Leaves

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Introduction

Leaves, the lateral organs of the shoot, serve multiple functions in plants. Their primary task is photosynthesis: harvesting light energy and combining it, in the form of high-energy electrons, with carbon dioxide gas taken from the atmosphere. The carbon is reduced and the high-energy electrons in the resulting sugars are the source of the vast majority of chemical energy used by the biosphere. Photosynthesis requires that leaves be able to effectively harvest light and CO_2. Light passes directly through the epidermis and can penetrate effectively into the leaf interior. Indeed, internal leaf morphology serves to maximize the penetration and harvesting of light. CO_2, however, enters the leaf by diffusing through stomatal pores (refer to ► Chap. 9, Epidermis), and stomatal opening is also the cause of water loss from the leaf via transpiration. The competing demands of maximizing carbon uptake while minimizing water usage, combined with a plant's habitat, have provided strong selective pressures during terrestrial plant evolution. The result of 480 million years of land plant evolution has been an extremely wide range of leaf anatomical forms and adaptations.

## 12.1   Leaves Have a Variety of Shapes and Functions

While leaves can take on many different forms and functions (see below), a typical foliage leaf has a more or less flattened form with two obvious surfaces, which is to say it is **bifacial**. The upper (adaxial or toward the shoot axis) and lower (abaxial or away from the shoot axis) surfaces of the leaf are easily distinguished (◘ Fig. 12.1a, b). In most plants, leaves are dorsiventral in their internal structure, meaning there is a different internal tissue morphology between the adaxial direction and the abaxial direction (◘ Fig. 12.1c). This contrasts with organs with a cylindrical axis such as stems and roots which are radially symmetrical. The main part of the leaf is its **lamina** (or leaf blade) to which the above characteristics belong. In addition, leaves often have a **petiole** and a pair of **stipules**. A petiole attaches the lamina to the stem. Stipules are usually paired flaps or projections of tissue found beneath the petiole, if present at all.

In terms of leaf variants, a **frond** is a large, usually compound, deeply divided leaf with individual leaflets. The name is commonly applied to the leaves of ferns (◘ Fig. 12.1d) and palms, although much reduced monocots in the family Lemnaceae (duckweeds, watermeal, and mud midgets) are also said to have (actually be almost entirely composed of) fronds (refer to ◘ Fig. 11.1h, i). Conifers have flattened and elongated leaves: scalelike in the junipers and cedars and needlelike in the spruces and pines (◘ Fig. 12.1e). Many grasses have a leaf with a long and stiff leaf sheath that connects the base of the plant to the leaves. Lacking a true stem, the individual **leaf sheaths** wrap around and form the stalk. A leaf blade extends from the top of the sheath (◘ Fig. 11.2i). The "trunk" of a 10-meter tall banana (*Musa*) "tree" is a **pseudostem** composed entirely of leaf sheaths that originate at the soil surface and terminate in the leaf blade proper.
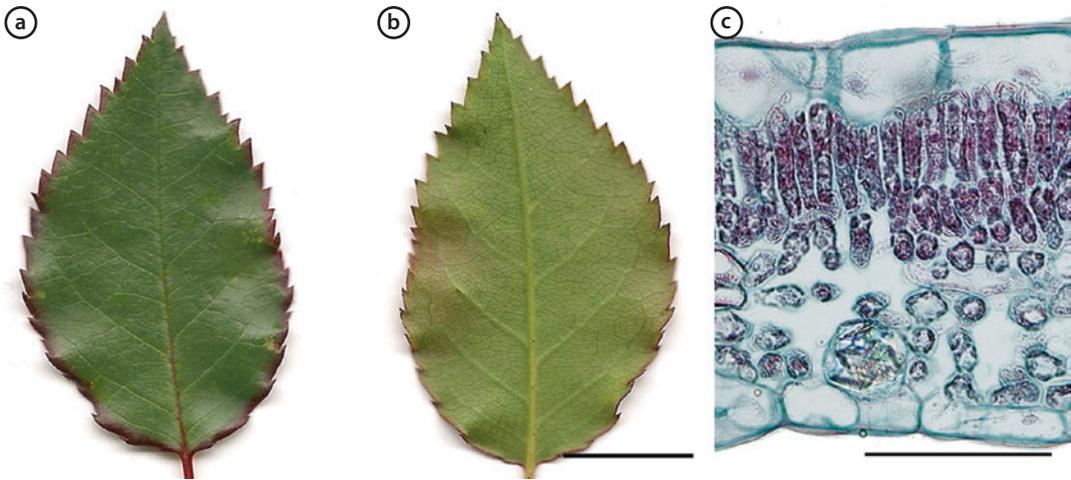
◘ Fig. 12.1   A typical foliage leaf, i.e., macrophyll, from a rose plant (*Rosa* sp.) showing **a** the adaxial surface, **b** the abaxial surface with ridges formed by the major veins, and **c** a cross-section with the adaxis toward the top. A portion of the leaf petiole is visible at the bottoms of **a** and **b**. Scale bar = 1 cm in **a** and **b**, 50 μm in **c** (**a**–**c** RR Wise)
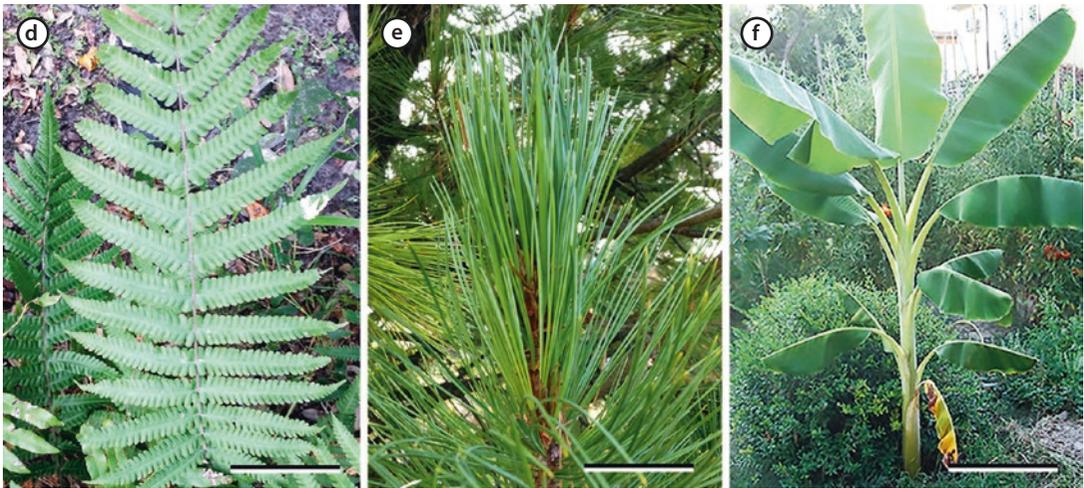
◘ Fig. 12.1   Leaves of **d** southern wood fern (*Dryopteris ludoviciana*), **e** Swiss pine (*Pinus cembra*), and **f** banana (*Musa* sp.)*. The entire "trunk" of the banana "tree" is composed of tightly wrapped leaf sheaths. Scale bars = 10 cm in **d**, 5 cm in **e**, and 1 m in **f** (**d**, **e** RR Wise; **f** I Lorenzini, CC BY-SA 3.0)

Box 12.1   Modern Plant Science Research Relies on Plant Anatomical Studies

Rice (*Oryza sativa*) is one of the most heavily cultivated crop plants in the world. Achieving maximum production and yield relies on a host of conditions that serve to increase water efficiency, nutrient uptake, carbon partitioning, and light interception. Leaf angle plays a large role in the latter. Rice leaves have a long leaf sheath that encircles the stem and

leads to a leaf blade that extends free of the stem. The greater the angle between the leaf blade and the stem, the more the leaf blade extends away from the stem, increasing light absorption but shading neighboring plants. The lesser the leaf angle, the more erect the leaf blade, the less light is absorbed, and the more light is available to neighboring plants. The leaf blade attaches to the leaf sheath at the lamina joint, the shape of which dictates the leaf angle. Therefore, understanding the anatomy and developmental control of the lamina joint can yield insights into leaf erectness, photosynthesis efficiency, and grain yield. Zhou et al. (2017) investigated the anatomical development of the rice lamina joint. They found that leaf angle was determined by the patterns and location of cell divisions and expansions, cell wall thickenings, and programmed cell death at the abaxial or adaxial sides of the joint. A wide range of genes involved in cell division, cell growth, hormone signaling, transcription factors, and signaling protein kinases were expressed in distinct spatial patterns. Their detailed study of the rice lamina joint, as it relates to crop-level productivity, combined anatomical observations with molecular studies and highlights the intersection of traditional plant anatomy approaches with cutting-edge molecular techniques.
Reference: Zhou et al. (2017).

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Modified leaves have a variety of functions. **Cataphylls** are modified leaves that serve a function other than photosynthesis. **Cotyledons** which store food reserves in the eudicotyledon seed and the **scutellum** of grass seeds are also considered to be a form of a cataphyll. The functions of both are discussed in ▶ Chap. 19. Bud scales protect overwintering buds (◘ Fig. 12.1g) and are a form of cataphyll as well. Clematis (*Clematis* sp.) has compound leaves with each leaflet attached to a central rachis. The rachis acts as a tendril (◘ Fig. 12.1h) that wraps around any available substrate to achieve effective climbing. The traps on the carnivorous pitcher plant are also modified tubular leaves (◘ Fig. 12.1i). Cacti have thick photosynthetic stems and modified leaves called spines that serve as effective antiherbivory defenses (◘ Fig. 12.1j). Onion bulb scales (◘ Fig. 12.1k) are modified leaves that store food reserves.

Finally, it should be pointed out that some plants lack leaves entirely. Parasitic plants such as dodder (*Cuscuta* sp., ◘ Fig. 14.7a) and love vine (*Cassytha filiformis*, ◘ Fig. 14.7b, c) derive all their nutrition from their host plant and have no need for photosynthesis, not even in their stems, which lack chlorophyll and are yellow.
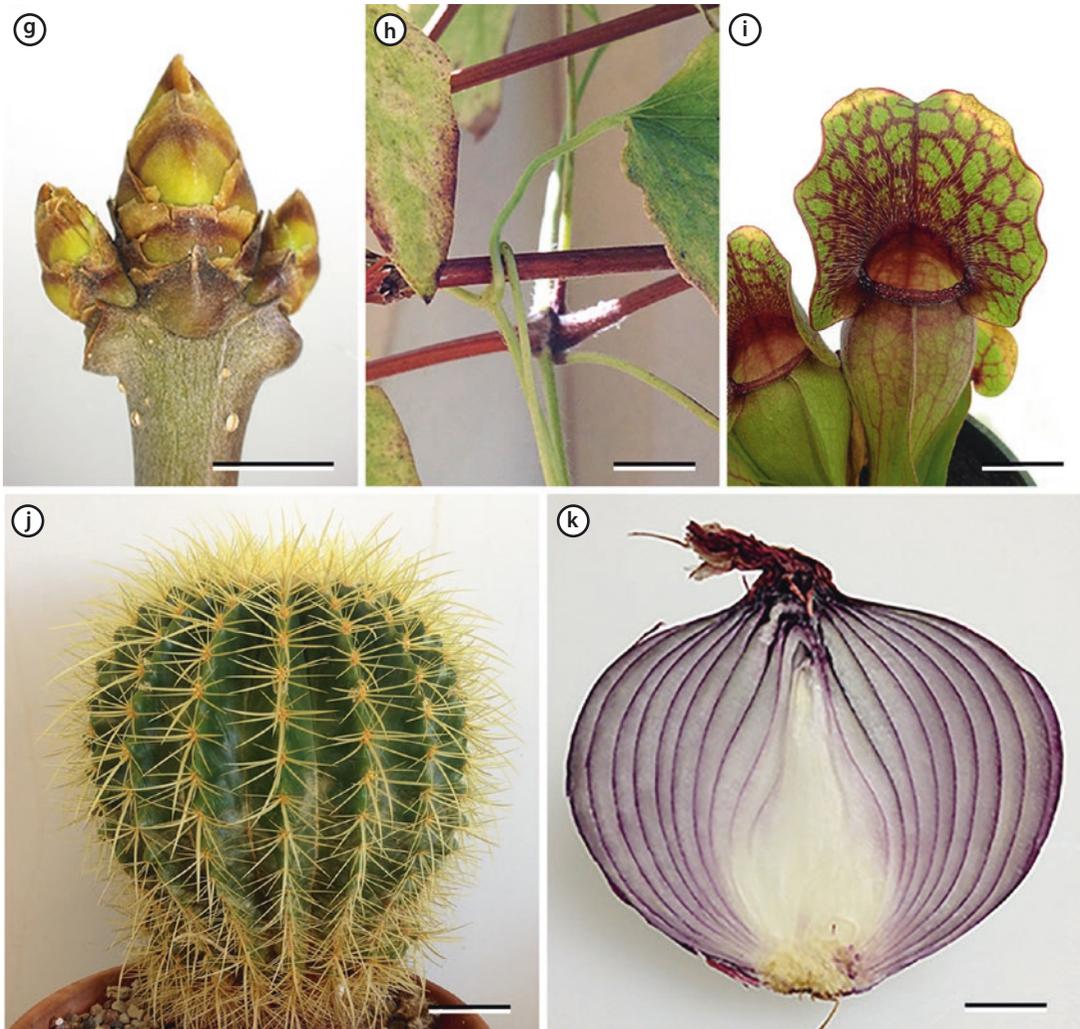
◘ Fig. 12.1   Modified leaves. **g** Cataphylls (bud scales) surrounding three terminal buds on a lilac (*Syringa* sp.) stem, **h** clematis (*Clematis* sp.) tendrils, **i** an insect-trapping pitcher on *Sarracenia* sp., **j** golden barrel cactus (*Echinocactus grusonii*) stem covered with spines, **k** an onion (*Allium* sp.) bulb (stem and leaves) cut in radial section to show individual bulb scales. Scale bars = 1 cm in **g**, 0.5 cm in **h**, 1 cm in **i**, 3 cm in **j**, and 2 cm in **k** (**g**–**k** RR Wise)

12.2   The Vascular System Spreads Throughout the Leaf

Leaf evolution reflects increased complexity of venation, the arrangement of **veins** in a leaf. One hypothesis on the origin of leaves with considerable, but not unanimous, support poses that the progenitors to leaves were small, planar outgrowths of a photosynthetic stem called **enations**. Enations lack a vascular connection to the stem and may be found on stems of the extant, primitive fern *Psilotum nudum* (◘ Fig. 12.2a). Subsequently, a vascular strand emerged from the stele and entered the enation resulting in a microphyll, a leaf with a single, unbranched vascular strand as may
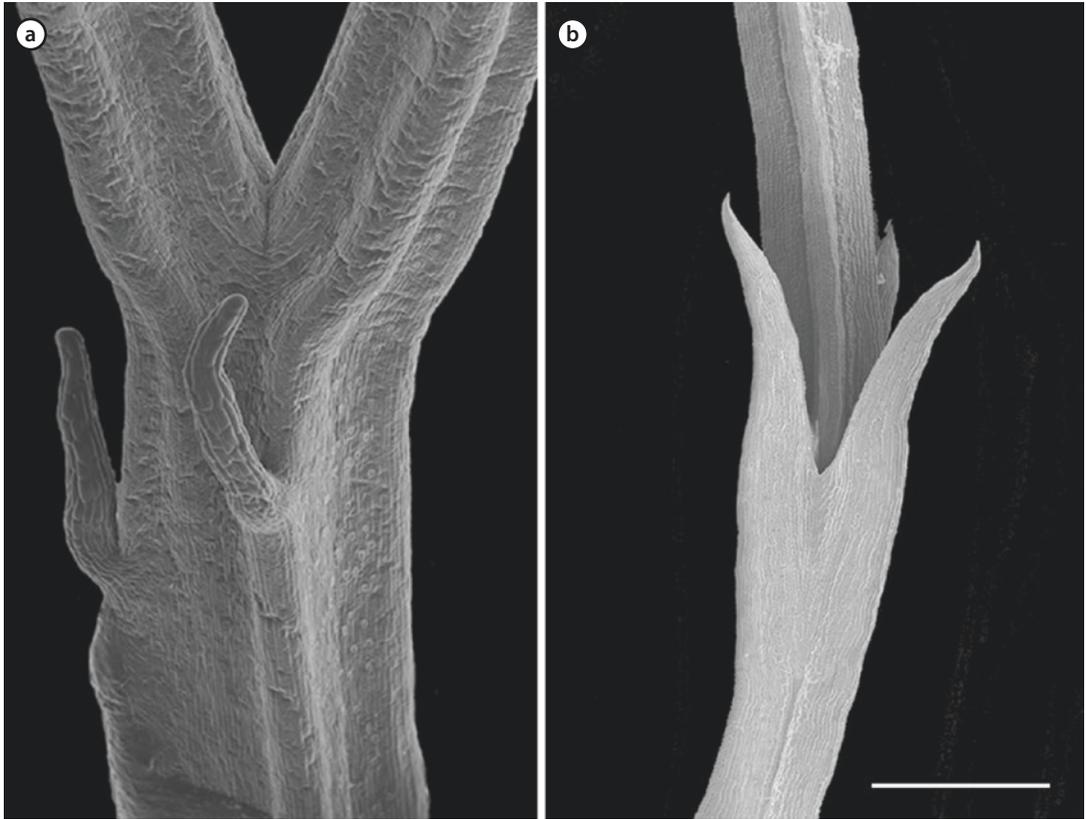
■ **Fig. 12.2   a**, **b** Leaf evolution. **a** Two enations on a skeleton fork fern (*Psilotum nudum*) stem, **b** two microphylls on a horsetail (*Equisetum* sp.) vegetative stem. Although it cannot be seen in these scanning electron micrographs, the *Psilotum* enations lack vasculature, and the *Equisetum* microphylls have a single, unbranched vascular strand. Scale bar = 1 mm and applies to both panels (**a**, **b** RR Wise)

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be seen in many modern-day ferns (■ Fig. 12.2b). Finally, over the course of time, the vascular system branched out forming the complex pattern of vascular angiosperm leaf blades or megaphylls (■ Fig. 12.1c–g).

The vascular system of leaves consists of veins which branch and form a continuous network extending throughout the leaf and passing into the stem. Each vein contains both xylem and phloem tissue; the two tissues are always found together. There are several types of venation, but the most common patterns are the **dichotomous venation** pattern in ferns and ginkgo, **parallel venation** found in the monocots, and the **reticulate venation** of most eudicot leaves.

