



KARPAGAM ACADEMY OF HIGHER EDUCATION
(Deemed University Established Under Section 3 of UGC Act 1956)
Coimbatore - 641021.

(For the candidates admitted from 2015 onwards)

DEPARTMENT OF BIOCHEMISTRY

SUBJECT : PLANT BIOCHEMISTRY
SEMESTER : V
SUBJECT CODE : 15BCU504 **CLASS : III B.Sc. Biochemistry**

Programme Objective: Understanding of the plant cell organelles and their functions. To gain a wide knowledge about plant growth substances, plant nutrition and photo morphogenesis.

Programme learning outcome:

The students after completion of this course would have

- Gained knowledge on plant cell organelles and their functions.
- Obtained knowledge on different transport systems in plants.
- Understood the basics of photosynthesis and different cycles of elements.
- Gained knowledge on the significance of plant nutrients and plant growth regulators.

UNIT-I

Plant Cell and Transport mechanisms: cell wall-structure and functions. Plastids: types and functions. Transport mechanisms: diffusion, vacuole and turgor pressure, osmosis and imbibition, conditions necessary for imbibition. Absorption and translocation of water: Mechanism-Active and passive absorption, Factors affecting absorption of water. Ascent of sap: Mechanism and theories.

UNIT-II

Photosynthesis: Photosynthetic pigments: Chlorophyll, carotenoids and phycobillin: Photosynthetic apparatus-structure and functions of chloroplasts: light absorption. Light reactions-Two kinds of chemical system-Photosystem I&II, evidences in support of light reaction. Hill's reaction; Emerson effect, cyclic and non-cyclic phosphorylation. Dark reaction : Calvin's cycle (C3 plants). Hatch-slack cycle (C4 plants) and CAM plants. Factors affecting photosynthesis. Photorespiration.

UNIT-III

Cycles of elements and Plant Nutrition: Nitrogen cycle-Ammonification, nitrification, nitrate reduction and denitrification. Nitrogen fixation: Symbiotic and non-symbiotic nitrogen fixation. Sulfur cycle: Release of sulfur from organic compounds; Oxidation of sulfur compounds; Reduction of sulfate. Phosphorus cycle and carbon cycle. Plant nutrition: Specific roles of essential elements and

their deficiency symptoms in plants. Macronutrients: Carbon, Hydrogen, Oxygen, Nitrogen, Sulfur, Phosphorus, Calcium and Potassium. Micronutrients: Manganese, Boron, Copper, Zinc, Molybdenum and Chlorine.

UNIT-IV

Plant growth regulators: Auxins: Chemistry, biosynthesis, mode of action, bioassay, practical applications of synthetic auxins. Gibberellins: Chemistry, biosynthesis and mechanism of action, role of endogenous gibberellins, bioassay and practical applications. Chemistry, mode of action and physiological role of cytokinins, abscisic acid and ethylene.

UNIT-V

Photomorphogenesis

Photoperiodism. Photochrome-function in growth and development of plant. Biochemistry of seed germination. Senescence: Biochemical changes during senescence. Senescence process in life cycle of plants.

TEXT BOOKS

Verma, S.K., & Mohit Verma. (2013). *A Text Book of Plant Physiology, Biochemistry and Biotechnology*. (6th ed.). New Delhi. S. Chand and Co.

Goodwin, T.W., & Mercer, E.I. (1990). *Introduction to Plant Biochemistry*. (2nd ed.). New York, NY: Robert Maxwell.M.C Publisher.

Bonner, J., & Varner, F.J. (2012). *Plant Biochemistry*. (3rd ed.). Burlington: Elsevier Science.

REFERENCES

Buchanan, B.B. (2015). *Biochemistry and Molecular Biology of Plants*. New Jersey, NJ: John Wiley.

Hans-Valter Heldt. (2005). *Plant Biochemistry and Molecular Biology*. England: Oxford University Press.

Wink, M. (2010). *Functions and Biotechnology of Plant Secondary Metabolites*. (2nd ed.), London: Blackwell Publishing Ltd,.



KARPAGAM ACADEMY OF HIGHER EDUCATION
(Deemed University Established Under Section 3 of UGC Act 1956)
Coimbatore - 641021.

(For the candidates admitted from 2015 onwards)

DEPARTMENT OF BIOCHEMISTRY

LECTURE PLAN

STAFF NAME : Dr. L. HARIPRASATH
SUBJECT : PLANT BIOCHEMISTRY **SUBJECT CODE : 15BCU504**
SEMESTER : V **CLASS : III B.Sc. Biochemistry**

Sl. No	Duration of Period	Topics to be Covered	Books Referred	Page No	Web Page referred
UNIT – I					
1.	2	Cell wall-structure and functions	T1	5-11	
2.	2	Plastids: types and functions Transport mechanisms: diffusion, vacuole pressure	T2	115-118	
3.	2	Turgor pressure, osmosis and imbibition,	T1	116-122	
4.	1	Conditions necessary for imbibition	T1	137-141	
5.	2	Absorption and translocation of water: Mechanism-Active and passive absorption,	T1	141-145	
6.	1	Factors affecting absorption of water	T1	145-146	
7.	2	Ascent of sap: Mechanism and theories.	T1	148-158	
8.	1	Revision			
9.	1	Class test			
Total number of hours planned for Unit I: 14					
UNIT – II					
1.	2	Photosynthetic pigments: Chlorophyll, carotenoids and phycobillin	T3	76-82	
2.	1	Photosynthetic apparatus-structure and functions of chloroplasts	T1	34-40	
3.	1	Light absorption.	T3	90-103	
4.	2	Light reactions-Two kinds of chemical system-Photosystem I&II,			
5.	1	Evidences in support of light reaction.			
6.	1	Hill's reaction; Emerson effect, cyclic and non-cyclic phosphorylation.	T1 R1	119-122 82-84	
7.	2	Dark reaction : Calvin's cycle (C3 plants).	T3	105-118	
8.	2	Hatch-slack cycle (C4 plants) and CAM	T1	283-288	

		plants.			
9.	1	Factors affecting photosynthesis	T1	309-315	
10.	1	Photorespiration	R1	195-211	
11.	1	Revision			
12.	1	Class test			
Total number of hours planned for Unit II: 16					
UNIT – III					
1.	2	Nitrogen cycle-Ammonification, nitrification, nitrate reduction and denitrification..	T3	217-222	
2.	2	Nitrogen fixation: Symbiotic and non-symbiotic nitrogen fixation.	T2	889-895	
3.	2	Sulfur cycle: Release of sulfur from organic compounds; Oxidation of sulfur compounds; Reduction of sulfate.	T2	612-619	
4.	1	Phosphorus cycle	T1	196	
5.	1	Carbon cycle	T1	313	
6.	1	Plant nutrition: Specific roles of essential elements and their deficiency symptoms in plants.	T1	194-201	
7.	1	Macronutrients: Carbon, Hydrogen, Oxygen and Nitrogen,			
8.	1	Macronutrients: Sulfur, Phosphorus, Calcium and Potassium.			
9.	1	Micronutrients: Manganese, Boron, and Copper	T1	201-205	
10.	1	Micronutrients: Zinc, Molybdenum and Chlorine.			
11.	1	Revisions			
12.	1	Class test			
Total number of hours planned for Unit III: 15					
UNIT – IV					
1.	2	Auxins: Chemistry, biosynthesis, mode of action, bioassay.	R1	465-468	
2.	1	Auxins: practical applications of synthetic auxins.			
3.	1	Gibberellins: Chemistry, biosynthesis	T2	750-764	
4.	2	Gibberellins: Mechanism of action, role of endogenous gibberellins,			
5.	1	Gibberellins: bioassay and practical applications.			
6.	2	Chemistry, mode of action and physiological role of cytokinins	T3	325-327	
7.	2	Chemistry, mode of action and			

		physiological role of abscisic acid			
8.	1	Chemistry, mode of action of ethylene.	R1	474-476	
9.	1	physiological role of ethylene			
10.	1	Revision			
11.	1	Class test			
Total number of hours planned for Unit IV: 15					
UNIT V					
1.	2	Photoperiodism.	T2	683-697	
2.	2	Photochrome-function in growth and development of plant.	T1	621-624	
3.	2	Biochemistry of seed germination.	T1	625-626	
4.	2	Senescence: Biochemical changes during senescence.	T1	626-632	
5.	2	Senescence process in life cycle of plants.	T3	341-342	
6.	1	Revision			
7.	1	Class test			
Total number of hours planned for Unit V: 12					
Previous year ESE Question Paper Discussion					
1.	1	Previous year ESE question paper discussion 1			
2.	1	Previous year ESE question paper discussion 2			
3.	1	Previous year ESE question paper discussion 3			
Total Hours Planned: 75					

Support Materials

T1: Verma, S.K., & Mohit Verma. (2013). *A Text Book of Plant Physiology, Biochemistry and Biotechnology*. (6th ed.). New Delhi. S. Chand and Co.

T2: Bonner J and Varner JE, 1976. *Plant Biochemistry*. 3rd edition. Academic press Inc., New Delhi

T3: Goodwin, T.W., & Mercer, E.I. (1990). *Introduction to Plant Biochemistry*. (2nd ed.). New York, NY: Robert Maxwell.M.C Publisher.

R1: Hans-Valter Heldt. (2005). *Plant Biochemistry and Molecular Biology*. England: Oxford University Press.



KARPAGAM ACADEMY OF HIGHER EDUCATION
(Deemed University Established Under Section 3 of UGC Act 1956)
Coimbatore - 641021.

(For the candidates admitted from 2015 onwards)

DEPARTMENT OF BIOCHEMISTRY

COURSE MATERIAL

STAFF NAME	:	Dr. L. HARIPRASATH	
SUBJECT	:	PLANT BIOCHEMISTRY	SUBJECT CODE : 15BCU504
SEMESTER	:	V	CLASS : III B.Sc. Biochemistry

UNIT-I

Plant Cell and Transport mechanisms: cell wall-structure and functions. Plastids: types and functions Transport mechanisms: diffusion, vacuole and turgor pressure, osmosis and imbibition, conditions necessary for imbibition. Absorption and translocation of water: Mechanism-Active and passive absorption, Factors affecting absorption of water. Ascent of sap: Mechanism and theories.

TEXT BOOKS

Verma, S.K., & Mohit Verma. (2013). *A Text Book of Plant Physiology, Biochemistry and Biotechnology*. (6th ed.). New Delhi. S. Chand and Co.

Bonner, J., & Varner, F.J. (2012). *Plant Biochemistry*. (3rd ed.). Burlington: Elsevier Science.

Goodwin, T.W., & Mercer, E.I. (1990). *Introduction to Plant Biochemistry*. (2nd ed.). New York, NY: Robert Maxwell.M.C Publisher.

REFERENCES

Buchanan, B.B. (2015). *Biochemistry and Molecular Biology of Plants*. New Jersey, NJ: John Wiley.

Hans-Valter Heldt. (2005). *Plant Biochemistry and Molecular Biology*. England: Oxford University Press.

Wink, M. (2010). *Functions and Biotechnology of Plant Secondary Metabolites*. (2nd ed.), London: Blackwell Publishing Ltd.,

Unit I

1. Plant Cell and Transport mechanisms

1.1. Cell wall-structure and functions

Cell is the structural and functional unit of living organisms. On the basis of number of these units, living organisms have been divided into two groups:

1. Unicellular
2. Multicellular

Unicellular organisms are those which start their life from a single cell or unit and till death continue it in the same manner. i.e., their body is made up of a single cell. The examples of unicellular organisms are yeast, diatoms, bacteria and protozoans. Multicellular organisms are those which are made up of more than one cell. These organisms also start their lives from a single cell but later on it undergoes continued divisions and develops into a multicellular body. All higher plants and animals are multicellular. The cells in multicellular organisms get arranged in a definite manner to form different parts of the body. These parts perform different functions and all are governed by the common activity of each cell present in that part of the body.

Electron microscopic studies have revealed that each cell contains in it several living sub-cellular particles called cell-organelles which perform different functions. The chief cell organelles are mitochondria, endoplasmic reticulum, ribosome's, plastids, liposome's, Golgi body and nucleus etc. Thus it can be said that despite being the unit of living organisms, cell has a much complex structure.

Types of cells

The cells are generally classified into two main groups

1. Prokaryotic cells
2. Eukaryotic cells

Cell wall

Cell wall is the characteristic feature of all the plant cells. It is the outermost layer and covering of the plasma membrane. The cell wall is entirely lacking in animal cells. The cell wall is rather rigid, strong, thick, porous and non-living structure and is secreted by the living matter of the cell.

It is laid down at the telophase stage and is believed to be formed by the fragments of the endoplasmic reticulum. In young cells, the cell wall is thin, elastic and about 1 to 3 μ thick while in old and mature cells the cell wall becomes stiff and 5 to 10 μ thick. The cell wall of parenchymatous cells are comparatively thinner than those of collenchymas, sclerenchyma and xylem vessels.

Structure of cell wall

The cell walls are complex and highly differentiated in some tissues. They also possess special sequence of their arrangement. The wall of a solitary cell can be differentiated into the following layers:

- i. Primary cell wall
- ii. Secondary cell wall
- iii. Tertiary cell wall
- iv. Middle lamella

Generally, primary and secondary cell walls are found in the cell. Tertiary cell walls are rare and are found in the xylem tracheids of Gymnosperms. These walls are deposited in layers one after the other during growth and differentiation.

1. Primary cell wall

The outermost wall layer of the cell is termed as primary cell wall and is regarded to be the first deposition product. It develops in the young growing or meristematic cells and parenchymatous cells. The primary cell wall is comparatively thin and permeable. Certain epidermal cells of leaf and the stem also possess the cutin and cutin wax, which make the primary wall impermeable. At first the primary wall is rather elastic and able to extend as the cell grows, but when more cellulose is deposited, it becomes more rigid. In many fungi the cell wall is composed of chitin.

2. Secondary cell wall

Next to the primary cell wall is another layer known as secondary cell wall. It is comparatively thicker, than the primary wall. The secondary cell wall is generally found in the mature, permanent or non-growing cells. The outer surfaces of the epidermal cells with secondary wall of some leaves and the stem may have a cuticle rich in fats and waxes which tends to limit water loss. The primary cell wall is composed of cellulose but the secondary wall in addition to cellulose contains pectin's, non-cellulose polysaccharides, lignin and a phenolic polymer which imparts hardness and mechanical rigidity to the wood. In certain cells, it is further differentiated into outer, middle and inner layers.

3. Tertiary cell wall

Rarely in certain cells is a third layer added inside the secondary wall which is known as tertiary cell wall. The presence of tertiary cell wall was described by Bucher (1953) in the xylem tracheids of Gymnosperms where it is quite thin and produces many warty outgrowths. The tertiary cell wall differs from the primary and secondary walls in its morphology, chemical composition and staining properties. Tertiary cell-wall is mainly composed of xylan instead of cellulose.

4. Middle lamella

The primary walls of the two adjacent cells are often separated by a layer or a structure known as middle lamella. During the development of cell-walls, the middle lamella is formed first. It is composed of calcium and magnesium pectates. The pectates are viscous and gelatinous. Middle lamella binds the adjoining cells firmly. In mature and aged cells the middle lamella is dissolved and consequently the cells are loosened. During the maturation of fruits the pectic substances of the middle lamella become soluble due to the action of pectolytic enzymes. The ripe fruits are therefore, soft.

Chemical composition of cell-walls

The primary cell wall consists of intertwined cellulose fibres. It may have a deposition of pectin, lignin, hemicelluloses etc. The cellulose molecules are polymers of disaccharide cellobiose having approximately 3000 glucose units. The glucose molecules are arranged in the form of chain which are joined by β 1 \rightarrow 4 linkages. The cellulose fibrils are about 0.25 μ wide and up to 1 μ long. They are woven into an irregular net with a mesh of about 0.3 μ .

Many chains of cellulose molecules lie parallel to each other to form the bundles. A bundle of 100 cellulose molecular chains forms the elementary fibril known as micelle. The 20 parallelly arranged micelles form another bundle known as microfibril. It is about 250 A° thick. Similarly, 250 microfibrils form the large sized bundle known as macrofibril. The macrofibrils ultimately form the main framework of the cell-wall. The hemicelluloses are the polysaccharides of pentose as well as hexose sugars like arabinose, xylose, mannose and galactose. The pectic substances are the long chains of repeating units

of uronic acid derivatives of hexoses, glucuronic and galacturonic acids. Lignin is composed of coniferyl alcohols and cutin of many fatty acids. Chitin is a polymer of glucosamine.

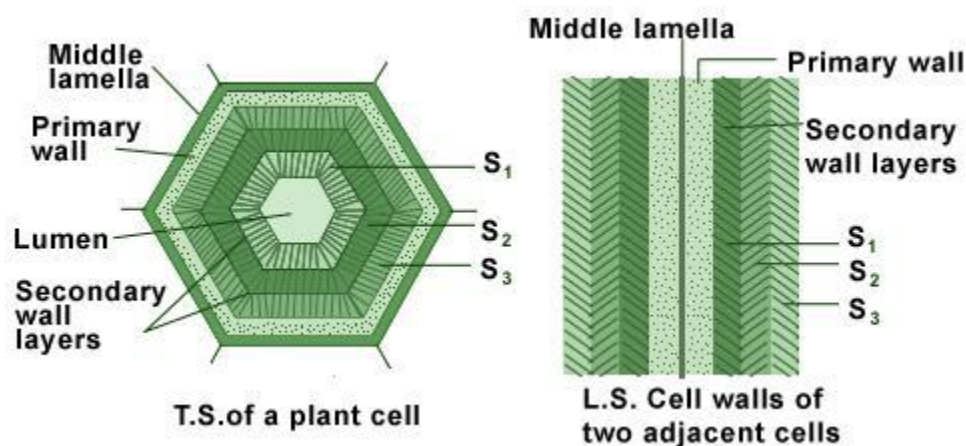
The secondary wall is also composed of cellulose and contains lignin. The formation and structure of microfibrils and macrofibrils are the same as in case of primary cell wall. The only difference is in their arrangement. In the secondary walls the cellulose microfibrils in a macrofibril are comparatively more compactly arranged.

The tertiary wall is composed of xylan instead of cellulose and the middle lamella is composed of calcium and magnesium pectates.

The relative concentration of cellulose, hemicelluloses and pectic substances in the primary cell-wall is not constant in all the cells. It varies from one cell type to another cell-type. Bishop (1958) reported the presence of high concentration of hemicelluloses. Jensen (1960) found that the provascular cells had high concentrations of pectic substances and hemicelluloses in their cell-walls. The cells of the cortex and protoderm had lower concentrations.

Average analysis of primary cell wall

S.No	Substance	Percent of fresh weight
1	Water	60
2	Hemicelluloses	5-15
3	Cellulose	10-15
4	Pectic substances	2-8
5	Proteins	1-2
6	Lipids	0.5-30



Functions of cell wall

Cell wall performs the following important functions:

- It protects the cell from adverse environment conditions.
- It provides a definite shape of the cell.
- It provides strength to the cell.
- It permits the entry of molecules of different sizes.
- It determines the manner of cell division and growth.

- vi. It possesses plasmodesmata through which cells remain connected with adjacent cells.
- vii. It separates one cell from the other.

1.2. Plastids: Types and functions

Plastids are characteristic structures of almost all the plant cells and a few animal cells like Euglena. They represent the largest cytoplasmic cell organelles and are intimately related with the synthesis and storage of carbohydrates, proteins and lipids.

Classification of plastids:

The plastids are generally classified into three main groups depending upon the presence or absence of colour contents.

(A) *Leucoplasts*

These plastids are devoid of pigments. They are mainly meant for storing the food materials like starch (carbohydrates), lipids and proteins. The leucoplasts vary in shape and size and are usually rod like spheroid. They are of common occurrence in embryonic cells, sex cells and meristematic cells. Depending upon the presence of storage material, they are further classified into following heads:

(i) Amyloplasts – The starch storing leucoplasts are known as amyloplasts. They are found in those cells which store the starch and particularly in storage tubers, cotyledons and endosperm.

(ii) Elaeoplasts – The oil storing leucoplasts are known as elaioplasts. They are found in the seeds of both monocotyledonous and dicotyledonous plants. In most of the monocotyledonous plants the chloroplasts after losing their chlorophyll start storing oil. In the epidermal cells of orchidaceae and Liliaceae the disorganized plastids fuse to form oil droplets.

(iii) Proteinoplasts – The proteins storing plastids are known as proteinoplasts. They have been reported in the epidermal cells of Helleborus and seeds of Ricinus.

(B) *Chromoplasts*:

The coloured plastids are known as chromoplasts. They contain variety of pigments and most of them synthesize food through the process of photosynthesis. The chromoplasts are found in the cells of leaves, many flowers and fruits. Some of the common chromoplasts of plant cells are as follows:

(i) Chloroplast – The green plastids are known as chloroplasts. They contain the pigments chlorophyll A and chlorophyll B, DNA and RNA. The chloroplasts are found in green algae and higher plants where they play a role in the photosynthesis.

(ii) Phaeoplast – The brownish colour plastids are known as pheoplasts. They are found in the brown algae, diatoms and dinoflagellates. These appear brown due to masking effect of brown carotenoids. In brown algae, it is the pigment fucoxanthin which is responsible for the absorption of light and transfer of energy to the chlorophyll.

(iii) Rhodoplasts – They are red colour plastids and contain the pigment phycoerythrin. This pigment plays the role similar to fucoxanthin. Rhodoplasts are found in the members of rhodophyceae, the red algae.

(C) *Chromotophores*:

In the cells of blue green algae, fungi and bacteria the pigments are not organized within a discrete plastid body but are often arranged on lamellar structures in concentric rings or plates. Various types of chromotophores are as follows:

(i) Blue green chromatophores – They are found in blue green algae which contain the pigments c-phyocyanin and c-phycoerythrin along with chlorophyll A and carotenoids. These accessory pigments impart bluish hue in addition to green colour and perform photosynthesis.

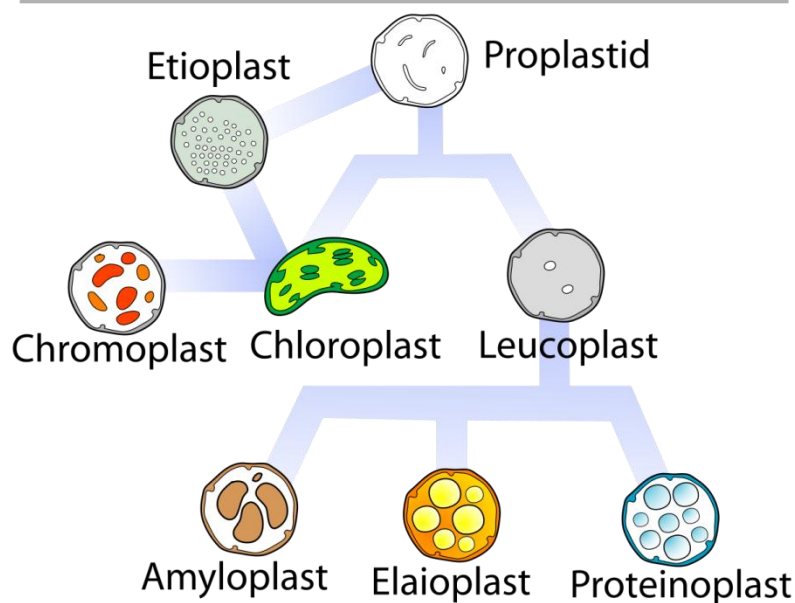
(ii) Bacterial chromatophores – Bacterial chromatophores are found in certain bacteria, eg., purple sulphur bacteria and contain the pigment bacteriochlorophyll. The green sulphur bacteria contain the pigment bacterio-viridin in their chromatophores. Bacterio-chlorophyll is very important in the absorption of infra-red light.

(iii) Carotenoids – They are generally found in certain bacteria, fungi, red coloured ripe tomato and pepper, flowers and fruits. Usually the pigment capsanthin occurs in the carotenoids of bacteria and fungi. Other carotenoids contain certain other pigments like fucoxanthin, xanthophylls and carotenes either singly or along with chlorophyll in the algae and other plants.

The various types of plastids are apparently homologous with each other. They can be transformed from one to another.

Among of various plastids, the chloroplasts are of most common occurrence in plants and have greatest biological importance, since they produce most of the chemical energy used by the living organisms through photosynthesis.

Plastids

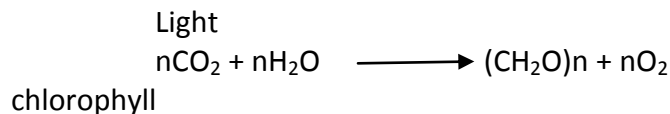


Functions of chloroplasts

(1) Photosynthesis

The most important function of chloroplast is photosynthesis. During the process of photosynthesis, the chlorophyll contained in the chloroplasts traps energy of sunlight emitted as photons and transforms it into chemical energy. Because this process takes place inside the chloroplast, they are considered as the centres of photosynthetic activity. The chemical energy is stored in the chemical bonds that are produced during the synthesis of various food stuffs like carbohydrates, lipids and proteins.

The photosynthesis can be defined with the help of following equation:



The equation shows that photosynthesis is the process in which carbon dioxide and water are converted into carbohydrates in presence of light and chlorophyll. The O₂ is evolved during the process.

The photosynthesis consists of two stages, the light or photochemical reaction and a dark or thermochemical reaction. The photochemical reaction is light sensitive and thermochemical (dark reaction) reaction is temperature sensitive. The photochemical reaction takes place in the following steps:

- (i) Photophosphorylation, in which ATP is formed from ADP through a chain of electron carriers.
- (ii) Hydrolysis and ionization of water, in which NADP is reduced to NADPH₂.

In dark reaction CO₂ is fixed and reduced by thermochemical mechanisms. This mechanism also involves a number of reactions in which the conversions of one product into another takes place in presence of certain specific enzymes. The initial enzyme, carboxydismutase, is responsible for the formation of phosphoglyceric acid molecules from ribulosediphosphate and CO₂.

(2) Protein synthesis

Special ribosomes are associated with chloroplasts and that chloroplasts contain a specific protein synthesizing system. The chloroplasts contain sufficient amounts of mRNA of its own. In the presence of CO₂, as the sole source of carbon, chloroplasts actively incorporate amino acids into proteins.

(3) Cytoplasmic heredity

The presence of DNA in chloroplasts has been related to the presence of a special non chromosomal genetic system. Thus, the chloroplasts act as carrier for genetic material.

(4) Role in Kreb's cycle and fatty acid synthesis

It is assumed that chloroplasts contain enzymes necessary for Kreb's cycle and for synthesis of fatty acids.

(5) Mutation

Some of the plastids even chloroplasts undergo mutation which is known as plastid-mutation. After mutation the plastids may perform altered functions. The genes of the plastids are known as plastogenes.

1.3. Transport mechanisms

1.3.1. Diffusion

The movement of free molecules of gases, liquids and solids from the region of higher concentration to the lower concentration due to internal or external forces is called diffusion. Blowing of wind, dispersal of good smell of agarbattis, intake of O₂ and liberation of CO₂ during respiration and dissolution of KMnO₄ particles in water are all the examples of diffusion. Such a diffusion of molecules in any region continues till it spreads throughout the available area. The movement of molecules depends upon the internal kinetic energy and that is why they show fast movement. During the movement all the molecules collide themselves and produce a pressure in the medium called diffusion pressure which is directly proportional to the concentration and temperature of diffusing molecules. It means if the temperature and concentration of diffusing molecules will be more, the diffusion pressure

will also be more and if the temperature and concentration is less, the diffusion pressure will also be less. Due to this reason, the diffusion always takes place from the region of higher diffusion pressure (conc.) to the region of lower diffusion pressure.

Mostly gases, liquids and solutes diffuse at different rates in different direction at the same place without affecting each other. This is the reason that O₂ and CO₂ both diffuse in different directions from a single stoma of leaf during respiration. The rate of diffusion is maximum in gases and minimum in solids when dissolved in solvent. Liquids stand on intermediate position in this regard.

Diffusion pressure of liquids

Like gases, solvents and liquids also possess diffusion pressure. The diffusion pressure of a pure solvent is always maximum and when solute particles are added in it, the diffusion pressure of the solution is reduced. The difference between the diffusion pressure of a solvent and its solution is called diffusion pressure deficit (DPD). When a deficiency in diffusion pressure of the solution is created, it starts absorbing more solvent particles to overcome this deficiency. Thus, can be found out the absorbing capacity of a solution through DPD and DPD of a solution gives an idea about the absorbing capacity of that solution. That is why DPD is also called suction pressure (SP).

Factors affecting the rate of diffusion

1. Temperature

It is directly proportional to the rate of diffusion. On increasing the temperature, the rate of diffusing particles is also increased because of increase in the velocity of these particles.

2. Concentration of the medium

It is inversely proportional to the rate of diffusion. On increasing the concentration of the medium, the rate of diffusion is reduced and on decreasing the concentration, it is increased.

3. The size and mass of the diffusing particles

If the size and mass of the diffusing particles is smaller, the rate of their diffusion will always be faster.

4. Solubility of solutes

The rate of diffusion increases with the rate of dissolution of solute in solution. Thus, more the solubility of any solute in solution, faster will be the rate of diffusion.

