Republic of Iraq Ministry of Higher Education &Scientific Research University of Anbar College of Science



Lecture 8

Photosynthesis

Cyclic photophosphorylation

Dark reaction

C₃ pathway or Calvin cycle

For Dr. Enas Fahd Naji Department of Biology Almost all the chemical processes that make up the light reactions of photosynthesis are carried out by four major protein complexes: photosystem II, the cytochrome b6f complex, photosystem I, and the ATP synthase. These four integral membrane complexes are vector ally oriented in the thylakoid membrane to function as follows:

1. Photosystem II oxidizes water to O2 in the thylakoid lumen and in the process releases photons into the lumen

2. Cytochrome b6f oxidizes plastohidroquinone (PQH₂) molecules that were reduced by PSII and delivers electrons to PSI. The oxidation of plastohidroquinone is coupled to proton transfer into the lumen from the stroma, generating a proton motive force.

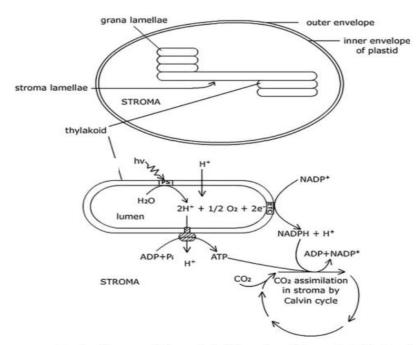
3. Photosystem I reduces NADP⁺ to NADPH in the stroma by the action of ferredoxin (Fd) and the flavoprotein ferredoxin-NADP reductase (FNR).

4. ATP synthase produces ATP as protons diffuse back through it from the lumen into the stroma.

Cyclic photophosphorylation

One way of excluding noncyclic photophosphorylation is to illuminate chloroplasts with wavelengths of light greater than 680 nm. Under these conditions only photosystem I is activated and electrons are not removed from H_2O , as illustrated by the lack of oxygen evolution under these circumstances. When the flow of electrons from H_2O is stopped, noncyclic photophosphorylation is also stopped and as a consequence CO_2 assimilation is retarded as a result of which oxidized NADP is no longer available as an electron acceptor. Activation of photosystem I by wavelengths of light greater than 680 nm causes electrons to flow from P700 to A (FeS). When electrons are not passed to NADP⁺, they may be lost to cytochrome b6 which in turn passes electrons back to P700 via cytochrome f and plastocyanin. There is some evidence that plastoquinone instead of cytochrome b6 may act as the primary acceptor of electrons from A (FeS). This possibility is quite likely because plastoquinone is necessary for proton transport across the thylakoid

membrane for the generation of ATP. There is possibility of production of two ATPs, one between (FeS) and cytochrome b6 and one between cytochrome b6 and cytochrome f which is not very likely without plastoquinone mediation. Nevertheless, the term photophosphorylation is used to denote the cycling of electrons from the donor (excited P700 system) to an acceptor (possibly FeS) and back to the P700 trap with some generation of ATP. If cyclic photophosphorylation does not indeed operate appreciably in certain organisms, it only produces limited ATP.



Overview of two steps of photosynthesis, light reaction which occurs in thylakoids and $\rm CO_2$ assimilation which occurs in the stroma of chloroplasts

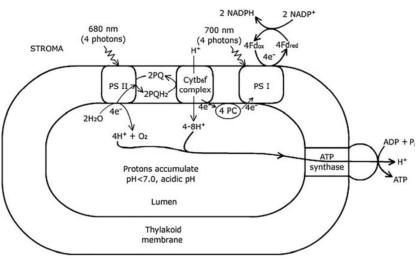


Figure showing positioning of the photosynthetic electron transport complexes in thylakoids and proton accumulation in lumen

pH gradient across the thylakoid membrane stimulated ATP production when chloroplasts were maintained in darkness. He also demonstrated that under normal light conditions an H⁺ concentration gradient is established in actively photosynthesizing chloroplasts. As figure illustrates, the electron carriers are located in the granum membrane. ATP and NADPH are produced on the stroma side surface of the thylakoid. An important aspect of the model is the mobility of plastoquinone. This carrier presumably transfers electrons to cytochrome f and in addition pick up H+ ions on the outside and releases protons to the thylakoid channel. The transfer of protons to the inside and the production of protons from the photolysis of water incurs buildup of protons inside and a pH gradient across the thylakoid membrane to the outside (stroma side), where the hydrogen concentration is relatively low. The membrane itself is not permeable to protons concentrated on the channel side, which represents a source of energy. It is believed that protons flow from the inside of the stroma side of the membrane through special pathways of CF (stalks) that terminate as knobs at the outer (stroma side) surface. These stalks and knobs are the sites of photophosphorylation. The proton flow along the gradient provides the necessary energy for the following reaction:

ADP+ Pi \longrightarrow ATP + H₂O +8,000 cal/ mole

ATPase

The proton flow and phosphorylation are thought to be brought together (coupled) by the activity of the enzyme ATPase (also called coupling factor) which is associated with the destruction of ATP, but it will operate in the reverse situation as long as sufficient energy is supplied (in this case from the proton flow). For every two electrons passing through the transport system, two protons are transported by the reduced plastoquinone, a water molecule is photolyzed. Theoretically, one molecule of ATP is produced for every three protons passing through the CF.