Leaves of many pteridophytes and the maidenhair tree (*Ginkgo biloba*) have dichotomous venation (■ Fig. 12.2c, d). From an evolutionary perspective, such venation pattern is considered to be ancestral. Each vein branches dichotomously into two others of equal size, and they in turn divide in the same way resulting in a dense venation pattern, all of the same size order. Veins lack anastomoses and end blindly in the mesophyll or approach the leaf margin.
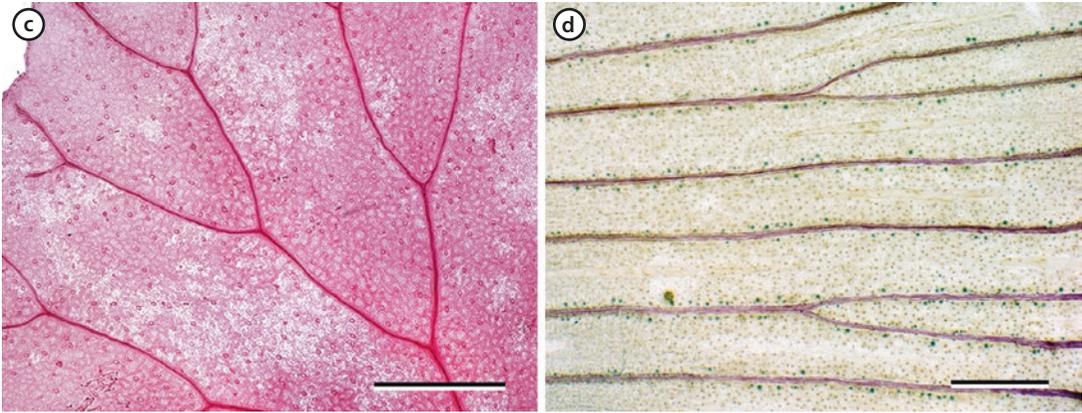
◻ **Fig. 12.2** **c**, **d** Clearings showing dichotomous leaf venation in **c** the sensitive fern (*Onoclea sensibilis*) and **d** ginkgo (*Ginkgo biloba*). The leaves were treated with bleach to remove pigments and then stained with safranin to reveal the vasculature. The mesophyll tissue is intact. Scale bars = 0.5 mm in **c** and 1 mm in **d**. (**c**, **d** RR Wise; specimens prepared by J.D. Curtis)

Ginkgo is a primitive gymnosperm that is now widely cultivated (◻ Fig. 1.16c), but virtually unknown in the wild state. The leaves are deciduous and can be highly variable—even on the same tree. The order, Ginkgoales, was most abundant during the Jurassic period (approx. 200 million years ago) when it was found almost worldwide. *Ginkgo biloba* is believed to be the oldest existing tree specimen. It became nearly extinct with few specimens left in western China until it was rediscovered in China and Japan during the late seventeenth century and subsequently became cultivated worldwide. The leaves are distinctive due to their lobed appearance and dichotomous branching parallel venation.

Monocot leaves have parallel venation (◻ Fig. 12.2e, f). While some veins are larger, most are of similar size. Careful observation of such leaf lamina shows that veins may indeed branch and converge in places, especially near the base and apex of the blade. It is also noteworthy that, at the microscopic level, a reticulate arrangement may exist with the vascular bundles. The small veins that interconnect the larger parallel veins in grasses are called **commissural veins**. It is believed that an intercellular signal induces the ground meristem cells into progenitor cells which subsequently form the commissural veins.

In the reticulate venation pattern (◻ Fig. 12.2g), the major veins spread out in a somewhat palmate pattern across the leaf. From these, minor veins branch off to join with other minor veins, thus forming a conspicuous network as shown in the inset of ◻ Fig. 12.2g. The network that is formed by minor veins varies in size and shape. Accordingly, the veins subdivide the lamina surface into small areas called **areoles** (sing. = areole) delineated by the thinnest veins. Areoles may contain one or more blind terminal veins (◻ Fig. 12.2g, h) in which the terminal vessel element lacks a perforation plate.

Two main types of veins are recognized according to their size and function, major veins and minor veins. Major veins conduct water and photosynthates and provide mechanical support to the delicate mesophyll. Major veins are typically composed of vascular
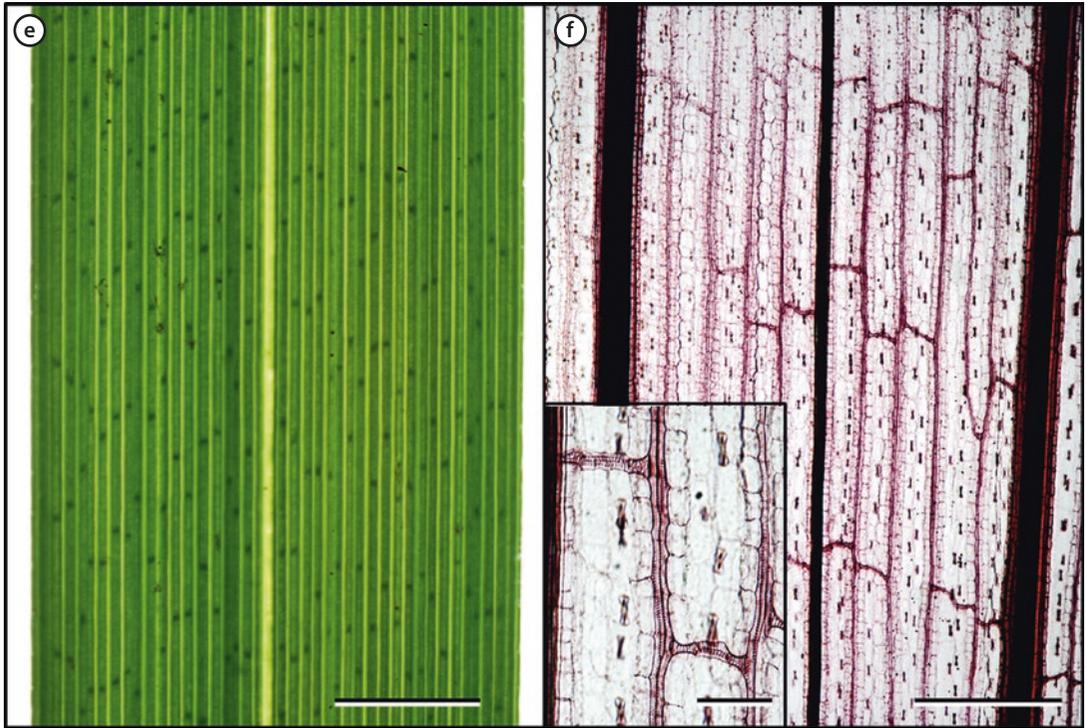
◘ **Fig. 12.2   e**, **f** Parallel venation in monocot leaves. **e** Lemon grass (*Cymbopogon citratus*) seen in macrophotography, **f** a leaf clearing of maize (*Zea mays*). The inset in **f** shows commissural veins connecting the main veins. The rows of red dots are stomata. Scale bars = 5 mm in **e**, 500 µm in **f**, and 100 µm in inset to **f** (**e**, **f** RR Wise)

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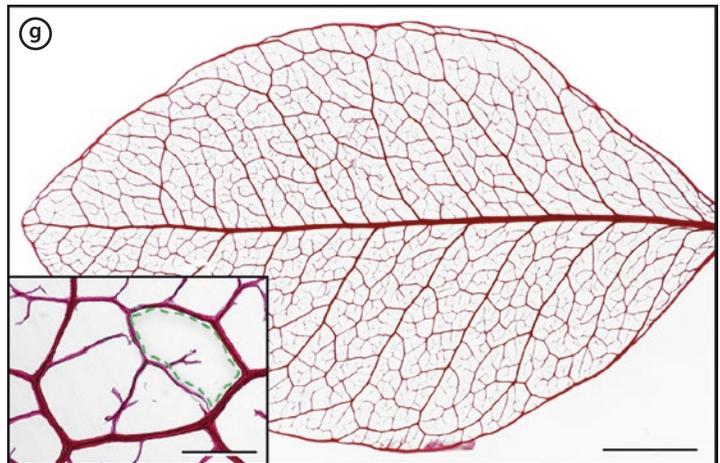
◘ **Fig. 12.2   g** Reticulate venation in a skeletonized leaf of heavenly bamboo (*Nandina domestica*) as shown in a low magnification view with a medium magnification insert. The area in the inset bounded by a green dashed line is a single areole. Note the blind terminal minor veins inside the areoles. The leaf was processed by treating in hot sodium carbonate and using a soft brush to remove the epidermal and mesophyll tissues, leaving only the vasculature. The leaf was then stained in safranin. Scale bar = 1 mm in main figure, 250 µm in inset. (RR Wise; specimen prepared by J.D. Curtis)
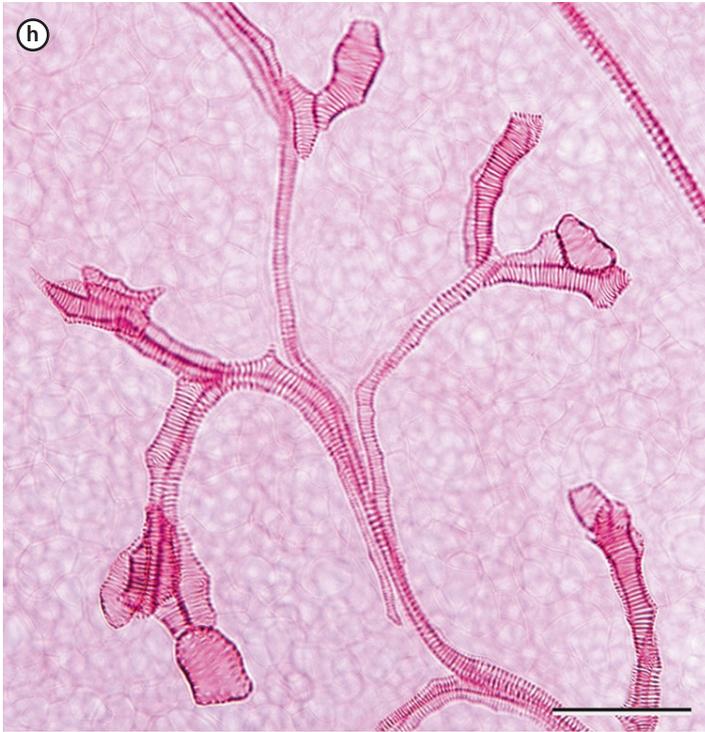
◲ Fig. 12.2   h Blind terminal minor veins as shown in a cleared leaf of crown of thorns (*Euphorbia milii*). Scale bar = 50 μm. (RR Wise; specimen prepared by J.D. Curtis)

bundles, **bundle sheaths**, undifferentiated parenchyma, sclerenchyma, and collenchyma. Most major veins in eudicots usually appear as ribs on the abaxial side of the lamina, and it is the major veins that determine the venation pattern of a leaf (◲ Fig. 12.1b). The major vein in the center of the leaf is called the midrib. By contrast, minor veins are completely enclosed in the mesophyll and not visible from the leaf surface.

The xylem and phloem in the major and minor leaf veins are not in direct contact with mesophyll cells or intercellular air spaces. The vascular tissues are delimited from them by a bundle sheath composed of specialized parenchyma cells. Major leaf veins serve as the main conduit for water and photosynthate. The functions of the minor veins are to distribute the transpiration stream throughout the mesophyll and to load the phloem with photosynthates produced by the chlorenchyma cells in the leaf mesophyll. Accordingly, no mesophyll cell is more than a few cells away from a minor vein. A minor vein consists of a small vascular bundle containing a few phloem sieve tube elements and a few xylem tracheary elements enclosed in a bundle sheath. Accordingly, all photosynthate produced in the mesophyll chlorenchyma must pass through the minor vein bundle sheath to be loaded into a phloem sieve tube element.

Two main types of minor veins are distinguished based on the method by which they take up photosynthate from the mesophyll into the bundle sheath cells. [Note: Phloem loading is a separate process. The photosynthate in the bundle sheath travels to the phloem companion cells and is loaded into the sieve tube elements either symplastically or apoplastically [Refer to ▶ Chap. 8 – Phloem]. **Open veins** have symplastic connections with the leaf mesophyll cells as evidenced by large fields of plasmodesmata in their shared walls (◘ Fig. 12.2i–k). Sugars pass directly from a mesophyll cell to an adjacent bundle sheath cell via the plasmodesmata. Plasmodesmata also connect the bundle sheath cell to the phloem sieve tube member. In contrast, **closed veins** lack plasmodesmatal connections and take up photosynthate apoplastically. In this case, the sugars are exported from the mesophyll cell to the apoplast and then taken up from the apoplast by the bundle sheath cell. A similar process occurs at the bundle sheath cell—phloem sieve tube element boundary. Extensive cell wall ingrowths increase the surface area for uptake and accompany the apoplastic uptake pathway in the bundle sheaths of closed veins (◘ Fig. 12.2l–n).

In eudicot leaves, veins may effectively divide the leaf into areas with separate air spaces. In some instances, the bundle sheath is limited to a single layer of cells surrounding the vascular bundle (◘ Fig. 12.2p, q). This architecture allows for lateral flow of air and water vapor within the leaf, primarily in the spongy mesophyll where most of the air space is found. Leaves with this style of bundle sheath are said to be **homobaric** (homo = the same, baric = atmosphere). Vascular bundle sheaths in the leaves of other plants, however, have vertical rows of clear parenchyma cells extending from the vascular bundle to both leaf surfaces (◘ Fig. 12.2o, q). These **bundle sheath extensions** (BSEs) effectively seal off individual areas of the leaf and result in a **heterobaric** leaf (different atmosphere). In forest foliar strata, heterobaric leaves are typically found in higher strata and homobaric leaves in lower strata. In terms of advantage, heterobaric leaves in the highest part of the canopy have higher light intensity and temperature, whereas homobaric leaves are more common in the lower strata with less light but higher humidity. The bundle sheath also plays specialized roles in C_4 photosynthesis as detailed in ▶ Sect. 12.5, Kranz anatomy.

In leaf cross-sections, veins occupy the middle of the leaf. Leaf vascular bundles are typically closed (meaning no secondary growth). However, an active vascular cambium capable of producing up to 45 years of growth has been reported in long-lived leaves of bristlecone pine (Ewers and Schmid 1981). In all vascular bundles—root, shoot, or leaf—the xylem is oriented facing the adaxial direction and the phloem toward the abaxis. That arrangement is indicated in ◘ Fig. 12.2q and may be seen in numerous other images in this text.
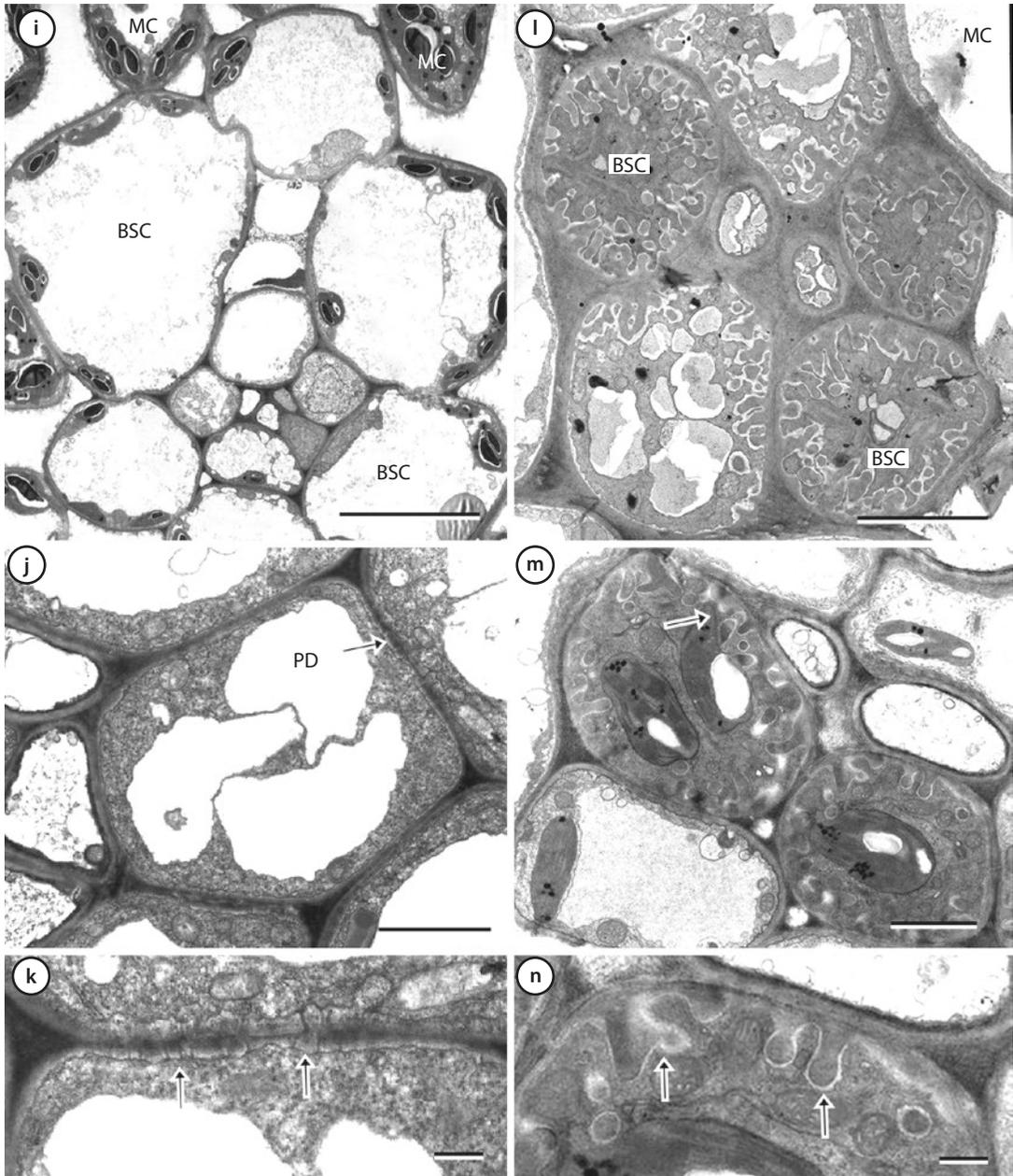
◩ **Fig. 12.2   i–n** Transmission electron micrographs of **i**, **j**, and **k** open and **l**, **m**, and **n** closed minor leaf veins. **i** A low magnification view of an open bundle sheath showing the relationship between the mesophyll cells (MC), bundle sheath cells (BSC), and the few cells of the minor vein in a leaf of *Paederia scandens* (Rubiaceae). **j** A single bundle sheath cell from *Hamelia patens* (Rubiaceae) with the plasmodesmata-containing wall indicated (PD). **k** A higher magnification view of **j** showing an area of cell wall with numerous plasmodesmata (*arrows*). **l** The bundle sheath cells (BSC) surrounding a closed vascular bundle in *Onosma gmelinii* (Boraginaceae) contain numerous cell wall ingrowths. **m** Two bundle sheath cells in a *Asperula kryloviana* (Rubiaceae) leaf. **n** A higher magnification view of **m** showing the cell wall ingrowths (*arrows*). Scale bars = 20 μm in **i** and **l**, 5 μm in **j** and **m**, and 1 μm in **k** and **n** (**i–n** Denis Batashev and Olga V Voitsekhovskaja, Komarov Botanical Institute, St. Petersburg, Russia)

