
4. In the microsporophyll are present many mucilage ducts, regularly scattered, among the rounded mesophyll-like cells forming the tissue of the sporophyll.

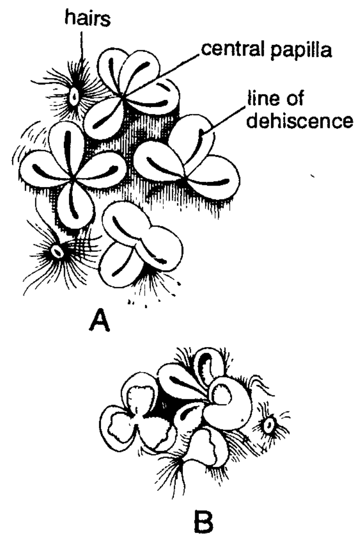


Fig. 16. ***Cycas.*** Microsporangia in sori. **A.** Before dehiscence, **B.** After dehiscence.

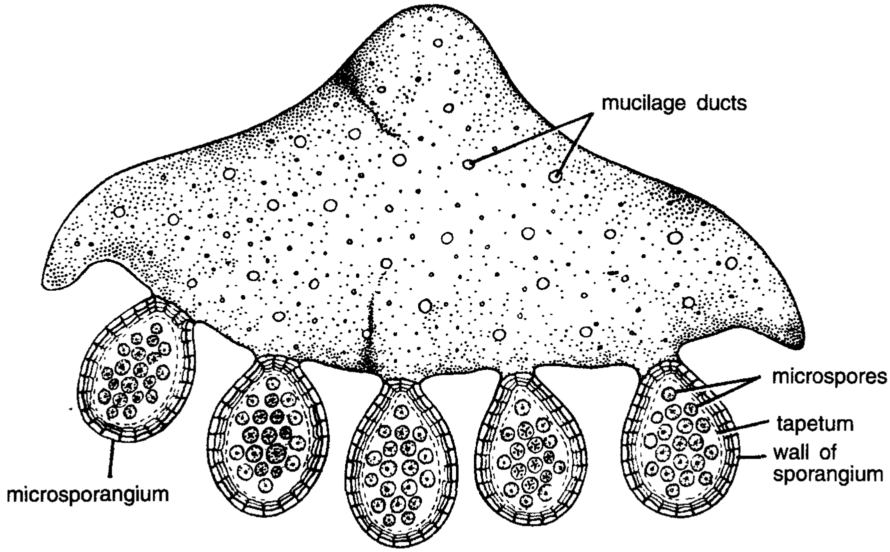


Fig. 17. ***Cycas.*** T.s. of microsporophyll.

Exercise 14

***Object* : Study of megasporophyll.**

Work procedure

Since there is no female cone, megasporophylls form a crown at the apex like foliage leaves. Only a megasporophyll can be studied as a specimen.

Comments

1. Female reproductive body consists of megasporphylls arranged spirally and arising in acropetal succession on the stem.
2. Megasporophylls appear as a rosette or a crown, leaving the apical meristem unaffected to grow further. A crown of megasporophyll is formed each year. Numerically they are more than the leaves.

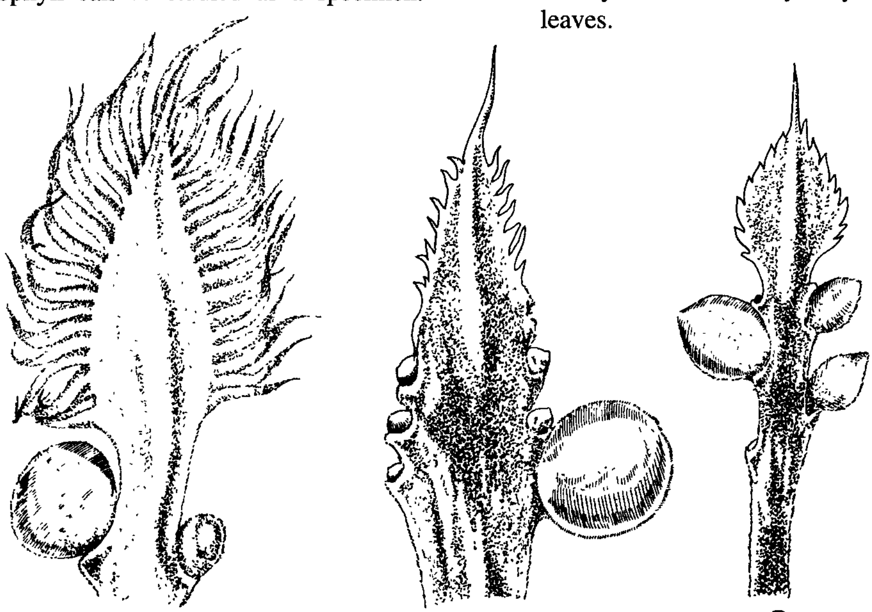


Fig. 18. ***Cycas.*** Megasporophylls of different species with ovules. **A.** *C. revoluta,* **B.** *C. circinalis,* **C.** *C. rumphii.*

3. They leave their persistent bases on the stem.
4. Each megasporophyll is leaf-like and densely covered with brown hairs. It varies in size from 6 to 12 inches.
5. Each megasporophyll is distinguished into a proximal (lower) petiole, a middle ovule bearing portion and a distal (upper) pinnately dissected sterile part.
6. The nature of upper sterile part varies with species.
   (i) In *C. revoluta*, the upper part is very much dissected, forming many pinnae.
   (ii) In *C. rumphii*, the upper part bears only short spines which represent reduced pinnae.
   (iii) In *C. circinalis*, the pinnate character is altogether absent and upper part shows only dentate or serrate margins.
7. The middle portion of sporophyll bears ovules which are borne in two rows, one on either side. The ovules of the two rows may be opposite or alternate.
8. Ovules are generally yellow or orange or dark green coloured, shortly stalked, oval and smooth. Number and size of the ovules differ from species to species.
   (i) In *C. revoluta*, ovules are many and orange coloured,
   (ii) In *C. circinalis* also, they are numerous, but these are dark green and attain a large size,
   (iii) In *C. siamensis*, the number of ovules is reduced to only two and
   (iv) In *C. thouarsii*, the ovules become still larger and may be that they are the largest ovules in the plant kingdom.
9. All the ovules do not develop fully. Some of those which remain unpollinated and small, finally abort.

Exercise 15

***Object* : Study of L.s. of mature ovule.**

Work procedure

Study the slide showing L. s. of mature ovule.

Comments

1. The section shows that the ovule is orthotropous.
2. It is unitegmic (possesses a single integument). The integument is very thick. It remains fused with the nucellus except for the nucellar beak leaving a small and narrow micropyle.

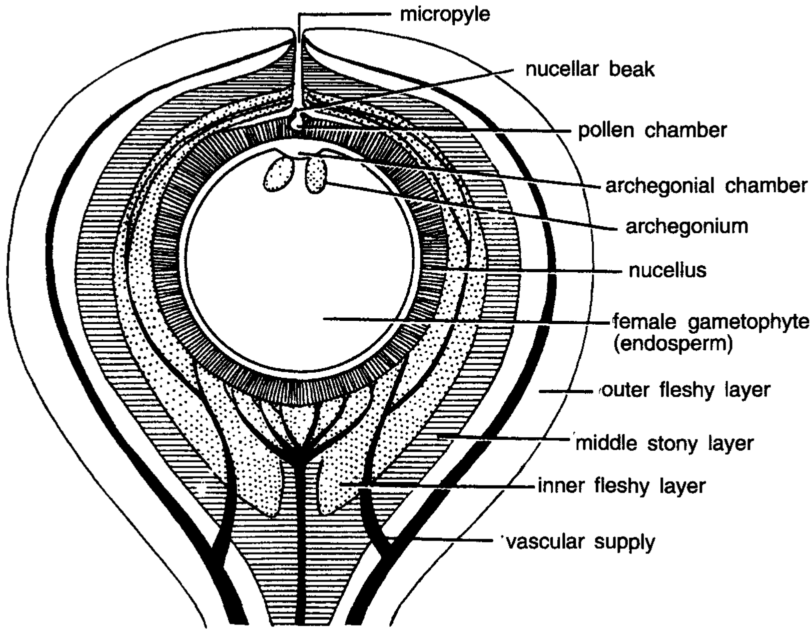


Fig. 19. ***Cycas.*** L.s. of ovule.

3. **The integument** consists of three distinct layers- an outer fleshy layer, middle stony layer and an inner fleshy layer. The outer and inner flshy layers are supplied with vascular strands but the middle stony layer receives no vascular supply.
4. **The nucellus** lies just below the integument and forms a nucellar beak in the region of the micropyle.
5. A few cells of this nucellar beak dissolve themselves and form a pollen chamber that lies in the tissue in the central region of the beak.
6. **Female gametophyte.** The innermost region of the ovule is filled with the tissue of female gametophyte, wherein lie two archegonia, situated opposite the pollen chamber.
7. **Archegonial chamber.** Just above the archegonia is the archegonial chamber.
8. **Micropyle.** The orange coloured, fleshy ovules are oval in shape and each shows a small point at the distal end which represents the remnant of the micropyle.

Exercise 16

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

Work procedure

Study a double-stained preparation of the L.s. of seed.

Comments

1. It shows seed coat, nucellus, embryo and the female gametophyte.
2. **Seed coat** consists of sarcotesta (from outer fleshy layer of integument), and middle sclerotesta (from middle stony layer). The inner fleshy layer of the integument appears as thin and papery structure in the seed.
3. **Nucellus** is papery and is situated inside the seed coat.
4. **Endosperm** and female gametophyte form the inner part of seed.
5. **A straight embryo** remains embedded in the endosperm. It has two unequl cotyledons.

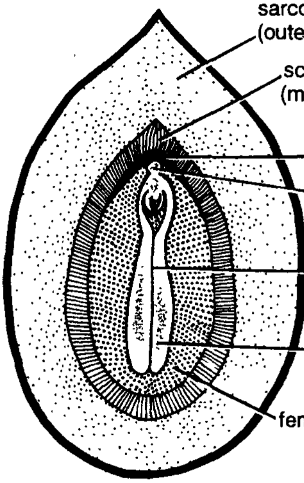


Fig. 20. *Cycas.* L.s. of seed.

Identification

Division—**Gymnosperms.** (1) Absence of vessels, (2) Ovules naked, (3) Seeds attached with woody acales, (4) Scales generally form a cone.

Class—**Cycadopsida.** (1) Wood manoxylic, (2) Large frond-like leaves (3) Seeds with radial symmetry.

Order—**Cycadales.** (1) Plants woody, stem unbranched, (2) Wood manoxylic, (3) Pressence of mucilage canals, (4) Leaf trace diploxylic, (5) Dioecious, (6) Ovules orthotropous, (7) Sperm with band of flagella.

Family—**Cycadaceae.** (1) Leaves with circinate vernation, (2) Presence of coralloid roots and endophytic blue green algae, (3) Megasporophylls foliar.

Genus—***Cycas.*** (1) Two types of leaves, (2) Foliage leaves pinnately compound, circinately coiled when young, (3) Presence of trasfusion tissue and diploxylic pascular bundle in leaf, (4) Secondary xylem in stem manoxlic, (5) Two types of roots, (6) Vascular bundles arranged in an inverted omega-shaped manner in the rachis, (7) Male cone large and single.

Hints for Collection

In India the genus grows naturally only in the north-east (East Nepal, Champaran-Bihar, Sikkim, Assam and East Bengal) and South (Orissa, Andhra Pradesh and Tamil Nadu). Six species are found in India of which *C. circinalis, C. pectinata. C. rumphii,* and *C. beddomei* grow wild but *C. revoluta,* a native of Japan and *C. siamensis* found in Burma and Siam are cultivated.

Pinus (Pine)

Classification

Division	–	**Gymnosperms**
Class	–	**Coniferopsida**
Order	–	**Coniferales**
Family	–	**Pinaceace**
Genus	–	***Pinus***

Exercise 1

Object **: Study of external morpohology.**

Work procedure

Note the pattern of branching, the two types of branches, two types of leaves, and male and female cones.

Comments

1. It is a tall conical tree and, therefore, commonly grouped under conifers.
2. **The plant body** is differentiated into root, stem and leaves.
3. **Underground root system** is formed by tap roots which disappear early and only lateral roots persist later on.
4. **The younger roots** are generally surrounded by fungal hyphae- the ectotrophic mycorrhizae.
5. **Aerial branch system** consists of cylindrical, rough (being covered with scaly bark) and branched stem.
6. **The branching** is monopodial and the branches are arranged in whorls.
7. **The branches** are dimorphic (of two types)- branches of unlimited growth or long shoots and branches of limited growth or dwarf shoots.
8. **Branches of unlimited growth** or long shoots are present on the main trunk. These are produced at regular intervals.
9. **Branches of limited growth** or dwarf shoots are borne on the main stem and on long shoots in the axils of scale leaves. Dwarf shoots also possess many scale leaves and bear group of foliage leaves at the apex.

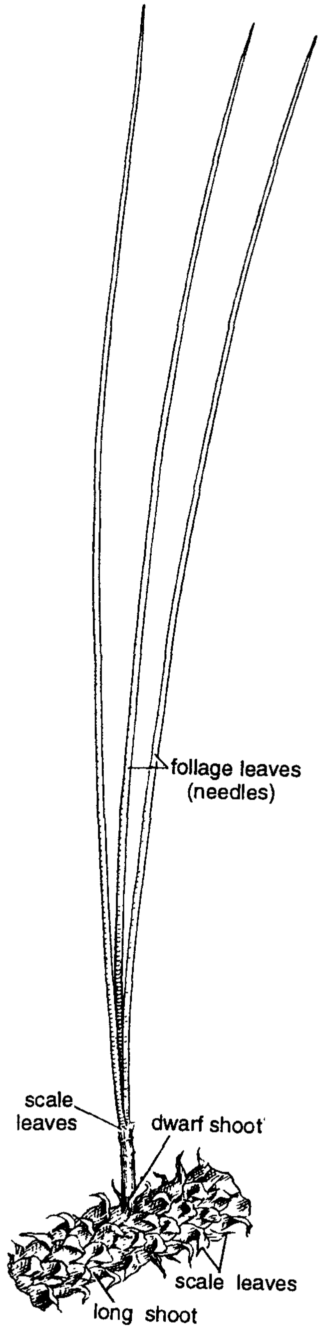


Fig. 1. ***Pinus.*** A part of stem showing two types of branches.

10. **The leaves** are also dimorphic (of two types)- scale leaves and foliage leaves.
11. **Scale leaves** are brown, membranous and small. They are present on both the types of branches (i.e. long and dwarf shoots).

12. **Foliage leaves** are green, acicular and needle-like. They are borne only by the dwarf shoots.
13. **A dwarf shoot** with a group of needle-like foliage leaves is known as a foliar spur. The number of needles in a group varies from species to species. *P. monophylla* has a single leaf and spur is known as monofoilar, while in *P. sylvestris*, two leaves are present and spur is called as bifoiiar. In *P. longifolia* and *P. gerardiana*, they are three in number, the spur being called as trifoliar. Quadrifoilar spur occurs in *P. quadrifolia* and pentafoliar in *P. excelsa*.
14. **The shape of the needle** varies, according to their number in a spur. In *P. sylvestris* (with bifoliar spur), single needle is semi-circular in T.s. while in *P. longifolia* (with trifoliar spur), single needle is almost triangular in shape.
15. ***Pinus* is monoecious.** Plant bears male and female reproductive parts in cones on the same plant .
16. **The male cones** are borne on lateral branches of unlimited growth. They are produced in clusters and replace the dwarf shoots. Also, they are formed earlier in the season than the female cones.
17. **The female cones** are borne terminally on branches of unlimited growth. They are produced singly and replace the long shoot. The female cone appears after every three years.
18. Generally male and female cones are not formed on one and same branch.

Exercise 2

***Object* : Study of anatomy of young root.**

Work procedure

Study a double stained prepared slide of T.s. of young part of root.

Comments

1. **The section** is almost circular in outline.
2. The tissues are differentiated into epiblema cortex and vascular tissues.
3. **Epiblema** is outermost single layer. It gives out many thin and unicellular root hairs.

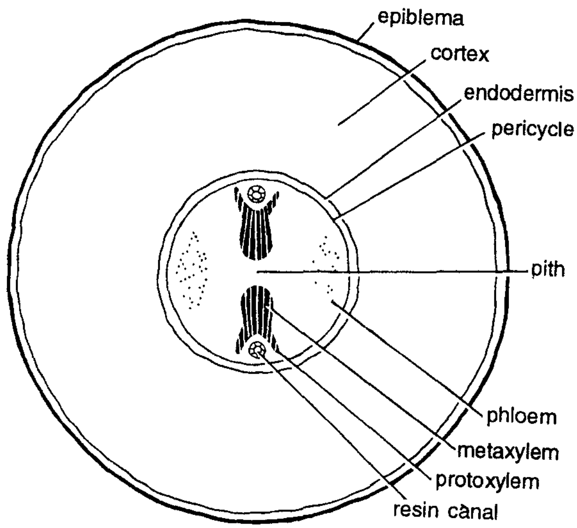


Fig. 2. ***Pinus.*** T.s. of young root (diagrammatic).

4. **Cortex** is multilayered and parenchymatous.
5. **Endodermis** separates outer cortex and central vascular cylinder. It is single layered and cells are radially thickened.
6. **Pericycle** follows endodermis. It is multilauered.
7. **Vascular bundles** are radial, exarch and diarch to hexarch.
8. **Protoxylem** is generally Y-shaped and a resin canal is present in between the arms of Y.
9. **Pith** is very small and lies between the groups of xylem.

Exercise 3

***Object* : Study of anatomy of the old root.**

Work procedure

Study a double stained prepared slide of T.s. of old part of root.

Comments

1. **The section** shows cork, cortex, primary and secondary vascular tissues and a small pith.
2. **Cork** forms the outermost several layers. (developed from pericycle and hence primary cortex is completely peeled off).
3. **Stone cells** occur in many groups scattered just below the zone of cork.

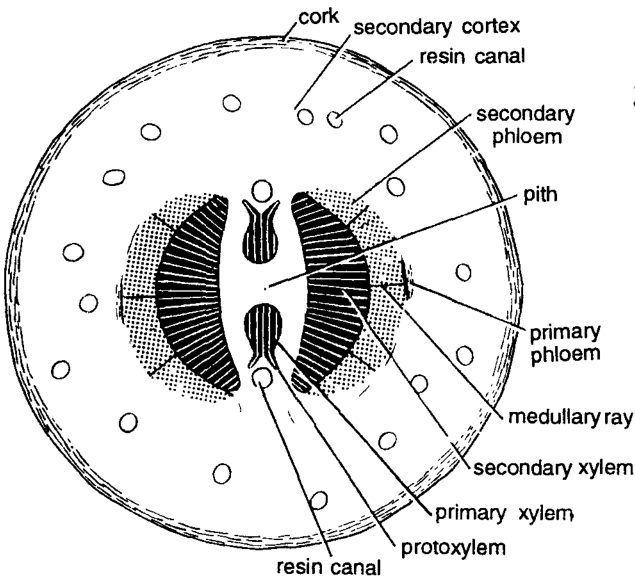


Fig. 3. ***Pinus.*** T.s. of old root (diagrammatic).

