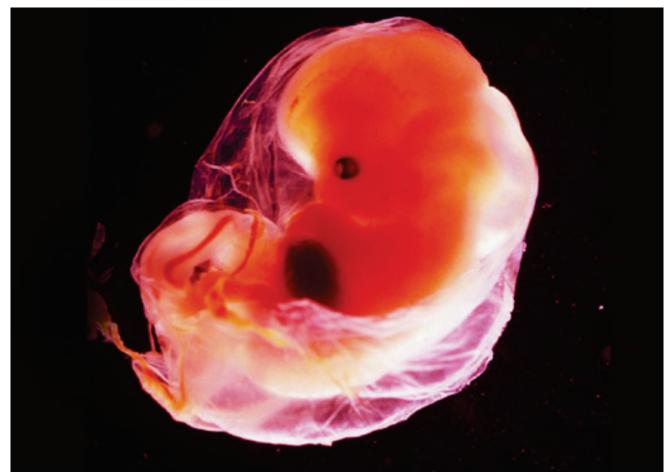


# Embryology, Anatomy and Elementary Morphogenesis

Swarupa. V Uzma Noor Shah





EMBRYOLOGY, ANATOMY, AND ELEMENTARY MORPHOGENESIS

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#### First Published 2022

A catalogue record for this publication is available from the British Library

Library of Congress Cataloguing in Publication Data

Includes bibliographical references and index.

Embryology, Anatomy, and Elementary Morphogenesis by Swarupa. V, Uzma Noor Shah

ISBN 978-1-64532-332-7

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# CHAPTER 1

# TOOLS AND TECHNIQUES IN PLANT ANATOMY

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Research in plant anatomy relies on the laboratory techniques that may be used to examine cell structure and function, much as in other experimental disciplines. The creation of new techniques that opened up fresh research directions was promptly followed by several significant advancements in our knowledge of cells. Understanding both the present situation and the future orientations of this fast-developing field of study requires a grasp of the experimental methods accessible to the cell biologist. The membrane and protoplast are components of a plant cell. The cytoplasm, nucleus, plastids, mitochondria, and other organelles are all present in the protoplast. In the past, the vegetative organs were the primary research topics in plant anatomy; however, the structure of flowers, fruits, and seeds is also a focus. Physiological plant anatomy is a subfield of plant anatomy that examines the connections between a plant's external structure and its interior functions. Environmental influences on plant structure are studied via ecological plant anatomy. Studying the impact of disease-causing chemicals with biological, physical, and chemical properties on plant structure is called pathological plant anatomy. Comparative or systematic plant anatomy is the study of specimens from various systematic groupings, such as species, genera, families, and so on, in order to clarify their phylogenetic relationships[1]–[3].

The fabrication of thin slices that are then examined under a microscope is the fundamental technique employed in plant anatomy, or the study of interior plant structure. The science "derives its name from this. The discovery and development of the microscope are directly tied to the establishment of the science of plant anatomy. Using a microscope of his own improved construction, the English scientist R. Hooke investigated the cellular structure of thin slices of cork, elder pith, and wood from diverse trees in 1665. The first and second books on this topic, which revealed the findings of a systematic microscopic investigation of plant material, were written by the English botanist N. Grew and the Italian biologist M. Malpighi. These two scientists are regarded as the true pioneers of plant anatomy. Only at the beginning of the 19th century did things start to get better. With the process of maceration, the German researcher J. Moldenhawer and the French researcher R. Dutrochet were able to separate plant tissue into its individual cells in 1812 and 1824, respectively. The discovery of the cell nucleus by the English botanist R. Brown in 1831, together with the research of the German botanist M. J. Schleiden, greatly contributed to the development of the cellular theory, which was developed by the German biologist T. Schwann. The German scientists Antony de Bary, Carl Von Nageli, K. Sanio, J. Hanstein, and S. Schwendener, as well as the French biologist Edward van Tieghem, made significant contributions to the study of plant anatomy.

#### **Resources for Plant Anatomy**

Without application, theoretical knowledge is insufficient. Plants are a readily accessible source of lab research material, and their study in the lab significantly advances our understanding of the topic. The practical labour creates the reasonable approach based on facts and numbers and develops the scientific viewpoint. We use a variety of equipment and procedures in the lab for better observation and describing the anatomical characteristics of the plants. Applied Microscopy: Plant cells are so small and microscopic that they are invisible to the naked eye. These things can only be seen through a microscope. Our eyes are unable to identify things smaller than 0.1 mm due to their low magnification or resolution capacity. In addition, living cells are transparent to ordinary light, making it impossible to distinguish between different cellular components. The most crucial instruments for studying plant anatomy are microscope, which use a variety of lenses to magnify objects. Without the invention of the microscope, the fascinating world of microorganisms and many anatomical aspects would not have been discovered.

The earliest description of a lens was made by Roger Bacon. His insight wasn't quickly followed up on, however. By fusing two lenses together, glass polishers Hans and Zacchrius Jensen created a primitive sort of basic microscope in 1590 that allowed them to view tiny things. Galileo created the first straightforward microscope with a focusing mechanism in 1609–1610 and used it to see a water flea. The first double lens microscope with a single convex objective and ocular occurred between 1617 and 1619, and C. Drebbel is generally credited as its creator.

The study of cells, plant and animal tissue, as well as tiny live creatures, was done with this microscope. This instrument had not previously been given the name microscope; Faber initially suggested the term in 1625. The English scientist Robert Hooke is credited with creating the first compound microscope with several lenses. Antony van Leeuwenhoek, a Delft cloth manufacturer, didn't begin producing microscopes as a hobby until until 1670. The microscope had significant advancements in the nineteenth century[4]–[6].

#### **Compound Microscope:**

The main instrument used in anatomy is a compound microscope. All anatomy students must thus have a thorough grasp of the construction, use, and usage of a compound microscope.

#### **Anatomical Techniques for Plants**

To determine the distribution of the different tissues within a solid material, it should be sectioned in a number of planes.

A transverse section at one or more levels, radial and tangential longitudinal sections at various depths from the surface to the centre, and a transverse section at one or more levels are often necessary for a thorough analysis of axial structures like stems and roots. Transverse, paradermal, and rarely vertical longitudinal sections are also required for foliar structures. Several methods are utilised to see the cells for anatomical investigation. The following list of approaches includes a few.

Epidermal peels: Several plant parts' surface tissues may be peeled away in strips thin enough to be examined under a microscope. Break or cut the plant's surface apart to create this kind of peel. Pull the outer tissue layer away from the wound by grasping the epidermis with forceps at one of the cut edges. The resultant epidermal peel should be mounted in alcohol if it is very hydrophobic or water with a wetting agent if it is not.

#### Macerations:

When a cell is cut away from the neighbouring tissue cells, its three-dimensional shape is most clearly seen. This is achieved by macerating fluids hydrolyzing the middle lamella. A tactic that is delicate yet powerful is the one that follows: In a solution of 1 part hydrogen peroxide, 4 parts distilled water, and 5 parts glacial acetic acid, add tiny pieces of the tissue. During 24 hours, bake the mixture in an oven set at 56–60 degrees. Replace the old liquid with a new combination and macerate the tissues for a further 24 hours if necessary. Continue the procedure until the material is mainly colourless and can be separated with a dissecting probe without much difficulty. Rinse the tissues with water in an open container when the maceration is finished. Stain in water with 0.25 percent safranin, then mount in diluted glycerin.

#### **Squashes:**

You may crush something to examine it cytologically on a slide. The most frequent applications of this method are for chromosome counts and the inspection of mitotic structures. Remove any non-meristematic tissues, slice the meristem with a scalpel, cover the tissue with a cover slip, lay paper towels over the cover slip, and push vertically through your thumb.

#### **Free-Hand Sectioning:**

While sectioning, the material should be maintained wet. In order for the parts to float as they are sliced, liquid should be maintained on the razor blade. In general, it is not a good idea to pay close attention to specific portions. Cutting a lot of slices quickly and selecting the finest ones typically produces better results. In most cases, uniformly thin sections are not essential. Slices in the form of a wedge that taper from opaque, excessively thick borders to very thin edges will reveal portions that are thick enough to be useful. It's crucial to use a small piece of material that's not much longer than broad when cutting longitudinal sections. By using the freehand approach, it is difficult to produce good longitudinal sections of any size. During sectioning, flexible structures like leaves need some support? If enough leaves are rolled or folded such that at least ten thicknesses are cut with each stroke, they will provide satisfactory transverse and vertical longitudinal sections. The substance may be put into the cut of a young carrot that has been pickled in alcohol if some additional support is required. The substance is then simultaneously cut from the carrot tissue all around it. Results from this procedure should be better than those using the more traditional elderberry pith method. Bend a leaf over a finger and cut thin slices from the bent surface to get paradermal pieces. To soften and eliminate air from the cells, dried material should first be soaked in hot water or alcohol[7]-[9].

# Staining

Staining cells primarily serves to improve microscope visibility of the cell or certain cellular components. Staining of cells is another method for highlighting metabolic processes or identifying live from dead cells in a sample. Counting cells may also be done by staining them to find out how many there are in a certain environment. One can choose to preferentially stain a cell's nucleus, cell wall, or the entire cell by using various stains. Only a small number of stains can be used on living cells, while the majority of stains can be applied to both living and non-living cells.

# **Regular Biological Stains**

Various stains react or concentrate in various locations inside a cell or tissue, and these characteristics are used to best advantage to highlight certain regions or components. The following list includes some of the most typical biological stains. All of these dyes can be used on fixed cells and tissues unless otherwise noted; essential dyes are noted.

# Carmine

Glycogen is stained with carmine, an extremely red dye, whereas nuclear staining is done with carmine alum. Aluminum is often used as a mordant to remove carmine stains.Cell walls become purple when stained with crystal violet and an appropriate mordant. The stain used in Gram staining is crystal violet. The acidic parts of the neuronal cytoplasm are stained violet by crystal violet, which is often used in brain research.

#### Eosin

Most often employed as a counterstain to haematoxylin, eosin gives cytoplasmic material, cell membranes, and certain extracellular structures a pink or red hue. Red blood cells are also given a bright red tint by it. In many different methods as well as certain variations of Gram staining, eosin may also be employed as a counterstain. The term "eosin" really refers to two different yet very similar substances. Eosin Y, which has a very faintly yellow hue, is the most often used stain. Eosin B, the other eosin component, has a very little blue tint. The choice between the two dyes may be made based more on personal taste and custom.

#### The fuchsin acid

To stain collagen, smooth muscle, or mitochondria, use acid fuchsine. The nuclear and cytoplasmic stain in Mallory's trichrome technique is acid fuchsine. In certain forms of Masson's trichrome, the cytoplasm is stained by acid fuchsine. Collagen fibres in Van Gieson's picro-fuchsine get their red hue from the acid fuchsine. Another common stain for mitochondria is acid fuchsine.

#### Haematoxylin

Nuclear stain haematoxylin. Haematoxylin stains nuclei brown or blue-violet when used with a mordant. One of the most typical histology processes, H & E staining, uses it most often in conjunction with eosin.

# Iodine

In chemical, iodine is employed as a starch indicator. A deep, dark blue hue appears when starch and iodine are combined in solution; this hue represents the starch/iodine combination. As most plant cells contain starch, a mild iodine solution will stain the starch that is present in the cells. Iodine is one of the ingredients of the Gram staining method, which is used in microbiology. Iodine is also used in Gram's staining as a mordant, facilitating dye entry through the membrane or cell wall pore.

## Meta Green

Cell chromatin is often stained with methyl green in bright-field microscopes to make the cells easier to see.

#### **Blue Methylene**

Animal cells, including cheek cells from humans, may be stained with methylene blue to make their nuclei more visible. utilised in cytology and to stain the blood film as well.

# Safranin

Nuclear stains include safranin. It creates red nuclei and is mostly used as a counter stain. Collagen may also be coloured yellow using safranin.

# **Anatomical illustrations**

The capacity to analyse complicated information, recognise diagnostic traits that differentiate between similar structures, and express and convey this knowledge visually are all talents you learn when you create anatomical drawings.

Since they are so little, plant cells are invisible to the human eye. Many methods, including microscopy, sectioning, staining, etc., are used to comprehend the anatomical characteristics of the plant. The xylem and phloem of stems, branches, roots, rhizomes of dicots and monocots, needles, leaves, and underground stems may be employed for anatomical investigations. Because to their incredible resolution power, microscopes are crucial instruments for observation. The magnifying power of a microscope is calculated by multiplying its eyepiece and objective lens magnifications. Plant material is being chopped in several planes to show the cellular structure. Typically, the study's cross and longitudinal sections are obtained. These sections are stained using chemical dyes, and after mounting them for the research, we place them under the microscope. Several kinds of stains, which are often chemical dyes, are used for staining in order to differentiate tissues. By permanently preparing the slide, we may also maintain our portions[10]–[12].

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**CHAPTER 2** 

#### TYPES OF TISSUES, ANATOMY OF ROOT, SHOOT AND LEAF

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Students should be aware that the study of a plant's interior structure is known as plant anatomy, also known as phytotomy. Plant anatomy is the study of the organised plant body's structure as revealed by dissection. Plant anatomy, in general, refers to the study of internal morphology in relation to various tissues. The internal structure of angiosperms, with a focus on primary tissues, is the topic of this chapter. While it initially encompassed plant morphology the description of the physical shape and exterior structure of plants the study of plant anatomy is now regarded as a separate, independent science, and plant anatomy only refers to the internal parts of plants. Higher plants have a more complex plant body and have different types, shapes, and origins of cells. The vascular cryptogams and the spermatophyte contain a variety of cell types that cluster together to create different kinds of tissue systems, according to an analysis of their internal structure. Root, stem, leaf, and flower make up the various parts of the plant body in angiosperms. All of these components are composed of several tissues with various cell types. A mass of similar or different cells carrying out a single function makes up a tissue.

Primary plant body and primary tissue both refer to the initially produced plant body. The plant body is always the principal organ in lower plants and monocotyledons. Although secondary thickening in stem and roots or secondary growth occurs in the Gymnosperms, most dicots, and in certain monocots, its tissues are referred to as secondary tissues. The main structure is not significantly altered by the secondary expansion. The relative distribution of the vascular and ground tissue systems distinguishes the main plant structures, which are the root, stem, and leaves. Before we talk about the many forms of tissue, let's define tissues. If used broadly, a tissue may be described as "a collection of similar or distinct cells that have a shared origin and purpose." The body of a plant performs a variety of activities, and as a result, it contains several tissue systems. For instance, the vascular tissue system aids in the conduction of food and water, the ground tissue system accomplishes photosynthesis, and the epidermal tissue system protects inner cells and aids in gaseous exchange. Meristematic, permanent, and secretary tissues are the three classes into which the many kinds of tissues are divided[1]-[3].

# The Meristematic Tissue or Meristem

All embryonic cells initially have the ability to divide and proliferate, but when the embryo transforms into a plant body, this ability to divide is only available to certain meristems, which remain active throughout the duration of the plant body. As meristematic cells split, some of the daughter cells, known as derivatives, develop into distinct tissue components while the other daughter cells, known as meristematic cells, stay meristematic. Typically, cells grow and protoplasmic and cell wall material are added before any cell division takes place. A collection of cells that are continuously dividing or have the ability to divide are said to be in a meristmatic tissue.

Meristems, which are found in the growth zones, continuously divide to create new cells, which subsequently mature to form the anatomical sections. Differentiation is the process through which freshly formed cells are transformed into mature, permanent cells. Apical meristems, also known as primary meristems, are those that develop at the tips of stems, roots, and other branches. They are responsible for the basic growth of plants. It is divided into main and secondary meristems according to its origin. During the embryonic stage of development, the primary meristem generates the main body of the plant. Later on, the secondary meristems, which are primarily in charge of a plant's growth, are now well known to you. Workers categorise meristems differently depending on their origin, location inside the plant, plane of division, and function.

# **Mature or Permanent Tissues**

Meristem-derived cells eventually undergo structural, metabolic, and chemical changes as well as gain specialised characteristics via a variety of differentiation processes. Not all cells are completely distinct from meristems. Although some cells can divide, others are unable to do so. In a strict sense, only cells that have lost the ability to divide must be considered permanent tissues; but, in a wide sense, cells originating from meristems that have picked up a specific function, such as photosynthesis, secretion, or storage, are viewed as a component of mature tissue. These tissues' cells might have thin or thick walls and can be alive or dead. Tissue with thin walls is often alive, but tissue with thick walls might be either dead or alive.Mature or permanent tissues come in two varieties: Simple and Complex.

# **Basic Tissue**

These tissues or collections of cells are comparable and straightforward types with various structural constituents that mostly make up the vegetative plant body. One kind of cell makes up a consistent cell system that makes up simple tissues. They fall into one of the three categories below based on structural differences:

# Parenchyma

The parenchyma is the primary tissue that makes up a plant's body. Every portion of the plant, including the pith and cortex of the stem and root, the mesophyll of the leaves, the meat of the fruits, the floral sections, and even the xylem and phloem, contains it. Primary cell walls are thin and feature polyhedral forms. Cells are organised either closely together or, more often, far apart with intercellular gaps, as in the cortex and pith. Cells have active metabolic and have thick cytoplasm.Isodiametric cells with thin walls and equal sizing make up a cell. Oval, spherical, and polygonal in form, parenchyma cells have well-defined gaps between them. The cytoplasm in the cells is adequate, and they are living organisms. Typically, a cell has one or more nuclei. The storage of starch, sucrose, protein, water, phenol derivatives, several mineral compounds, etc. is often carried out by parenchyma cells.

Moreover, parenchymatous cells may carry out specific tasks and undergo structural modifications. The stiffness of the plat body is aided by the turgid parenchyma. These cells continue to sustain some water conduction. The many parenchymal kinds are listed below. Aerenchyma: Aquatic plants' parenchyma undergoes modification, and the cortical cells have well-developed air gaps. Aerenchyma is a tissue type that has wide and many intercellular gaps filled with air. While cells are smaller and take up less space, they nonetheless provide

aquatic plants the necessary vigour. These plants often have air gaps that aid in aeration and buoyancy. Air gaps may also be observed in the petioles of canna, aroids, and grasses, among other places. Chlorenchyma: Chloroplasts are abundant in the cells of photosynthetic parenchyma. Chlorenchyma is the chloroplast that develops in parenchymatous cells when they are exposed to sunlight. These cells are often seen in leaves and sometimes in new shoots. Large intercellular gaps are also seen in chlorenchyma cells. There are two categories of chlorenchyma cells:

- 1. Compactly organised, elongated palisade cells.
- 2. Large, irregularly formed, spaciously organised, spongy cells.

# Collenchyma

A basic living tissue made up of elongated cells is called collenchyma. Since it just contains one kind of cell, it is a straightforward tissue morphologically. Pectin deposits cause cell membranes to thicken. Dicot stems, leaves, and floral components are supported primarily by collenchyma, while monocot stems and leaves often lack collenchyma. The capacity of this tissue to modify and develop early in a rapidly expanding organ is its most crucial quality. The tissue's main purpose is to sustain the body of the plant; hence it is crucial that it be present in peripheral areas like the stem, petiole, and leaf midrib.

# Sclerenchyma

Sclerenchyma cells have thick walls and are often lignified. The secondary wall's development is what causes the thickness. The secondary wall is at least initially independent of the main wall. As a cell reaches maturity, it often lacks protoplast, indicating that it is dead. A hollow lumen is enclosed by the cell wall, which often has pits on it. This is a kind of supporting tissue that endures different stresses brought on by the stretching and bending of plant organs without suffering any harm. They come in a variety of sizes and shapes; some of these cells are among the longest in the plant world. This tissue's main job is to sustain the body mechanically. Typically, fibres and sclereids are used to categorise sclerenchyma cells.

Fibers: They have blunted or tapered ends and are often long, spindle-shaped structures. Boehmeria nivea has the longest fibre. They're set up in groupings. 90% of the area of the cell may be secondary thickening; the lumen is small. Little, circular, slit-like, and often oblique pits are found inside of cells. The two types of fibres are extraxylary and xylary fibres. Xylary fibres, commonly known as wood fibres, are components of xylem and are the longest xylem elements.

The origin of the fibre cells determines how they are categorised. They may be divided into three categories:

- 1. Several plants, including cotton, coconuts, Calotropis species, and others, have surface fibres in the testa of their seeds and the covering of their fruits. They are cellulose-based and used to stuff pilose, among other things.
- 2. Surface fibres are the first, followed by wood and bast fibres.
- 3. Wood fibres, often referred to as xylary fibres, and are found in the xylem of the stem and roots. Libriform fibre and fibre tracheids are the two kinds. The lengths, wall thicknesses, and types of pit chambers of the two structures are different from one another. The wall of libriform cells is thick, whereas the wall of tracheids is thinner.

Tracheids feature bordered pits, but simple pits do not surpass the diameter of libriform cells' pit apertures.

Bast Fibers or phloem: Fibers are derived from the phloem and pericycle of the plant and may be lignified or non-lignified. Fibers allow the plant body to resist diverse stresses because of their suppleness. Corchorus capsularis, Hibiscus cannabis, Tilia sp., Nerium sp., Vinca sp., and Crotolaria juncea are a few examples of plants that produce phloem fibres. These fibres are used to make bags, carpets, cordage, coarse fabric, and other items. Extraxylary fibres include industrial textiles like jute, flax, and ramie.

Sclereids: They have thick walls, spherical, oval, or cylindrical shapes, and are shorter than fibres. Sclereids may be found alone or in groups, and they have lignified cell walls and empty interiors. Sclereids are often found in the fruit wall, seed coat, epidermal scales, and petiole of submerged aquatics. They may also infrequently be found in the cortex, pith, mesophyll, and petiole. They may be found in the hard seed coverings of various leguminous seeds as well as the endocarp of almond and coconut[4]–[6].

#### **Complicated Tissue:**

A complex tissue is made up of several distinct cell types that have similar functions. Since they are constructed of several cell types, the xylem and phloem serve as illustrations of complicated tissues. Both of these formations are made up of variously shaped and sized cells that are both alive and nonliving cell assemblages. Vascular tissue is the aggregate name for xylem and phloem tissues.

# **Circulating Tissue**

This tissue is diverse in nature and complicated, including several cell types. Its function is to transport nutrients, water, and minerals throughout the body of the plant, and its main components are xylem and phloem. Xylem: Vascular plants have developed a highly specialised tissue called xylem that functions as the plant's mechanical support system as well as a conduit for the movement of water, minerals, and phytohormonal signals. There is still much to learn about the structure, function, development, and evolution of xylem as well as the genes that control the processes, despite the fact that it is the most prevalent biological tissue on Earth. The tissue that transports water from roots to leaves is called xylem. Together with these fibres, it also contains live cells like parenchyma and dead cells like tracheary components.

The four cell types that make up xylem are tracheids, vessels, xylem fibres, and xylem parenchyma. Tracheids are lacking in Pteridophytes and Gymnosperms, but this arrangement is present in Angiosperms. This is not a ubiquitous combination of xylem since tracheids or vessels are lacking in other angiosperms as well. There are two categories of tracheary components: vessels and tracheids. Growth of vessels is limited, and they are linked end to end to create continuous tubular constructions with holes in the cross walls. These holes effectively transmit water and minerals. Most angiosperms have vessels, and some lower plants like Gnetum, Marsilea, and Selaginella also have them.

The majority of the storage capacity of the xylem is provided by living cells known as xylem parenchyma cells. Particularly in woody plants, many xylem parenchyma cells contain secondary lignified walls. In other instances, these cells feature major pit fields, which are

regions of plasmodesmata that allow water and mineral nutrients to pass from cell to cell. Active xylem tissue contains mature xylem parenchyma cells that can store starch-like carbohydrates and preserve functioning protoplasm. These cells may develop to replenish useful xylem cells, and they also play a significant part in the healing of wounds by generating callus.

Sclerenchymatous cells, or xylem fibres, are present in both main and secondary xylem tissue. Primary xylem is made up of xylem components that separate from an apical meristem, whereas secondary xylem separates from vascular cambium. Due to the growth of lignified secondary walls, xylem is the tissue that has been preserved the best in fossils. An organ gains stiffness and strength from fibres.

Principal Xylem: Protoxylem and metaxylem are the two types. Protoxylem is located in roots further from the centre and in stems closest to the central axis. The protoxylem components feature secondary thickenings that are circular, spiral, and sometimes reticulate. Protoxylem is devoid of fibres. Protoxylem is followed by metaxylem. Only secondary walls have been pitted by metaxylem. Protoxylem's elements are narrower and less complicated than those of metaxylem.

Secondary Xylem: Fusiform and ray initials make up the vascular cambium that produces secondary xylem. Secondary xylem is more complicated and exhibits orderly growth when compared to primary xylem. There is development of pitted and scalariform secondary thickenings. Phloem. The intricate food-conducting tissue known as bast is called phloem. It is made up of fibres, parenchyma cells, partner cells, and sieve elements. All of these cell types are not universally present, much like xylem. Gymnosperms and Pteridophytes do not have partner cells. Some hydrophytes do not clearly differentiate these cells. Primary phloem is distinguished from procambium, while secondary phloem is started from vascular cambium[7], [8].

#### **Secretary Tissue**

The term "secretory tissues" refers to a cell or group of cells that secrete a range of fluids. The secreted material may be expelled, or discharged from the cell, or it may be deposited within the secretory cell itself. Materials may be discharged to the plant's surface or into intercellular channels or cavities. Some of the various compounds found in secretions are not used further by the plant, while others are involved in its processes. Secretory structures may be simple arrangements of a single cell interspersed among other types of cells or complicated arrangements of numerous cells; the latter are sometimes referred to as glands.

Given that the first land plants lived on or near the water and that many of their early inventions were aimed at enhancing photosynthesis via the development of stems and leaves, it is possible that the root was the last of the three primary vegetative organs to arise. Angiosperm root systems often have two very distinct developmental and structural characteristics. Dicots typically have a main root system that develops from the radicle and gives birth to lateral roots with varying degrees of branching. Although the main root of monocots is often transient, their root systems are made up of adventitious roots and seminal roots, where they also develop lateral roots. The root apical meristem is an additional apical meristem found in the root system. This causes extension development in a manner similar to how the shoot apical meristem does. The primary distinction is that roots—rather than leaves and branches—come from the root apical meristem and that development occurs below the surface of the earth. Since they are always underground, roots do very vital duties without receiving the proper recognition for their labour of love. The plant's roots are in charge of anchoring it to the soil, absorbing water and nutrients, storing nutrients, and forming symbiotic partnerships with soil bacteria.

When roots spread out, they go downward through the earth, evading any potential obstructions like rocks. While playing hockey or riding a motorbike, you should always wear a helmet, and roots have their own specific form of helmet called a root cap. When the root penetrates through the earth, the root cap guards the root apical meristem. Moreover, the root secretes a sticky slime that lubricates the soil around the tip of the root as it travels through the abrasive dirt. The anatomy of a root is less complex than that of a stem, and roots exhibit several distinguishing characteristics. They are positively geotropic, devoid of chlorophyll, and resistant to light. The apex of roots has a root cap and is surrounded by root hairs. Vascular bundles are of the radial and exarch type, with protoxylem positioned at the periphery and metaxylem positioned in the core[9].

Root hairs are long, tubular, unicellular hairs. Despite the fact that some species have stomata, the root epidermis does not have any. Specialized pores in the epidermis are present in the breathing roots of halophytes. They are known as lenticels, and roots like these are known as pneumatophores. The epidermis' primary roles are those of protection and water and solute absorption.Root cortex is made up of cells with thin walls and many intercellular gaps. Cells often have a specific form of organisation and are polygonal, rounded, and oval in shape. Certain herbaceous dicots, which lack secondary growth, have mechanical tissues permanently preserved in the cortex. It has been revealed that Tinospora spcortex .'s contains chloroplasts. The cortical portion of dicot roots also contains latex, tannin, and mucilage cells. Due to secondary growth, the cortex in the majority of dicots is replaced by suberized cells. The cork cambium is produced by cortical cells, which also aid in gaseous exchange, the transit of absorbed water, root pressure maintenance, and the recovery of meristmatic activity during secondary development.

The centre cylinder of the plant is made up of the uniseriate layer known as the endodermis. A separate layer known as the endodermis contains live cells that have casparian stripes on their radial and transverse walls. Casparian strips are really bands of suberin that have been deposited on the endodermis's walls. After secondary growth has occurred, the endodermis is often removed. The casparian strip keeps resources moving through the root and into the xylem cells. The starch grain is contained in the endodermis, which also serves as a storage area and controls how materials travel through the root and into the xylem. A thin-walled parenchymatous cell layer called the pericycle is adjacent to the epidermis. It may be mono or multiseriate, and it is in charge of causing the lateral roots to grow. In order to create lateral roots, phellogen, and a portion of vascular cambium, it retains its meristmatic activity.

#### Vascular bundles:

There are radial and tetrahedral vascular bundles. Each xylem and phloem bundle has four bundles that alternate. Exarch xylem is defined as metaxylem in the centre and protoxylem near the pith, which is missing in older roots. The following tissues are visible in the cross section of a monocot plant, together with the significant anatomical aspects of a monocot root: **Epiblema:** The epidermis of the root is made up of a single layer of tightly packed, barrelshaped parenchyma cells. As they are engaged in water absorption, the cells have typical thin walls. Stomata and a cuticle are not present. Some epidermal cells develop into lengthy, single-celled extensions known as root hairs. Hence, epiblema is another name for the piliferous layer.

A significant part of the root's ground tissue is the cortex. It is shown by a number of layers of haphazardly positioned parenchyma cells. Significant intercellular spaces exist. The cortex's primary function is water storage. Water may flow freely into the xylem channels thanks to the cells. The deepest layer of the cortex is called the endodermis, and it is made up of tightly packed barrel-shaped cells. The term "passage cells" refers to some of the endodermis' thin-walled cells. Water may enter the xylem vessels via the passage cells. The endodermis' surviving cells may be identified by the thickening of their radial walls. Casparian thickenings are the name for these enlargements. They are created by the deposit of suberin, a waxy material. A physical force known as root pressure is created and maintained in large part by the casparian thickenings. The root's centre cylinder, known as the stele, is made up of the pith, pericycle, conjunctive tissue, and vascular bundles.

# **Pericycle:**

A single layer of parenchyma cells serves as the stele's outermost coating to symbolise the pericycle. Loosely organised parenchyma cells located in between the arterial bundles make up conjunctive tissue. The cells have been designed with water storage in mind. The pith, which represents the centre axis, is the core part of the root. It consists of a few parenchyma cells that are dispersedly distributed.

# Vascular bundles

The arrangement of vascular bundles is radial. Each xylem and phloem bundle has eight bundles. Polyarch is the term used to characterise the situation. Exarch is how Xylem is classified.Except for certain metamorphic stems, which develop towards the earth instead of towards the light, stems are typically above-ground organs that grow upward. The embryo's plumule serves as the primary stem, while auxillary or adventitious buds serve as the sources for lateral branches. Normal stems have distinct internodes and nodes, the latter of which are the areas where the leaves are connected. Stomata are seen in the epidermis of younger stems, while lenticels are visible in adult stems. One can also tell a woody stem from a herbaceous stem based on the toughness of the stem. The internal architecture of juvenile dicotyledonous and monocotyledonous stems, secondary thickening in dicot stems, and distinctions between dicots and monocots will all be covered in this section.

One of a vascular plant's two primary structural axes, the other being the root, is the stem. Normally, the stem is split into nodes and internodes; the nodes contain one or more leaves as well as buds that have the potential to develop into branches. The nodes may also give rise to accidental roots."Shoots" often refers to new, fresh plant growth, which may include stems as well as other structures like leaves or flowers. While some plants have subterranean stems, the majority of plant stems are found above the soil's surface.

New living tissue is produced each year by stem cells called meristems, which are responsible for this. Stems, together with their appendages, leaves, lateral buds, blooming stems, and flower buds make up shoots. A shoot that emerges from the germination of a seed

is where leaves will eventually grow. Perennial plant shoots, which appear in the spring, are the fresh growth that emerges from the soil in herbaceous plants or the fresh stem and/or blossom development on woody plants.