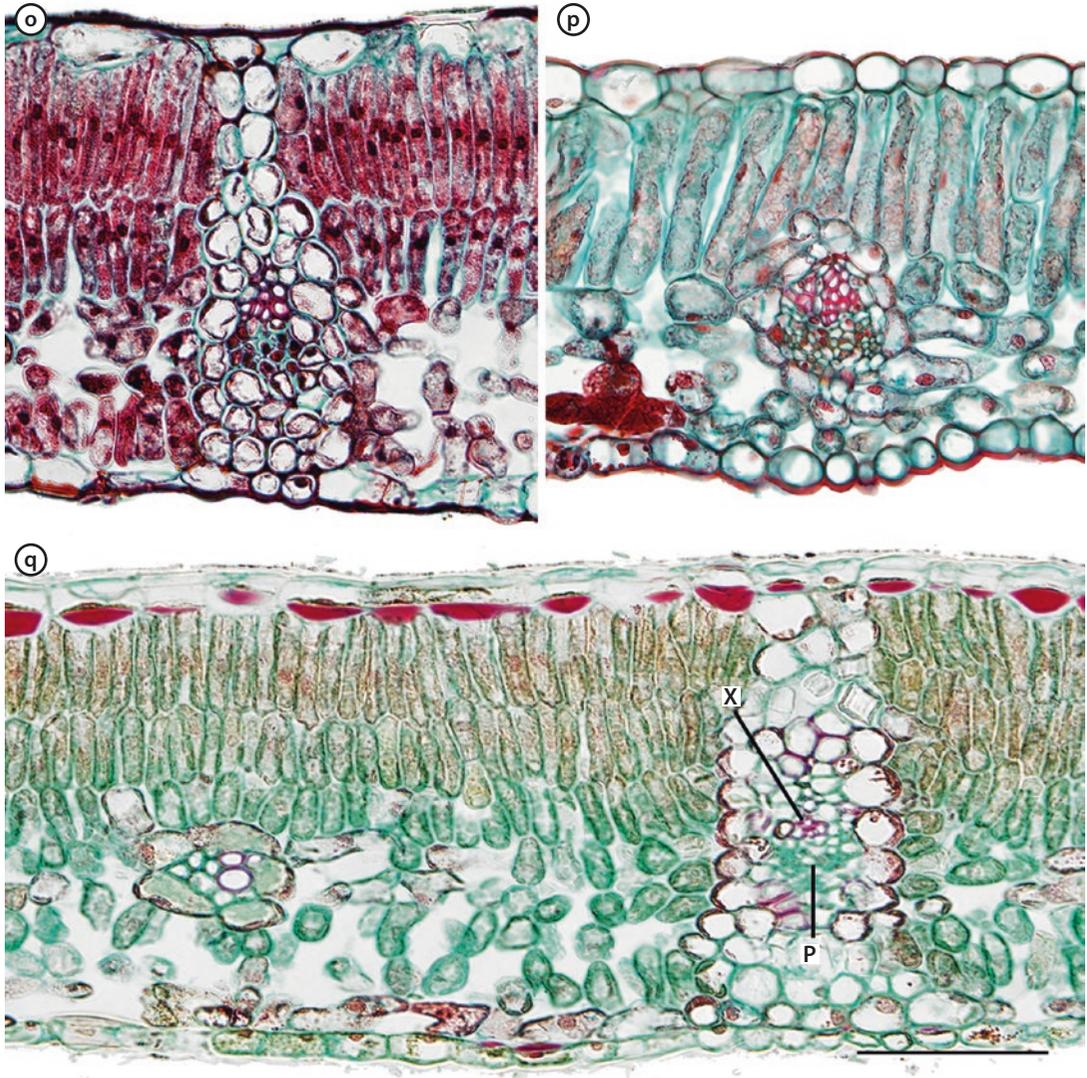■ **Fig. 12.2**   **o–q** Heterobaric and homobaric leaves seen in cross-section. **o** Bundle sheath extensions in a heterobaric Osage orange (*Maclura pomifera*) leaf. **p** Homobaric leaf of privet (*Ligustrum* sp.) which lacks bundle sheath extensions. **q** Bundles with and without sheath extensions in a cherry (*Prunus* sp.) leaf. Note the xylem (X) is toward the adaxis and the phloem (P) is toward the abaxis. Also note the single palisade layer in **p**, double in **o**, and triple in **q**. Scale bar in **q** = 50 μm and applies to all panels (**o–q** RR Wise)

The vascular system may play a role of providing support to leaves via the development of collenchyma, sclerenchyma fibers, or individual sclereids. If the leaf bundle has collenchyma, that tissue may protrude below the abaxial surface of the lamina in the form of a prominent rib (■ Fig. 12.2r). Fibers are most often associated with large veins, either surrounding them completely or forming abaxial or (to a lesser extent) adaxial caps. Collenchyma is also often found at the periphery of the major veins and on the leaf margin where it helps to prevent tearing of the leaf.
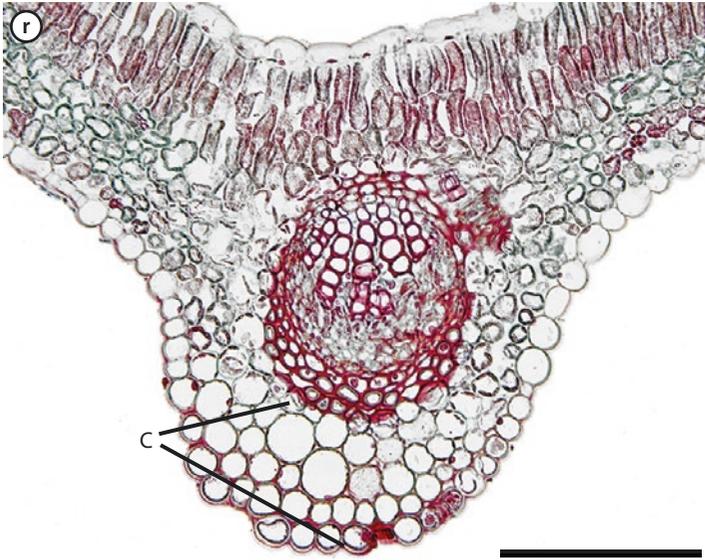
◘ Fig. 12.2 **r** Cross-section of an alfalfa (*Medicago sativa*) leaf midrib. Collenchyma (C) forms a bulge at the abaxial side of the midrib. Scale bar = 100 μm (RR Wise)

12.3 Leaf Morphology Is Optimized for Light Absorption, Gas Exchange, and Water Conservation

While many leaf modifications may be found in the kingdom Plantae (refer to ◘ Fig. 12.1g–k), leaves on most plants are engaged primarily, if not exclusively, in photosynthesis. In such leaves, called foliage leaves, the leaf epidermis consists of a single (or occasionally multiple) layer of cells covering the entire leaf surface and is continuous with the surface of the stem. In addition, the epidermis contains a variety of cells including guard cells, pavement cells, trichomes (glandular and nonglandular), and idioblasts, whose descriptions have been given in the chapters on the epidermis (► Chap. 9) and secretory structures (► Chap. 13).

12.3.1 Typical Dorsiventral Eudicot Leaves

The anatomy of the leaf lamina is normally best shown in cross-sectional view. A typical mature, C_3, eudicot leaf (◘ Fig. 12.3a) has an adaxial (upper) epidermis, an abaxial epidermis, and a zone of photosynthetic tissue in between called the mesophyll. In the epidermis, only guard cells have chloroplasts, which are used to drive stomatal opening and closing (► Sect. 3.5.10).

Comparing a cross-section with an accompanying paradermal section reveals the leaf structure in three dimensions (◘ Fig. 12.3b, c). Mesophyll is, as a rule, a multilayered parenchyma (also classified as
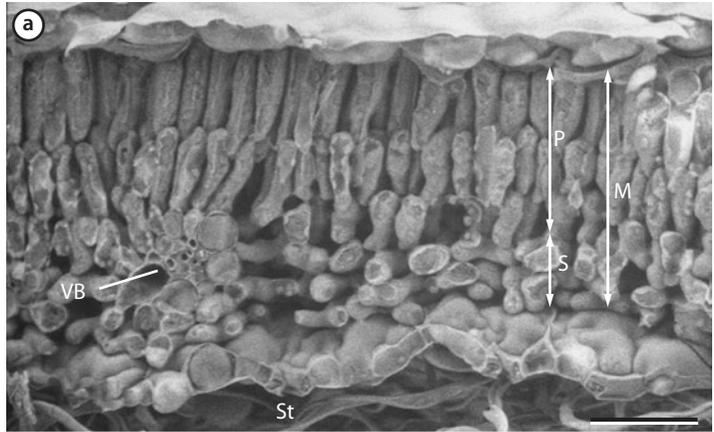
Fig. 12.3   a Colorado cinquefoil (*Potentilla subjuga*). M mesophyll, P palisade mesophyll, S spongy mesophyll, St stoma, VB vascular bundle. Scale bar = 50 μm (RR Wise)
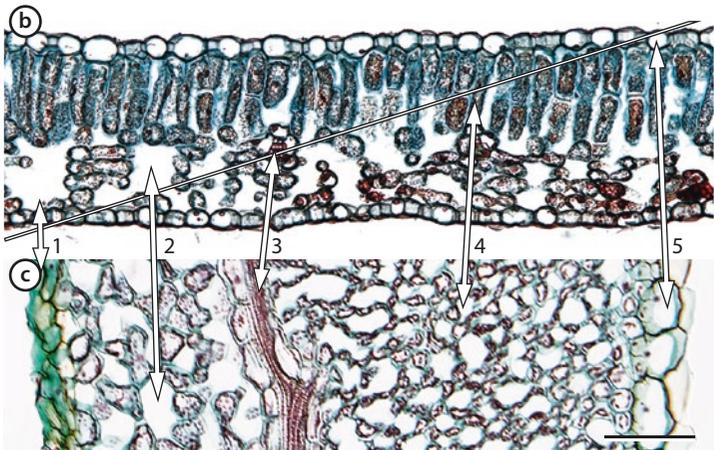

Fig. 12.3   b, **c** Comparison of **b** cross and **c** paradermal sections in privet (*Ligustrum* sp.), a C$_3$, eudicot species with homobaric leaves. **b** In cross-section, the internal structure of palisade and spongy mesophyll layers is easily seen. The diagonal line in **b** indicates the plane of the paradermal section. **c** The paradermal section cuts through the different levels of tissue semi-parallel to the leaf surface. 1 abaxial epidermis, 2 spongy mesophyll with intercellular spaces (at *arrow heads*), 3 minor vein, 4 palisade mesophyll with vacuoles (at *arrow heads*), 5 adaxial epidermis. Scale bar = 100 μm (**b**, **c** RR Wise)

chlorenchyma) consisting of cells which are similar in their structure and functional specialization for photosynthesis. Their most important specific feature is the extraordinary development of chloroplasts. There is no other tissue where there are so many chloroplasts or such a high level of organization. The presence of a highly developed anastomosing system of intercellular air spaces in which more than half of the cell surfaces are in contact with the gas phase is a characteristic of mesophyll. The remaining surfaces are in cell-to-cell contact and have numerous plasmodesmata that allow for symplastic continuity among mesophyll cells and the vascular system.

In the eudicot leaf, the mesophyll is typically divided into two layers of chlorenchyma (◨ Fig. 12.3a, b). The layer toward the adaxis, called the **palisade mesophyll**, is composed of long, columnar, tightly packed cells with little intercellular air space. Palisade cells have a large vacuole and numerous chloroplasts, because those cells are exposed to the highest irradiance level and photosynthesize at a high rate. The **spongy mesophyll** toward the abaxis has mostly isodiametric cells and a large amount of intercellular air space. Spongy mesophyll cells are smaller than palisade mesophyll cells, with fewer chloroplasts, in accordance with the lower light level toward the abaxis. This style of leaf morphology is common in plants that hold their leaves directly from the stem and more or less perpendicular to the sun's rays, exposing the adaxis to incoming light. The form of leaf anatomy is called dorsiventral (or bifacial) to indicate that the leaf has distinctly different internal anatomy in a abaxial/adaxial direction (dorsiventral), and the two layers of the mesophyll are composed of morphologically different tissues (bifacial).

All of these anatomical features of the eudicot leaf relate leaf orientation and to the three competing demands placed on a leaf of 1) light absorption, 2) CO_2 uptake, and 3) water conservation. The light environment inside the leaf is quite heterogeneous; not all chloroplasts receive that same quantity and quality of light. Leaf anatomy is maximized to take advantage of that heterogeneity to maximize photosynthesis during diurnal and seasonal variations in light level (Xiao et al. 2016).

Both the palisade layer and the spongy layer are actively engaged in photosynthesis, which requires constant supplies of light and carbon dioxide. At the adaxial surface, which receives more irradiance than the abaxial surface, the columnar shape of the palisade cells, combined with their large vacuoles, serves to bend and focus light to the leaf interior. Some of the incoming light is absorbed by chloroplasts and used for photosynthesis, but some is focused to the spongy layer. The palisade cells, therefore, are anatomically specialized to not only absorb light but also to facilitate the penetration of light to chloroplasts in the lower, spongy mesophyll layer. The spongy mesophyll layer, with its fewer chloroplasts and larger exposure to internal air spaces, is specialized in CO_2 absorption, which enters the leaf via the numerous abaxial stomata. Some consider the spongy mesophyll to be a form of aerenchyma. Placing the stomata on the abaxial surface has another advantage in that the atmospheric humidity is lower in the shaded, underside of the leaf. A higher external humidity reduces transpirational water loss.

12.3.2 Variations in Palisade Parenchyma

The palisade mesophyll is typically a single layer of long and tubular cells at the leaf adaxis (◨ Fig. 12.3a, b); however other shapes and arrangements exist. Individual palisade parenchyma may be Y-shaped as in *Lilium* or X-shaped as in *Lactuca* (◨ Fig. 12.3d, e). The arrangement and numbers of palisade layers in a leaf vary as
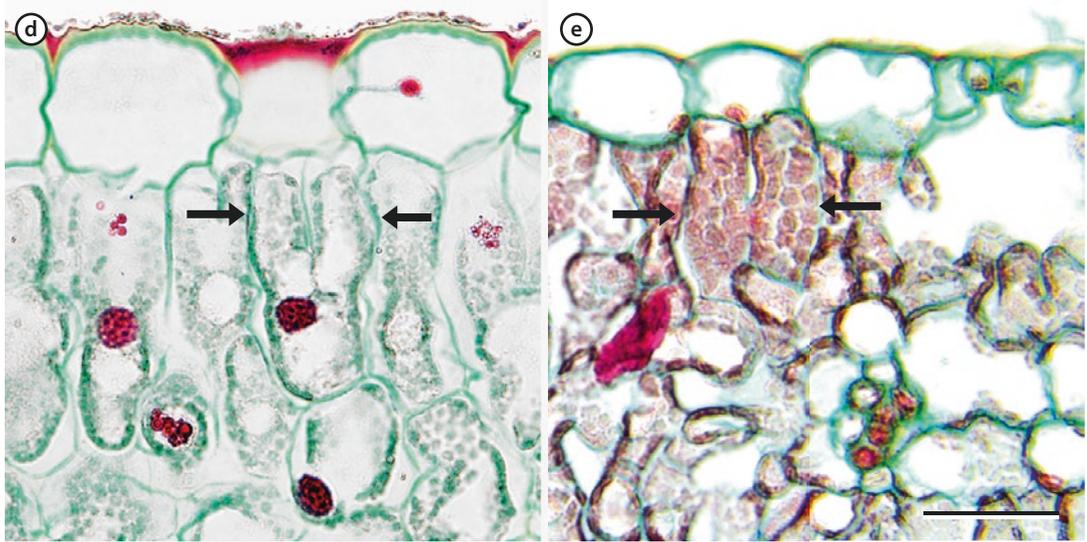
◘ Fig. 12.3   d, e Cross section of **d** lily (*Lilium* sp.) leaf showing a Y-shaped palisade cell and **e** a lettuce (*Lactuca* sp.) leaf with an X-shaped palisade cell (between arrows). Scale bar in **e** = 25 μm and applies to both panels (**d**, **e** RR Wise)

◘ Fig. 12.3   f Cross-section of a willow (*Salix* sp.) leaf composed entirely of palisade parenchyma mesophyll cells. Note also the vascular bundle extension in this heterobaric leaf. Scale bar = 50 μm (RR Wise)

well. A bifacial/dorsiventral eudicot leaf may have one, two, or three palisade layers, as can be seen in ◘ Fig. 12.2o, p, and q. **Isobilateral** or **isolateral** (meaning "same both sides") leaves are strictly defined as having the same "face" at both sides of the leaf. There are three different anatomical arrangements that qualify as isobilateral. (1) Leaves may lack a spongy layer and be composed of only a palisade mesophyll layer; *Salix* is an example of a palisade-only, isobilateral leaf (◘ Fig. 12.3f). (2) Leaves may lack a palisade layer and be composed of only a spongy mesophyll; grasses (► Sect. 12.3.3) and hydrophytes (► Sect. 12.8) commonly have spongy-only, isobilateral leaves. (3) Isobilateral leaves may have two palisade layers, one at each surface, with a spongy layer in between; this arrangement is common in water-storing xerophytic plants (► Sect. 12.6).
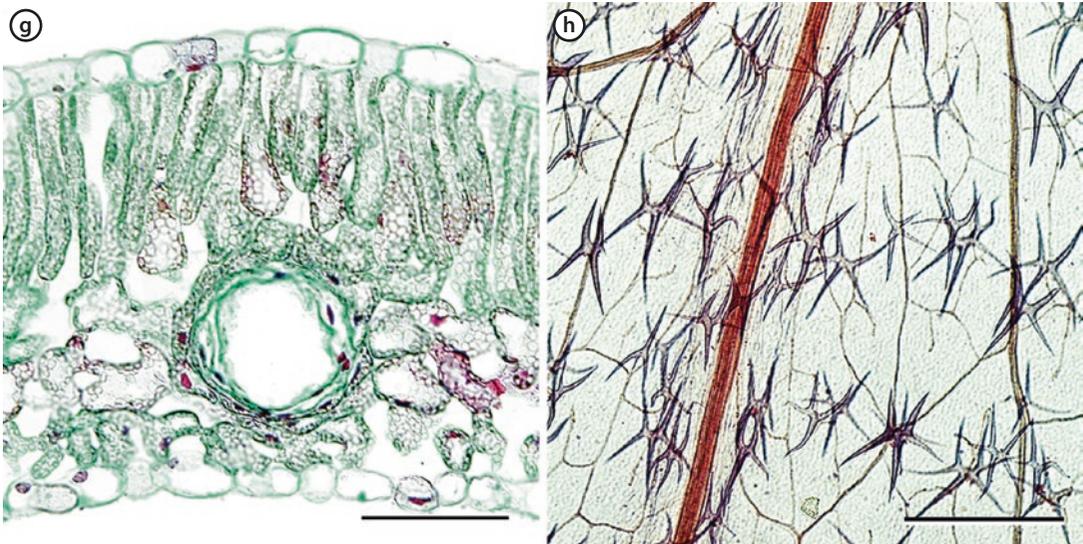
◨ Fig. 12.3   g A resin canal in a cotton (*Gossypium hirsutum*) leaf cross-section. The canal is lined with an epithelium of secretory parenchyma cells. **h** Astrosclereids as seen in a clearing of a shepherd's purse (*Capsella bursa-pastoris*) leaf. Scale bars = 50 μm in **g** and 250 μm in **h**. (**g** RR Wise; specimen in **h** prepared by J.D. Curtis)

In addition to the chlorenchyma of the palisade and spongy layers, non-photosynthetic cells in the mesophyll may include oil cells, crystal-containing idioblasts, and mucilage cells. Resin canals and sclereids may be interspersed among the mesophyll cells (◨ Fig. 12.3g, h).

