

PRACTICAL

SUBJECT: BIO-BOTANY

STANDARD: 11

Topic

Preparation and Demonstration of Slides

1 Mitotic cell division stages

2 Anatomical structure -
Dicot & Monocot (Root, Stem & Leaf)

3 Plasmolysis and Deplasmolysis

Fresh or preserved specimens

4 Phylloclade - Opuntia

5 Special inflorescence - Cyathium

Taxonomy - Flower Dissection

6 Fabaceae - Clitoria ternatea

7 Solanaceae - Datura metel

Bio molecules - Nutrient test

8 Test for reducing sugar-Benedict test

9 Starch - Iodine test

10 Protein - Biuret test

11 Lipid - Saponification test

Plant Physiology Experiments

12 Paper Chromatography

13 Wilmott's Bubbler

14 Demonstration of production of CO₂ during respiration



a. Mitosis – Stage : Metaphase

Diagnostic features:

- The spindle fibres attached to the kinetochore region of centromere of chromosomes
- Chromosomes are arranged at the equator region of the cell (metaphase plate)
- Chromosomes are distinctly visible in this stage.

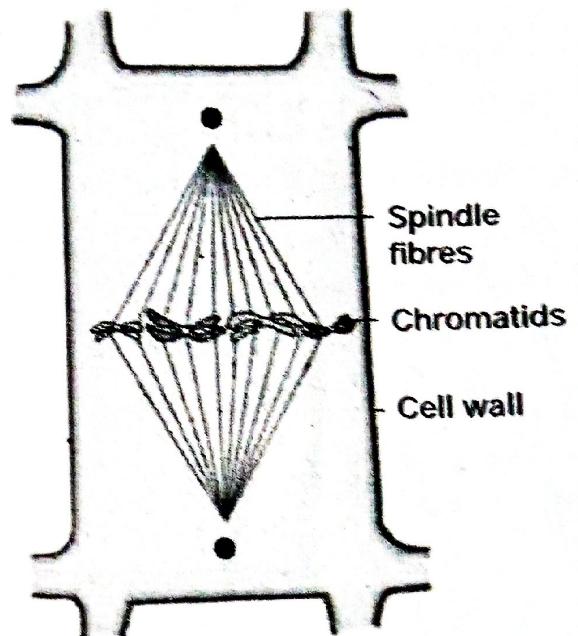


Figure 4a: Metaphase

b. Mitosis – Stage : Anaphase

Diagnostic features:

- Each chromosome splits and two daughter chromatids begin to move towards opposite poles.
- Shortening of spindle fibre and longitudinal splitting of centromere creates a pull which divide the chromosomes.

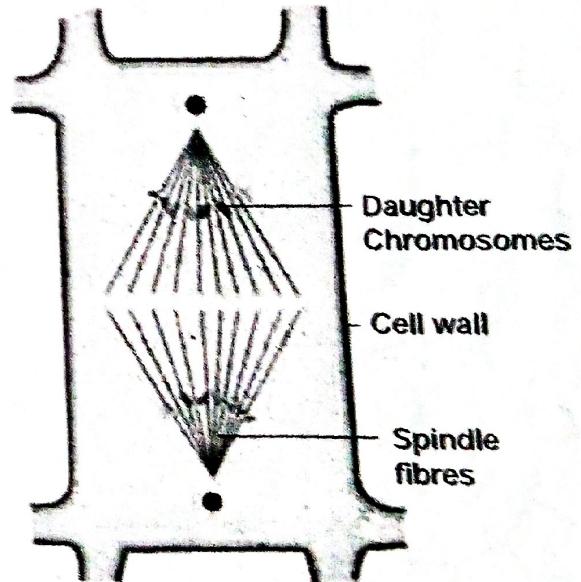


Figure 4b: Anaphase

a. Dicot Root (T.S)

