Lecture 17-18

BT 203

Biochemistry

3-0-0-6

Prof. Ajaikumar B. Kunnumakkara

CANCER BIOLOGY LABORATORY
Department of Biosciences and Bioengineering
Indian Institute of Technology (IIT) Guwahati
Assam, INDIA

Key Concepts

- What is carbon reactions? How does plants assimilate carbon from the environment ?
- What is Calvin-Benson cycle?
- What is the major component of C3 cycle?
- What is C2 cycle and how it is important in plant metabolism ?
- What is C4 cycle and how it is different from other plant bioenergetics?
- What is the relevance of CAM pathways?

Photosynthesis

General Information

•Photosynthesis- Synthesis using light

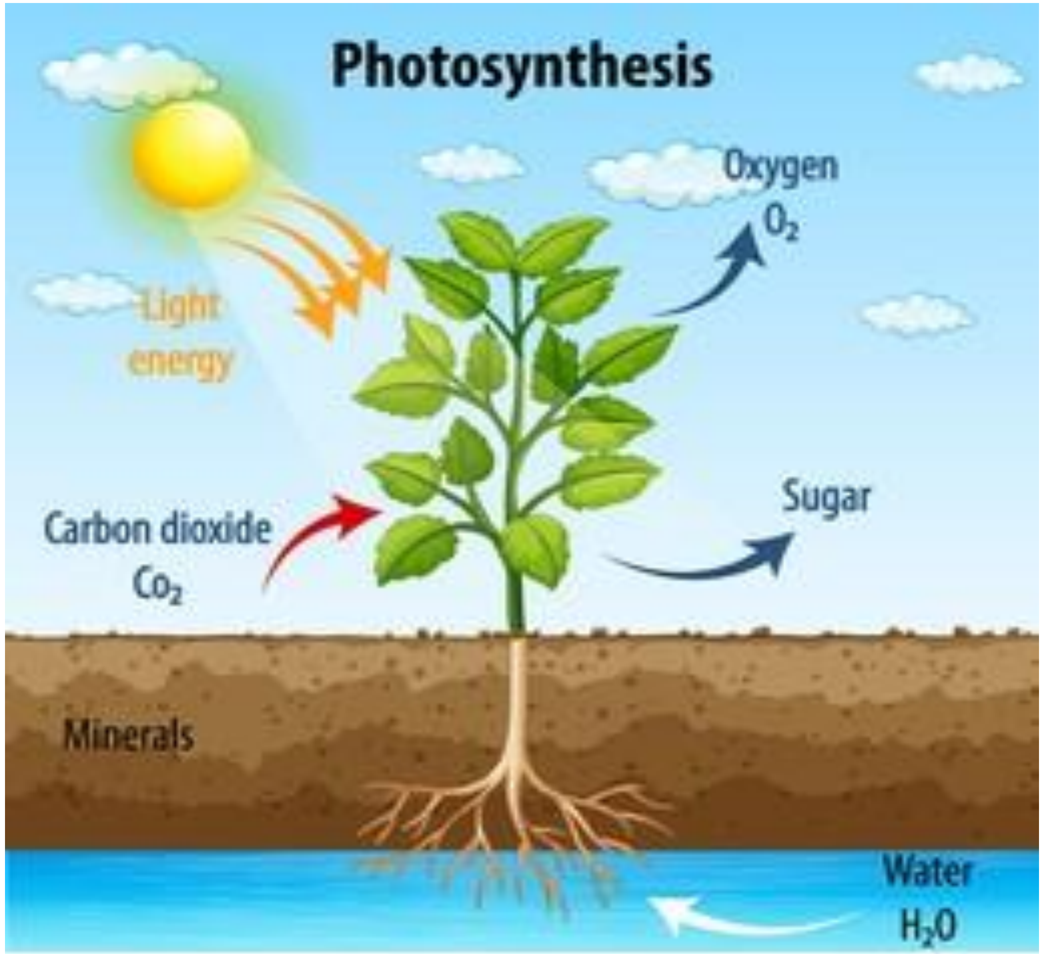
•Synthesis of carbohydrates from carbon dioxide and water with the generation of oxygen

•Photosynthetic organisms- use solar energy to synthesize carbon compounds that cannot be formed without the input of energy

•Photosynthetic tissue- mesophyll of leaves contain chlorophylls

•Plant uses solar energy to oxidize water, thereby releasing oxygen, and to reduce carbon dioxide, thereby forming large carbon compounds, primarily sugars.

Photosynthetic Reaction

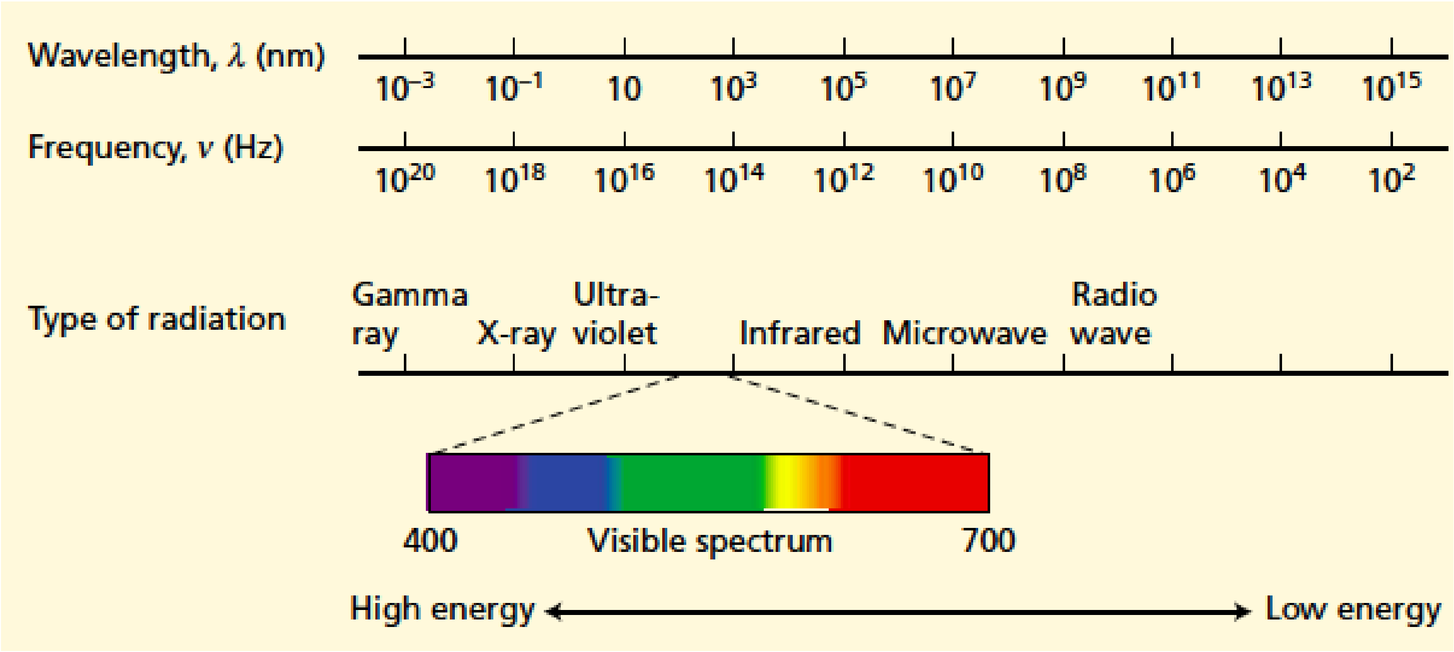


$$6\,CO_2 + 6\,H_2O \rightarrow C_6H_{12}O_6 + 6\,O_2$$

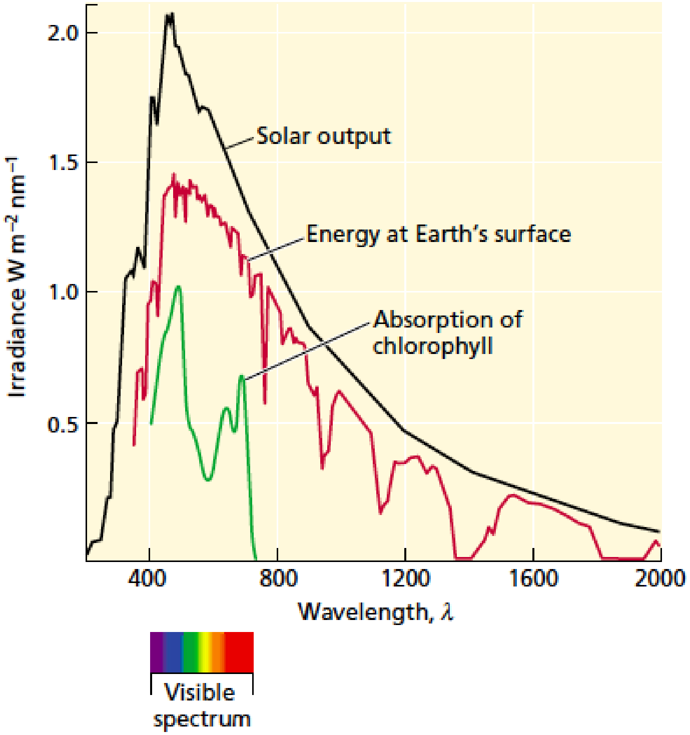
Carbon dioxide | Water | Carbohydrate | Oxygen

Characteristics of light

Electromagnetic Spectrum



Absorption Spectrum of Chlorophyll



- ✓ An absorption spectrum displays the amount of light energy taken up or absorbed by a molecule or substance as a function of the wavelength of the light

