CHAPTER NINE

9.0 ANATOMY OF ROOTS

CHAPTER OBJECTIVES

By the end of this chapter, students should be able to:

(i) identify two types of root as tap and fibrous roots;
(ii) describe the transverse section of a dicot root;
(iii) differentiate between the three regions of a root tip;
(iv) state five functions of roots in plants;
(v) identify plant nutrients and the effects of their deficiency;
(vi) understand the differences in structure between primary and secondary tissues in dicot root.

9.1 General Information

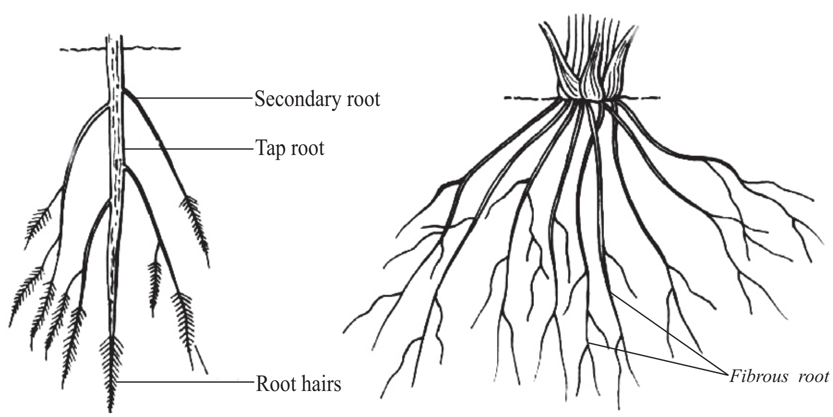
As soon as a seed begins to grow, it puts out its first root to draw water and nutrients from the soil. Other roots soon branch out from this first root, adding length and surface areas to the root system. The root is the simplest organ of a plant, but it has several important functions. In most plants, a root anchors the plant to the ground. If you have ever tried to pull a weed out of the soil, you know how strong a hold roots can have. Also, a root absorbs water and minerals from soil and then transports them upward to the rest of the plant. A third function of the root is storage of food. Plants often produce more food than they can use. This excess food is often stored in roots in the form of sugars or starches. Beets, carrots, and radishes are a few examples of roots that store food.

9.2 Types of Root system

The first root to appear when a seed germinates is a primary root called radical. In dicots, this root grows down into the soil and becomes both strong and thick. This mature root in a dicot is called a taproot.

An example of a taproot is shown in Figure 9.1a. A carrot is an edible taproot. Some plants in dry areas have taproots almost 20 meters long. These long roots enable the plants to reach deep sources of water and to survive during hot, dry weather.

As the plant grows, the taproot develops little side branches called secondary roots. These roots help hold the plant more firmly in the soil and increase the root's surface area for absorbing water. A taproot and its smaller secondary roots make up a taproot system. In monocots, such as grass and corn, the primary root is usually short-lived, and the root system does not develop from this root. Instead, numerous long, thin roots grow from the stem and spread out through the soil. These roots and their side branches are fibrous roots. Figure 9.1b shows that no single root is more prominent than the others in a fibrous root system. Fibrous roots do not grow as deep as taproots. However, they do grow rapidly at a shallow depth and cover a fairly large area. Fibrous root absorb water and dissolved minerals very efficiently because the total surface area of the root system is quite large. One scientist estimated the surface area of the roots of a single rye plant. Including the roots' hairlike projections, the surface area added up to 639 square meters larger than the area of a basketball court. Fibrous roots also help hold soil particles together and, as a result, are effective in preventing soil erosion. Some plants have thick fibrous roots that store food just as taproots do. The sweet potatoes that people eat are an example of these fleshy fibrous roots.



(a) ***Tap root*** (b) ***Fibrous root***

Figure 9.1: *Tap root and fibrous roots.*
Source: (Ewusie, 1973).

Note: (i) Type of roots refers to still roots, prop roots and storage roots etc.

Note: *(ii) All plants first form a primary root and in most plants, including the gymnosperms, primitive dicotyledons and eudicotyledons, this persists as a taproot. The lateral roots arise from it at various points. In the monocotyledons the primary root dies and adventitious roots are formed which grow out of the stem. These give rise to a mass of fine, fibrous roots, all of similar size and prominence.*

9.3 The Root tip and its Growth

If one were to examine a thin section of a young carrot root under a microscope, you would see root cells similar to those shown in Figure 9.2. Notice that the youngest cells of the tip are very small, while the older cells of the upper part of the root are much longer. Biologists divide roots into three main regions of growth (regions of cell division, elongation and maturation), although the boundaries between the regions are not distinct.

- **Region of cell division**

At the tip of the root is the root cap. This mass of cells acts like a shield protecting the apical meristem, where cell division occurs. As cells in the apical meristem divide, new cells are added both to the elongation region above and to the root cap below. The new cells of the root cap replace cells that are worn away as the root grows and pushes through rough soil.

- **Region of Elongation**

Above the apical meristem, in the region of elongation, cells do not divide. Instead, the newly formed cells grow longer. As the cells lengthen, they push the root tip downward, extending it farther into the soil. The enlargement of the cells in this region is responsible for most of the lengthening or primary growth of the root.

- **Region of Maturation**

Above the region of elongation, fully grown cells differentiate or change in form so that they can carry on special tasks. These specialised cells form various tissues in the mature root. Some cells become tubelike and transport water or food through the root. Other specialised cells absorb water from the ground surrounding the root.

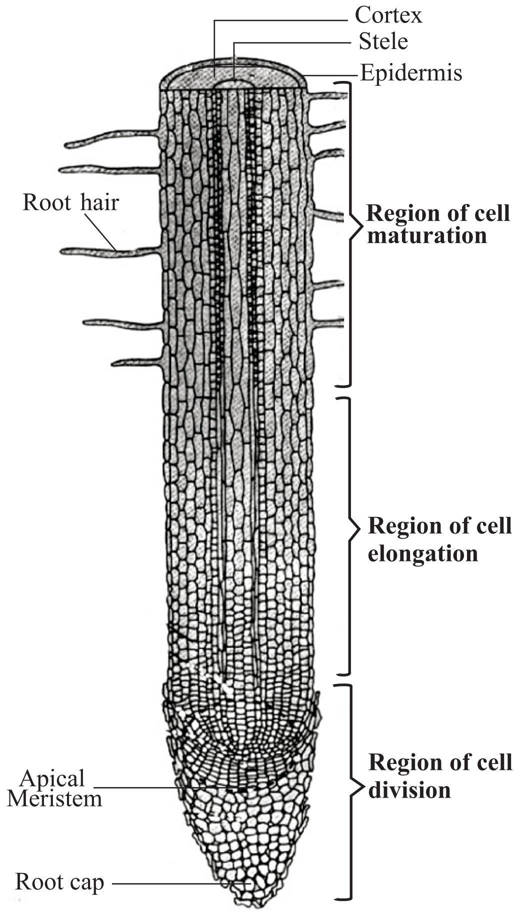


Figure 9.2: *Primary tissues in a dicot root (* Longitudinal section).

Source: (Campbell, 1993).

9.4 Structure of a Young Dicot Root

The root is a plant organ which usually attaches the plant firmly to the soil and absorbs water and mineral salts from it. The diagram in Figure 9.3 illustrates the structure of a young dicotyledonous root. It is bounded on the outside by a layer which is made up of tabular closely fitted cells. This is called the piliferous layer and it is protective in function. The young root is perforated at intervals, some epidermal cells are elongated at right angles to the axis of the root. These extension of epidermal cells are called root hairs. Root hairs are important in the absorption of water and mineral salts from the soil.

Within the epidermis is the cortex which is made up of parenchymatous cells. The cortex is usually many cell layers deep. Food or water may be stored in the cortex. It is bounded on the inside by a single layer of special cells. The layer is the endodermis and its cells are distinctive in that they are thickened on the inner and lateral walls only.

The tissues that occupy the centre of the root constitute the vascular or conducting tissues. These are the food-conducting phloem tissue and xylem tissues, which are concerned with the conduction of water and mineral salts within the plant. The xylem occurs as a stellate (star-shaped) structure, i.e. it has radiating arms. The tips of the arms are occupied by young xylem vessels called protoxylem and metaxylem. This arrangement in which the protoxylem is on the inside is called exarch. The xylem is said to be diarch, tetrarch, or polyarch depending on whether there are 2, 4 or many xylem arms i.e. protoxylem groups. The phloem occurs in groups which alternate with the protoxylem groups. There are usually the same number of phloem groups as those in protoxylem .

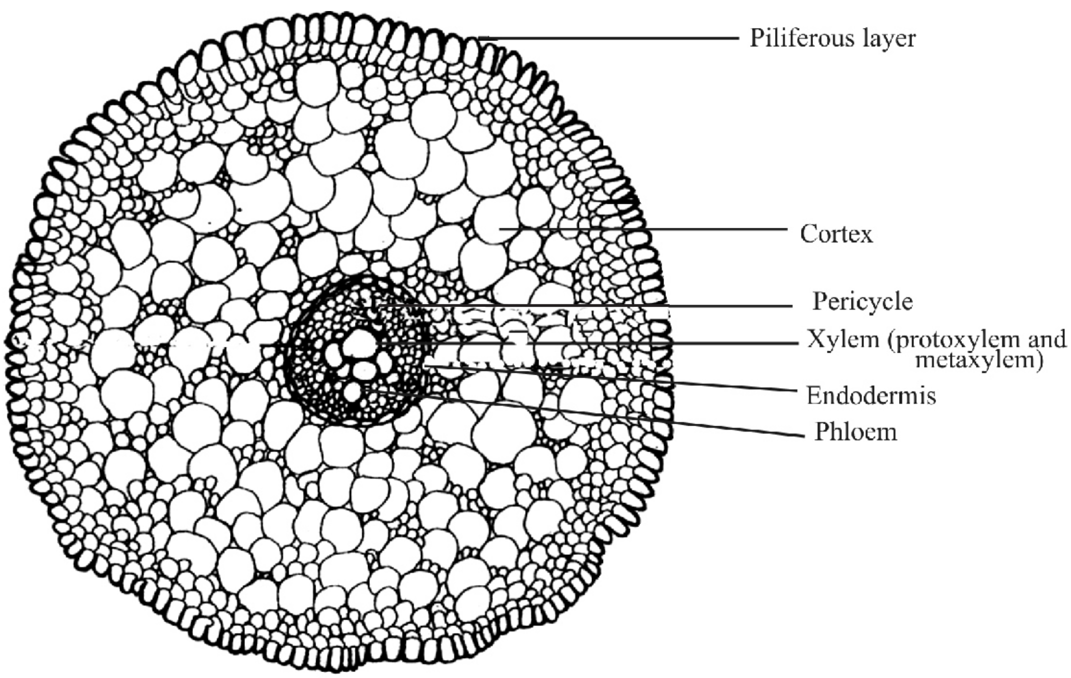


Figure 9.3: *Structure of a young dicot root.*
Source: (Walter, (1988).

9.5 Functions of Root

Roots anchor a plant to the soil and absorb water and dissolved nutrients from it. How does a root go about the job of absorbing water and mineral salts from the soil? Water does not just "soak" into the root from soil although it might seem so. It takes energy on the part of the plant to absorb water. The explanation of this process begins with a description of soil and plant nutrients.

(i) Uptake of Plant Nutrients

Soil help plants to grow and function properly. Soil is a complex mixture of sand, silt, clay, air and bits of decaying animal and plant tissues (organic matter). Soil in different places and at different depth contains varying amounts of these ingredients. Sandy soil, for example, is made of large particles that retain few nutrients, whereas the finely textured silt and clay soils of the South - East and South-South of Nigeria are high in nutrients. These nutrients define the soil and determine, to a large extent, the kinds of plants that can grow in it.