Adaxial and abaxial epidermises in eudicot leaves often differ from each other in the size and shape of ground epidermal cells and in the frequency of trichomes and stomata. In leaves which are oriented with the adaxial epidermis toward the light (as described above for the typical eudicot leaf), stomata mostly occur within the abaxial epidermis (i.e., designated as hypostomatous leaves). Such a location allows for the reduction of water loss during transpiration. In vertical or upright leaves without a preferable orientation with respect to light, the stomata may be found in both epidermises (**amphistomatous leaves**). Many grass leaves, with upright positioning, are amphistomatous, as is wheat (◨ Fig. 12.3i). In other cases, such as leaves of **xeromorphic** grasses (◨ Fig. 12.6a) or floating leaves of water plants (◨ Fig. 12.8b), the stomata are confined to the upper epidermis (i.e., designated as epistomatic **l**eaves). Leaf anatomical adaptations to various environmental conditions are discussed in more detail in ▶ Sects. 12.6–12.9.

### 12.3.3 Isobilateral and Unifacial Monocot Leaves

The leaves of most monocot species are more or less upright and crowded to either side by neighboring leaves. In this orientation, a single light-focusing palisade layer and the dorsiventral/bifacial
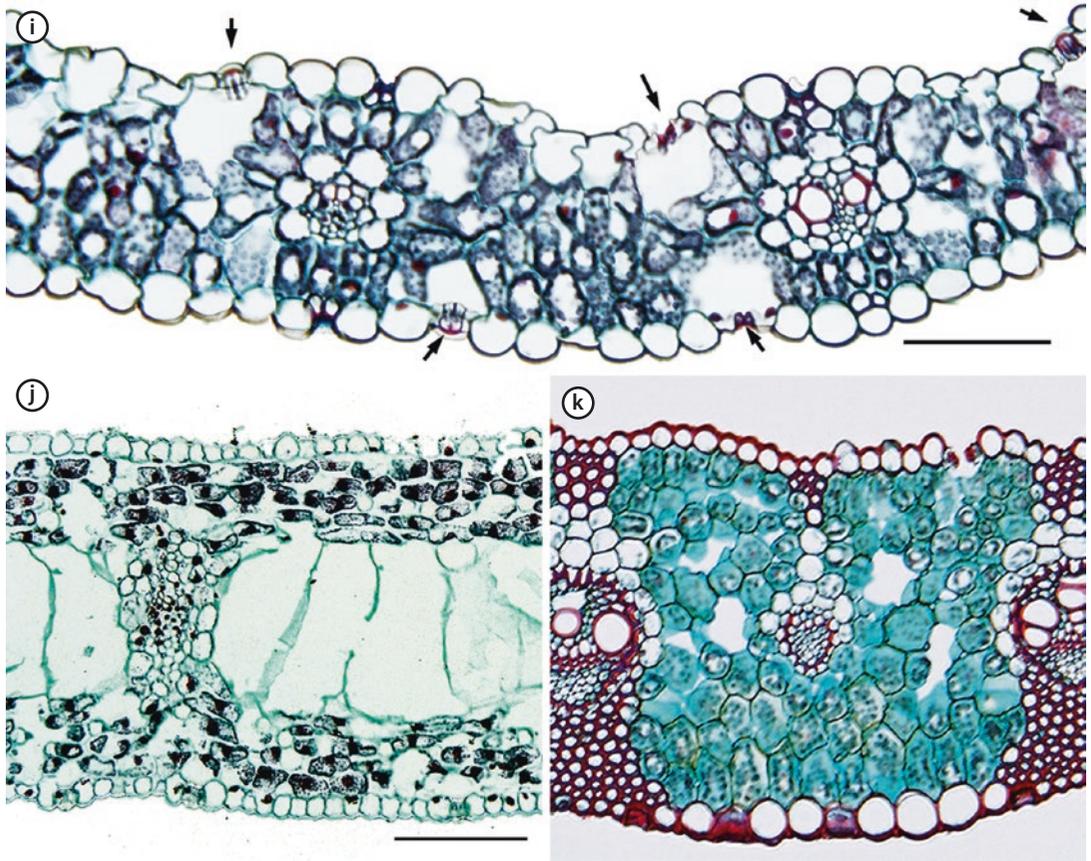
◙ Fig. 12.3   i–k Isobilateral monocot leaves with identical abaxial and adaxial surfaces and a homogeneous mesophyll of isodiametric loosely arranged cells. **i** Wheat (*Triticum aestivum*) (arrows indicate stomata), **j** Amaryllis (*Amaryllis belladonna*) and **k** Kentucky bluegrass (*Poa pratensis*). The abaxis is to the bottom of all three images. Scale bars = 100 μm in all three panels. (**i–k** RR Wise)

tissue arrangement described above for eudicots would not be adaptive. In general, therefore, the majority of monocots, such as grasses, lilies, and irises, have parallel-veined isobilateral leaves. Therefore, an isobilateral, mesophyll-only arrangement can be seen in monocots such as wheat, amaryllis, and *Poa* (◙ Fig. 12.3i–k).

A **unifacial** leaf has an unusual anatomy. Leaf development at the apical meristem usually starts as a leaf buttress growing off of one side of the apex as shown in ◙ Fig. 4.7b for an *Elodea* shoot tip. In this way, the leaf assumes an adaxial (toward the axis) and an abaxial (away from the axis) symmetry very early on in its development, and the two faces develop accordingly. However, in some monocots, only the abaxial side continues development, and an adaxial side never develops. In iris, the leaf is flattened and is oriented along the vertical axis of the plant (◙ Fig. 12.3l). As noted above, vascular bundles are always arranged with their phloem toward the abaxial surface. Therefore, in the iris leaf, vascular bundles on both sides of the leaf are oriented with their phloem facing the outside of the leaf. The middle of the leaf is filled with non-photosynthetic parenchyma (in some iris varieties, mucilaginous cavities may
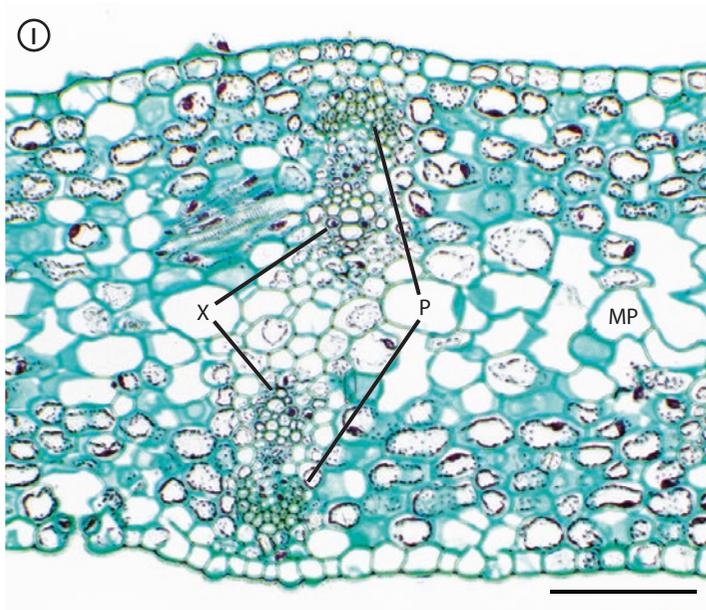
◘ **Fig. 12.3   I** Anatomy of a unifacial monocot leaf of (*Iris domestica*). Note that the abaxial orientation of the vascular bundles as evidenced by the phloem (P) tissues lying outside the xylem (X) tissues. The location of the missing adaxial surface is occupied by mesophyll parenchyma (MP). Scale bar = 100 μm (RR Wise)

occupy the leaf interior). There is only an abaxial side, which is on both sides. This same basic developmental pathway can be seen in the tubular leaves of onion (◘ Fig. 9.1i) or bulrush (◘ Fig. 12.3q, r). The two differences are as follows: (1) the leaves are cylindrical and not flattened and (2) the internal parenchyma breaks down, leaving a hollow, tubular leaf.

12.3.4 Centric Eudicot Leaves

Centric leaves are circular in cross-section, with a solid interior, in which the palisade parenchyma layer forms a continuous ring around the spongy mesophyll. The centric leaf shape minimizes the surface area to volume (SA/V) ratio. Because transpiration occurs at the surface and the water lost comes from the interior, a low SA/V ratio reduces transpiration, and centric leaves are often found on xerophytes or halophytes. In the pincushion tree (*Hakea drupacea*, ◘ Fig. 12.3m, n), the palisade forms a double layer surrounding a central core of parenchyma with small vascular bundles scattered throughout. *Hakea* is a drought-resistant evergreen native to southwestern Australia and naturalized elsewhere. Saltwort (*Salsola* sp.) is a C_4 plant with centric leaves and a prominent bundle sheath lying to the interior of a single layer of palisade cells (◘ Fig. 12.3o, p). Saltwort is known to store water in the leaf interior (Carolin et al. 1975).
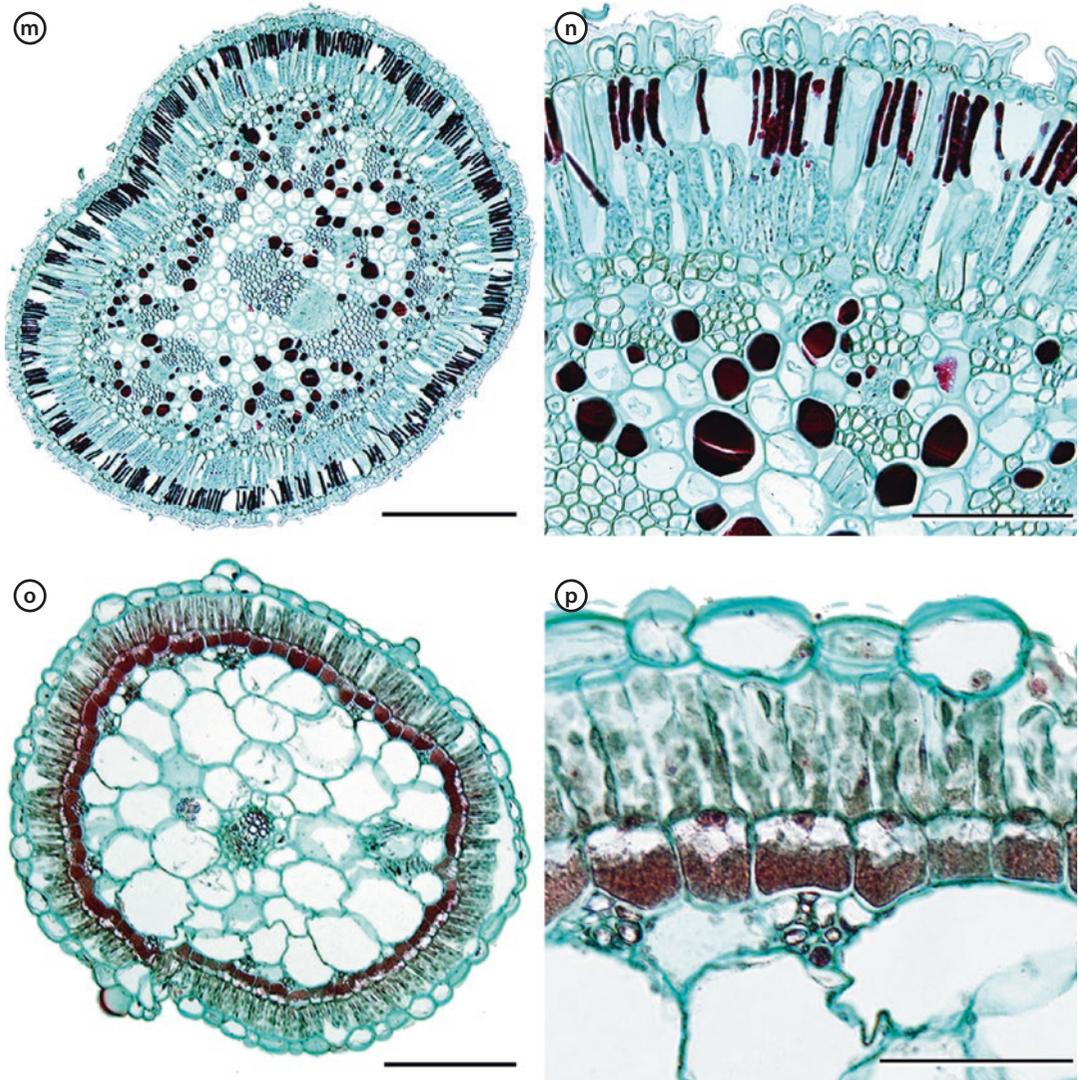
◻ Fig. 12.3   m–p Eudicot centric leaves. **m**, **n** Sweet hakea (*Hakea drupacea*) has a double layer of palisade parenchyma surrounding a central core of parenchyma with small vascular bundles scattered throughout. **o**, **p** The saltwort (*Salsola* sp.) centric leaf has a single layer of palisade cells to the exterior of a dense bundle sheath. Scattered minor bundles lie just inside the bundle sheath. A larger major bundle sits at the center of the water-storing parenchyma. Scale bars = 250 μm in **m**, 100 μm in **m** and **o**, and 25 μm in **p** (**m–p** RR Wise)

12.3.5 Tubular Leaves

Tubular leaves are round, like centric leaves, but they have a hollow interior. *Juncus* leaf is tubular with multiple palisade layers (◻ Fig. 12.3q, r). The palisade mesophyll appears next to the epidermis around the complete cylinder. Below it is the non-photosynthetic spongy mesophyll in which vascular bundles are found. Note that the phloem of vascular bundles faces the exterior of the leaf, as in the stem. The central part of the leaf, which is hollow, was originally composed of thin-walled parenchyma which was a stable tissue and therefore torn apart as the outer part of the tubular leaf expanded. The
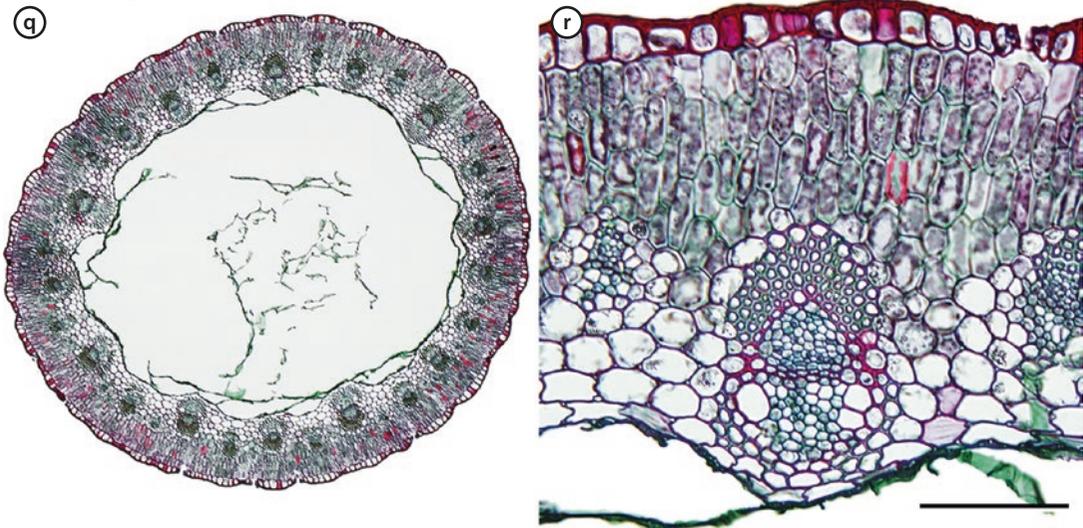
■ **Fig. 12.3**   **q**, **r** Tubular leaf of bulrush (*Juncus* sp.). Scale bars = 250 μm in **q** and 50 μm in **r** (**q**, **r** RR Wise)

adaxial side is to the inside of the leaf. It is unifacial because an adaxial epidermis never developed. *Juncus* is an aquatic plant with emergent leaves, meaning the roots are submerged below the surface, in oxygen poor soil, while the leaves are extended above the surface. The open center of the *Juncus* leaf allows for the diffusion of oxygen, which is photosynthetically generated by the chloroplasts of the leaf, down to the roots. Onion (*Allium cepa)* which is shown in ■ Fig. 9.1i also has a tubular leaf. In this case, the tubular shape of the onion provides a structural advantage (Schulgasser and Witzum 1992).

Technically speaking, the carnivorous trap of the carnivorous pitcher plant *Sarracenia* sp. (■ Fig. 12.1i) is a tubular leaf modified to make a trap. It, however, is bifacial (Shreve 1906). In addition, the leaf sheaths which compose much of the support structure of *Miscanthus* sp. grass (■ Fig. 11.2i) and wheat are tubular leaves.