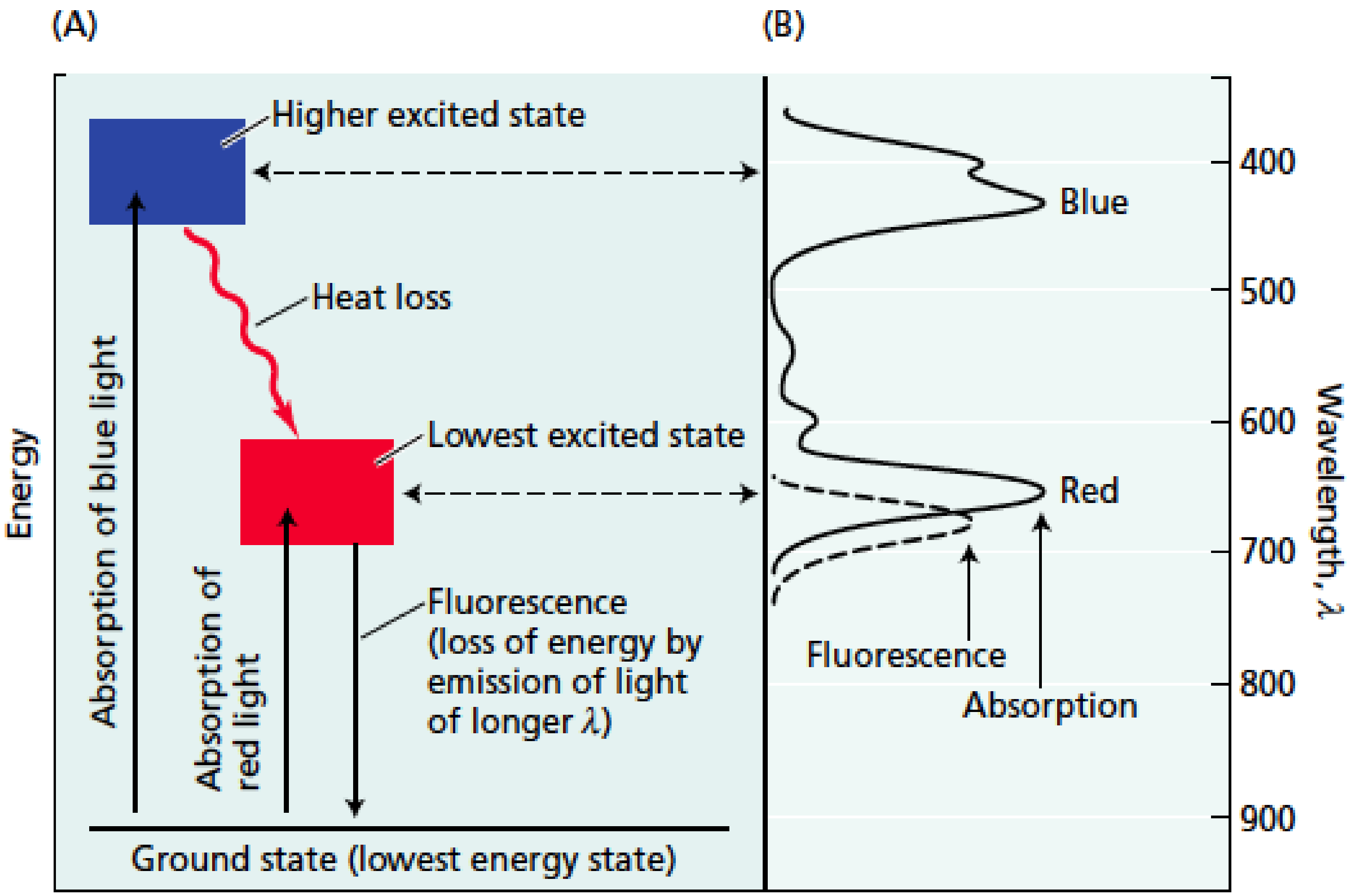
- ✓ The absorption spectrum of chlorophyll a indicates approximately the portion of the solar output that is utilized by plants

Electronic state of a molecule after light absorption

- Chlorophyll appears green to our eyes because it absorbs light mainly in the red and blue parts of the spectrum, so only some of the light enriched in green wavelengths (about 550 nm) is reflected into our eyes
- Chlorophyll (Chl) in its lowest-energy, or ground state absorbs a photon (represented by hn) and makes a transition to a higher-energy, or excited, state (Chl*)

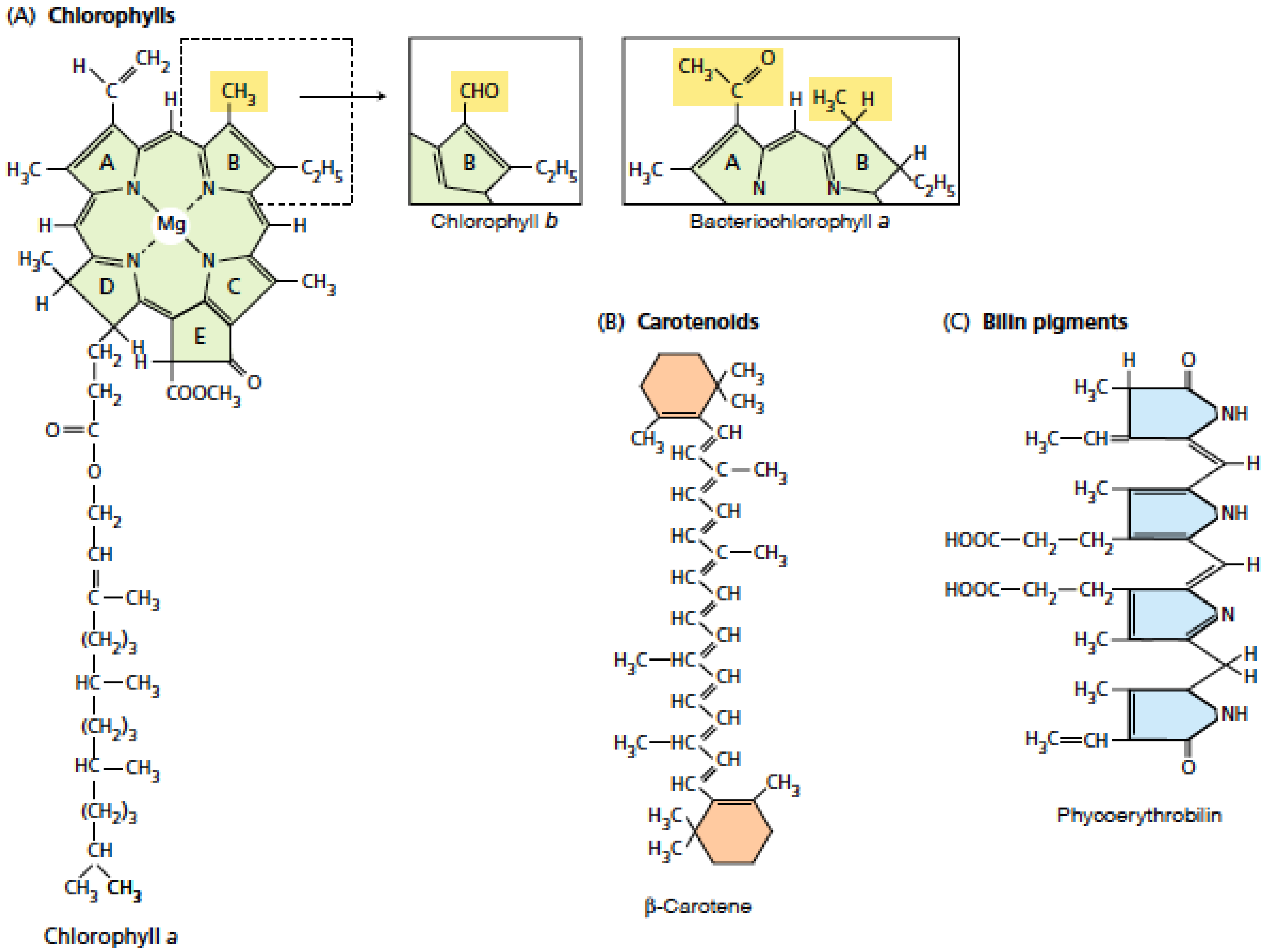
$$\text{Chl} + \text{hn} \rightarrow \text{Chl*}$$

- The distribution of electrons in the excited molecule is different from the distribution in the ground state molecule
- The excited chlorophyll has four alternative pathways for disposing of its available energy- florescence, emission of heat, transfer of energy, photochemistry



Molecular structure

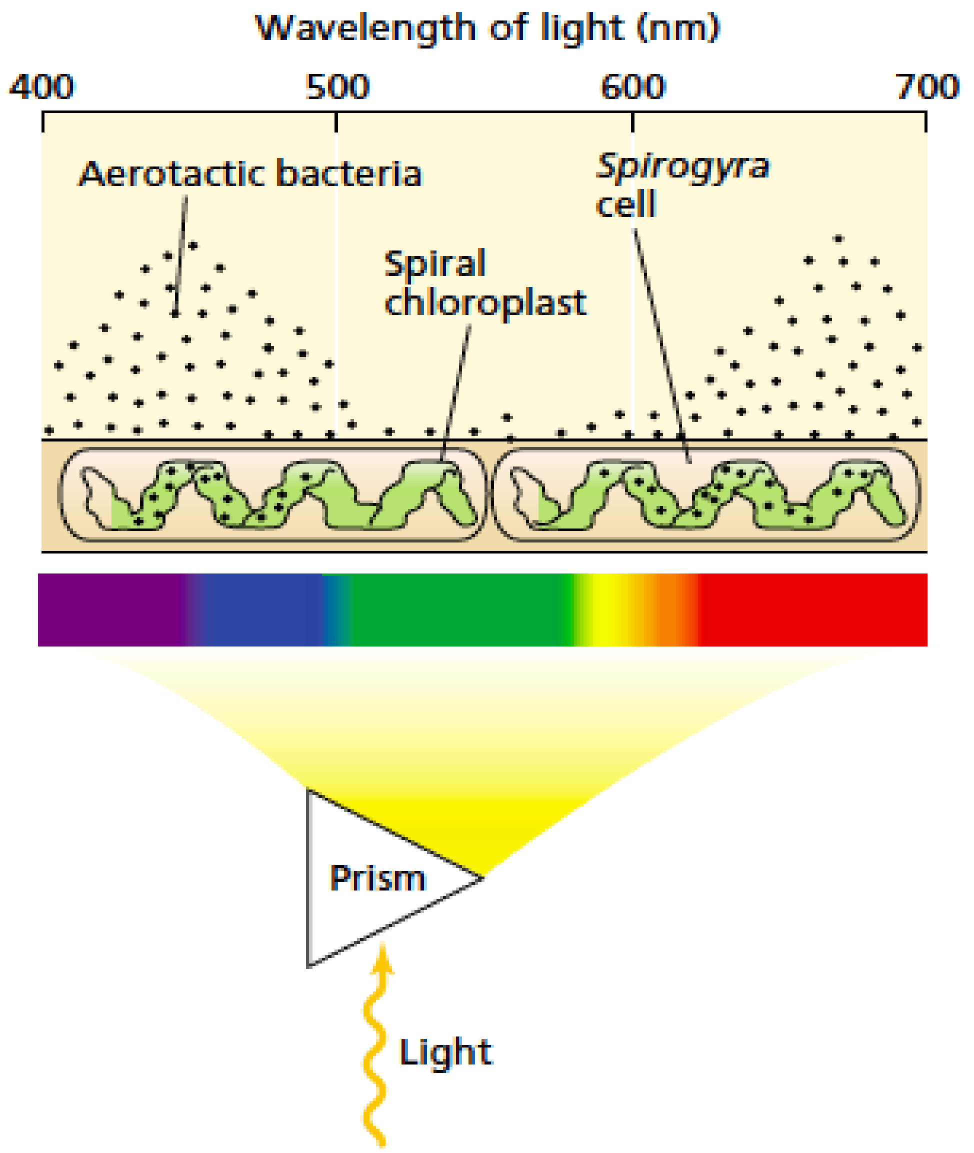
- Chlorophylls have a porphyrin-like ring structure with a magnesium atom coordinated in the center
- They have long hydrophobic hydrocarbon that anchors them in the photosynthetic membrane
- Porphyrin-like ring is the site of the electron rearrangements that occur when the chlorophyll is excited and of the unpaired electrons when it is either oxidized or reduced
- Chlorophylls a and b are abundant in green plants, and c and d are found in some protists and cyanobacteria.



