5.2 Pathways of Photosynthesis

Figure 1 Although they do not have chloroplasts, prokaryotes like these cyanobacteria photosynthesize using the same chemical processes that eukaryotes use.

Solar energy is the most abundant and widespread source of energy on Earth. In a single day, enough solar energy strikes Earth's surface to supply all human energy needs for 55 years. With such an abundant supply, scientists are researching and developing technologies to capture and use sunlight more efficiently to meet our growing energy demands. In fact, life on Earth evolved a pathway to harness this same energy source to meet its needs—a pathway living organisms have been using for well over 2 billion years.

This section examines the details of this extraordinary pathway. It will focus on how eukaryotes carry out photosynthesis in chloroplasts inside specialized photosynthetic cells. Keep in mind that these same chemical processes occur in photosynthetic prokaryotes, even though they do not have chloroplasts and other specialized structures that eukaryotes have. In prokaryotes, the photosynthesis reactions occur both in the cytosol and along folds in the cell membrane (**Figure 1**).

Light-Dependent Reactions

Photosystem II and photosystem I are the two light-capturing complexes used by most photoautotrophs. As in all electron transport systems, the electron carriers of the photosynthetic system consist of non-protein organic groups that alternate between being oxidized and being reduced as electrons move through the system. The carriers include compounds that are similar in structure and function to those in mitochondrial electron transport chains.

Photosystem II

As life emerged on Earth, it began to substantially change the environment. Earth's early atmosphere may have consisted of as little as 3 % oxygen gas and as much as 10 % carbon dioxide. We know that the composition of today's atmosphere is 21 % oxygen and only 0.04 % carbon dioxide. When simple organisms began using photosynthesis as a way to store energy in chemical compounds, they used water as their source of hydrogen atoms and their electrons. A by-product of this process was oxygen gas. As early photosynthetic organisms flourished, they generated an abundance of oxygen gas that accumulated in the atmosphere. This oxygen enabled the evolution of the rich diversity of aerobic life forms that we see on Earth today. CAREER LINK

The splitting of water into electrons, protons, and oxygen gas occurs in photosystem II. When a photon of light strikes the antenna complex, it is absorbed and its energy is transferred to the molecule P680, and one of its electrons changes from the ground state to an excited state, resulting in the energized molecule P680* (**Figure 2(a),** next page). The excited electron is then transferred to the primary acceptor molecule, which becomes negatively charged, while the P680 now carries a positive charge (**Figure 2(b)**, next page).

The resulting positive ion $P680^+$ is now extremely electronegative and can exert forces strong enough to remove an electron from a molecule of water! $P680^+$ is the strongest oxidant known in biology. The reduction of $P680^+$ to P680 by electrons from water is facilitated by an enzyme subunit of photosystem II called the water-splitting complex. This complex is inside the thylakoid membrane, facing the lumen.

Driven by this powerful electronegative pull, the water-splitting complex oxidizes a molecule of water, passing an electron to the $P680^+$ to make it neutral again (**Figure 2(c)**, next page). The acceptor molecule also transfers an electron to a molecule of plastoquinone (PQ) and becomes neutral, allowing the photon absorption process to start all over again. Note that this entire process occurs twice for each water molecule that is completely oxidized.

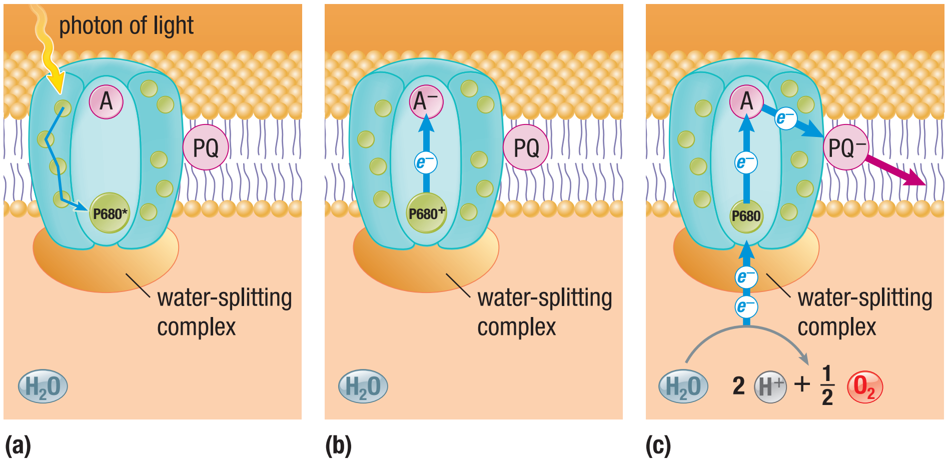

Figure 2 (a) The actions of photosystem II begin when a photon energizes an electron in P680, forming P680*. (b) The energized chlorophyll (P680*) then transfers the high-energy electron to acceptor A in the reaction centre. P680 is now positively charged. (c) The positive P680$^+$ ion then oxidizes water, and the high-energy electron is transferred from the reaction centre to the carrier molecule plastoquinone (PQ). This process releases both oxygen gas and protons into the lumen.

