Photosynthesis in Higher Plants

Photosynthesis

Early Discoveries

- **Joseph Priestley**: Concluded that air is necessary for the growth of a plant. He discovered the fact that plants restore oxygen in the air, fouled because of burning and respiration of living organisms.

- **Jan Ingenhousz**: Concluded that sunlight is essential for plant processes that purify the air. He also discovered that the green parts of plants release oxygen.

- **Julius Von Sachs**: Discovered that the green substance in plants (chlorophyll) is located in special bodies (chloroplast). Green parts of plants are where glucose is made, which is stored as starch.

- **T.W. Engelmann**: Discovered the first action spectrum of photosynthesis using green alga *Cladophora*. It roughly resembles the absorption spectra of chlorophylls *a* and *b*.

- **Cornelius van Niel** : Demonstrated that photosynthesis is a light-dependent reaction in which hydrogen from an oxidisable compound reduces carbon dioxide to carbohydrates.

$$2H_2A + CO_2 \xrightarrow{\text{Light}} 2A + CH_2O + H_2O$$

- He worked on green and purple bacteria.

- When he worked on sulphur bacteria, he found that sulphur is the oxidation product in them, and not oxygen, since H_2S is the hydrogen donor there. Hence, he concluded that oxygen comes from H_2O, and not from CO_2.

- Finally, the correct equation for photosynthesis was discovered.

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

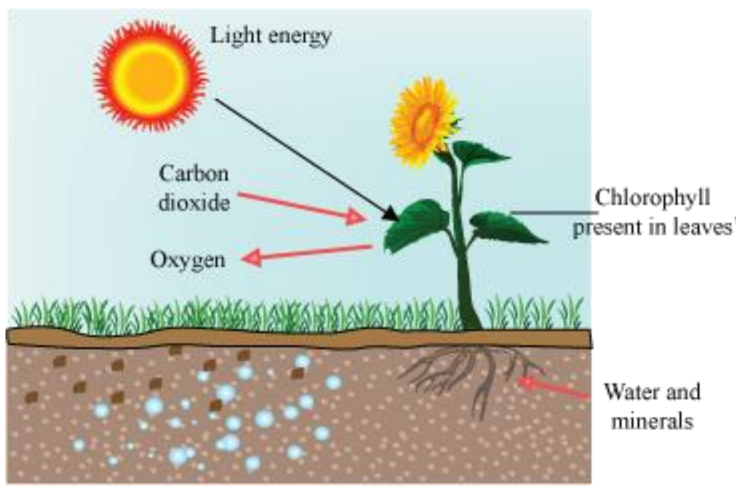
We know that plants manufacture their own food. They are known as **autotrophs** and those organisms that cannot produce their own food are called **heterotrophs**.

Autotrophs can be further classified into

- **Photoautotrophs:** they are organisms that utilize solar energy for the production of food.
- **Chemoautotrophs:** they are organisms that use other chemicals to make their own food by some chemical reactions.

But where does the production of food take place in plants?

Leaves are the food factories of plants. They are the sites where the synthesis of food occurs in plants. **But why do only leaves manufacture food for plants? What process is involved in the synthesis of food?** The leaves of plants contain a green pigment called chlorophyll. This pigment captures the sun's energy, which is used to prepare food from carbon dioxide and water. The process of synthesis of food using sunlight, carbon dioxide, and water is known as **photosynthesis.**



Since solar energy is essential for plants to prepare food, we can say that sun is the ultimate source of energy for plants and otherwise.

