41

How Plants Grow in Response to Their Environment

Concept Outline

41.1 Plant growth is often guided by environmental cues.

Tropisms. Plant growth is often influenced by light, gravity, and contact with other plants and animals.
Dormancy. The ability to cease growth allows plants to wait out the bad times.

41.2 The hormones that guide growth are keyed to the environment.

Plant Hormones. Hormones are grouped into seven classes.
Auxin. Auxin is involved in the elongation of stems.
Cytokinins. Cytokinins stimulate cell division.
Gibberellins. Gibberellins control stem elongation.
Brassinosteroids and Oligosaccharins. There are several recent additions to the plant hormone family.
Ethylene. Ethylene controls leaves and flower abscision.
Abscisic Acid. Abscisic acid suppresses growth of buds and promotes leaf senescence.

41.3 The environment influences flowering.

Plants Undergo Metamorphosis. The transition of a shoot meristem from vegetative to adult is called phase change.
Pathways Leading to Flower Production. Photoperiod is regulated in complex ways.
Identity Genes and the Formation of Floral Meristems and Floral Organs. Floral meristem identity genes activate floral organ identity genes.

41.4 Many short-term responses to the environment do not require growth.

Turgor Movement. Changes in the water pressure within plant cells result in quick and reversible plant movements.
Plant Defense Responses. In addition to generalized defense mechanisms, some plants have highly evolved recognition mechanisms for specific pathogens.

FIGURE 41.1
Plant growth is affected by environmental cues. The branches of this fallen tree are growing straight up in response to gravity and light.

All organisms sense and interact with their environment. This is particularly true of plants. Plant survival and growth is critically influenced by abiotic factors including water, wind, and light. In this chapter, we will explore how a plant senses such factors, and transduces these signals to elicit an optimal physiological, growth, or developmental response. Hormones play an important role in the internal signaling that brings about environmental responses, and is keyed in many ways to the environment. The effect of the local environment on plant growth also accounts for much of the variation in adult form within a species (figure 41.1). Precisely regulated responses to the environment not only allow a plant to survive from day to day but also determine when a flowering plant will produce a flower. The entire process of constructing a flower in turn sets the stage for intricate reproductive strategies that will be discussed in the next chapter.

41.1 Plant growth is often guided by environmental cues.

Tropisms

Growth patterns in plants are often guided by environmental signals. **Tropisms** (from *trope*, the Greek word for "turn") are positive or negative growth responses of plants to external stimuli that usually come from one direction. Some responses occur independently of the direction of the stimuli and are referred to as nastic movements. For example, a tendril of a pea plant will always coil in one direction when touched. Tropisms, on the other hand, are directional and offer significant compensation for the plant's inability to get up and walk away from unfavorable environmental conditions. Tropisms contribute the variety of branching patterns we see within a species. Here we will consider three major classes of plant tropisms: phototropism, gravitropism, and thigmotropism. Tropisms are particularly intriguing because they challenge us to connect environmental signals with cellular perception of the signal, transduction into biochemical pathways, and ultimately an altered growth response.

FIGURE 41.2
Phototropism. *Impatiens* plant growing toward light.

Phototropism

Phototropic responses involve the bending of growing stems and other plant parts toward sources of light (figure 41.2). In general, stems are positively phototropic, growing toward a light source, while most roots do not respond to light or, in exceptional cases, exhibit only a weak negative phototropic response. The phototropic reactions of stems are clearly of adaptive value, giving plants greater exposure to available light. They are also important in determining the development of plant organs and, therefore, the appearance of the plant. Individual leaves may display phototropic responses. The position of leaves is important to the photosynthetic efficiency of the plant. A plant hormone called auxin (discussed later in this chapter) is probably involved in most, if not all, of the phototropic growth responses of plants.

The first step in a phototropic response is perceiving the light. Photoreceptors perceive different wavelengths of light with blue and red being the most common. Blue light receptors are being characterized and we are beginning to understand how plants "see blue." Much more is known about "seeing red" and translating that perception into a signal transduction pathway leading to an altered growth response. Plants contain a pigment, **phytochrome,** which exists in two interconvertible forms, P_r and P_{fr}. In the first form, phytochrome absorbs red light; in the second, it absorbs far-red light. When a molecule of P_r absorbs a photon of red light (660 nm), it is instantly converted into a molecule of P_{fr}, and when a molecule of P_{fr} absorbs a photon of far-red light (730 nm), it is instantly converted to P_r. P_{fr} is biologically active and P_r is biologically inactive. In other words, when P_{fr} is present, a given biological reaction that is affected by phytochrome will occur. When most of the P_{fr} has been replaced by P_r, the reaction will not occur (figure 41.3). While we refer to phytochrome as a single molecule here, it is important to note that several different phytochromes have now been identified that appear to have specific biological functions.

Phytochrome is a light receptor, but it does not act directly to bring about reactions to light. The existence of phytochrome was conclusively demonstrated in 1959 by Harry A. Borthwick and his collaborators at the U.S. Department of Agriculture Research Center at Beltsville, Maryland. It has since been shown that the molecule consists of two parts: a smaller one that is sensitive to light and a larger portion that is a protein. The protein component initiates a signal transduction leading to a particular tropism. The phytochrome pigment is blue, and its light-sensitive portion is similar in structure to the phycobilins that occur in cyanobacteria and red algae. Phytochrome is present in all groups of plants and in a few genera of green algae, but not in bacteria, fungi, or protists (other than the few green algae). It is likely that phytochrome systems for measuring light evolved among the green algae and were present in the common ancestor of the plants.

Phytochrome is involved in many plant growth responses. For example, seed germination is inhibited by far-red light and stimulated by red light in many plants. Because chlorophyll absorbs red light strongly but does not absorb far-red light, light passing through green leaves inhibits seed germination. Consequently, seeds on the

ground under deciduous plants that lose their leaves in winter are more apt to germinate in the spring after the leaves have decomposed and the seedlings are exposed to direct sunlight. This greatly improves the chances the seedlings will become established.

A second example of these relationships is the elongation of the shoot in an *etiolated* seedling (one that is pale and slender from having been kept in the dark). Such plants become normal when exposed to light, especially red light, but the effects of such exposure are canceled by far-red light. This indicates a relationship similar to that observed in seed germination. There appears to be a link between phytochrome light perception and brassinosteroids in the etiolation response. Etiolation is an energy conservation strategy to help plants growing in the dark reach the light before they die. They don't green-up until there is light, and they divert energy to growing as tall as possible through internode elongation. The de-etiolated (*det2*) *Arabidopsis* mutant has a poor etiolation response. It does not have elongated internodes and greens up a bit in the dark. It turns out that *det2* mutants are defective in an enzyme necessary for brassinosteroid biosynthesis. Researchers suspect that brassinosteroids play a role in how plants respond to light through phytochrome. Thus, because *det2* mutants lack brassinosteroids, they do not respond to light, or lack of light, as normal plants do, and the *det2* mutants grow normally in the dark.

Red and far-red light also are used as signals for plant spacing. The closer plants are together, the more likely they are to grow tall and try to outcompete others for the sunshine. Plants somehow measure the amount of far-red light being bounced back to them from neighboring trees. If their perception is messed up by putting a collar around the stem with a solution that blocks light absorption, the elongation response is no longer seen.

Gravitropism

When a potted plant is tipped over, the shoot bends and grows upward (figure 41.4). The same thing happens when a storm pushes over plants in a field. These are examples of **gravitropism**, the response of a plant to the gravitational field of the earth. We saw in chapter 40 that brown algae orient their first cell division so the rhizoid grows downward. Rhizoids also develop away from a unilateral light source. Separating out phototropic effects is important in the study of gravitropisms.

Gravitropic responses are present at germination when the root grows down and the shoot grows up. Why does a shoot have a negative gravitropic response (growth away from gravity), while a root has a positive gravitropic response? The opportunity to experiment on the space shuttle in a gravity-free environment has accelerated research in this area. Auxins play a primary role in gravitropic responses, but they may not be the only way gravitational information is sent through the plant. When John
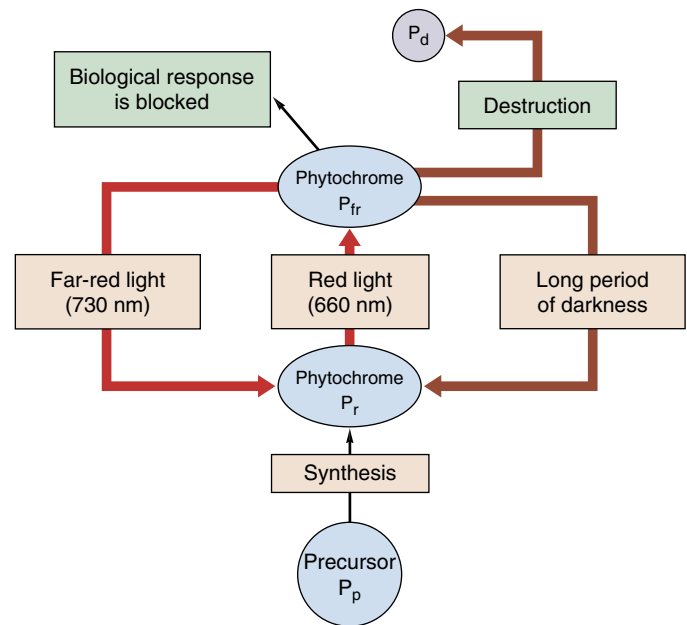
FIGURE 41.3
How phytochrome works. Phytochrome is synthesized in the P_r form from amino acids, designated P_p for phytochrome precursor. When exposed to red light, P_r changes to P_{fr}, which is the active form that elicits a response in plants. P_{fr} is converted to P_r when exposed to far-red light, and it also converts to P_r or is destroyed in darkness. The destruction product is designated P_d.
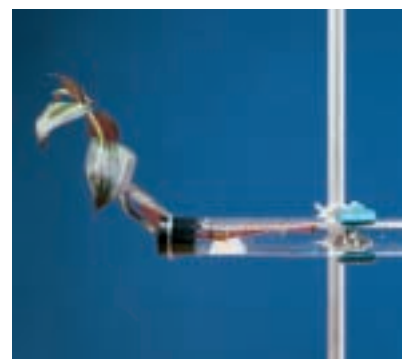
FIGURE 41.4
Plant response to gravity. This plant (*Zebrina pendula*) was placed horizontally and allowed to grow for 7 days. Note the negative gravitational response of the shoot.

Glenn made his second trip into space, he was accompanied by an experiment designed to test the role of gravity and electrical signaling in root bending. Analysis of gravitropic mutants is also adding to our understanding of gravitropism. There are four steps that lead to a gravitropic response:

1. Gravity is perceived by the cell
2. Signals form in the cell that perceives gravity
3. The signal is transduced intra- and intercellularly
4. Differential cell elongation occurs between cells in the "up" and "down" sides of the root or shoot.

One of the first steps in perceiving gravity is that amyloplasts, plastids that contain starch, sink toward the gravitational field. These may interact with the cytoskeleton, but the net effect is that auxin becomes more concentrated on the lower side of the stem axis than on the upper side. The increased auxin concentration on the lower side in stems causes the cells in that area to grow more than the cells on the upper side. The result is a bending upward of the stem against the force of gravity—in other words there is a *negative gravitropic response*. Such differences in hormone concentration have not been as well documented in roots. Nevertheless, the upper sides of roots oriented horizontally grow more rapidly than the lower sides, causing the root ultimately to grow downward; this phenomenon is known as *positive gravitropism*. In shoots, the gravity-sensing cells are in the endoderm. Mutants like *scarecrow* and *short root* in *Arabidopsis* that lack normal endodermal development fail to have a normal gravitropic response. These endodermal cells are the sites of the amyloplasts in the stems.

In roots, the gravity-sensing cells are located in the root cap and the cells that actually do the asymmetric growth are in the distal elongation zone which is closest to the root cap. How the information gets transferred over this distance is an intriguing problem. Auxin may be involved, but when auxin transport is suppressed, there is still a gravitropic response in the distal elongation zone. Some type of electrical signaling involving membrane polarization has been hypothesized and this was tested aboard the space shuttle. So far the verdict is still not in on the exact mechanism.

It may surprise you to learn that in tropical rain forests, roots of some plants may grow up the stems of neighboring plants, instead of exhibiting the normal positive gravitropic responses typical of other roots. The rainwater dissolves nutrients, both while passing through the lush upper canopy of the forest, and also subsequently as it trickles down tree trunks. Such water functions as a more reliable source of nutrients for the roots than the nutrient-poor soils in which the plants are anchored. Explaining this in terms of current hypotheses is a challenge. It has been proposed that roots are more sensitive to auxin than shoots and that auxin may actually inhibit growth on the lower side of a root, resulting in a positive gravitropic response. Perhaps in these tropical plants, the sensitivity to auxin in roots is reduced.

Thigmotropism

Thigmotropism is a name derived from the Greek root *thigma*, meaning "touch." A thigmotropism is a response of a plant or plant part to contact with the touch of an object, animal, plant, or even the wind. (figure 41.5). When a tendril makes contact with an object, specialized epidermal cells, whose action is not clearly understood, perceive the contact and promote uneven growth, causing the tendril to

FIGURE 41.5 Thigmotropism. The thigmotropic response of these twining stems causes them to coil around the object with which they have come in contact.

curl around the object, sometimes within as little as 3 to 10 minutes. Both auxin and ethylene appear to be involved in tendril movements, and they can induce coiling in the absence of any contact stimulus. In other plants, such as clematis, bindweed, and dodder, leaf petioles or unmodified stems twine around other stems or solid objects.

Again, *Arabidopsis* is proving valuable as a model system. A gene has been identified that is expressed in 100-fold higher levels 10 to 30 minutes after touch. Given the value of a molecular genetics approach in dissecting the pathways leading from an environmental signal to a growth response, this gene provides a promising first step in understanding how plants respond to touch.

Other Tropisms

The tropisms just discussed are among the best known, but others have been recognized. They include *electrotropism* (responses to electricity); *chemotropism* (response to chemicals); *traumotropism* (response to wounding which we discuss on page 834); *thermotropism* (response to temperature); *aerotropism* (response to oxygen); *skototropism* (response to dark); and *geomagnetotropism* (response to magnetic fields). Roots will often follow a diffusion gradient of water coming from a cracked pipe and enter the crack. Some call such growth movement *hydrotropism*, but there is disagreement whether responses to water and several other "stimuli" are true tropisms.

While plants can't move away or toward optimal conditions, they can grow. Phototropisms are growth responses of plants to a unidirectional source of light. Gravitropism, the response of a plant to gravity, generally causes shoots to grow up (negative gravitropism) and roots to grow down (positive gravitropism). Thigmotropisms are growth responses of plants to contact.

Dormancy

Sometimes modifying the direction of growth is not enough to protect a plant from harsh conditions. The ability to cease growth and go into a dormant stage provides a survival advantage. The extreme example is seed dormancy, but there are intermediate approaches to waiting out the bad times as well. Environmental signals both initiate and end dormant phases in the life of a plant.

In temperate regions, we generally associate dormancy with winter, when freezing temperatures and the accompanying unavailability of water make it impossible for plants to grow. During this season, buds of deciduous trees and shrubs remain dormant, and apical meristems remain well protected inside enfolding scales. Perennial herbs spend the winter underground as stout stems or roots packed with stored food. Many other kinds of plants, including most annuals, pass the winter as seeds.

