PHOTOSYNTHESIS IN HIGHER PLANTS

INTRODUCTION

- Green plants synthesise their food by photosynthesis and all other organisms depend on plants for their needs.
- Photosynthesis is a physico-chemical process by which plants use light energy to drive the synthesis of organic compounds.
- The use of energy from sunlight by plants doing photosynthesis is the basis of life on Earth
- Photosynthesis is important due to two reasons : (a) It is the primary source of all food on earth and (b) It is also responsible for the release of oxygen into the atmosphere.

WHAT DO WE KNOW?

- Experiment for starch formation on variegated leaf or a leaf that was partially covered with black paper & exposed to light showed that photosynthesis occurred only in green part of leaves in the presence of light
- Experiment where a part of leaf is enclosed in a test-tube with some KOH soaked cotton (which absorbs CO_2), while other half is exposed to air and set-up kept in light proved that CO_2 is needed for photosynthesis.

EARLY EXPERIMENTS

1) Joseph Priestley

Using a burning candle, a mouse, mint plant and a bell jar for closed space, hypothesised that plants restore the air whatever burning candles or breathing animals remove

(2) Jan Ingenhousz

In an elegant experiment with an aquatic plant, showed that in bright sunlight plants produce oxygen.

(3) Julius Von Sachs

Found that glucose is made in green plant parts and stored as starch

(4) T W. Engelmann

Using a prism, green alga *Cladophora* and aerobic bacteria, described the action spectrum of photosynthesis, which roughly resembles the absorption spectrum of chlorophyll- a and b.

(5) Cornelius Van Niel

- Demonstrated that photosynthesis Is essentially a light dependent reaction in which hydrogen from suitable oxidisable compound reduces CO_2 to carbohydrates.
- H_2S is hydrogen donor for purple & green sulphur bacteria H_2O the hydrogen donor in green plants which is oxidised to O_2.
- The oxidation product is sulphur or sulphate in purple & green sulphur bacteria and not O_2. Hence it Was inferred that O_2 evolved by green plants comes from H_2O and not from CO_2. This was later proved by using radioisotopic techniques. The correct equation, for the overall process.

$$6CO_2 + 12H_2O \xrightarrow{\text{Light}} C_6H_{12}O_6 + 6H_2O + 6O_2$$

WHERE DOES PHOTOSYNTHESIS TAKE PLACE

- In green parts of the plants, mainly in the mesophyll cells in the leaves, which have large number of chloroplasts.
- Usually the chloroplasts align themselves along the walls of mesophyll cells to get optimum quantity of the incident light.

CHLOROPLAST ALIGNMENT

PARALLEL

In low or optimum light intensity to get maximum incident light

PERPENDICULAR

In extremely high light intensity to avoid photo-oxidation.

There is a clear division of labour within the chloroplast.

CHLOROPLAST

MEMBRANOUS SYSTEM

- (Grana + Stromal lamellae)
- Responsible for trapping light & synthesis of ATP & NADPH
- Directly light driven, called light reaction (photochemical reactions)

STROMA

- Enzymatic reactions to synthesise sugar, which in turn forms starch, takes place
- Dependent on products of light reactions (ATP & NADPH)
- By convention called dark reactions (Carbon reactions)

However, this does not to mean that the dark reaction occur in darkness or that they are not light-dependent

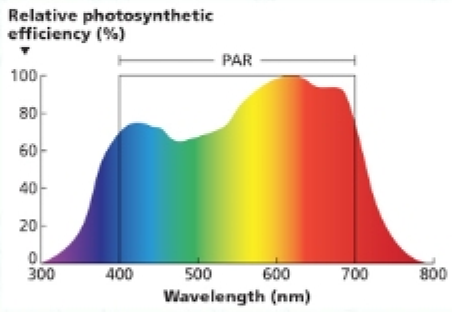
HOW MANY TYPES OF PIGMENTS ARE INVOLVED IN PHOTOSYNTHESIS

Leaf-pigments of any green plant can be separated through paper chromatography

The colour in leaves is due to four pigments, that have the ability to absorb light, at specific wavelengths

COLOUR OF THE PIGMENTS IN THE CHROMATOGRAPHY

i. Chlorophyll-a = Bright or blue green
ii. Chlorophyll-b = Yellow green
iii. Xanthophyll = Yellow
iv. Carotenoids = Yellow to yellow- orange



- The wavelength of light at which there is maximum absorption by chlorophyll-a i.e.. in blue and red regions, also shows higher rate of photosynthesis.
- Hence, we can conclude that Chl-a is the chief pigment associated with photosynthesis.
- Chl-b, carotenoids and xanthophyll are accessory pigments. They absorb light and transfer the energy to Chl-a. They enable a wider range of wavelength of incoming light to be utilised for photosynthesis and also protect chlorophyll-a from Photo-oxidation.

WHAT IS LIGHT REACTION?