What happens during photosynthesis? During the process of photosynthesis, the leaves containing chlorophyll convert carbon dioxide and water into carbohydrates in the presence of sunlight. This process can be represented in the form of the following equation:

$$\text{Carbon dioxide} + \text{Water} \xrightarrow[\text{Chlorophyll}]{\text{Sunlight}} \text{Carbohydrates} + \text{Oxygen}$$

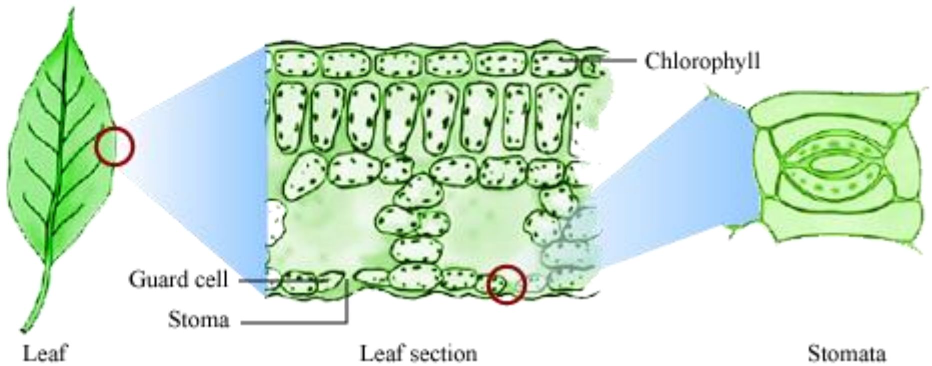
Carbohydrates, which are produced during photosynthesis, are ultimately converted into starch to be stored in plants. The slimy green patches seen on the surface of ponds are organisms called **algae.** They too contain chlorophyll and can prepare their own food through the process of photosynthesis.

Can you tell why potted plants are advised to be placed in well-lit areas?

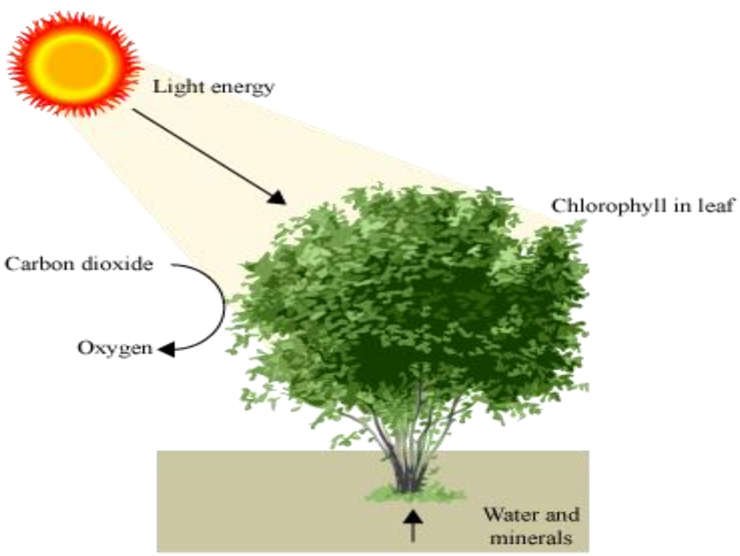
Plants require sunlight to manufacture food. Without sunlight, photosynthesis cannot take place in plants, and can even lead to the death of plants. Hence, it is advisable to place potted plants in areas receiving sufficient sunlight.

Let us perform the following activity to test the production of food in plants.

We now know that plants require water and carbon dioxide, in the presence of sunlight, to carry out photosynthesis. **How are raw materials supplied to plants?**



The tiny pores found on the underside of leaves are called **stomata.** Carbon dioxide enters leaves through stomata.



Water and minerals enter plants through the roots. They are transported to leaves by the tiny vessels present throughout the plant body. Therefore, the carbohydrates produced in leaves are converted into starch, while the oxygen produced in the process is released into the atmosphere through the stomata.

Site of Photosynthesis

Chloroplasts are the sites of photosynthesis. They are present in the mesophyll cells of the leaves.

Chlorophyll is the green pigment found in the chloroplasts.

The chlorophyll pigments trap the photons of light and get excited and thus initiate the process of photosynthesis.