In some seasonally dry climates, seed dormancy occurs primarily during the dry season, often the summer. Rainfalls trigger germination when conditions for survival are more favorable. Annual plants occur frequently in areas of seasonal drought. Seeds are ideal for allowing annual plants to bypass the dry season, when there is insufficient water for growth. When it rains, they can germinate and the plants can grow rapidly, having adapted to the relatively short periods when water is available. Chapter 40 covered some of the mechanisms involved in breaking seed dormancy and allowing germination under favorable circumstances. These include the leaching from the seed coats of chemicals that inhibit germination, or mechanically cracking the seed coats, a procedure that is particularly suitable for promoting growth in seasonally dry areas. Whenever rains occur, they will leach out the chemicals from the seed coats, and the hard coats of other seeds may be cracked when they are being washed down along temporarily flooded arroyos (figure 41.6).

Seeds may remain dormant for a surprisingly long time. Many legumes (plants of the pea and bean family, Fabaceae) have tough seeds that are virtually impermeable to water and oxygen. These seeds often last decades and even longer without special care; they will eventually germinate when their seed coats have been cracked and water is available. Seeds that are thousands of years old have been successfully germinated!

A period of cold is necessary before some kinds of seeds will germinate, as we mentioned in chapter 40. The seeds of other plants will germinate only when adequate water is available and the temperatures are relatively high. For this reason, certain weeds germinate and grow in the cooler part of the year and others in the warmer part of the year. Similarly, a period of cold is needed before the buds of some trees and shrubs will break dormancy and develop normally. For this reason, many plants that normally grow in temperate regions do not thrive in warmer regions near the equator, because even at high elevations

(a)

(b)

FIGURE 41.6
Palo verde *(Cercidium floridum)*. This desert tree (*a*) has tough seeds (*b*) that germinate only after they are cracked.

in the tropics it still does not get cold enough, and the day-length relationships are different from those of temperate regions.

Mature plants may become dormant in dry or cold seasons that are unfavorable for growth. Dormant plants usually lose their leaves and drought-resistant winter buds are produced. Long unfavorable periods may be bypassed through the production of seeds, which themselves can remain dormant for long periods.

Plant Hormones

While initial responses of plants to environmental signals may rely primarily on electrical signaling, longer-term responses that alter morphology rely on complex physiological networks. Many internal signaling pathways involve plant hormones, which are the focus of this section. Hormones are involved in responses to the environment, as well as internally regulated development (examples of which you saw in chapter 40).

Hormones are chemical substances produced in small, often minute, quantities in one part of an organism and then transported to another part, where they bring about physiological or developmental responses. The activity of hormones results from their capacity to stimulate certain physiological processes and to inhibit others (figure 41.7). How they act in a particular instance is influenced both by the hormone and the tissue that receives the message.

In animals, hormones are usually produced at definite sites, usually organs. In plants, hormones are not produced in specialized tissues but, instead, in tissues that also carry out other, usually more obvious, functions. There are seven major kinds of plant hormones: auxin, cytokinins, gibberellins, br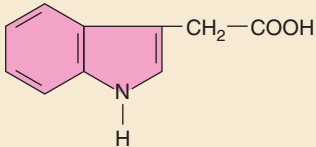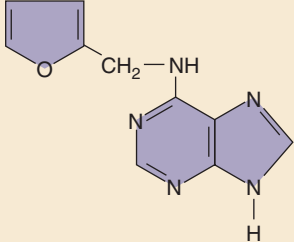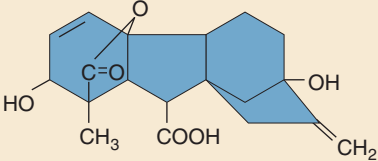assinosteroids, oligosaccharins, ethylene, and abscisic acid (table 41.1). Current research is focused on the biosynthesis of hormones and on characterizing the hormone receptors that trigger signal transduction pathways. Much of the molecular basis of hormone function remains enigmatic.

Because hormones are involved in so many aspects of plant function and development, we have chosen to integrate examples of hormone activity with specific aspects of plant biology throughout the text. In this section, our goal is to give you a brief overview of these hormones. Use this section as a reference when specific hormones are discussed in the next few chapters.

There are seven major kinds of plant hormones: auxin, cytokinins, gibberellins, brassinosteroids, oligosaccharins, ethylene, and abscisic acid.
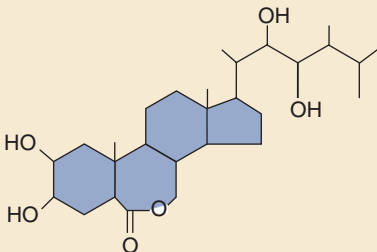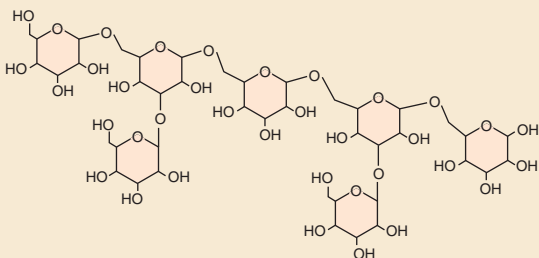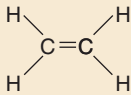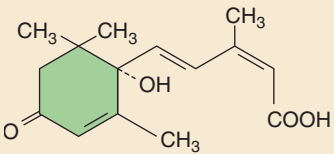
(a)

(b)

FIGURE 41.7
Effects of plant hormones. Plant hormones, often acting together, influence many aspects of plant growth and development, including (*a*) leaf abscission and (*b*) the formation of mature fruit.

Table 41.1 Functions of the Major Plant Hormones

Hormone		Major Functions	Where Produced or Found in Plant
Auxin (IAA)		Promotion of stem elongation and growth; formation of adventitious roots; inhibition of leaf abscission; promotion of cell division (with cytokinins); inducement of ethylene production; promotion of lateral bud dormancy	Apical meristems; other immature parts of plants
Cytokinins		Stimulation of cell division, but only in the presence of auxin; promotion of chloroplast development; delay of leaf aging; promotion of bud formation	Root apical meristems; immature fruits
Gibberellins		Promotion of stem elongation; stimulation of enzyme production in germinating seeds	Roots and shoot tips; young leaves; seeds
Brassinosteroids		Overlapping functions with auxins and gibberellins	Pollen, immature seeds, shoots, leaves
Oligosaccharins		Pathogen defense, possibly reproductive developmentz	Cell walls
Ethylene		Control of leaf, flower, and fruit abscission; promotion of fruit ripening	Roots, shoot apical meristems; leaf nodes; aging flowers; ripening fruits
Abscisic acid		Inhibition of bud growth; control of stomatal closure; some control of seed dormancy; inhibition of effects of other hormones	Leaves, fruits, root caps, seeds

Auxin

More than a century ago, an organic substance known as **auxin** became the first plant hormone to be discovered. Auxin increases the plasticity of plant cell walls and is involved in elongation of stems. Cells can enlarge in response to changes in turgor pressure when their cell walls have enhanced plasticity from auxin action. The discovery of auxin and its role in plant growth is an elegant example of thoughtful experimental design. The historical story is recounted here for that reason. Recent efforts have uncovered an auxin receptor. Transport mechanisms are also being unraveled. As with all the classes of hormones, we are just beginning to understand, at a cellular and molecular level, how hormones regulate growth and development.

Discovery of Auxin

In his later years, the great evolutionist, Charles Darwin, became increasingly devoted to the study of plants. In 1881, he and his son Francis published a book called *The Power of Movement of Plants*. In this book, the Darwins reported their systematic experiments on the response of growing plants to light—responses that came to be known as **phototropisms.** They used germinating oat *(Avena sativa)* and canary grass *(Phalaris canariensis)* seedlings in their experiments and made many observations in this field.

Charles and Francis Darwin knew that if light came primarily from one direction, the seedlings would bend strongly toward it. If they covered the tip of the shoot with a thin glass tube, the shoot would bend as if it were not covered. However, if they used a metal foil cap to exclude light from the plant tip, the shoot would not bend (figure 41.8). They also found that using an opaque collar to exclude light from the stem below the tip did not keep the area above the collar from bending.

In explaining these unexpected findings, the Darwins hypothesized that when the shoots were illuminated from one side, they bent toward the light in response to an "influence" that was transmitted downward from its source at the tip of the shoot. For some 30 years, the Darwins' perceptive experiments remained the sole source of information about this interesting phenomenon. Then Danish plant physiologist Peter Boysen-Jensen and the Hungarian plant physiologist Arpad Paal independently demonstrated that the substance that caused the shoots to bend was a chemical. They showed that if the tip of a germinating grass seedling was cut off and then replaced with a small block of agar separating it from the rest of the seedling, the seedling would grow as if there had been no change. Something evidently was passing from the tip of the seedling through the agar into the region where the bending occurred. On the basis of these observations under conditions of uniform illumination or of darkness, Paal suggested that


(a)
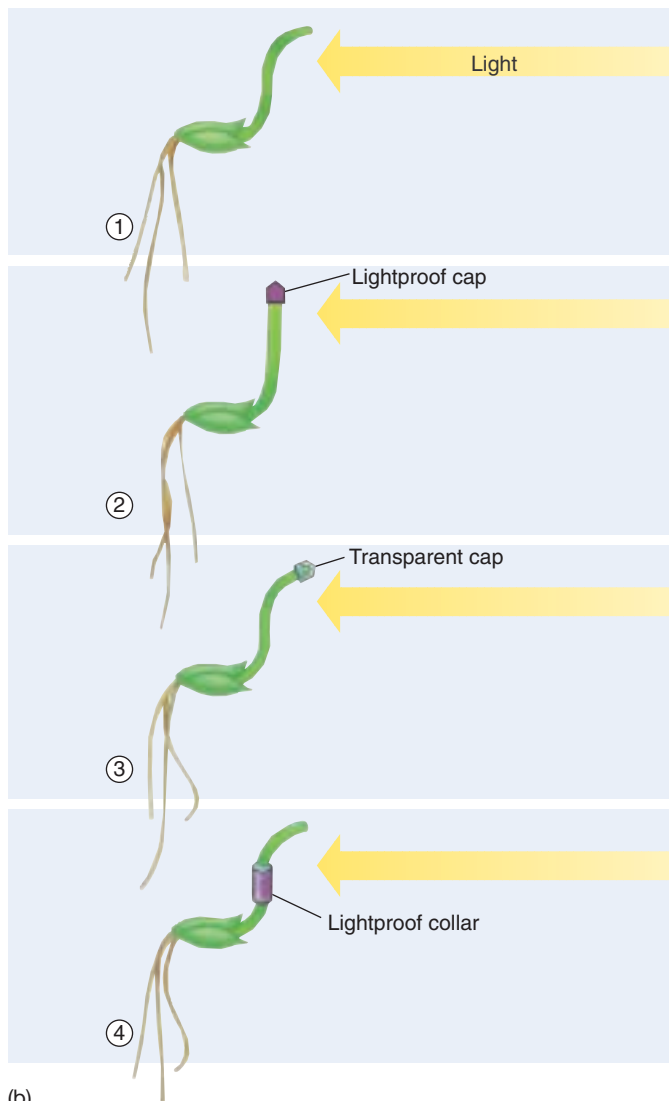
(b)

FIGURE 41.8
The Darwins' experiment. (*a*) Young grass seedlings normally bend toward the light. (*b*) The bending (*1*) did not occur when the tip of a seedling was covered with a lightproof cap (*2*), but did occur when it was covered with a transparent one (*3*). When a collar was placed below the tip (*4*), the characteristic light response took place. From these experiments, the Darwins concluded that, in response to light, an "influence" that caused bending was transmitted from the tip of the seedling to the area below, where bending normally occurs.

FIGURE 41.9
Frits Went's experiment. (*1*) Went removed the tips of oat seedlings and put them in agar, an inert, gelatinous substance. (*2*) Blocks of agar were then placed off-center on the ends of other oat seedlings from which the tips had been removed. (*3*) The seedlings bent away from the side on which the agar block was placed. Went concluded that the substance that he named *auxin* promoted the elongation of the cells and that it accumulated on the side of an oat seedling away from the light.

Auxin in tip of seedling

Agar

Auxin diffuses into agar block

Auxin
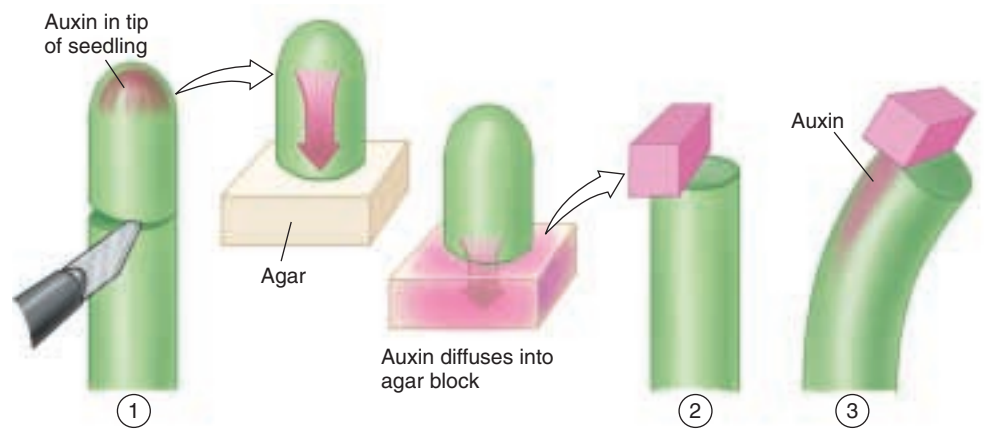
an unknown substance continually moves down from the tips of grass seedlings and promotes growth on all sides. Such a light pattern would not, of course, cause the shoot to bend.

Then, in 1926, Dutch plant physiologist Frits Went carried Paal's experiments an important step further. Went cut off the tips of oat seedlings that had been illuminated normally and set these tips on agar. He then took oat seedlings that had been grown in the dark and cut off their tips in a similar way. Finally, Went cut tiny blocks from the agar on which the tips of the light-grown seedlings had been placed and placed them off-center on the tops of the decapitated dark-grown seedlings (figure 41.9). Even though these seedlings had not been exposed to the light themselves, they bent away from the side on which the agar blocks were placed.

Went then put blocks of pure agar on the decapitated stem tips and noted either no effect or a slight bending toward the side where the agar blocks were placed. Finally, Went cut sections out of the lower portions of the light-grown seedlings to see whether the active principle was present in them. He placed these sections on the tips of decapitated, dark-green oat seedlings and again observed no effect.

As a result of his experiments, Went was able to show that the substance that had diffused into the agar from the tips of light-grown oat seedlings could make seedlings curve when they otherwise would have remained straight. He also showed that this chemical messenger caused the cells on the side of the seedling into which it flowed to grow more than those on the opposite side (figure 41.10). In other words, it enhanced rather than retarded cell elongation. He named the substance that he had discovered **auxin,** from the Greek word *auxein*, which means "to increase."

