

**SEETHALAKSHMI RAMASWAMI COLLEGE (AUTONOMOUS)**  
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**AFFILIATED TO BHARATHIDASAN UNIVERSITY**  
**TIRUCHIRAPPALLI – 620 002**



**LAB MANUAL**  
**III B.Sc., BOTANY**  
**PRACTICAL PAPER- IV**

**PLANT PHYSIOLOGY, BIOCHEMISTRY, BIOPHYSICS,  
ENVIRONMENT, BIODIVERSITY AND CONSERVATION**

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## PREFACE

Observation cum practical learning is a supplement to the theoretical class room knowledge. In plant physiology, fundamental experiments are dealt with labelled diagrams for self understanding. we hope, the method of presentations would boost the memory of the students and their performance in practical examinations. Environment is the assemblage of material factors and conditions surrounding the living organisms to which they show morphological and anatomical adaptations for their survival. This manual covers the characters and adaptations of hydrophytes, xerophytes, halophytes, epiphytes and parasites. The biotic and abiotic components of pond ecosystem, pyramid of numbers, food chain and food web are also illustrated.

We are highly indebted to our management, Seethalakshmi Ramaswami College for valuable support and also to Dr. (Mrs.) Kanaka Bhashyam, Principal, Seethalakshmi Ramaswami College for her fruitful guidance. We sincerely thank the authorities of Star College Scheme, DBT, New Delhi for the Financial Assistance.

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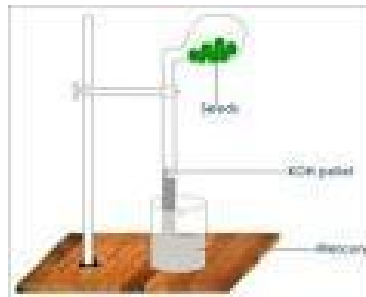
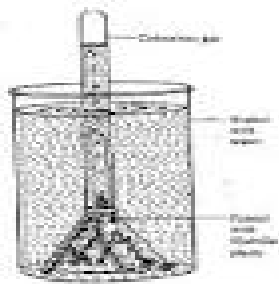
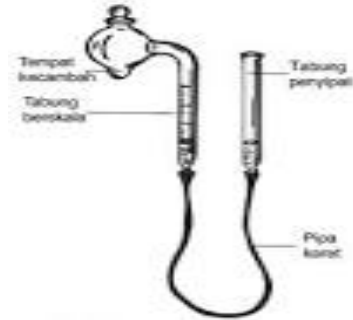
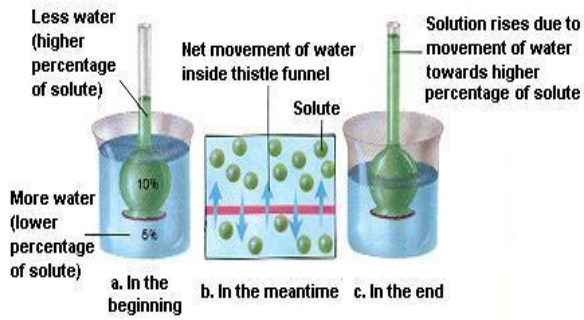
# PRACTICAL PAPER- IV

## PLANT PHYSIOLOGY, BIOCHEMISTRY, BIOPHYSICS, ENVIRONMENT, BIODIVERSITY AND CONSERVATION

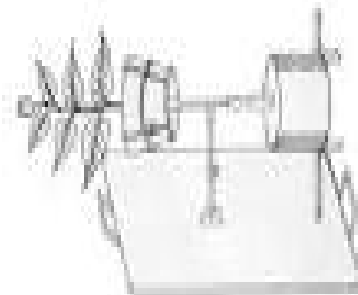
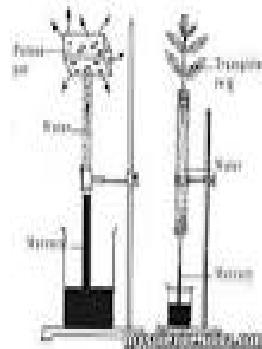
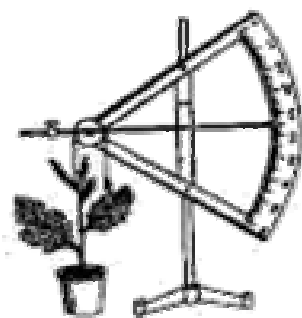
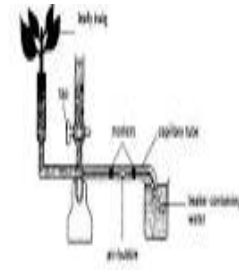
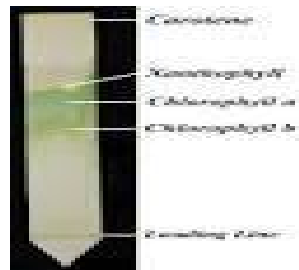
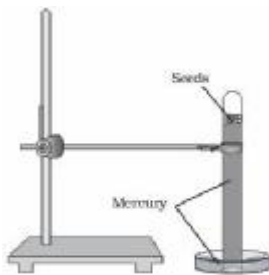
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## PLANT PHYSIOLOGY, BIOCHEMISTRY AND BIOPHYSICS



## **DILATOMETER**

### **AIM**

To demonstrate imbibition.

### **REQUISITES**

Dilatometer, water, seeds. Dilatometer consists of a glass jar with the movable piston to which a pointer is attached. A pointer slides over the graduated scale.

### **PROCEDURE**

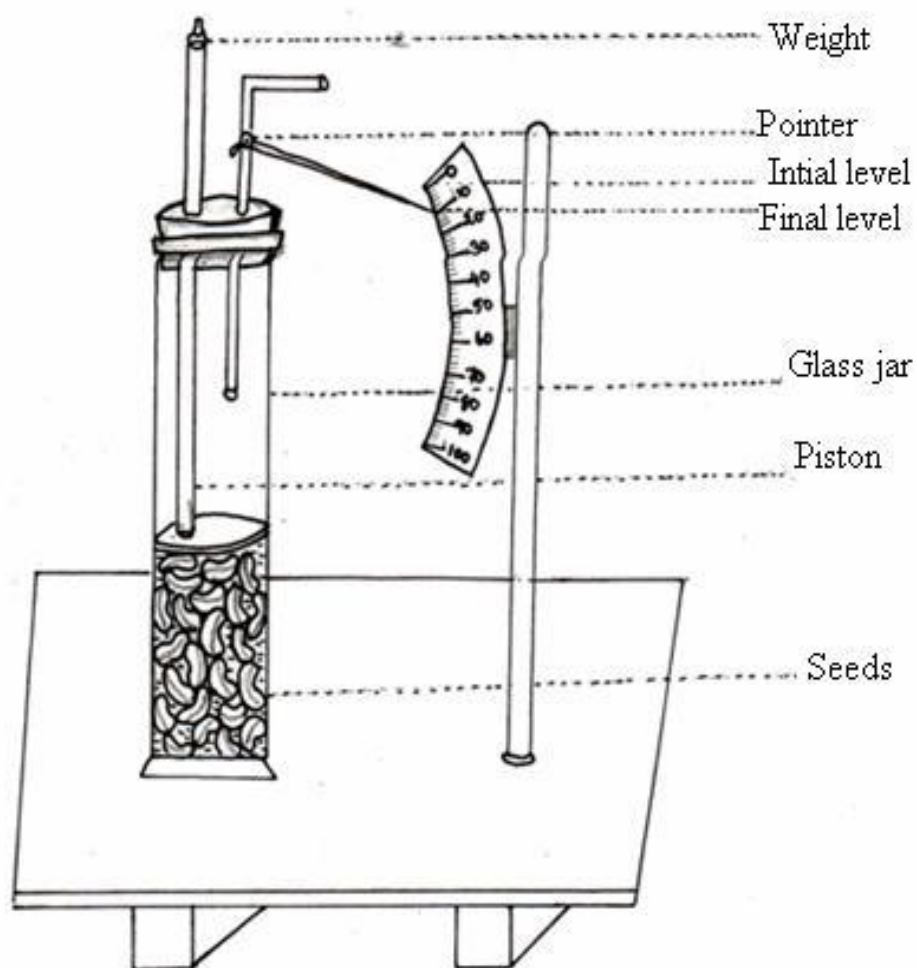
The dry seeds are taken in the jar and water is poured into the jar to immerse the seeds. The jar is tightly closed with lid which is attached to the movable piston resting on the germinating seeds. Weight is placed on the disc of the piston.

### **OBSERVATION**

The pointer moves down to the graduated scale.

### **INFERENCE**

The dry seeds contain colloidal substance which shows specific affinity towards water. As a result the seeds imbibe water and swells. This creates a pressure which is responsible for lifting up of the piston. This in turn pushes the pointer over the scale.





## **THISTLE FUNNEL EXPERIMENT**

### **AIM**

To demonstrate osmosis by thistle funnel experiment.

### **REQUISITES**

Thistle funnel, sheep's bladder, water, beaker, sugar solution and stand.

### **PROCEDURE**

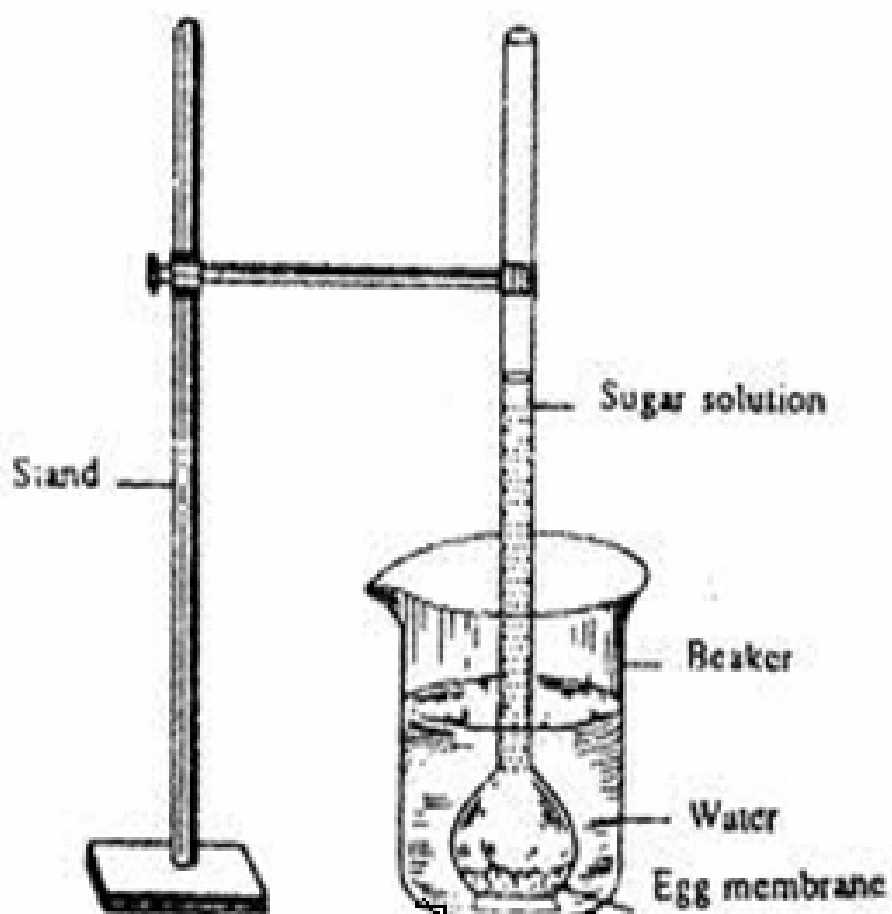
Thistle funnel is tied with sheep's bladder (It acts as semi permeable membrane).The thistle funnel is filled with concentrated sugar solution and the initial level is marked. The thistle funnel is placed in a beaker of water with the help of the stand .This step is kept for some times.

### **OBSERVATION**

The level of sugar solution in the thistle funnel is increased.

### **INFERENCE**

Osmotic pressure of the sugar increases in the thistle funnel. To decrease the osmotic pressure water moves from the beaker into thistle funnel through the semi permeable membrane.



## **POTATO OSMOSCOPE**

### **AIM**

To demonstrate osmosis in the living cells of potato.

### **REQUISITES**

Water, coloured sugar solution, potatoes, pins, scalpel, petridishes and graph paper.

### **PROCEDURE**

A large potato tuber is taken and the skin is removed. It is cut into two equal halves. The central portion is scooped and a hollow cavity is made, so as to leave a thin wall at the base. The potato cups were called as potato osmoscope. In the first osmoscope, the cavity is filled with water. A narrow strip of graph paper is placed in the cavity and the initial level is marked with pin. The osmoscope is placed in to a petridish containing water. In the second set up coloured sugar solution is taken in the potato cup and water is taken in the petridish. In the third set up water is taken in the potato cup and coloured sugar solution is taken in the petridish. The first set up treated as control.

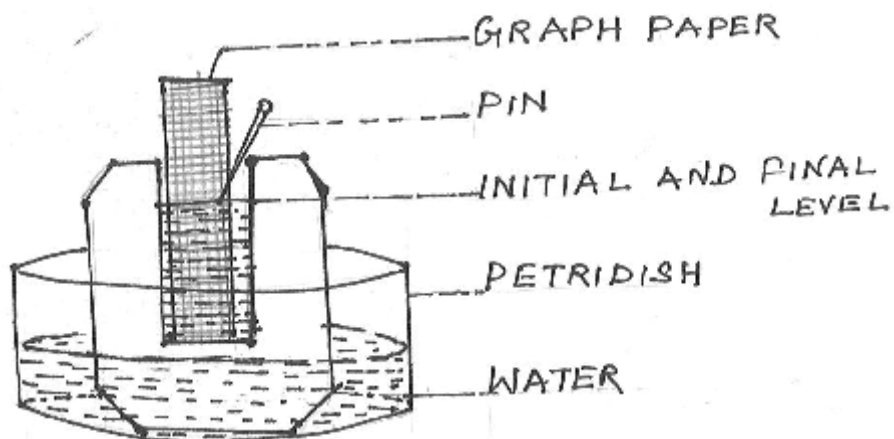
### **OBSERVATION**

There is no change in the control set up. The sugar solution of the second osmoscope rises whereas the level of water decreases in the third potato cup.

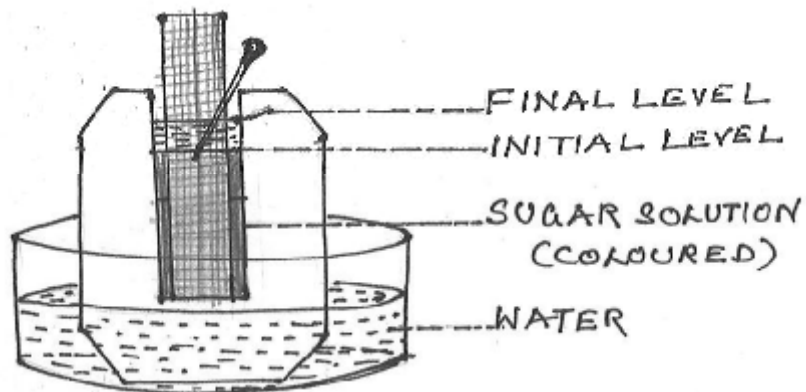
### **INFERENCE**

The rise in the level of coloured sugar solution in the second osmoscope is due to endosmosis. In the third set up, the decrease in level is due to exosmosis. There is no change in the control set up because the concentration of the solvent is equal on either side.

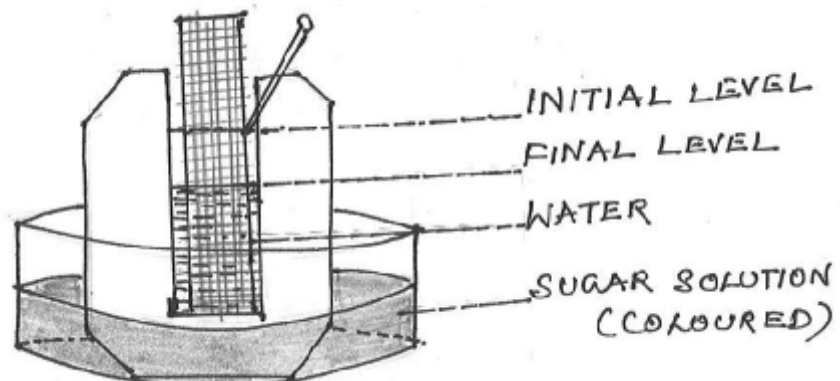
### CONTROL SETUP



### ENDOSMOSIS



### EXOSMOSIS



## **DETERMINATION OF OSMOTIC PRESSURE**

### **AIM**

To determine the osmotic pressure of the cell sap of Rheo leaf epidermal peeling by plasmolytic method.

### **REQUISITES**

Rheo leaf, 0.5 molar sugar solution, distilled water, petridishes, cover glasses, slides and microscope.

### **PROCEDURE**

0.5 molar sugar solution is prepared by dissolving 17.1 grams of sugar in 100ml of distilled water. From this different concentration of solutions such as 0.16, 0.18, 0.20, 0.22 and 0.24M were prepared. These solutions were poured in to the petridishes and they were labelled correctly. The epidermal peeling is separated from this leaf and are kept in each molar solution. After half an hour these epidermal peelings are mounted on the slides with their corresponding sugar solution.

### **OBSERVATION**

Epidermal peelings are examined under the microscope. The total number of cells and plasmolysed cells per field are counted and the results are tabulated.

Concentration of sugar solution (M)	Total no. of cells per field	No. of plasmolysed cells per field	% of plasmolysed cells.
0.16			
0.18			
0.20			
0.22			
0.24			

### INFERENCE

The cells in isotonic solution shows 50% plasmolysed cells indicating that their solution is isotonic with the cell sap. It is calculated by using the following formula.

$$\text{Osmotic pressure} = 22.4 \times M \times (T + t)/T.$$

M = Molar concentration,

T = Standard temperature,

t = Room temperature.

Osmotic pressure = ----- atm.

## **GANONG'S POTOMETER**

### **AIM**

To measure the rate of transpiration by using Ganong's potometer under two environmental conditions such as direct sunlight and diffused sunlight.

### **REQUISITES**

Ganong's potometer apparatus, leafy twig, one holed rubber cork, beaker/vial, water, vaseline, cotton and stand.

### **APPARATUS**

It consists of graduated capillary tube. The one end of the apparatus is dilated to form the shoot chamber in which leafy twig is introduced into the one holed rubber cork and the other end is bent with a small hole through which the air bubble is introduced. The reservoir is connected to the capillary tube with stop cock.