Significance of Photosynthesis

The process of photosynthesis is essential to plants as well as the other living organisms.

- All heterotrophs depend on plants for their energy requirements.
- The level of carbon dioxide is maintained at a constant.
- The byproduct of photosynthesis, oxygen is the main factor responsible for the maintenance of life on earth.

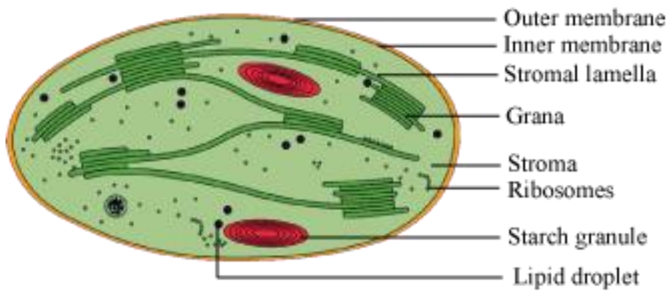
Do You Know?

Do you know that aquatic plants perform 90% of the world's photosynthesis! The amount of photosynthesis is almost ten times the rate of respiration during the day.

Chloroplast- The site of Photosynthesis

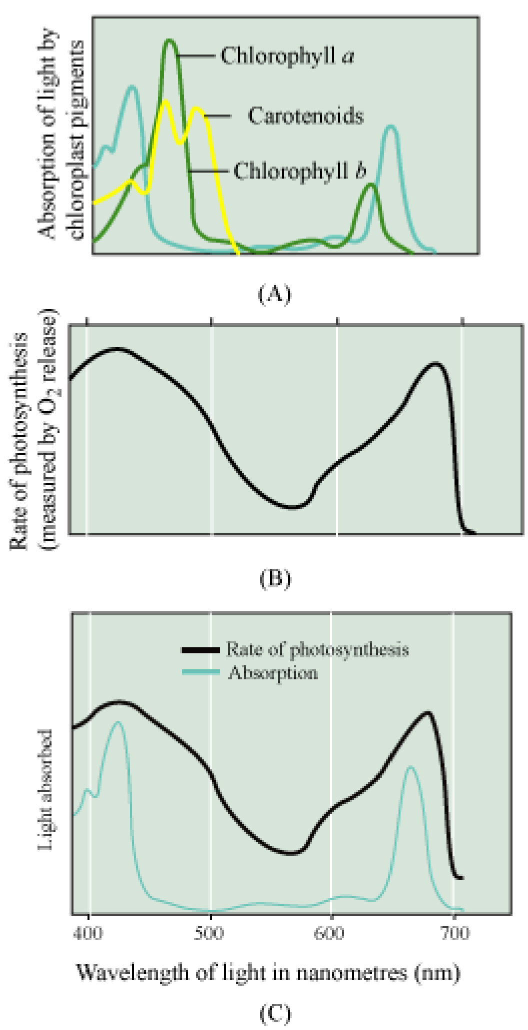
Site of Photosynthesis

- Mesophyll cells in green leaves have large number of **chloroplasts**, which are the site of photosynthesis.
- Chloroplast consists of the grana and the stroma lamellae (forming a membranous system) and the fluid stroma.
- Membrane system of chloroplast – traps light, and synthesises ATP and NADPH (site of light-dependent reaction of photosynthesis)
- Stroma – CO_2 is incorporated into the plant by enzymatic reactions, leading to the synthesis of sugar (site of light-independent reaction of photosynthesis)
- Chloroplasts are aligned along the walls of mesophyll cells so as to get optimum light.



Pigments Involved in Photosynthesis

- An **absorption spectrum** is the graph plotted against the fraction of light absorbed by the pigment.
- An **action spectrum** is the rate of a physiological activity plotted against the wavelength of light.
- The similarity of the action spectrum of photosynthesis and the absorption spectrum of chlorophyll tells us that chlorophylls are the most important pigments in the process.