Went's experiments provided a basis for understanding the responses that the Darwins had obtained some 45 years earlier. The oat seedlings bent toward the light because of
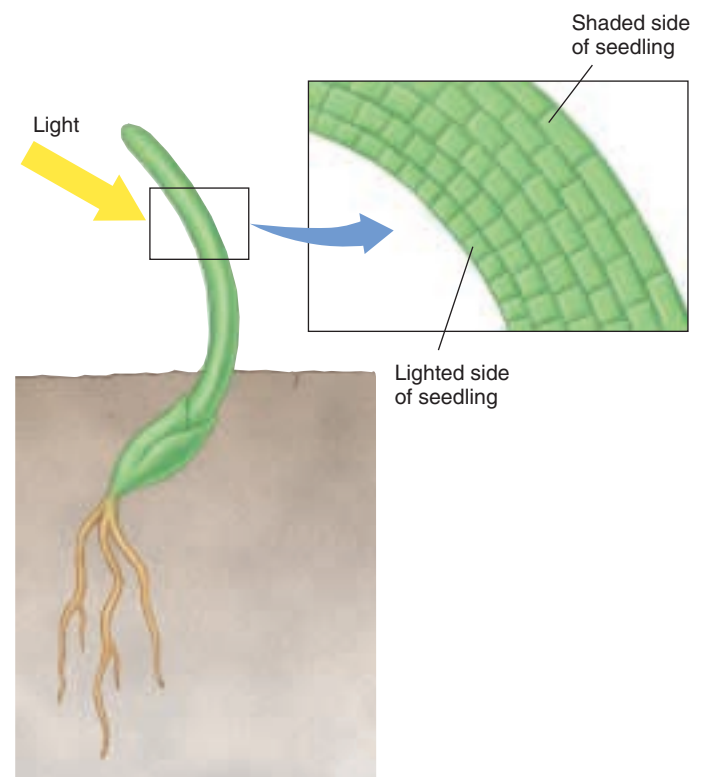

Light

Shaded side of seedling

Lighted side of seedling

FIGURE 41.10
Auxin causes cells on the dark side to elongate. Went determined that a substance called auxin enhanced cell elongation. Plant cells that are in the shade have more auxin and grow faster than cells on the lighted side, causing the plant to bend toward light. Further experiments showed exactly why there is more auxin on the shaded side of a plant.

differences in the auxin concentrations on the two sides of the shoot. The side of the shoot that was in the shade had more auxin, and its cells therefore elongated more than those on the lighted side, bending the plant toward the light.

The Effects of Auxins

Auxin acts to adapt the plant to its environment in a highly advantageous way. It promotes growth and elongation and facilitates the plant's response to its environment. Environmental signals directly influence the distribution of auxin in the plant. How does the environment—specifically, light—exert this influence? Theoretically, it might destroy the auxin, decrease the cells' sensitivity to auxin, or cause the auxin molecules to migrate away from the light into the shaded portion of the shoot. This last possibility has proved to be the case.

In a simple but effective experiment, Winslow Briggs inserted a thin sheet of mica vertically between the half of the shoot oriented toward the light and the half of the shoot oriented away from it (figure 41.11). He found that light from one side does not cause a shoot with such a barrier to bend. When Briggs examined the illuminated plant, he found equal auxin levels on both the light and dark sides of the barrier. He concluded that a normal plant's response to light from one direction involves auxin migrating from the light side to the dark side, and that the mica barrier prevented a response by blocking the migration of auxin.

The effects of auxin are numerous and varied. Auxin promotes the activity of the vascular cambium and the vascular tissues. Also, auxins are present in pollen in large quantities and play a key role in the development of fruits. Synthetic auxins are used commercially for the same purpose. Fruits will normally not develop if fertilization has not occurred and seeds are not present, but frequently they will if auxins are applied. Pollination may trigger auxin release in some species leading to fruit development occurring even before fertilization.

How Auxin Works

In spite of this long history of research on auxin, its molecular basis of action has been an enigma. The chemical structure of IAA resembles that of the amino acid tryptophan, from which it is probably synthesized by plants (figure 41.12). Unlike animal hormones, a specific signal is not sent to specific cells, eliciting a predictable response. There are most likely multiple auxin perception sites. Auxin is also unique among the plant hormones in that it is transported toward the base of the plant. Two families of genes have been identified in *Arabidopsis* that are involved in auxin transport. For example, one protein is involved in the top to bottom transport of auxin; while two other proteins function in the root tip to regulate the growth response to gravity. We are still a ways from linking the measurable and observable effects of auxin to events that transpire after it travels to a site and binds to a receptor.
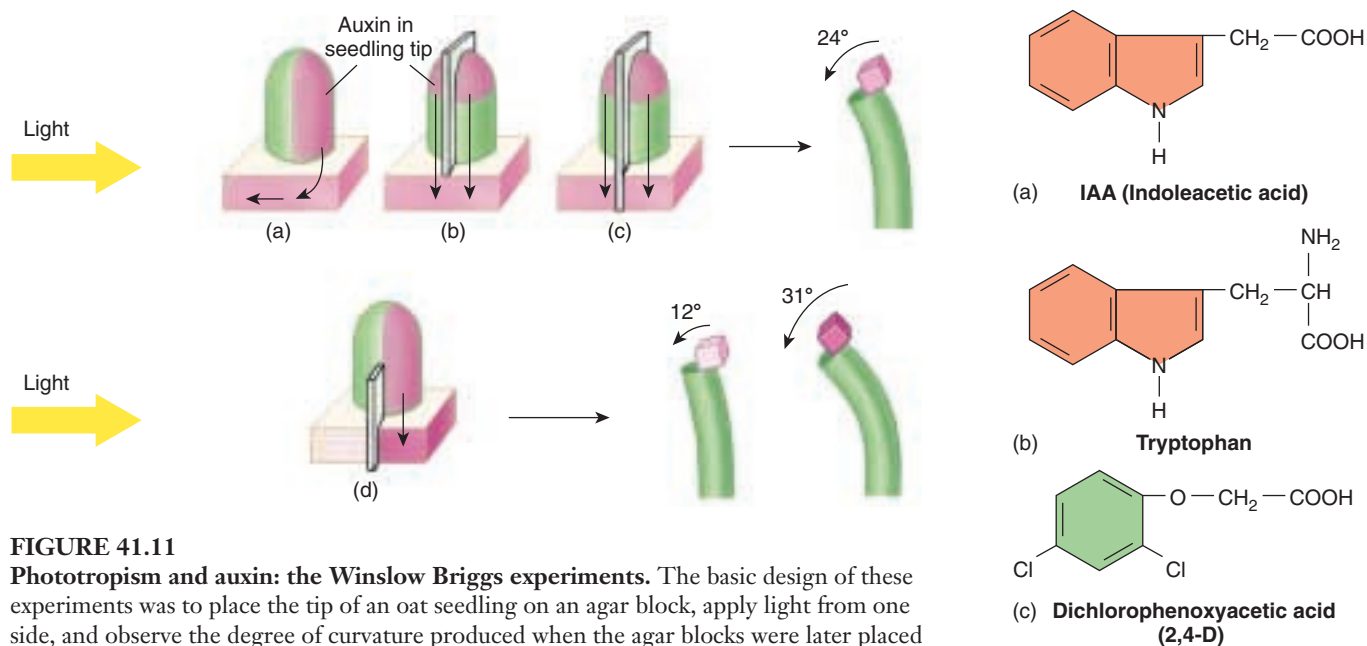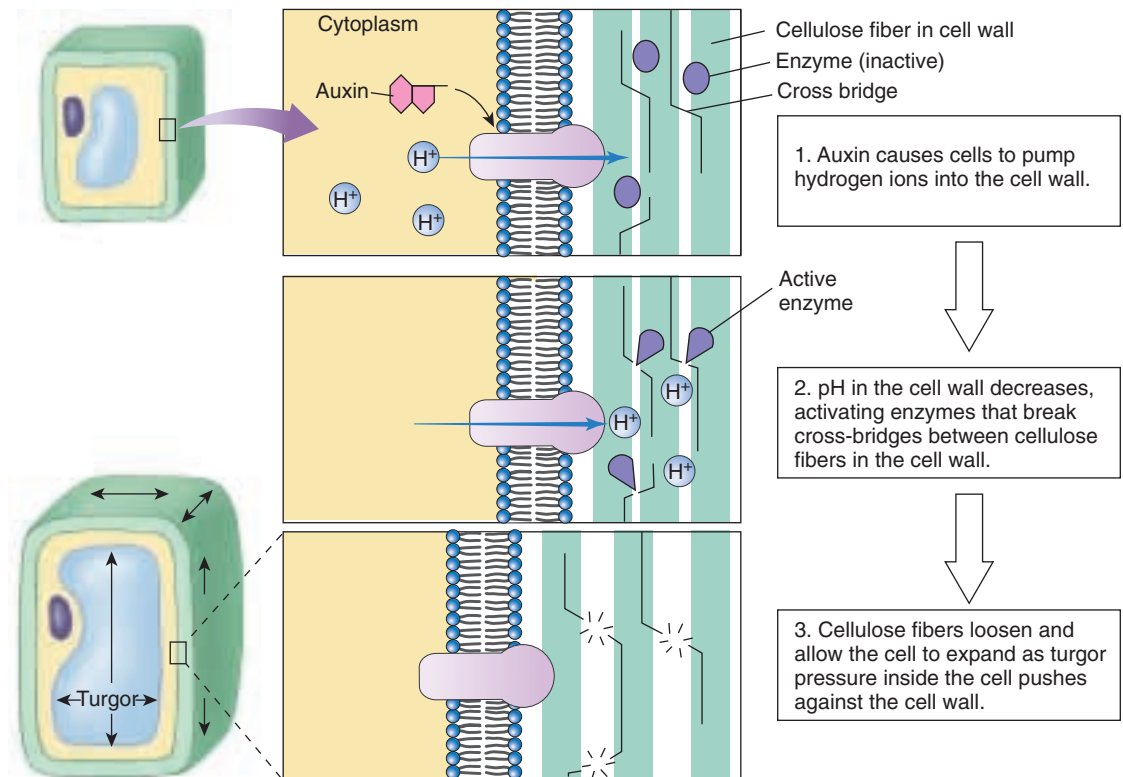

FIGURE 41.11
Phototropism and auxin: the Winslow Briggs experiments. The basic design of these experiments was to place the tip of an oat seedling on an agar block, apply light from one side, and observe the degree of curvature produced when the agar blocks were later placed on the decapitated seedlings. However, Briggs inserted a barrier in various places and noted how this affected the location of auxin. A comparison of (*a*) and (*b*) with similar experiments performed in the dark showed that auxin production does not depend on light; all produced approximately 24° of curvature. If a barrier was inserted in the agar block (*d*), light caused the displacement of the auxin away from the light. Finally, experiment (*c*) showed that it was displacement that had occurred, and not different rates of auxin production on the dark and light sides, because when displacement was prevented with a barrier, both sides of the agar block produced about 24° of curvature.

FIGURE 41.12
Auxins. (*a*) Indoleacetic acid (IAA), the principal naturally occurring auxin. (*b*) Tryptophan, the amino acid from which plants probably synthesize IAA. (*c*) Dichlorophenoxyacetic acid (2,4-D), a synthetic auxin, is a widely used herbicide.

FIGURE 41.13
Acid growth hypothesis. Auxin stimulates the release of hydrogen ions from the target cells which alters the pH of the cell wall. This optimizes the activity of enzymes which break bonds in the cell wall, allowing them to expand.

Cytoplasm
Cellulose fiber in cell wall
Enzyme (inactive)
Cross bridge
Auxin
H⁺
H⁺
H⁺

1. Auxin causes cells to pump hydrogen ions into the cell wall.

Active enzyme
H⁺
H⁺

2. pH in the cell wall decreases, activating enzymes that break cross-bridges between cellulose fibers in the cell wall.

Turgor

3. Cellulose fibers loosen and allow the cell to expand as turgor pressure inside the cell pushes against the cell wall.

One of the downstream effects of auxin is an increase in plasticity of the plant cell wall. This will only work on young cell walls without extensive secondary cell wall formation. A more plastic wall will stretch more while its protoplast is swelling during active cell growth. The **acid growth hypothesis** provides a model linking auxin to cell wall expansion (figure 41.13). Auxin causes responsive cells to release hydrogen ions into the cell wall. This decreases the pH which activates enzymes that can break bonds between cell wall fibers. Remember that different enzymes operate optimally at different pHs. This hypothesis has been experimentally supported in several ways. Buffers that prevent cell wall acidification block cell expansion. Other compounds that release hydrogen ions from the cell can also cause cell expansion. The movement of hydrogen ions has been observed in response to auxin treatment. This hypothesis explains the rapid growth response. There are also delayed responses which most likely involve auxin-stimulated gene expression.

Synthetic Auxins. Synthetic auxins such as NAA (naphthalene acetic acid) and IBA (indolebutyric acid) have many uses in agriculture and horticulture. One of their most important uses is based on their prevention of abscission, the process that causes a leaf or other organ to fall from a plant. Synthetic auxins are used to prevent fruit drop in apples before they are ripe and to hold berries on holly that is being prepared for shipping. Synthetic auxins are also used to promote flowering and fruiting in pineapples and to induce the formation of roots in cuttings.

Synthetic auxins are routinely used to control weeds. When used as herbicides, they are applied in higher concentrations than IAA would normally occur in plants. One of the most important synthetic auxin herbicides is 2,4-dichlorophenoxyacetic acid, usually known as 2,4-D (see figure 41.12c). It kills weeds in grass lawns by selectively eliminating broad-leaved dicots. The stems of the dicot weeds cease all axial growth.

The herbicide 2,4,5-trichlorophenoxyacetic acid, better known as 2,4,5-T, is closely related to 2,4-D. 2,4,5-T was widely used as a broad-spectrum herbicide to kill weeds and seedlings of woody plants. It became notorious during the Vietnam War as a component of a jungle defoliant known as Agent Orange and was banned in 1979 for most uses in the United States. When 2,4,5-T is manufactured, it is unavoidably contaminated with minute amounts of dioxin. Dioxin, in doses as low as a few parts per billion, has produced liver and lung diseases, leukemia, miscarriages, birth defects, and even death in laboratory animals. Vietnam veterans and children of Vietnam veterans exposed to Agent Orange have been among the victims.

Auxin is synthesized in apical meristems of shoots. It causes young stems to bend toward light when it migrates toward the darker side, where it makes young cell walls more plastic and thereby promotes cell elongation. By interacting with other hormones, auxin can promote an increase in girth and is involved in growth responses to gravity and fruit ripening.

Cytokinins

Cytokinins comprise another group of naturally occurring growth hormones in plants. Studies by Gottlieb Haberlandt of Austria around 1913 demonstrated the existence of an unknown chemical in various tissues of vascular plants that, in cut potato tubers, would cause parenchyma cells to become meristematic, and would induce the differentiation of a cork cambium. The role of cytokinins, active components of coconut milk, in promoting the differentiation of organs in masses of plant tissue growing in culture later led to their discovery. Subsequent studies have focused on the role cytokinins play in the differentiation of tissues from callus.

A cytokinin is a plant hormone that, in combination with auxin, stimulates cell division and differentiation in plants. Most cytokinins are produced in the root apical meristems and transported throughout the plant. Developing fruits are also important sites of cytokinin synthesis. In mosses, cytokinins cause the formation of vegetative buds on the gametophyte. In all plants, cytokinins, working with other hormones, seem to regulate growth patterns.