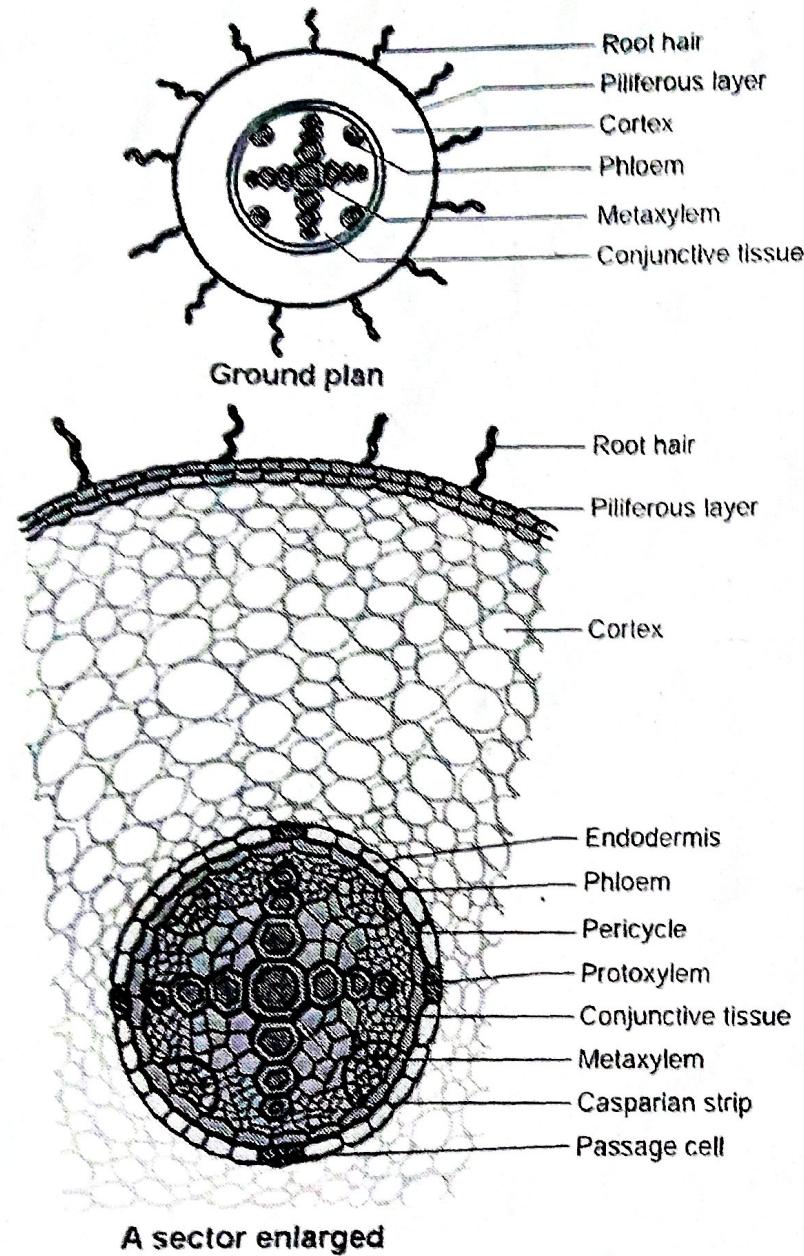


Figure 5a: T.S of Dicot root (Bean root)

b. Dicot Stem (T.S)

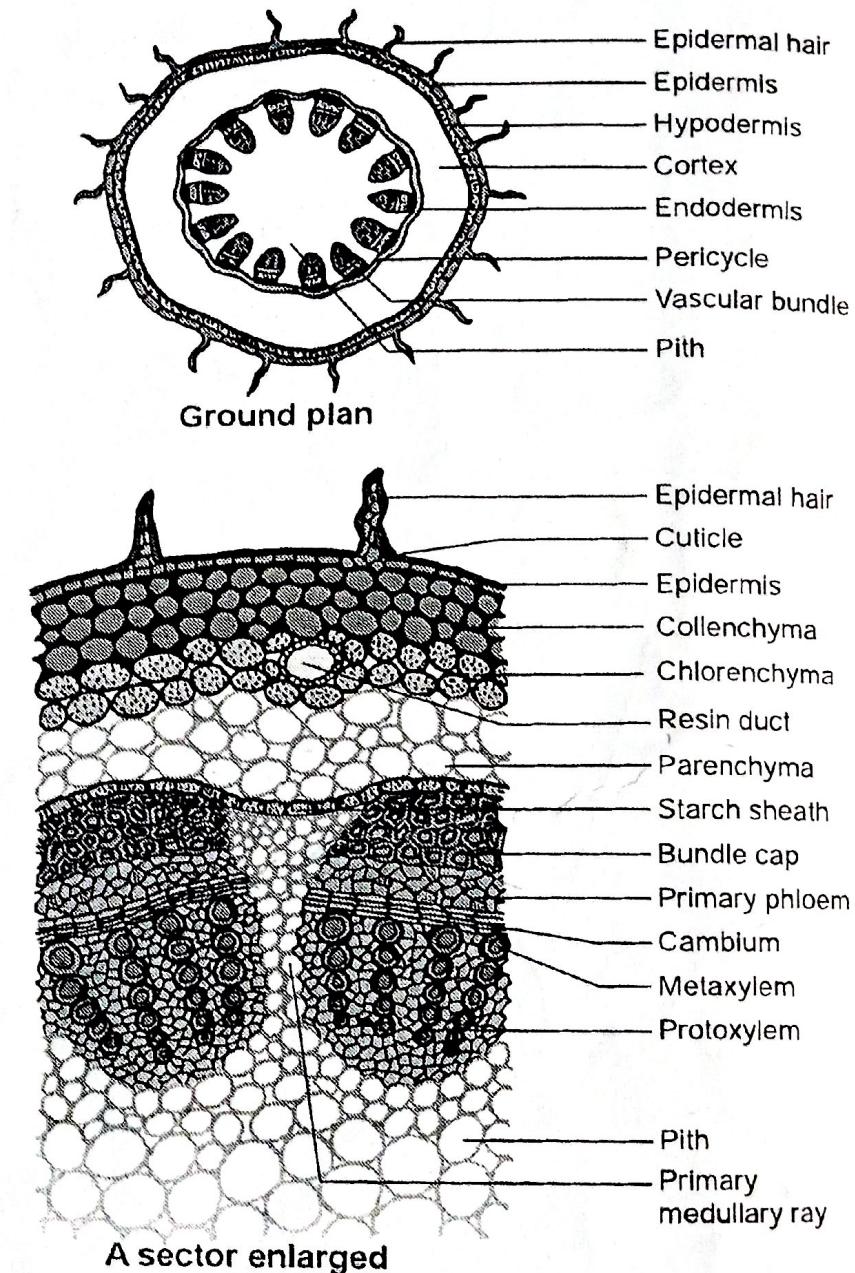


Figure 5b: T.S of Dicot stem (Sun flower stem)

Dicot Root (T.S)

Diagnostic features:

- Radial vascular bundle, exarch and tetrarch xylem.
- Parenchymatous conjunctive tissue is present.
- Pith is absent.

Dicot Stem (T.S)

Diagnostic features

- Cortex differentiated, hypodermis made up of collenchyma cells.
- Conjoint, Collateral and Open vascular bundle (Cambium present)
- Vascular bundle arranged like a ring, wedge shaped vascular bundle.
- Presence of pith and primary pith rays.

c. Dicot Leaf (T.S)

Diagnostic features

- Conjoint, Collateral and closed vascular bundle.
- Mesophyll tissue differentiated into upper palisade parenchyma and lower spongy parenchyma. (Dorsiventral leaf)
- Stomata are more in number on the lower epidermis.
- Stomata surrounded by bean shaped guard cells.