(A) Absorption spectrum of chlorophylls *a*, *b*, and carotenoids

(B) Action spectrum of photosynthesis

(C) Action spectrum superimposed on absorption spectrum of chlorophyll a

- 4 types of pigments may be present in leaves:
- Chlorophyll *a* (blue-green)
- Chlorophyll *b* (yellow-green)
- Xanthophylls (yellow)
- Carotenoids (yellow to yellow-orange)
- Chlorophyll *a* is the main pigment in photosynthesis.
- In VIBGYOR spectra, chlorophyll *a* shows maximum absorption, and hence, the rate of photosynthesis is the highest at the blue and red regions.
- Accessory pigments: Chlorophyll *b*, xanthophylls and carotenoids
- Absorb a wider range of light, and transfer the energy to chlorophyll *a*
- Protect chlorophyll *a* from photo-oxidation

Light Reaction of Photosynthesis and Cyclic and Non-Cyclic Photophosphorylation

Light Reaction (Photochemical Phase)

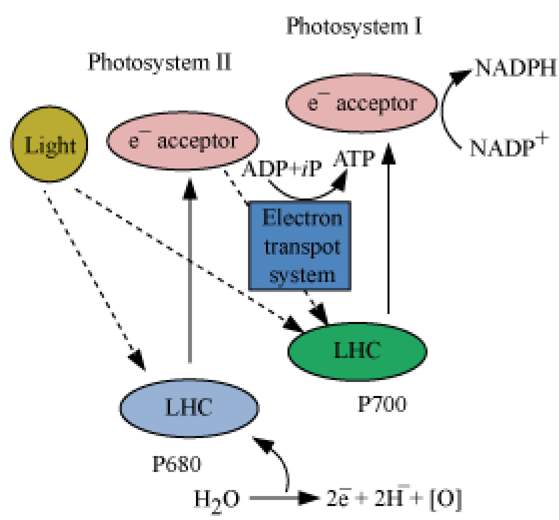
- This phase directly depends on light. The pigments absorb light energy and produce ATP.
- Includes:
- Light absorption
- Water splitting
- Oxygen release
- Formation of ATP and NADPH, which is then used in the biosynthetic phase
- Pigment molecules bound to the proteins form LHC (light harvesting complexes). LHC are located within two photosystems – PSI and PSII
- Each photosystem has two parts:
- Reaction centre – consisting of chlorophyll *a* molecule
- Antennae – consisting of accessory pigments, which increase the efficiency of photosynthesis by absorbing different wavelengths of light

- Reaction centre is different in both photosystems:
- PSI – P700; since chlorophyll *a* has absorption peak at 700 nm here
- PSII – P680; since chlorophyll *a* has absorption peak at 680 nm here.

Photo-Phosphorylation

- The process of formation of ATP in chloroplast in the presence of sunlight
- Photo-phosphorylation is of two types:
- Non-cyclic photo-phosphorylation
- Cyclic photo-phosphorylation

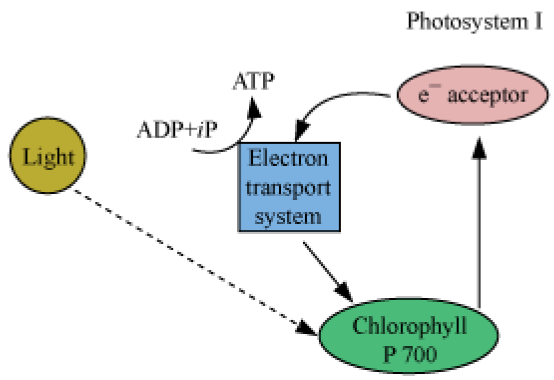
Non-Cyclic Photo-Phosphorylation



- PSII absorbs 680 nm wavelength of red light, causing electrons to become excited and jump into an orbit farther from the nucleus.
- These electrons are then accepted by an electron acceptor, which sends them to an electron transport system.
- Electron transport system transfers the electrons to PSI.
- Electrons in PSI are simultaneously excited on receiving a wavelength of 700 nm. These electrons are again transferred to another electron acceptor having a greater redox potential.