In common speech, stems and shoots are commonly used interchangeably. Buds, fruits, and leaves all have an axis on stems, which are a crucial part of shoots. Animals often consume immature shoots because they are softer and simpler to chew and digest since the fibres in the new growth have not yet finished developing secondary cell walls. Shoots generate secondary cell walls with a robust and rigid structure as they mature and expand. Several plants release poisons that render their shoots unpalatable or unfit for human consumption.

#### **Flower Anatomy**

Flower anatomy refers to the structure and arrangement of different parts of a flower, which is the reproductive organ of flowering plants. Understanding flower anatomy is essential for plant identification, classification, and reproduction. Here are the different parts of a flower and their functions:

**Petals:** These are the colorful, often fragrant structures that attract pollinators like bees, butterflies, and birds to the flower. The number, shape, and size of petals vary depending on the plant species.

**Sepals:** These are the green, leaf-like structures that protect the flower bud before it blooms. They are usually found underneath the petals and can be the same color as the petals or a different color.

**Stamens:** These are the male reproductive parts of the flower, and they produce pollen. Each stamen consists of a slender stalk called the filament and an anther, which is the part that produces pollen.

**Pistil:** This is the female reproductive part of the flower, and it consists of three parts: the stigma, style, and ovary. The stigma is a sticky surface that captures pollen, the style is a long, slender tube that connects the stigma to the ovary, and the ovary contains the ovules, which will eventually develop into seeds if fertilized.

**Receptacle:** This is the base of the flower, and it supports all the other parts. It can be flat or swollen, and it's where the flower attaches to the stem. The main lateral appendage of the stem and an organ of vascular plants, the leaf. The shoot is made up of the stem and leaves combined. A mass noun that refers to all leaves is called foliage. A leaf is often an organ that is thin, dorsiventrally flattened, carried above ground, and designed specifically for photosynthesis. The majority of leaves have separate top and bottom surfaces that vary in colour, hairiness, stomatal density, and other characteristics. The majority of plant species have flat, wide leaves. Broad-leaved plants are the term used to describe these species. The thin, needle-like leaves of several Gymnosperm species may be helpful in cold locations with regular snowfall and frost. In certain conifer species, leaves may take on additional forms and structures, like scales. Several leaves are below the surface. While succulent plants often have large, juicy leaves, some of these leaves lack significant photosynthetic function and may even be dead by the time they reach maturity, like certain cataphylls and spines. In most leaves, the primary site of photosynthesis almost always occurs on the upper side of the blade or lamina of the leaf; however, in some species, such as mature Eucalyptus palisade foliage,

the primary site of photosynthesis occurs on both sides, and the leaves are referred to as being isobilateral. Tissues are divided into meristematic, simple, and complex tissues based on the various cell structure and function kinds. Actively proliferating cells that are isodiametric in form, rich in cytoplasm, with tiny vacuoles or no vacuoles, and cells that are actively metabolic make up meristematic tissues. Basic tissues have cells that are identical in form, function, and origin. Parenchyma, collenchyma, and sclerenchyma are the three divisions. Complex tissues are made up of many cell types that vary in their structure, function, and origin while working together to execute a single task, such as vascular tissues, secretary tissues, etc[10], [11].

The apex of roots has a root cap and is surrounded by root hairs. Vascular bundles are of the radial and exarch type, with protoxylem positioned at the periphery and metaxylem positioned in the core. Typically, the stem is split into internodes and nodes: One or more leaves, as well as buds that may develop into branches, are held in the nodes. Dermal tissue, ground tissue, and vascular tissue make up the three tissues that make up a stem. The dermal tissue covers the stem's exterior and serves as a protective layer, a means of controlling gas exchange, and a means of water resistance. Around the vascular tissue, the ground tissue fills up and often comprises mostly of parenchyma cells. Based on the characteristics of the vascular bundles, monocot and dicot stems are distinguished. Vascular bundles are dispersed in monocots and grouped in rings in dicots. Mesophyll cells, or palisade and spongy parenchyma, are found in dicot leaves. Whereas the mesophyll in monocot leaves solely consists of spongy cells.

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# **CHAPTER 3**

# STRUCTURE OF VASCULAR TISSUES

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Vascular tissues are a crucial component of plants, responsible for the transport of water, nutrients, and other essential substances throughout the plant. There are two types of vascular tissues in plants: xylem and phloem. Xylem tissue is responsible for the transport of water and minerals from the roots to the rest of the plant. It is composed of four types of cells: tracheids, vessel elements, fibers, and parenchyma cells. Tracheids and vessel elements are the main water-conducting cells in xylem tissue. Tracheids are long, thin cells with tapered ends that overlap each other and form a continuous tube. Vessel elements are shorter and wider than tracheids, and they are arranged end-to-end to form vessels. Fibers provide structural support to the xylem tissue, while parenchyma cells are involved in storage and other metabolic functions.

Phloem tissue, on the other hand, is responsible for the transport of organic nutrients, such as sugars, from the leaves to other parts of the plant. It is composed of four types of cells: sieve-tube elements, companion cells, fibers, and parenchyma cells. Sieve-tube elements are the main conducting cells in phloem tissue, and they are arranged end-to-end to form sieve tubes. Companion cells are specialized cells that are closely associated with sieve-tube elements and are involved in regulating their function. Fibers and parenchyma cells have similar roles to their counterparts in xylem tissue.

Both xylem and phloem tissues are arranged in a specialized manner within the plant. Xylem tissue is typically found on the interior of the plant, while phloem tissue is located on the exterior. Together, these two types of vascular tissue are critical for the survival and growth of plants.Both the human body and plant bodies need a circulatory system to survive. So whereas our own arteries and veins are often quite well known to us, we often ignore plant vessels. Vascular tissue is not present in all plants. As they are submerged in their supply of nourishment and hydration, algae do not need it. Neither do mosses contain vascular tissue. Diffusion is the process that moves water and other elements throughout these plants. Yet, higher plants like fir trees, conifers, and seed-bearing plants as well as ferns contain vascular tissue[1]–[3].

The biggest tree you have ever seen comes to mind. How do you suppose the tree transports food and water via its enormous trunk? Certain plant species, referred to as vascular plants, have a network of internal arteries that transport nutrients and water throughout the plant. The plant's roots, stems, and leaves contain these vessels. Based on the materials they convey, the vascular arteries are split into two categories. Vascular veins enable plants to grow bigger in addition to facilitating the more effective movement of food and water throughout the plant. These vessels let plants to transport essential resources further, allowing them to expand in size. Since both systems move nutrients and enable the organisms to grow bigger owing to the capacity to transport further, these vascular veins are comparable to the human closed circulatory system.

Vascular plants may be further subdivided into more precise groups since they share many common traits with a wide variety of other plants. Vascular plants may be classified according on how they reproduce. Ferns are classed as vascular plants that employ spores for reproduction. A seedless vascular plant is another name for this kind of vascular plant. The bulk of vascular plants, which fall under the Gymnosperm or Angiosperm classification, reproduce by producing seeds rather than spores. The vascular plants known as gymnosperms produce cones to hold their seeds. Large trees such as cedars, hemlocks, pines, and spruces are examples of common gymnosperms. The term "flowering plants" is often used to describe angiosperms, which are vascular plants that produce their seeds within fruits or flowers. Sunflowers, dogwood trees, elm trees, lilies, and maple trees are a few examples of angiosperms that are often seen in nature.

With over 250,000 known species, angiosperms are a tremendously diverse category of plants and are often divided into monocots and dicots.Ground tissue, which essentially occupies the area surrounding the vascular tissue, makes up the bulk of the tissue in a plant stem. After discussing the three categories of ground tissue—parenchyma, collenchyma, and sclerenchyma—we will discuss vascular tissue. The most prevalent kind of tissue in plants, parenchyma, serves a number of purposes, including storing food and water. Young stems and roots are supported by the collenchyma tissue. Finally, the plant stem is rigidly supported and shielded by the sclerenchyma tissue. Sclera, a Greek word, meaning "hard." This might help you keep in mind that sclerenchyma is stiff support provided by hard tissue.

Keep in mind that vascular tissue is the tissue that carries nutrients and water throughout a plant. It moves the nutrients and water the plant needs in a manner similar to how roads and pipes work. Vascular tissue comes in two varieties: xylem and phloem. Phloem transfers food, while xylem transports water and dissolved minerals. The fact that the words "phloem" and "food" both start with the same sound makes it simple to recall which vascular tissue is which, as was previously mentioned. This could help you recall that the xylem tissue transfers water and the phloem tissue moves food[4].

A complex conducting tissue made up of several cell types, vascular tissue is only present in vascular plants. The xylem and phloem are the two main parts of vascular tissue. Internally, these two tissues transfer fluid and nutrients. Moreover, two meristems called the vascular cambium and the cork cambium are connected to vascular tissue. The vascular tissue system of a specific plant is made up of all of its vascular tissues. Vascular tissue often has long, thin cells. It is not unexpected that the xylem and phloem resemble pipes since they are responsible for transporting water, minerals, and nutrients throughout the plant. Phloem's individual cells are joined end to end, much as a pipe's sections may be. When the plant expands, new vascular tissue differentiates in the expanding tips of the plant. The new tissue maintains a link with the plant's existing vascular tissue by aligning with it. Plants' vascular tissue is organised into vascular bundles, which are long, distinct threads. These bundles include supporting and protecting cells, as well as xylem and phloem. Phloem normally occupies the inside of stems and roots, whereas xylem tends to be closer to the outside. Phloem may also be found within the xylem in the stems of several Asteridae dicots. The vascular bundles are found in the spongy mesophyll of leaves. Phloem is orientated towards the leaf's abaxial surface, whereas xylem is directed towards the leaf's adaxial surface. Since the phloem transfers plant-produced glucose and is located closer to the lower surface, aphids are often found on the underside of leaves rather than the top ones.

After learning the fundamentals of xylem and phloem, let's take a closer look at their architecture. First, let's examine the xylem. Tracheids, which are non-living, elongated cells that enable fluid transfer, make up the xylem. Sometimes the stem might be supported by the xylem. In plants, fluids typically travel from the roots through the stem and out to the leaves. Phloem, which always consists of living cells and carries nutrients from the leaves via the stem, is the opposite of xylem, which is formed of non-living cells. The sieve components that make up phloem, such as sieve cells, plates, and tubes, are designed specifically to transfer food through plants. Phloem, which are tubes on the outer layer of the stem, transports food components like glucose from storage tissues or the leaves, where they are created, to the rest of the plant. When a tree is cut, sap, which is the phloem's contents, often seeps out of the tree. If you've ever tried maple syrup, you know that it is the refined version of the sap that maple trees produce in their phloem. Botanists categorise plants by the pattern of vascular tissue in plant stems. We'll concentrate on the variations between monocots and dicots[5], [6].

The main function of phloem is to transport the sucrose produced in the leaves to the remainder of the plant. Moreover, it transports compounds required for defence and development. The contents of phloem, which we typically refer to as "sap," migrate as required to various sections of the plant, in contrast to xylem, which transmits water upward. Phloem, for instance, may transport sucrose from the leaves to the roots for storage throughout the summer and then back up to the leaves for the plant's energy requirements during the spring's blossoming process. Phloem cells are living, in contrast to xylem cells.

#### **Physical Form of Xylem**

Xylem is a type of vascular tissue that is responsible for transporting water and minerals from the roots to the rest of the plant. The physical form of xylem varies depending on the type of plant, but generally, it consists of two types of cells: tracheids and vessel elements. Both tracheids and vessel elements are long, hollow cells with thick cell walls.Tracheids are the most primitive type of xylem cells and are found in all vascular plants. They are long and thin cells that taper at the ends and have pits in their cell walls. These pits allow water to move from one tracheid to another, allowing for water to be transported throughout the plant. Tracheids are particularly important in conifers, which lack vessel elements.

Vessel elements, on the other hand, are found in angiosperms, which are flowering plants. They are wider and shorter than tracheids and have openings on their ends called perforations. These perforations allow water to flow freely through the cells, making them more efficient at transporting water than tracheids. Vessel elements are arranged end-to-end to form long tubes, called vessels, that make up the bulk of the xylem tissue in angiosperms.Both tracheids and vessel elements have thick cell walls that are reinforced with lignin, a complex polymer that makes them rigid and strong. The lignified cell walls of xylem cells provide support to the plant, helping it to stand upright and resist the forces of wind and gravity.One of the two forms of transport tissue found in vascular plants is xylem, the other being phloem. While it is present throughout the whole plant, the best-known xylem tissue is wood. The name "xylem" is derived from the Greek word "wood." Water is transported via the plant's xylem from the roots to the shoots and out the leaves. The majority of xylem cells are dead cells that create a hollow cylinder that indirectly moves from root to leaf across the whole plant. By transpiration, which is the process of water loss by evaporation, water

continually leaves plants via their leaves. The moisture absorbed via the roots is transported up through xylem to the leaves in order to replenish the water that has been lost since water molecules have a tendency to stay together owing to their molecular structure. Moreover, xylem transports dissolved minerals, and since the cells have strong cell walls, it gives the plant some means of support. The lengthy tracheary components that carry water are the xylem cells that are most easily recognised. There are over 600 species of conifers, and tracheids and vessel elements differ from one another in morphology; vessel elements are shorter and joined together to form long tubes known as vessels. All species contain secondary xylem, and this group's species are all rather similar in terms of structure. The secondary xylem of many conifers that grow to be tall trees is utilised and sold as soft wood[7]–[9].

**Angiosperms:** there are between 25,000 and 400,000 different species of angiosperms. The monocots in this category seldom have secondary xylem. The secondary xylem of many monocot Angiosperms that become trees is utilised and sold as hard wood.

Plants need a more effective water transport system to release them from the limitations of tiny size and consistent moisture imposed by the parenchymatic transport system. They evolved into specialised cells during the early Silurian, which were lignified to prevent implosion. This process also caused the cells to die, enabling their insides to be emptied for water to move through them. These larger, empty, dead cells were a million times more conductive than inter-cell transfer, allowing for greater  $CO_2$  diffusion rates and the possibility of transport across greater distances.

Regulation of water movement is necessary, and stomata offer dynamic control. They may limit the quantity of water lost via transpiration by controlling the rate of petrol exchange. As stomata are found in non-vascular hornworts, they seem to have developed before tracheids, playing a crucial function in environments where water supply is not consistent.

While the earliest fossil evidence for an endodermis may be found in the Carboniferous, this structure most likely first formed during the Silu-Devonian. The water transport tissue is covered by this route structure, which also controls ion exchange. When transpiration is insufficient as a motor, the endodermis may also create an upward pressure, driving water out of the roots. As plants reached this stage of regulated water transport, they were genuinely homoiohydric, able to draw water from their surroundings via organs resembling roots rather than depending on a thin layer of surface wetness. This allowed plants to reach considerably larger sizes. They lost their capacity to endure desiccation as a consequence of their independence from their environment, which was an expensive feature to keep.

Higher water transport pressures are achievable with broader tracheids with strong walls, but cavitation is a bigger issue as a result. Cavitation happens when an air bubble arises within a vessel, rupturing the bonds that hold chains of water molecules together and prevents them from drawing up additional water with their cohesive force. Once cavitated, a tracheid cannot have its embolism removed and be put back in use. Hence, it is beneficial for plants to prevent cavitation from occurring. Because of this, tracheid wall pits have extremely tiny widths to keep out air and avoid the formation of bubbles. It is crucial that many tracheids function simultaneously since air leakage and cavitation are virtually always caused by damage to a tracheid's wall.

Cavitation is difficult to prevent, but once it has happened, plants have a variety of defences to limit the harm. Tiny pits connect neighbouring conduits so that fluid may move between them but not air. Ironically, these pits are a primary source of embolisms since they both stop them from spreading and enable fluid to do so. The flow of water through the xylem is further decreased by these pitted surfaces by up to 30%. Some plants just accept cavitation; for instance, oaks begin each spring by developing a ring of broad vessels, but none of them survive the winter frosts. Each spring, maple trees employ root pressure to push sap upward from the roots and squeeze out any air bubbles.

Another characteristic of tracheids used in height-gain was the support provided by their lignified walls. Inactive tracheids were kept in place to create a sturdy, woody stem, which was often made by a secondary xylem. Tracheids, on the other hand, kept a central location in early plants because they were too mechanically weak, and the outside rim of the stems had a covering of hard sclerenchyma. Sclerenchymatic tissue supports the tracheids even when they do have a structural function[10].

Tracheids have walls at their ends, which significantly restrict flow. The vessel parts are stacked in series and act as if they were one continuous vessel thanks to their perforated end walls. End walls, which were present by default in the Devonian, likely served to prevent embolisms. When an air bubble forms in the trachea, it is called an embolism. Gases may dissolve out of solution or freeze, causing this to occur. Once an embolism has occurred, it is often impossible to remove since the damaged cell is rendered incapable of pulling water up.

Tracheids can only be a single cell, which restricts their length and, in turn, their maximum practical diameter, which is 80 m. increased diameter offers significant benefits since conductivity rises with the fourth power of diameter. Vessel elements, which are made up of many cells linked at their ends, were able to overcome this restriction and build bigger tubes with diameters up to 500 m and lengths up to 10 m.

Horsetails, ferns, and Selaginellales all individually underwent the early stages of vessel evolution during the dry, low  $CO_2$  times of the late Permian, and angiosperms subsequently displayed them in the mid-Cretaceous. Compared to tracheids, vessels are able to carry a hundred times more water than the same cross-sectional area of wood! It also gave vines a new market opportunity since they could now convey water without having to be as thick as the tree they were growing on. This enabled plants to cover more of their stems with structural fibres. Despite these benefits, tracheid-based wood is significantly lighter and less expensive to produce since vessels need more reinforcement to prevent cavitation.

#### Development

Centrarch, exarch, endarch, and mesarch are four terminologies that may be used to describe xylem development. Its nature changes from protoxylem to metaxylem as immature plants grow. In the study of plant morphology, the patterns of protoxylem and metaxylem arrangement are significant.

# The metaxylem and protoxylem

One or more principal xylem strands develop in the stems and roots of a young vascular plant as it matures. Protoxylem is the name given to the earliest xylem to form. Protoxylem may often be identified by its narrower vessels, which are made of smaller cells. These cells include walls in some of them feature ring- or helical-shaped thickenings. Protoxylem may stretch in a functional sense since the cells can expand in size and mature when a stem or root lengthens. Eventually, the strands of xylem produce "metaxylem." The veins and cells of the metaxylem are normally bigger, and the cells contain thickenings that are typically either transverse bars that resemble ladders or continuous sheets, with the exception of holes or pits. After elongation stops and the cells are no longer required to increase in size, the metaxylem functionally completes its development.

#### Protoxylem and metaxylem patterns

Centrarch describes the situation in which the primary xylem forms a single cylinder in the stem's middle and grows outward from there. Hence, the metaxylem is located in a cylinder surrounding the central core and the protoxylem is located there. There are no live plants that have this pattern, despite it being ubiquitous in early terrestrial plants like "Rhyniophytes."

Where there are multiple primary xylem strands, the other three terms are used.Exarch is utilised when a stem or root has more than one strand of primary xylem and the xylem grows centripetally, or from the outside inward. As a result, the protoxylem is closest to the middle of the stem or root while the metaxylem is closest to the outside. Typically, it is thought that vascular plant roots exhibit exarch development.Endarch is employed when there are many strands of primary xylem in a stem or root and the xylem grows centrifugally, or from the inside out. Hence, the protoxylem is closest to the stem's or root's core, whereas the metaxylem is closest to the perimeter. Endarch development is often seen in the stems of seed plants.When a stem or root has several strands of main xylem that emerge from the centre of a strand in opposite directions, the term mesarch is employed. Hence, the protoxylem is sandwiched between the metaxylem on both the peripheral and central sides of the strand. A lot of ferns exhibit mesarch development in their leaves and stems.

#### Phloem

Phloem in vascular plants is the living tissue that transports organic nutrients, particularly sucrose, and a sugar, to all areas of the plant where they are required. Phloem, a term derived from the Greek word for "bark," refers to the innermost layer of the bark of trees. The transfer of soluble organic material produced during photosynthesis is the major issue of the phloem. Translocation is the term for this kind of transportation.Conducting cells, also known as sieve elements, parenchyma cells, which include both specialist companion cells or albuminous cells and unspecialized cells, and supporting cells, such fibres and sclereids, make up phloem tissue.

The sort of cells called sieve elements are in charge of moving sugars across the plant. They lack a nucleus and have few organelles when they reach adulthood, therefore they mostly depend on partner cells or albuminous cells for their metabolic requirements. Before they develop, sieve tube cells do have vacuoles and other organelles like ribosomes, but they often migrate to the cell wall and disappear, ensuring that there is nothing to obstruct fluid flow. The smooth endoplasmic reticulum, one of the few organelles they do have at maturity, is located at the plasma membrane, often close to the plasmodesmata that link them to their partner or albuminous cells. All sieve cells feature collections of holes growing from altered and enlarged plasmodesmata, known as sieve regions, near their ends. Platelets of the carbohydrate callose strengthen the pores.

# Associated cells

Sieve-tube members must maintain a strong relationship with partner cells, a particular kind of parenchyma cell, in order for their metabolism to work. The companion cell, which is similar to a regular nucleate plant cell but typically contains more ribosomes and mitochondria, performs all of the biological tasks of a sieve-tube element.

The sieve-tube element is coupled to a partner cell's thick cytoplasm via plasmodesmata. There are several plasmodesmata that create sieve regions on the sidewall that the companion cell and sieve tube components have in common.While the transmission of sugars is the phloem's main purpose, it may also include cells that serve as mechanical supports. Fibers and sclereids are the two main classifications of these. Since them both contain a secondary cell wall, both cell types are dead when they reach maturity. Their stiffness and tensile strength are increased by the secondary cell wall.

# Fibers

The long, thin structural units known as fibres provide tensile strength without compromising flexibility. They are the primary constituent of many textiles, including paper, linen, and cotton, and are also present in the xylem.

# Sclereids

Sclereids are atypically formed cells that increase compression strength but may somewhat decrease flexibility. They also act as anti-herbivory structures since the herbivores' chewing will wear down their teeth more due to their uneven form and hardness. For instance, they are in charge of giving pears their grittier texture.Phloem, which transports sap, is made up of still-living cells as opposed to xylem. Water-based sap contains a lot of carbohydrates produced by the photosynthetic regions. These sugars are sent to storage organs like tubers or bulbs or to non-photosynthetic plant sections like the roots.Storage organs like the roots, which store sugar, serve as sources of sugar throughout the plant's development phase, which typically occurs in the spring. Phloem cells travel in several directions, but xylem cells only move in one direction. The leaves are sources and the storage organs are sinks after the growth phase, when the meristems are quiescent. Organs that produce seeds are always sinks. It is common for sap in neighbouring sieve-tubes to be flowing in the opposing directions due to this multi-directional flow and the difficulty sap has moving easily across sieve-tubes.

Whereas the majority of the time negative hydrostatic pressures drive the passage of water and minerals through the xylem, positive hydrostatic pressures drive the movement via the phloem. Phloem loading and unloading, also known as translocation, is the mechanism responsible for carrying out this action. A sieve-tube element is "loaded" by cells in a sugar source by actively transferring solute molecules into it. Through osmosis, water is forced into the sieve-tube component as a result, producing pressure that forces the sap through the tube. Cells aggressively transport solutes out of the sieve-tube components in sugar sinks, which has the exact opposite effect.

# Girdling

A tree or other plant may be successfully destroyed by removing the bark in a ring on the boot or stem since phloem tubes often sit on the exterior of the xylem in most plants. Phloem damage prevents nutrients from getting to the roots, which causes the tree or plant to die.

Since beavers nibble off the bark at a pretty exact height, trees in regions where animals like beavers live are particularly susceptible. Girdling is a technique that may be used in agricultural settings. For instance, girdling is used to create the huge fruits and vegetables that people see at fairs and carnivals. A farmer would pluck all but one fruit or vegetable from a big branch by placing a girdle at its base. Since there are no other fruits or vegetables for all the sugars produced by the leaves on that branch to go to, it grows several times its usual size.

## Vascular bundle types

Depending on their internal structure, plants have diverse arrangements for their vascular bundles. The vascular bundles of plants that are categorised as dicots and monocots are organised in a circular inside the stem, with phloem on the outside and xylem on the inside. Dicots have a layer of cambium between each bundle, but monocots don't. In many instances, the xylem of these plants has the potential to develop into woody tissue. The vascular bundles are divided into several categories based on the relative positions of xylem and phloem.

#### Vascular Bundles' Purpose

In vascular plants, a vascular bundle is a component of the transport system. Plants' vascular bundles are made up of a variety of distinct tissue types, much like your own veins, arteries, and capillaries. The actual transfer takes place in vascular tissue, which comes in the forms of xylem and phloem. A vascular bundle, which also contains supporting and protecting tissues, contains both of these tissues.

Phloem is often positioned abaxial to the xylem. This indicates that in a stem or root, the xylem is closer to the interior while the phloem is closer to the exterior. The upper side of a leaf is typically the adaxial surface, and the lower side is the abaxial surface. Since the phloem, which is located closer to the lower surface, transports the sugars produced by the plant, aphids are typically found there rather than on the upper side of a leaf. Vascular bundle positions in relation to one another can vary greatly.

Going up, the xylem transports water and dissolved minerals from the roots to the plant's various parts as needed. Since the function of water transportation depends on transpiration, water's hydrogen bonds, and tension. When a cell reaches functional maturity, it is dead. The phloem transports sugar and any other photosynthetic products to the parts of that plant that need it. When they work, phloem cells are alive and able to transport food either upward or downward. The plant tissues that make up the bulk of the 'filling' spaces in plants are known as parenchyma, which grow with the plant and also help in the storage of various substances. Also involved in growth is the cambium tissue, which creates new xylem and phloem as the plant stems increase in girth. All of these tissues serve to ensure that critical substances are transported through the plant.

# Vascular Cambium

The vascular cambium is a plant tissue located between the xylem and the phloem in the stem and root of a vascular plant, and is the source of both the secondary xylem growth inwards, towards the pith and the secondary phloem growth outwards to the bark. It is a cylinder of unspecialized meristem cells that divide to give new cells which then specialise to form secondary vascular tissues.Vascular cambia are found in dicots and Gymnosperms but not in monocots, which usually lack secondary growth. A few leaf types also have a vascular cambium. Vascular cambium does not transport water, minerals, or dissolved food through the plant. It does, however, produce the phloem and xylem, which do perform these functions. For successful grafting, the vascular cambia of the rootstock and scion must be aligned so they can grow together. In wood, the vascular cambium is the obvious line separating the bark and wood.

The cambium present between primary xylem and primary phloem is called intrafascicular cambium. At the time of secondary growth, cells of medullary rays, in a line with intrafascicular cambium, become meristematic and form interfascicular cambium. The intrafascicular and interfascicular cambia, therefore, represent a continuous ring which bisects the primary xylem and primary phloem and is known as cambial ring. The vascular cambium then produces secondary xylem on the inside of the ring, and secondary phloem on the outside, pushing the primary xylem and phloem apart.

Fusiform cells are elongated with tapering ends and their size is variable as per the species. They are much longer then wide, broad in middle and tapering at both ends. While Ray initial are isodiametric and much smaller. They constitute the axial system and form radial system of vascular cambium. The tracheary element fibres, xylem and phloem parenchyma, sieve elements develops from fusiform initials. The vascular rays developed from ray initials. Cambial cells are highly vacuolated and their thin cell walls possess primary pit fields with plasmodesmata. The radial walls of xylem and phloem mother cells are thicker than the tangential ones because cambial cells predominantly divide periclinally during which the thickening of the radial walls is continuous.

Plant vasculature forms a network of interconnected cells spanning the plant's body in an organised manner, from the root tip immersed deep within the soil to the highest tree-tops. The vascular system of multicellular land plants fulfils two main functions, long distance transport and mechanical support. Xylem cells, with thick secondary cell walls rich in lignin, cellulose and hemicellulose, are mainly responsible for providing support to the plant, as well as bulk transport of water, nutrients and minerals from the root system to the shoot. Phloem mediates the shoot-to- root transport of the autotrophic energy source, photoassimilates, as well as signalling molecules, such as plant hormones and peptides.

Plant growth arises from mitotic cell divisions taking place in growth foci called meristems. The earliest meristems are of embryonic origin, such as the root apical meristem and shoot apical meristem, which contributes to root and shoot elongation, respectively. These meristems produce the primary plant body, including the primary vasculature. In the primary shoot, the vasculature is located in separate collateral vascular bundles with primary xylem towards the pith parenchyma cells. In roots, the vascular tissue is arranged in a bisymmetric pattern; primary xylem forms a central axis flanked by two poles of primary phloem. Procambial cells intervene between the primary xylem and phloem in both root and shoot vasculature; at the onset of secondary growth, these begin to divide periclinally , giving rise to secondary xylem , secondary phloem , and a secondary meristem called vascular cambium, which forms a continuous ring in an organ- specific manner, discussed in detail later). The vascular cambium is responsible for the lateral growth of plants, a process which must be carefully regulated in order to ensure holistic development of the plant vasculature.

Meristematic cells are small, cytoplasmic and undifferentiated. As these cells divide, the outermost cells are pushed away from the meristem, where they cease division, initiate

turgor- driven cell expansion and differentiate into specialised cell types. The balance between cell proliferation and differentiation into other cell types is crucial for meristem indeterminacy, and it is evident that both of these aspects of growth are under genetic control.

Vascular meristems generate cells which differentiate into xylem and phloem. The apical meristems in the shoot and root contain procambium, the primary vascular meristem. Vascular tissue in the primary root and hypocotyl originates from embryonic provascular tissue, whereas shoot vascular tissue, located in vascular bundles, is derived from the shoot apical meristem. In Arabidopsis and other species which undergo secondary growth, a lateral vascular meristem called cambium develops mainly from the procambium embedded between the differentiated xylem and phloem. In the shoot, the cambium between the vascular bundles arises from parenchyma and endodermis tissues. Consequently, the complete ring of vascular cambium is formed early on in root/hypocotyl, whereas in shoot the interfascicular cambium between the vascular bundles[11], [12].

Stem cells are located in the meristems, where they maintain the undifferentiated state of the other meristematic cells. A classical stem cell niche consists of a group of cells called an organising centre which keeps the adjacent stem cells from differentiating. In vascular plants, a vascular bundle is a component of the transport system. The transport itself happens in vascular tissue, which exists in two forms as xylem and phloem. Vascular tissue is made of xylem and phloem tissue which transports water and nutrients; they always lie next to each other, forming a structure called a vascular bundle in stems. Vascular tissue is made of xylem tissue which transports water and nutrients from the roots to different parts of the plant and phloem tissue which transports organic compounds from the site of photosynthesis to other parts of the plant. Xylem transports and stores water and water-soluble nutrients in vascular plants. Phloem is responsible for transporting sugars, proteins, and other organic molecules in plants. Vascular bundles are the criteria to identify the plant anatomy. By the help of vascular bundles we can determine whether the section is of stem or root. Along with this by the position of vascular bundles we can also determine whether the plant is monocot or dicot. In between the vascular bundles the cambial cells help to produce the wood and by counting the annual rings produced due to the activity of cambium we determine the age of the trees.

The vascular bundles are divided into several categories based on the relative positions of xylem and phloem. When xylem and phloem tissues occur in separate groups on alternate radial positions are known as Radial vascular bundle. This is seen in roots. When xylem and phloem tissues are present on the same radius and just opposed to each other in conjoint vascular bundles they are called conjoint vascular bundles. It is a common occurrence in dicot stems.In between xylem and phloem cambial cells are present so that vascular bundles are called open type of vascular bundles. Cambium divides and increases the plant in girth called secondary growth and in the peripheral region forms cork cambium which gives rise to the bark.

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# CHAPTER 4

# NORMAL AND ANOMALOUS GROWTH

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Remember that all plant stem growth occurs at the meristems of the shoot system because this is where cell division occurs. There are two types of meristems in the plant stem: apical and lateral. As we just reviewed, primary growth occurs at the apical meristem and increases plant stem length. We have previously looked at the basic structures of the shoot system as well as primary growth of the stem. We will now look at another form of growth known as secondary growth of the stem. Before we do, let us review a few key components of the shoot system, which is the above ground structures of plants, including the leaves, buds, stems, flowers and fruits.