Linear Electron Transport and ATP Synthesis

The first steps in the electron transport chain within the thylakoid membrane involve the transfer of the high-energy electron to the primary acceptor and then to plastoquinone (PQ). Here, the light energy that is transferred to the electrons is used to generate a proton gradient. At the end of the electron transport chain, a second photosystem energizes the electrons a second time. The high-energy electrons can then be transferred to NADPH carrier molecules. The major steps of this electron transport system are illustrated in **Figure 3** and described below.

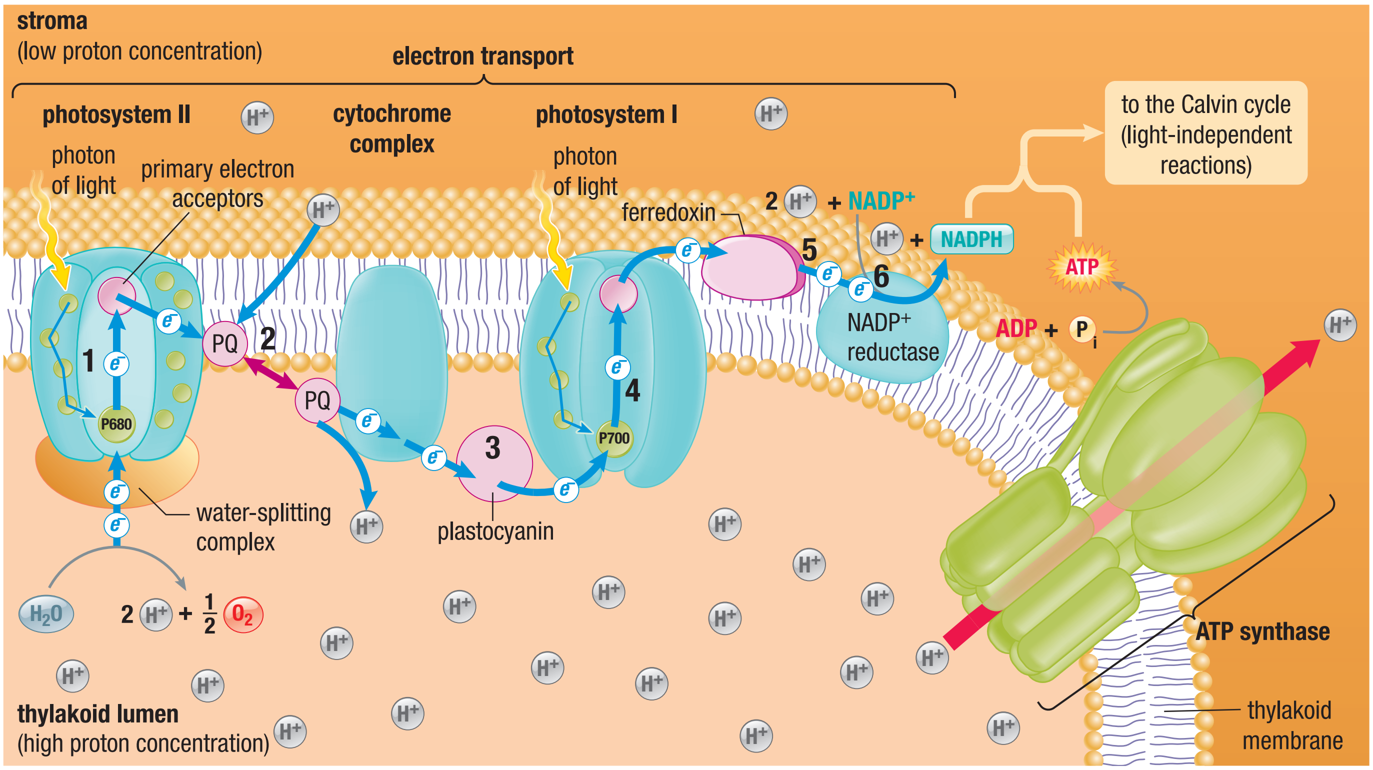

Figure 3 This model of the eukaryotic thylakoid membrane illustrates the major protein and redox cofactors required for non-cyclic electron transport and ATP synthesis by chemiosmosis. The four major protein complexes are photosystem II, the cytochrome complex, photosystem I, and ATP synthase. The blue arrow illustrates the pathway of electron transport.