- Light reactions or the Photochemical phase include
(a) Light absorption (b) Water splitting
(c) Oxygen release, and (d) ATP and NADPH formation

- Several protein complexes are involved in the process

- The pigments are organised into two photosystems

PHOTO SYSTEM

PS-I
- LHC-I (Hundreds of pigments)
- Reaction centre → Absorption peak at 700nm(P_{700})

One molecule of Chl-a

PS-II
- LHC-II
- Reaction centre → Absorption peak at 680 nm (P_{680})

Named the sequence of their discovery and not in the sequence of their function.

SPLITTING OF WATER

PS-II continuously supplies electrons which becomes available by splitting of water

Water splitting complex is associated with PS-II which itself is physically located on inner side of membrane of thylakoid

Water split into $2H^+ + 1/2\ [O_2]$ + electrons

This produces oxygen, one of the net products of photosynthesis

THE ELECTRON TRANSPORT

- The whole scheme of transfer of electrons starting from PS-II → uphill to the acceptor → down the ETC to PS-I → Excitation of electrons → transfer to another acceptor → finally downhill NADP+ → reducing it to NADPH → H^+ is called the Z-scheme due to its characteristic shape.
- This shape is formed when all the carriers are placed in a sequence on a redox potential scale.

CYCLIC AND NON-CYCLIC PHOTO-PHOSPHORYLATION

- When both PS-1 and PS-II are involved the process is non-cyclic producing ATP, NADPH+ H^+ and oxygen
- When only PS-I is functional, cycle flow of electron takes place to produce only ATP.
- A possible location for cyclic flow is the stroma lamellae membranes which lack PS-II and NADP reductase enzyme.

Cyclic photo-phosphorylation also occurs when light only of wavelengths beyond 680 nm are available for excitation

The membrane or lamellae of the grana have both PS I and PS-II

CHEMIOSMOTIC HYPOTHESIS

- ATP synthesis during photosynthesis is linked to the development of a proton gradient across the membranes of thylakoid and protons accumulate in the lumen of thylakoids
- The proton gradient is caused by:

(a) Protons or hydrogen ions produced by splitting of water, accumulate in the lumen of the thylakoids.

(b) The primary acceptor of electron located towards outer side of membrane transfers its electron to a H carrier, which removes a proton from stroma while transporting an electron, to the electron carrier on the inner side of membrane

(c) The NADP reductase enzyme located on stroma side of membrane removes protons from stroma, while reducing $NADP^+$ to $NADPH + H^+$.

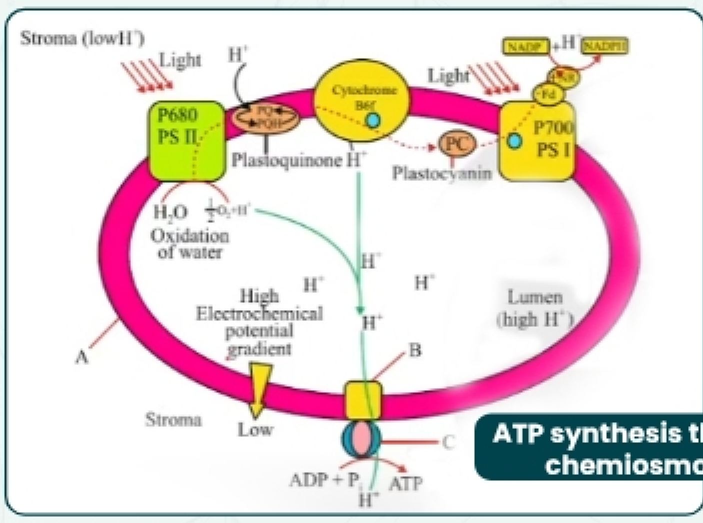
- Within chloroplast, protons decrease in stroma and accumulate in lumen. This creates a proton-gradient across thylakoid membrane as well as a measurable decrease in pH in the lumen.
- Breakdown of this gradient leads to synthesis of ATP. When protons move across the membrane to the stroma through transmembrane channel of the CF_0 of the ATP synthase.

ATP SYNTHASE

CF_0 = Embedded in the thylakoid membrane. A transmembrane channel for facilitated diffusion of protons

CF_1 = Protrudes on outer surface of thylakoid membrane on the side that faces stroma. It synthesize ATP

- Chemiosmosis requires - a membrane, a proton pump, a proton gradient and ATP synthase



ATP synthesis through chemiosmosis

WHERE ARE THE ATP AND NADPH USED?

Of the products of light reaction- ATP, NADPH and O_2. O_2 diffuses out of chloroplast while ATP and NADPH are used to synthesise sugars in the biosynthetic phase of photosynthesis Melvin Calvin used radioactive ^{14}C in algal photosynthesis studies to discover the first CO_2 fixation product, the 3-C organic acid (3-PGA) (C_3- pathway).

In another group of plants, the first stable product was 4 carbon, oxaloacetic acid OAA(C4-pathway)

THE CALVIN CYCLE

- The Calvin cycle occurs in all photosynthetic plants whether they have C_3 or C_4 (or any other) pathways.
- Calvin cycle can be described under three stages.