5. Diffusion pressure gradient (DPG)

The rate of diffusion of molecules of gases and liquids also depends upon the diffusion pressure gradient (DPG). When the difference in the diffusion pressure is more, faster is the rate of diffusion. In other words, steeper the rate of diffusion pressure gradient, the faster is the rate of diffusion.

6. Density of the diffusing particles

The rate of diffusion of gases is related with the density of diffusing particles. According to Graham's law of diffusion, 'The rate of diffusion of any gas particle is inversely proportional to the square root of the density.

Importance of diffusion in plants

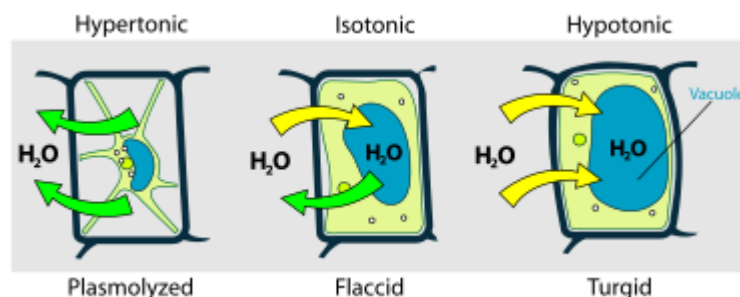
- i. The exchange of O₂ and CO₂ gases in the atmosphere through stomata of leaves takes place by the process of diffusion, O₂ gas participates in respiration whereas CO₂ in photosynthesis

- ii. During stomatal transpiration the water vapours from intercellular spaces diffuse in the atmosphere through stomata by the process of diffusion.
- iii. The diffusion of ions of mineral salts during passive absorption also takes place by this process.
- iv. The absorption of water through roots is also performed by diffusion.

1.3.2. Vacuole and turgor pressure

- Vacuoles are membrane-bound organelles that are found in cells of plants, animals and fungi.
- Plant cells tend to have a large central vacuole, while the vacuoles in animal cells tend to be smaller.
- The main function of vacuoles is to contain the waste products of the cells and isolate them from the rest of the organelles and the cytoplasm.
- Vacuoles maintain an acidic pH internally.
- This allows them to denature misfolded proteins transferred to the vacuole from the cytoplasm.
- The vacuoles can also help remove unwanted or toxic materials from the cells by exporting them to the cell membrane, where they are released to the outside surroundings of the cell.

Turgor pressure



- Turgor pressure is the pressure of water against the inside wall of a plant cell.
- Turgor pressure pushes the plasma membrane against the cell wall of plant, bacterial, and fungal cells as well as those protist cells which have cell walls.
- This pressure, turgidity or turgidness, is caused by the osmotic flow of water from an area of low solute concentration outside the cell into the cell's vacuole, which has a higher solute concentration.
- Healthy plant cells are turgid and plants rely on turgidity to maintain rigidity.

1.3.3. Osmosis

- Plant cells are surrounded by rigid cellulose walls, (unlike animal cells), but plant cells still take in water by osmosis when placed in pure water.
- However, plant cells do not burst because their cellulose cell walls limit how much water can move in.
- The cell walls exert pressure, called turgor pressure, as the cells take up water. Turgor pressure is analogous to the air pressure of an inflated tire.
- These physical forces can be described by a simple mathematical equation: $\psi = P + \pi$, where ψ ("Psi") is the water potential, a measure of the overall tendency of water to move into a cell; P is the pressure potential, a measure of the turgor pressure exerted by the cell walls; and π is the osmotic potential (see above). Water always moves from regions of higher ψ to areas of lower ψ . Animal cells do not have cellulose cell walls, so $P = 0$ and $\psi = \pi$ for these cells.

1.3.4. Imbibition

- Imbibition the uptake of water by substances that do not dissolve in water, so that the process results in swelling of the substance.
- Imbibition is a property of many biological substances, including cellulose (and other constituents of plant cell walls), starch, and some proteins.
- One example of imbibition that is found in nature is the absorption of water by hydrophilic colloids.
- Matrix potential contributes significantly to water in such substances. Examples of plant material which exhibit imbibition are dry seeds before germination.

Conditions necessary for imbibitions

- Water potential gradient between the absorbing surface & the liquid imbibed.
- The affinity between the absorbent & the imbibed liquid

1.4. ABSORPTION AND TRANSLOCATION OF WATER

Absorption and translocation of water: Mechanism-Active and passive absorption, Factors affecting absorption of water.

Importance of water to the plants

Water plays major roles in plants. It includes

1. The amount of water present in the soil changes the morphology and anatomy of the plants. Accordingly the plants are mesophytes, hydrophytes and xerophytes.
2. Water is a good solvent. It also acts as a reactant in various chemical reactions in plant cells.
3. Water helps in the formation of protoplasm.
4. The absorption and translocation of mineral salts and dissolved substances take place through water.
5. During photosynthesis, water is oxidized and O₂ is produced.
6. Water effects transpiration, seed germination, respiration, dispersal of fruits and seeds, activation of enzymes, hydrolysis of ATP growth and all other metabolic processes.
7. Water maintains the temperature in plant tissues.
8. Water maintains the turgidity of plant body.

1.4.1. Mechanism of Active and Passive absorption

According to Renner (1912 and 1915), the mechanism of water absorption is of two types:

1. Active absorption

When roots are involved actively in water absorption and water absorbing forces in plants are developed primarily in roots, such type of absorption is called active absorption. It is found in those plants where transpiration is less and water is present in sufficient amounts. Active absorption requires ATP released during respiration. It is also of two types:

- (a) Osmotic absorption: In this type of absorption the roots act like osmometer and water is absorbed according to osmotic gradient.
- (b) Non osmotic absorption: In this type of water absorption more ATP is required and water absorption takes place against osmotic gradient.

2. Passive absorption

When roots are inactive in water absorption and the water absorption forces develop primarily in leaves and stem and then reach to root through xylem, this type of water absorption is called passive absorption. It takes place mainly due to transpiration. Passive absorption is found in those plants where transpiration is very fast and it does not require ATP.

(a) Osmotic Active Absorption

Soil is made up of irregularly arranged soil particles. Well-developed air spaces are found among these particles. The spaces contain capillary water where air and mineral salts remain dissolved. The root hairs remain spread in this water. It has already been described earlier that each root hair contains a vacuole which remains filled with mineral salts, sugar and organic acids. The cell wall of root hair is permeable and it does not create any hindrance in the entry and exit of the liquids in the cell. Plasma membrane is semipermeable and it allows only the diffusion of water and important dissolved salts into the cytoplasm. The cell wall of root hair being hydrophilic in nature first absorbs soil water through imbibitions. The cytoplasm of root hair is usually concentrated than the capillary water of soil. The OP of cell-sap of root is also greater ($OP = 3 - 8 \text{ atm}$) than the OP of capillary water of soil ($OP = 1 \text{ atm}$). Thus, the DPD and SP become more in root hairs resulting in osmotic diffusion or endosmosis of water and its dissolved substances into the root hair.

After a definite time, the cells of root hairs become turgid and their OP, DPD and SP are reduced and TP is increased due to continuous absorption of water into the root hair. At this time, the cytoplasm of root hair becomes thin and its OP, DPD and SP are reduced in comparison to the cytoplasm of its adjacent first cortex cell-resulting in osmotic diffusion of water and its dissolved substances from root hair into the first cortex cell. Now, the OP, DPD and SP of this first cortex cell are also reduced and TP is increased. Similarly the osmotic diffusion of water and its dissolved substances from first cortex cell to second adjacent cell takes because the second cortex cell possesses greater OP, DPD and SP than the first cortex cell. This process continues till the water reaches to pericycle through all cortex cells and endodermal cell. The endodermis possesses passage cells and casparian striped cells. The passage cells are thin walled and found against protoxylems. Through these passage cells the water diffuses into cells of pericycle and ultimately into xylem vessels. As the water diffuses from root hair to first cortex cell, the cytoplasm of root hair cell again becomes concentrated and its OP, DPD and SP are also increased resulting again in osmotic diffusion of capillary water of soil into the root hair. This phenomenon continues for long time and thus, osmotic active water absorption takes place.

Pathway of water in root

The pathway of water in root cells is as follows:

Root hair → Epidermal cell → Various successive cortex cells → Endodermal cell (passage cell) → cells of pericycle → xylem cells → Xylem duct → Upward movement of water.

Root pressure

When the water enters from the turgid pericycle cells into xylem vessels, a pressure is created in the xylem of roots due to which the water rises to a certain height in the xylem. This pressure is called root pressure. Actually the root pressure is a type of hydrostatic pressure which is produced in the cell-sap of xylem vessels.

Demonstration of Root pressure

For the demonstration of root pressure, take a potted plant of tomato, Bryophyllum or zenia and cut the main stem in water with the help of knife about 4-5 cm above the soil. Now connect the cut end

of stem with a manometer glass tube through rubber-ring as shown in figure and fill the tube first with some water and then with mercury. Leave the experiment for some time and note the rise in mercury level in manometer which is mainly due to root pressure. The water absorbed by the root hairs is also pushed up in xylem vessels due to root pressure. If the upper portion of stem of any plant is cut, some liquid is oosed out due to root pressure.

The well known example of demonstration of root pressure is the production of toddy juice from the cuts of upper portion of stem.

(b) Non osmotic active absorption

According to Thimann (1951) and Kramer (1959) the water absorption is an active process which takes place against the osmotic gradient. In other words, sometimes the water absorption also takes place when the osmotic pressure of soil water, is greater than the OP of cytoplasm of root hair. This is against osmosis. Such type of water absorption is called non-osmotic active absorption. And it requires ATP produced during respiration of root cells. How this energy is utilized, it is not well worked out. The energy may be utilized directly, or indirectly. The theory of non-osmotic active absorption is supported by the following facts:

- (i) Correlation between water absorption and rate of respiration: Usually in presence of respiration reducing factors the rate of water absorption is also reduced. Thus, both the processes are inter-related.
- (ii) Reduction of water absorption in presence of respiratory inhibitors: It has been observed when a plant is placed in the solution of respiratory inhibitors like KCN, the rate of water absorption is reduced and from cut regions of plant the water secretion is also reduced or stopped. This indicated that respiration and water absorption processes are inter related. KCN directly, destroys the coenzymes of respiratory chain like NAD, FAD and cytochromes and thus effects respiration and other metabolic activities.
- (iii) Wilting of plants in oxygen deficient soil: In oxygen deficient soil, the plants do not get sufficient oxygen for respiration and ultimately wilt.
- (iv) Effects of auxins: In presence of auxins, the rates of metabolisms along with water absorption are increased.
- (v) The occurrence of water absorption and respiration only in living cells: Both these processes are found only in living plants and not in dead plants.

Passive absorption

It takes place mainly due to transpiration. In passive absorption, the roots remain inactive and the water absorbing forces are first produced in the cells of leaves. When DPD increases in the cells of leaves due to transpiration, the water diffuses from the xylem cells of leaves to all mesophyll cells. When the rate of transpiration is high, a tension is created in the water column of xylem which increases the DPD of water. This tension is like negative pressure and it moves from leaves to roots. At this stage, the DPD of peripheral cells of young roots becomes very high. The DPD in roots increases from xylem cells to epidermal root hairs. The rate of passive water absorption is directly proportional to the rate of transpiration and it requires sufficient amount of water in the soil.

Differences between Active and Passive absorption

Active absorption	Passive absorption
1. It requires energy or ATP	It does not require energy

2.Active absorption creates root pressure	Root pressure is not created due to passive absorption
3.It requires oxygen	It does not require oxygen
4.The movement of water takes place from the solution of higher concentration to the solution of lower concentration i.e., against the concentration gradient	The movement of water takes place from the solution of lower concentration to the solution of higher concentration i.e., according to osmotic gradient

Apoplast and symplast concept

The two terms, apoplast and symplast were used by Munch (1930) to explain the flow of water and minerals in plants. He used the term apoplast for the non-living or dead portions of the plants like interconnecting cell-walls, intercellular spaces, cell-walls of endodermal cells excluding casparian strips, cell-walls of pericycle, xylem tracheids and vessels. All these non-living portions constitute a single system called apoplast. This system is continuous throughout the plant and through this system the water moves chiefly due to capillary action or free diffusion along the gradient. The term symplast was used by Munch for the living portions of the plants like cytoplasm of all living cells connected through plasmodesmata present in the cell-walls. Since only the living system is included under symplast, here the water moves chiefly due to osmosis.

1.4.2. Factors affecting the rate of water absorption

- (i) **Available soil water:** Soil water is found in many forms such as gravitational, capillary, hygroscopic and chemically bound water. But out of these only capillary water is available for the absorption of plants. The rate of water absorption in plants remains constant the same in between field capacity and permanent wilting percentage. If the amount of water is increased than the field capacity, it creates a bad effect on soil aeration and also reduces the rate of water absorption. Similarly, on shortage of soil water or reduction in permanent wilting percentage, the water absorption rate is also reduced.
- (ii) **Soil aeration:** The rate of water absorption increases when the aeration in soil is high. In other words, the rate of water absorption is decreased in absence of oxygen. The reduction in rate of water absorption is due to lack of O₂ and accumulation of CO₂ which effects the plants in following manner:
 - a) **CO₂ reduces metabolic activities and respiration rates.**
 - b) **It reduces the size and growth of roots.**
 - c) **It increases the concentration of protoplasm which results in the reduction of permeability.**

Due to above effects of CO₂ the rate of water absorption is decreased. This situation is created in water logged soil where the aeration is quite poor. Such soil is usually physiologically dry and is not fit for water absorption.

- (iii) **Concentration of the soil solution:** the osmotic pressure of any solution is directly proportional to its concentration. In other words the OP of more concentrated solutions is always greater than the OP of less concentrated solutions. If the soil water contains more salts its concentration and OP will also be more. The rate of water absorption will be reduced if the OP of soil solution will be more than the OP of cell sap of roots. Due to this

reason the plants growing in alkaline and marsh soil show a very little or no water absorption.

- (iv) Soil temperature: Generally, the rate of water absorption is maximum between 20-30°C. It is reduced if the temperature is less than 20°C or more than 30°C. at very low temperature or at 0°C the water absorption is almost stopped and its rate becomes 0. On very much reduction in temperature, the rate of water absorption is affected directly or indirectly in the following manners.
- It increases the concentration of protoplasm.
 - It reduces the permeability of plasma membrane.
 - The growth roots is reduced which results in reduction of their length.
 - The rate of diffusion of soil water in to roots is also reduced.
 - Metabolic activities of root cells are also reduced.
- (v) Root system: root system also affects the rate of water absorption. Those plants which possess hairy and well developed root system show higher rates of water absorption in comparison to those which possess very small roots and less number of root hairs.

1.5. Ascent of sap

1.5.1. Mechanism and theories

Introduction

The water and soluble mineral salts absorbed by the roots reached to the leaves through roots, stem and branches of the plant. The phenomenon of ascending of absorbed water against gravitation through the vessels and tracheids of xylem is called ascent of sap.

Path of ascent of sap

Path of ascent of sap can be demonstrated by the following experiments.

Experiment 1

It can be studied easily in the white flowered lupin plant or rose plant. Cut a flowering twig of either lupin plant or rose plant in water to avoid the entry of air bubbles and place it in beaker containing a solution of eosin in water. After a few hours, the minute veins of the petals of flower become reddish. Now cut the TS of stem and observe that xylem vessels and tracheids are red. It proves that ascent of sap in plants takes place through vessels and tracheids of xylem.

Experiment 2

Cut the lower portion of white balsam stem carefully in water. So that the air bubbles could not enter into it. Place this portion vertically in eosin solution and observed after a few hours that the stem possesses many minute vertical red linings due to ascent of eosin solution. Now cut the TS of the stem and observe that only the vessels and tracheids of xylem become red whereas other tissues have no effect of eosin and remain white. It again proves that ascent of sap in plants takes place through vessels and tracheids of xylem.

Experiment 3

Ringing experiment

This experiment was performed for the first time by Stephen Hales. It also demonstrates the ascent of sap in the stem through vessels and tracheids of xylem. For this experiment take a potted plant and remove in circular fashion at some place of stem, all the ousted tissues (epidermis, cortex, endodermis, pericycle and phloem) except vessels and tracheids of xylem and pith. Leave the experiment for a few days and observe that the leaves of the plant are still green. It proves that the water and soluble salts ascend through xylem tissues. The question may arise that the water may also ascend through pith cells instead of xylem cells. To confirm that water and dissolved salts ascend through xylem tissues and not through pith cells, destroy the pith cells also with the help of a large needle or wire and set the experiment. After a few days you will observe again that the leaves of this plant are still green. This confirms that the water ascends only through the xylem tissues and not through any other tissue.

From all the above experiments, it becomes clear that the water and dissolved mineral salts ascends in the stem through vessels and tracheids of xylem (xylem tissues).

KARPAGAM ACADEMY OF HIGHER EDUCATION
COIMBATORE - 21
DEPARTMENT OF BIOCHEMISTRY
III B.Sc BIOCHEMISTRY
BATCH: 2015 - 2018

15BCU504 - Plant Biochemistry

PART A (20 X 1 = 20 MARKS) - Online MCQ questions

SL.NO	QUESTION	OPTION 1	OPTION 2	OPTION 3	OPTION 4	ANSWER
Unit 1						
1	What features do plant cells have that animal cells lack?	Nucleus and cell membrane	cell wall and chloroplasts	cytoplasm and cell wall	cell membrane and chloroplasts	cell wall and chloroplasts
2	Which two plant cell structures work together, like security guards, for the cell?	Cytoplasm and Nucleus	Cell Membrane and Cell Wall	Nucleus and Oxygen	Oxygen and Cell Membrane	Cell Membrane and Cell Wall
3	The movement of molecules from an area of high concentration to an area of lower concentration is known as :	Osmosis	Diffusion	Active transport	Phagocytosis	Diffusion
4	The movement of water molecules from an area of high concentration to an area of low concentration through a semipermeable membrane is known as :	Osmosis	Diffusion	Active transport	Phagocytosis	Osmosis
5	The movement of molecules from an area of low concentration to an area of high concentration against the concentration gradient is known as :	Osmosis	Diffusion	Active transport	Phagocytosis	Active transport
6	Identify the process that requires energy in order to take place	Osmosis	Diffusion	Active transport	Phagocytosis	Active transport
7	Essential salts dissolved in body fluids are known as :	Phagocytes	Erythrocytes	Electrolytes	Podocytes	Electrolytes
8	Due to low atmospheric pressure, the rate of transpiration will	increase	decrease rapidly	decrease slowly	remain unaffected	increase
9	Guard cells help in	Transpiration	Protection against grazing	Fighting against infection	Guttation	Transpiration
10	The transpiration is regulated by the movements of	Subsidiary cells of the leaves	Guard cells of the stomata	Mesophyll tissue cells	Epidermal cells of the leaves	Guard cells of the stomata

SL.NO	QUESTION	OPTION 1	OPTION 2	OPTION 3	OPTION 4	ANSWER
Unit 1						
11	Living cells placed in isotonic solution (0.9% saline) retain their size and shape. This is based on the concept of	Facilitated diffusion	Diffusion	Osmosis	Transpiration	Osmosis
12	Passive absorption of water by the root system is the result of	Tension on the cell sap due to transpiration	Increased respiratory activity in root cells	Forces created in the cells of the root	Osmotic force in the shoot system	Tension on the cell sap due to transpiration
13	In which of the following plants, there will be no transpiration?	Plants growing in hilly regions	Aquatic, submerged plants	Plants living in deserts	Aquatic plants with floating leaves	Aquatic, submerged plants
14	Which one of the following theories for ascent of sap was proposed by eminent Indian scientist J. C Bose?	Pulsation theory	Transpiration Pull theory	Root pressure theory	Atmospheric pressure theory	Pulsation theory
15	Munch hypothesis is based on	Translocation of food due to TP gradient and	Translocation of food due to turgor pressure (TP)	Translocation of food due to imbibitions force	None of the above	Translocation of food due to turgor pressure (TP) gradient
16	The first process by which water enters into the seed coat when a seed is placed in a suitable environment for germination is	Osmosis	Active transport	absorption	Imbibition	Imbibition
17	Which of the following plant material, is an efficient water imbibant?	lignin	agar	pectin	cellulose	agar
18	Which of the following experiment is called physiological demonstration of osmosis?	Thistle funnel- whose mouth is tied with egg membrane	Thistle funnel- whose mouth is tied with parchment	bell jar experiment	potometer	Thistle funnel- whose mouth is tied with parchment paper
19	Water is lost in a liquid state in some plants through hydathodes. These hydathodes	Remain closed at night	Remain closed during day	Remain always open	Do not show any specificity in opening and closing	Remain always open
20	Passive absorption of water by the root system is the result of	Forces created in the cells of the root	Increased respiratory activity in root cells	Osmotic force in the shoot system	Tension on the cell sap due to transpiration	Tension on the cell sap due to transpiration
21	Which of the following helps in ascent of sap?	Root pressure	Transpiration	Capillarity	all of above	all of above
22	The translocation of organic solutes in sieve tube members is supported by	cytoplasmic streaming	P proteins	mass flow involving a carrier and ATP	root pressure and transpiration pull	P proteins

SL.NO	QUESTION	OPTION 1	OPTION 2	OPTION 3	OPTION 4	ANSWER
Unit 1						
23	Potometer works on the principle of	Osmotic pressure	Root pressure	Amount of water absorbed equals the amount	potential difference between the tip of the tube and that of the	Amount of water absorbed equals the amount transpired
24	The water readily available to plants for absorption by roots is	Gravitational water	Capillary water	Rain water	Hygroscopic water	Capillary water
25	The water potential of pure water at atmospheric pressure is	-2.3 bar	+2.3 bar	Zero bar	One bar	Zero bar
26	Loss of water from the stomata of leaves are known as	Guttation	Exudation	Transpiration	Evaporation	Transpiration
27	During rainy season wooden doors are difficult to open or closure because of	Plasmolysis	Imbibition	Osmosis	Diffusion	Imbibition
28	Plasmolysis occurs due to	Absorption	Osmosis	Endoosmosis	Exosmosis	Exosmosis
29	The marine animals that kept in fresh water burst. It shows the process of	Exosmosis	Endoosmosis	Plasmolysis	Deplasmolysis	Endoosmosis
30	Cooling of plants is caused by	Guttaion	Photorespiration	Transpiration	Assimilation	Transpiration
31	Active uptake of minerals by roots mainly depends on the	Availability of oxygen	Temperature	Light	Availability of CO	Availability of oxygen
32	The hormone which signals the closure of stomata is	Auxins	Cytokinin	Gibberelline	Abscisic acid	Abscisic acid
33	Water absorption takes place through	Lateral roots	Root cap	Root hairs	Tap root	Root hairs
34	Which of the following is an anti-transpirant	PMA	PAN	IAA	AUG	PMA

SL.NO	QUESTION	OPTION 1	OPTION 2	OPTION 3	OPTION 4	ANSWER
Unit 1						
35	What is the action spectrum of transpiration?	Orange and red	Green and ultraviolet	Blue and red	None of these	Blue and red
36	Which one of the following is used for measuring the rate of transpiration?	Porometer	Osmometer	Moll's experiment	Potometer	Potometer
37	Transpiration is least in	High atmospheric humidity	good soil moisture	high wind velocity	dry environment	High atmospheric humidity
38	Stomata open at night and close during day time in	Xerophytes	Mesophytes	Succulents	Hydrophytes	Succulents
39	The magnitude of diffusion pressure deficit in a non turgid cell is equal to	osmotic pressure-wall pressure	osmotic pressure+wall pressure	turgor pressure	osmotic pressure	osmotic pressure-wall pressure
40	DPD may be defined as the amount by which diffusion pressure of	a solution is lower than that of its solvent	a solution is higher than that of its solvent	a solvent is higher than that of its solutes	a solvent is lower than that of its solutes	a solution is lower than that of its solvent
41	Dehiscence of fruits and sporangia are due to	osmosis	diffusion	active transport	passive transport	osmosis
42	Flowers cut under water remain fresh for a longer time because	it has sufficient supply of water	water column is not blocked by air bubbles	transpiration is lowered	there is no vascular supply	water column is not blocked by air bubbles
43	The pulsatile theory is given by	J.C.Bose	Dixon	Strassburger	Sachs	J.C.Bose
44	Water available to plants is	runoff water	hygroscopic water	gravitational water	capillary water	capillary water
45	Which of the following is a rapid type of absorption?	passive absorption	active osmotic absorption	active non osmotic absorption	salt absorption	passive absorption
46	Wilting of plant occurs when	phloem is blocked	xylem is blocked	both xylem and phloem are blocked	few old roots are removed	xylem is blocked

SL.NO	QUESTION	OPTION 1	OPTION 2	OPTION 3	OPTION 4	ANSWER
Unit 1						
47	The following statements are true for active absorption except	It requires ATP	It occurs in plants where transpiration is less	It requires oxygen	Water absorbing force developed primarily in leaves	Water absorbing force developed primarily in leaves
48	The following statements are true for passive absorption except	It does not require ATP	Water absorbing forces developed primarily in leaves	It occurs in plants where transpiration is less	It takes place due to transpiration	It occurs in plants where transpiration is less
49	The rate of water absorption is maximum at	0 - 20°C	20 – 30°C	40 – 50°C	0 - 10°C	20 – 30°C
50	Vital force theory is proposed by	Godlewski	J.C.Bose	Unger	Sachs	J.C.Bose
51	Cohesion theory of Ascent of sap is proposed by	Dixon and Jolly	J.C.Bose	Unger	Sachs	Dixon and Jolly
52	The plastid found in both monocotyledonous and dicotyledonous plants are	amyloplasts	elaioplasts	proteinoplasts	chloroplasts	elaioplasts
53	The type of plastid found in the epidermal cells of <i>Helleborus</i> is	amyloplasts	elaioplasts	proteinoplasts	chloroplasts	proteinoplasts
54	The plastid found in the seeds of <i>Ricinus</i> is	amyloplasts	elaioplasts	proteinoplasts	chloroplasts	proteinoplasts
55	The plastid found in Brazil nut is	amyloplasts	elaioplasts	proteinoplasts	chloroplasts	proteinoplasts
56	The pigment present in chloroplast is	fucoxanthin	chlorophyll	phycoerythrin	phycocyanin	chlorophyll
57	The plastid found in dinoflagellates is	chloroplast	phaeoplast	amyloplast	proteinoplast	phaeoplast
58	The pigment present in phaeoplast is	fucoxanthin	chlorophyll	phycoerythrin	phycocyanin	fucoxanthin

SL.NO	QUESTION	OPTION 1	OPTION 2	OPTION 3	OPTION 4	ANSWER
Unit 1						
59	The pigment present in rhodoplast is	fucoxanthin	chlorophyll	phycoerythrin	phycocyanin	phycoerythrin
60	The plastid found in red algae is	chloroplast	rhodoplast	amyloplast	proteinoplast	rhodoplast



KARPAGAM ACADEMY OF HIGHER EDUCATION
(Deemed University Established Under Section 3 of UGC Act 1956)
Coimbatore - 641021.

(For the candidates admitted from 2015 onwards)

DEPARTMENT OF BIOCHEMISTRY

COURSE MATERIAL

STAFF NAME	:	Dr. L. HARIPRASATH	
SUBJECT	:	PLANT BIOCHEMISTRY	SUBJECT CODE : 15BCU504
SEMESTER	:	V	CLASS : III B.Sc. Biochemistry

UNIT-II

Photosynthesis: Photosynthetic pigments: Chlorophyll, carotenoids and phycobillin: Photosynthetic apparatus-structure and functions of chloroplasts: light absorption. Light reactions-Two kinds of chemical system-Photosystem I&II, evidences in support of light reaction. Hill's reaction; Emerson effect, cyclic and non-cyclic phosphorylation. Dark reaction : Calvin's cycle (C3 plants). Hatch-slack cycle (C4 plants) and CAM plants. Factors affecting photosynthesis. Photorespiration.

TEXT BOOKS

- Verma, S.K., & Mohit Verma. (2013). *A Text Book of Plant Physiology, Biochemistry and Biotechnology*. (6th ed.). New Delhi. S. Chand and Co.
- Bonner, J., & Varner, F.J. (2012). *Plant Biochemistry*. (3rd ed.). Burlington: Elsevier Science.
- Goodwin, T.W., & Mercer, E.I. (1990). *Introduction to Plant Biochemistry*. (2nd ed.). New York, NY: Robert Maxwell.M.C Publisher.