Primary growth occurs at the apical meristem and allows the plant stem to increase in length. However, some plants need more than just growth in the length of the stem. We will now look at this type of growth. Remember that all plant stem growth occurs at the meristems of the shoot system because this is where cell division occurs. There are two types of meristem in the plant stem: apical and lateral. As we just reviewed, primary growth occurs at the apical meristem and increases plant stem length. Primary growth occurs when plants grow toward the sunlight necessary for photosynthesis and also sink roots deep into the soil to anchor them and enable them to absorb water and nutrients. This 'up and down' growth is possible due to apical meristem, stem cell like tissue that, upon division, creates an undifferentiated cell that will become either a new root or shoot tip[1]–[3].

Secondary growth happens when stems or branches grow outward this type of growth is possible because some plants have lateral meristem, another stem cell like tissue. Instead of causing the plant to grow up or down, lateral meristematic tissue causes the plant to increase in girth by adding rings of growth. Now we know how a plant gets taller and its roots get longer. But what about being wider? Even a big tree with an enormous trunk starts out as a puny seedling. So when the width of a plant or its girth increases is called secondary growth and it arises from the lateral meristems in stems and roots.

As with apical meristems, lateral meristems are regions of high cell division activity. However, the cells they make grow outward rather than upward or downward. Dicots use lateral meristems to add to their width; monocots, however, do not experience secondary growth. We will come back to them later. The lateral meristems that produce secondary growth are called cambia, which just mean a tissue layer that adds to plant growth. The two important ones for secondary growth are the vascular cambium and the cork cambium. The vascular cambium produces more vascular tissue, which provide support for the shoot system in addition to transporting water and nutrients. Because the xylem and phloem that come from the vascular cambium replace the original xylem and phloem, and add to the width of the plant, they are called secondary xylem and secondary phloem.

Secondary growth is growth at the lateral meristem and increases the girth of the stem. This type of growth is only found in dicots and is not found in monocots. In order to understand why it does not occur in monocots, let us review the structure of vascular tissue in both types of flowering plants. There are two types of vascular tissue: xylem, which moves water and dissolved minerals, and phloem, which moves food in the plant stem. In monocots and dicots, these structures are organized a bit differently. In monocots, the xylem and phloem are found in paired bundles and are scattered throughout the stem. Remember that monocots are simple flowering plants such as grasses. However, in dicots - which are more advanced flowering plants such as roses and apple trees - the xylem and phloem are found in rings with the xylem on the inside and the phloem on the outside. This organization allows for secondary growth of plant stems.

# Cambium

Meristematic tissue responsible for lateral growth in plants is known as cambium. There are two kinds of cambium in woody plant stems, both of which increase the diameter of stems. First type of cambium is vascular cambium found in the center of the stem; its division produces the plant's secondary vascular tissue. The outer ring or near the epidermis the bark of a woody plant also contains a cambium called secondary cambium or cork cambium, which creates cork cells of the outer layer and responsible to give rise the bark.

The cambium layer consists of a single layer of cell and these cells divide in a direction parallel with epidermis. Each time it divides into two cells and one of the two new cells one remains meristematic and the other differentiates into permanent tissue. If the newly formed cell is near the xylem, it will form secondary xylem and if newly formed cell is towards phloem it will develop in secondary phloem. The activity of cambium thus increases and the enlargement of stem takes place and the activity of cambium remains for a considerable long period of time[4]–[6].

#### Normal Behaviour of Cambium

Cells of apical meristems divide, differentiate and develop to form primary tissues. As a result, the plant grows in length this is called primary growth. While by the activity of secondary lateral meristems, increase in the circumference/girth of the plant organs due to the formation of secondary tissues in stelar and extra stelar regions, is called as secondary growth. Normally secondary growth takes place in roots and stem of dicots & Gymnosperms. Due to lack of cambium in monocots, secondary growth is absent. But exceptionally secondary, growth takes place in some monocots such as Palm, Yucca, Dracaena, Smilax, Agave, Coconut etc.

## Formation of ring of vascular cambium

Vascular bundle comprises xylem and phloem in a bundle and in case of dicot stem these are conjoint, collateral and open type i.e., cambium cells are present in between xylem and phloem cells. A cambium which is present inside the vascular bundle is called intrafascicular cambium. This is a type of primary meristem. When plants become mature then the secondary growth starts and the first step of secondary growth is the formation of cambial ring. For this first of all the cells of medullary rays present in between the vascular bundles become meristematic to form interfascicular cambium this is secondary lateral meristem. Interfascicular cambium is the meristematic cells present outside the vascular bundle and these cells are developed from the medullary cells. Intrafascicular and interfascicular cambia are collectively known as vascular cambial ring. Vascular cambium is formed in the form of a complete ring which is made up of single layer of cells. In dicot stem some part of vascular cambium is primary and some part is secondary

Fusiform initials are long with pointed ends, while ray initials are spherical. Amount of fusiform initials is more in vascular cambium. Continuous periclinal divisions or tangential division takes place in fusiform initials. The plane of division in periclinal divisions is parallel to longitudinal axis of a cell. Through this type of activity few cells are formed towards the radius and these cells differentiate into secondary phloem or bast and some of the cells are formed towards the central axis and these cells are differentiated into secondary xylem or wood.

Now the complete cambial ring starts producing cells towards inside and outside by division. Normally more secondary xylem is formed as compared to secondary phloem due to unequal distribution of hormones. By the pressure of secondary phloem; primary phloem is pushed towards the outside and gets crushed. By the pressure of secondary xylem, all the primary tissues such as primary xylem, pith and old secondary xylem degenerates in the centre of the stem. Due to this central part of the stern becomes woody. These activities keep going on continuously in plants throughout.

Before secondary growth the sequences of cells from center towards outside remains pith, primary xylem, cambium, primary phloem, pericycle and endodermis. But due to secondary growth the sequence of the vascular bundle from center changes to primary xylem, secondary xylem, cambium, secondary phloem, primary phloem and then endodermis. Pith crushes due to the pressure created by the newly formed secondary xylem. Secondary xylem forms in the plant regularly and primary tissues degenerate continuously. This new secondary xylem also degenerates the old secondary xylem.

Waste materials are formed in the stem such as lignin, suberin, tannin, resin-gums etc. due to degeneration of the cells. All these waste materials are filled in the lumen of tracheids and vessels of secondary xylem. Because of this, wood in the central region of the stem becomes dark colored. It is called heart wood or Duramen. The peripheral or outer wood which looks light in color is known as Sap wood or Alburnum. As a result of growing of secondary xylem, the diameter of heart wood increases. Physiologically active wood is sapwood and the main function of sap wood is water conduction. Heart wood provides maximum mechanical strength to stem.

#### **Classification of Wood**

On the basis of amount of parenchyma wood is classified into two groups:

#### Manoxylic wood:

Such type of wood contains more living parenchyma. It is soft and loose wood e.g., Cycas. In these secondary vascular tissues with large amounts of softer storage cells mixed with the wood or xylem cells. The stems of these plants are softer than the wood of trees we use for lumber. Examples of plants with manoxylic wood are sago palms or cycads, the spurs or short shoots of Ginkgo trees, as well as many extinct seed fern groups.

## **Pycnoxylic wood:**

Such wood contains less amount of living parenchyma. It is hard wood. Such types of wood are found in most of the plants and in these secondary vascular tissues with copious amount of xylem cells and little parenchyma. This wood is much stronger and durable. Examples of plants with pycnoxylic wood are conifers or cone- bearing trees, the long shoots of Ginkgo, and Angiosperms.

These terms should not be confused with the terms "hard" versus "soft" woods. These terms are used by agro-foresters to make a distinction between conifer trees and angiosperm trees. Conifers have wood that is light-weight, light in color, and strong in tension, but weak in shear Therefore this is called "soft wood", which is usually cheaper and used for building inexpensive furniture or used for paper pulp. Flowering plants have wood that is darker in color, heavier in weight, and strong in compression, tension, and shear. Therefore, this is called "hard wood", which is used to make durable furniture, flooring, and building structures

On the basis of distribution of parenchyma wood is classified into three groups:

Apotracheal: In this type of wood parenchyma is in scattered form e.g., Gymnosperms.

**Paratracheal wood:** In this wood parenchyma is arranged or distributed in the form of masses or groups e.g., Dicot plants.

**Syntracheal wood:** In this wood parenchyma is collected around the vessels e.g., Terminalia arjuna.

### Classification based on vessels:

On the 'basis of presence or absence of vessels, wood is classified in two categories

- 1. Non-porous soft wood: Vessels are absent in such type of wood e.g., Gymnosperms
- 2. Porous wood: Vessels are present in such type of wood. On the basis of arrangement of vessels porous wood is divided into two groups.

**Ring porous wood:** Vessels are arranged in the form of a ring in this type of wood.Such wood conducts water more efficiently e.g., in temperate region as in Dalbergia.

Diffused porous wood: Asystematical distribution of vessels is found in this type of wood in tropical region as in Azadirachta.

Annual rings are formed due to unequal activity of vascular cambium. The activity of cambium does not remain same; it is changeable in the whole year. Activity of vascular cambium is affected by physiological and environmental factors. In winter or autumn season the activity of the cambium is less and the secondary xylem or wood formed is not extensive through the vascular cambium. Cells formed during this period are small thick walled and have narrow lumens. This is called autumn wood or late wood. The vascular cambium is highly active in spring or summer season and secondary xylem formed during this period is extensive and cells of secondary xylem are larger, thin walled and have wider lumen. This wood is known as spring wood or early wood. The spring wood is lighter in color and exhibits low density whereas the autumn wood is darker and has higher density.

The autumn and spring wood is formed in the form of rings. The ring of any type of wood is called growth ring. Thus, two growth rings are formed in one year. A ring of autumn wood and a ring of spring wood are collectively/known as annual ring. Thus, are annual ring consisting of two growth rings. The number of annual rings, formed in a tree gives the idea of the age of the tree. The study of determination of age of the plant by these techniques is called Dendrochronology. The annual rings are counted from the base of the stem because basal part has maximum annual rings and upper part has less. Therefore, counting from the basal region can give the correct idea. A piece is taken from the stem up to central region from the base of stem with the help of increment borer instrument. The annual rings are counted from that piece and again inserted into the same stem at the same place.

More distinct annual rings are formed in those regions where climatic variations are sharp, as in temperate plants. Distinct annual rings are not formed in tropical plants. Distinct annual rings are not formed in India except Himalayan regions. Least distinct annual rings are formed in seashore regions because the climate remains same throughout the year. One more thing clearer annual ring are formed in deciduous plants as compared to evergreen plants. Similarly in deserts annual rings are less distinct. Annual rings are bands of secondary xylem and xylem rays. Sometimes drought conditions prevail during the middle of a growing season resulting in formation of more than one annual ring these are called pseudo annual rings.

#### **Secondary Growth in Dicot Root**

In dicot roots the vascular bundles are of radial type and in this condition the xylem and the phloem are present in different radii. So, there are no cambial cells in between xylem and phloem as in the case of stem. So, for the secondary growth in roots first of all, conjunctive tissue becomes meristematic during the secondary growth in a dicot root and form separate curved strips of vascular cambium below phloem bundles. Then after, the cells of pericycle lying opposite to protoxylem also become meristematic to form additional strips of cambium. So, the cells present at the base of phloem and the top of xylem first of all become meristematic. Very soon the cells present near to these cells also attain the meristematic behaviour and, in this way, a complete ring of vascular cambium is formed. The portion of vascular cambium formed by pericycle is less. The main portion of vascular cambium is formed by conjunctive tissue.

The shape of ring of vascular cambium is wavy in the beginning, but later on it becomes circular due to the pressure of secondary xylem. The portion of vascular cambium formed by conjunctive tissue becomes meristematic first and forms the secondary xylem towards the center. Ultimately the ring becomes circular by the pressure of secondary xylem. The activity of vascular cambium of root is the same as the activity of vascular cambium of stem. Secondary xylem is formed towards the inner side and secondary phloem is formed towards the outer side by vascular cambium. The portion of vascular cambium which is formed by pericycle is responsible for the formation of pith rays. These are made up of parenchyma. These pith rays are known as primary medullary rays. A few medullary or pith rays are also formed from remaining vascular cambium. These are called secondary medullary rays. Thus, two types of medullary rays is basic characteristic feature of roots. Only secondary medullary rays are found in stem after the secondary growth. Both of them conduct water and food in radial direction.

Cork cambium is developed from the pericycle in roots. Cork is formed towards the outside and secondary cortex is formed towards the inner side by the cork cambium. Lenticels are also found in roots but less in number as compared to stem. Cortex completely degenerates in roots after the secondary growth of one or two years. This falls down due to the pressure of cork, whereas in stem, it degenerates after the long duration. Secondary growth is essential in roots to provide strength to the growing aerial parts of the plants and fulfill the requirement of water and minerals. Annual rings are not formed in roots because these are not affected by the changes of environment. Secondary growth is not found in monocot roots.

# Functions of Secondary Meristem

# 1. Healing of wounds

When any plant part gets injured wound is formed there. Boundary of the wound is raised outside and composed of similar type of living cells called callus. Living cells of wound are responsible to form a cambium. This is called wound cambium. It is also called inducible cambium. This newly formed cambium forms cork towards the outside. This cork covers the wound entirely. Wound cambium is a secondary lateral meristem.

# 2. Abscission

Falling of any plant organ is called as abscission. Abscission takes place due to formation of abscission layer at the base of plant organ and it is composed of parenchyma. Middle lamella is dissolved in abscission layer during abscission and primary walls also dissolve partially or completely. Sign of leaf fall on stem is called leaf scar and it is a type of wound. The living cells of leaf scar are responsible to form cork cambium, which produce cork. Cork covers the wound. At the site of abscission protective layer is found which is suberized.

# 3. Knots

Knot is formed when branches are embedded inside the main stem. In most cases knots are caused by the natural growth of the tree, though the specific circumstances under which they form determines how they will appear. As a tree grows and increases the circumference of its trunk, the growing trunk begins to overtake the branches that grow out from it. Knots form around these branches, building up trunk material as the tree continues to expand. The wood of the knot is typically tougher than the surrounding wood and may form a bulge around the branch emerging from its center and known as tight knot. If a branch becomes injured or otherwise dies while still attached to the tree, a loose knot forms as the trunk grows larger. Loose knots are similar to tight knots, but instead of having living wood in the center of the knot there is only a dark plug of dead or decaying material.

# **Abnormal Behaviour of Cambium**

The word anomalous means deviating from the general or common order or type. Thus, the term, anomalous growth reflects a growth condition which is not commonly seen and which is present in a limited number of families or genera. Plants showing anomalous secondary growth can be studied in two main groups. Those in which cambium of normal type is present and persists but by peculiarity or irregularity in its activity develop vascular tissues of unusual arrangement. Those in which the normal cambium either does not develop or is soon replaced by another cambium. This abnormal cambium may either develop from cortex or pericycle and shows abnormal activity.

### Bougainvillea stem

Bougainvillea is a member of the Nyctaginaceae and is an example of a dicotyledonous stem which displays anomalous secondary growth. In the T.S. of Bougainvillea, near the centre of the stem, you will see some primary vascular bundles embedded in lignified pith parenchyma. Move the slide towards the outer regions, and you will notice that there has been fairly extensive production of secondary vascular tissue. Secondary phloem and secondary xylem lie on either side of it. The secondary xylem is composed of tracheids, fibers and narrow-diameter vessels. Interspersed with the secondary xylem you will be able to see small pockets of phloem and look like large-diameter metaxylem vessels. These are reminiscent of the primary bundles towards the centre of the stem. These are in fact primary vascular bundles embedded within the secondary xylem, hence the use of the term, anomalous growth in this instance.

The phloem is described as being included phloem, which by definition is phloem tissue which lies between regions of secondary xylem. Whilst the physiological advantage of the formation of included phloem has not yet been studied, one could speculate that in this instance, the included phloem would be well-protected from predators and pests and, of course, be well-supplied with water and nutrient. The anomalous growth results as a result of differential cambial activity. Newly-produced vascular cambia result in the outer lateral meristem becoming quiescent and this cambium returns to activity only when the internal vascular cambium becomes less active. Vascular cambia are said to not produce rays in Nyctaginaceae but do produce vessels and associated, axial parenchyma and sometimes fibers to the inside and variable secondary phloem to the outside.

### Nyctanthes stem

In Nyctanthes arbor-tristis stem which is a dicot plant, apart from normal vascular bundles which occur in a ring in the central region, there are four inversely oriented vascular bundles at the four ridges of stem. These cortical bundles are collateral and open. So in addition to the normal ring of stelar bundle some vascular bundles are also present in the cortex, they are known as cortical bundles. Morphologically these are the leaf traces which traverse through the cortical region of the stem before entering into the petiole. These types of vascular bundles are also found in family Crassulaceae, Casuarinaceae and Oleaceae. These cortical bundles are equally active producing cells and helping in secondary growth of the plant[7], [8].

## Dracaena stem

Palm trees are monocots that grow quite tall and thick, yet they lack "normal" secondary growth. Dracaena is a monocot but not a true palm, as palms lack the peripheral secondary thickening meristem such as is found in Dracaena and Cordyline. Dracaena is an unusual plant, in that the vascular bundles are surrounded by very prominent fiber bundles. In this sense, Dracaena is not anomalous. The stems undergo a specialized secondary growth, which manifests itself in the production of additional parenchymatous elements. Their later growth pattern is termed diffuse secondary growth, and consists mostly of a proliferation of ground parenchyma cells and additional vascular bundles near the periphery.

The young Dracaena stem has typical structure i.e., epidermis is followed by sclerenchymatous hypodermis. A large number of closed collateral bundles are scattered in

ground tissue. One of the outer layers of cells from the ground tissue becomes meristematic and functions as cambium. The cambium formed in the region which has ceased elongating. The activity of this cambium is more on the inner side and very little on the outside where it forms only parenchyma. On the inner side it forms xylem and parenchyma in alternate patches. The inner parenchymatous cells are called conjunctive tissue. After a short while the activity of cambium on inner side changes and above the xylem it starts forming phloem and then again xylem. Thus phloem becomes encircled by xylem and ring of leptocentric vascular bundle is formed. The xylem formed earlier has bigger vessels and around each vascular bundle is developed a sclerenchymatous sheath. The cambium after sometime alter its activity and forms xylem on the inner side, at those places where it was previously forming the parenchyma and parenchyma in place of xylem. Similar to earlier case again by change in activity it forms a ring of vascular bundles. Activity of cambium goes on changing regularly and more rings of vascular bundles are formed. The last one or two rings of vascular bundles lie in conjunctive tissue. Cork cambium is formed below hypodermis and forms cork and cork cambium in normal fashion.

#### **Ficus root**

Ficus is a pantropical genus of trees, shrubs and vines occupying a wide variety of ecological niches; most are evergreen, but some deciduous species are endemic to areas outside of the tropics and to higher elevations. Fig species are characterized by their unique inflorescence and distinctive pollination syndrome, which utilizes wasp species belonging to the Agaonidae family for pollination.

In this furrow of secondary phloem are present in the cylinder of secondary xylem. A peculiar secondary growth takes place due to development of unidirectional and bidirectional arcs of cambium. Unidirectional arc of cambium is that portion of the cambium which produces little or no xylem but extensive amount of phloem, bidirectional arc if cambium produces as much or more xylem then phloem. In the initial stages cambial cylinder produces secondary vascular tissues that have a cylindrical configuration. But subsequently four grooves or furrows of phloem are formed. Other portion of the cambial cylinder except for these four arcs show bicambial activity i.e., they produce as much or more xylem than the phloem. As secondary growth continues furrows of phloem become truncated and the unidirectional and bidirectional arcs of cambium become separated. Initially four furrows of phloem are formed but in the older stems additional furrows may be formed.

#### **Tinospora stem**

Tinospora cordifolia commonly known as Guduchi is an Indian medicinal plant and has been used in Ayurvedic preparations for the treatment of various ailments throughout the centuries. It is a glabrous, succulent, woody climbing shrub native to India. It thrives well in the tropical region, often attains a great height, and climbs up the trunks of large trees. The stem is gray or creamy - white, deeply cleft spirally and longitudinally with the space between spotted with large rosette - like lenticels. The wood is white, soft, and porous, and the freshly cut surface quickly assumes a yellow tint when exposed to air.

Vascular zone is composed of discrete vascular strands with 10 to 12 or more wedge shaped strips of xylem, externally surrounded by semicircular strips of phloem alternating with wide medullary rays; phloem parenchyma contains calcium oxalate crystals; cambium is of 1-2

layers; xylem consists of vessel elements, tracheids, parenchyma and fibres. Vessel elements cylindrical in shape bearing bordered pits. Medullary rays 15 to 20 cells wide. Pith mostly made up of large thin-walled cells containing starch grains[9], [10].

The presence of discrete vascular strands in the mature stem of Tinospora cordifolia is one of the anomalous secondary structures found in Menispermaceae. The cambium forms secondary vascular tissue only in the fascicular region, whereas in the interfascicular areas parenchyma is produced. Thus, in the old stem the xylem becomes fissured due to the development broad parenchymatous rays. In such stem parenchyma acts like a shock absorber. It also enables the stem to resist the pulling and compression due to the pressure of high winds. This anomaly is thus an adaptation to the climbing habit of the plant.

#### **Activity of Cork Cambium**

In plants, primary growth occurs in the epidermis, which is the outermost layer. This layer is sufficient in plants without lateral development to aid in shielding the interior tissues. Yet, when a stem thickens, this epidermis cracks and detaches. Without the cork cambium, the plant would be vulnerable to disease and water loss. Because to the activity of the cork cambium, secondary growth occurs in the extra-stelar zone. Other names for cork cambium include phellogen and extra stelar cambium. The phellogen has a much simpler structure than vascular cambium and only consists of one kind of cell. Since they become meristematic, the hypodermis or the outer layer of the cortex give birth to cork cambium. The first phellogen appears at the time of their formation in the subepidermal area. The cambium in cork may also develop as a single-layered ring.

Many vascular plants include cork cambium, which is a periderm tissue. A lateral meristem called the cork cambium is in charge of the secondary growth that takes the place of the epidermis in roots and stems. Gymnosperms, several monocots, and many herbaceous dicots—which often lack secondary growth—all include it. One of the meristems, a group of tissues made up of embryonic cells from which the plant develops, is the cork cambium. The layer of bark between the cork and major phloem is one of several. Cork production is the role of the cork cambium.

In plant science, primary growth is growth that results from cell division at the tips of stems and roots, which causes them to elongate, and which gives rise to primary tissue, whereas secondary growth is growth that results from cell division in the cambia or lateral meristems and causes the stems and roots to thicken. Most seed plants have secondary growth, while monocots often don't. If they do experience secondary development, it is not like other seed plants' regular pattern.

The action of the two lateral meristems, the cork cambium and vascular cambium, is what causes secondary growth in many vascular plants. Secondary growth, which develops from lateral meristems, widens the plant's root or stem rather than lengthening it. The stem or root will keep expanding in diameter as long as the lateral meristems continue to create new cells. This process creates wood in woody plants, transforming them into trees with thicker trunks.

Plants with secondary growth often also develop a cork cambium, which ruptures the epidermis of the stems or roots as a result of this development. Thickened cork cells are produced by the cork cambium to protect the plant's surface and reduce water loss. This technique could result in a layer of cork if it is continued for a long period of time. The cork

oak will produce cork that can be harvested. Several non-woody plants also experience secondary development, including the tomato, potato, carrot, and sweet potato tuberous roots. Some leaves with a lengthy lifespan also develop secondary growth.

The pattern of a single vascular cambium generating xylem for the interior and phloem for the exterior is not followed by abnormal secondary growth. Certain dicots exhibit abnormal secondary development, such as the Bougainvillea plant, which develops a succession of cambia outside the oldest phloem.

The majority of monocots either have no secondary development or have secondary growth that is abnormal in some way. For instance, diffuse secondary growth occurs when parenchyma cells multiply and proliferate, increasing the diameter of the trunk of palm palms. A cambium develops in several other monocot stems with abnormal secondary development, but it only creates parenchyma on the outside while producing vascular bundles and parenchyma within. Certain monocot stems thicken as a result of a main thickening meristem's activity, which develops from the apical meristem. Plants can exhibit both normal and anomalous growth, which are influenced by genetic and environmental factors. Normal growth is the typical growth pattern observed in most plants, whereas anomalous growth is a deviation from the normal growth pattern. Normal growth involves the sequential development of cells, tissues, and organs, leading to the formation of a functional plant. This process is regulated by various hormones, such as auxins, cytokinins, and gibberellins. The growth of plants can be divided into three distinct phases: cell division, cell elongation, and cell differentiation. During cell division, cells undergo rapid proliferation, leading to the formation of new cells. In the cell elongation phase, cells increase in size, leading to the elongation of tissues and organs. Finally, in the cell differentiation phase, cells become specialized and acquire specific functions, leading to the formation of different plant organs[11], [12].

Anomalous growth, on the other hand, refers to the deviation from the normal growth pattern, which can occur due to genetic mutations or environmental factors. Anomalous growth can result in various abnormalities, such as fasciation, witch's broom, and phyllody. Fasciation is a condition where the plant stem becomes flattened, resulting in a ribbon-like structure. Witch's broom is a condition where a plant develops a mass of abnormal branches, resembling a broom. Phyllody is a condition where the flower develops into a leaf-like structure, instead of a typical flower. Anomalous growth can also occur due to environmental factors such as nutrient deficiencies, water stress, and exposure to pathogens. For example, nutrient deficiencies can lead to stunted growth and abnormal development of plant organs. Water stress can cause wilting and premature aging of plant tissues, leading to reduced growth and productivity. Exposure to pathogens, such as viruses and bacteria, can also result in abnormal growth and development of plants.

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# **CHAPTER 5**

# MALE GAMETOPHYTE

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A living thing can't last indefinitely. Each species must continue to exist, and for that to happen, each individual must create more of its own kind. Students, you are all aware that plants may reproduce asexually, vegetatively, and sexually. Asexual reproduction is the process of creating new people from a single parent's cell or cells. With the exception of some instances of "automixis," the kids will be precise genetic replicas of the parents.Many kinds of asexual reproduction exist, and vegetative propagation is one of them. Vegetative reproduction or vegetative propagation is the regeneration of a plant from any vegetative portion of it, such as a stem cutting, rhizome, tuber, bulb, leaves, etc. Whereas sexual reproduction involves the union of two separate gametes from two different parents to create a new combination of genes, asexual reproduction, including vegetative reproduction, is an auxiliary means of propagation and does not require genes from different cell lineages.

A dominating sporophytic generation and a significantly diminished gametophytic generation alternate throughout the life cycle of flowering plants. It is haploid in the decreased gametophytic generation and diploid in the dominant sporophytic generation. Two crucial mechanisms are involved in the typical sexual cycle:

### **Embryo Fertilization and Meiosis**

The production of haploid spores, which are the byproducts of meiosis, is the primary purpose of diploid sporophytic generation. To become gametophytes, spores go through cell proliferation and differentiation. The production of haploid gametes is the primary purpose of gametophytic generation. The life cycle is completed when the egg and sperm fuse to form the zygote, which is the precursor to the formation of diploid sporophytes. The sporophyte generates microspores and megaspores, two different kinds of spores, during the angiosperm life cycle. Male and female gametophytes are produced by these spores, respectively. The sporophytic tissues that make up the flower's sexual organs are where the angiosperm gametophyte grows[1]–[3].

The male gametophyte, also known as the pollen grain or microgametophyte, is made up of two sperm cells wrapped in a vegetative cell and grows within the stamen's anther. The ovule, which is located within the ovary of the carpel, is where the female gametophyte, also known as the embryo sac or megagametophyte, develops. Important stages in the sexual reproductive cycle that occur in the flower include spore production and gamete development. The stamen is the floral organ that deals with male sexual reproduction, and the anther is the area of the stamen where male sexual reproduction activities take place. Similarly, the pistil, a floral organ involved in female sexual reproduction, contains the ovule, which is where female sexual reproduction activities take place within the ovary. You now understand that haploid spores are produced by meiosis or reduction division in angiosperm plants, which are diploid sporophytes, or spore-bearing plants. The female spores, known as megaspores, develop via meiosis inside the megasporangium whereas the male spores, known as microspores, do so within the microsporangium. They produce endosporous gametophytes in both the male and female genders in turn.

While the multicellular male and female gametophytes are generated inside the sporophyte's blooms, the sporophyte is the dominant generation. To create microspores, microsporangium cells inside the anther go through meiosis. While there are few more mitotic divisions, the outcome is a pollen grain. The ovary wall and two layers of integuments guard the megasporangium. Meiosis produces four megaspores—three little and one large—within the megasporangium. The only megaspore that produces the embryo sac is the huge one. As the pollen germinates and the pollen tube advances towards the embryo sac, fertilisation takes place. The seed coat's protection allows the sporophytic generation to be maintained in a latent condition.

## Anther

An anther is a part of a stamen that is fertile. Actually, the stamen is a thin organ with a proximal sterile portion and a filament containing a fertile component, the anther, at its distal end.Each of an anther's two lobes, which are joined by a connective, has two pollen chambers. Each microsporangium contains pollen grains, which help produce the male gametes, or you might say they are generated within an anther.An anther is referred to as dithecous when it has two lobes, similar to how citrus has four microsporangia. The word "monothecous" refers to an anther that has a single lobe rather than two, as in the case of the two microsporangia seen in Hibiscus rosa-sinensis. A clump of undifferentiated, thin-walled cells surrounded by epidermis makes up a newborn anther.

# Microsporangium

The anther's microsporangium, a structure, is where the male gametophyte and microspores are produced. A microsporangium, also known as a future pollen sac, is a cylindrical sac that looks round when seen crosswise. The exterior wall and the core homogenous mass of sporogenous tissue make up this structure. There are four distinct layers in the microsporangial wall:

- 1. Epidermis
- 2. Endothecium
- 3. Two to three midlayers, and
- 4. Tapetum

The anther is first seen as a homogenous mass of meristematic cells, oblong in cross section, and surrounded by a clearly defined epidermis during the formation of the microsporangium. Thereafter, it becomes roughly four-lobed, with certain hypodermal cells standing out more than others in each lobe due to their bigger size, more deeply stained cytoplasm, and obvious nuclei. These cells make up the beginnings of the archesporial system. As in Boerhaavia, there could be only one archesporial cell in each of the four lobes.

The epidermis, which is the top layer, only experiences anticlinal divisions. In order to keep up with the anther's growth, its cells flatten and significantly stretch. The epidermis fulfils its typical protective role.

The endothecium, or fibrous layer, is found underneath the epidermis. The radially extended endothecium cells eventually shed their cell contents, typically become fibrous, and create the adult anther's dry covering. As an anther is prepared to dehisce for the release of pollen, this layer has reached its maximal development. During maturity, anthers dehisce due to the presence of fibrous bands, differential expansion of the outer and inner tangential walls, and hygroscopic nature of the endothecial cells. Along the course of each anther lobe's dehiscence, the endothecium's thin-walled cells may be seen. Stomium refers to the orifice by which the pollen grains exit the pollen sac. As the anther reaches maturity, the stomium is put under stress by the endothecium cells losing water, which causes the stomium to rupture and the anther to dehisce. A mature anther typically dehisces via an apical hole or slit. Two or three middle layers are found next to the endothecium. By the time the sporogenous cells go through true meiosis, the intermediate layers are often crushed. In many species, the cells in the inner layers act as storage units for reserves of starch and other nutrients that are subsequently mobilised during pollen formation[4]–[6].

The anther wall's innermost layer, known as the tapetum, is distinguished by its rich cytoplasm and large nuclei. Tapetum has important physiological implications. It develops to its fullest potential during the tetrad stage of microsporogenesis. Any food item that is intended to feed the sporogenous tissue must pass through this nutritive tissue in order for the microspores to form.

### **Spherical tissue**

The sporogenous cells may act immediately as microspore mother cells or pollen mother cells, or they can divide a small number of times via mitosis to increase their size before going through meiosis. Although while every sporogenous cell in the anther has the ability to produce microspores, many of them regularly degenerate and are taken up by other cells.

### Microsporogenesis

Microsporogenesis is the process of forming microspores in plants. Microspores are the small, haploid cells that develop into male gametophytes, which are necessary for sexual reproduction in plants. The process of microsporogenesis begins with the formation of microsporocyte cells within the anther of the flower. These cells undergo meiosis, which results in the formation of four haploid microspores. The microspores are released from the anther and eventually develop into pollen grains, which contain the male gametophyte.

