

Photosynthesis



THE HARNESSING OF LIGHT ENERGY BY PLANTS

DEVELOPING A GENERAL EQUATION FOR PHOTOSYNTHESIS

*Early Observations Showed the Roles of Raw Materials and Products
Comparative Studies Showed That Several Molecules May Reduce CO₂ in the Light*

LIGHT REACTIONS AND ENZYMATIC REACTIONS (PREVIOUSLY CALLED DARK REACTIONS) CHLOROPLASTS: SITES OF PHOTOSYNTHESIS

Chloroplast Structure Is Important in Trapping Light Energy

Experiments Reveal a Division of Labor in Chloroplasts

CONVERTING LIGHT ENERGY TO CHEMICAL ENERGY

*Light Has the Characteristics of Both Waves and Particles
Reactions in Thylakoid Membranes Transform Light Energy into Chemical Energy*

THE REDUCTION OF CO₂ TO SUGAR: THE CARBON CYCLE OF PHOTOSYNTHESIS

*Enzymes Catalyze Many Light-independent Reactions in Photosynthesis
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The C₃ Pathway Is the Major Path of Carbon Reduction and Assimilation in Plants*

PHOTORESPIRATION

ENVIRONMENTAL STRESS AND PHOTOSYNTHESIS

*Some Succulents Trap CO₂ at Night
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FACTORS AFFECTING PRODUCTIVITY

*Greater Productivity Can Be Bred into Plants
Fluctuations in the Environment Alter the Rate of Photosynthesis*

SUMMARY

IN DEPTH: Chemosynthesis

KEY CONCEPTS

1. Photosynthesis is the primary energy-storing process on which almost all life, both plant and animal, depends. The energy from sunlight is stored as chemical energy in organic compounds through a series of light- and temperature-sensitive reactions. Carbon dioxide and water are the raw materials, and the products are sugar and oxygen.
2. Chlorophyll in green plants absorbs light energy, which activates electrons in special chlorophyll *a* molecules. These electrons move along a chain of electron carriers, and some of their energy is stored in the production of adenosine triphosphate (ATP) or reduced nicotinamide adenine dinucleotide phosphate (NADPH). These reactions take place in association with thylakoid membranes.
3. Temperature-sensitive enzymatic reactions in the stroma use the ATP and NADPH to reduce CO₂ and to produce sugar.
4. The C₃ cycle is the major path of carbon assimilation in green plants. In this cycle, two three-carbon-atom molecules of phosphoglyceric acid are formed, hence the name C₃.
5. Some plants have adapted to extreme environmental conditions by evolving variations on the carbon assimilation pathway. Some plants use Crassulacean acid metabolism (CAM). They absorb CO₂ through open stomata at night and form organic acids. During the day, when their stomata are closed, CO₂ is released from the organic acids and converted into carbohydrate by the C₃ cycle.
6. Other plants have evolved another variation of carbon assimilation, the C₄ pathway. These plants effectively concentrate CO₂ in their leaf mesophyll chloroplasts by forming four-carbon organic acids. The organic acids are transported into the bundle sheath chloroplasts and lose the CO₂, which is used in the C₃ cycle there.
7. In the light and at high temperatures, chloroplasts take up O₂ and release CO₂ (photorespiration). Photorespiration is reduced in C₄ plants.
8. Usually only about 0.3% to 0.5% of light energy that strikes a leaf is stored during photosynthesis, but under ideal conditions this may increase several-fold. Photosynthesis may be limited by CO₂ concentration, light, temperature, minerals, and other environmental and hereditary factors.

10.1 THE HARNESSING OF LIGHT ENERGY BY PLANTS

With few exceptions, all living cells require a continuous supply of energy, which comes directly or indirectly from the sun. Although terrestrial green plants use large amounts of energy directly from the sun in both transpiration and photosynthesis, only in photosynthesis is light energy stored as chemical energy for future use. Billions of years ago, Cyanobacteria (also known as blue-green algae) created an oxygen-rich atmosphere through their photosynthetic activity. Since that time, photosynthetic organisms have continued to support life by being the original source of energy for other organisms.

The great importance of photosynthesis is twofold: the liberation of oxygen as an end product and the transformation of low energy compounds (carbon dioxide and water into high-energy compounds (sugars). Perhaps someday humans will use other sources of energy to drive the energy-requiring steps in the production of food and fiber. Currently, however, except for a few species of bacteria (see IN DEPTH: Chemosynthesis" sidebar), all life is dependent of the energy-storing reaction of photosynthesis.

Although the subject of this chapter is primarily "photosynthesis in green plants" you should keep in mind that the photosynthesis carried out by aquatic life--including photosynthetic bacteria and red, green, yellow, golden, and brown algae--liberates at least as much oxygen per day as that produced by terrestrial green plants.

10.2 DEVELOPING A GENERAL EQUATION FOR PHOTOSYNTHESIS

Like respiration and other complex processes occurring in living cells, photosynthesis consists of many reaction steps. It is easier to approach photosynthesis, or any biochemical process, by looking at an overview. Indeed, this is how scientific knowledge about photosynthesis evolved; therefore, this section traces the discoveries that led to a general understanding of photosynthesis. Later sections detail the steps of specific reactions.

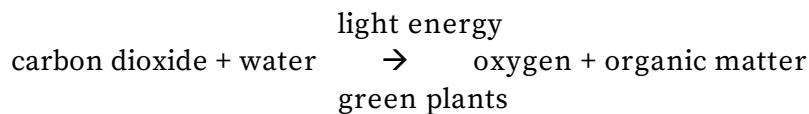
Early Observations Showed the Roles of Raw Materials and Products

Until the early 17th century, scholars believed that plants derived the bulk of their substance from soil humus. A simple experiment performed by Flemish physician and chemist Joannes van Helmont disproved this idea. He planted a 2.27-kg (5-lb) willow (*Salix*) branch in 90.7 kg (200 lb) of carefully dried soil and supplied rainwater to the plant as needed. In 5 years, it grew to a weight of 67.7 kg (169 lb), but according to van Helmont's measurements, the soil had lost only 57 g (2 oz). Consequently, he reasoned that the plant substance must have come from water. This was a logical deduction, though not entirely correct. Almost two centuries elapsed before van Helmont's findings were correctly explained.

Our knowledge of photosynthesis begins with the observations of a religious reformer, philosopher, and spare-time naturalist, Joseph Priestley. In 1772, Priestley reported that a sprig of mint could restore confined air that had been made impure by a burning candle. The plant changed the air so that a mouse was able to live in it.

The experiment was not always successful, probably because Priestley, who did not know about the role of light in photosynthesis, did not always provide adequate illumination for his plants. In 1780, a Geneva pastor, Jean Senebier, published his own research and pointed out another important part of the process: that "fixed air," carbon dioxide, was required. Thus in the new terminology of French chemist Antoine Lavoisier, it could be said that green plants in the light use carbon dioxide and produce oxygen.

But what was the fate of the carbon dioxide? A Dutch physician, Jan Ingen-Housz, answered this question in 1796, when he found that carbon went into the nutrition of the plant. In 1804, 32 years after Priestley's early observations, the final part of the overall reaction of photosynthesis was explained by the Swiss botanist and physicist Nicolas de Saussure, who observed that water was involved in the process. Now the experiment performed by van Helmont almost 200 years earlier could be explained:

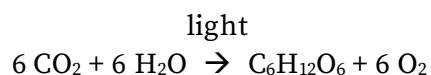


Almost 50 years elapsed before scientists identified carbohydrates as the organic matter formed during photosynthesis in most plants. Between 1862 and 1864 a German plant physiologist, Julius von Sachs, observed that starch grains occur in the chloroplasts of higher plants, and that if leaves containing starch are kept in darkness for some time, the starch disappears. If those leaves are exposed to light, starch reappears in the chloroplasts. van Sachs was the first to connect the appearance of starch, a carbohydrate, with both the fixation of carbon in chloroplasts and the presence of light.

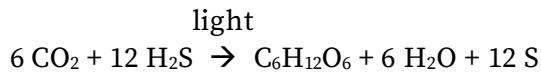
It is easy to demonstrate in the laboratory that starch forms during photosynthesis; however, it is much more difficult to show that sugar forms *before* starch does. Proof that sugar is the first carbohydrate produced by photosynthesis had to await the availability of radioactive carbon (^{14}C). This development is discussed in a later section of this chapter.