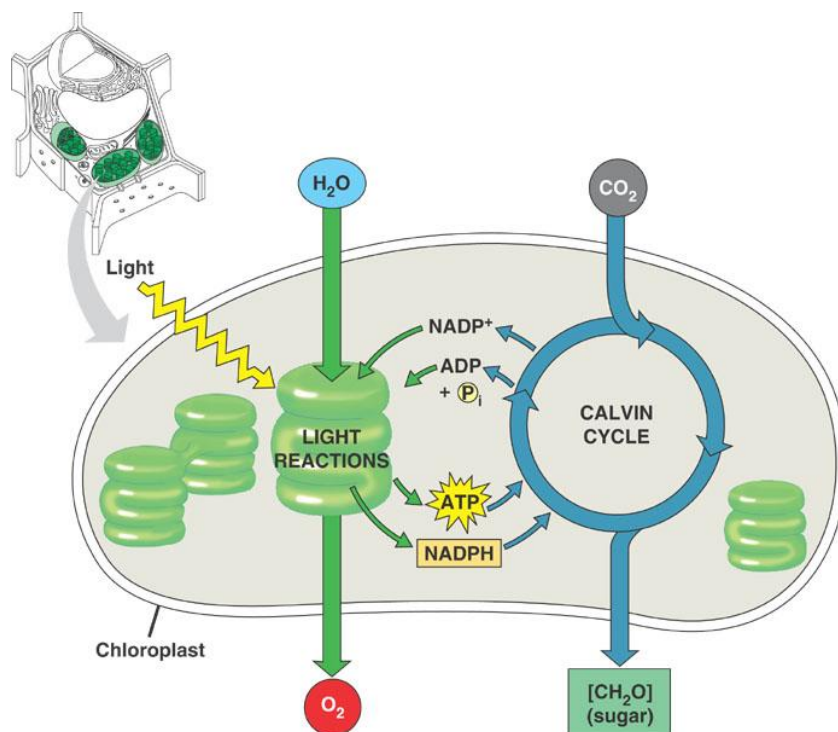
REFERENCES

- Buchanan, B.B. (2015). *Biochemistry and Molecular Biology of Plants*. New Jersey, NJ: John Wiley.
- Hans-Valter Heldt. (2005). *Plant Biochemistry and Molecular Biology*. England: Oxford University Press.
- Wink, M. (2010). *Functions and Biotechnology of Plant Secondary Metabolites*. (2nd ed.), London: Blackwell Publishing Ltd.,

Unit II

2. Photosynthesis

PHOTOSYNTHESIS-OVERVIEW



2.1 Photosynthetic pigments

2.1.1 Chlorophyll

Chlorophylls are greenish pigments which contain a porphyrin ring. This is a stable ring-shaped molecule around which electrons are free to migrate. Because the electrons move freely, the ring has the potential to gain or lose electrons easily, and thus the potential to provide energized electrons to other molecules. This is the fundamental process by which chlorophyll "captures" the energy of sunlight.

There are several kinds of chlorophyll, the most important being chlorophyll "a". This is the molecule which makes photosynthesis possible, by passing its energized electrons on to molecules which will manufacture sugars. All plants, algae, and cyanobacteria which photosynthesize contain chlorophyll "a". A second kind of chlorophyll is chlorophyll "b", which occurs only in "green algae" and in the plants. A third form of chlorophyll which is common is (not surprisingly) called chlorophyll "c", and is found only in the photosynthetic members of the Chromista as well as the dinoflagellates. The differences between the chlorophylls of these major groups were one of the first clues that they were not as closely related as previously thought.

2.1.2 Carotenoids

Carotenoids are usually red, orange, or yellow pigments, and include the familiar compound carotene, which gives carrots their color. These compounds are composed of two small six-carbon rings connected by a "chain" of carbon atoms. As a result, they do not dissolve in water, and must be

attached to membranes within the cell. Carotenoids cannot transfer sunlight energy directly to the photosynthetic pathway, but must pass their absorbed energy to chlorophyll. For this reason, they are called accessory pigments. One very visible accessory pigment is fucoxanthin the brown pigment which colors kelps and other brown algae as well as the diatoms.

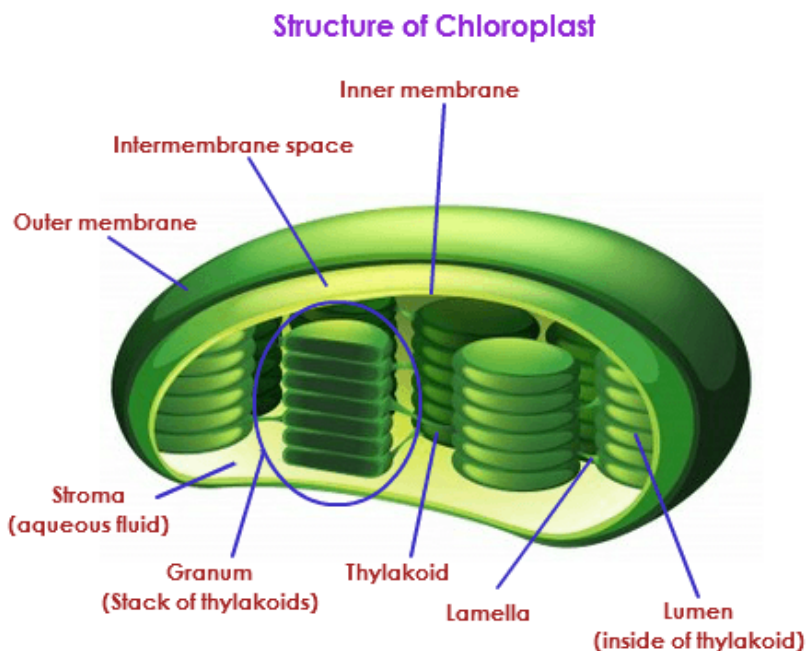
2.1.3 Phycobillin

Phycobilins are water-soluble pigments found in the stroma of chloroplast organelles that are present only in Cyanobacteria and Rhodophyta. The two classes of phycobilins include phycocyanin and phycoerythrin. Phycocyanin is a bluish pigment found in primarily cyanobacteria (blue-green algae) to aid in absorption of light in photosynthesis, while phycoerythrin is a pigment found in Rhodophyta (red algae) that is responsible for its characteristic red color. It is an accessory pigment that allows red algae to carry out photosynthesis in deep water where wavelengths of blue light are most abundant by absorbing blue light and reflecting red light.

2.2. Photosynthetic Apparatus

Chloroplast Structure

- Chloroplasts found in higher plants are generally biconvex or planoconvex shaped. In different plants chloroplasts have different shapes, they vary from spheroid, filamentous saucer-shaped, discoid or ovoid shaped.
- They are vesicular and have a colorless center. Some chloroplasts are in shape of club, they have a thin middle zone and the ends are filled with chlorophyll. In algae a single huge chloroplast is seen that appears as a network, a spiral band or a stellate plate.
- The size of the chloroplast also varies from species to species and it is constant for a given cell type. In higher plants, the average size of chloroplast is 4-6 μ m in diameter and 1-3 μ m in thickness.



The chloroplast are double membrane bound organelles and are the site of photosynthesis. The chloroplasts have a system of three membranes: the outer membrane, the inner membrane and the thylakoid system. The outer and the inner membrane of the chloroplast enclose a semi-gel-like fluid

known as the stroma. This stroma makes up much of the volume of the chloroplast, the thylakoids system floats in the stroma.

Outer membrane - It is a semi-porous membrane and is permeable to small molecules and ions, which diffuses easily. The outer membrane is not permeable to larger proteins.

Intermembrane Space - It is usually a thin intermembrane space about 10-20 nanometers and it is present between the outer and the inner membrane of the chloroplast.

Inner membrane - The inner membrane of the chloroplast forms a border to the stroma. It regulates passage of materials in and out of the chloroplast. In addition of regulation activity, the fatty acids, lipids and carotenoids are synthesized in the inner chloroplast membrane.

Stroma

Stroma is an alkaline, aqueous fluid which is protein rich and is present within the inner membrane of the chloroplast. The space outside the thylakoid space is called the stroma. The chloroplast DNA, chloroplast ribosomes and the thylakoid system, starch granules and many proteins are found floating around the stroma.

Thylakoid System

- The thylakoid system is suspended in the stroma. The thylakoid system is a collection of membranous sacks called thylakoids. The chlorophyll is found in the thylakoids and is the site for the process of light reactions of photosynthesis to happen. The thylakoids are arranged in stacks known as grana.
- Each granum contains around 10-20 thylakoids.
- Thylakoids are interconnected small sacks, the membranes of these thylakoids is the site for the light reactions of the photosynthesis to take place. The word 'thylakoid' is derived from the Greek word "thylakos" which means 'sack'.
- Important protein complexes which carry out light reaction of photosynthesis are embedded in the membranes of the thylakoids. The Photosystem I and the Photosystem II are complexes that harvest light with chlorophyll and carotenoids, they absorb the light energy and use it to energize the electrons.
- The molecules present in the thylakoid membrane use the electrons that are energized to pump hydrogen ions into the thylakoid space, this decreases the pH and becomes acidic in nature. A large protein complex known as the ATP synthase controls the concentration gradient of the hydrogen ions in the thylakoid space to generate ATP energy and the hydrogen ions flow back into the stroma.
- Thylakoids are of two types - granal thylakoids and stromal thylakoids. Granal thylakoids are arranged in the grana as pancake shaped circular discs, which are about 300-600 nanometers in diameter. The stromal thylakoids are in contact with the stroma and are in the form of helicoid sheets.
- The granal thylakoids contain only photosystem II protein complex, this allows them to stack tightly and form many granal layers with granal membrane. This structure increases stability and surface area for the capture of light.
- The photosystem I and ATP synthase protein complexes are present in the stroma. These protein complexes act as spacers between the sheets of stromal thylakoids.

2.2.2. Functions of chloroplast:

- In plants all the cells participate in plant immune response as they lack specialized immune cells. The chloroplasts with the nucleus and cell membrane and ER are the key organelles of pathogen defense.
- The most important function of chloroplast is to make food by the process of photosynthesis. Food is prepared in the form of sugars. During the process of photosynthesis sugar and oxygen are made using light energy, water, and carbon dioxide.
- Light reactions takes place on the membranes of the thylakoids.
- Chloroplasts, like the mitochondria use the potential energy of the H^+ ions or the hydrogen ion gradient to generate energy in the form of ATP.
- The dark reactions also known as the Calvin cycle takes place in the stroma of chloroplast.
- Production of $NADPH_2$ molecules and oxygen as a result of photolysis of water.
- BY the utilization of assimilatory powers the 6-carbon atom is broken into two molecules of phosphoglyceric acid.

2.3. Light reaction

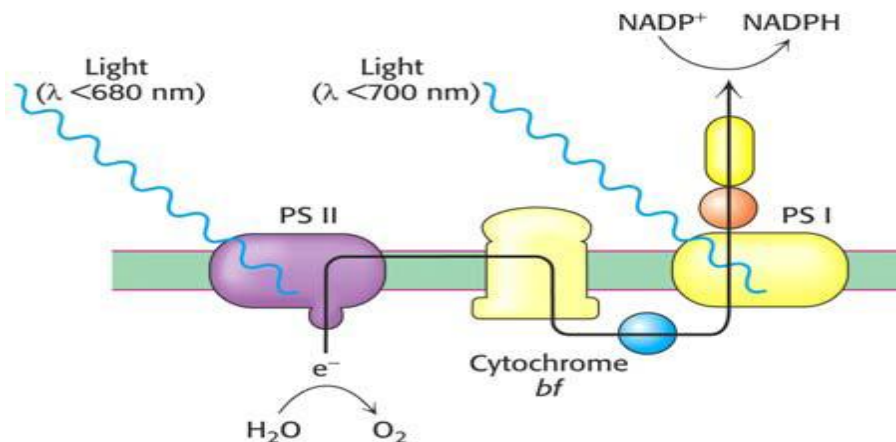
A photosynthetic reaction center is a complex of several proteins, pigments and other co-factors assembled together to execute the primary energy conversion reactions of photosynthesis.

The reaction center of photosystem I is P700

The reaction center of photosystem II is P680

2.3.1. Two kinds of chemical system-Photosystem I&II

Within the thylakoid membranes of the chloroplast, are two photosystems. Photosystem I optimally absorbs photons of a wavelength of 700 nm. Photosystem II optimally absorbs photons of a wavelength of 680 nm. The numbers indicate the order in which the photosystems were discovered, not the order of electron transfer. Under normal conditions electrons flow from PSII through cytochrome *bf* (a membrane bound protein analogous to Complex III of the mitochondrial electron transport chain) to PSI. Photosystem II uses light energy to oxidize two molecules of water into one molecule of molecular oxygen. The 4 electrons removed from the water molecules are transferred by an electron transport chain to ultimately reduce $2NADP^+$ to $2NADPH$. During the electron transport process a proton gradient is generated across the thylakoid membrane. This proton motive force is then used to drive the synthesis of ATP. This process requires PSI, PSII, cytochrome *bf*, ferredoxin- $NADP^+$ reductase and chloroplast ATP synthase.

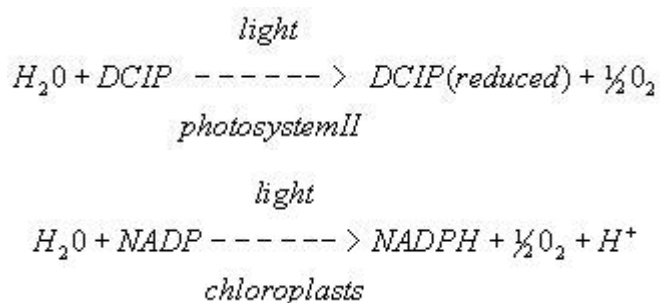


2.3.2. Evidences in support of light and dark reactions:

1. Evidence from intermittent light indicates that the rate of dark reaction is reduced due to continuous supply of light.
2. From temperature coefficient – It also indicates that light and dark reactions are although independent but are interlinked.
3. From CO₂ reduction in dark – It indicates that this phase is definitely a dark phase.

2.4 Hill's reaction

Photolysis of water and production of reducing power can be demonstrated by the Hill reaction. DCPIP can substitute for NADP, receiving electrons and becoming reduced.



In the presence of illuminated chloroplasts DCPIP becomes reduced. It does this by accepting excited electrons released from chlorophyll molecules. It may also accept hydrogen ions from the photolysis of water. We know this occurs due to a colour change. DCPIP is blue whereas reduced DCPIP is colourless.

In the Hill reaction the isolated chloroplasts in the experiment have to be suspended in an ice-cold buffered 2% sucrose solution

- The low temperature is to slow down enzyme activity
- 2% sucrose solution is to prevent water leaving entering or leaving the chloroplasts by osmosis

2.5 Emerson effect

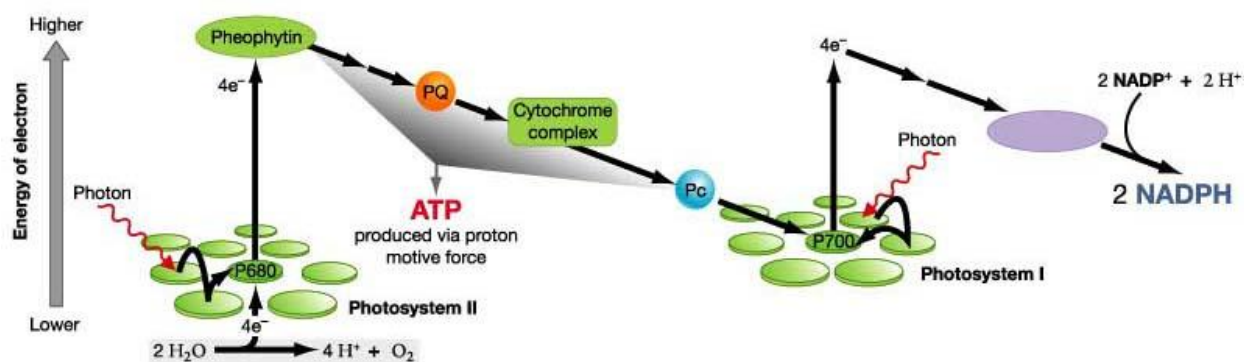
- The phenomenon of red drop shown by Emerson and Lewis was quite puzzling. This is not due to decrease in light absorption because the quantum yield measures only light that has actually been absorbed.
- This indicates that light of λ f. wavelengths greater than 680 nm is much less efficient than light of shorter wavelengths. In subsequent experiments, Emerson and his colleagues measured photosynthesis using red and far-red light after adjusting their fluence rates to give equal rates of photosynthesis.
- They observed that the quantum yield obtained using both red and far-red light simultaneously was much higher than the sum of the yields obtained with red and far-red light separately
- This phenomenon is known as Emerson enhancement effect or often as Emerson effect.
- These puzzling phenomena of red drop and enhancement effect led to the conclusion that two different reaction centers or photochemical events are involved in photosynthesis. One event is driven by red light (≈ 680 nm) and the other driven by far-red light (>680 nm). Optimal photosynthesis occurs when both events are driven simultaneously or in rapid succession. These two photochemical events are now known as Photosystem II and Photosystem I and they operate in series to carry out photosynthesis optimally. Photosystem II absorbs red light of 680 nm well and is driven very poorly by far-red light. On the other hand Photosystem I absorb preferentially far-red light of wavelengths greater than 680 nm.
- Electron transport actually begins with the arrival of excitation energy at the PS-II reaction centre chlorophyll, P680. The excited P680 (written as P680*) passes an electron to the pheophytin (Pheo), which is considered as the primary electron acceptor of PS-II. (Pheo is a form of chlorophyll-a in which the magnesium has been replaced by two hydrogen atoms) the result of this photo-oxidation event is the formation of P680 and Pheo⁺ (due to charge separation).
- The electron flows from Pheo through plastoquinone (PQ) to another multiprotein complex, cytochrome b_6/f complex. From this complex the electrons are picked up by a copper-binding protein, plastocyanin (PC).
- P680, formed by initial charge separation, is very strong oxidant and is able to extract electrons from water. The electron that reduces P680 is supplied by Y_Z which is called as the first electron donor of PS-II. Y_Z is a tyrosine residue in the D₁ protein of PS-II, which in turn receives the electron from a cluster of four manganese ions bound to a small complex of proteins called the oxygen-evolving complex (OEC). This complex is responsible for the splitting (oxidation) of water by drawing electrons from water with the evolution of molecular oxygen and the formation of O₂. Photolysis of water takes place as per the following equation:
- Light-driven charge separation similar to that involving PS-II reaction centre (P680) also takes place in the reaction centre of PS-I. The PS-I reaction centre chlorophyll, P700, is first excited to P700*, then photo-oxidized to P700⁺.
- The primary electron acceptor in PS-I is special chlorophyll-a molecule (A₀) which then passes the electron to ferredoxin. Ferredoxin subsequently is used to reduce XADP* to XADPH, a reaction mediated by the enzyme ferredoxin-NADP⁺-oxidoreductase. The oxidized P700* is reduced by withdrawing an electron from reduced plastocyanin.

- The complete photosynthetic electron transport chain in which there is a continuous flow of electrons starting from water to NADP⁺, passing through the two different photosystems and cytochrome b₆/f complex is shown schematically in. The scheme, in which the components of the photosynthetic electron transport chain are arranged as per their redox potential, is named as the Z-scheme because of its characteristic shape.

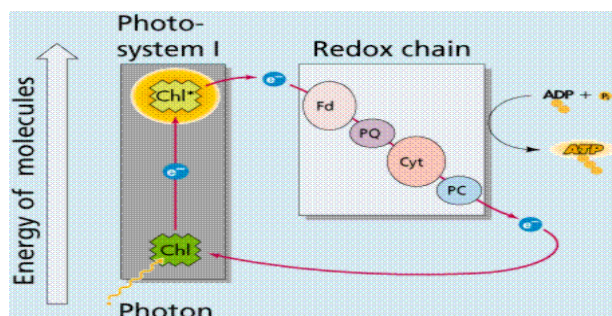
2.6. Cyclic and non cyclic photophosphorylation:

Photophosphorylation is the process of creating ATP using a Proton gradient created by the Energy gathered from sunlight. The process of creating the Proton gradient resembles that of the electron transport chain of Respiration. But since formation of this proton gradient is light- dependent, the process is called Photophosphorylation.

Noncyclic Photophosphorylation really refers to the ATP generated by Protons moved across the Thylakoid membranes during the Z-scheme. The Cytb₆-f complex acts as an electron transport chain. As the electrons lose Energy (during a series of re/dox reactions) Protons are moved into the Thylakoid space. This Proton gradient can be used to generate ATP chemiosmotically.

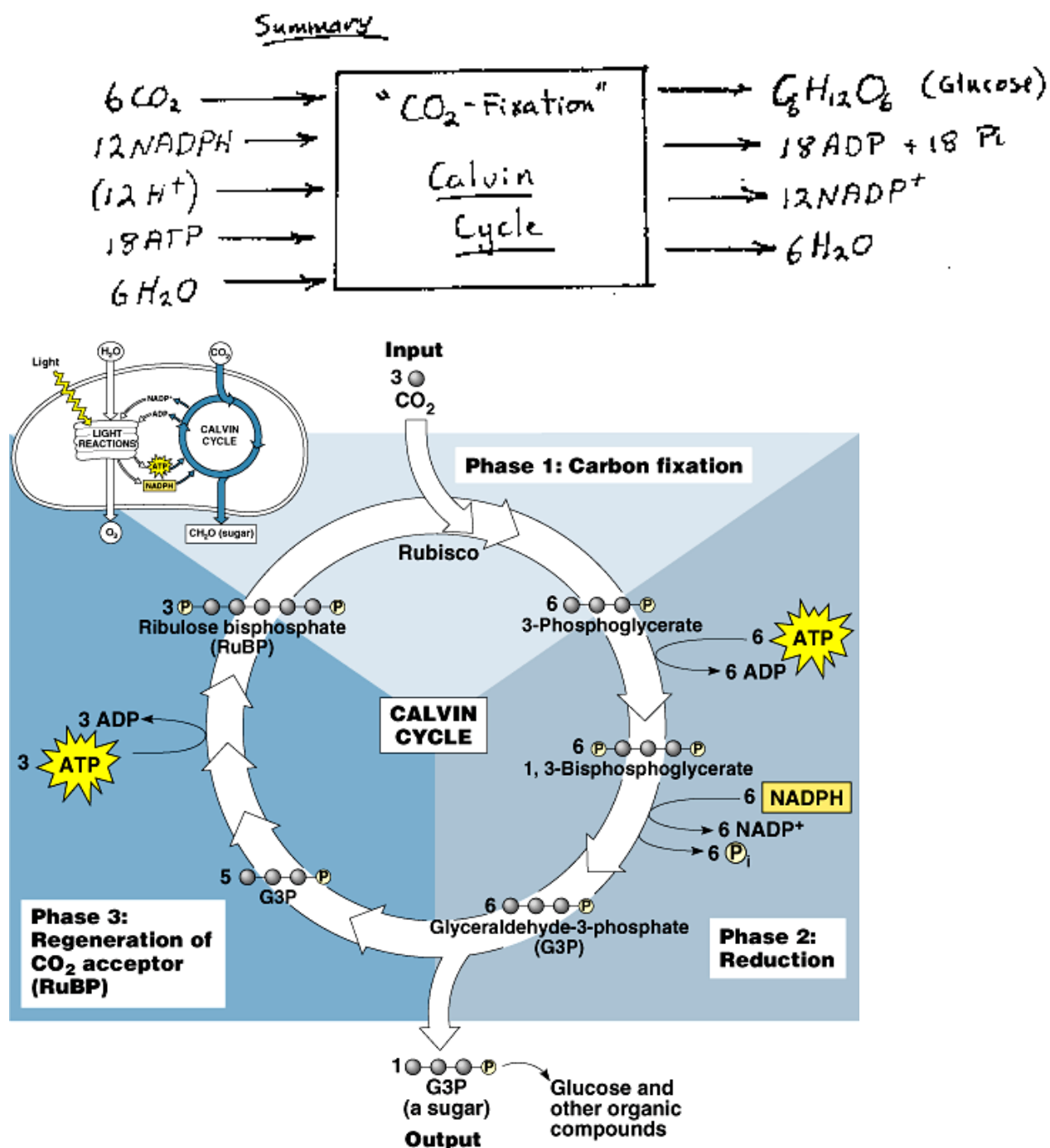


During Cyclic Photophosphorylation the electrons are recycled, hence the name cyclic photophosphorylation. The excited electrons resulting from the absorption of light in photosystem I are received by the primary electron acceptor and then transferred to the cytb₆-f complex which acts as an electron transport chain. The electrons return back to the reaction center of Photosystem I, where the cycle is ready to start all over. The electrons are used to translocate Protons which the ATPase uses to synthesize ATP. No reduction of NADP⁺ occurs in Cyclic Photophosphorylation.



2.7. The Light Independent Reactions-Dark reactions

2.7.1. The Calvin Cycle(C3 plants):



This part of photosynthesis occurs in the stroma of the chloroplasts called carbon dioxide fixation.

The fixation of the CO_2 is carried out by a giant enzyme ribulosebiphosphate carboxylase/oxidase (RUBISCO) which is the most abundant enzyme on earth. This enzyme is very

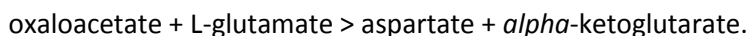
sluggish it works much slower than most other enzymes. (i.e. ~ 3 molecules of substrate per sec. compared with $\sim 1000/\text{sec}$ for others). Therefore, there are many copies of this enzyme in the stroma $\sim 50\%$ of chloroplast protein.

The first fixation reaction of the cycle uses a five carbon sugar ribulose 1-5 biphosphate and adds to it a CO_2 molecule to form 2 (3 carbon) molecules of 3 - phosphoglycerate. These are rearranged through a series of energy requiring reactions, using up ATP and NADPH to generate 2 molecules of glyceraldehyde 3 - phosphate. (If this were done six (6) times we now would have 12 molecules of glyceraldehyde - 3 - phosphate (G3P). Two (2) of the G3Ps are removed to make one glucose while the rest 10 G3Ps go back into the cycle to regenerate six (6) of the five (5) carbon sugars ribulose 1-5 biphosphate.

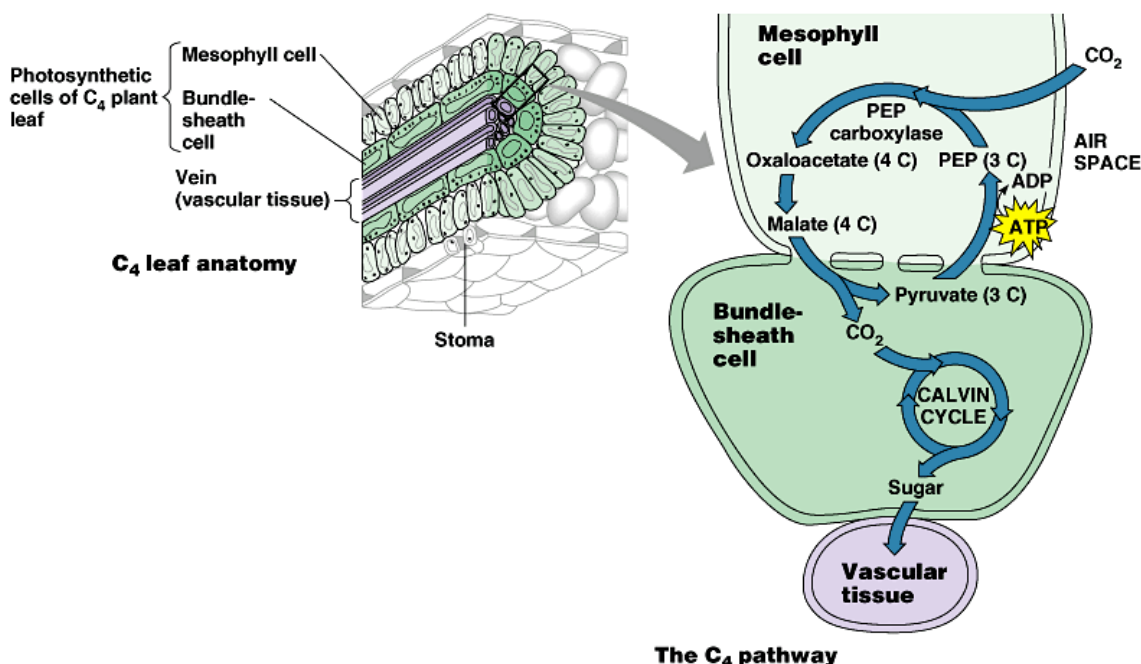
2.7.2. The HATCH- SLACK-cycle and "C4 Plants"

A number of plants display an increased and more efficient net photosynthesis during strong light intensities. A prime example are the Gramineae of warmer regions like maize or sugar-cane.

At the beginning of the sixties observed that the first product of photosynthesis in sugar-cane is not the C_3 unit 3-phosphoglycerate but a unit with four C-atoms. The Australian plant physiologist M. D. HATCH and his English colleague C. R. SLACK confirmed this result and identified the compound as oxaloacetate (OAA). It is produced by the addition of one molecule of carbon dioxide to phosphoenolpyruvate (PEP). The cycle is also known as the HATCH- SLACK-cycle or the C_4 cycle. Plants with this cycle are called C_4 -plants (and CAM plants, respectively) in contrast to C_3 plants where the carbon dioxide is directly fed into the CALVIN cycle. The oxaloacetate is usually converted into malate of which the carbon dioxide is split off again with the help of an enzyme. This carbon dioxide is now bound by ribulose-1,5-diphosphate and assimilated *via* the CALVIN cycle. : Some species use malate instead of aspartate

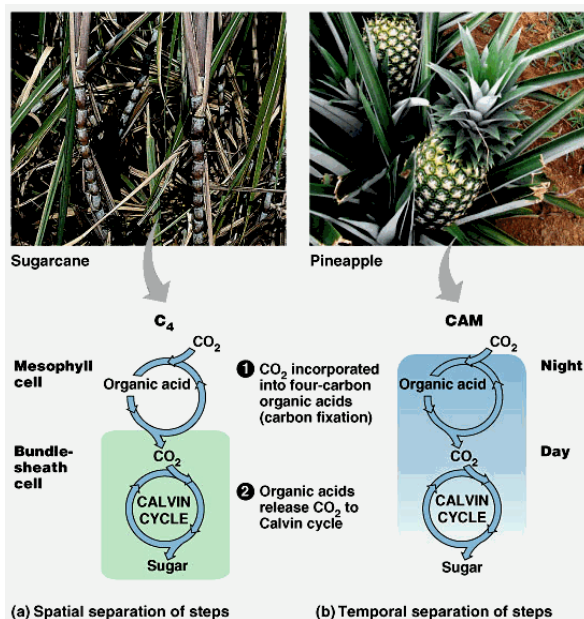


The reversible binding of carbon dioxide has the function to accumulate and store CO_2 . The process consumes energy, so that it could also be spoken of a carbon dioxide pump. It should be mentioned that the HATCH-SLACK cycle requires two molecules of ATP are per fixed carbon dioxide.