### **PROCEDURE**

The apparatus is filled with water without any air bubble. The leafy twig is introduced into the one holed rubber cork and is fitted with the shoot chamber. An air bubble is introduced into the capillary tube through the hole and the initial level is marked. The bent tube is immersed in a small beaker (vial) containing water. The apparatus is made air tight by applying vaseline. The set up is kept in the direct sunlight as well as in the diffused sunlight.

## OBSERVATION

The air bubble moves in the graduated capillary tube.

Source of light	Time (min)	Initial level (cm)	Final level (cm)	Distance travelled by the air bubble (cm)
Direct sunlight.				
Indirect sunlight				

## INFERENCE

Due to transpiration a vacuum and suction pressure is developed in the leafy twig. In order to fill this, water moves in the capillary tube along with the air bubbles. The distance travelled by the air bubble was more in the direct sun light than the diffused sunlight indicates that the rate of transpiration was found to be greater in the direct sunlight than the diffused sunlight.

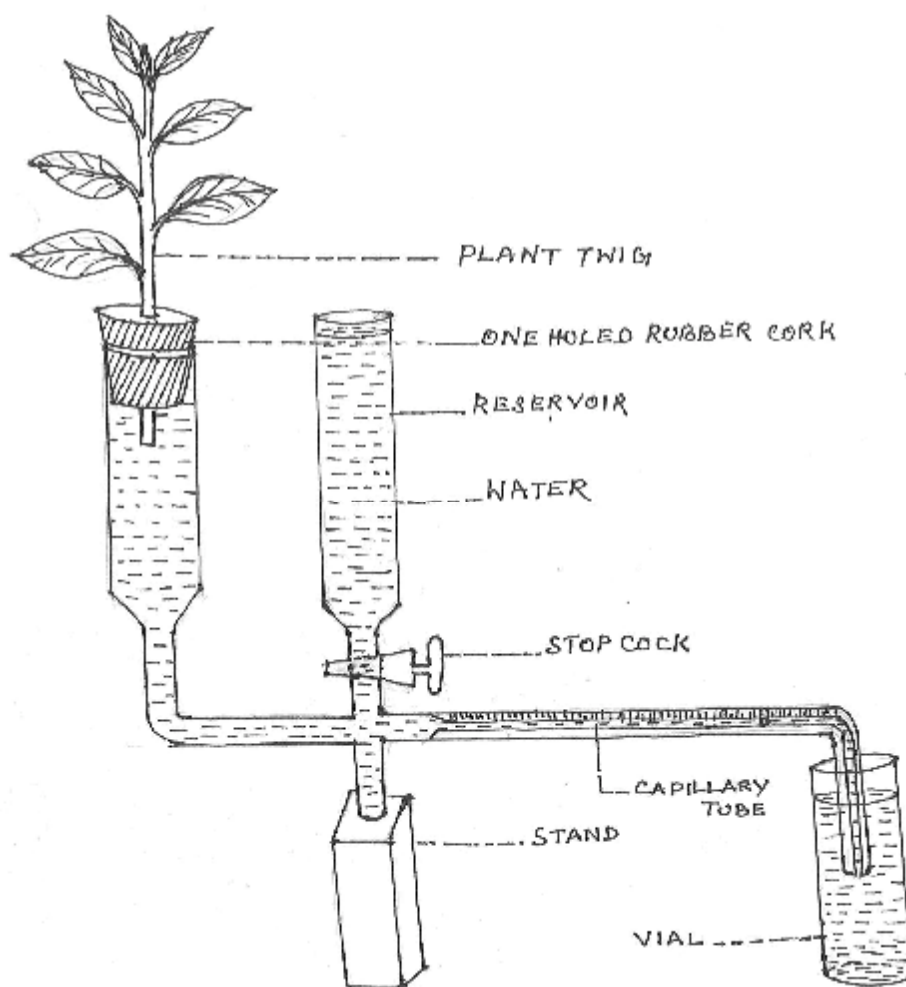
## RESULT

Distance travelled by air bubble in direct sunlight = ----- cm

Distance travelled by air bubble in diffused sunlight = ----- cm



## GANONG'S POTOMETER



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## **TRANSPIRATION ABSORPTION BALANCE**

### **AIM**

To show that the amount of water transpired is equal to the amount of water absorbed.

### **REQUISITES**

Transpiration absorption balance apparatus, one-holed rubber cork, rooted plant, water, and oil. The apparatus consists of a wide mouthed bottle which is connected to a graduated narrow side tube.

### **PROCEDURE**

The apparatus is filled with water. The rooted plant is introduced into the glass jar through one holed rubber cork. The initial level of water in the graduated side tube is noted. To prevent the evaporation of water in the side tube, a drop of oil is poured. The apparatus is kept in the sunlight for an hour.

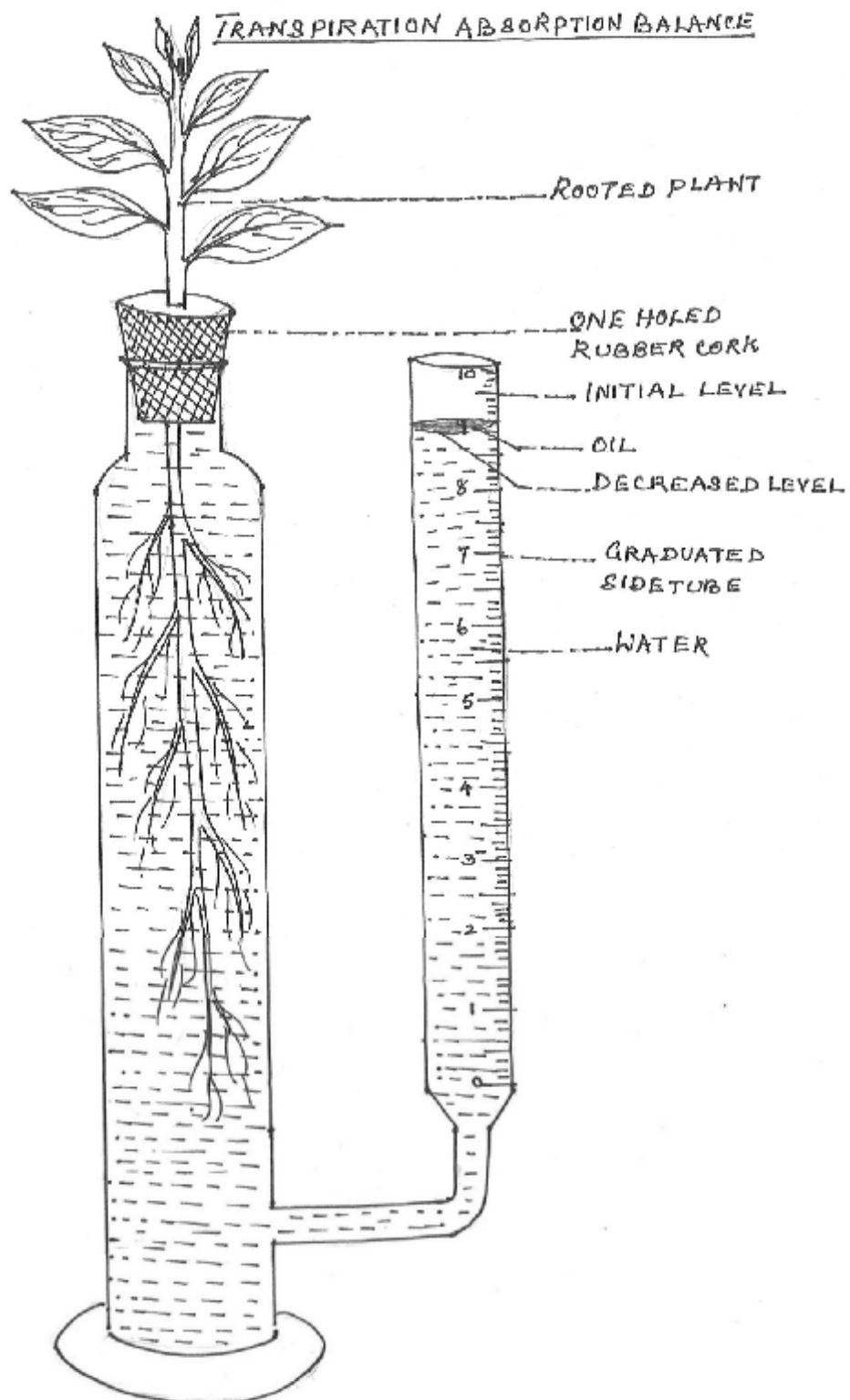
### **OBSERVATION**

The water level in the graduated side tube falls down.

<b>Time</b>	<b>Level of water in ml</b>		<b>Actual level in ml</b>
	<b>Initial</b>	<b>Final</b>	

### **INFERENCE**

The decrease in water level is due to the absorption of water by the rooted plant. It clearly indicates that the amount of water transpired is equal to the amount of water absorbed.



## **TRANSPIRATION INDEX**

### **AIM**

To find out the stomatal frequency and transpiration index of a leaf.

### **REQUISITES**

Potted plant, cobalt chloride solution, filter paper, two glass plates, two clips and vaseline.

### **PROCEDURE**

Filter paper strips are soaked in 3% cobalt chloride solution and dried in sunlight. The dried papers are blue in colour. But they form pink when wet. Filter paper strips were placed in the upper and lower surface of the leaf with the help of glass plates and clips. Vaseline is applied to prevent the evaporation of water through the slit between the leaf and glass plates. A third filter paper is kept on moist paper and the time taken for this to turn pink is noted. This gives the standard time.

### **OBSERVATION**

Time taken for the cobalt chloride paper to turn pink colour is noted and tabulated.

<b>Surface of the leaf</b>	<b>Time taken for cobalt chloride paper to change its colour</b>
Lower surface	
Upper surface	

Transpiration index of lower surface =

$$\frac{\text{Time Taken by the cobalt chloride paper to turn pink in lower surface}}{\text{Standard time}}$$

Transpiration index of upper surface =

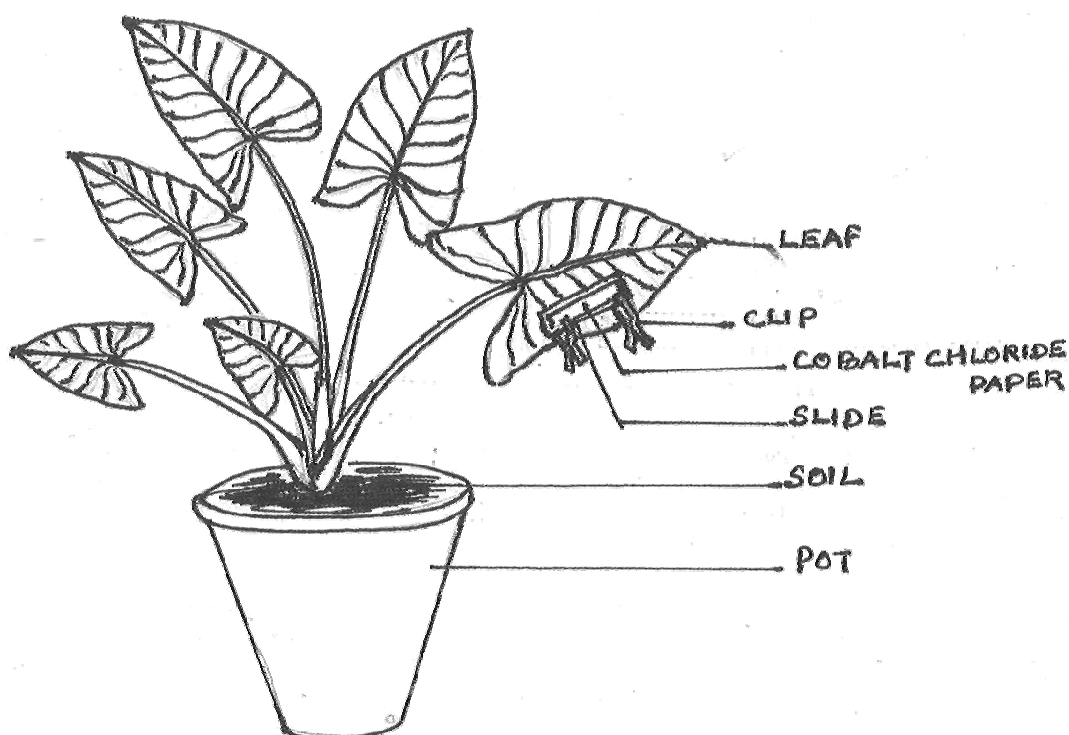
$$\frac{\text{Time Taken by the cobalt chloride paper to turn pink in upper surface}}{\text{Standard time}}$$

Transpiration index of lower surface =

Transpiration index of upper surface =

## INFERENCE

The rate of transpiration is found to be greater in the lower surface than the upper surface. The rate of transpiration is found to be greater in the lower surface is due to higher frequency of stomata. Transpiration index clearly indicates that the stomatal frequency is found to be greater in lower surface than the upper surface.



## **TRANSPIRATION PULL**

### **AIM**

To demonstrate the suction pressure developed during transpiration.

### **REQUISITES**

Transpiration twig, graduated glass tube with shoot chamber, one holed rubber cork, petridish, stand, water, mercury and vaseline.

### **PROCEDURE**

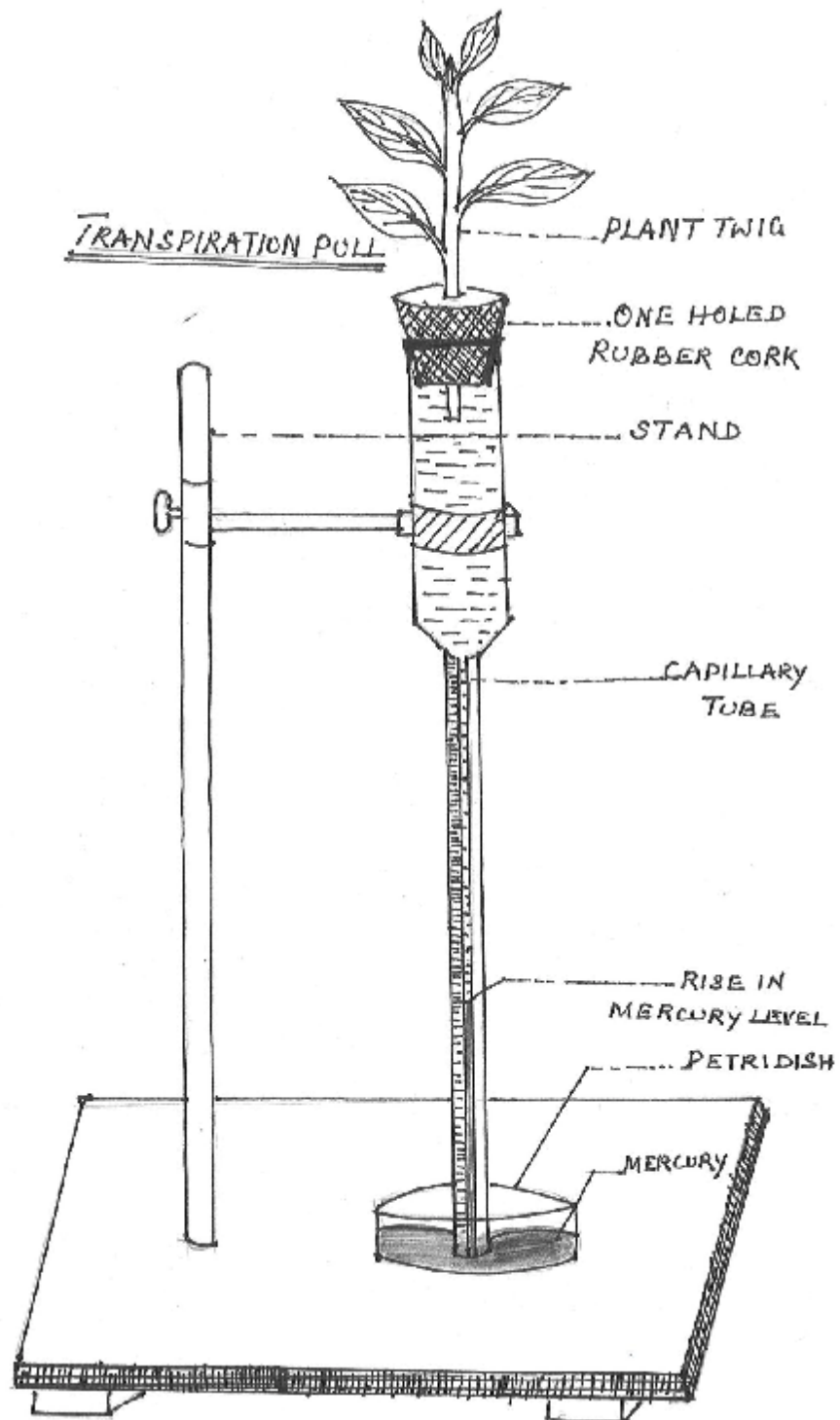
The apparatus is filled with water. A leafy twig is introduced into the shoot chamber through one holed rubber cork. The set up is made air tight. The lower end of graduated tube is immersed in a petridish of mercury.

### **OBSERVATION**

The mercury level rises in the graduated tube.

### **INFERENCE**

Due to transpiration (water loss), suction pressure is developed in the leaf cells. The pressure which lifts the water against the gravitational force is known as transpiration pull. It is so strong that it can lift not only the water but also the mercury.





## **EFFECT OF CARBON DI OXIDE CONCENTRATION ON THE RATE OF PHOTOSYNTHESIS**

### **AIM**

To show the effect of carbon dioxide concentration on the rate of photosynthesis by using test tube funnel experiment.

### **REQUISITES**

Beakers, funnels, test tubes, water, hydrilla twigs, different concentration of sodium bi carbonate (100, 200 and 300 mg).

### **PROCEDURE**

Hydrilla twigs are introduced into the mouth of the funnel and the funnel is inverted into the beaker containing water. The graduated test tube is filled with water and it is inverted over the stem of funnel. In the same way the other setups are made. The set up without sodium bi carbonate is used as control. In the other set ups, different concentration of sodium bi carbonate such as 100, 200 and 300 mg are added. All of them are kept in the direct sunlight.