12.4 The Light Environment During Development Can Modify Leaf Anatomy

Besides availability of water (refer to following sections), light intensity is an especially important ecological factor affecting leaf anatomy. The various placements of the leaf palisade layer(s) discussed above reflect the adaptation of a plant to its light environment. Different light intensities intercepted by a leaf during development can also have an impact on the anatomy of the mature leaf. Where light intensity during leaf development is low, as in the interior of a tree's crown, **shade leaves** are formed. They are usually larger in area than **sun leaves**—leaves that develop at the light-exposed periphery of the crown—but thinner. Shade leaves have a thinner cuticle, less internal air space, and a single palisade layer (compare ■ Fig. 12.4a, b). The chloroplasts of shade leaves possess larger grana (i.e., more thylakoids; refer to ■ Fig. 3.5v, w) and contain more
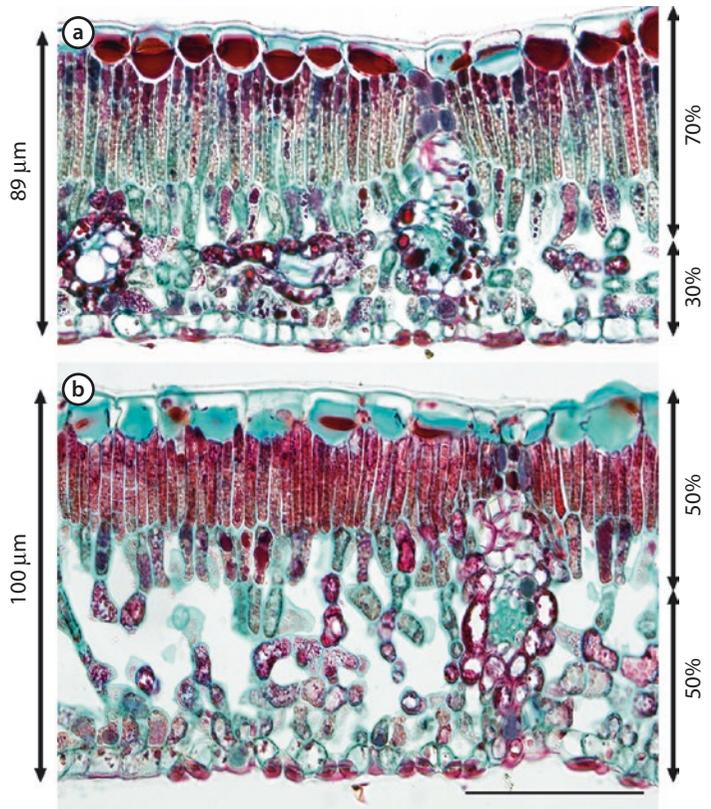
Fig. 12.4 **a** Shade and **b** sun leaves from oak (*Quercus* sp.). Images are labeled to indicate leaf, palisade mesophyll, and spongy mesophyll thicknesses. Scale bar in **b** = 50 μm and applies to both panels (**a**, **b** RR Wise)

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chlorophyll. Sun leaves often have two or more palisade layers and considerably more internal air space.

The sun/shade developmental plasticity allows plants to optimize photosynthesis based on the light environment. Photosynthesis requires two substrates—light and carbon dioxide (water is being ignored for this discussion)—and one of them is always limiting. At the periphery of the crown, CO_2 is limiting and light is saturating. That is, the leaf has all the light it can use, and the maximal rate of photosynthesis is limited by the supply of carbon dioxide. Thus, such leaves have a thin light-absorbing layer (the palisade) and a robust, airy CO_2-absorbing layer (spongy mesophyll). Shade leaves develop the opposite anatomy: a thick palisade to capture any available light and a thinner spongy layer because there is less need for CO_2 uptake. The rate of photosynthesis in a sun leaf can be as much as five times that of a shade leaf.

Shade and sun leaves may often be found in different parts of the crown of the same tree, but keep in mind these differences are only expressed during leaf development. Transferring a shade leaf to the sun by trimming back outer crown foliage will not cause a mature leaf to thicken.
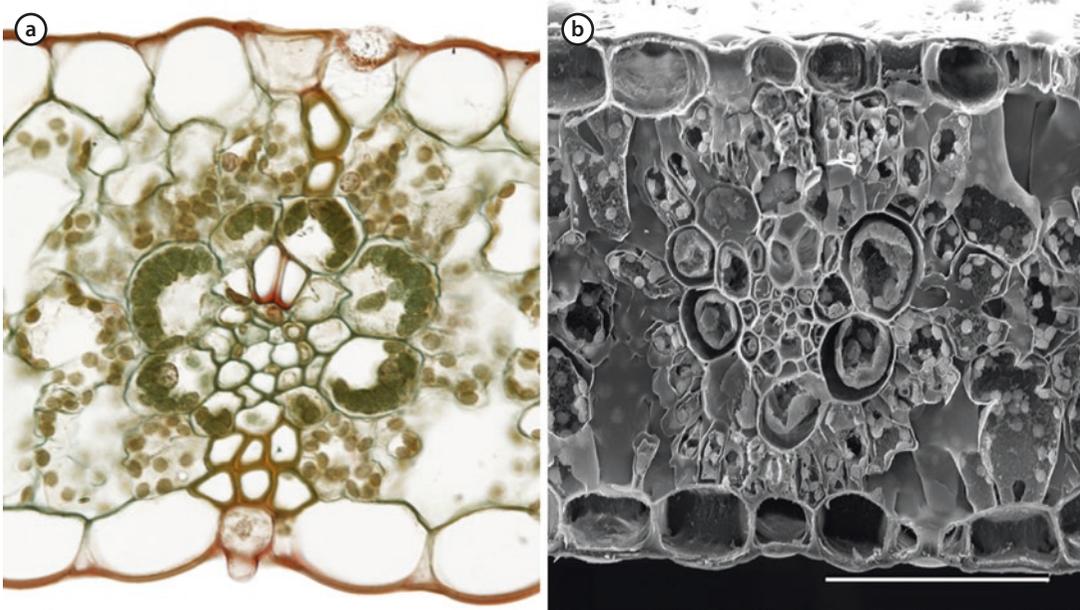
Fig. 12.5 **a**, **b** Maize (*Zea mays*) leaf in cross-section. **a** Light micrograph showing a single leaf vein surrounded by a ring of large bundle sheath cells. Note the centrifugal orientation of the bundle sheath chloroplasts, placing them closer to the surrounding mesophyll chloroplasts (refer to ▣ Fig. 3.5 q, r). **b** Scanning electron micrograph of a similar view. The bundle sheath cell contents are slightly shrunken in this preparation. Scale bar in **b** = 100 μm and applies to both panels (**a**, **b** RR Wise)

12.5 Kranz Leaf Anatomy Is a Specialization of the C$_4$ Photosynthetic Pathway

As described in ▶ Sect. 12.3, the bundle sheath cells of C$_3$ plants are rather inconspicuous. In contrast, C$_4$ bundle sheath cells are large and bright green due to the presence of numerous chloroplasts. This anatomical variant is called **Kranz anatomy** (German for "halo" or "wreath") and is unique to plants that utilize the C$_4$ photosynthetic pathway (▣ Fig. 12.5a, b).

C$_4$ photosynthesis and its dimorphic chloroplasts were introduced in ▶ Chap. 3 (▶ Sect. 3.5.9). Because the two chloroplasts have such different biochemical roles to play in the leaf, they typically must be kept in different compartments in plants. Therefore, in addition to having two different chloroplast types, C$_4$ plants also have unique bundle sheath cells that are used to provide the necessary spatial separation of those two organelles (▣ Fig. 12.5). No other part of the anatomy is affected in the C$_4$ syndrome.

C$_4$ photosynthesis has independently evolved multiple times and is found in both monocots and eudicots, being more common in the former. Kranz leaf anatomy is found in most, but not all, C$_4$ plants. Indeed, a few members of the Chenopodiaceae engage in single-cell C$_4$ photosynthesis, in which elements of the cytoskeleton separate the chloroplasts involved in the primary CO$_2$ fixation and those performing secondary CO$_2$ fixation into distinct compartments in the leaf cells (Voznesenskaya et al. 2001).

Box 12.2   Single-Cell C4 Photosynthesis Relies on Spatial Separation, No Matter How Small

C_4 photosynthesis is predicated on a spatial separation of the primary carboxylating enzyme, phospho*enol*pyruvate carboxylase (PEPcase), from the secondary carboxylating enzyme, ribulose-*bis*phosphate carboxylase/oxygenase (Rubisco). This is because the former operates well in a low $[CO_2]$ and high $[O_2]$ environment while the latter requires the opposite. In leaves with Kranz anatomy, this spatial separation is achieved with PEPcase being found in the mesophyll cells and Rubisco in the bundle sheath cells. Oddly, some plants, notably members of the genus *Bienertia* (Chenopodiaceae), are capable of single-cell C_4 photosynthesis. In this case, the leaf mesophyll cells are quite large, 80–110 µm in diameter, and a cluster of chloroplasts in the center of the cell is separated by scattered chloroplasts at the cell periphery by a large vacuole. PEPcase and primary carbon fixation are found in the periphery of the cell, while Rubisco and secondary fixation are localized to the chloroplasts in the central cluster (Voznesenskaya et al. 2001). Jurić et al. (2017) subsequently modeled the diffusion of CO_2 and O_2 within *Bienertia* mesophyll cells and demonstrated that a spatial separation of only 10 µm, well within the dimensions of the cells, could provide diffusive resistance necessary to spatially separate the primary carbon fixing reactions catalyzed by PEPcase from the secondary fixation reactions catalyzed by Rubisco.
Reference: Jurić et al. (2017).

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There are two basic variants of bundle sheath anatomy in C_4 plants. The single-layer bundle sheath of maize (◘ Fig. 12.5a, b) is in direct contact with leaf mesophyll cells to the exterior and vascular tissue to the interior, and the individual bundle sheath cells are quite large. Sugar cane (◘ Fig. 12.5c) likewise has a single bundle sheath layer, but its bundle sheath cells are smaller and more numerous than in maize. Single layer bundle sheaths are most common in the subfamily Panicoideae or the "warm season" grasses. Grama grass, a member of the "cool season" grass subfamily Pooideae, has two layers of bundle sheath cells (◘ Fig. 12.5d). The outer layer is parenchymatous, while the inner layer, called the **mestome**, has thickened cell walls. The mestome walls may be suberized and contain numerous plasmodesmatal connections to the vascular tissues to the interior and the outer layer of the sheath to the exterior. The mestome appears to allow the leaf a certain degree of differential control over the opposite movements of water from the xylem to the mesophyll and of photosynthate from the mesophyll to the phloem.
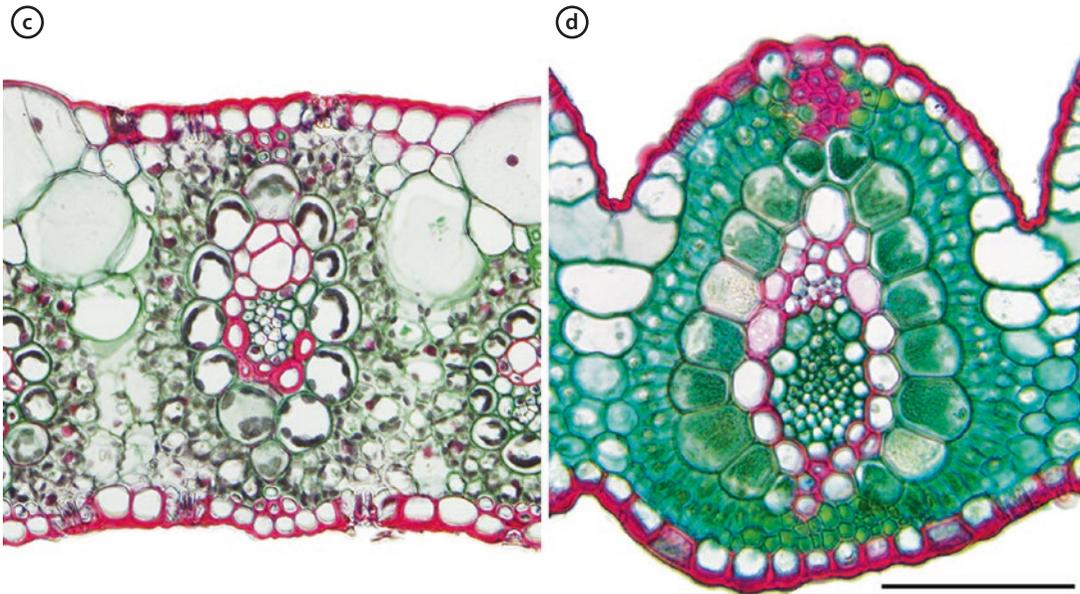
◧ Fig. 12.5   c, **d** Grass leaves with Kranz anatomy. **c** Sugar cane (*Saccharum officinarum*) has a single layer of photosynthetic bundle sheath cells, **d** while grama grass (*Bouteloua breviseta*) has a double layer of bundle sheath cells; the inner layer (mestome) is sclerified while the outer is parenchymatous. Gama grass is a xerophyte and shows features such as bulliform cells and a thick cuticle which are discussed in the next section. Scale bar in **d** = 50 μm and applies to both panels (**c**, **d** RR Wise)

12.6 Xeromorphic Angiosperm Leaves Conserve Water in Arid Environments

Water is of vital importance to plant survival and has been a strong force driving the evolution of leaf anatomy. Angiosperms may be divided into three basic categories based on their major adaptations to water supply. Leaves are typically the most plastic of organs in their ability to change their structure as an adaptation to a particular ecological factor. Xerophytes ("xeric" = dry, "phyte" = plant) are adapted to habitats with seasonal or permanent restrictions in water supply. Hydrophytes ("water plants") live in or on lakes, ponds, and streams. **Mesophytes** (literally "middle plants") are adapted to more favorable conditions and lack the specialized adaptations to overly dry or wet growing conditions. Mesophytic leaves, while not named as such, have been the subject of the much of the chapter so far. Xerophytic leaves are discussed in this section, in ▶ Sect. 12.7 (succulent leaves), and in ▶ Sect. 12.9 (gymnosperm leaves). Hydrophytic leaves are covered in ▶ Sect. 12.8.

Water loss through transpiration is a necessary evil for plants. Stomata must remain open for the diffusion of carbon dioxide to feed photosynthesis. However, the leaf interior is saturated with water—water that diffuses out through the open stomata as a gas (or vapor). Diffusion is a passive process driven by the difference in

concentration between the area of high concentration and the area of low concentration and is dependent on temperature (which is ignored in this discussion) and the mass of the molecules that are diffusing. Because CO_2 (which is diffusing into the leaf) and water vapor (which is diffusing out of the leaf) have different concentration gradients and different molecular masses, any resistance in their shared diffusional pathway (i.e., from the leaf to the atmosphere or vice versa, via the stomata) will have a different impact on the total flux of gases. As a consequence, leaf anatomical adaptations that inhibit diffusion, such as sunken stomata, dense coverings of trichomes, and leaf rolling, have a greater impact on H_2O diffusion than CO_2 diffusion. The diffusion of both gases is inhibited, but the diffusion of H_2O is inhibited more than the diffusion of CO_2, and water use efficiency (the ratio of water lost to carbon gained) is improved. Growth may be slowed, but competitiveness is enhanced. This is the evolutionary bargain xerophytes have struck.

Beach grasses are prototypical xerophytic plants. Sandy beach soil holds little water. Exposure to full sunlight and steady winds places a high transpirational demand on the leaves while requiring a tough, flexible leaf design. In addition to evolutionarily driven anatomical adaptations, short-term water status can actually affect leaf shape. Many grasses are capable of rolling or folding their leaves during drought due to the presence of specialized motor (or **bulliform**) cells in the adaxial epidermis (◼ Fig. 12.6a–c) (Kadioglu and Terzi 2007). If the water supply is sufficient, these cells are turgid and keep the leaf open. In the case of water deficit, the cells lose turgor and shrink. The leaf, like a spring, rolls into a tube with the adaxial surface and stomata oriented to the inside of the tube. The leaf rolling involves not only the obvious changes in the size of the bulliform cells but also takes advantage of the entire leaf surface as

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◼ **Fig. 12.6   a** Typical xeromorphic leaf of European beachgrass (*Ammophila arenaria*) in the rolled-up configuration. Scale bar = 0.5 mm (RR Wise)
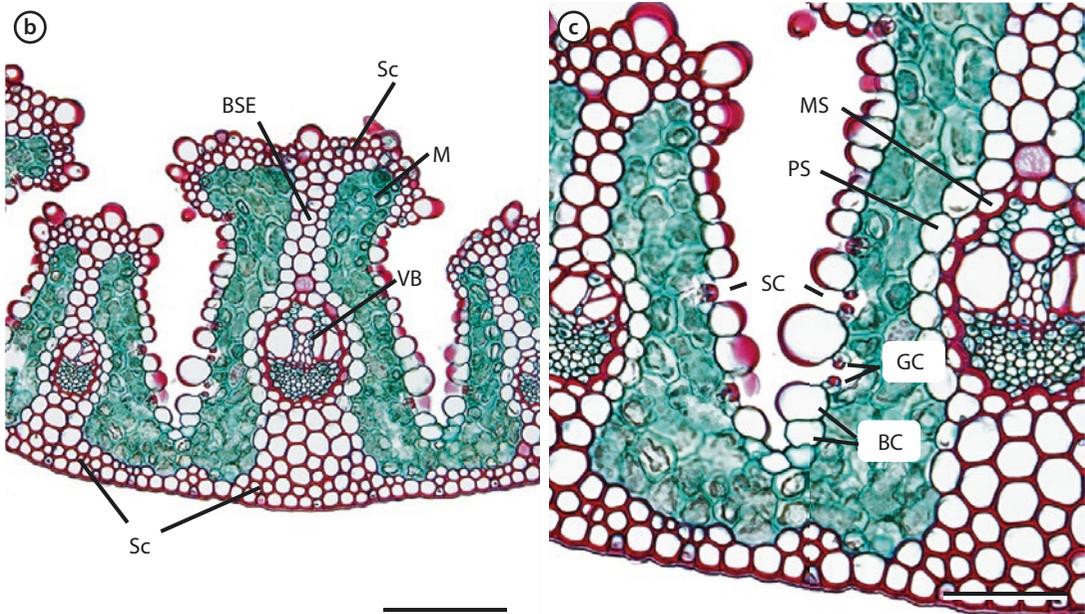
☉ Fig. 12.6 Details of the xeromorphic European beachgrass (*Ammophila arenaria*) leaf. **b** Vascular bundles (VB) have large bundle sheath extensions (BSE) which isolate areas of the compact mesophyll (M) into a heterobaric leaf. Both the outside (abaxial) and inside (adaxial) epidermal cells are heavily sclerified (Sc). **c** Stomatal guard cells (GC) sit below the leaf surface in stomatal crypts (SC). Thin-walled, non-sclerified bulliform cells (BC) occupy the hinge area of the leaf. Note the compact arrangement of mesophyll cells. There are two bundle sheaths in this species, an inner sclerenchymatous sheath designated the mestome sheath (MS) and an outer parenchymatous sheath (PS). The cell walls of the mestome sheath have "U-like" secondary thickenings. Scale bars = 50 μm in **b** 25 μm in **c** (**b**, **c** RR Wise)

a structure (Moulia 2000). Rolling of leaves allows them to reduce transpiration. Bulliform cells in *Poa pratensis* are particularly large (☉ Fig. 12.6d). Like many grasses, *Ammophila and Poa* leaves are epistomatous, with stomata on the upper, or adaxial, surface only. All stomata are found on the adaxial leaf surface and are concentrated in depressions called stomatal crypts that are found on the inner, unexposed side of the rolled leaves. A thick cuticle and heavy sclerification (a condition called **sclerophylly**) of the abaxial surface protect the rolled-up leaf from abrasion by windblown sand particles.