2.7.3. Crassulacean Acid Metabolism (CAM)

The ultimate prevention of CO_2 loss is found in desert plants like cactus. CAM In these plants the stomata are open at night. The plant fixes CO_2 into C_4 carbon compounds during the night, then transfers the carbon to the Calvin cycle during the daylight hours while the stomata are completely closed therefore reducing H_2O loss. This is all done in the same cell.



C₃, C₄ and CAM. Regulation of The Activity of Photosynthesis

Photosynthesis of C_4 plants. CO_2 is bound to phosphoenolpyruvate (PEP) in mesophyll cells. The product is oxaloacetate. The next step generates malate. In the cells of the vascular bundle sheath, the 'Kranz' cells, is carbon dioxide split off the malate and fed into the CALVIN cycle. The pyruvate is transported back into the mesophyll cells (active transport) and is with the help of additional ATP phosphorylated to PEP.

The anatomy of C_4 leaves with so-called 'Kranz' cells differs fundamentally from that of C_3 plants. The chloroplasts of C_3 plants are of homogeneous structure, while two types of chloroplasts occur in C_4 plants. The mesophyll cells contain normal chloroplasts, that of the vascular bundle sheath have chloroplasts without grana, i.e. they are partially impaired in function. This peculiarity does not affect the CALVIN cycle, it concerns only the light reactions of photosynthesis. The first binding of carbon dioxide (the HATCH-SLACK reaction) occurs in the mesophyll cells, the incorporation into carbohydrates (the CALVIN cycle) in the cells of the vascular bundle sheath. Both processes of photosynthesis are spatially separated.

The Crassulacean Acid Metabolism (CAM)

CAM is the abbreviation of Crassulacean acid metabolism. The name points at the fact that this pathway occurs mainly in Crassulacean species (and other succulent plants). The chemical reaction of the carbon dioxide accumulation is similar to that of C_4 plants but here are carbon dioxide fixation and its assimilation not separated spatially but in time. CAM plants occur mainly in arid regions. The opening of the stomata to take up carbon dioxide is always connected with large losses of water. To inhibit this loss during intense sun (the transpiration *via* the cuticle remains intact) has a mechanism developed that allows the uptake of carbon dioxide during the night. The prefixed carbon dioxide is stored in the vacuoles as malate (and isocitrate) and is used during the daytime for photosynthesis.

Which Metabolism Goes With Which Conditions?

The enzyme that catalyzes the primary carbon dioxide fixation of C_4 and CAM plants is phosphoenolpyruvate carboxylase (PEPC). Its affinity for carbon dioxide is by far higher than that of Rubisco, the first enzyme of the CALVIN cycle. As a consequence are C_4 plants able to use even trace amounts of carbon dioxide. PEPC occurs in small amounts (roughly 2 - 3 %) also in C_3 plants, where it, too, has a key position in the metabolic regulation.

CAM: Advantages and Disadvantages.

CAM has been detected in more than 1000 angiosperms of 17 different families. It is usually accompanied by succulence, though not all Crassulaceae, for example, display CAM and succulence is no precondition of CAM. *Tillandsia usneoides* of the bromelia family is not succulent, but uses CAM. *Mesembryanthemum crystallinum* (a plant with succulent leaves) can use the C_3 pathway but switches to CAM when growing in saline soils. Under experimental conditions can the shift be achieved by increasing the NaCl concentration of the nutrient medium. While the advantage of C_4 plants comes in useful under high light intensities, is the degree of the CAM influence in CAM plants regulated mainly by temperature, atmospheric humidity and salinity. Both strong and weak CAM plants are known. In weak CAM plants becomes CAM only apparent at certain differences between day and night temperatures. CAM plants that store a lot of malate and due to the thus high osmotic value also a lot of water, are usually less frost resistant than C_3 plants. Because of the high concentration of acid are they less heat resistant, too. Species of arid regions are therefore forced to break their pool of malate down during the daytime (R. LÖSCH and H. KAPPEN, Universität Kiel, 1985). Usually do the C_4 pathway and CAM exclude each other. An exception is the succulent C_4 dicotyledon *Portulaca oleracea* that is able to choose the optimal pathway under natural conditions.

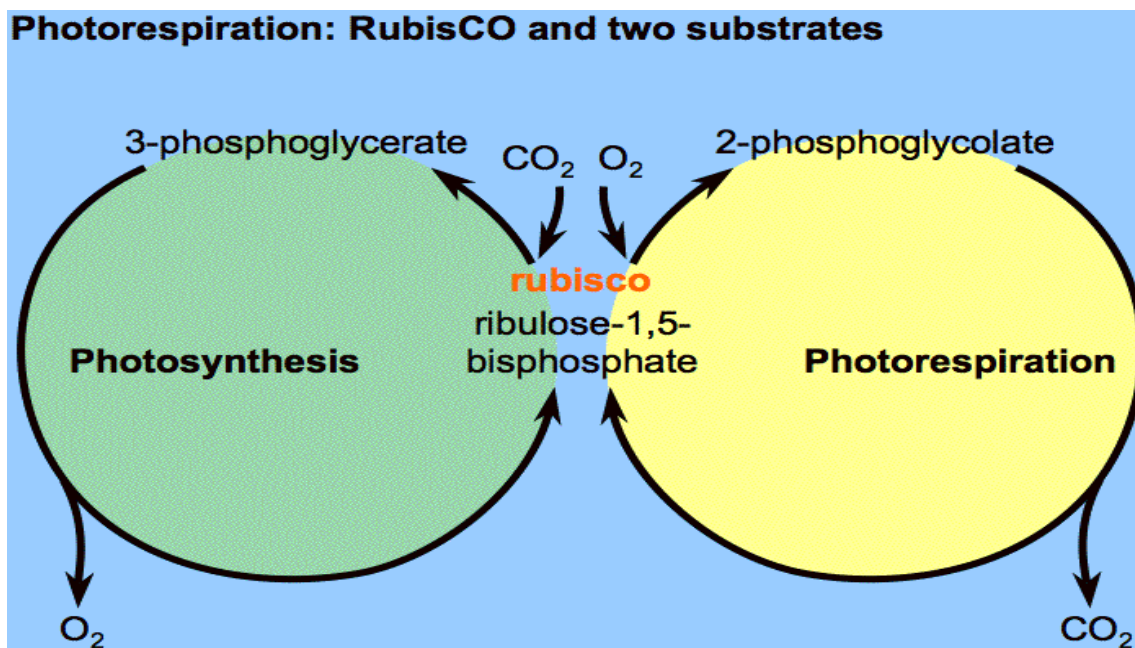
2.8. Factors affecting photosynthesis

By understanding the factors that affect photosynthesis, one can better understand the rate of the process.

- **Light:** Without light, a plant cannot photosynthesize very quickly, regardless of whether there are water and CO_2 or not. But overdoing light is also not a good idea. In nature, balance is crucial. But increasing the intensity of light to a prudent limit will speed up the process.
- **Carbon Dioxide:** It happens to be the major limiting factor. The problem arises because the concentration of carbon dioxide in the air is less. Even if there is plenty of light, the plant cannot photosynthesize in the absence of sufficient amount of carbon dioxide.
- **Temperature:** The plants are affected lesser by temperature in comparison to light and CO_2 . Nevertheless, if the temperature is too hot or too cold, the rate of photosynthesis is adversely affected. C_4 plants have an affinity towards higher temperatures while C_3 have a much lower optimum temperature.
- **Water:** It makes its presence felt more through its effect on the plant rather than directly on photosynthesis. It is found that slight deficiency of water shows a considerable reduction in plant yield.

As is obvious each of these factors is interrelated and a fine balance is crucial.

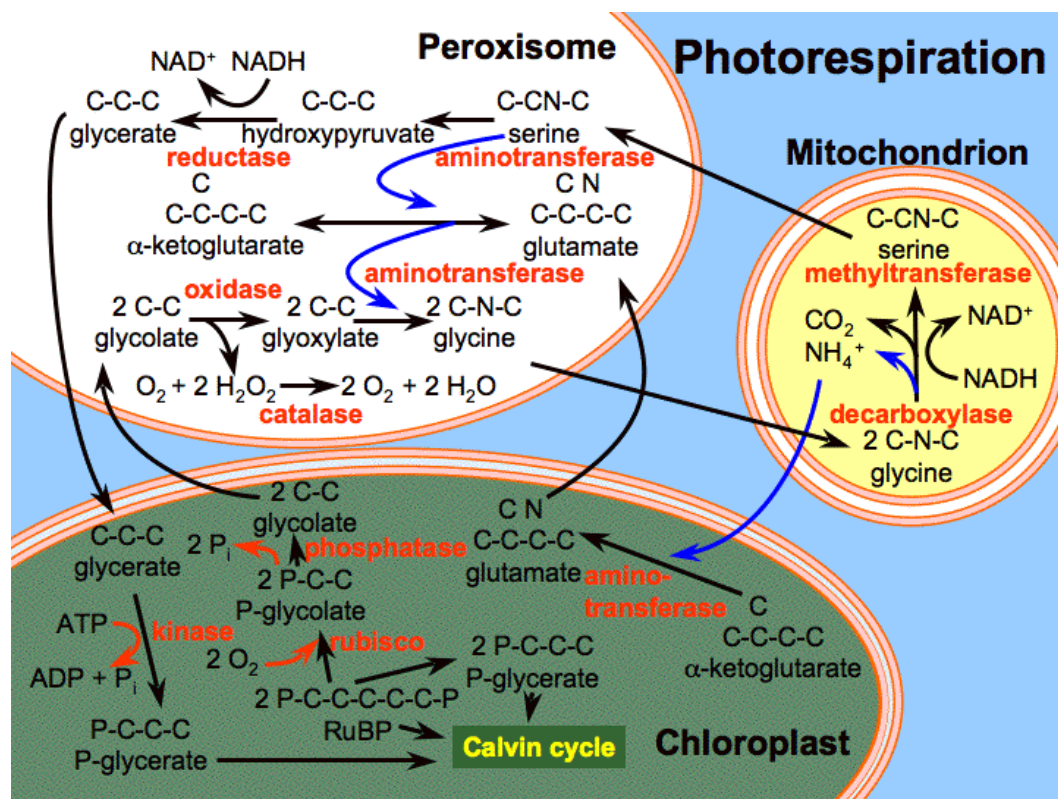
Photorespiration



The enzyme, rubisco, not only initiates carbon fixation in the Calvin cycle; it also combines with oxygen to initiate photorespiration. As its name suggests (rubsico) the enzyme is both a carboxylase and an oxygenase. The active site of rubisco cannot distinguish the two similar substrates: $\text{O}=\text{C}=\text{O}$ and $\text{O}=\text{O}$. As we shall see, the two reactions catalyzed by the same enzyme are diametrically opposed to each other.

Each reaction pathway undoes the other, and both reactions can operate in a cell simultaneously depending upon the environmental conditions. As both substrates combine with the active site of rubisco, they are competitive inhibitors of each other's reactions. One might recall our earlier discussions about competitive inhibition. The relative concentration of the two substrates and the differential affinity of the enzyme for each substrate will determine which of the reactions (Calvin cycle or Photorespiration) predominate. Fortunately for plants (and for us indirectly!) rubisco has an affinity for carbon dioxide that is 80 times higher than its affinity for oxygen. However, the relatively low ratio of CO_2 to O_2 of mesophyll fluids in contact with air (0.04) means that, in a typical plant, the Calvin cycle only occurs about three times faster than photorespiration. Temperature also influences the relative rates of photorespiration and the Calvin cycle. Because increased temperature more efficiently removes carbon-dioxide from solution than it does oxygen, high temperatures favor photorespiration.

The photorespiration pathway is an enzymatic one that is not coupled to any electron transfer system. It does not generate ATP. It does use oxygen and it does produce carbon dioxide, and it uses a sugar-phosphate as its primary fuel. The complete pathway is depicted here.



It is worthy to note that this diagram, as others of its type, show the organelles tightly oppressed to each other. Indeed there are some famous electron micrographs (example above) that show this, but other micrographs do not show them this way. I say this just to comment that this positioning may be more an efficient design for communication to students than a realistic portrayal of life in a typical cell.

In the chloroplast, rubisco, combines with ribulose-1,5-bisphosphate (RuBP) and oxygen. The five-carbon RuBP is split into the two-carbon 2-phosphoglycolate and the three-carbon 3-phosphoglycerate (PGA). The enzymes of this pathway are enumerated in the diagram above.

The 2-phosphoglycolate is converted to glycolate by phosphoglycolate phosphatase in the chloroplast. The phosphate liberated is returned to the local phosphate pool. The glycolate is transported from the chloroplast into a nearby peroxisome.

In the peroxisome, the glycolate is oxidized by oxygen gas to glyoxylate and hydrogen peroxide by glycolate oxidase. The peroxide is converted to water and oxygen gas by catalase. So the consumption of oxygen in the oxidation is replaced by catalase activity in the peroxisome.

The glyoxylate is converted to the amino acid glycine in the peroxisome. The amino group is transferred to the glyoxylate from glutamate (another amino acid) by glyoxylate:glutamate aminotransferase. The glutamate is converted to α-ketoglutarate (we will remember and come back to that later!). The glycine is transported to the mitochondrion.

In the mitochondrion, glycine decarboxylase carves off carbon dioxide gas from the glycine. This requires NAD⁺ to park the hydrogen atom. It also cleaves off the amino group. If you are paying attention to the chemical structures, you realize that the two-carbon amino acid has had both its amino and acid groups removed! There is only one carbon left! This methylene group is parked on a folate molecule in the mitochondrion.

When a second glycine arrives into the mitochondrion from the peroxisome, it combines with the methylene-folate to release the three-carbon amino acid serine through the action of serine hydroxymethyltransferase. Also released for re-use by this enzyme reaction is the folate. The serine is transported to the peroxisome.

In the peroxisome, the serine loses its amino group to α -ketoglutarate (remember, we would get back to that!) to regenerate the glutamate required in an earlier step in the pathway. This amino-transfer is accomplished by serine aminotransferase. In this reaction the serine is converted to hydroxypyruvate.

The peroxisome reduces the hydroxypyruvate to glycerate by hydroxypyruvate reductase. The reducing power for this comes from NADH; if you recall this was produced in an earlier step in the mitochondrion. The glycerate is transported to the chloroplast.

In the chloroplast, the glycerate is converted by glycerate kinase to 3-phosphoglycerate. The phosphate comes from ATP. Instead of producing ATP, photorespiration uses ATP. The 3-phosphoglycerate from the beginning and this new one from the end of photorespiration enter the chloroplast pool of PGA that is used to regenerate RuBP.

Photorespiration loses 25% of the carbon it takes from the Calvin cycle

The photorespiration pathway siphons carbon away from the Calvin cycle, but it also returns some of what it takes. Because it takes two glycines in photorespiration to complete the pathway, two glycolates must be taken from the Calvin cycle. Of these four carbons taken, one is lost as carbon dioxide and three are returned to the Calvin cycle. This 25% loss of carbon is going to give measurement errors for photosynthesis in whole-cells or leaves. We are probably under-estimating the photosynthetic rate by 25%.

Rubisco evolution

The wastefulness of photorespiration is probably a consequence of just two factors. Early in the evolution of photosynthesis there was a higher carbon dioxide to oxygen gas ratio in the ancient atmosphere. Indeed the atmosphere was likely anaerobic in the earliest times on Earth. Rubisco evolved its active site when oxygen was rare and carbon dioxide was common. Since ancient times, rubisco has not yet evolved a mechanism to discriminate between the two similar substrates ($\text{O}=\text{C}=\text{O}$ and $\text{O}=\text{O}$). The reactions are similar too; the substrate is attached at a point along RuBP resulting in its splitting into organo-monophosphates. 3-phosphoglycerate is a common product of both reactions. So the difficulty of a protein to distinguish such similar molecules and to catalyze one reaction but not the other just has not happened yet. Photorespiration losses have not been intolerable either; the selection pressure is probably not severe in most environments. But in hot, dry, heavily-populated environments where plants effectively reduce the local carbon-dioxide content of the atmosphere, selection should have resulted in a few adaptations to overcome photorespiration.

KARPAGAM ACADEMY OF HIGHER EDUCATION
COIMBATORE - 21
DEPARTMENT OF BIOCHEMISTRY
III B.Sc BIOCHEMISTRY
BATCH: 2015 - 2018

15BCU504 - Plant Biochemistry

PART A (20 X 1 = 20 MARKS) - Online MCQ questions

SL.NO	QUESTION	OPTION 1	OPTION 2	OPTION 3	OPTION 4	ANSWER
Unit 2						
1	Both Photo systems I and II are involved in	Cyclic photophosphorylation	Non-cyclic Photophosphorylation	Pseudophotophosphorylation	photorespiration	Non-cyclic Photophosphorylation
2	Chlorophyll a and b can be excited to the second excited electronic states by wavelength	650-700nm	420-460	700-740nm	250-300nm	650-700nm
3	The reaction center of Photo system I is	P680	P700	P800	P750	P700
4	Which of the following chlorophylls is found in all plants and cyanobacteria?	Chl a	Chl b	Chl c1	Chl c2	Chl b
5	Chlorophyll b differs from chlorophyll a	in having a formyl group on ring II	in having a formyl group on ring I	in having a formyl group on ring III	in having a methyl group on ring II	in having a formyl group on ring II
6	The source of oxygen evolved during photosynthesis is	water	CO ₂	Organic acid	glucose	water
7	Photosynthetic enhancement is referred as	Red drop	Quantum yield	Emerson effect	Hills reaction	Emerson effect
8	The central atom that is covalently and coordinately bound to the N- atom of tetrapyrrole system of chlorophyll is	Mg ⁺⁺	Fe ⁺⁺	Fe ³⁺	K ⁺	Mg ⁺⁺
9	Chlorophyll is found in	Plasma membrane	Thylakoid membrane	Peroxisomes	Glyoxysomes	Thylakoid membrane
10	One of the following is not a photo synthetic pigment	Chlorophyll	Carotenoid	Phycobilin	xanthin	xanthin
11	The Chlorophylls absorb light of wavelength	650 – 700nm	450 – 500nm	500 – 600 nm	350-400 nm	650 – 700nm
12	The Carotenoids absorb light in the wavelength	450 – 500nm	650 – 700nm	500 – 600nm	350-400nm	450 – 500nm
13	The Phycobilins absorb light in the wavelength	500 – 600nm	450 – 500nm	650 – 700nm	~ 450nm	500 – 600nm
14	The most potent reagents which block non-cyclic photophosphorylation in green plants and algae are	CMU & DCMU	Octyl guanidine & grameodine	urea	cyanogen bromide	CMU & DCMU
15	The primary electron donar in PS II is	P700	Cyt C	P680	Chl a	P680
16	In cyclic photo phosphorylation	Only PS I is functional	Both PSI and PSII are functional	O ₂ is evolved	NADPH ₂ is formed	Only PS I is functional
17	The main function of light harvesting complex is to	Increase the temperature	Capture solar energy	Enhance ATP formation	transfer of electrons	Capture solar energy
18	The skeleton of chlorophyll is made of a	pentanone	Tetrapyrrole	Per hydro phenanthrene	Hexagon	Tetrapyrrole
19	The following are C ₄ plants except	Chlorella	Sugarcane	maize	sorghum	Chlorella

SL.NO	QUESTION	OPTION 1	OPTION 2	OPTION 3	OPTION 4	ANSWER
20	Which of the following is the substrate for photorespiration	Oxaloacetate	PEP	succinate	glycollate	PEP
21	The stomata of CAM plants remain	Opened during the day and night	closed during day and opened at night	closed during the day and night	opened during the day and closed at night	closed during day and opened at night
22	The following organelles are involved in photo respiration except	mitochondria	chloroplasts	peroxisomes	golgi bodies	golgi bodies
23	Kranz anatomy is a special feature in the leaves of	C3 plants	C4 plants	C2 plants	CAM plants	C4 plants
24	In C ₃ plants the primary CO ₂ acceptor is -----	ribulose 1,5 bis phosphate	Ribulose-5-phosphate	ribulose-1-phosphate	ribose -1-phosphate	ribulose 1,5 bis phosphate
25	The sequence of dark reaction in photosynthesis was established by	A.A Bensen	J.Bassham	Melvin Calvin	J.C.Bose	Melvin Calvin
26	The Benson – Calvin cycle takes place in	Chloroplasts	etioplasts	Mitochondria	cytoplasm	Chloroplasts
27	Sugarcane & cynodon dactylon are	C4 plants	C3 Plants	C2 plants	CAM plants	C4 plants
28	The primary acceptor of CO ₂ in C ₄ plant is	PEP	Pyruvate	Alanine	oxaloacetate	PEP
29	In CAM plants CO ₂ assimilation occurs during	Day	Night	Day & Night	Evening	Night
30	In C ₄ plants Pyruvate, Phosphate dikinase is located mainly in the	mesophyll cells of chloroplast	bundle sheath cells of chloroplast	mitochondria	peroxisomes	mesophyll cells of chloroplast
31	Plants in which the Hatch slack pathway takes place are called as	C2 plants	C3 plants	C4 plants	CAM plants	C4 plants
32	In bundle sheath cells, malate is decarboxylated to form	oxaloacetate	citrate	pyruvate	PEP	pyruvate
33	The calvin cycle enzymes are present in	Stroma	Thylakoid lumen	Grana	Thylakoid membrane	Stroma
34	During photophosphorylation the NADPH and ATP are	Absorbed	Released	Reduced	Oxidised	Released
35	Thylakoids of grana possess the	enzymes of calvin cycle	enzymes of photophosphorylation	enzymes for C3 cycle	enzymes of C4 cycle	enzymes of photophosphorylation
36	Hydrogen peroxide is formed during	Calvin cycle	Hatch-Slack cycle	CAM cycle	photorespiration	photorespiration
37	ATP molecules are synthesized in all except	cyclic photophosphorylation	non cyclic photophosphorylation	dark respiration	photorespiration	photorespiration
38	The optimum temperature of photorespiration is	10 - 20°C	25 - 35°C	40 - 60°C	35 - 45°C	25 - 35°C
39	The nocturnal opening of stomata is the characteristic feature of	water plants	C4 plants	CAM plants	C3 plants	CAM plants
40	The first stable compound formed in C ₃ cycle is	DHAP	phosphoglyceric acid	oxalo acetic acid	glycolic acid	phosphoglyceric acid
41	The first stable compound formed in C ₄ cycle is	DHAP	phosphoglyceric acid	oxalo acetic acid	glycolic acid	oxalo acetic acid
42	The optimum temperature for the growth of C ₄ plants is	30 - 45°C	25 - 35°C	0 - 20°C	40 - 60°C	30 - 45°C

SL.NO	QUESTION	OPTION 1	OPTION 2	OPTION 3	OPTION 4	ANSWER
43	For the synthesis of each molecule of glucose from CO ₂ in a photosynthesis how many ATP molecules are required?	18	12	3	2	18
44	For the synthesis of each molecule of glucose from CO ₂ in a photosynthesis how many NADPH molecules are utilized	18	12	3	2	12
45	Number of ATP molecules synthesized in non cyclic photophosphorylation is	2	3	1	4	1
46	Number of ATP molecules synthesized in cyclic photophosphorylation is	2	3	1	4	2
47	The following statement is true with cyclic photo phosphorylation	PS I is involved	PS II is involved	photo oxidation of water takes place	NADP ⁺ is reduced to NADPH + H ⁺	PS I is involved
48	Carotenes are usually found in	PS I	PSII	Both PS I and PSII	neither PS I nor PSII	PS I
49	Xanthophylls are usually found in	PS I	PSII	Both PS I and PSII	neither PS I nor PSII	PSII
50	In C ₄ cycle oxalo acetic acid is converted to malic acid by the enzyme	malic dehydrogenase	malate decarboxylase	malic oxidase	transaminase	malic dehydrogenase
51	In C ₄ plants malic enzyme is present in	mesophyll cells	bundle sheath cells	xylem	phloem	bundle sheath cells
52	Photosynthetic yield is more in	C4 plants	C3 plants	C2 plants	CAM plants	C4 plants
53	CAM cycle is observed in all the plants except	cactus	orchid	chlorella	bryophyllum	chlorella
54	In C ₄ cycle the enzyme involved in conversion of oxalo acetic acid to malic acid is	malic enzyme	malate dehydrogenase	transaminase	malate decarboxylase	malate dehydrogenase
55	Only mesophyll cells are involved in	C3 cycle	C4 cycle	CAM cycle	C2 cycle	C3 cycle
56	In CAM plants carbohydrate synthesis takes place during	night time	day time	both night and day time	only in the evening	day time
57	Which of the following is formed during photo respiration	H ₂ O ₂	O ₂	OH ⁻ ions	O ₃	H ₂ O ₂
58	In photo respiration glyoxylic acid is converted to glycine by	transaminase	decarboxylase	dehydrogenase	reductase	transaminase
59	In photo respiration the enzyme involved in detoxification of H ₂ O ₂ is	catalase	decarboxylase	transaminase	dehydrogenase	catalase
60	When the atmospheric CO ₂ is less than 1% ribulose di phosphate is converted to	3 phospho glyceric acid	glycolic acid	glyoxylic acid	glycine	glycolic acid



KARPAGAM ACADEMY OF HIGHER EDUCATION
(Deemed University Established Under Section 3 of UGC Act 1956)
Coimbatore - 641021.

(For the candidates admitted from 2015 onwards)

DEPARTMENT OF BIOCHEMISTRY

COURSE MATERIAL

STAFF NAME	:	Dr. L. HARIPRASATH	
SUBJECT	:	PLANT BIOCHEMISTRY	SUBJECT CODE : 15BCU504
SEMESTER	:	V	CLASS : III B.Sc. Biochemistry

UNIT-III

Cycles of elements and Plant Nutrition: Nitrogen cycle-Ammonification, nitrification, nitrate reduction and denitrification. Nitrogen fixation: Symbiotic and non-symbiotic nitrogen fixation. Sulfur cycle: Release of sulfur from organic compounds; Oxidation of sulfur compounds; Reduction of sulfate. Phosphorus cycle and carbon cycle. Plant nutrition: Specific roles of essential elements and their deficiency symptoms in plants. Macronutrients: Carbon, Hydrogen, Oxygen, Nitrogen, Sulfur, Phosphorus, Calcium and Potassium. Micronutrients: Manganese, Boron, Copper, Zinc, Molybdenum and Chlorine.

TEXT BOOKS

Verma, S.K., & Mohit Verma. (2013). *A Text Book of Plant Physiology, Biochemistry and Biotechnology*. (6th ed.). New Delhi. S. Chand and Co.

Bonner, J., & Varner, F.J. (2012). *Plant Biochemistry*. (3rd ed.). Burlington: Elsevier Science.

Goodwin, T.W., & Mercer, E.I. (1990). *Introduction to Plant Biochemistry*. (2nd ed.). New York, NY: Robert Maxwell.M.C Publisher.

REFERENCES

Buchanan, B.B. (2015). *Biochemistry and Molecular Biology of Plants*. New Jersey, NJ: John Wiley.

Hans-Valter Heldt. (2005). *Plant Biochemistry and Molecular Biology*. England: Oxford University Press.

Wink, M. (2010). *Functions and Biotechnology of Plant Secondary Metabolites*. (2nd ed.), London: Blackwell Publishing Ltd.,

Unit III

3. Cycles of elements and Plant Nutrition

3.1. Nitrogen cycle

- All life requires nitrogen-compounds, e.g., proteins and nucleic acids.
- Air, which is 79% nitrogen gas (N_2), is the major reservoir of nitrogen.
- But most organisms cannot use nitrogen in this form.
- Plants must secure their nitrogen in "fixed" form, i.e., incorporated in compounds such as:
 - nitrate ions (NO_3^-)
 - ammonium ions (NH_4^+)
 - urea ($(NH_2)_2CO$)
- Animals secure their nitrogen (and all other) compounds from plants (or animals that have fed on plants)

3.1.1. Ammonification

During ammonification, the plant or animal that you are a part of dies. You are left to be converted back into ammonium by decomposers (bacteria and fungi that break down dead organisms). You are returned back into the soil and can then reenter the cycle. You can go through nitrification (step two, the previous step) again to be used by more plants and animals, or you can go through the next step, denitrification.