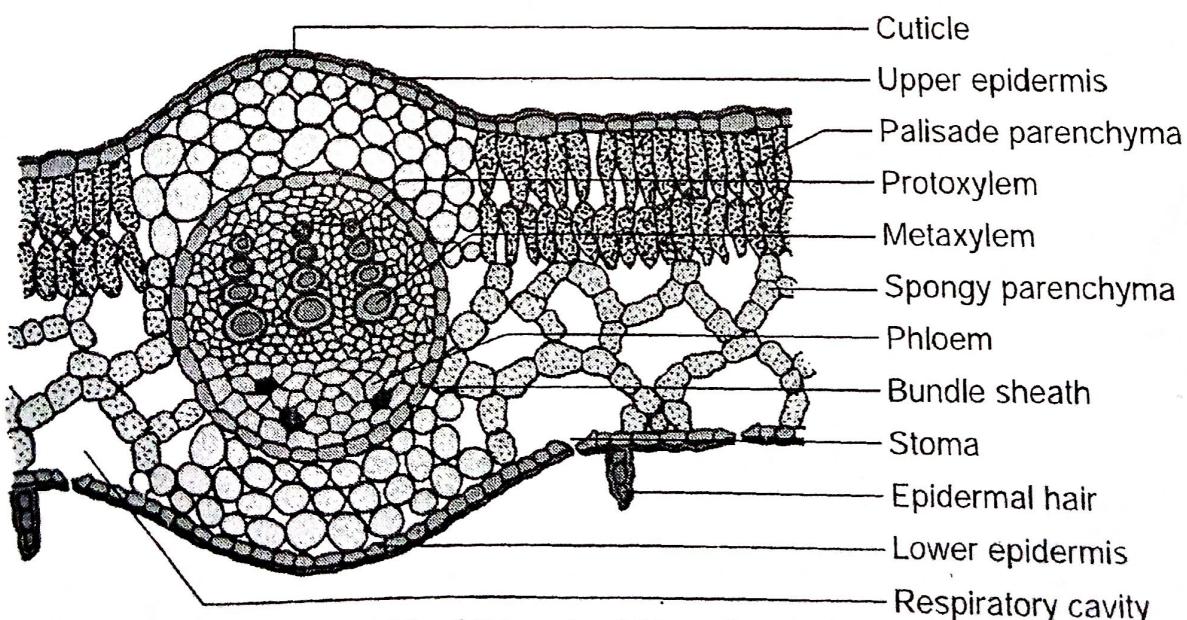


Figure 5c: T.S of Dicot leaf (Sun flower leaf)

d. Monocot Root (T.S)

Diagnostic features:

- Radial vascular bundle, exarch and Polyarch xylem.
- Pith is Present.

e. Monocot Stem (T.S)

Diagnostic features

- Conjoint, Collateral and Closed vascular bundle. (Cambium absent)
- Skull shaped and scattered vascular bundle.

- Sclerenchymatous conjunctive tissue is present.

- Pith absent, homogenous ground tissue.

- Ground tissue is not differentiated into cortex and pith. Hypodermis made up of Sclerenchyma cells.

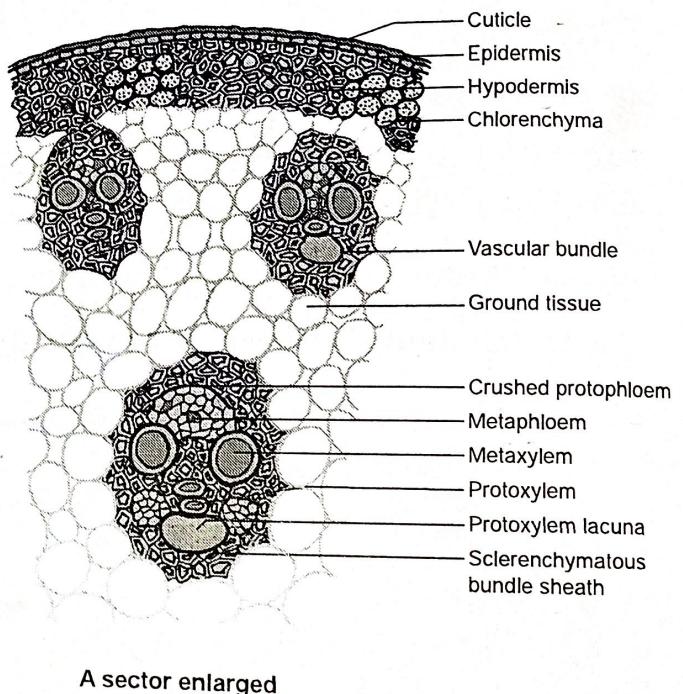
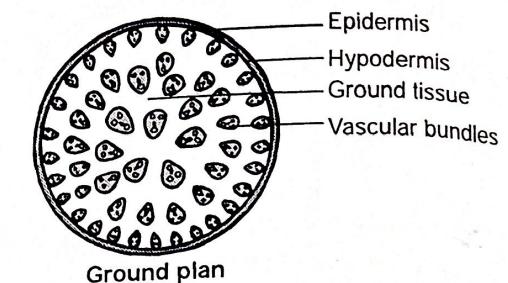
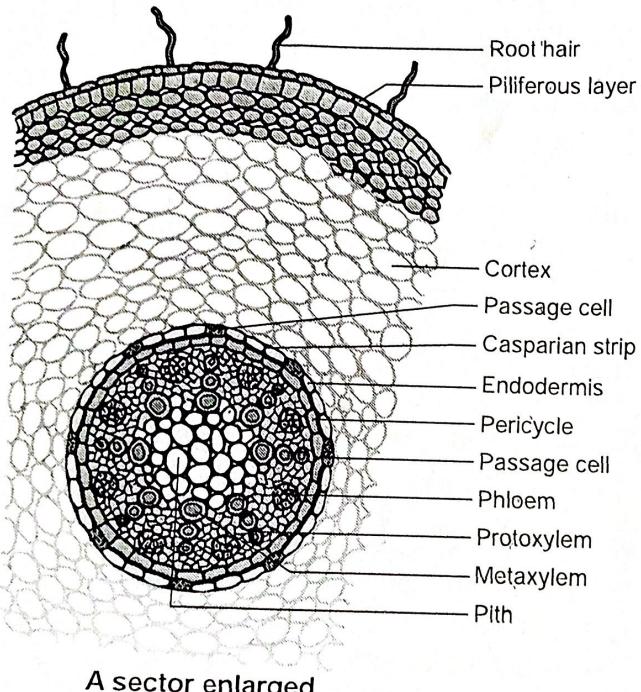
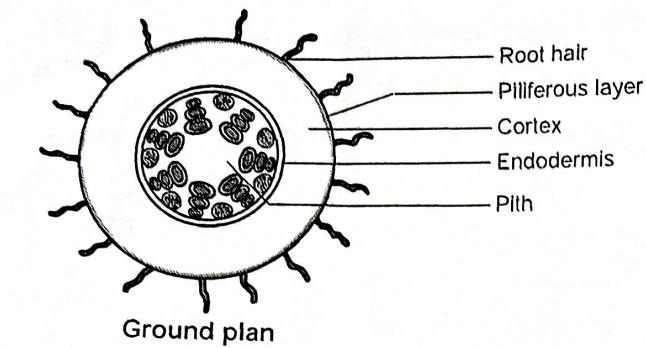