- From the electron acceptor, electrons are transferred to the molecule of $NADP^+$.
- Addition of these electrons reduces the $NADP^+$ to $NADPH+ H^+$.
- During this process, electrons move downhill, i.e., towards the systems at greater redox potential.
- The flow of electrons assumes the shape of the letter 'Z' when all carriers are placed according to their redox potential. Hence, the process is called Z scheme.
- Since the electrons lost by PSII do not come back to it, this process of formation of ATP is called non-cyclic photo-phosphorylation.

Cyclic Photo-Phosphorylation



- In this scheme, only PSI is functional. Hence, the electrons are circulated within the photosystem.
- This results in a cyclic flow of electrons.
- This scheme could possibly be occurring in *stroma lamellae* because it lacks both PSII and NADP reductase enzyme.
- This cyclic flow results only in the synthesis of ATP, and not of $NADPH + H^+$.

Splitting Of Water

- The electrons being transferred in the photo-phosphorylation reactions are generated by the splitting of water.
- Water splitting complex is associated with PSII.
- Manganese, chlorine, etc., play an important role.

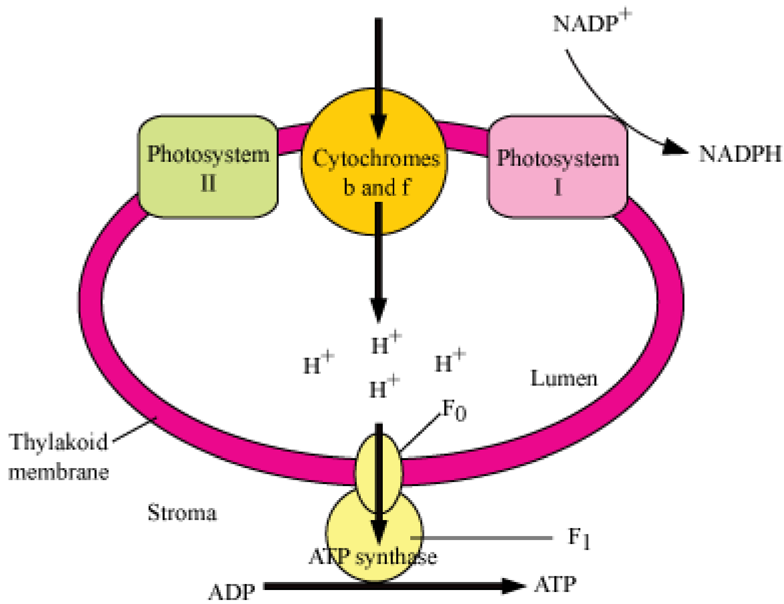
- The light-dependent splitting of water is called photolysis
  $2H_2O \rightarrow 4H^+ + O_2 + 4e^-$
- Electrons formed are used for replacing the electrons lost from P680.
- Oxygen is liberated as a byproduct of photosynthesis.
- Protons are used for the formation of reducing power NADP to $NADPH^+$.

Chemiosmotic Hypothesis

Chemiosmotic Hypothesis

- Chemiosmotic hypothesis explains how ATP is synthesised in chloroplast.
- Chemiosmosis requires a membrane, a proton pump, a proton gradient and ATPase.
- Membrane: Thylakoid membrane; protons accumulate towards the inside of the membrane; in the lumen
- Proton gradient develops because of the following reasons:
- Water splitting reaction takes place on the inner side of the membrane. So H^+ produced in the reaction accumulates within the lumen.
- During the photo-phosphorylation steps when the electron-carrier molecule passes on its electrons to the electron-carriers on the inner side of the membrane, protons are released into the inner side or the lumen of the membrane.
- NADP reductase enzyme is located on the stroma side of the membrane. Protons are removed from the stroma and provided to the lumen because protons are necessary for the reduction of $NADP^+$ to $NADPH + H^+$, along with the electrons that come from the electron-accepter located in PSI.
- As a result, the protons in the stroma decrease in number, while the protons in the lumen, increase in number, thus creating a proton gradient across the thylakoid membrane.
- The proton gradient is broken down due to the movement of H^+ from the lumen towards the stroma through the transmembrane channel to the F_0 of ATPase.
- ATPase: This enzyme has two parts
- F_0 – embedded in the membrane and carry out facilitated diffusion of H^+