Comparative Studies Showed That Several Molecules May Reduce CO_2 in the Light

By glancing at the overall equation for photosynthesis,

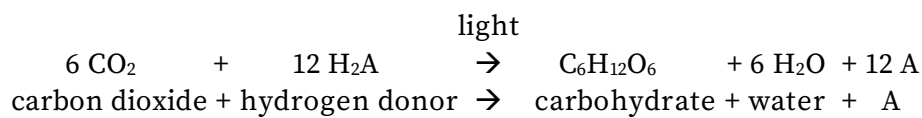


you might conclude that the carbon dioxide molecule splits, liberating the oxygen molecule. Indeed, most scientists believed that the reaction proceeded in this manner until the early 1930s, when Cornelis van Niel, working at Stanford University, compared photosynthesis in a number of different groups of photosynthetic bacteria. The green and purple sulfur bacteria use hydrogen sulfide instead of water to reduce carbon dioxide, for example, and van Niel found that sulfur instead of oxygen is liberated, as follows:



The sulfur can come only from the hydrogen sulfide. Because the hydrogen sulfide serves the same role in these bacteria as water does in higher plants, van Niel reasoned that the oxygen evolved by higher plants comes from water, not from carbon dioxide. Experiments using radioactive tracers have since verified van Niel's insight.

After comparing similar reactions in other organisms, van Niel concluded that a general equation for photosynthesis should be written as:



H₂A can be H₂O, H₂S, H₂, or any other molecule capable of donating an electron, and the reaction requires an input of energy. When H₂A give up its electron, it is oxidized to A.

10.3 LIGHT REACTIONS AND ENZYMATIC REACTIONS (PREVIOUSLY CALLED DARK REACTIONS)

Although the general equation for photosynthesis identifies the reactants and products, it tells us nothing about the individual reactions that, taken together, make up this complex process. To supply food and fiber to the increasing world population, we need to be able to increase crop yields. We need to know, among other things, the specific reactions of photosynthesis. Research spanning 100 years has shown that photosynthesis involves both light absorption and enzymatic reactions.

Between 1883 and 1885, a German physiologist, T.W. Engelmann, in a remarkably simple experiment, demonstrated which colors of light are used in photosynthesis. The spectrum of visible light varies from violet to red, as can be observed when white light is broken into its components by passing it through a prism. Engelmann placed together on a microscope slide a living filament of a green alga and some bacteria that would migrate toward high concentrations of dissolved oxygen. He reasoned that the bacteria would cluster near regions of the alga generating the most oxygen from photosynthesis. When he placed a filament of the green alga, *Spirogyra*, in a spectrum produced by passing light through a prism, he found that the bacteria migrated to the sections of the alga exposed to the red and blue light. This demonstrated that red and blue light supplied energy to drive photosynthesis and liberate oxygen (Fig. 10.1).

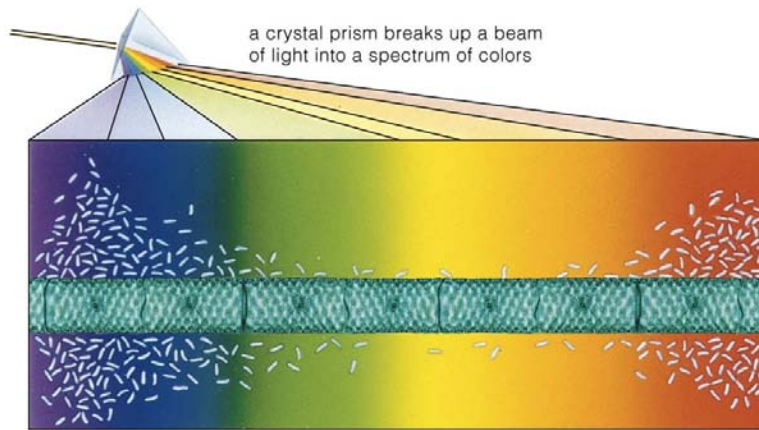


Figure 10.1. Engelmann's experiment, demonstrating that red and blue light are effective in photosynthesis. The bacteria migrate to the portion of the algal filament where oxygen is produced.

At approximately the same time, J. Reinke, another German scientist, was studying the effect of changing the intensity of light on photosynthesis. Reinke observed that the rate of photosynthesis increased proportionally to an increase in the intensity of light, but only at low-to-moderate light intensities. At greater intensities, the rate of photosynthesis was unaffected by increases in light. This indicated that the reaction was then proceeding at its maximum rate; it had become light-saturated.

A further study and a more comprehensive interpretation of this phenomenon were conducted in 1905 by F.F. Blackman, a British plant physiologist. Blackman realized that the rate of many light-stimulated reactions is relative unaffected by a change in temperature, but that the rate of many light-insensitive reactions approximately doubles with every 10°C increase in temperature, over a temperature range of about 10 to 25°C.

He found that when photosynthesis was proceeding rapidly under adequate levels of CO₂ and high light intensity, the rate of photosynthesis more than doubled with a temperature increase of 10°C. However, if the light intensity was low, an increase in temperature had little effect on the rate of photosynthesis. Blackman reasoned that photosynthesis is divided into two general parts: (1) photochemical reactions, *light reactions*, which are insensitive to temperature changes, and (2) temperature-sensitive reactions, previously called *dark reactions*.

The temperature-sensitive reactions do not depend directly on light. They can occur either in the light or in the dark. However, in the chloroplast, the activities of several of the enzymes that catalyze temperature-sensitive reactions are affected by products of light reactions, such as changes in pH and NADPH concentrations. In addition, there are special chloroplast proteins known as thioredoxins that regulate the activities of some dark reaction enzymes. Thus the rate of the dark reactions depends indirectly on the presence of light.

10.4 CHLOROPLASTS: SITES OF PHOTOSYNTHESIS

Because many of the individual reaction steps of photosynthesis are dependent on the specific cellular structure in which they occur, we need to describe this structure. Early studies with intact plants showed that oxygen is liberated and starch is formed in chloroplasts. It does not necessarily follow, however, that all of the reaction steps take place in chloroplasts. To find out, scientists isolated intact chloroplasts and

parts of chloroplasts. One of the earliest successful attempts to do this was by Robin Hill in Cambridge, England. In 1932, he demonstrated that isolated chloroplasts could trap light energy and liberate oxygen. Then in 1954, Daniel Arnon, at the University of California, and others proved that isolated chloroplasts could convert light energy to chemical energy and use this energy to reduce CO_2 .

Chloroplast Structure Is Important in Trapping Light Energy

Not all chloroplasts have the same shape, but they do have a universally similar structural organization that is critically important for photosynthesis. Chloroplasts, seen with the light microscope in living cells, appear to be homogeneously green, but in an electron micrograph, the double-membrane envelope and the complex internal membranes are apparent (Fig. 10.2). There are two types of internal membranes: those forming the **grana** (singular, *granum*) and those interconnecting the grana, the **stroma lamellae**. Together, these two membrane types constitute the **thylakoids** in the chloroplasts.

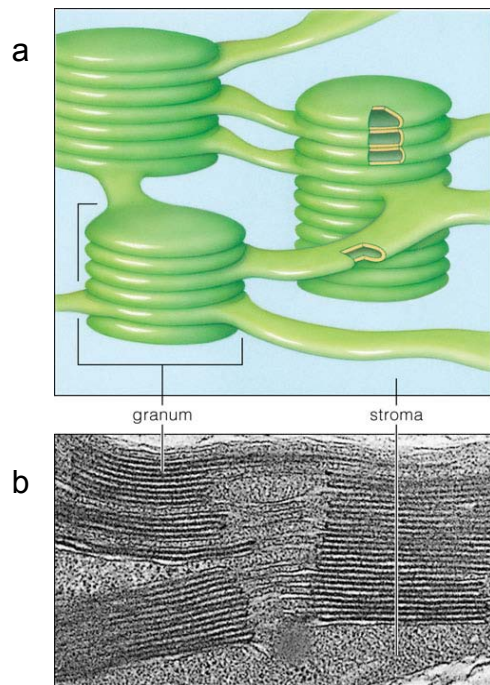


Figure 10.2. Chloroplast membrane systems. (a) Diagram, (b) electron micrograph, showing grana and stroma lamellae.