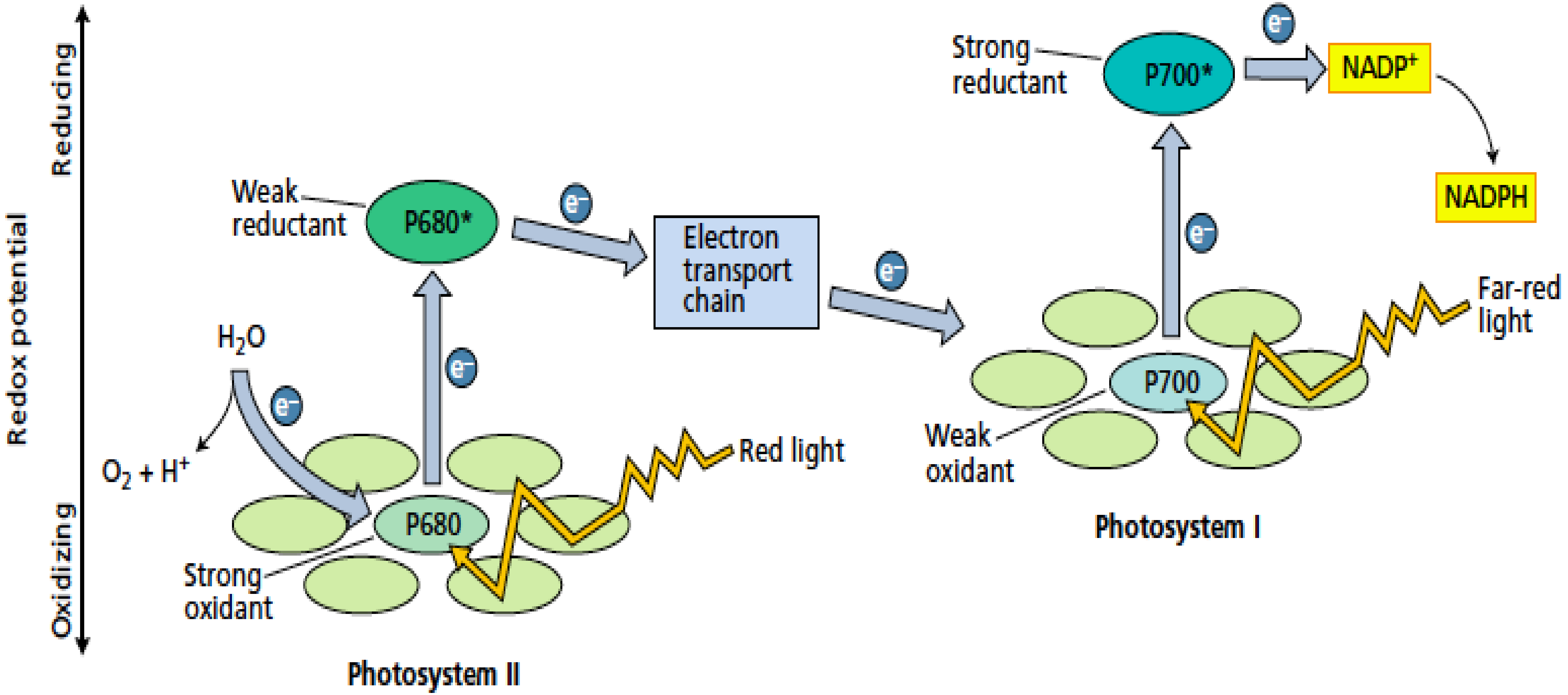
Understanding photosynthesis

Key experiments

- Establishing the overall chemical equation of photosynthesis required several hundred years and contributions by many scientists
- In 1771, Joseph Priestley observed that a sprig of mint growing in air in which a candle had burned out improved the air so that another candlecould burn. He had discovered oxygenevolution by plants.
- A Dutchman, Jan Ingenhousz, documented the essential role of light in photosynthesis in 1779
- The chemical reactions of photosynthesis are complex. In fact, at least 50 intermediate reaction steps have now been identified, and undoubtedly additional steps will be discovered
- From his studies on these bacteria, C. B. van Niel concluded that photosynthesis is a redox (reduction–oxidation) process

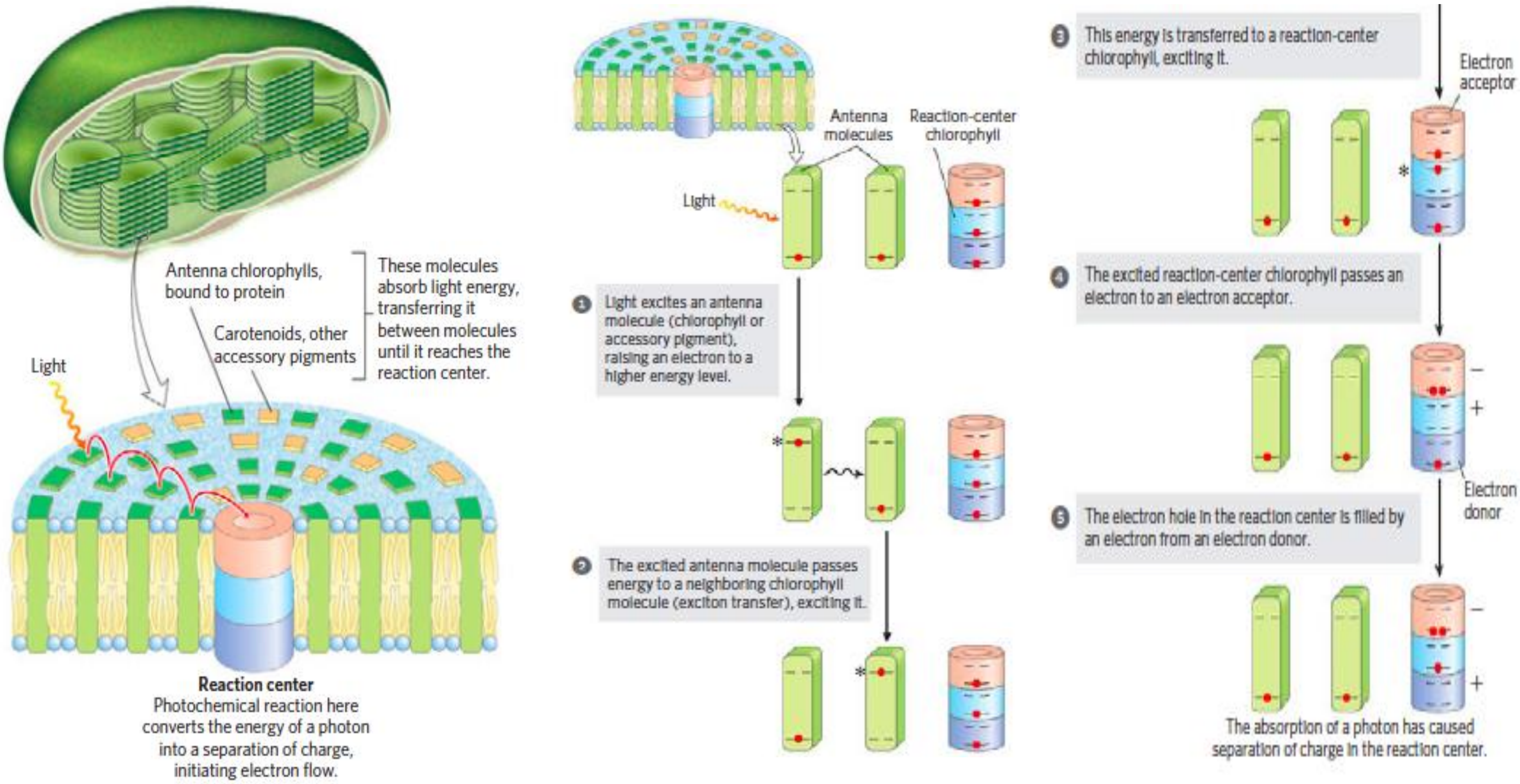


Photosystems



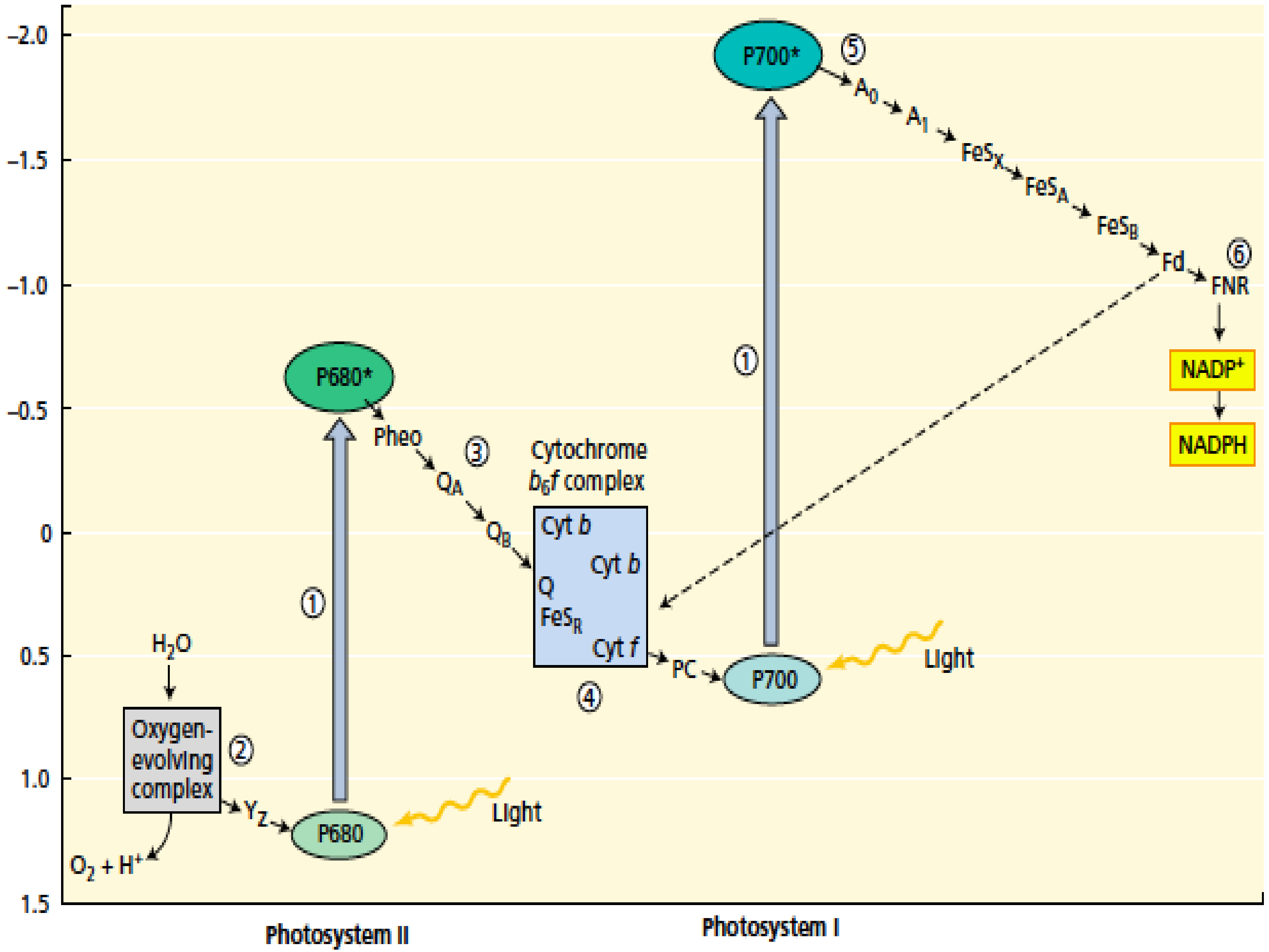
- Photosystem I produces a strong reductant, capable of reducing NADP+, and a weak oxidant.
- Photosystem II produces a very strong oxidant, capable of oxidizing water, and a weaker reductant than the one produced by photosystem I.