1. **Oxidation of P680:** The absorption of light energy by photosystem II results in the formation of an excited-state P680 (P680*) molecule. This molecule is rapidly oxidized, transferring a high-energy electron to the primary acceptor.
2. **Oxidation-reduction of plastoquinone:** From the primary acceptor, the electrons transfer to plastoquinone (PQ), which moves through the lipid bilayer and acts as an electron shuttle between photosystem II and the cytochrome complex. As plastoquinone accepts electrons from photosystem II, it also gains protons (H^+) from the stroma. When PQ donates electrons to the cytochrome complex, it also releases protons into the lumen, increasing the proton concentration there.
3. **Electron transfer from the cytochrome complex and shuttling by plastocyanin:** From the cytochrome complex, electrons pass to the mobile carrier plastocyanin, which shuttles electrons from the cytochrome complex to photosystem I.
4. **Oxidation-reduction of P700:** When a photon of light is absorbed by photosystem I, an electron is excited and P700* forms. The P700* chlorophyll transfers its electron to the primary electron acceptor of photosystem I, forming $P700^+$. $P700^+$ can now act as an electron acceptor and is reduced back to P700 by the oxidation of plastocyanin.
5. **Electron transfer to $NADP^+$ by ferredoxin:** The first electron from P700* is transported by a short sequence of carriers within photosystem I. It is then transferred to ferredoxin, an iron-sulfur protein. The oxidation of ferredoxin results in the transfer of the electron to $NADP^+$, reducing it to NADP.
6. **Formation of NADPH:** A second electron is transferred to NADP by another molecule of ferredoxin. This second electron and a proton (H^+) from the stroma are added to NADP by the $NADP^+$ reductase to form NADPH. NADPH is now carrying two high-energy electrons. The concentration of protons in the stroma decreases as a result of this NADPH formation. Along with the movement of protons from stroma to lumen by plastoquinone and the splitting of water into protons, these three processes create a much higher proton concentration inside the lumen than outside in the stroma.

This pathway is referred to as linear (or non-cyclic) to distinguish it from an alternative process in which electrons are not passed on to NADPH. The proton gradient is used to generate ATP with the same kind of ATP synthase complexes found in mitochondrial membranes.

CHEMIOSMOTIC SYNTHESIS OF ATP

Recall from Chapter 4 that during cellular respiration, a proton gradient across a membrane is used as a source of energy to generate ATP by chemiosmosis. In the electron transport of photosynthesis just described, a similar proton gradient is established across the thylakoid membrane by three mechanisms (**Figure 4,** next page).

1. Protons are taken into the lumen by the reduction and oxidation of plastoquinone as it moves from photosystem II to the cytochrome complex and back again.
2. The concentration of protons inside the lumen is increased by the addition of two protons for each water molecule that is split in the lumen.
3. The removal of one proton from the stroma for each NADPH molecule formed decreases the concentration of protons in the stroma outside the thylakoid.

The result of these three mechanisms is a substantial proton gradient across the membrane. The higher concentration of protons inside the membrane creates a substantial proton-motive force that drives protons out of the lumen, back into the stroma. However, the thylakoid membrane allows protons to pass out into the stroma only through the pores in the protein complexes of ATP synthase, which are embedded in the membrane (Figure 4). This process, called chemiosmosis, is the same process that occurs in cellular respiration. The chloroplast's ATP synthase is, in fact,