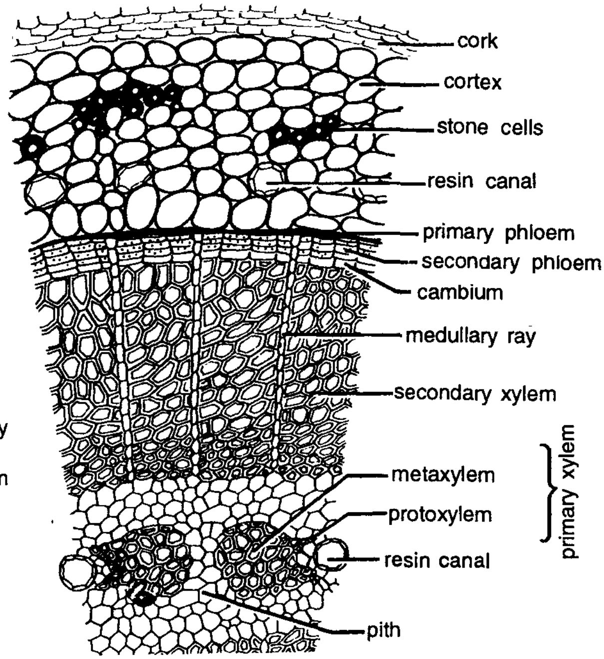


Fig. 4. ***Pinus.*** T.s. of old root (a part cellular).

4. **Secondary cortex** follows cork. It is parenchymatous and a few layered deep.
5. **Many resin canals** are found in the secondary cortex.
6. **Primary phloem** occurs in two patches. The tissues are mostly crushed and obliterated.
7. **Secondary phloem** that follows is a few layered deep ring. It consists of sieve tubes, sieve plates, phloem parenchyma and albuminous cells.
8. Secondary phloem and secondary xylem are separated by a cambium.
9. **Secondary xylem** is composed of tracheids arranged in regular rows. It is traversed by uniseriate medullary rays.
10. **Pith** is small and parenchymatous. Two groups of primary xylem are situated on opposite radii.
11. **Each primary xylem** group is Y shaped. The divided arm faces the outer side (away from the pith).
12. **Resin canal.** The characteristic of the pine root is the presence of large resin canal between the divided arm of Y, close to each primary protoxylem group.

Exercise 4

Object **: Study of anatomy of the young long shoot.**

Work procedure

Cut a T.s. of younger part of the long shoot towards the apex, stain in safranin-fast green combination, mount in glycerine and study.

Comments

1. **Outline** is wavy due to the presence of scaly leaves.
2. The stem is differentiated into epidermis, cortex and stele.
3. **Epidermis** is the outermost single layer. It is thickly cuticularized.
4. **Cortex** is multilayered and lies below the epidermis. The outer few layers forming hypodermis are sclerenchymatous. Inner layers are thin walled and parenchymatous in which

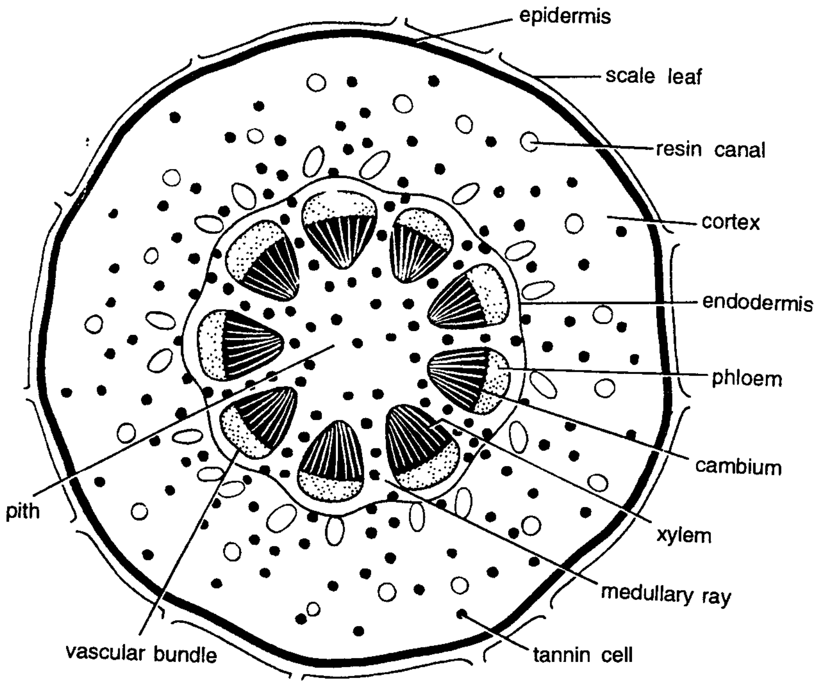


Fig. 5. ***Pinus.*** T.s. of young long shoot (diagrammatic).

large number of resin canals and leaf traces are distributed irregularly.

5. **Resin canal.** The cavity of resin canal is bounded by a glandular, resin secreting epithelial layer. Outer to this layer are one or two layers of sclerotic cells.
6. In *Pinus*, resin canals are present in the cortex and secondary wood of both stem and root and on margins of the primary xylem in the root.
7. **The stele** is ectophloic siphonostele.
8. **Endodermis** is present but is undistinguishable and so also a few layered pericycle located inner to it.
9. **Vascular cylinder** is composed of 5-8 vascular bundles, separated by medullary rays. Vascular bundles are arranged in a ring.
10. **Each vascular bundle** is conjoint, collateral endarch and open.
11. **Xylem** is composed of tracheids and xylem parenchyma only, vessels are absent.
12. **The phloem** is made up of sieve tubes, sieve plates and phloem parenchyma. Albuminous cells are also present.
13. **Pith** lies in the centre and is parenchymatous. It is connected with the cortex but narrow medullary rays separate the vascular bundles.

Exercise 5

Object **: Study the anatomy of the old long shoot.**

Work procedure

Cut a T.s. of the old part of the stem, stain with safranin-fast green combination, mount in glycerine and study.

Comments

1. **The section** shows cork, cortex, primary and secondary vascular tissues and pith.
2. **Cork.** The outmost region is formed by the successive layers of cork. It consists of thick and suberized cells.
3. **Cork cambium** follows cork. It is made of a few layers of regularly arranged cells.
4. **Secondary cortex** present below is parenchymatous.

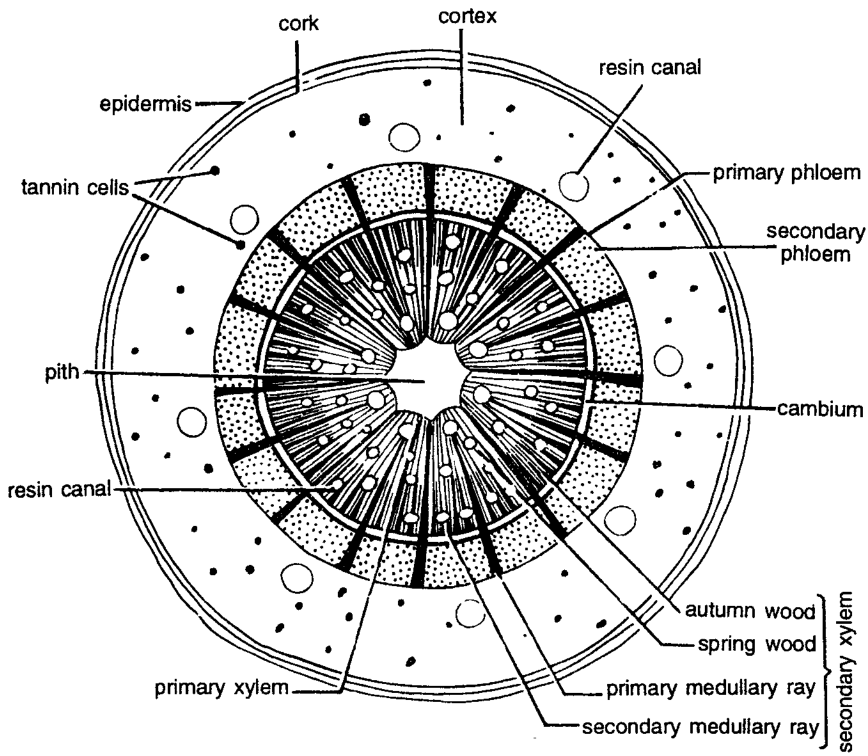


Fig. 6. ***Pinus.*** T.s. of old long shoot (diagrammatic).

5. **Primary cortex** is parenchymatous and many layered. The resin canals occur irregularly distributed in this region.
6. **Primary phloem** that lies inner to primary cortex occurs as small patches of crushed tissues.
7. **Secondary phloem** occurs as a well distinguished ring.
8. **Phloem** is composd of sieve tubes and phloem parenchyma.
9. **Cambium** separates the secondary phloem on its outer side and secondary xylem on its inner side.
10. **Secondary xylem** shows distinct and sharp annual rings. Thin walled and large xylem elements form a ring of spring wood. Thick walled and small xylem elements form a ring of autumn wood. The wood is pyconoxylic (compact).
11. **Rings of secondary xylem** — autumn and spring wood alternate one another and together form annual ring.
12. **Secondary xylem** (wood) is composed of tracheids and xylem parenchyma. Vessels are completely absent. Hence it is called non-porous wood.
13. **Medullary rays** traverse xylem and phloem. Primary medullary rays run from primary xylem to secondary phloem.
14. **Primary xylem** groups are endarch and lie just near the pith.
15. **Resin canals** are scattered in the primary and secondary xylem as in the cortex.
16. **Pith** is small, parenchymatous and many cells are filled with tannin.

Exercise 6

Object **: Study of R.L.s. of the wood.**

Work procedure

Cut a thin section of wood along any one of the radii, stain in safranin-fast green combination, mount in glycerine and study.

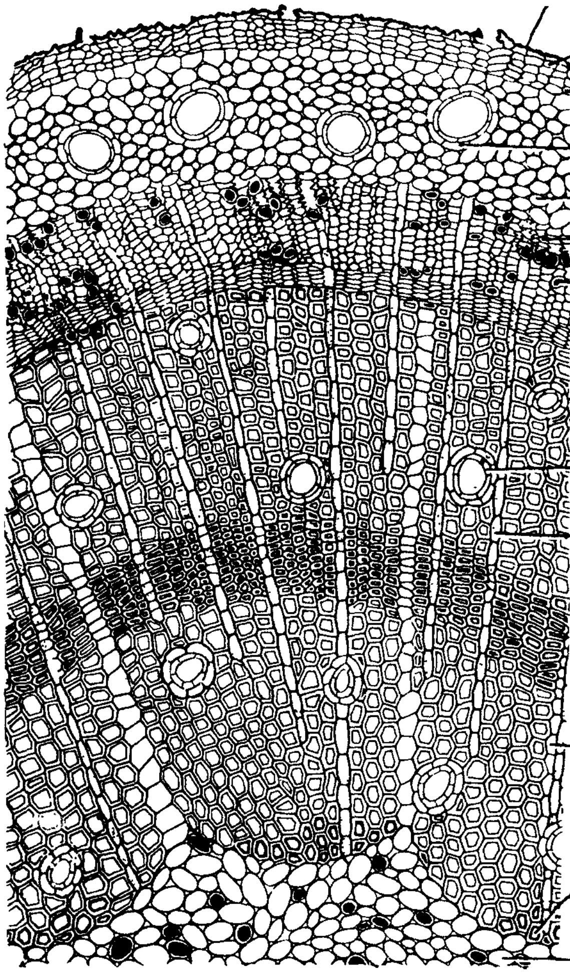


Fig. 7. ***Pinus.*** T.s. of old long shoot a part cellular.

Comments

1. It shows presence of secondary xylem, ray tracheids and medullary rays.
2. **Xylem** is composed of tracheids with bordered pits on their radial walls. The bordered pits in this section are seen in surface view.
3. **Bordered pits** are circular areas surrounded by special cellulose thickenings called crassulae or Bars of Sanio. If pits are close to one another, the bars fuse to form Rims of Sanio.
4. **Medullary rays** run horizontally. In radial longitudinal plane they are cut length-wise and their length and height can be noticed. They are uniseriate.
5. **Each medullary ray** is made up of ray cells, ray tracheids and parenchyma.
6. **Ray tracheids** are present on both the sides of the medullary ray cells, only in the region of

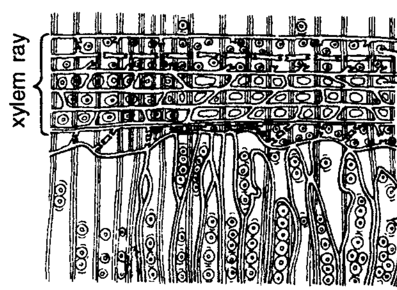


Fig. 8. ***Pinus.*** R.L..s. of wood (a part cellular).

xylem. These cells are thick, narrow and long. They show bordered pits.

7. **Ray parenchyma** occurs between the tracheids. These cells are thin, broad, small and living.
8. **Medullary ray**, in the region of phloem replaces ray tracheids with albuminous cells. They are small and contents are dense. (Ray parenchyma associated with these cells is filled with large amount of starch).

Exercise 7

***Object* : Study of T.L.s. of wood.**

Work procedure

Cut a thin section of wood along the tangent in the outer region, stain in safranin-fast green combination, mount in glycerine and study.

Comments

1. Tracheids and medullary rays are cut transversely in this plane.
2. **The bordered pits** are cut to show ovearching borders, forming a dome-like structure. It encloses in the centre a small disc, called torus.
3. **Medullary rays** are uniseriate. Since they are cut transversely, their height and breadth can thus be determined.
4. **Each medullary ray** appears to be a short row of more or less rounded cells, three or four cells high.
5. Composition of medullary ray reveals centrally placed, thin-walled and living cells-the albuminous cells (in the phloem region) and the ray cells (in the xylem region).
6. These are surrounded on the lower and upper sides by thick walled and dead cells known as ray tracheids.

Exercise 8

***Object* : Study of T.s. of dwarf shoot at the base (before secondary growth).**

Work procedure

Take out a spur, invert it with needles downwards, the base is now the uppermost, cut T.s., at the base mount in glycerine and study.

Comments

1. **The section** almost resembles with that of the main stem.

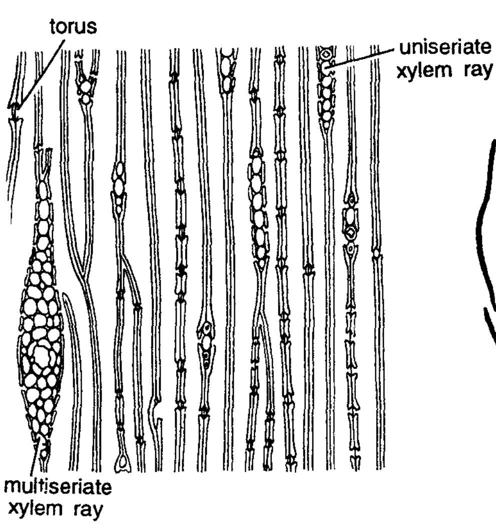


Fig. 9. ***Pinus.*** T.L.s. of wood (a part cellular).

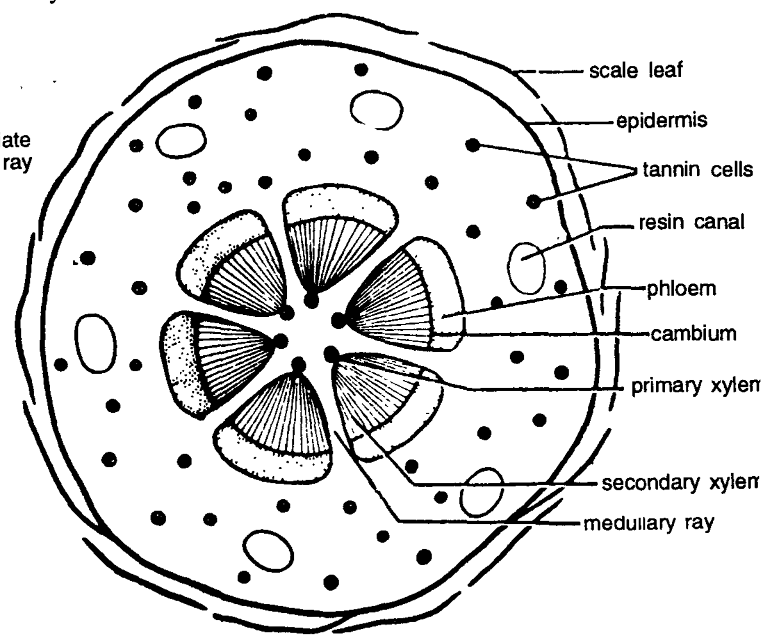


Fig. 10. ***Pinus.*** T.s. dwarf shoot showing secondary growth (diagrammatic).

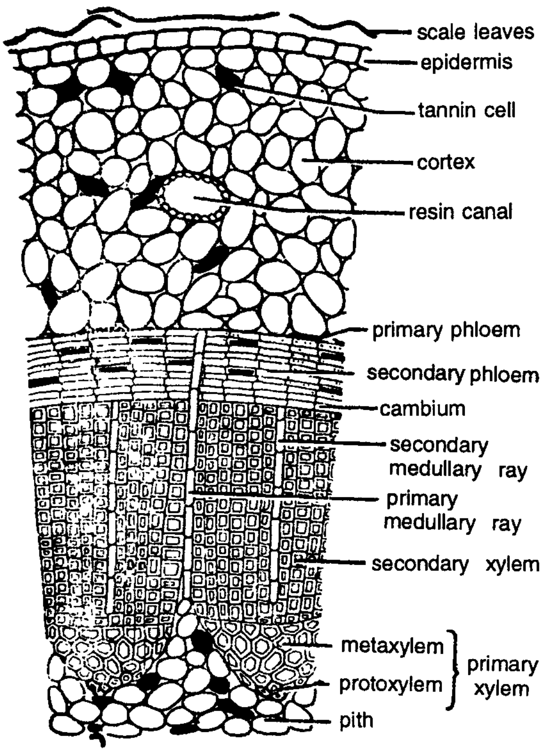


Fig. 11. ***Pinus.*** T.s. of dwarf shoot showing secondary growth (a part cellular).

2. **The outline** is wavy, due to ensheathing scaly leaves.
3. **The tissues** are differentiated into epidermis. cortex and stele.
4. **Epidermis** is made of single layer of thick walled cells.
5. **Cortex** follows epidermis. Outer few layers, close to epidermis are thick walled, while the inner layers are thin walled and parenchymatous.
6. **Resin canals** are present in the cortex. These are about six in number. Tannin cells are also irregularly scattered in this region.
7. **Stele** is an ectophloic siphonostele.
8. **Endodermis** is single layered and is followed by pericycle. Both the layers are indistinguishable.
9. **Vascular bundles** vary in number. They are generally six. Each vascular bundle is conjoint, collateral, endarch and open.
10. **Pith** is small. The cells are thick walled.
11. **Medullary rays** connect the pith and cortex and separate vascular bundles from one another.

Exercise 9

Object : **Study of T.s. of dwarf shoot at base.**

Work procedure

Cut a T.s. of dwarf shoot above the base, stain in safranin-fast green combination, mount in glycerine and study.

Comments

1. Dwarf shoot also shows a little amount of secondary growth.
2. **The section** shows scale leaves, single-layered thick walled epidermis, few layers of cork cells and tannin-filled cells.
3. **Primary phloem** is crushed and form patches. Secondary phloem underlies it and forms a complete ring.
4. **Secondary xylem** is small and is separated by a thin ring of cambium from the phloem region. Medullary rays traverse the secondary xylem.
5. **Endarch protoxylem** group lies just near the pith. It is small and consists of thick-walled cells. Few cells are tannin-filled.