Proteolysis:

Plants use the ammonia produced by symbiotic and non-symbiotic Nitrogen fixation to make their amino acids & eventually plant proteins. Animals eat the plants and convert plant proteins into animal proteins. Upon death, plant and animals undergo microbial decay in the soil and the nitrogen contained in their proteins is released. Thus, the process of enzymatic breakdown of proteins by the microorganisms with the help of proteolysis enzymes is known as "proteolysis".

The breakdown of proteins is completed in two stages. In first stage proteins are converted into peptides or polypeptides by enzyme "proteinases" and in the second stage polypeptides / peptides are further broken down into amino acids by the enzyme "peptidases".

Proteins	----->	Peptides	----->	Amino	Acids
	Proteinases		Peptidases		

The amino acids produced may be utilized by other microorganisms for the synthesis of cellular components, absorbed by the plants through mycorrhiza or may be deaminated to yield ammonia.

The most active microorganisms responsible for elaborating the proteolytic enzymes (Proteinases and Peptidases) are *Pseudomonas*, *Bacillus*, *Proteus*, *Clostridium Histolyticum*, *Micrococcus*, *Alternaria*, *Penicillium* etc.

2. Ammonification (Ammonia acid degradation): Amino acids released during proteolysis undergo deamination in which nitrogen containing amino ($-NH_2$) group is removed. Thus, process of deamination which leads to the production of ammonia is termed as "ammonification". The process of ammonification is mediated by several soil microorganisms. Ammonification usually occurs under

aerobic conditions (known as oxidative deamination) with the liberation of ammonia (NH₃) or ammonium ions (NH₄) which are either released to the atmosphere or utilized by plants (paddy) and microorganisms or still under favorable soil conditions oxidized to form nitrites and then to nitrates.

The processes of ammonification are commonly brought about by *Clostridium* sp, *Micrococcus* sp, *Proteus* sp. etc. and it is represented as follows.

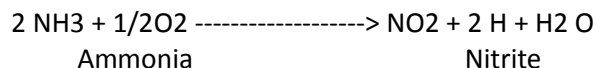


3. Nitrification:

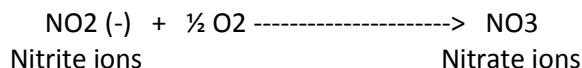
Ammonical nitrogen / ammonia released during ammonification are oxidized to nitrates and the process is called "nitrification". Soil conditions such as well aerated soils rich in calcium carbonate, a temperature below 30 ° C, neutral PH and less organic matter are favorable for nitrification in soil.

Nitrification is a two stage process and each stage is performed by a different group of bacteria as follows.

Stage I: Oxidation of ammonia of nitrite is brought about by ammonia oxidizing bacteria viz. *Nitrosomonas europaea*, *Nitrosococcus nitrosus*, *Nitrospira briensis*, *Nitroso vibrio* and *Nitrocystis* and the process is known as nitrosification. The reaction is presented as follows.



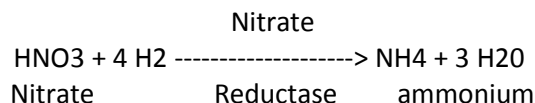
Stage II: In the second step nitrite is oxidized to nitrate by nitrite-oxidizing bacteria such as *Nitrobacter winogradsky*, *Nitrospiragracilis*, *Nitrosococcus mobilis* etc, and several fungi (eg. *Penicillium*, *Aspergillus*) and actinomycetes (eg. *Streptomyces*, *Nocardia*).



The nitrate thus, formed may be utilized by the microorganisms, assimilated by plants, reduced to nitrite and ammonia or nitrogen gas or lost through leaching depending on soil conditions. The nitrifying bacteria (ammonia oxidizer and nitrite oxidizer) are aerobic gram-negative and chemoautotrophic and are the common inhabitants of soil, sewage and aquatic environment.

4. Nitrate Reduction:

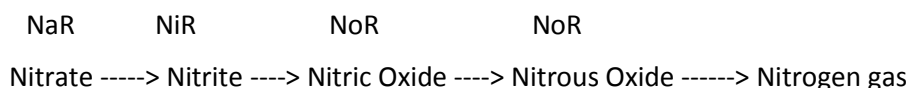
Several heterotrophic bacteria (*E. coli*, *Azospirillum*) are capable of converting nitrates to nitrites and nitrites to ammonia. Thus, the process of nitrification is reversed completely which is known as nitrate reduction. Nitrate reduction normally occurs under anaerobic soil conditions (water logged soils) and the overall process is as follows:



Nitrate reduction leading to production of ammonia is called "dissimilatory nitrate reduction" as some of the microorganisms assimilate ammonium for synthesis of proteins and amino acid.

This is the reverse process of nitrification. During denitrification nitrates are reduced to nitrites and then

to nitrogen gas and ammonia. Thus, reduction of nitrates to gaseous nitrogen by microorganisms in a series of biochemical reactions is called "denitrification". The process is wasteful as available nitrogen in soil is lost to atmosphere. The overall process of denitrification is as follows:



This process also called dissimilatory nitrate reduction as nitrate nitrogen is completely lost into atmospheric air. In the soils with high organic matter and anaerobic soil conditions (waterlogged or ill-drained) rate of denitrification is more. Thus, rice / paddy fields are more prone to denitrification.

The most important denitrifying bacteria are *Thiobacillus denitrificans*, *Micrococcus denitrificans*, and species of *Pseudomonas*, *Bacillus*, *Achromobacter*, *Serratia paracoccus* etc.

Denitrification leads to the loss of nitrogen (nitrate nitrogen) from the soil which results into the depletion of an essential nutrient for plant growth and therefore, it is an undesirable process / reaction from the soil fertility and agricultural productivity. Although, denitrification is an undesirable reaction from agricultural productivity, but it is of major ecological importance since, without denitrification the supply of nitrogen including N_2 of the atmosphere, would have not got depleted and NO_3^- (which are toxic) would have accumulated in the soil and water.

3.3. Nitrogen fixation

Introduction

Living organisms need nitrogen because it is a part of the amino acids that make up proteins, and the nucleic acids that make up DNA (deoxyribonucleic acid) and RNA (ribonucleic acid). Nitrogen (N_2) is highly inert gas. Because the nitrogen atoms in dinitrogen are bound by a very strong triple bond, this gas is very stable and cannot be utilized as a source of nutrition by any but a few highly specialized microorganisms. Nitrogen within living organisms is eventually decomposed and converted to atmospheric nitrogen (N_2). Molecular nitrogen (N_2) is the major component (approximately 80%) of the earth's atmosphere but most organisms cannot use free nitrogen, to build the chemicals required for growth and reproduction. But it has to be combined with C, H, N, O to form compounds. Before its incorporation into plants, N_2 must first be "fixed" (combined) in the form of ammonium (NH_4^+) or nitrate (NO_3^-) ions. This process of reduction of N_2 , commonly known as "nitrogen fixation" (N-fixation).

Nitrogen fixation is the process by which atmospheric nitrogen gas is converted into salts of nitrogen such as, ammonia, nitrate and nitrogen dioxide.

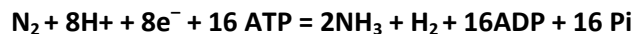


Although ammonia (NH_3) is the direct product of this reaction, it is quickly ionized to ammonium (NH_4^+). The reaction is mediated by an oxygen-sensitive enzyme nitrogenase and requires energy, as indicated by the consumption of adenosine triphosphate (ATP). This complex process is carried out by nitrogen-fixing bacteria present in the soil.

3.3.1. Symbiotic Nitrogen Fixation:

Biological nitrogen fixation was discovered by the German agronomist Hermann Hellriegel and Dutch microbiologist Martinus Beijerinck.

Biological nitrogen fixation can be represented by the following equation, in which two moles of ammonia are produced from one mole of nitrogen gas, at the expense of 16 moles of ATP and a supply of electrons and protons (hydrogen ions):



This reaction is performed exclusively by prokaryotes (the bacteria and related organisms), using an enzyme complex termed **Nitrogenase**. This enzyme consists of two proteins – an iron protein and a molybdenum-iron protein.

The reactions occur while N_2 is bound to the nitrogenase enzyme complex. The Fe protein is first reduced by electrons donated by ferredoxin. Then the reduced Fe protein binds ATP and reduces the molybdenum-iron protein, which donates electrons to N_2 , producing $\text{HN}=\text{NH}$. In two further cycles of this process (each requiring electrons donated by ferredoxin) $\text{HN}=\text{NH}$ is reduced to $\text{H}_2\text{N}-\text{NH}_2$, and this in turn is reduced to 2NH_3 .

In free-living diazotrophs, the nitrogenase-generated ammonium is assimilated into glutamate through the glutamine synthetase/glutamate synthase pathway.

Biological nitrogen fixation is of two types: Symbiotic nitrogen fixation and Non-symbiotic nitrogen fixation.

Symbiotic nitrogen fixation occurs in plants that harbor nitrogen-fixing bacteria within their tissues. The best-studied example is the association between **legumes** and **bacteria** in the genus *Rhizobium*. A symbiotic relationship in which both partners benefit is called mutualism. A mutualistic symbiosis is an association between two organisms from which each derives benefit. It is usually a longer-term relationship, and with symbiotic nitrogen (N_2) fixation, often involves a special structure to house the microbial partner. Each N_2 -fixing symbiotic association involves an N_2 -fixing prokaryotic organism, the microsymbiont (eg. *Rhizobium*, *Klebsiella*, *Nostoc* or *Frankia*) and a eukaryotic, usually photosynthetic, host (e.g., leguminous or nonleguminous plant, water fern or liverwort).

History of the legume-*Rhizobium* symbiosis:

Legumes have been used in crop rotations since the time of the Romans. However, it was not until detailed N balance studies became possible, that they were shown to accumulate N from sources other than soil and fertilizer. In 1886 Hellriegel and Wilfarth demonstrated that the ability of legumes to convert N_2 from the atmosphere into compounds which could be used by the plant was due to the presence of swellings or nodules on the legume root, and to the presence of particular bacteria within these nodules.

Mechanism of Symbiotic Nitrogen Fixation:

Root Nodule Formation:

Rhizobia can infect their hosts and induce root nodule formation using following mechanisms:

- Root hair penetration and infection thread formation, as occurs in clovers and beans.
- Entry via wounds or sites of lateral root emergence, as found in peanut and the pasture legume *Stylosanthes*.
- Penetration of root primordia found on the stem of plants such as *Sesbania*.

NODULE INITIATION AND DEVELOPMENT

The process of root hair penetration and infection thread formation is treated in greater detail by Hirsch (1992) and Lhuissier et al (2001). The Fahraeus slide technique (Fahraeus, 1957) allowed repeated observation of the infection process and The Root-tip marking procedure (Bhuvaneswari et al., 1981) showed differences in the susceptibility of immature and mature root hairs to infection, and focused research on those parts of the root where infection by *Rhizobium* was most common.

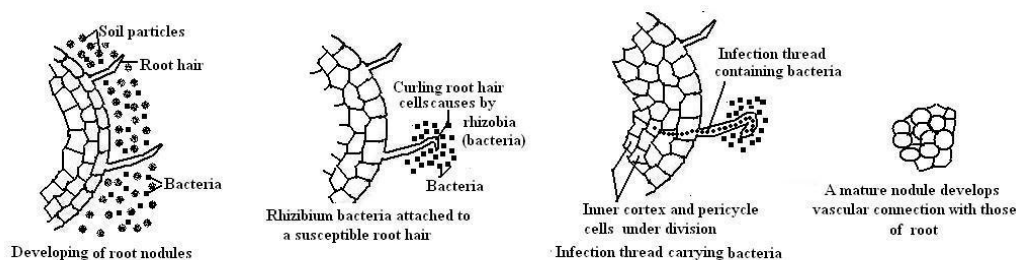


Fig 4. Development of root nodule

Rhizobium secretes Nod factors (Some nod genes encode enzymes that make Nod factors) the leguminous plant recognizes Nod factors.

Visible changes during root hair infection:

Infection begins with rhizobial attachment to immature, emerging root hairs of a compatible host. This occurs within minutes of inoculation, with attached rhizobia capping the root-hair tip, and often oriented end on to their host. Deformation and curling of the root hair follows, with the root hair surface at the point of infection hydrolyzed to permit penetration of the rhizobia. Rhizobia then move down the root hair toward the root cortex. Rhizobia never really gain free intracellular access to their host. During infection, and as they move down the root hair, they remain enclosed within a plant-derived material, and in some plants, for example *Chamaecrista*, may never escape from this infection thread.

Examination of a nodule under the microscope reveals four distinct zones:

- Meristematic region in which host cells undergo active division but show little infection by rhizobia
- A region in which many plant cells are infected but in which the bacteria have not undergone changes in size and shape, and N_2 fixation is limited
- Region of active N_2 fixation, often red or pink in color due to the presence of leghemoglobin. Host cells will contain many rhizobia and these may be misshapen. Such bacteria are referred to as bacteroids.
- Region of nodule senescence in which the symbiosis is breaking down. Bacteroids may undergo lysis, and the degradation of leghemoglobin results in a green or brown coloration.

The Infection Thread:

The interaction between a particular strain of rhizobia and the “appropriate” legume is mediated by:

- a “Nod factor” secreted by the rhizobia.
- Transmembrane receptors on the cells of the root hairs of the legume.
- Different legumes produce receptors of different specificity.
- Different strains of rhizobia produce different Nod factors, and

If the combination is correct, the bacteria enter an epithelial cell of the root; then migrate into the cortex. Their path runs within an intracellular channel that grows through one cortex cell after another.

This **infection thread** is constructed by the root cells, not the bacteria, and is formed only in response to the infection.

When the infection thread reaches a cell deep in the cortex, it bursts and the rhizobia are engulfed by endocytosis into membrane-enclosed **symbiosomes** within the cytoplasm. At this time the cell goes through several rounds of mitosis — without cytokinesis — so the cell becomes polyploid. The cortex cells then begin to divide rapidly forming a **nodule**. This response is driven by the translocation of cytokinins from epidermal cells to the cells of the cortex.

The rhizobia also go through a period of rapid multiplication within the nodule cells. Then they begin to change shape and lose their motility. The **bacteroids**, as they are now called, may almost fill the cell. Only now does nitrogen fixation begin. Root nodules are not simply structureless masses of cells. Each becomes connected by the xylem and phloem to the vascular system of the plant. Thus the development of nodules, while dependent on rhizobia, is a well-coordinated developmental process of the plant. The benefit to the legume host is clear. The rhizobia make it independent of soil nitrogen.

3.3.2. Non-Symbiotic N₂-Fixing Bacteria:

The non-symbiotic nitrogen fixing bacteria do not require a host plant. In 1891, Winogradsky observed that when soil was exposed to the atmosphere, the nitrogen content of the soil was recorded to be increased.

The anaerobic bacterium *Clostridium pasteurianum* was found responsible for such an increase of the nitrogen content in soil. In 1901, Beijerinck proved that there were also free-living aerobic bacteria, *Azotobacter chroococcum* that could fix atmospheric nitrogen.

Another bacterial group, *Granulobacter* (purple colour) obtains nitrogen directly from the atmosphere. The amounts of atmospheric nitrogen fixed by these bacteria are largely variable because of divergent nature of soils.

In aerobic soils of tropical climatic regions, the acid tolerant N₂-fixer *Azotobacter beijerinckia* is most abundant *Azospirillum* spp. also fix N₂-non-symbiotically and help to many crops for their growth and yield.

3.4.Sulphur Cycle:

Sulphur is one of the components that make up proteins and vitamins. Proteins consist of amino acids that contain sulphur atoms. Sulphur is important for the functioning of proteins and enzymes in plants, and in animals that depend upon plants for sulphur.

3.4.1. Release of sulfur from organic compounds

It enters the atmosphere through both natural and human sources. Natural recourses can be for instance volcanic eruptions, bacterial processes, evaporation from water, or decaying organisms. When sulphur enters the atmosphere through human activity, this is mainly a consequence of industrial processes where sulphur dioxide (SO₂) and hydrogen sulphide (H₂S) gases are emitted on a wide scale.

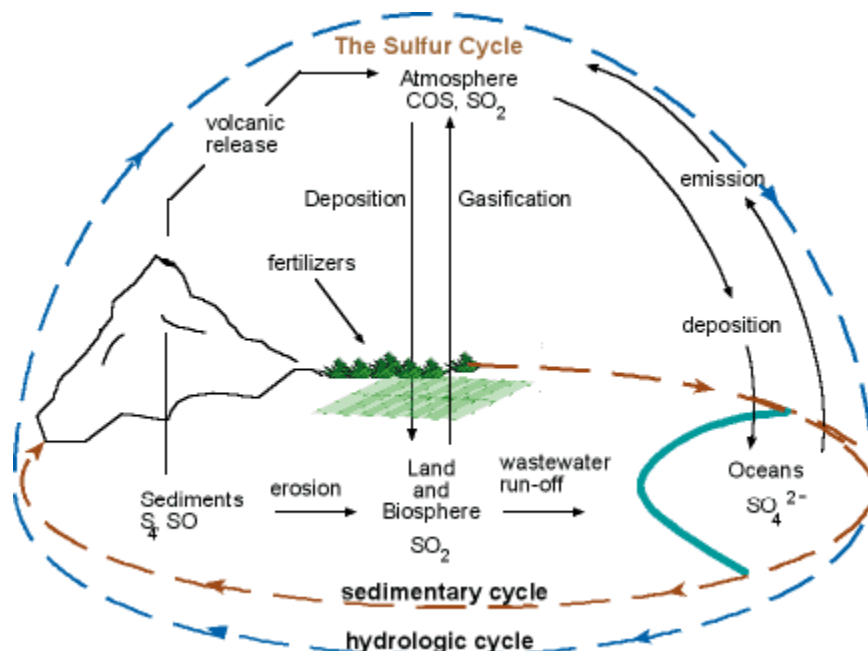
3.4.2. Oxidation of sulfur compounds

When sulphur dioxide enters the atmosphere it will react with oxygen to produce sulphur trioxide gas (SO₃), or with other chemicals in the atmosphere, to produce sulphur salts. Sulphur dioxide may also react with water to produce sulphuric acid (H₂SO₄). Sulphuric acid may also be produced from demethyl-sulphide, which is emitted to the atmosphere by plankton species.

3.4.3. Reduction of sulfate

All these particles will settle back onto earth, or react with rain and fall back onto earth as acid deposition. The particles will then be absorbed by plants again and are released back into the atmosphere, so that the sulphur cycle will start over again.

- i. Fossil fuels like coal and petroleum are extremely important energy resources which are getting exhausted.
- ii. Hydrocarbon fuel based resources create pollution levels and green house gases. Their management is related to improved technology and finding alternative energy sources taking this into account.
- iii. An overall prudent and sustainable uses of resources both at an individual and collective level can benefit a wide cross section of society as well meet the future generations.



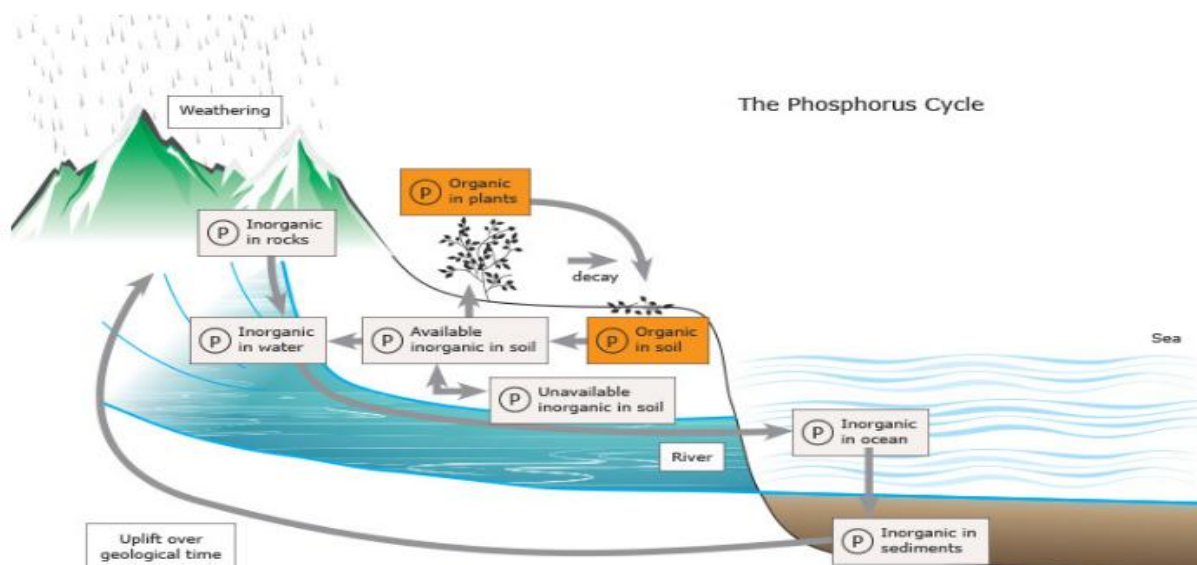
3.5. Phosphorus cycle

The phosphorus cycle

Phosphorus moves in a cycle through rocks, water, soil and sediments and organisms.

Here are the key steps of the phosphorus cycle

- Over time, rain and weathering cause rocks to release phosphate ions and other minerals. This inorganic phosphate is then distributed in soils and water.
- Plants take up inorganic phosphate from the soil. The plants may then be consumed by animals. Once in the plant or animal, the phosphate is incorporated into organic molecules such as DNA. When the plant or animal dies, it decays, and the organic phosphate is returned to the soil.
- Within the soil, organic forms of phosphate can be made available to plants by bacteria that break down organic matter to inorganic forms of phosphorus. This process is known as mineralisation.
- Phosphorus in soil can end up in waterways and eventually oceans. Once there, it can be incorporated into sediments over time.



Phosphorus moves in a cycle through rocks, water, soil and sediments and organisms.

Most phosphorus is unavailable to plants

Since most of our phosphorus is locked up in sediments and rocks, it's not available for plants to use. A lot of the phosphorus in soils is also unavailable to plants.

Soil phosphorus becomes unavailable to plants through several routes:

- **Bacteria:** Bacteria convert plant-available phosphate into organic forms that are then not available to plants. Although other bacteria make phosphate available by mineralisation, the contribution of this is small.
- **Adsorption:** Inorganic (and available) phosphorus can be chemically bound (adsorbed) to soil particles, making it unavailable to plants. Desorption is the release of adsorbed phosphorus from its bound state into soil solution.
- **pH:** Inorganic phosphorus compounds need to be soluble to be taken up by plants. This depends on the acidity (pH) of the soil. If soils are less than pH 4 or greater than pH 8, the phosphorus starts to become tied up with other compounds, making it less available to plants.

Many plant crops need more phosphorus than is dissolved in the soil to grow optimally. In addition, crops are usually harvested and removed – leaving no decaying vegetation to replace phosphorus. Therefore, farmers replenish the phosphorus 'pool' by adding fertilisers or effluent to replace the phosphorus taken up by plants.

3.6. Carbon Cycle:

The carbon cycle is the biogeochemical cycle by which carbon is exchanged among the biosphere, pedosphere, geosphere, hydrosphere, and atmosphere of the Earth. It is one of the most important cycles of the earth and allows for carbon to be recycled and reused throughout the biosphere and all of its organisms.

The Carbon Cycle is a complex series of processes through which all of the carbon atoms in existence rotate. The wood burned just a few decades ago could have produced carbon dioxide which through photosynthesis became part of a plant. When you eat that plant, the same carbon from the wood which was burnt can become part of you. The carbon cycle is the great natural recycler of carbon atoms.

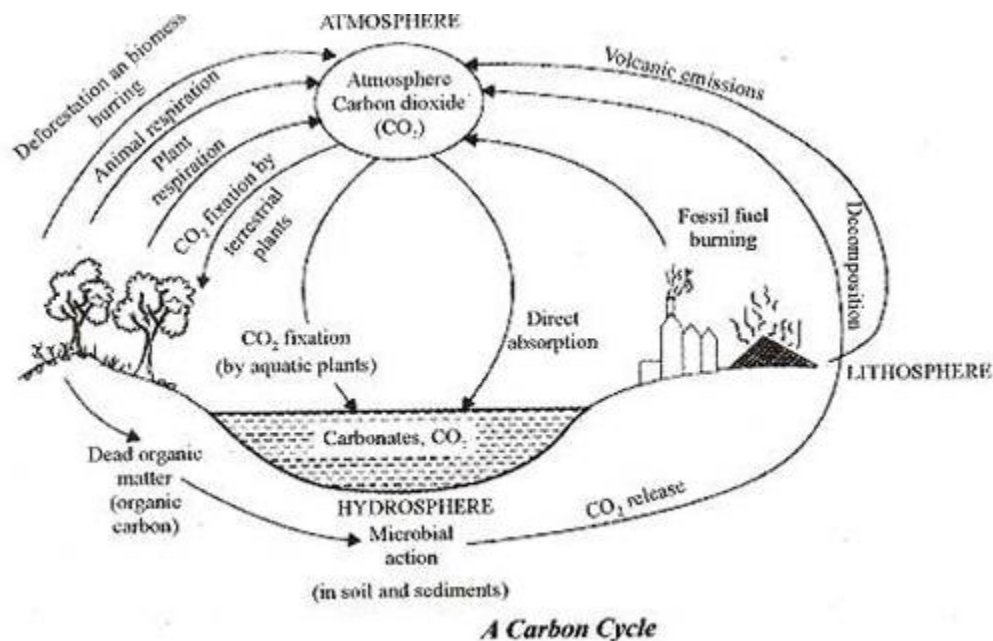
Without the proper functioning of the carbon cycle, every aspect of life could be changed dramatically. Plants, animals, and soil interact to make up the basic cycles of nature. In the carbon cycle, plants absorb carbon dioxide from the atmosphere and use it, combined with water they get from the soil, to make the substances they need for growth. The process of photosynthesis incorporates the carbon atoms from carbon dioxide into sugars.

Animals, such as the rabbit eat the plants and use the carbon to build their own tissues. Other animals, such as the fox, eat the rabbit and then use the carbon for their own needs. These animals return carbon dioxide into the air when they breathe, and when they die, since the carbon is returned to the soil during decomposition. The carbon atoms in soil may then be used in a new plant or small microorganisms. The following major reservoirs of carbon interconnected by pathways of exchange:

- i. The atmosphere.
- ii. The terrestrial biosphere, which is usually defined to include fresh water systems and non-living organic material, such as soil carbon.

ADVERTISEMENTS:

- iii. The oceans, including dissolved inorganic carbon and living and non-living marine biota.
- iv. The sediments including fossil fuels
- v. The Earth's interior, carbon from the Earth's mantle and crust is released to the atmosphere and hydrosphere by volcanoes and geothermal systems.



The annual movements of carbon, the carbon exchanges between reservoirs, occur because of various chemical, physical, geological, and biological processes. The ocean contains the largest active pool of carbon near the surface of the Earth, but the deep ocean part of this pool does not rapidly exchange with the atmosphere in the absence of an external influence, such as an uncontrolled deep-water oil well leak.

The global carbon budget is the balance of the exchanges (incomes and losses) of carbon between the carbon reservoirs or between one specific loop the carbon cycle.

Carbon is released into the atmosphere in several ways:

- i. Through the respiration performed by plants and animals. This is an exothermic reaction and it involves the breaking down of glucose (or other organic molecules) into carbon dioxide and water.
- ii. Through the decay of animal and plant matter. Fungi and bacteria break down the carbon compounds in dead animals and plants and convert the carbon to carbon dioxide if oxygen is present, or methane if not.
- iii. Through combustion of organic material which oxidizes the carbon it contains, producing carbon dioxide (and other things, like water vapour). Burning fossil fuels such as coal, petroleum products releases carbon dioxide. Burning agro fuels also releases carbon dioxide
- iv. Volcanic eruptions and metamorphism release gases into the atmosphere. Volcanic gases are primarily water vapour, carbon dioxide and sulphur dioxide.
- v. Carbon is transferred within the biosphere as heterotrophs feed on other organisms or their parts (e.g., fruits). This includes the uptake of dead organic material (detritus) by fungi and bacteria for fermentation or decay.
- vi. Most carbon leaves the biosphere through respiration. When oxygen is present, aerobic respiration occurs, which releases carbon dioxide into the surrounding air or water, following the reaction $C_6H_{12}O_6 + 6O_2 \rightarrow 6CO_2 + 6H_2O$. Otherwise, anaerobic respiration occurs and releases methane into the surrounding environment, which eventually makes its way into the atmosphere or hydrosphere (e.g., as marsh gas or flatulence).

Circulation of carbon dioxide:

- i. Plants absorb the carbon dioxide from the atmosphere.
- ii. During the process of photosynthesis, plants incorporate the carbon atoms from carbon dioxide into sugars.
- iii. Animals, such as the rabbit eat the plants and use the carbon to build their own tissues, chain the carbon content
- iv. Through the food chain, carbon is transferred into foxes, lions etc.
- v. The animals return carbon dioxide into the air when they breathe, and when they die, since the carbon is returned to the soil during decomposition

In Case of Ocean:

In regions of oceanic upwelling, carbon is released to the atmosphere. Conversely, regions of down welling transfer carbon (CO_2) from the atmosphere to the ocean. When CO_2 enters the ocean, it participates in a series of reactions which are locally in equilibrium:

- i. Conversion of CO_2 (atmospheric) to CO_2 (dissolved).
- ii. Conversion of CO_2 (dissolved) to carbonic acid (H_2CO_3).
- iii. Conversion of carbonic acid (H_2CO_3) to bicarbonate ion.
- iv. Conversion of bicarbonate ion to carbonate ion.