All naturally occurring cytokinins are purines that appear to be derivatives of, or have molecule side chains similar to, those of adenine (figure 41.14). Other chemically diverse molecules, not known to occur naturally, have effects similar to those of cytokinins. Cytokinins promote growth of lateral buds into branches (figure 41.15); though, along with auxin and ethylene, they also play a role in apical dominance (the suppression of lateral bud growth). Con-
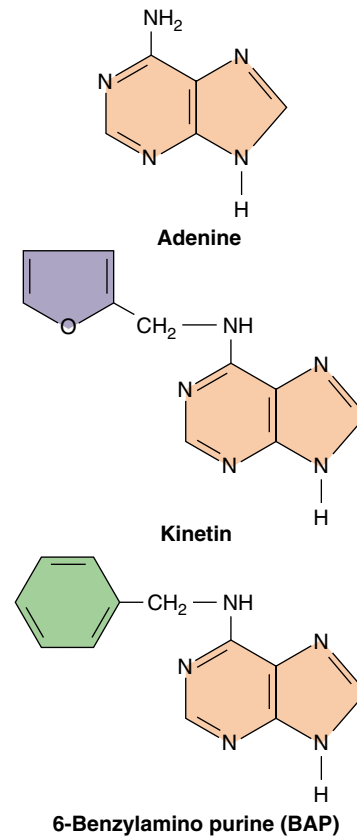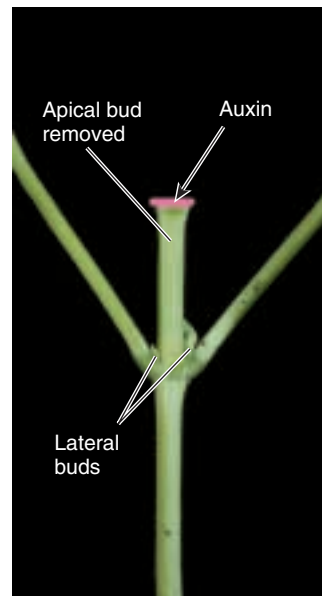

FIGURE 41.14
Some cytokinins. Two commonly used synthetic cytokinins: kinetin and 6-benzylamino purine. Note their resemblance to the purine base adenine.
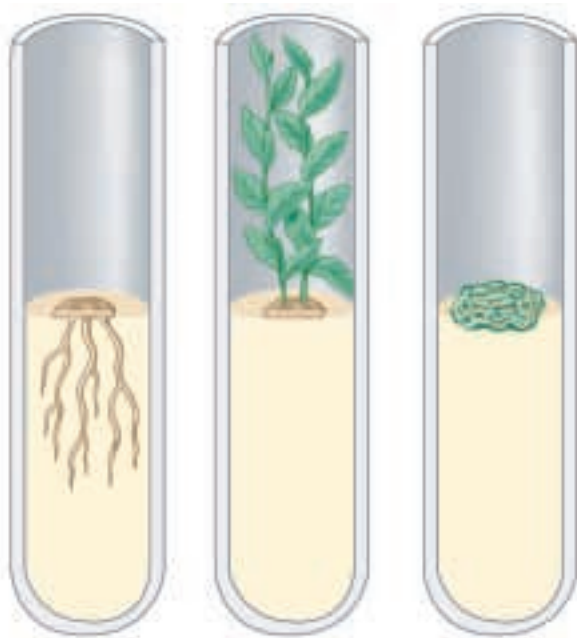

FIGURE 41.15
Cytokinins stimulate lateral bud growth. (*a*) When the apical meristem of a plant is intact, auxin from the apical bud will inhibit the growth of lateral buds. (*b*) When the apical bud is removed, cytokinins are able to produce the growth of lateral buds into branches. (*c*) When the apical bud is removed and auxin is added to the cut surface, axillary bud outgrowth is suppressed.

FIGURE 41.16
Relative amounts of cytokinins and auxin affect organ regeneration in culture. In the case of tobacco, (*a*) high auxin to cytokinin ratios favor root development; (*b*) high cytokinin to auxin ratios favor shoot development; and (*c*) intermediate concentrations result in the formation of undifferentiated cells. These developmental responses to cytokinin/auxin ratios in culture are species specific.

| Auxin: | High | Low | Intermediate |
| Cytokinin: | Low | High | Intermediate |

FIGURE 41.17
Crown gall tumor. Sometimes cytokinins can be used against the plant by a pathogen. In this case *Agrobacterium tumefaciens* (a bacteria) has incorporated a piece of its DNA into the plant genome. This DNA contains genes coding for enzymes necessary for cytokinin and auxin biosynthesis. The increased levels of these hormones in the plant cause massive cell division and the formation of a tumor.

versely, cytokinins inhibit formation of lateral roots, while auxins promote their formation. As a consequence of these relationships, the balance between cytokinins and auxin, along with other factors, determines the appearance of a mature plant. In addition, the application of cytokinins to leaves detached from a plant retards their yellowing. They function as anti-aging hormones.

The action of cytokinins, like that of other hormones, has been studied in terms of its effects on growth and differentiation of masses of tissue growing in defined media. Plant tissue can form shoots, roots, or an undifferentiated mass of tissues, depending on the relative amounts of auxin and cytokinin (figure 41.16). In the early cell-growth experiments coconut "milk" was an essential factor. Eventually, it was discovered that coconut "milk" is not only rich in amino acids and other reduced nitrogen compounds required for growth, but it also contains cytokinins. Cytokinins seem to be essential for mitosis and cell division. They apparently promote the synthesis or activation of proteins that are specifically required for mitosis.

Cytokinins have also been used against plants by pathogens. The bacteria *Agrobacterium*, for example, intro-

duces genes into the plant genome that increase the rate of cytokinin, as well as auxin, production. This causes massive cell division and the formation of a tumor called crown gall (figure 41.17). How these hormone biosynthesis genes ended up in a bacterium is an intriguing evolutionary question. Coevolution does not always work to the plant's advantage.

Cytokinins are plant hormones that, in combination with auxin, stimulate cell division and, along with a number of other factors, determine the course of differentiation. In contrast to auxins, cytokinins are purines that are related to or derived from adenine.

Gibberellins

Gibberellins are named after the fungus *Gibberella fujikuroi*, which causes rice plants, on which it is parasitic, to grow abnormally tall. Japanese plant pathologist Eiichi Kurosawa investigated Bakane ("foolish seedling") disease in the 1920s. He grew *Gibberella* in culture and obtained a substance that, when applied to rice plants, produced bakane. This substance was isolated and the structural formula identified by Japanese chemists in 1939. British chemists reconfirmed the formula in 1954. Although such chemicals were first thought to be only a curiosity, they have since turned out to belong to a large class of more than 100 naturally occurring plant hormones called gibberellins. All are acidic and are usually abbreviated to **GA** (for gibberellic acid), with a different subscript (GA_1, GA_2, and so forth) to distinguish each one. While gibberellins function endogenously as hormones, they also function as pheromones in ferns. In ferns gibberellin-like compounds released from one gametophyte can trigger the development of male reproductive structures on a neighboring gametophyte.

Gibberellins, which are synthesized in the apical portions of stems and roots, have important effects on stem elongation. The elongation effect is enhanced if auxin is also present. The application of gibberellins to dwarf mutants is known to restore the normal growth and development in many plants (figure 41.18). Some dwarf mutants produce insufficient amounts of gibberellin; while others lack the ability to perceive gibberellin. The large number of gibberellins are all part of a complex biosynthetic pathway that has been unraveled using gibberellin-deficient mutants in maize (corn). While many of these gibberellins are intermediate forms in the production of GA_1, recent work shows that different forms may have specific biological roles.

In chapter 41, we noted the role gibberellins stimulate the production of $\propto$-amylase and other hydrolytic enzymes needed for utilization of food resources during germination and establishment of cereal seedlings. How are the genes encoding these enzymes transcribed? Experimental studies in the aleurone layer surrounding the endosperms of cereal grains have shown that transcription occurs when the gibberellins initiate a burst of messenger RNA (mRNA) and protein synthesis. GA somehow enhances DNA binding proteins, which in turn allow DNA transcription of a gene. Synthesis of DNA does not seem to occur during the early stages of seed germination but becomes important when the radicle has grown through the seed coats.

Gibberellins also affect a number of other aspects of plant growth and development. These hormones also hasten seed germination, apparently because they can substitute for the effects of cold or light requirements in this process. Gibberellins are used commercially to space grape

FIGURE 41.18
Effects of gibberellins. This rapid cycling member of the mustard family plant *(Brassica rapa)* will "bolt" and flower because of increased gibberellin levels. Mutants such as the rosette mutant shown here *(left)* are defective in producing gibberellins. They can be rescued by applying gibberellins. Other mutants have been identified that are defective in perceiving gibberellins and they will not respond to gibberellin applications.

FIGURE 41.19
Applications of gibberellins increase the space between grapes. Larger grapes develop because there is more room between individual grapes.

flowers by extending internode length so the fruits have more room to grow (figure 41.19).

> **Gibberellins are an important class of plant hormones that are produced in the apical regions of shoots and roots. They play the major role in controlling stem elongation for most plants, acting in concert with auxin and other hormones.**

Brassinosteroids and Oligosaccharins

Brassinosteroids

Although we've known about brassinosteroids for 30 years, it is only recently that they have claimed their place as a class of plant hormones. They were first discovered in *Brassica* pollen, hence the name. Their historical absence in discussions of hormones may be partially due to their functional overlap with other plant hormones, especially auxins and gibberellins. Additive effects among these three classes have been reported. The application of molecular genetics to the study of brassinosteroids has led to tremendous advances in our understanding of how they are made and, to some extent, how they function in signal transduction pathways. What is particularly intriguing about brassinosteroids are similarities to animal steroid hormones (figure 41.20). One of the genes coding for an enzyme in the brassinosteroid biosynthetic pathway has significant similarity to an enzyme used in the synthesis of testosterone and related steroids. Brassinosteroids have been identified in algae and appear to be quite ubiquitous among the plants. It is plausible that their evolutionary origin predated the plant-animal split.

Brassinosteroids have a broad spectrum of physiological effects—elongation, cell division, bending of stems, vascular tissue development, delayed senescence, membrane polarization, and reproductive development. Environmental signals can trigger brassinosteroid actions. Mutants have been identified that alter the response to brassinosteroid, but signal transduction pathways remain to be uncovered. From an evolutionary perspective, it will be quite interesting to see how these pathways compare with animal steroid signal transduction pathways.

Oligosaccharins

In addition to cellulose, plant cell walls are composed of numerous complex carbohydrates called oligosaccharides. There is some evidence that these cell wall components function as signaling molecules as well as structural wall components. Oligosaccharides that are proposed to have a hormonelike function are called oligosaccharins. Oligosaccharins can be released from the cell wall by enzymes secreted by pathogens. These carbohydrates are believed to signal defense responses, such as the hypersensitive response discussed later in this chapter. Another oligosaccharin has been shown to inhibit auxin-stimulated elongation of pea stems. These molecules are active at concentrations one to two orders of magnitude less than the traditional plant hormones. You have seen how auxin and cytokinin ratios can affect organogenesis in culture. Oligosaccharins also affect the phenotype of regenerated tobacco tissue, inhibiting root formation and stimulating flower production in tissues that are competent to regenerate flowers. How
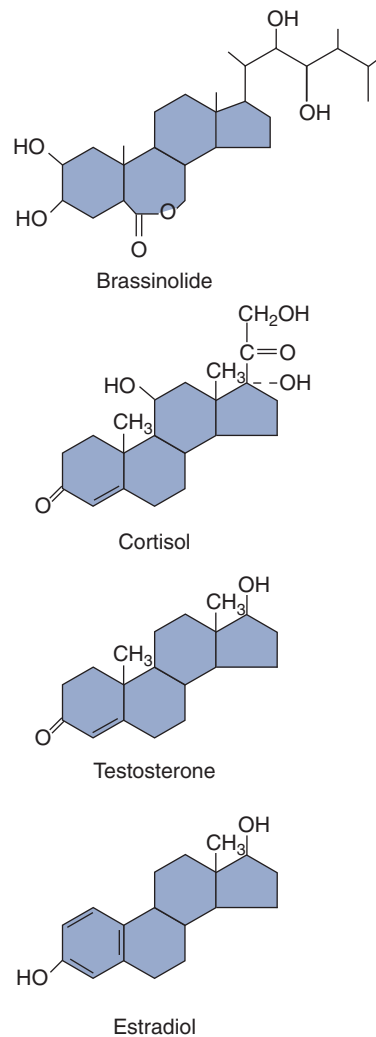

FIGURE 41.20
Brassinosteroids, such as brassinolide, have structural similarities to animal steroid hormones.

the culture results translate to in vivo systems is an open question. The structural biochemistry of oligosaccharins makes them particularly challenging molecules to study. How they interface with cells and initiate signal transduction pathways is an open question.

Brassinosteroids are structurally similar to animal steroid hormones. They have many effects on plant growth and development that parallel those of auxins and gibberellins. Oligosaccharins are complex carbohydrates that are released from cell walls and appear to regulate both pathogen responses and growth and development in some plants.

Ethylene

Long before its role as a plant hormone was appreciated, the simple, gaseous hydrocarbon ethylene ($H_2C=CH_2$) was known to defoliate plants when it leaked from gaslights in streetlamps. Ethylene is, however, a natural product of plant metabolism that, in minute amounts, interacts with other plant hormones. When auxin is transported down from the apical meristem of the stem, it stimulates the production of ethylene in the tissues around the lateral buds and thus retards their growth. Ethylene also suppresses stem and root elongation, probably in a similar way. An ethylene receptor has been identified and characterized. It appears to have evolved early in the evolution of photosynthetic organisms, sharing features with environmental-sensing proteins identified in bacteria.

Ethylene plays a major role in fruit ripening. At first, auxin, which is produced in significant amounts in pollinated flowers and developing fruits, stimulates ethylene production; this, in turn, hastens fruit ripening. Complex carbohydrates are broken down into simple sugars, chlorophylls are broken down, cell walls become soft, and the volatile compounds associated with flavor and scent in ripe fruits are produced.

One of the first observations that led to the recognition of ethylene as a plant hormone was the premature ripening in bananas produced by gases coming from oranges. Such relationships have led to major commercial uses of ethylene. For example, tomatoes are often picked green and artificially ripened later by the application of ethylene. Ethylene is widely used to speed the ripening of lemons and oranges as well. Carbon dioxide has the opposite effect of arresting ripening. Fruits are often shipped in an atmosphere of carbon dioxide. A biotechnology solution has also been developed (figure 41.21). One of the genes necessary for ethylene biosynthesis has been cloned, and its antisense copy has been inserted into the tomato genome. The antisense copy of the gene is a nucleotide sequence that is complementary to the sense copy of the gene. In this transgenic plant, both the sense and antisense sequences for the ethylene biosynthesis gene are transcribed. The sense and antisense mRNA sequences then pair with each other. This blocks translation, which requires single-stranded RNA; ethylene is not synthesized, and the transgenic tomatoes do not ripen. Sturdy green tomatoes can be shipped without ripening and
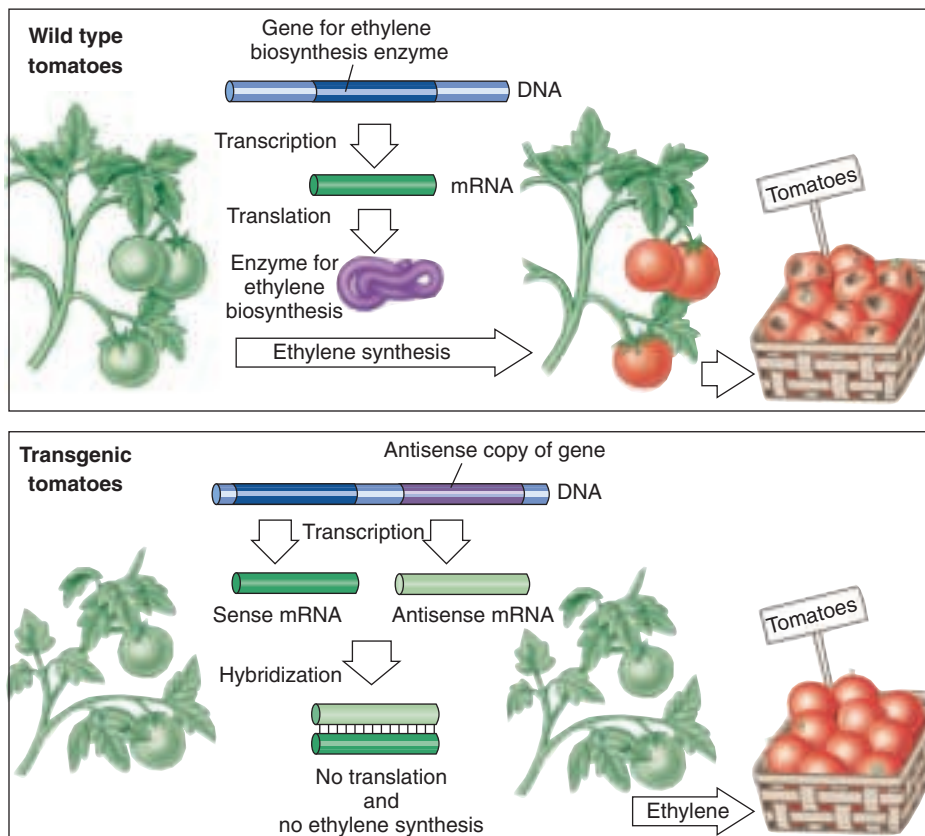

FIGURE 41.21
Genetic regulation of fruit ripening. An antisense copy of the gene for ethylene biosynthesis prevents the formation of ethylene and subsequent ripening of transgenic fruit. The antisense strand is complementary to the sequence for the ethylene biosynthesis gene. After transcription, the antisense mRNA pairs with the sense mRNA, and the double-stranded mRNA cannot be translated into a functional protein. Ethylene is not produced, and the fruit does not ripen. The fruit is sturdier for shipping in its unripened form and can be ripened later with exposure to ethylene. Thus, while wild-type tomatoes may already be rotten and damaged by the time they reach stores, transgenic tomatoes stay fresh longer.

rotting. Exposing these tomatoes to ethylene later will allow them to ripen.