The development of microspores and pollen grains is crucial for plant reproduction, as they are responsible for delivering the male gametes to the female reproductive structure. Anomalies in microsporogenesis can result in male sterility or reduced fertility in plants, which can have significant effects on crop yield and reproductive success. Several factors can influence microsporogenesis, including genetics, environmental conditions, and stress. Research on microsporogenesis is ongoing and is important for understanding plant reproduction and developing strategies for improving crop yield and reproductive success. Microsporogenesis is the process through which pollen or microspores form.

Microsporogenesis is the name given to the process of microspore formation. Each microspore mother cell or pollen mother cell undergoes meiosis or reduction division throughout the microsporogenesis process, resulting in the formation of four haploid microspores. Four haploid microspores are contained in a single callose wall at the

conclusion of meiosis. The callose barrier that divides each individual spore from the others lacks a wall of its own. Microspore tetrads are collections of four microspores. Each spore eventually develops its own wall.

Via the use of a callose wall, a tetrad's four spores are totally cut off from one another and from the spores in other tetrads of the locule.Tetrads are divided into several categories according on how the spores are arranged. A tetrad's microspore configuration is often tetrahedral or isobilateral. However various configurations, such as decussate, linear, and T-shaped tetrads, are also discovered.

## **Tetrahedral:**

Dicots often have this kind of tetrad. When seen from an angle, only three of the four microspores are visible because they are positioned like the quadrants of a sphere, and the fourth one is located towards the rear.

## Isobilateral:

Monocots often form this kind of tetrad. The four microspores are positioned in a single plane at the four corners of a square.

T-shaped: The tetrad is shaped like a "T" because two of the four microspores are perpendicular to the others. As in, for instance, Aristolochia and Butomopsis.

### Linear:

Transverse division in the mother cell caused the four microspores to arrange themselves linearly. For instance, in several genera of the Asclepiadaceae and the Hydrocharitaceae genus Halophila.

### **Decussate:**

Magnolia has been shown to have cells arranged in a decussate manner.All five forms of tetrads in Aristolochia elegans have been documented. After meiosis, a tetrad's microspores often separate from one another as the anther develops. They are now known as pollens. As pollen grains split from the tetrad, they are uninucleate and have their own wall. Yet, it has been shown that in certain plants, the microspores of a tetrad prefer to stay together and form compound structures, such as compound pollen grains and pollinium, etc.

### Specialised pollen grains

Microspores may sometimes stay clumped together in clusters and fail to disperse. Compound pollen grains refer to these collections. Drosera, Annona, Elodea, and Typha are examples.

Development of Male Gametophyte in Angiosperm: In angiosperms, the development of the male gametophyte, or pollen grain, involves a process known as microgametogenesis. This process begins with the formation of microspores, which are produced by meiosis in the anthers of the flower.

Each microspore undergoes two mitotic divisions, resulting in a three-celled structure: a generative cell and two sperm cells. The generative cell is responsible for producing the sperm cells through mitosis. Once the pollen grain is mature, it is released from the anther

and carried by wind or animals to the stigma of the female flower. The pollen grain then germinates, sending a pollen tube down through the style and into the ovary[7]–[9].

The generative cell divides once again, producing two sperm cells. One of these sperm cells fertilizes the egg cell, forming a zygote, while the other combines with the two polar nuclei to form the endosperm, which provides nutrients to the developing embryo.Overall, the development of the male gametophyte in angiosperms is a complex process that involves multiple rounds of cell division and differentiation, culminating in the formation of mature pollen grains that are capable of fertilizing the female reproductive structures of the flower.

## Gametogenesis

Gametogenesis, the process by which male gametophytes grow, and haploid microspores or pollen, which are produced from diploid the initial cell of a male gametophyte is called the microspore mother cell as a consequence of meiosis. MMC is the last cell in the sporophytic generation, or sporophyte. Pollen grains are the soon-to-be-released microspores from the tetrad.

## Pollen grain/microspores:

The microsporangia contain pollen grains. They resemble tiny dust particles because of their very small size. A newly produced pollen grain has a conspicuous, centralised nucleus and is densely cytoplasmic. The mature pollen grain has layered walls. There are two layers in it. The inner layer is referred to as intine and the outer layer as exine. Fritsch suggested the terms exine and intine.

**Exine:** Thick, stiff cutinized layer that sometimes has smooth, sometimes spiny outgrowths. A complicated molecule termed sporopollenin makes up the exine.Within: Inside the exine lies a thin, smooth, and fragile pecto-cellulosic layer.

The pollen is more or less water resistant due to its waxy covering. The pollen grain walls are often maintained for a long time in fossil deposits because sporopollenin is resistant to both physical and biological breakdown. The pollen wall guard's pollen as it travels from one place to the stigma. Exine is absent at one or more locations. They are referred to as germ pores or slits. These are known as germ holes if they seem circular, and germinal furrows if they appear elongated. In general, pollen grains are monocolpate in monocots and tricolpate in dicots. While the male gametophyte is developing, the germ pore helps the pollen tube to emerge through it.

Several insect-pollinated species have an oily layer that forms a thick, viscous coating on the pollen particle surface. The pollenkitt is what gives the grains their stickiness, flavour, and colour. It mostly consists of carotenoid or flavonoid pigments, which give the pollen its distinctive yellow or orange colour. The glycoproteins, lipids, glycolipids, and monosaccharides found in the pollenkitt or surface pollen cement are also responsible for the pollen's stickiness. The following are some ways that pollenkitt is thought to be helping: • Serving as an insect attractant.

- 1. Serving as an adherent to the insect body due to its adhesive nature; shielding the pollen from the harmful effects of UV radiation.
- 2. Given its hydrophobic nature, it could possibly be connected to pollen distribution.
- 3. Serving as pollen-borne components of sporophytic incompatibility.

In angiosperms, the development of the male gametophyte is surprisingly consistent. The first cell of a male gametophyte is called a microspore. Just two cell divisions occur in this organism. Only generative cells are involved in the second division, which can occur in either the pollen grain or the pollen tube. Compared to the sporophyte, the male gametophyte has a much shorter lifespan[10]–[12].

A notable feature of the life cycle of all higher plants is the alternation of generation. Angiosperm plants are diploid sporophytes, or spore-bearing plants, and meiosis is the process by which haploid spores are produced. We have covered the definitions of terminology like gametophyte, male gametophyte, microsporogenesis, etc. as well as the anther's structure in this unit. Also, we learned about the stages of gametophyte growth and the mechanism of microsporogenesis.

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# **CHAPTER 6**

# FEMALE GAMETOPHYTE

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In angiosperms, the female gametophyte, also known as the embryo sac, is a crucial component of the reproductive process. It is responsible for the production of the egg cell, which is fertilized by a sperm cell to form the zygote, which eventually develops into the embryo. The female gametophyte is a complex structure that undergoes multiple rounds of cell division and differentiation to produce the seven cells and eight nuclei that make up the mature embryo sac. In this article, we will discuss the anatomy, development, and function of the female gametophyte in angiosperms.

#### Anatomy of the Female Gametophyte

The female gametophyte is typically located within the ovule, which is the structure that ultimately develops into the seed. The ovule is surrounded by integuments, which are layers of tissue that protect the developing embryo sac. The female gametophyte itself is a multicellular structure that is typically composed of seven cells and eight nuclei. These cells and nuclei are arranged in a specific pattern that is critical for successful fertilization.

The mature female gametophyte can be divided into three main regions: the chalazal region, the micropylar region, and the central region. The chalazal region is located near the base of the embryo sac and contains three antipodal cells. The micropylar region is located near the opening of the ovule and contains two synergids, which are specialized cells that are involved in guiding the pollen tube to the egg cell. The central region is located between the chalazal and micropylar regions and contains two polar nuclei, which will eventually fuse with a sperm cell to form the endosperm.[1]–[3]

### **Development of the Female Gametophyte**

The development of the female gametophyte begins with the formation of a megaspore mother cell within the ovule. This cell undergoes meiosis to produce four haploid megaspores, but only one of these megaspores survives and develops into the female gametophyte. This process is known as megasporogenesis. The megaspore then undergoes multiple rounds of mitosis and cell differentiation to produce the mature female gametophyte. The exact number of cell divisions and differentiation events can vary between different species, but in general, the female gametophyte undergoes three rounds of mitosis to produce eight nuclei that are enclosed within seven cells. The first mitotic division occurs in the megaspore to produce two nuclei. One of these nuclei migrates to the chalazal end of the embryo sac, while the other migrates to the micropylar end. The second mitotic division occurs in each of these two nuclei, producing four nuclei that are located at opposite poles of the embryo sac.

The third and final mitotic division occurs in the four nuclei, producing a total of eight nuclei that are enclosed within seven cells. The three antipodal cells are located at the chalazal end

of the embryo sac, the two synergids are located at the micropylar end, and the two polar nuclei are located in the central region.

### **Function of the Female Gametophyte**

The female gametophyte plays a critical role in the reproduction of angiosperms. It is responsible for producing the egg cell, which is fertilized by a sperm cell to form the zygote, which eventually develops into the embryo. The female gametophyte also plays a role in the development of the endosperm, which is a nutritive tissue that provides nutrients to the developing embryo. The multicellular haploid gametophyte and multicellular diploid sporophyte, which are both different physically and functionally throughout the life cycle of plants, alternate. According to Gifford and Foster, a plant's life cycle includes the following steps:

- 1. Spores grow into gametophytes from haploid spores produced by reduction division in a diploid sporophyte.
- 2. Haploid gametes are produced by gametophytes.

The zygote, the first stage of the diploid sporophyte, is created by the union of the female and male gametes. Whenever we discuss the angiosperm life cycle: Microspores and megaspores are the two kinds of spores produced by diploid sporophytes.

As was explained in the unit before, microspores grow into male gametophytes, whereas megaspores generate female gametophytes, which we shall investigate in this unit. You learned about the construction of the anther, microsporogenesis, and the growth of the male gametophyte in the previous unit. You should now be able to see that the pollens or microspores, which are the initial cell of the male gametophyte, are produced by reduction division or meiosis in the microspore mother cell inside the microsporangium.

In angiosperms, the sporophytic tissue, which houses the flower's sexual organs, is where the gametophyte grows. Inside the anther, the fertile portion of the stamen, the male gametophyte grows. The ovary's ovule, which houses the female gametophyte, is where it develops.

We discuss the structure and many ovule types in this unit, as well as megasporogenesis, megagametogenesis, and several kinds of female gametophytes. The female reproductive system of a flower is represented by the gynoecium or pistil, of which the carpel is a component. A carpel is made up of an ovary with a swelling basal portion that contains one or more ovules, a receptive stigma, and often a stalk-like style in the middle. [4]–[6]

As you have read, the ovarian wall encloses the ovules. The distribution of ovules in the ovary is referred to as placentation, and the placenta is the portion of the carpellary tissue to which the ovules are connected. The megaspore and the female gametophyte are both formed in the ovule, also referred to as the megasporangium. The latter creates the embryo and endosperm after fertilisation, while the complete megasporangium with its contained structure develops into the seed and the ancestor of the next generation.

## **Condition of Ovule**

The nucellus with its integuments, which serve as protection, make up the megasporangium or ovule. On the inner wall of the ovary, it is joined to the placenta by a stalk known as the funiculus, and the point where the ovule's body connects to the funiculus is known as the hilum.A mature ovule that is prepared for fertilisation has a nucellus that is nearly entirely encased by one or two integuments, leaving a little hole at the apical end. Micropyle is the term for this opening. This side is referred to as the chalazal end because the ovule's basal portion, where the funicle attaches it to the placenta, is known as the chalaza. The major pathway for the pollen tube's entrance into the ovule lies at the opposite end, which is known as the micropylar end. The female gametophyte, or embryo sac, is found in the nucellus.

The wall of the megasporangium is represented by nucellar tissue, which is parenchymatous. The growing embryo sac or endosperm consumes the nucellus to a large extent. There is just one nucellus per ovule. Yet, as has been shown in Aegle marmelos, two nuclei may develop abnormally inside a shared fold of integuments. Ovules with a single integument are referred to as unitegmic, those with two integuments as bitegmic, and those without an integument as ategmic.

## **Types of ovules**

On the basis of the position of the micropyle with respect to the funiculus, mature ovule can be classified into six main types. These are:

- 1. Orthotropous
- 2. Anatropous
- 3. Campylotropous
- 4. Amphitropous
- 5. Hemianatropous
- 6. Circinotropous
- 1. Orthotropous ovule, sometimes referred to as an atropous ovule. It stands erect. As in Polygonaceae and Piperaceae, the micropyle, chalaza, and funiculus all lay in a single straight line in this form.
- 2. Anatropous ovule: This form has a lengthy funiculus and an entirely inverted ovule body, causing the micropyle to be located near to the funiculus' base. The ovule's unilateral development is the cause of this. Micropyle, chalaza, and funiculus all lay in a single line because the nucellus is still straight. In Angiosperms, it is the most prevalent kind of ovule.
- 3. Campylotropous ovule: Unlike anatropous ovules, campylotropous ovules have a less pronounced curvature and a partially inverted body. As in Chenopodiaceae and Capparaceae, the micropyle and chalaza do not lay in a straight line, and the funiculus is positioned at a right angle to the chalaza.
- 4. Amphitropous ovule: Like campylotropous, but in this instance the nucellus/embryo-sac is also affected by the ovule's curvature, causing it to bend like a "horse shoe" as in Alismaceae and Butomaceae.
- 5. The hemitropous ovule is often referred to as hemianatropous. The funiculus of this form of ovule is at a straight angle to the nucellus and integuments. On the same plane as Ranunculus, Nothoscordum, and Tulbaghia are Micropyle and Chalaza.
- 6. Circinotropous ovule: Certain species of the Plumbaginaceae family have this very unusual form of ovule. Here, the nucellar protuberance initially aligns with the axis, but because to the fast development on one side, it has become anatropous. The ovule continues to curve until it has entirely turned over, at which time the micropylar end will

once again point upwards. This type of ovule, which is also found in Opuntia, has been suggested to be distinctive enough to merit its own name, Circinotropous.

Depending on the degree of nucellus development and the location of the sporogenous cell, an ovule may alternatively be classed as:

- Tenuinucellate kind 1
- Type Crassinucellate 2
- 7. Type 1 tenuinucellate the sporogenous cell is also hypodermal since the archesporial cell serves directly as the megaspore mother cell. These ovules are referred to be tenuinucellate when the sporogenous cell is hypodermal and the nucellar tissue around it is still one layer.
- 8. Type of crassinucellate: A transverse division creates an inner sporogenous cell and an outer parietal cell in the hypodermal archesporial cell. The parietal cell may split a few times or stay undivided to allow the sporogenous cell to get entrenched in the large nucellus. By splitting the nucellar epidermis, the sporogenous cell may get entrenched in the large nucellus. Any such ovules in which the sporogenous cell undergoes either of the two aforementioned processes are referred to be crassinucellate.

#### Growth of the ovule

The ovule first develops as a primordium on the placenta within the ovarian cavity. Subsequently, the protuberance becomes noticeable and develops into a mass of tissue called the nucellus as a result of the ovular primordia cells' meristematic activity. At the base of the nucellus, the initials of two integuments appear. The outer integument begins either dermally or subepidermally, whereas the inner integument, which often forms first, originates from the epidermal layer. The ovule starts to curve with the development of integuments, and by the megaspore tetrad stage, it has taken on its final form. While they begin later, the integuments expand more quickly than the nucellus. The nucellus is quickly covered by the integuments, leaving the micropyle—a tiny opening—at the tip.

There are two stages to the formation of female gametophytes:

- 1. Megasporogenesis
- 2. Megagametogenesis

The Polygonum type refers to the developmental pattern that is shown by the majority of species since Polygonum divaricatum was the first species to describe it.

#### Megasporogenesis

Megasporogenesis is the process by which the megaspore develops within the ovule.A differentiated hypodermal cell in the nucellus serves as the archesporium. Due to its size, thick cytoplasm, and sizeable nucleus, it stands out from the other cells and may be distinguished from them.As you can see from the previous section, ovules may be divided into two sorts based on where the sporogenous cell is located. As a result, the archesporial cell directly acts as the megaspore mother cell in tenuinucellate ovules, but in crassinucellate ovules, the archesporial cell does not directly act as the MMC and instead splits periclinally into two cells. a main sporogenous cell within and a primary parietal cell outside. The megaspore mother cell now performs the activities of the main sporogenous cell.

Megaspore mother cells, which have diploid chromosome numbers, are also known as megasporocytes. Meiosis, or reduced division, takes place. Four haploid megaspores are produced as a consequence. A dyad is formed when the wall is put down transversely after the initial meiotic division. In the two dyad cells, the second meiotic division is likewise transverse. This results in the formation of a row of four haploid megaspore cells. Two integuments are simultaneously growing from the ovule's base. The lowermost megaspore in the linear tetrad grows and starts to function. Three remaining megaspores from a tetrad do not contribute to the development of a female gametophyte and degenerate.[7]–[9]

The female gametophyte is now formed by the functioning megaspore. Megaspores with haploid tetrads may be T-shaped, isobilateral, or tetrahedral. A T-shaped tetrad forms when a micropylar dyad cell divides vertically and a chalazal dyad cell divides transversely.

## **Operating megaspore**

Megasporogenesis is the process by which the diploid megaspore mother cell divides into four haploid nuclei during meiosis. The three primary megasporogenesis patterns that angiosperms display are monosporic, bisporic, and tetrasporic. The key distinction between the three kinds is whether or not wall construction follows these divisions, which affects how many meiotic products contribute to the development of the adult female gametophyte.Four one-nucleate megaspores are produced as a consequence of both meiotic divisions and wall development in the monosporic pattern. Just one develops into a functioning female gametophyte. Three megaspores, often the megaspores with the greatest micropylar structure, then degenerate. The chalazal megaspore of the tetrad is the active component. As shown in Onagraceae, the micropylar megaspore may sometimes generate the female gametophyte.Embryo Sac Development in the Female with Special Reference to the Polygonum Type

#### Megagametogenesis

The adult female gametophyte is produced during megagametogenesis by the functioning megaspore. The functional megaspore, also known as the mother cell of the female gametophyte, is the initial cell of the female gametophyte, as we have previously explained. It expands in size and develops into an embryo sac. The megaspore's haploid nucleus splits during mitosis, resulting in a clear organisation inside the embryo sac. The remaining two move to the centre of the embryo sac, leaving three nuclei at the micropylar end, three at the chalazal end, and three elsewhere. At the micropylar end, three nuclei assemble into an egg-shaped structure. The egg cell, which makes up the bulk of the egg apparatus, is partly encircled by the two lateral synergid cells. Chalazal end cells with three nuclei are called antipodal cells. Polar nuclei are the two nuclei that move to the centre. A single diploid secondary nucleus is created later when these polar nuclei combine.

These processes lead to the development of a mature seven-celled structure known as the female gametophyte or embryo sac, which consists of one egg cell, two synergid cells, one central cell with two polar nuclei, and three antipodal cells. This form of embryo sac is known as a monosporic eight nucleate embryo sac or Polygonum type of embryo sac because it arises from a single megaspore and contains eight nuclei. The female gametophyte displays polarity along its chalazal-micropylar axis throughout development.

### Analysis With Respect to Tetrasporic and Bisporic Types

### **Bisporic egg sac**

According to its name, this kind of embryo sac is formed by two megaspore nuclei. A dyad is created by the creation of walls after the initial meiotic division. One of the dyad cells only progresses through the second meiotic division, while the other one degenerates just before the micropyle. Since wall construction does not occur after cell division in the functional dyad, both megaspore nuclei contribute to the creation of the embryo sac. Each megaspore nucleus experiences two rounds of mitosis, resulting in eight nuclei. The mature embryo sac is organised similarly to that of the Polygonum type.

Hence, it may be said that the two dyad cells created during meiosis I, from which the bisporic embryo sacs are derived, are 8-nucleate. In MMC, the meiotic tetrad contains four genetically distinct nuclei. A bisporic embryo sac's nuclei are genetically distinct since they are a derivation of two meiotic products.[10]–[12]

The construction of the ovule, the several forms of ovule based on the position of the micropyle in relation to the funiculus, the degree of nucellus development, and the location of the sporogenous cell have all been covered in this unit. Megasporogenesis and further ovule development, as well as female gametophyte development specifically with regard to Polygonum type, were characterised. In addition to this monosporic embryo sac, bisporic and tetrasporic embryo sacs have also been covered.

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# CHAPTER 7

### FERTILIZATION AND POST FERTILIZATION

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By going through the male gametophyte and female gametophyte units, it is now clear that gametophytic generation is haploid. The first male gametophytic cell is microspore or pollen grain. The first female gametophytic cell is functional megaspore. The pollen grains are liberated at the 2-celled or 3-celled stage. Female gametophyte is also known as embryo sac and in most of the species it is of Polygonum type. After development of male and female gametophyte, the next biological phase is pollination, which is must for fertilization. Pollination ends in a copious dusting of the stigma surface with pollen grains.

In this unit we will discuss about fertilization and post fertilization developments along with some very important phenomena occurring in the life cycle of Angiosperm plants i.e., apomixis, adventive embryony, polyembryony and parthenocarpy. The capacity to reproduce is one of the most important characteristics of life and is aimed to sustain the individual species. Reproduction methods are mainly of two types- asexual and sexual. In flowering plants sexual method of reproduction requires fusion of two gametes, one from male organ and other from female organ of the plant. The product of the fusion of two different gametes is zygote and this fusion process is known as fertilization.

In Angiosperms fertilization initiates with the compatible type pollen reaching the stigma and ends with the fusion of male and female gametes in the embryo sac. The pollens received by the female reproductive organ i.e., gynoecium is held at the stigma. There is no such way by which the pollen can reach to the egg in the embryo sac. So, to overcome this difficulty pollen germinate on the stigma and forms pollen tube which penetrate the stigmatic tissue, grows down the style, enters the ovary and finally finds its way into the embryo sac through ovule. Here it releases two sperms in the vicinity of the female gametes. Out of the two sperms, one fuses with the egg and forms zygote. The other one fuses with the polar nuclei or the secondary nucleus and forms primary endosperm nucleus. This phenomenon is known as double fertilization and is a characteristic unique feature of the Angiosperms[1]–[3].

After series of divisions primary endosperm nucleus forms endosperm. Endosperm is very nutritive tissue that nourishes the developing embryo. Zygote or oospore forms embryo, either dicotyledonous or monocotyledonous embryo, as the case may be. "Fertilization is the process of fusion of two dissimilar reproductive units, called gametes."

In flowering plants, the process of fertilization was first discovered by Strasburger in 1884. As described in Unit 6, the female gametophyte of Angiosperms is situated in the ovule, at a distance from the stigma. There is no such device developed in the gynoecium which facilitates transfer of pollen from stigma to embryo sac. Therefore, the pollen after reaching to the stigma produces a pollen tube which facilitates transport of male gametes deep into the embryo sac from stigma.

In Angiosperms, the fertilization is being completed as follows:

Germination of pollen grains and growth of pollen tube.

When the pollen is shed from anther it has usually two cells:

- A generative cell
- A vegetative cell

The generative cell forms two male gametes. Once the pollen has landed on compatible receptive stigma as a result of pollination, its germination starts. On the surface of stigma the pollen hydrates. This means pollen absorbs water from the surrounding and swells. After that the vegetative cell forms a pollen tube.

The stigmatic fluid secreted by the stigma contains sugars, lipids and resins, etc. which provides suitable medium for the germination of pollen grains. Pollen grains as well as pollen tube contain an enzyme cutinase which helps in the penetration of pollen tube into the stigmatic tissue. Cutinase as the name indicates degrades the cutin of the stigma at the point of contact with the pollen tube. The entire content of the pollen including two male gametes of generative cell move into the pollen tube.

The growing pollen tube penetrates the stigmatic tissue and pushes its way through the style and then down the wall of the ovary. The style may be hollow or solid. If it is hollow, then the pollen tube grows along the epidermal surface but in case of solid style, the pollen tube travels through intercellular spaces between the cells which lie in its path.

### Entry of pollen tube into ovule

After arriving in the ovary, the pollen tube finds its way into the ovule. The pollen tube may enter into the ovule via three routes.

- 1. through the micropyle
- 2. through the chalazal end
- 3. through the integument

On that basis of modes of entry of pollen tube into the ovule, three terms are given as follows:

**Porogamy:** When the pollen tube enters the ovule through the micropyle, the condition is known as porogamy. This is the most common mode of pollen tube entry into the ovule and so the most common type of fertilization.

**Chalazogamy:** When the pollen tube enters the ovule through the chalazal end, the condition is known as chalazogamy. This type of pollen tube entry into the ovule and so the type of fertilization is observed in Casuarina, Betula and Juglans regia. The chalagogamy was first reported by Treub in Casuarina.

**Mesogamy:** When the pollen tube enters the ovule through the integument or through the funiculus, the condition is known as mesogamy. This type of pollen tube entry into the ovule and so the type of fertilization is observed in Cucurbita, and Pistacia.

Therefore, depending on the place of pollen tube entry into the ovule, fertilization may also be called of three types:

- 1. Porogamous
- 2. Chalazogamous
- 3. Mesogamous

## Entry of pollen tube into the embryo sac

It does not matter through which way pollen tube enters into the ovule; it always enters in the embryo sac from the micropylar end means entry of pollen tube in the embryo sac is irrespective of pollen tube entry into the ovule. So, we can say that synergids not only play an important role in determining the entry of pollen tube in the embryo sac but them also affect dissemination of male gametes in the embryo sac.

## Discharge of male gametes from pollen tube

After reaching the embryo sac the pollen tube burst at its tip and deliver the male gametes. Just prior to bursting of pollen tube the tube nucleus disorganizes. Immediately after releasing, the male gametes show amoeboid movement and one male gamete moves toward the egg and other one move to the polar nuclei. As the one of the male gametes reached the egg, it fuses with it. As a result of this fusion diploid zygote/oospore forms. The fusion of male and female gametes is known as fertilization. This is also known as syngamy. One of the most significant discoveries was made by Strasburger in 1884, as mentioned above. He observed the actual fusion of the male gamete with the female gamete in Monotropa[4]–[6].

## **Post Fertilization Developments**

After fertilization, development of the embryo and the endosperm within the ovule goes side by side. The oospore, formed as a result of fusion of one male gamete with the egg, develops into the embryo while the primary endosperm nucleus- product of triple fusion, develops the endosperm. The other nuclei or cells within the embryo sac disorganize sooner or later.

# **Development of the Endosperm**

The primary endosperm nucleus is a product of triple fusion. This undergoes a series of divisions and ultimately forms endosperm. The Angiospermic endosperm is a triploid tissue as it is a product of triple fusion. It is formed either by the fusion of one haploid male gamete and one diploid secondary nucleus or by the fusion of three haploid nuclei.

It is therefore distinct from the endosperm of heterosporous Pteridophytes and Gymnosperms where the endosperm is a simple haploid tissue of the gametophyte not involving any triple fusion like in Angiosperms. Endosperm is a highly nutritive tissue which provides nourishment to the developing embryo.In Orchidaceae and Podostemonaceae, the product of double fertilization soon disintegrates and endosperm development is completely suppressed.

Depending upon mode of development three types of endosperms has been recognized:

- Nuclear endosperm
- Cellular endosperm
- Helobial endosperm

Of this nuclear endosperm is the most common type which occurs in about 56% families of Angiosperms. It is followed by cellular endosperm and then by helobial endosperm.

### Nuclear endosperm

In this type of endosperm, the division of primary endosperm nucleus and number of subsequent nuclear divisions are not accompanied by wall formation and the nuclei thus produced remain free in the cytoplasm of the embryo sac. They remain in the peripheral layer of the cytoplasm surrounding a large central vacuole. Wall formation occurs at later stage around nuclei. The wall formation is mostly centripetal i.e., from the periphery towards the centre and usually begins from the basal periphery e.g., Arachis hypogea.

In some cases, the central vacuole may not be filled up even in the mature seed. This is seen in the palms. Cocus nucifera is the classical example of this type of nuclear endosperm. Development of endosperm in it deserves special mention. The primary endosperm nucleus undergoes a number of free nuclear divisions. Then the embryo sac gets filled with a clear fluid in which numerous nuclei float. It is known as liquid syncytium. Gradually nuclei start settling at the periphery with the beginning of peripheral cell wall formation. This forms the coconut meat. In mature coconuts the liquid endosperm becomes milky. The watery endosperm of coconut contains growth promoting "coconut milk factor $\Box$  and that is why it is used as a nutrient medium in plant tissue culture experiments. Nuclear endosperm is commonly found in polypetalous dicotyledons.

## **Cellular endosperm**

In this type of endosperm, division of the primary endosperm nucleus is immediately followed by wall formation so that the endosperm is cellular from the beginning. The first wall is laid down transversely but the subsequent divisions are irregular. Adoxa, Peperomia, Villarsia etc. are some common examples.

### **Helobial endosperm**

Among members of Helobiales there is type of endosperm the development which is intermediate between the nuclear and the cellular type. Here the first division of the primary endosperm nucleus is accompanied by the formation of transverse wall. This divides the embryo sac unequally into two compartments - a small chalazal chamber and a large micropylar chamber. This step is followed by free nuclear division in both the chambers but there are relatively more free nuclear divisions in micropylar chamber in comparison to chalzal one. The chalazal chamber often degenerates. The free nuclear divisions in the micropylar chamber are followed by wall formation and thus a cellular endosperm tissue is formed and usually found in the members of the order Helobiales.

It is the endosperm, on the basis of which seeds can also be categorized into two categories.

- 1. Non-endospermic seeds
- 2. Endospermic seeds
- 3. Non- endospermic seeds

In plants where the entire endosperm consumed or utilized in the nutrition of the developing embryo, the mature seeds thus formed are without endosperm. Such seeds are termed as nonendospermic seeds. Example are seeds of beans, peas etc. The non-endospermic seeds store their food material in cotyledons.

## **Endospermic seeds**

In plants where the seeds retain endosperm even at maturity and do not consumed or utilized the endosperm completely in the nutrition of the developing embryo. Such seeds are said to be endospermic seeds. Example are seeds of coconut, castor etc. The endosperm present in the seed is utilized after germination in the establishment of young seedlings.

## **Development of the embryo**

After fertilization, a series of changes occurs in the ovule and finally seed is formed. Side by side with the development of the endosperm, the oospore is developing the embryo after a period of rest.

The process of development of mature embryo from diploid oospore is called embryogenesis.Both dicotyledons and monocotyledons begin embryo development in the same way but there is considerable difference in later differentiation. Before proceeding let us discuss about the dicotyledonous and monocotyledonous embryo.The dicotyledonous embryo as the name reflects, has two cotyledons attached laterally to an embryonical axis, whereas in the monocotyledonous embryo, the embryonical axis has a single cotyledon at its apex. Due to this organographic difference, it is very easy to distinguish the two types of embryo but there is no fundamental difference in their early stage of development. The development is very similar till the globular stage.

In all Angiosperms the embryogenesis starts with the division in oospore and it divides to develop a two-celled proembryo by forming a transverse wall. The cell near the micropyle is termed the basal cell and the cell facing towards the centre of the embryo sac is called the terminal cell. The basal cell forms the suspensor and may or may not contribute in rest activities so sometimes called as suspensor cell, whereas terminal cell is responsible for further development of embryo so called embryo cell[7]–[9].

### Types of embryo development

On the basis of plane of division of the terminal cell in the 2-celled proembryo and the contribution of the basal cell and terminal cells in the formation of embryo proper, six types of embryogeny have been reported by Johansen among the Angiosperms.

### Development of dicotyledonous embryo

The classical example is Capsella bursa-pastoris of Brassicaceae. The ovule is campylotropous so that the embryo sac and the later developed endosperm as well as embryo are horseshoe-shaped. Here the development of embryo is Onagrad or Crucifer type.

- 1. Zygote divides transversely. As a result of this a two-celled proembryo is formed.
- 2. The larger basal cell at the micropylar end is called suspensor cell. The smaller one, away from it termed as terminal cell or embryo cell.
- 3. The suspensor cell divides transversely a few times to produce a filamentous suspensor of 6- 10 cells. The suspensor helps in pushing the embryo in the endosperm.
- 4. The first cell of the suspensor becomes swollen and called haustorium or vesicular cell.
- 5. The last cell of suspensor is known as hypophysis. It forms radicle and root cap.