- F_1 – protrudes towards the stroma
- Conformational changes are induced in the F_1 particle of the ATPase. This makes ATPase synthesise molecules of ATP.



Dark Reaction- C3 Pathway

- Products of light reaction: ATP, NADPH, CO_2 and H_2O
- Next stage is the biosynthetic phase. In this, ATP and NADPH are used for synthesising the food.
- CO_2 is fixed to form CO_2 fixation product.
- This stage is also called the dark phase as it is independent of light, but deals with the products of light reaction.
- The reaction involved in the biosynthetic phase takes place in the stroma of chloroplasts.
- In some plants, the first product of CO_2 fixation is a 3-carbon compound called 3-phosphoglyceric acid (PGA). These plants are said to adopt the C_3 pathway.
- In other plants, the first CO_2 fixation product is a 4-carbon compound called oxaloacetic acid. These plants are said to adopt the C_4 pathway.

Calvin Cycle (C_3 Cycle)

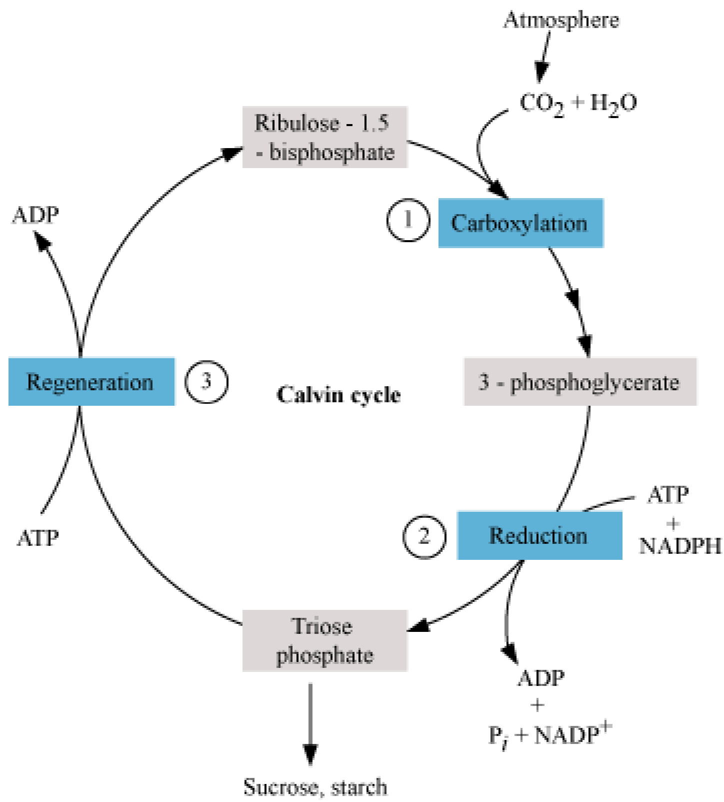
- The path of carbon in the dark reaction was traced by Melvin Calvin using radioactive carbon (^{14}C).

- Calvin pathway occurs in all photosynthetic plants, irrespective of whether they have C_3 or C_4 pathway.

- The primary acceptor of CO_2 was found to be a 5-carbon ketose sugar called Ribulose bisphosphate (RuBP). This was surprising as it was believed that for the formation of a 3-carbon product, CO_2 would have to be accepted by a 2-carbon compound.