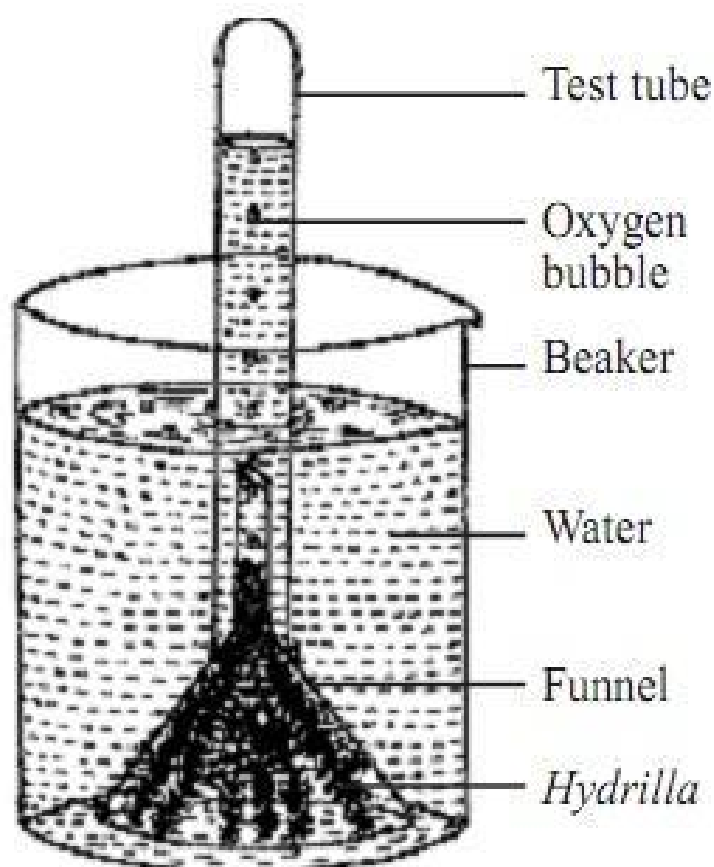
### **OBSERVATION**

Water level in all the set ups, decreases in the graduated test tube. It is due to the accumulation of oxygen which is evolved during photosynthesis.

Amount of $\text{NaHCO}_3$ (mg)	Level of water		Decrease in amount of water (ml)
	Initial (ml)	Final (ml)	
100			
200			
300			
Control			

### INFERENCE

The decreased level of water in test tube indicates the rate of photosynthesis. The rate of photosynthesis was found to be greater in 200 mg than to 100 and 300 mg.



## **EFFECT OF MONOCHROMATIC LIGHT ON THE RATE OF PHOTOSYNTHESIS**

### **AIM**

To show the effect of monochromatic light on the rate of photosynthesis by using test tube funnel experiment.

### **REQUISITES**

Beakers, funnels, test tubes, water, hydrilla twigs, a pinch of sodium bi carbonate, different colour papers (Red, Blue and Green).

### **PROCEDURE**

Hydrilla twigs are introduced into the mouth of the funnel and the funnel is inverted into the beaker containing water. The graduated test tube is filled with water and it is inverted over the stem of funnel. In the same way the other setups are made. A pinch of sodium bi carbonate is added to all the setups. They are covered with different coloured papers such as red, blue and green. One of the setup without colour paper is used as control. All of them are kept in the direct sunlight.

### **OBSERVATION**

Water level in all the set ups, decreases in the graduated test tube. It is due to the accumulation of oxygen which is evolved during photosynthesis.

<b>Monochromatic light used</b>	<b>Level of water</b>		<b>Decrease in amount of water (ml)</b>
	<b>Initial (ml)</b>	<b>Final (ml)</b>	
Control Red Blue Green			

### **INFERENCE**

Decreased level of water in test tubes indicates the rate of photosynthesis found to be greater in red light when compared to blue, green and control set ups. Minimum rate of photosynthesis was observed in green light.

## **WILMOTT'S BUBBLER**

### **AIM**

To determine the rate of photosynthesis by using Wilmott's bubbler.

### **REQUISITES**

Wilmott's bubbler apparatus, Hydrilla twigs, sodium bi carbonate, water. The apparatus is a wide mouthed bottle fitted with one holed rubber cork. Nozzle tube is inserted through the hole and the reservoir is fixed around the nozzle tube.

### **PROCEDURE**

The apparatus is filled with water. Hydrilla twigs introduced at the lower open end of the bubbler without any air bubbles. A pinch of sodium bi carbonate is added to enhance the supply of carbon di oxide. The reservoir is filled with water. The setup is made air tight with vaseline and is exposed to sun light.

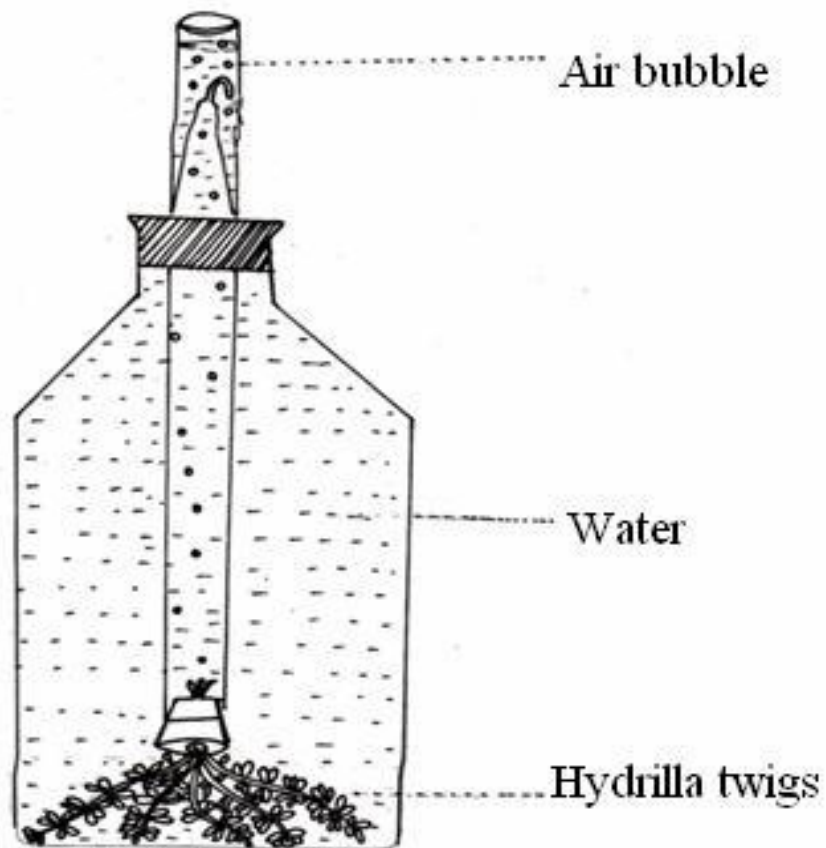
### **OBSERVATION**

The bubbles of gas (oxygen) escape from the nozzle. The number of bubbles evolved per minute is counted which is equal to the amount of oxygen liberated.

### **INFERENCE**

Since the hydrilla plant is a hydrophyte, it takes carbon dioxide from water for photosynthesis. The oxygen is evolved during the process escapes as bubbles. The rate of photosynthesis is calculated by counting the number of bubbles with in the given time.

## WILMOTT'S BUBBLER



## **PAPER CHROMATOGRAPHY**

### **AIM**

To separate the leaf pigments by paper chromatographic technique.

### **REQUISITES**

Fresh grass leaves, pestle and mortar, Whatmann filter paper no:1, glassjar, one holed rubber cork, micropipette,10 ml measuring jar, acetone and petroleum ether.

### **PROCEDURE**

Fresh grass leaves are homogenised in the pestle and mortar with equal amount of petroleum ether and acetone and the extract is prepared. A narrow strip of Whatmann paper no.1 is taken. Near the pointed end, the extract is spotted and it is called as loading spot. The spot is loaded again and again and air dried. The pointed tip of the whatmann paper is dipped in a glass jar containing solvent mixture of petroleum ether and acetone in the ratio of 9:1. The loading spot should not touch the solvent.

### **OBSERVATION**

As the solvent ascends the pigments are carried with the solvent. Depending upon the adsorption capacity, the components get separated and form distinct zones. The orange colour indicates the carotene which lies near the solvent front followed by yellow colour xanthophyll, bluish green chlorophyll-a and yellowish green chlorophyll-b. The solvent front (SF) is marked and the distance travelled by the pigment is measured by using this, RF value is calculated.

Rf value is calculated by using the formula.

$$\mathbf{Rf} = \frac{\text{Distance travelled by the pigment from the origin spot}}{\text{Distance travelled by the solvent from the origin spot}}$$

**S.F.** =

Rf value of carotene =

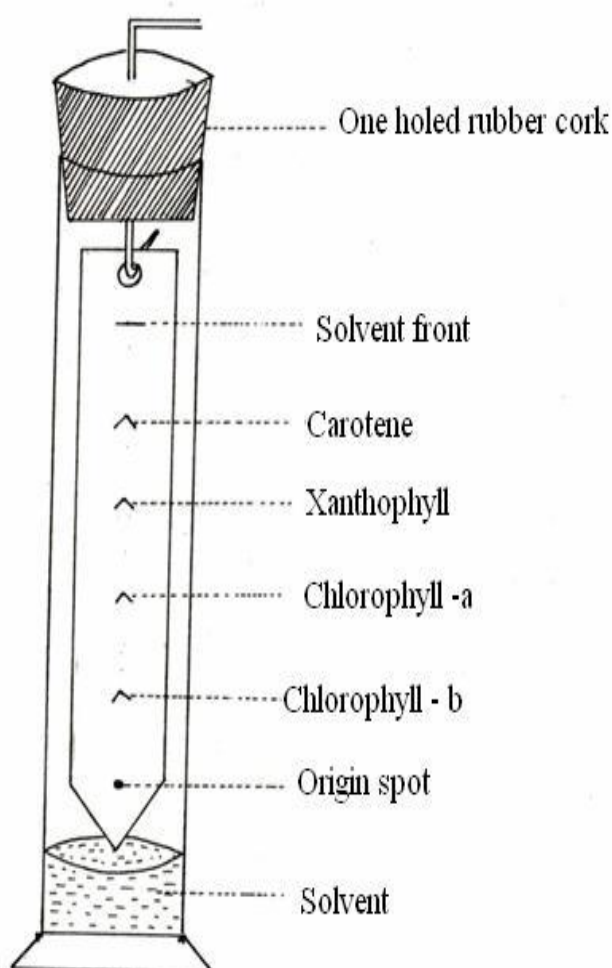
Rf value of xanthophyll =

Rf value of chlorophyll a =

Rf value of chlorophyll b =

### INFERENCE

Rf value indicates the differential solubility and differential adsorption characteristic of the leaf pigments.





## **THIN LAYER CHROMATOGRAPHY**

### **AIM**

To separate the leaf pigments by thin layer chromatographic technique.

### **REQUISITES**

Fresh grass leaves, pestle and mortar, chloroform, methanol, micropipette, 10 ml measuring jar, acetone and petroleum ether, slides and coupling jar.

### **PROCEDURE**

Fresh grass leaves are homogenised in the pestle and mortar with equal amount of petroleum ether and acetone and the extract is prepared. A uniform suspension of silica gel is prepared by dissolving it in a mixture of chloroform and methanol in the ratio of 2:1. The suspension is stirred well with glass rod. Two slides are dipped into the suspension and slowly drawn out and drained to the excess. The slides are completely dried. The extract is spotted towards the corner of the slide leaving space from the edge. Spot is loaded again and again and dried. The slides are kept immersed in a coupling jar containing a solvent mixture of petroleum ether and acetone in the ratio of 4:1. The setup is kept undisturbed.

### **OBSERVATION**

As the solvent ascends the pigments are carried with the solvent. Depending upon the adsorption capacity, the components get separated and form distinct zones. The orange colour indicates the carotene which lies near the solvent front followed by yellow colour xanthophylls, bluish green chlorophyll-a and yellowish green

chlorophyll-b. The solvent front (SF) is marked and the distance traveled by the pigment is measured by using this, RF value is calculated.

Rf value is calculated by using the formula.

$$R_f = \frac{\text{Distance travelled by the pigment from the origin spot}}{\text{Distance travelled by the solvent from the origin spot}}$$

S.F. =

Rf value of carotene =

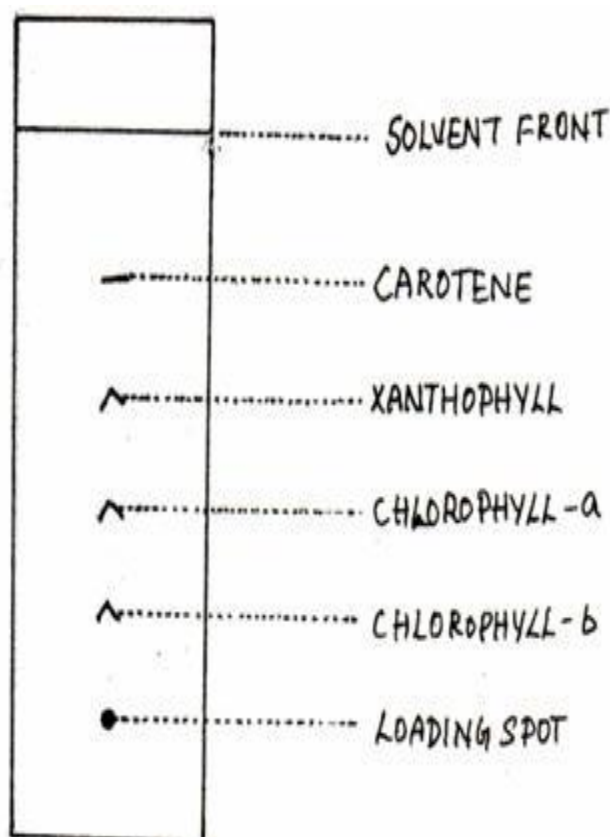
Rf value of xanthophyll =

Rf value of chlorophyll a =

Rf value of chlorophyll b =

## INFERENCE

Rf value indicates the differential solubility and differential adsorption characteristic of the leaf pigments.



## **GANONG'S RESPIROSCOPE**

### **AIM**

To prove that carbon dioxide is released during respiration by using Ganong's respiroscope.

### **REQUISITES**

Ganong's respirosopes, pyrogallol solution, potassium hydroxide solution, water, stand, beaker, flower buds and cotton.

### **PROCEDURE**

Equal amount of flower buds are taken in the bulbs of respirosopes. A piece of cotton is introduced into the bulb to prevent the flow of buds into the beaker. The open ends of the bulbs are kept immersed in a beaker containing water, pyrogallol and potassium hydroxide solution respectively.

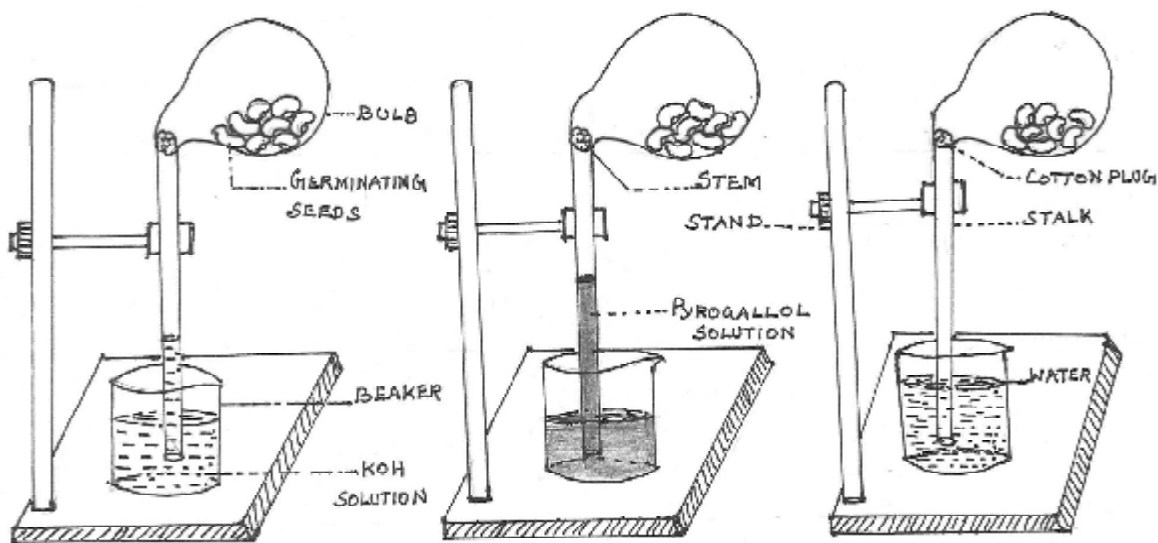
### **OBSERVATION**

There is no increase in the level of respiroscope with water. Potassium hydroxide solution and pyrogallol solution rises up in their corresponding respirosopes.

### **INFERENCE**

In the first setup the pyrogallol solution rise immediately in the respiroscope because pyrogallol solution absorbs the oxygen in the respiroscope (Here there is no respiration). In the second setup the potassium hydroxide solution rises in the respiroscope. It is because

of the respiration of flower buds. During respiration, carbon dioxide is released which is absorbed by potassium hydroxide solution. As a result the level of potassium hydroxide (KOH) rises in the respiroscope. In the third setup there is no change in the level of water because the rate of exchange of gases is equal.



## **DEMONSTRATION OF ANAEROBIC RESPIRATION**

### **AIM**

To determine respiration in the absence of O<sub>2</sub>.

### **REQUISITES**

Mercury, petridish, small test tube, a few germinating seeds.

### **PROCEDURE**

The test tube filled with mercury is inverted over a petridish containing mercury. The germinating seeds are introduced into the test tube. The seed coat should be removed because it is impermeable to gases. It may prevent the evaluation of CO<sub>2</sub> from coming out.