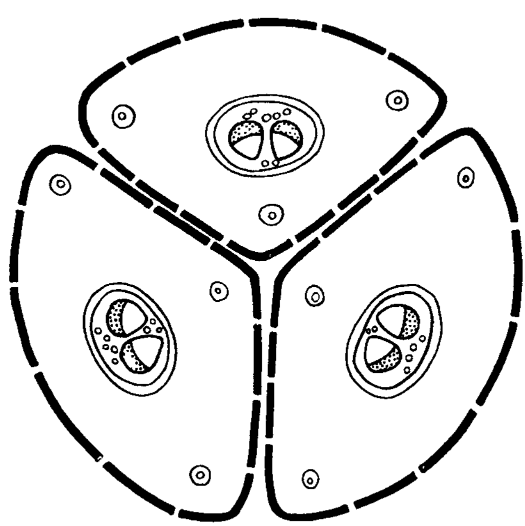


Fig. 12. ***Pinus.*** T.s. of dwarf shoot upper region (diagrammatic).

Exercise 10

***Object* : Study of T.s. of the dwarf shoot at upper end.**

Work procedure

Hold the dwarf shoot in upright position, cut a T.s., stain in safranin and fast green combination, mount in glycerine and study.

Comments

1. The structure is essentially similar to one found at its base.
2. The tissues of the dwarf shoot, towards the upper part, gradually become separated into equal parts, corresponding to the number of leaves in a spur (e.g. *Pinus gerardiana,* with trifoliar spur shows division of the dwarf shoot into three equal parts while in *P. quadrifolia,* with quadrifoliar spur, gets separated into four equal parts).
3. Each part shows distinct epidermis with stomata present all over.
4. Parenchymatous cortex fills most part of the section. Resin canals are located in the corners.
5. In the centre two conjoint, collateral and endarch vascular bundles are present. These are surrounded by distinct endodermis and pericycle.

Exercise 11

***Object* : Study of T.s. needle (leaf).**

Work procedure

Cut a thin T.s. of a needle, stain with safranin-fast green combination, mount in glycerine and study.

Comments

1. **The outline** of the section varies according to the species. (Triangular if spur is trifoliar, semi-circular if spur is bifoliar)
2. The needle is differentiated into epidermis, mesophyll and stele.
3. **Epidermis** is single with tangentially elongated and thickly cuticularized cells.

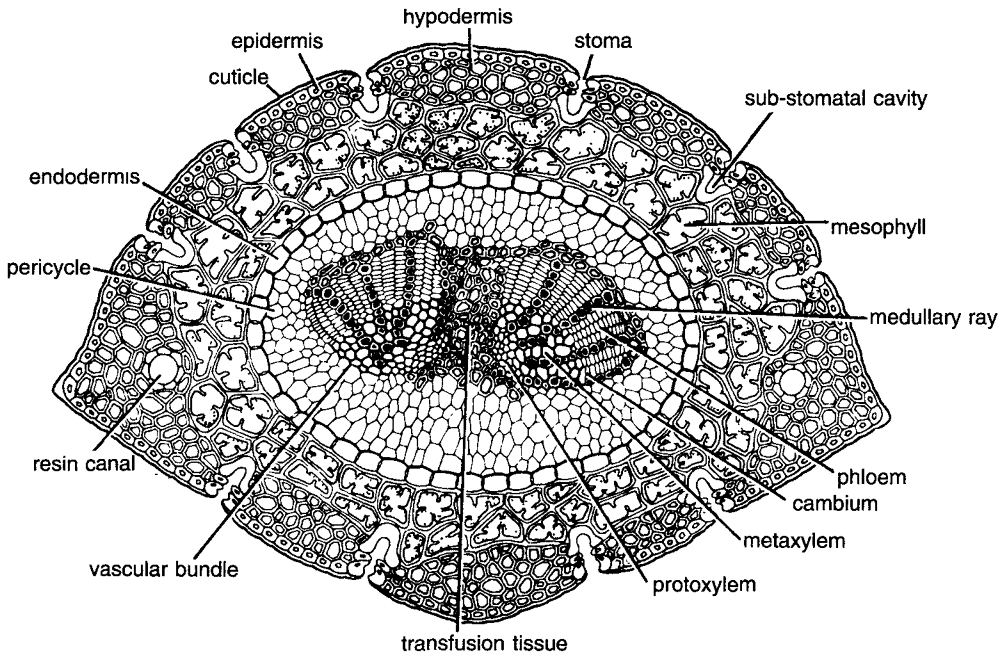


Fig. 13. ***Pinus.*** T.s. of needle (foliage leaf)-cellular details.

4. **Stomata** are sunken. These are present on all the faces of epidermis. The needle is thus said to the amphistomatic.
5. **Epidermis** is followed by hypodermis. It is few layered thick at the corners and 1-2 layered in other parts. Sub-stomatal chambers occur in this region. Cells are sclerenchymatous and fibrous.
6. **Mesophyll** lies below the hypodermis. It is made up of polygonal parenchymatous cells, densely filled with the chloroplasts. Numerous plate-like or peg-like infoldings project into the cell lumen (cavity) from the wall of the mesophyll cells.
7. **Resin canals** generally occur in the sclerotic hypodermis but also occur in the mesophyll tissue.
8. **Endodermis** is conspicuous. Cells are barrel-shaped and tangentially thickened. It is followed by a many layered, parenchymatous pericycle.
9. Generally two vascular bundles remain surrounded by this tissue. (In *P. strobus* there is only one vascular bundule).
10. **The vascular bundles** are separated from one another by a T-shaped thick walled transfusion tissue.
11. **Each vascular bundle** is conjoint, collateral and open. Protoxylem faces adaxial side. Phloem is located on the abaxial side.
12. **Xylem and phloem** groups are separated from one another by cambium at the base of the needle and by parenchymatous cells in the upper region.
13. **Secondary growth** is very little during which the medullary rays run between xylem and phloem.

Features of special interest. It shows the following xerophytic characters —

1. Narrow acicular form of the leaf.
2. Presence of thick cuticle.
3. Aphistomatic nature.
4. Sunken stomata.
5. Thick and sclerenchymatous hypodermis.
6. Infolded peg-like structures in mesophyll.
7. Presence of transfusion tissue.
8. Simple vascular system.

Exercise 12

***Object* : Study of male cone, microsporophylls and microsporangia.**

Work procedure

Dissect out the male cone, separate the microsporophylls, study the shape, size and microspores.

Comments

1. Male cones replace the dwarf shoots. Each male cone arises in the axile of a scale leaf. The main shoot, on which these are produced, continues to grow further.
2. Male cones are grouped in clusters on the shoots of the same year only.
3. Each male cone has single, centrally located cone axis around which many scaly microsporophylls are spirally arranged.
4. Each microsporophyll has an expanded triangular central part and stalk-like base. Terminal part projects into a tip.
5. Few lowermost sporophylls are sterile, and do not bear any male reproductive structures.
6. On the abaxial side, each microsporophyll bears two ovoid microsporangia or pollen sacs on its lateral sides.
7. Each microsporangium has its own wall which encloses many microspores
8. The young microspore is globular or spherical in shape and is uninucleate.
9. A mature microspore or pollen grain shows two wall layers- exine and intine, 2 prothallial cells and antheridial cell.
10. Pollen grain has a thick expanded exine in the form of wings on the sides, followed by a smooth intine.

Exercise 13

***Object* : Study of L.s. of male cone.**

Work procedure

Study a slide showing L.s. of male cone.

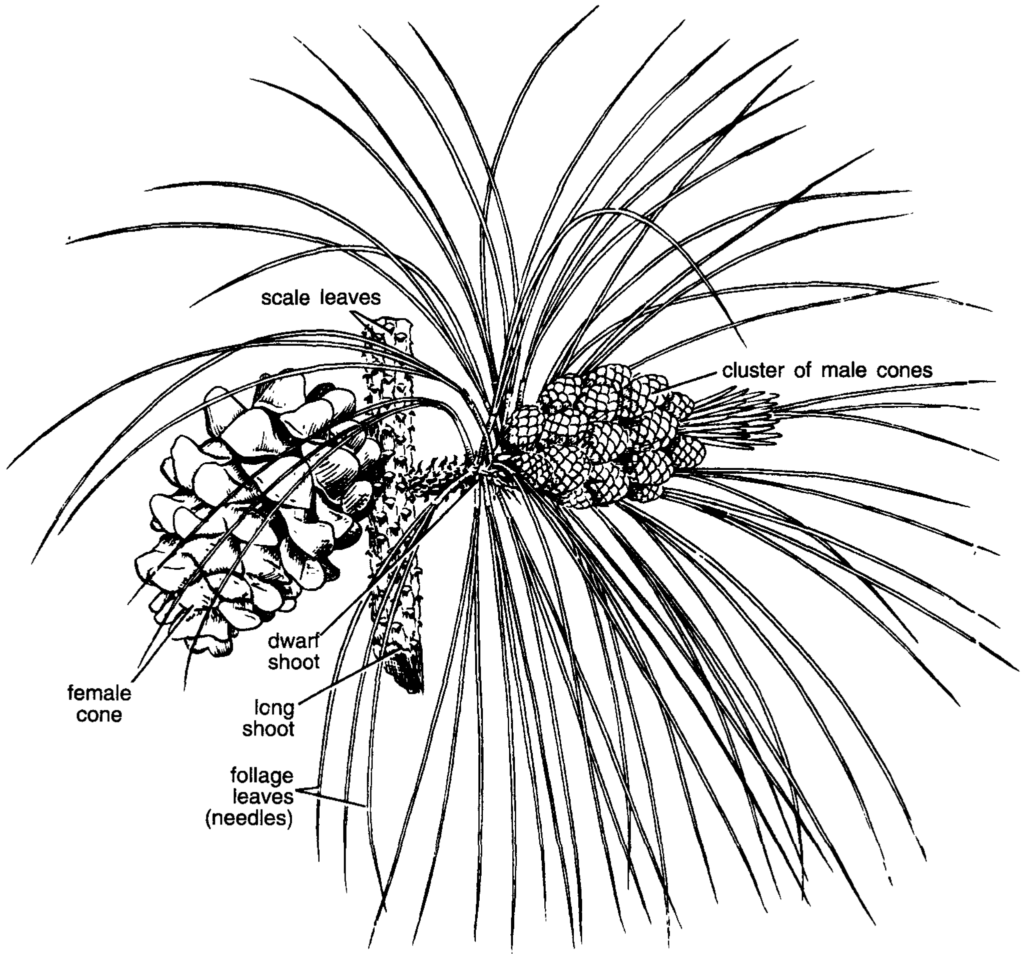


Fig. 14. *Pinus*. A twig with male and female cones.

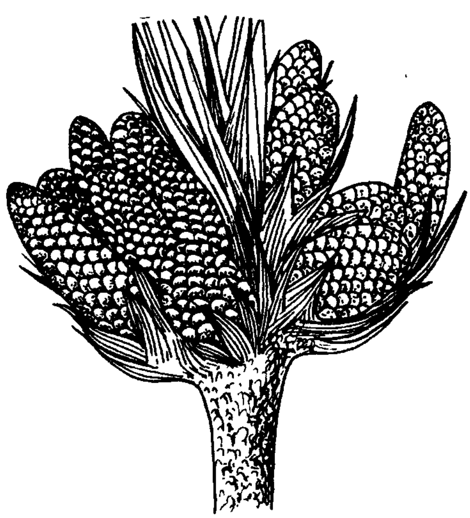


Fig. 15. *Pinus*. Male cones in cluster.

Comments

1. It shows a cone axis bearing microsporophylls.
2. The cone axis is centrally located.
3. Microsporophylls are spirally arranged. These are scaly, triangular and expanded.
4. It is attached to the cone axis by a stalk-like base.
5. The outer expanded part is sterile and is known as apophysis.
6. Microsporangia are present on the lower or abaxial surface.
7. Each microsporangium has a wall that encloses a cavity.
8. The wall consists of epidermis, wall layers and tapetum.
9. The cavity shows numerous microspores in various stages of development.

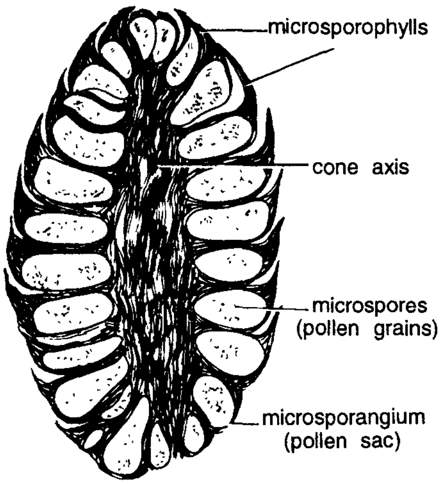


Fig. 16. ***Pinus.*** L.s. of male cone.

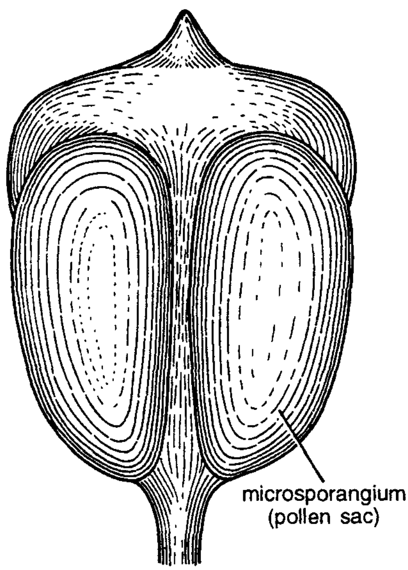


Fig. 17. ***Pinus.*** A microsporophyll with two microsporangia as seen from lower side.

Exercise 14

Object **: Study of morphology of the female cone.**

Work procedure

Study the external features of 1st, 2nd and 3rd year female cones. Note the position, arrangement and structure of sporophylls.

Comments

1. Female cones are larger than the male cones. They are borne at the apices of the young elongated shoots, replacing the shoot of unlimited growth (long shoots).
2. Single shoot may bear one to four female cones which are reddish-green in colour and mature in three years.
3. In the first year, cones are compact and sporophylls are closely arranged.
4. The second year cones are large in size and woody in nature but sporophylls are still compactly arranged.
5. In the third year, cone becomes loose. Sporophylls separate from one another due to elongation of the cone axis.
6. Each female cone consists of many sporophylls, arranged spirally around the cone axis.

Exercise 15

Object **: Study L.s. of female cone.**

Work procedure

Study a prepared slide of L.s. of female cone.

Comments

1. Female cone is made of centrally located cone axis and spirally arranged sporophylls.
2. Each sporophyll consists of two kinds of paired scales : (i) bract scale or cone scale and (ii) ovuliferous scale or seminiferous scale.
3. Many small and thin bract scales are arranged spirally around the cone axis. They are directly borne on the cone axis. Each of these is present on the abaxial (lower) side of the ovuliferous scale.
4. On the adaxial (upper) side of the bract scale, a thick, large, woody and triangular ovuliferous scale is present.
5. The ovuliferous scales in the middle part of the cone are the largest and get gradually smaller towards its base and apex.
6. Ovuliferous scale and bract scale are fused for a little distance near the cone axis while free at a distance away from it.

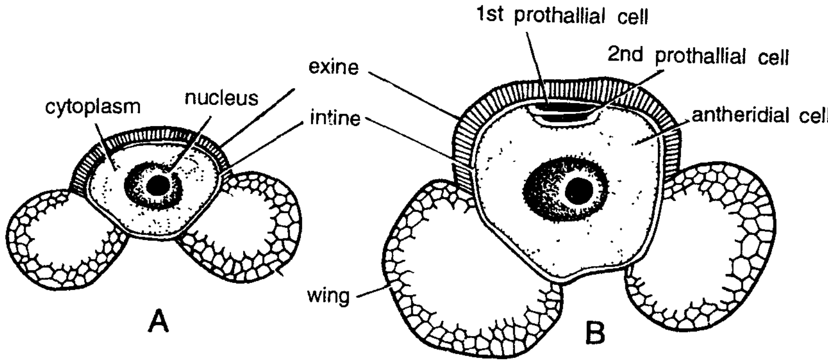


Fig. 18. ***Pinus.*** Microspores (pollen grains), **A.** Young. **B.** Old.

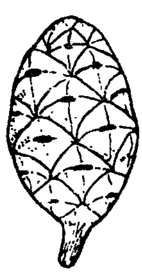


Fig. 19. ***Pinus.*** 1st year female cone.

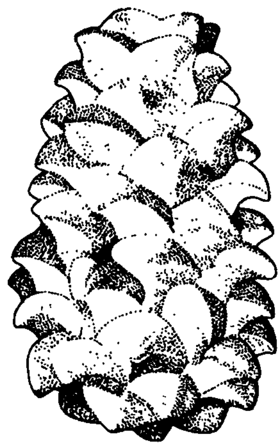


Fig. 20. ***Pinus.*** 2nd year female cone.

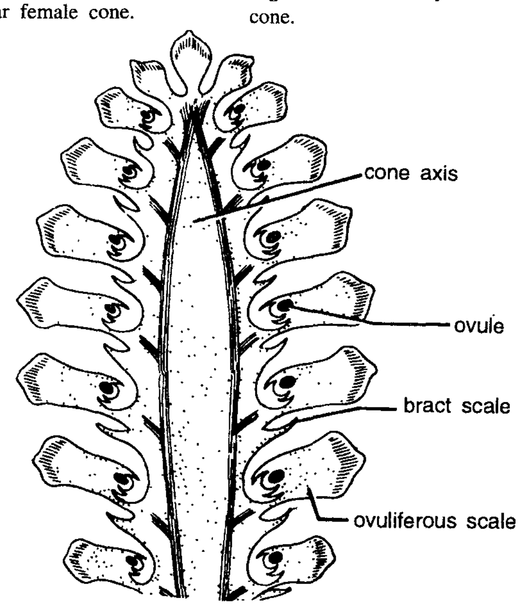


Fig. 22. ***Pinus.*** L.s. of female cone.

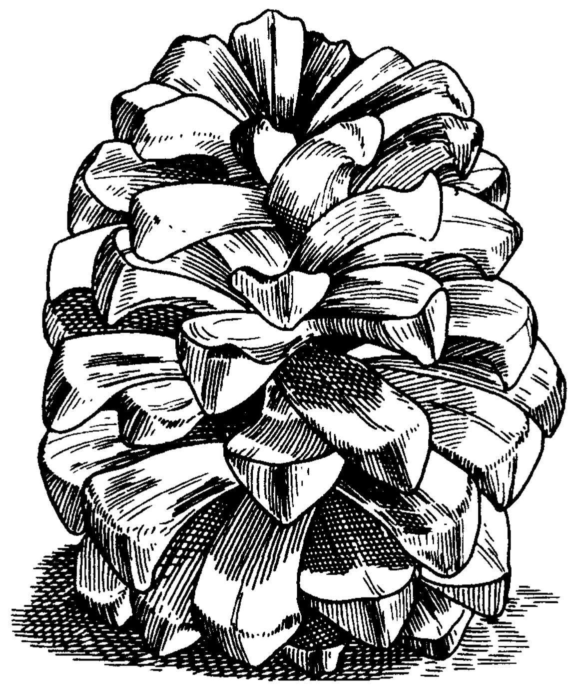


Fig. 21. ***Pinus.*** 3rd year female cone.