In the oceans, dissolved carbonate can combine with dissolved calcium to precipitate solid calcium carbonate, $CaCO_3$, mostly as the shells of microscopic organisms. When these organisms die, their shells sink and accumulate on the ocean floor. Over time these carbonate sediments form limestone which is the largest reservoir of carbon in the carbon cycle.

The dissolved calcium in the oceans comes from the chemical weathering of calcium-silicate rocks, during which carbonic and other acids in groundwater react with calcium-bearing minerals liberating calcium ions to solution and leaving behind a residue of newly formed aluminium-rich clay minerals and insoluble minerals such as quartz.

The flux or absorption of carbon dioxide into the world's oceans is influenced by the presence of widespread viruses within ocean water that infect many species of bacteria. The resulting bacterial deaths spawn a sequence of events that lead to greatly enlarged respiration of carbon dioxide, enhancing the role of the oceans as a carbon sink.

3.7. Plant nutrition

3.7.1. Specific roles of essential elements and their deficiency symptoms in plants

Elements are required by plants at a particular concentration. Each element has a specific role in the growth of plant, and an inadequate supply leads to deficiency symptoms. Carbon, hydrogen and oxygen serve as principle structural components of organic matter. Deficiency of carbon leads to quick death of plant and deficiency of water leads to wilting and desiccation.

3.7.2. Macro Nutrients

Primary (macro) nutrients are nitrogen, phosphorus, and potassium. They are the most frequently required in a crop fertilization program. Also, they are need in the greatest total quantity by plants as fertilizer.

Nitrogen

- Necessary for formation of amino acids, the building blocks of protein
- Essential for plant cell division, vital for plant growth
- Directly involved in photosynthesis
- Necessary component of vitamins
- Aids in production and use of carbohydrates
- Affects energy reactions in the plant

Phosphorus

- Involved in photosynthesis, respiration, energy storage and transfer, cell division, and enlargement
- Promotes early root formation and growth
- Improves quality of fruits, vegetables, and grains
- Vital to seed formation
- Helps plants survive harsh winter conditions
- Increases water-use efficiency
- Hastens maturity

Potassium

- Carbohydrate metabolism and the break down and translocation of starches
- Increases photosynthesis
- Increases water-use efficiency
- Essential to protein synthesis
- Important in fruit formation
- Activates enzymes and controls their reaction rates
- Improves quality of seeds and fruit
- Improves winter hardiness
- Increases disease resistance

Secondary Nutrients

The secondary nutrients are calcium, magnesium, and sulphur. For most crops, these three are needed in lesser amounts than the primary nutrients. They are growing in importance in crop fertilization programs due to more stringent clean air standards and efforts to improve the environment.

Calcium

- Utilized for Continuous cell division and formation
- Involved in nitrogen metabolism
- Reduces plant respiration
- Aids translocation of photosynthesis from leaves to fruiting organs
- Increases fruit set
- Essential for nut development in peanuts
- Stimulates microbial activity

Magnesium

- Key element of chlorophyll production
- Improves utilization and mobility of phosphorus
- Activator and component of many plant enzymes
- Directly related to grass tetany
- Increases iron utilization in plants
- Influences earliness and uniformity of maturity

Sulphur

- Integral part of amino acids
- Helps develop enzymes and vitamins
- Promotes nodule formation on legumes
- Aids in seed production
- Necessary in chlorophyll formation (though it isn't one of the constituents)

3.7.3. Micronutrients

The micronutrients are boron, chlorine, copper, iron, manganese, molybdenum, and zinc. These plant food elements are used in very small amounts, but they are just as important to plant development and profitable crop production as the major nutrients. Especially, they work "behind the scene" as activators of many plant functions.

Boron

- Essential for germination of pollen grains and growth of pollen tubes
- Essential for seed and cell wall formation
- Promotes maturity
- Necessary for sugar translocation
- Affects nitrogen and carbohydrate

Chlorine

- Not much information about its functions
- Interferes with P uptake
- Enhances maturity of small grains on some soils

Copper

- Catalyzes several plant processes
- Major function in photosynthesis
- Major function in reproductive stages
- Indirect role in chlorophyll production
- Increases sugar content
- Intensifies color
- Improves flavor of fruits and vegetables

Iron

- Promotes formation of chlorophyll
- Acts as an oxygen carrier
- Reactions involving cell division and growth

Maganese

- Functions as a part of certain enzyme systems
- Aids in chlorophyll synthesis
- Increases the availability of P and CA

Molybdenum

- Required to form the enzyme "nitrate reductase" which reduces nitrates to ammonium in plant
- Aids in the formation of legume nodules
- Needed to convert inorganic phosphates to organic forms in the plant

Zinc

- Aids plant growth hormones and enzyme system
- Necessary for chlorophyll production
- Necessary for carbohydrate formation
- Necessary for starch formation
- Aids in seed formation

In addition to the 13 nutrients listed above, plants require carbon, hydrogen, and oxygen, which are extracted from air and water to make up the bulk of plant weight.

KARPAGAM ACADEMY OF HIGHER EDUCATION
COIMBATORE - 21
DEPARTMENT OF BIOCHEMISTRY
III B.Sc BIOCHEMISTRY
BATCH: 2015 - 2018

15BCU504 - Plant Biochemistry

PART A (20 X 1 = 20 MARKS) - Online MCQ questions

SL.NO	QUESTION	OPTION 1	OPTION 2	OPTION 3	OPTION 4	ANSWER
Unit 3						
1	Chlorosis of leaves is due to the deficiency of	Carbon	nitrogen	sulphur	phosphorus	nitrogen
2	The release of NH_4^+ ion in the soil through the decomposition of organic matter by microbial decay is called	Nitrification	biological nitrogen fixation	Ammonification	denitrification	Ammonification
3	The conversion of ammonium ions into nitrites is performed by	Nitrobacter	nitrococcus	azotobacter	clostridium	nitrococcus
4	Oxidation of nitrites into nitrates is performed by	Nitrobacter	nitrosomonas	nitrococcus	clostridium	Nitrobacter
5	The process by which NH_3 is converted to nitrates is called as	Nitrification	denitrification	ammonification	nitrogen fixation	Nitrification
6	Nitrate reductase is found in	Mitochondria	cytoplasm	vacuoles	golgi complex	cytoplasm
7	The electron transfer in the enzyme nitrite reductase is mediated by	Mo	FAD	sirohaem	cyt b557	sirohaem
8	Which of the following is a free living chemo synthetic bacteria?	Chlorobium	clostridium	azotobacter	desulphovibrio	desulphovibrio
9	Which of the following is a free living anaerobic nitrogen fixing bacteria?	Desulphovibrio	clostridium	chlorobium	azotobacter	clostridium
10	The growth factor secreted by the roots of pism sativum is	Serine	homoserine	glycine	alanine	homoserine

SL.NO	QUESTION	OPTION 1	OPTION 2	OPTION 3	OPTION 4	ANSWER
11	Which is the symbiotic nitrogen fixing bacteria?	Rhizobium	azotobacter	clostridium	nitrosomonas	Rhizobium
12	The pink colour of nitrogen fixing nodule is due to the presence of	Xanthophylls	carotenoids	leghaemoglobin	bacteroids	leghaemoglobin
13	Nutrients that are required by plants in smaller quantities are considered as	micronutrients	macronutrients	mega nutrients	chemical nutrients	micronutrients
14	Nutrients that are required by plants in large quantities are considered as	mega nutrients	chemical nutrients	micronutrients	macronutrients	macronutrients
15	Micronutrient which is important in transport of sugar, synthesis of enzymes and cell division is	phosphorus	boron	potassium	Sulphur	boron
16	Insectivorous plants use _____ as nitrogen source	Nitrate	molecular nitrogen	ammonia	nitrite	molecular nitrogen
17	Distinctive odour and flavor to garlic, onion and mustard oil is due to	phosphorus	boron	potassium	Sulphur	Sulphur
18	Suppression of fruit formation and delaying in ripening is due to deficiency of	phosphorus	boron	potassium	Sulphur	Sulphur
19	Reduction in the number of stroma lamellae and increase in the number of grana lamellae is due to the deficiency of	phosphorus	boron	potassium	Sulphur	Sulphur
20	Promotion of fruit ripening is due to the deficiency of	phosphorus	boron	potassium	Sulphur	phosphorus
21	Accumulation of carbohydrates is due to the deficiency of	phosphorus	boron	potassium	Sulphur	phosphorus
22	Toxic effect of calcium is antagonized by	phosphorus	boron	potassium	Sulphur	potassium

SL.NO	QUESTION	OPTION 1	OPTION 2	OPTION 3	OPTION 4	ANSWER
23	Mottled chlorosis of leaves is due to the deficiency of	phosphorus	boron	potassium	Sulphur	potassium
24	The main constituent of middle lamellae of the cell is	phosphorus	boron	calcium	Sulphur	calcium
25	Oxalic acid is neutralized by	phosphorus	boron	calcium	Sulphur	calcium
26	Arginine kinase is activated by	phosphorus	boron	calcium	Sulphur	calcium
27	Phospho lipase is activated by	phosphorus	boron	calcium	Sulphur	calcium
28	Indole acetic acid is oxidized by	phosphorus	boron	manganese	Sulphur	manganese
29	Enzymes of nitrogen metabolism is activated by	phosphorus	boron	manganese	Sulphur	manganese
30	_____ acts as a cofactor in oxidative phosphorylation	phosphorus	boron	manganese	Sulphur	manganese
31	Formation of seeds is slowed down by the deficiency of	phosphorus	boron	manganese	Sulphur	manganese
32	Retardation in nitrogen assimilation is due to the deficiency of	phosphorus	boron	manganese	Sulphur	manganese
33	Chlorosis in pine apple and citrus is caused by the deficiency of	phosphorus	manganese	calcium	Sulphur	manganese
34	_____ participates in the synthesis of tryptophan and auxin	phosphorus	boron	zinc	Sulphur	zinc

SL.NO	QUESTION	OPTION 1	OPTION 2	OPTION 3	OPTION 4	ANSWER
35	Formation of carbonic anhydrase is enhanced by	zinc	boron	calcium	Sulphur	zinc
36	Metabolism of alcohol dehydrogenase is activated by	zinc	boron	calcium	Sulphur	zinc
37	Rosette disease of walnut is due to the deficiency of	zinc	boron	calcium	Sulphur	zinc
38	White bud disease of maize is due to the deficiency of	zinc	boron	calcium	Sulphur	zinc
39	Antibiotic production in <i>Fusarium</i> is promoted by	zinc	boron	calcium	Sulphur	zinc
40	Nicotine production in tobacco plants is promoted by	zinc	boron	calcium	Sulphur	zinc
41	Easy transport of sugar in phloem is helped by	zinc	boron	calcium	Sulphur	boron
42	“Top sickness” disease in tobacco is caused due to the deficiency of	zinc	boron	calcium	Sulphur	boron
43	“heart rot disease of sugar beet” is due to the deficiency of	zinc	boron	calcium	Sulphur	boron
44	_____ participates in the formation of phenolase lactase and ascorbic acid oxidase	zinc	boron	copper	Sulphur	copper
45	Black pigmentation in the spores of <i>Aspergillus niger</i> is imparted by	zinc	boron	copper	Sulphur	copper
46	_____ acts as fungicide to prevent the disease “late blight of potato”	copper	boron	calcium	Sulphur	copper

SL.NO	QUESTION	OPTION 1	OPTION 2	OPTION 3	OPTION 4	ANSWER
47	Nitrate reductase enzyme is activated by	molybdenum	boron	calcium	Sulphur	molybdenum
48	“Whip tail” disease of cauliflower is due to the deficiency of	zinc	boron	calcium	molybdenum	molybdenum
49	Cysteine desulfurase converts	Cysteine into pyruvate and H ₂ S	Cysteine into carbohydrate	Cysteine into sulfur	cysteine into cystine	Cysteine into pyruvate and H ₂ S
50	When chlorophyll is burst, which one is obtained?	iron	Magnesium	calcium	Manganese	Magnesium
51	The element which is required in largest quantities by plant is	nitrogen	calcium	sulphur	phosphorus	nitrogen
52	Which of the following elements is responsible for maintaining turgor?	sodium	potassium	Calcium	Magnesium	potassium
53	Which of the following is not an essential element for plants?	iron	zinc	potassium	iodine	iodine
54	Ascorbic acid synthesis is controlled by	Molybdenum	copper	Boron	Zinc	Molybdenum
55	Carbohydrate metabolism is increased by	Molybdenum	copper	Boron	Zinc	Molybdenum
56	Auxin synthesis is enhanced by	Molybdenum	copper	Boron	Zinc	Zinc
57	Toxic effect of NaCl is antagonized by	calcium	copper	Boron	Zinc	Calcium
58	Phospholipase is activated by	calcium	copper	Boron	Zinc	Calcium

SL.NO	QUESTION	OPTION 1	OPTION 2	OPTION 3	OPTION 4	ANSWER
59	More lignifications of cells occurs due to deficiency of	Potassium	copper	Boron	Zinc	Potassium
60	Binding of nucleic acid with protein is enhanced by	Calcium	copper	Boron	Zinc	Calcium



KARPAGAM ACADEMY OF HIGHER EDUCATION
(Deemed University Established Under Section 3 of UGC Act 1956)
Coimbatore - 641021.

(For the candidates admitted from 2015 onwards)

DEPARTMENT OF BIOCHEMISTRY

COURSE MATERIAL

STAFF NAME	:	Dr. L. HARIPRASATH	
SUBJECT	:	PLANT BIOCHEMISTRY	SUBJECT CODE : 15BCU504
SEMESTER	:	V	CLASS : III B.Sc. Biochemistry

UNIT-IV

Plant growth regulators: Auxins: Chemistry, biosynthesis, mode of action, bioassay, practical applications of synthetic auxins. Gibberellins: Chemistry, biosynthesis and mechanism of action, role of endogenous gibberellins, bioassay and practical applications. Chemistry, mode of action and physiological role of cytokinins, abscisic acid and ethylene.

TEXT BOOKS

- Verma, S.K., & Mohit Verma. (2013). *A Text Book of Plant Physiology, Biochemistry and Biotechnology*. (6th ed.). New Delhi. S. Chand and Co.
- Bonner, J., & Varner, F.J. (2012). *Plant Biochemistry*. (3rd ed.). Burlington: Elsevier Science.
- Goodwin, T.W., & Mercer, E.I. (1990). *Introduction to Plant Biochemistry*. (2nd ed.). New York, NY: Robert Maxwell.M.C Publisher.

REFERENCES

- Buchanan, B.B. (2015). *Biochemistry and Molecular Biology of Plants*. New Jersey, NJ: John Wiley.
- Hans-Valter Heldt. (2005). *Plant Biochemistry and Molecular Biology*. England: Oxford University Press.
- Wink, M. (2010). *Functions and Biotechnology of Plant Secondary Metabolites*. (2nd ed.), London: Blackwell Publishing Ltd.,

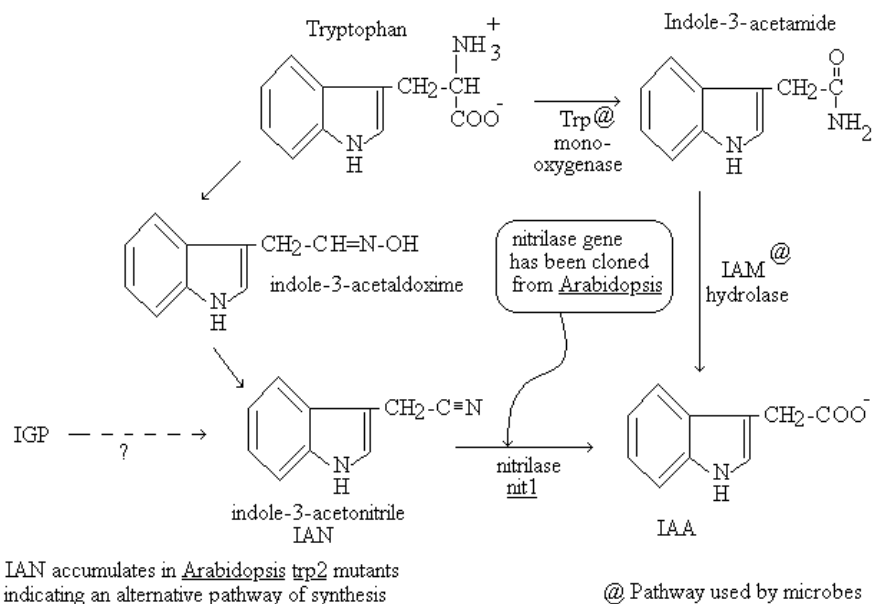
Unit IV

4. Plant growth regulators

4.1. Auxins

4.1.1. Chemistry: Indole-3-acetic acid (IAA) is the main auxin in most plants.

4.1.2. Biosynthesis: IAA is synthesized from tryptophan or indole primarily in leaf primordia and young leaves, and in developing seeds.



4.1.3. Mode of action:

The mechanism by which the plant hormone auxin regulates plant growth has puzzled scientists since Darwin's time. Auxin is known to regulate gene expression by binding to its receptor TIR1 and promoting ubiquitin-dependent degradation of Aux/IAA repressor proteins. Now the determination of the crystal structures of TIR1 in complexes with three different auxins and an Aux/IAA peptide shows auxin to act as a 'molecular glue' promoting interactions between the receptor and proteins targeted for degradation. As well as revealing auxin's mechanism, this work establishes the first structural model of a plant hormone receptor. Also, the discovery that a small molecule like auxin can regulate ubiquitin ligases suggests a novel strategy for developing therapeutics for human disorders associated with ubiquitin ligase defects. On the cover, auxin (shown as a spacefilling model) is seen in the cavity between TIR1 (blue) and IAA7 peptide (orange).

4.1.4. Bioassay of Auxins:

It is testing of a biological activity like growth response of a substance by employing a living material like plant or plant part. Auxin bioassay is quantitative test as it measures concentration of auxin to produce the effect and the amount of effect.

1. Avena Curvature Test :

The test is based upon experiments of Went (1928). 10° curvature is produced by auxin concentration of $150 \mu\text{g/litre}$ at 25°C and 90% relative humidity. The test can measure auxin upto 300 pg/litre .

Auxin from a shoot tip or any other plant organ is allowed to diffuse in a standard size agar block (generally 2 x 2 x 1 mm). Auxin can also be dissolved directly in agar. 15-30 mm long oat coleoptile grown in dark is held vertically over water. 1 mm tip of coleoptile is removed without injuring the primary leaf.

After 3 hours a second decapitation is carried out for a distance of 4 mm. Primary leaf is now pulled loose and agar block supported against it at the tip of decapitated coleoptile. After 90-110 minutes, the coleoptile is found to have bent. The curvature is measured. It can also be photographed and the curvature known from shadow graph.

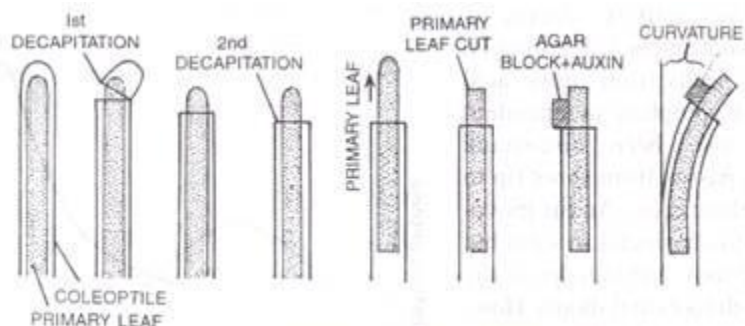


Fig. 15.20. Method of Avena Curvature Test.

2. Root Growth Inhibition:

Sterilized seeds of Cress are allowed to germinate on moist filter paper. As the roots reach a length of 1 cm or so, root lengths are measured. 50% of the seedlings are placed in a test solution while the remaining is allowed to grow over moist paper.

Lengths of the roots are measured after 48 hours. It is seen that the seedlings placed in test solution show very little root growth while root growth is normal in control seedlings.

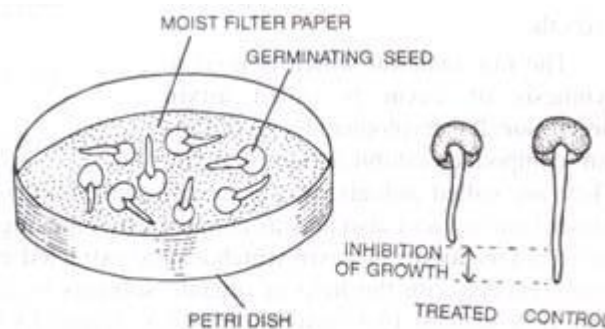


Fig. 15.21. Root growth inhibition bioassay of auxin.

4.1.5. Practical applications of synthetic auxins and Physiological Effects:

- The uses of synthetic auxins in horticulture can be traced directly to the natural roles of IAA in the plant. In general, compounds such as α -naphthalene acetic acid (NAA) are used because they resemble IAA in action but are resistant to degradation by plant enzymes. Auxins are used for a variety of agricultural purposes, including: Promotion of rooting of cuttings (e.g., Rootone). The base of the cutting is dipped in a powder containing NAA or indolebutyric acid (IBA) prior to planting. Induction of flowering in pineapple (actually caused by the auxin-induced production of ethylene).

- NAA is generally employed as the auxin. Increased fruit set and induction of the pericarp in the absence of fertilization. Prevention of preharvest fruit drop. Auxin type herbicides (e.g., 2-4-D).
- Cell enlargement - auxin stimulates cell enlargement and stem growth.
- Cell division - auxin stimulates cell division in the cambium and, in combination with cytokinin, in tissue culture.
- Vascular tissue differentiation - auxin stimulates differentiation of phloem and xylem.
- Root initiation - auxin stimulates root initiation on stem cuttings, and also the development of branch roots and the differentiation of roots in tissue culture.
- Tropistic responses - auxin mediates the tropistic (bending) response of shoots and roots to gravity and light.
- Apical dominance - the auxin supply from the apical bud represses the growth of lateral buds.
- Delayed leaf senescence.
- Leaf and fruit abscission - auxin may inhibit or promote (via ethylene) leaf and fruit abscission depending on the timing and position of the source.
- Delayed fruit ripening.
- In several systems (e.g., root growth) auxin, particularly at high concentrations, is inhibitory. Almost invariably this has been shown to be mediated by auxin-produced ethylene. If the ethylene synthesis is prevented by various ethylene synthesis inhibitors, then auxin is no longer inhibitory.

4.2. Gibberellins

Gibberellins are a plant growth substance (phytohormone) involved in promotion of stem elongation, mobilisation of food reserves in seeds and other processes. Its absence results in the dwarfism of some plant varieties. Chemically all known gibberellins are gibberellic acids, a family of diterpene acids that are synthesized by the terpenoid pathway in plastids and then modified in the endoplasmic reticulum and cytosol until they reach their biologically active form.

4.2.1. Chemistry

It is chemically $C_{19}H_{22}O_6$. GA_3 is one of the most intensively studied gibberellin. A mixture of GA_4 and GA_7 is used commercially. Until now 125 different gibberellins have been identified. Many of them occur naturally in plants and fungi. *Gibberella fujikori* has as many as 15 gibberellins.

A single plant also possesses a number of gibberellins. This is in contrast to auxin, where a single natural hormone occurs. Gibberellins are synthesized in the apical shoot buds (young leaves), root tips and developing seeds. The precursors for their synthesis is mevalonic acid (derived from acetyl coenzyme A). Gibberellin transport occurs through simple diffusion as well as through conducting channels.

4.2.2 Biosynthesis

Biosynthesis of Gibberellins in Plants:

The gibberellins which are chemically related to terpenoids (natural rubber, carotenoids & steroids) are thought to be formed by the condensation of a 5-C precursor—an isoprenoid unit called as isopentenyl pyrophosphate (IPP) through a number of intermediates to give rise to gibberellins. The primary precursor for the formation of this isoprenoid unit and synthesis of gibberellins is however, acetate.

(Besides gibberellins, carotenoids, rubber, steroids, Absciscic acid (ABA) and part of cytokinins are also derived from 5-C isoprenoid unit.).

Biosynthesis of gibberellins in plants is shown schematically in Fig

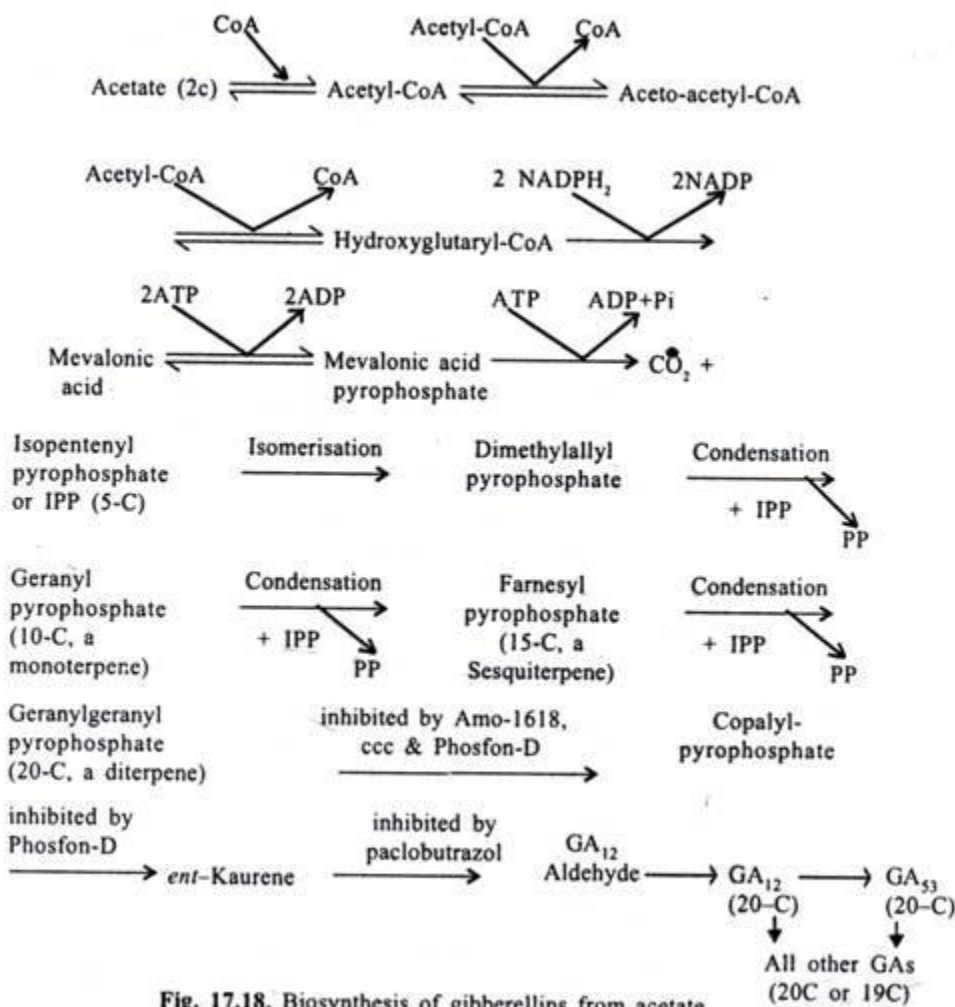


Fig. 17.18. Biosynthesis of gibberellins from acetate.

In plants GAs are biosynthesized in apical tissues and there are three main sites of their biosynthesis, (i) developing seeds and fruits, (ii) young leaves of developing apical buds and elongating shoots and (iii) the apical regions of roots. The pathway of GA biosynthesis can be divided into three stages each of which is accomplished in a different cellular compartment.

(a) Stage I. Formation of terpenoid precursors and entkaurene in plastids:

GAs are biosynthesized from a 5-C precursor IPP. The IPP may be synthesized either in plastids or cytosol. From IPP, 10-C (GPP), 15-C (FPP) and 20-C (GGPP) precursors of terpenoids are formed by condensation of 5-C units (IPP). After the formation of GGPP, the pathway becomes specific for GAs.

GGPP is converted by two cyclization reactions through copalyl pyrophosphate into entkaurene. These reactions are catalysed by the enzymes cyclases which are located in pro-plastids and not in mature chloroplasts and in-fact constitute the first step that is specific for GAs. This step of GA biosynthesis is inhibited by compounds such as Amo-1618, Phosphon D and CCC.

(In more recent literature, various pyro-phosphorylated compounds such as IPP, DPP, FPP and GGPP etc. involved in terpenoid biosynthesis are referred to as diphosphates instead of pyrophosphates. However, their old abbreviated forms are still retained (e.g.) IPP now means isopentenylidiphosphate).

(b) Stage II. Oxidations to form GA₁₂ and GA₅₃ on ER through GA₁₂ aldehyde:

The entkaurene is transported from plastids to ER (endoplasmic reticulum). Now a methyl group on entkaurene at 19th-carbon position is oxidised to carboxylic group which is followed by contraction of ring B from 6-C to 5-C ring structure to form GA₁₂-aldehyde. GA₁₂-aldehyde is subsequently oxidised to give GA₁₂ which is precursor to all other GAs in plants. Hydroxylation of GA₁₂ at C -13 results in the formation of GA₅₃.