Photochemical reaction centers

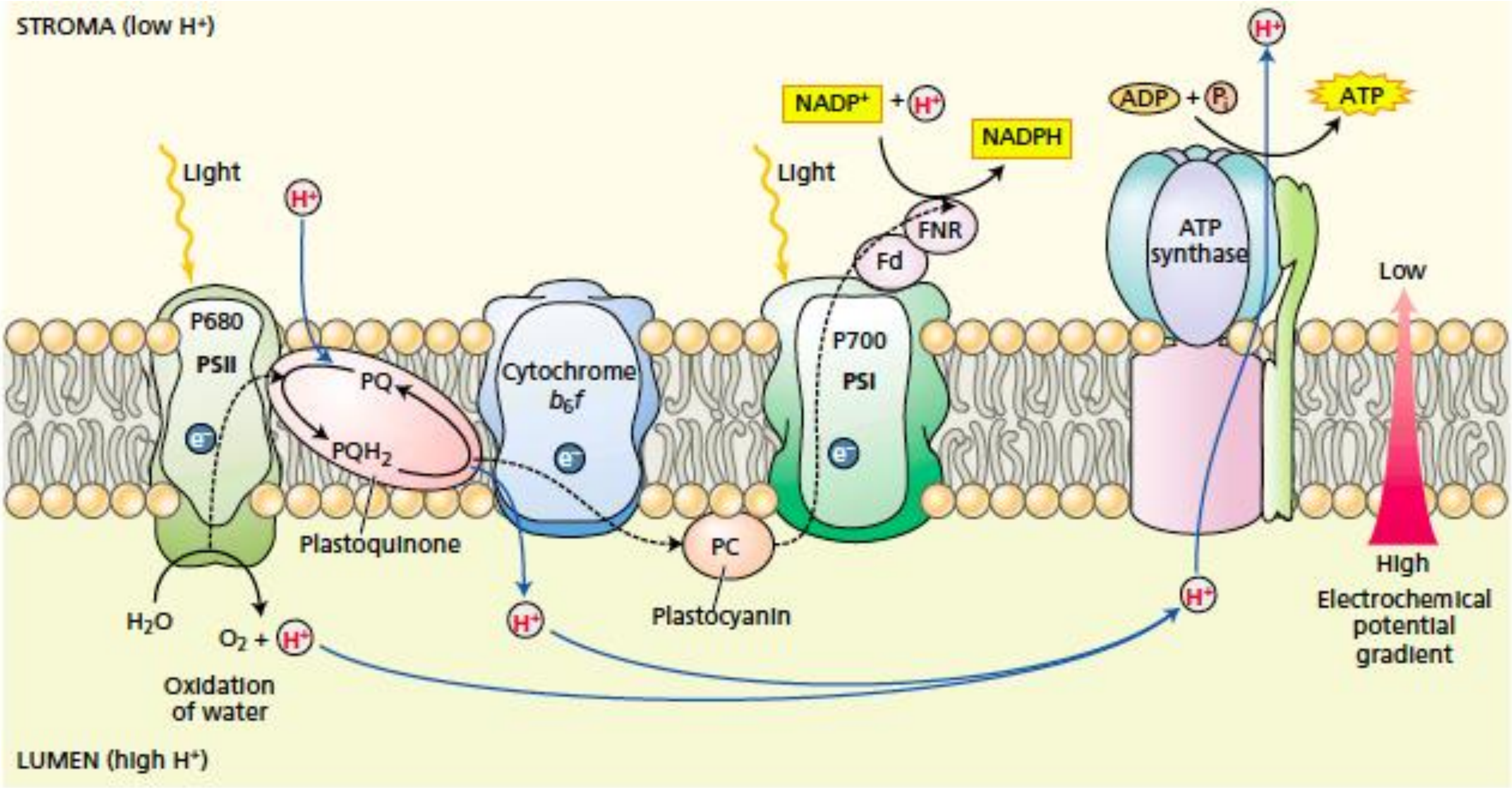


The central photochemical event-Light driven electron flow

- Photosystem II oxidizes water to O2 in the thylakoid lumen and in the process releases protons into the lumen.
- Cytochrome b6 f receives electrons from PSII and delivers them to PSI. It also transports additional protons into the lumen from the stroma.
- Photosystem I reduces NADP+ to NADPH in the stroma by the action of ferredoxin (Fd) and the flavoprotein ferredoxin–NADP reductase (FNR).
- ATP synthase produces ATP as protons diffuse back through it from the lumen into the stroma.