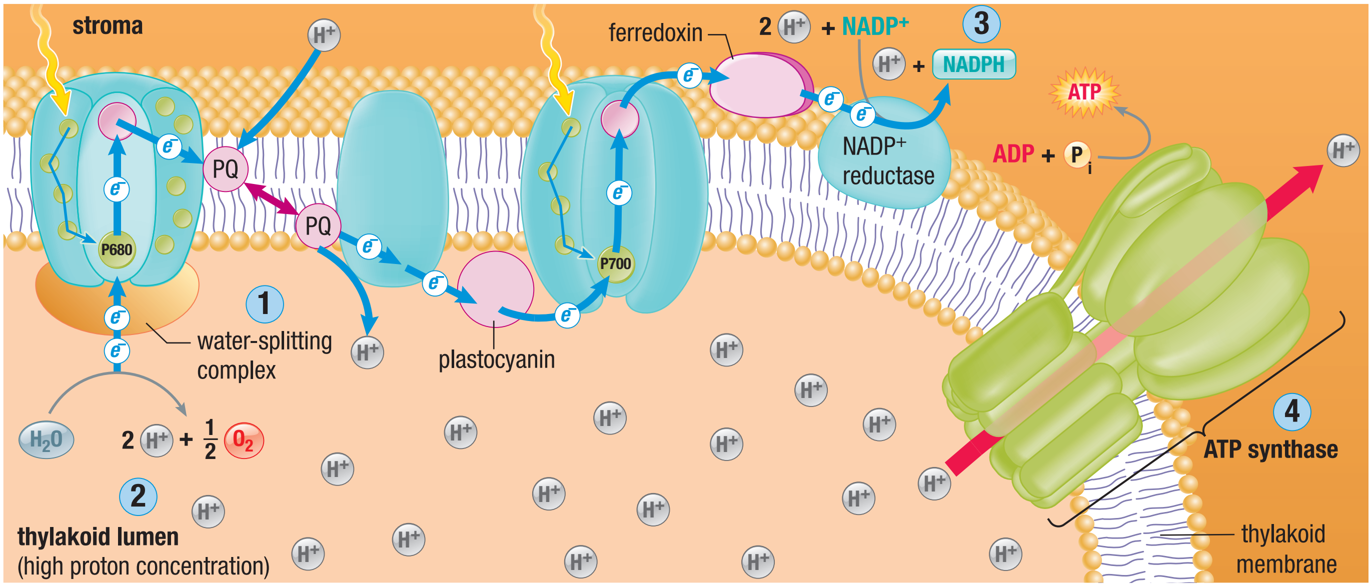

Figure 4 (1) A proton gradient is established by the carrying of protons across the membrane by PQ, (2) by the releasing of protons into the lumen during the oxidation of water, and (3) by the removal of protons from the stroma during the reduction of $NADP^+$. (4) ATP is then synthesized as protons move through the ATP synthase complex.

identical to the ATP synthase used in cellular respiration. As protons move through the channels in the ATP synthase, some of their free energy is captured and used to synthesize ATP from ADP and P_i.

THE ROLE OF LIGHT ENERGY

All electron transport chains operate by electrons being pulled spontaneously "downhill" from molecules with high-energy electrons (molecules that are easily oxidized) to molecules with progressively stronger attractions to electrons (molecules that are more electronegative). In mitochondrial respiration, the electron flow is "downhill" from high-energy NADH to very electronegative O_2. In photosynthesis, electron transport occurs by the same principle. The electron transport chain in respiration begins with high-energy NADH and finishes with low-energy H_2O. The opposite occurs in the electron transport chain in photosynthesis, which begins with low-energy H_2O and ends with high-energy NADPH. Therefore, to drive photosynthetic electron transport, low-energy electrons in water must be given enough potential energy to establish a proton gradient and enough energy to form NADPH. As you have seen, this dual function can be accomplished by the combined actions of photosystems I and II.

Figure 5 (next page) is a representation of the energy level changes that occur as electrons move along the electron transport chain of photosynthesis. First, by absorbing a photon of light, an electron in a P680 chlorophyll molecule of photosystem II gets excited and moves farther away from its nucleus. This high-energy electron is now held much less strongly by the P680* chlorophyll molecule. This enables the electron to be transferred to the primary electron acceptor.

A series of redox reactions follows, beginning with the oxidation of the acceptor molecule by plastoquinone and ending with the oxidation of plastocyanin by photosystem I. Each of these steps results in a small decrease in the free energy of the electron as oxidizing agents establish a stronger and stronger force of attachment to the electron. As you saw in Figure 4, these first steps released enough free energy to help establish a proton gradient across the thylakoid membrane. However, the electron is now bound to the strongly electronegative P700 chlorophyll molecule in photosystem I.

At this point, the primary acceptor of photosystem I is unable to pull away the strongly held low-energy electron from P700. This is overcome when photosystem I