CARBOXYLATION: Most crucial step

$$\underset{(5C)}{\textbf{RuBP}} \xrightarrow[CO_2 + H_2O]{\text{RuBisCo}} \underset{(3C)}{\textbf{2 x 3 - PGA}}$$

REDUCTION: A series of reactions that lead to formation of glucose. Utilises 2 ATP and 2 NADPH per CO_2. (The fixation of $6CO_2$, and 6 turns of the cycle are needed to form one molecule of glucose from the pathway)

REGENERATION: Regeneration of RUBP is crucial for the cycle to continue. This step require one ATP so, to produce one molecule of glucose in Calvin cycle an input of $6CO_2$, 18 ATP & 12 NADPH are required.

THE C_4-PATHWAY

Plants adapted to dry tropical regions have the C_4-pathway.

C_4-plants are special: They have special type of leaf anatomy, tolerate higher temperatures show response to high light intensities, lack photorespiration & have greater biomass productivity.

C_4-plants have leaves showing kranz anatomy the, particularly have large cells around the vascular bundles, which may form several layers and are called bundle sheath cells characterised by having a large number of chloroplasts, thick walls impervious to gaseous exchange & no intercellular spaces.

The pathway is cyclic & called the Hatch and Slack Pathway. It is partly completed in mesophyll & partly in bundle sheath cell.

MESOPHYLL CELL	BUNDLE¬SHEATH CELLS
(1) Primary CO_2 acceptor is a 3-C compound PEP (2) Enzyme for this fixation is PEPcase. (3) Lacks RuBisCO (4) C_4-acid formed is OAA which forms malic acid or aspartic acid and transported to bundle sheath cells.	(1) Malic Acid/Aspartic Acid → CO_2 → Enters – Calvin cycle a pathway common to all plants. (1) Malic Acid/Aspartic Acid → 3-Carbon molecule → Transported to mesophyll & converted to PEP (2) Rich in RuBisCO, but lack PEP case.

PHOTORESPIRATION

- RuBisCo has the active site that can bind to both CO_2 and O_2. This binding is competitive. It is the relative concentration of O_2 and CO_2 that determines which of the two will bind to the enzyme.
- In photo-respiration there is neither synthesis of sugars, nor of ATP. It results in release of CO_2 with utilisation Of ATP.
- RuBisCo has a much greater affinity for CO2, when the CO_2 & O_2 is nearly equal than for O_2.
- The biological function of photorespiration is not known yet.
- In C_3-plants some O_2 does bind to RuBisCO, and hence CO_2 fixation is decreased, due to the following reaction.

  RuBP + O_2 $\longrightarrow$ 3PGA (3C) + 2-phosphoglycolate (2C)
- In C_4-plants photo-respiration does not occur, as they have a mechanism that increases the concentration of CO_2 at the enzyme site. This ensures that the RuBisCO functions as a carboxylase minimising the oxygenase activity.

FACTORS AFFECTING PHOTOSYNTHESIS

Photosynthesis is under the influence of severs' factors both internal (plant) & external

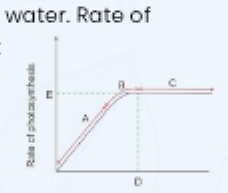
Internal Factors

• The plant factors include the number, size, age & orientation of leaves, mesophyll cells and chloroplasts, internal CO_2 concentration & the amount of chlorophyll.

• The plant or internal factors are dependent on the genetic predisposition & growth of the plant.

External factors

include availability of sunlight, temperature CO_2 concentration and water. Rate of photosynthesis vs Light Intensity



Blackman's Law of Limiting Factor

If a chemical process is affected by more than one factor, then its rate will be determined by the factor which is nearest to its minimal value It is the factor which directly affects the process if its quality is changed

Light: Light saturation occurs at 10% of the full sunlight except for plants in shade or in dense forests, light is rarely a limiting factor in nature

• There is a linear relationship between incident light & CO_2 fixation rates at low light intensities. At higher light intensities, gradually the rate does not show further increase as other factors become limiting.

CO_2 concentration: Major limiting factor. The concentration of CO_2 is very low in the atmosphere (0.03 & 0.04%). Increase in concentration upto 0.05% can cause increase in CO_2 fixation rates, beyond this levels can become damaging over longer periods.

• At low light conditions neither group(C_3 & C_4)responds to high CO_2 conditions. C_4 plants show saturation at 360μlL^{-1} CO_2 while in c_3, saturation is seen at 450 μlL^{-1}. Some greenhouse crops like tomatoes and bell pepper show higher yields in CO_2 enriched atmosphere.

Temperature: Dark reactions being enzymatic are temperature controlled, Light reactions are also temperature sensitive but to much lesser extent C_4 plants show higher yield at high temperature while C_3 plants have a much lower temperature optimum.

Water: Effect of water as a factor is more through its effect on the plant rather than directly on photosynthesis. Water stress causes the stomata to close hence reducing CO_2 availability Water stress also makes leaves wilt, thus, reducing the surface area of leaves and their metabolic activity as well.