7. Ovuliferous scale is shortly stalked and rest of the part is expanded.
8. At the base of this expanded, triangular part, two naked and sessile ovules are present. These are situated on the adaxial, (upper) surface of the ovuliferous scale, at its base, with their micropyles directed towards cone axis.
9. The terminal part of the ovuliferous scale is broad and sterile and is known as apophysis.

Exercise 16

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

Work procedure

Study a prepared slide of L.s. of ovule. Note integuments, nucellus, female gametophyte and archegonia.

Comments

1. Ovule is elongated in shape.
2. It is unitegmic and the integument is three layered. The outermost layer is thin. The middle layer is stony and prominent. The innermost layer is fleshy and well developed.
3. Nucellus is fused with inner layer of the integument, except at its tip where it forms an elongated and slender micropyle, directed towards the cone axis.
4. In the nucellar region lies a small cavity just opposite the micropyle. It is known as pollen chamber.
5. Female gametophyte (endosperm) is differentiated from nucellus. About 2-5 archegonia are situated in this region at the micropylar end near the base of the archegonial chamber

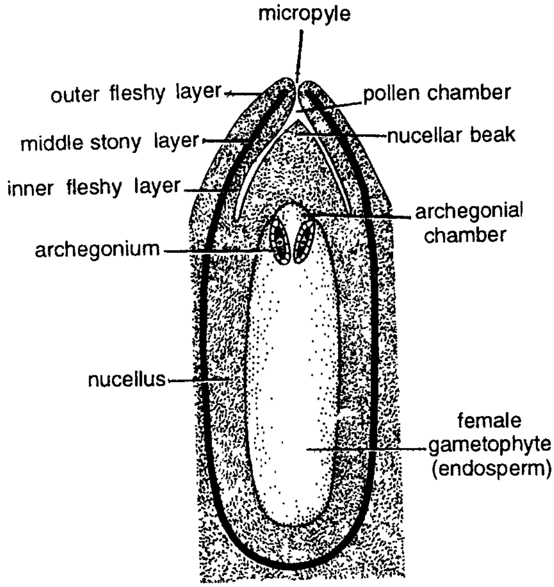


Fig. 23. ***Pinus.*** L.s. of ovule.

Exercise 17

Object **: Study of seed.**

Work procedure

Study the position and arrangement of seeds on ovuliferous scale, also study a prepared slide of L.s. of seed

Comments

1. Fertilized ovules get transformed into seeds which are situated on the adaxial side of the ovuliferous scale at its base near the cone axis.
2. Seeds are small, elongated and winged. The wing is a thin layer of tissue which splits off from the adaxial face of the ovuliferous scale. (Seed can be best studied by cutting longitudinal section of the seed of *P. gerardiana*; vern. chilgoza).

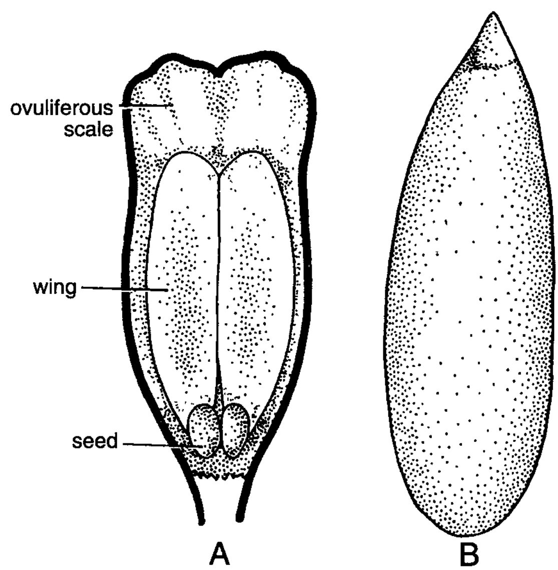


Fig. 24. ***Pinus.*** **A.** Ovuliferous scale bearing two winged seeds. **B.** seed.

3. The seed is covered with red and brown testa.
4. Inner fleshy layer of the integument still persists. It is membranous, thin and papery, termed as tegmen.
5. The nucellus is present as a thin layer and forms a nucellar cap at the micropylar end.
6. The larger part of the seed consists of oily endosperm.

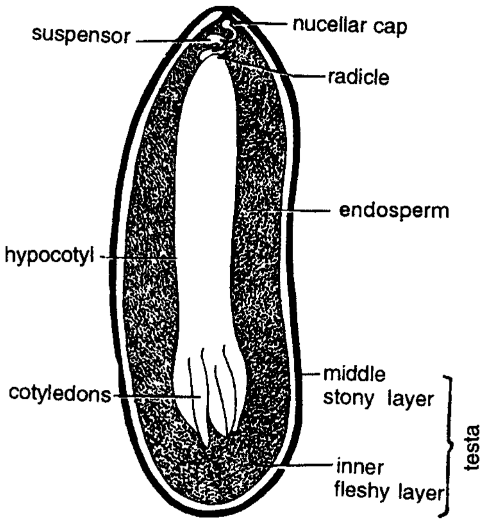


Fig. 25. ***Pinus.*** L.s. of seed.

7. The suspensor is long and becomes coiled. Embryo is differentiated into radicle, plumule and cotyledons (3-8 in number).
8. In between the radicle and plumule, is present a well developed hypocotyl.

Exercise 18

Object **: Study of seedling.**

Work procedure

Study a newly germinated seed. Observe various organs.

Comments

1. Seedling shows three parts — (i) roots (ii) hypocotyl and (iii) leaves.
2. The **roots** are well branched and arise from the radicle.
3. **Hypocotyl** gives rise to unbranched, slender and thin primary shoot.
4. **Leaves** are green and needle-like which are borne in whorls on the primary shoot. These are cotyledonary leaves.
5. **The primary leaves** or first spur shoots arise in the axils of some of these juvenile (cotyledonary) leaves and are borne in spiral series on the primary shoot.

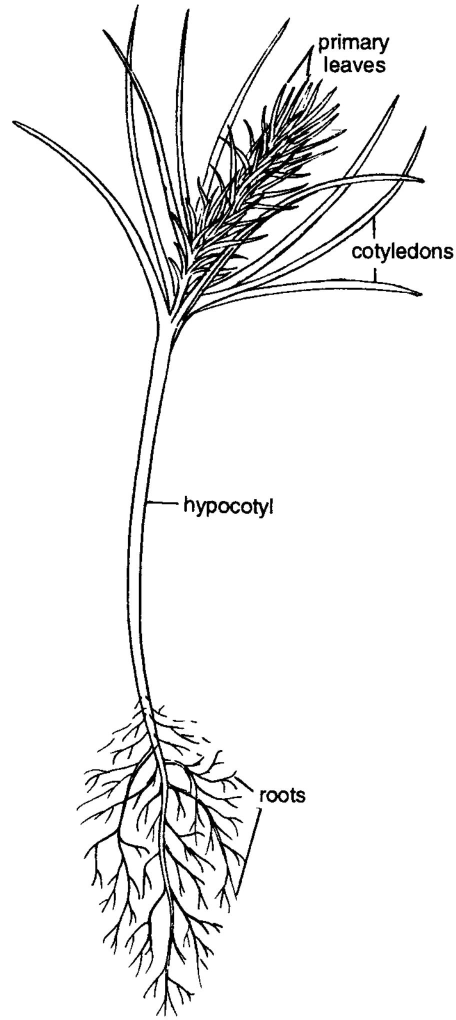


Fig. 26. ***Pinus.*** Older seedling to show cotyledons and spirally arranged primary leaves.

Identification

Division—**Gymnosperms**. (1) Absence of vessels, (2) Ovules naked, (3) Seeds attached to woody scales, (4) Scales generally form a cone.

Class—**Coniferopsida**. (1) Leaves needle shaped, (2) Wood pycnoxylic (compact), (3) Presence of resin canals, (4) Compact male and female cones, (5) Non-flagellate male gametes, (6) Seeds bilaterally symmetrical.

Order—**Coniferales**.

Family— **Pinaceace**. (1) Resinous wood, (2) Plants monoecious, (3) Sporophylls spirally arranged, (4) Microsporophylls with two microsporangia, (5) Pollen grains winged, (6) Female cone woody, (7) Polyembryony present, (8) Seed dry and winged.

*Genus—**Pinus***. (1) Plants sporophytic and monoecious. Male and female reproductive organs in cones, (2) Branches dimorphic, (3) Long shoots with secondary xylem, annual rings are formed, wood pycnoxylic and resinous, (4) Dwarf shoots with a little secondary growth, (5) Leaves are of two types, (6) Scale leaves brown and membranous, (7) Foliage leaves are acicular, xerophytic, mesophyll cells with peg-like ingrowths, 2 resin canals and T-shaped transfusion tissue, (8) Male cones borne laterally, in clusters, microsporophyll bears two microsporangia on abaxial side, (9) Pollen grains winged, (10) Female cones borne single and terminal, (11) Bract scales and ovuliferous scales spirally arranged, (12) Two naked ovules on the adaxial side of the ovuliferous scale, (13) Seeds dry and winged.

Hints for Collection

In India *Pinus* is represented by five species which grow wild in north-east and north-west Himalayas. The species are *P. gerardiana* (Chilgoza in Hindi), *P. roxburghii (=P. longifolia), P. wallichiana* (*P. excelsa*), *P. insularis* (= *P. khasya*) and *P. armandi.* In plains it is cultivated for its ornamental value.

Ephedra
(Jointed Fir)

Classification

Division	–	**Gymnosperms**
Class	–	**Gnetopsida**
Order	–	**Gnetales**
Family	–	**Ephedraceae**
Genus	–	***Ephedra***

Exercise 1

Object **: Study of external morphology.**

Work procedure

Study the external features of the plant, note the jointed nature of stem, scale leaves and underground tap root, observe the positions of male and female strobili.

Comments

1. Plants are small, bushy, trailing or climbing shrubs attaining a height of not more than 2 meters. However, *E. antisiphilitica* is a small tree, reaching a height of 3-5 meters.
2. The plant body is branched and posseses only minute leaves at the nodes. It therefore, resembles superficially with the species of *Psilotum* and *Equisetum.*
3. It is differentiated into stem, leaves and underground roots.
4. The stem remains anchored by a deep tap root and many adventitious roots.
5. The stem is delicate, slender and green when young. It is ribbed irregularly and is differentiated into short nodes and long internodes.
6. Two or three branches arranged in whorls arise from the nodes in the axils of leaves. The branches are shed off during dry season.
7 Older part of the stem may bear many branches. It becomes hard and woody due to secondary growth.
8. Leaves are borne in a whorl of 2-4 at each node.

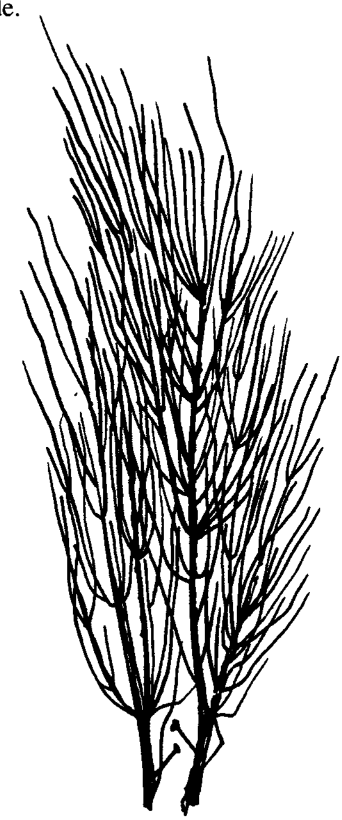
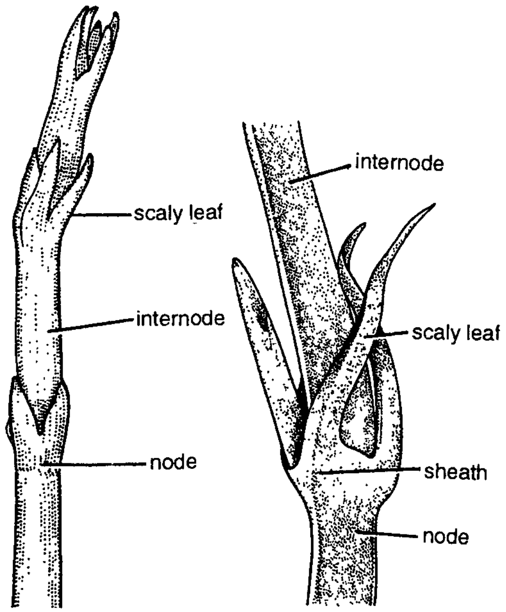


Fig. 1. *Ephedra*. Plant showing shrubby habit.



A B

Fig. 2. ***Ephedra.*** A part of stem showing **A.** Nodes and internodes. **B.** A node with scaly leaves.

9. Leaves that are small and scale-like are connate at the base and thus form a sheath around the node.
10. Each scale leaf is traversed by two parallel and unbranched veins.
11. Foliage leaves are completely absent.
12. The male and female reproductive organs are borne in small strobill.
13. The plants are mostly dioecious and bear only one type of reproductive organs. They may also be monoecious when they bear both kinds of strobilii.

Exercise 2

Object **: Study of T.s. of young stem.**

Work procedure

Cut a T.s. of the younger part of the stem, stain in safranin-fast green combination, mount in glycerine and study.

Comments

1. **The outline** of the section shows ribbed nature of the stem.

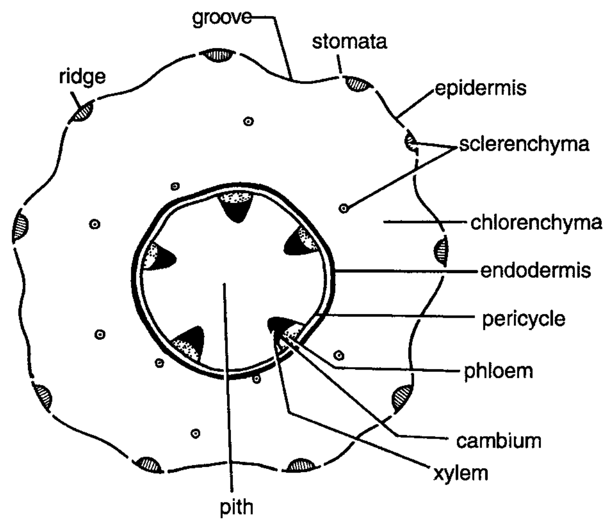


Fig. 3. ***Ephedra.*** T.s. of young stem (diagrammatic).

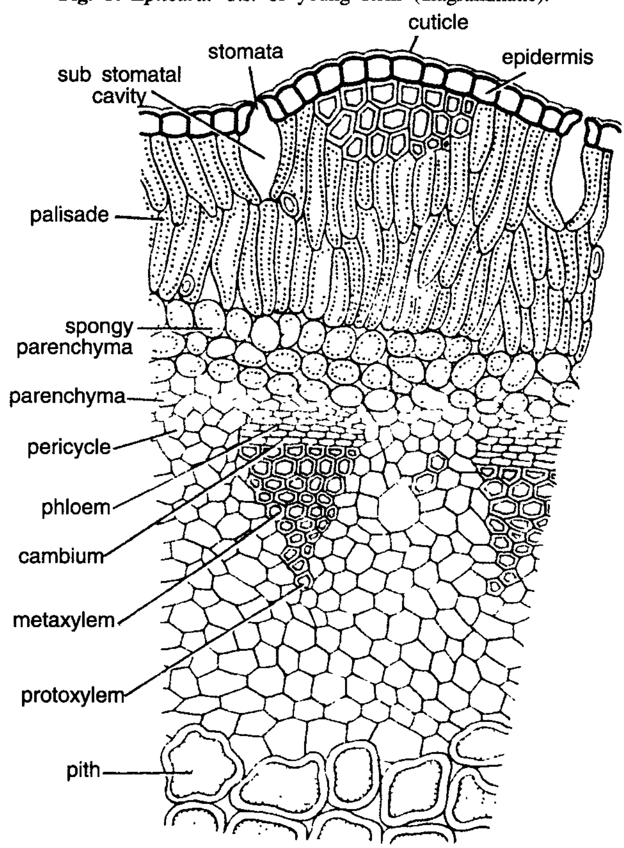


Fig. 4. ***Ephedra.*** T.s. of young stem (part cellular).

2. The tissues are differentiated into epidermis, cortex and stem.

3. **Epidermis** is the outermost layer. It is very thick and heavily cuticularized.
4. **The stomata** are sunken. These interrupt epidermis frequently and occupy a position just on the slopes of the ridges.
5. **Hypodermis** is sclerenchymatous and occur in small groups below the ridges.
6. **Cortex.** Rest of the cortical tissue is cholrenchymatous. The cells are often radially elongated and contain abundant chloroplasts. Large intercellular spaces are present between these cells.
7. A few patches of sclerenchyma occur dispersed in the cortex (specially in young axis rendering hardness and resistance).
8. **Stele** is ectophloic siphonostele. It is composed of many vascular bundles, their number being variable.
9. **Endodermis** is single layered and is followed by a pericycle.
10. **A few vascular bundles** are arranged in a ring. Each is conjoint, collateral, endarch and open.
11. **External phloem** group is separated from internal xylem group by a narrow layer of cambium.
12. **Pith** is parenchymatous and occurs in the central region.
13. **Nodal diaphragm.** The characteristic anatomical feature is the presence of diaphragm-like plate of cells at the base of each internode or at the node (nodal diaphragm). This helps the plant to shed off the branches at the nodes.

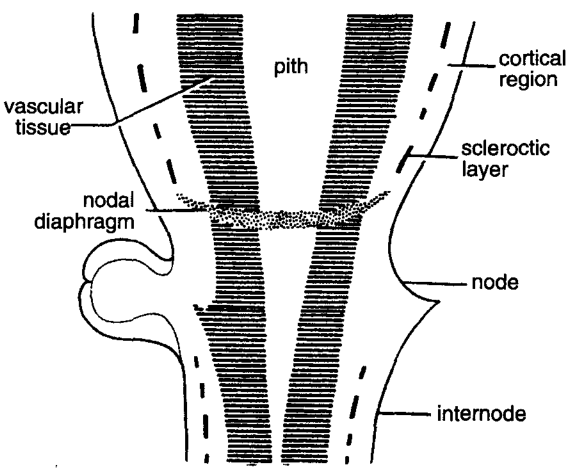


Fig. 5. ***Ephedra.*** L.s. nodal region (diagrammatic).

Exercise 3

Object **: Study of T.s. of stem showing secondary growth.**

Work procedure

Cut a T.s. of the older part of the stem, stain in safranin-fast green combination, mount in glycerine and study.

Comments

1. **The section** shows epidermis, cortex and primary and secondary vascular tissues.
2. **The epidermis** and cortex remain unchanged. However, after (3-4 Years) of growth, cork develops just outside the phloem and outer tissues (epidermis, cortex, etc.) are, therefore, cast off.
3. **Sclerotic cells** (stone cells) develop just above the zone of secondary tissue.
4. **Primary phloem** occurs as obliterated patches.
5. **Secondary phloem** forms a zone below. Phloem is composed of sieve tubes and phloem parenchyma.
6. **Annual rings** are distinct comprising autumn and spring wood each. These are formed in the secondary xylem (wood).
7. **The secondary** xylem shows a thin walled spring wood and thick walled autumn wood, successively formed in alternating zones.
8. **Antumn** wood is made of smaller cells, while those of spring wood are bigger in size.
9. **The tracheidal cells** of the secondary wood are associated with broad vessels. Though absence of vessels is characteristics of the Gymnosperm, *Ephedra*, (i.e. order Gnetlaes) itself is an exception.
10. **Vessels** are most abundant in the spring wood and a few or none at all in the autumn wood. Spring wood is often ring porous.
11. **Tracheids and vessels** have uniseriatey or irregularly distributed bordered pits. Protoxylem elements of primary xylem show spiral, annular or reticulate tracheids.