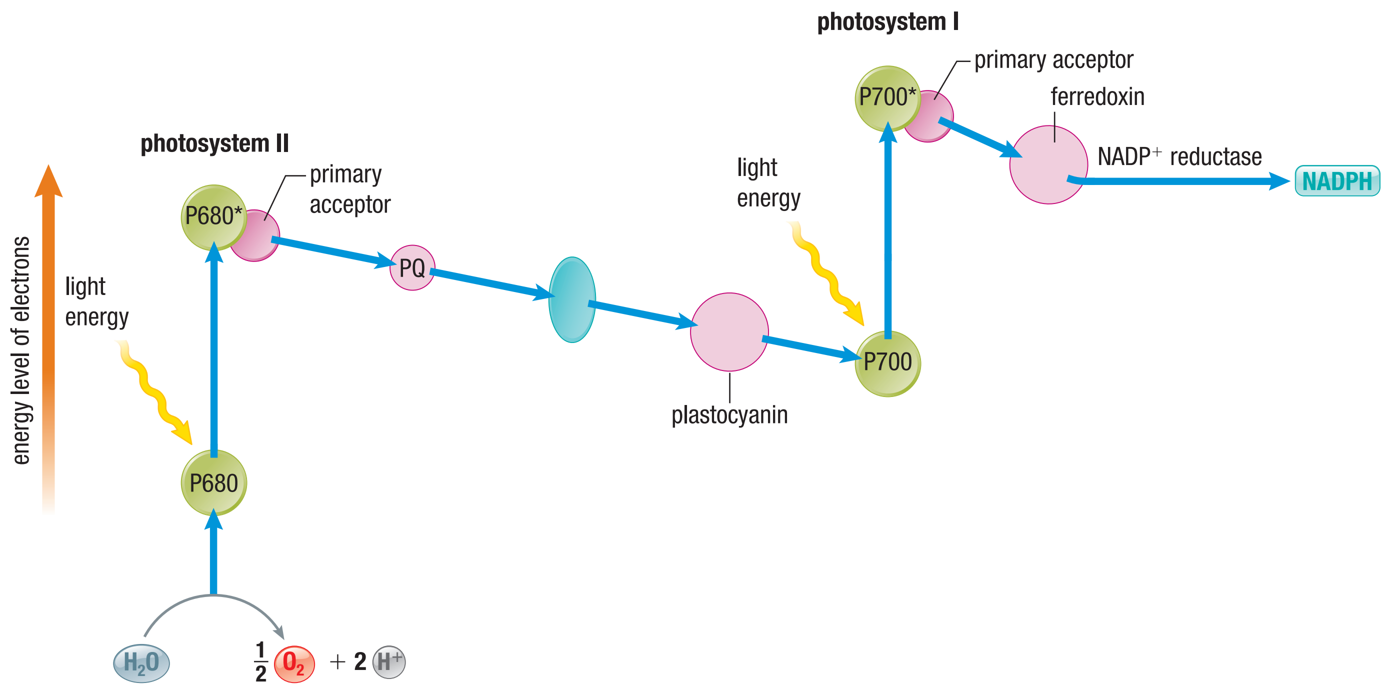

Figure 5 In the electron transport chain of the light-dependent reactions, the energy level of the electrons is increased dramatically by the light-absorbing actions of both photosystems II and I. The high-energy electrons at the end of the electron transport chain ultimately produce NADPH.

absorbs a photon of light, producing P700*. As was the case in photosystem II, the electron gains energy as it moves farther away from its nucleus. Since the electron is held less strongly it can be transferred to the acceptor molecule and along a second short redox pathway to $NADP^+$, which takes final possession of this high-energy electron.

Thus, two photons of light—one absorbed by photosystem II and another absorbed by photosystem I—are required to span the energy difference between H_2O and NADPH. Note that two electrons must be transported for every one molecule of NADPH produced. The conceptual chart shown in Figure 5 is sometimes referred to as the "Z scheme" because of the overall shape of the energy path of the electrons.

In respiratory electron transport, the electron flow is spontaneous from NADH to O_2, producing H_2O. By comparison, in photosynthetic electron transport, the electron flow is essentially the opposite, from H_2O to $NADP^+$. This electron flow does not occur spontaneously but must be boosted twice through the absorption of light energy.

Consider a physical analogy to the chemical pathway in Figure 5. Imagine that a cyclist travels along a path over two hills. The cyclist begins at the bottom of the first hill and must work hard, expending energy, to get to the top. Having reached the top of this hill, he stops pedalling and spontaneously coasts downhill a short distance, releasing a small amount of energy. Before getting back down to his original level, however, he comes to the base of another hill and must once again expend energy to climb. The top of this second hill is even higher than the first. At the top of the second hill, he starts to coast downhill again. The bottom of the second hill is higher than the start of the first hill. In this analogy, the energy to climb uphill is supplied by the cyclist rather than photons of light, but as in photosynthesis, the downhill parts of the pathway can happen spontaneously as energy is released.

LINEAR ELECTRON TRANSPORT—A BALANCE SHEET

You know that passing a single electron through the electron transport chain from photosystem II to $NADP^+$ requires two photons of light: one photon absorbed by photosystem II and a second photon absorbed by photosystem I. You also know that this process began with the oxidation of water and the production of oxygen gas. How many photons need to be absorbed by the photosynthetic apparatus to produce a single molecule of O_2? First, write out a balanced equation. This shows that two molecules of H_2O must be oxidized to energize and remove four electrons:

$$2\ H_2O \rightarrow 4\ H^+ + 4\ e^- + O_2$$

To move a single electron down the chain requires the absorption of two photons. Therefore, to get four electrons (and yield one O_2 molecule), the photosynthetic apparatus needs to absorb eight photons of light, four by each photosystem.

Cyclic Electron Transport

Photosystem I can function independently of photosystem II in what is called cyclic electron transport (**Figure 6**). In cyclic electron transport, the electron transport from photosystem I to ferredoxin is not followed by electron donation to the $NADP^+$ reductase complex. Instead, reduced ferredoxin donates electrons back to plastoquinone. In this way, plastoquinone gets continually reduced and oxidized, and keeps moving protons across the thylakoid membrane without the involvement of photosystem II. The net result of cyclic electron transport is that the energy absorbed from light is converted into the chemical energy of ATP without the oxidation of water or the reduction of $NADP^+$ to NADPH. Cyclic electron transport is sometimes referred to as cyclic photophosphorylation, because the light energy captured in this cycle is ultimately used to drive the phosphorylation of ADP to ATP.