To grow, flower, and produce seed, plants require a variety of inorganic nutrients in addition to carbon (iv) dioxide and water. The most important of these nutrients are nitrogen, phosphorus, potassium, magnesium, and calcium. The functions of these essential nutrients within a plant are described in Table 9.1. These nutrients are located in varying amounts in the soil and are drawn up by the roots of a plant. In addition to these essential nutrients are trace elements. These elements are required in small quantities to maintain proper plant growth. Trace elements include sulphur, iron, zinc, molybdenum, boron, copper, manganese, and chlorine. Large amounts of trace elements in the soil can be poisonous.

Table 9.1: Essential Plant Nutrients

Nutrient element	Role in plant	Effect of deficiency
Nitrogen	Proper leaf growth and colour; synthesis of amino acids, proteins, nucleic acids and chlorophyll	Stunted plant growth; pale yellow leaves
Phosphorus	Synthesis of DNA; development of roots, stems, flowers and seeds	Poor flowering; stunted growth
Potassium	Synthesis of proteins and carbohydrates; development of roots, stem and flowers; resistance to cold and disease.	Weak stems and stunted roots; edges of leaves turn brown
Magnesium	Synthesis of chlorophyll	Thin stems; mottled, pale leaves
Calcium	Cell growth and division; cell wall structure; cellular transport; enzyme action	Stunted growth; curled leaves.

Source: (Taylor, Green and Stout, 1997).

(ii) Active Transport of Mineral Salts

The cell membranes of root hairs and other cells in the root epidermis contain active transport proteins. These proteins use Adenosine Triphosphate ATP (an energy source) to pump mineral ions from the soil into the plant. The high concentration of mineral ions in the plant cells cause water molecules to move into the plant by osmosis, as shown in Figure 9.4.

Osmosis is the movement of water across a membrane toward an area where the concentration of dissolved minerals is higher. By using active transport to accumulate ions from the soil, cells of the root epidermis create conditions under which osmosis causes water to 'follow' those ions and flow into the root. Note that the root does not actually pump water. But the pumbing of dissolved minerals into its own cells, the end result is almost the same - the water moves from the epidermis through the cortex into the vascular cylinder.

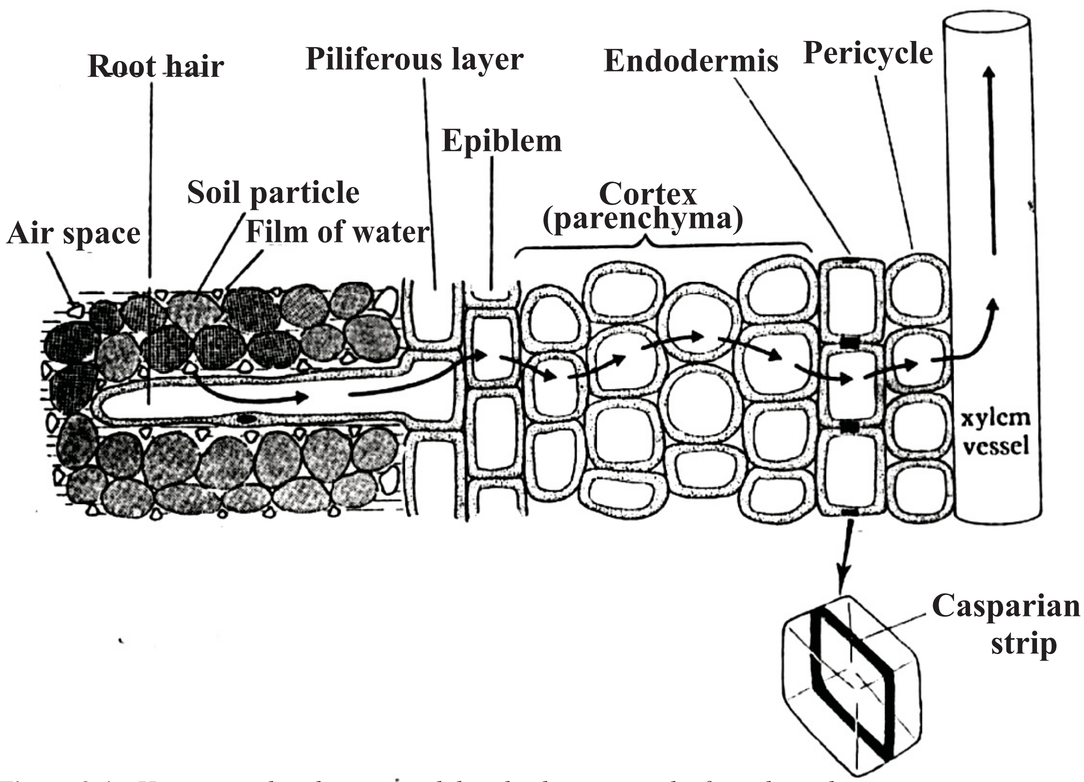


Figure 9.4: *How roots absorb water and dissolved nutrient salts from the soil.*
Source: (Roberts,1976).

(iii) Movement into the Vascular Cylinder

Both osmosis and active transport cause water and mineral salts to move from the root epidermis into the cortex. From there, the water and dissolved minerals pass the inner boundary of the cortex and enter the endodermis. This process is shown in Figure 9.4.

The endodermis encloses the vascular cylinder and stretches up and down the entire length of the root, like a cylinder. It is composed of many individual cells, each shaped a bit like a brick. Each of these cells is surrounded on four sides by a water-proof strip called a casparian strip. To imagine what the casparian strip looks like, think of a brick with a thick rubber band stretched around it. The rubber bands stick together like mortar between the bricks. Imagine many of these bricks placed edge to edge to build a cylinder. When a root is viewed in cross section, the endodermis forms a circle.

(iv) Osmosis

Water moves into the vascular cylinder by osmosis, since there is a one-way passage of materials into the vascular cylinder in plant roots. Water and mineral salts cannot pass through the waxy casparian strip, once they pass through the endodermis, they are trapped in the vascular cylinder. Hence, there is a continuous flow of water and mineral salts up the cylinder (Figure 9.4).

(v) Root Pressure

Why do plants "need" a system that ensures the one-way movement of water and mineral salt? That system is how the plant generates enough pressure to move water out of the soil and up into the body of the plant. As mineral salts are pumped into the vascular cylinder, more and more water follows by osmosis, producing a strong pressure. If the pressure were not contained, roots would expand as they are filled with water.

Instead, constrained by the casparian strip, the water has just one place to go and that is up. Root pressure, produced within the cylinder by active transport, forces water through the vascular cylinder and into the xylem. As more water moves from the cortex into the vascular cylinder, more water in the xylem is forced upward through the root into the stem. Root pressure is the starting point for the movement of water through the vascular system of the entire plant. But it is just the beginning. Once you have learned about stems and leaves, you will see how water and other materials are transported within a plant.

9.6 Primary Tissues in Dicot Root

The primary tissues of the root are involved in primary growth - or lengthening. They include the apical meristem and the tissues it produces, namely, the epidermis, the vascular tissues, and the parenchyma. Study the cross section of the root in Figure 9.5.

Epidermis consists of a single layer of cells that protects the outer surface of the root. Similar epidermal cells protect the surface of other parts of the plant. In stems and leaves, the epidermis prevents water loss. But in the region of root hairs, the epidermis of roots must allow for absorbing water and its dissolved mineral salts. In roots, epidermal cells have thread-like projections called root hairs which aid in water absorption. Millions of root hairs increase the surface area of the root, enabling the root to absorb more water. If you were to examine a young root, you would see that root hairs give the root a fuzzy appearance, as shown in Figure 9.2. Root hairs are most dense just above the root tip.

Vascular tissues are grouped together at the centre of the root. These tissues extend from the roots, through the stem, and up to the leaves. Vascular tissues are composed of two kinds of tissue: xylem and phloem. In the cross section shown in Figure 9.5, the xylem looks like a thick "plus" sign. The phloem is located between the arms of the "plus" sign.

Xylem is made of thick-walled cells that are fused end-to-end. Often the end of xylem cells dissolve, leaving continuous hollow tubes through which water and dissolved mineral salts move upward from the roots. These cells do not live very long. Phloem is made of thin-walled, living cells. The ends of phloem cells are perforated, making the cells sieve-like. Like xylem cells, phloem cells form continuous tubes from root to leaves. Food manufactured in the leaves moves down to the roots through phloem. Also, food stored in the roots moves back up through the phloem to the rest of the plant.

Parenchyma cells are found between the epidermis and vascular tissues. Notice that thin-walled pericycle cells form a thin layer around the xylem and phloem. Pericycle cells are part of the parenchyma. The pericycle together with the vascular tissues make up the vascular cylinder, which provides rigidity for the root. Also, root branches can develop from pericycle tissues.

Surrounding the vascular cylinder and inside the epidermis is another layer of parenchyma cells, called the cortex. Food is stored in the cortex. Cells of the cortex are not packed tightly together. As a result, water flows easily from the root hairs around these cells toward the xylem. Water and dissolved substances must then pass through the innermost layer of the cortex - the endodermis. This layer of thick, waxy cells surrounds the vascular cylinder. The cell membranes of these endodermal cells regulate the passage of all dissolved substances into the vascular cylinder. Thus, these cells control which substances are transported to the rest of the plant.

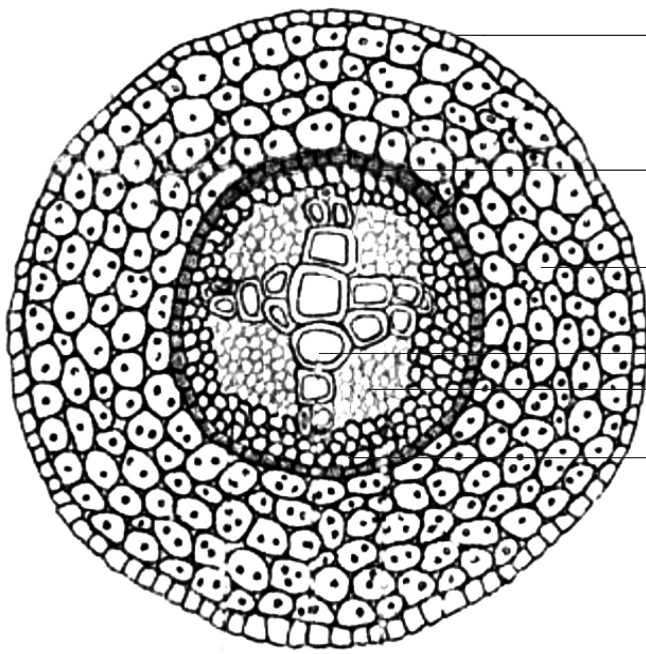


Figure 9.5: *Primary tissues in a dicot root (Transverse section).*
Source : (Slesnick. *et. al,* 1985).

Note: Primary growth of a root
Mitosis is concentrated in the zone of cell division, where the apical meristem and its products, the three primary meristems, are located. The apical meristem also maintains the root cap by generating new cells that replace those that are sloughed off. If the apical meristem is damaged its quiescent centre is activated and restores the meristem by means of cell division. Most cell growth takes place in the zone of cell elongation. Cells become functionally mature in the zone of cell differentiation, which is sometimes known as the root hair zone, because root hairs extend from epidermal cells.

9.7 Secondary Root Tissues

Another meristem tissue is found in the roots of dicots. This tissue is called lateral meristem because it produces an increase, laterally (in thickness) rather than in length. Xylem and phloem that the lateral meristem produces are secondary tissues. Cells of lateral meristem, also known as vascular cambium, develop as a root grows older. They form a layer between the xylem and phloem, as shown in Figure 9.6. Cells of the vascular cambium divide continually and live as long as the root lives.