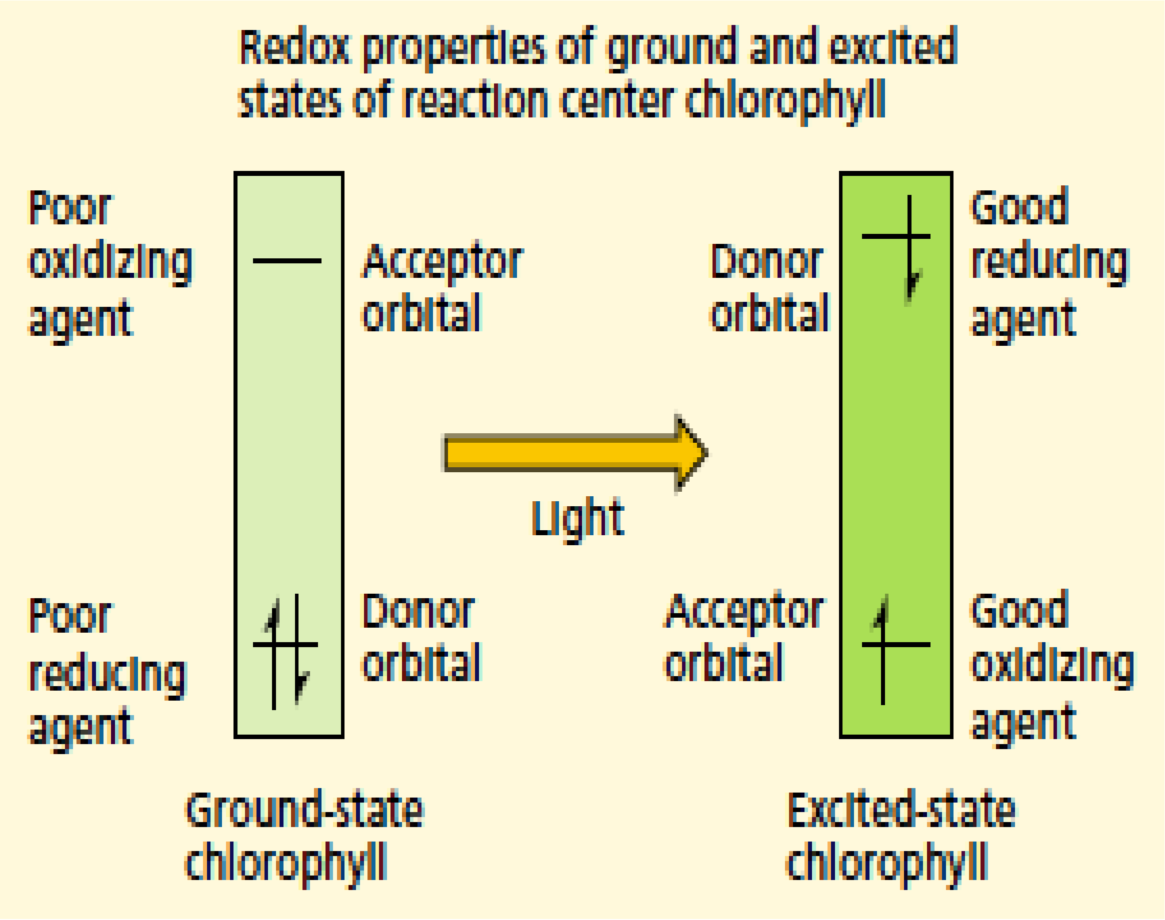


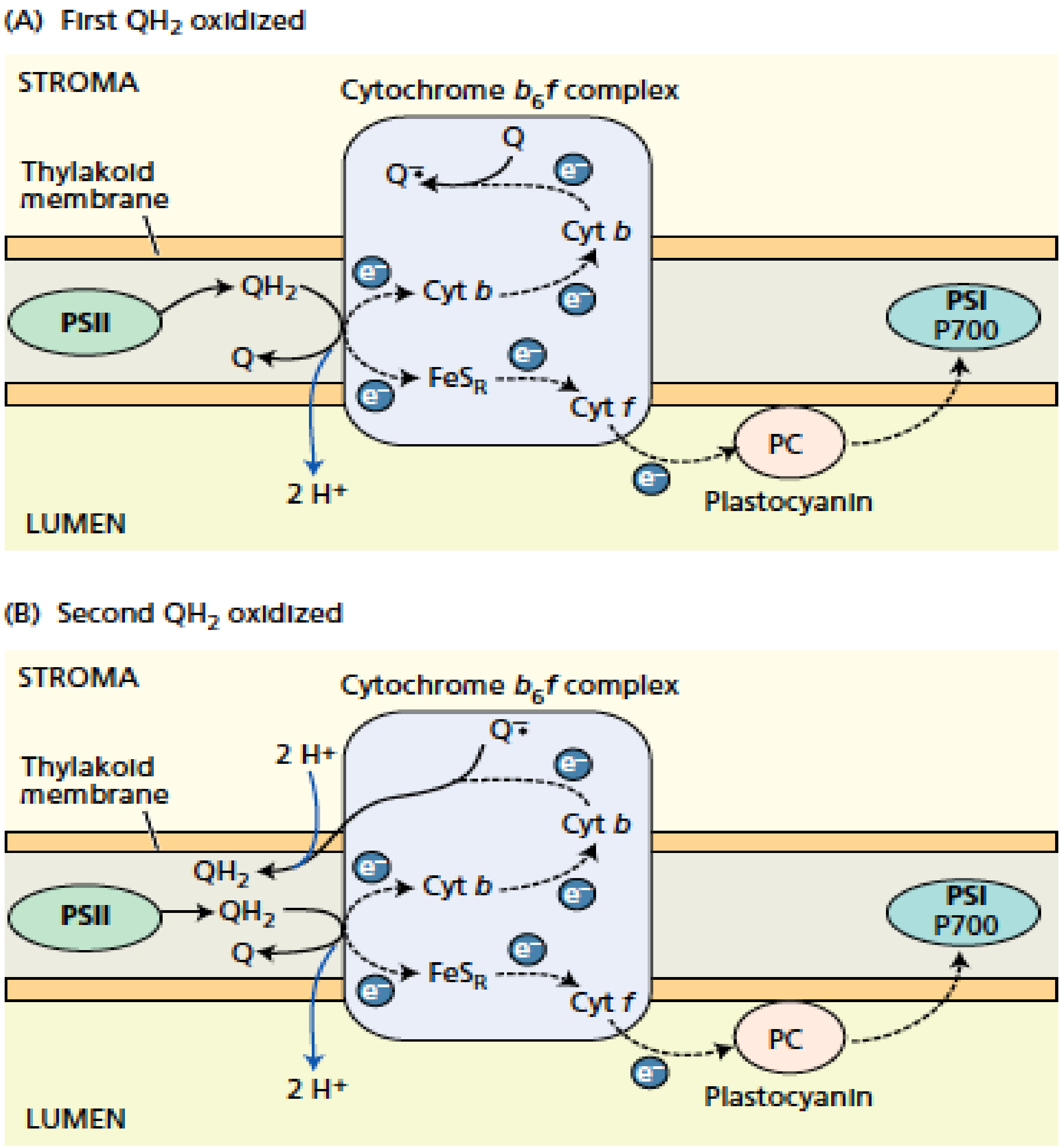
The central photochemical event



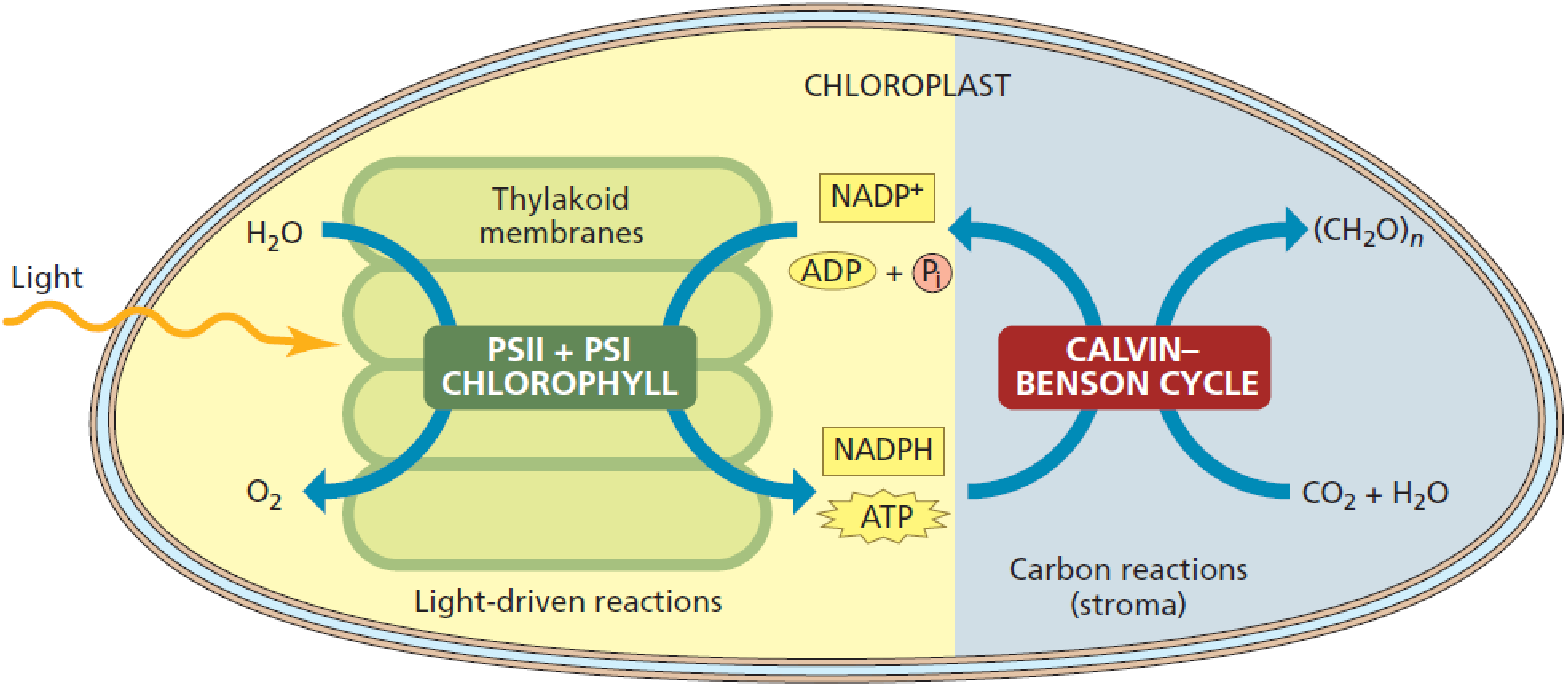
The central photochemical event

Orbital occupation diagram for the ground and excited states of reaction center chlorophyll.



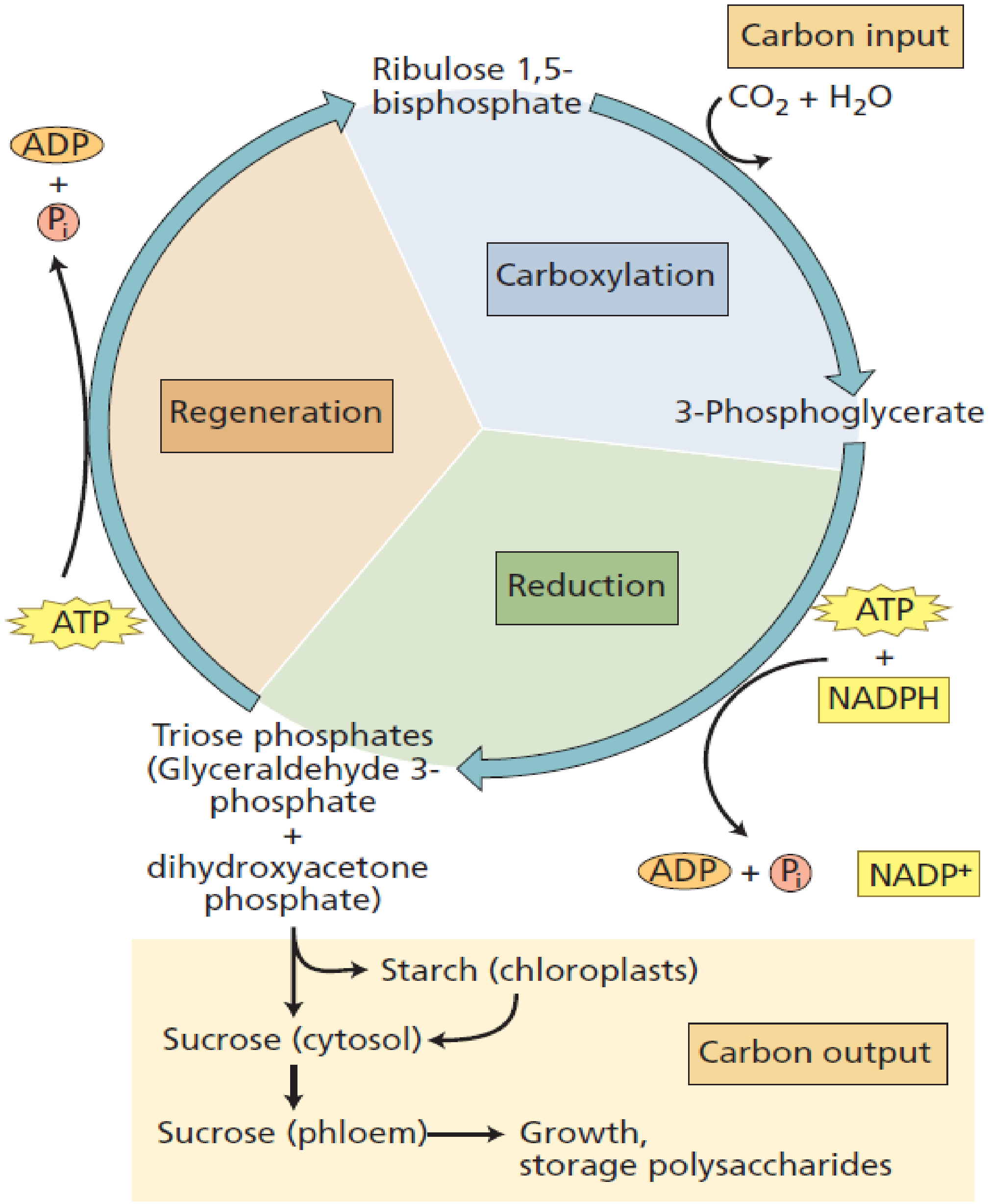


Photosynthesis-Dark cycles (Carbon reactions)



The light and carbon reactions of photosynthesis. Light is required for the generation of ATP and NADPH. The ATP and NADPH are consumed by the **carbon reactions**, which reduce carbon dioxide to carbohydrate.

Calvin-Benson Cycle (C3 cycle)



1. ***Carboxylation of the CO2 acceptor molecule***: reaction of CO2 and water with a five-carbon acceptor molecule (ribulose 1,5-bisphosphate) to generate two molecules of a three-carbon intermediate (3-phosphoglycerate)

2. ***Reduction of 3-phosphoglycerate***. The 3-phosphoglycerate is converted to three-carbon carbohydrates (triose phosphates)

3. ***Regeneration of the CO2 acceptor ribulose 1,5-bisphosphate***. The cycle is completed by regeneration of ribulose 1,5-bisphosphate through a series of ten enzyme-catalyzed reactions, one requiring ATP.