6. The embryo cell undergoes two vertical divisions and one transverse division to form quadrant and then octant stage. In octant, eight cells arranged in two tiers- epibasal and hypobasal.

The epibasal cells eventually form the two cotyledons and the plumule. The hypobasal cells produce the hypocotyl. For this the octant embryo undergoes periclinal divisions producing outer layer of protoderm, procambium and ground meristem.

Protoderm forms epidermis, procambium gives rise to steal or vascular strand and ground meristem produces cortex and pith. It is initially globular but with the growth of cotyledons it becomes heart-shaped and then assumes the typical shape, e.g., Capsella bursa-pastoris.

## **Structure of Dicot Embryo**

The mature embryo consists of an embryonal axis having two cotyledons. Embryonal axis above the level of cotyledons forms the plumule and below the cotyledons, the radical. Upon germination the plumule forms the shoot and the radical gives rise the root system. The reserve food material in the cotyledons is used in the establishment of young seedlings.

## Development of monocotyledonous embryo

There is no essential difference between the embryogeny of monocotyledons and that of dicotyledons but as a single cotyledon develops instead of two from the embryo in monocotyledons, there is some difference in later stages.

We are taking an example of Luzula forsteri of Juncaceae for describing the development of monocotyledonous embryo. Here the development of embryo is also Onagrad or Crucifer type.

- 1. The early development of dicot and monocot embryos are similar upto octant stage. Later on, differentiation starts.
- 2. The zygote or oospore elongates and then divides transversely to form basal and terminal cells.
- 3. The basal cell produces a large swollen, vesicular suspensor cell. It may function as haustorium.
- 4. The terminal cell divides by another transverse wall to form two cells.
- 5. The top cell after a series of divisions forms plumule and a single cotyledon.
- 6. Cotyledon called scutellum, grows rapidly and pushes the terminal plumule to one side. The plumule comes to lie in a depression.
- 7. The middle cell, after many divisions' forms hypocotyl and radicle. It also adds a few cells to the suspensor.
- 8. In some cereals both plumule and radicle get covered by sheaths developed from scutellum called coleoptile and coleorhiza respectively.

# **Structure of Monocot Embryo**

The embryos of monocotyledons have only one cotyledon. In grass family, this cotyledon is called scutellum. It is situated towards lateral side of embryonal axis. This axis at its lower end has radicle and root cap enclosed in a sheath called coleorhiza. The part of axis above the level of attachment of scutellum is called epicotyl. It has shoot apex and few leaf primordia

enclosed in a hollow foliar structure called coleoptile. Epiblast represents rudiments of second cotyledon.

# Apomixis

# "Reproduction without fertilization"

Apomixis in flowering plants is defined as the asexual formation of a seed from the maternal tissues of the ovule, avoiding the processes of meiosis and fertilization, leading to embryo development.

The term Apomixis was first coined by Hacke in 1893. Apomixis, derived from two Greek word "Apo" and "mixis". Winkler explained the term apomixis as the substitution of sexual reproduction by any such method which does not involve meiosis and syngamy. or we can say that Winkler used the term apomixis to signify any asexual method of propagation not involving the normal production of embryo by fertilization. It includes even propagation by bulbils.

When we are talking about asexual formation of seed, in this sense apomixis is synonymous with agamospermy: seed formation without fertilization of the egg cell. In some plants meiosis which converts a diploid sporophytic cell into four haploid gametophytic cells and fertilization where two haploid gametes of opposite sex fuse to re-establish the diploid sporophytic generation, the two very important necessary processes of sexual cycle are interrupted. Even then a viable embryo if formed resulting into asexual seeds. When these asexual seeds produce plants identical to the female parent are called apomictic seeds and the phenomenon is known as apomixis.

**Non-recurrent apomixes:** non-recurrent means which cannot be repeated. In this type of apomixis, the megaspore mother cell undergoes normal meiotic division and one of the four megaspores thus formed develops into haploid female gametophyte. However, there is no fertilization and the embryo arises directly from normal egg-cell. Since an egg cell is haploid, the resulting embryo will also be haploid and so sterile, therefore the process cannot be repeated in the next generation[10], [11].

Haploid parthenogenesis and haploid apogamy are non- recurrent apomixis. Such types of apomixis are of rare occurrence.

**Recurrent apomixes:** Recurrent means which can be repeated. In recurrent apomixis, the nuclei of the embryo sac are usually diploid. Such embryo sac may arise either from a cell of the archesporium due to disturbance in meiosis or from some other cell of the nucellus due to disintegration of megaspore mother cell.

The embryo subsequently develops directly from the diploid egg-cell without fertilization. Somatic apospory, diploid parthenogenesis and diploid apogamy are recurrent apomixis. However, diploid parthenogenesis/apogamy occurs only in aposporic embryo-sacs. Therefore, it is the somatic or diploid apospory that constitutes the recurrent apomixis. Such apomixis occurs in some species of Crepis, Taraxacum, Paa, and Allium without the stimulus of pollination.

Adventive apomixes: In it, the development of embryo takes place from any diploid cell of the ovule lying outside the embryo sac. Since it takes place outside the embryo sac, it is not

grouped with recurrent apomixis, though this is regenerated with the accuracy. In addition to such embryos, regular embryo within the embryo sac may also develop simultaneously, thus giving rise to polyembryony condition as in Citrus, Opuntia.

**Vegetative apomixes:** In some cases, like Poa bulbosa and some Allium, Agave and grass species, vegetative buds or bulbils, instead of flowers are produced in the inflorescence. They can also be reproduced without difficulty. However, Russian workers do not group this type of vegetative reproduction with apomixis.

Apomixis does not involve meiosis, so there is no segregation and recombination of chromosomes. Therefore, it could be useful in preserving desirable characters for indefinite period.

## Parthenogenesis

Parthenogenesis means development of an embryo directly from an egg cell or a male gamete or it may be defined as - the development of female gamete into a new individual without fertilization. Parthenogenesis may be haploid or diploid as the case may be.Haploid parthenogenesis: Generally, normal haploid egg develops into an embryo, so the embryo and resultant plant are haploid. This type of parthenogenesis is termed as haploid parthenogenesis e.g., Oenothera, Datura. Plants thus produces are sterile.

Diploid parthenogenesis: When the cells of embryo sac including egg cell are already diploid as a result of apospory. This diploid egg when develops parthenogenetically into diploid embryo, termed as diploid parthenogenesis e.g., Taraxacum.

## Apogamy

Apogamy is the development of a sporophyte out of any gametophytic cell without fertilization i.e., the union of gametes. Plants formed in this way are sterile because they are haploid. Example - Lilium, Nicotiana. One of the two synergids develops into embryo in Lilium while male gamete forms embryo in Nicotiana by apogamy.

### Apospory

Apospory was discovered by Rosenberg in Hieracium species. In this type, megaspores are formed by usual process but all the four megaspores degenerate gradually. At the same time, somatic cells, usually nucellar cells enlarge and functions as initials of embryo sac. These initials enlarge, undergo mitotic divisions and develop embryo sacs. This type of apospory is also called as somatic apospory. Aposporic embryo sacs are diploid. It means the formation of gametophyte on a sporophyte without any reduction division.

# **Adventive Embryony**

Adventive embryony is an embryony where an embryo develops directly from any diploid sporophytic cell for example- cells of nucellus, integument etc., without formation of gametophyte. This is also known as adventitious embryony or nucellar embryony. This may be considered as vegetative growth of the category of bulbils. Sometimes this is called sporophytic budding.Or in simple way we can describe it as - cells outside the embryo sac also develop into embryos. These embryos are known as adventives embryos and the process as adventives embryony.Adventive embryony has great significance in horticulture and plant

breeding. It provides uniform seedlings of the parental line as obtained through vegetative propagation by cuttings.

### **Polyembryony and Parthenocarpy**

After fertilization, ovules mature into seeds. In normal case, a single embryo is present in each seed but sometimes more than one embryo may present in a seed. When a seed contain more than one embryo, this condition is termed as polyembryony. Therefore, polyembryony has been defined by many workers as the occurrence of more than one embryo in a seed or "The development of several embryos within the same ovule."Polyembryony is very common among Gymnosperms but when we are talking about Angiosperms, it is very rare. You can find it in Citrus species like lemons, oranges and also in few Quercus species. Additional embryos do not always mature. They may degenerate during the course of development. The mature seed thus has only one embryo. The first case of polyembryony was reported by Antoni van Leeuwenhoek in 1719 in certain orange seeds. Since then, it has been observed in large number of plants[12].

### **Classification of Polyembryony**

In broad sense it is of two types:

- 1. Spontaneous- includes instances of naturally occurring polyembryony.
- 2. Induced- includes instances of experimentally induced polyembryony.

Ernst divides spontaneous polyembrony into two categories:

- 1. True polyembryony- development of two or more embryo in same embryo sac
- 2. False polyembryony development of embryos in more than one embryo sac within the same ovule

Yakovlev divides spontaneous polyembrony into two categories on genetic basis-

- 1. Gametophytic: arising from any gametic cell of the embryo sac after or without fertilization.
- 2. Sporophytic: arising from the zygote, proembryo or the initial sporophytic cells of the ovule.

In this unit we have discussed about fertilization, pathway of pollens to their destination for fertilization. After that we have also learnt post fertilization development process. Along with these topics light have been thrown on apomixis, adventives embryony, polyembryony as well as on parthenocarpy.

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# CHAPTER 8

### PLANT MORPHOGENESISAND MORPHOGENETIC FACTORS

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Morphogenesis is defined as a process concerned with formation and development of whole plant, a part of plant or a specific structure. During very early developmental stages polarity is established at the zygote stage due to which a polar difference is developed at both the ends of zygote. Cytological differences at the two ends determines the position of first cell division and also the fate of the structure which will be produced by the two cells. Polarity is not limited to initial developmental stages but polarity is maintained throughout the growth. Plant axis also exhibits polarity. If a portion of shoot is excised and allowed to regenerate the end toward shoot tips will always form shoot whereas end towards root will regenerate roots. As in stem polarity is also exhibited in other organs like upper and lower surface of leaf, petals, sepals etc. Different parts of plant have different type of morphology. This diversity in different parts of plant is produced due to variation in growth rate of different parts and also because different parts show growth in different dimension. Rate of cell division, cell elongation along with orientation of plane of division and axis of cell elongation altogether establish the form of structure of plant.

Different factors affect growth and development of plants. These factors can be environmental such as light, temperature, water or nutritional factor, physical factors such as gravity, pressure and genetic factor. Genes are considered to be the ultimate factor which growth but they do not regulate growth independently. Instead, genes interact with existing environmental conditions to control plant development[1]–[3].

During their growth and development plant, cells exhibit a specific phenomenon called as totipotency. It is the ability of a cell to give rise to different types of cells and eventually lead to regeneration of a complete plant. Meristematic cells get differentiated to attain specific functions once the cell gets differentiated. They lose their ability to divide. However, differentiated plant cells can undergo a process of dedifferentiation and can again become meristematic. Now, such dedifferentiated cells can again redifferentiate to form cells and tissues with specific structure and function.

#### **Basic Idea of Morphogenesis and Concept of Differentiation**

The word morphogenesis comes from Greek words morphe and genesis to indicate a process of formation of a particular structure with a specific shape and size. Morphogenesis is considered to be a biological process which causes an organism to develop its shape. Morphogenesis is concerned with development of particular part or structure. Plants possess a longer period of morphogenesis. During development plants do not exhibit a distinct body plan. Plants may grow and develop on and on till they die. This is because plants have meristematic tissue composed of actively dividing cells which result in formation of more and more new tissues, organs and structures throughout the life of plant. The term differentiation was first of all used by Karl Willhelm. Differentiation refers to a process in which distinct types of cells are formed from a precursor cell. Differentiation is a permanent localized qualitative change in size, biochemistry, structure and function of cells, tissues or organs. A cell which has ability to get differentiated into different cell types of an adult organism is called pluripotent. In plants such cells are also called as meristematic cells.

Different type of structural changes occurs inside a cell during the process of differentiation. These changes may occur in cell wall, protoplasm or both. For example, when a cell gets differentiated into tracheary elements it loses its cytoplasm and the cells develop an elastic, strong, lingo cellulosic secondary cell wall to carry out transport of water.

Hence, meristematic cells are group of unspecialized cells which are capable of dividing throughout the life of plant and can get differentiated into different types of cells. When a cell gets differentiated it acquires specific morphological, physiological as well as biochemical properties. During growth and development of plant meristematic tissue give rise differentiated tissue where each cell has specified structure and function. Differentiated tissue loses its differentiated state and becomes undifferentiated. Such undifferentiated tissue can again undergo the process of differentiation known as redifferentiated cell can divide and produce new cells. Dedifferentiation is a commonly observed phenomenon during secondary growth in plants and also during the process of differentiation to give rise to different and possesses an inherent ability to undergo process of differentiation to give rise to different types of cells, which ultimately form different organs in plant system[4]–[6].

#### Polarity

The term polarity means specific orientation of plant activity and morphogenesis in space. Plants are multicellular organisms made up of cells, tissue and organs. As we already know that in a multicellular organism cells, tissue and organs are integrated with one another to bring about overall functioning of an organism. There are many factors which regulate and control this integrated functioning. Among different factors polarity is one the most important factor of plant integrity. In plants axial polarity, dorsiventral polarity and radial polarity are known. However, when we talk of polarity in plants we generally refers to axial polarity. Axial polarity means presence of a well-defined longitudinal axis which bears lateral organs such as lateral branches, roots, leaves and flowers. The radial axis is most clearly evident in dicotyledonous species as the concentric rings of cell layers stem, hypocotyl and root with an increase in size across this axis can arise from the generation of new cell layers following divisions in the vascular cambium in the older plant.

There are several factors which influence polarity in plants. Physical factors like light, gravity, electric and magnetic field, chemical agents such as plant growth regulators and ions influence polarization in plants. Polarization is related to axial gradient of bioelectric potential which develop from gradient of Ca2+, K+, H+ etc. Changes in membrane permeability to these ion generates a dielectric potential. Results obtained from studies conducted on plants such as Arabidopsis, Capsella bursa-pastoris have made it clear that apical-basal polarity is determined even before the first zygotic division in the egg.

Early events of zygote polarization have been very well studied in Fucus . In Fucus polarization of zygote is initiated and influenced by various types of stimuli such as unidirectional light, temperature, electric field or chemical gradient. Axis formulation is associated with redistribution of plasma membrane components. Ca++ is the most important component which gets accumulated toward basal end during axial axis function.

In Arabidopsis during axial polarization, zygote divides by an asymmetric transverse division resulting in formation of two daughter cells of unequal size. One is the basal cell which is derived from vacuolar region and is larger in size and another cell is smaller upper cell which is derived from cytoplasmic region. Upper cell divides to form suspensor. Only the upper most cell of suspensor called hypophysis is actually the part of embryo proper.

Although suspensor cells are known to have different functions such as they physically project the embryo into endosperm, avail a source of hormone and nutrient to the developing embryo, the suspensor cells undergo programmed cell death when embryo reaches its torpedo stage of development. In Fucus the larger upper cell is known to form thallus cell from which develops the thallus structure of mature alga. On the other hand, the small basal cell forms rhizoid which undergoes polarized growth. In ferns and mosses polarity can be induced by membrane bound biliprotein phytochrome. There are two system under which induction of polarity in plants have been studied. The first system of polarity in plants is ROOT- RHIZOID POLARITY, this type of system studied in phaeophycean zygotes and in pteridophytic spores. Development of polarity occurs parallel to ionic gradient of calcium, potassium and sodium. During polarization an increase influx of calcium ions occurs into the cell present in the future rhizoid pole. On the contrary a decreases influx of calcium ions occurs in the opposite pole. Another system of polarity is Shoot-Root Polarity found in higher plants:

The earliest work related to shoot-root polarity was done by Marquis Duhamel du monceau in eighteenth century. In his work existence of two morphogenetic factors was proposed, one was a heavy root sap and another a light shoot sap. Both morphogenetic factors were directed by gravity to their respective poles, where they got accumulated and shoot sap-initiated formation of shoots and root sap gave rise to roots. Zygote displays a specific cell polarity with a vacuolar pole present at micropylar site and an opposed cytoplasmic pole. Establishing polarity is an important event for morphogenesis and development of plant. Particularly in plants polar differences can be identified at very early stage of development after the formulation of zygote. During the process of development of plant, polarity can also be seen in plant axis i.e. in shoot and root tips. This means that once a polarity is established it does not gets altered naturally. So, if a part of shoot or root is existed and allowed to regenerate the end toward shoot tips always regenerates into shoot and the opposite end will develop roots. However, during the process of development either removal of one part of plant or changes in a part of plant significantly affect morphogenesis of one or more other parts of plant. This process is called as correlation and is generally mediated through nutrient and plant growth regulators[7]–[9].

# Totipotency

### **Basic concept of totipotency**

Totipotency refers to inherent genetic potential of a plant cell to regenerate into complete plant. Plant cells can follow a developmental pathway similar to that of a zygote resulting in

formation of new plant. The concept of regeneration the entire plant from a single cell or tissue was conceptualized by G. Haberlandt in 1902, who is known as father of plant tissue culture.

F. C. Steward along with his colleagues developed a method for growing carrot tissue by taking small part, from the secondary phloem region of carrot root. This part was utilized as explants in the experiment. The explants were cultured by placing it onto a liquid medium under aseptic conditions. During the culture process the phloem tissue began to grow. Initially some single cells and some groups of cells became loosened from the surface of growing tissue and started growing separately. Some single cells developed somatic embryos or embryoids by a process now known as somatic embryogenesis. The embryo ultimately gave rise to shoot and root and the complete plant was regenerated.

## **Importance or significance of Totipotency**

- 1. The most important aspect/application of totipotency is reconstruction or regeneration of complete plant from any tissue or organ.
- 2. Regeneration of plants from somatic cells through their ability totipotency has been utilized for vegetative propagation of many medicinal, aromatic and ornamental plants with economic importance.
- 3. With the development of plant tissue culture technology large number of plants can be produced in short time interval. Totipotency is the underlying principle of regeneration of plants through plant tissue culture. Hence, endangered, rare and scarce plants can be mass propagated through the technique.
- 4. Advancements made in plant science have resulted in development of genetically modified plants. Production of homozygous plants, haploid plants, somatic embryogenesis, somatic hybridization, protoplast culture etc. Totipotency is the basic of all the above mentioned developments made in plant science.
- 5. In vitro regenerated cells, tissue, callus with totipotency potential can be preserved for long periods under liquid nitrogen. The process is known as cryopreservation. Whenever required
- 6. These cells can be retrieved thawed and can be utilized for regeneration

## Totipotency and plant tissue culture

Plant tissue culture also known as in vitro micropropagation is a technique utilized for regeneration of plants under controlled conditions. The technique has been successfully utilized for regeneration, conversation of large number of medicinal, aromatic, ornamental and other plants on a large scale. The entire success of plant tissue culture technology is based upon the totipotency of plant cells. Normally, we grow plants mainly through seeds or by methods of vegetative propagation including cutting, grafting, layering etc. But through the technique of tissue culture plants can be regenerated by culturing any part of the plant. The part of plant excised to culture is called as explant. Explants are transferred to a culture medium aseptically. The cultures are then incubated under suitable temperature with proper light.

Now, during the process of incubation the explants which are differentiated tissue undergo the process of dedifferentiation and become undifferentiated and totipotent. Explants now undergo the process of redifferentiation and start growing to regenerate a new plant. When totipotent cells undergoes the process of differentiation to form different types of cells and organ there are basically three types of pathways which can be followed:

**Embryogenesis:** Totipotent cells can divide and differentiate to give rise to embryoid structure. Formation of embryo is a bipolar structure and same structure give rise to root as well as shoot i.e. complete plant.

**Organogenesis:** division and differentiation of totipotent cells may also result in formation of organs. If totipotent cell give rise to only shoot it is known as caulogenesis. However, rhizogenesis is process of formation of roots. In another process called as Caulorizogenesis formation of shoot as well as root occurs simultaneously.

**Histogenesis / Cyto differentiation:** Totipotent cells may divide and differentiate and give rise to tissues like xylem, phloem etc.

Dedifferentiation & redifferentiation occurs, different type of growth response can be obtained from explants. In tissue culture process growth can be of two types direct and indirect. In direct growth formation of organs occurs directly from explant whereas in indirect growth first a callus is formed. By further sub culturing this callus regeneration of shoot and roots can be obtained. The plant regenerated in laboratory conditions are then transferred to soil through a process known as hardening or acclimatization.Hence, totipotency forms the basis of plant tissue culture through which large number of plants can be regenerated in comparatively shorter duration of time.

There are several advantages of plant tissue culture.

- 1. Production of large number of plants.
- 2. Conservation of endangered species.
- 3. Production of hybrid plants.
- 4. Synthesis of secondary metabolites.
- 5. Production of virus resistant plants through meristem culture.

### **Morphogenetic Factors**

Basic concept and effect of morphogenetic factors

Morphogenetic factors are physiological factors which induce regulate and coordinate morphogenetic events in plants. These factors can be a part of inner or outer environment of the plants.

Morphogenetic factor can be divided into two groups:

- 1. Environment factors
- 2. Genetic factors

Plants are multicellular organisms which survive in an environment which is complex and keeps on changing. Genetic makeup remains unchanged except for rarely occurring somaclonal variation. Now, even since there is no change in genetic constitution of plant but plants do exhibit phenotypic changes i.e., their appearance changes or gets modified with changes in their environment. Such phenotypic changes which occur in plants are considered to have occurred due to environmental factors. However, it is quite difficult to judge whether

the morphogenetic change occurring in plant is due to a genetic factor or an environmental factor since both environmental as well as genetic factors are operating simultaneously.

Responses such as flowering, thickness of cuticle, height of plant are greatly influenced by environment and gets altered according to the changing environment.Whereas characters such as formation of pits on side walls of vessels, arrangement of leaves etc. do not change with the change in environmental conditions. The degree of lobbing in leaves is greatly influenced by changes in temperature.Another class of factors which influence plant growth or morphogenesis are nutrients. They act as chemical factors, come into plant body from outside and participate in biochemical process occurring inside the plant. There are several growth substances which significantly influence morphogenesis in plants.There are three possible attributes of action of morphogenesis factors.

It is not necessary that a morphogenetic factor may directly result in a response but it may act as a stimuli to trigger other biochemical reaction in an organism.One morphogenetic factor can significantly influence or modify the action of another factor. No factor can act independently; response mediated by each factor is dependent upon the environment as well as on the status of plant.A plant is not a constant system i.e., character of plant changes from one phase of life cycle to another and also from one part of plant to other part of the plant. Hence, plants may exhibit different response to same morphogenetic factor in different phases of life cycle. And also, different part of plant may respond differently to same morphogenetic factor.

#### Morphogenetic effect of light

As we all know that light is one of the most crucial factors for growth, development and survival of plants. Light is required by the plants for vital processes such as photosynthesis, photo morphogenesis etc. Beside these processes, light also influences several other physiological processes. One of the most prominent effects of light as morphogenesis factor is that any plant reaches its maximum height with optimum growth only when the plant is exposed to sufficient amount of light. If insufficient light is provided the plants exhibited retarded growth even if supplied with sufficient water, nutrients and temperature.

There are three aspects of light which influence growth of plants:

**Intensity:** It is the measure of brightness of light or other illumination i.e., actual energy of the radiation.

Quality: It refers to wavelength of the light perceived by plants.

**Duration:** By duration it means the length of lightness and darkness to which a plant is exposed.

The effect of light can have different effect on different parts of plant. Some of the effect of light on plants are:

- 1. Rate of photosynthesis generally increases with increase in intensity of light to a certain extent.
- 2. Intensity of light also affects qualitative traits such as strength of stem, development of xylem and phloem etc.
- 3. Plants grown in shade have comparatively small root system.

- 4. Light intensity is directly proportional to width of stem.
- 5. Some herbaceous plants show zig-zag growth pattern in light but grow straight if same plants are grown in darkness.
- 6. Whenever we think of light and plants. We get an image that light is required for photosynthesis by plants. But light is also needed by plants which lack chlorophyll.
- 7. Etiolation is an important effect of light intensity. Plant grown in darkness are somewhat with pale leaves, weak roots and poorly developed xylem and phloem.
- 8. Longer wavelength of light enhances elongation cells and tissues whereas blue light tends to present elongation.
- 9. Quality of light also effects flowering in plants.
- 10. Beside quality duration of light also effects flowering in plants.
- 11. The length of photoperiod may also affect differentiation of sex e.g., in Cannabis sativa, when 16-hour photoperiod is given flowering occurs within 4-6 week. About half plants are male and half females. However, same plants when provided with 8 hours photoperiod, enhanced and fast development occurs with flowering occurring within 3-4 weeks an about half the plants are hermaphrodites and half females.

### Morphogenetic effect of water

Water is another important morphogenetic factor which influences growth, development and morphogenesis in plants. Water is one of the keys requires for photosynthesis to occur. Deficiency of water results in phenomenon known as xeromorphy. On the contrary presence of excess amount of water results in small roots. Poor development of mechanical and vascular tissue, leaves become then stomata are reduced or absent. These traits are generally regarded as adaptation to survive in aquatic environment.

It has been found through several studies conducted by different scientist that there exists a definite correlation between the amount of water passing through the vascular tissue and the amount of vascular tissue developed.Water also exerts other morphogenetic effects. Development of positive hydrostatic pressure generally occurs at early and rapid leaf growth and leads to formation of larger leaves. When the hydrostatic pressure is low at later stage smaller leaves are developed.

### Morphogenetic effect of temperature

For all living organisms including plants temperature is a crucial factor which influences morphogenesis as well as metabolic processes occurring inside the organism. A peculiar feature about temperature is that most of the response mediated by temperature are equally affected by light. The most important effect of temperature is on the growth of plant. Like any other living organisms plants also need an optimum temperature for growth and development. However, the optimum temperature may vary from one plant species to another, same plant may require different temperature during different phase of life cycle and moreover optimum temperature may be different for different region of plant. As we study the concept of photoperiodism similarly there exists thermoperiodism. It refers to daily rhythm in reaction to temperature. If plant is provided with constant temperature throughout 24 hours, many plant show slow growth as compared to the growth obtained when the same plants are grown in comparatively warmer days and cooler nights.

A plant usually contains many buds out of which several buds do not develop. Significant amount of study has been conducted to find out which factors or growth substances decide that which bud will develop and which will not. Temperature is one the crucial factors which influences breaking of bud dormancy. Low temperature is considered to be an effective treatment for breaking dormancy.

Another morphogenetic effect of temperature is observed in form of vernalization. Vernalization is a process of providing low temperature for induction or acceleration of flowering. For some plants vernalization is a must for flower to occur. In Horticulture practice, seeds and seedlings are intentionally given treatment of low temperature to induce early flowering. In a study conducted by Burstrom it was found that exposure to high temperature results in reduction in length of root cells. This is due to shorter period of cell elongation.

#### **Morphogenetic effect of Mechanical Factors**

Physical factors such as compression, tension, bending and swaying, gravity also effect growth and development of plants. These factors are also referred to as mechanical factors. These factors may be called as mechanical but they are quite simple in character as compared to temperature and light. Mechanical factors influence morphogenesis indirectly by affecting the physiological process occurring in plants.

There are plants which display thigmotropism. This type of response also involves morphogenetic changes. For example, when a tip of a tendril is touched by another branch or wire any other material tendril tends to coil around the wire or branch to provide support to the plant. This response involves enhanced growth of tendril in the direction of the support. When the stem of herbaceous plant bents, smaller cells are formed on convex side where as thick-walled cells are formed on concave side. This difference is due to mechanical strain. Cells on convex side are under tension and cells on the concave side are under compression.

Gravity is another factor which influences growth of plants. Unlike other morphogenetic factors gravity is continuous, unchanging in intensity and also constant in direction. Downward growth of primary root, upward growth of main stem, etc are considered to be manifestations of geotropic growth reaction. Effect of gravity and light appears to be indistinguishable from one another. A change in relation to one generally produces a change in relation to the other. However, unlike light gravity exerts an indirect effect on plant. Gravity is also known to play an important role in distribution of growth substances.

#### Morphogenetic effect of chemical factors

Chemical factors also affect morphogenesis in plants. Normally chemical factor is known to execute their effect on physiological processes occurring inside an organism but beside this they do affect form and structure of plant. Till now we have studied about factors such as light, temperature, water, gravity which execute their effect on plant through external environment. But chemical factors influence morphogenesis through external as well as internal environment. There is another peculiar feature of chemical factors that their effect can be localized to a particular part of plant instead of affecting the whole plant. Effect of chemical substance varies from time to time and from one phase of plant life cycle to another. Different elements are required by living organisms for several physiological functions. Elements such as O, N, K, Mg, C, Ca are considered to be macro elements. Since they are

needed in larger amounts on the other hand elements such as B, Cu, Zn, Co, Mn is known as microelements as they are required in micro quantities by living organisms.

Nitrogen is essential constituent of all the proteins. Nitrogen is also reported to enhance growth of plants. In a study conducted by Burkholder and Mc Veigh in maize plant displayed better meristematic growth and enhanced length and diameter of stem when cultivated in presence sufficient quality of nitrogen. Nitrogen is also known to enhance differentiation in phloem with increased growth of sieve tube and vessels.

The ratio of C/N is also known to affect morphogenesis. Nitrogen is known to support vegetative growth hence plants having low C/N ratio tends to possess few flowers or fruits. Whereas when the ratio of C/N is high abundant flowering and fruiting occurs. Studies conducted have also related C/N ratio to the ratio of shoot length and root length.Phosphorus is another element which is a prime constituent of nucleic acid. Besides being an important part of DNA and RNA, phosphorus also promotes cell division in roots but has little effect on elongation of stem. If we compare effect of phosphorus to that of nitrogen, elongation of stem is promoted by nitrogen but nitrogen does not directly affect cell division.

Calcium is known to support formation of cell wall. However, calcium is not directly a part of composition of cell wall but it produces its effect by bringing changes in cytoplasm. Zn is a trace element but is known to have an indirect effect on maintaining auxin in its active state. Boron is also required for cell wall formation. Deficiency of Boron causes hypertrophy and hyperplasia of tissue.Plant hormones better known as plant growth regulators also control and coordinate morphogenesis in plants. Auxin and cytokinin remain to be the most significant plant hormones, along with them ethylene is crucial for fruit ripening, Gibberellic acid for germination. Almost all the morphogenesis response or growth shown by plants is mediated by one or the other hormone.

### **Morphogenetic Effect of Genetic Factor**

Genes are known to have specific response to a specific environment. We are very well familiar with George Mendel $\Box$ s law of genetics. In his first law called as Law of dominance he described how inheritance of genes governs formation of tall or short plants in Pisum sativum. Both types of plant can be easily differentiated from one another based upon their genetic composition. Transcription and translation of genes leads to synthesis of enzymes which directly regulate or control growth and morphogenesis in plants. Generally, any morphogenetic trait is not entirely controlled or affected by a single gene but many genes or polygenes collectively affect morphogenesis. One of the key effects of genes on morphology is seen in extent of growth as well as on distribution of growth. Several examples are available where shape of leaves, flowers, and fruits is inherited and controlled by gene expression[10].

Lamprent in his study found that in pea plant there is a long distance between first and second flower as compared to the total length of inflorescence. This is believed to be controlled by three genes. In corn grass due to a single gene dominant mutation result in formation of narrow leaves, many tillers and a smaller number of male flowers as compared to normal plant. In another plant Aquilegia canadensis a dwarf race with bushy and compact growth differs from normal plant by a single gene. There is another example of Acetabularia in which control of gene over form and morphology was determined. This alga has a branching,

rhizoid base from this base rises a stalk which has a hat A single large nucleus is found to be located in the basal rhizoids.

There are two species of Acetabularia one is longer and another shorter. Both the species also differ in form of hat. In Hammering  $\Box$ s grafting experiment stalk was excised from longer species and grafted onto the basal portion of shorter species, now a new hat formation will begin from the stalk, at initial stages the newly formed hat may look like the hat to the species to which stalk belongs but finally the hat formed was similar to the hat of species which contributed rhizoid. Hence, it was clear that formation of hat in Acetabularia is controlled by gene present in nucleus.