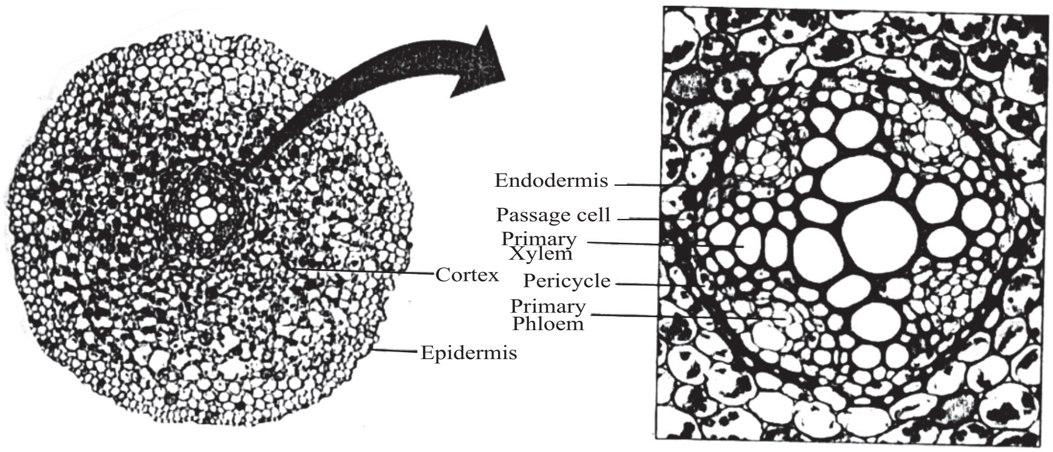


Figure 9.6: *Secondary tissues in a dicot root*

Source: (Stern, 2000).

9.8 Summary

A root is the downward part of the plant that grows through the soil designed to draw water and dissolve nutrients from the soil. There are two major types of root systems found in plants viz; taproot system and fibrous root system. Taproots and its lateral branches are found mainly in trees like mango and orange while fibrous roots exist in plants such as maize, oil palm and coconut. The major functions of root include: absorption of water and dissolved nutrients from the soil, anchoring of the plant to the soil, storage of excess food, etc. Three regions of root growth are identified: region of cell division, region of elongation and region of maturation with dermal, vascular and ground tissues as the root growth tissues. The surface of the epidermal sub-system is covered by root hairs resulting in large surface area through which water can enter. The root cap protects the root as it forces its way through the soil. As the root grows, the root cap forces its way through the soil as it secretes a slippery substance that lubricates the progress of the root through the soil. The cortex is made up of parenchymatous cells within the epidermis. Food or water may be stored in the cortex. Phloem and xylem tissues conduct water and dissolved mineral salts within the plant. Inorganic nutrients within the soil like nitrogen, phosphorus potassium, magnesium and trace elements like sulphur, iron, zinc, molybdenum, *boron, copper, manganese, and chlorine dissolved in the soil are drawn up by the roots of the plant. Active transport proteins in cell membranes of root hairs and other cells in the root epidermis absorb mineral ions from the soil and flow into the root. This process is called osmosis. Both osmosis and ATP cause water and dissolved mineral salt to flow from the root epidermis into the cortex resulting in root pressure. Root pressure forces water through the vascular cylinder and into the vascular system hence more water in the xylem is forced upward through the root into the stem.*

9.9 Evaluation

Having read this chapter, answer the following questions:

1. (a) What is a root?
   (b) Describe two types of roots you have studied.

2. Use a labeled diagram to describe the structure of a young dicot root.

3. List and explain the functions of roots in plants.

4. Carefully explain the process of osmosis in the transportation of soil nutrients by roots.

5. Differentiate between the three regions of a root tip.

6. List the plant nutrients you have studied and state the effect of its deficiency.

7. Name the primary tissues of a root and state their functions.

8. List the secondary tissues of a root and state their functions.

9. Explain the movement of water by osmosis and root pressure into the vascular cylinder.

10. (a) What are secondary root tissues?
    (b) State the function of secondary root tissues.

9.10 TUTORIAL QUESTIONS

Fill in the gap in the sentences provided below:

1. As soon as a seed begins to grow, it puts out its first root to draw ___________ and ___________ from the soil.

2. Roots contain cells from the three tissue systems: dermal, vascular and___________ tissues.

3. As the root grows, the root cap secretes a __________ substance that lubricates the progress of the root through the soil.

4. The process by which unspecialised cells change to become specialised in structure and function is known as _____________ .

5. _____________ are important in the absorption of water and mineral salts from the soil.

6. Within the epidermis is the cortex which is made up of___________ cells.

7. The tip of the arms are occupied by young xylem vessels called _____________.

8. The cell membrane of root hairs and other cells in the root epidermis contain _________________.

9. The high concentration of mineral ions in the plant cells causes ______________ to move into the plant by osmosis.

10. Both osmosis and ______________ cause water and mineral salts to move from the root epidermis into the cortex.

11. Water moves into the vascular cylinder by ___________.

12. Epidermis consists of a single layer of cells that protects the ____________ of the root.

13. _______ cells protects the surface of other parts of the plants.

14. In stems and leaves, the epidermis prevents ___________.

15. In roots, epidermal cells have thread - like projections called ___________ which aid in water absorption.

16. Xylem is made of ______________ that are fused end-to-end.

17. Pericycle cells are part of the ________________.

18. The pericycle together with the vascular tissues make up the ____________, which provides rigidity for the root.

19. Surrounding the vascular cylinder and inside the epidermis is another layer of parenchyma cells called ___________.

20. Xylem and phloem that the lateral meristem produces are ______________.

CHAPTER TEN

10.0 MICROSCOPY

CHAPTER OBJECTIVES

By the end of this chapter, students should be able to:

(i) list and explain the types of microscopes;

(ii) identify the parts of light microscope and transmission electron microscope;

(iii) draw and label parts of a light microscope;

(iv) explain the process of focusing the light microscope;

(v) list the rules for making effective use of light microscope;

(vi) compute the high power diameter of light microscope;

(vii) practically use a microscope to observe specimens.

10.1 General Information

When students think of laboratory equipment, one of the first things that come to mind is the microscope. Without microscopes, very little would be known about cells. Our present vast knowledge of cells and all aspects of biological investigations associated with them is directly related to the development of these instruments. Light microscopes can produce clear images of objects at a magnification of about 1,500 times. Thinly sliced materials can be viewed with compound microscopes. Compound light microscopes allow light to pass through the specimen and use two lenses to form an image. Light microscopes make it possible to study dead organisms and their parts, and to observe some tiny organisms and cells while they are still alive. Microscopes, such as the light microscope in Figure 10.1(a) are devices that produce magnified images of objects that are too small to see with the unaided eye. Light microscopes produce magnified images by focusing visible light rays. Electron microscope produce magnified images by focusing beams of electrons.

The difference between electron and compound light microscopes, however, is not simply a matter of magnification; it is also the electron microscope's ability to show detail. The electron microscope has a resolving power. The use of electron rather than light gives electron microscope a much greater resolving power. Since the first microscope was invented, microscope manufacturers have had to deal with two problems: what is the instrument's magnification? that is, how much larger can it make an object appear compared to the object's real size? And how sharp an image can the instrument produce?

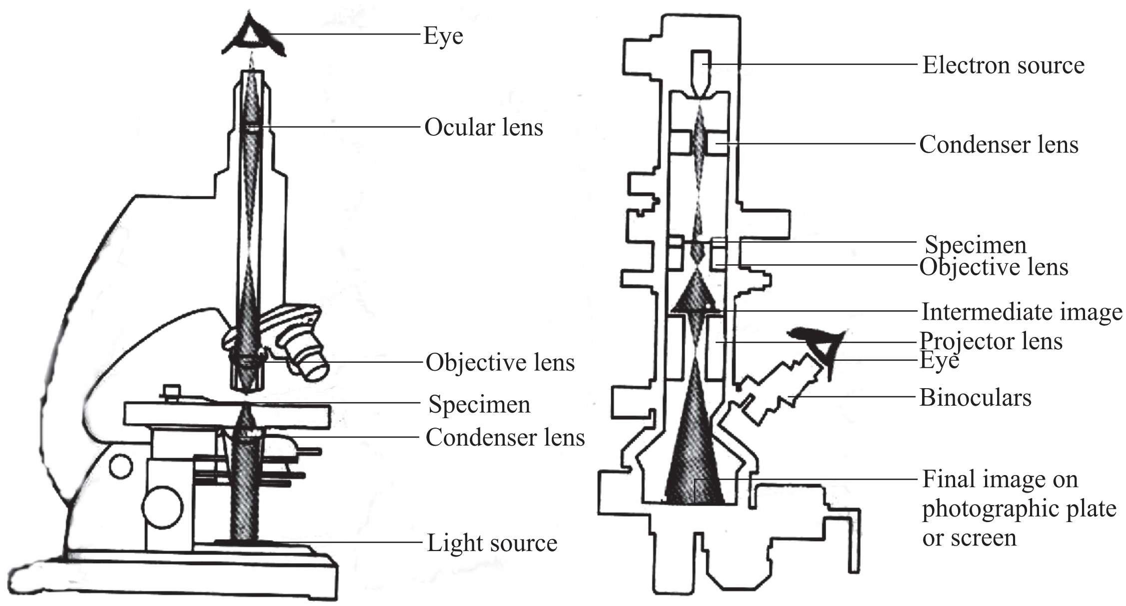
10.2 Light Microscope

The most commonly used microscope is the light microscope (Figure 10.1a). Light microscopes are of two basic types: compound microscope, which require the material being examined to be sliced thinly enough for light to pass through, and dissecting microscopes (stereomicroscopes), which allow three dimensional viewing of opaque objects. The best compound microscope in use today can produce useful magnifications of up to 1,500 times under ideal conditions. Many dissecting microscope used in teaching laboratories magnify up to 30 times, but higher magnifications are possible with both types of microscopes. Magnification of more than 1,500 times, however, are considered "empty" because resolution (the capacity of lenses to aid in separating closely adjacent tiny objects) does not improve with magnification beyond a certain point. Light microscopes will continue to be useful particularly for observing living cells, into the foreseeable future. Biologists have developed techniques and procedures to make light microscopes more useful. Chemical stains, also called dyes, can show specific structures in the cell. Fluorescent dyes have been combined with video cameras and computer processing to produce moving three-dimensional images of processes such as cell movement.

10.3 Electron Microscopes

Since the 1950s, the development of high resolution electron microscopes has resulted in observation of much greater detail than is possible with light microscopes. Resolution is the minimum distance between two objects at which they can still be seen, or resolved, as two separate objects. Electron microscopes use a beam of electrons (as shown in Figure 10.1b) that is magnified and focused on a photographic plate by means of electromagnets. The Transmission Electron Microscope (TEM) is analogous to the compound light microscope. The scanning electron microscope is analogous to the dissecting light microscope.

Electron microscopes are of two basic types: the transmission electron microscope and scanning electron microscope. Transmission electron microscope can produce magnification of 200,000 or more times, but the material to be viewed must be sliced extremely thin and introduced into the column's vacuum. Scanning electron microscope usually do not attain such high magnifications (3,000 to 10,000 times is the usual range), but thick objects can be observed when a scanner makes the object visible on a cathode tube like a television screen. The techniques for observation with electron microscope have become so refined that even preserved materials can appear exceptionally lifelike, and high resolution three-dimensional images can be obtained.



(a) Light Microscope **(b) Electron Microscope**

Figure 10.1: *The Light Microscope and Transmission Electron Microscope.*
Source: (Campbell, 1993).