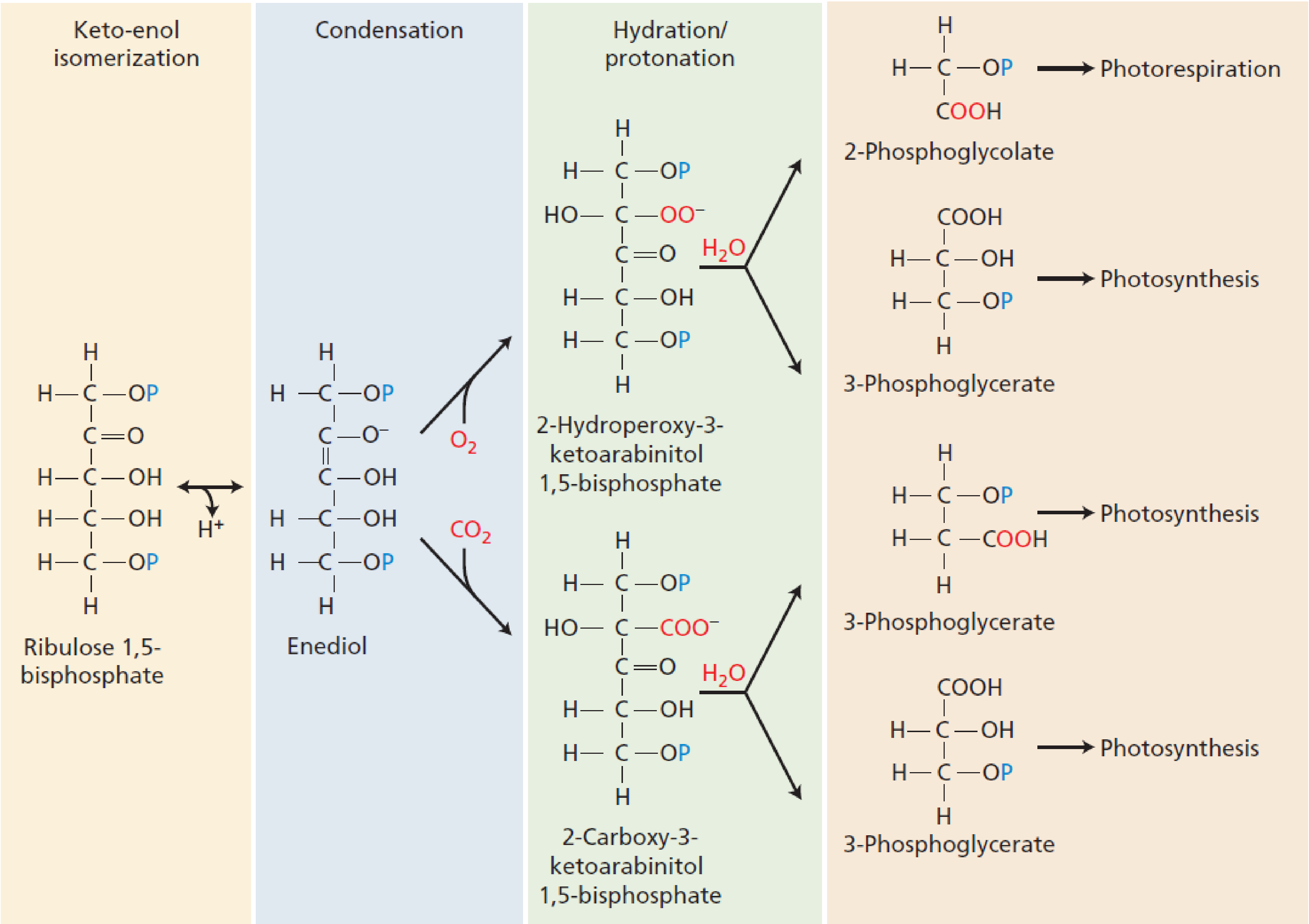
Calvin-Benson Cycle (C3 cycle)

TABLE 8.1 Reactions of the Calvin–Benson cycle

Enzyme	Reaction
1. Ribulose 1,5-bisphosphate carboxylase/oxygenase (rubisco)	Ribulose 1,5-bisphosphate + CO_2 + H_2O → 2 3-phosphoglycerate
2. 3-Phosphoglycerate kinase	3-Phosphoglycerate + ATP → 1,3-bisphosphoglycerate + ADP
3. NADP–glyceraldehyde-3-phosphate dehydrogenase	1,3-Bisphosphoglycerate + NADPH + H^+ → glyceraldehyde 3-phosphate + $NADP^+$ + P_i
4. Triose phosphate isomerase	Glyceraldehyde 3-phosphate → dihydroxyacetone phosphate
5. Aldolase	Glyceraldehyde 3-phosphate + dihydroxyacetone phosphate → fructose 1,6-bisphosphate
6. Fructose 1,6-bisphosphatase	Fructose 1,6-bisphosphate + H_2O → fructose 6-phosphate + P_i
7. Transketolase	Fructose 6-phosphate + glyceraldehyde 3-phosphate → erythrose 4-phosphate + xylulose 5-phosphate
8. Aldolase	Erythrose 4-phosphate + dihydroxyacetone phosphate → sedoheptulose 1,7-bisphosphate
9. Sedoheptulose 1,7-bisphosphatase	Sedoheptulose 1,7-bisphosphate + H_2O → sedoheptulose 7-phosphate + P_i
10. Transketolase	Sedoheptulose 7-phosphate + glyceraldehyde 3-phosphate → ribose 5-phosphate + xylulose 5-phosphate
11a. Ribulose 5-phosphate epimerase	Xylulose 5-phosphate → ribulose 5-phosphate
11b. Ribose 5-phosphate isomerase	Ribose 5-phosphate → ribulose 5-phosphate
12. Phosphoribulokinase (ribulose 5-phosphate kinase)	Ribulose 5-phosphate + ATP → ribulose 1,5-bisphosphate + ADP + H^+

Note: P_i stands for inorganic phosphate.

Calvin-Benson Cycle (C3 cycle)



Calvin-Benson Cycle (C3 cycle)

1 In summary, the Calvin-Benson cycle produces, six triose phosphates, NADP and ADP

$$3\ CO_2 + 3\ \text{ribulose 1,5-bisphosphate} + 3\ H_2O + 6\ NADPH + 6\ H^+ + 6\ ATP \\ \downarrow \\ 6\ \text{Triose phosphates} + 6\ NADP^+ + 6\ ADP + 6\ P_i$$

2

From these six triose phosphates, five are used in the regeneration phase that restores the CO2 acceptor (ribulose 1,5-bisphosphate)

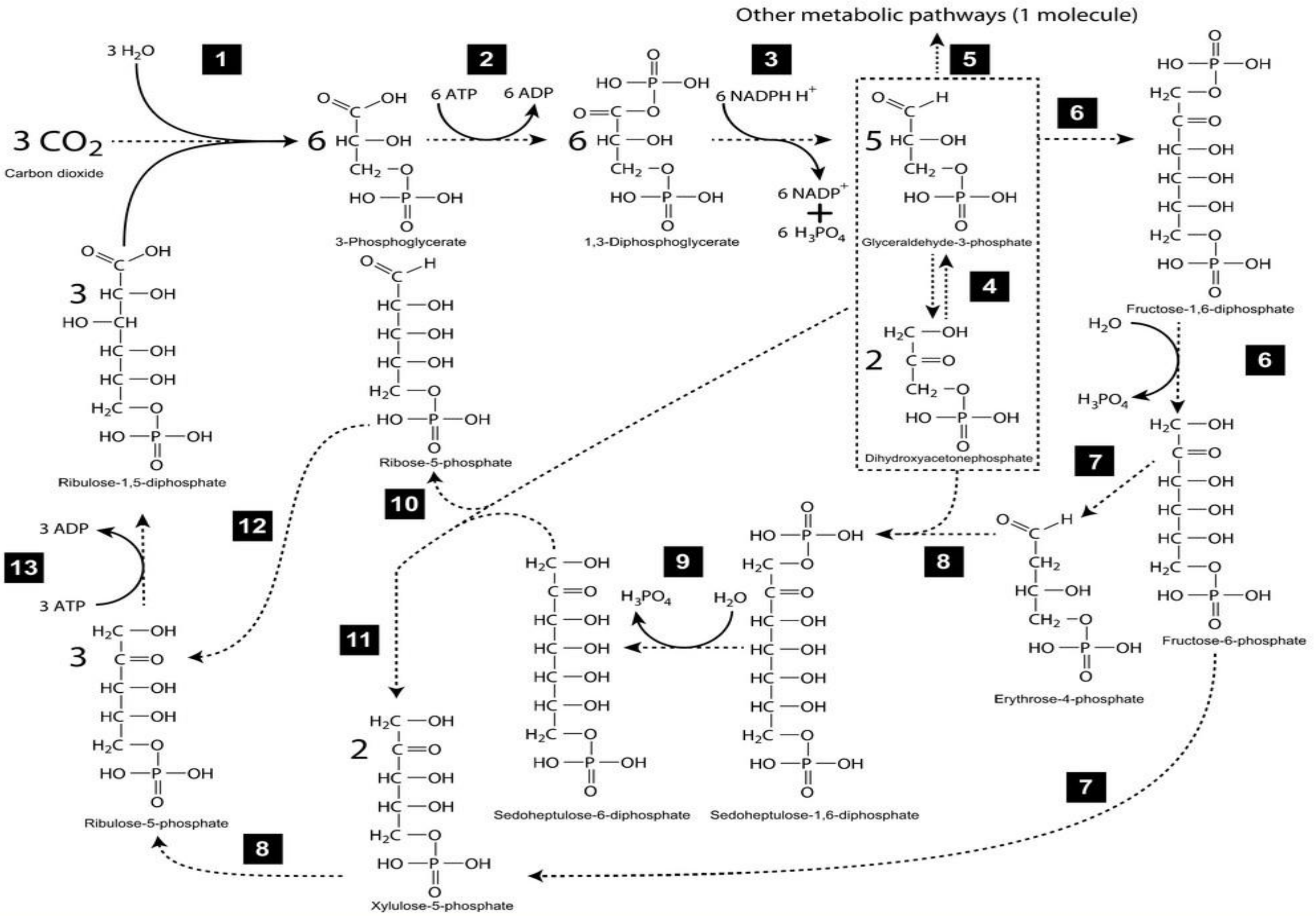
$$5\ \text{Triose phosphates} + 3\ ATP + 2\ H_2O \rightarrow 3\ \text{ribulose 1,5-bisphosphate} + 3\ ADP + 2\ P_i$$

3 Net, the fixation of three CO2 into one triose phosphate uses 6 NADPH and 9 ATP

$$3\ CO_2 + 5\ H_2O + 6\ NADPH + 9\ ATP \\ \downarrow \\ \text{Glyceraldehyde 3-phosphate} + 6\ NADP^+ + 9\ ADP + 8\ P_i$$

The Calvin–Benson cycle uses two molecules of NADPH and three molecules of ATP to assimilate a single molecule of CO_2.

Calvin cycle - C3 pathway of photosynthesis (dark phase)



Photorespiration- C2 cycle

RuBP oxygenase-carboxylase (**rubisco**), a key enzyme in photosynthesis, is the molecular equivalent of a good friend with a bad habit.

In the process of **carbon fixation**, rubisco incorporates carbon dioxide into an organic molecule during the first stage of the Calvin cycle.