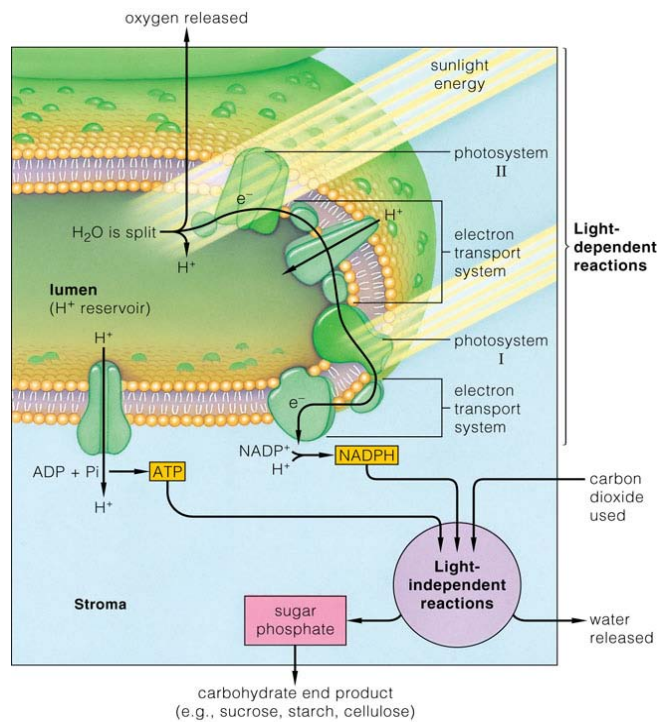


Figure 10.3. Diagram of a section of chloroplast granum showing where reactions take place.

All biological membranes have high concentrations of both lipids and proteins. Biochemists believe that the chlorophylls and other pigments that are located in the grana and stroma lamellae are in close contact with the photosynthetic enzyme systems, forming definite lipid, pigment, protein patterns (Fig. 10.3). Such a structural arrangement improves the cell's ability to trap light

energy and transform it into chemical energy during photosynthesis. Light energy initiates a series of reactions in which electrons from chlorophyll flow to other compounds--electron acceptors--in the internal membranes.

Experiments Reveal a Division of Labor in Chloroplasts

To determine what role chloroplasts play in photosynthesis, researchers need to study them independently of the rest of the plant cell. To isolate intact, functional chloroplasts, cells are carefully cut or ground in a solution containing a pH buffer and an osmotic stabilizer. Chloroplasts are separated from the rest of the cell contents by placing the mixture in a centrifuge tube and spinning it at moderate speeds. Large and dense particles--nuclei, starch grains, cell wall fragments--collect at the bottom of the tube. The liquid containing the chloroplasts and other, smaller cellular components is then poured into another test tube and centrifuged at a slightly greater speed. The chloroplasts, free of most of the cellular debris, collect at the bottom of the tube. Electron micrographs of these plastids show some to be complete, with internal membranes embedded in stroma and surrounded by the outer envelope. The others have lost their envelope and stroma and consist of just the thylakoids, which still contain all the light-absorbing pigments (Fig. 10.4).

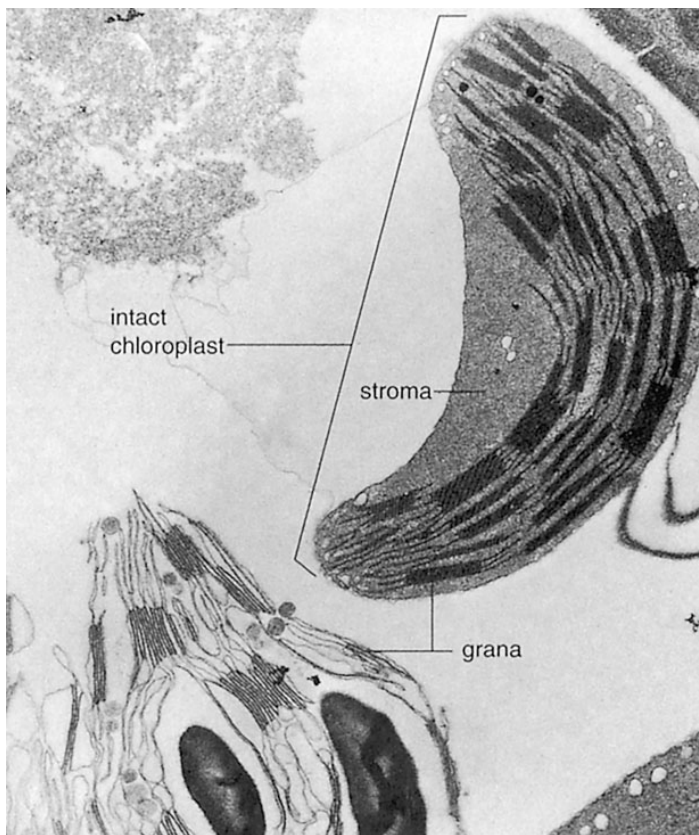
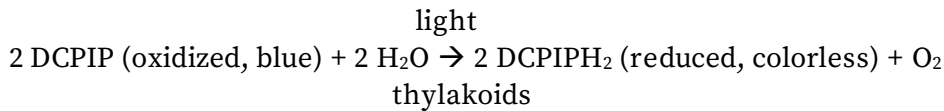


Figure 10.4. Electron micrograph of isolated chloroplast of broad bean (*Vicia faba*), showing one intact chloroplast and chloroplasts that have lost their outer envelopes and stroma.

The thylakoids of the broken plastids will carry out part of the reactions of photosynthesis. They will liberate oxygen, as Hill demonstrated. A simple

experiment analogous to Hill's uses dichlorophenol indophenol, which is blue in its oxidized form (DCPIP) and colorless in its reduced form (DCPIPH₂). If DCPIP is mixed with broken chloroplasts and the mixture is illuminated, the blue color fades. The reaction depends on the thylakoids and light and releases O₂.



Experiments with the isolated chloroplasts and isolated thylakoids reveal a division of labor within the chloroplasts. The green thylakoids capture light, liberate oxygen from water, form ATP from ADP and phosphate, and reduce NADP⁺ to NADPH. The colorless stroma contains water-soluble enzymes, captures carbon dioxide, and uses the energy from ATP and NADPH in sugar synthesis.

10.5 CONVERTING LIGHT ENERGY TO CHEMICAL ENERGY

It is not necessary to know the details of photosynthesis to appreciate the significance of the process. However, the more we know about the process, the closer we can come to optimizing its rate and quality. This section focuses on the light reactions of the thylakoids.

Light Has the Characteristics of Both Waves and Particles

Physicists have two models describing light, and both are needed to understand the role of light in photosynthesis. The first model interprets light as electromagnetic waves. Light is only a small part of the **electromagnetic energy spectrum** that comes to us from the sun or outer space. The longest waves, radio waves, may be hundreds or thousands of meters in length, whereas gamma rays have wavelengths a fraction of a nanometer in length (1 nanometer [nm] = 10⁻⁹ meters). When white light passes through a prism or through water droplets, it is resolved into its separate component colors, forming the visible spectrum (Fig. 10.5).

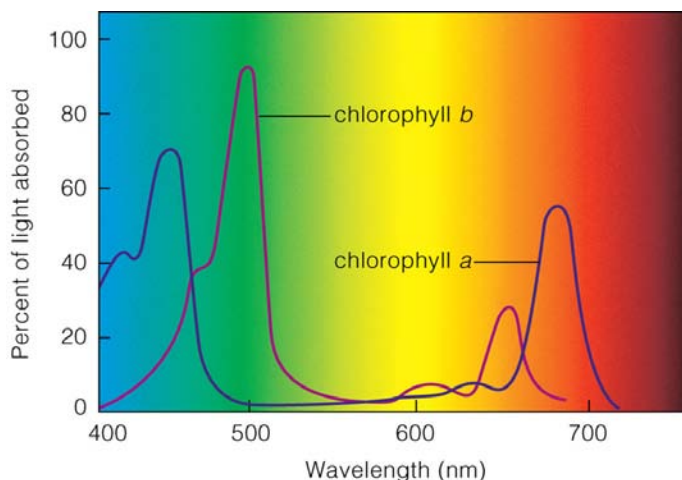


Figure 10.5. Absorption spectra of chlorophylls *a* and *b*. Curves show the fraction of received light that is absorbed when chlorophyll *a* and chlorophyll *b* are exposed to various wavelengths. The colors associated with different wavelengths are also shown.