Some plants develop perfect or complete flower having both male and female flowers. Such flowers are also known as hermaphrodite or bisexual and when male and female flowers develop on the same plant i.e. some flowers will be male and some flowers will be female such plant is called monoecious and the condition is known as monoecism whereas when male and female flowers develop on separate plant as seen in case of animals and the plant is called dioecious and the condition is known as dioecism. In this case a male plant will develop only male flowers and female plant will develop only female flowers. Some common examples of dioecious is strawberry.

These types of sexual development in flowers are controlled by specific gene. However, environmental factors equally contribute to development of sex of flowers.Genes also play a crucial role in production and distribution of growth substances which in turn affects morphology of plant.

Genes also control photoperiodic effect which regulates flowering in plants. As a result of gene mutation, the flower time and season may get altered[11], [12].

Most plant are haploid i.e., two sets of chromosomes in each nucleus. In some plants number of sets of chromosomes is multiplied. Such plants are called polyploids. Polyploids plants are believed to exhibits better growth in terms of leaves size, enhanced number and size of fruits and flower etc. But this increase is restricted to certain level only. Plants with ploidy level higher than tetra or pentaploid show negative growth in terms of number and size of leaves, flower, fruit and other growth parameters.Morphogenesis is defined as a process concerned with formation and development of whole plant, a part of plant or a specific structure. During very early developmental stages polarity is established at the zygote stage. Due to this polarity difference at both the ends of zygote is established according to which different structures are developed at different poles. Axial polarity is most significant polarity pattern in plants which is represented by longitudinal axis which bears lateral organs such as lateral branches, roots, leaves and flowers.

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# **CHAPTER 9**

# PLANT GROWTH REGULATORS

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The plant growth regulators are small, simple molecules of diverse chemical composition. They are organic substances synthesized in one part of the plant body and transported to another part where they are active. PGRs are also known as plant hormones or phytohormones. They are the regulators produced by the plants which in low concentrations regulate the metabolic process. The PGRs can be broadly classified into two categories:

- 1. Growth promoters Growth inhibitors, depending on their growth promoting or inhibiting activities respectively.
- 2. Generally, five types of chemical growth regulating systems have been identified by plant physiologists namely Auxins Gibberellins or GAs Cytokinins Abscissic acid or ABA Ethylene. In this chapter PGRs of major importance are being described.

### Auxin

## Tryptophan

The term auxin is derived from the Greek word auxein which means to grow. Compounds are generally considered auxins if they can be characterized by their ability to induce cell elongation in stems and otherwise resemble Indole Acetic Acid in physiological activity. Auxins usually affect other processes in addition to cell elongation of stem cells but this characteristic is considered critical of all auxins and thus "helps" define the hormone. Although there is only one naturally occurring auxin: indole-3-acetic acid, there are many synthetic auxins as we shall see in laboratory.

### **Discovery of Auxin**

In the late 1870s, Charles Darwin and his son Francis were one of the earliest scientists who studied phototropism. The Darwin studied coleoptiles of canary grass and oats. They discovered that both plants grow toward the light source.

The Darwin followed up this discovery with these experiments:

- 1. The tips of coleoptiles were covered with a metal foil. This blocked the incoming light and the coleoptiles did not grow toward light. When the foil was removed, they grew toward the light.
- 2. The growing region of the coleoptiles rather than their tips was covered and they discovered that the coleoptiles grew toward the light. The conclusion was made that the growth of coleoptiles toward light was controlled by the tip of the coleoptiles.
- 3. The Darwin suggested that, "phototropism was due to an 'influence' produced in the tip of a coleoptile that moved to the growing region, where it caused the coleoptile to grow toward light." Their discovery helped later scientists discover plant hormones.

Peter Boysen-Jensen further developed on the Darwin  $\Box$ s experiments:

- 1. He cut off the tip of a coleoptile and noticed that it stopped its growth, which showed that something within the tip of the coleoptile controlled growth.
- 2. He then separated the tip from the coleoptile with a tiny piece of agar. He observed that the coleoptiles grew and curved toward the light. He concluded that the tips of the coleoptiles did not have to be in their normal position to affect growth, and the chemical that controlled phototropism moved through agar therefore it was a water-soluble chemical.
- 3. He replaced the agar block with butter. Since water is insoluble in butter, any watersoluble chemical from the tip could not move through the butter into the growing region. He observed that there was no growth, and from this concluded that the chemical was water- soluble.
- 4. To test if the signal was electrical he replaced the agar blocks with pieces of foil, and there was no growth. Therefore, the signal was chemical rather than electrical
- 5. In 1918, Arpad Paal continued on with Boysen-Jensen's experiments to identify the chemical. He studied coleoptiles grown in the dark:
- 6. He cut off the tips of coleoptiles grown in the dark, and placed them on one side of the cut surface. These curved away from the side onto which the tips were placed, despite them being in the dark. The curvature was identical to that of the plants growing toward light . Paal concluded that the coleoptile's tip produces something that travels down and stimulates growth, and that light causes the accumulation of the chemical on the shaded side of the coleoptile.
- 7. Frits Went finalized all these experiments: He cut off the tips and placed the cut surfaces onto agar. The tips were removed after an hour and the agar was placed on the cut tips of the coleoptiles grown in the dark. Went's different experiments and results:

Cut off coleoptiles & without agar blocks, did not grow. This confirmed that the tips produced something essential for growth.Agar blocks that contacted cut tips were placed on the center of the cut off coleoptiles and they grew straight up. Therefore, the chemical diffused into the agar from the coleoptile tips, and stimulated their growth.Agar blocks that did not contact the cut tips of coleoptiles did not show any response. Therefore, nothing in the agar caused growth of the coleoptile.Agar blocks that had contacted the cut tips when placed on one side of the cut off coleoptiles, curved away from the agar blocks. This confirmed that the agar blocks had a chemical that stimulated growth of coleoptiles.Went came to the conclusion that the phototrophic response was due to a chemical coming from the coleoptile's tip. He named this chemical auxin which comes from a Greek word meaning "to grow."

### Synthesis

The most active auxin in plants is indole-3-acetic acid and its most active areas of synthesis are in young leaves, fruits, flowers, shoot tips, embryos, and pollens. Some synthetic compounds have auxin like effects; such as 2, 4-D and NAA. 2, 4-D is used as an herbicide because it is relatively cheap and non-toxic to humans. Other uses of synthetic auxins are that they are used to produce roots on cuttings, prevent pre-harvest dropping of fruits and prevent lateral buds from growing[1]–[3].

### **Effects of Auxin**

**Apical dominance:** In higher plants the apical bud is far more active than the lateral buds. For certain period the growth of the lateral buds is suppressed. This phenomenon is called as apical dominance. According to Thimann and co-worker's auxin is responsible for the dominance of apical bud. Apical dominance seems to result from the downward transport of auxin produced in the apical meristem. In fact, if the apical meristem is removed and IAA applied to the stumps, inhibition of lateral buds is maintained.

The common white potato is really a portion of the underground stem of the potato plant. It has a terminal bud or "eye" and several lateral buds. After a long period of storage, the terminal bud usually sprouts but the other buds do not. However, if the potato is sliced into sections, one bud to a section, the lateral buds develop just as quickly as the terminal bud

**Parthenocarpy:** As a result of pollination the auxin level of ovary is raised resulting in fruit formation, when the ovary is converted into fruit without occurrence of fertilization, the phenomenon is called as parthenocarpy. The auxins are applied in low concentration in a lanolin paste to the stigma of the flower and, as a result parthenocarpy is induced.

**Root initiation:** The auxins induce rooting in stem cuttings. Application of IAA in low concentration at the cut end of stem induces formation of adventitious roots. Besides, IBA, NAA, and 2, 4-D are also successfully used for this purpose. This property of auxins is of great economic importance for multiplying plants by cutting in nurseries.

**Prevention from abscission:** Auxin also plays a role in the abscission of leaves and fruits. Young leaves and fruits produce auxin and so long as they do so, they remain attached to the stem. When the level of auxin declines, a special layer of cells - the abscission layer - forms at the base of the petiole or fruit stalk. Soon the petiole or fruit stalk breaks free at this point and the leaf or fruit falls to the ground.

If the blade of the leaf is removed, as shown in the figure, the petiole remains attached to the stem for a few more days. The removal of the blade seems to be the trigger as an undamaged leaf at the same node of the stem remains on the plant much longer, in fact, the normal length of time. If, however, auxin is applied to the cut end of the petiole, abscission of the petiole is greatly delayed. Fruit growers often apply auxin sprays to cut down the loss of fruit from premature dropping

**Removal of weeds:** A concentration of auxins like 2, 4-D and 2, 4, 5-T destroys dicot weeds. The roots are sensitive to auxins. They block their sieve elements and disturb mitosis. The plant is ultimately destroyed.

**Stimulation of respiration:** The auxins induce respiration, perhaps by providing more ADP to be converted to ATP.

The auxins also bring about shortening of internodes. This property of auxins is useful in apple where flowers and fruits are borne on dwarf shoots. By auxin sprays more dwarf shoots are formed.

### Gibberellins

Some Japanese farmers observed a particular disease in rice seedlings on account of which they become thin, tall and pale. They called the disease as bakanae or foolish seedling. It was later on found that these seedlings were infected with the fungus Gibberella fujikuroi. Sawada thought that the disease was caused due to some substance secreted by the fungus. This was experimentally confirmed by Kurosawa. Yabuta and Sumiki, were first to extract a crystalline substance from the fungus which they named as gibberellin.

All gibberellins are acidic in nature whose carboxyl group is marked by esterification. Hence, they are chemically called as gibberellic acid and abbreviated as G.A. Cross and co- workers isolated six gibberellins from the fungus Gibberella which are identified as GA.1, GA.2, GA.3, GA.4, GA.7 and GA.9.

In same year Mac Millan and co-worker isolated three gibberellins from bean seeds which they identified as GA.5, GA.6, and GA.8. So far about sixty gibberellins have been isolated, of which some 15 have been obtained from the fungus Gibberella. The seed plants contain about 51 gibberellins. All the gibberellins contain gibbane ring skeleton.

The gibberellins are present in all groups of plants i.e., from algae to Angiosperms but rarely in fungi and bacteria. They may be synthesized at places where they are needed or they may be transported from other regions. The transport is done by diffusion through xylem as also through phloem.Their transport is non-polar. The young leaves are the main sites of gibberellin synthesis. In contrast, the older leave shave only a little ability to do so. The gibberellin synthesis in young leaves renews the activity of the vascular cambium. The roots synthesize gibberellins in sufficient amount. If the roots are repeatedly excised, a marked decreased in the concentration of the gibberellins is observed in the shoot. This suggests that dicot the shoot's gibberellin is derived from the root's via xylem elements. The immature seed contain a higher percentage of gibberellin in comparison to mature seeds. This increased protection is due to synthesis and not due to transport. In grass seeds the gibberellin synthesis mainly occurs in the scutellum and probably in others parts too[4]–[6].

### Mechanism of action

It stimulates cell division in shoot tip. The cell growth is promoted by the increase in the hydrolysis of starch, fructose, and sucrose. It further well plasticity. Most of the workers are the opinion that the effect of gibberellin is indirect since they act by altering the auxin status. In the aleurone layer of barley the gibbrerllin increase the transcription of genes that code of protease and amylase enzymes. The protease activity produces tryptophan which is translocated to coleoptile tip where it is converted to IAA. The IAA shows polar movement.

### **Physiological effects**

- 1. Stem elongation: The gibberellin induces internodal elongation. When treated with gibberellins, the lettuse plant became vine like.
- 2. Light induced stem growth: Dark grown etiolated plants are lean, tall, and yellow. Perhaps light has inhibiting effect on stem growth. It is thought that light induced inhibition of stem growth is overcome by exogenous application of gibberellins.
- 3. Genetic dwarfism: By the application of gibberellins, Lang observed a rapid growth or 'bolting' on some dwarf plant. Similar behavior has been observed in dwarf rice, maize, watermelon, squash, and cucumber.
- 4. Promotion of flowering in long-day plants: The long day plants generally possess a basal rosette of leaves. Before flowering they show significant internodal growth. After receiving the minimum hours of day light requirement, they bolt and flower.

The continuation of rosette form or bolt and flower is linked with the amount of native gibberellins present in the plant.

- 5. Increase in flower and fruit size: By the exogenous application of gibberellin the size of flower of Geranium and Camellia increased. Similar application of gibberellin also increased the size of the fruit of Vitis.
- 6. Parthenocarpy: Like auxins, the exogenous application of gibberellins also induced the production of parthenocarpic fruits. They are also applied to the stigma in a lanolin paste.
- 7. Substituting cold treatment: By exogenous application of gibberellins, many biennials can be induced to behave as annuals and they no more require the natural chilling treatment for their flowering.
- 8. Breaking of dormancy: The exogenous application of gibberellins has been shown to be capable of breaking the dormancy of potato tubers and buds of trees in winter.

In addition to above, the gibberellins promote hypocotyl growth, increase in the number and size of leaves, expression of apical dominance, delay the senescence of leaves and Citrus fruits, sexual development of flowers particularly the maleness as well as the enzyme activity.

### **Commercial uses of Gibberellins**

Commercially the gibberellins are employed in the following:

- 1. Increase in the size of Thompson seedless grape fruits as also the distance in between them.
- 2. Increase in the height of sugar cane plant and more sugar yield.
- 3. Increase in the fresh weight of pastures and hay crops.
- 4. In storage of oranges, gibberellin prevents rind disorder by delaying senescence.

## Cytokinin

Haberlandt observed that the vascular sap is capable of including cell division activity in wounded potato tubers. Similar capability was observed by Van Overbeek et al. in coconut milk. Miller isolated an active substance from autoclaved DNA from Herring sperms which stimulated cell division. He named this substance as Kinetin. It was identified as 6-furfurylamino purine. Several names were given to this substance such as Kinetenoid by Busstorm; Phytokinin by Osborne; Cytokinin by Letham and so on. However, the name cytokinin was preferred over other names and abbreviated as CK. Skoog et al. Defined cytokinin as 'chemicals' which regardless of their activities promote cytokinesis in various plant organs'

The most common and physiologically active cytokinins are zeatin, dihydrozeatin, and isopentanyl adenine. Benzyladenine was regarded as a synthetic cytokinin but Ernst found 6-benzyladenine riboside in the seeds of Pimpinella anisum.

### Zeatin

It was obtained by Letham in crystalline form from immature maize grain who named it as zeatin. Later, Letham et al. Identified as 6 aminopurine. Its synthesis was done by Wilson. According to Letham the zeatin and zeatin ribose are most active compound. The higher activity of zeatin is perhaps due to a very reactive allylic-OH group in its side chain.

# Distribution

Beside higher plants, the cytokinins occur in diatoms, red and brown algae as well as mosses. In these plants they promote growth. A few pathogenic bacteria and fungi also contain cytokinins. In non-pathogenic bacteria they influence nodulation and formation of mycorrhiza. In higher plants cytokinin occurs in the root extract of sunflower, pea seedling, flower and fruits of apple, pear, plum, and tomato, cambial tissue of eucalyptus and tobacco, young fruits of maize, banana and walnut, fruit and embryo of peach, liquid endosperm of coconut and tumor tissue of tobacco.

# **Biological activities**

- 1. **Cell division:** The cytokinins induce cell division activity. It is the most characteristic property of cytokinin. By its application a normal cell of Vinca was converted to a tumor cell. They also induce cell division activity in bacteria such as E. coli.
- 2. **Cell elongation:** They induced cell enlargement. It was observed in disc etiolated leaves as well as tobacco pith and cortical cells. This property is enhanced in combination with auxins.
- 3. **Morphogenesis:** By applying cytokinin and IAA in balanced combination, tobacco pith cells produced callus. If the amount of cytokinin is increased, differentiation of buds was observed in the callus.
- 4. **Breaking dormancy:** The dormancy of seeds of Lactuca sativa is broken by a spray of cytokinin. It is thought that the site of cytokinin action on seed germination in the cotyledon.
- 5. **Suppression of apical dominance:** The cytokinin counteract the phenomenon of apical dorminance. In fact, this phenomenon is controlled by a balance of concentration between endogenous IAA and cytokinins.
- 6. **Delay in senescence:** Richmond and Lang observed that the degradation of proteins and chlorophyll was delayed in the detached leaves of Xanthium, if there was cytokinin in the medium. The phenomenon of delay in senescence by cytokinin treatment is also caused as Richmond-Lang effect.

# Abscissic Acid

When a fruit ripens or before a leaf falls, a special zone of cells is formed at the base of the pedicel or petiole. This zone is called as abscission zone. It is delimited by a protection layer on the stem side and a separation layer on the organ side. The fruit or leaf is ultimately separated and the phenomenon is called as abscission. A Swedish botanist, Hemberg observed that the extract of dormant potato tubers and the buds of Fraxinus excelsior inhibit the growth of Avena coleoptiles. Osborn also observed that the extract of old leaves causes premature fall of young leaves in bean plants. Addicott and co-workers isolated two compounds, Abscissin I and Abscissin II, responsible for the abscission of cotton fruits. Robinson and warming isolated a substance responsible for dormancy in Acer pseudoplatanus and named it as dormin. It was later on found that abscissin II and dormin were identical compounds. Addicott suggested the name Abscissic acid which was widely accepted. It is a naturally occurring hormone which appears to be present in all vascular plants and some mosses but not in bacteria, algae, most of fungi and liverworts[7]–[9].

## Structure of Abscissic acid

The ABA is a terpenoid having an asymmetric carbon. It is a C-15 sesquiterpene. Most of the hormone is synthesized in leaves and fruits. Some of ABA in chloroplast can arise from xanthophyll, violaxanthin.

## Physiological Effects of Abscissic Acid

- 1. Seed and bud dormancy: Abscissic acid induces dormancy of buds towards the approach of winter. Abscissic acid accumulates in many seeds during maturation and apparently contributes to seed dormancy.
- 2. Senescence: ABA acts as a general inducer of senescence. The onset of senescence is correlated with stomatal closure. The ABA content of aging leaves increases markedly as senescence is initiated.
- 3. Flowering: In long-day plants, the effect of gibberellins on flowering is counteracted by ABA, which accumulated in the leaves during the short winter days. This ABA acts as inhibitor of flowering in long-day plants. On the other hand ABA induces flowering in short-day plants.
- 4. Starch hydrolysis: The GA-induced synthesis of a-amylase and other hydrolytic enzymes in barley aleurone cells is inhibited by abscissic acid. This inhibition can be reversed by increasing the amount of GA supplied.

### Ethylene

It was observed in 1864 that the gas illuminating the streets in German cities, due to leakage to pipes caused leaf fall in road side shade trees. This gas was thought to be responsible for causing senescence and abscission of leaves. Later, in 1879 it was observed that an illuminating gas promoted ripening of oranges. The gas was identified as ethylene in both the cases. Galston and Davis recognized it as a growth regulator.

Physiological Effects of Ethylene

- 1. Unripe fruits can be made to ripe before proper time if they are kept in ethylene atmosphere.
- 2. In some plants, it stimulates germination of seed.
- 3. It inhibits root and stem elongation but induces root hair formation.
- 4. It also induces cellulose activity leading to promotion of leaf abscission.
- 5. It induces petal discoloration.

The plant growth regulators are small, simple molecules of diverse chemical composition. They are organic substances synthesized in one part of the plant body and transported to another part where they are active.

The immature seed contain a higher percentage of gibberellin in comparison to mature seeds. This increased concentration is due to synthesis and not due to transport. In grass seeds the gibberellin synthesis mainly occurs in the scutellum and probably in others parts too. The gibberellin induces internodal elongation. By the application of gibberellins, Lang observed a rapid growth or 'bolting' on some dwarf plant. Similar behavior has been observed in dwarf rice, maize, watermelon, squash, and cucumber it is also effective in genetic dwarfism, promotion of flowering in long day plants, increase in flower and fruit size, parthenocarpy and breaking of dormancy. Skoog et al. Defined cytokinin as 'chemicals' which regardless of

their activities promote cytokinesis in various plant organs'. The most common and physiologically active cytokinins are zeatin, dihydrozeatin, and isopentanyl adenine. Beside higher plants, the cytokinins occur in diatoms, red and brown algae as well as mosses. In these plants they promote growth.

The cytokinins induce cell division activity. It is the most characteristic property of CK. They induced cell enlargement morphogenesis. They also effective in breaking dormancy, suppression of apical dominance and delay in senescence. The phenomenon of delay is senescence by CK treatment is also caused as Richmond-Lang effect. Robinson and warming isolated a substance responsible for dormancy in Acer pseudoplatanus and named it as dormin.

It was later on found that abscissin II and dormin were identical compounds. Addicott suggested the name Abscissic acid which was widely accepted. Abscissic acid induces dormancy of buds towards the approach of winter. Abscissic acid accumulates in many seeds during maturation and apparently contributes to seed dormancy.ABA acts as a general inducer of senescence. It is also effective in flowering and starch hydrolysis. Ethylene gas are helpful for unripe fruits which can be made to ripe before proper time if they are kept in ethylene atmosphere[10]–[12].

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# **CHAPTER 10**

## PHYSIOLOGY OF FLOWERING

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After attaining certain growth, the plants begin to flower. Flower is reproductive organ of plants and most of the plants utilize the process of flowering as a mode of reproduction. Flowering is followed by pollination, fertilization which ultimately leads to formation of fruits/seeds. The time taken by a plant to flower varies from species to species. For example, so many fruiting trees which you commonly see around such as mango tree, guava tree etc., take many years before they begin to flower and fruit. Many herbs begin to flower in few months. Such plants have very short vegetative phase and the reproductive phase begins early. Different plant species may exhibit different pattern of growth before they begin to flower, for example corn plants does not begin to flower until they have produced certain number of leaves. Plant of bamboo takes several years to flower. Flowering in plants crucially depends upon season. Each plant displays a strict and definite pattern of their vegetative and reproductive growth depending upon season. It means that every plant requires specific seasonal/ environmental condition before they begin to flower. In this chapter you will come to know how seasons, length of day and night control flowering in plants.

Flowering is the process by which a plant produces its reproductive structures, such as flowers, fruits, and seeds. Flowering is essential for the survival of angiosperms, or flowering plants, as it allows them to reproduce sexually and produce offspring. The process of flowering is regulated by various internal and external factors, including hormones, light, temperature, and day length. The process of flowering can be divided into several stages, beginning with the induction phase, in which the plant receives signals to initiate the flowering process. These signals may come from various sources, such as photoperiod (the length of daylight and darkness), temperature, and plant hormones such as gibberellins and cytokinins. Once the plant has received the necessary signals, it enters the developmental phase, in which the meristem (the growing tip of the plant) transforms from a vegetative to a reproductive state. This is achieved through the activation of floral meristem identity genes, which initiate the formation of floral buds[1]–[3].

The next stage is the differentiation phase, in which the floral meristem differentiates into the various organs of the flower, including sepals, petals, stamens, and carpels. The differentiation of these organs is regulated by various genes and hormones, which control their growth and development. Once the flower has formed, it enters the maturation phase, in which the various organs mature and become capable of performing their reproductive functions. The sepals and petals become colorful and attract pollinators, while the stamens and carpels become capable of producing and receiving pollen. Pollination occurs when pollen is transferred from the stamen (the male reproductive organ) to the carpel (the female reproductive organ), either by wind, water, or animal pollinators. Once pollination has occurred, fertilization can take place, in which the male gamete fuses with the female gamete to form a zygote, which develops into a seed.

## **Basic Concept of Flowering**

After attaining certain vegetative growth, plants undergo structural and functional changes and reproductive growth begins leading to flowering. Flowering in plants is influenced by various experimental factors. Plant will respond to environmental factors only when the plant has reached certain stage of maturity. It means that if a plant is provided with all the favourable conditions required for flowering but if the plant is not mature enough it will not flower. Flowering crucially depends upon developmental status of plants among different environmental factors, length of days, quality and intensity of light and temperature are among the most important factors which control flowering in plants.

After sufficient amount of vegetative growth, if plant is provided with suitable environmental conditions, the development of plant shifts towards reproductive growth. Several changes occur at metabolic level including changes in kind and number of hormones produces, production of metabolites required for reproductive growth, etc. Stimulus for flowering is perceived by leaves, the flowering stimulus from leaves in form of hormones is transferred from leaves to shoot tips / nodes. Formation of floral buds occurs at shot tips which ultimately results in flowering.

### Photoperiodism

The term photoperiodism was suggested by Garner and Allard and the term photoperiodism refers to effect of length of day and night on growth and development of plants. Photoperiod is the favourable day length required by plants mainly for flowering to occur.Garner and Allard first of all reported the phenomena of photoperiodism. They observed that mutant tobacco plant and soyabean follow seasonal dependent pattern of flowering. Soyabean plant flower only in late summer irrespective of the time when the seeds were sown. Effect of various environmental factors such as nutrition, soil moisture on flowering has been analyzed and it was found that none of these factors played a key role in regulating flowering. When plants were placed in dark and provided with shorter light period, flowering was obtained in plants. After this, similar experiments were conducted on different plant species under different photoperiods and found that it was length of day which control flowering in plant.

### Short - day flowering plants

These are those plants which flower when length of day is shorter than a critical period. These plants need a day length shorter than a critical period to flower. If day length exceeds a critical value then short-day flowering plants fail to flower. For example, in soyabean day length of more than 12 hours effectively reduced number of flowers. These plants are also called as long night plants.

### Characteristic features of short-day flowering plants

Short - day flowering plants need continuous /uninterrupted long period of darkness to flower. Hence, you can say that in SDP length of day is not as important as period of darkness.SDP will fail to flower if the continuous period of darkness is interrupted by weak intensity of light given for some time. The plant will also not flower even if a flash of light is given during period of darkness. Moreover, even if weak intensity is given to plant for sometimes during the period of darkness flowering is inhibited. These plants can be made to

flower in long day conditions as well by transferring to plants to darkness for sufficient duration.

It is obvious that length of night is more crucial for flowering in SDP than day length. If plants are kept in complete darkness and provided with sucrose externally. They exhibit normal pattern of flowering indicating that the photoperiod is required only for the process of photosynthesis.

SDP do not flower under alternating cycles of lightness and darkness. The period of darkness which is needed by SDP for flowering showed be continuous. Suppose if a plant requires 16 hrs of darkness for flowering and the plant is given 16 hrs of darkness but not continuously instead in four instalments of 4 hrs each. Now the total period of darkness is 16 hrs but the plant fails to flower because period of darkness is not given continuously. Some examples of SDP are: tobacco, soyabean, strawberry, coffee, rice, Bryophyllum, maize [4]–[6].

## **Types of SDP**

**Qualitative short - day plants:** Also called as absolute or obligatory short-day plants. These plants will flower only under short- day conditions and will never flower under unsuitable photoperiod e.g., strawberry, coffee, maize.

**Quantitative short-day plant:** Also called as facultative short- day plant. These plants best flower under short-day conditions. However, they may also flower under long- day conditions but with delayed flowering e.g., cotton.

**Short long-day plants:** Plant which flower when placed under short -day conditions followed by long days e.g., white clover.

## **Long-day Flowering plants**

These are those plants which flower when provided with longer photoperiods. They need day length longer than a critical period to flower. More than, the requirement of longer photoperiod these plants require short period of darkness for flowering because larger period of darkness inhibits flowering in this plant. Hence, long-day plants are also called as short night plants.

Characteristic features of long-day flowering plants:

- 1. Long- day flowering plants flower best in continuous light. They either need little or no darkness for flowering.
- 2. Long period of darkness exhibits an inhibitory effect on flowering in long- day plants.
- 3. Long day- plants can flower in short -day conditions if the period of darkness is interrupted by flash of light.
- 4. Unlike short-day plants long- day plants flower normally if light and dark period are provided alternately. Flowering occurs because dark period is not maintained for longer duration and hence cannot exhibit its inhibitory effect on flowering e.g. pea, peppermint, barley, rye Grass, wheat, radish.
- 5. Long- day's plants flower when provided with a photoperiod of more then critical length. Period of darkness is believed to have somewhat inhibitory effect on flowering, hence if period of darkness extends beyond a limit flowering is inhibited. However, if a flash of light is given during the period of darkness, the inhibitory effect of darkness is

compensated and their plant exhibits normal pattern of flowering. LDP also exhibits flowering kept under continuous light. LDP will also flower if exposed to alternate period of light and darkness.

## **Types of LDP**

**Qualitative long-day plants:** These plants are also called as absolute long-day plants or obligatory long day plants. These are those plants which flower under only under long day conditions and will never flower under unsuitable / improper) photoperiod e.g., oat, radish

**Quantitative long-day plants:** Also called as facultative long day plant. These are basically long day flowering plant and flower best under long day conditions. However, they may also exhibit flowering under short day delayed conditions e.g., turnip, garden pea, spring wheat.

**Long short-day plant:** plants which exhibited flowering when provided with long days followed by short day treatment / exposure e.g., Aloe, Kalanchoe.

### **Day- neutral Plants**

These are those plants which can flower in all photoperiods. There is no seasonal preference for flowering in these plants e.g., tomato, bean, cucumber.Normally short-day plants flower when day length is shorter than 11 hours and for long-day plants day length period of 14-16 hours is sufficient for flowering. However, day length may vary from species to species.

**Ambiphotoperiodic plants:** These are those plants which flower when photoperiods are shorter than certain length or longer than certain period. Madia elegans flowers when photoperiod is either shorter than 14 hours or longer than 18 hours.

**Intermediate-day plant:** These are those plants which flower when exposed to dark period of certain length. For example, Salsola komarovii flower when provided with darkness period of 12 hours. The plant fails to flower of under short as well longer night length. You can consider these plants as modification of short-day plants.

### **Quality of light**

Green color of visible spectrum has been reported to be ineffective in inducing flower and orange red color is by far the most effective wavelength to inhibit flowering. Blue light is known to induce poor flowering. Red portion of the visible spectrum with wavelength 580nm-680nm have been found to be the best portion of spectrum to initiate flowering in short day as well as long-day plants.

## Site of photoperiodic induction

M.K. Chailakhyan demonstrated that stimulus for flowering was perceived by leaves. He utilized Chrysanthemum plant for the purpose. Leaves were removed from above portion of the plant. After defoliation plants were divided in four groups A, B, C, D.Group a plants were exposed to long days. Upper defoliated portion of group B plants was exposed to long days where lower leafy region was given short day treatment. Contrary to this, upper defoliated portion was provided with long days. Group D plants were provided with short day treatment. Plants of Group B and D exhibited development of flower since leaves in both the group were exposed to short day photoperiod. Leaves perceived the stimulus for flowering, synthesized

flowering hormone, which was transported to defoliated upper part and as a result flowering occurred.

In an experiment three short day plant were taken. In the first plant no changes were done, whereas in second plant the plant was completely defoliated and in third plant only one leaf was left intact rest all leaves were removed. All the three plants were exposed to appropriate conditions required by short-day plants to flower. As you might expect in normal plants exhibited flowering and the plant which was completely defoliated failed to develop flowers. Interestingly, the third plant in which only one leaf was left also exhibited normal flowering. Observations made from the above experiment confirmed that flowering stimulus is perceived by leaves and also even a single leaf is sufficient to perceive the flowering stimulus which will make the whole plant flower.

### **Flowering Hormone**

There has been ever-existing evidence of presence of a flowering hormone in plants. The presence of flowering hormone and its translocation has been demonstrated through grafting experiment.

**Experiment I:** In a grafting experiment one short day plant was provided with short day treatment and other with long day treatment. Obviously, plant exposed to short day treatment exhibited flowering and plant exposed to long day failed to flower.

SDP which was given long day treatment which did not flower could be made to flower if grafted with a SDP exposed to proper photoperiod. From the result obtained you can clearly predict that the plant exposed to proper photoperiod synthesized a flowering hormone which was transferred to the grafted plant which resulted in flowering response

**Experiment II:** In another experiment several short-day plants were grafted with one another. All the plants were given long day treatment except one leaf of one plant. Flowering was observed in all the plants. The results obtained shows that if single leaf receives proper stimulus, the flowering hormone will be synthesized and was transferred from one plant to another. Hence, even if one leaf of the plant receives correct day length, the plant exhibits flowering regardless of the conditions surrounding rest of plant.