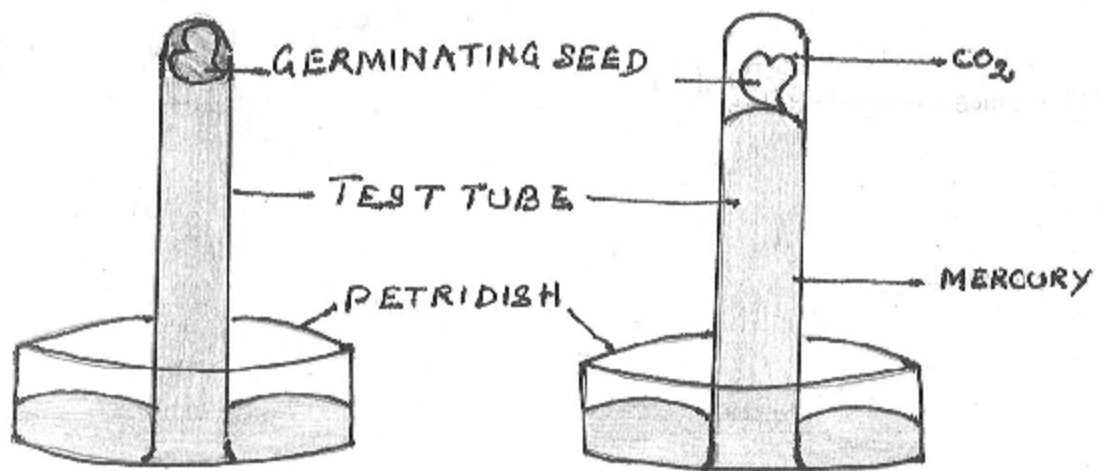
### **OBSERVATION**

The mercury level in the test tube falls down due to the accumulation of CO<sub>2</sub>.

### **INFERENCE**

In the absence of O<sub>2</sub> the seed respire and CO<sub>2</sub> is liberated. Anaerobic respiration is exhibited by germinating seed.

## ANAEROBIC RESPIRATION



## **KUHNE'S BULB**

### **AIM**

To demonstrate alcoholic fermentation of sugar by microorganisms and anaerobic respiration.

### **REQUISITES**

Kuhne's bulb, water, sugar, yeast powder, cover slip and pellets of potassium hydroxide. The apparatus consists of a vertical tube and a bulb which are connected to each other.

### **PROCEDURE**

The Kuhne's bulb is filled with sugar solution without any air bubbles. Thus anaerobic condition is provided. The yeast powder is added. The sugar solution is mixed well. The bulb is closed by the cover slip.

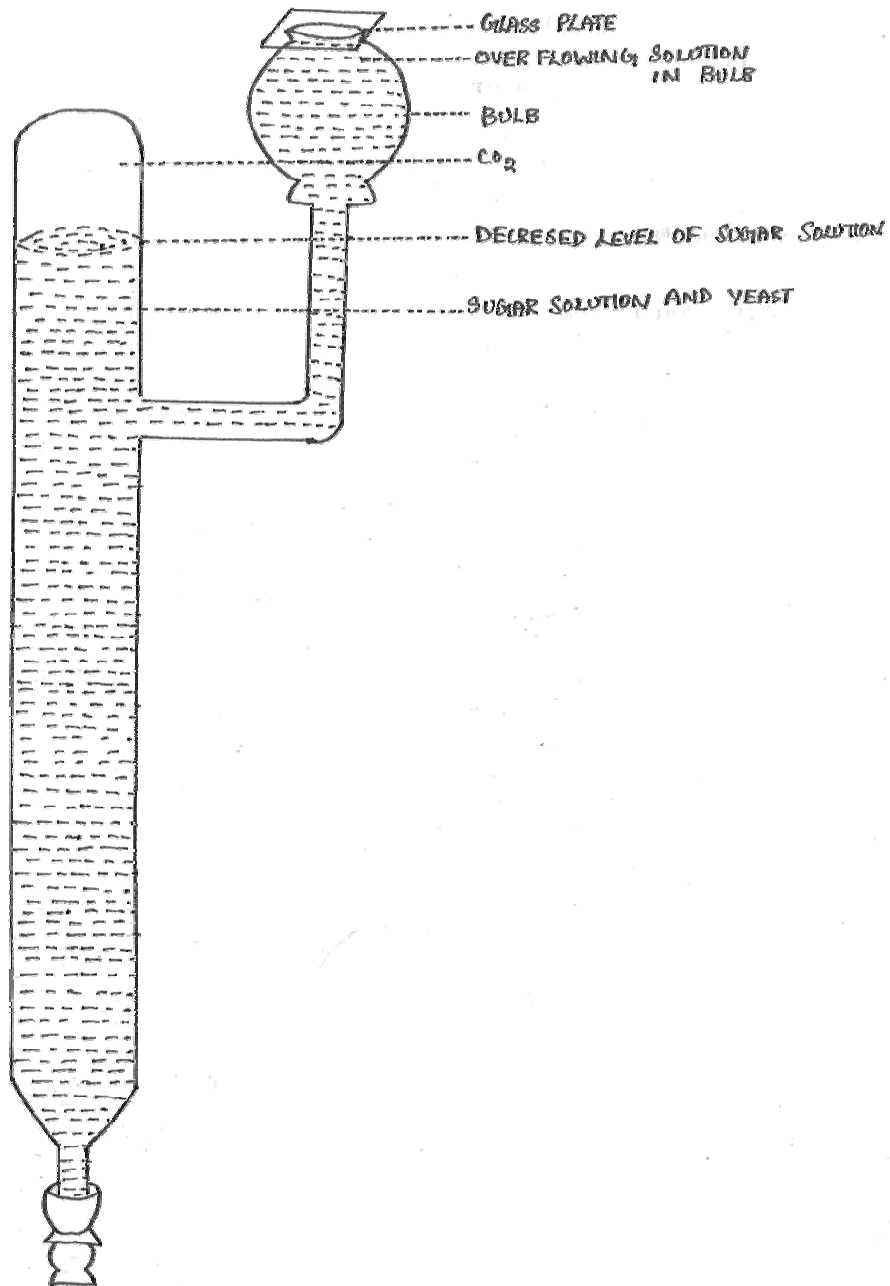
### **OBSERVATION**

Bubbles of gas get collected in the tube pushing the solution down. So, the level of the solution in the vertical tube falls down. Alcoholic smell is emitted by the solution. When the pellets of potassium hydroxide is added the solution again rises indicating the presence of carbon dioxide.

### **INFERENCE**

Thus under anaerobic condition sugar is incompletely oxidized to form alcohol and carbon dioxide by yeast. This process of incomplete oxidation of sugar by yeast is called alcoholic fermentation.

## KUHNE'S BULB





## **ARC AUXANOMETER**

### **AIM**

To measure the rate of growth of a plant using arc auxanometer.

### **REQUISITES**

Arc auxanometer, potted plant, thread and weight. Arc auxanometer consists of a long pointer or lever fixed to the axis of a small pulley. The free end of pointer moves on the graduated arc scale.

### **PROCEDURE**

The thread is tied to the tip of the stem of the plant and the thread is passed over the pulley in the arc auxanometer. A small weight is tied to the free end of the thread and it is kept in tension. The pointer is kept in zero

### **OBSERVATION**

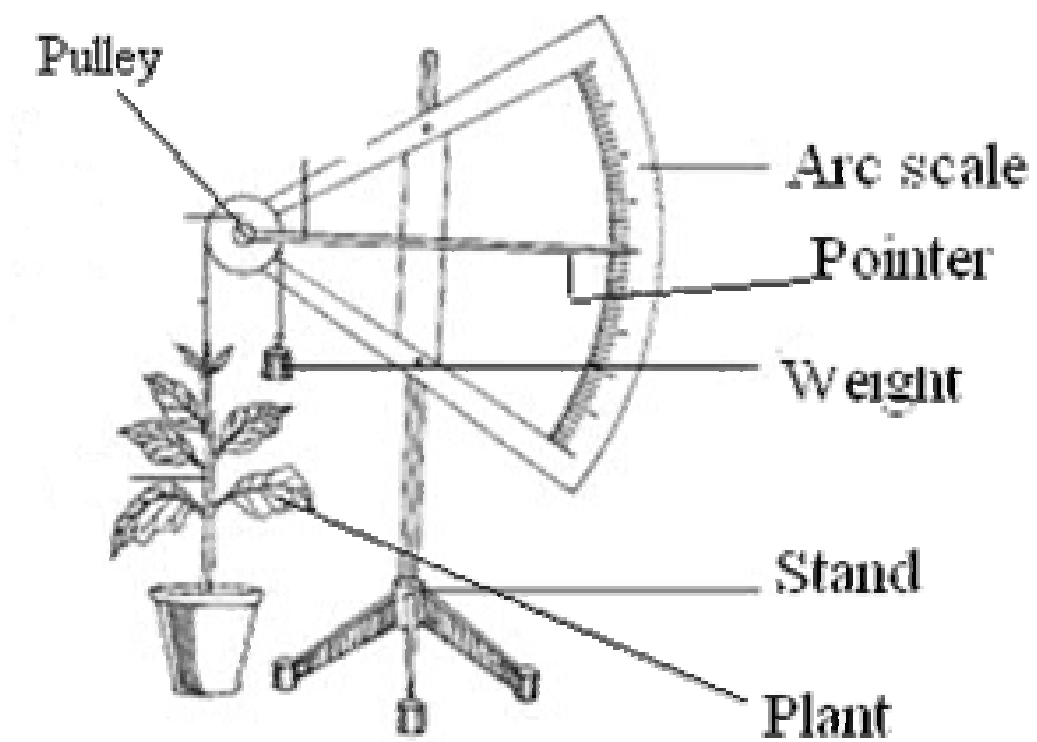
The pointer moves on the scale.

### **INFERENCE**

The stem tip grows in length. This turns the pulley and pushes the weight down. So, the pointer moves over the scale. The rate of growth is measured.

$$\text{The actual rate of growth} = \frac{\text{Number of divisions moved}}{\text{Magnitude of the divisions of scale}}$$

## ARC AUXANOMETER



## CLINOSTAT

### AIM

To show that the negative geotropism of shoot is due to unilateral effect of gravity.

### REQUISITES

Clinostat, water, soil and seedling.

### PROCEDURE

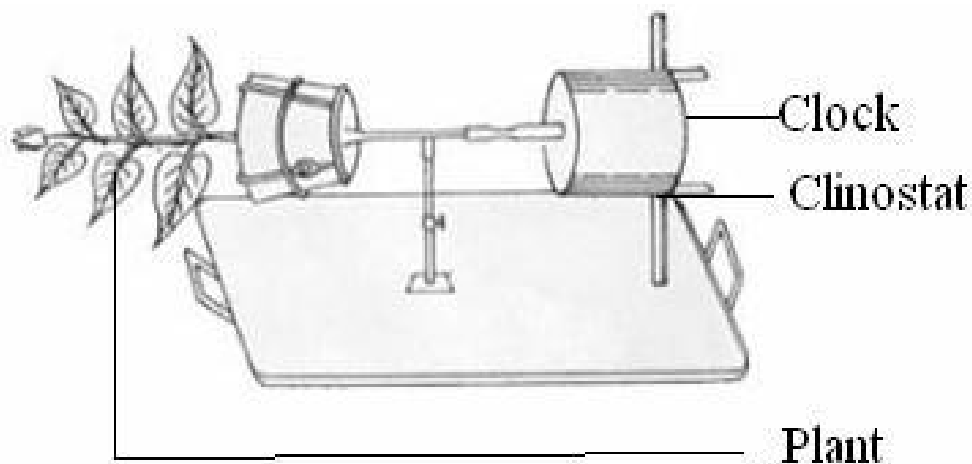
Clinostat consists of a clock and a metallic pot in which plant is fixed. (When the clock is operated the metallic pot rotates slowly)

### OBSERVATION

The plants grow horizontally without producing a geotropic curvature of the stem tip when the clock works. But when the clock work is stopped; the stem tip is turned upwards.

### INFERENCE

Rotation of plants nullifies the unilateral effects of gravity that felt on all sides. So the negative geotropic curvature is not seen. But when the clock work stops, unilateral effects of gravity causes geotropic curvature.



## ESTIMATION OF SUGAR

### AIM

To estimate the sugar content of the given sample of orange fruit by colorimetric method.

### REQUISITES

Orange fruit, pestle and mortar, distilled water, measuring cylinder, muslin cloth, funnel, beaker and Benedict's reagent and standard graph.

### PROCEDURE

50 mg of fleshy tissue of orange was homogenized with 4ml of distilled water. The extract was filtered and boiled in hot water bath for 10 minutes. After 10 minutes, it was cooled and after cooling 1ml of Benedict's reagent was added. The mixture was boiled in water bath again for 5 minutes and cooled. The optical density (O.D) was measured at 540 nm.

### OBSERVATION

The optical density of the sample at 540 nm is tabulated.

S. No.	Sample	Optical Density
1.	Orange	

When the optical density is -----, the concentration of the sugar is ----- as it is in the standard graph.

The concentration of the sugar is ----- mg.

So 50 mg of orange fruit tissue contain ----- mg of sugar.

## **ESTIMATION OF CHLOROPHYLL**

### **AIM**

To estimate the chlorophyll content of given sample.

### **PRINCIPLE**

Chlorophyll is extracted in 80% acetone and the absorption of solution at 645 and 663 nm are read in a spectrophotometer. Using the absorption co-efficient the amount of chlorophyll is calculated.

### **REQUISITES**

Leaf, mortar and pestle, 80% acetone, centrifuge, cuvette and colorimeter.

### **PROCEDURE**

Weigh 1 gm of finely cut and well mixed sample of leaf. Transfer it into a clean mortar. Grind the tissue to fine pulp with 20ml of 80% acetone. Centrifuge (5000 rpm) for 5 minutes and transfer the supernatant to 100ml beaker. Grind the residue with 200ml of 80% acetone. Centrifuge and transfer the supernatant to the beaker. Repeat this procedure until the residue is colourless and make up the volume to 100ml with 80% acetone. Read the absorption of the solution at 645 and 663 nm against the solvent (80% acetone) or blank.

### **CALCULATION**

Calculate the amount of chlorophyll present in the extract as mg chlorophyll gm tissue using the following formula.

$$\text{Chlorophyll a / gm tissue} = 12.7(\text{O.D at 663}) - 2.96(\text{O.D at 645}) \times V/1000 \times W$$

$$\text{Chlorophyll b / gm tissue} = 22.9(\text{O.D at 645}) - 4.68(\text{O.D at 663}) \times V/1000 \times W$$

$$\text{Total chlorophyll/ gm tissue} = 20.5(\text{O.D at 645}) + 8.02(\text{O.D at 663}) \times V/1000 \times W$$

Where,

V = final volume of chlorophyll extract in 80% acetone.

W = Fresh weight of tissue extracted.

$$\text{O.D at 645 nm} = \text{----- O.D at 663 nm} = \text{-----}$$

### **INFERENCE**

$$\text{Chlorophyll a / gm tissue} = 12.7(\text{O.D at 663}) - 2.96(\text{O.D at 645}) \times V/1000 \times W$$

$$= \text{-----mg/gm tissue}$$

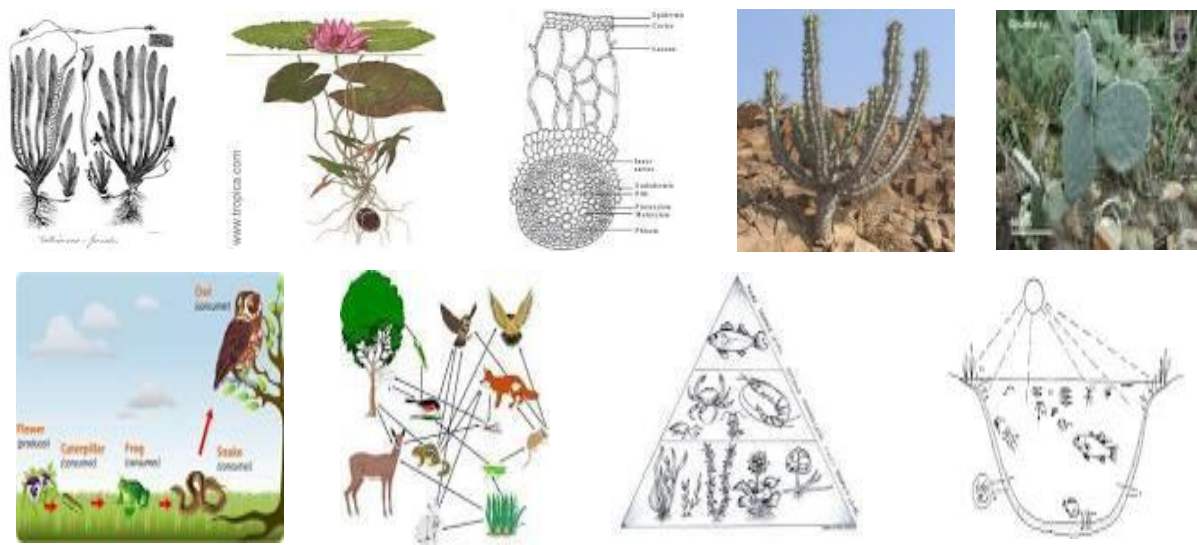
$$\text{Chlorophyll b / gm tissue} = 22.9(\text{O.D at 645}) - 4.68(\text{O.D at 663}) \times V/1000 \times W$$

$$= \text{-----mg/gm tissue}$$

$$\text{Total chlorophyll/ gm tissue} = 20.5(\text{O.D at 645}) + 8.02(\text{O.D at 663}) \times V/1000 \times W$$

$$= \text{-----mg/gm tissue}$$





## ENVIRONMENT, BIODIVERSITY AND CONSERVATION







## SUBMERGED HYDROPHYTE

### *HYDRILLA*

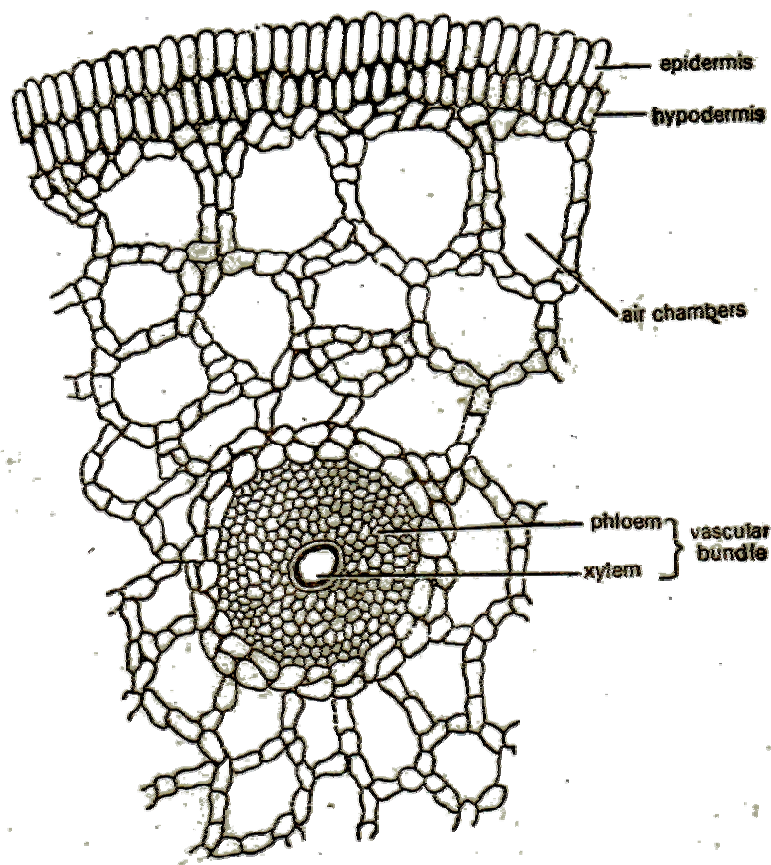
#### HABIT

- *Hydrilla* is a submerged aquatic plant.
- Stem is soft, slender and delicate
- The leaves are small, flimsy and are borne in whorls at the node.