Note:
(i) In light microscopy, light is focused on a specimen by a glass condenser lens; the image is then magnified by an objective lens and an ocular lens, for projection on the eye or photographic film.
(ii) In electron microscopy, a beam of electrons (top of the microscope) is used instead of light, and electromagnets instead of glass lenses. The electron beam is focused on the specimen by a condenser lens; the image is magnified by an objective lens and a projector lens, for projection on a screen or photographic film.

10.4 Use of the Compound Light Microscope

As mentioned, the name compound light microscope indicates that it uses light and both ocular and objective lenses to view an object.

10.5 Identifying the Parts

After your instructor has explained how to carry a microscope, obtain one from the storage place and place it securely on the table. Identify the following parts on your microscope, and label them.

(i) **Eyepieces (ocular lenses):** what is the magnifying power of the ocular lenses on your microscope?

(ii) **Body tube:** Holds nosepiece at one end and eyepiece at the other end; conducts light rays.

(iii) **Arm:** Support upper parts and provides carrying handle.

(iv) **Nosepiece:** Revolving device that holds objectives.

(v) **Objectives** (objective lenses)

(a) Scanning power objective

This is the shortest of the objective lenses and is used to scan the whole slide. The magnification is stamped on the housing of the lens. It is a number followed by an X. what is the magnifying power of the scanning lens on your microscope?

(b) Low-power objective: This lens is longer than scanning lens and is used to view objects in greater detail. What is the magnifying power of the low-power objective lens on your microscope?

(c) High-power objective: If your microscope has three objectives lenses, this lens will be the longest. It is used to view an object in even greater detail. What is the magnifying power of the high-power objective lens on your microscope?

(d) Oil immersion objective: (on microscopes with four objective lenses): Hold a 95x (to 100x) lens and is used in conjunction with immersion oil to view objects with greatest magnification. Does your microscope have an oil immersion objective? If this lens is available, your instructor will discuss its use when the lens is needed.

(vi) **Coarse-adjustment knob:** knob used to bring object into approximate focus; used only with low-power objective.

(vii) **Fine-adjustment knob:** knob used to bring object into final focus.

(viii) **Condenser:** Lens system below the stage used to focus the beam of light on the object being viewed.

(ix) **Diaphragm or diaphragm control level:** controls amount of illumination used to view the object.

(x) **Light Source:** an attached lamp that directs a beam of light up through the object.

(xi) Base: the flat surface of the microscope that rests on the table.

(xii) Stage: holds and supports microscope slides.

(xiii) Stage clip: hold slides in place on the stage.

(xiv) Mechanical stage: (optional): a moveable stage that aids in the accurate positioning of the slide. Does your microscope have a mechanical stage?

(xv) Mechanical stage control knobs (optional): Two knobs that are usually located below the stage. One knob controls forward/reverse movement, and the other controls right/left movement.

10.6 Focusing the Microscope - Lowest power

(i) Turn the nosepiece so that the lowest power lens is in straight alignment over the stage.

(ii) Always begin focusing with the lowest power objective lens (4 x [scanning] or 10 x [low power]).

(iii) With the coarse-adjustment knob, lower the stage (or raise the objectives) until it stops.

(iv) Place a slide on the stage, and stabilize it with the clips. (if your microscope has a mechanical stage, pinch the spring of the slide arms on the stage, and insert the slide). Center the slide as best you can on the stage or use the two control knobs located below the stage.

(v) Again, be sure that the lowest objectives is in place. Then as you look from the slide, decrease the distance between the stage and the tip of the objective lens until the lens comes to an automatic stop or is no closer than 3mm above the slide.

(vi) While looking into the eyepiece, rotate the diaphragm (or diaphragm control lever) to give the maximum amount of light.

(vii) Using the coarse-adjustment knob, slowly increase the distance between the stage and the objective lens until the object comes into view, or focus.

(viii) Once the object is seen, you may need to adjust the amount of light. To increase or decrease the contrast, rotate the diaphragm slightly.

(ix) Use the fine-adjustment knob to sharpen the focus if necessary.

(x) Practice having both eyes open when looking through the eyepiece, as this greatly reduces eyestrain.

10.7 Focusing the Microscope - Highest Power

Compound light microscopes are parfocal, that is, once the object is in focus with the lowest power, it should also be almost in focus with the higher power.

(1) Bring the object into focus under the lowest power by following the instructions in the previous section.
(2) Make sure that the letter e is centered in the field of the lowest objective.
(3) Move to the next higher objective (low power (10x) or high power (40x) by turning the nose-piece until you hear it click into place. Do not change the focus; parfocal microscope objectives will not hit normal slides when changing the focus if the lowest objective is initially in focus. (if you are on low power (10x), proceed to high power (40x) before going to the next step).
(4) If any adjustment is needed, use only the fine-adjustment knob, (Note: always use only the fine-adjustment knob with high power). On your drawing of the letter e, draw a circle around the portion of the letter that you are now seeing with high-power magnification.
(5) When you have finished your observations of this slide (or any slide), rotate the nosepiece until the lowest power objective clicks into place, and then remove the slide.

10.8 Rules for Effective use of Microscope

(1) The lowest power objective (scanning or low) should be in position both at the beginning and end of microscope use.
(2) Use only lens paper for cleaning lenses.
(3) Do not tilt the microscope when viewing a wet mount.
(4) Keep the stage clean and dry to prevent rust and corrosion.
(5) Do not remove parts of the microscope.
(6) Keep the microscope dust-free by covering it after use.
(7) Report any malfunctioning.

10.9 Total Magnification

Total magnification is calculated by multiplying the magnification of the ocular lens (eye piece) by the magnification of the objective lens.

Observation: Total Magnification

Calculate total magnification figures for your microscope, and record your findings in the table in Table 4.

Table 10.1: Total Magnification

Objectives	Ocular lens	Objective lens	Total magnification
Scanning power (if present)			
Low power			
High power			
Oil immersion (if present)			

10.10 Field of View

A microscope's field of view is the circle visible through the lens. The diameter of field is the length of the field from one edge to the other.

Observation: Field of View

Low power (10 x) Diameter of Field

(1) Place a clear, plastic ruler across the stage so that the edge of the ruler is visible as a horizontal line along the diameter of the low-power (not scanning) field. Be sure that you are looking at the millimeter side of the ruler.

(2) Estimate the number of millimeters, to tenths, that you can see along the field: _________ mm. (Hint: start with one of the millimeter markers at the edge of the field). Covert the figure to micrometers: _________ um. This is the low-power diameter of field (LPD) for your microscope in micrometers.

High power (40 x) diameter of field

(1) To compute the high-power diameter of field (HPD), substitute these data into the formular given:

(a) LPD = lower power diameter of field (in micrometers) = ______________

(b) LPM = low power total magnification = ____________________________

(c) HPM = high power total magnification = ____________________________

$$HPD = \frac{LPD \times LPM}{HPM}$$

$$HPD = (\quad) \times \frac{(\quad)}{(\quad)} = \underline{\qquad\qquad\qquad}$$

Conclusion

(i) How does low power or high power have a larger field of view (one that allows you to see more of the object)? ____

(ii) Which has a smaller field but magnified to a greater extent? __________
(iii) To locate small objects on a slide, first find them under low power; then place them in the center of the field before rotating to high power.

10.11 Depth of Field

The vertical distance that remains in focus at one time is called the depth of field.

Observation: Depth of Field

(i) Obtain a prepared slide with three or four coloured threads mounted together, or prepare a wet-mount slide with three or four crossing threads or hairs of different colours.
(ii) With low power, find a point where the thread or hairs cross. Slowly focus up and down. Notice that when one thread or hair is in focus, they seem blurred. Determine the order of the threads or hairs. Remember, as the stage moves upwards (or the objectives move downward), objects on top come into focus first.

Table 10.2. Order of Thread (or hairs)

Depth	Thread (or hair) color
Top	
Middle	
Bottom	

(iii) Switch to high power, and notice that the depth of field is more shallow with high power than with low power. Constant use of the fine-adjustment knob when viewing a slide with high power will give you an idea of the specimen's three-dimensional form. For example, viewing a number of sections allows reconstruction of the three dimensional structure as demonstrated in Figure 10.2.

10.12 Microscopic Examination: Temporary or Wet mount

(i) On a clean slide place a small drop of water. If the material is already in liquid, place a drop of the suspension on the slide.
(ii) Place the specimen in the drop of water (Figure 10.2b).
(iii) Cover the specimen with a cover glass by first placing one edge of the cover glass on the slide to touch the water and then lowering the opposite edge with a dissecting needle. This will minimise the trapping of air bubbles

(iv) Water should fill the space between the slide and cover glass. If excess water spreads over any other part of the slide, remove it by carefully blotting with a strip of paper towelling (Figure 10.2d).

If the preparation begins to dry during examination, place a small drop of water at the edge of the cover glass. Capillary forces will cause the water to move between the cover glass and slide. The slide need not be removed from the microscope stage during this operation.

The water may be replaced with another medium without disturbing the preparation. Place a piece of absorbent paper at one edge of the cover glass and a drop of the new medium at the opposite edge (Figure 10.2e). Again capillary forces will cause the water to be absorbed by the paper and the new medium to flow between cover glass and slide.

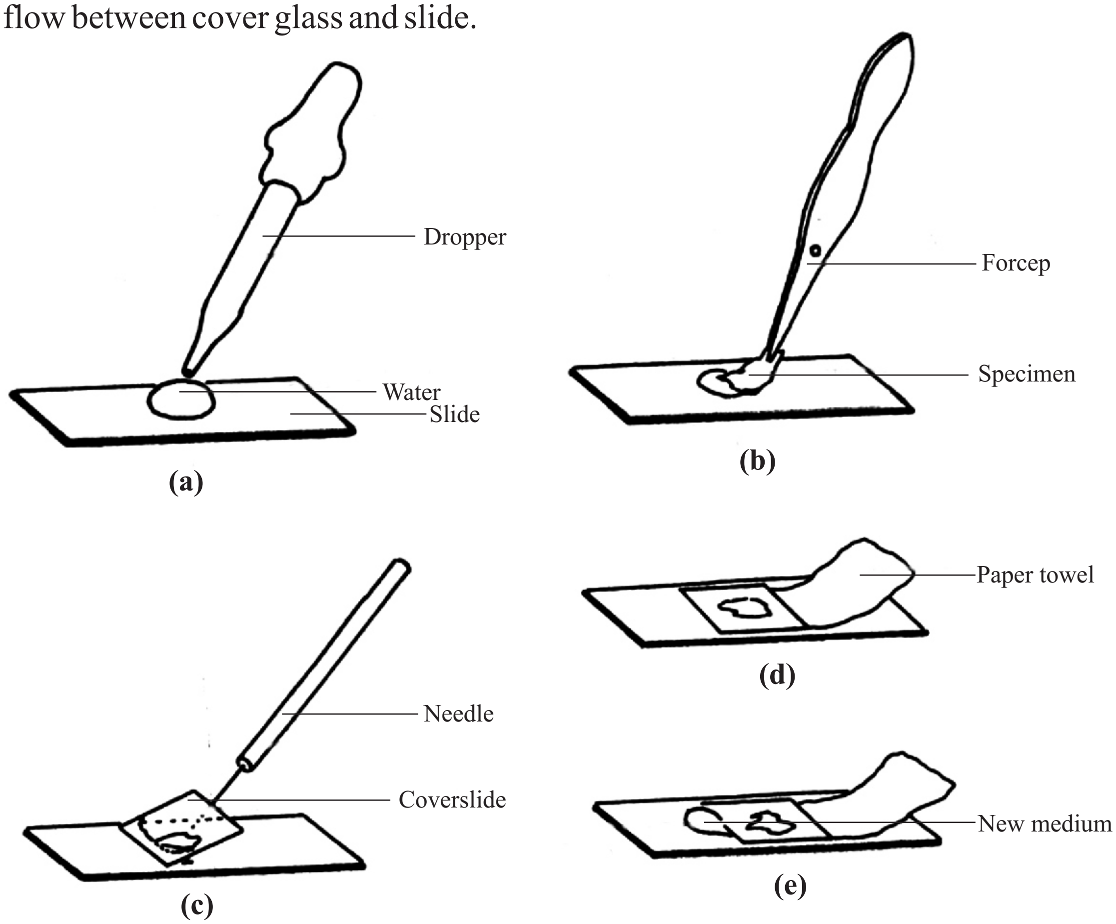


Figure 10.2: *Preparation of Temporary or Wet Mount.*

10.13 Onion Epidermal Cells

Epidermal cells cover the surface of plant organs such as leaves. The bulb of an onion is made up of fleshy leaves. A typical onion epidermal cell is shown in Figure 10.3.