White light is composed of wavelengths ranging from red (wavelengths of 640 - 740 nm) to violet (400 - 425 nm). We cannot see wavelengths longer than red--that is, the infrared and radio waves--or shorter than violet--that is, the ultraviolet, X-rays, and gamma rays. The visible spectrum is only a small part of the complete electromagnetic spectrum, and only a part of the visible spectrum provides the energy for photosynthesis.

The second model of light states that light acts as if it were composed of discrete particles or packets of energy called **photons**. Each photon contains an amount of energy that is inversely proportional to the wavelength of light characteristic for that photon. Therefore, the short wavelength of blue light has more energy per photon than does the longer wavelength of red light. When light is absorbed by a pigment--as in photosynthesis, light is absorbed by chloroplast pigments--only one photon is absorbed by one pigment molecule at a time. The energy of the photon is absorbed by an electron of the pigment, giving this electron more energy. The additional energy moves the electron away from its nucleus into a different and higher "orbital."

Reactions in Thylakoid Membranes Transform Light Energy into Chemical Energy

ABSORPTION OF LIGHT ENERGY BY PLANT PIGMENTS Chlorophyll appears green because it absorbs some of the blue and red wavelengths of white light, leaving proportionally more green light to be transmitted or reflected. *It is the absorbed light that is used in photosynthesis.* Scientists can measure the amount of any specific wavelength of light that is absorbed by a pigment by using a **spectrophotometer**, an instrument that measures and then plots the percentage of light absorbed for each wavelength of the visible spectrum. The resulting graph is an **absorption spectrum**.

Vascular plants contain two major types of chlorophyll: *a* and *b*. The absorption spectra of chlorophylls *a* and *b* in solution (Fig. 10.5) show that they absorb much of the red, blue, indigo, and violet light; thus these are the wavelengths that are used most in photosynthesis. Part of the red and most of the yellow, orange, and green light are scarcely absorbed at all, unless the chlorophyll solution is very concentrated. If, instead of a chlorophyll solution, a very thin green leaf is placed in the spectrophotometer, the absorption spectrum is quite similar, although not identical, to that of the chlorophyll solution. The difference can be attributed in part to other pigments in the leaf, such as **carotenoids**, which absorb other wavelengths of light. Also, chlorophyll in the leaf is in close association with proteins in the chloroplasts; chlorophyll in solution is not.

What happens when the chlorophyll molecules absorb light energy? How is light energy converted into chemical bond energy during photosynthesis? Recall from Chapter 2 that molecules are composed of atoms having positively charged nuclei and one or more electrons spinning around each nucleus. When a photon is absorbed by a chlorophyll molecule, the energy of the photon causes an electron from one of its atoms to be moved into a higher energy state. The chlorophyll molecule now is in an excited state. This condition is unstable, and the electron tends to move rapidly, usually within a billionth of a second, back to its original energy level. As it does this, the absorbed energy can be transferred to an adjacent

pigment molecule by a process called resonance. Eventually the energy can be transferred to a special chlorophyll *a* molecule (reaction center, Fig. 10.6). From the reaction center an excited electron can move to another molecule, which can in turn pass it on. This represents the first step in the conversion of light energy to chemical energy.

THE TWO PHOTOSYSTEMS Two photosystems are involved in the trapping of light energy during photosynthesis. In the 1950s, Robert Emerson at the University of Illinois found that red light of about 680 nm or longer wavelengths was inefficient in photosynthesis, but that adding light of shorter wavelength increased the efficiency of the longer wavelengths. This observation was explained by the hypothesis that there are two light reactions and two pigment systems, designated photosystems I and II, in thylakoids of plants that produce oxygen. Photosystem I absorbs light wavelengths longer than 680 nm; but photosystem II requires light of shorter wavelengths. The fact that the longer wavelengths were inefficient in photosynthesis, but could be used in conjunction with shorter wavelengths, suggested that both photosystems had to work together, in series, to accomplish photosynthesis.

Each photosystem has its own reaction center, together with chlorophyll *a* and *b* and accessory pigments, grouped together in **light-harvesting complexes**. The energy absorbed by these pigments is transferred to a specific chlorophyll molecule in the reaction center. The reaction centers of photosystems I and II are called P_{700} and P_{680} , respectively, because they absorb light maximally at those wavelengths. In addition to being distinguished by their reaction centers, photosystems I and II differ in their light-harvesting complexes. Photosystem I has a greater proportion of chlorophyll *a* than chlorophyll *b*; photosystem II has almost equal amounts of the two chlorophyll types.

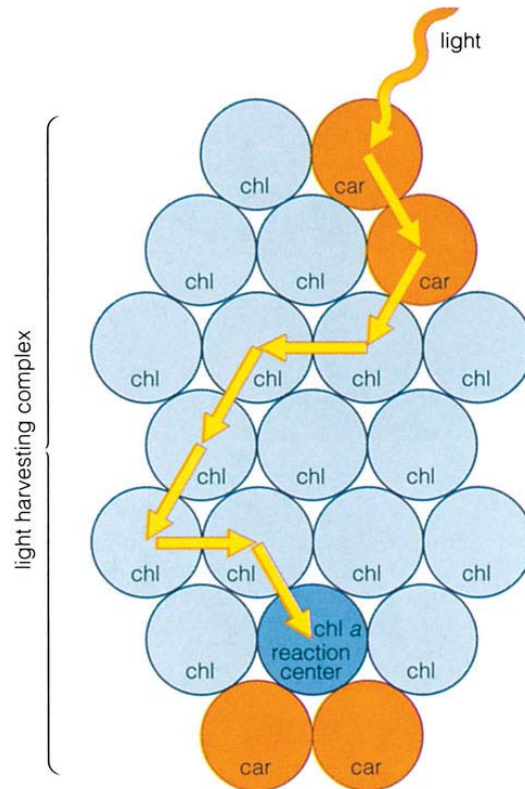


Figure 10.6. Accessory pigments in the light-harvesting complex absorb light and transfer it to the reaction center. Car, carotene; chl, chlorophyll.

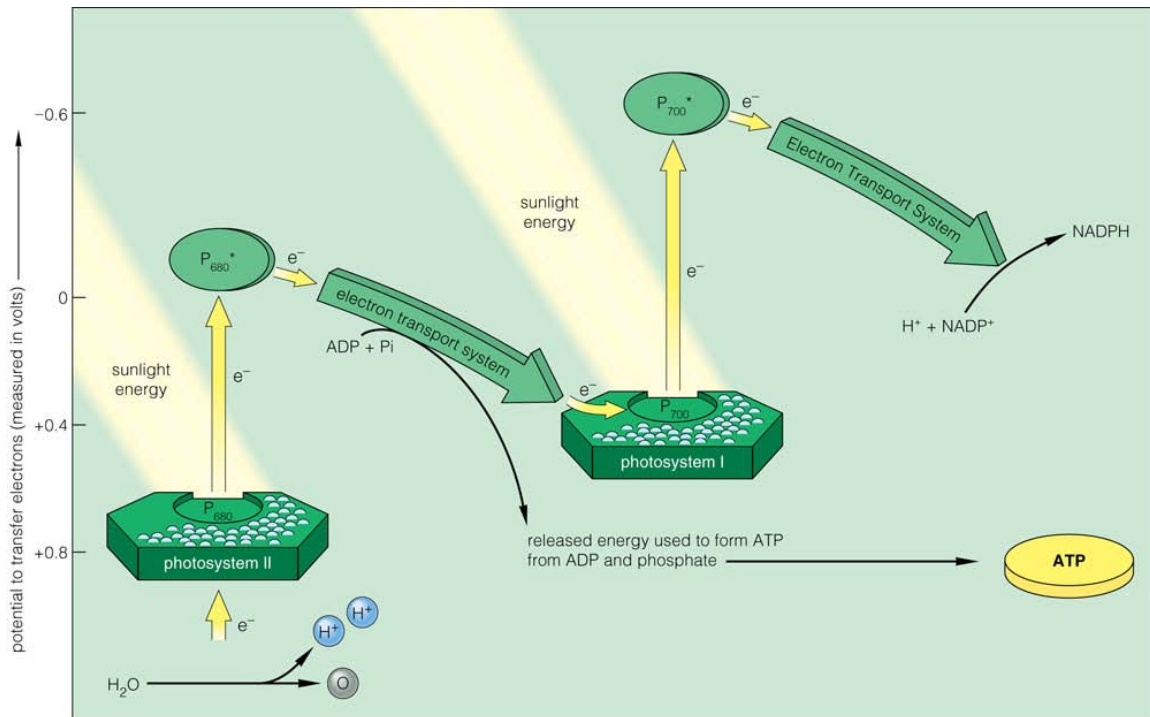


Figure 10.7. The pathway of noncyclic electron transport from water to NADPH, with associated ATP synthesis.