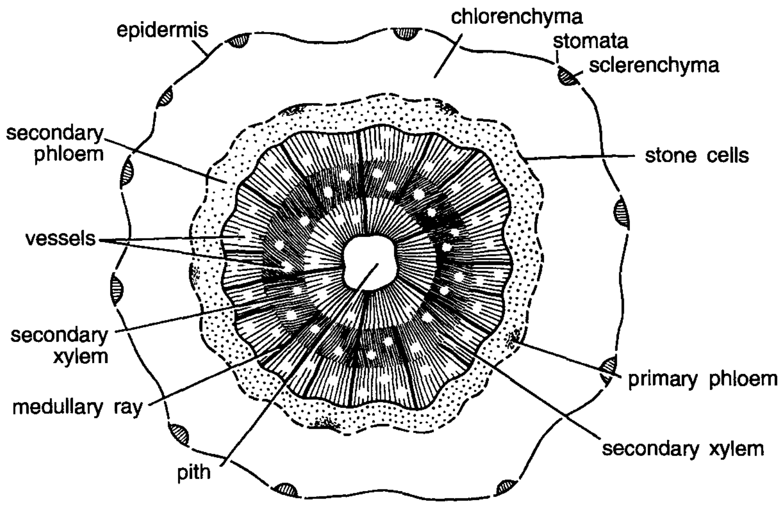


Fig. 6. ***Ephedra.*** T.s. of old stem (diagrammatic).

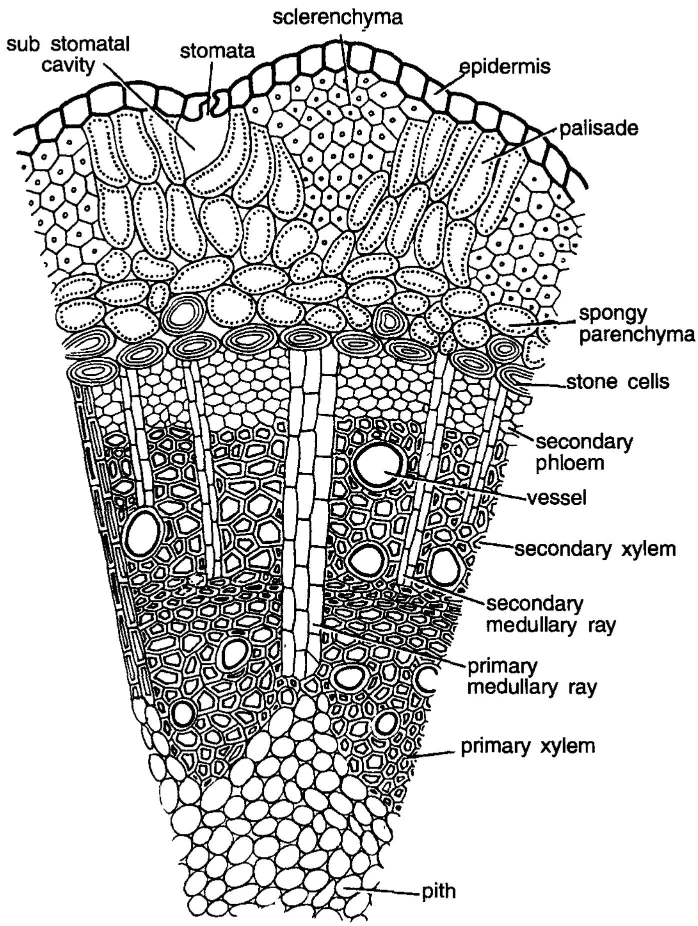


Fig. 7. ***Ephedra.*** T.s. of old stem (a part cellular).

12. **Medullary rays** traverse the wood. Primary medullary rays run from primary xylem to primary phloem while secondary medullary rays run from secondary xylem to secondary phloem.
13. **Medullary rays** are uniseriate in the young stem but are very broad and long (multiseriate) in the old stem.
14. **Primary** xylem groups are present at the end of the secondary wood near the pith. These are endarch.
15. **Pith** is large and parenchymatous. It occupies the centre.

Features of special interest. It shows the following xerophytic characters.

1. Thickly cuticularized epidermis.
2. Sunken stomata.
3. Palisade and spongy parenchyma in the cortex.
4. Patches of sclerenchyma.
5. Shedding of branches.
6. Presence of nodal diaphragm.
7. Vessel in the secondary wood.

Exercise 4

***Object* : Study of R. L.s. of wood.**

Work procedure

Cut a R.L.s. of wood along any one of the radii. Stain in safranin-fast green combination, mount in glycerine and study.

Comments

1. It shows the presence of secondary xylem and medullary rays.
2. Secondary xylem consists of tracheids with bordered pits on their radial walls.
3. The bordered pits are circular or slightly elliptical. These may form reticulations (mostly due to the dissolution of walls of cavities of the pits) and such perforations, being characteristic of *Ephedra*, are known as Ephedroid perforations. Bordered pits are either scattered or arranged in 2 or 3 tight rows. (They are never polygonal due to mutual compression).
4. Special cellulose thickening - Bars of Sanio are also present below the pits.
5. A few vessels present show bordered pits which are scattered or may remain arranged in 2 or 3 rows. The apertures of the bordered pits are commonly horizontally oriented. End walls are also perforated.
6. Medulllary rsays are uniseriate or multiseriate. These run horizontally. In this plane medullary rays are cut lengthwise and their length and height can be observed.
7. Medullary rays may range up to 40-50 cells in height.
8. Each medullary ray in the region of secondary xylem is composed of ray cells and ray tracheids dispersed regularly.

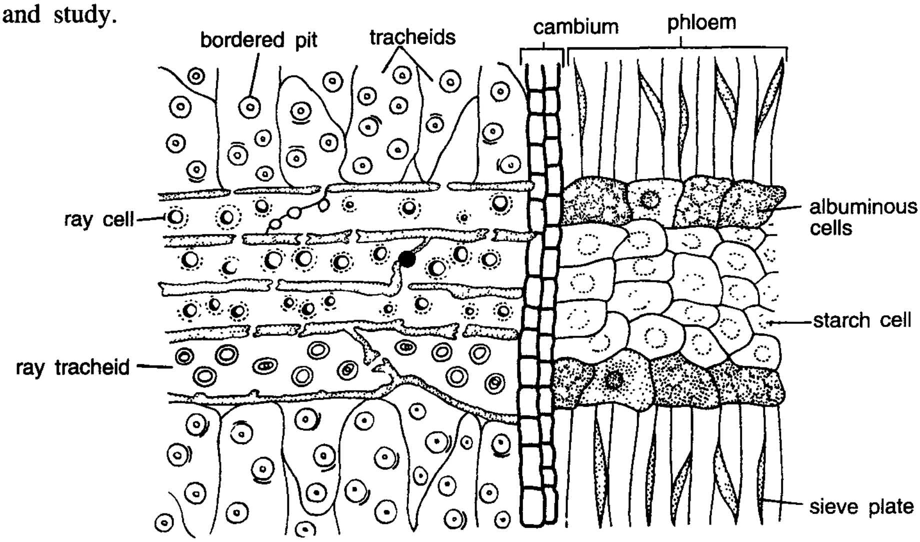


Fig. 8. ***Ephedra.*** R.L.s. of wood.

9. Ray cells are thick walled as well as thin walled. These occur in the same medullary ray. Their tangential walls possess bordered pits or slit-like openings.
10. Ray tracheids are thick walled. Their radial and tangential walls are pitted, pits being bordered.
11. In the region of phloem, medullary ray is made of starch cells surrounded by albuminous cells on both sides.

Exercise 5

Object : **Study of T.L.s. wood.**

Work procedure

Cut a T.L.s. of wood by passing the razor along any one of the tangents, stain in safranin-fast green combination mount in glycerine and study.

Comments

1. In this plane tracheids, vessels and medullary rays are cut transversely.
2. Bordered pits are seen in surface view.
3. The bordered pits show usual over-arching dome-shaped structure and a small disc-torus.
4. Medullary rays are transversely cut and as such their height and breadth can be determined.

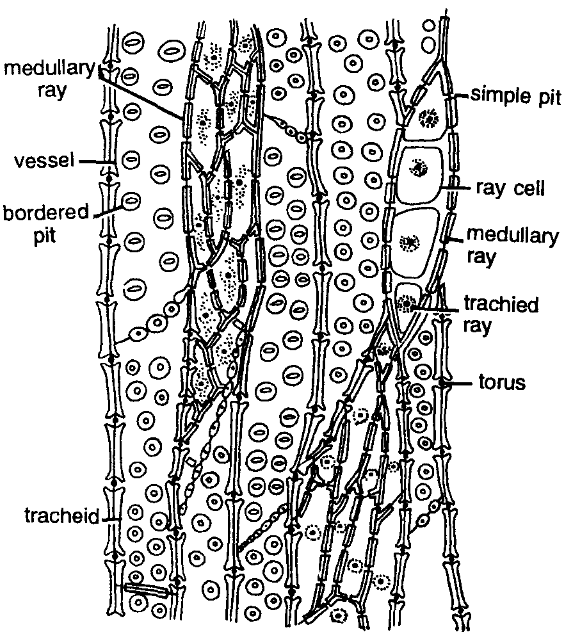


Fig. 9. ***Ephedra.*** T.L.s of wood.

5. Rays are elongate and many tangential walls show simple slit-like pits.

Exercise 6

Object : **Study of male strobilus.**

Work procedure

Study the position, structure and organization of male strobilus. This can be done by studying external morphology of the strobilus and a slide of its longitudinal section.

Reproductive parts are borne in strobili. One of the following conditions may be found :

(i) Usually male and female strobili are different; in such a case, the strobilus may be termed as monosporangiate. These strobili may be borne on two different plants (dioecious sps.).

(ii) Sometimes one plant may bear both the strobili (monoecious sps.).

(iii) A few plants, sometimes, bear both the reproductive parts in one strobilus only (bisporarangiate strobilus), *e.g. E. foliata* and *E. intermedia.* In such cases, male flowers are situated below the female flowers which occur at the higher level in the same strobilus.

Comments

1. The strobilus resembles inflorescence, spike.
2. Each stobilus consists of an axis which bears decussately arranged sterile scales and stamens.
3. Male spike (male strobilus or staminate strobilus) arises in the axil of scale leaf.
4. Each spike is generally round in shape but may be ovoid or spherical.
5. A spike has a short axis with many scaly bracts. The bracts are arranged in decussate pairs. The number of pairs varies from 2-12.
6. In the axil of each bract arises a single male flower.

Exercise 7

Object : **Study of male flower.**

Work procedure

Place a male strobilus under the dissecting microscope, separate the sterile scale to isolate a

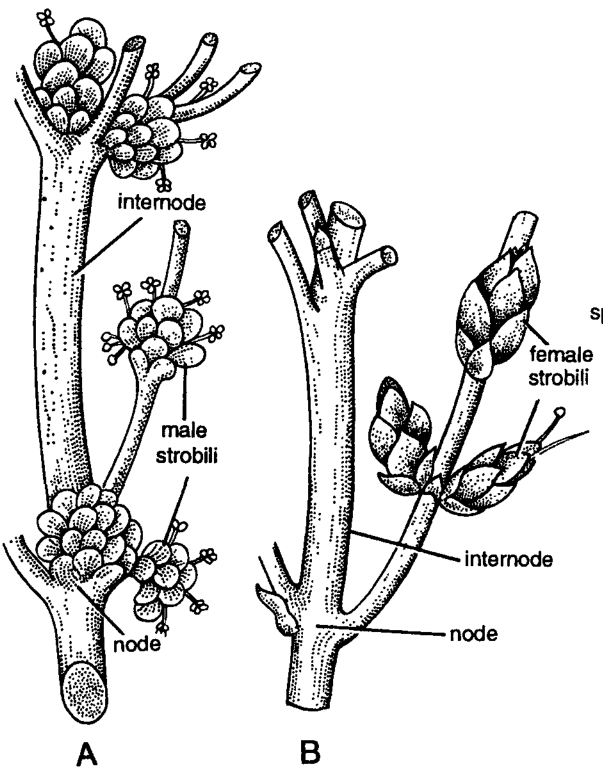


Fig. 10. ***Ephedra.*** Reproductive structures. **A.** A branch with male strobili. **B.** A branch with female strobili.

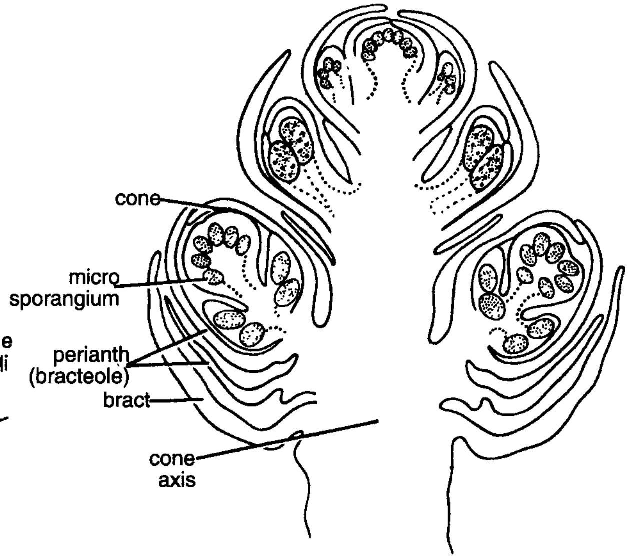


Fig. 12. ***Ephedra.*** L.s. of male strobilus.

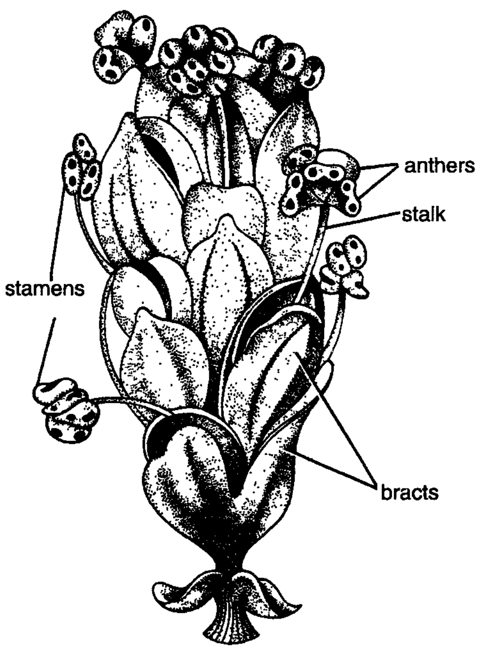


Fig. 11. ***Ephedra.*** A male strobilus.

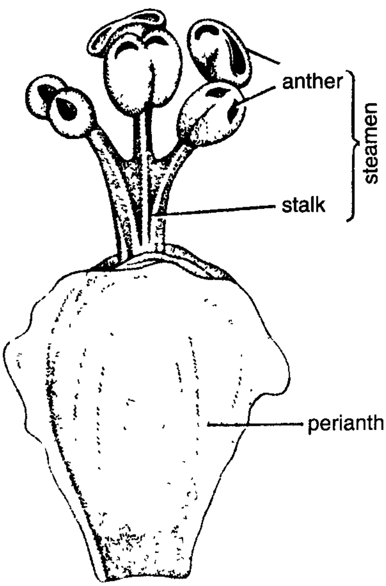


Fig. 13. ***Ephedra.*** A male flower

single male flower, stain in safranin, mount in glycerine and study. Also study slide of L.s. of a stamen.

Comments

1. **A male flower** has a perianth of bract scale which encloses a stamen.

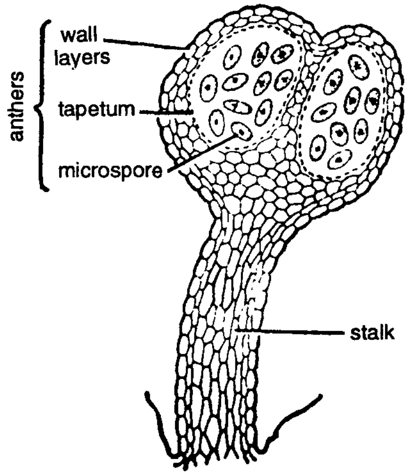


Fig. 14. ***Ephedra.*** L.s. of stamen.

2. **Stamen** consists of a stalk (variously termed as column or antherophore). It bears 2-5 microsporangia or anthers at its tip.
3. **Each microsporangium** is a bilocular structure. It has two wall layers and a prominent tapetal layer which encloses pollen grains or microspores.
4. **Each microspore** is elliptical and has an outer thick and ribbed exine and a thin intine.
5. A microsporangium opens by apical part (apical dehiscence).

Exercise 8

Object **: Study of female strobilus.**

Work procedure

Study the position, structure and organization of the female strobilus. This is done by studying the external features and a slide of its longitudinal section.

Comments

1. The strobilus resembles spike inflorescence.
2. Each strobilus consists of an axis bearing decussately arranged sterile bracts (scales) and ovules.
3. The female or ovulate strobili arise in the axil of scale leaves. The female strobilus is also sessile and not so richly branched as the male.

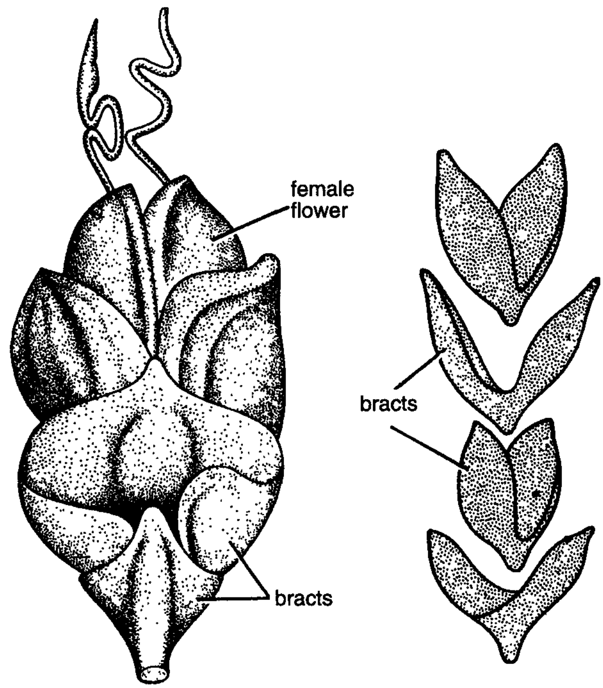


Fig. 15. ***Ephedra.*** Female strobilus. **A.** Female strobilus as it appears on plant. **B.** Bracts separated to show arrangement.

4. The apex of ovulate strobilus is mostly acute.
5. The spike has a short axis on which about 4-7 pairs of bracts are arranged decussately.
6. Lower most 1 or 2 pairs are sterile while terminal pairs bear short stalked ovules. The bracts are generally dry, winged and may be variously coloured.
7. Each bract mostly encloses two ovules out of which one may be abortive.

Exercise 9

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

Work procedure

Study a prepared slide of L.s. of ovule.