## T.S. OF STEM

- The outermost tissue is epidermis.
- The cells are thin walled and elongated. The cells contain chloroplast.
- There is no cuticle.
- The cells of the hypodermis are thin walled, parenchymatous, compactly arranged.
- Just behind the hypodermis, cortex is aerenchymatous.
- Distinct endodermis and the xylem are represented by xylem cavities with non lignified cell walls.
- The xylem is surrounded by the delicate phloem cells.

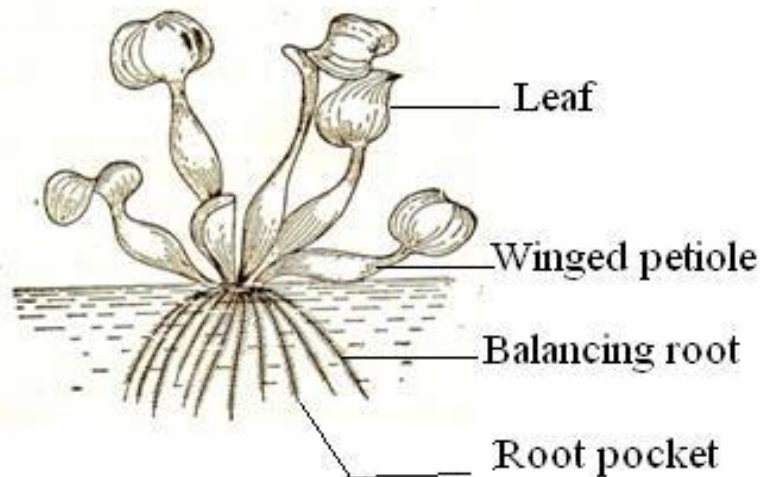


## FLOATING HYDROPHYTE

### *EICHHORNIA*

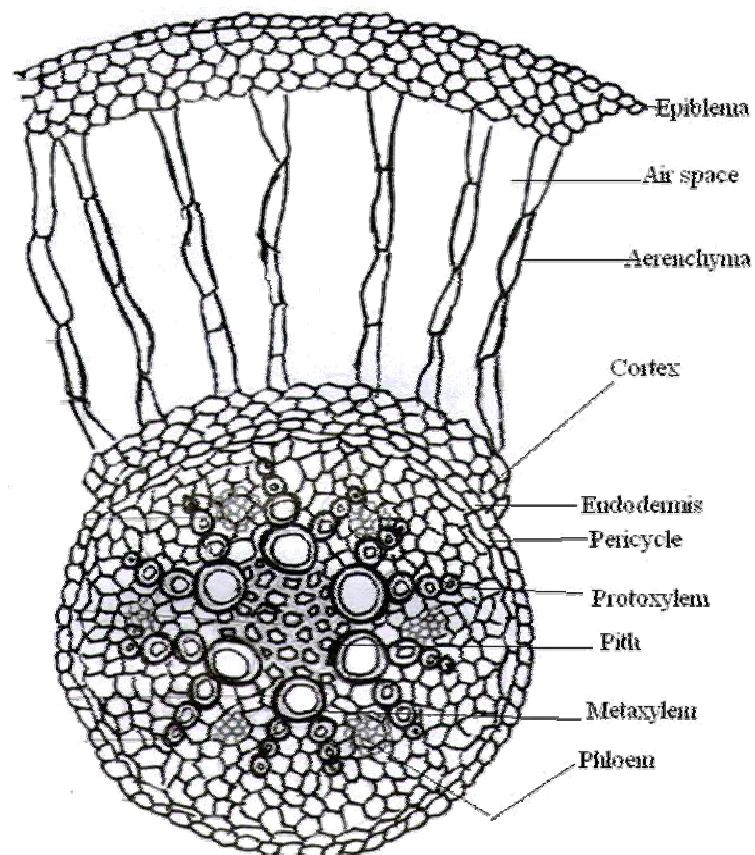
#### HABIT

- *Eichhornia* is a free floating hydrophyte.
- The stem is an offset which gives rise to cluster of leaves.
- Each leaf has a swollen petiole which is filled with air.
- The lamina is broad.
- The root system is extensively developed with numerous roots.
- Each root has a root pocket at its end.



## T.S OF ROOT

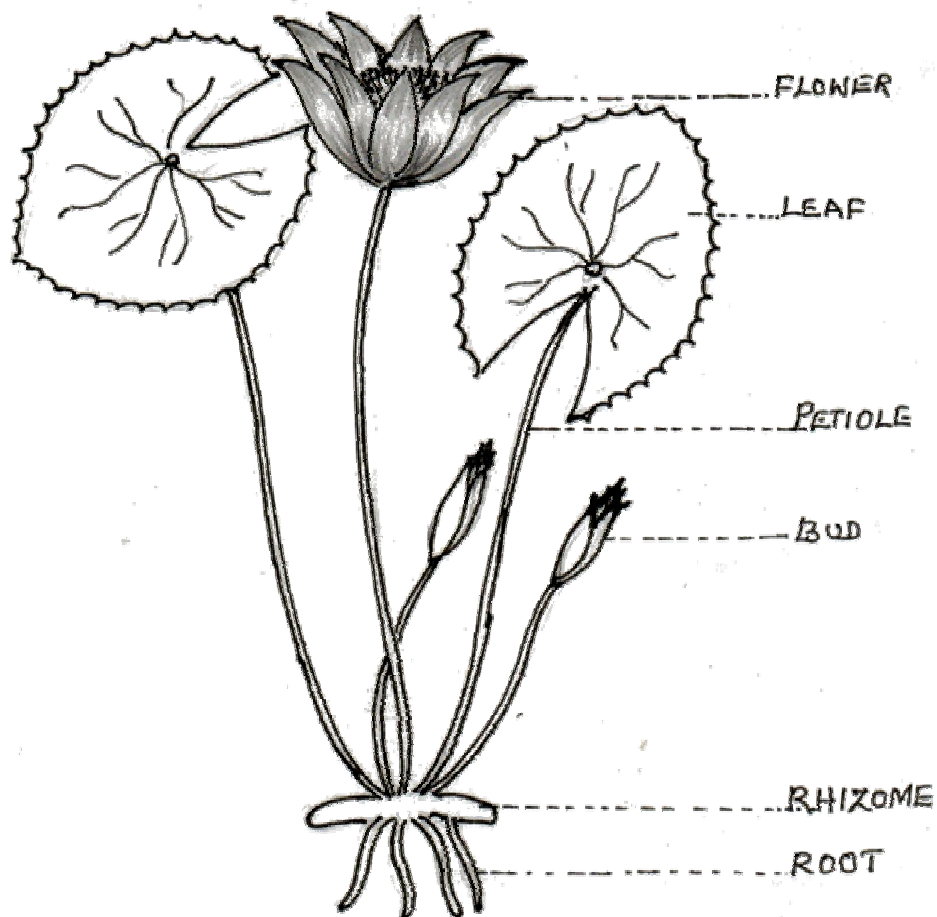
- The transverse section of *Eichhornia* root shows an outer epidermis which is non-cuticularised.
- The outer cortex is made up of compactly arranged thin walled parenchymatous cells.
- The middle cortex is composed of loosely arranged strands of aerenchyma separating long narrow chamber.
- The inner cortex is made up of regularly arranged cells. Single layered endodermis and pericycle enclose the xylem strands, alternating with phloem patches.
- Vascular bundle is radial, closed with exarch xylem.



## FLOATING BUT ROOTED HYDROPHYTE

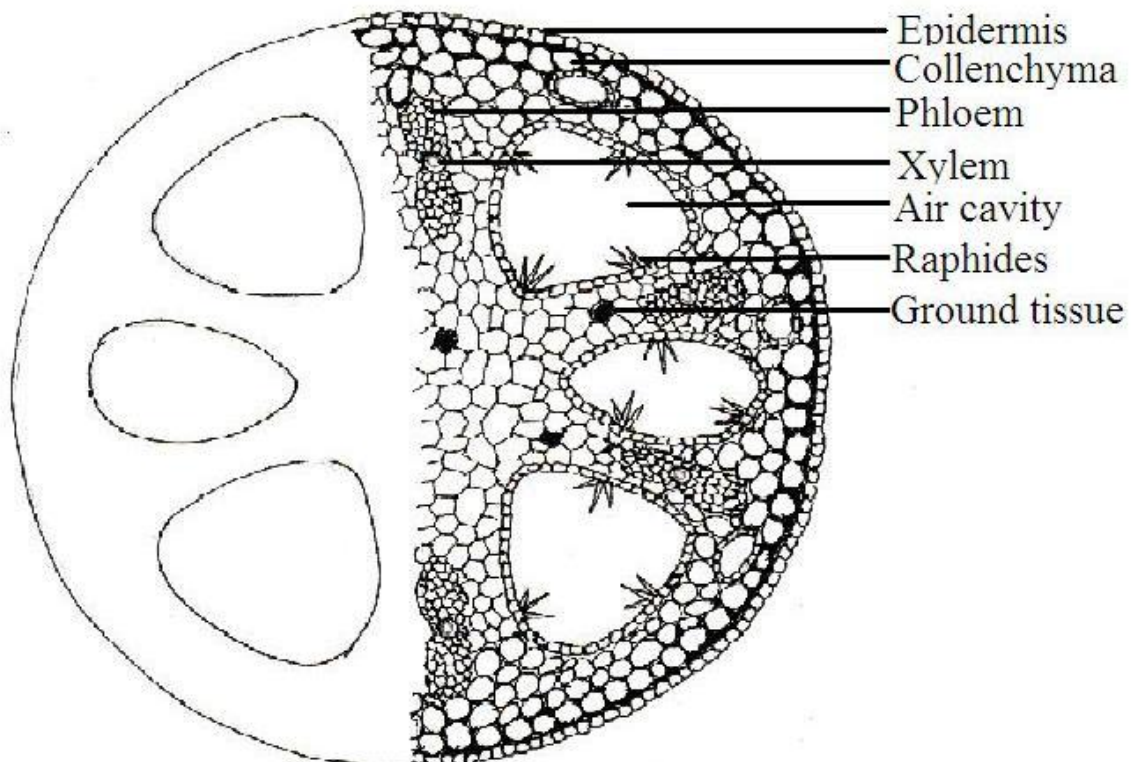
### *NYMPHAEA*

- The parts of the plant- leaves and flower- float on the surface of water.
- Remaining parts attached to the shallow bottom of the reservoir.



## T.S. OF PETIOLE

- *Nymphaea* is a floating but rooted aquatic plant.
- Presence of waxy layer above the epidermis.
- Presence of large air spaces.
- Reduction in mechanical tissue.
- Poorly developed vascular tissue.
- Mechanical strength is provided by raphides.
- Xylem represented by small cavities.

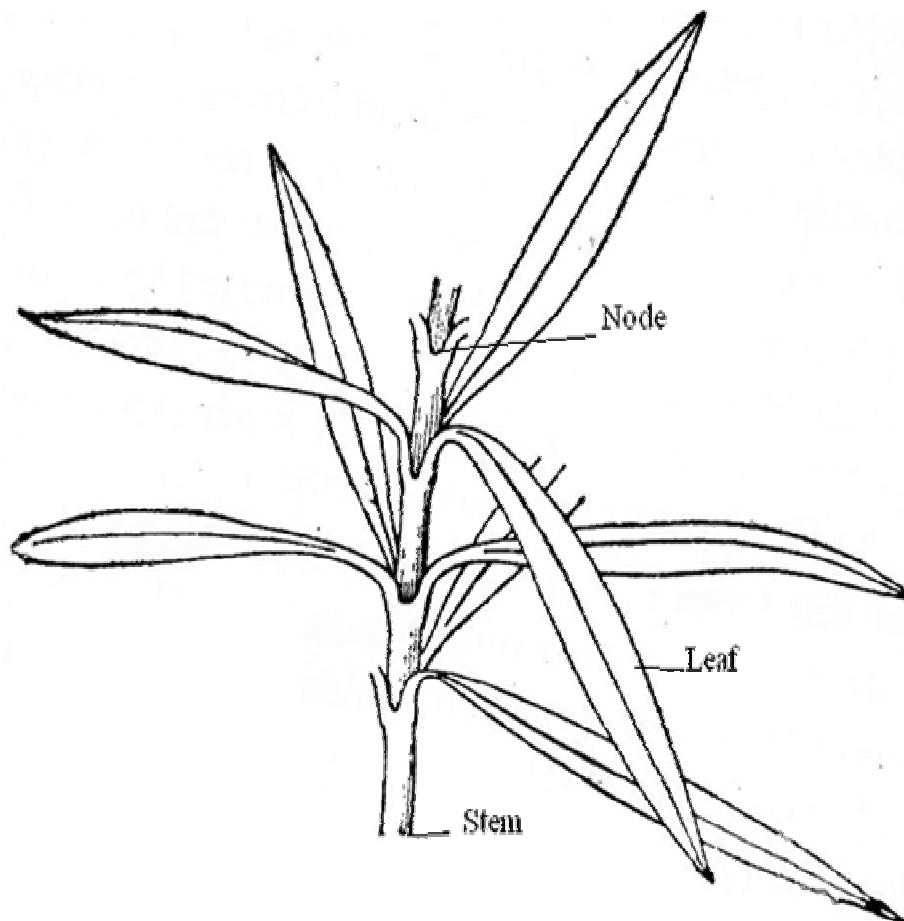


## XEROPHYTE

### *NERIUM*

#### HABIT

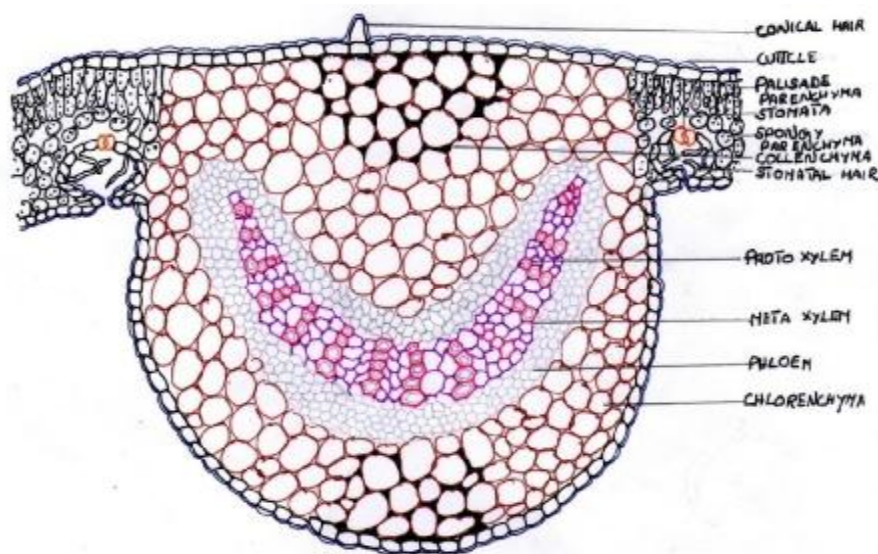
- *Nerium* shows xerophytic adaptations like shiny thick leaves which are vertically oriented.
- The phyllotaxy is ternate.





## T.S OF LEAF

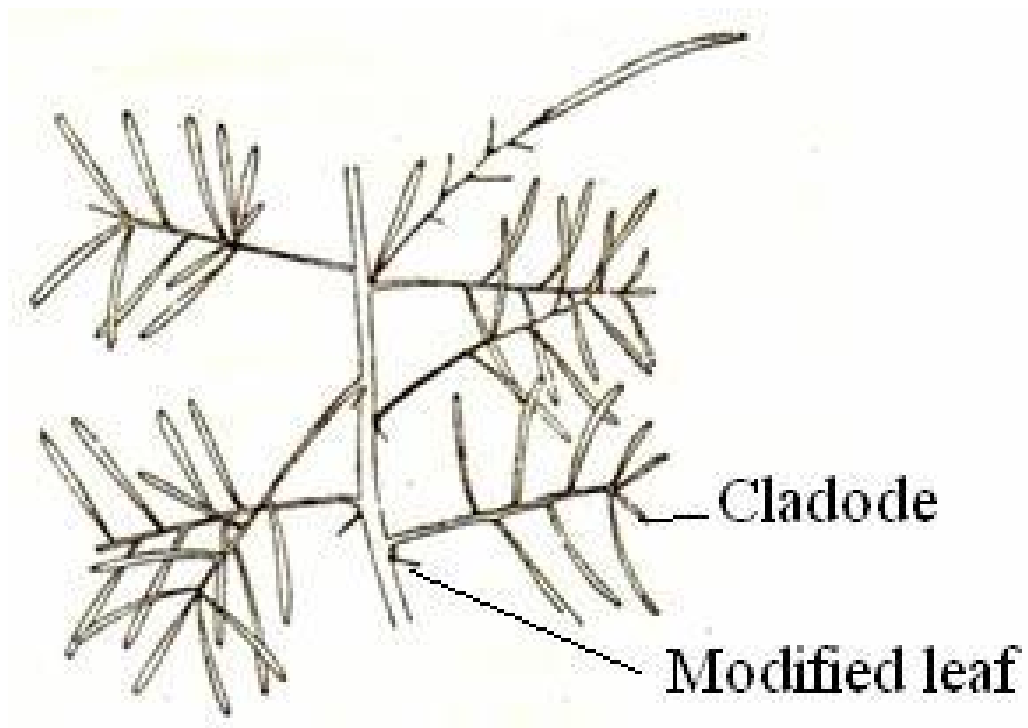
- Transverse section of *Nerium* leaf shows upper and lower epidermal layers.
- Cuticle is thick, sunken stomata situated in the stomatal pits on the lower epidermis.
- Dense hairs cover the stomata in depression.
- Mesophyll is differentiated into palisade and spongy parenchyma.
- The palisade tissue is arranged in 2 to 3 layers below the upper epidermis and lower epidermis which contain numerous chloroplasts.
- Spongy parenchyma is located between palisade of lower and upper epidermal cells.
- Calcium oxalate crystals are found throughout the mesophyll.
- The vascular bundle in the midrib is larger in size than those in the wings.
- Bundles in the wings are arranged almost in a parallel series.
- Each vascular bundle has xylem and phloem which is surrounded by a parenchymatous sheath.