FORMATION OF REDUCED NICOTINAMIDE ADENINE DINUCLEOTIDE PHOTPHATE

The light reactions of photosynthesis are oxidation-reduction processes involving the transfer of electrons in a series of steps. These transfers are initiated by the absorption of light energy by one of the photosystems, exciting an electron in the reaction center. The electron moves along a series of electron carriers, distinguished by their increasing ability to accept (and decreasing ability to donate) electrons. The relative tendency of electron carriers to donate and accept electrons is expressed in volts on the vertical scale of Fig. 10.7. A carrier higher on the scale will spontaneously transfer an electron to a carrier lower on the scale. Each time an electron is transferred from one molecule to the next, some energy is lost as heat, but the rest of the energy is then in the electron acceptor.

When a photon is absorbed by the light-harvesting complex of photosystem I, the energy is transferred to the reaction center P_{700} . An electron in P_{700} moves to a higher energy level, symbolized by P_{700}^* . In this excited state, the P_{700}^* will lose the electron to an adjacent carrier. This electron is transferred quickly from one carrier molecule to another, illustrated by moving down the vertical axis of Fig. 10.7, and finally arriving at an $NADP^+$ molecule. The donation of two electrons plus one H^+ to $NADP^+$ forms NADPH, an energy-rich molecule now available for enzymatic reactions of photosynthesis.

Meanwhile, the reaction center of photosystem I is left as a positive ion. It holds its remaining electrons very strongly in this low-energy state. It cannot accept another photon until the lost electron is replaced. Photosystem I must await events in photosystem II.

When a photon is absorbed by the light-harvesting complex of photosystem II, the energy is transferred to the reaction center P_{680} , producing the excited P_{680}^* . A electron is lost from P_{680}^* and moves down an electron transport chain, finally joining P_{700}^+ and regenerating P_{700} . But now P_{680} has a positive charge and must recover an electron. In its oxidized state, P_{680}^+ , has a very strong affinity for electrons, so strong that it can gain electrons from water. This reaction, known as photolysis, forms O_2 and releases H^+ .

The photosystem II reaction probably takes place near the inner surface of the thylakoid membrane, and the hydrogen ions produced contribute to the establishment of a proton gradient across the thylakoid membrane. In addition, hydrogen ions are absorbed on the stromal side of the thylakoid membrane in the formation of NADPH. An additional movement of H^+ from stroma to the thylakoid lumen occurs during the photosystem II electron flow.

In summary, the absorption of two photons pushes an electron from water to $NADP^+$. Four photons are needed to remove two electrons from water and form NADPH. Eight photons will be needed to remove four electrons from two water molecules and form O_2 . Light energy has been used to synthesize NADPH and O_2 . The light energy has also been used to form an H^+ gradient across the thylakoid membrane.

SYNTHESIS OF ADENOSINE TRIPHOSPHATE During electron flow, hydrogen ions are moved from the stroma to the fluid inside the thylakoids. The result is a proton gradient—a difference in H^+ concentration across the thylakoid membrane. Driven by this concentration gradient, H^+ ions flow back to the stroma through an ATP synthetase, forming ATP from ADP plus P_i , a process exactly analogous to the formation of ATP in mitochondria (Chapter 9, Fig. 9.8).

The light-driven production of ATP in chloroplasts is called photophosphorylation. Physiologists refer to two processes: non-cyclic photophosphorylation and cyclic photophosphorylation. Non-cyclic photophosphorylation is associated with the movement of electrons from water to $NADP^+$ and thus the formation of O_2 and NADPH (Fig. 10.7). Cyclic photophosphorylation is associated with "cyclic" electron flow from excited photosystem I's P_{700}^* through electron carriers and back to reform P_{700} (Fig. 10.8). No NADPH is formed in cyclic electron flow, but this electron flow promotes movement of H^+ into the thylakoid lumen, contributing to the H^+ gradient and subsequent formation of ATP.

10.6 THE REDUCTION OF CO_2 TO SUGAR: THE CARBON CYCLE OF PHOTOSYNTHESIS

The preceding section dealt with the transformation of electromagnetic energy (light) into the chemical energy of NADPH and ATP. This section considers how energy stored in NADPH and ATP is used to reduce CO_2 to sugar.

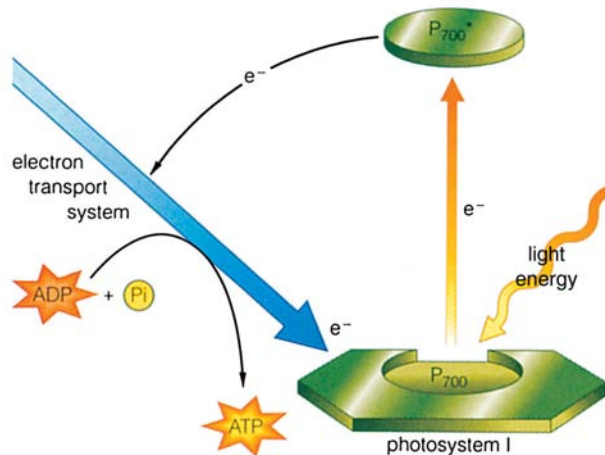
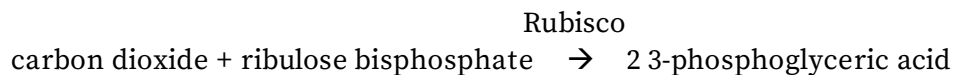


Figure 10.8. The pathway of cyclic electron transport, which contributes to the H⁺ gradient and the formation of ATP from ADP and inorganic phosphate (Pi).

Enzymes Catalyze Many Light-independent Reactions in Photosynthesis

The process of reducing carbon dioxide to carbohydrate requires many enzymatic reactions. All the enzymes that directly participate function in the chloroplasts. Most of them are water-soluble and are found in the stroma.

One of the most extensively studied enzymes of photosynthesis, and the enzyme that probably occurs in greatest concentration in many leaf cells, is **ribulose biphosphate carboxylase/oxygenase (Rubisco)**. This enzyme catalyzes the first step in the incorporation of carbon into organic compounds:



In this reaction, CO₂ combines with the five-carbon sugar, ribulose biphosphate (RuBP), in the plastid stroma to produce two molecules of the three-carbon acid, phosphoglyceric acid (PGA). This is a spontaneous reaction involving little change in free energy.

The C₃ Pathway Is the Major Path of Carbon Reduction and Assimilation in Plants

Photosynthetic carbon reduction involves a series of reactions that can be described as a cycle, because each intermediate compound in the pathway can be converted into another compound of the pathway (Fig. 10.9). The pathway is often called the Calvin cycle (after its discoverer) or the C₃ pathway (for the number of carbons in the first product, PGA). This pathway is used by almost all plants, although it is modified in some plants as an adaptation to stressful environments. The key points of the Calvin cycle are the following:

1. CO₂ enters the cycle when it combines with RuBP in the stroma. Two molecules of PGA are produced.

2. NADPH is used to reduce the PGA to phosphoglyceraldehyde (PGAL). Hydrolysis of ATP provides energy to move this reaction forward. The NADPH and ATP represent the contributions of the light reactions to the carbon fixation process.

3. A proportion of the PGAL and some additional ATP are used to reform RuBP. This completes the cycle.

3. Other molecules of PGAL can be converted to another three-carbon sugar phosphate, dihydroxyacetone phosphate (DHAP). PGAL and DHAP can combine to form a sugar phosphate, fructose-1,6-bisphosphate.

4. Fructose-1,6-bisphosphate can be converted into other carbohydrates, including glucose, sucrose, and starch. These reactions are not part of the Calvin cycle.