The light reaction phase of photosynthesis may be summarized by the following equation, which represents the photochemical, photophosphorylation, photo reduction and photo oxidation (splitting of water) events:

chloroplasts

$2H_2O + 2NADP^+ + (ADP)n + (Pi)n \longrightarrow (ATP)n + 2NADPH + 2H + O_2$

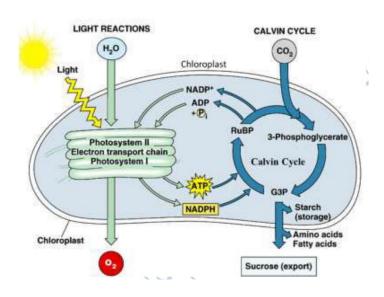
The summary equation also indicates that the stoichiometry of the overall equation is not exact, particularly for ATP production and the quanta required .We do not know the number of ATP molecules produced per oxygen molecule liberated. Some investigators claim 2 molecules of ATP are produced for every oxygen molecule liberated, and others maintain 4. According to plant scientists 8 to 12 quanta (photons) appear to be necessary to produce NADPH and ATP sufficient for CO_2 fixation. Approximately 2NADPH and 3ATP molecules are required to incorporate 1 molecule of CO_2 into sugar phosphate

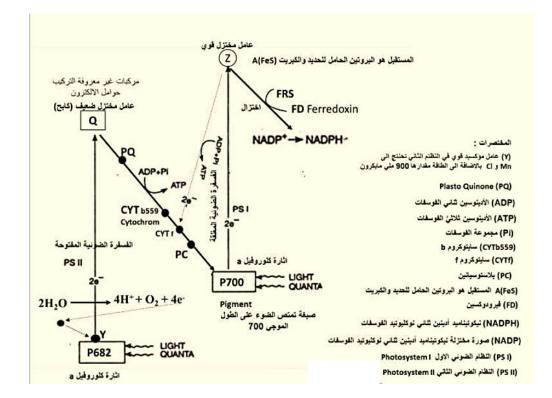
Summary of light reaction:

- Eight light photons are required to oxidize 4H2O.
- 2 e⁻ and 1 H⁺ are required to reduce 1 molecule of NADP to NADPH2. 1H+ remains in the medium.
- Transfer of 1 electron from H_2O to NADP requires 2 photons as excitation of both the photosystems is necessary.
- Photolysis of $4H_2O$ produces $2H_2O$, O_2 , $4 e^-$ and $4H^+$.

• Reduction of 1 molecule of CO₂ requires 2NADPH₂ and 3ATP.

• 4 molecules of water after photolysis forms 3ATP and $2NADPH_2$ and cycle repeats 6 times producing 18 ATP and 12 $NADPH_2$ during light reaction of photosynthesis.





Dark reaction :

 CO_2 is fixed in one of the following pathways in green plants.

- 1. C₃ pathway or Calvin cycle
- 2. C₄ pathway or Hatch and Slack cycle.

3. C₂ or Glycolate pathway.

4. Crassulacean acid metabolism (CAM plants).

C₃ pathway or Calvin cycle:

• First stable compound formed after the entry of CO_2 in plant system is triose sugar viz; Phosphoglyceric acid (PGA) (Calvin 1961).

• Pathway of photosynthetic CO_2 assimilation beginning with carboxylation of RuBP by Rubisco.

• The biochemical pathway for the reduction of CO_2 to carbohydrate. The cycle involves three phases, the carboxylation of ribulose-1,5bisphosphate with atmospheric CO_2 catalyzed by rubisco, the reduction of the formed 3-phosphoglycerate to triose phosphate by 3phosphoglycerate kinase and NADP-glyceraldehyde -3-phosphate dehydrogenase, and the regeneration of ribulose-1,5-bisphospate through the concerted action of ten enzymatic reactions.

It is called C_3 pathway because the first stable compound formed after the entry of CO_2 in plant system is triose sugar i.e. PGA. Calvin by using isotope of carbon C^{14} could identify various products formed during reduction of CO_2 by paper chromatography and radio autography.