New Zealand hemp has **sclerophyllous leaves** with extensive columns of fibers at each vascular bundle (☉ Fig. 12.6e). These fibers are classified as hard and are highly lignified. Strands of these fibers are commercially used for cordage. Through lysigenous breakdown of the mesophyll, large central air cavities are formed. The photosynthetic mesophyll is concentrated beneath both the adaxial and abaxial epidermal layers. The leaf surfaces are covered with a thick layer of cuticular waxes (☉ Fig. 12.6f, g).

Eudicot leaves, as well, may have several xeromorphic adaptations. The surface of the Russian olive leaf is covered with a dense layer of thin trichomes that obscure the leaf surface (☉ Fig. 12.6h). *Nerium oleander* leaves have several xeromorphic adaptations. First, their stomata are housed in large, abaxial **stomatal crypts**
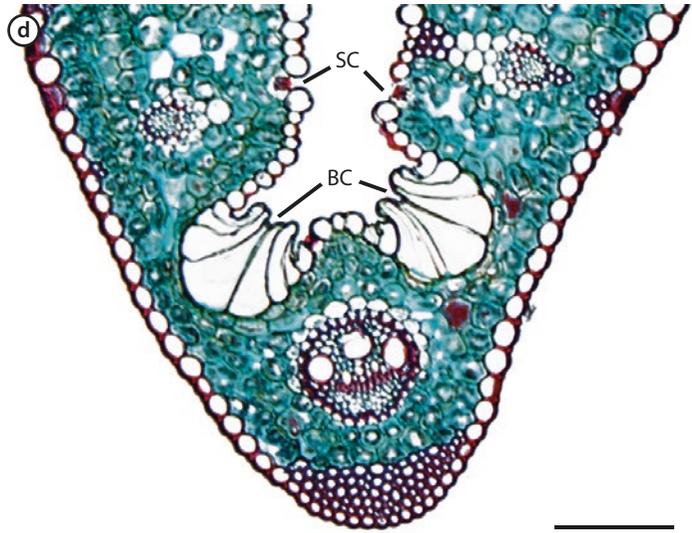
◘ Fig. 12.6   d Cross-section of the midrib of an involute Kentucky bluegrass (*Poa pratensis*) leaf showing stomatal crypts (SC) and bulliform cells (BC). Scale bar = 100 μm (RR Wise)
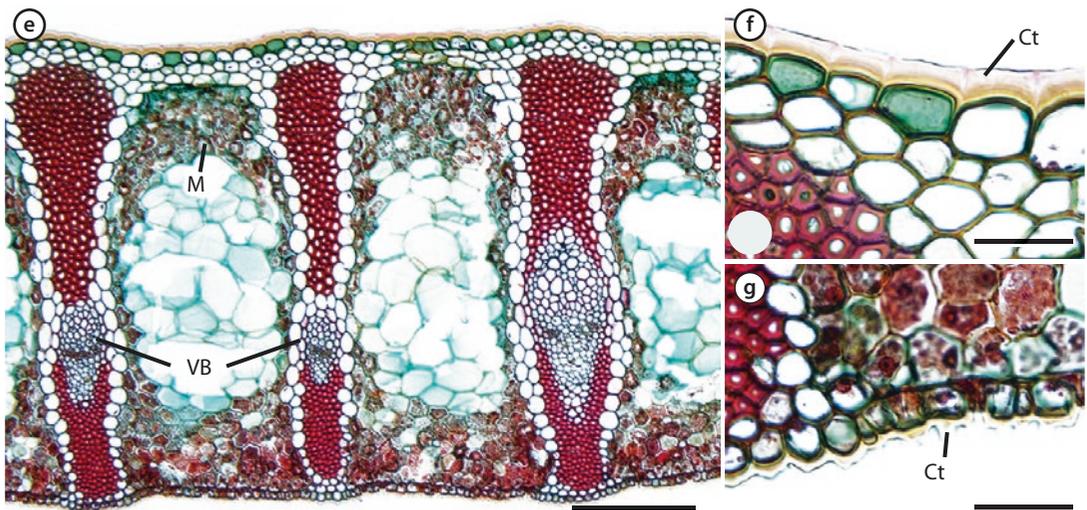
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◘ Fig. 12.6   e–g Xeromorphic leaf of a non-grassy monocot, New Zealand hemp (*Phormium tenax*). **e** Note the massive columns of fibers in the bundle sheath extensions, which provide significant support to this sclerophyllous leaf. Vascular bundles (VB) and the photosynthetic mesophyll (M) are labeled. **f** and **g** Higher magnification views of **f** the adaxial surface and **g** the abaxial surface showing the thick cuticle (Ct). Scale bars = 100 μm in **e** and 20 μm in **f** and **g** (**e–g** RR Wise)

(◘ Fig. 12.6i–k); each crypt may have five to ten stomata. The tissue lining the crypt is epidermal (as is obvious from the presence of stomata). Epidermal trichomes grow into the crypts and fill the crypts with a dense forest of trichomes (◘ Fig. 12.6i–k), with the effect of creating a pocket of water vapor, and thus reducing water loss by transpiration, especially in the wind. *Nerium* also has a thick cuticle and multiple layers of adaxial palisade (not shown), which are both xerophytic traits.
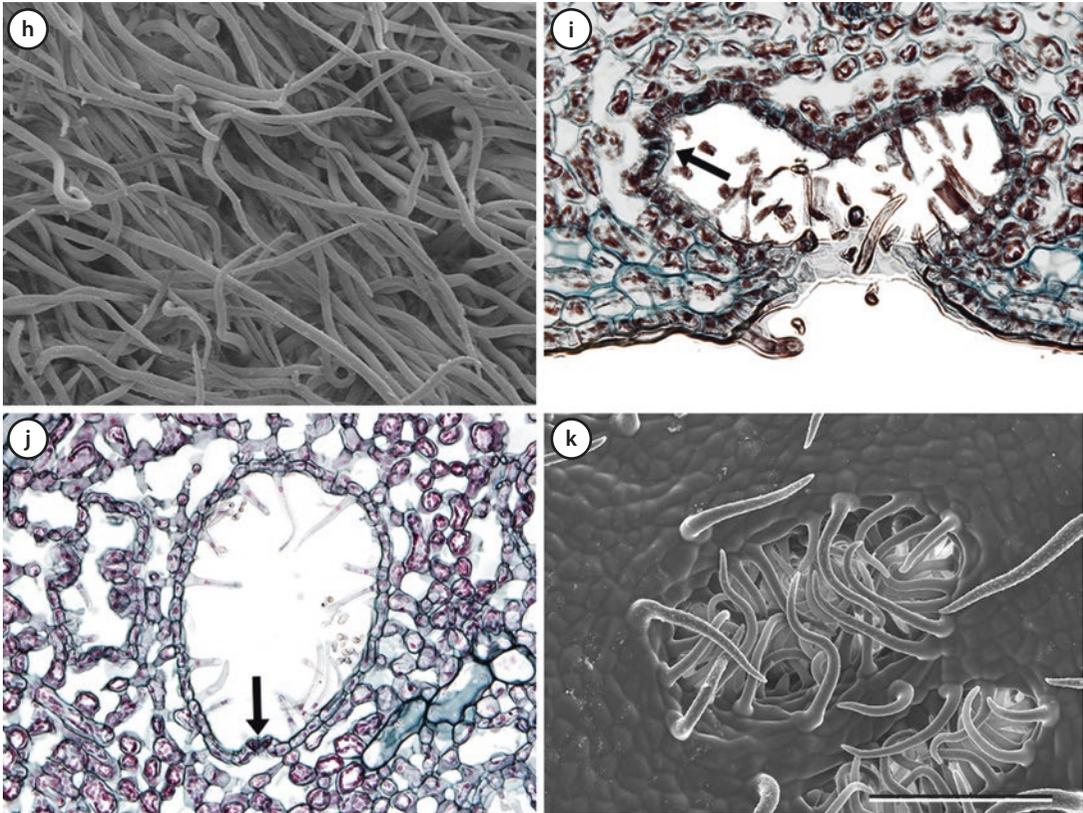
◘ Fig. 12.6   **h**–**k** Xeromorphic leaf surfaces. **h** SEM of the surface of a Russian olive (*Elaeagnus angustifolia*) leaf showing a dense layer of hair-like trichomes, so dense they obscure the underlying stomata. **i** A cross-section of a single stomatal crypt on the abaxial surface of an oleander (*Nerium oleander*) leaf lined with dense epidermal cells. One stoma is indicated by the arrow. All other stomata are out of the plane of section. **j** The same structure as described in **i**, except in paradermal section. Arrow marks a stoma and trichomes are evident. Note the epidermal layer lining the crypt and the surrounding spongy mesophyll. **k** SEM of a *Nerium* leaf abaxial surface. The forest of trichomes filling the crypt is clearly seen. Scale bar in **k** = 100 μm and applies to all panels (**h**–**k** RR Wise)

12.7 Succulent Leaves Are Adapted for Water Storage

The xerophytes discussed above have adapted to dry conditions largely by limiting the amount of water lost via transpiration. A second strategy used by some xerophytes is to store water internally in the leaf or stem, which imparts a character called **succulence**. The cactus shown in ◘ Fig. 12.1j is a stem succulent. The cactus stem has a photosynthetic exterior and a fleshy, water-storing interior while its leaves are non-photosynthetic, spiny, protective structures. Leaf succulence is a second water-storing strategy and will be discussed here.

The desert plants *Yucca*, *Agave*, and *Aloe* are typical, desert-dwelling leaf succulents. The saltwort shown in ◘ Fig. 12.3o–p also has succulent leaves and is found in salt marshes which, from the standpoint of water availability, can mimic desert conditions.

Yucca leaves can be a millimeter or more in thickness (◘ Fig. 12.7a). Compare that to the 100–200 μm mesophytic leaves
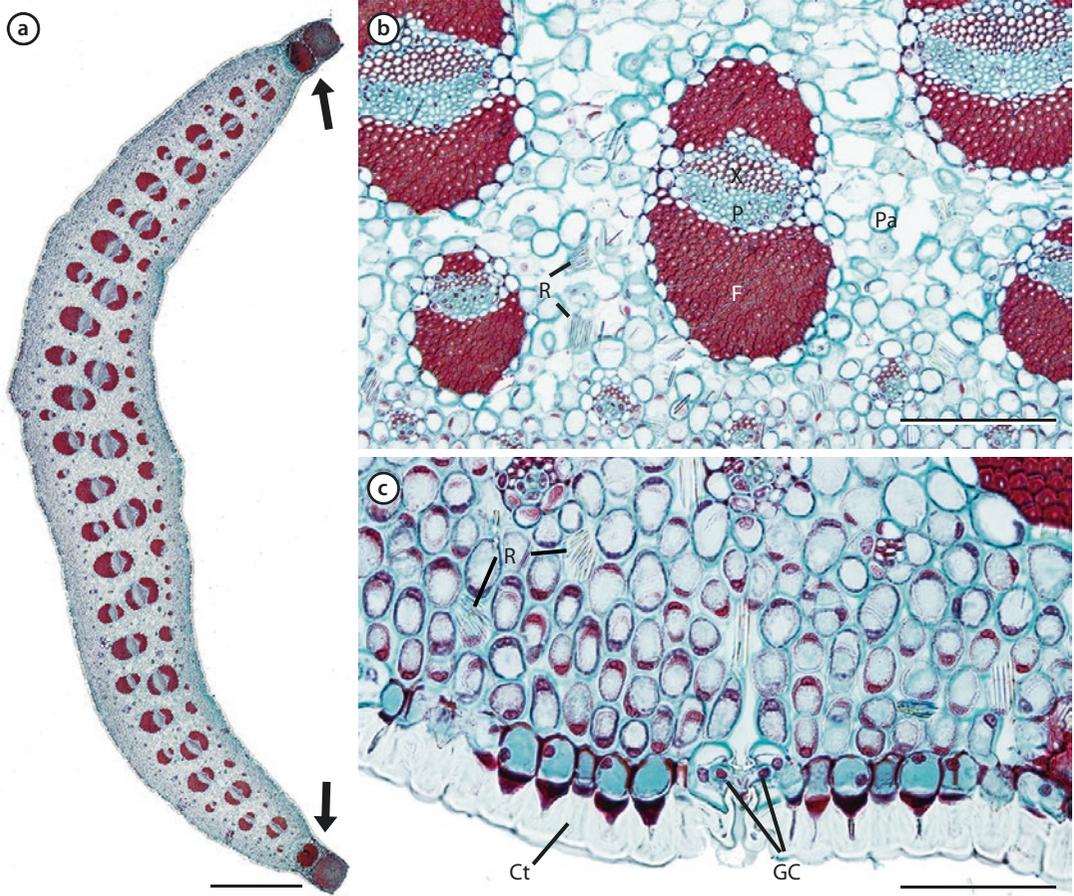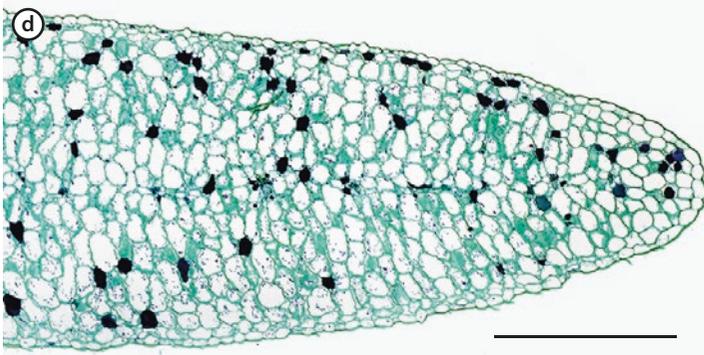
Fig. 12.7 **a–c** Cross-sections of an isobilateral succulent leaf, yucca (*Yucca* sp.). **a** Large fiber bundles strengthen the leaf margins (*arrows*) and cap the vascular bundles. **b** The interior of the leaf has numerous vascular bundles containing xylem (X) and phloem (P) with large fiber (F) caps to both sides. The water-storing parenchyma (Pa) has few organelles. **c** The mesophyll cells closest to the leaf surface are small with a dense cytoplasm (note the difference in scale between **b** and **c**. A thick waxy cuticle (Ct) covers the leaf, and guard cells (GC) are sunken beneath the epidermis. There is little internal air space, even in the photosynthetic tissue. Scale bars = 500 μm in **a**, 50 μm in **b**, and 100 μm in **c** (**a–c** RR Wise)

introduced in ▶ Sects. 12.3, 12.4, and 12.5. Succulent leaves accumulate water in the vacuoles of large, thin-walled water storage parenchyma cells in the mesophyll (�“ Fig. 12.7b). The photosynthetic mesophyll lies near the leaf surface on both the abaxial and adaxial sides (�“ Fig. 12.7c). Large fiber bundles protect both the leaf margins and the vascular bundles to the interior. Stomata are sunken below the epidermis, which is covered with a thick layer of cuticular wax. *Sedum*, another leaf succulent, has many of the same leaf characteristics (�“ Fig. 12.7d).

Begonia and canna leaves have an enlarged, water-storing hypodermis on both sides of the leaf, lying just underneath the epidermises (�“ Fig. 12.7e, f). Canna is grown as a food crop for its large, starch-filled tubers. Under well-watered conditions, the hypodermal cells are filled with water and turgid. During drought, the stored water is used by the plant, and the hypodermis shrinks significantly (Brück et al. 2001).

◼ **Fig. 12.7   d** Cross-section of an isobilateral *Sedum* sp. leaf filled with large, water-storing parenchyma cells. Scale bar = 500 µm (RR Wise)
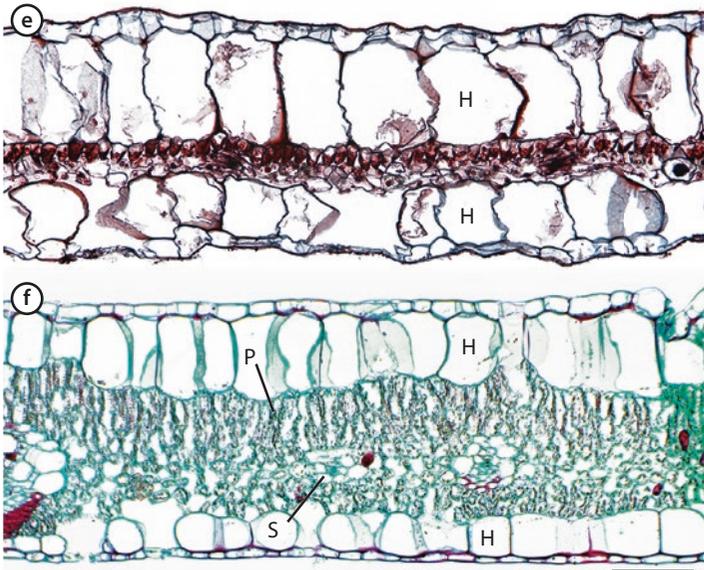
◼ **Fig. 12.7   e**, **f** Begonia (*Begonia* sp.) **e** and **f** canna (*Canna* sp.) leaves have a hypodermis modified for water storage. The leaves are bifacial in that they have a thin band of distinct palisade (P) and spongy (S) mesophyll sandwiched between an upper and lower hypodermis (H). The layers are not distinct in the *Begonia* leaf. Scale bar in **f** = 100 µm and applies to both panels (**e**, **f** RR Wise)

12.8 Aquatic Angiosperms Have Hydromorphic Leaves

Aquatic plants are found in the ferns, monocots, and eudicots. They may have leaves that are fully submerged, float on the surface, or emerge from the water. As an example, northern wild rice (*Zizania palustris*) has all three forms of leaves. Seeds germinate underwater, and the first leaves to emerge from the embryo remain submerged. Subsequent leaves grow to and float across the water surface. A third set of leaves emerges from the water and extends up to 1 meter above the surface (Sculthorpe 1967). Aquatic leaf shape varies.
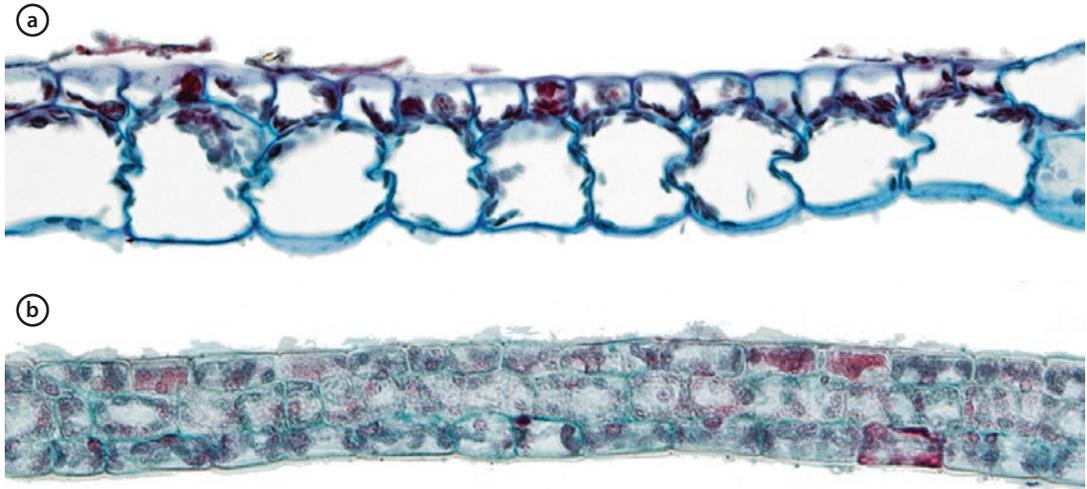
Fig. 12.8   a Submerged leaves of Canadian waterweed (*Elodea canadensis*) are only two cell layers thick. Chloroplasts are found in both layers. **b** Submerged leaf of pondweed (*Potamogeton* sp.) has three layers, two epidermal layers, and one mesophyll layer. The leaves of both plants are thin, with a minimal cuticle, no internal gas spaces, and no xylem. Scale bar in **b** = 50 μm and applies to both panels (**a**, **b** RR Wise)

Typically, leaves found in or on flowing water such as rivers and streams are long and narrow, while those found in or on quiet waters, ponds and lakes, may be oval or round.