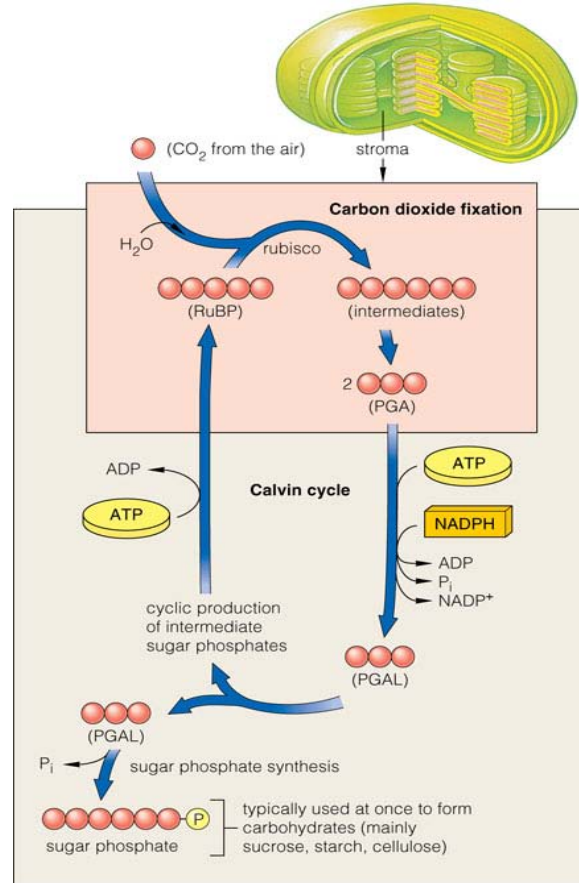
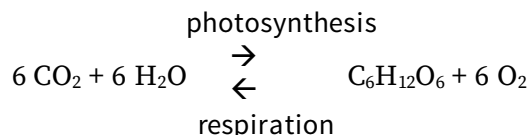


Figure 10.9. Some major steps in the C₃ pathway of photosynthesis. Carbon atoms are shown in red. All the intermediates have one or two phosphates; for simplicity, only the phosphate on the last product is shown.

For every one CO₂ that enters the Calvin cycle, 2 NADPH and 2 ATP molecules are required to reduce PGA to PGAL and a third ATP is needed to reform RuBP. Thus the formation of a six-carbon glucose molecule requires 12 NADPH and 18 ATP. This represents ideal conditions. Under stressful conditions, extra energy may be needed, as described in the next sections.

10.7 PHOTORESPIRATION

Formally, and considering just the reduction and oxidation of carbon, photosynthesis and respiration are opposite processes:



If respiration occurred during photosynthesis, it would release the CO₂ just fixed by photosynthesis. Light actually does stimulate the loss of CO₂ from carbohydrate, but the process, called photorespiration, yields no NADH or ATP and does not occur in the dark.

Photorespiration starts when Rubisco acts on RuBP. The activity of Rubisco depends on the ratio of O₂ to CO₂ in the cell. When CO₂ concentration in the leaf is high, Rubisco catalyzes the addition of CO₂ to RuBP, as described for the Calvin cycle. When the concentration of O₂ is high and/or the concentration of CO₂ is low, or when the temperature is high, Rubisco instead catalyzes the addition of O₂ to RuBP. One of the products of this reaction, a two-carbon molecule, is oxidized to CO₂ without any formation of NADH or ATP. Under some conditions, such as hot, sunny days, 50% of the carbon reduced during photosynthesis may be reoxidized to CO₂ during photorespiration in C₃ plants.

C₃ plants may have very high rates of photorespiration, particularly on hot, bright days, but some other plants with special adaptations show little or no photorespiration. The adaptations in C₄ plants raise the effective concentration of CO₂ and reduce the concentration of O₂ in Rubisco-containing cells (see the next section). Consequently, C₄ plants may produce two or three times as much photosynthate as C₃ cells during hot days. This is part of the reason that the C₄ plants sugar cane (*Saccharum officinarum*) and corn (*Zea mays*) are so productive. Under milder conditions with less photorespiration, C₃ plants are more efficient than C₄ plants, in part because they expend less energy to concentrate CO₂.

10.8 ENVIRONMENTAL STRESS AND PHOTOSYNTHESIS

Plants have adapted in various ways to environments that are not ideal--too cold, too hot, too dry. Some adaptations include variations in the process of photosynthesis. The C₄ adaptation, described briefly above and in more detail in this section, is a trait useful in both hot and dry conditions. Crassulacean acid metabolism is found in plants in extremely dry environments.

The C₄ Pathway Concentrates CO₂

We can determine how well a plant absorbs CO₂ if we place it in a closed container in the light and measure the CO₂ content of the air in the chamber. At some point, the CO₂ produced by respiration will just balance or compensate for the CO₂ absorbed during photosynthesis. The concentration of CO₂ remaining in the chamber under these conditions is known as the CO₂ **compensation point**, and it varies among different plants. If a bean plant and a corn plant are placed together in a chamber in the light, the corn plant will successfully compete with the bean for the limited CO₂. Both will eventually die, but the bean will die before the corn plant does (Fig. 10.10). This is because plants such as corn have very low CO₂ compensation points.

In general, such plants seem to be adapted to grow in habitats with high light intensities and high temperatures, and they use water more efficiently. They also have certain structural features, such as the arrangement of large parenchyma cells in a "bundle sheath" around veins (Fig. 10.11). Often, though not always, the chloroplasts of the bundle sheath parenchymal cells have greatly reduced grana and

frequently store starch. In contrast, the mesophyll cells between veins contain chloroplasts that have typical grana but little or no starch.



Figure 10.10. Corn (*Zea mays*, right), a C₄ plant with a low CO₂ compensation point is able to survive at a lower CO₂ concentration than bean (*Phaseolus vulgaris*, left), a C₃ plant, when they are grown in a lighted closed chamber for 10 days.

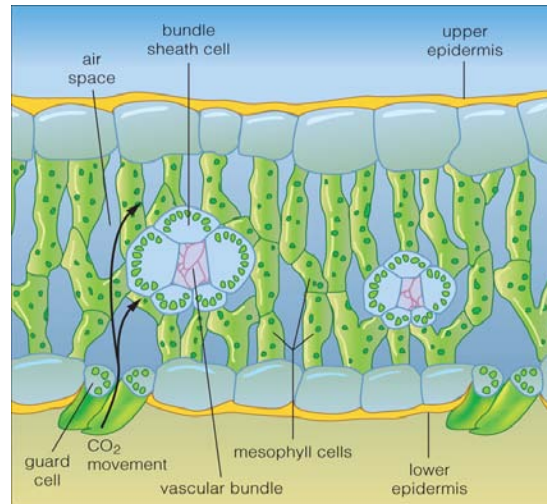


Figure 10.11. A section through a corn (*Zea mays*) leaf shows the concentric arrangement of bundle sheath around vascular bundles. See also the micrograph in Figure 6.10.

Plants that have these specialized bundle sheath and mesophyll cells rely on a special variation of photosynthesis. Instead of combining CO₂ directly with RuBP, these plants initially catalyze the addition of CO₂ to a three-carbon compound, phosphoenolpyruvate (PEP), to form the four-carbon compound, oxaloacetate (Fig. 10.12). Thus this pathway is called C₄ photosynthesis. This initial addition relies on the enzyme, PEP carboxylase, which has a strong affinity for CO₂ and partially explains the low CO₂ compensation point.

Oxaloacetate is generally reduced in the mesophyll cells to other four-carbon compounds, malate or aspartate. These compounds move from the mesophyll cells to bundle sheath cells, where they release CO₂, are reoxidized to oxaloacetate, and return to the mesophyll. This process effectively pumps CO₂ and electrons into the bundle sheath chloroplasts, where they are available to form sugars in the normal C₃ cycle.

The strong affinity of PEP carboxylase for CO₂ reduces the CO₂ concentration inside the leaf, which speeds the diffusion of CO₂ from outside air into the leaf even if stomata are partially closed. This explains how C₄ plants can reduce water loss under hot, dry conditions. Pumping CO₂ to the bundle sheath raises its concentration relative to that of O₂, which explains the very low level of photorespiration, even at high temperatures.