Comments

1. **Ovule** is covered by two integuments.
2. **Outer integument** (involucre or perianth) is a cup-like structure, attached at the base of the ovule and free above.
3. **Inner integument** is delicate, composed of two segments. It prolongs into a tubular process and

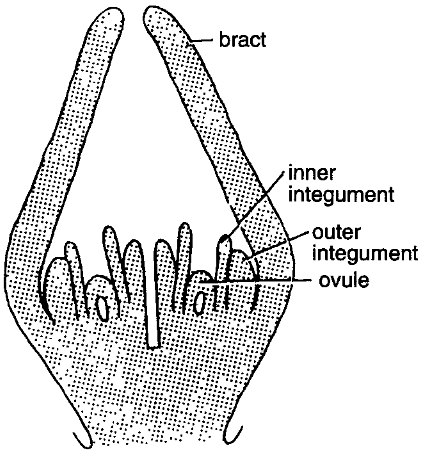


Fig. 16. ***Ephedra.*** L.s. of female flower.

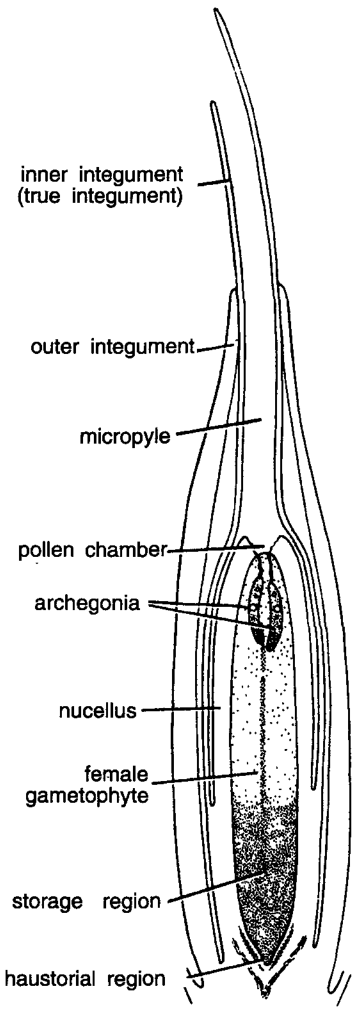


Fig. 17. ***Ephedra.*** L.s. of ovule

comes out beyond the bracts and involucre at the time of pollination.

4. **Micropyle** is an opening in between the integuments, in the upper region of the ovule.
5. **Nucellus** lies below the integuments. A small pollen chamber is present just below the micropyle in the tissue of nucellus.
6. **Female gametophyte** is a tissue situated below the pollen chamber. Two archegonia are present, just below the pollen chamber, in the female gametophyte.
7. **Haustorial region** lies opposite the micropylar end. It is occupied by tissue filled with stored food material. It also gives out haustorial processes for the absorption of food and is known as haustorial region.

Exercise 10

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

Work procedure

Study a double stained prepared slide of L.s. of seed. (A seed is formed as a result of fertilization).

Comments

1. **Outer integument encloses** entire seed. It is thick walled.

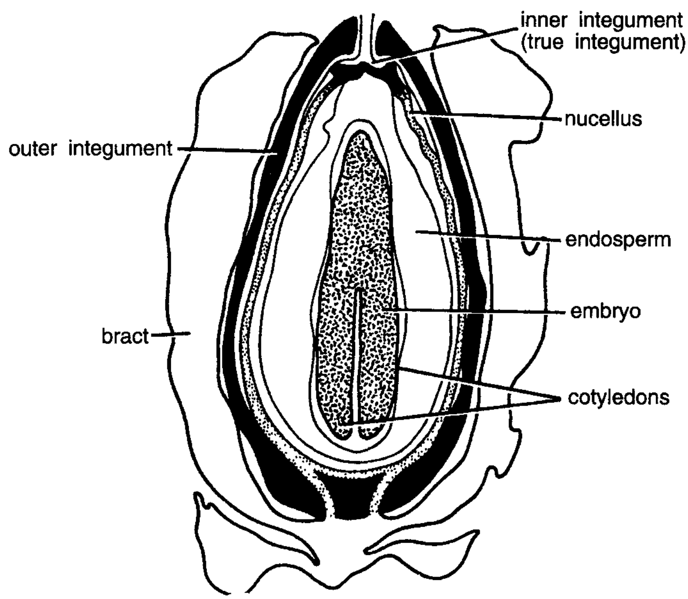


Fig. 18. ***Ephedra.*** L.s. of seed.

2. **The inner integument** ('true integument') persists at the micropylar end only.
3. **Nucellus** forms shrivelled layer in the form of a disorganized sheath of cells. It is located inside the inner integument.
4. **Female gametophyte** (endosperm) surrounds a big embryo which has two large cotyledons.
5. **Bracts** adjacent to strobilus are fleshy and thick in a completely mature seed. These form an additional envelope.

Identification

Division—**Gymnosperms**. (1) Ovules naked, (2) Seed attached to a scales, (3) Scales form a strobilus.

Class—**Gnetopsida**. (1) Wood with vessels, (2) Flowers in compound strobili or 'inflorescence', unisexual, usually dioecious, (3) Ovules surrounded by several envelops.

Order—**Gnetales**. (1) Plants woody trees, shrubs or lianes, (2) Leaves opposite or whorled, simple.

Family—**Ephedraceae**. (1) Plants either shrubs or woody climbers, (2) Leaves scaly, foliage leaves absent, (3) Nodal diaphragm present, (4) Stamens enclosed by bract, (5) Seeds covered with fleshy bracts.

Genus—***Ephedra***. Single genus.

Hints for Collection

Six species of *Ephedra* viz. *E. pachyclada, E. intermedia* var. *tibetica, E. saxatilis* var. *sikkimensis, E. gerardiana E. nebrodensis* var. *procera* and *E. regeliana* grow wild in the north west Himalayan region and are all shrubby. *E. foliata* var. *ciliata* grows widely as a scandent shrub, climbing over small trees in the southern part of the Punjab and Rajasthan. It is usually cultivated in gardens and in other places.

Palaeobotany

10

Chapter

Preamble

Palaeobotany is the study of plant life of geological past (i.e. the study of plant fossils). The earth has undergone large climatic changes over billion years of its geologic past. During this period of drastic weather changes, most of the plants died and their remains were decomposed. However, a few plants which escaped decay and decomposition, remained preserved. These preserved plant specimen are called plant fossils. Generally, only certain plants made of harder tissues are preserved while those made of soft tissues get decomposed. The preserved plant parts are discovered independently at different times and places, hence each part is given a status of genus, representing form genus or artificial genus. Later, all these related parts are placed together and a full plant is visualized and reconstructed.

The age of the plant can be determined by using radioactive elements like uranium-238, uranium-235, thorium-232, potassium-40 and rubidium-87. Carbon-14 has also been used for time-measuring technique.

The major stratigraphic and time divisions used are given below.

The process of fossil formation is called fossilization. This had begun ever since deposition of sedimentary rocks began. According to the nature of fossilization, following fossil types have been recognized.

1. **Petrifaction fossil.** These fossils have well preserved external form and internal structure. This is due to replacement of plant material by some 20 minerals.
2. **Cast or incrustation.** This type of fossils are formed when a plant part gets covered by sand or mud. The cast or coat is hard and, therefore, used in the study of external morphology.
3. **Impression.** These are impressions of plant or plant parts when these fall on partially hard clay. Such types of fossils show clear venation patterns.
4. **Compression.** These are formed as a result of complete burial and the constant pressure of continuing sedimentation above it. The organs generally become flat.
5. **Coal, Amber, Graphite, Diatomaceous Earth,** etc. These are also types of fossils.

The fossils are of great importance in the study of evolution. These make it possible to know the time and the type of flora and fauna that flourished. It has, therefore, been possible to trace evolutionary series and reconstruct the changes which might have occurred during the past.

In this chapter, few fossils belonging to Pteridophytes and Gymnosperms are described.

Classification

1 Division. PTERIDOPHYTA

Sub-division	Order	Family	Example
1. Psilophytopsida	Psilophytales	Rhyniaceae	*Rhynia, Horneophyton*
2. Lycopsida	Lepidodendrales	Lepidodendraceae	*Lepidodendron, Lepidocarpon*
3. Sphenopsida	Calamitales	Calamitaceae	*Calamites*

II Division. GYMNOSPERMAE

Class	Order	Family	Example
1. Cycadopsida	Pteridospermales	Lyginopteridaceae	*Lyginopteris*
	Bennettitales	Williamosoniaceae	*Williamsonia*
		Cycadeoidaceae	*Cycadeoidea*

Distinguishing Characters of Taxa

1 DIVISION. **PTERIDOPHYTA**

(1) True roots generally present
(2) Plant body differentiated into stem, roots and leaves
(3) True vascular strand present

Sub-division. **Psilophytopsida**

(1) True roots absent
(2) Sporangia borne at the tips of erect branches either singly or in pairs

Order. **Psilophytales**

(1) Sporophyte dichotomously branched
(2) Sporangia generally borne singly

Family. **Rhyniaceae**

(1) Rhizoids unicellular, present on rhizomes
(2) Aerial portion leafless

Examples. ***Rhynia, Horneophyton***

	Geological periods			Age (in million years)
	Present Day			0
CENOZOIC	**Quaternary**	Post-Glacial Glacial		
	Tertiary	Pliocene Miocene Oligocene Eocene	Angiosperms dominant	1
MESOZOIC	**Cretaceous**	Upper Lower		
	Jurassic	Upper Middle Lower	Gymnosperms and Pteridophytes dominant	58
	Triassic	Upper Middle Lower		
PALAEOZOIC	**Permian**	Upper Lower		182
	Carboniferous	Pennsylvanian Mississipian	Pteridophytes dominate Gymnospserms	
	Devonian	Upper Middle Lower		255
	Silurian		Earliest records of land plants	313
	Ordovician		Algae	350
	Cambrian		Algae	430
	Precambrian	Upper Lower	Traces of algae	510

Sub-division. **Lycopsida**

(1) Leaves microphyllous
(2) Sporangia borne singly on adaxial face of the sporophyll or in its axil
(3) Sporophyll borne in strobili

Order. **Lepidodendrales**

(1) Plants tree-like
(2) Secondary tissues formed due to cambium
(3) Leaves microphyllous and ligulate
(4) Strobili heterosporous

Family. **Lepidodendraceae**

(1) Aerial portion freely branched
(2) Strobili at the tips of branches
(3) Trunk and branches with spirally arranged leaf scars

Examples. ***Lepidodendron, Lepidocarpon***

Sub-division. **Sphenopsida**

(1) Stem branched, articulated, ridged and furrowed with distinct nodes and internodes
(2) Leaves microphyllous, small, scaly and arranged in whorls at nodes

Order. **Calamitales**

(1) Tree-like sporophytes with considerable secondary thickening of stem and branches

Family. **Calamitaceae**

(1) Stem branched, branches in whorls at nodes
(2) Stem shows endarch siphonostele

Example. ***Calamites***

II DIVISION. **GYMNOSPERMAE**

(1) Ovules naked
(2) Seeds attached to a scale
(3) Scales forming a strobilus

Class. **Cycadopsida**

(1) Wood manoxylic
(2) Large frond-like leaves
(3) Seeds with radial symmetry

Order 1. **Pteridospermales**

(1) Leaves large, frond-like, pinnately compound
(2) Large leaf traces with one or more strand
(3) Spores formed in sporangia, aggregated in synangia

Family 1. **Lyginopteridaceae**

(1) Stem monostelic
(2) Petioles with single vascular strand
(3) Seeds small

Examples. ***Heterangium, Lyginopteris***

Family 2. **Glossopteridaceae**

(1) Leaves with a strong midrib
(2) Stelar structure unusual, showing several plates of vascular tissues
(3) Reproductive structure cupulate and bisexual

Example. ***Glossopteris***

Order 2. **Bennettitales**

(1) Tree trunk covered by a mantle of persistent leaf-bases
(2) Microsporophylls in groups, at the tip, leaves frond-like
(3) Megasporophylls in cone-like organization

Family 1. **Williamsoniaceae**

(1) Stem delicate, branched
(2) Inflorescence stalked or sessile, not sunk in the scales of persistent leaf bases

Example. ***Williamsonia***

Family 2. **Cycadeoideaceae**

(1) Trunk colummar
(2) Trunk covered by mantle of leaf bases
(3) 'Flowers' sunk in the distal part of the trunk

Example. ***Cycadeoidea***

PTERIDOPHYTES

Rhynia

Classification

Division	—	**Pteridophyta**
Sub-division	—	**Psilophytopsida**
Order	—	**Psilophytales**
Family	—	**Rhyniaceae**
Genus	—	***Rhynia***

Exercise 1

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

Work procedure

Study the reconstruction of plant. Observe the differentiation of plant body into rhizoids, rhizome and leaf-less aerial branches.

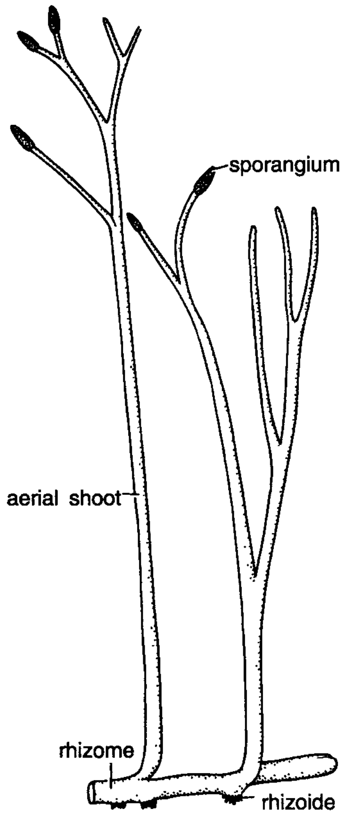


Fig. 1. ***Rhynia.*** External features of the plant.

Comments

1. *Rhynia* is a fossil member (not found living in the present age), discovered from Rhynichert Beds (or upper Devonian era) in Aberdeenshire of Scotland, by Kidston and Lang in 1917.
2. The two species *R. major*, about 40-50 cms in height and *R. gwynne-vaughani*, about 20 cms in height, found at this station, were well preserved, hence their form and structure are well known.
3. The plant grew in swampy marshes. It was differentiated into horizontally creeping rhizome and an upright branched shoot without leaves.
4. There were no roots. Unicellular rhizoids were borne in patches, on underside of the rhizome.
5. The upright branches were dichotomously branched and gradually tapering. The sporangia terminated these branches.

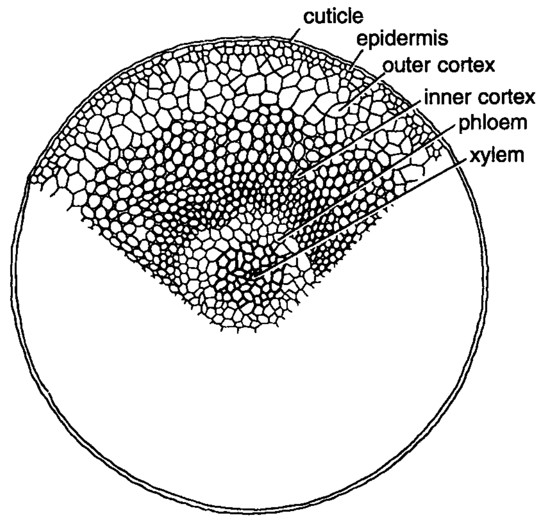


Fig. 2. *Rhynia.* T.s. of rhizome (a part is shown cellular).

Exercise 2

***Object* : Study of anatomy of rhizome and aerial shoot.**

Work procedure

Study the prepared slides of transverse section of rhizome and aerial branch.

Comments

1. Both aerial branches and rhizome were differentiated into epidermis, cortex and stele.
2. **The epidermis** was cuticularized with stomata scattered on the aerial shoot. They were absent from the rhizome.
3. **The cortex** was differentiated into outer and inner zone.
4. **The outer cortex** was only 1-4 layered and had compact parenchymatous cells.
5. **The inner cortex** had spherical cells with large intercellular spaces. The inner cortex is believed to be the chief photosynthetic region of the plant.
6. **The stele** was a typical protostele with xylem completely surrounded by phloem.

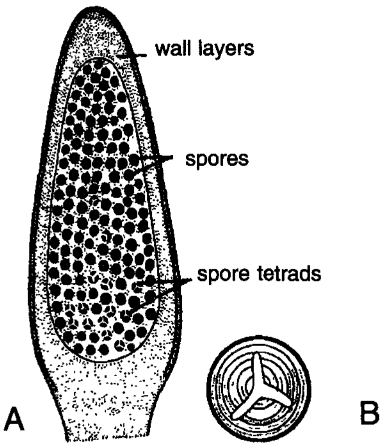


Fig. 3. ***Rhynia.*** Reproductive structures : **A.** A.L.s. sporangium, **B.** Single spore.

Exercise 3

Object **: Study of reproductive organ.**

Work procedure

It is sporangium borne at the tip of the branch. Study a slide of L.s. of sporangium.

Comments

1. It was nearly oblong in shape, broad at the base and pointed at the apex.
2. The wall of the sporangium was several layered, of which two could be easily distinguished.
3. The outer wall was cuticularized and the innermost acted as tapetum.
4. Inside the sporangium were spores, all of which were of the same size. Spores in tetrads were also seen in some specimens.
5. Columella was absent from the sprangium.

Identification

Division—**Pteridophyta**. (1) Generally true roots present. (except in Psilopsida), (2) True vascular strand present.

Sub-division—**Psilophytopsida**. (1) True roots absent, (2) Shoot differentiated into subterranean rhizome and apical portion, (3) Sporangia borne terminally, (4) Homosporous.

Order—**Psilophytales**. (1) Sporophyte dichotomously branched, (2) Sporangia generally borne singly.

Family—**Rhyniaceae**. (1) Rhizoids unicellular, on rhizomes, (2) Aerial portion leafless.

Genus—***Rhynia***. (1) Subterranean portion not corm-like, (2) No columella in sporangia.

Horneophyton lignieri

Classification

Division	—	**Pteridophyta**
Sub-division	—	**Psilophytopsida**
Order	—	**Psilophytales**
Family	—	**Rhyniaceae**
Genus	—	***Horneophyton***

Exercise 1

Object **: Study of the external features of the reconstructed plant.**

Work procedure

Study the features from a drawing of a reconstructed plant.

Comments

1. *Horneophyton* with one species *H. lignieri* was found with *Rhynia* in the Rhynichert bed of Aberdeenshire, Scotland. It was previously named as *Horneo*.
2. The rhizome was a lobed parenchymatous body bearing non-septate rhizoids but with no continuous vascular strand of its own.

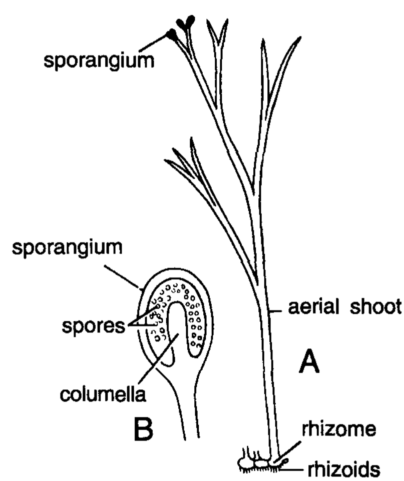


Fig. 1. ***Horneophyton lignieri.*** **A.** Restoration. **B.** L.s. sporangium.

3. The rhizome gave rise to upright shoots which ranged from 1 to 2 mm. in diameter and were dichotomously branched.
4. Sporangia were borne on the tips of these branches and were 1 to 2 mm in diameter and 2 or more mm in length.