Observation: Onion Epidermal Cells

(i) With a scalpel, strip a small, thin, transparent layer of cells from the inside of a fresh onion leaf.
(ii) Place it gently on a clean, dry slide, and add a drop of iodine solution (or methylene blue). Cover with a cover slip.
(iii) Observe under the microscope.
(iv) Locate the cell wall and the nucleus.
(v) Count the number of onion cells that line up and end in a single line across the diameter of the high-power (40 x) field.

Based on what you learned in this course about measuring diameter of field, what is your high-power diameter if field (HPD) in micrometers? ________ μm
Calculate the length of each onion cell (HPD: number of cells): ________ μm
(vi) Note the shapes of the cells and cell walls in the diagram below.

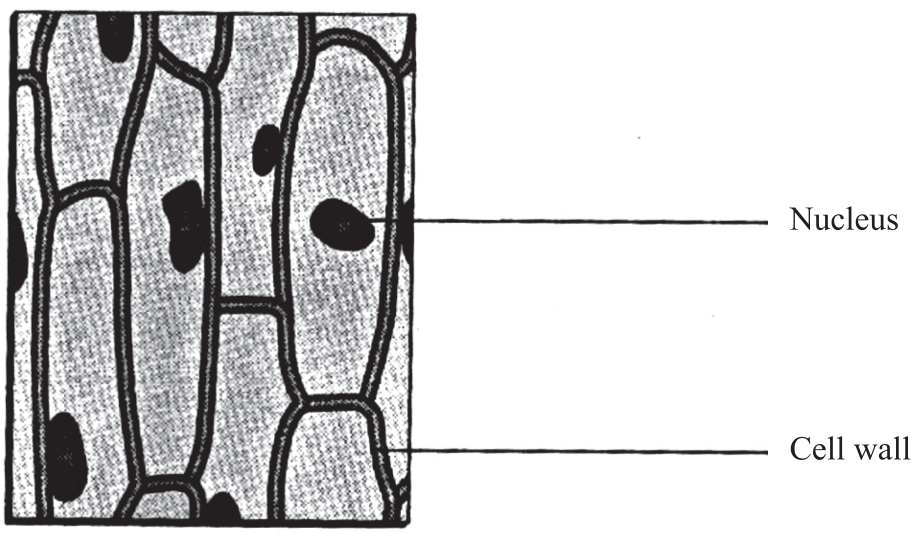


Figure 10.3: *Onion epidermal cells.*

Source: (Seiter, 2004).

10.14 Summary

Microscopes are scientific devices that produce magnified images of structures that are too small to be seem with unaided eyes. Two problems that manufacturers of microscopes have to deal with are (i) the instruments magnification and (ii) the sharpness of the image produced by the instrument. Types of microscopes include the light microscope and the electronic microscopes. Light microscopes can produce images of objects at a magnification of 1000 times. Compound light microscopes allow light to pass through the specimen while using two lenses to form an image. Electron microscopes use a beam of electrons that is magnified and focused on a photographic plate by means of electron magnets. The transmission electron microscope (TEM) is analogous to the compound light microscope. Parts of the light microscope include eye-piece (ocular lenses), body tube, arm, nosepiece, objective lenses, coarse-adjustment knob, fine adjustment knob, diaphragm control level, light source, base and stage. Others are stage clip, mechanical stage and mechanical stage control knobs which are optional. There are ten (10) steps involved in focusing the microscope in the lowest power while there are five steps involved in focusing the microscope in the higher power. Compound light microscopes are parfocal, that is once the object is in focus with the lower power, then it should be in focus with the higher power. There are five rules to observe while using the microscope.

Total magnification (TM)

TM = Moci x Mobi, where

Moci = Magnification of ocular lenses

Mobi = magnification of objective lenses.

A microscopes field of view is the circle visible through the lens. The diameter of the field is the length of the field from one edge to the other. Practical observation of the field of view involves determining the low-power (10 x) diameter of the field by estimating the number of mm to tenths that you can see on the edge of a plastic ruler placed on the stage. This figure is converted to micrometer (μm) which is the low-power diameter of field (LPD) for your microscope in micrometers. To compute the high-power diameter of field (HPD) use the formular below:

$$HPD = \frac{LPD \times LPM}{HPM}$$

HPM where

LPD = low power diameter of field (um)

LPM = Low power total magnification

HPM = high power total magnification.

Observation of depth of field can be done by obtaining a prepared slide with colored threads or hairs, mounting it on the stage and using low power to find a point where the thread or hairs cross. As you switch to high power, you will notice that the depth of field is more shallow with high power than with low power .

10.15 Evaluation

Having read this chapter, answer the following questions:

1. (a) What is a microscope?
   (b) Why do Laboratory scientist use the microscope?

2. In few sentences, differentiate between light microscope and transmission electron microscope.

3. Draw and label the compound light microscope.

4. Explain the various parts of a compound light microscope.

5. State the rules for effective use of a light microscope.

6. Discuss the steps to be followed in focusing the microscope (a) Lowest power (b) Higher power.

5. Explain the process of wet mount preparation.

10.16 TUTORIAL QUESTIONS

Fill in the gap in the sentences provided below:

1. When people think of scientific equipment, one of the first tools that come to mind is the ________________.

2. Light microscopes produce ____________ images by focusing visible light rays.

3. The most commonly used ___________ is the light microscope.

4. Electron microscope uses a beam of light that is magnified as focused on the photographic plate by means of _____________.

5. The use of electron rather than light gives electron microscope a much greater ___________.

6. The name compound light microscope indicates that it uses _________ and both ocular and objective lens to view an object.

7. Compound light microscopes are ____________, that is once the object is in focus with the lowest power, it should also almost in focus with the _____________.

8. The vertical distance that remains in focus at one time is called the ____________.

9. When a specimen must be prepared for observation, the object should always be viewed as a ____________

10. The transmission electron microscope (TEM) is __________ to the compound light microscope.

11. Compound light microscope allow light to pass through the __________ and use two ____________ to form an images.

CHAPTER ELEVEN

11.0 MAKING BIOLOGICAL DRAWINGS AND WRITING OF EXPERIMENTAL REPORTS

CHAPTER OBJECTIVES

By the end of this chapter, students should be able to:

(i) enumerate the qualities of a good drawing;

(ii) identify the materials needed for successful drawing;

(iii) state the major weaknesses in reporting plant anatomy practicals;

(iv) explain the steps to be followed in cutting a freehand section;

(v) outline the pattern followed for writing reports of an experiment.

11.1 Making Biological Drawing/Illustration

It cannot be over emphasized that the purpose of an illustration is to record observations. A single illustration may convey more information than a hundred (or thousand) words. Emphasis must always be placed on accuracy and neatness.

A drawing is a representation of an object and may include as many details as possible. It should be made after a careful study of the object. A good drawing should have the following qualities:

(i) It should be a large and well labeled (not less than 15cm) to show clearly all structures and should not be over crowded;

(ii) It should have a title and purpose;

(iii) It should have horizontal labeling and the labeling lines should not cross each other and must be done with a ruler;

(iv) All good drawings should have the magnification written below the drawing (e.g x 3);

(v) If it is on a single sheet of paper, the drawing should have the students name, laboratory section, registration number and department on the upper right hand corner of the sheet;

(vi) It should be drawn on a drawing paper with a HB pencil;

(vii) It should have the date written on the top right hand corner and the title should be written centrally on the top of the paper and underlined boldly;

(viii) A good drawing should be centrally located so as to give enough room on the right for labeling;

(ix) It should be drawn to specification;

(x) A good drawing should recognise the different views of the specimen;

(xi) The Drawing Lines of a good drawing should be continuous and smooth, it must not be wavy, wooly and unnecessarily thick, or double dotted or broken;

(xii) The guidelines must touch the structure to be labeled and must not go beyond it or fall short of the structure;

(xiii) Cut surface should be represented with double lines;

(xiv) Shading should be avoided.

11.2 Materials required for Successful Drawing

The under listed materials are required for successful drawing.

(a) A thick plain sheet of drawing paper.

(b) Sharp pencil (HB type preferable).

(c) Eraser, preferably the type that can clean without leaving dirty stains on the drawing.

(d) Sharpener or razor blade.

(e) A meter rule

(f) A dissecting kit including pins

(g) A flat piece of thick cardboard of 30cm square

(h) Watch glasses of various sizes.

(i) A microscope with microscope slides obtained from the laboratory store.

11.3 Major Weaknesses of Students in Reporting Practicals in Plant Anatomy

Major weaknesses of students in reporting practicals in plant anatomy include:

(i) Poor observation.

(ii) General weakness in drawing and labeling.
(iii) Students inability to compare the specimens.
(v) Weakness in interpretation of data.
(vi) Poor methodology in carrying out experiments.

11.4 Cutting of Freehand Sections

A sharp single-edge razor blade should be used. Cutting sections dulls the blade, hence it is unwise to use one blade more than three or four times.

(i) Hold the material between the thumb and forefinger of the less dextrous hand. Make sure the thumb is a little below the level of the index finger. Steady the arms by keeping them close to the body. Hold the razor blade between the thumb and forefinger of the other hand (see the diagram in Figure 11.1).

(ii) Place a drop of water on the material to be sectioned. Rest the flat side of the razor blade on the upper edge of the material to be sectioned.

(iii) Cut toward the body, with a long slicing motion. Do not attempt to get uniform thickness, rather start the section as thin as possible and taper it to greater thinness. It is not necessary to have a complete section, a quarter to half a section is ample.

(iv) Cut many thin sections and float them off the razor blade into a drop of water. If necessary, use the needle probe to push them off the blade. Transfer the thinnest sections to the water drop on the slide and cover with cover slip. More water may be added to the edge of the cover slip if necessary.

(v) If the material to be sectioned is quite small, it may be supported in a piece of elderberry pith. Slit a two-inch piece of pith length wise with the razor. Place the material in the center of the pith, even with the top. Cut sections according to the above instructions, discarding the small sections of pith when mounting.