Figure 5d: T.S of Monocot root (Maize root)

Figure 5e: T.S of Monocot stem (Maize Stem)

f. Monocot Leaf (T.S)

Diagnostic features:

- Conjoint, Collateral and closed vascular bundle.
- Mesophyll is not differentiated into Palisade and Spongy parenchyma. (Isobilateral leaf)
- Number of Stomata are more or less equal on both epidermis, Stomata surrounded by dumb-bell shaped guard cells.

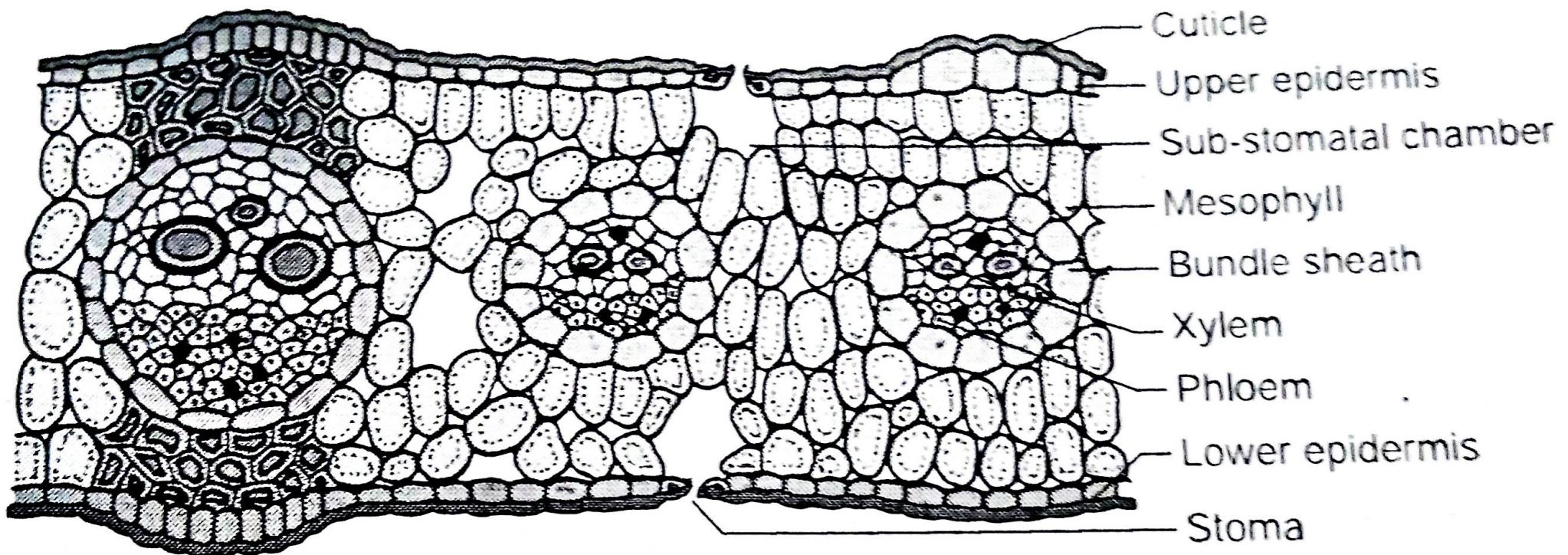


Figure 5f: T.S Monocot leaf (Grass leaf)

Exercise: 6

Plasmolysis and Deplasmolysis

Figure 5f: T.S Monocot leaf (Grass leaf)

Diagnostic features: Plasmolysis

- Cell membrane is pulled away from the cell wall.
- Cells becomes flaccid due to loss of water by exosmosis, when a plant cell is kept in a hypertonic solution.

Diagnostic features: Deplasmolysis

- It is reverse of plasmolysis.
- It is swelling of shrunked protoplasm to regain its original unplasmolysed shape when cell is placed in hypotonic solution. It is a type of endosmosis.

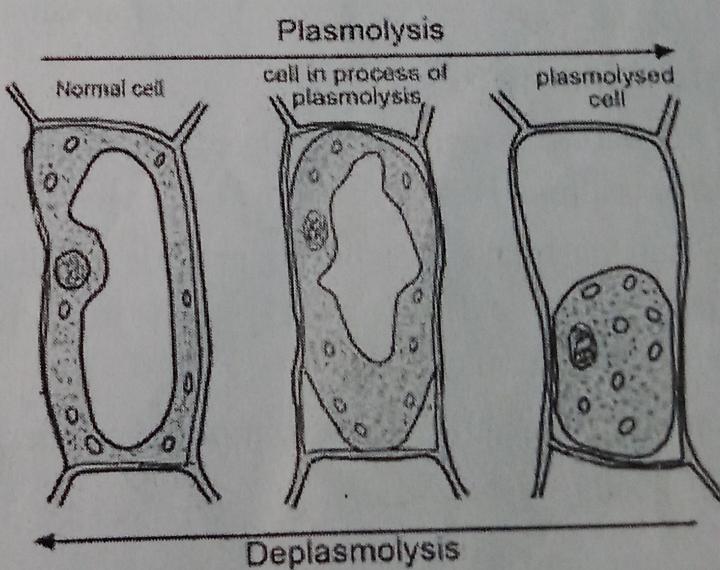


Figure 6: Different Stages of Plasmolysis and Deplasmolysis in a plant cell

Exercise: 9

Phylloclade - *Opuntia*

Diagnostic features:

- It is a green, flattened stem.
- Phylloclade (Cladophyll) is the stem modification, perform the function of leaves.
- Leaves are modified into spines for xerophytic adaptation.