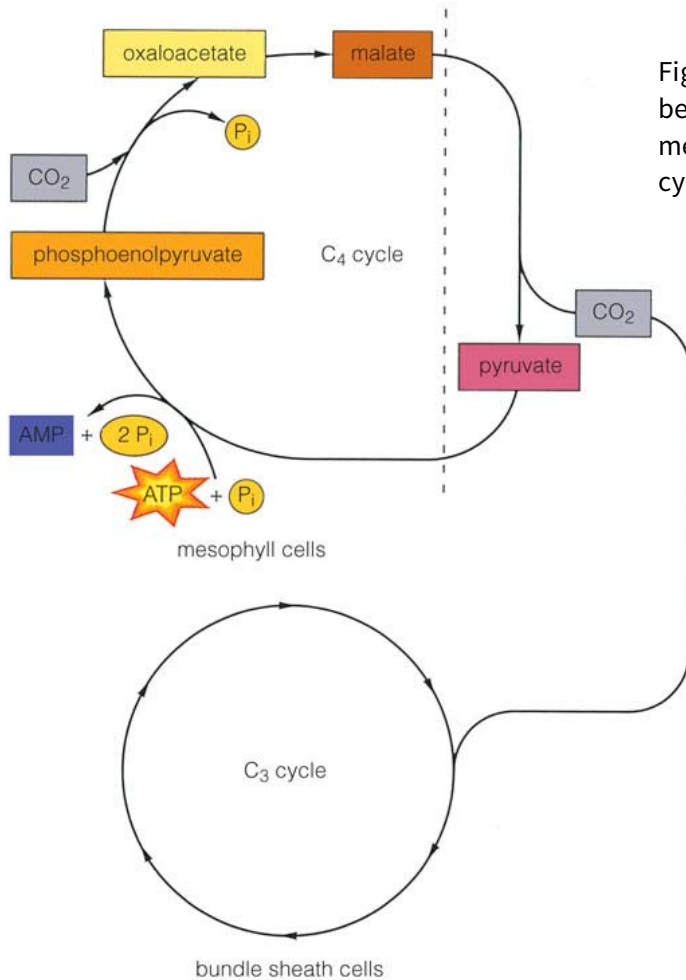


Figure 10.12. Interaction between the C₄ cycle in mesophyll cells and the C₃ cycle in bundle sheath cells.

Some Succulents Trap CO₂ at Night

In contrast to most mesophytes, which have stomata open during the day and closed at night, many succulents have their stomata closed during the day and open at night. This adaptation reduces water loss during the day, when water stress is high. It could, however, be disadvantageous for photosynthesis by reducing CO₂ uptake in the daylight, which is when light reactions occur. These plants minimize this disadvantage by using a process called **Crassulacean acid metabolism (CAM)**. Botanists first observed it in plants belonging to the family Crassulaceae, but they now know it is common among plants in 10 other families, including the Cactaceae, Euphorbiaceae, Liliaceae, and Orchidaceae.

The major features unique to CAM are the following:

1. During the night stomata are open, and the leaves rapidly absorb CO₂. During the day stomata are closed, greatly reducing CO₂ absorption and water loss.
2. At night, when CO₂ is being absorbed, the enzyme PEP carboxylase initiates the fixation of CO₂. Generally, a four-carbon compound, often malate, is produced.

3. The total amount of organic acids rapidly increases at night, as malate is stored in leaf-cell vacuoles.
4. During the day, the stored malate is decarboxylated and the CO₂ is released. Leaf acidity rapidly decreases.
5. The released CO₂ is incorporated into carbohydrate by C₃ photosynthesis.

These features give CAM plants an effective mechanism for trapping CO₂ at night, when temperatures and transpiration are low, and using it for photosynthesis during the day, when temperatures are high but transpiration can still be controlled. The daily rate of photosynthesis in CAM plants is limited by the amount of organic acid that can be formed and stored during the night, which means that growth rates of CAM plants are often low.

10.4 FACTORS AFFECTING PRODUCTIVITY

The photosynthetic leaf is a highly evolved unit with many features that promote an efficient capture of light and carbon dioxide. Nevertheless, only about 0.3% to 0.5% of the light energy that strikes a leaf is stored as photosynthate. The yield may be increased by a factor of 10 under ideal conditions. In a hungry world, there is much to be gained by exploring and controlling the conditions that limit plant productivity, the amount of living tissue produced per unit of time by a plant or population of plants.

Greater Productivity Can Be Bred into Plants

Plant productivity is determined partly by the environment and partly by the hereditary traits of the plant. One avenue toward greater productivity is to breed more efficient plants. This approach has already been quite successful in the case of cereal grains. Norman Borlaug received the Nobel Prize in 1970 for developing high-yielding wheat (*Triticum*) strains so productive as to constitute a *green revolution* in tropical countries. Unfortunately, these strains require high levels of fertilizer application--an expensive and pollution-creating practice. There also are potential genetic problems associated with these grains.

Photorespiration is one example of a hereditary trait that reduces plant productivity. The C₄ system offers a compensating hereditary advantage. Perhaps breeding programs or recombinant DNA technology will lead to new C₄ varieties or to C₃ plants that are less prone to photorespiration.

Fluctuations in the Environment Alter the Rate of Photosynthesis

The environment strongly influences the rate of photosynthesis and plant productivity. To a certain degree, some environmental factors are under the control of a plant grower--for instance, water and mineral nutrients. Other factors--

temperature, light intensity, duration, and quality, and CO₂ concentration--offer opportunities to increase productivity but require special equipment.

TEMPERATURE Plants are capable of photosynthesis over a wide temperature range. Plants native to Arctic regions may photosynthesize at temperatures below 0°C. Some lichens from the Antarctic carry out photosynthesis at -18°C and have a photosynthetic temperature optimum near 0°C. Cyanobacteria in hot springs may photosynthesize at a temperature as high as 70°C. C₄ species generally have higher temperature optima for photosynthesis than C₃ species.

Nevertheless, most plants function best between temperatures of 10°C and 25°C. If there is adequate light and a normal supply of CO₂, the rate of photosynthesis increases with temperature up to about 25°C; above this temperature there is a decrease as temperature increases. The longer the exposure to a given high temperature, the greater is the decrease in photosynthetic rate.

LIGHT Both the intensity and the wavelength of light affect the rate of photosynthesis. Assuming moderate temperatures and sufficient CO₂, the rate of carbohydrate production increases with increasing light intensity, up to a maximum, after which production decreases. Several factors influence the light-dependence of photosynthesis:

1. Leaf structure. Surface hairs, thick cuticle, thick epidermis, and other structural features of a leaf affect the light intensity at the chloroplasts.
2. Community structure. Leaves on the surface of the plant canopy receive more intense light than those beneath, which are shaded. Therefore, some of the leaves receive light of optimum intensity, whereas others get light greater or less than the optimum.

Intense light appears to retard the rate of photosynthesis. Many plants living in places where the light is very bright often have structural adaptations that tend to diminish the intensity reaching the chloroplasts. The usual light intensity in arid and semiarid regions is considerably greater than the optimum for photosynthesis, especially for crop plants. In these places, the light intensity is probably nearer the optimum on days when the sky is overcast than on clear, sunny days.

A special situation exists in a dense stand of plants such as a forest, where the vertical distribution of light varies dramatically--from full sunlight on the upper leaves of the canopy to very dim light at the floor. Plants growing below a canopy receive full sunlight for relatively short periods, when pulses of light--called sunflecks--penetrate through holes in the canopy. When breezes move the leaves, these brief exposures to direct sunlight may become very short indeed. One may wonder whether plants can use light from these sunflecks efficiently. Research shows that as much as 60% of the daily carbon gain by seedlings of Hawaiian understory trees comes from sunflecks.

Under many natural conditions, the quality of light varies widely. Such variations may determine seedling survival and growth on forest floors. The leaves of the overstory canopy absorb predominantly red and blue light. Thus, lower

leaves, screened from direct sunlight by upper leaves, receive light rich in green wavelengths. Marine algae growing in deep water also receive light rich in blue-green wavelengths. Many such plants develop strong accessory pigment systems that absorb light and pass it to photosystem reaction centers.

CARBON DIOXIDE Earth's atmosphere contains approximately 0.039% to 0.040% carbon dioxide. The amount of this gas used by plants is very great. A hectare (2.5 acres) of corn (*Zea mays*) may contain 24,700 plants, which during a growing season of 100 days can accumulate 6,200 kg (6.83 tons) of CO₂. All this carbon is derived from the atmosphere.

The concentration of atmospheric CO₂ limits the rate of photosynthesis for C₃ plants. This may be particularly true in greenhouses kept closed in the winter. Under these conditions, the CO₂ may be reduced to much less than the average value of outside air. An artificial increase in the CO₂ concentration--up to 0.6%--may increase the rate of photosynthesis, but only for a limited period. It appears that this high a level of CO₂ is injurious to some plants after 10 to 15 days of exposure.

The concentration of CO₂ in the atmosphere has been rising for several years (Fig. 10.13) due to human activities, predominantly deforestation and the burning of coal and oil as energy sources. In 1960, the average concentration was less than 0.032%. In 2014, as mentioned above, the concentration topped 0.040%. Some plant physiologists have hypothesized that an increase in photosynthesis, produced in response to rising CO₂ concentrations, will limit the atmospheric increase, but experiments in growth chambers and controlled outside environments do not yet support such speculations.