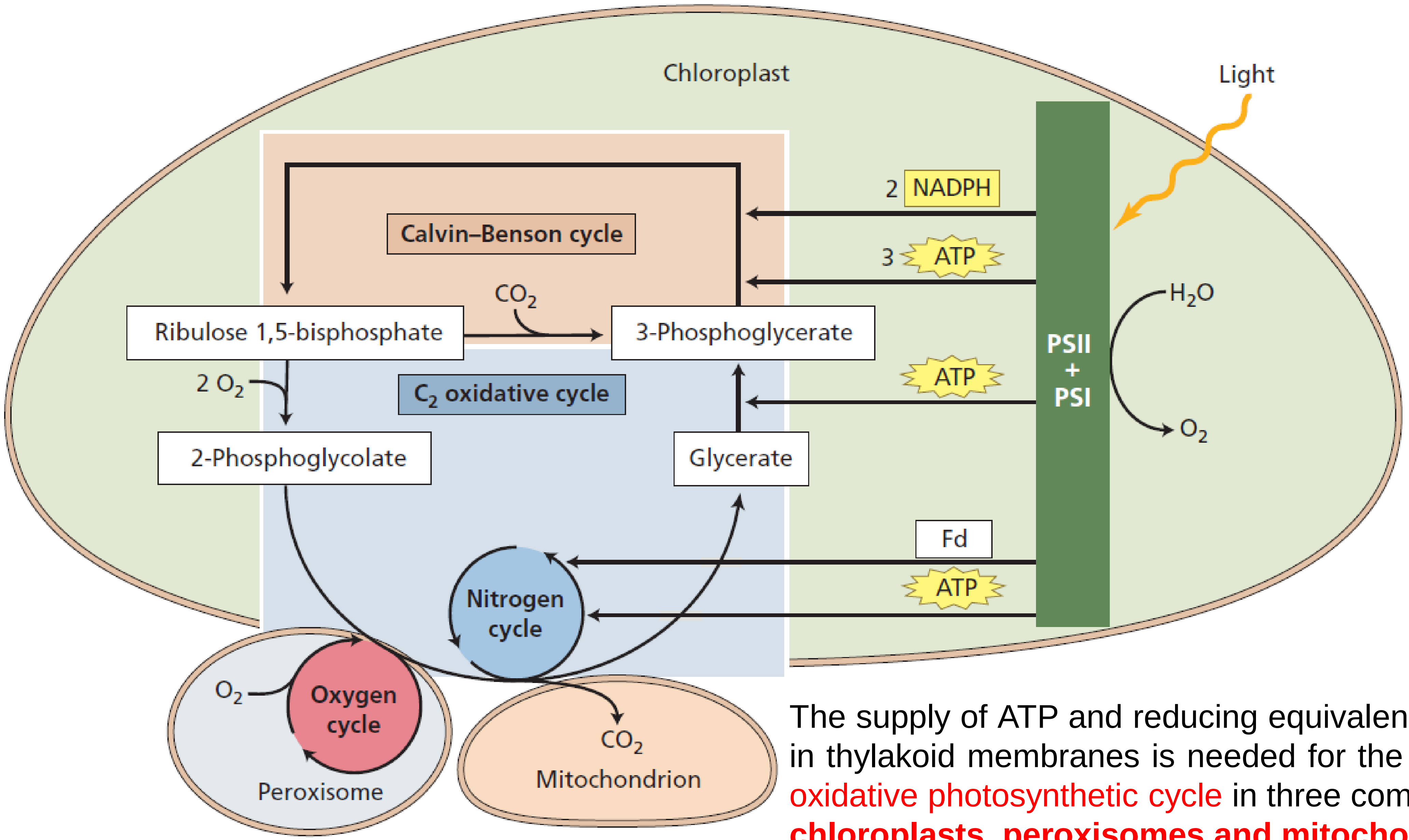
Rubisco is so important to plants that it makes up 30% or more of the soluble protein in a typical plant

But rubisco also has a major flaw: instead of always using CO2 as a substrate, it sometimes picks up O2 instead.

This side reaction initiates a pathway called **photorespiration**, which, rather than fixing carbon, actually leads to the loss of already-fixed carbon as CO2.

Photorespiration wastes energy and decreases sugar synthesis, so when rubisco initiates this pathway, it's committing a serious molecular *faux pas*.

Photorespiration- C2 cycle



The supply of ATP and reducing equivalents from light reactions in thylakoid membranes is needed for the functioning of the C2 oxidative photosynthetic cycle in three compartments: **chloroplasts, peroxisomes and mitochondria**.

Photorespiration- C2 cycle

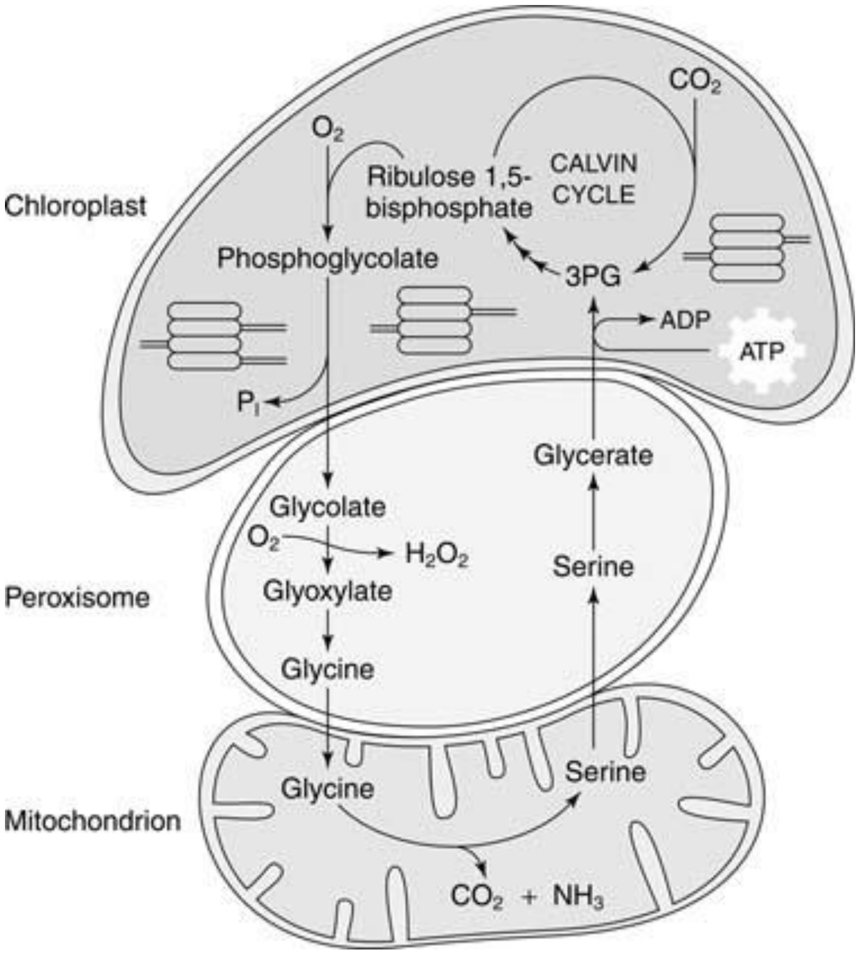


TABLE 8.2 Reactions of the C_2 oxidative photosynthetic carbon cycle

Reaction[a]	Enzyme
1. 2 Ribulose 1,5-bisphosphate + 2 O_2 → 2 2-phosphoglycolate + 2 3-phosphoglycerate	Rubisco
2. 2 2-Phosphoglycolate + 2 H_2O → 2 glycolate + 2 P_i	Phosphoglycolate phosphatase
3. 2 Glycolate + 2 O_2 → 2 glyoxylate + 2 H_2O_2	Glycolate oxidase
4. 2 H_2O_2 → 2 H_2O + O_2	Catalase
5. 2 Glyoxylate + 2 glutamate → 2 glycine + 2 2-oxoglutarate	Glutamate:glyoxylate aminotransferase
6. Glycine + NAD^+ + [GDC] → CO_2 + NH_4^+ + NADH + [GDC-THF-CH_2]	Glycine decarboxylase complex (GDC)
7. [GDC-THF-CH_2] + glycine + H_2O → serine + [GDC]	Serine hydroxymethyl transferase
8. Serine + 2-oxoglutarate → hydroxypyruvate + glutamate	Serine:2-oxoglutarate aminotransferase
9. Hydroxypyruvate + NADH + H^+ → glycerate + NAD^+	Hydroxypyruvate reductase
10. Glycerate + ATP → 3-phosphoglycerate + ADP	Glycerate kinase
11. Glutamate + NH_4^+ + ATP → glutamine + ADP + Pi	Glutamine synthetase
12. 2-Oxoglutarate + glutamine + 2 Fd_{red} + 2 H^+ → 2 glutamate + 2 Fd	Ferredoxin-dependent glutamate synthase (GOGAT)

[a]Locations: Chloroplasts; peroxisomes; mitochondria. Fd: ferredoxin.

C4 cycle (Hatch–Slack cycle)