Studies have shown that ethylene plays an important ecological role. Ethylene production increases rapidly when a plant is exposed to ozone and other toxic chemicals, temperature extremes, drought, attack by pathogens or herbivores, and other stresses. The increased production of ethylene that occurs can accelerate the loss of leaves or fruits that have been damaged by these stresses. Some of the damage associated with exposure to ozone is due to the ethylene produced by the plants. The production of ethylene by plants attacked by herbivores or infected with diseases may be a signal to activate the defense mechanisms of the plants. This may include the production of molecules toxic to the pests.

Ethylene, a simple gaseous hydrocarbon, is a naturally occurring plant hormone. Among its numerous effects is the stimulation of ripening in fruit. Ethylene production is also elevated in response to environmental stress.

Abscisic Acid

Abscisic acid, a naturally occurring plant hormone, appears to be synthesized mainly in mature green leaves, fruits, and root caps. The hormone earned its name because applications of it appear to stimulate leaf senescence (aging) and abscission, but there is little evidence that it plays an important role in this process. In fact, it is believed that abscisic acid may cause ethylene synthesis, and that it is actually the ethylene that promotes senescence and abscission. When abscisic acid is applied to a green leaf, the areas of contact turn yellow. Thus, abscisic acid has the exact opposite effect on a leaf from that of the cytokinins; a yellowing leaf will remain green in an area where cytokinins are applied.

Abscisic acid probably induces the formation of winter buds—dormant buds that remain through the winter—by suppressing growth. The conversion of leaf primordia into bud scales follows (figure 41.22a). Like ethylene, it may also suppress growth of dormant lateral buds. It appears that abscisic acid, by suppressing growth and elongation of buds, can counteract some of the effects of gibberellins (which stimulate growth and elongation of buds); it also promotes senescence by counteracting auxin (which tends to retard senescence). Abscisic acid plays a role in seed dormancy and is antagonistic to gibberellins during germination. It is also important in controlling the opening and closing of stomata (figure 41.22b).

Abscisic acid occurs in all groups of plants and apparently has been functioning as a growth-regulating substance since early in the evolution of the plant kingdom. Relatively little is known about the exact nature of its physiological and biochemical effects. These effects are very rapid—often taking place within a minute or two—and therefore they must be at least partly independent of gene expression. Some longer-term effects of abscisic acid involve the regulation of gene expression, but the way this occurs is poorly understood. Abscisic acid levels become greatly elevated when the plant is subject to stress, especially drought. Like other plant hormones, abscisic acid probably will prove to have valuable commercial applications when its mode of action is better understood. It is a particularly strong candidate for understanding desiccation tolerance.

> Abscisic acid, produced chiefly in mature green leaves and in fruits, suppresses growth of buds and promotes leaf senescence. It also plays an important role in controlling the opening and closing of stomata. Abscisic acid may be critical in ensuring survival under environmental stress, especially water stresses.

(a)
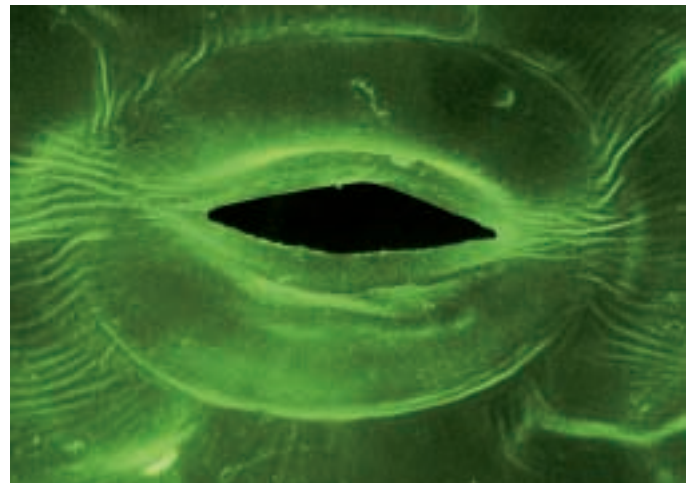
(b)

FIGURE 41.22
Effects of abscisic acid. (*a*) Abscisic acid plays a role in the formation of these winter buds of an American basswood. These buds will remain dormant for the winter, and bud scales—modified leaves—will protect the buds from desiccation.
(*b*) Abscisic acid also affects the closing of stomata by influencing the movement of potassium ions out of guard cells.

Plants Undergo Metamorphosis

Overview of Initiating Flowering

Carefully regulated processes determine when and where flowers will form. Plants must often gain competence to respond to internal or external signals regulating flowering. Once plants are competent to reproduce, a combination of factors including light, temperature, and both promotive and inhibitory internal signals determine when a flower is produced (figure 41.23). These signals turn on genes that specify where the floral organs, sepals, petals, stamens, and carpels will form. Once cells have instructions to become a specific floral organ, yet another developmental cascade leads to the three-dimensional construction of flower parts.

Phase Change

Plants go through developmental changes leading to reproductive maturity just like many animals. This shift from juvenile to adult development is seen in the metamorphosis of a tadpole to an adult frog or caterpillar to a butterfly that can then reproduce. Plants undergo a similar metamorphosis that leads to the production of a flower. Unlike the frog that loses its tail, plants just keep adding on structures to existing structures with their meristems. At germination, most plants are incapable of
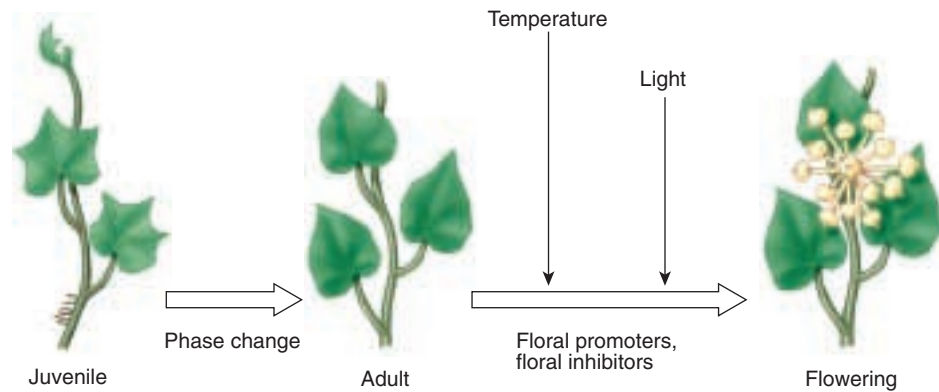
FIGURE 41.23
Factors involved in initiating flowering. This is a model of environmentally cued and internally processed events that result in a shoot meristem initiating flowers.

producing a flower, even if all the environmental cues are optimal. Internal developmental changes allow plants to obtain **competence** to respond to external and/or internal signals that trigger flower formation. This transition is referred to as **phase change**. Phase change can be morphologically obvious or very subtle. Take a look at an oak tree in the winter. The lower leaves will still be clinging to the branches, while the upper ones will be gone (figure 41.24*a*). Those lower branches were initiated by a juvenile meristem. The fact that they did not respond to environmental cues and drop their leaves indicates that they are young branches and have not made a phase change. Ivy also has distinctive juvenile and adult phases of growth (figure 41.24*b*). Stem tissue produced by a juvenile meristem initiates adventitious roots that can cling to walls. If

FIGURE 41.24
Phase change. (*a*) The lower branches of this oak tree represent the juvenile phase of development and cling to their leaves in the winter. The lower leaves are not able to form an abscission layer and break off the tree in the fall. Such visible changes are marks of phase change, but the real test is whether or not the plant is able to flower. (*b*) Juvenile ivy (*left*) makes adventitious roots and has an alternating leaf arrangement. Adult ivy (*right*) cannot make adventitious roots and has leaves with a different morphology that are arranged on an upright stem in a spiral.

you look at very old brick buildings that are covered with ivy, you will notice the uppermost branches are falling off because they have transitioned to the adult phase of growth and have lost the ability to produce adventitious roots. It is important to remember that even though a plant has reached the adult stage of development, it may or may not produce reproductive structures. Other factors may be necessary to trigger flowering.

Generally it is easier to get a plant to revert from an adult to vegetative state than to induce phase change experimentally. Applications of gibberellins and severe pruning can cause reversion. There is evidence in peas and *Arabidopsis* for genetically controlled repression of flowering. The *embryonic flower* mutant of *Arabidopsis* flowers almost immediately (figure 41.25), which is consistent with the hypothesis that the wild-type allele suppresses flowering. It is possible that flowering is the default state and that mechanisms have evolved to delay flowering. This delay allows the plant to store more energy to be allocated for reproduction.

The best example of inducing the juvenile to adult transition comes from the construction of transgenic plants that overexpress a gene necessary for flowering, that is found in many species. This gene, *LEAFY*, was cloned in *Arabidopsis* and its promoter was replaced with a viral promoter that results in constant, high levels of *LEAFY* transcription. This gene construct was then introduced into cultured aspen cells which were used to regenerate plants. When *LEAFY* is overexpressed in aspen, flowering occurs in weeks instead of years (figure 41.26). Phase change requires both sufficient signal and the ability to perceive the signal. Some plants acquire competence in the shoot to perceive a signal of a certain intensity. Others acquire competence to produce sufficient promotive signal(s) and/or decrease inhibitory signal(s).

FIGURE 41.25
In *Arabidopsis*, the *embryonic flower* gene may repress flowering. The *embryonic flower* mutant flowers upon germination.

Plants become reproductively competent through changes in signaling and perception. The transition to the adult stage of development where reproduction is possible is called phase change. Plants in the adult phase of development may or may not produce reproductive structures (flowers), depending on environmental cues.
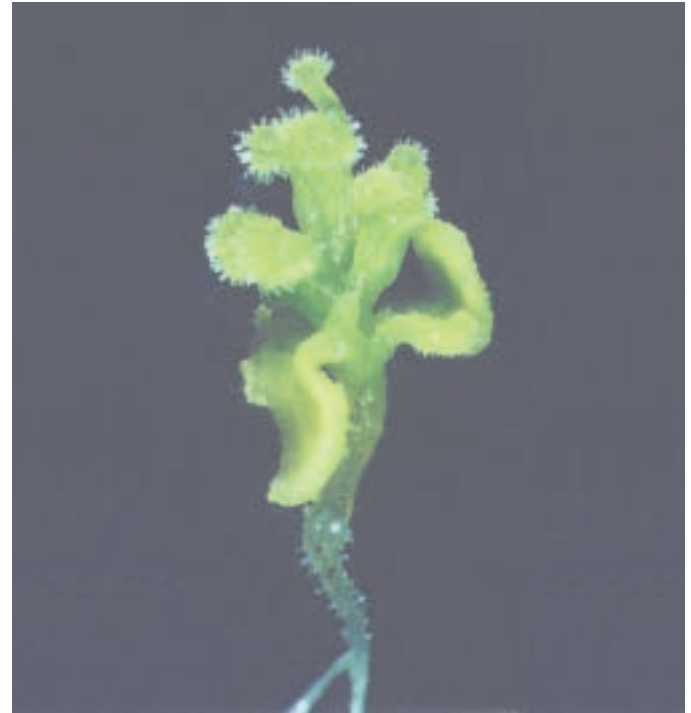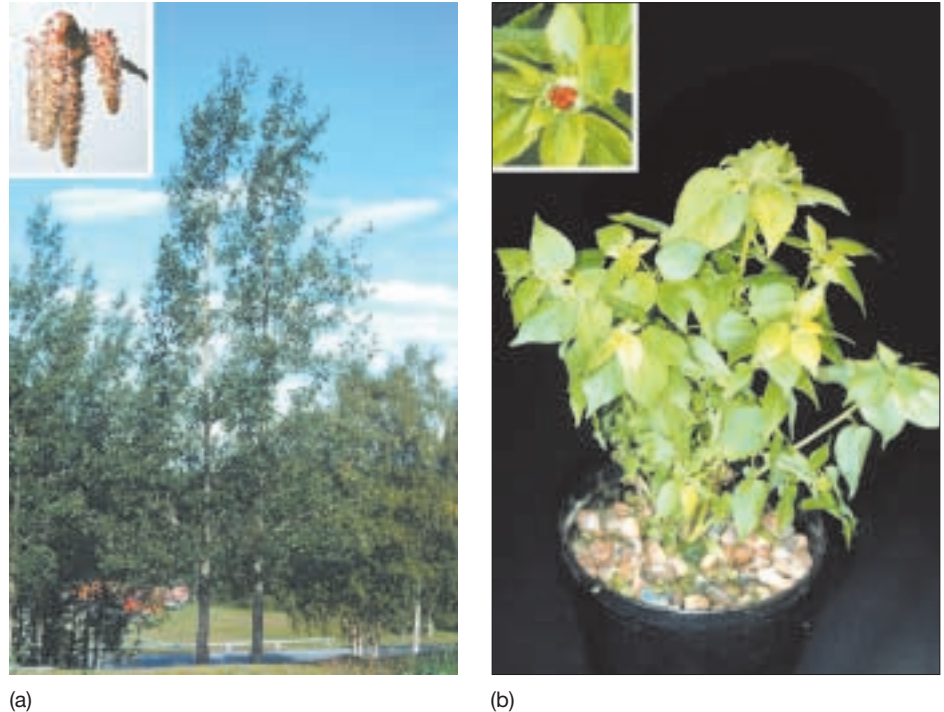

(a)

(b)

FIGURE 41.26
Overexpression of a flowering gene can accelerate phase change. (*a*) An aspen tree normally grows for several years before producing flowers. (*b*) Overexpression of the *Arabidopsis* flowering gene, *LEAFY*, causes rapid flowering in a transgenic aspen.