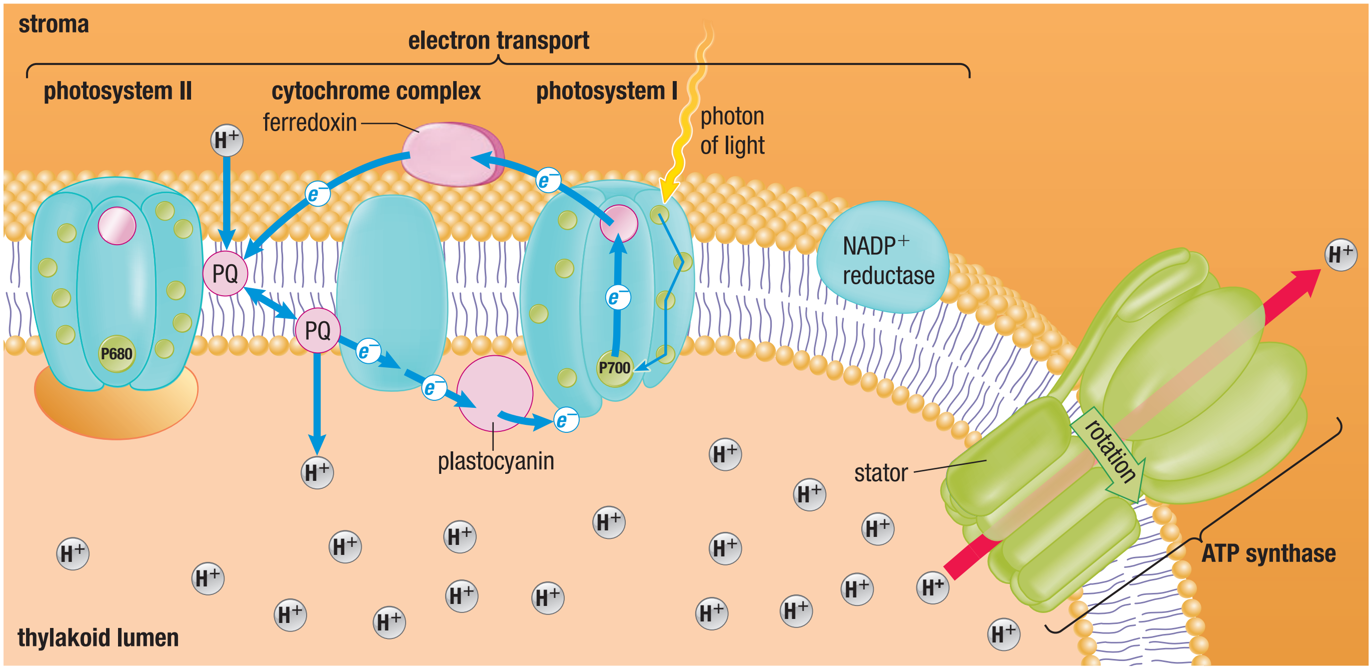

Figure 6 During cyclic electron transport, electrons move in a circular pathway from photosystem I, through ferredoxin to plastoquinone, through the cytochrome complex and plastocyanin, and then back to photosystem I. In cyclic electron transport, photosystem II does not operate. The pathway generates proton pumping, and thus leads to ATP production, but does not result in the synthesis of NADPH.

Cyclic electron transport plays an important role in overall photosynthesis. The reduction of carbon dioxide by the Calvin cycle requires more ATP than NADPH, and the additional ATP molecules are provided by cyclic electron transport. Other energy-requiring reactions in the chloroplast are also dependent on ATP produced by the cyclic pathway.

The Light-Independent Reactions

Recall, from the last chapter, that CO_2 is a fully oxidized carbon molecule and thus contains no usable energy. On the other hand, sugar molecules such as glucose and sucrose are highly reduced. They contain many C–H bonds and thus are an abundant source of energy. In the stroma of the chloroplast, a series of 11 reactions uses NADPH to reduce CO_2 into sugar. The overall process is endergonic, requiring energy supplied by the hydrolysis of ATP. These 11 enzyme-catalyzed (or light-independent) reactions are collectively known as the Calvin cycle. The Calvin cycle is by far the most dominant pathway on Earth by which CO_2 is fixed into carbohydrates. As you will learn in Section 5.4, many plants use a small number of additional light-independent steps immediately prior to the Calvin cycle.

How the Calvin Cycle Produces Carbohydrates

The Calvin cycle can be divided into three phases: fixation, reduction, and regeneration (**Figure 7**).