Exercise 2

Object : **Study the fossil slide of L.s. of sporangium.**

Work procedure

Slide of fossil of sporangium is available. Study various characters using microscope.

Comments

1. The sporangium was oval or slightly cylindrical spore sac, somewhat pointed distally.
2. The sporangial wall consisted of a cuticularised epidermis followed by several layers of thin walled cells. The innermost layer was a well defined tapetum.
3. The most striking feature was the presence of central columella projecting into the spore cavity from the base. The columella was continuous with the phloem of the main stem.
4. Surrounding the columella and within the sporangial wall were many spores.
5. The spores were about 50 m in diameter and occur as tetrads.

Identification

Division—**Pteridophyta**. (1) Generally true roots present (except Psilopsida), (2) True vascular strand present.

Sub-division—**Psilophytopsida**. (1) True roots absent, (2) Shoot differentiated into subterranean rhizome and apical portion, (3) Sporangia borne terminally.

Order—**Psilophytales**. (1) Sporophyte dichotomously branched, (2) Sporangia generally borne singly.

Family—**Rhyniaceae**. (1) Rhizoids unicellular, (2) Aerial portion leafless.

Genus—***Horneophyton***. (1) Rhizome—a lobed parenchymatous body, (2) Rhizome without a continuous vascular strand of its own, (3) Presence of columella in the sporanigum.

Lepidodendron

Classification

Division	—	**Pteridophyta**
Sub-division	—	**Lycopsida**
Order	—	**Lepidodendrales**
Family	—	**Lepidodendraceae**
Genus	—	***Lepidodendron***

Exercise 1

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

Work procedure

Study the reconstruction of the plant. Observe the differentiation into stigmarian root system, Lepidophylloids leaves and *Lepidostrobus* strobili.

Comments

1. *Lepidodendron* with over 100 species, is the largest known genus. It is found in the shales and sandstones of carboniferous coal bearing formations.
2. The root system is included in the form genus *Stigmaria*, leaves in form genus *Lepidophylloides* and the strobili in the form genus *Lepidostrobus*.
3. *Lepidodendron* was a tall tree reaching 40 meters. The trunk was straight that branched dichotomously only at the top. The branches were shorter.
4. The branches were covered with spirally arranged leaves, placed in the form genus *Lepidophylloides*. These were simple, ligulate, acicular to linear. A single vein traversed the length of a leaf.
5. Leaves were attached to the summit of pyramidal cushion-like leaf bases which were rhombic or diamond shaped. The leaf scars formed oblique or spiral rows on the stem. Actual leaf scar was above the middle line of rhombus. In the leaf scar, the bundle scar mark

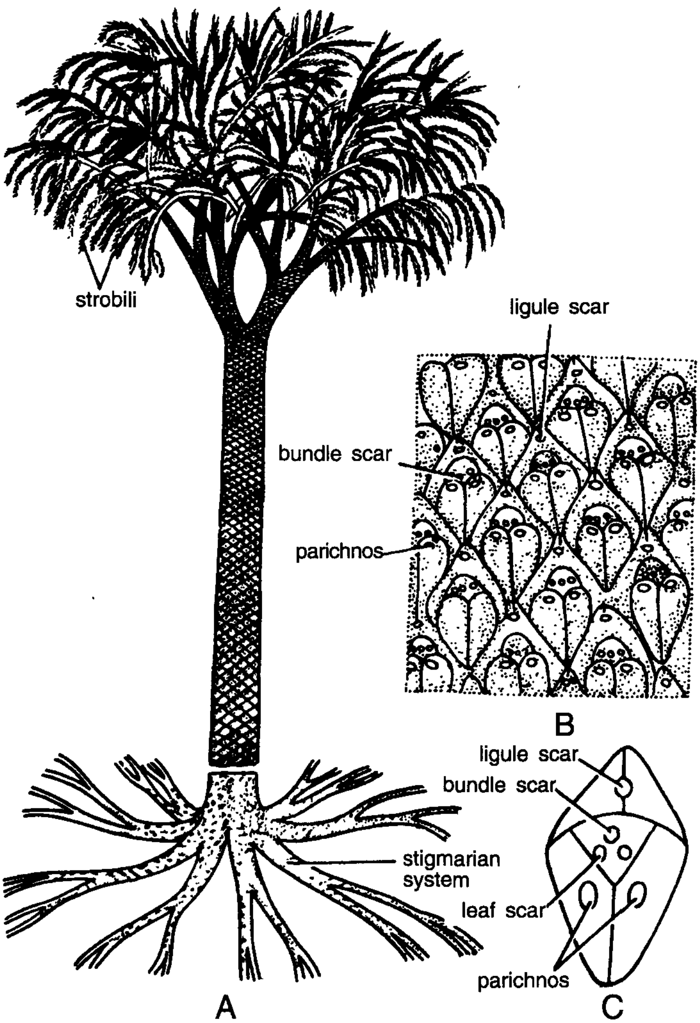


Fig. 1. ***Lepidodendron.*** **A.** Reconstruction of tree. **B.** Surface of the trunk showing scars formed by the leaf bases **C.** A leaf scar.

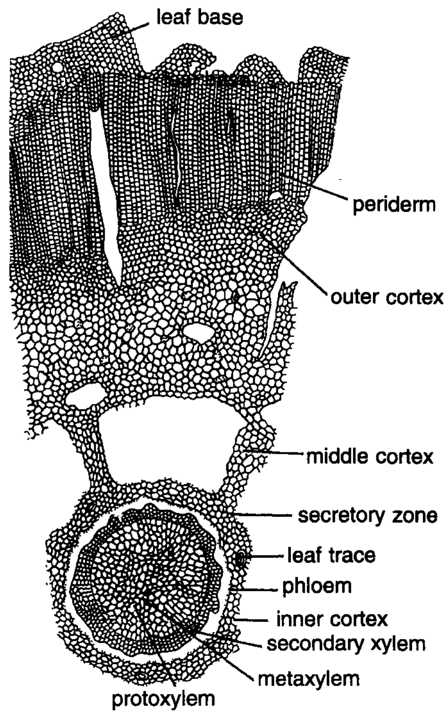


Fig. 2. ***Lepidodendron.*** *L. selaginoides* (= *L. vasculare*). T.s. of stem.

was flanked by two scars and at the lower level by two more large scars which represented parichnos scars (strands of loosely arranged parenchyma).

6. Typical *Lepisdodendron* trunk was attached to a typical stigmarian root system. The base of the trunk was divided into four massive rhizophores which later again divided dichotomously.
7. The strobili borne terminally were found attached to a branch identical to *Lepidodendron* and are placed under the form genus *Lepidostrobus.*

Exercise 2

Object **: Study of the anatomy of stem.**

Work procedure

Study the prepared slide of transverse section of the main trunk or a branch.

Comments

1. It shows a protostele or siphonostele with exarch and polyarch protoxylem.
2. Metaxylem was present in the centre with many protoxylem points on periphery.
3. A ring of cambium situated outside produced a narrow zone of secondary xylem. Both primary

and secondary xylem were made of scalariform and spiral tracheids.

4. A large cortex surrounded the vascular tissues. It showed four regions-
   (i) Inner cortex : homogeoneous and made of parenchyma except for leaf traces.
   (ii) Secretory zone made of intermingled large and small cells, many of which were filled with a dark coloured substance.
   (iii) Middle cortex that consisted of delicate cells which were often destroyed.
   (iv) Outer cortex made of alternate radial masses of thick and thin walled elements.
5. Outermost was the periderm formed by phellogen. It consisted of many elements on its inner side forming endophelloderm than on the outer side that formed exophelloderm.

Exercise 3

***Object* : Study the strobilus.**

Work procedure

Study the structure of a strobilus by observing and studying the slide of L.s. of strobilus.

Comments

1. The strobili borne by Paleozoic lepidodendrids are placed in organ genus *Lepidostrobus*. All known species are heterosporous.
2. Strobili occurred on terminal parts of certain smaller branches. The strobili were elliptical, 1 to 7.5 cm in diameter and 2.5 to 30 cm or more long.
3. Each consisted of a central axis around which sporophylls were found in spirals or whorls (in some cases verticillate).
4. The sporophyll was peltate with the upper terminal lobe overlapping the sporophyll above. The sporophylls were attached at right angles to the axis.
5. Each sporophyll bore a single sessile elongate sporangium on its adaxial face. Just beyond the sporangium, the sporophyll had a small ligule.
6. There was also a shorter downward prolongation of the lamina called heel, completely covered by the lamina of the sporophyll below.
7. The sporangium was a large sac-like structure wider than the sporophyll. The wall was of a single layer of prismatic cells. Some radiating flaps of tissue called trabeculae extended upward from the base of the sporangium.
8. The microsporangia and megasporangia are of the same size.
9. Microsporangium enclosed several hundred microspores. Megasporangium enclosed 4, 8, or 16 megaspores.
10. Megaspores showed gametophytic development while still within the megasporangium.

Identification

Division—**Pteridophyta**. (1) True roots present, (2) Plant body differentiated into stem, roots and leaves, (3) True vascular strand present.

Sub-division—**Lycopsida**. (1) Leaves microphyllous, (2) Sporangia borne singly on adaxial face of the sporophyll or in its axil, (3) Sporophylls borne in strobili.

Order—**Lepidodendrales**. (1) Plants tree-like, (2) Secondary tissues formed by means of a cambium, (3) Leaves microphyllous, ligulate, (4) Strobili heterosporous.

Family—**Lepidodendraceae**. (1) Aerial portion freely branched, (2) Strobili at the tips of branches, (3) Trunk and branches with spirally arranged leaf scars.

Genus—***Lepidodendron***. (1) The vertical diagonal of the rhombus longer than the tranverse, (2) Cones borne on short lateral branches.

Calamites

Classification

Division	—	**Pteridophyta**
Sub-division	—	**Sphenopsida**
Order	—	**Calamitales**
Family	—	**Calamitaceae**
Genus	—	***Calamites***

Exercise 1

***Object* : Study the reconstructed plant.**

Work procedure

Study reconstruction from the standard books.

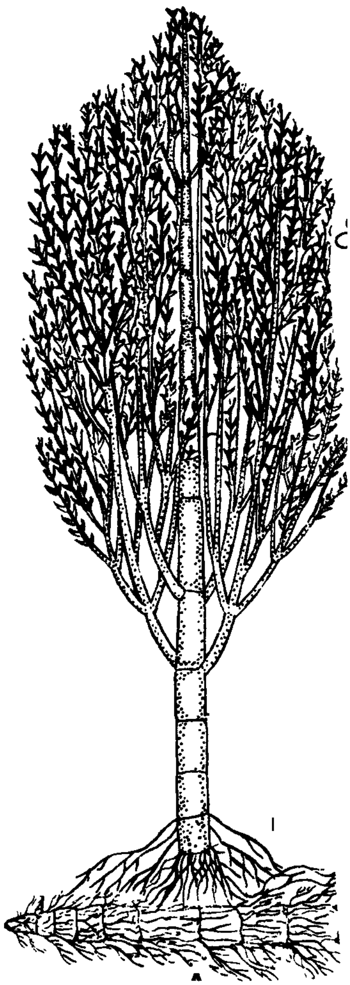


Fig. 1. ***Calamites.*** Reconstruction of the plant.

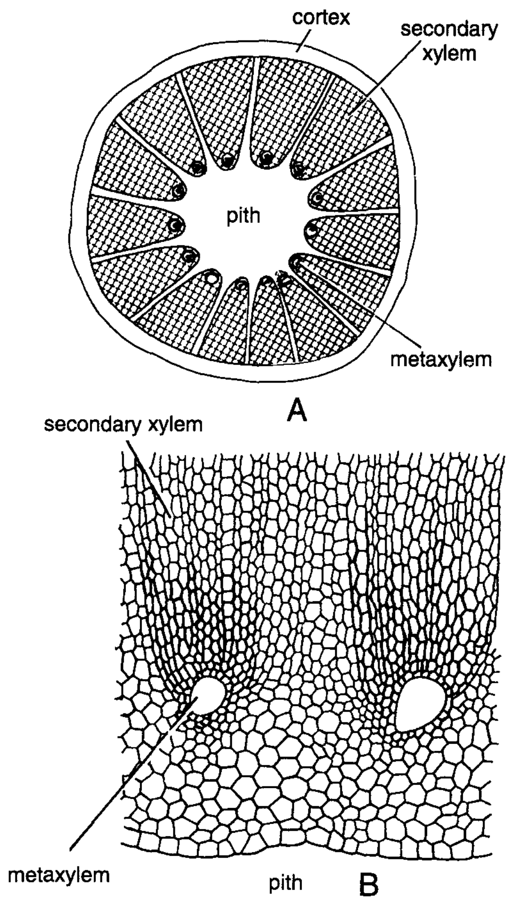


Fig. 2. ***Calamites.*** T. s. of stem. **A.** Outlines, **B.** A part cellular.

Comments

1. Genus *Calamites* appeared in Upper Devonian, and was most abundant during Carboniferous and became extinct early in the Triassic.
2. The plant was 20 to 30 meters tall tree.
3. The tree was arborescent with horizontal rhizome, aerial shoots and whorls of leaves.
4. Rhizomes were differentiated into nodes and internodes. Nodes bore whorl of adventitious roots.
5. Aerial branches were borne on the upper side of rhizome. These wee generally constricted at the place where these joined the thizome.
6. The lowermost nodes of the erect branches produced whorls of adventitious roots.
7. The aerial branches were highly articulated. The leaves were borne in whorls at the upper nodes. These also had branches borne in pairs.
8. Each node had a single whorl of leaves. The leaves were smaller and perhaps were photosynthetic organs. These were lanceolate, linear or spathulate with a single vein.
9. Genus *Calamites* is divided into subgenera on the basis of mode of branching and general growth pattern.
   (i) *Eucalamites*—bore one to many branches at the node.
   (ii) *Calamitina*—bore branches in whorls which were separated by a series of branchless nodes.
   (iii) *Diplocalamites*—2 to 3 branches produced with branches alternating from node to node.

(iv) *Crucicalamites*—large number of branches produced at each node.
(v) *Stylocalamites*—wihout branches or branches fewer and irregularly scattered.

Exercise 2

Object **: Study of the stem anatomy.**

Work procedure

Study the prepared slide showing stem anatomy.

Comments

1. The petrified calamitean stem of three form genera viz. *Arthropitys, Calamodendron* and *Arthroxyulonn* have been recognised. They have been established to belong to *Calamites* and their special names are used only when necessary.
2. Petrified stems of *Calamites* are frequent in the coal balls and other carboniferous petrifactions.
3. The stem was ridged and furrowed which alternate at each successive internodes.
4. The transverse section of internode shows endarch siphonostele with secondary xylem produced by vascular cambium.
5. There was a large hollow central pith cavity. It became solid at nodes forming a diaphragm.
6. Surrounding the pith was a ring of more than a dozen primary collateral vascular bundles.
7. The protoxylem was endarch and was represented by carinal canal formed due to dissolution of many elements.
8. The metaxylem was very less in amount. It was composed of scalariform and pitted tracheids.
9. Cambium occurred in between xylem and phloem.
10. Secondary xylem was formed on the inner face of cambium. It was made of tracheids and rays. Tracheids were arranged in radial rows. These showed scalariform thickenings or multiseriate bordered pits on the radial walls. Secondary xylem formed wedge-shaped zones separated by interfascicular rays.
11. The rays show considerable diversity in structure and serve as the basis for generic distinction.

12. The tissues outside the secondary xylem showed inner cortex made of thin walled parenchyma with resin canals.
13 The cells of the outer cortex were also thin walled but were much smaller than the cells of the inner zone.
14. The cortex was as wide as xylem and formed thick bark (also termed periderm) that was smooth externally.

Identification

Division—**Pteridophyta**. (1) True roots present, (2) Plant body differentiated into stem, roots and leaves, (3) True vascular strand present.

Sub-division—**Sphenopsida**. (1) Stem branched, articulated, ridged and furrowed with distinct nodes and internodes, (2) Leaves microphyllous, small, scaly and arranged in whorls at nodes.

Order—**Calamitales**. (1) Tree-like sporophytes with considerable secondary thickening of stem and branches, (2) Strobilus with central articulated axis which bore alternate whorls of sporangiophores and sterile bracts.

Family—**Calmitaceae**. (1) Stem branched, branches in whorls at nodes, (2) Stem shows endarch siphonostele.

Genus—***Calamites***. (1) Subterranean rhizome present, (2) Lower nodes producing adventitious roots, (3) Vallecular canal absent, (4) Periderm may or may not be formed.

GYMNOSPERMS

Lyginopteris

Classification

Division	—	**Gymnospermae**
Class	—	**Cycadopsida**
Order	—	**Pterdospermales**
Family	—	**Lyginopteridaceae**
Genus	—	***Lyginopteris***

Exercise 1

Object **: Study of external morphology of the stem.**

Work procedure

Study the diagrammatic representation of the stem genus from standard books. Summarise the major points of significance.

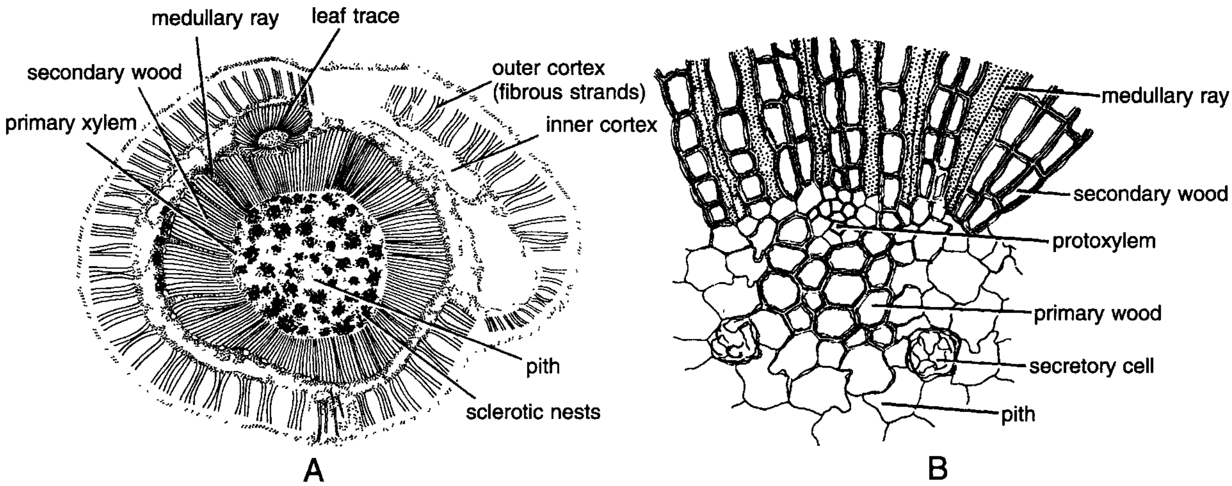


Fig. 1. ***Lyginopteris.*** **A.** T.s. of stem (outlines), **B.** T.s. stem— a part cellular.

Comments

1. *Lyginopteris* is a palaeozoic pteridosperm. It is a stem genus of *Calymatotheca Hoeninghausi.* The descriptions of *Lyginopteris Oldhamia* are available.
2. The genus was described under *Dadoxylon oldhamium* by Binney in 1886. Later t h e genus was transferred to *Lyginodendron* on the basis of cortical impressions. Genus *Lyginopteris* was established by Potonie in 1899.
3. Petrifications and compressions of the stem were found in the lower and middle. Coal measures of Britain, continental Europe and north America.
4. The stem was long and generally slender. Diameter varied from 2 mm to 40 mm, sometimes up to 4 cms . Some specimen showed branching. The branches were axillary when present. These were arranged spirally.