Pathways Leading to Flower Production

The environment can promote or repress flowering. In some cases, it can be relatively neutral. Light can be a signal that long, summer days have arrived in a temperate climate and conditions are favorable for reproduction. In other cases, plants depend on light to accumulate sufficient amounts of sucrose to fuel reproduction, but flower independently of the length of day. Temperature can also be used as a clue. Gibberellins are important and have been linked to the vernalization pathway. Clearly, reproductive success would be unlikely in the middle of a blizzard. Assuming regulation of reproduction first arose in more constant tropical environments, many of the day length and temperature controls would have evolved as plants colonized more temperate climates. Plants can rely primarily on one pathway, but all three pathways can be present. The complexity of the flowering pathways has been dissected physiologically. Now analysis of flowering mutants is providing insight into the molecular mechanisms of the floral pathways. The redundancy of pathways to flowering ensures that there will be another generation.

Light-Dependent Pathway

Flowering requires much energy accumulated via photosynthesis. Thus, all plants require light for flowering, but this is distinct from the **photoperiodic,** or light-dependent, flowering pathway. Aspects of growth and development in most plants are keyed to changes in the proportion of light to dark in the daily 24-hour cycle (day length). This provides a mechanism for organisms to respond to seasonal changes in the relative length of day and night. Day length changes with the seasons; the farther from the equator, the greater the variation. Flowering responses of plants to day length fall into several basic categories. When the daylight becomes shorter than a critical length, flowering is initiated in **short-day plants** (figure 41.27). When the daylight becomes longer than a critical length, flowering is initiated in **long-day plants.** Other plants, such as snapdragons, roses, and many native to the tropics (for example, tomatoes), will flower when mature regardless of day length, as long as they have received enough light for normal growth. These are referred to as **day-neutral plants.** Several grasses (for example, Indian grass, *Sorghastrum nutans*), as well as ivy, have two critical
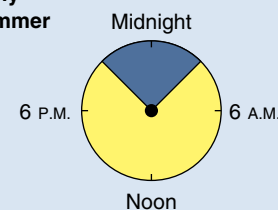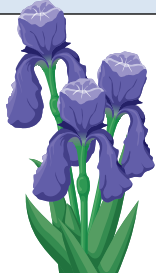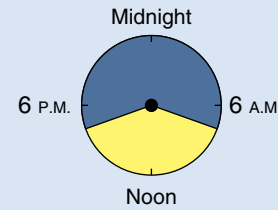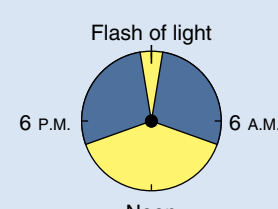

FIGURE 41.27
How flowering responds to day length.
(*a*) This iris is a long-day plant that is stimulated by short nights to flower in the spring. The goldenrod is a short-day plant that, throughout its natural distribution in the northern hemisphere, is stimulated by long nights to flower in the fall. (*b*) If the long night of winter is artificially interrupted by a flash of light, the goldenrod will not flower, and the iris will. In each case, it is the duration of uninterrupted darkness that determines when flowering will occur.

photoperiods; they will not flower if the days are too long, and they also will not flower if the days are too short. In some species, there is a sharp distinction between long and short days. In others, flowering occurs more rapidly or slowly depending on the length of day. These plants rely on other flowering pathways as well and are called facultative long- or short-day plants. The garden pea is an example of a facultative long-day plant. In all of these plants, it is actually the length of darkness (night), not the length of day, that is physiologically significant. Using light as a cue permits plants to flower when abiotic environmental conditions are optimal, pollinators are available, and competition for resources with other plants may be less. For example, the spring ephemerals flower in the woods before the canopy leafs out, blocking sunlight necessary for photosynthesis.

At middle latitudes, most long-day plants flower in the spring and early summer; examples of such plants include clover, irises, lettuce, spinach, and hollyhocks. Short-day plants usually flower in late summer and fall, and include chrysanthemums, goldenrods, poinsettias, soybeans, and many weeds. Commercial plant growers use these responses to day length to bring plants into flower at specific times. For example, photoperiod is manipulated in greenhouses so poinsettias flower just in time for the winter holidays (figure 41.28). The geographic distribution of certain plants may be determined by flowering responses to day length.

Photoperiod is perceived by several different forms of phytochrome and also a blue-light-sensitive molecule (cryptochrome). The conformational change in a light receptor molecule triggers a cascade of events that leads to the production of a flower. There is a link between light and the circadian rhythm regulated by an internal clock that facilitates or inhibits flowering. At a molecular level the gaps between light signaling and production of flowers are rapidly filling in and the control mechanisms are quite complex. Here is one example of how day length affects a specific flowering gene in *Arabidopsis*, a facultative long-day plant that flowers in response to both far-red and blue light. Red light inhibits flowering. The gene *CONSTANS* (*CO*) is expressed under long days but not short days. The loss of *CO* product does not alter when a plant flowers under short days, but delays flowering under long days. What happens is that the gene is positively regulated by cryptochrome that perceives blue light under long days. Cryptochrome appears to inhibit the inhibition of flowering by phytochrome B exposed to red light. Simply put, flowering is promoted by repressing a gene that represses flowering! *CO* is a transcription factor that turns on other genes which results in the expression of *LEAFY*. As discussed in the section on phase change, *LEAFY* is one of the key genes that "tells" a meristem to switch over to flowering. We will see that other pathways also converge on this important gene.

FIGURE 41.28
Manipulation of photoperiod in greenhouses ensures that short-day poinsettias flower in time for the winter holidays. Note that the colorful "petals" are actually sepals. Even after flowering is induced, there are many developmental events leading to the production of species-specific flowers.

The Flowering Hormone: Does It Exist? The Holy Grail in plant biology has been a flowering hormone, quested unsuccessfully for more than 50 years. A considerable amount of evidence demonstrates the existence of substances that promote flowering and substances that inhibit it. Grafting experiments have shown that these substances can move from leaves to shoots. The complexity of their interactions, as well as the fact that multiple chemical messengers are evidently involved, has made this scientifically and commercially interesting search very difficult, and to this day, the existence of a flowering hormone remains strictly hypothetical. We do know that *LEAFY* can be expressed in the vegetative as well as the reproductive portions of plants. Clearly, information about day length gathered by leaves is transmitted to shoot apices. Given that there are multiple pathways to flowering, several signals may be facilitating communication between leaves and shoots. We also know that roots can be a source of floral inhibitors affecting shoot development.

Temperature-Dependent Pathway

Lysenko solved the problem of winter wheat seed rotting in the fields in Russia by chilling the seeds and planting them in the spring. Winter wheat would not flower without a period of chilling, called **vernalization.** Unfortunately a great many problems, including mistreatment of Russian geneticists, resulted from this scientifically significant discovery. Lysenko erroneously concluded that he had converted one species, winter wheat, to another, spring wheat, by simply altering the environment. There was a shift from science to politics. Genetics and Darwinian evolution were suspect for half a century. Social history aside, the valuable lesson here is that cold temperatures can accelerate or permit flowering in many species. As with light, this ensures that plants flower at more optimal times.

Vernalization may be necessary for seeds or plants in later stages of development. Analysis of mutants in *Arabidopsis* and pea indicate that vernalization is a separate flowering pathway that may be linked to the hormone gibberellin. In this pathway, repression may also lead to flowering. High levels of one of the genes in the pathway may block the promotion of flowering by gibberellins. When plants are chilled, there is less of this gene product and gibberellin activity may increase. It is known that gibberellins enhance the expression of *LEAFY*. One proposal is that both the vernalization and autonomous pathways share a common intersection affecting gibberellin promotion of flowering. Weigel has shown that gibberellin actually binds the promoter of the *LEAFY* gene, so its effect on flowering is direct. The connection between gibberellin levels and temperature also needs to be understood.

Autonomous Pathway

The autonomous pathway to flowering is independent of external cues except for basic nutrition. Presumably this was the first pathway to evolve. Day-neutral plants often depend primarily on the autonomous pathway which allows plants to "count" and "remember." A field of day-neutral tobacco will produce a uniform number of nodes before flowering. If the shoots of these plants are removed at different positions, axillary buds will grow out and produce the same number of nodes as the removed portion of the shoot (figure 41.29). At a certain point in development shoots become committed or **determined** to flower (figure 41.30). The upper axillary buds of flowering tobacco will remember their position when rooted or grafted. The terminal shoot tip becomes florally determined about four nodes before it initiates a flower. In some other species, this commitment is less stable and/or occurs later.

How do shoots know where they are and at some point "remember" that information? It is clear that there are inhibitory signals from the roots. If bottomless pots are continuously placed over a growing tobacco plant and filled

(a) Upper axillary bud removed

(b) Lower axillary bud removed
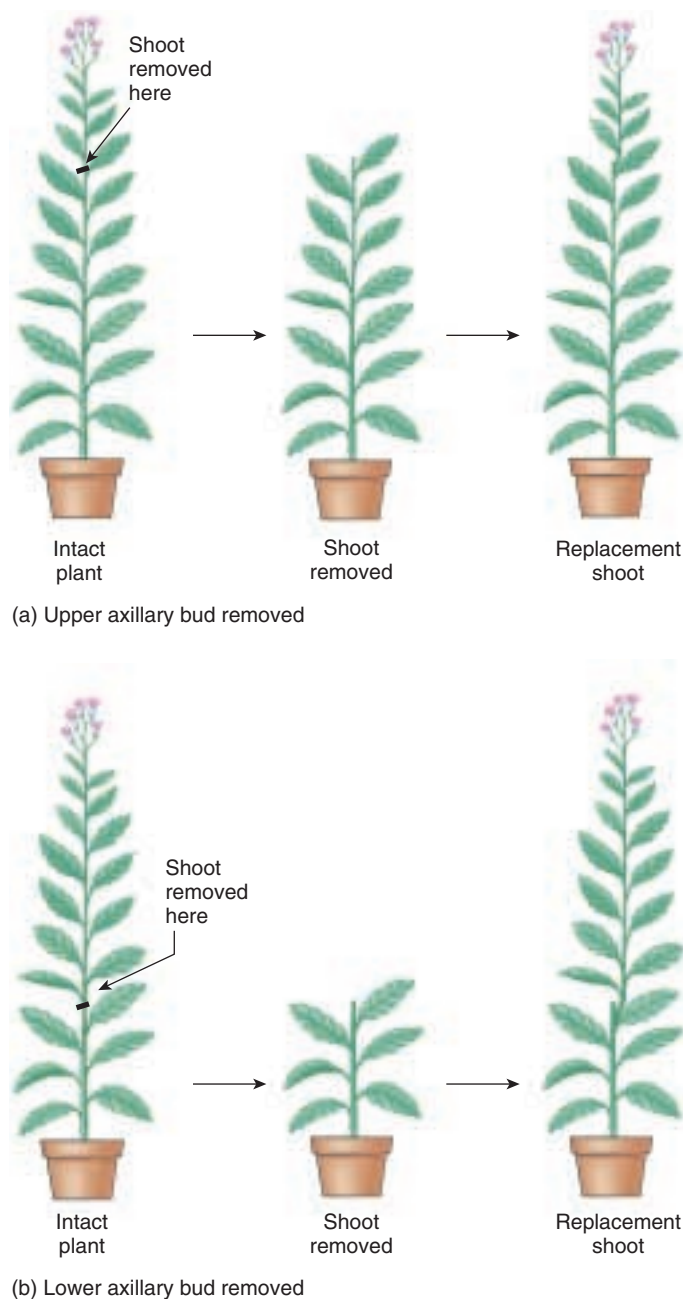
FIGURE 41.29
Plants can count. When axillary buds of flowering tobacco plants are released from apical dominance by removing the main shoot, they replace the number of nodes that were initiated by the main shoot. (After McDaniel 1996.)

with soil, flowering is delayed by the formation of adventitious roots (figure 41.31). Control experiments with leaf removal show that it is the addition of roots and not the loss of leaves that delays flowering. A balance between floral promoting and inhibiting signals may regulate when flowering occurs in the autonomous pathway and the other pathways as well.

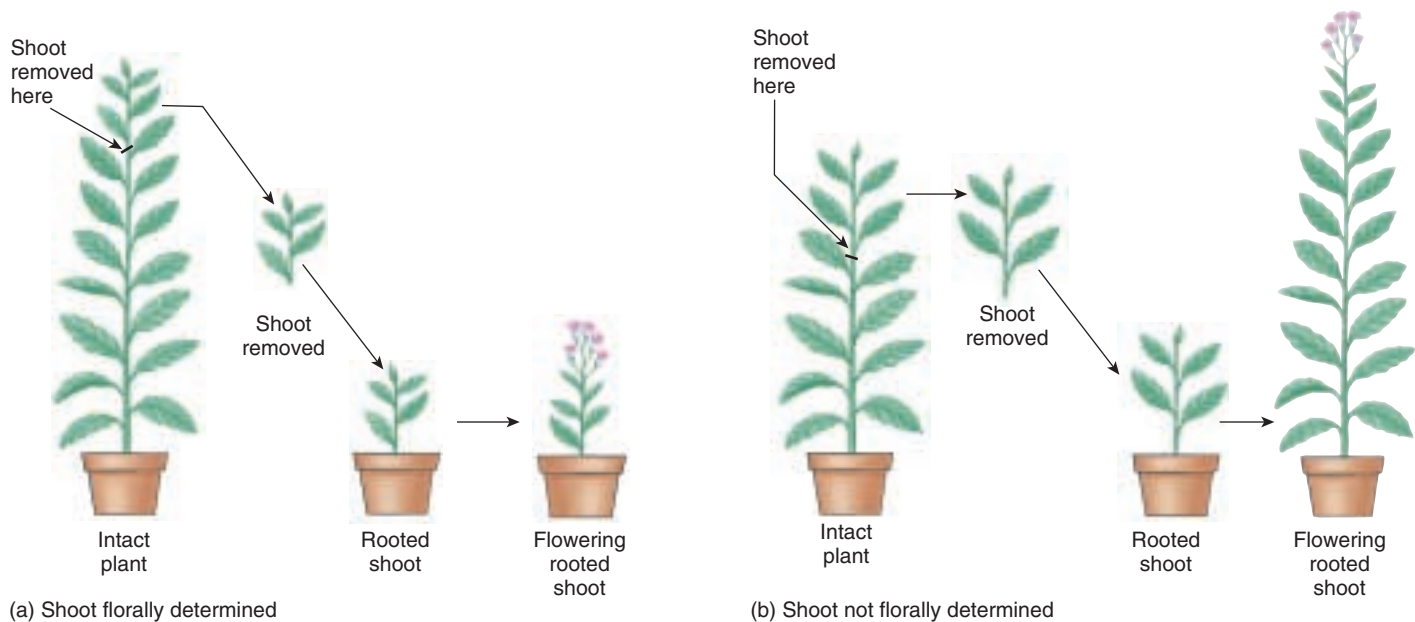(a) Shoot florally determined

(b) Shoot not florally determined

FIGURE 41.30
Plants can remember. At a certain point in the flowering process, shoots become committed to making a flower. This is called floral determination. (*a*) Florally determined axillary buds "remember" their position when rooted in a pot. That is, they produce the same number of nodes that they would have if they had grown out on the plant, and then they flower. (*b*) Those that are not yet florally determined cannot remember how many nodes they have left, so they start counting again. That is, they develop like a seedling and then flower. (After McDaniel 1996.)

Determination for flowering is tested at the organ or whole plant level by changing the environment and ascertaining whether or not the fate has changed. How does floral determination correlate with molecular level changes? In *Arabidopsis*, floral determination correlates with the increase of *LEAFY* gene expression and has occurred by the time a second flowering gene, *APETALA1*, is expressed. Because all three flowering pathways appear to converge with increased levels of *LEAFY*, this determination event should occur in species with a variety of balances among the pathways.