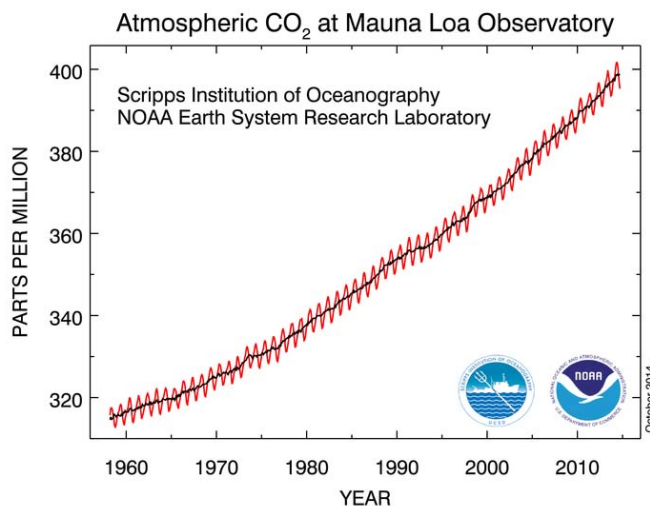


Figure 10.13. Rise in the concentration of CO₂ in the atmosphere at Mauna Loa, Hawaii, from 1958 to 2014.

WATER Most of the water absorbed by plant roots is lost in transpiration. A significant amount of the rest is used in the growth of the highly hydrated living cells. Only 1% or less is actually used in photosynthesis. However, the rate of photosynthesis may be changed by small differences in the water content of the chlorophyll-bearing cells. Drought reduces the rate of photosynthesis in some plants, such as long pod bean (*Vicia faba*), because the turgor of guard cells is

reduced and the stomata close. The decreased diffusion of CO₂ into the leaf limits the rate of photosynthesis.

MINERAL NUTRIENTS All of the essential elements for normal plant growth and development are required for an actively photosynthesizing plant. Several elements are specifically required for the development of photosynthetic systems. For instance, magnesium and nitrogen are part of the chlorophyll molecule, and iron is necessary for its synthesis. The water-splitting, oxygen-evolving system requires manganese, chloride, and calcium. Some electron carriers contain iron, and all proteins contain nitrogen. Sodium is needed for C₄ and CAM photosynthesis. Consequently, poor soils can result in plants with poorly developed photosynthetic capabilities. In these cases, yields can be greatly increased by effective fertilizer programs.

KEY TERMS

absorption spectrum
ribulose biphosphate
carboxylase/oxygenase (Rubisco)
carotenoids
chemosynthesis
compensation point
Crassulacean acid metabolism (CAM)
cyclic electron transport
cyclic photophosphorylation
electromagnetic energy spectrum
grana

light-harvesting complexes
noncyclic electron transport
noncyclic photophosphorylation
P₆₈₀
P₇₀₀
photons
photophosphorylation
photosystems I and II
spectrophotometer
stroma lamellae
thylakoids

SUMMARY

1. Photosynthesis is the primary energy-storing process of life. Light energy is stored as chemical energy in organic compounds.
2. Carbon dioxide and water are the raw materials. The products of photosynthesis are sugar and oxygen.
3. Light reactions and electron transport occur in thylakoid membranes. Thylakoids of green plants that evolve oxygen have two photosystems, photosystems I and II, with reaction centers P₇₀₀ and P₆₈₀, respectively.
4. Light energy absorbed by light-harvesting pigment complexes in the two photosystems is funneled to special chlorophyll molecules in the reaction centers. A

reaction center becomes excited as one of its electrons is driven into a higher energy level.

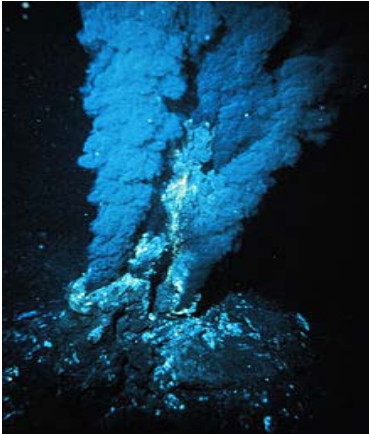
5. The electron that has gained energy escapes from the chlorophyll molecule and passes down an electron transport chain.
6. NADP^+ acts as the final electron acceptor of P_{700} and forms NADPH, thereby storing some of the energy acquired by the electron through the absorption of light.
7. During electron transport, a proton (H^+) gradient is formed across the thylakoid membrane. This proton gradient drives the synthesis of ATP.
8. Electrons move from water to replace those lost by P_{680} . H^+ and O_2 are formed.
9. An enzyme in the stroma catalyzes the fixation of inorganic carbon by attaching CO_2 to an organic molecule.
10. In the stroma, the H^+ and electrons of NADPH are transferred to organic compounds and used in a series of reactions to produce sugar. ATP is used as an energy source to push the reactions forward.
11. In C_3 plants at high temperatures, photorespiration releases up to 50% of the previously captured CO_2 . This loss does not occur in C_4 plants, which therefore are more productive at high temperatures.
12. In CAM plants, CO_2 is captured at night when stomata are open. The resulting organic acid releases CO_2 to photosynthesizing cells during the day, when stomata are closed.
13. Photosynthesis may be limited by CO_2 , light, temperature, water, minerals, and the plant's hereditary information.
14. The atmosphere's concentration of CO_2 has been increasing because of the removal of forests and the burning of fossil fuel. As a result, global temperatures are expected to increase.

Questions

1. Write the general equation for photosynthesis in symbols and words.
2. Describe an experiment to demonstrate which colors of light are used in photosynthesis.
3. When light is absorbed by chlorophyll in a leaf, what happens to an electron in the reaction center?

4. Where in the chloroplast is a proton gradient established? What is its significance? Is electron transport required? Is O₂ synthesis required?
5. What are the first two molecules that store energy from the light absorbed during photosynthesis?
6. Name and briefly distinguish between the three major carbon cycles of photosynthesis.
7. Under what environmental conditions would CAM and C₄ metabolism be advantageous to a plant?
8. A person was growing plants in a greenhouse in the winter. The greenhouse was closed and heated to keep it warm. The plants grew very slowly. Offer some reasonable hypotheses to explain the slow growth.

IN DEPTH: *Chemosynthesis*



In the 1970s, scientists discovered vents deep in the Pacific Ocean associated with the upwelling of hot sea water--hydrothermal vents. Rich animal populations including large clams, crabs, mussels, and worms are clustered around these hydrothermal vents. Some of these animals live deep within the vents. It was shown that organic materials produced by photosynthetic organisms near the ocean surface do not provide adequate food to sustain the life of these organisms. Instead, they obtain their food from dense microbial mats consisting primarily of bacteria.

Many of these bacteria use reduced inorganic compounds such as H_2S , that are in high concentrations in the vents. These microorganisms oxidize the reduced inorganic compounds and use the energy liberated to assimilate CO_2 into organic compounds in a process known as **chemosynthesis**. Chemosynthesis is similar to photosynthesis in several respects, but chemical energy rather than light energy drives the synthetic reactions. In addition to obtaining organic matter by grazing on the microbial mass, some of the animal species in the vents live in symbiotic association with chemosynthetic organisms.

In 1996, the bacteria in a cave in southern Romania were studied using radioactive ^{14}C bicarbonate to determine their source of carbon. It was found that the radioactive carbon was incorporated into microbial lipids. Because light was not available to these bacteria, chemosynthetic fixation must have occurred. Organisms that can produce their own food from inorganic materials using reduced inorganic compounds instead of light as a sources of energy are *chemoautotrophic organisms*. Just as chemoautotrophic bacteria serve as a food base for the marine life in the hydrothermal vents in the ocean, the chemoautotrophic bacteria in the cave in Romania serve as a food base for a variety of cave-dwelling organisms.

CO <http://en.wikipedia.org/wiki/Leaf>

Figure 10.1. Art by Raychel Ciemma

Figure 10.2(a). Art by Raychel Ciemma

Figure 10.2(b). Micrograph by David Fisher

Figure 10.10. Thomas L. Rost

Figure 10.11. Art by Raychel Ciemma

Figure 10.12. NOAA and Scripps Institute of Oceanography, October 2014