## ***ASPARAGUS***

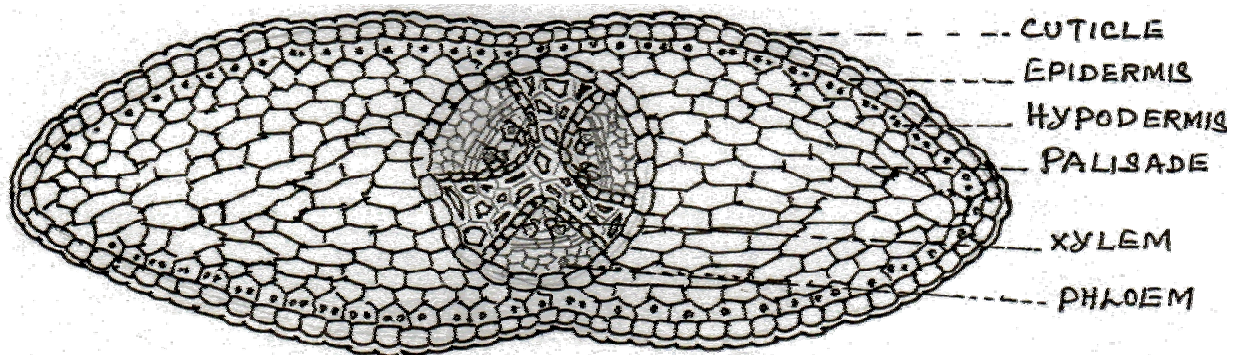
### **HABIT**

- In *Asparagus* the leaves are partly modified into scale and partly into spines.
- In the axils of leaves flat green cladodes are produced.
- They have green flattened stem which perform the functions of the leaf and hence the name cladode.



## T.S OF CLADODE

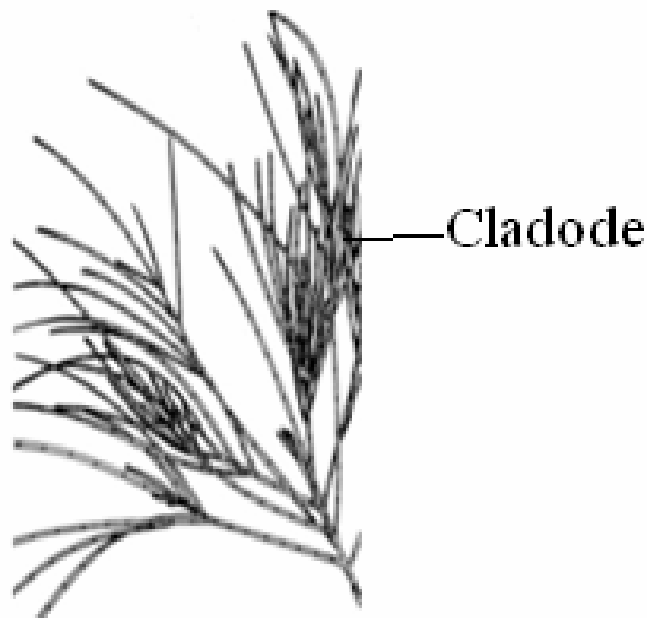
- T.S of cladode of *Asparagus* shows outermost single layered epidermis, covered with cuticle.
- Next layer is the palisade layer with chloroplast.
- The vascular bundle is located in the centre with three groups of xylem and phloem.
- In between the vascular bundles are palisade layer consisting of elongated parenchyma cells, which are water storage in function.
- Thick cuticle and water storage tissues are the xerophytic adaptations.



## **CASUARINA**

### **HABIT**

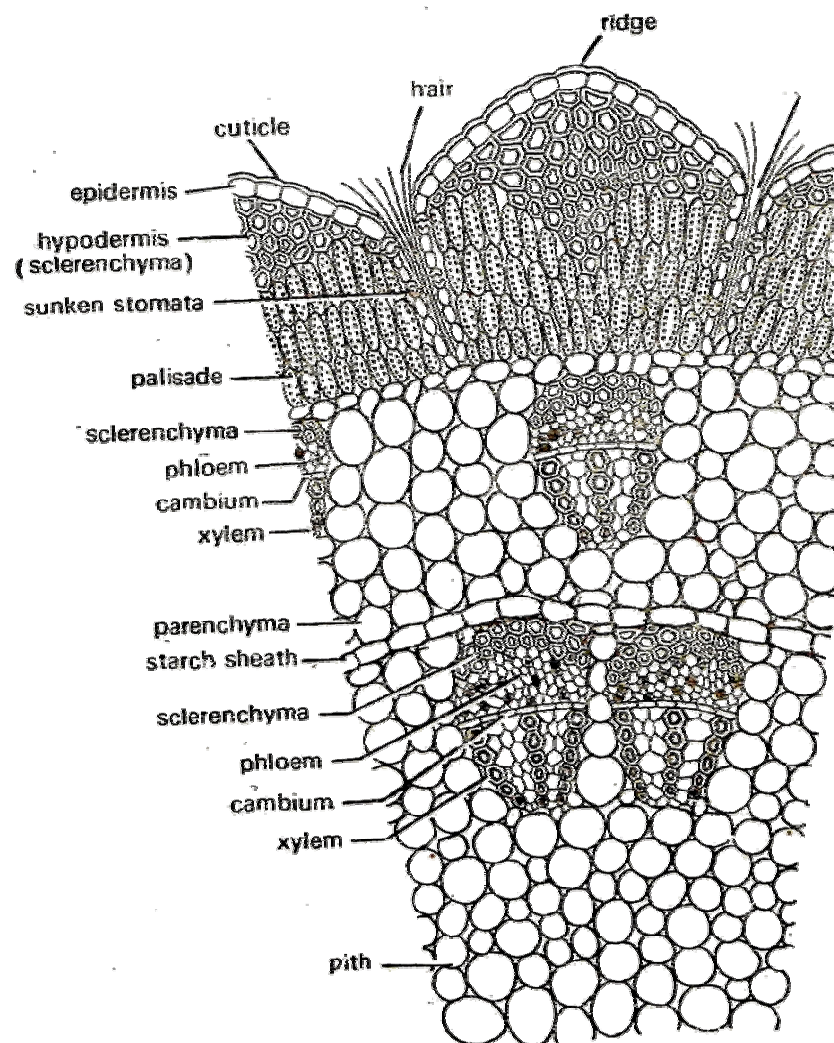
- *Casuarina* is a highly branched cylindrical stem with whorls of branches arising at the nodes of the stem.
- The leaves are reduced into scale leaves and the function of leaf is similar to the stem and its branches.
- The internodes are long and grooved.



### **T.S OF CLADODE**

- The stem of *Casuarina* has got an irregular outline showing the presence of ridges and furrows.
- The outer epidermal layer is cuticularised.
- The sunken stomata are confined to the grooves.
- Some of the epidermal cells grow out into hairs.
- The hypodermis is sclerenchymatous and it is developed very well along the ridges.

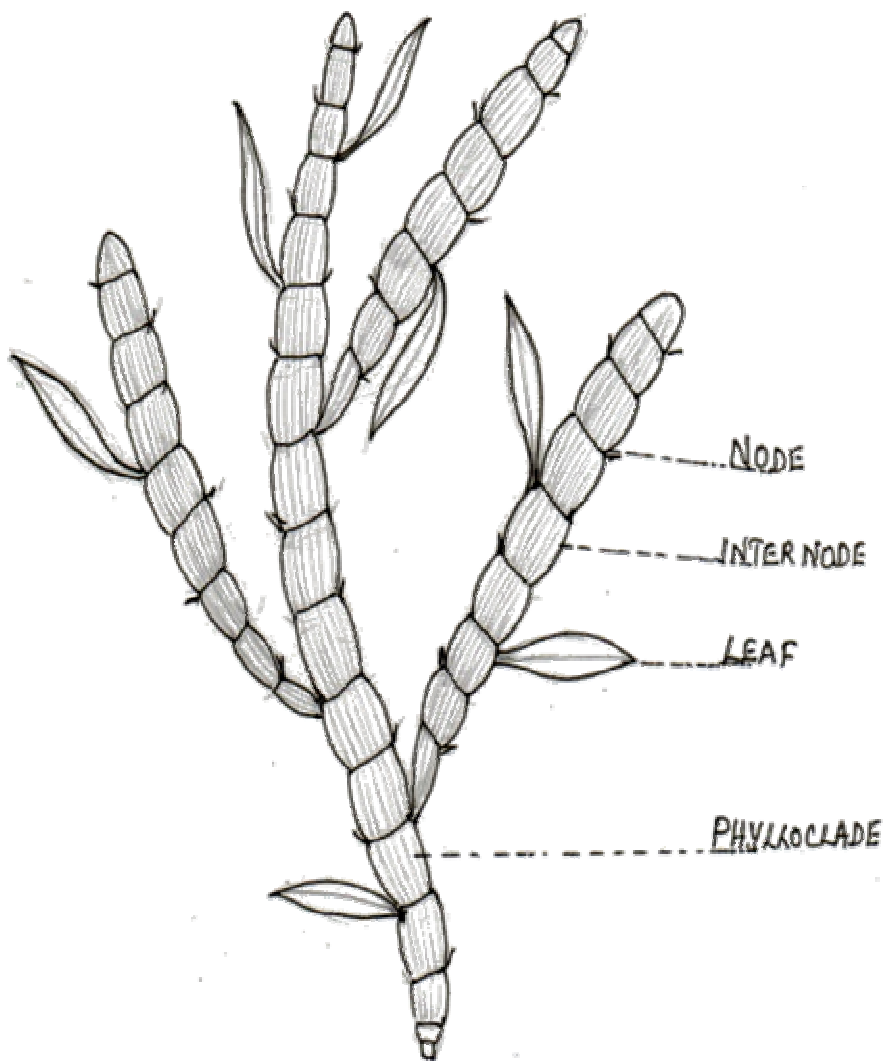
- Two or three layers of palisade like parenchyma cells lie beneath the sclerenchymatous region of the ridges.
- The rest of the cortex is parenchymatous.
- Cortical bundles are present in the cortical region.
- The vascular bundles which surround the pith region and open, endarch and collateral.
- They have sclerenchymatous cap.
- The medullary rays are seen in between the bundles.
- The pith is solid and parenchymatous.



## ***MUEHLENBECKIA***

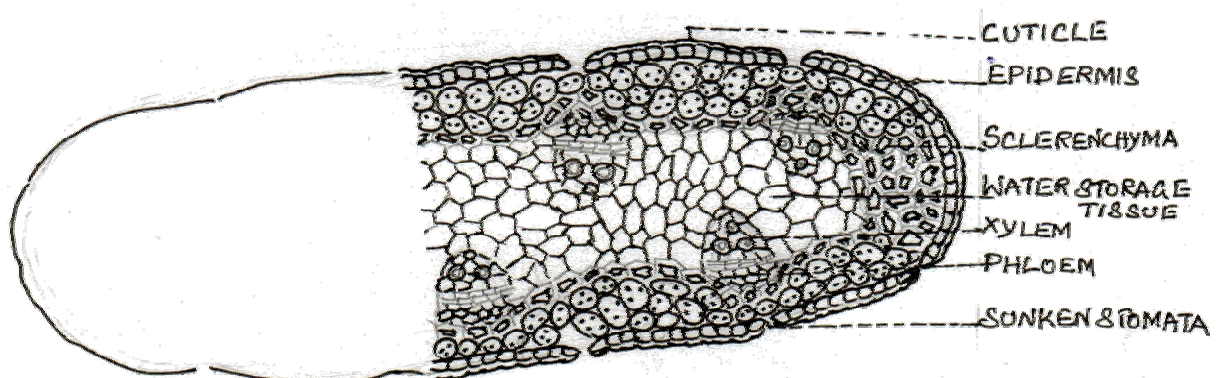
### **HABIT**

- It is flattened, ribbon-shaped phylloclade.
- Nodes and internodes are seen.
- Leaves develop at the nodes in an alternate manner.



## PHYLLOCLADE T.S

- The phylloclade is bounded by a cuticularised epidermis.
- This is followed by the hypodermis which is partially consisting of sclerenchymatous cells and partially consisting of chlorenchymatous cells.
- Below this there is a single layer of sclerenchymatous cells.
- Vascular bundles are restricted to the lower portion of sclerenchyma cells of the hypodermis.
- Each bundle is capped by sclerenchyma.
- The bundles are collateral, open with endarch xylem.
- The margins of the phylloclade have sclerenchymatous hypodermis.
- The rest of phylloclade is composed of large parenchymatous cells which help to store water.



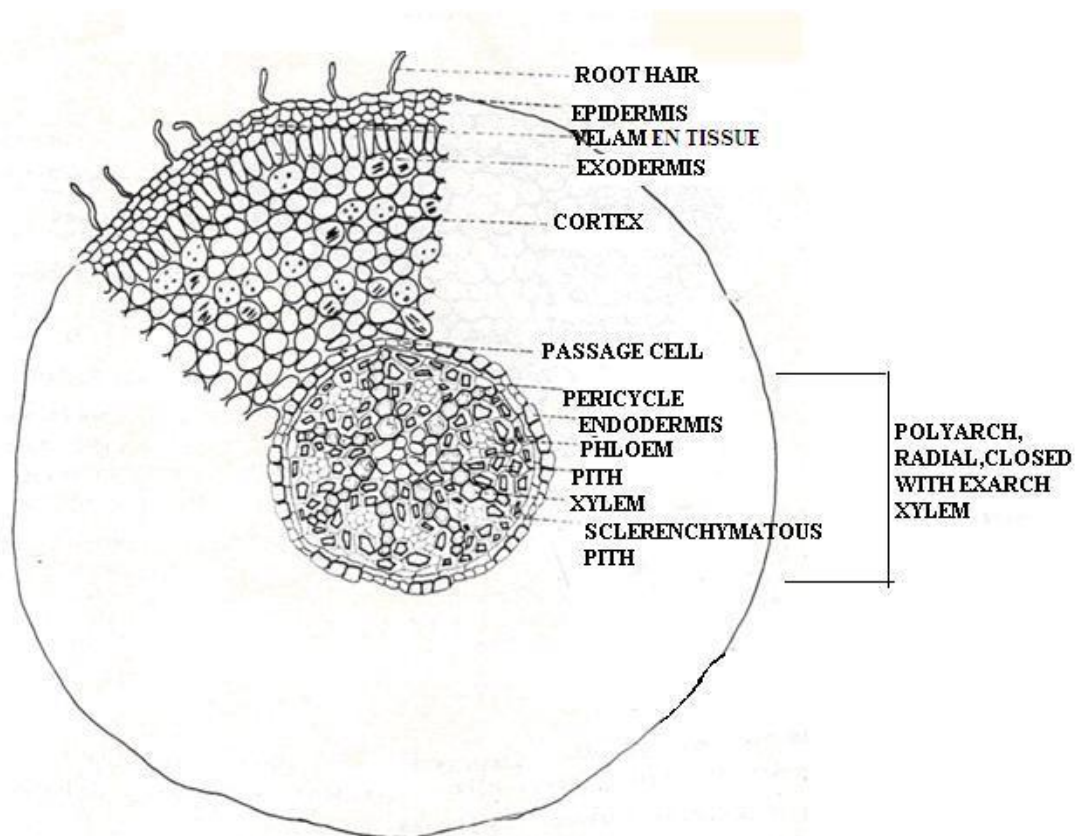


## EPIPHYTIC ROOT

### ORCHID

#### T.S OF ROOT

- Transverse section of an orchid root has a multiple epidermis forming the velamen tissue delimited from the cortex by submerged cells of exodermis.
- The outer most layer is epiblema produce many root hairs which are unicellular.
- The cortex is broad and parenchymatous with starch grains and raphides are seen in the cortex.
- There is a well defined endodermis having the thin walled passage cells.
- The protoxylem lie opposite to passage cells.
- The pericycle is parenchymatous.
- Vascular bundle is polyarch, radial, closed with exarch xylem.
- Pith is parenchymatous.