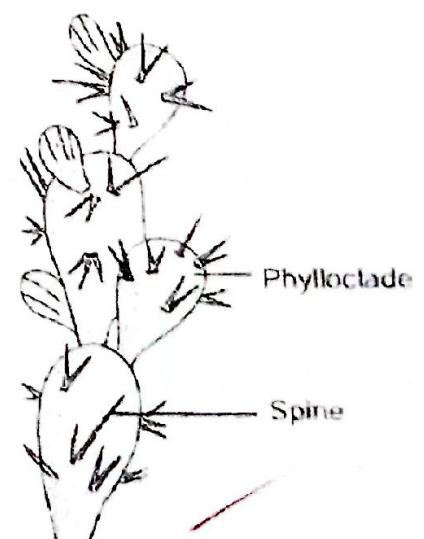


Figure 9: Phylloclade - *Opuntia*

Excise: 10

Special inflorescence- Cyathium

Diagnostic features:

- Special type of inflorescence consists of small unisexual flowers.
- Centrally located single female flower surrounded by male flowers.
- Male flower represented by only stamen and female flower represented only by pistil.
- Involucres protect flowers and consist of nectar.

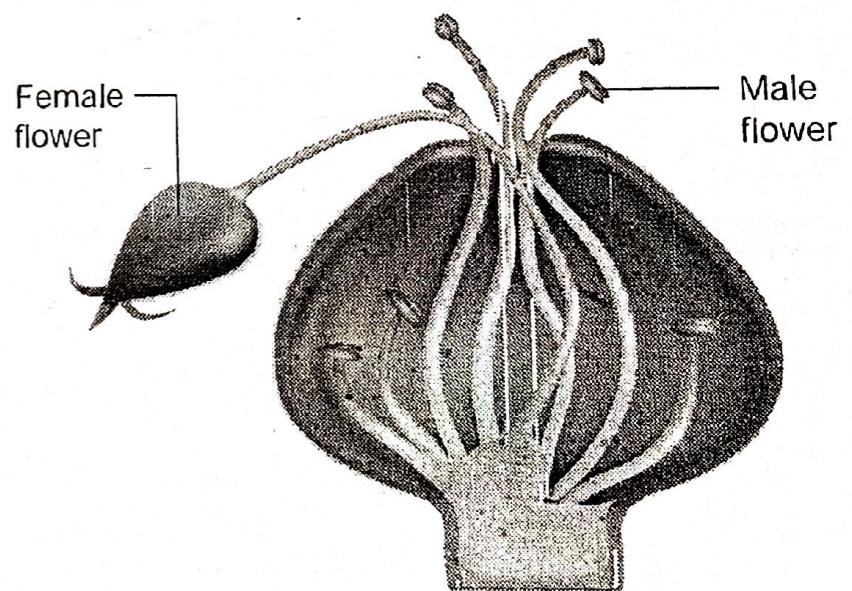


Figure 10: Cyathium inflorescence

Exercise: 12

Fabaceae - *Clitoria ternatea*

Systematic position

Kingdom : Plantae

Clade : Angiosperms

Clade : Eudicots

Clade : Rosids

Order : Fabales

Family : Fabaceae

Floral characters:

Inflorescence : Solitary and axillary cyme.

Flower : Bractate, bracteolate, bisexual, zygomorphic, pentamerous and hypogynous.

Calyx : Sepals 5, synsepalous, Valvate aestivation, odd sepal is anterior in position.

Corolla : Petals 5, apopetalous, Papilionaceous corolla and descendingly imbricate.

Androecium : Stamens 10, diadelphous, (9) + 1.

Gynoecium: Monocarpellary, unilocular and ovules on marginal placentation, Superior ovary.