- RuBP is used in a cyclic manner (regenerated) and a sugar is synthesised.

- 3 stages of Calvin cycle: Carboxylation, Reduction and Regeneration



- **Carboxylation**

- Ribulose 1, 5-bisphosphate combines with CO_2, and fixes it to a stable organic intermediate (3-phosphoglycerate – 2 molecules).

- Reaction catalysed by the enzyme RuBisCO (RuBP Carboxylase-Oxygenase)

- 3 PGA is the first stable product of this cycle.

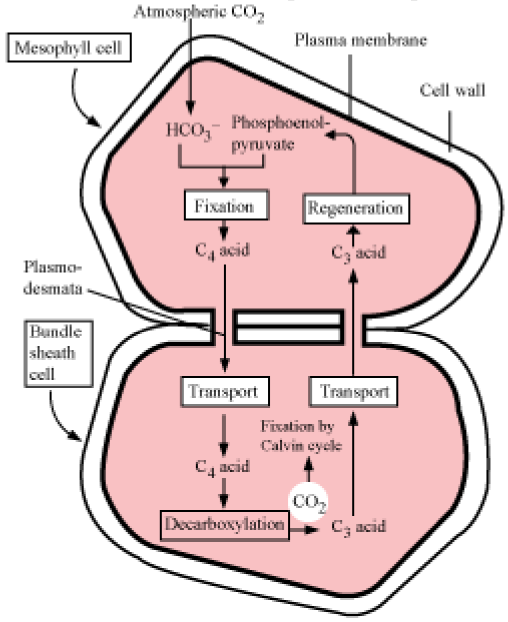
- **Reduction**
- Here, 2 molecules each of ATP and NADPH are required for fixing 1 molecule of CO_2.
- This stage contains a series of reactions.
- Glucose is formed as a result of this series of reactions.
- **Regeneration**
- RuBP regenerates to enable the cycle to continue uninterrupted.
- 1 ATP molecule is required.
- For the formation of one molecule of glucose, six molecules of CO_2 need to be fixed; hence, six cycles are required.
- ATP required:
  For fixing 1 molecule of CO_2 – 3 (2 for reduction and 1 for regeneration)
  For fixing 6 molecules of CO_2 – 3 × 6 = 18 ATP
- NADPH required:
  For fixing 1 molecule of CO_2 – 2 (for reduction)
  For fixing 6 molecules of CO_2 – 2 × 6 = 12 NADPH
- Thus, the synthesis of 1 molecule of glucose requires 18 ATP and 12 NADPH.

Dark Reaction: Biosynthetic Phase- C4 Pathway

C_4 Pathway (Hatch and Slack Pathway)

- Occurs in plants like maize, sugarcane – plants adapted to dry tropical regions
- The leaves of C_4 plants have Kranz anatomy. They have special large cells around their vascular bundles called bundle sheath cells. These cells form several layers around the vascular bundles and have large number of chloroplasts, thick walls impermeable to gaseous exchange, and lack intercellular spaces.
- First CO_2 fixation product is a 4-carbon compound called oxaloacetic acid, but C_3 cycle is used as the main biosynthetic phase.
- Mesophyll cells: Occur in the C_4 cycle
- Bundle Sheath Cells: Occur in the C_3 cycle

- C_4 plants can tolerate high temperature and high light intensity, show greater productivity of biomass, and lack photorespiration.



Hatch and Slack pathways

- Primary CO_2 acceptor: Phosphoenol pyruvate (PEP) – a 3-carbon molecule

- PEPcase (PEP Carboxylase) fixes CO_2 in the mesophyll cells. It forms the 4-carbon compound oxaloacetic acid (OAA), and then other 4-carbon compounds like malic acid and aspartic acid.

- These compounds are transported to the bundle sheath cells. There, C_4 acid breaks down to form C_3 acid and CO_2 (released only in the bundle sheath cells, and enters the C_3 cycle).