The enzymes catalysing the above oxidation reactions are mono-oxygenases which are located on ER and utilize cytochrome P450 in these reactions. Activity of these enzymes is inhibited by paclobutrazol and other inhibitors before GA₁₂-aldehyde

(c) Stage III. Formation of all other GAs from GA₁₂ or GA₅₃ in cytosol:

All other steps in the biosynthesis of GAs from GA₁₂ or GA₅₃ are carried out in cytosol by soluble enzymes called dioxygenases. These enzymes require molecular O₂ and 2-oxoglutarate as cosubstrates and use ferrous iron (Fe⁺⁺) and ascorbic acid as cofactors. Activity of these enzymes is inhibited by cyclohexanetriones.

Environmental factors such as temperature and photoperiod are known to affect biosynthesis of gibberellins.

4.2.4 Mode of action

- Stimulates shoot and cell elongation
- Delays senescence of leaves
- Inhibits root growth
- Inhibits adventitious root growth
- Produces seed germination
- Antagonist promotes root growth and GA reverses this
- Promotes root initiation in low concentration in pea cuttings
- Stimulates bolting and flowering in biennials
- Regulates production of hydrolytic enzymes for digesting starches
- Inhibits CK bud growth on calluses
- Inhibits bud formation
- Inhibits leaf formation
- Breaking of dormancy
- Induces extra Chlorophyll production or more efficient methods of photosynthesis (C4Photosynthesis).
- Stimulates root senescence
- Directly or indirectly induces CK at high levels
- (From Theory II) Inhibits the rate of metabolism of cells in the roots (who are not already at their lowest metabolism rates) in response to an decrease in the levels sugar and/or essential gases

4.2.4. Role of endogenous gibberellins

Effects of gibberellins on growth and development

1. Gibberellin stimulates stem growth in dwarf and rosette plants.
2. GAs regulate the transition from Juvenile to adult phases
English ivy (GA_3) and conifers ($GA_4 + GA_7$)
3. GAs influence floral initiation and sex determination.
 - (1). Primary role of GA in sex determination: to suppress stamen development.
 - a. Exposure to short days and cool nights increases endogenous GA levels in the tassel flowers 100-fold and causes feminization of the tassel flowers.
 - b. Application of GA to the tassels can also induce pistillate flowers.
 - (2). In dicots such as cucumber and spinach, GA seems to have opposite effect.
4. GAs promote fruit set.
5. GAs promote seed germination.
6. GAs have commercial applications.
 - (1). Fruit production: GAs increase the size of seedless grapes and the distance between branches and delay senescence in citrus plants, Gas.
 - (2). Malting of barley and increase sugarcane growth and sugar yield.
 - (3). Uses in plant breeding
 - a. GA_4 and GA_7 mixture is used to enhance seed production in *Pinaceae*.
 - b. GA_3 is used for *Taxodiaceae* and *Cupressaceae*.
 - b. The promotion of male flowers in cucumber.
 - c. The stimulation of bolting in biennial vegetables such as beet and cabbage.

4.2.5. Bioassay of Gibberellins:**1. Dwarf Pea:**

Seeds of dwarf pea are allowed to germinate till the formation of coleoptile. GA solution is applied to some seedlings. Others are kept as control. After 5 days, epicotyl length is measured. GA stimulates epicotyl growth with a concentration as low as 1 Nano gram.

2. Barley Endosperm:

Endosperms are detached from embryos, sterilized and allowed to remain in 1 ml of test solution for 1-2 days. There is a build-up of reducing sugars. The content of reducing sugar is proportional to gibberellin concentration. Reducing sugars are not formed in control experiment where endosperms are kept in plain water.

4.2.6. Practical applications**1. Fruit Growth:**

Application of gibberellins increases the number and size of several fruits, e.g., Grape, Tomato. The hormone creates more room by increasing the size of stalks so that fruits can grow in size. Size and shape of Apple fruits is enhanced by application of GA_4 and GA_7 mixture.

2. Parthenocarpy:

Seedless pomaceous fruits can be produced by application of gibberellins to un-pollinated flowers.

3. Malt:

Gibberellins (e.g., GA₃) increase the yield of malt from barley grains.

4. Overcoming Dormancy:

Gibberellins can be employed for breaking seed and bud dormancy. They induce germination of positively photoblastic seeds of Tobacco and Lettuce in complete darkness.

5. Delayed Ripening:

GA₇ delays senescence so that fruit can be left on the tree for longer period. It extends period of marketing. Ripening of Citrus fruits can be delayed with the help of gibberellins. This is useful in storing the fruits.

6. Flowering:

Gibberellins can be used in inducing offseason flowering in many long day plants as well as plants requiring vernalisation.

7. Sugarcane:

Spraying of sugarcane crop with gibberellins increases length of stem and yield of sugarcane to as much as 20 tonnes/acre.

8. Early Maturity:

Juvenile conifers sprayed with mixture of GA₄ and GA₇ reach maturity quite early resulting in early seed production.

4.3. Cytokinins

4.3.1. Chemistry: CKs are adenine derivatives characterized by an ability to induce cell division in tissue culture (in the presence of auxin). The most common cytokinin base in plants is zeatin.

Sites of Biosynthesis:

Cytokinin biosynthesis is through the biochemical modification of adenine. It occurs in root tips and developing seeds.

Transport: Cytokinin transport is via the xylem from roots to shoots.

4.3.2. Mode of action: The action of Cytokinins is still poorly understood and insufficient evidence exists to conclusively identify any biochemical point of action.

4.3.3. Physiological role of cytokinins:

Cell division - applications of cytokinins induce cell division in tissue culture in the presence of auxin. The presence of cytokinins in tissues with actively dividing cells (e.g., fruits, shoot tips) indicates that cytokinins may naturally perform this function in the plant. Morphogenesis - in tissue culture, Cytokinins promote shoot initiation. Growth of lateral buds - Cytokinin applications can cause the release of lateral buds from apical dominance. Leaf expansion - resulting solely from cell enlargement. This is probably the mechanism by which the total leaf area is adjusted to compensate for the extent of root growth, as the amount of CKs reaching the shoot will reflect the extent of the root system. Cytokinins delay leaf senescence. Cytokinins may enhance stomatal opening in some species.

Chloroplast development - the application of Cytokinin leads to an accumulation of chlorophyll and promotes the conversion of leukoplasts into chloroplasts.

Commercial uses:

The major use for cytokinins derives from their ability to delay senescence and maintain greenness. The artificial, highly active cytokinin, benzyladenine, is the main compound used. The treatment of holly for festive decorations enables its harvest many weeks prior to use.

Post-harvest sprays or dips are now available to prolong the storage life of green vegetables such as asparagus, broccoli, and celery.

4.4. Absciscic acid:**4.4.1. Chemistry:**

The name absciscic acid is rather unfortunate. The first name given was "abscisin II" because it was thought to control the abscission of cotton bolls. At almost the same time another group named it "dormin" for a purported role in bud dormancy. By a compromise the name absciscic acid was coined. It now appears to have little role in either abscission or bud dormancy, but we are stuck with this name. As a result of the original association with abscission and dormancy, ABA has become thought of as an inhibitor. While exogenous applications can inhibit growth in the plant, ABA appears to act as much as a promoter (e.g., storage protein synthesis in seeds) as an inhibitor, and a more open attitude towards its overall role in plant development is warranted.

Sites of Biosynthesis: ABA is synthesized from mevalonic acid in roots and mature leaves, particularly in response to water stress. Seeds are also rich in ABA which may be imported from the leaves or synthesized.

4.4.2. Mode of action

The mechanism:

- ABA binds to receptors at the surface of the plasma membrane of the guard cells.
- The receptors activate several interconnecting pathways which converge to produce
 - a rise in pH in the cytosol;
 - transfer of Ca^{2+} from the vacuole to the cytosol.
- These changes
 - stimulate the loss of negatively-charged ions (anions), especially NO_3^- and Cl^- , from the cell and also
 - the loss of K^+ from the cell.
- The loss of these solutes in the cytosol reduces the osmotic pressure of the cell and thus turgor.
- The stomata close.

Transport: ABA is exported from roots in the xylem and from leaves in the phloem. There is some evidence that ABA may circulate to the roots in the phloem and then return to the shoots in the xylem.

4.4.3. Physiological Effects:

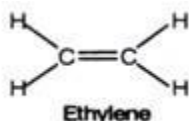
Stomatal closure - water shortage brings about an increase in ABA which leads to stomatal closure. ABA inhibits shoot growth (but has less effect on, or may promote, root growth). This may

represent a response to water stress. ABA induces storage protein synthesis in seeds. ABA counteracts the effect of gibberellin on α -amylase synthesis in germinating cereal grains. ABA affects the induction and maintenance of some aspects of dormancy in seeds. It does not, however, appear to be the controlling factor in "true dormancy" or "rest," which is dormancy that needs to be broken by low temperature or light.

4.5. Ethylene:

4.5.1. Chemistry

Ethylene (C_2H_4) with a molecule weight of 28, is a well known and simplest olefin gas and has the following structural formula,



Ethylene is flammable and highly volatile substance that readily undergoes oxidation to produce ethylene oxide. In many plant tissues, ethylene can be fully oxidised to CO_2 through ethylene oxide. It is colorless, lighter than air at room temp, and under physiological conditions and is sparingly soluble in water. Ethylene is readily absorbed by potassium permanganate ($KMnO_4$). The latter is frequently used to remove excess ethylene from the storage chambers.

Sites of Biosynthesis: Ethylene is synthesized by most tissues in response to stress. In particular, it is synthesized in tissues undergoing senescence or ripening.

Transport: Being a gas, ethylene moves by diffusion from its site of synthesis.

4.5.2. Mode of Action:

As mentioned earlier, ethylene exhibits wide range of physiological effects in plants. However, earlier steps are assumed to be similar in all cases that include,

- (i) Binding of ethylene to a receptor,
- (ii) Activation of one or more signal transduction pathways and
- (iii) Modulation of gene expression leading to cellular response.

i. In *Arabidopsis thaliana* and other plants, ethylene responses are negatively regulated by a receptor gene family.

ii. An ethylene binding receptor protein has been identified in *A. thaliana*, tomato and other plants which is known as ETR1. Four other proteins are also known that serve as ethylene receptors which are called as ETR2, ERS1, ERS2 and EIN4.

iii. Like cytokin receptor, the ethylene receptor (ETR1 & others) is also similar to bacterial two component sensor histidine kinases. It is a dimer of two polypeptides which are held together by disulphide (-S-S-) bonds and is located on ER (endoplasmic reticulum).

iv. Each polypeptide of the dimer receptor protein consists of three domains,

- (i) Amino terminal domain which spans the ER membrane at least thrice and contains the ethylene binding site,
- (ii) A middle histidine kinase domain and

- (iii) A receiver domain towards the carboxyl terminus of the polypeptide.
- v. Ethylene binds to trans membrane domain of the receptor through a copper cofactor. The latter is incorporated into the receptor protein by RAN1 protein.
- vi. Binding of ethylene to receptor inactivates a protein kinase such as CTR1 (a member of RAF family of protein kinases) in the cytosol.
- vii. Inactivation of CTR1 allows a trans-membrane protein (on ER) called EIN2 to function.
- viii. EIN2 now activates a cascade of transcription factors in the nucleus such as EIN3, ERF1 etc.
- ix. These transcription factors in turn modulate gene expression and ultimately ethylene response occurs.

4.5.3. Physiological role of ethylene

- Release from dormancy.
- Shoot and root growth and differentiation.
- Adventitious root formation.
- Leaf and fruit abscission.
- Flower induction in some plants.
- Induction of femaleness in dioecious flowers.
- Flower opening.
- Flower and leaf senescence.
- Fruit ripening.

Commercial uses:

- Ethylene enjoys a wide variety of uses, but its gaseous nature precludes its use in nonenclosed spaces.
- Ethylene itself can be used to enhance the ripening of fruits such as bananas in storage following their shipment in an unripe condition; this is of great benefit, since the green bananas are rugged and do not bruise or spoil easily.
- The tender ripe bananas can then be carried safely to market from the nearby warehouse. Recently, an ethylene-producing liquid chemical, 2 chloroethylphosphonic acid (commercially called Ethrel or Ethepon) has been introduced into commerce. This compound is sprayed onto the plant at a slightly acid pH. When it enters the cells and encounters the cytoplasm at about neutral pH, it breaks down to release gaseous ethylene.
- Numerous commercial applications for this compound have appeared, mostly in relation to the natural effects of ethylene: The most important commercial use involves enhancing latex flow in rubber trees in Southeast Asia. When a rubber tree is "tapped," the latex flows for a certain period before the cut seals and the flow stops.
- Ethepon delays the healing of the cut so that the latex flow continues for a longer period, thus yielding more latex with less tapping. Enhancement of uniform fruit ripening and coloration. This has been shown to be of particular value in field tomatoes picked at a single time by machine.
- Acceleration of fruit abscission for mechanical harvesting. This provides a potential area of use in a wide variety of fruits such as grapes, cherries, and citrus. Promotion of female flower production in cucurbits (cucumber, squash, melon) so as to increase the number of fruits produced per plant. Promotion of flower initiation and controlled ripening in pineapples.

KARPAGAM ACADEMY OF HIGHER EDUCATION
COIMBATORE - 21
DEPARTMENT OF BIOCHEMISTRY
III B.Sc BIOCHEMISTRY
BATCH: 2015 - 2018

15BCU504 - Plant Biochemistry

PART A (20 X 1 = 20 MARKS) - Online MCQ questions

SL.NO	QUESTION	OPTION 1	OPTION 2	OPTION 3	OPTION 4	ANSWER
Unit 4						
1	Auxin is found in which region of the plants?	Meristematic region	mature tissue	flowers	fruits	Meristematic region
2	Chemically auxin is	Indole pyruvic acid	Indole 3 acetic acid	Indole butyric acid	2,4 dichloro phenoxy acetic acid	Indole 3 acetic acid
3	Auxin isolated from human urine is	Indole acetic acid	auxonotriolic acid	auxonolonic acid	alpha naphthalene acetic acid	auxonotriolic acid
4	The only true natural auxin of higher plants is	Indole 3 acetic acid	alpha naphthalene acetic acid	2,4 dichloro phenoxy acetic acid	Indole butyric acid	Indole 3 acetic acid
5	The precursor of indole acetic acid is	Tyrosine	methionine	tryptophan	phenyl alanine	tryptophan
6	The enzyme involved in conversion of tryptophan to indole 3 pyruvic acid is	indole pyruvic acid decarboxylase	indole acetaldehyde decarboxylase	amino transferase	tryptophan decarboxylase	amino transferase
7	In tryptamine pathway tryptophan is converted to tryptamine by	tryptophan decarboxylase	indole pyruvic acid decarboxylase	indole acetaldehyde decarboxylase	amino transferase	tryptophan decarboxylase
8	The indole acetaldoxime pathway is characteristic of the family	cruciferae	solanacea	marvacea	malvacea	cruciferae
9	The only non indole auxin is	phenyl pyruvic acid	phenyl acetic acid	alpha naphthalene acetic acid	2,4 dichloro phenoxy acetic acid	phenyl acetic acid
10	Phenyl alanine is converted to phenyl pyruvic acid by the enzyme	aromatic amino transferase	indole pyruvic acid decarboxylase	indole acetaldehyde decarboxylase	amino transferase	aromatic amino transferase

SL.NO	QUESTION	OPTION 1	OPTION 2	OPTION 3	OPTION 4	ANSWER
11	Absorption of water is increased by	gibberellins	auxin	cytokinin	ethylene	auxin
12	Shortening of internodes and production of dwarf varieties in apple and pear is due to	gibberellins	auxin	cytokinin	ethylene	auxin
13	The hormone used widely to break seed dormancy is	gibberellins	auxin	cytokinin	ethylene	auxin
14	_____ are weed killers	gibberellins	auxin	cytokinin	ethylene	auxin
15	Early flowering and fruiting is induced by	gibberellins	auxin	cytokinin	ethylene	auxin
16	The precursor for gibberellin biosynthesis is	mevalonic acid	tyrosine	tryptophan	alanine	mevalonic acid
17	Genetic dwarfism is overcome by	gibberellins	auxin	cytokinin	ethylene	gibberellins
18	Bolting and flowering in <i>Brassica</i> is induced by	gibberellins	auxin	cytokinin	ethylene	gibberellins
19	Light induced inhibition of stem growth is increased by	gibberellins	auxin	cytokinin	ethylene	gibberellins
20	The production of parthenocarpic fruit in stone fruits is induced by	gibberellins	auxin	cytokinin	ethylene	gibberellins
21	Reduction in number of male flowers and increase in number of female flowers in cucumber is induced by	gibberellins	auxin	cytokinin	ethylene	gibberellins
22	Chemically gibberellins are	triterpenoid acids	sesquiterpenoid acids	diterpenoid acids	monoterpenoid acids	diterpenoid acids

SL.NO	QUESTION	OPTION 1	OPTION 2	OPTION 3	OPTION 4	ANSWER
23	Seed germination is promoted by	gibberellins	auxin	cytokinin	ethylene	gibberellins
24	Cytokinin is a derivative of	6 furfuryl amino purine	pyrimidine	isopentenyl pyro phosphate	geranyl pyro phosphate	6 furfuryl amino purine
25	Kinetin is formed from	adenosine	guanosine	deoxy adenosine	deoxy guanosine	deoxy adenosine
26	Cell enlargement is induced by all the hormones except	abscisic acid	auxin	cytokinin	ethylene	abscisic acid
27	Enzyme synthesis in plants is regulated by	abscisic acid	auxin	cytokinin	ethylene	cytokinin
28	Sex reversal is induced by	abscisic acid	auxin	cytokinin	ethylene	cytokinin
29	RNA synthesis in elongating zones of onion roots is stimulated by	abscisic acid	auxin	cytokinin	ethylene	cytokinin
30	Abscisic acid is a	triterpene	sesquiterpene	diterpene	monoterpene	sesquiterpenes
31	The precursor for abscisic acid biosynthesis is	mevalonic acid	tyrosine	tryptophan	alanine	mevalonic acid
32	The following hormone is a growth inhibitor	abscisic acid	auxin	cytokinin	gibberellins	abscisic acid
33	Senescence is promoted by	abscisic acid	auxin	cytokinin	ethylene	abscisic acid
34	Loss of chlorophyll and turgor is caused by	abscisic acid	auxin	cytokinin	ethylene	abscisic acid

SL.NO	QUESTION	OPTION 1	OPTION 2	OPTION 3	OPTION 4	ANSWER
35	Inhibition of gibberellins induced synthesis of α – amylase and ribonuclease is caused by	abscisic acid	auxin	cytokinin	ethylene	abscisic acid
36	Transcinnamic acid is	gibberellins	auxin	cytokinin	antiauxin	antiauxin
37	RNA synthesis is inhibited by	abscisic acid	auxin	cytokinin	ethylene	abscisic acid
38	Nuclease production is increased by	abscisic acid	auxin	cytokinin	ethylene	abscisic acid
39	_____ is a ripening hormone	abscisic acid	auxin	cytokinin	ethylene	ethylene
40	Precursor for ethylene biosynthesis is	tyrosine	tryptophan	alanine	methionine	methionine
41	Methionine is oxidatively deaminated to	methionol	methionyl phosphate	cystine	cysteine	methionol
42	The cofactor involved in ethylene biosynthesis is	ortho coumaric acid	meta coumaric acid	para coumaric acid	delta coumaric acid	para coumaric acid
43	Resistance to pathogenic infection is induced by	abscisic acid	auxin	cytokinin	ethylene	ethylene
44	The enzyme peroxidase and Polyphenol oxidase in the tissues is stimulated by	abscisic acid	auxin	cytokinin	ethylene	ethylene
45	Epinastic movements is induced by	abscisic acid	auxin	cytokinin	ethylene	ethylene
46	Flowering in pine apple is induced by	abscisic acid	auxin	cytokinin	ethylene	ethylene

SL.NO	QUESTION	OPTION 1	OPTION 2	OPTION 3	OPTION 4	ANSWER
47	Radial growth in stems and roots is induced by	abscisic acid	auxin	cytokinin	ethylene	ethylene
48	The level of endogenous auxin is regulated by	abscisic acid	auxin	cytokinin	ethylene	ethylene
49	Leaf abscission is promoted by	abscisic acid	auxin	cytokinin	ethylene	ethylene
50	Genetically dwarf plants can be made taller by	gibberellins	auxin	cytokinin	ABA	Gibberellins
51	Bio assay for gibberellins is	Avena curvature test	soybean callus test	Amylase activity test	Barley leaf disc test for chlorophyll	Amylase activity test
52	During adverse conditions, plants develop a stress hormone	ABA	IAA	ethylene	2,4 – D	ABA
53	Fruit ripening is a	reversible process	irreversible process	light controlled phenomenon	response to light stimulus	irreversible process
54	----- acts as a wound hormone	Auxin	Gibberellins	Traumatins (traumatic acid)	Cytokinins	Cytokinins
55	Cytokinins ----- the leaf senescence.	Delay	Induce	Promote	Enhance	Delay
56	One of the most important biological effects of kinetin on plants is	Respiration	Photosynthesis	cell division	Root growth	cell division
57	The following are synthetic analog of auxin molecule except one	Indole acetic acid	2, 4- dichloro phenoxy acetic acid	indolyl butyric acid	indolyl propionic acid	Indole acetic acid
58	Among the following which is antigibberellins	Phenol	Phosphan D	Kaurene	GA 36	Phosphan D

SL.NO	QUESTION	OPTION 1	OPTION 2	OPTION 3	OPTION 4	ANSWER
59	Dwarfism can be overcome by the application of	Auxin	Zeatin	Gibberellins	Ethylene	Gibberellins
60	Decarboxylation of tryptophan leads to	Tyrosine	Tryptamine	Indole acetaldehyde	IAA	Tryptamine



KARPAGAM ACADEMY OF HIGHER EDUCATION
(Deemed University Established Under Section 3 of UGC Act 1956)
Coimbatore - 641021.

(For the candidates admitted from 2015 onwards)

DEPARTMENT OF BIOCHEMISTRY

COURSE MATERIAL

STAFF NAME	:	Dr. L. HARIPRASATH	
SUBJECT	:	PLANT BIOCHEMISTRY	SUBJECT CODE : 15BCU504
SEMESTER	:	V	CLASS : III B.Sc. Biochemistry

UNIT-V

Photomorphogenesis

Photoperiodism. Photochrome-function in growth and development of plant. Biochemistry of seed germination. Senescence: Biochemical changes during senescence. Senescence process in life cycle of plants.

TEXT BOOKS

Verma, S.K., & Mohit Verma. (2013). *A Text Book of Plant Physiology, Biochemistry and Biotechnology*. (6th ed.). New Delhi. S. Chand and Co.

Bonner, J., & Varner, F.J. (2012). *Plant Biochemistry*. (3rd ed.). Burlington: Elsevier Science.

Goodwin, T.W., & Mercer, E.I. (1990). *Introduction to Plant Biochemistry*. (2nd ed.). New York, NY: Robert Maxwell.M.C Publisher.

REFERENCES

Buchanan, B.B. (2015). *Biochemistry and Molecular Biology of Plants*. New Jersey, NJ: John Wiley.

Hans-Valter Heldt. (2005). *Plant Biochemistry and Molecular Biology*. England: Oxford University Press.

Wink, M. (2010). *Functions and Biotechnology of Plant Secondary Metabolites*. (2nd ed.), London: Blackwell Publishing Ltd.,

Unit V

V. Photomorphogenesis

5.1. Photoperiodism:

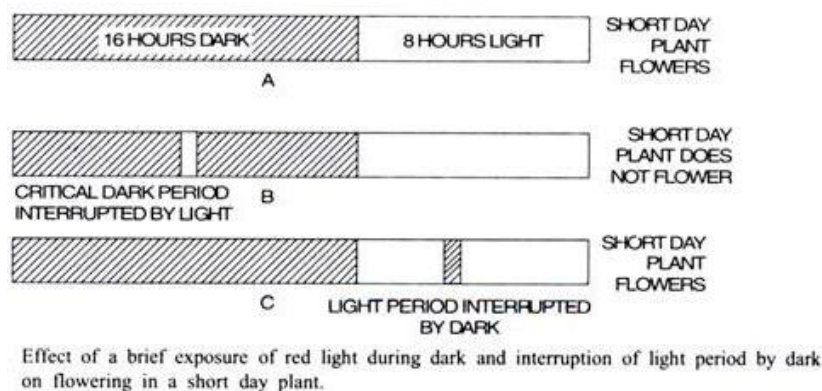
The plants in order to flower require a certain day length i.e., the relative length of day and night which is called as photoperiod. The response of plants to the photoperiod expressed in the form of flowering is called as photoperiodism.

The phenomenon of photoperiodism was first discovered by Garner and Allard (1920, 22) who observed that the Biloxi variety of Soybeans (*Glycine max*) and 'Maryland Mammoth' variety of tobacco (*Nicotianatabacum*) could be made to flower only when the daily exposure to the light was reduced below a certain critical duration and after many complex experiments concluded that 'the relative length of the day is a factor of the first importance in the growth and development of plants'. Depending upon the duration of the photoperiod, they classified plants into three categories.

(1) Short Day Plants (SDP):

These plants require a relatively short day light period (usually 8-10 hours) and a continuous dark period of about 14-16 hours for subsequent flowering. Some examples of these plants which are also known as long-night-plants are Maryland Mammoth variety of tobacco (*Nicotianatabacum*) Biloxi variety of Soybeans (*Glycine max*), Cocklebur (*Xanthium pennsylvanicum*).

i. In short day plants the dark period is critical and must be continuous. If this dark period is interrupted even with a brief exposure of red light (660-665 mμ wavelength), the short day plant will not flower



ii. Maximum inhibition of flowering with red light occurs at about the middle of critical dark period.

iii. However, the inhibitory effect of red light can be overcome by a subsequent exposure with far-red light (730-735 mμ wavelengths).

iv. Interruption of the light period by dark does not have inhibitory effect on flowering in short day plants.

v. Prolongation of the continuous dark period initiates early flowering in short day plants.

(2) Long Day Plants (LDP):

These plants require a longer day light period (usually 14-16 hours) in a 24 hours cycle for subsequent flowering. Some examples of these plants which are also called as short night plants are *Hyoscyamusniger* (Henbane) *Spinacea* (spinach) *Beta vulgaris* (Sugar beet).

i. In long day plants the light period is critical.

ii. A brief exposure in the dark period or the prolongation of the light period stimulates flowering in long day plants.

(3) Day Neutral Plants:

These plants flower in all photoperiods ranging from 5 hours to 24 hours continuous exposure. Some of the examples of these plants are tomato, cotton, sunflower, cucumber and certain varieties of peas and tobacco.

During recent years certain intermediate categories of plants have also been recognised. They are,

Long Short Day Plants:

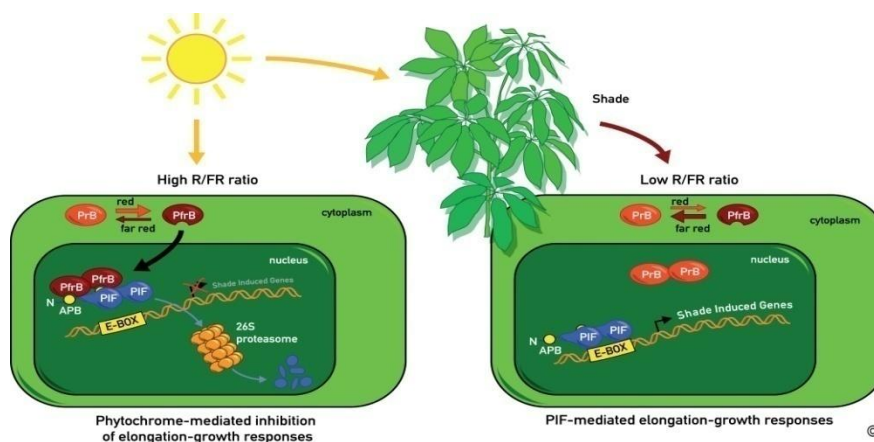
These are short day plants but must be exposed to long days during early periods of growth for subsequent flowering. Some of the examples of these plants are certain species of *Bryophyllum*.

Short-Long Day Plants:

These are long day plants but must be exposed to short days during early periods of growth for subsequent flowering. Some of the examples of these plants are certain varieties of wheat (*Triticum*) and rye (*Secale*).