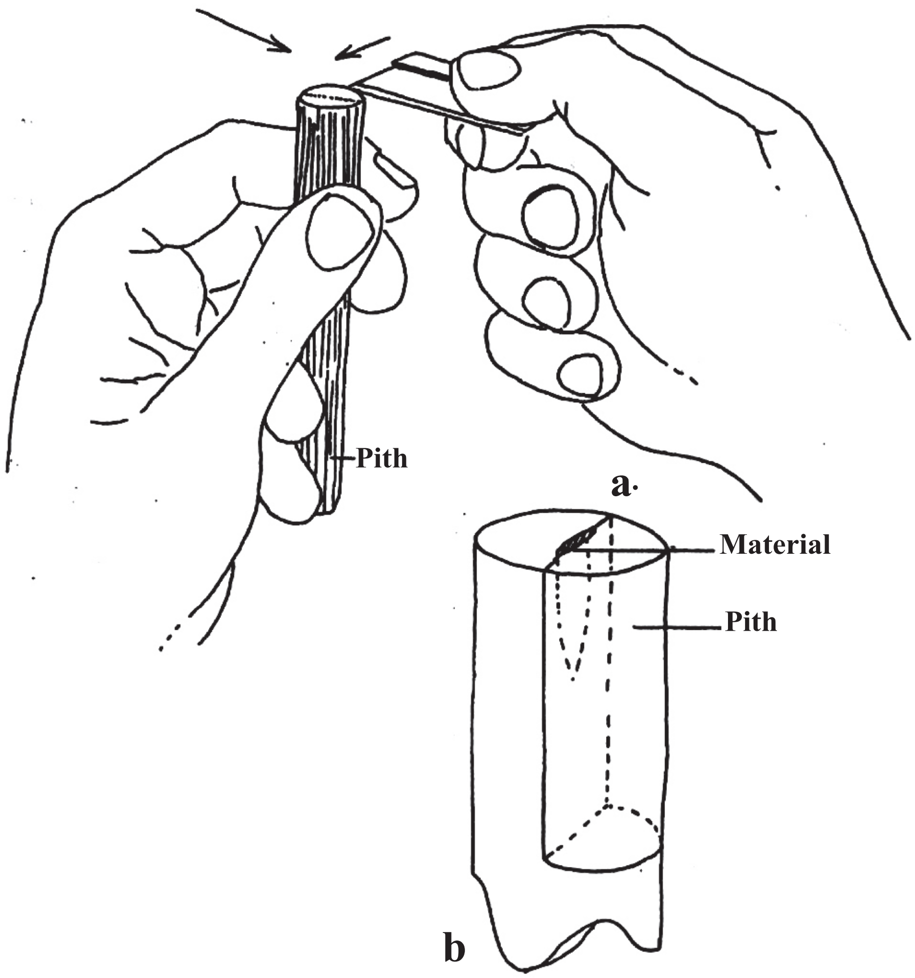


Figure 11.1: *Cutting freehand section.*

11.5 Writing Reports of Experiment

Experimental reports are to be written in a language that will show that the experiments were performed. You are not to write the reports of the experiment as if you are instructing someone to follow certain steps in carrying out these experiments. This instruction should be heeded when reporting all your experiment.

Reports to most experiments follow this pattern:

Aim: Under aim you are expected to write the purpose of an experiment. This is also a heading for the experiment.

Materials: All items used in carrying out the experiment should be listed including laboratory wares, chemicals and plant materials. This is followed by diagram of the set up.

Methods: The procedure or the steps taken in carrying out the experiments after setting up the materials are written, in reported speech.

Observation: All the things observed during the experiment are reported here. All weighing done and readings taken are presented in a tabular form.

Result: If necessary calculations are presented here.

Conclusion: Here the conclusion drawn on the basis of the observations made and the results obtained are reported.

Discussion: Reasons for the success or failure in performing the experiment are hereby listed. Also included here are the precautions taken and the principles on which the experiment is based.

11.6 Summary

A biological drawing is an accurate representation of an object and may include as many details as possible. It should be made after a careful study of the object. Qualities of a good drawing include the following: large and well labeled, title and purpose written, horizontal labeling done with a ruler, magnification written below the drawing. Student's name, department, registration number, and laboratory section. Other qualities are that it should be drawn on good quality drawing paper with HB pencil, the date written on the top right corner, title written and underlined on top of the paper, drawing centrally located on paper, drawn to specification recognise the different views of the specimen and should have continuous and smooth drawing line, labeling lines must touch the parts, shading should be avoided and cut surfaces should be represented with double lines. Materials required for successful drawing are drawing paper, HB pencil, an eraser, razor blade, a meter rule, dissecting kit, a flat piece of cardboard paper, watch glasses and a microscope with slides. Major weaknesses of students in reporting plant anatomy practicals include *poor observation, general weakness in drawing and labeling, inability to compare specimens and poor methodology in carrying out experiments. Cutting freehand sections, involves the use of a razor blade. By holding the material between the thumb and forefinger of one hand and razor blade in the other hand, thin slices of the specimen can be cut and floated from razor into a drop of water. The thinnest sections are transferred from the water drops on to the slide and covered with cover glass. Experimental reports should be written in clear language, it should not be written as if one is instructing someone to follow certain steps in carrying out the experiment. The report should include aims, materials, methods, observation, results, conclusion and discussion.*

11.7 Evaluation

Having read this chapter, answer the following questions:

1. List ten (10) qualities of a good biological drawing.

2. Name seven (7) materials required for successful drawing.

3. State the major weaknesses in reporting plant anatomy practicals.

4. Explain three steps that should be followed in cutting free hand sections.

5. Explain the sub-headings to be included in writing reports of an experiments.

11.8 TUTORIAL QUESTIONS

Fill in the gap in the sentences provided below:

1. A drawing is an accurate ____________ of an object and may include as many details as possible.

2. A good drawing should be _____________ located so as to give enough room on the right for labeling.

3. Under aim you are expected to write the ___________ of an experiment.

4. The procedure or steps taken in carrying out the experiments after setting up the materials are written in ____________

5. All weighing done and readings taken are presented in a _______________

6. All good drawing should have ___________ written below the drawing.

7. A thick plane sheet of drawing paper, sharp pencil (HB type preferable) and a meter rule are materials require for ______________ drawing.

8. Experimental reports are to be written in a __________ that will show that the experiments were performed.

9. If the material to be _________ is quite small, it may be supported in a piece of elderberry.

10. All ___________ used in carrying out the ____________ should be listed including laboratory wares, chemicals and plant materials.

CHAPTER TWELVE

12.0 PRACTICAL EXERCISES IN PLANT ANATOMY AND HISTOLOGY

CHAPTER OBJECTIVES

By the end of this chapter, students should be able to carry out simple laboratory exercises like:

(I) section cutting;
(ii) mounting;
(iii) microscope examination;
(iv) sectioning and staining techniques in plant tissues;
(v) observation of primary and secondary thickening in roots and stems;
(vi) examining the internal structure of young monocot stems and roots;
(vii) examining the internal structure of young dicot stems and roots.

12.1 Section Cutting

(i) Use a sharp razor to cut thin section from fresh material like Irish potato which contains a large proportion of water.

(ii) Hold the razor lightly in the right hand and wet the blade. Hold the piece of potato between the index finger and thumb of the left hand so that the finger is in the same level with the cutting surface and thumb some what below.

(iii) Rest the blade horizontally on the tip of the index finger and move it diagonally across the cutting surface which should also be in the horizontal plane. The movement of the razor should be a sweeping one, towards the body but also obliquely towards the left arm or shoulder. In the movement, the thinnest possible slice of potato is cut. The thinner the slice, the more transparent it will be. Repeat this process several times, cutting a section each time.

12.2 Mounting: Practical -1

Preparation and observation of temporary slide (transverse section) of stems, leaves and roots of dicots and monocots permanent slide would also be observed

(i) Remove the sections from the razor into a watch glass containing water.
(ii) Separate the sections, pick out the thinnest and transfer this to a glass slide.
(iii) Add a drop of water and cover with a cover slip. The latter should rest gently on the section which should be immersed in water. Do not let the cover-slip "float" on too much water.
(iv) Soak up any excess water with blotting paper. Exclude air bubbles by carefully lowering the cover slip over the mount.

Precaution: The stage of the microscope and the objective lens should never come into contact with mounting medium. The high power objective is the most likely to come into contact with mounting liquid on the cover slip, should this happen, the lens must be cleaned and dried.

12.3 Microscope Examination: Practical -2

(i) Examine the section first with low and then with the high power objective.
(ii) Observe and draw the tissue of cells of which the potato tuber is built up. Note the way in which the starch grains are packed into cells.
(iii) Irrigate with the iodine solution. The starch grains will stain dark blue and appear almost black. The cell wall and also the cytoplasm will stain yellow. It may be possible to observe a nucleus in some of the cells. This will also stain yellow.
(iv) If your section is sufficiently thin, a number of the cell will appear empty, this is because they have been cut through and their contents have escaped into the mounting liquid.
(v) Make a large labeled drawing of what you have observed.

12.4 The Leaves (Dicot/Monocot): Practical -3

Materials: A leaf of relatively thin texture

(a) Leaf of guinea grass (*Panicum maximum*)/ leaf of maize (*Zea mays*).
(b) Leaf of any dicot, e.g. water leaf (*Talinum triangulare*) or green (*Amaranthus spp.*)

Preparation for Examination (Section cutting)

(i) Cut vertical sections of the leaf at right angles to the midrib or parallel to it.

(ii) A thin section can then be cut parallel to the surface exposed. In cutting fresh materials the razor should be wet with water.

(iii) Transfer the cut sections to water or dilute alcohol and pieces of pith separated. This is best accomplished in a watch glass with the aid of a hand lens.

(iv) Pick thin sections out, and transfer to a slide mounted in water.

(v) Draw the sections as observed under the microscope.

Note: (i) It is only necessary to draw a few cells of each type. Concentration should be on careful and accurate drawing visible through critical observation of the objects to be drawn at a given magnification. (a) water leaf (b) guinea grass.
(ii) In the same way in which leaf/stem sections were cut, cut the root in the same way and pick out a thin root section and mount on a slide in glycerin after staining with iodine solution. Draw what you observe under the microscope.

12.5 Observation of Primary and Secondary Thickening in roots: Practical -4

Materials: bean seedlings at 12th day of germination are suitable

Whole root systems (dicot/monocot) e.g water leaf, maize/guinea grass. Roots generally as absorbing organs also serve to anchor the plants in the soil. The root system of different plants differ much in their external form and roots sometimes carry out a variety of functions.

(i) Examine and draw the root system of
 (a) Water leaf.
 (b)Guinea grass / Maize

(ii) In the same way in which leaf/stem sections were cut, cut the root in the same way and pick out a thin root section and mount on a slide in glycerin after staining with iodine solution. Draw what you observe under the microscope. There should be observation of permanent slides of roots, stems and leaves of dicots and monocots.

12.6 Field observation of Primary and Secondary Thickening in Stems

Materials: Bean seedling at 12th day of germination are suitable .

Practical -5: Observation of Dicotyledonous Stem: Perennial shrubs and trees

After the primary tissues are fully formed, the cambium becomes active and begins to cut out new (secondary) tissues in the steler region. Sooner or later, another strip of meristem, the cork cambium makes its appearance in the peripheral region and begins to form other secondary tissues, viz. secondary cortex, secondary phloem/xylem, etc. In the region. All those secondary tissues are added on the primary one and as a result the stem increases in thickness. This increase in thickness due to the addition of secondary tissues cut off by the cambium and the cork cambium in the steler and extra-steler regions, respectively is called secondary growth/thickening.

(i) For the development of thickening in a dicotyledonous stem, successive internodes downwards of hibiscus stem should be examined (a transverse section at an early stage will show the inception of the interfascicular cambium, slightly later stages show the cambium as a continuous ring. While this has been going on, fascicular cambium has already formed and added to its secondary xylem and to the primary xylem.

(ii) In order to observe the increase in the amount of xylem in successive internodes, make a smooth transverse section across the middle of each internode, smearing with hydrochloric acid (HCl) (note the lignified tissues including sclerenchyma which standout clearly, and the sections should be examined under a hand lens. It is useful and saves time. Make these preliminary observations before cutting sections for microscopic examination.

(iii) Make thin sections as directed in (i) and (ii) and make large labeled drawings of them.

12.7 Observation of Dicotyledonous Roots

As in the stem, the secondary growth or thickening of the root is due to the addition of new tissues cut off by the cambium and the cork cambium in the interior as well as in the peripheral regions. In the root, the secondary growth commences a few centimeters below the region of maturation.