> **Plants use light receptor molecules to measure the length of night. This information is then used to signal pathways that promote or inhibit flowering. Light receptors in the leaves trigger events that result in changes in the shoot meristem. Vernalization is the requirement for a period of chilling before a plant can flower. The autonomous pathway leads to flowering independent of environmental cues. Plants integrate information about position in regulating flowering and both promoters and inhibitors of flowering are important.**

Control plants: no treatment   Experimental plants: pot-on-pot treatment   Control plants: Lower leaves were continually removed

FIGURE 41.31
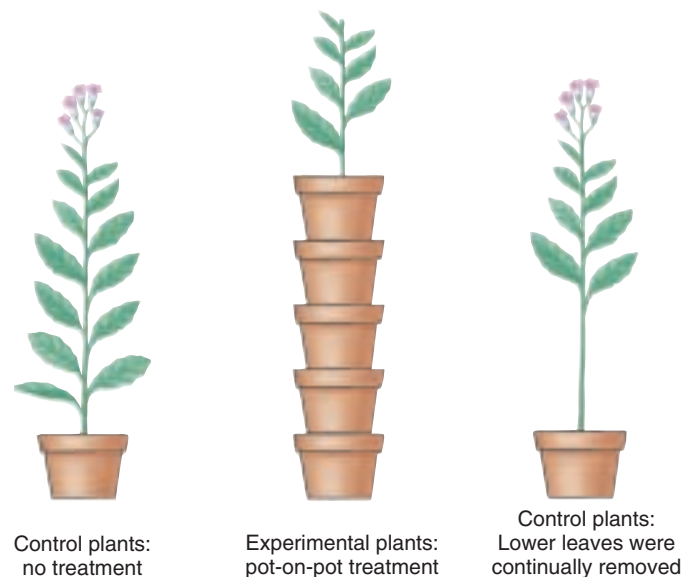**Roots can inhibit flowering.** Adventitious roots formed as bottomless pots were continuously placed over growing tobacco plants, delaying flowering. The delay in flowering is caused by the roots, not the loss of the leaves. This was shown by removing leaves on control plants at the same time and in the same position as leaves on experimental plants that became buried as pots were added.

Identity Genes and the Formation of Floral Meristem and Floral Organs

Arabidopsis and snapdragon are valuable model systems for identifying flowering genes and understanding their interactions. The three pathways, discussed in the previous section, lead to an adult meristem becoming a floral meristem by either activating or repressing the inhibition of floral meristem identity genes (figure 41.32). Two of the key floral meristem identity genes are *LEAFY* and *APETALA1*. These genes establish the meristem as a flower meristem. They then turn on floral organ identity genes. The floral organ identity genes define four concentric whorls moving inward in the floral meristem as sepal, petal, stamen, and carpel. Meyerowitz and Coen proposed a model, called the ABC model, to explain how three classes of floral organ identity genes could specify four distinct organ types (figure 41.33). The ABC model proposes that three classes of organ identity genes (*A, B,* and *C*) specify the floral organs in the four floral whorls. By studying mutants the researchers have determined the following:

1. Class *A* genes alone specify the sepals.
2. Class *A* and class *B* genes together specify the petals.
3. Class *B* and class *C* genes together specify the stamens.
4. Class *C* genes alone specify the carpels.

The beauty of their ABC model is that it is entirely testable by making different combinations of floral organ identity mutants. Each class of genes is expressed in two whorls, yielding four different combinations of the gene products. When any one class is missing, there are aberrant floral organs in predictable positions.

It is important to recognize that this is actually only the beginning of the making of a flower. These organ identity genes are transcription factors that turn on many more genes that will actually give rise to the three-dimensional flower. There are also genes that "paint" the petals. Complex biochemical pathways lead to the accumulation of anthocyanin pigments in vacuoles. These pigments can be orange, red, or purple and the actual color is influenced by pH and by the shape of the petal.

The Formation of Gametes

The ovule within the carpel has origins more ancient than the angiosperms. Floral parts are modified leaves, and within the ovule is the female gametophyte. This next generation develops from placental tissue in the ovary. A megaspore mother cell develops and meiotically gives rise to the embryo sac. Usually two layers of integument tissue form around this embryo sac and will become the seed coat. Genes responsible for the initiation of integuments and also those responsible for the formation of the integument have been identified. Some also affect leaf structure. This chapter has focused on the complex and elegant process that gives rise to the reproductive structure called the flower. It is indeed a metamorphosis, but the subtle shift from mitosis to meiosis in the megaspore mother cell leading to the development of a haploid, gamete-producing gametophyte is perhaps even more critical. The same can be said for pollen formation in the anther of the stamen. As we will see in the next chapter, the flower houses the haploid generations that will produce gametes. The flower also functions to increase the probability that male and female gametes from different (or sometimes the same plant) will unite.

Floral structures form as a result of floral meristem identity genes turning on floral organ identity genes which specify where sepals, petals, stamens, and carpels will form. This is followed by organ development which involves many complex pathways that account for floral diversity among species.
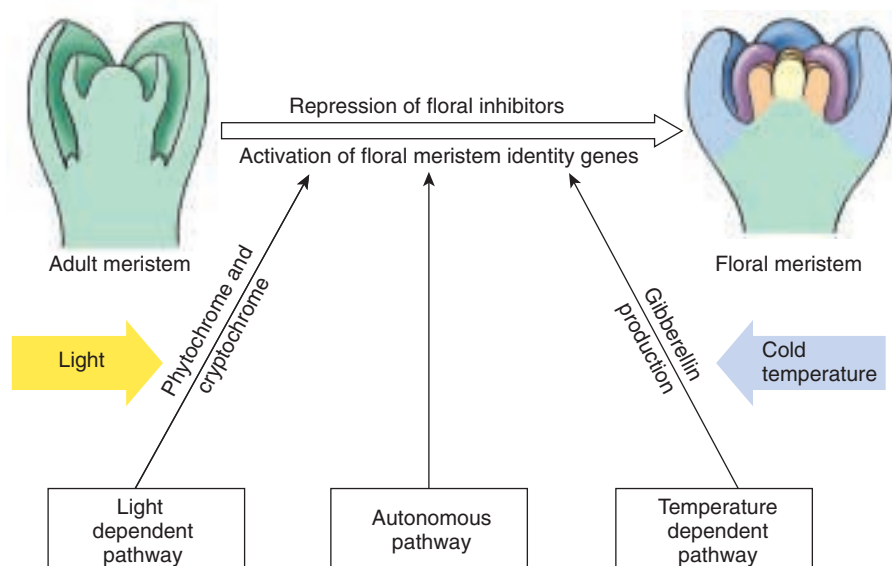

FIGURE 41.32
Model for flowering. The light-dependent, temperature-dependent, and autonomous flowering pathways promote the formation of floral meristems from adult meristems by repressing floral inhibitors and activating floral meristem identity genes.
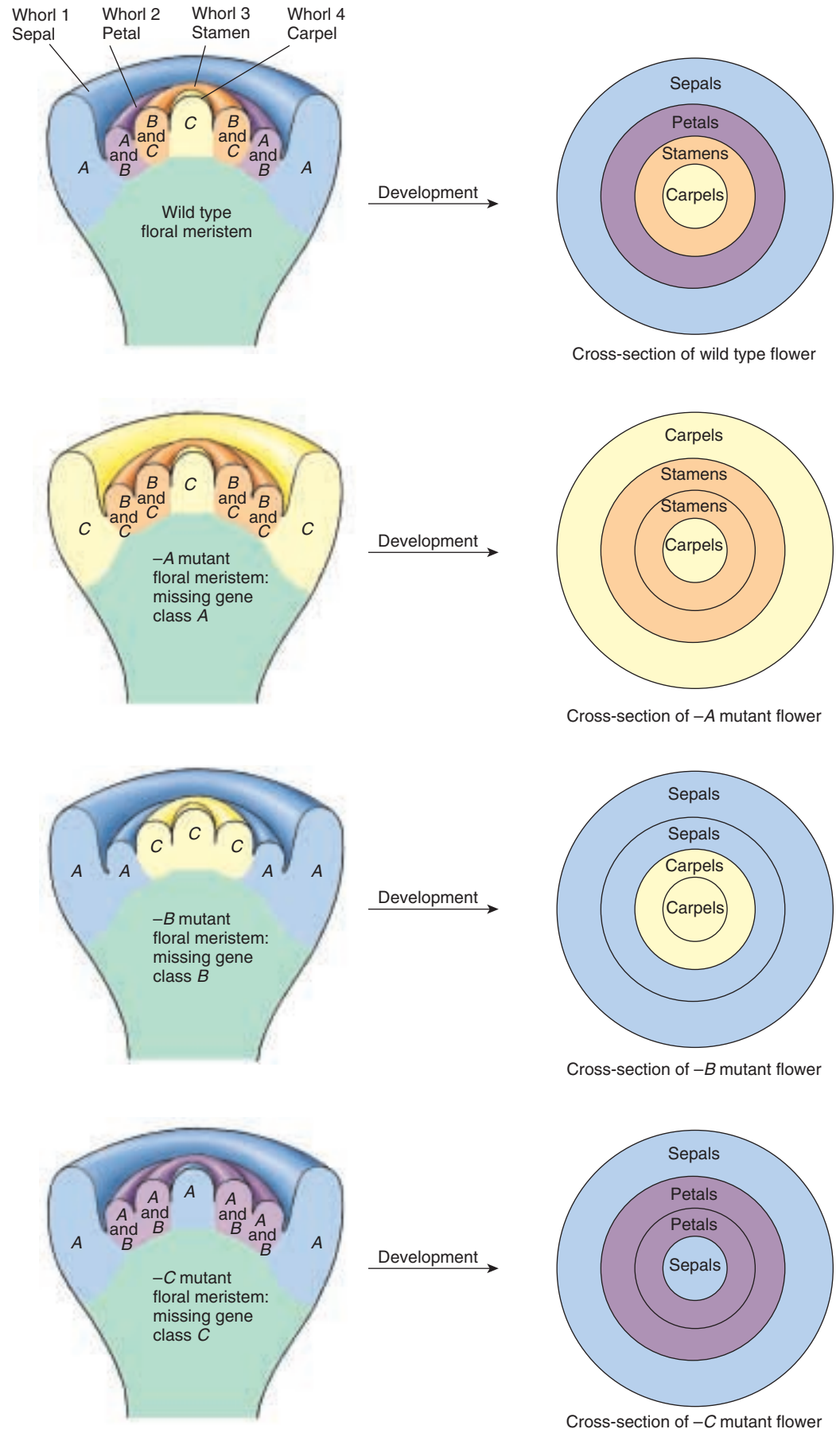
FIGURE 41.33
ABC model for floral organ specification.
Letters labeling whorls indicate which gene classes are active. When *A* function is lost (-*A*), *C* expands to the first and second whorls. When *B* function is lost (-*B*), both outer two whorls have just *A* function, and both inner two whorls have just *C* function; none of the whorls have dual gene function. When *C* function is lost (-*C*), *A* expands into the inner two whorls. These new combinations of gene expression patterns alter which floral structures form in each whorl. (Model proposed by Coen and Meyerowitz, 1991.)

41.4 Many short-term responses to the environment do not require growth.

Larger predators, microbes, water, and wind often present a plant with rapid immediate stress. Response, to be effective, must also be immediate. There is little time for growth, and plants instead invoke a variety of other kinds of responses. Many environmental cues trigger rapid and reversible localized plant movements, for example. The rapid folding of leaves can startle a potential predator. Leaf folding can also prevent water loss by reducing the surface area available for transpiration. Some localized plant movements are triggered by unpredictable environmental signals. Other movements are tied into daily internal rhythms established by cyclic environmental signals like light and temperature. Plants lack a nervous system in the conventional sense. Some of the rapid signaling, however, is the result of electric charge moving through an organ as a wave of membrane ion exchange, not unlike that seen in animals. This is translated into movement by changing the turgor pressure of cells.

Turgor Movement

Turgor is pressure within a living cell resulting from diffusion of water into it. If water leaves turgid cells (ones with turgor pressure), the cells may collapse, causing plant movement; conversely, water entering a limp cell may also cause movement as the cell once more becomes turgid.

Many other plants, including those of the legume family (Fabaceae), exhibit leaf movements in response to touch or other stimuli. After exposure to a stimulus, the changes in leaf orientation are mostly associated with rapid turgor pressure changes in **pulvini** (singular: pulvinus), which are multicellular swellings located at the base of each leaf or leaflet. When leaves with pulvini, such as those of the sensitive plant *(Mimosa pudica)*, are stimulated by wind, heat, touch, or, in some instances, intense light, an electrical signal is generated. The electrical signal is translated into a chemical signal, with potassium ions, followed by water, migrating from the cells in one half of a pulvinus to the intercellular spaces in the other half. The loss of turgor in half of the pulvinus causes the leaf to "fold." The movements of the leaves and leaflets of the sensitive plant are especially rapid; the folding occurs within a second or two after the leaves are touched (figure 41.34). Over a span of about 15 to 30 minutes after the leaves and leaflets have folded, water usually diffuses back into the same cells from which it left, and the leaf returns to its original position.