Experiments showed that flowering hormone exists in plants and also stimulus for synthesis of flowering hormone is perceived by leaves which is transmitted to buds for flowering to occur. Chailakhyan has named the flowering hormone as florigen. He gave the name florigen to unknown chemical stimulus which acts as floral inducer.

### Significance of Photoperiodism

Photoperiodism determines the flowering season of a plant.Knowledge of photoperiodism can be utilized in keeping plant in vegetative phase to obtain high yield of tubers rhizomes etc. or the plants may be maintained in reproductive stage to yield more flower and fruits.Annuals can be grown more than once in a year by regulating photoperiod.By increasing light hours winter dormancy and autumnal leaf fall can be prevented.By providing requirement photoperiod plants can be made to flower throughout the year under green house.Knowledge of photoperiodism is also useful in setting up of gardens, orchards etc.The difference between photoperiodism and vernalization.

# Phytochrome

Phytochrome is a pigment found in plants which is known to control development of plants. Phytochrome is a protein with chromatophore. The pigment exists in two interconvertable forms PR and PFR. The type of phytochrome which absorbs red light is called as PR and the type of phytochrome which absorbs far-red light if called PFR. PR is red light sensitive and PFR is far red light sensitive. Plant utilizes phytochrome to sense the seasonal changes in night length or photoperiod.Phytochrome is mainly produced during darkness and firstly exists as PR. When exposed to light of wavelength 660nm it is converted into PFR. PFR can be reconverted to PR if exposed to wavelength of 730nm. Among both the forms of phytochrome, PR is biologically inactive whereas PFR is biologically active. Many of the physiological changes occurring in plants such as pigmentation, hypocotyls - hook opening, unfolding of leaves, photomorphogenesis, photoperiodism and many others are influenced by phytochrome.

# Effect of phytochrome on flowering in SDP

As you have already seen that short-day plants are those plants which show normal flowering when exposed to shorter photoperiod and longer period of darkness. It has already been discussed that longer period of darkness is crucial for flowering to occur in SDP. If the longer period of darkness is interrupted by red light, SDP fail to flower. However, if red light treatment is followed by far red, the inhibitory effect of red right is compensated and plants show normal flowering. If SDP are given alternate treatment of far and far-red light then the treatment given in the last will show its effect[7], [8].

## Effect of phytochrome on flowering in LDP

We have already discussed that long-day plants are those which flower when provided with longer period of light and shorter period of darkness. You have also seen that longer period of darkness has an inhibitory effect on flowering in LDP. If LDP is exposed to longer period of darkness it will not flower. However, this longer period of darkness is interrupted by red light, plants show normal flowering as because the period of continuous darkness is not maintained. Red light breaks the longer period of darkness into two shorter periods and hence period of darkness loses its inhibitory effect and flowering occurs. But if red light is followed by far red, the effect of red light is counter acted by far red and red light does not show its effect. As a result, the longer period of darkness can maintain its inhibitory effect and plant fails to flower. As seen in the case of SDP, if Red and far-red light are given alternately than the last treatment provided to the plant will exert its effect.

Gibberellic acid has been reported to have a positive influence on flowering in long-day plants. There are several long-day plants which under unfavourable conditions provided will gibberellic acid exhibit normal flowering. Many scientists namely Brian, Chailakhyan, Naylor have supported association of gibberellin with flowering hormone. It is believed that CO<sub>2</sub> leads to formation of a precursor which is then converted into a hormone. This gibberellin - like hormone as finally converted into florigen. Flowering inducing effect of gibberellin have been observed only in long-day plants and not in short-day plants. However, application of GA in SDP will cause stem elongation.

In the presence of red light, the precursor is converted into GA like hormone and far red light inhibits the action of Gibberellic acid as in the presence of far red light gibberellin like hormone is converted back to the precursor. Arabidopsis thaliana, Hyoscyamus niger, Lactuca sativa are some of long-day plants which can be made to flower under short day condition by exogenous application of gibberellic acid. Plants belonging to family Cupressaceae, Taxodiaceae and Pinaceae also show flowering induced by GA.

Although, it is clear that GA influenced flowering especially in long-day plants, there are other hormones also which have been found to effect flowering in plants. Auxin and Cytokinin are two type of plant hormones which are known to control growth and development of plants. These hormones have also been found to have effect on flowering. Auxins have been reported to induce flowering in pine apple, Hysocyamus niger, wintex barley. Flower inducing effects of auxin on pine apple was discovered in 1942 and since then auxin have been commercially utilized to induce flowering in pine apple varieties. However, auxin has been found to have different response in different plant species. In some plants auxin helps in inducing flowering whereas in other plant it inhibits flowering or have no effect on flowering. It is expected that the plants in which auxin inhibits flowering is via. Ethylene production as it is well known that application of auxin leads to production of ethylene[9], [10].

Similarly, Cytokinin have been reported to induce flowering in plant varieties such as Lemna paucicostata, Perilla, Wolffia, Chrysanthemum etc. flowering in this plant have been achieved even under non inductive photoperiods by utilization of cytokinin. However, as you have seen in case of auxin that in some plants it induces flowering and, in some plants, it inhibits flowering. Similarly, cytokinin may also inhibit flowering in some plants. One such plant is Chenopodium in which flowering is inhibited in presence of cytokinin.

Abscissic acid cannot induce flowering if the required photoperiod is not provided, however application of abscissic acid under favourable photoperiod enhances reproductive development. Chenopodium and Pharbitis nil are two such plant species in which flowering in enhanced in favourable season by application of abscissic acid.Salicylic acid has been known to act as plant growth regulator. Most of the plants do not have a requirement of salicylic acid for flowering however it has been reported to enhance flowering in plants such as Lemna. Ascorbic acid has also been reported to induce in flowering in plants like Brassica and Lemna.

### **Increased Carbon: Nitrogen Ratio**

Many scientists including Kraus and Bill have proposed that C/N ratio is also significant in determination of flowering in plants. They conducted their study on tomato plants and have proposed following effects of C/N ratio on plants.Plants such as Pharbitis nil and Lemna paucicostata show increased flowering when C/N ratio is high, as you have already seen that short-day plants require shorter photoperiod for flowering, however short-day plant Lemna was induced to flower even in presence of continuous light when cultured onto nitrogen free medium. When plants are supplied with sucrose, ratio of C/N increases which result in increased flowering in plants such as Pharbitis nil, Anagallis arvensis.

### Vernalization

Vernalization can be defined as a process or method of inducing early flowering in plants. It is achieved mainly by treatment of seeds at very low temperature.Generally, you may consider growth and development to be more or less similar; however, both are different processes. Growth generally refers to increase in size and weight whereas development includes processes such as differentiation in flowering, pollination and fertilization which ultimately leads to reproduction. Lysenko in 1920-30 postulated the main principle of vernalization. The basic concept remains that by providing specific treatment either to germinating seed or to the plant one of the two phase of life cycle of plant can be favored. For example, winter wheat is normally grown in winter season but if the seeds of the plant are allowed to germinate in ice box with appropriate suitable light moisture and air, they can be grown in summer as well along with normal flowering.

Flowering is one of the most important processes in life cycle of plant since it is the key event for reproductive succession in plants. Most of the plants flower only when they are exposed to proper period of light. Long-day flowering plants need short period of darkness to flower whereas short-day plants require longer period of darkness to exhibit flowering. However, day neutral plants are independent of photoperiod and flower irrespective of day night length. There is no doubt about the photoperiod remains to the most crucial important factor for following to occur. Beside this, temperature is another factor which also has significant effect on flowering.

If you consider flowering in annuals and biennials, for annuals photoperiod is most crucial for flowering, followed by temperature. However, in the case of biennials as you know that biennials are those plants which show vegetative growth in first season and when they have gone through prolonged exposure to low temperature during winter season, they exhibit flowering in next season. If due to any reason these plants do not get exposure to low temperature they fail to flower and will continue to grow vegetatively. However, they can be made to flower of the plants that are exhibited to cold treatment following suitable photoperiod. Many biennials such as carrot, cabbage, beet, glove needs cold treatment for flowering to occur. If young plants or moistened seeds of biennial plants are provided with chilling treatment, they can be made to flower in single growing season. Winter annuals respond to low temperature early in their life and can be vernalized before germination. However, most biennials grow during first season and flower in summer. Hence, they must reach a minimum size before they become responsive towards vernalization treatment

Examples:

- In case of winter rye, if the seeds are treated with the temperature of 1 C for about 4 weeks the plants exhibit flowering in after 11 weeks of plantation. However, if the seeds are treated and germinated at temperature about 18 C no flowering can be obtained after same period of growth after which vernalized plants flowered.
- Hyoscyamus niger is a biennial plant and requires exposure to cold treatment for flowering, besides that there is another interesting fact about flowering in Henbane. Plants of Henbane will show flowering only if when vernalization is followed by a long day treatment. However, if vernalized plants are exposed to short day treatments, plant fail to flower. There is another variety of Henbane which is annual and does not require cold treatment for flowering.

### **Process of Vernalization**

Vernalization can be achieved by a very simple process. Seeds to be vernalized should be soaked in water properly, vernalization can never be achieved in dry seeds, it has been reported that seed should contain about 90% water of their dry weight. Seeds then allowed to germinate, followed by treatment of low temperature for suitable period of time. Treated seedlings are slightly dried and then sown for further growth.

### **Requirement of vernalization**

- 1. Low temperature: Normally vernalization is achieved in temperature range of zero degree to  $10\Box C$ , when the temperature is decreased below  $0\Box C$  the effectiveness of vernalization generally decreases and at about  $-6\Box C$  there is no effect and vernalization cannot be achieved.
- 2. Duration of treatment: The time period required for vernalization may vary from species to species and can range from few days to few weeks. Normally the time period of treatment is long since vernalization is considered to be a slow process.
- 3. Actively dividing cells: Since vernalization cannot occur in dry seeds due to absence of active embryo, after germination embryo becomes active and can perceive vernalization. In whole plant vernalization signal is perceived by meristematic cells shoot apical meristem of Chrysanthemum have been demonstrated to perceive vernalization.
- 4. Water: Water is an essential requirement for germination and that seeds provided with vernalization treatment also requires water for germination and growth.

### **Devernalization and Revernalization**

Devernalization can be defined as a process in which vernalized seeds / seedlings loses their vernalized states and becomes devernalized. Over drying of vernalized seed / seedling, heat treatment of vernalized seed/ seedling may result in devernalization. An atmosphere of nitrogen in presence of high concentration of  $CO_2$  may also result in devernalization. However, devernalized seeds can be easily revernalized by subjecting them to low temperature. Vernalization response critically depends upon the temperature to which seeds are exposed and also on the duration for which the treatment is maintained. If either of them falls short of the requirements then either vernalization may not occur properly or the vernalized seed may be easily devernalized.

### Advantages of Vernalization

Vernalization shortens the Juvenile or vegetative phase and induces early flowering.Vernalization can be applied to temperate as well as tropical plants.It can be used in Horticulture. Flowering can be induced into non vernalization plant by grafting a vernalized shoot open.When proper cell treatment is provided a stimulus is perceived by the dividing cell. Researchers have named the stimulus as vernalin. Formation of vernalin alone is not enough for flowering to occur. After vernalization plants appropriate photoperiod is also required. Following proper photoperiod either vernalin is converted into florigen or vernalin regulates florigen synthesis. Florigen once produced directs reproductive development and flowering is initiated in the plants.

When plant has completed certain vegetative growth, it makes a transition from juvenile stage of maturity.Most of the plants require favourable environmental to occur. Photoperiod and temperature are the most crucial environmental factors for flowering.The response of plants to day length is known as photoperiodism.Depending upon requirement of photoperiod plants can be short -day plants, long-day plants and day neutral plants.Short - day plants flower when a critical period of darkness is exceeded. Long-day plants exhibit flowering when period of darkness is less than a critical period. When exposed to suitable photoperiod, the photoperiodic stimulus is perceived by leaves and a flowering hormone is synthesized[11], [12].

Flowering hormone is translocated to shoot apex where bud formation and flowering takes place. The presence of a hormone which can be transmitted from leaves to short apex have been demonstrated by grafting experiments.Phytochrome is a protein with chromatophore found in plants in two interconvertable forms: PR and PFR.PR is red light sensitive and PFR is far red light sensitive. Among the two forms of phytochrome PFR is biologically active form.Some plants require exposure to low temperature for flowering to occur.The effect of temperature on flowering is more profound in biennials as compared to annuals.Temperature in range of 0-5°C is considered to be most effective to achieve vernalization.Effect of vernalization can be reversed by exposing vernalized seed or plant to high temperature.

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# **CHAPTER 11**

# **REGENERATION AND REPAIR**

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Regeneration and repair are essential processes in biology that allow organisms to recover from tissue damage and restore normal function. These processes involve a complex interplay between various cellular and molecular mechanisms that act to repair damaged tissues, replace lost cells, and restore normal morphology and function. In this essay, we will explore the mechanisms of regeneration and repair in the context of elementary morphogenesis, focusing on the cellular and molecular mechanisms that underlie these processes.

#### **Regeneration and Repair**

Regeneration and repair are often used interchangeably to describe the process by which damaged tissues are replaced or repaired. However, these terms have distinct meanings and implications. Regeneration refers to the process by which lost or damaged tissues are replaced with new tissues that are similar in structure and function to the original tissues. Regeneration can occur through the proliferation and differentiation of stem cells or through the dedifferentiation and reprogramming of existing cells.Repair, on the other hand, refers to the process by which damaged tissues are repaired or replaced with scar tissue, which is composed of fibroblasts and collagen. Repair is typically a less efficient process than regeneration, as it often results in the loss of tissue function and can lead to long-term complications.

#### **Regeneration and Repair in Elementary Morphogenesis**

Regeneration and repair are essential processes in elementary morphogenesis, as they allow organisms to recover from tissue damage and maintain normal function. The cellular and molecular mechanisms underlying regeneration and repair are complex and involve a wide range of cellular and molecular processes.

### **Cellular Mechanisms of Regeneration and Repair**

The cellular mechanisms of regeneration and repair involve the proliferation and differentiation of stem cells, the migration and activation of immune cells, and the activation of signaling pathways that regulate cell growth, differentiation, and survival. Stem cells play a critical role in regeneration and repair, as they have the capacity to differentiate into a wide range of cell types and can be mobilized to repair damaged tissues.Immune cells also play an important role in regeneration and repair, as they are involved in the clearance of damaged tissues and the recruitment of stem cells and other repair cells to the site of injury. Immune cells are also involved in the production of cytokines and growth factors that promote tissue repair and regeneration[1]–[3].

### **Molecular Mechanisms of Regeneration and Repair**

The molecular mechanisms of regeneration and repair involve the activation of signaling pathways that regulate cell growth, differentiation, and survival. These pathways are activated in response to tissue damage and act to promote the proliferation and differentiation of stem cells, the migration and activation of immune cells, and the production of cytokines and growth factors that promote tissue repair and regeneration.

One of the key signaling pathways involved in regeneration and repair is the Wnt signaling pathway, which is involved in the regulation of stem cell proliferation and differentiation. The Wnt signaling pathway is activated in response to tissue damage and promotes the proliferation and differentiation of stem cells, leading to the regeneration of damaged tissues.

Another key signaling pathway involved in regeneration and repair is the TGF- $\beta$  signaling pathway, which is involved in the regulation of immune cell activation and the production of cytokines and growth factors that promote tissue repair and regeneration. The TGF- $\beta$  signaling pathway is activated in response to tissue damage and acts to promote the recruitment and activation of immune cells and the production of cytokines and growth factors that promote tissue repair and regeneration.

### **Regeneration and Repair in Different Tissues and Organs**

Regeneration and repair occur differently in different tissues and organs, as each tissue and organ has its own unique cellular and molecular mechanisms. Some tissues and organs, such as the liver and bone marrow, have a high capacity for regeneration and can regenerate quickly in response to injury. Other tissues and organs, such as the heart and nervous system, have a limited capacity for regeneration.

#### **Mechanisms of Regeneration**

Regeneration can occur through different mechanisms, depending on the organism and the tissue type involved. In some cases, regeneration involves the activation of stem cells that are normally present in the tissue. These stem cells can differentiate into various cell types and replace the damaged or lost tissue. In other cases, regeneration occurs through the dedifferentiation of mature cells, which then proliferate and differentiate into the necessary cell types. This process is often accompanied by changes in the extracellular matrix, which can provide the necessary signals for cell proliferation and differentiation. Regeneration is the ability of a plant to replace damaged or lost cells, tissues, or organs. Unlike animals, plants possess the remarkable ability to regenerate whole organs, such as roots, stems, leaves, and even whole plants from small fragments or individual cells. Regeneration in plants involves complex physiological and genetic mechanisms that are still not fully understood. However, recent advances in molecular biology and genetics have provided some insights into the mechanisms of plant regeneration.

The process of regeneration in plants can be classified into two main types: vegetative regeneration and reproductive regeneration. Vegetative regeneration is the ability of a plant to regenerate new organs, such as roots and stems, from its vegetative tissues, such as leaves and stems. Reproductive regeneration, on the other hand, is the ability of a plant to regenerate its reproductive structures, such as flowers, fruits, and seeds. Both types of regeneration involve a series of cellular and molecular events that are regulated by various signaling

pathways and genetic factors. One of the main mechanisms of regeneration in plants is the activation of meristematic cells. Meristematic cells are undifferentiated cells that have the potential to divide and differentiate into any type of specialized cells. In plants, meristematic cells are located in the apical meristem, which is the growing tip of the plant. When a plant is injured or damaged, the apical meristem is activated, and the meristematic cells start dividing and differentiating to form new tissues and organs.

Another mechanism of regeneration in plants is the activation of cell dedifferentiation. Dedifferentiation is the process by which specialized cells lose their identity and revert to an undifferentiated state. In plants, dedifferentiation can be induced by various stimuli, such as hormonal signals, stress, or injury. When dedifferentiation is induced, the cells lose their specialized functions and become meristematic, allowing them to regenerate new tissues and organs[4], [5].

Plant regeneration also involves the activation of various signaling pathways and genetic factors. One of the keys signaling pathways involved in regeneration is the WNT signaling pathway. The WNT signaling pathway is a conserved signaling pathway that is involved in many developmental processes, including embryonic development, tissue homeostasis, and regeneration. In plants, the WNT signaling pathway is involved in the regulation of meristem activity and the activation of cell dedifferentiation.

Another key factor involved in plant regeneration is the activation of transcription factors. Transcription factors are proteins that regulate gene expression by binding to specific DNA sequences. In plants, various transcription factors have been identified that are involved in the regulation of meristem activity and cell dedifferentiation. For example, the transcription factor WUSCHEL (WUS) is involved in the maintenance of the apical meristem, while the transcription factor LEC1 is involved in the activation of cell dedifferentiation.

### **Factors that Affect Regeneration**

The ability of an organism to regenerate is influenced by various factors, including age, environment, and genetics. Generally, younger organisms have a greater capacity for regeneration than older organisms, as their cells are more plastic and have a greater ability to proliferate and differentiate.

Environmental factors, such as temperature and nutrition, can also affect regeneration by influencing the metabolism and growth of cells. Genetic factors can also play a role in regeneration, as some organisms are naturally better equipped for regenerative processes than others.

#### **Comparison of Regeneration and Repair**

While regeneration and repair are both mechanisms for restoring damaged tissue, there are some key differences between the two processes. Regeneration typically involves the replacement of damaged tissue with new, functional tissue that is similar to the original tissue in terms of structure and function. Repair, on the other hand, involves the formation of scar tissue to fill in the damaged area. This scar tissue is often structurally and functionally different from the original tissue, and can lead to long-term complications, such as reduced flexibility or impaired organ function.

#### **Potential Applications of Regeneration**

The ability to regenerate tissue has important implications for medical research and treatment. For example, researchers are exploring the use of stem cells and other regenerative techniques to repair damaged tissues and organs, such as the heart, liver, and spinal cord. In some cases, regeneration may even be used to replace entire organs, such as the liver or pancreas. Additionally, researchers are investigating the potential of regeneration to treat age-related diseases, such as Parkinson's and Alzheimer's, by restoring damaged neural tissue.

## **Challenges and Future Directions**

While regeneration holds great promise for medical research and treatment, there are also significant challenges that must be addressed. For example, researchers must develop techniques for controlling and directing the differentiation of stem cells, in order to ensure that they differentiate into the appropriate cell types. Additionally, the extracellular matrix plays a crucial role in regeneration, and researchers must develop methods for controlling and modifying this matrix to facilitate tissue repair. Finally, ethical considerations must be taken into account, particularly when working with embryonic stem cells or other sensitive tissues.

Another important aspect of regeneration and repair in reference to elementary morphogenesis is the role of signaling pathways. These pathways involve a series of molecular signals that regulate cellular processes and can play a crucial role in the regeneration and repair of tissues. One such signaling pathway is the WNT signaling pathway, which has been shown to play a critical role in the regeneration of several tissues, including bone, muscle, and skin. The WNT pathway is activated by the binding of WNT ligands to Frizzled receptors, leading to the activation of a cascade of intracellular signaling events that ultimately regulate gene expression and cellular behavior[6]–[8].

Similarly, the Notch signaling pathway has been implicated in the regeneration of tissues such as the liver and pancreas. This pathway is activated by the binding of Notch receptors to Delta or Jagged ligands, leading to the cleavage of the Notch receptor and the release of its intracellular domain. The intracellular domain then translocates to the nucleus, where it regulates gene expression and promotes cellular proliferation and differentiation. In addition to these signaling pathways, the extracellular matrix (ECM) also plays a crucial role in the regeneration and repair of tissues. The ECM is a complex network of proteins and carbohydrates that surrounds and supports cells within tissues. It provides mechanical support, regulates cell behavior, and can influence cell fate and differentiation.

During tissue repair and regeneration, the ECM undergoes dynamic changes, including the deposition of new ECM components and the remodeling of existing ones. These changes can affect cellular behavior and contribute to the formation of new tissue.Finally, the immune system also plays an essential role in tissue repair and regeneration. Immune cells, such as macrophages and neutrophils, are recruited to the site of injury or damage, where they phagocytose cellular debris and release cytokines and growth factors that promote tissue repair and regeneration.In conclusion, the process of regeneration and repair is a complex and dynamic process that involves a variety of cellular and molecular mechanisms. These mechanisms include stem cell activation, cellular proliferation and differentiation, signaling pathways, the extracellular matrix, and the immune system. Understanding the interplay

between these mechanisms is crucial for developing effective strategies for tissue repair and regeneration in the context of elementary morphogenesis.

Regeneration and repair are important mechanisms for maintaining tissue homeostasis and responding to injury. These processes rely on a complex interplay between cells, extracellular matrix, and signaling molecules, and are influenced by a variety of factors, including age, environment, and genetics. While regeneration holds great promise for medical research and treatment, there are also significant challenges that must be addressed. By understanding the mechanisms of regeneration and repair, and by developing new techniques and technologies for facilitating these processes, researchers can work towards improving human health and well-being[9]–[11].

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# CHAPTER 12

### MORPHOGENETIC MOVEMENTS AND MIGRATION

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Morphogenetic movements and migration play crucial roles in the process of embryonic development, which is a key component of elementary morphogenesis. These processes are responsible for shaping the embryo into its final form and ensuring that all organs and tissues are in their correct locations. Morphogenetic movements refer to the physical changes that occur during embryonic development, including cell division, cell migration, and differentiation. These movements are tightly regulated by signaling pathways and interactions between cells, which guide cells to their correct locations and ensure that the developing embryo has the correct shape and structure.

Cell migration is an essential morphogenetic movement that occurs during embryonic development. This process involves the movement of cells from one location to another, which is important for the proper organization and development of tissues and organs. There are several different types of cell migration, including radial migration, tangential migration, and directed migration.Radial migration is the process by which cells move from the inner layer of the embryo to the outer layer, or vice versa. This process is crucial for the development of the central nervous system, as it ensures that all neurons are in their correct locations and able to form the proper connections. During radial migration, cells move along radial glial cells, which are specialized cells that provide a scaffold for migrating neurons. The radial glial cells extend from the ventricular zone, which is the site of neurogenesis, to the outer layers of the developing brain. As neurons are generated in the ventricular zone, they migrate along the radial glial cells to their final destination in the outer layers of the brain. Radial migration is a complex process that is regulated by a variety of molecular signals. One of the key regulators of radial migration is a protein called Reelin, which is secreted by the radial glial cells. Reelin binds to receptors on migrating neurons and helps to guide them to their final destination.

Mutations in genes that regulate radial migration can lead to a variety of developmental disorders, including lissencephaly and double cortex syndrome. In lissencephaly, neurons fail to migrate properly, resulting in a smooth brain surface and severe neurological defects. In double cortex syndrome, neurons migrate too far, resulting in the formation of two distinct layers of cortex. Understanding the mechanisms that regulate radial migration is important for understanding the development of the nervous system and for developing new therapies[1]–[3].

Tangential migration, on the other hand, involves the movement of cells within a layer of tissue. This process is important for the proper organization of different cell types within a tissue and ensures that all cells are in their correct locations. Tangential migration, also known as lateral migration, is a biological process in which cells move parallel to a surface or plane. This type of migration is commonly observed during the development of the nervous system, particularly during the formation of the cerebral cortex. In the developing brain, neural

progenitor cells divide and differentiate into neurons and glial cells. These newly generated cells must migrate to their appropriate locations to form the complex network of the brain. While some cells migrate radially, others migrate tangentially to reach their final destination. During tangential migration, cells move along a path that is perpendicular to their axis of elongation. This process is guided by a variety of signaling cues, including chemokines, growth factors, and adhesion molecules. These cues are recognized by receptors on the cell surface, which initiate a cascade of intracellular signaling events that ultimately drive cell movement.

In the cerebral cortex, tangential migration is particularly important for the development of interneurons, a type of inhibitory neuron that plays a critical role in regulating the activity of excitatory neurons. Interneurons originate from the ganglionic eminences, which are regions of the embryonic brain that give rise to the basal ganglia. These cells then migrate tangentially along the ventral telencephalon, a region adjacent to the developing cortex. As interneurons migrate tangentially, they encounter a number of obstacles, including other migrating cells and physical boundaries such as blood vessels. To navigate these obstacles, interneurons use a variety of strategies, including changing their direction of movement and moving around physical barriers.

The precise mechanisms that control tangential migration are still being studied, but recent research has identified several key signaling pathways that are involved. For example, the Reelin signaling pathway has been shown to play an important role in guiding tangential migration of interneurons. Mutations in genes associated with this pathway have been linked to a number of neurological disorders, including autism and schizophrenia. Overall, tangential migration is a complex and dynamic process that plays a critical role in the development of the nervous system. By understanding the mechanisms that control this process, researchers hope to gain insight into a variety of neurological disorders and develop new therapies for these conditions.

Directed migration, also known as chemotaxis, is a process by which cells are guided to a specific location by chemical signals. This process is important for the proper organization of cells during embryonic development and is also involved in tissue repair and regeneration in adults. Another key morphogenetic movement is cell division, which is responsible for the growth and expansion of tissues and organs during embryonic development. This process is tightly regulated by signaling pathways and interactions between cells, which ensure that the correct number of cells is produced and that tissues and organs are of the correct size and shape[4].

In addition to morphogenetic movements, migration, and cell division, other processes also play important roles in elementary morphogenesis. These processes include cell differentiation, apoptosis, and the formation of extracellular matrix.Cell differentiation refers to the process by which cells become specialized and take on specific functions within a tissue or organ. This process is essential for the proper functioning of tissues and organs and ensures that each cell type is able to carry out its specific role.Apoptosis, or programmed cell death, is another process that plays an important role in elementary morphogenesis. This process is responsible for the elimination of excess or damaged cells during embryonic development and ensures that tissues and organs are properly formed and functioning. Finally, the formation of extracellular matrix, which provides support and structure to tissues and organs, is also a key component of elementary morphogenesis. This process involves the production and organization of proteins such as collagen and elastin, which form the structural framework of tissues and organs. In conclusion, morphogenetic movements, migration, and cell division are all essential processes that play crucial roles in elementary morphogenesis. These processes are tightly regulated by signaling pathways and interactions between cells, which ensure that tissues and organs are properly formed and functioning. Understanding these processes is critical for the development of new therapies for tissue repair and regeneration, as well as for the treatment of developmental disorders and birth defects. Another morphogenetic movement involved in elementary morphogenesis is invagination. Invagination is the inward folding or buckling of a cell sheet that creates a pocket or pouch. This process is critical in the formation of many organs, including the lungs, digestive tract, and kidneys. During invagination, a group of cells begins to fold inward, creating a depression or pocket. This pocket then elongates and deepens, forming a tube or pouch that will eventually become an organ[5].

Conversely, evagination is the outward extension or protrusion of a cell sheet. This morphogenetic movement is essential in the development of organs such as the lungs, eyes, and mammary glands. Evagination begins when a group of cells at the surface of the embryo begins to extend outward, forming a bulge or protrusion. This bulge or protrusion then elongates and expands, creating a tube or sac that will develop into the organ. Another critical aspect of elementary morphogenesis is cell migration. Cell migration is the movement of cells from one location to another within an organism. During development, cells migrate to form tissues and organs and establish their proper location and orientation. Cell migration also plays a crucial role in wound healing and tissue repair.

Cell migration is regulated by a complex network of molecular signals, including chemokines and growth factors. These signaling molecules activate intracellular signaling pathways that control the movement of cells by regulating cytoskeletal dynamics, cell adhesion, and the formation of cellular protrusions such as filopodia and lamellipodia.Elementary morphogenesis is a fundamental process in embryonic development that involves a series of morphogenetic movements and cell behaviors. These movements, including gastrulation, neurulation, invagination, and evagination, play a crucial role in the formation of organs and tissues. Cell migration is also a critical aspect of elementary morphogenesis, allowing cells to establish their proper location and orientation during development and repair damaged tissues. Understanding the mechanisms of elementary morphogenesis is crucial for advancing our understanding of embryonic development and for developing new strategies for tissue repair and regeneration[6], [7].

Morphogenetic movements and migration play crucial roles in embryonic development, tissue repair, and organogenesis. These processes involve the coordinated movement and rearrangement of cells and tissues, which ultimately determine the final shape and structure of developing organisms. In this article, we will provide an overview of the key concepts and mechanisms of morphogenetic movements and migration, with a focus on their roles in elementary morphogenesis. Morphogenetic movements refer to the changes in cell shape, position, and organization that occur during embryonic development. These movements are essential for the formation of complex structures and organs, such as the brain, heart, and limbs. The cellular rearrangements that occur during morphogenetic movements are driven

by a combination of biochemical and mechanical signals, which coordinate the behavior of cells and tissues. One of the well-studied examples of morphogenetic movements is gastrulation, which occurs during early embryonic development. Gastrulation involves the invagination of cells at the blastopore, which creates the three primary germ layers: the endoderm, mesoderm, and ectoderm. The movements of cells during gastrulation are driven by a combination of signaling pathways, including WNT, BMP, and FGF, which regulate cell adhesion, polarization, and migration.

Another example of morphogenetic movements is neurulation, which is the process by which the neural tube is formed. The neural tube is the precursor to the brain and spinal cord, and its formation is critical for the development of the nervous system. Neurulation involves the folding of the neural plate to form the neural groove, which eventually closes to form the neural tube. The movements of cells during neurulation are also regulated by signaling pathways, including WNT, BMP, and Shh, which control the patterning and differentiation of neural progenitor cells.

Migration, on the other hand, refers to the movement of cells from one location to another. Cell migration is essential for many biological processes, including wound healing, immune response, and tissue repair. In embryonic development, cell migration is critical for the formation of many organs and structures, including the heart, blood vessels, and limbs.

One of the key mechanisms of cell migration is the formation of focal adhesions, which are specialized structures that anchor cells to the extracellular matrix (ECM). Focal adhesions are composed of integrins, which are transmembrane proteins that link the ECM to the actin cytoskeleton. By forming and disassembling focal adhesions, cells can generate the forces necessary to move through the ECM.