Practical 6(a): Arrangement of tissue in a dicot root e.g. bean root

The process of secondary thickening in a dicotyledonous root can be best studied by taking a length of root e.g. a germinating bean seedling, and cutting transverse sections at intervals of about one centimetre behind the root apex.
(Note how the cells, provided by the successive divisions of a cambial cells are arranged in regular radial files. This arrangement often enables one to distinguish secondary tissues from primary tissues. Draw and label your observation).

Practical 6 (b)

(i) Examine a bottle cork. Note the faintly annual regions on the smooth transverse face, and the passages (lenticels filled with dark-brown tissues of loose texture, running in the radial direction. These lenticels will indicate the planes along which radial and tangential sections may be cut. Observe carefully with hand lens.

(ii) Examine the cut sections microscopically and cork cells drawn from these three view points. Bottle cork is an example of product of continued growth of cork from the same phelogen.

12.8 Sectioning and Staining Techniques of Plant tissues

Practical 7: Sectioning Cutting

Sections of the stems, roots, leaves may be transverse, radial or longitudinal depending on the symmetry of the object from which they are cut. For hand sectioning, a sharp cutting razor or scalpel is required.

(i) Hold the object in which sections are to be cut, between the thumb and forefinger of the left hand or it may be necessary to embed it in a cork or paraffin wax in order to give it rigidity. It is sometimes helpful to rest the hand against the edge of the bench.

(ii) Thoroughly moisten the tissue and the razor with dilute alcohol (30 or 50%) and keep them moist. This prevents sections from drying and air bubbles spreading in the mounted specimen.

(iii) Holding the razor between the thumb and forefinger of right hand with the remaining three fingers alongside the latter and resting the blade on the forefinger of the left hand make a long oblique strike. (note: the sections must be thin and of uniform thickness). Ideally they should be one cell thick. Several sections should be cut and considerable practice will be required for perfection.

(iv) Select the thinnest section and transfer it to a watch glass, place on a microslide, stain and leave until they are stained to the required depth (normal sections take only two or three minutes to be stained).

(v) Remove excess of the stain afterwards by placing the section in the solvent used for the stain. Mount and observe under the microscope (note the structure of the xylem and phloem vessels). Draw and label your observation.

12.9 Essential Laboratory Experiments
Experiment 1: Observing Plant Cell under the Light Microscope

Equipment/materials
Microscope
Cover slip
Slide
Pin
Onion bulb or Rheo

chemical
iodine solution

Precaution
Handle needle with care

Diagram

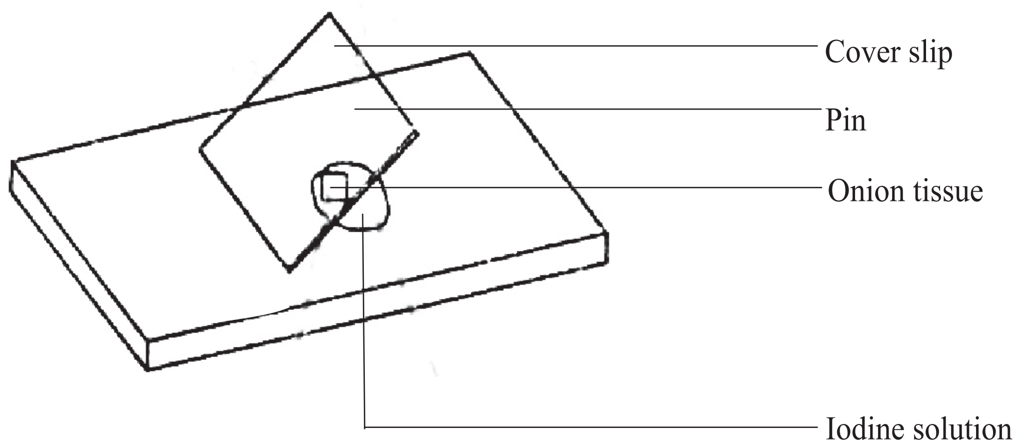


Figure 12.1: *The use of slide and cover slip*

Method
Plant cell: General structure
1. Strip off a piece of epidermis from one of the inner 'fleshy' leaves of an onion bulb.
2. Mount a small piece of the epidermis in iodine solution (as in Fig. 12.1). Slowly remove the pin as you lower the cover slip.
3. Observe one cell under low and high power of the light microscope.
4. Identify the nucleus.
5. Observe the distribution of granular cytoplasm surrounding the vacuole.

Discussion

(a) Notice the cellulose cell wall.
(b) The onion bulb is a plant organ but it does not contain chloroplasts: explain why.
(c) What can you say about the three dimensional shape of the cells.

Experiment 2: Examining the Internal Structure of Leaves

Equipments/materials
Cork polystyrene
Young dicotyledonous leaf
Young monocotyledonous leaf
Petri dish
Paintbrush
Microscope
Slides and cover slip
Razor Blade

Chemical
iodine solution

Precaution
Handle razor blade with care

Diagram

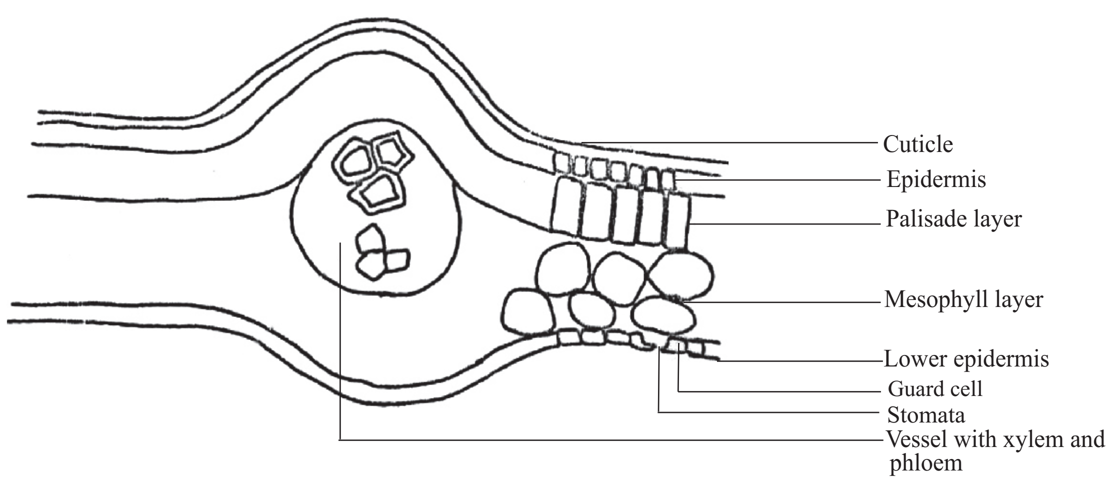


Figure 12.2: *Cross section of a leaf*

Method

1. Cut a narrow strip of cork or polystyrene, this helps to cut thin sections.
2. Split it into two and fit a young dicotyledonous leaf in-between the two halves
3. Cut thin sections of the cork/polystyrene and lead
4. Float them in a petri dish of water
5. With a paintbrush, transfer the thinnest transverse sections of the leaf into a petri dish containing iodine solution and leave them there for three minutes.
6. Mount a thin section of the leaf in water on a microscope slide and observe under the low power of the microscope.
7. Using Figure 12.2 as a guide, identify the various structures of your section.
8. Collect prepared slides of transverse sections of dicotyledonous and monocotyledonous leaves from your teacher and observe too.
9. Using Figure 12.2 above as a guide, draw and label them.

Discussion

(a) In the form of a table, compare the tissue arrangement of a monocotyledonous and dicotyledonous leaves.

(b) Explain how the structure of each tissue in the leaf is adapted to its specific function.

Experiment 3: Examining the Internal Structure of Young Monocotyledonous Stem and Root

Equipments/materials

Razor blade
Petri dish
Paint brush
Dropping pipette
Plain microscope slide
Cover slip
Young monocotyledonous plant e.g. maize, *Commelina* sp.
Microscope

Chemical

iodine solution

Precaution

Handle razor blade with care

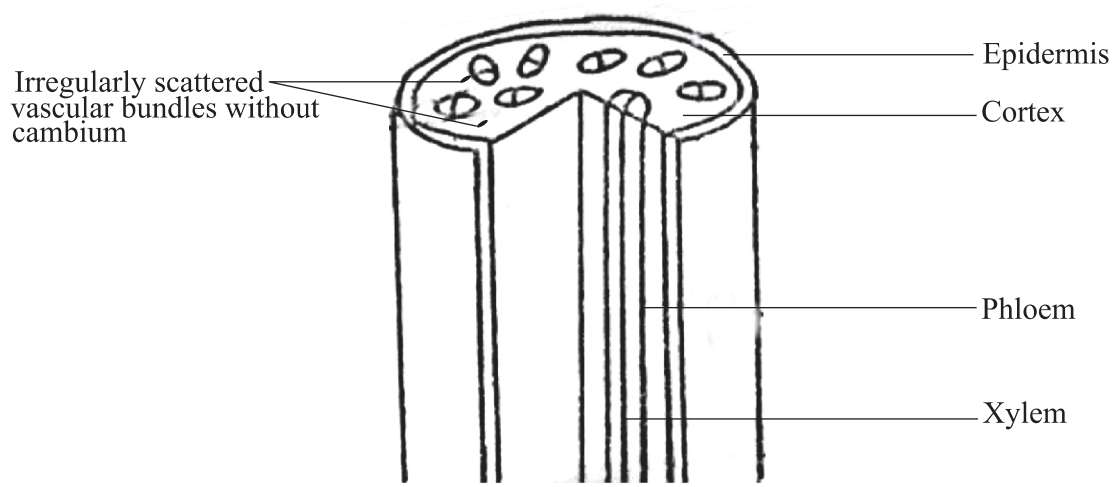


Figure 12.3: *Cross / longitudinal sections of a Monocot Root*

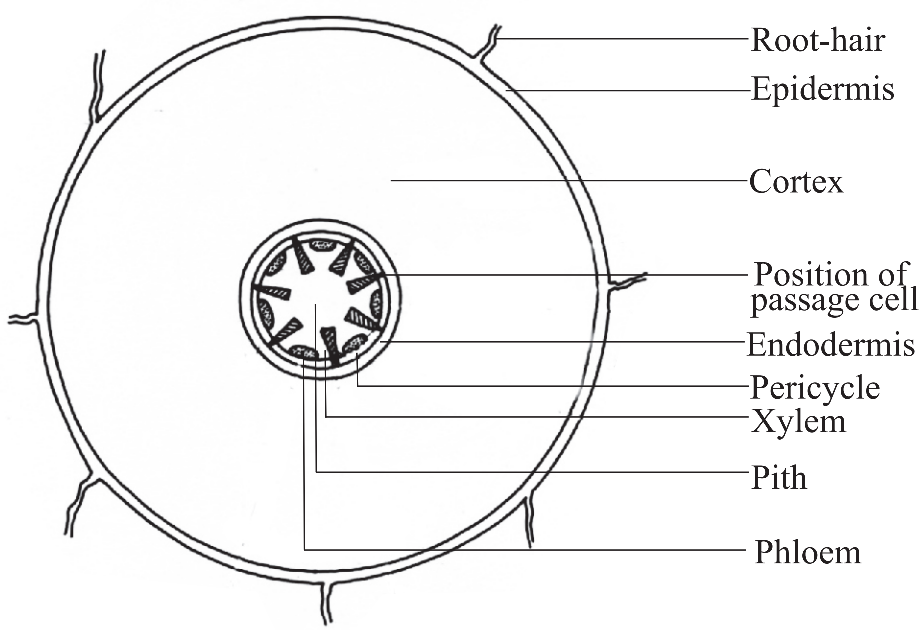


Figure 12.4: *Transverse section of maize root*

Method

1. With a sharp razor blade, cut very thin transverse sections of the stem of a young plant and put them in a petri dish containing iodine solution.
2. Using Figure 12.3 as a guide, identify the various parts of the transverse section of the stem.
3. Following Step 1 above, prepare a transverse section of maize root.
4. Using Figure 12.4 as a guide, identify the various parts of the transverse section of a maize root.
5. Collect permanent slides of a monocotyledonous stem and root in transverse section from your teacher.
6. Examine each one to show the distribution of the various tissues in the transverse section of the monocotyledonous stem and root.