(a)   Pulvinus   Vascular tissue

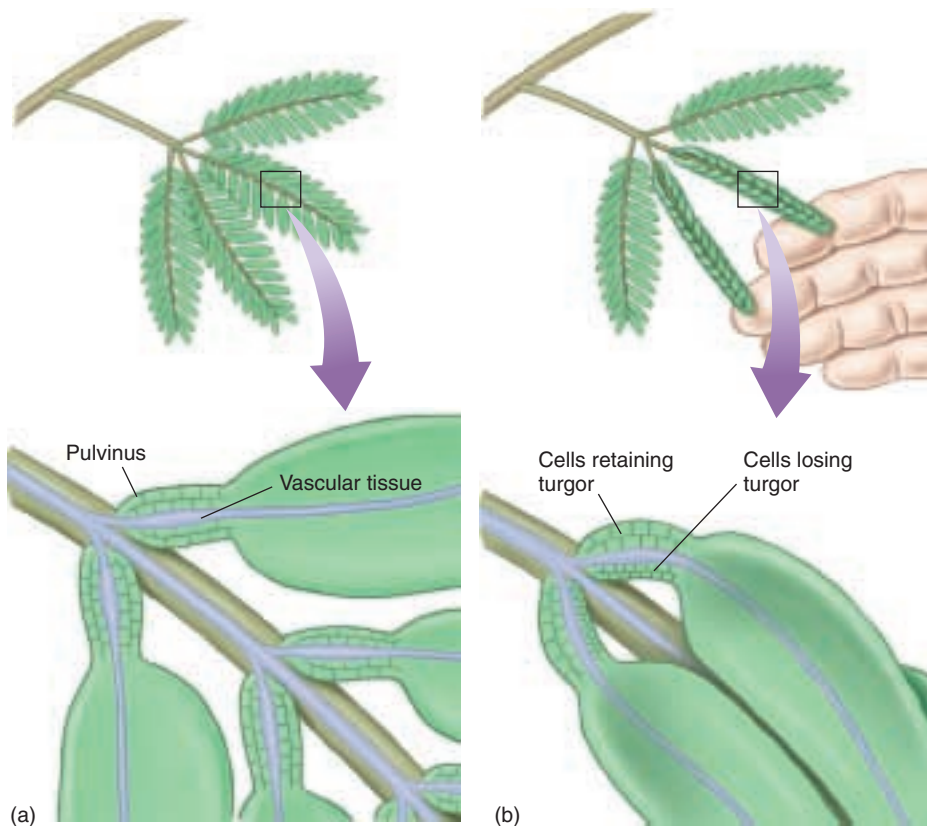(b)   Cells retaining turgor   Cells losing turgor

FIGURE 41.34
Sensitive plant *(Mimosa pudica)*. (*a*) The blades of *Mimosa* leaves are divided into numerous leaflets; at the base of each leaflet is a swollen structure called a pulvinus. (*b*) Changes in turgor cause leaflets to fold in response to a stimulus. (*c*) When leaves are touched (center two leaves above), they fold due to loss of turgor.
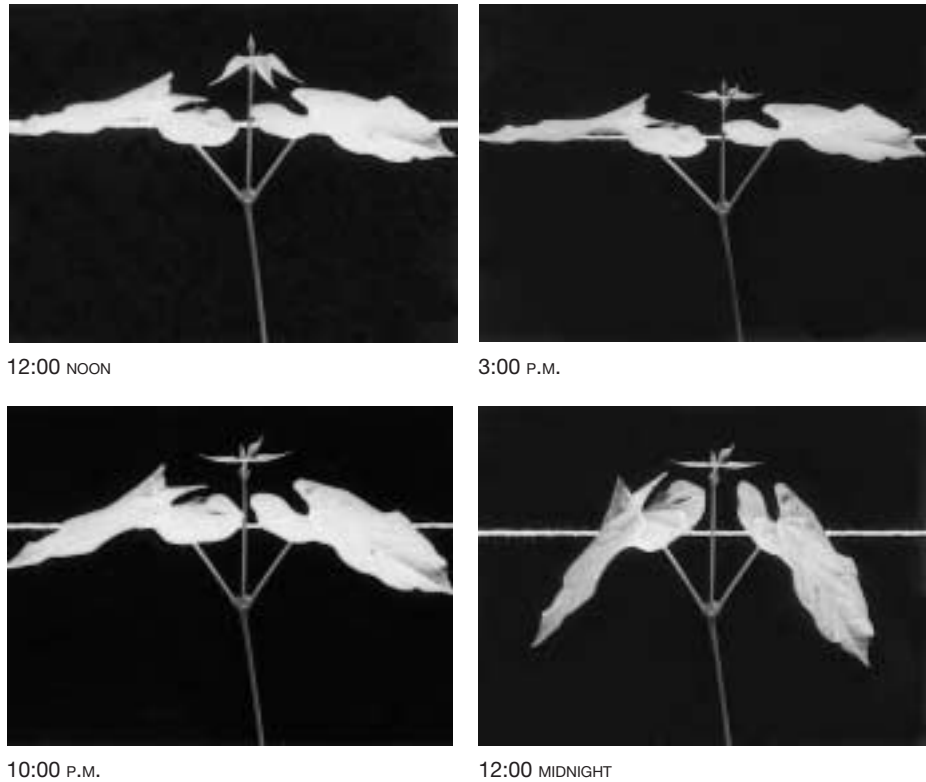
12:00 NOON

3:00 P.M.

10:00 P.M.

12:00 MIDNIGHT

FIGURE 41.35
Sleep movements in bean leaves. In the bean plant, leaf blades are oriented horizontally during the day and vertically at night.

The leaves of some plants with similar mechanisms may track the sun, with their blades oriented at right angles to it; how their orientation is directed is, however, poorly understood. Such leaves can move quite rapidly (as much as 15° an hour). This movement maximizes photosynthesis and is analogous to solar panels that are designed to track the sun.

Some of the most familiar of these reversible changes are seen in leaves and flowers that "open" during the day and "close" at night. For example, the flowers of four o'clocks open at 4 P.M. and evening primrose petals open at night. The blades of plant leaves that exhibit such a daily shift in position may not actually fold; instead, their orientation may be changed as a result of **turgor movements.** Bean leaves are horizontal during the day when their pulvini are turgid, but become more or less vertical at night as the pulvini lose turgor (figure 41.35). These sleep movements reduce water loss from transpiration during the night, but maximize photosynthetic surface area during the day. In these cases, the movement is closely tied to an internal rhythm.

Circadian Clocks

How do leaves know when to "sleep"? They have endogenous circadian clocks that set a rhythm with a period of about 24 hours (actually it is closer to 22 or 23 hours). While there are shorter and much longer, naturally occur-

ring rhythms, circadian rhythms are particularly common and widespread because of the day-night cycle on earth. Jean de Mairan, a French astronomer, first identified circadian rhythms in 1729. He studied the sensitive plant which, in addition to having a touch response, closes its leaflets and leaves at night like the bean plant described above. When de Mairan put the plants in total darkness, they continued "sleeping" and "waking" just as they did when exposed to night and day. This is one of four characteristics of a circadian rhythm: it must continue to run in the absence of external inputs. It must be about 24 hours in duration and can be reset or entrained. (Perhaps you've experienced entrainment when traveling to a different time zone in the form of jet-lag recovery.) The fourth characteristic is that the clock can compensate for differences in temperature. This is quite unique when you consider what you know about biochemical reactions; most rates of reactions vary significantly based on temperature. Circadian clocks exist in many organisms and appear to have evolved independently multiple times. The mechanism behind the clock is not fully understood, but is being actively investigated at the molecular level.

Turgor movements of plants are reversible and involve changes in the turgor pressure of specific cells. Circadian clocks are endogenous timekeepers that keep plant movements and other responses synchronized with the environment.

Plant Defense Responses

Interactions between plants and other organisms can be symbiotic (for example, nitrogen-fixing bacteria and mycorrhizae) or pathogenic. In evolutionary terms, these two types of interactions may simply be opposite sides of the same coin. The interactions have many common aspects and are the result of coevolution between two species that signal and respond to each other. In the case of pathogens, the microbe or pest is "winning," at least for that second in evolutionary time. In chapter 38, we discussed surface barriers the plant constructs to block invasion. In this section, we will focus on cellular level responses to attacks by microbes and animals.

Recognizing the Invader

Half a century ago, Flor proposed that there is a plant resistance gene (*R*) whose product interacts with that of a pathogen avirulence gene (*avr*). This is called the gene-for-gene model and several pairs of *avr* and *R* genes have been cloned in different species pathogenized by microbes, fungi, and insects, in one case. This has been motivated partially by the agronomic benefit of identifying genes that can be added to protect other plants from invaders. Much is now known about the signal transduction pathways that follow the recognition of the pathogen by the *R* gene. These pathways lead to the triggering of the hypersensitive response (HR) which leads to rapid cell death around the source of the invasion and also a longer-term resistance (figure 41.36). There is not always a gene-for-gene response, but plants still have defense responses to pathogens and also mechanical wounding. Some of the response pathways may be similar. Also, oligosaccharins in the cell walls may serve as recognition and signaling molecules.

While our focus is on invaders outside the plant kingdom, more is being learned about how parasitic plants invade other plants. There are specific molecules released from the root hairs of the host that the parasitic plant recognizes and responds to with invasive action. Less is known about the host response and so far the different defense genes that are activated appear to be ineffective.

Responding to the Invader

When a plant is attacked and there is gene-for-gene recognition, the HR response leads to very rapid cell death around the site of attack. This seals off the wounded tissue to prevent the pathogen or pest from moving into the rest of the plant. Hydrogen peroxide and nitric oxide are produced and may signal a cascade of biochemical events resulting in the localized death of host cells. They may also have negative effects on the pathogen, although antioxidant abilities have coevolved in pathogens. Other antimicrobial agents produced include the phytoalexins which are chemical defense agents. A variety of pathogenesis-related genes (*PR* genes) are expressed and their proteins can either function as antimicrobial agents or signals for other events that protect the plant.

In the case of virulent invaders (no *R* gene recognition), there are changes in local cell walls that at least partially block the movement of the pathogen or pest farther into the plant. In this case there is not an HR response and the local plant cells are not suicidal.

When an insect takes a mouthful of a leaf, defense responses are also triggered. Mechanical damage causes responses that have some similar components, but the reaction may be slower. Biochemically, it is distinct from some of the events triggered by signals in the insect's mouth. Such responses are collectively called wound responses. Wound responses are a challenge in designing other types of experiments with plants that involve cutting or otherwise mechanically damaging the tissue. It is important to run control experiments to be sure you are answering your question and not observing a wound response.

Preparing for Future Attacks

In addition to the HR or other local responses, plants are capable of a systemic response to a pathogen or pest attack. This is called a systemic acquired response (SAR). Several pathways lead to broad-ranging resistance that lasts for a period of days. The signals that induce SAR include salicylic acid and jasmonic acid. Salicylic acid is the active ingredient in aspirin too! SAR allows the plant to respond more quickly if it is attacked again. However, this is not the same as the human immune response where antibodies (proteins) that recognize specific antigens (foreign proteins) persist in the body. SAR is neither as specific or long lasting.

Plants defend themselves from invasion in ways reminiscent of the animal immune system. When an invader is recognized, localized cell death seals off the infected area.
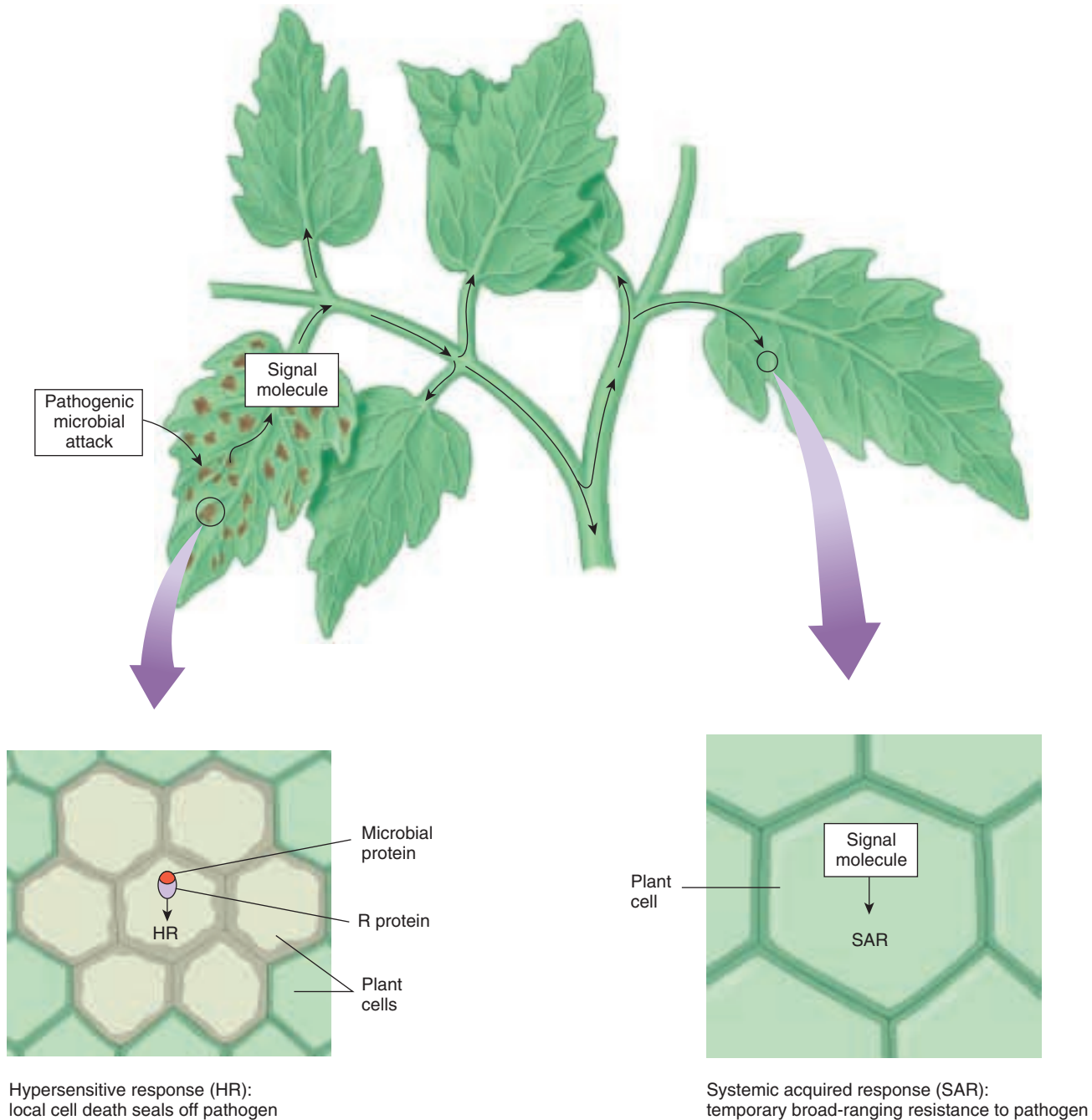
Signal
molecule

Pathogenic
microbial
attack

Microbial
protein

R protein

HR

Plant
cells

Hypersensitive response (HR):
local cell death seals off pathogen

Plant
cell

Signal
molecule

SAR

Systemic acquired response (SAR):
temporary broad-ranging resistance to pathogen

FIGURE 41.36
Plant defense responses. In the gene-for-gene response, a cascade of events is triggered leading to local cell death (HR response) and longer-term resistance in the rest of the plant (SAR).

Summary	Questions	Media Resources

41.1 Plant growth is often guided by environmental cues.

- Tropisms in plants are growth responses to external stimuli, such as light, gravity, or contact.
- Dormancy is a plant adaptation that carries a plant through unfavorable seasons or periods of drought.

1. In general, which part of a plant is positively phototropic? What is the adaptive significance of this reaction?

- Photoperiod

41.2 The hormones that guide growth are keyed to the environment.

- Auxin migrates away from light and promotes the elongation of plant cells on the dark side, causing stems to bend in the direction of light.
- Cytokinins are necessary for mitosis and cell division in plants. They promote growth of lateral buds and inhibit formation of lateral roots.
- Gibberellins, along with auxin, play a major role in stem elongation in most plants. They also tend to hasten the germination of seeds and to break dormancy in buds.

2. How does auxin affect the plasticity of the plant cell walls?

3. Where are most cytokinins produced? From what biomolecule do cytokinins appear to be derived?

4. What plant hormones could be lacking in genetically dwarfed plants?

- Hormones

- Student Research: Plant Growth

41.3 The environment influences flowering.

- The transition of a shoot meristem from vegetative to adult development is called phase change. During phase change, plants gain competence to produce a floral signal(s) and or perceive a signal.
- The light-dependent pathway uses information from light receptor molecules integrated with a biological clock to determine if the length of night is sufficient for flowering.
- The autonomous path functions independently of environmental cues. Internal floral inhibitor(s) from roots and leaves and floral promoter(s) from leaves move through the plant.

5. A plant has undergone phase change. Although it is an adult, it does not flower. How might you get this plant to flower?

6. You have recently moved from Canada to Mexico and brought some seeds from your favorite plants. They germinate and produce beautiful leaves, but never flower. What went wrong?

- Student Research: Selection in Flowering Plants

41.4 Many short-term responses to the environment do not require growth.

- Changes in turgor pressure reflect responses to environmental signals that can protect plants from predation.
- Other reversible movements in plants are caused by changes in turgor pressure that are regulated by internal circadian rhythms.
- Plants have the ability to recognize and respond to invaders through cellular level recognition and response.

7. How are motor cells involved in the function of the pulvinus? What happens in the motor cells of the sensitive plant (*Mimosa pudica*) when its leaves are touched?

8. In what ways can a plant protect itself from pathogenic microbes? From animals?