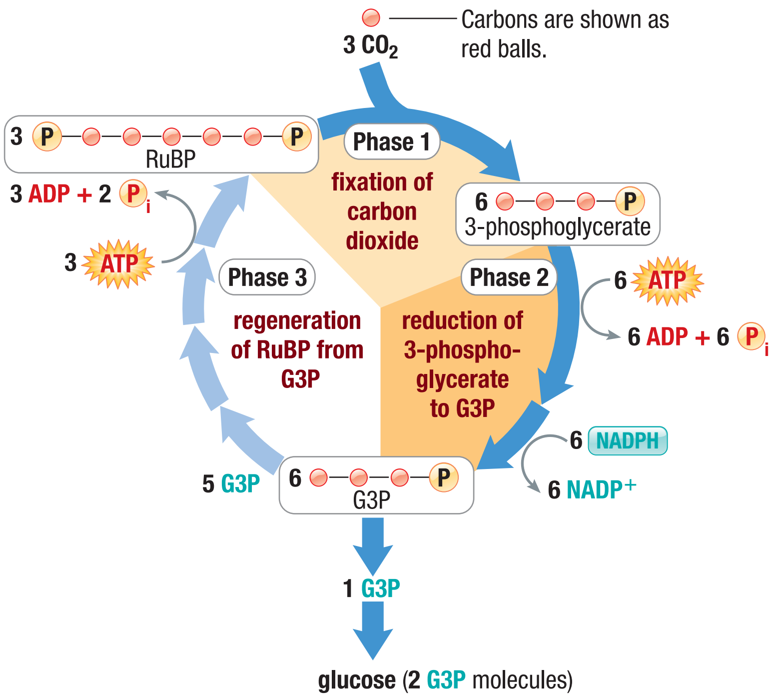

Figure 7 The Calvin cycle reactions consist of three phases: carbon fixation (each CO_2 is incorporated into a 6-C compound), reduction (G3P is produced), and regeneration (RuBP is re-formed). The sum of three turns of the cycle (three CO_2 molecules) ultimately produces one molecule of a 3-carbon sugar, G3P, which can be used to make other organic molecules. The cycle also produces $NADP^+$, ADP, and P_i, which can then return to participate in the light-dependent reactions.

Phase 1: Carbon fixation. Carbon fixation is the conversion of carbon from an inorganic to an organic form. At the beginning of the Calvin cycle, CO_2, an inorganic compound, reacts with a molecule of ribulose-1,5-bisphosphate (RuBP), a 5-carbon sugar, to produce two 3-carbon molecules of 3-phosphoglycerate. This single step has monumental significance for life on Earth. Every carbon atom in every cell of virtually all living things has taken part in this chemical reaction. This type of photosynthesis is called a C_3 metabolism from the two 3-carbon molecules formed. In Section 5.4 you will learn about alternative mechanisms of photosynthesis.

Phase 2: Reduction. In Phase 2, each molecule of 3-phosphoglycerate gets an additional phosphate added from the hydrolysis of ATP. This molecule is subsequently reduced by high-energy electrons from NADPH, producing glyceraldehyde-3-phosphate (G3P).

Phase 3: Regeneration. In a multi-step process, some of the G3P molecules are combined and rearranged to regenerate the RuBP that is required to start the cycle over again.

In each complete Calvin cycle, one molecule of CO_2 is converted into one reduced carbon—essentially one CH_2O unit of carbohydrate. However, it takes three cycles to produce something the cell can actually use—one extra molecule of the 3-carbon sugar, G3P. To account for this, Figure 7 is a summary of three cycles, tracking the fate of three carbon atoms.

Examine Figure 7 again, keeping track of the carbons in each phase. In 3 complete turns of the cycle, 3 CO_2 (3 carbons) are combined with 3 molecules of RuBP (15 carbons), to produce 6 molecules of 3-phosphoglycerate (18 carbons). These go on to yield 6 molecules of G3P (totalling 18 carbons). Five of the 6 molecules of G3P (totalling 15 carbons) are used to regenerate the 3 RuBP molecules (15 carbons). Thus, the cycle generates 1 molecule of G3P (3 carbons) after 3 turns.

The production of this one molecule of G3P is the ultimate goal of photosynthesis. G3P is a high-energy carbon that contains raw material from which all other organic plant compounds are synthesized. For the synthesis of this one G3P, the Calvin cycle requires a total of nine molecules of ATP and six molecules of NADPH. The $NADP^+$, ADP, and P_i that are formed during the reduction phase, as well as the ADP and P_i formed during the regeneration phase, of the Calvin cycle, are regenerated to NADPH and ATP by the light reactions. These numbers must be doubled to 18 ATP and 12 NADPH for a single glucose molecule to be produced from two G3P.