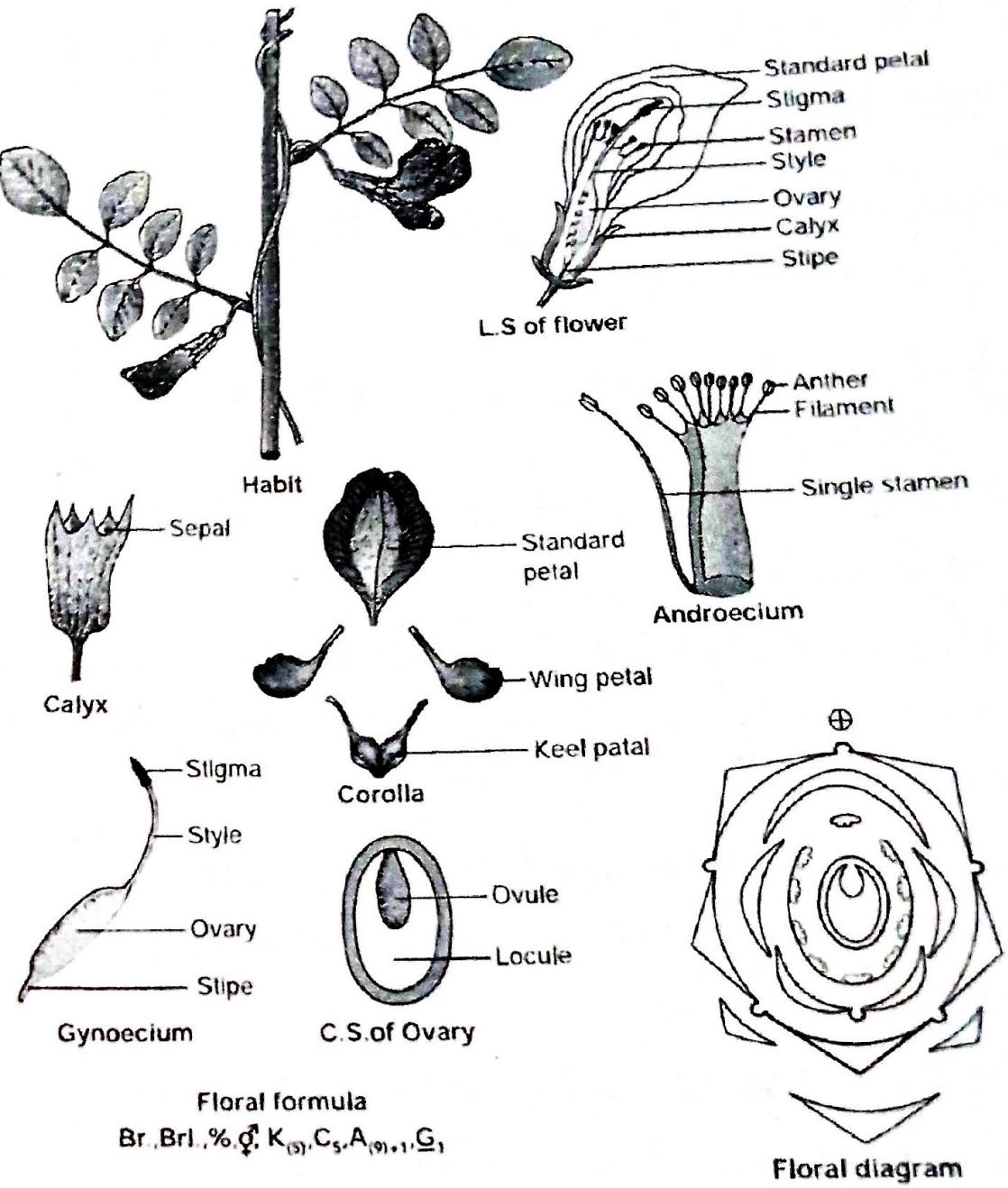


Figure 12 : *Clitoria ternatea*

Exercise: 13

Solanaceae – *Datura metel*.

Kingdom: Plantae

Clade: Angiosperms

Clade: Eudicots

Clade: Asteridae

Order: Solanales

Family: Solanaceae

Floral characters

Inflorescence: Solitary and axillary cyme.

Flower: Bractate, ebracteolate, bisexual, actinomorphic, pentamerous and hypogynous.

Calyx: Sepals 5, synsepalous, Valvate aestivation, persistent calyx and odd sepal posterior.

Corolla: Petals 5, Synpetalous, twisted aestivation and plicate.

Androecium: Stamens 5, epipetalous and alternipetalous.

Gynoecium: Bicarpellary, syncarpous and superior ovary, bilocular due formation of false septum looks tetra locular.