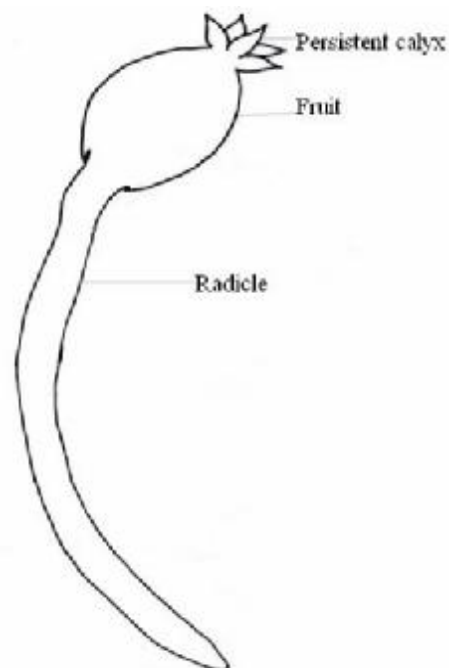


## HALOPHYTE

### *AVICENNIA*

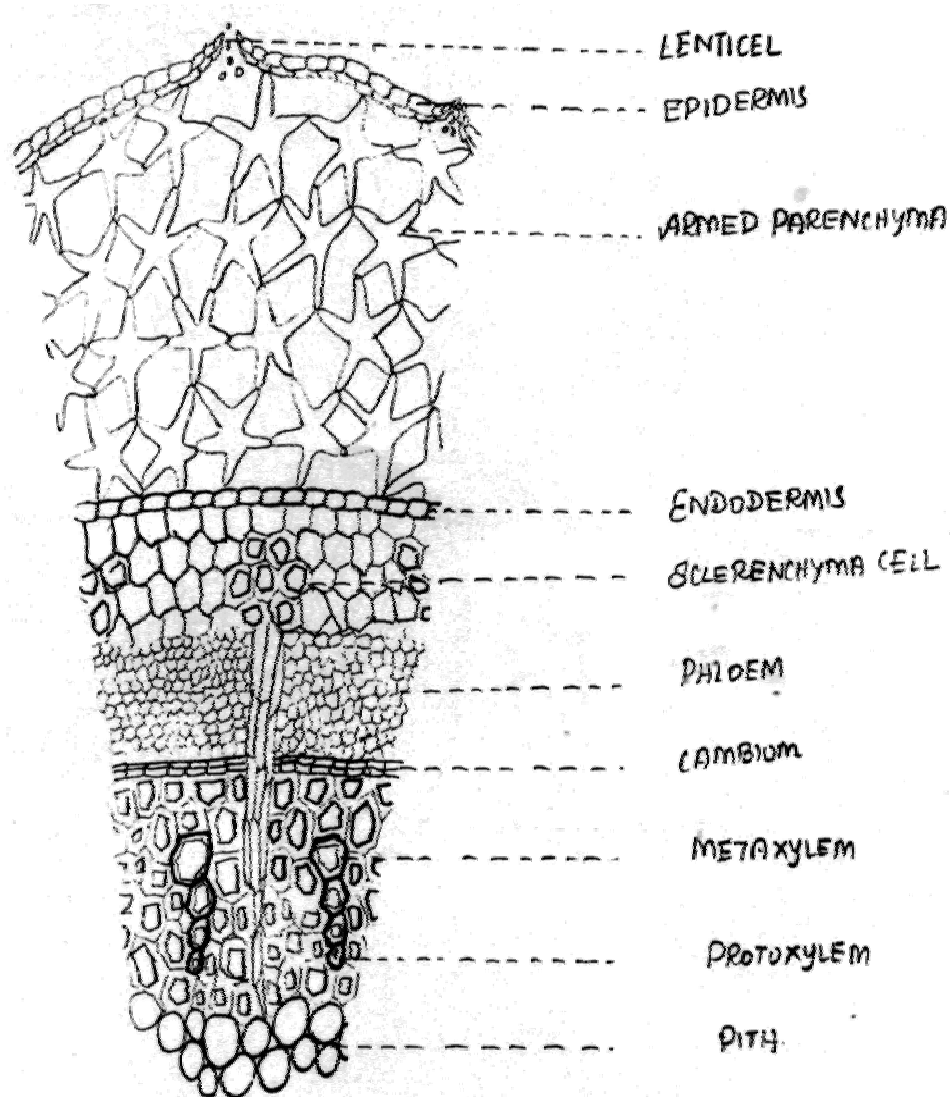
#### VIVIPAROUS GERMINATION

- *Avicennia* is a halophyte growing in marshy areas, which do not afford conditions favourable for germination of seeds.
- The plant shows a specialized type of germination known as vivipary.
- The seed starts germination within the fruit, while it is attached to the mother plant.
- The embryo does not undergo any period of rest.
- The radicle comes first outside the seed and it hangs in the air for a while, when it happens to touch the soil, it becomes anchored to the substratum.
- The hypocotyl is large and swollen and the plumule develops above the water level.



## T.S OF PNEUMATOPHORE

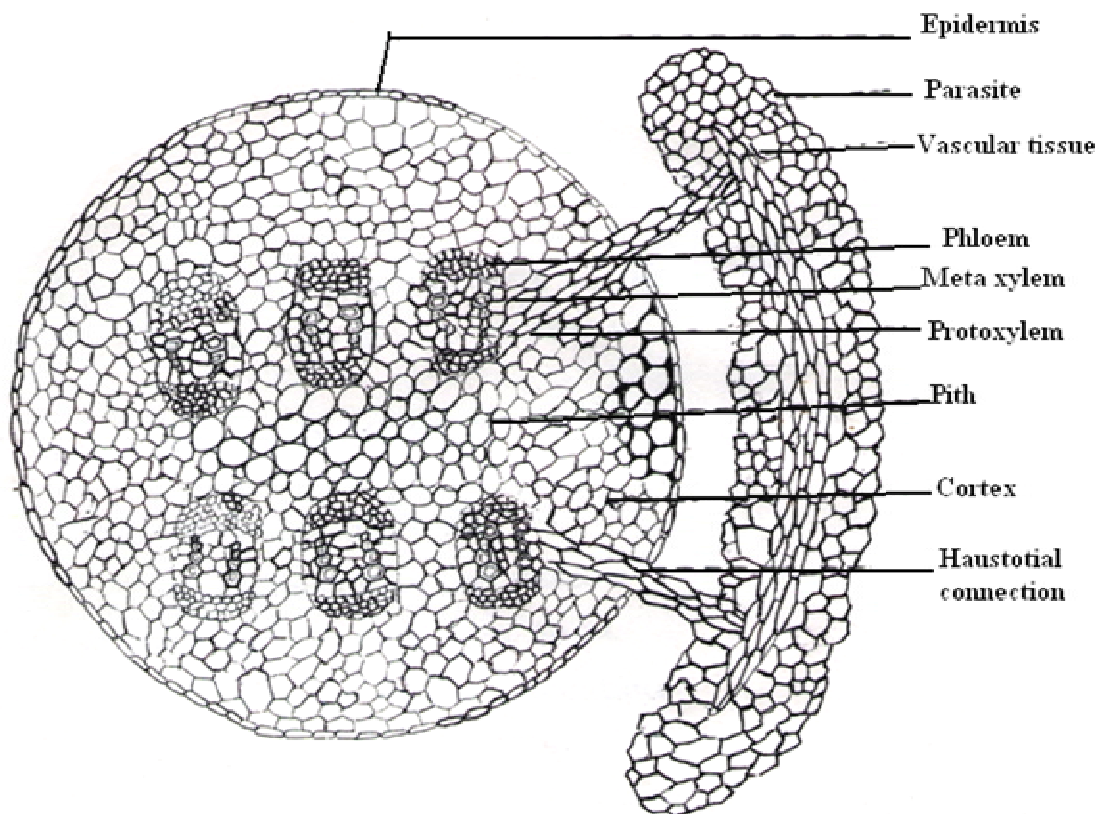
- The periderm shows the presence of many lenticels, having complementary cells behind. The cortex is parenchymatous with large air cavities, helping to store air.
- The vascular bundles are open with collateral endarch xylem.
- Pith is parenchymatous.
- Since the germination is above the soil it resembles the stem.



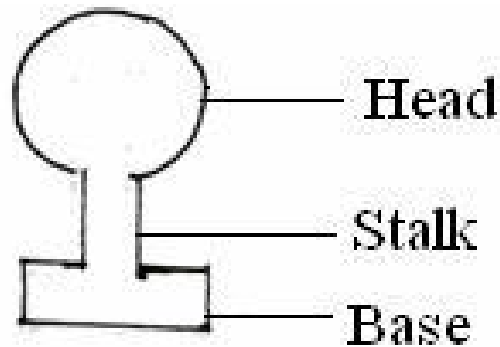
## *CUSCUTA*

### T.S OF HAUSTORIA

- *Cuscuta* is an example of ectoparasite. It is a total stem parasite too growing on the stem of the host.
- The parasite maintains vascular connection with the host through the haustoria.

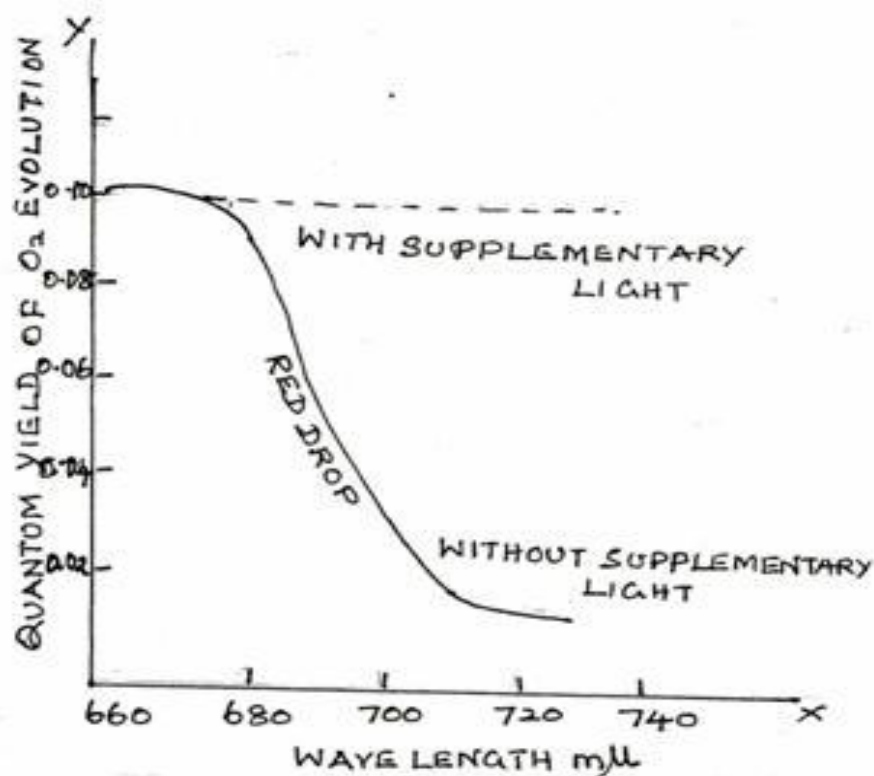


**SPOTTERS**  
**PLANT PHYSIOLOGY**  
**ETP OR F<sub>1</sub> PARTICLE**



- Under electron microscope inner and outer membrane of mitochondria have small particles called elementary particles or F<sub>1</sub> particles or oxysomes or eletrontransport particles.
- The F<sub>1</sub> particles of the outer membrane are stalkless.
- The F<sub>1</sub> particles of the inner membrane are stalked.
- Each stalked F<sub>1</sub> particles consists of a base, a stalk and a head.
- They are regularly placed at a distance of 100 Å.
- They store the energy rich molecule in the form of ATP released during oxidative phosphorylation.

## RED DROP

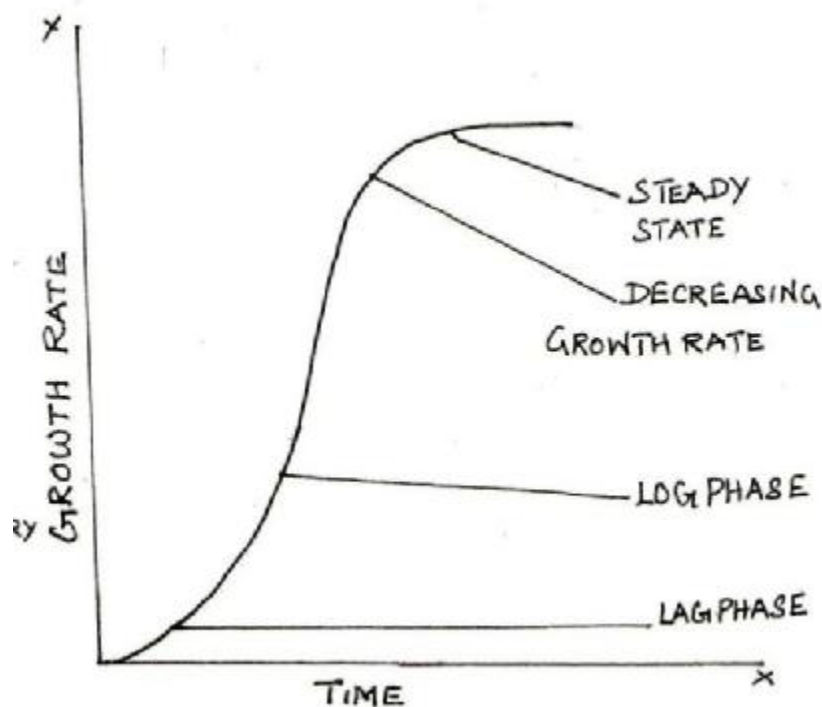


This graph shows red drop Emerson's Enhancement Effect.

While determining the quantum yield of photosynthesis in *Chlorella* by using monochromatic light at different wavelength,

- Robert Emerson noticed the sharp decrease in quantum yield at wavelength beyond 680 nm in the far red part of the spectrum called red drop.
- He found that the inefficient red light in *Chlorella* is made fully efficient, if the shorter wavelengths are used.
- This is known as Emerson's Enhancement Effect.

## GROWTH CURVE



The graph by plotting growth rate against time is called growth curve. This curve is 'S' shaped and is called sigmoid curve. The total period of growth is divided into

- a) Lag period
- b) Log period
- c) Slow period
- d) Steady period

Lag period-Growth starts from this period and the growth rate is very slow. Log period- The growth is maximum. Slow period- Metabolic processes are slow, so the growth rate is very slow. Steady period-during this period, growth is almost complete and static.

## **IAA-INDOLE ACETIC ACID**

- It is a natural auxin.
- They are synthesized in the tips of the stem and roots.
- It is a growth regulator.
- It promotes cell elongation, apical dominance.
- Root initiation in stem cuttings.
- Induces parthenocarpic fruit.
- Enhances respiration.
- IAA is used to prolong the dormancy of buds in potato.
- Induces callus formation in tissue culture.
- Prevent abscission.

## **CYTOKININ**

- Cytokinins are plant growth hormones.
- Chemically they are identified as 6-furfuryl aminopurine.
- It is richly found in liquid endosperm of coconut milk.
- Zeatin is more active Cytokinin.

## **PHYSIOLOGICAL ROLE**

- Promotes cell division and cell enlargement.
- Initiation of interfascicular cambium
- Counteraction of apical dominance.
- Break seed dormancy.
- Delay of senescence or Richmand Lang effect.

- Richmand Lang showed that senescence could be postponed in *Xanthium* leaves by kinetin treatment, hence the name Richmand Lang effect

## **GIBBERELLINS**

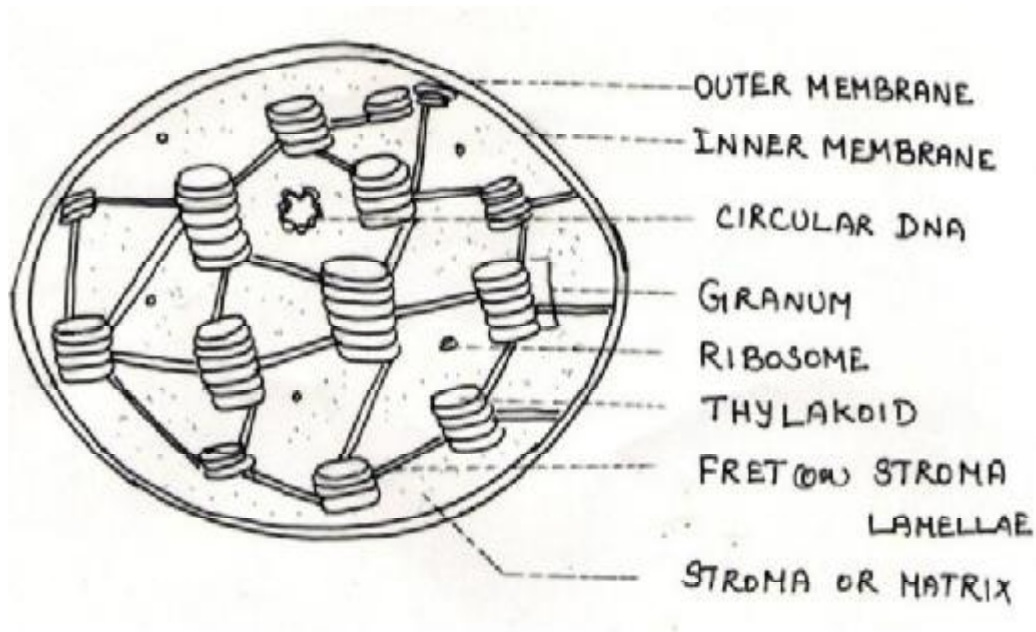
- Gibberellins are growth hormones
- Isolated from infected rice seedlings by the fungus *Gibberella fugikuroi*.
- 60 gibberellins have been isolated from different plants.
- It has common skeleton called gibbane ring.
- GA<sub>3</sub> is called as gibberellic acid.
- They are synthesized in shoot, roots and leaves.

## **PHYSIOLOGICAL ROLE**

- Induces seed germination.
- Break seed and bud dormancy.
- Elongation of internodes in dwarf pea, dwarf maize etc to overcome the genetic dwarfism.
- Bolting and flowering-induces shoot elongation and flowering in rosette type of plants.
- Induces parthenocarpy.
- Promotes synthesis of  $\alpha$ -amylase in maize.

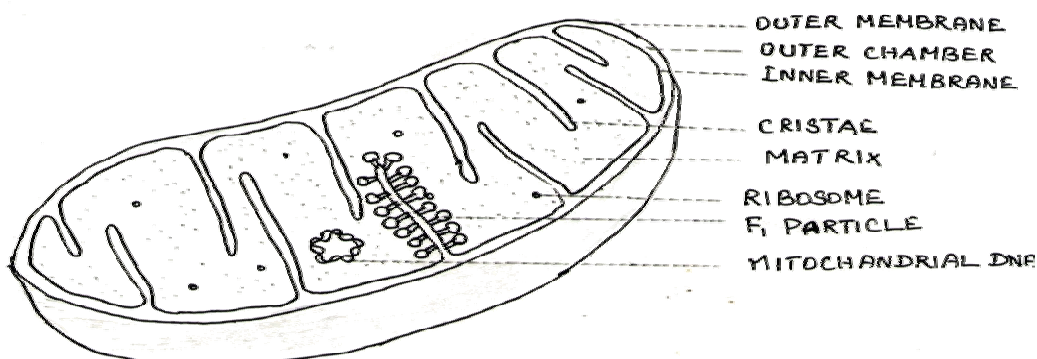


## CHLOROPLAST



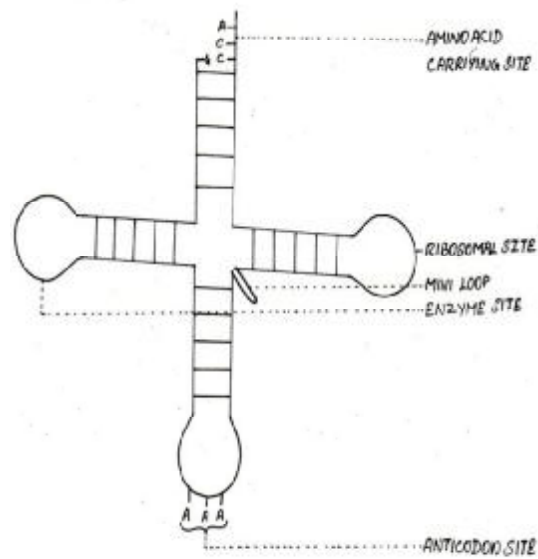
- Chloroplast is a cytoplasmic organelle.
- It contains green pigments-chlorophylls.
- It is bounded by two lipoproteinaceous membranes.
- Inner region is filled with colloidal substance-matrix or stroma.
- Many closed, flat sacs like thylakoids are arranged one above the other called granum.
- Grana are interconnected by frets.
- Presence of circular DNA.
- Function - photosynthesis.