Submerged leaves clearly do not face the same constraints on transpiration and gas exchange as aerial leaves and usually have a thin cuticle and lack stomata and intercellular air spaces (■ Fig. 12.8a). Light levels under water are reduced, photosynthetic rates are low, and dissolved gasses diffuse into the leaf from the surrounding water. With no need for transpiration and buoyancy provided by the water, xylem and fibers are usually absent. The thin cuticle probably serves as a barrier to bacterial and fungal infection, rather than as a barrier to water loss.

Because diffusion is only effective over short distances, most submerged leaves have a single-layered mesophyll, such as *Potamogeton* (■ Fig. 12.8b), or lack a mesophyll entirely. *Elodea* leaves are so thin that photosynthesis takes place in the cells of the bilayer leaf, each of which qualifies as an epidermis (■ Fig. 12.8a).

In other, larger submerged plants such as *Myriophyllum* (■ Fig. 12.8c), the leaves are finely divided into cylindrical **pinnae**. Such structure improves diffusion of carbon dioxide from water into the leaf for photosynthesis in the absence of gas spaces. The leaves and petioles are centric and unifacial which may allow for a greater and more uniform surface for the diffusion of carbon dioxide from the water into the leaf for photosynthesis. The movement of the surrounding water through the fingerlike leaves also helps circulate dissolved gases.

In floating leaves, stomata are found on the upper surface exposed to the air but not on the lower surface in contact with the

◘ Fig. 12.8 c Cross-section of a pinna of a leaf from the submerged hydrophyte water milfoil (*Myriophyllum* sp.). The plant has pinnately divided leaves which lack cuticle, stomata, sclerenchyma, and air spaces in the mesophyll. The inset shows the centric and unifacial structure of a single leaf. Scale bars = 2 mm in main panel and 200 μm in inset (RR Wise)

water (◘ Fig. 12.8d, e). Palisade mesophyll consists of several layers of rather short cells at the adaxial surface. There is little shade on the surface of a pond and little CO_2 uptake from the bottom leaf surface which is floating on the water. Therefore, there is no need for an abaxial spongy mesophyll layer. Air spaces in spongy mesophyll constitute an aerenchyma that provides buoyancy. Structural support is provided by astrosclereids that extend throughout the mesophyll in all three dimensions.

Emergent leaves of aquatic plants face different constraints. Here, aerenchyma does not serve for buoyancy, but as a diffusional pathway for the movement of gases, oxygen primarily, from aerial leaves to the submerged stem and roots which are exposed to low environmental oxygen concentrations. Large galleries, or **lacunae**, run the length of the leaf (◘ Fig. 12.8f, g). While some of the oxygen in the diffusional pathway originates from the air,
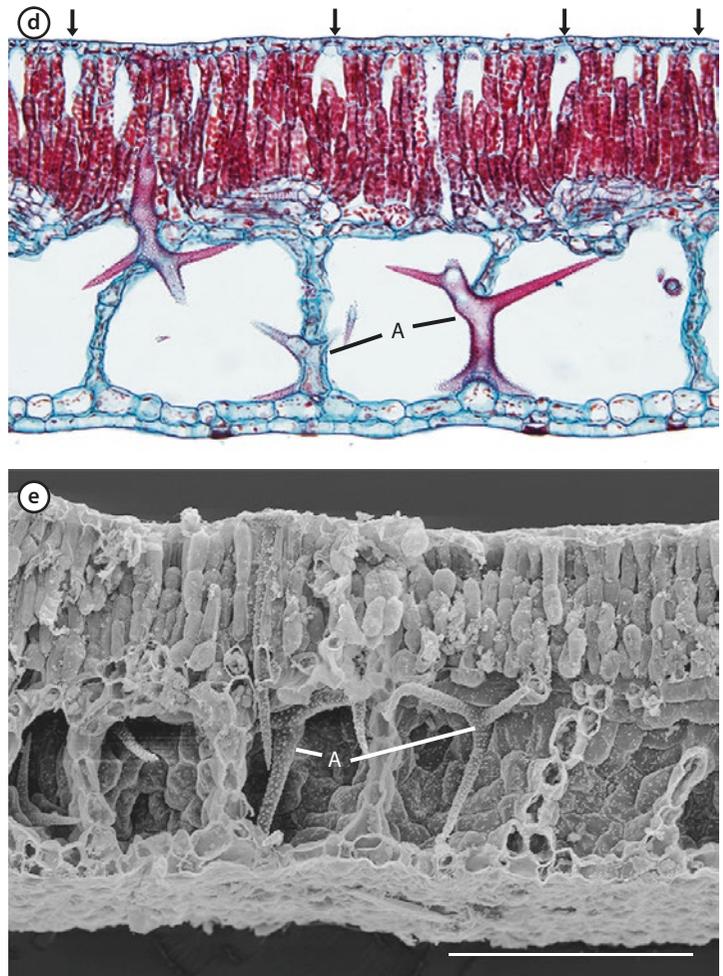
◘ **Fig. 12.8   d**, **e** Cross-sections of bifacial floating leaves from the fragrant water lily (*Nymphaea odorata*). **d** Palisade parenchyma lies on the adaxial surface, which has numerous stomata (*arrows*). Note the substomatal cavity underneath each stoma. Astrosclereids (A) provide support to the aerenchyma tissue on the abaxial surface. **e** The air spaces of the aerenchyma can be seen to extend throughout the leaf. Scale bar in **e** = 200 μm and applies to both panels (**d**, **e** RR Wise)

most of it is generated by the chloroplasts in the leaf itself (Sand-Jensen and Prahl 1982). Thus, much of the oxygen consumed by the roots is generated by photosynthesis and recycled from the leaves.

Being aerial, the leaves must have some means of support, usually in the form of bundles of fibers (◘ Fig. 12.8h) in the vascular bundle sheath extensions and structural cross bracing in the leaf lacunae (◘ Fig. 12.8i).
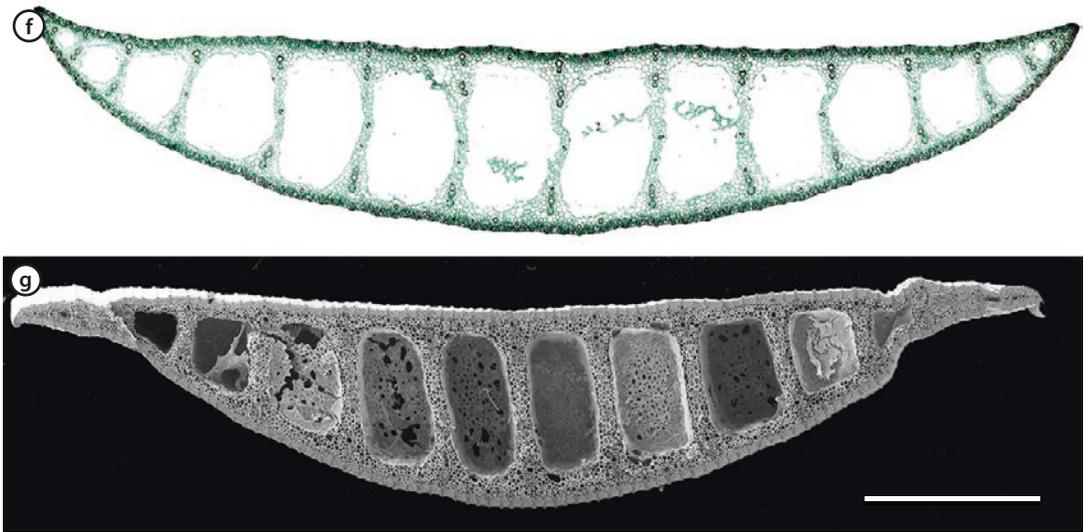
◘ Fig. 12.8   f, **g** Cross-sections of cattail (*Typha latifolia*) leaves. Large areas of aerenchyma form lacunae for gas exchange and facilitate the diffusion of oxygen from the atmosphere to the submerged portions in *Typha latifolia* LM and SEM. Scale bar = 2 mm (**f**, **g** RR Wise)
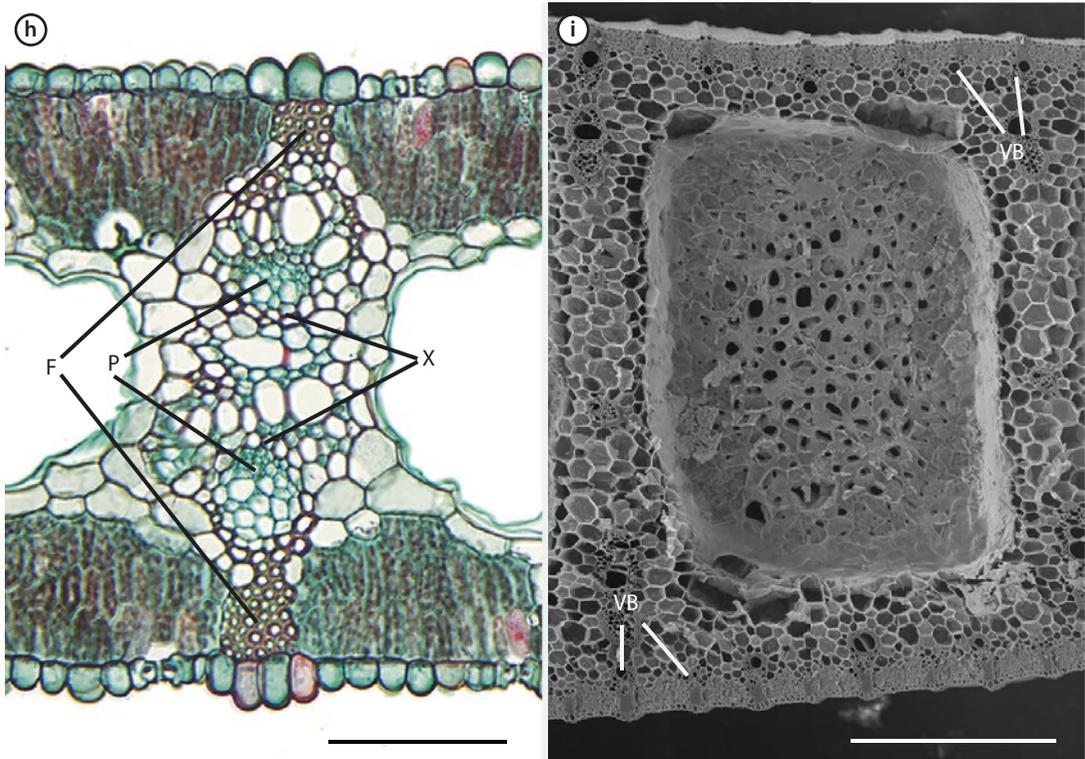
◘ Fig. 12.8   h Cross-section of a relatively thin portion of a cattail (*Typha latifolia*) leaf. Fibers (F) cap the xylem (X) and phloem (P) bundles in this isobilateral leaf. **i** In a much thicker cattail leaf, vascular bundles (VB) are arranged near the leaf surface, and a wall of porous aerenchyma provides structural support by cross bracing the leaf lacuna. Scale bars = 50 μm in **h** and 500 μm in **i** (**h**, **i** RR Wise)

12.9   Xeromorphic Conifer Leaves Conserve Water During Winter

Conifers (Coniferophyta) are the cone-bearing gymnosperms (refer to ► Sect. 1.16 for an overview of gymnosperms) and include species of pine, cedar, cypress, juniper, fir, redwood, spruce, and yew. Most of the conifers retain their leaves for multiple growing seasons and are thus termed "evergreens" with individual leaves living for 2–4 years, on average, or as long as 30 years in some species. The deciduous larches and tamaracks (*Larix* sp.) are among the few exceptions to the evergreen habit. Many conifers are native to temperate climate zones where low winter temperatures and heavy snowfall are common. In terms of water availability, winter conditions mimic drought in that there is very little liquid water in the soil. Therefore, xeromorphy—or a leaf with structural adaptations to dry conditions—is a hallmark of the gymnosperms. The epidermis consists of pavement cells with thick lignified secondary walls and a heavy cuticle. Stomata are deeply sunken with overarching subsidiary cells. The **epistomatal cavity** (depression over guard cells of stomata) is usually filled with dense deposits of wax particles which greatly reduce water loss. A sclerenchymatous hypodermis layer underlies the epidermis. It is interrupted only by substomatal cavities (◘ Fig. 12.9a–c).

Conifer leaves may be needle-shaped (pines), scalelike (cedars, cypress), or flattened (spruces and firs), thus reducing the surface-to-volume ratio and likewise transpiration. Pine needles are arranged in bundles of one to five individual leaves called a fascicle. In this case, each leaf assumes a section of the circle (◘ Fig. 12.9d–f). Larger, scalelike leaves are flattened in the dorsiventral direction and may have an adaxial palisade mesophyll and an abaxial spongy mesophyll (◘ Fig. 12.9g). The mesophyll may be homogeneous and compact as in pines or differentiated into palisade and spongy parenchyma as in fir, larch, and cycads. Conspicuous resin ducts are found in all conifer leaves. (◘ Fig. 12.9d–i).

Conifer leaves contain a central cylinder of vascular tissues and fibers called a **fibrovascular bundle** (◘ Fig. 12.9j). The bundle is bounded by an endodermis, which separates and isolates it from the leaf mesophyll. Interior to the endodermis is a layer or two of cells called **transfusion tissue**. Transfusion tissue is composed of living parenchyma cells and dead tracheids, and its presence and role are unique to gymnosperms (Wordsell 1897). The transfusion tissue lies between the xylem and the mesophyll and serves to recover solutes from the transpiration stream before they leave the fibrovascular bundle and return them to the adjacent phloem (Canny 1993).
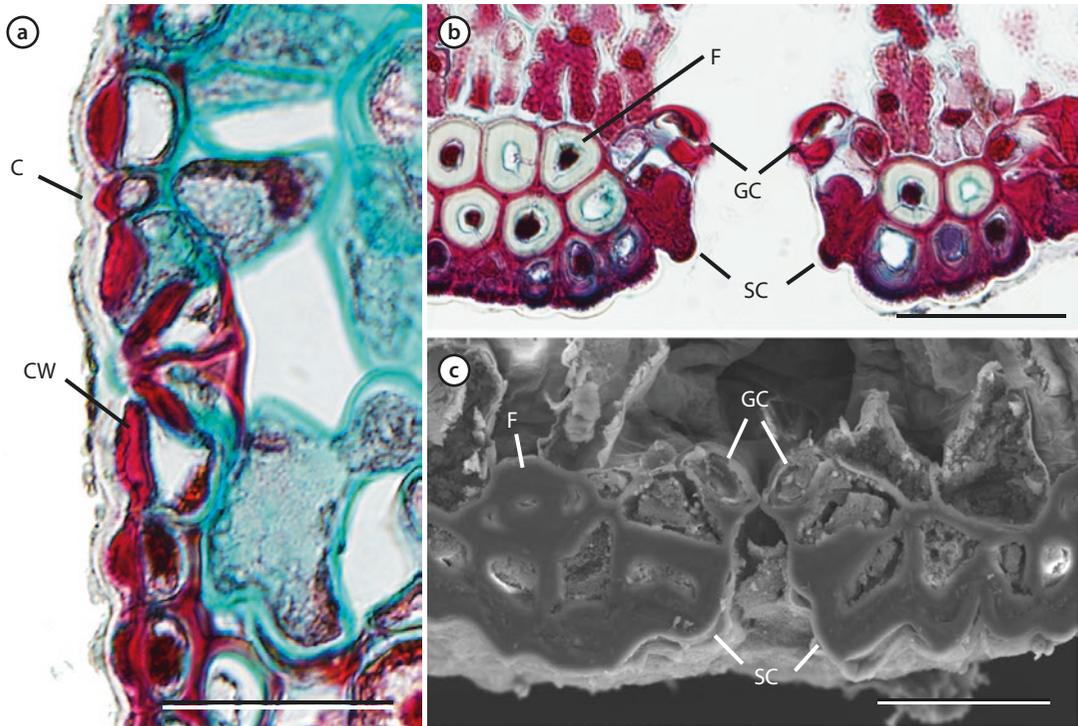
◨ Fig. 12.9   a–c Xeromorphic characters of gymnosperm leaves. **a** Eastern hemlock (*Tsuga canadensis*) epidermis showing a thick cuticle (C) and thick epidermal cell walls (CW). Mesophyll cells are to the right in the figure. **b** and **c** show hypodermal fibers (F) and sunken guard cells (GC) overarched by subsidiary cells (SC) in **b** single-leaf pinyon (*Pinus monophylla*) and **c** white fir (*Abies concolor*). Scale bars = 20 μm in **d** and 50 μm in **e** and **f** (**a–c** RR Wise)

12.10   Leaf Abscission Is a Timed and Genetically Controlled Process

In many plants, the leaf is a temporary organ, designed to take advantage of favorable conditions during the growing season and then discarded as environmental conditions become less favorable. **Drought deciduous** plants, such as *Euphorbia splendens*, drop their leaves at the start of seasonal dry periods whereas winter deciduous plants, such as many temperate, eudicot trees, drop their leaves in the fall, prior to winter, in a process called **autumnal senescence**. The actual shedding of the leaf is called abscission ("ab" = away from and "scise" meaning to cut). Leaf abscission is preceded by multiple physiological and anatomical changes.