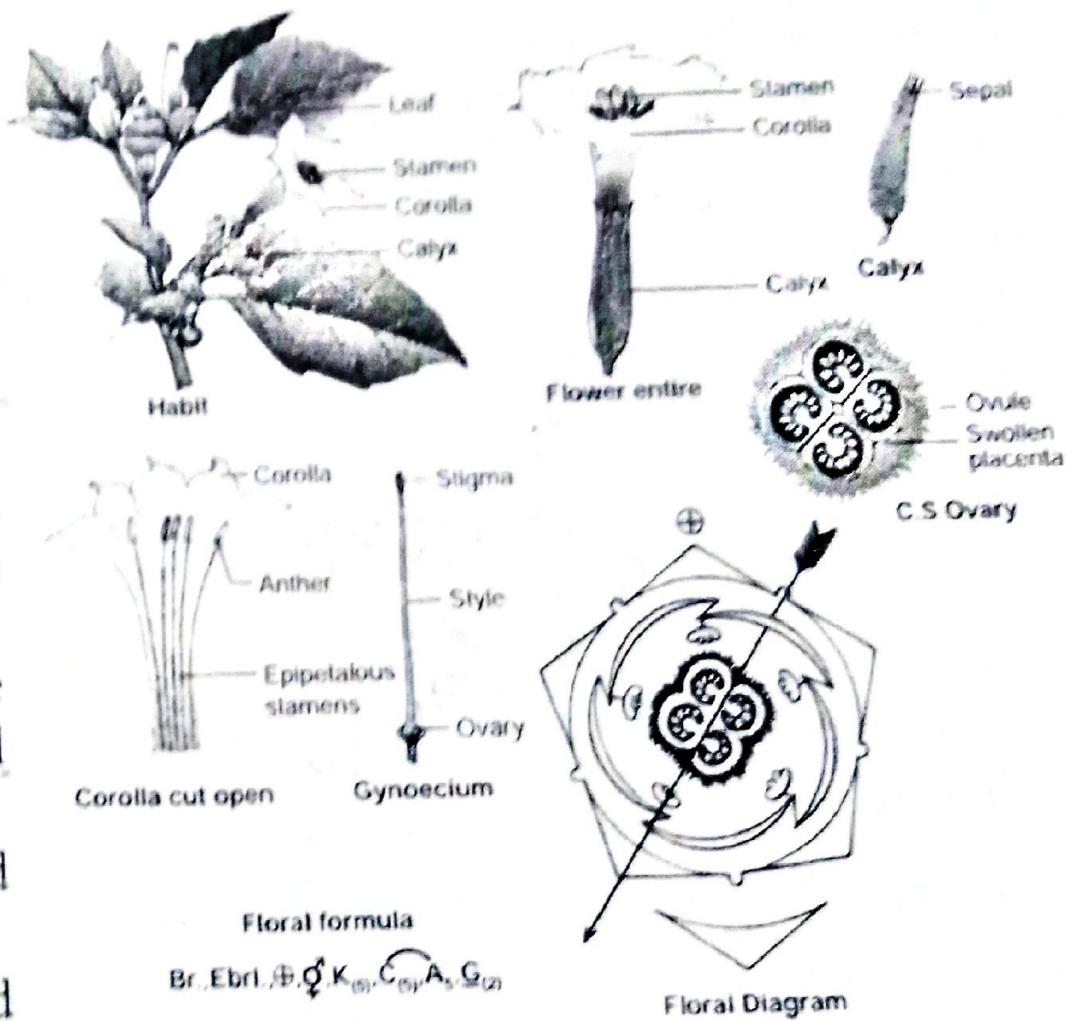


Figure 13 : *Datura metel*

V - Bio molecules-Nutrient test

Exercise: 14

Test for reducing sugar - Benedict reagent test

Aim:

To detect the presence of reducing sugar.

Basic Principle:

1. Aldoses and Ketoses are reducing sugars. Glucose is the reducing sugar and sucrose is the non-reducing sugar.
2. When reducing sugar is heated with an alkaline solution of Copper (II) sulphate (Benedict's solution) reduces Cu^{2+} into Cu^+ forming brick red precipitate of Copper (I) oxide.

Requirements:

Test tube, test tube stand, test tube holder, Samples for test- Fruit juices of apples/ banana/ leaves of onion, sugar cane extract, milk etc., Benedict's solution, spirit lamp, water bath.

Procedure:

1. Take 1 ml of sample solution in a clean test tube
2. Add 1 ml of Benedict's solution
3. Keep the test tube in the boiling water bath.
4. Appearance of brick red colour depends on concentration of reducing sugar.

Table:

Procedure	Observation	Inference
1ml of sample solution + 1ml of Benedict's solution, Heated	Appearance of brick red colour	Reducing sugar is present (Glucose is the reducing sugar)

Exercise: 15

Test for starch - Iodine test

Aim:

To detect the presence of starch in the given sample solution.

Basic Principle:

1. Starch is the storage polysaccharide of plants.
2. It consists of two components a. amylose (linear, unbranched polymer, soluble in water)
b. amylopectin (a branched polymer)
3. Amylose portion of starch reacts with Iodine (Potassium iodide) produces deep blue-black colour.

Requirements:

Test tube, Iodine solution, Extract of sample foodstuff (potato, rice, wheat or maize grits)

Procedure:

1. Take 1 ml of sample solution in a test tube.
2. Add 1 ml of Iodine (Potassium iodide).
3. Appearance of blue-black colour.

Table:

Procedure	Observation	Inference
1ml of sample solution + 1ml of Iodine solution	Appearance of deep blue-black colour	Starch is present

Exercise: 16**Test for protein - Biuret test****Aim :**

To detect the presence of proteins.

Basic Principle:

1. Proteins are polymer of amino acids. (Polypeptide).
2. Amino group of one amino acid binds with carboxylic group of another amino acid to form peptide bond. (NH-CO linkage)
3. In alkaline medium CuSO_4 reacts with peptide bond and gives a purple colour .
4. All proteins do not contain the same amino acids, and hence they do not respond to all colour reactions. (Biuret test is for peptide bond in the molecule of a protein, xanthoproteic test is specific for protein containing aromatic amino acids).

Requirements:Test tube, NaOH , CuSO_4 solution, milk/albumin of egg / gram seed extract.**Procedure:**

1. Take 2 ml of sample solution.
2. Add 1 ml of sodium hydroxide solution.
3. Add 1 or 2 drops of 1% copper (II) sulphate and mix it well.
4. Appearance of Purple colour (Increase with increase in concentration)

Table:

Procedure	Observation	Inference
2 ml of sample solution + 1 ml of Sodium hydroxide + 1 or 2 drops of 1% Copper (II) sulphate and mix it well.	Appearance of Purple colour	Protein is present

Exercise: 17

Test for Lipids – Saponification test

Aim:

To detect the presence of fats (lipid) in different plants and animal materials.

Basic Principle:

1. Lipids are esters of fatty acid and alcohol
2. Lipids are not soluble in water and soluble in organic solvent like benzene, ether and chloroform.
3. Major groups of lipids are triglycerides, phospholipids, Steroids and Waxes.
4. Soapy appearance due break down of ester bonds by NaOH.

Requirements:

Test tubes, test tube stands, NaOH, oil/ghee/butter.

Procedure:

1. Take 1 ml of sample solution in a test tube.
2. Add 1 ml of 5% NaOH and mix it well.
3. Appearance of soapy solution.

Procedure	Observation	Inference
1 ml of sample solution + 1ml 5% NaOH solution and mix it well.	Appearance of Soapy solution	Lipid is present

Exercise: 19**Paper chromatography experiment****Aim:**

To separate and study the photosynthetic pigments (chloroplast pigments) by paper chromatography method.

Requirements:

Fresh spinach leaves, chromatography paper (whatman No.1), a wide long test tube, a split sand etc.,

Procedure:

1. Grind a few spinach leaves with little fine sand and about 5 ml of acetone in a mortar and pestle. Filter it to get acetone extract of the leaf pigments.
2. Take a narrow strip of chromatographic paper (Whatman No.1). Cut one end of the strip into a narrow notch.
3. Put a drop of the pigment extract in the middle of the strip near the notch with the help of capillary tube. Allow the drop to dry and repeat till four or five drops are placed on the paper.
4. Take the test tube and pour about 5 ml of ether acetone solvent (9 ether : 1 acetone) in it. Now hang the pigment extract loaded chromatographic strip in the test tube with the help of a split cork, in such a way that the loading spot lies about 1 cm above the solvent level.
5. Make the cork air tight and place the test tube undisturbed for some time, when solvent rises about 3/4th of the strip, take out the strip carefully and let it dry.