Discussion

In a tabular form, compare the structure of:

(a) the monocotyledonous root with the monocotyledonous stem;

(b) the monocotyledonous stem with the dicotyledonous stem;

(c) the monocotyledonous root with the dicotyledonous root.

Experiment 4: Examining the Internal Structures of Young Dicotyledonous Stems and Roots

Equipments/materials	**Chemicals**
Razor blade	iodine solution
Model of conducting tissue	other stains can be used
Petri dish	
Paintbrush	
Dropping pipette	
Plain microscope slide	
Cover slip	
Bean seedling (or young plant of *Talinum* sp. *or Helianthus* sp.*)*	

Precaution

Handle razor blade with care

Diagram

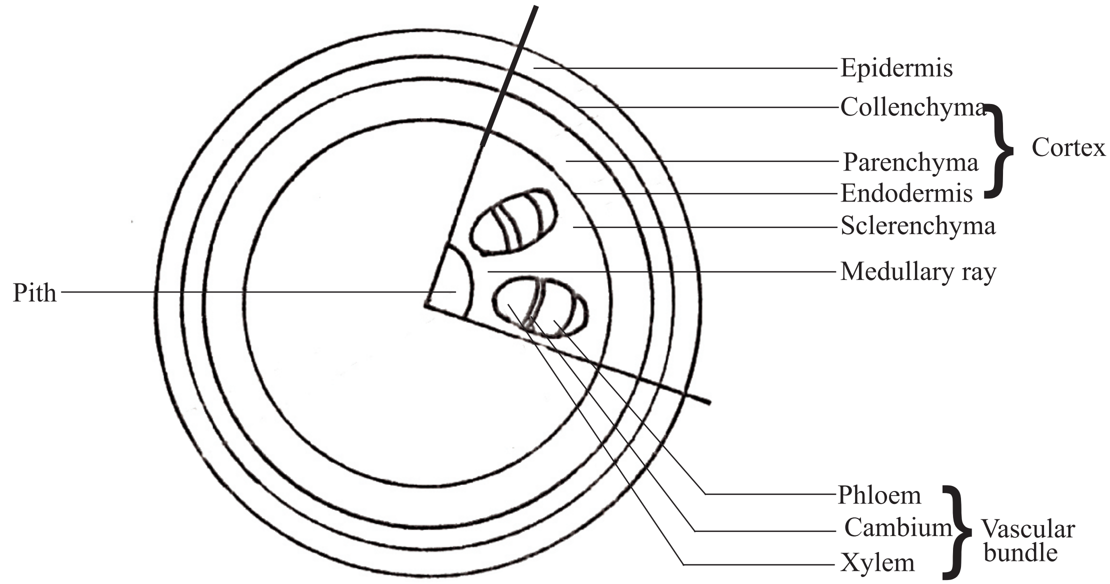


Figure 12.5: *Transverse section of dicotyledonous stem.*

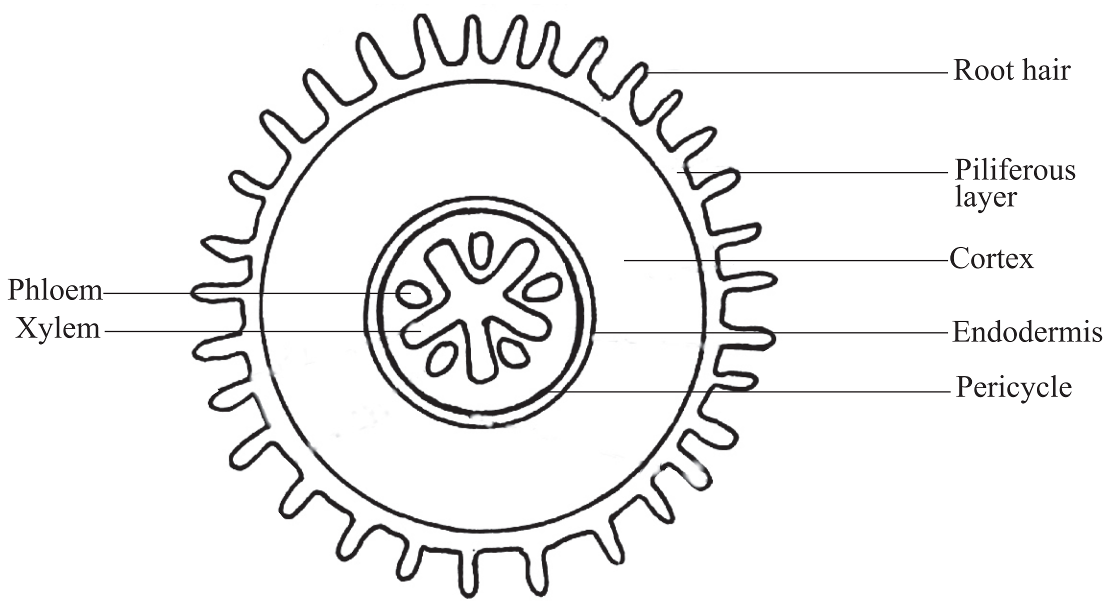


Figure 12.6: *Transverse section of young dicotyledonous root .*

Method

1. Obtain young bean seedling (or a young plant of *Tithonia* or *Talinum*).
2. With a sharp razor blade, cut very thin transverse section of stem of the above named plants.
3. Float the sections in a petri dish of water.
4. Pour some iodine solution into a petri dish
5. Using a paint brush, transfer one of your thinnest sections into the iodine solution and leave it there for three minutes.
6. Lift the section out of the iodine and mount it in a drop of water on a microscope slide.
7. Cover the section with a cover slip and examine it under the low power of the microscope.
8. Try cutting thin longitudinal sections of the stem. Stain one thin section with iodine, mount in water and examine under the low power of the microscope using steps 3 to 7 above.
9. Using a razor blade carefully cut several very thin transverse sections of the root of a young bean seedling.
10. Repeat steps 3 to 7 above. Compare your section with Figure 12.6
11. Obtain permanent slides of a young dicotyledonous stem and a dicotyledonous root from your teacher.
12. Examine each one of the slides under the low power microscope and make labelled drawings of each slide to indicate the arrangement of the various tissues.

Discussion

(i) How do the drawings of the transverse sections of your young bean stem and root compare with Figure 12.5 and 12.6?

(ii) What was the purpose of placing your sections in iodine solution for three minutes?

(iii) Tabulate as many differences as you can between the transverse sections of a young dicotyledonous stem and root.

(iv) Identify the numbered structures in the model of conducting tissue.

Experiment 5: Investigating Secondary Thickening in Stems and Roots

Equipment/materials
Old plant e.g. *Talinum* or *Helianthus*
Microscope
Razor blade
Slides and cover slip

Chemical
Phloroglucinol in HCl Solution

Precaution
Handle razor blade with care

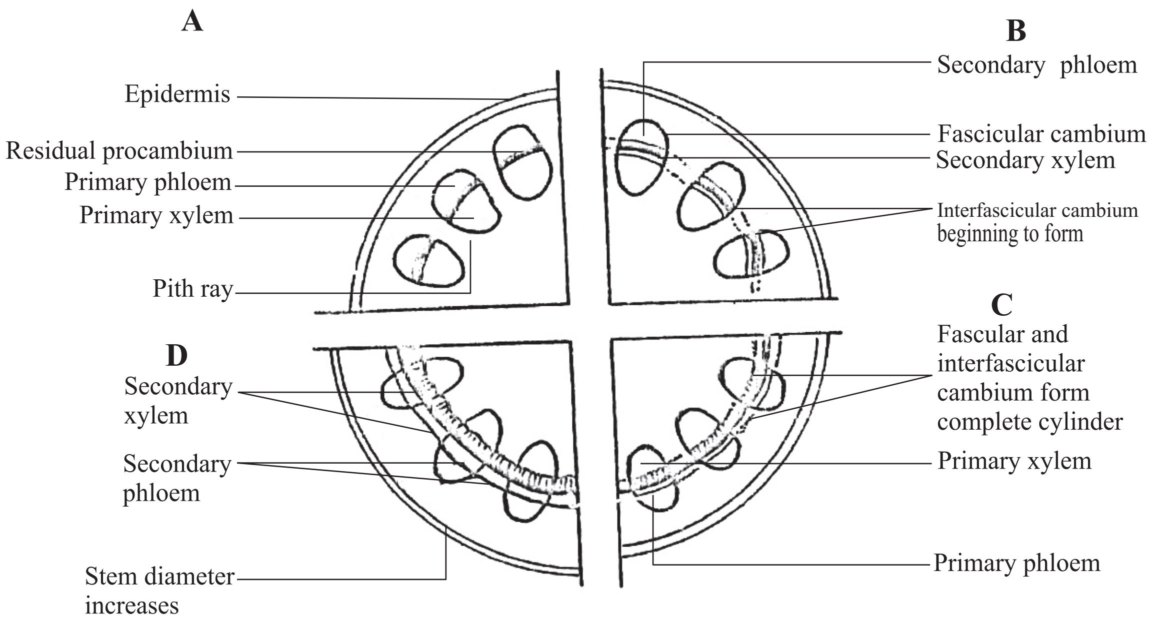


Figure 12. 7: *Stages in secondary growth of a dicot stem*

Method
1. Select an old *Tithonia* sp. plant and uproot it carefully.
2. Cut very thin sections in TS of the older parts of the stem and root.
3. Stain cut sections with Phloroglucinol in hydrochloric acid.
4. Mount in water and observe under low and high power of the microscope.

5. Identify the various tissues as shown in Figure 12.7.
6. Obtain permanent slides of older shoot and root tips of *Talinum* sp., from your teacher and observe using the microscope under low power.
7. Make a labelled drawings of each permanent slide to indicate the arrangement of the various tissues.

12.10 Evaluation

Having read this chapter, answer the following questions:

1. Describe the steps to be followed in section cutting of fresh Irish potato.
2. List the steps to be followed in the section cutting of a leaf.
3. Examine and draw the root system of a water leaf plant.
4. Differentiate using diagram, between transverse, radial and longitudinal sections of stems.
5. Examine the internal structure of young monocot stems and roots.
6. Examine the internal structure of young dicot stems and roots.

12.11 TUTORIAL QUESTIONS

Fill in the gaps in the sentences provided below:

1. Use a sharp __________ to cut thin section from fresh___________ like potato which contains a large proportion of water.

2. Sections of the stems, roots and leaves may be transverse, radial, longitudinal depending on the ___________ of the object from which they are cut.

3. Remove the sections from razor into a _________ glass containing water.

4. The stage of the microscope and the ___________ should never come into _________ with mounting medium.

5. Examine the section first with ___________and then with _________ power of a microscope.

6. All those secondary tissues are added on the ____________ one and as a result the stem increases in ____________

7. The process of continuous ______________ in a dicotyledonous root can be best studied by taking a length of root.

8. Remove excess of the stain afterwards by placing the section in the _________ used for the stain.

9. With a sharp razor blade, cut very thin __________ sections of the stem of a young plant and put them in a petri dish containing iodine solution.

10. Phloroglucinol in HCl solution is the chemical required in investigating _________________ in stems and roots.