Cell migration is also regulated by chemotaxis, which is the directed movement of cells towards a chemical gradient. Chemotaxis is critical for many biological processes, including the migration of immune cells to sites of infection and the migration of cancer cells during metastasis. In embryonic development, chemotaxis is involved in the migration of cells to specific locations, such as the dorsal root ganglia, which are clusters of sensory neurons that innervate the skin and other organs.

In addition to morphogenetic movements and migration, cellular differentiation is another critical process in embryonic development. Cellular differentiation refers to the process by which stem cells give rise to specialized cell types, such as neurons, muscle cells, and blood cells. The differentiation of stem cells is regulated by a combination of intrinsic and extrinsic signals, including transcription factors, growth factors, and ECM components.

The differentiation of stem cells is critical for the formation of many organs and tissues, including the brain, heart, and blood vessels. For example, the differentiation of neural stem cells is essential for the formation of the different regions of the brain, which have distinct functions and characteristics. Another important morphogenetic movement is cell elongation, which occurs when cells increase in length. This process is important for tissue elongation and the shaping of organs. For example, in plants, cell elongation is responsible for the growth of stems and roots, while in animals, it plays a role in the elongation of limbs and the development of the nervous system.

Cell migration is another key morphogenetic movement that involves the movement of cells from one location to another. This process is critical during embryonic development when cells must move to their final destinations to form tissues and organs. Cell migration is also important during wound healing and tissue repair, where cells move to damaged areas to promote healing and regeneration.

There are two main types of cell migration: collective and individual. Collective cell migration involves groups of cells moving together as a cohesive unit, while individual cell migration involves single cells moving independently. Both types of cell migration are essential for proper tissue formation and repair.

During collective cell migration, cells interact with each other through various mechanisms such as cell-cell adhesion and cell-matrix interactions. These interactions allow cells to coordinate their movements and move as a group. Collective cell migration is important for processes such as epithelial sheet migration, which occurs during wound healing and tissue repair.In contrast, individual cell migration involves a single cell moving independently of other cells. This type of migration is important for processes such as axon guidance during the development of the nervous system. During axon guidance, individual neurons extend their axons to connect with other neurons and form neural circuits. Individual cell migration is also important for immune cell migration, where immune cells move independently to reach sites of infection or inflammation.

In conclusion, morphogenetic movements and cell migration are critical processes during embryonic development, tissue formation, and repair. These movements involve complex cellular interactions and signaling pathways that regulate cell behavior and shape tissue and organ structures.

Understanding the mechanisms underlying these processes is essential for developing new therapies for tissue repair and regeneration. In conclusion, morphogenetic movements and cell migration are critical processes in embryonic development, tissue repair, and disease progression. These movements involve coordinated changes in cell shape, cell adhesion, and cytoskeletal organization that allow cells to move and rearrange in response to extracellular signals and mechanical forces[8], [9].

Morphogenetic movements, such as convergent extension and epithelial-to-mesenchymal transition, play important roles in shaping tissues and organs during embryonic development. They involve complex interactions between signaling pathways, transcription factors, and cytoskeletal regulators that are still being unraveled. Cell migration is also essential for tissue repair, wound healing, and immune responses. It involves multiple steps, including protrusion of the leading edge, adhesion to extracellular matrix or other cells, contraction of the cell body, and detachment at the rear end. Dysregulation of cell migration is associated with various diseases, including cancer metastasis, developmental disorders, and chronic inflammation.

Recent advances in imaging techniques, genetics, and bioengineering have provided new insights into the mechanisms of morphogenetic movements and cell migration. However, many questions still remain unanswered, such as how cells integrate multiple signals and how mechanical forces are transmitted across tissues.Further research on morphogenetic movements and cell migration is needed to understand their roles in embryonic development,

tissue repair, and disease progression. This knowledge could lead to the development of new therapies for various diseases and improve our understanding of the fundamental processes that shape life[10]–[12].

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## **CHAPTER 13**

## GENETIC CONTROL OF MORPHOGENESIS

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Morphogenesis is the biological process of shaping the physical form of organisms during development. This process involves a series of complex cellular events, including cell division, differentiation, and movement. The genetic control of morphogenesis is essential to ensure that organisms develop correctly, and any errors in this process can lead to developmental disorders and disease. The genetic control of morphogenesis involves the regulation of gene expression, which determines the activity of proteins that control cell behavior. These proteins interact with the cytoskeleton, a network of filaments that provide structural support and shape to cells. The cytoskeleton also plays a critical role in morphogenesis by facilitating cell migration and division.

#### The Role of Signaling Pathways in Morphogenesis

Morphogenesis is the process by which an organism develops its shape and form. It involves the coordinated movement and differentiation of cells, as well as changes in cell shape and behavior. Signaling pathways play a crucial role in regulating these processes, allowing cells to communicate with each other and respond to their environment. In this article, we will explore the role of signaling pathways in morphogenesis and how they contribute to the development of complex organisms.Signaling pathways are networks of proteins and other molecules that transmit information between cells. They allow cells to respond to their environment and communicate with each other, coordinating their behavior and regulating their development. In morphogenesis, signaling pathways are involved in a variety of processes, including cell migration, cell proliferation, and cell differentiation.

One important signaling pathway in morphogenesis is the WNT pathway. This pathway is involved in a wide range of developmental processes, including the formation of the nervous system, the development of limbs and organs, and the maintenance of stem cells. In the WNT pathway, signaling molecules called WNT proteins bind to receptors on the surface of target cells, activating a series of intracellular signals that regulate gene expression and cell behavior.

Another important signaling pathway in morphogenesis is the Hedgehog pathway. This pathway is involved in a variety of processes, including the development of the nervous system, the formation of limbs and organs, and the maintenance of stem cells. In the Hedgehog pathway, signaling molecules called Hedgehog proteins bind to receptors on the surface of target cells, activating a series of intracellular signals that regulate gene expression and cell behavior[1]–[3].

The Notch pathway is another important signaling pathway in morphogenesis. This pathway is involved in a variety of processes, including the development of the nervous system, the formation of organs, and the maintenance of stem cells. In the Notch pathway, signaling molecules called Notch proteins bind to receptors on the surface of target cells, activating a series of intracellular signals that regulate gene expression and cell behavior.

These signaling pathways are not independent of each other and often interact to regulate morphogenesis. For example, the Wnt and Hedgehog pathways can work together to regulate the formation of the neural tube, a structure that gives rise to the brain and spinal cord. In addition, the Notch pathway can interact with the WNT pathway to regulate the proliferation and differentiation of stem cells.

Dysregulation of signaling pathways can lead to developmental defects and disease. For example, mutations in the WNT pathway have been linked to several forms of cancer, while mutations in the Hedgehog pathway have been linked to a variety of developmental disorders, including holoprosencephaly and polydactyly. Similarly, mutations in the Notch pathway have been linked to a variety of developmental disorders, including Alagille syndrome and Adams-Oliver syndrome.Signaling pathways are critical in controlling morphogenesis by transmitting signals from outside the cell to the nucleus. These signals trigger a cascade of events that lead to changes in gene expression and ultimately regulate cellular behavior.

One important signaling pathway in morphogenesis is the WNT pathway. WNT proteins bind to cell surface receptors and activate a series of intracellular signaling events that result in the activation of transcription factors, which bind to DNA and control gene expression. The WNT pathway is involved in many processes, including embryonic development, tissue regeneration, and stem cell maintenance. Another important signaling pathway is the Hedgehog pathway. This pathway is involved in the development of many organs, including the brain, limbs, and skin. The Hedgehog pathway is activated by binding to the receptor Patched, which results in the activation of a transcription factor that controls gene expression[4].

## **Genetic Control of Cell Division and Proliferation**

The genetic control of cell division is essential for proper morphogenesis. Cell division is regulated by a family of proteins called cyclins and cyclin-dependent kinases (CDKs). Cyclins and CDKs work together to control the progression of the cell cycle, which involves DNA replication and chromosome segregation.

The activity of cyclins and CDKs is tightly regulated by other proteins that act as inhibitors or activators. For example, the protein p53 is a critical regulator of the cell cycle and is often referred to as the "guardian of the genome." p53 responds to DNA damage by activating genes that stop the cell cycle and allow for DNA repair.Cell division and proliferation are critical processes in the development of multicellular organisms. The control of these processes is regulated by a variety of genetic mechanisms that ensure the proper timing and location of cell division, as well as the maintenance of genetic stability. The cell cycle is a highly regulated process that controls the replication and division of a cell. It is composed of several phases, including G1, S, G2, and M. During the G1 phase, the cell undergoes a period of growth and preparation for mitosis during the G2 phase. Finally, during the M phase, the cell undergoes mitosis, resulting in the formation of two genetically identical daughter cells.

The control of cell division and proliferation is essential for proper development and the maintenance of tissue homeostasis. Aberrant cell division and proliferation can lead to a

variety of developmental disorders, including cancer. One of the key genetic mechanisms that regulate cell division and proliferation is the activity of cyclin-dependent kinases (CDKs). CDKs are a family of protein kinases that are activated by the binding of cyclins. Cyclins are a family of proteins that are produced and degraded in a cyclic manner during the cell cycle. The binding of cyclins to CDKs activates the kinase activity of the enzyme, which then phosphorylates a variety of proteins involved in cell cycle progression.

Another critical genetic mechanism that regulates cell division and proliferation is the activity of tumor suppressor genes. These genes encode proteins that inhibit cell cycle progression and can induce apoptosis in cells that have undergone DNA damage or other abnormalities. Loss or mutation of these genes can lead to the uncontrolled proliferation of cells and the development of cancer.

In addition to genetic mechanisms, a variety of extracellular signals can also regulate cell division and proliferation. These signals are often mediated by signaling pathways, which involve the activation of receptors on the cell surface and the subsequent activation of intracellular signaling cascades. One example of a signaling pathway that regulates cell division and proliferation is the Wnt pathway. The Wnt pathway is activated by the binding of Wnt ligands to receptors on the cell surface. This activation leads to the stabilization and activation of  $\beta$ -catenin, which then translocates to the nucleus and activates the expression of a variety of genes involved in cell cycle progression[5]–[7].

Another signaling pathway that regulates cell division and proliferation is the Notch pathway. The Notch pathway is activated by the binding of Notch ligands to receptors on adjacent cells. This activation leads to the cleavage and release of the intracellular domain of the receptor, which then translocates to the nucleus and activates the expression of a variety of genes involved in cell cycle progression.

## **Genetic Control of Cell Differentiation**

Cell differentiation is the process by which cells become specialized to perform specific functions. The genetic control of cell differentiation involves the regulation of gene expression, which determines the activity of proteins that control cell behavior. One important family of proteins involved in cell differentiation is the homeobox (HOX) genes. These genes are involved in determining the body plan of organisms by controlling the development of different body segments. Mutations in HOX genes can lead to developmental disorders, such as limb abnormalities. Another important family of genes involved in cell differentiation is the Notch signaling pathway. This pathway is involved in many processes, including cell fate determination, tissue regeneration, and cancer. Notch signaling is mediated by the interaction between the Notch receptor and its ligands, which results in the activation of transcription factors that control gene expression.

#### **Genetic Control of Cell Migration**

Cell migration is a critical process in morphogenesis, as it allows cells to move to their correct positions during development. The genetic control of cell migration involves the regulation of gene expression, which determines the activity of proteins that control cell behavior. One important family of proteins involved in cell migration is the Rho family of GTPases. These proteins regulate the organization of the cytoskeleton, which is essential for

cell migration. Rho GTPases also control the formation of cell adhesions and the activity of enzymes that break down extracellular matrix, allowing cells to move through tissues.

Cell migration is an essential process in many physiological and pathological events, such as embryonic development, wound healing, and cancer metastasis. Cell migration is tightly regulated by various genetic and molecular mechanisms, including signaling pathways and cytoskeleton dynamics. In this article, we will discuss the genetic control of cell migration, including the genes and molecular pathways involved in regulating cell migration and the roles of these genes in normal and pathological conditions.

## Genes and Molecular Pathways Involved in Cell Migration:

Cell migration involves complex interactions between cells and their microenvironment, including extracellular matrix (ECM) molecules, growth factors, and other signaling molecules. These interactions lead to the activation of signaling pathways that regulate the cytoskeleton dynamics and adhesion molecules, ultimately leading to cell movement. Several key genes and signaling pathways have been identified as crucial regulators of cell migration.

## **Rho Family of GTPases:**

Rho family of GTPases, including RhoA, Rac1, and Cdc42, are important regulators of actin cytoskeleton dynamics and cell migration. RhoA promotes the formation of actin stress fibers and focal adhesions, which are essential for cell adhesion and migration. Rac1 and Cdc42, on the other hand, promote the formation of lamellipodia and filopodia, respectively, which are essential for cell protrusion and migration. Dysregulation of Rho family of GTPases has been implicated in many pathological conditions, including cancer metastasis.

## **Integrins:**

Integrins are transmembrane receptors that mediate cell adhesion to ECM molecules, such as fibronectin and collagen. Integrins play a critical role in regulating cell migration by controlling the formation and turnover of focal adhesions, which are essential for cell attachment and migration. Dysregulation of integrin signaling has been implicated in many pathological conditions, including cancer metastasis.

#### **Growth Factors and Receptor Tyrosine Kinases:**

Growth factors, such as epidermal growth factor (EGF) and platelet-derived growth factor (PDGF), promote cell migration by activating receptor tyrosine kinases (RTKs), such as EGFR and PDGFR. RTK activation leads to the activation of downstream signaling pathways, such as the PI3K/Akt and MAPK/ERK pathways, which promote cytoskeleton dynamics and cell migration. Dysregulation of RTK signaling has been implicated in many pathological conditions, including cancer metastasis.

## Wnt Signaling Pathway:

The Wnt signaling pathway plays a critical role in regulating cell migration during embryonic development and in many pathological conditions, including cancer metastasis. Wnt ligands bind to frizzled receptors, leading to the activation of downstream signaling pathways, such as the  $\beta$ -catenin pathway. B-catenin activation leads to the activation of downstream target genes, such as cyclin D1 and c-Myc, which promote cell migration and proliferation.

The Wnt signaling pathway is a highly conserved signaling pathway that plays a critical role in various developmental processes, including cell fate determination, tissue patterning, and morphogenesis. The name "Wnt" is derived from a combination of "wingless" and "integrated," reflecting the original discovery of the pathway in Drosophila melanogaster as a homolog of the wingless gene in mice.

The WNT signaling pathway is initiated by the binding of WNT ligands to the Frizzled (Fz) family of receptors on the cell surface. This binding leads to the activation of a signaling cascade that ultimately results in the translocation of  $\beta$ -catenin from the cytoplasm to the nucleus, where it acts as a transcription factor to regulate the expression of target genes[8], [9].

The canonical WNT pathway is the well-studied branch of the WNT signaling pathway and is characterized by the activation of  $\beta$ -catenin-mediated transcription. In the absence of Wnt ligands, cytoplasmic  $\beta$ -catenin is phosphorylated by a destruction complex that includes the proteins Axin, Adenomatous polyposis coli (APC), and glycogen synthase kinase 3 $\beta$  (GSK3 $\beta$ ). Phosphorylated  $\beta$ -catenin is targeted for proteasomal degradation, preventing its accumulation in the nucleus.

Binding of Wnt ligands to the Fz receptor leads to the recruitment of Disheveled (Dvl) to the membrane and the inhibition of the destruction complex, allowing  $\beta$ -catenin to accumulate and translocate to the nucleus. Once in the nucleus,  $\beta$ -catenin binds to members of the T-cell factor/lymphoid enhancer factor (TCF/LEF) family of transcription factors to activate the expression of target genes, including those involved in cell proliferation, differentiation, and migration.

In addition to the canonical pathway, there are several non-canonical WNT signaling pathways that regulate cell migration and morphogenesis. These pathways include the planar cell polarity (PCP) pathway, which regulates cell polarity and directional cell movements, and the Wnt/Ca2+ pathway, which regulates calcium-dependent cellular processes such as cell adhesion and motility.

The Wnt signaling pathway has been implicated in a variety of developmental processes, including gastrulation, neural crest formation, limb development, and organogenesis. Aberrant activation or inhibition of the pathway has been associated with various developmental disorders and diseases, including cancer, osteoporosis, and Alzheimer's disease.

Overall, the Wnt signaling pathway plays a critical role in the genetic control of morphogenesis by regulating cell fate determination, tissue patterning, and cell migration. Further understanding of this pathway and its interactions with other signaling pathways will provide insights into the mechanisms underlying normal development and disease. Roles of Genetic Control of Cell Migration in Normal and Pathological Conditions:

The genetic control of cell migration plays a critical role in many physiological and pathological events. In normal conditions, cell migration is essential for embryonic development, tissue repair, and immune response. In pathological conditions, dysregulation of cell migration can lead to various diseases, including cancer metastasis, chronic inflammation, and autoimmune disorders[10]–[12].

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CHAPTER 14

# EVOLUTIONARY DEVELOPMENTAL BIOLOGY ANDFUTURE IN EMBRYOLOGY, ANATOMY, AND ELEMENTARY MORPHOGENESIS

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Evolutionary developmental biology, or Evo-Devo, is a field of biology that aims to understand how changes in developmental processes contribute to the evolution of new species. Evo-Devo combines principles from evolutionary biology and developmental biology to explore the genetic and molecular mechanisms that drive the formation and differentiation of complex organisms.One of the key concepts in Evo-Devo is the idea that small changes in the timing, location, or expression of genes during development can lead to significant changes in an organism's anatomy, behavior, or physiology. For example, variations in the genes that control the number or shape of limb buds can produce different types of limbs, such as wings, fins, or legs, depending on the evolutionary pressures and environmental conditions that shaped the evolution of each species.

Evo-Devo researchers use a variety of techniques to study the molecular and cellular processes that underlie development and evolution. These include comparative genomics, which involves comparing the genomes of different species to identify key genes and regulatory elements that control development; gene expression analysis, which measures the levels of specific genes and proteins during development; and experimental manipulation of developmental processes, such as gene knockouts, tissue grafts, or hormone treatments, to test their effects on morphogenesis.

One of the major discoveries in Evo-Devo is the role of homeobox genes, or Hox genes, in specifying the body plan of animals. Hox genes are a family of transcription factors that control the expression of other genes in a spatially and temporally specific manner, depending on their position along the anterior-posterior axis of the developing embryo. Hox genes are highly conserved across different animal phyla, suggesting that they played a key role in the diversification of animal body plans during evolution[1]–[3].

Another important aspect of Evo-Devo is the study of developmental constraints and plasticity. Developmental constraints refer to the limits or biases imposed by the genetic and molecular interactions that shape the developmental processes of an organism. For example, the molecular signaling pathways that control the formation of the head and the tail in a vertebrate embryo are highly conserved, which may limit the degree of variation in these structures across species. Developmental plasticity, on the other hand, refers to the ability of organisms to adapt to changing environments or conditions by altering their developmental trajectories or phenotypes. For example, some species of tadpoles can develop into different types of frogs, depending on whether they are exposed to predators, parasites, or different types of food.

Evo-Devo has also contributed to our understanding of the evolution of developmental processes and the origins of key innovations in animal morphology. For example, the evolution of feathers in birds is thought to have involved modifications of existing developmental pathways and structures, such as the scales of reptiles, to produce a novel and functional adaptation for flight and thermoregulation.

In summary, Evo-Devo is a rapidly evolving field of biology that integrates concepts and methods from developmental biology and evolutionary biology to explore the genetic and molecular mechanisms that control morphogenesis and the evolution of complex organisms. By studying the developmental processes and constraints that shape the diversity of life on Earth, Evo-Devo provides insights into the origins of biological form and function, as well as potential applications in biotechnology, medicine, and conservation.

Evolutionary developmental biology, or Evo-Devo, is a relatively new field that combines evolutionary biology and developmental biology to investigate the processes that shape the diversity of life on Earth. By studying the development of organisms, Evo-Devo researchers aim to understand how genetic and environmental factors interact to create the range of morphological and behavioral characteristics seen in different species.

Evolutionary developmental biology emerged in the 1980s as a response to the need for a better understanding of the genetic and developmental mechanisms underlying the evolution of complex traits. At the time, traditional evolutionary biology focused largely on the role of natural selection in shaping traits, while developmental biology focused on the mechanisms that produce these traits. Evo-Devo seeks to bridge the gap between these two fields by investigating the genetic and developmental processes that underlie evolutionary change.

Evo-Devo research draws on a range of disciplines, including genetics, developmental biology, paleontology, ecology, and evolutionary biology. By studying the development of organisms and how it varies between different species, Evo-Devo researchers can identify the genetic and developmental changes that have occurred over the course of evolution. This, in turn, can shed light on the processes that drive evolutionary change and the factors that constrain it.

#### The Role of Gene Regulation in Evo-Devo

One of the key insights of Evo-Devo research is the importance of gene regulation in shaping the development and evolution of organisms. While all organisms share many of the same genes, the ways in which these genes are regulated can differ greatly between species. This can result in significant differences in morphology and behavior.

For example, in the early 20th century, the British geneticist J.B.S. Haldane proposed that changes in the timing of developmental processes, such as the rate of cell division, could lead to evolutionary change. This idea was later developed into the concept of "heterochrony", which refers to changes in the timing of developmental events between different species.Heterochrony can occur in a number of ways. For example, changes in the timing of the onset of cell division can lead to differences in the overall size and shape of an organism. Similarly, changes in the timing of the cessation of cell division can affect the final size of an organism.

More recent research has identified specific genes that play a key role in regulating developmental timing. For example, the homeobox genes are a family of genes that are involved in the formation of body segments and other key developmental processes. Changes in the regulation of these genes can lead to significant differences in morphology between species.

#### **Evo-Devo and the Evolution of Novel Traits**

Evo-Devo, or Evolutionary Developmental Biology, is a field that combines principles of evolutionary biology and developmental biology to understand how the diversity of life has arisen through evolution. One of the key questions in Evo-Devo is how novel traits arise and how they are fixed in a population.

A novel trait is a feature that is not found in the ancestral species and is therefore considered new to the species. These traits can arise through various mechanisms, such as mutations or changes in gene regulation during development. The study of Evo-Devo has shed light on the processes by which these novel traits arise and how they can be fixed in a population.

One example of a novel trait is the development of feathers in birds. Feathers are thought to have evolved from reptilian scales, and the process by which they arose is thought to have been gradual, with intermediate stages that may have served different functions. The discovery of genes involved in feather development has allowed researchers to study the developmental mechanisms that underlie the evolution of this novel trait.

Another example of a novel trait is the evolution of the mammalian middle ear, which allows for improved hearing. The middle ear is derived from the jawbones of ancestral reptiles, and the process of its evolution involved changes in the structure of the bones and the development of new muscles and nerves. The study of the developmental pathways involved in the evolution of the middle ear has provided insights into the mechanisms by which novel traits can arise.

The study of Evo-Devo has also revealed that the evolution of novel traits often involves the modification of existing developmental pathways rather than the evolution of entirely new ones. This is because developmental pathways are highly conserved across different species, and modifications to these pathways can result in novel traits without disrupting the overall developmental program. In addition, Evo-Devo has highlighted the importance of gene regulatory networks in the evolution of novel traits. These networks are responsible for controlling the expression of genes during development, and changes in these networks can lead to the evolution of new developmental programs and the emergence of novel traits[4]–[6].

Overall, the study of Evo-Devo has provided important insights into the evolution of novel traits and how they are fixed in populations. By combining principles of developmental biology and evolutionary biology, researchers have been able to uncover the developmental mechanisms that underlie the evolution of novel traits and shed light on the diversity of life on earth. Another area of Evo-Devo research focuses on the evolution of novel traits, such as wings in insects or feathers in birds. These traits are thought to have arisen through the modification of existing structures, rather than through the evolution of entirely new structures.

One example of this process is the evolution of the wings of insects. Insects are thought to have evolved from crustacean-like ancestors that lived in the water. Over time, some of these ancestral organisms began to develop specialized outgrowths on their bodies that allowed them to move more efficiently through the water. These outgrowths eventually became wings, which allowed insects to take to the air. Studies of the genetic and developmental mechanisms underlying the evolution of insect wings have revealed the importance of the BMP and Wnt signaling pathways in this process. These pathways play a key role in the regulation of cell growth and differentiation, and changes in their regulation can lead to significant differences in morphology.

Embryology, anatomy, and elementary morphogenesis are crucial fields of study that provide us with an understanding of the development and structure of living organisms. These fields have come a long way since their inception, but there are still challenges and opportunities that need to be addressed to advance the field further.

One of the significant challenges in embryology and anatomy is the lack of understanding of how different genes interact and contribute to development. Although advances in genomics and transcriptomics have enabled the identification of several genes and pathways involved in embryonic development, there is still much to be learned about how these genes interact and function in different contexts. Understanding these interactions will help us develop better models for predicting developmental abnormalities and diseases.

Another significant challenge in these fields is the complexity of developmental processes, which are difficult to observe and study. Imaging technologies like confocal microscopy and live imaging have made significant contributions to understanding the mechanisms of embryonic development. However, there is still a need for improved imaging techniques that allow us to observe cellular dynamics and interactions in greater detail and with higher resolution. Furthermore, the field of morphogenesis faces significant challenges in understanding how cells, tissues, and organs interact to form complex structures in different organisms. While we have a basic understanding of the molecular and cellular mechanisms underlying morphogenesis, there is still a lack of knowledge about how these processes differ between species and how they can be manipulated to achieve desired outcomes. Understanding these processes will be essential for fields like tissue engineering and regenerative medicine.

Lack of understanding of complex genetic regulation: One of the biggest challenges in embryology, anatomy, and elementary morphogenesis is the lack of understanding of complex genetic regulation. The regulation of gene expression during embryonic development is complex and not yet fully understood. This complexity makes it difficult to study how different genes contribute to the development of specific structures and organs.

**Limited ability to study early development:** Another challenge is the limited ability to study early development. Embryonic development occurs rapidly, and it is difficult to obtain samples from early stages of development. This limitation makes it difficult to study the earliest events that shape the developing organism.

**Difficulty in studying three-dimensional structures:** The complexity of three-dimensional structures presents another challenge. Many structures and organs are highly complex,

making it difficult to study them in their entirety. Researchers need to develop new techniques and technologies to study these structures in detail.

Lack of standardization in nomenclature and techniques: A lack of standardization in nomenclature and techniques presents another challenge. Embryonic development and anatomical structures are described differently by different researchers, making it difficult to compare and combine results from different studies. Standardization is required to facilitate collaboration and make it easier to build upon existing research.

**Ethical concerns and limitations on research:** Ethical concerns and limitations on research present a significant challenge to embryology, anatomy, and elementary morphogenesis. Many studies involve the use of animals or human embryos, raising ethical concerns. These concerns can lead to restrictions on research, making it difficult to study certain developmental processes and structures.

Finally, the future of embryology, anatomy, and elementary morphogenesis is closely tied to advances in technology and computational methods. With the rise of big data and artificial intelligence, there is a need for novel computational methods to handle the enormous amounts of data generated by genomic, transcriptomic, and imaging techniques. Moreover, there is a need for novel technological developments that can aid in the study of development and morphogenesis.

There are several solutions to the major challenges faced in embryology, anatomy, and elementary morphogenesis. Here are five possible solutions:

**Collaboration:** Collaboration among researchers from different disciplines can help to bridge the gap between biology and physics, and lead to a more comprehensive understanding of developmental processes. Collaborative efforts can also facilitate the sharing of resources, data, and knowledge. Collaboration is essential in addressing major challenges in Embryology, Anatomy, and Elementary Morphogenesis. Here are some ways collaboration can help address these challenges

**Shared expertise:** Collaboration allows experts in different fields to work together and share their knowledge and expertise. By bringing together researchers from different backgrounds, collaboration can help overcome disciplinary boundaries and lead to innovative solutions to complex problems.

Access to resources: Collaboration can provide access to resources and facilities that may not be available to individual researchers or institutions. This can include specialized equipment, research funding, and access to research databases and archives.

**Multidisciplinary approaches:** Many of the challenges in Embryology, Anatomy, and Elementary Morphogenesis require a multidisciplinary approach. Collaboration allows researchers from different fields to work together and combine their expertise to tackle complex problems from different angles.

**Networking:** Collaboration can help build networks and partnerships between researchers, institutions, and organizations. This can lead to new collaborations and partnerships, as well as opportunities for joint research projects and funding.

**Sharing data and results:** Collaboration can promote the sharing of data and research results. This can help ensure that research is reproducible and can lead to new insights and discoveries. Overall, collaboration is critical for addressing major challenges in Embryology, Anatomy, and Elementary Morphogenesis. By working together, researchers from different fields can combine their expertise and resources to develop innovative solutions to complex problems.

**Technological Advances:** Advances in technology, such as 3D imaging and printing, tissue engineering, and gene editing, have the potential to revolutionize our understanding of embryology, anatomy, and morphogenesis. These technologies can provide new insights into the complex mechanisms that underlie development, and offer new opportunities for the creation of models and interventions.

**Interdisciplinary Training:** There is a need for interdisciplinary training programs that can equip students and researchers with the skills and knowledge necessary to work across disciplines. Such programs can help to foster a more collaborative and integrated approach to research, and prepare researchers to tackle the complex challenges faced in embryology, anatomy, and morphogenesis.

Interdisciplinary training can play a vital role in addressing the major challenges in embryology, anatomy, and elementary morphogenesis. Bridging gaps between fields: Interdisciplinary training can bridge the gap between different fields of study, such as genetics, developmental biology, and anatomy. This can facilitate the exchange of ideas and promote collaboration among researchers, leading to a more holistic approach to understanding embryology and morphogenesis[7]–[9].

**Innovating new techniques:** By combining different techniques from various fields, researchers can develop new methods for studying embryology and morphogenesis. For example, advances in imaging technologies have allowed researchers to study the development of tissues and organs in real-time, providing new insights into the process of morphogenesis.

**Improving experimental design:** Interdisciplinary training can help researchers design experiments that are more comprehensive and rigorous. For example, a researcher with a background in genetics and developmental biology can design experiments that incorporate both genetic and developmental approaches to study the same phenomenon.

Advancing personalized medicine: Interdisciplinary training can help researchers understand the individual differences in embryonic development that contribute to disease. By combining knowledge from genetics, developmental biology, and anatomy, researchers can develop personalized therapies for patients with developmental disorders.

**Improving communication and public engagement:** Interdisciplinary training can also improve communication between researchers and the public. By developing a common language and understanding of complex scientific concepts, researchers can better communicate their findings to a wider audience, promoting greater public engagement and understanding of embryology and morphogenesis.

Ethical Guidelines: As research in embryology and anatomy progresses, it is important to establish ethical guidelines and regulations to ensure that research is conducted in a

responsible and ethical manner. Guidelines can help to ensure that research is conducted in a manner that is respectful of the rights and welfare of all animals and humans involved in the research.

**Public Education:** Public education is essential for promoting public understanding of the importance and potential benefits of research in embryology, anatomy, and morphogenesis. Greater public understanding can lead to increased funding, support, and collaboration for research efforts, as well as better dissemination of research findings to the public. One way to promote public education is through outreach programs, such as workshops, seminars, and public lectures. These events can provide opportunities for researchers to interact with the public, share their work, and answer questions. They can also be tailored to specific audiences, such as students, teachers, and community members. Another way to promote public education is through social media and other online platforms. Researchers can use these platforms to share their work, engage with the public, and provide updates on the latest advancements in the field. They can also use these platforms to address common misconceptions and ethical concerns. In addition, integrating embryology, anatomy, and elementary morphogenesis into school curricula can help educate the next generation of researchers and promote public understanding of the field. This can include developing ageappropriate lesson plans and providing teacher training programs. Finally, collaboration with science museums and other public institutions can also help promote public education. These institutions can provide interactive exhibits, workshops, and other educational resources that can engage the public and provide a better understanding of embryology, anatomy, and elementary morphogenesis[10]-[12].

Public education is crucial for promoting understanding and support for embryology, anatomy, and elementary morphogenesis research. By using outreach programs, social media, school curricula, and public institutions, we can educate the public and promote ethical considerations, interdisciplinary collaboration, and scientific advancement in the field. In conclusion, the challenges faced by embryology, anatomy, and elementary morphogenesis are complex and multifaceted. Researchers need to develop new techniques, technologies, and standardization methods to overcome these challenges and make new discoveries in this field. At the same time, they must navigate ethical concerns and limitations on research to ensure that their work is responsible and beneficial to society.

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