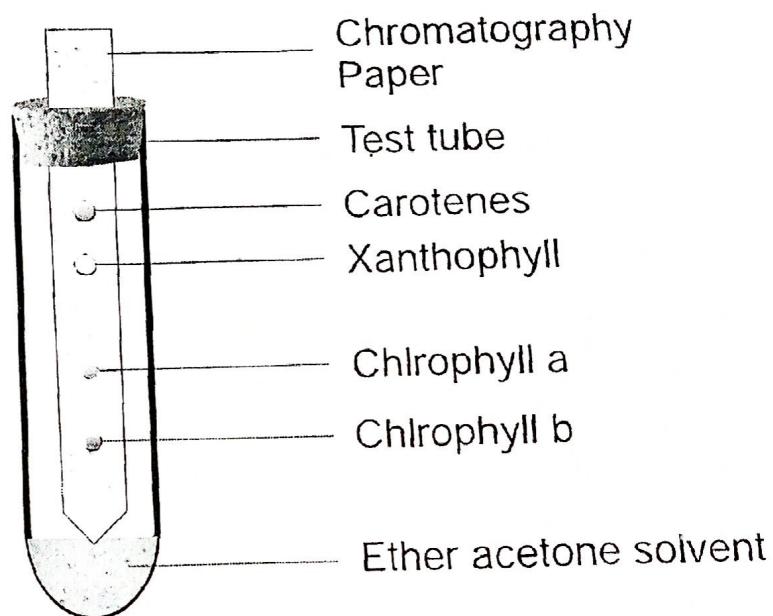


Figure 15 : Paper chromatography experiment

Observation:

After one hour observe the chromatographic paper. The Photosynthetic pigments being separated into four distinct bands. Different leaf pigments can be identified by their colours.

Carotene

Xanthophyll

Chlorophyll a

Chlorophyll b

Yellow Orange

Yellow

Bluish Green

Greenish Yellow

Inference:

Photosynthetic pigments chlorophyll b, chlorophyll a, xanthophyll and carotenes are separated on the chromatographic paper. Presence of different photosynthetic pigments in chloroplast is proved.

Exercise: 20

Wilmott's bubbler experiment

Aim :

To determine rate of photosynthesis by Wilmott's bubbler

Requirements :

Wilmott's bubbler apparatus, Hydrilla twig, water.

Procedure :

1. Fill the bottle with water and insert Hydrilla twig into the wider part of the tube
2. Hydrilla plant should be cut inside the water to avoid entry of air bubbles
3. Fix the tube with jar which acts as water reservoir
4. Keep the apparatus in sunlight
5. Count the bubbles when they are in same size.
6. Repeat the experiment in different light intensity.

Observation :

When there is an increase in photosynthesis, bubble count also increased.

Inference :

Rate of photosynthesis increases with increase of light intensity is proved

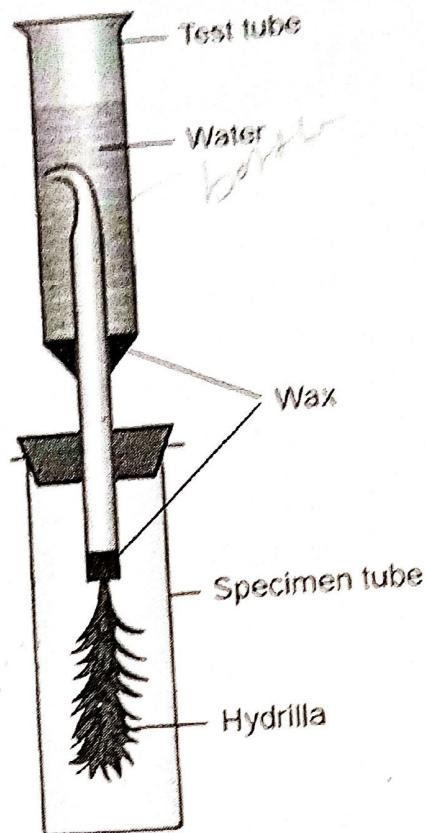


Figure 16 : Wilmott's bubbler

Exercise: 21

Experiment to demonstrate the production of CO₂ in aerobic respiration.

Aim:

To prove carbon dioxide is released by germinating seeds during respiration.

Requirements:

A conical flask, cork, beaker, a twice bent glass tube, a small test tube, thread, water KOH, germinating seeds of bean / gram/ groundnut seeds.

Procedure :

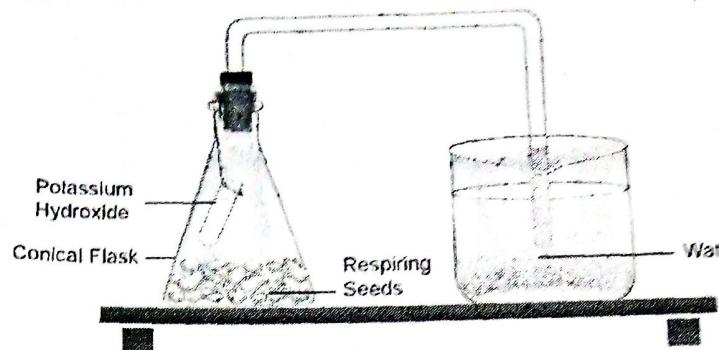


Figure 17: Demonstration of production of CO_2 during aerobic respiration

1. Take a definite quantity (i.e 10 gm) of germinating seeds of bean/gram/groundnut in the conical flask and hang a small test tube containing Potassium hydroxide (KOH) crystal inside the flask with the help of a thread.
2. Introduce one end of the bent glass tube into the conical flask through the cork. Dip the free end of the tube in a beaker containing water.
3. Make the apparatus air tight and fix the apparatus with the help of a stand.
4. Note the initial level of water in the bent glass tube and keep the apparatus undisturbed.

Observation :

After two hours the level of water rises in the glass tube.

Inference :

Carbon dioxide released by the germinating seeds is absorbed by KOH solution. It creates vacuum, to fill up the vacuum water raised in the tube. Liberation of carbon dioxide during respiration by germinating seeds is proved.