- C_3 acid, so formed, is again transported to the mesophyll cells and regenerated back into PEP.

- C_3 cycle cannot directly occur in the mesophyll cells of C_4 plants because of the lack of the enzyme RuBisCO in these cells

- RuBisCO is found in abundance in the bundle sheath cells of C_4 plants. They lack PEPcase.

Photorespiration

- It is a process in which there is no formation of ATP or NADPH, but there is utilization of ATP with release of CO_2. It is also considered a wasteful process.

- Photorespiration is responsible for the difference between C_3 and C_4 plants.

Do C_4 plants have better yield than C_3 plants?

- To understand this, let us first recall the first step of Calvin pathway.

  $$RuBP + CO_2 \xrightarrow{RuBisCO} 2 \times 3PGA$$

  This reaction is catalyzed by an enzyme RuBisCO. The active site of RuBisCO can bind to both O_2 and CO_2, though it has a greater affinity for CO_2.

- In C_3 plants, some of the O_2 binds to RuBisCO to form phosphoglycerate and phosphoglycolate (instead of PGA). There is no production of sugar or ATP. This mechanism in C_3 plants results in utilization of ATP with the release of O_2.

- In C_4 plants, photorespiration does not occur since they have mechanism to increase the CO_2 concentration at enzyme site. C_4 acids are broken in the bundle sheath cells to release CO_2, thus increasing the intracellular concentration of CO_2.
  This is the reason why C_4 plants are better yielding as compared to C_3 plants.

RuBisCO - A unique enzyme!

RuBisCO or ribulose bisphosphate carboxylase oxygenase is the most abundant enzyme in the world!

As the name suggests, it can bind to both carbon dioxide as well as oxygen. The binding to the enzyme is determined by the relative concentration of oxygen and carbon dioxide.

Factors Affecting Rate of Photosynthesis

- Photosynthesis is influenced by internal (plant) factors and external factors.
- **Internal factors:** Number, size and orientation of leaves, mesophyll cells and chloroplasts, internal carbon dioxide concentration and the amount of chlorophyll.
- **External factors:** Availability of sunlight, temperature, carbon dioxide concentration and water.
- **Law of Limiting Factors –** If a chemical process is affected by more than one factor, then its rate will be determined by factor which is nearest to its minimal value (factor which directly affects the process if its quantity is changed).
- This law of limiting factor was given by Blackman (1905).

- **Light**

- Incident light $\propto$ CO_2 fixation rate; but at higher light intensities, the rate does not increase further as other factors become limiting

- Light is rarely a limiting factor (with exception of the shade plants or plants of dense forest) because light saturation occurs at 10% of the full sunlight.
- Beyond a point, if incident light is increased, then it leads to decrease in photosynthesis due to breakdown of chlorophyll.

- **CO_2 Concentration**

- Major limiting factor
- Usually low in atmosphere (0.03 – 0.04%)
- Up to 0.05% – increases rate of CO_2 fixation
- > 0.05% – damaging effect
- Though both C_3 and C_4 show increase in rate of photosynthesis at high light intensities accompanied by high CO_2 concentration. The saturation point for C_3 is obtained at higher concentrations as compared to C_4. Therefore, CO_2 concentration is more of a limiting factor for C_3 plants.
- Increased CO_2 concentration is beneficial for greenhouse crops such as tomatoes and bell pepper.

- **Temperature**

- Dark reactions are more sensitive to an increase in temperature.
- C_4 plants respond more to an increase in temperature and show higher rate of photosynthesis as compared to C_3 plants.
- Adaptations according to habitat also affect temperature optimum for photosynthesis. Tropical plants have higher temperature optimum compared to plants growing in temperate climates.

- **Water**

- Water stress causes stomata to close and hence, less CO_2 is available.
- Water stress causes the leaves to wilt, thereby reducing their surface area and metabolic activity as well.