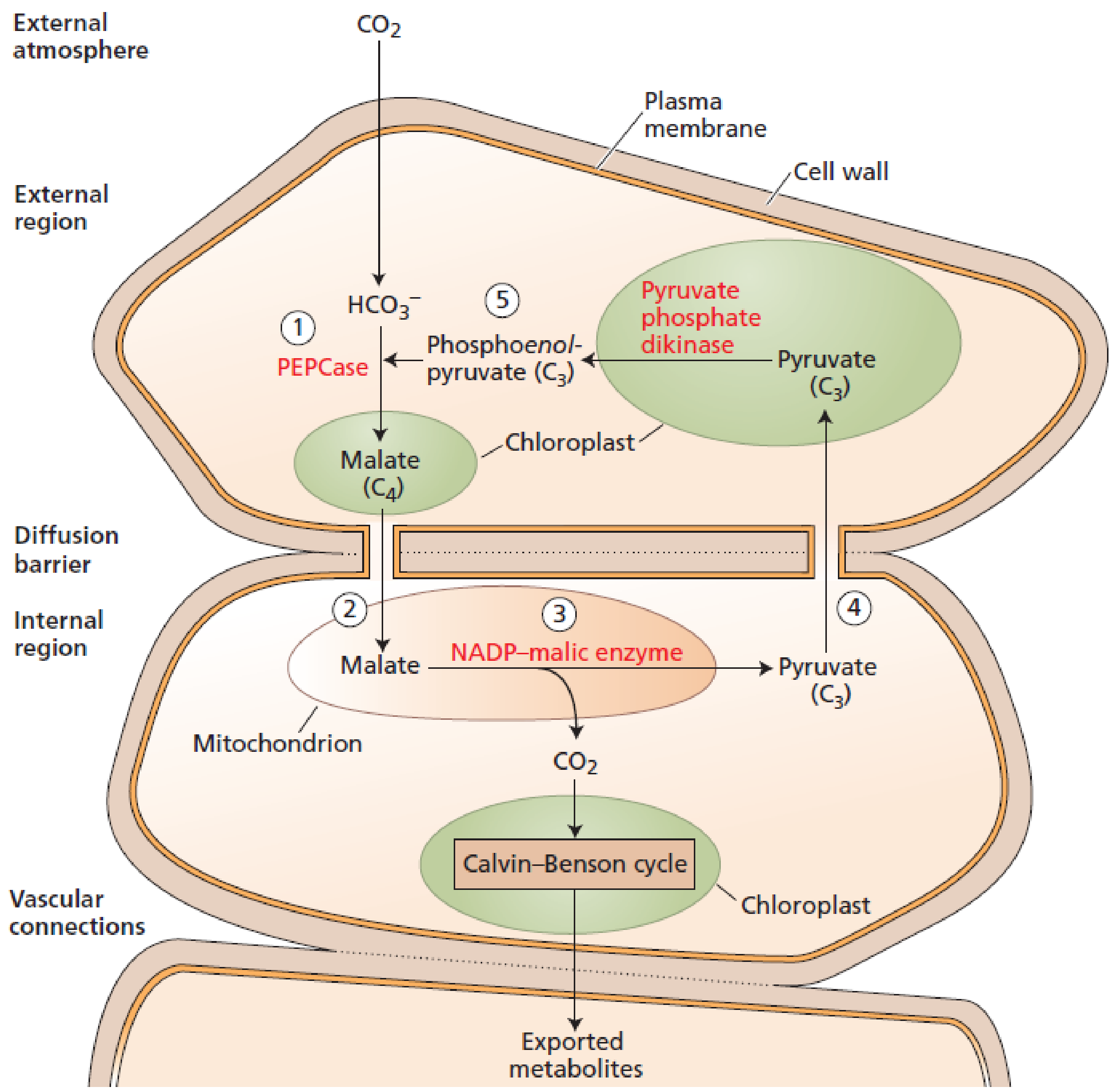


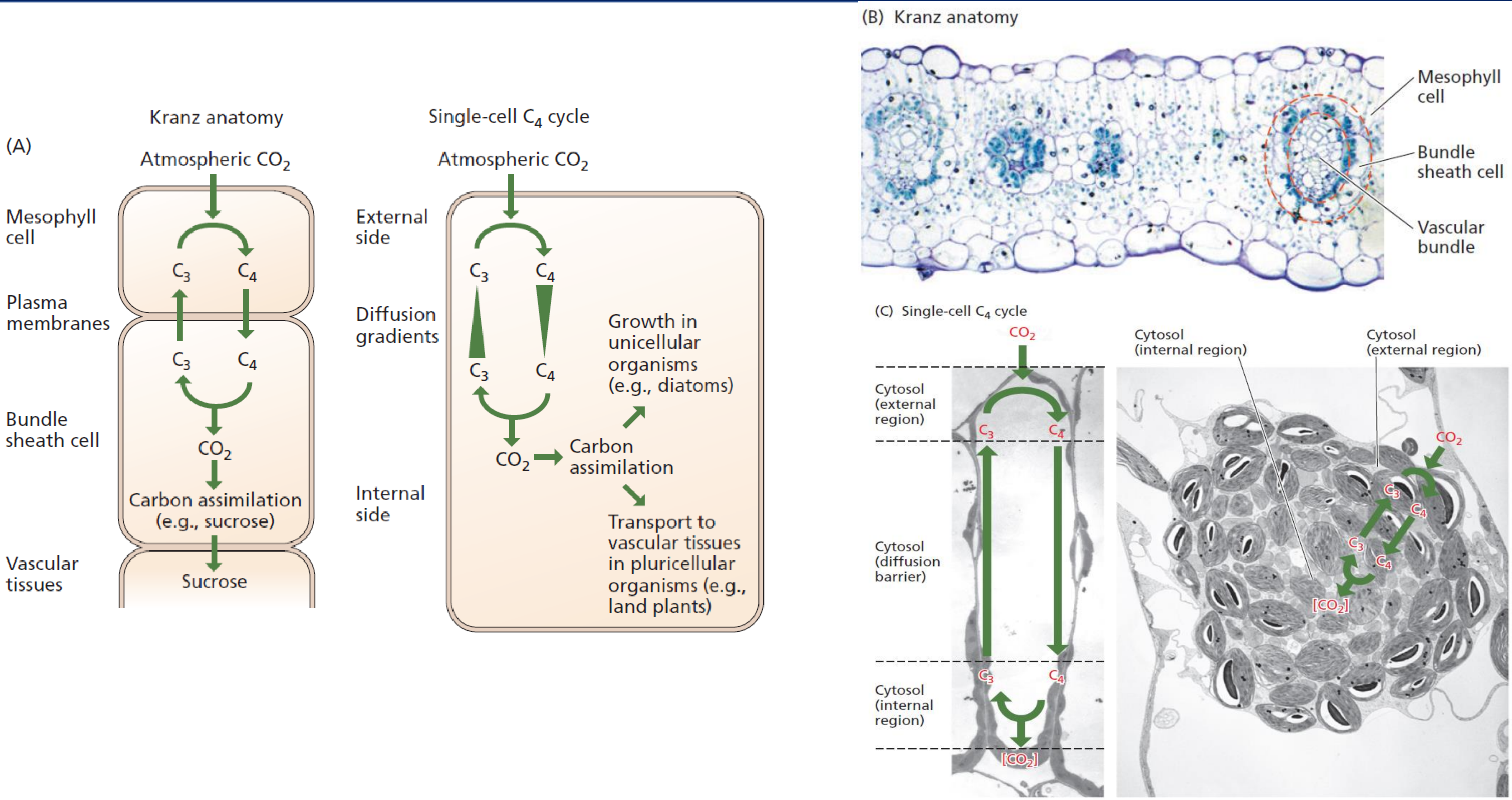
Table 8.4 Reactions of C_4 photosynthesis

Enzyme	Reaction
1. PEPCase	Phospho*enol*pyruvate + HCO_3^- → oxaloacetate + P_i
2. NADP–malate dehydrogenase	Oxaloacetate + NADPH + H^+ → malate + $NADP^+$
3. Aspartate aminotransferase	Oxaloacetate + glutamate → aspartate + 2-oxoglutarate
Decarboxylating enzymes	
4a. NADP–malic enzyme	Malate + $NADP^+$ → pyruvate + CO_2 + NADPH + H^+
4b. NAD–malic enzyme	Malate + NAD^+ → pyruvate + CO_2 + NADH + H^+
5. Phospho*enol*pyruvate carboxykinase	Oxaloacetate + ATP → phospho*enol*pyruvate + CO_2 + ADP
6. Alanine aminotransferase	Pyruvate + glutamate → alanine + 2-oxoglutarate
7. Pyruvate–phosphate dikinase	Pyruvate + P_i + ATP → phospho*enol*pyruvate + AMP + PP_i
8. Adenylate kinase	AMP + ATP → 2 ADP
9. Pyrophosphatase	PP_i + H_2O → 2 P_i

Note: P_i and PP_i stand for inorganic phosphate and pyrophosphate, respectively.

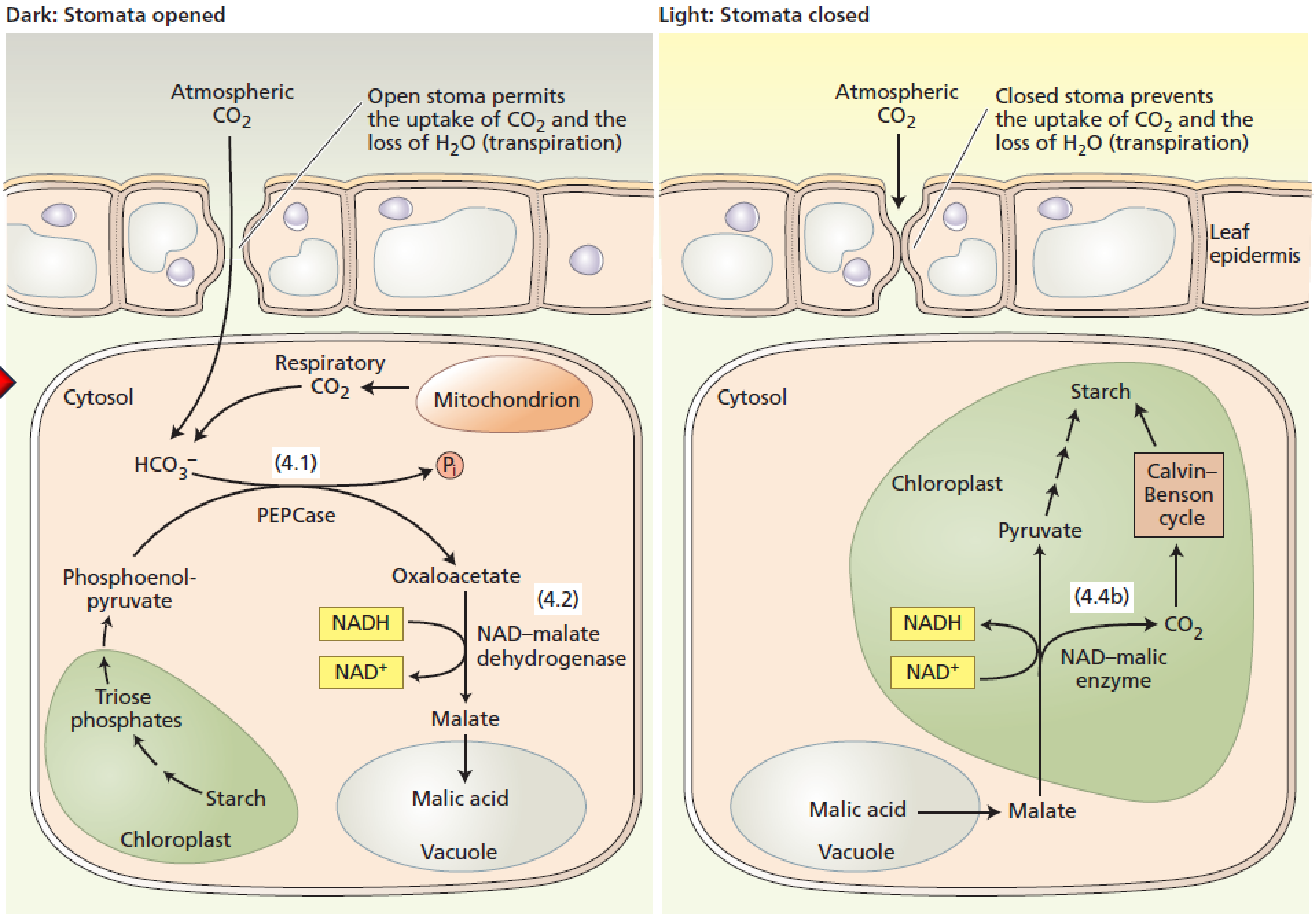
- Compensation for limitations associated with low levels of atmospheric CO2
- Most productive crops on the planet (e.g., corn; sugarcane, sorghum) use this mechanism to enhance the catalytic capacity of rubisco

C4 cycle (Hatch–Slack cycle)



Crassulacean Acid Metabolism (CAM) pathways

Plants inhabit arid environments with seasonal water availability, including commercially important plants such as pineapple (*Ananas comosus*), agave (*Agave* spp.), cacti (Cactaceae), and orchids (Orchidaceae)



Summary

- Dark cycles/ Carbon reactions
- Carbon assimilations
- C3 or Calvin-Benson cycle
- C2 cycle or Photorespiration
- C4 cycle (Hatch–Slack cycle) and its importance
- CAM pathways