Exercise 2

Object **: Study the anatomy of stem.**

Work procedure

Study the slide of T.s. of fossil stem or alternatively study the diagrams from the books and draw a list of important features of anatomy.

Comments

1. The section shows three distinct regions—
   (i) central pith,
   (ii) centrally located vascular tissue and
   (iii) outer region of cortex.
2. A large parenchymatous pith was present in the centre. Many sclerotic nests were present scattered in this region.
3. Along the margins of the pith were present many strands of primary xylem. Protoxylem was mesarch .
4. Primary xylem was made of tracheids with bordered pits on their radial walls.
5. The strands of primary xylem were surrounded by a wide region of secondary xylem. It was made of tracheids with multiseriate bordered pits. Wide medullary rays which alternated with groups of primary xylem were also present.
6. Continuity of secondary xylem was interrupted by outgoing leaf traces giving an appearance of four large and unequal masses.
7. Outside the secondary xylem were present cambium and secondary phloem. Pericycle was situated outside the vascular tissues and was represented by groups of stone cells.
8. A band of internal periderm was present outside pericycle.
9. The cortex situated outside was divisible into inner cortex and outer cortex.

10. The inner cortex was poorly preserved and, therefore, the tissues were not distinct. However, it gave an impression as if made of large parenchymatous cells.
11. Outer cortex was made of a wide region of radially arranged fibrous strands. These were anastomosed and formed a distinct reticulum.
12. Numerous glands were present on the stem surface.The tip of the gland was spherical and was made of small cells.These were sessile as well as stalked.

Identification

Division—**Gymnospermae**. (1) Ovules naked, (2) Seeds attached to scales, (3) Scales forming a strobilus.
Class—**Cycadopsida**. (1) Wood manoxylic, (2) Large frond-like leaves, (3) Seeds with radial symmetry.
Order—**Pteridospermales**. (1) Leaves large, frond-like, pinnately compound, (2) Large leaf traces with one or more strands,(3) Spores formed in sporangia, aggregated in synangia.
Family—**Lyginopteridaceae**. (1) Stem monostelic, (2) Petioles with single vascular strand, (3) Seeds small.
Genus—***Lyginopteris***. (1) Presence of stone cells in pith, (2) Wide fibrous bands in outer cortex.

Cycadeoidea

Classification

Division	—	**Gymnospermae**
Class	—	**Cycadopsida**
Order	—	**Bennettitales**
Family	—	**Cycadeoidaceae**
Genus	—	***Cycadeoidea***

Exercise 1

Object **: To study the external features of stem.**

Work procedure

Study the external features of the reconstructed plant from the figures and text available from standard text books. List various important features.

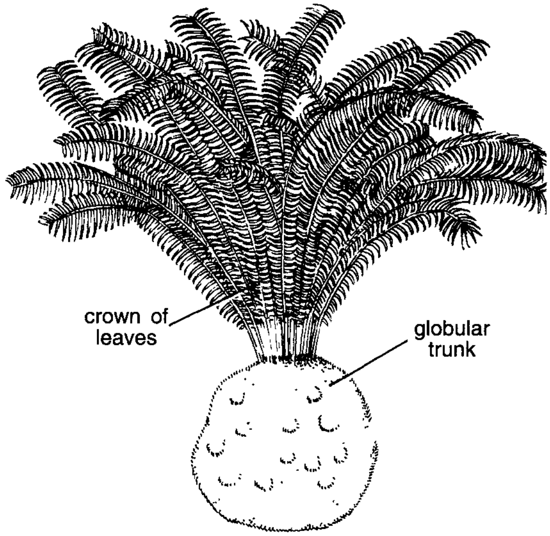


Fig. 1. ***Cycadeoidea.*** External features.

Comments

1. *Cycadeiodea* (= *Bennettitis)* is a fossil that includes many species. Many petrifications have been found of which some are complete plants. This fossil has been found in Jurassic and cretaceous strata of Dakota, America.
2. The stems were generally small (less than three feet in height, sometimes 10 to 12 feet,) and thick (about 2 feet). It remained covered with persistent leaf bases. A few specimen were branchless (though in some 3 to 4 small branches arose from in a group from a basal stalk).
3. Persistent leaf bases were closely placed and formed a thick mantle. In between them were present flat scale-like hairs called ramentum.
4. Only young leaves have so far been found. These were present close to the apex. Leaves were pinnately compound and the venation was parallel.
5. The flowers were borne in the axils of leaves and completely embedded in the armour of persistent leaf bases.

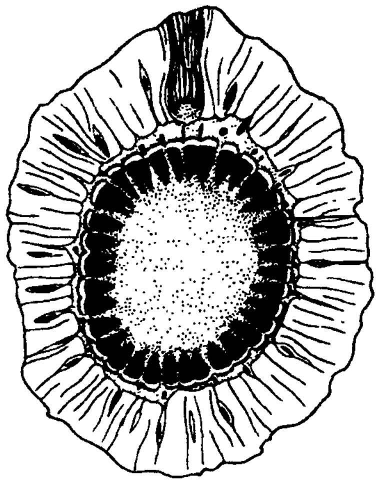


Fig. 2. ***Cycadeoidea.*** T.s. stem.

Exercise 2

Object **: Study of anatomy of transverse section of stem.**

Work procedure

Study the slide of T.s. of stem or study the diagrams available in the standard books.

Comments

1. The section is differentiated into three regions central pith, middle region consisting of wood and outer cortex.
2. Centrally located pith is smaller in diameter. It was surrounded by wood from all the sides.
3. Wood formed a small region that surrounded central pith. The region appeared broken by ray-like extensions projecting from central pith.
4. Endarch protoxylem was situated on the inner side of the wood , close to the pith. Vascular bundles were arranged in a ring and the xylem was arranged centrifugally.
5. Secondary xylem was made of scalariform tracheids and small rays. Rays were uniseriate or biseriate.
6. Medullary rays extended beyond cambium up to phloem. These alternate with parenchyma and vertical vascular tissues.
7. Cortex was made of parenchyma where gum canals and leaf traces were abundant.
8. Leaf trace occurred singly at the place of their origin but broke into many mesarch strands to arrange itself into horse-shoe shaped structure. The mouth of hose-shoe was directed outwards.

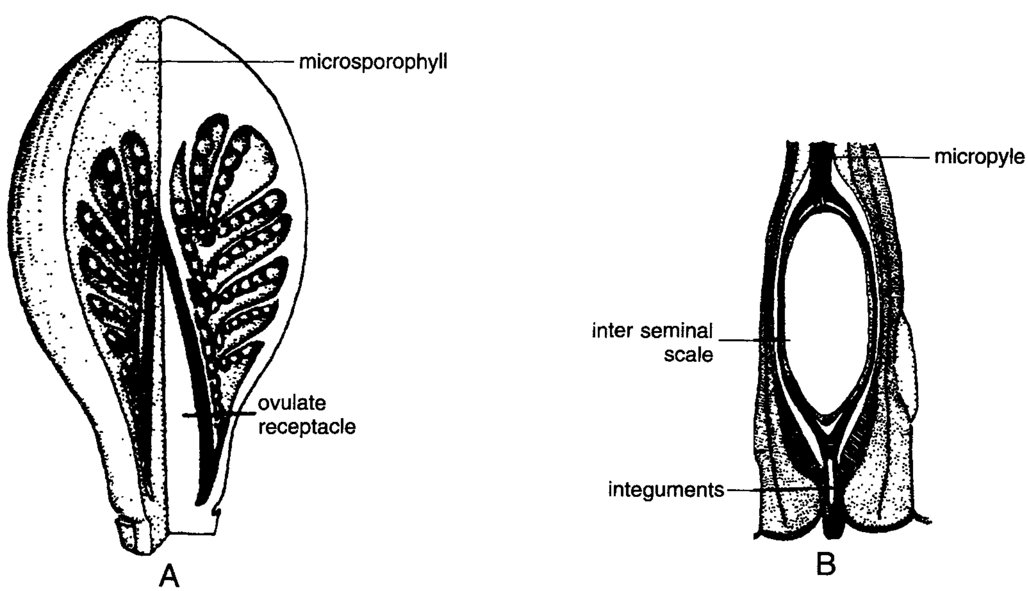


Fig. 3. ***Cycadeoidea.*** **A.** L.s. of cone, **B.** L.s. of Ovule.

Exercise 3

***Object* : To study the reproductive parts.**

Work Procedure

Study the diagrams from the book and prepare the list of main features.

Comments

1. Fruiting bodies were produced on the axillary shoots (In some species more than 100 fruiting bodies).
2. These appeared in a group between leaf bases. Fructifications were bisexual.
3. Many spirally arranged bracts were present on fertile shoots. These kept fructifications covered till they matured. On maturity the bracts of the flower opened and formed saucer-shaped perianth. The structures were modified androecium and gynoecium.
4. The pollen bearing structures bore a whorl of about 20 microsporophylls.. These were almost fused at the base. Each microsporophyll consisted of main rachis which bore two rows of trabeculae which in turn held two lateral rows of multicellular synangia.Each synangia was pear-shaped bearing 20 to 30 longitudinal pollen sacs.
5. The ovule bearing structure was made of conical or spherical receptacle with many stalked ovules and interseminal scales .
6. The ovule was orthotropous with a long micropylar beak. The integument of the ovule was fused with the nucellus except at the apex where it formed micropyle.
7. The seeds were small and invested with a basal cupule.It was somewahat elongated or oval in shape and possessed two cotyledons.

Identification

Division—**Gymnospermae**. (1) Ovules naked, (2) Seeds attached to scales, (3) Scales forming a strobilus.

Class—**Cycadopsida**. (1) Wood manoxylic, (2) Large frond-like leaves, (3) Seeds with radial symmetry.

Order—**Bennettitales**. (1) Tree trunk covered with mantle of persistent leaf bases, (2) Microsporophylls in groups, at the tip, leaves frond-like, (3) Megasporophylls in cone-like organization.

Family—**Cycadeiodaceae**. (1) Trunk columnar, (2) Trunk covered with mantle of leaf bases, (3) Flowers sunk in the distal part of the trunk.

Genus—***Cycadeoidea***. (1) Small, globular trunk,(2) Bisexual strobilus, (3) Orthotropous ovules.

Williamsonia

Classification

Division	—	**Gymnospermae**
Class	—	**Cycadopsida (Bennettitopsida)**
Order	—	**Cycadeoidales (Benettitales)**
Family	—	**Williamsoniaceae**
Genus	—	***Williamsonia***

Exercise 1

***Object* : To study the external features of the plant.**

Work procedure

Study the morphological features of the reconstructed plant, note the significant features and list them from standard books.

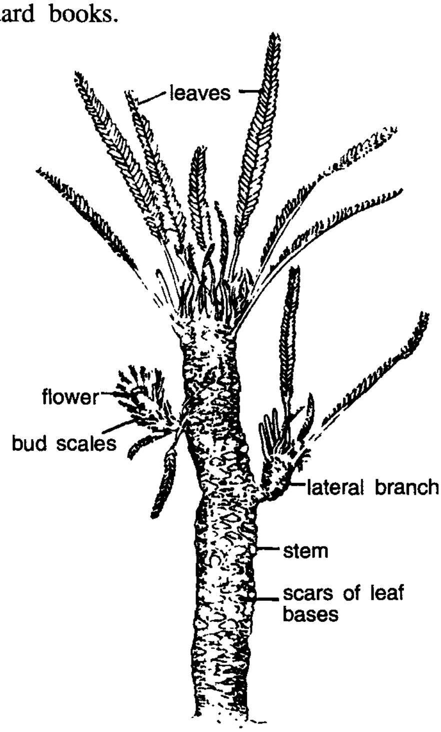


Fig. 1. ***Williamsonia.*** External features. Reconstruction.

Comments

1. Many species have so far been reported from upper Triassic and Jurassic beds of India, Europe and north America.
2. It was earlier placed under Zamia gigas by Williamson (1868). Later it was named in honour of its discoverer. *Williamsonia sewardiana* is the best known species described by late Professor Birbal Sahni from Rajmahal Hills, Bihar (India). Gupta (1953) later described another species from Rajmahal Hills and named it after Professor sahni.
3. *W. sewardiana* had a study columnar trunk similar to that of *Cycas.* It had a crown of pinnately compound leaves. The stem was covered with alternating areas of large (foliage) and small (scaly) leaf bases.
4. The leaf was placed in from genus *Ptilophyllum.* It has linear and parallel veined leaflets similar to those found in *Zamia.*
5. The lateral branches were borne through leaf bases and possessed flowers. These branches were also covered with scales and bracts.
6. The flowers were unisexual.

Identification

Division—**Gymnospermae**. (1) Ovules naked, (2) Seeds attached to the scale, (3) Scales forming a strobilus.

Class — **Cycadopsida**. (1) Wood manoxylic, (2) Large fornd-like leaves, (3) Seeds with radial symmetry.

Order — **Bennettitales**. (1) Tree trunk covered by a mantle of persistent leaf bases, (2) Microsporophylls in groups, at the tip, leaves frond-like, (3) Megasporophylls in cone-like organization.

Family —**Williamosniaceae**. (1) Stem delicate, branched, (2) Inflorescence stalked or sessile, not sunk in the scales of peristent leaf bases.

Genus—***Williamsonia***. (1) Stem study and columnar, (2) Leaves pinnately compound, (3) Lateral branches with flowers.

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Appendix

Fixing Agents and Preservatives

1. **Carnoy's fluid**

100% ethyl alcohol	...	30 cc
Glacial acetic acid	...	5 cc
Chloroform	...	15 cc

It is used for root tips, anthers, etc. and is preferred for its great penetrating power.

2. **Formalin-Aceto-Alcohol**

50% or 70% ethyl alcohol	...	90 cc
Glacial actic acid	...	5 cc
Formalin	...	5 cc

It is popularly known as FAA and is a standard universal fixative. It is the most extensively used fixing and killing agent.

3. **Formalin-Propino-Alcohol**
In the preparation of FAA use propionic acid instead of acetic acid.

4. **Randolph's modified Navashin fluid**

Solution A.	Chromic acid	...	5 gm
	Glacial acetic acid	...	50 cc
	Distilled water	...	320 cc
Solution B.	Natural formalin	...	200 cc
	Saponin	...	3 gm
	Distilled water	...	175 cc

At the time of use, mix solutions A and B in equal amounts. Recommended for buds, roots tips and similar objects.

5. **Bouin's fluid**

Picric acid (1.5% aq. solution)	...	75 cc
Formalin	...	25 cc
Glacial acetic acid	...	5 cc

This fixative is more useful than those with chromic acid.

Stains

1. **Acetocarmine**
Dissolve 1 gm of stain in 100 cc of boiling 45% acetic acid (or propionic acid). Cool and decant. Add a few drops of saturated aqueous solution of ferric acetate. Cool by keeping in ice for at least twelve hours. Filter and store the stock in refrigerator. For storage, use dropping bottle that is dark or covered with a black paper.

2. **Aniline blue (Cotton blue, China blue, Water blue)**

Aniline blue	...	1 gm
Alcohol 90% or water	...	100 cc

For better results stain or alcohol should be slightly acidified with hydrochloric acid.

3. **Crystal violet (Gentian violet)**

Crystal violet	...	1 gm
Distilled water	...	100 cc

4. **Erythrosine**

(a)	Erythrosine	...	1 gm
	Alcohol 90%	...	100 cc
(b)	Erythrosine	...	1 gm
	Absolute Alcohol	...	5 cc
	Clove oil	...	95 cc

5. **Fast green**

(a)	Fast green	...	0.5 gm
	Alcohol 90%	...	100 cc
(b)	Fast green	...	0.5 gm
	Absolute alcohol	...	25 cc
	Clove oil	...	75 cc

6. **Gram's iodine**

Iodine	...	2 gm
Potassium iodide (KI)	...	3 cc
Distilled water	...	300 cc

7. **Hematoxylin**
It is a chromogen derived from logwood *Haematoxlyon coampechianum* of Leguminosae. Two types of hematoxylins are commonly employed (a) Heidenhain's and (b) Delafield's hematoxylin.
(a) *Heidenhain's hematoxylin.* Half per cent solution of the stain is prepared in warm and distilled water. It is then stored in dark and

closed bottle to ripen for at least four days before use.

(b) Delafield's hematoxylin.

1. A saturated aqueous solution (100 cc) of ferric ammonium sulphate is prepared.
2. One gram of stain is dissolved in 6 cc of absolute alcohol.
3. Mixture of solutions 1 and 2 is prepared. Add to this solution 25 cc of glycerine and 25 cc of absolute alcohol.
4. The solution thus prepared is allowed to remain for sufficient time till the colour becomes dark red.

8. Safranin

(a) Safranin	...	1 gm
Alcohol 95%	...	50 cc
Distilled water	...	50 cc
(b) Safranin	...	1 gm
Distilled water	...	100 cc

Mounting Media

1. Glycerine jelly

Soak some gelatin for 2 to 3 hours in cold water, pour off the excess water and heat until melted. To 1 part of this, add $1\frac{1}{2}$ parts of glycerine and filter while still hot. Add 2 or 3 percent phenol. Still keeping the mixture hot and fluid, add drop by drop a saturated solution of methyl green in 50 percent alcohol, until the glycerine becomes fully as dark as green ink.

2. Lactophenol

Mix equal parts of phenol crystals, lactic acid, glycerine and distilled water. Cotton blue may be mixed to stain fungi.

Recommended Stains and Mounting Media

1. Algae :

For study, mostly temporary or semi-permanent mounts are prepared. However, sometimes permanent preparations may also be made but extreme care, is necessary, not to allow any shrinkage while dehydrating. This is done by passing the material through a very close dehydrating series e. g. alcohol 5%, 7%, 10%, 12%, 15%, and similar grades.

If a single stain is to be used in any of these preparations, the following are some of the most commonly used—

Stains :	Mounting media :
(a) Aniline blue	(a) Glycerine jelly
(b) Aceto-carmine	(b) Glycerine 10%
(c) Cotton blue	(c) Formo-glycerine
(d) Delafield's hematoxylin	(d) Lactophenol
(e) Safranin	

2. Fungi

Temporary and semi-permanent preparations; single-staining:

Stains :	Mounting media :
(a) Aniline blue	(a) Lactophenol
(b) Cotton blue	(b) Glycerine10%
(c) Hematoxylin	

3. Bryophytes

Temporary, semi-permanent and permanent preparations; single staining :

Stains :	Mounting media :
(a) Delafield's hematoxylin	(a) Glycerine jelly
(b) Fast green	(b) Glycerine 10%
(c) Safranin	(c) Canada balsam.

4. Pteridophytes

Temporary, semi-permanent and permanent perparations; double staining.

Stains and combinations :

Principal Stain :	Counter Stain :
(a) Delafield's hematoxylin	— Safranin
(b) Safranin	— Fast green
(c) Crystal violet	— Erythrosine

Mounting media :

(a) Glycerine 10% (b) Canada balsam

5. Gymnosperms and Angiosperms

Temporary, semi-permanent and permanent preparations; double staining;

Stains and combinations :

Principal Stain :	Counter Stain:
(a) Safranin	— Fast green
(b) Crystal violet	— Erythrosine
(c) Delafield's hematoxylin	— Safranin

Mounting media :

(a) Glycerine 10% (b) Canada balsam.

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