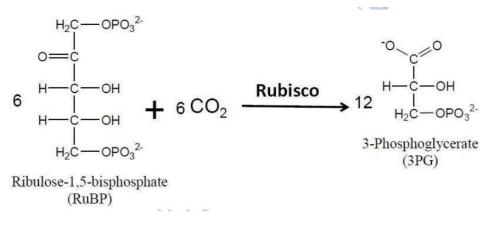
Calvin cycle occurs in three main parts:

1. Carboxylation (Carbon fixation):

Which involves addition of CO_2 and H_2O to form two molecules of 3 PGA.

Six molecules of CO_2 react with 6 molecules of Ribulose 1, 5 biphosphate or bis phosphate to form 12 molecules of 3PGA. The reaction is catalysed by RuBP carboxylase (Rubisco). First CO_2 is added

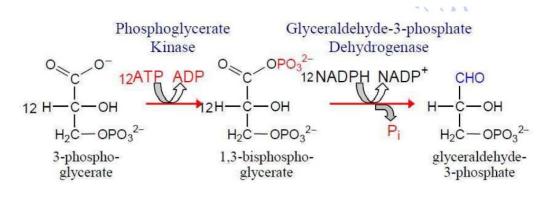
to a 5C sugar to form 6C sugar, which then splits into two molecules of 3 carbon compound.



2. Reduction:

In which COOH group in 3 PGA is reduced to an aldehyde group (3 PG aldehyde).

In next step 12 molecules of 3PGA are phosphorylated to 12 molecules of 1-3 di phospho glyceric acid. The reaction is catalyzed by phospho glycero kinase. In the next step 12 molecules of 1, 3 di phospho glyceric acid reduced to 12 molecules of 3 phospho glyceraldehyde by 12 molecules of NADPH produced in the light phase of photosynthesis.

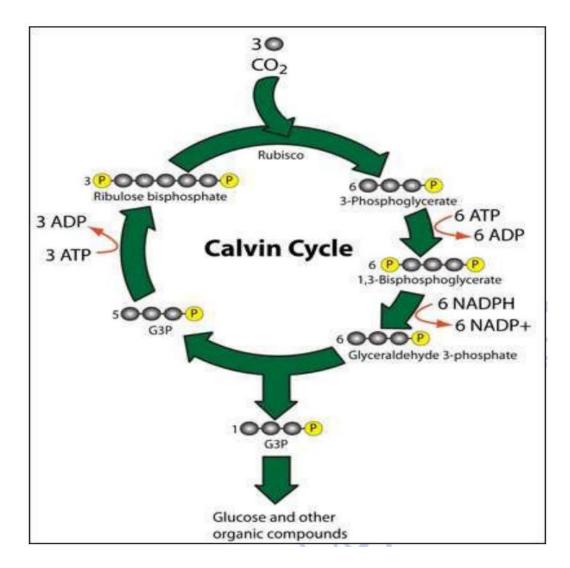


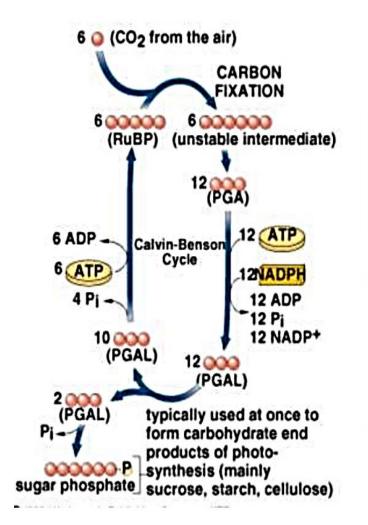
3. Regeneration of RuBP:

Out of the entry for every 6 CO2 molecules the output is one mole of glucose and rest go to the regeneration of RuBP. Six molecules of

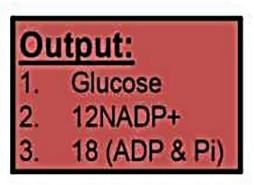
Ribulose 5 P are formed up to this stage. In the final step all 6 molecules of ribulose 5 phosphate are phosphorylated at the expense of 6 mol. of ATP in presence of enzyme phosphoribulo kinase to form 6 mol. of carbon acceptor ribulose 1-5 diphosphate. Then all 6 the molecules of ribulose 1-5 diphosphate are regenerated reenter into cycle. Whole reaction can be written as:

6CO₂ + 12 NADPH + 12 H ⁺ + 18 ATP + 11 H₂O 1 Mol. F - 6 P + 12 NADP ⁺ + 18 ADP + 18 Pi









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