5.2. Photochrome-function in growth and development of plant

Photochrome



- Phytochrome exists in two forms, P_r , which absorbs maximally at 660 nm, and P_{fr} , which absorbs maximally at 725–735 nm
- Absorption of light by either form results in phototransformation to the isomeric form.
- These properties make phytochrome a unique photoreceptor and raise important questions concerning the function of phytochrome in plants growing in the natural environment
- In the physiological experiments which have been so successful in elucidating the details of the structure and properties of the molecule, and which are beginning to provide evidence on its cellular mode of action, etiolated plants are usually used and are given abnormal irradiation treatments involving brief or prolonged exposure to radiation of restricted wavelengths.
- Such conditions do not occur in the natural environment and, therefore, such experiments provide no evidence on the role of phytochrome in plants growing in the field. Suggestions have been made that phytochrome may enable the plant to detect shading by other plants and thus induce the alteration of metabolism and development in an appropriate manner
- These proposals point out that radiant energy below 700 nm is almost completely reflected or absorbed by vegetation, whilst that between 700–800 nm (that is, the far red) is largely transmitted
- The relative enhancement of the far red will presumably alter the photoequilibrium of $P_r \rightleftharpoons P_{fr}$ in plants growing under vegetation canopies, presenting a possible mechanism for the detection of shading.
- No systematic attempts have yet been made, however, to determine the extent of the spectral energy changes in the natural environment, to correlate them with effects on phytochrome photoequilibria and to assess the possible regulatory role of phytochrome under these conditions.
- In this report we present preliminary evidence along these lines which is consistent with the above hypothesis.

5.3. Biochemistry of seed germination

Seed germination

The process of seed germination includes the following five changes or steps. Such five changes or steps occurring during seed germination are:

- (1) Imbibition
- (2) Respiration
- (3) Effect of Light on Seed Germination
- (4) Mobilization of Reserves during Seed Germination and Role of Growth Regulators
- (5) Development of Embryo Axis into Seedling.

Imbibition

- The first step in the seed germination is imbibition i.e. absorption of water by the dry seed.

- Imbibition results in swelling of the seed as the cellular constituents get rehydrated. The swelling takes place with a great force.
- It ruptures the seed coats and enables the radicle to come out in the form of primary root. Imbibition is accomplished due to the rehydration of structural and storage macromolecules, chiefly the cell wall and storage polysaccharides and proteins.
- Many seeds contain additional polysaccharides, not commonly found in vegetative tissues.
- Seeds packed dry in a bottle can crack it as they imbibe water and become swollen.

Respiration:

- Imbibition of water causes the resumption of metabolic activity in the rehydrated seed.
- Initially their respiration may be anaerobic (due to the energy provided by glycolysis) but it soon becomes aerobic as oxygen begins entering the seed.
- The seeds of water plants, as also rice, can germinate under water by utilizing dissolved oxygen.

Effect of Light on Seed Germination:

- Plants vary greatly in response to light with respect to seed germination. The seeds which respond to light for their germination are named as photoblastic. Three categories of photoblastic seeds are recognized: Positive photoblastic, negative photoblastic and non-photoblastic. Positive photoblastic seeds (lettuce, tobacco, mistletoe, etc.) do not germinate in darkness but require exposure to sunlight (may be for a brief period) for germination.
- Negative photoblastic seeds (onion, lily, Amaranthus, Nigella, etc.) do not germinate if exposed to sunlight. Non-photoblastic seeds germinate irrespective of the presence (exposure) or absence (non-exposure) of light.

Mobilization of Reserves during Seed Germination and Role of Growth Regulators

- The activated embryo cells induce the production of hormones and digestion of reserve food.
- Depending on the nature of seed, the resource may be stored chiefly in the endosperm(e.g. castor, cereal grains and other monocots) or in the cotyledons (egg. many dicotyledons such as pea, gram, bean etc.).
- The cells which are rich in proteins produce and secrete hydrolyzing enzymes.
- These enzymes bring about the digestion of the reserve foods.
- The latter are changed into sugars, amino acids and other soluble substances.
- They are translocated to the embryo.

Development of Embryo Axis into Seedling.

- On the receipt of soluble food, the cells of embryo axis undergo division and expansion.
- The radicle end of the embryo axis is the first to enlarge.
- It grows out of the seed coats and passes downwardly into the soil to establish itself as the primary root.
- The plumule also comes out of the seed and soil to establish itself as shoot.

5.4. Senescence

5.4.1. Biochemical changes during senescence

(i) Photosynthesis

- It is generally observed that the net photosynthetic rate increases as leaves grow and then declines gradually at the time of maximum leaf expansion.
- This indicates that the rate of photosynthesis is a function of individual leaf age. During the rapid phase of whole plant senescence, young and old leaves degenerate almost together.
- The rate of photosynthesis also declines with equal rapidity in young and old leaves. Simultaneous senescence of leaves occurs not only in monocarpic senescence but also in autumnal senescence of polycarpic plants.
- Although loss of chlorophyll content is a commonly used index of senescence, it does not always measure accurately either senescence or photosynthetic activity.
- Chlorophyll content usually decreases well after photosynthetic activity has begun to decline, and in some cases, chlorophyll content shows an increasing trend while the photosynthetic activity is dropping.
- Even then, chlorophyll loss during senescence offers a visual method of estimating the degree of senescence.

(ii) Respiration:

- The respiratory apparatus of senescing tissue remains active until late in senescence, and then declines rapidly.
- Loss of respiratory capacity can be a factor in the final stage of senescence, whereas there is evidence that the initial stage of senescence is not linked with respiratory decline.
- The mitochondria often swell and decrease in number, but these changes occur late in senescence.
- Amino acids have been found to accumulate, giving rise to a change in the respiratory quotient.
- A climacteric rise in respiration may occur in the senescence of both intact and detached leaves.
- From the fact that respiration decreases only late in senescence and that the lack of oxygen supply prevents the breakdown of chlorophyll and protein, it appears that cellular energy is required during senescence, possibly for the synthesis of the degradative enzymes.
- The respiratory climacteric of senescing leaves is correlated with a rise in RNA levels and an increase in protease activity.

(iii) Nitrogen Fixation and Mineral Uptake:

- Several studies have demonstrated that symbiotic nitrogen fixation in legumes directly depends upon the amount of photosynthetic available to the root nodules.
- It is interesting to note that depodding which increases the availability of photosynthetic can cause appreciable increase in nitrogen fixation and delay nodule senescence.
- In a similar way, the decline in whole plant photosynthesis parallels the decline in nitrogen fixation.
- Symbiotic nitrogen fixation decreases with the aging and senescence of leguminous plants.

- Such decline in nitrogen fixation is associated with the structural changes of individual nodules and bacteroids which is followed by their senescence.
- Root growth as well as root function usually slow early in the reproductive phase, just like nitrogen fixation, mineral uptake through the roots suffers a decline during the reproductive phase.

(iv) Protein and Nucleic Acids:

- The most basic of all events accompanying senescence is the decline in protein and nucleic acid levels.
- It has been shown that although there is a decline in protein, RNA and DNA of senescing tissues, these three metabolites do not decline at the same rate.
- There is an early gradual decline in protein and RNA about the time when vegetative growth ceases, while DNA decreases last.
- The initial decrease in protein and nucleic acid in the chloroplast involves a decrease in the major chloroplast enzyme.
- RuBP case and other enzymes which are synthesized on chloroplast ribosome.
- The loss of these enzymes is correlated with reduced ability of the chloroplast to synthesize protein and RNA coupled with a reduction in the number of chloroplast polysomes.
- The decline in protein and nucleic acids takes place in two stages. The period of gradual decline, characteristic of initial stage of senescence, is followed by more rapid senescence and rapid decline when chlorosis or leaf yellowing starts.
- During the rapid decline, protein and nucleic acid degradation is accelerated in both the chloroplasts and the cytoplasm.
- In addition, the synthetic capacity in the whole cell decreases.
- During this period, large quantities of hydrolytic enzymes like protease, RNase, phosphatase and chlorophyllase are synthesized on cytoplasmic ribosome.
- Thus, the prime candidates for central centres for a marked decline in protein and nucleic acids seem to be due to (a) release or activation of hydrolases present in the vacuoles or lysosomes, (b) a possible decrease in protease inhibitor, and (c) a decrease in protein and nucleic acid synthesis.
- In the whole plant, the senescing leaves before their abscission are depleted of materials like amino acids, sugars which are transported to other parts.

(v) Membranes and Organelles:

- Of all the cell organelles, the chloroplast shows the earliest symptoms of physiological decline.
- Decrease in RuBP case activity is noticed first with a corresponding early loss of chloroplast ribosomes and this precedes the decrease in cytoplasmic ribosomes.
- In later phases of senescence, however, pronounced ultra-structural changes in the thylakoids occur.

- Other changes occurring at this time include swelling, vesiculation and disappearance of endoplasmic reticulum (ER) and Golgi bodies together with the loss of ER-associated ribosome.
- The galactolipids and sulpho-lipids, the constituents of chloroplast lipids, decrease before the phospholipids that predominate in the non-chloroplast membranes, suggesting that the biochemical decline may occur relatively early in chloroplasts.
- Although the changes in chloroplasts occur early, they remain intact till the time when the tonoplast ruptures.
- As soon as the tonoplast disintegrates, the hydrolytic enzymes, acids and toxic compounds are released in the vacuole and this triggers the cellular degradation.
- In the final stage of cellular degradation, the plasma lemma disintegrates and the nucleus undergoes massive alteration, whereas the mitochondria may persist with intact shape even at this stage.
- It is generally acknowledged that membranes seem to play an important role in plant senescence.

(vi) Nutrient Deficiency Syndrome:

- Nutrient deficiencies may be important in the senescence programme of particular organs or the whole plant.
- This concept finds support in the senescence of monocarpic plants where the emergence of reproductive structures like flowers and fruits seems to impose a great demand on the vegetative body, thus causing nutrient deficiency.
- In such cases, the life of the plant can be spared or at least death can be delayed by removal of the reproductive structures, obviously, through avoidance of nutrient deficiency.

5.4.2. Senescence process in life cycle of plants

Plant senescence

Plant senescence is the study of aging in plants. It is a heavily studied subject just as it is in the other kingdoms of life. Plants, just like other forms of organisms, seem to have both unintended and programmed aging. Leaf senescence is the cause of autumn leaf color in deciduous trees.



The autumn senescence of Oregon Grape leaves is an example of programmed plant senescence.

Programmed senescence

Programmed senescence seems to be heavily influenced by plant hormones. The hormones abscisic acid and ethylene are accepted by most scientists as the main causes, but at least one source believes gibberellins and brassinosteroids are equally responsible. Cytokinins help to maintain the plant cell but when they are withdrawn or if the cell cannot receive the cytokinin it may then undergo apoptosis or senescence.

Annual versus perennial benefits - theory

Some plants have evolved into annuals which die off at the end of each season and leave seeds for the next, whereas closely related plants in the same family have evolved to live as perennials. This may be a programmed "strategy" for the plants.

The benefit of an annual strategy may be genetic diversity, as one set of genes does continue year after year, but a new mix is produced each year. Secondly, being annual may allow the plants a better survival strategy, since the plant can put most of its accumulated energy and resources into seed production rather than saving some for the plant to overwinter, which would limit seed production.

Conversely, the perennial strategy may sometimes be the more effective survival strategy, because the plant has a head start every spring with growing points, roots, and stored energy that have survived through the winter. In trees for example, the structure can be built on year after year so that the tree and root structure can become larger, stronger, and capable of producing more fruit and seed than the year before, out-competing other plants for light, water, nutrients, and space.

Plant self pruning - theory

There is a speculative hypothesis on how and why a plant induces part of itself to die off. The theory holds that leaves and roots are routinely pruned off during the growing season whether they are annual or perennial. This is done mainly to mature leaves and roots and is for one of two reasons; either both the leaves and roots that are pruned are no longer efficient enough nutrient acquisition-wise or that energy and resources are needed in another part of the plant because that part of the plant is faltering in its resource acquisition.

- Poor productivity reasons for plant self pruning - the plant rarely prunes young dividing meristematic cells, but if a fully grown mature cell is no longer acquiring nutrients that it should acquire, then it is pruned.
 - Shoot efficiency self pruning reasons - for instance, presumably a mature shoot cell must on average produce enough sugar, and acquire enough oxygen and carbon dioxide to support both it and a similar sized root cell. Actually, since plants are obviously interested in growing it is arguable, that the "directive" of the average shoot cell, is to "show a profit" and produce or acquire more than enough sugar and gases than is necessary to support both it and a similar sized root cell. If this "profit" isn't shown, the shoot cell is killed off and resources are redistributed to "promising" other young shoots or leaves in the hope that they will be more productive.
 - Root efficiency self pruning reasons - similarly a mature root cell must acquire on average, more than enough minerals and water needed to support both it and a similar sized shoot cell that does not acquire water and minerals. If this does not happen, the root is killed off and resources sent to new young root candidates.
- Shortage/need-based reason for plant self pruning - this is the other side of efficiency problems.
 - Shoot shortages - if a shoot is not getting enough root derived minerals and water, the idea is that it will kill part of itself off, and send the resources to the root to make more roots.
 - Root shortages - the idea here is that if the root is not getting enough shoot derived sugar and gases it will kill part of itself off and send resources to the shoot, to allow more shoot growth.

This is an oversimplification, in that it is arguable that some shoot and root cells serve other functions than to acquire nutrients. In these cases, whether they are pruned or not would be "calculated" by the plant using some other criteria. It is also arguable that, for example, mature nutrient-acquiring shoot cells would have to acquire more than enough shoot nutrients to support both it and its share of both shoot and root cells that do not acquire sugar and gases whether they are of a structural, reproductive, immature, or just plain, root nature.

The idea that a plant does not impose efficiency demands on immature cells is that most immature cells are part of so called dormant buds in plants. These are kept small and non-dividing until the plant needs them. They are found in buds, for instance in the base of every

Hormonal induction of senescence - theory

There is not a lot of theory on how plants induce themselves to senesce, although it is reasonably widely accepted that some of it is done hormonally. Plant scientists generally concentrate on ethylene and abscisic acid as culprits in senescence, but neglect gibberellin and brassinosteroid which inhibits root growth if not causing actual root pruning. This is perhaps because roots are below the ground and thus harder to study.

1. Shoot pruning - it is now known that ethylene induces the shedding of leaves much more than abscisic acid. ABA originally received its name because it was discovered to have a role in leaf abscission. Its role is now seen to be minor and only occurring in special cases.
 - Hormonal shoot pruning theory - a new simple theory says that ethylene induces senescence in leaves due to a run away positive feedback mechanism. What supposedly happens is that ethylene is released by mostly mature leaves under water and or mineral shortages. The ethylene acts in mature leaf cells however, by pushing out minerals, water, sugar, gases and even the growth hormones auxin and cytokinin (and possibly salicylic acid in addition). This causes even more ethylene to be made until the leaf is drained of all nutrients.
2. Root pruning - the concept that plants prune the roots in the same kind of way as they abscise leaves, is not a well discussed topic among plant scientists, although the phenomena undoubtedly exists. If gibberellin and brassinosteroid are known to inhibit root growth it takes just a little imagination to assume they perform the same role as ethylene does in the shoot, that is to prune the roots too.
 - Hormonal root pruning theory - in the new theory just like ethylene, GA/BA are seen both to be induced by sugar and gas shortages in the roots, and to push sugar and gases, as well as minerals, water and the growth hormones out of the root cell causing a positive feedback loop resulting the emptying and death of the root cell.
3. Parallels to cell division - the theory, perhaps even more controversially, asserts that just as both auxin and cytokinin seem to be needed before a plant cell divides, in the same way perhaps ethylene and GA/BA are needed before a cell would senesce.
4. Discussion of the complete mechanism - what really may occur is that because ethylene pushes out all nutrients out of the shoot cell including sugar and gases, eventually this causes a shortage of these nutrients in the organ (the shoot) which is supposed to procure them. This shortage leads to GA/BA synthesis in the rapidly declining shoot cell, and this simply adds fuel to the fire.

5. A role for abscisic acid - finally a question may be to ask, what is the role of abscisic acid, the hormone which was first thought to be the primary mover in this department? According to author of this theory, ABA is induced when plant cells are encountering stress other than that of a nutrient shortage kind. In this case a senescing cell experiencing a drain of nutrients may experience a strain which causes it to produce ABA. Indeed it may not be that ethylene and GA/BA alone are needed for programmed cell senescence, but that all three are needed.

Similarly, auxin and cytokinin may not be enough for plant cell division alone, but a proposed compliment of ABA, SA, may be needed in addition.

Senescence Process in Plants

Senesce processes are tightly controlled processes in which the sequence of events is usually highly ordered until the terminal stages (death) are underway. Various factors which control the processes of senescence in different organs during life cycle of a flowering plant.

- Senescence of root: which also includes senescence of vascular tissues, root cap and hairs, is regulated by auxins and cytokinins.
- Senescence of cotyledons: is regulated by axis, shoot, light fruit, root and all hormones.
- Senescence of leaf: which also includes senescence of abscission zone, is regulated by fruit, root, light, all hormones and ethylene.
- Senescence of stem: which also includes senescence of vascular tissues, is regulated by auxins, cytokinins and sugars.
- Senescence of fruit: which also includes senescence of seeds is controlled by all hormones and ethylene.
- Senescence of flower: is regulated by pollination, ABA (Absciscic acid), cytokinins and ethylene.
- Senescence of apex: is regulated by fruits, day-length and gibberellins.
- Senescence of whole plant (monocarpism): is regulated by fruits, roots, day-length, auxins, ABA (Absciscic acid) and cytokinins.

KARPAGAM ACADEMY OF HIGHER EDUCATION
COIMBATORE - 21
DEPARTMENT OF BIOCHEMISTRY
III B.Sc BIOCHEMISTRY
BATCH: 2015 - 2018

15BCU504 - Plant Biochemistry

PART A (20 X 1 = 20 MARKS) - Online MCQ questions

SL.NO	QUESTION	OPTION 1	OPTION 2	OPTION 3	OPTION 4	ANSWER
Unit 5						
1	The response of a plant to the relative length of light and dark period is called as	phytochrome	photoperiod	cytochrome	photoreceptor	photoperiod
2	The protein pigment that absorb red and far red light most strongly is	cytochrome	phytochrome	photochlorophyllide	bacterial chlorophyllide	phytochrome
3	Coumarin inhibits	seed germination	senescence	flowering	fruiting	seed germination
4	A Study of aging in plants is called as	dormancy	senescence	ripening	fruiting	senescence
5	The photoperiod required to induce flowering for Maryland Mammoth tobacco is	12 hours	15 hours	10 hours	8 hours	12 hours
6	In phytochrome Pr form is converted to Pfr form at the wavelength	730-735 nm	660-665 nm	440-455 nm	540-555 nm	660-665 nm
7	The seeds requiring single exposure of light for germination are called	non-photoblastic seeds	positive photoblastic seeds	negative photoblastic seeds	photoclastic seeds	positive photoblastic seeds
8	Senescence of detached leaves can be delayed by the use of	auxin	gibberellins	cytokinin	abscisic acid	cytokinin
9	<i>Capsicum annum</i> is a	short day plant	long day plant	photo-neutral plant	intermediate plant	photo-neutral plant
10	In phytochrome Pfr form is converted to Pr form at the wavelength	730-735 nm	660-665 nm	440-455 nm	540-555 nm	730-735 nm

SL.NO	QUESTION	OPTION 1	OPTION 2	OPTION 3	OPTION 4	ANSWER
11	The method employed in softening or weakening the seed coat is called	stratification	scarification	after-ripening	alternating temperature	scarification
12	Senescence is induced by the following hormones except	abscisic acid	ethylene	cytokinin	gibberellins	cytokinin
13	<i>Hibiscus syriacus</i> is a	long day plant	short day plant	intermediate plant	photo-neutral plant	long day plant
14	Phytochrome is a	lipoprotein	chromoprotein	glycoprotein	nucleoprotein	chromoprotein
15	Seed germination is induced by the chemical	CMU	DCMU	thiourea	guanidinium	thiourea
16	Rice is an example of	whole plant senescence	sequential senescence	shoot senescence	simultaneous senescence	whole plant senescence
17	Rice is a	long day plant	short day plant	intermediate plant	photo-neutral plant	short day plant
18	The active form of phytochrome is	Pr	Pfr	la	lb	Pfr
19	Respiration rate is increased during	senescence	flowering	fruiting	seed germination	seed germination
20	An example of a plant undergoing shoot senescence is	rice	wheat	banana	mustard	banana
21	The photoperiod required to induce flowering for <i>Xanthium strumarium</i> is called	12 hours	15.5 hours	10 hours	18 hours	15.5 hours
22	The photo period of short day plants is	less than 12 hours	less than 10 hours	less than 15 hours	less than 5 hours	less than 12 hours

SL.NO	QUESTION	OPTION 1	OPTION 2	OPTION 3	OPTION 4	ANSWER
23	<i>Xanthium pensylvanicum</i> is a	short day plant	long day plant	photo neutral plant	intermediate plant	short day plant
24	Chrysanthemum is a	short day plant	long day plant	photo neutral plant	intermediate plant	short day plant
25	Soybean is a	short day plant	long day plant	photo neutral plant	intermediate plant	short day plant
26	The photoperiod required by long day plants is	more than 12 hours	more than 10 hours	more than 15 hours	more than 5 hours	more than 12 hours
27	<i>Hyoscyamus</i> is a	short day plant	long day plant	photo neutral plant	intermediate plant	long day plant
28	<i>Spinacea oleracea</i> is a	short day plant	long day plant	photo neutral plant	intermediate plant	long day plant
29	<i>Hibiscus syriacus</i> is a	short day plant	long day plant	photo neutral plant	intermediate plant	long day plant
30	<i>Anethum graveolens</i> is a	short day plant	long day plant	photo neutral plant	intermediate plant	long day plant
31	<i>Lycopersicum esculentum</i> is a	short day plant	long day plant	photo neutral plant	intermediate plant	photo neutral plant
32	<i>Capsicum annum</i> is a	short day plant	long day plant	photo neutral plant	intermediate plant	photo neutral plant
33	The plants require a photoperiod of 12 to 15 hours for flowering is called	short day plant	long day plant	photo neutral plant	intermediate plant	intermediate plant
34	<i>Mechania</i> is a	short day plant	long day plant	photo neutral plant	intermediate plant	intermediate plant

SL.NO	QUESTION	OPTION 1	OPTION 2	OPTION 3	OPTION 4	ANSWER
35	<i>Bryophyllum</i> is a	short day plant	long day plant	photo neutral plant	long short day plant	long short day plant
36	Pr form is converted to Pfr form at	730-735 nm	660-665 nm	450-550 nm	400-500 nm	660-665 nm
37	Pfr form is converted to Pr form at	730-735 nm	660-665 nm	450-550 nm	400-500 nm	730-735 nm
38	The pigment responsible for the inhibition of flowering in short day plants and stimulation of flowering in long day plants is called	Chlorophyll	carotenoid	xanthophylls	phytochrome	phytochrome
39	Inactive form of phytochrome is	Pr	Pfr	la	lb	Pr
40	Active form of phytochrome is	Pr	Pfr	la	lb	Pfr
41	The conversion of Pfr to Pr is accelerated by	Dithionite	thiourea	CMU	DCMU	Dithionite
42	The conversion of Pr to Pfr is	Oxidation reduction process	decarboxylation process	transamination process	deamination process	Oxidation reduction process
43	Phytochrome is a	lipoprotein	nucleoprotein	chromoprotein	glycoprotein	chromoprotein
44	Number of disulphide linkage in in one phytochrome molecule is	3	6	8	2	6
45	The light absorbing property of phytochrome is due to	protein	amino acids	chromophore	disulphide linkage	chromophore
46	Stimulation of flowering in long day plants during day time is due to the accumulation of	Pr	Pfr	la	lb	Pfr

SL.NO	QUESTION	OPTION 1	OPTION 2	OPTION 3	OPTION 4	ANSWER
47	Inhibition of flowering in short day plants during day time is due to the accumulation of	Pr	Pfr	la	lb	Pfr
48	In short day plants flowering is induced by Pr form during	light period	dark period	in presence of sun light	only in evening	dark period
49	In lettuce and tobacco seeds seed germination is induced by	auxin	gibberellins	cytokinin	ethylene	gibberellins
50	The following chemicals induce seed germination except	KNO ₃	thiourea	ethylene	CMU	CMU
51	Seed germination is induced by	blue light	red light	yellow light	green light	red light
52	During germination the starch in cereal endosperm is converted to	amylase	amylopectin	sucrose	maltose	maltose
53	In cotton seeds lipase synthesis is triggered by	auxin	gibberellins	cytokinin	ethylene	gibberellins
54	The activity of peptidase is stimulated by the endogenous hormone	auxin	GA	cytokinin	ethylene	GA
55	Reserve pool of phosphorus in seeds is	phytin	inorganic phosphate	ortho phosphoric acid	meta phosphoric acid	phytin
56	Lotus seeds can survive for	200 years	400 years	50 years	5 years	400 years
57	The optimum temperature for seed germination is	25-35°C	40-50°C	15-20°C	60-75°C	25-35°C
58	Senescence of detached leaves can be delayed by the use of	auxin	gibberellins	cytokinin	ethylene	cytokinin

SL.NO	QUESTION	OPTION 1	OPTION 2	OPTION 3	OPTION 4	ANSWER
59	One of the following statement is false about senescence	cytokinin delay senescence	senescence is accompanied by losses of chlorophyll	gibberellins retard leaf senescence	senescence does not occur due to nutrient competition	senescence does not occur due to nutrient competition
60	The response of plant to the relative length of light and dark period is called	photoperiodism	vernalization	germination	dormancy	photoperiodism

Reg. No.....

[15BCU504]

KARPAGAM UNIVERSITY

Karpagam Academy of Higher Education
(Established Under Section 3 of UGC Act 1956)
COIMBATORE – 641 021
(For the candidates admitted from 2015 onwards)

B.Sc., DEGREE EXAMINATION, NOVEMBER 2017

Fifth Semester

BIOCHEMISTRY

PLANT BIOCHEMISTRY

Time: 3 hours

Maximum : 60 marks

PART – A (20 x 1 = 20 Marks) (30 Minutes)
(Question Nos. 1 to 20 Online Examinations)

PART B (5 x 8 = 40 Marks) (2 ½ Hours)
Answer ALL the Questions

21. a. Elaborate the various types of plastids and their functions?
Or
b. Explain the structure and functions of plant cell wall in detail.
22. a. Describe the structure and functions of photosynthetic apparatus.
Or
b. Explain the Hatch-Slack pathway with a neat sketch.
23. a. Describe phosphorus and carbon cycle.
Or
b. What are micronutrients? Explain the functions and deficiency manifestations of manganese, molybdenum and zinc.
24. a. Outline the chemistry, biosynthesis and mode of action of gibberellins.
Or
b. Write a note on the chemistry, mode of action and physiological role of cytokinins.

25. a. Discuss the metabolic changes during seed germination.

Or

b. Discuss the functions of phytochrome in plant growth and development.

Reg. No.....

[12BCU504]

KARPAGAM UNIVERSITY

(Under Section 3 of UGC Act 1956)

COIMBATORE – 641 021

(For the candidates admitted from 2012 onwards)

B.Sc. DEGREE EXAMINATION, NOVEMBER 2014

Fifth Semester

BIOCHEMISTRY

PLANT BIOCHEMISTRY

Time: 3 hours

Maximum : 100 marks

PART – A (15 x 2 = 30 Marks)

Answer ALL the Questions

1. Write any three functions of plastids.
2. What is imbibition?
3. Define: Passive absorption.
4. Describe phycobillin.
5. List out stomata function.
6. What is dark reaction?
7. Define: denitrification.
8. How is caffeine synthesized?
9. What are glycolipids? Give two examples.
10. How are gibberellins synthesized?
11. What are phytochromes?
12. List out the factors influencing endogenous growth?
13. What is germination?
14. Define: Photoperiodism.
15. Short note on senescence.

PART B (5 X 14= 70 Marks)

Answer ALL the Questions

16. a. Summarize types and functions of plastids.
Or
b. Make an account of active and passive absorption mechanism in water.
17. a. Compare the mechanism of Photosystem I & Photosystem II.
Or
b. Explain the assimilation: CO₂ assimilated by C₃ plants?

18. a. Discuss on Sulphite reduction and Sulphur cycle.

Or

b. Explain the Nitrogen cycle and its mechanism

19. a. Explain in detail the biosynthesis, mode of action and physiological role of Gibberellins.

Or

b. Explain in detail the biosynthesis, mode of action and physiological role of abscisic acid.

20. Compulsory : -

Discuss in detail the factors influencing the seed germination.

Reg. No.....

[14BCU504A]

KARPAGAM UNIVERSITY
Karpagam Academy of Higher Education
(Established Under Section 3 of UGC Act 1956)
COIMBATORE - 641 021
(For the candidates admitted from 2014 onwards)

B.Sc., DEGREE EXAMINATION, NOVEMBER 2016
Fifth Semester

BIOCHEMISTRY

PLANT BIOCHEMISTRY

Time: 3 hours

Maximum : 60 marks

PART - A (20 x 1 = 20 Marks) (30 Minutes)
(Question Nos. 1 to 20 Online Examinations)

PART B (5 x 8 = 40 Marks) (2 ½ Hours)
Answer ALL the Questions

21. a. Explain the structure and functions of cell wall.
Or
b. Elaborate the mechanisms involved in water absorption in plants.
22. a. Describe noncyclic photophosphorylation with neat sketch.
Or
b. What is the end product of Calvin cycle? Discuss the steps involved.
23. a. Describe in detail the symbiotic nitrogen fixation.
Or
b. Explain the functions and deficiency symptoms of calcium and iron in plants.
24. a. Outline the chemistry, mode of action and any two bioassay and applications of auxin.
Or
b. Write elaborate notes on plant growth inhibitors and retardants.
25. a. Discuss the biochemical changes during senescence.
Or
b. Explain the role of phytochrome in plants.