## MITOCHONDRIA



- Mitochondria-cytoplasmic organelle.
- Power house of the cell.
- Covered by outer and inner membrane, separated by outer chamber.
- Inner membrane has finger like projections called cristae.
- Cristae are covered with elementary particles or F1 particles.
- Small,circular,double standard DNA present in the matrix.
- **Functions:**
  - i) Cellular respiration.
  - ii) Synthesis energy rich compound ATP.

## STRUCTURE OF T-RNA



- Transfer RNA-single stranded, folded itself to form clover leaf.
- 60 types of t-RNA and it is made up of 73 to 95 nucleotides.
- It has 5' and 3'. 3' is ending with CCA and 5' is ending with G or C.
- t-RNA has 5 sites:
  - a. ribosomal site,
  - b. enzyme site,
  - c. anticodon site
  - d. aminoacid site carrying site.
  - e. mini loop.
- Each t-RNA shows specificity in carrying aminoacids.
- Mini loop or variable arm contains unusual aminoacids.
- **Function:**

Protein synthesis.

## ENVIRONMENT, BIODIVERSITY AND CONSERVATION

### FOOD CHAIN

- The sequence of eaters and being eaten is called a food chain.
- The various stages in a food chain are called trophic level.

Eg., producers form the food for herbivores; the herbivores form the food for the carnivores.

Producers    →    Herbivores    →    Carnivores.

- Producers are chiefly green plants. They trap solar energy and convert into chemical energy.
- Food manufactured by green plants is also eaten by herbivores.
- Herbivores fall prey to some carnivores.
- Food chain in any ecosystem runs directly in which green plants are eaten by herbivores, herbivores are eaten by carnivores and carnivores are eaten by top carnivores.

Marsh grass    →    Grasshopper    →    Bird    →    Hawk

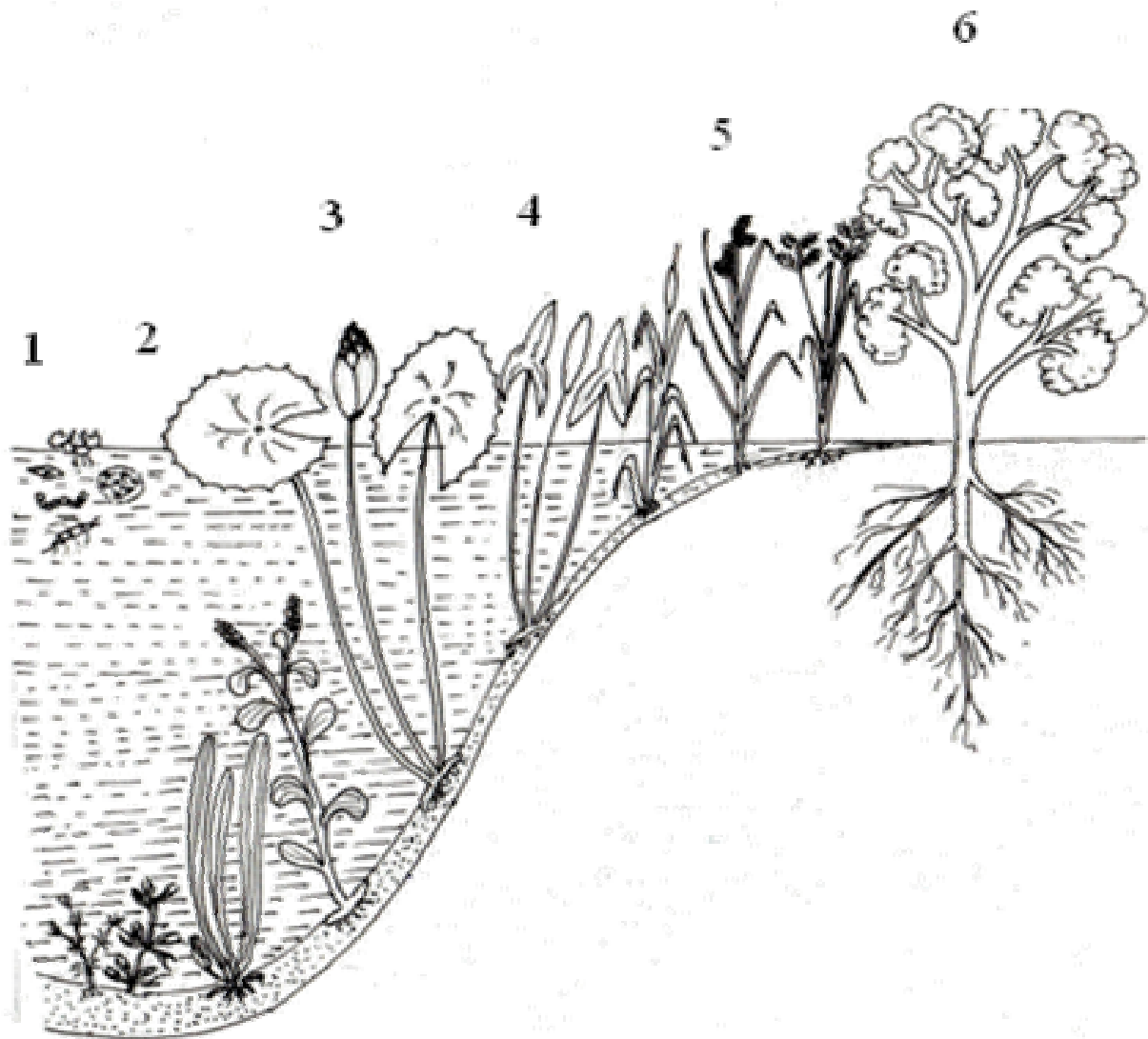
## FOOD WEB

- Complex of interrelated food chains is called food web.
- Food web maintains the stability of the ecosystem.
- In a grassland ecosystem, grass is eaten by grasshopper, rabbit and mouse; grasshopper is eaten by hawk or lizard which is eaten by hawk; Rabbit is eaten by Hawk; Mouse is eaten by snake which may be eaten by hawk; Besides these hawk also eats grasshopper and mouse.
- Thus there are five linear food chains which are interlocked.
  1. Grass → Grasshopper → Hawk.
  2. Grass → Grasshopper → Lizard → Hawk.
  3. Grass → Rabbit → Hawk.
  4. Grass → Mouse → Hawk.
  5. Grass → Mouse → Snake → Hawk.

## SUCCESSION

- Gradual replacement of one type of vegetation by the other till a climax community is formed is termed as succession.
- When the succession starts from a hydric medium, it is termed hydrosere.
- The phytoplanktons are the pioneers in hydrosere.
- The climax is the mesophytic forest.
- The various seral communities in a hydrosere are:
  - (i) Phytoplanktonic stage: Pioneer; simple forms like bacteria, algae and aquatic plants.

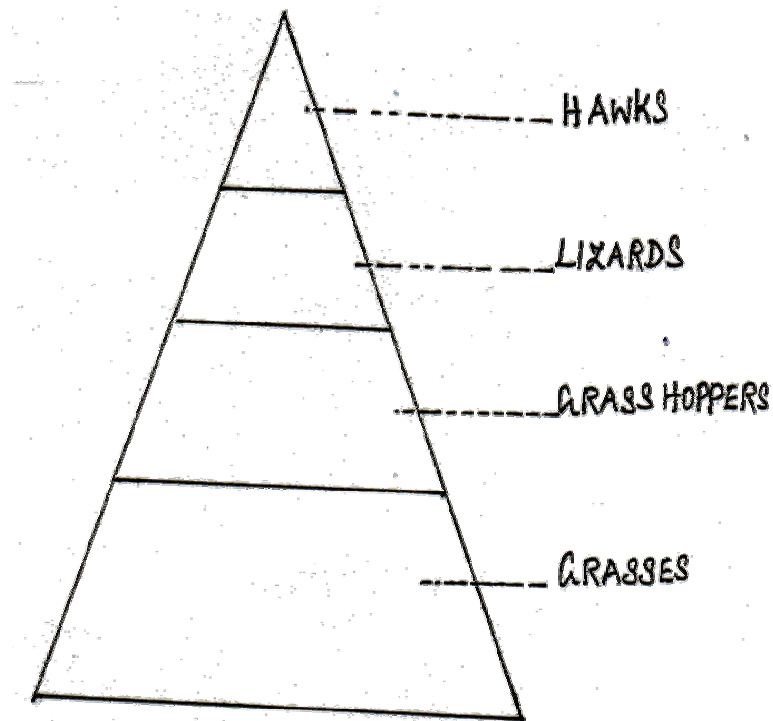
- (ii) Submerged stage: *Vallisneria*, *Chara*.
- (iii) Floating stage: *Nelumbium*, *Pistia*.
- (iv) Reed-swamp stage: *Typha*, *Bothrioclova*.
- (v) Sedge meadow stage: *Carux*, *Mentha*.
- (vi) Woodland stage: *Acasia*, *Cassia*.
- (vii) Climax forest: Herbs, shrubs, mosses.



1. Phytoplankton stage 2. Submerged stage 3. Floating stage  
4. Reed swamp stage 5. Sedge meadow stage and 6. Climax forest

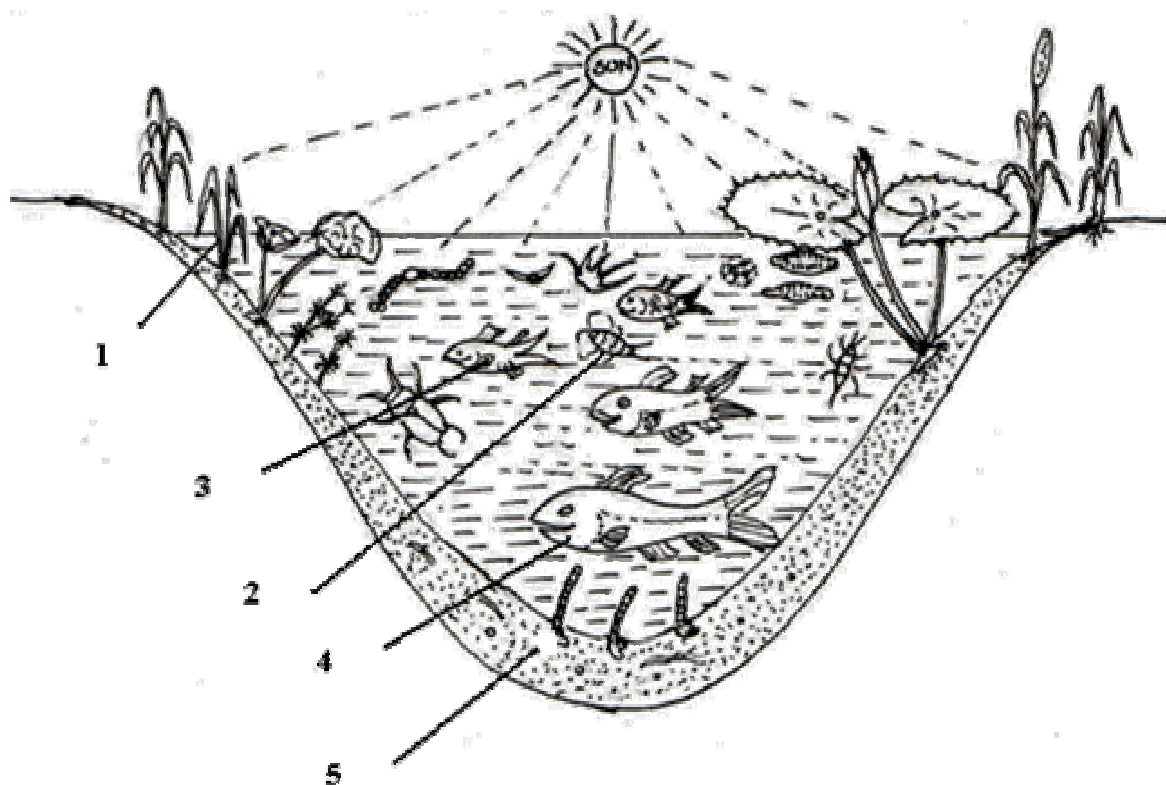
## PYRAMID OF NUMBERS

- The trophic structure of an ecosystem represented in the form of pyramid is called as ecological pyramid;
- The number of individuals at the trophic level decreases from the producer to the consumer level.
- The base of the pyramid is represented by producer, which is abundant.
- Successive levels of consumers, the number of organisms goes on decreasing rapidly.
- In a grassland ecosystem grasses are there in large numbers; number of consumers decreases in the following order.  
Grasses > Grasshopper > Lizards > Hawks.



## POND ECOSYSTEM

- Pond ecosystem is a lentic fresh water ecosystem.
- It is formed by abiotic and biotic factors.
- Abiotic components are water, CO<sub>2</sub>, inorganic compounds, organic compounds, light, temperature etc.,
- Biotic components are producers, consumers and decomposers.
- Producers include phytoplanktons, green algae, rooted plants, submergerd plants and floating plants.
- Consumers include primary consumers (Zooplanktons, insects) and secondary consumers(large fishes and snakes)
- Decomposers include microbes like bacteria and fungi.



1. Producer 2. Primary consumer 3. Secondary consumer 4. Tertiary consumer and 5. Decomposer



## **ORCHID ROOT**

- Presence of velamen tissue.
- Single layer of thickened cells inner to velamen tissue called as exodermis.
- Cortex consists of thin walled cells with intercellular space.
- Innermost layer of cortex is endodermis followed by pericycle.
- Conjunctive tissue is sclerenchymatous.
- Vascular bundles are polyarch, radial, closed and exarch.
- Pith consists of thin walled parenchymatous cells.
- Presence of starch grains and raphides in the cortex and pith.

## **PNEUMATOPHORE**

- Pneumatophores are special type of negatively geotropic roots found in halophytes.
- It is also called as breathing roots.
- Outermost layer is the periderm.
- Periderm has a number of lenticels.
- Cortex is spongy and consists of aerenchyma enclosing air spaces.
- Vascular bundles are open, conjoint, collateral with endarch xylem.
- Pith is parenchymatous.

## ***CUSCUTA***

- *Cuscuta* is an obligate total stem parasite.
- It grows on *Acacia*, *Zizyphus* and other angiospermic plants.
- It maintains vascular connection with the host through haustoria.

## **HYDROPHYTIC CHARACTERS**

### ***HYDRILLA* STEM**

- *Hydrilla* is a rooted submerged hydrophyte
- Absence of cuticle.
- Presence of aerenchymatous cortex.
- Only thin walled parenchymatous cells present.
- No supporting or mechanical tissue is found.
- Phloem is represented by broad zone.
- Central cavity represents the xylem.
- No vessels are found.

### ***EICHHORNIA* ROOT**

- *Eichhornia* is a free floating aquatic weed.
- Outer epiblema is non-cuticularised.
- Middle cortex made up of loosely arranged strands of parenchyma separated by air spaces.
- Absence of mechanical tissue.
- Only thin walled parenchymatous cells are present.
- Reduction in vascular tissues.

### ***NYMPHAEA* PETIOLE**

- *Nymphaea* is a floating but rooted aquatic plant.
- Presence of waxy layer above the epidermis.
- Presence of large air spaces.
- Reduction in mechanical tissue.
- Poorly developed vascular tissue.
- Mechanical strength is provided by raphides.
- Xylem represented by small cavities.

### **XEROPHYTIC CHARACTERS**

#### ***NERIUM* LEAF**

- Presence of thick cuticle.
- Multilayered epidermis present.
- Stomata are confined to lower epidermis, protected by hairs.
- Vascular system is well developed.
- Well differentiated mesophyll; more of palisade.
- Presence of mechanical tissue.

#### ***CASUARINA* CLADODE**

- Presence of thick cuticle.
- Ridges and furrows are found around the stem.
- Stomata are found in the depressions/ furrows
- Hairs are present in the furrows which protect the stomata.
- Palisade cells are present in the stem.
- Vascular system is well developed.
- Mechanical tissues are well developed.

## ***ASPARAGUS CLADODE***

- Well defined cuticularised epidermis.
- Hypodermis is parenchymatous.
- Vascular bundle consists of 3 groups of xylem and phloem.
- Bundle is surrounded by bundle sheath.
- Rest of the space is occupied by elongated thin walled chlorenchymatous cells.

## MODEL QUESTION PAPER

**Time: 3 hrs**

**Max. Marks: 100**

1. Set up an experiment of **A**. List the materials required. Submit the procedure, tabulate the data and record the results. Leave the set up for valuation.

**(Materials required-2; procedure-6; Results-2; set up-4; inference-3)** **(1 × 17 = 17)**

2. Cut transverse section of **B** and **C** stain and mount in glycerin. Draw diagrams and identify their ecological habitat with reasons. Submit the slides for valuation.

**(Slide-4; diagram-2; notes-2; ecological habitat-1)** **(2 × 9 = 18)**

3. Draw sketches, identify and write notes on **D, E, F, G, H, I, J** and **K**.

**(Identification-1; diagram-2; notes-3)** **(8 × 6 = 48)**

4. Comment on the physiology set up **L**.

**(Identification-1; diagram-2; notes-4)** **(1 × 7 = 7)**

<b>Total</b>	<b>90</b>
<b>Record</b>	<b>10</b>
<b>Total</b>	<b><u>100</u></b>

**KEY**

1. **A** - Individual set up
2. **B** and **C** - *Hydrilla* stem / *Nymphaea* petiole / Eichhornia root / Casuarina / Nerium / Asparagus.
3. **D, E, F & G** – ETP / Growth curve / Red drop / Auxin / Gibberellin / Cytokinin / Chloroplast / Mitochondria / tRNA.  
**H, I, J & K** - Epiphytic root / *Cuscuta* / Pneumatophore / Food chain / Food web / Hydrosere / Pyramid of number / Pond ecosystem / Vivipary.
4. **L** - Transpiration pull / Anaerobic respiration / Khune's bulb / Arc auxanometer / Clinostat / Respiroscope.