RUBISCO: THE MOST ABUNDANT PROTEIN ON EARTH

Before concluding this overview of the Calvin cycle, turn your attention to a special chemical—the chemical that is responsible for the very first step in Phase 1 and the fixing of carbon. Ribulose-1,5-bisphosphate carboxylase oxygenase, or RuBisCO (commonly spelled **rubisco**), is the enzyme that catalyzes the first reaction of the Calvin cycle. It is arguably the most important enzyme of the biosphere. By catalyzing CO_2 fixation in all photoautotrophs, it provides the source of organic carbon molecules for most of the world's organisms. It begins the conversion of about 100 billion tonnes of CO_2 into carbohydrates annually. There are so many rubisco molecules in chloroplasts that the enzyme makes up 50 % or more of the total protein of plant leaves. As such, rubisco is also the world's most abundant protein, estimated to total some 40 million tonnes worldwide—equivalent to about 6 kg per person on Earth.

rubisco ribulose-1,5-bisphosphate carboxylase oxygenase; a critical enzyme that acts as a catalyst for the reduction of carbon dioxide in the Calvin cycle of photosynthesis

A Diversity of Organic Products

The G3P molecule formed by three turns of the Calvin cycle is the starting point for the production of a wide variety of organic molecules. Complex carbohydrates, such as glucose and other simple sugars (monosaccharides), are made from G3P by reactions that, in effect, reverse the first half of glycolysis. Once produced, the monosaccharides may enter biochemical pathways that make disaccharides such as sucrose, polysaccharides such as starches and cellulose, and other complex carbohydrates. Other biochemical pathways manufacture amino acids, fatty acids and lipids, and nucleic acids. The reactions that form these products occur both within chloroplasts and in the surrounding cytosol and nucleus.

Sucrose, a disaccharide that consists of glucose bonded to fructose, is the main form in which the products of photosynthesis circulate from cell to cell in higher plants. In most higher plants, organic nutrients are stored as sucrose, starch, or a combination of the two in proportions that depend on the plant species. Sugar cane and sugar beets, which contain stored sucrose in high concentrations, are the main sources of the sucrose you use as table sugar.

5.2 Review

Summary

- In the light-dependent reactions, photosystem II absorbs light energy that oxidizes the reaction centre chlorophyll P680, producing the powerful oxidant $P680^+$. This molecule oxidizes water, removing electrons and releasing O_2.
- Electrons that are excited by the light energy in photosystem II pass through an electron transport system to photosystem I. There, the electrons become excited again by the absorption of light and are ultimately transferred to $NADP^+$ as the final electron acceptor forming NADPH.
- These processes establish a proton gradient across the thylakoid membrane, which is then used to generate ATP through chemiosmosis.
- ATP synthase complexes in the thylakoid membrane harness the energy of the moving protons to produce ATP reactions that are identical to those seen in cellular respiration.
- In cyclic electron transport, electrons can also flow cyclically through photosystem I, further building up the H^+ concentration inside the thylakoid membrane and allowing extra ATP to be produced, but no NADPH.
- In the Calvin cycle, CO_2 is fixed and converted into reduced high-energy organic substances by the addition of electrons and protons that are carried by the NADPH produced in the light reactions. ATP, also derived from the light reactions, provides additional energy. Rubisco catalyzes the reaction that first fixes CO_2 into organic compounds.
- For every 3 complete turns of the Calvin cycle, a single molecule of the 3-carbon molecule G3P is produced. G3P is the starting point for the synthesis of glucose, sucrose, starches, and many other organic molecules.

Questions

1. Describe how a low-energy electron in water becomes a high-energy electron on the primary acceptor of a reaction centre. K/U
2. Sketch the linear electron transport chain that ends with the passing of final electrons to $NADP^+$ to create NADPH. K/U C
3. Why is light energy essential for photosynthesis to occur? K/U
4. Describe three steps that contribute to the buildup of the proton gradient across the thylakoid membrane. K/U
5. Why and how do protons move through the thylakoid membrane from the lumen to the stroma? K/U
6. Use a t-chart to compare the electron transport chains found in mitochondria and chloroplasts. K/U C
7. Consider the analogy of the cyclist (page 224). T/I C
   (a) Illustrate the analogy. Include labels for photosystem I, photosystem II, and ATP production.
   (b) Is the analogy a good one? Why or why not?
8. Careful measurements indicate that the Calvin cycle in a plant leaf is occurring very slowly. In such a situation, there is a reduced demand for both NADPH and ATP. T/I A
   (a) What benefit (if any) would the light-dependent reactions still have for this plant?
   (b) Can you think of any environmental conditions under which such a situation might arise?
9. Analysis of a plant's leaf showed an unusually high amount of ATP, but relatively little NADPH. Which electron transport method is probably working much more predominantly in this leaf? Which photosystem is probably absorbing more light energy? Explain your answer. T/I
10. Rubisco is the world's most abundant protein, yet it is not found inside any animal cells. Suggest a reason why animals do not need or use this protein. T/I A