The senescence of deciduous leaves is the end result of a detailed and genetically controlled process of resource recovery. Chloroplasts
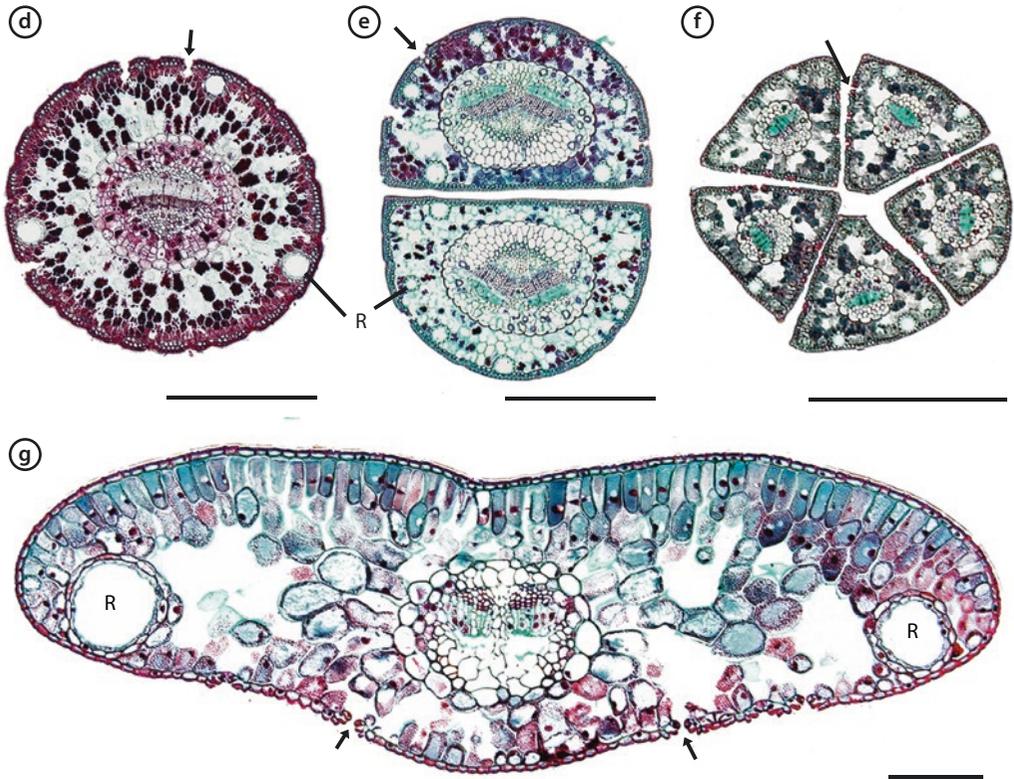
⬛ Fig. 12.9   Cross-sections of conifer leaves. **d** One leaf in a fascicle of the single-leaf pinyon (*Pinus monophylla*).
e Two-leaf fascicle of the Austrian pine (*Pinus nigra*). **f** Five-leaf fascicle of white pine (*Pinus strobus*). **g** This fir (*Abies* sp.) leaf
has a flattened shape with a palisade parenchyma to the adaxis and spongy mesophyll to the abaxis. In all images, resin
ducts are labeled (R) and arrows indicate stomata. Scale bars = 500 μm in **d** through **f** and 100 μm in **g** (**d–g** RR Wise)
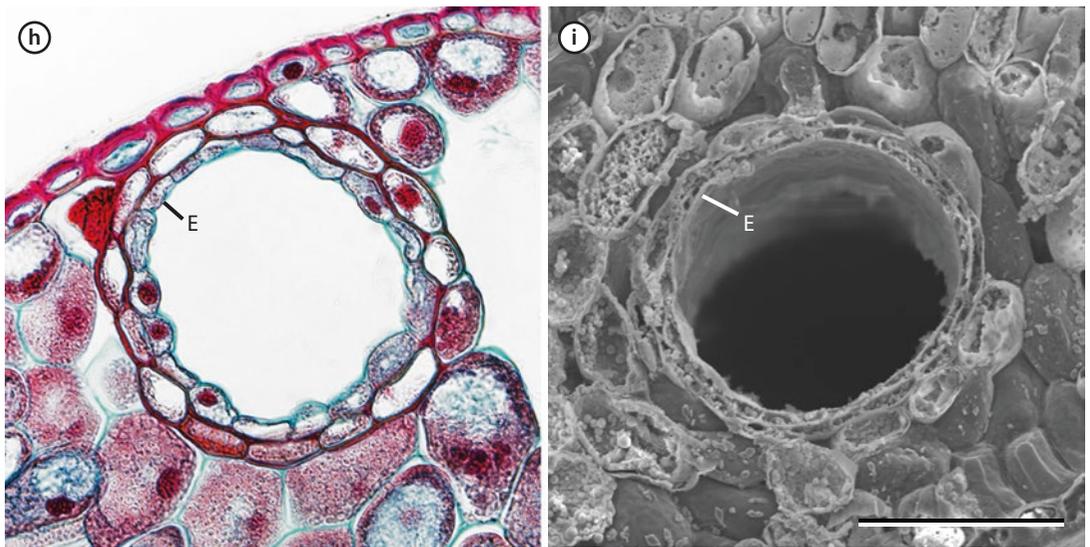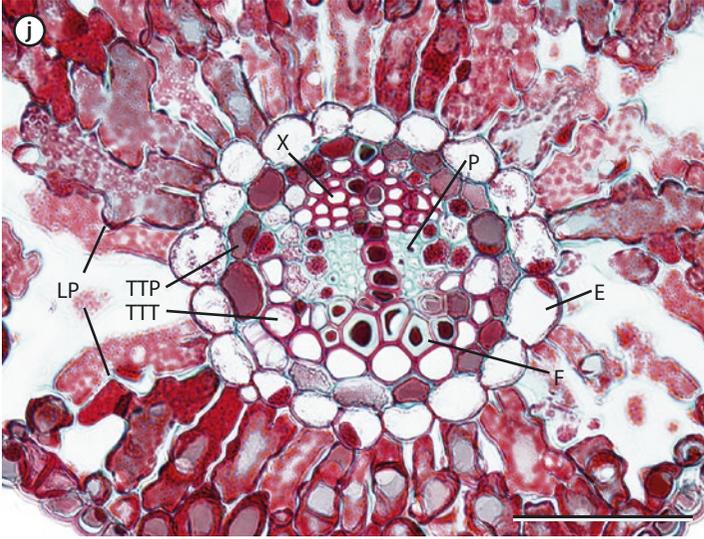
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⬛ Fig. 12.9   h, i Resin ducts in leaves of balsam fir (*Abies balsamea*) as seen in the light microscope **h** and the scanning
electron microscope **i**. The resin is secreted by the layer of epithelial cells (E). Scale bar in **i** = 100 μm and applies to both
panels (**h, i** RR Wise)

☐ **Fig. 12.9** **j** Central fibrovascular bundle from a tamarack (*Larix laricina*) leaf. The bundle contains xylem (X), phloem (P), fibers (F), and transfusion tissue (TT) and is bounded by an endodermis (E). The transfusion tissues consist of living parenchyma (TTP) and dead tracheids (TTT). Lobed parenchyma cells (LP) lie in the mesophyll. Scale bar = 100 μm (RR Wise)

contain the vast majority of protein in a leaf, and the valuable amino acids in those proteins must be recovered. Plants undergoing autumnal senescence use the environmental cues of shortening day length and cooler temperatures to initiate chloroplast breakdown, resulting in the conversion of that organelle to a gerontoplast (refer to ► Sect. 3.5.6). At the end of that process, the leaf is shed or abscised. The chloroplast-to-gerontoplast conversion begins prior to and accompanies the process of abscission. If timed correctly (the coordinated processes can be disrupted by a late summer drought, a heavy fall rainstorm, or an early winter freeze), all of the leaf protein will have been removed and recovered before the leaf is ultimately shed.

Leaf abscission starts with the formation of a specialized separation layer of mitotically active cells called an **abscission zone** (☐ Fig. 12.10a, b). Abscission zones are found at the site of connection of petioles with the stem. This layer of cells defines a transverse rupture zone. Similar zones are also found at the sites of connection for flowers and fruits, because they abscise as well. It typically starts with cell divisions that take place perpendicular to the axis of the petiole. The growth of the abscission layer does not extend through the vascular tissues. It is a region that has parenchyma cells with thin walls, tracheids instead of vessels, and a near lack of fibers. Differentiation of this zone may be early, or it may start just before activation. Initiation of the abscission zone involves the action of the plant growth regulator ethylene (a gas), to stimulate degradation of the middle lamella and hydrolysis of the cellulosic walls. Since lignin is not affected, everything at the juncture site is eventually severed except for xylem.

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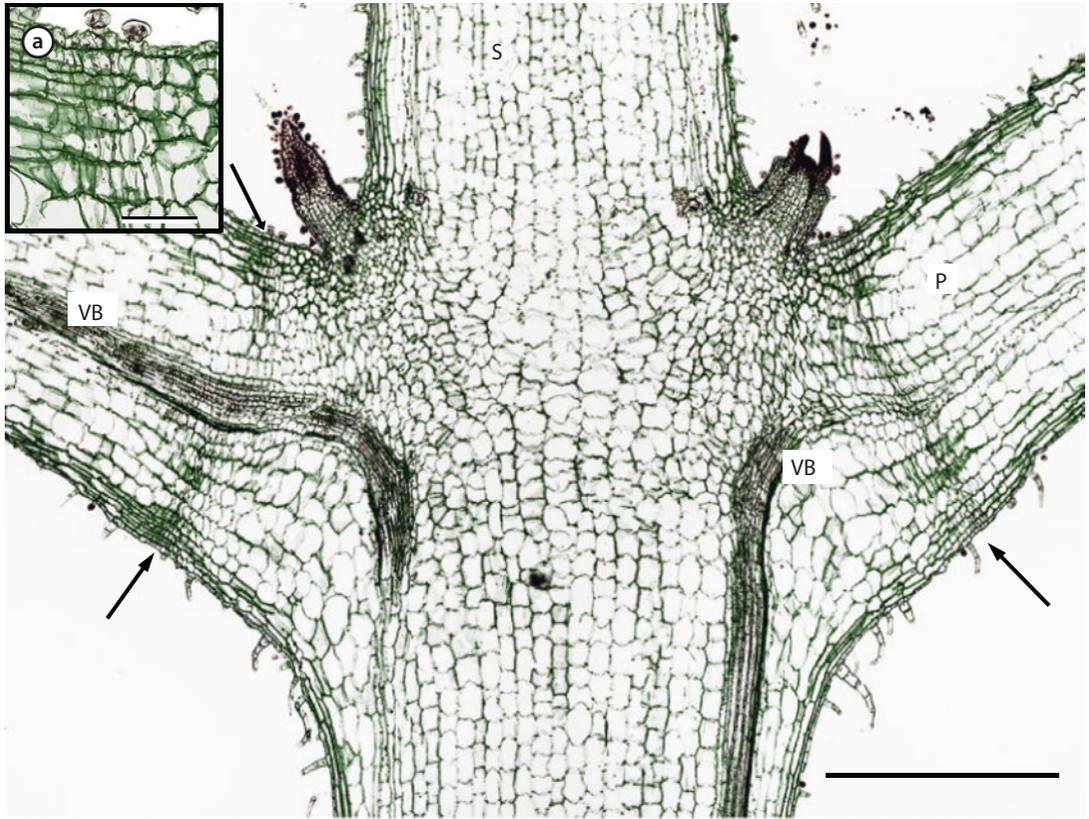
☐ **Fig. 12.10   a** General view of early abscission zones (*arrows*) in coleus (*Plectranthus scutellarioides*) leaves extending off a central stem (S). Vascular bundles (VB) can be seen. The one to the left extends into the petiole (P). A similar connection is not revealed, but exists, to the petiole on the right. Inset at top left shows the *tabular* cell shape of the early abscission zone. Scale bars = 500 μm in main panel and 50 μm in inset (RR Wise)

☐ **Fig. 12.10   b** Higher magnification view of an early abscission zone in a black walnut (*Juglans nigra*) petiole. Stem is to the left and petiole to the right. Scale bar = 250 μm (RR Wise)

◘ Fig. 12.10   c Mature abscission zone in American elm (*Ulmus americana*). Stem is to the left and petiole is to the right. The separation layer (SL) of thin-walled cells is bordered by protective corky layers (CL) to either side. Note that the vasculature (V) has not fully separated yet and that the corky layer is thicker on the stem s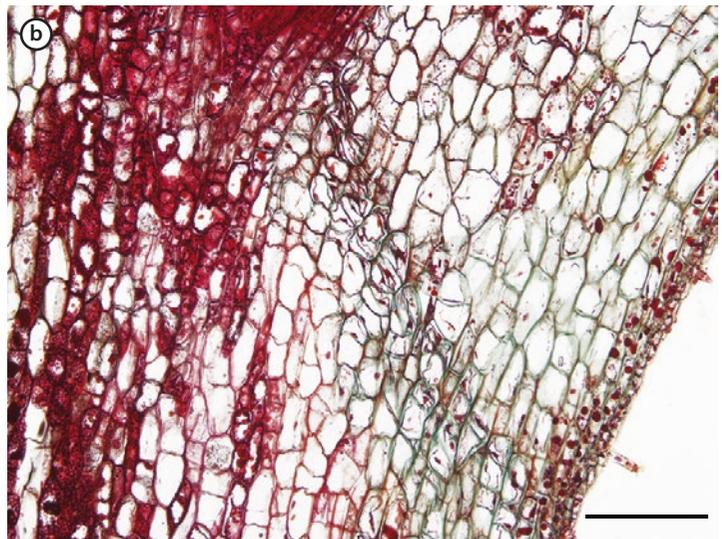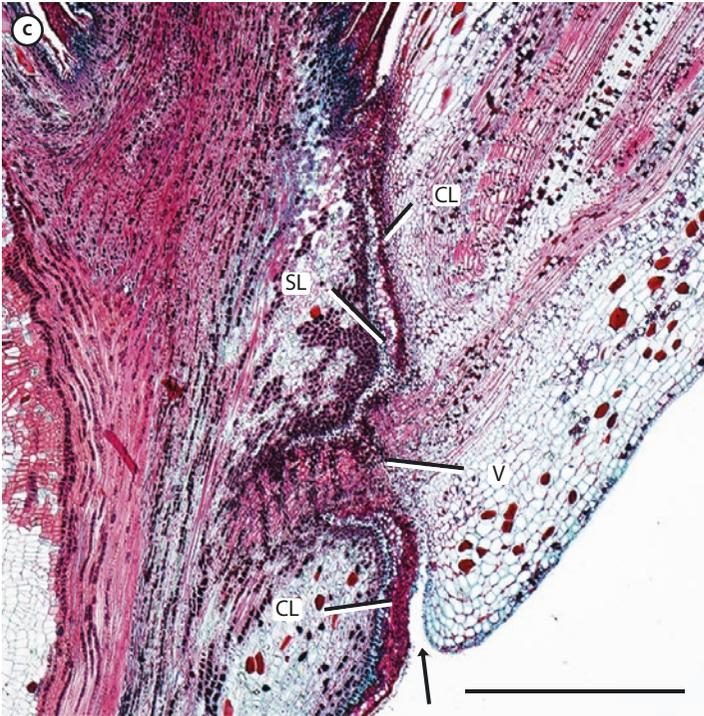ide than on the petiole side. Separation has begun at the bottom of the petiole (*arrow*) and will eventually propagate through the zone and result in leaf abscission. Scale bar = 500 μm (RR Wise)

As the abscission process progresses, the abscission zone is subdivided into a **separation layer** of thin-walled empty cells through which the actual break occurs and two **corky layers** at both sides of the separation layer (◘ Fig. 12.10c). Corky layers are cork cells that appear dark brown due to the deposition of specific "protective" substances (e.g., suberin) in their cells. Physical force, in time, breaks the xylem. The formation of the weak abscission zone prevents injury to the living tissues in the stem, and the cork layer left behind protects the newly exposed stem surface from desiccation and infection by microorganisms. Abscission of the leaf will leave a **leaf scar** appearing on the side of the stem (◘ Fig. 12.10d). The protective layer on the stem side is gradually transformed in each leaf scar with further periderm development beneath the scar.

ⓓ

Lateral bud

Leaf scar

Bundle
scars

Lenticels
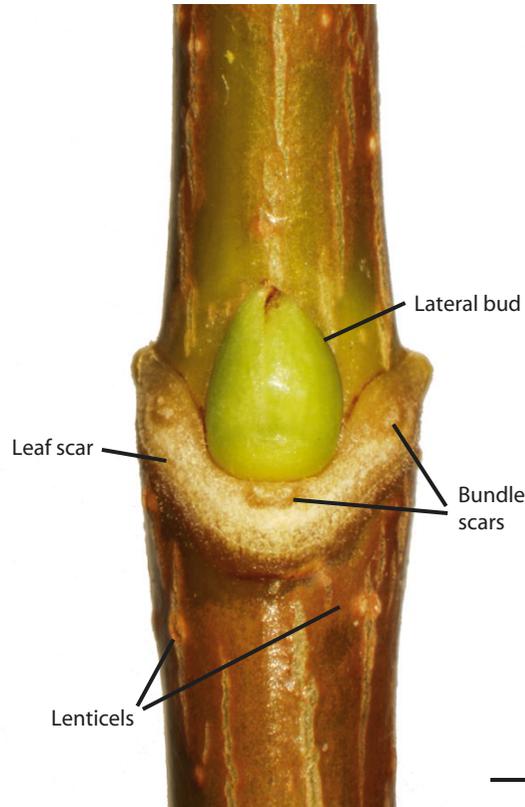
⏺ **Fig. 12.10   d** A leaf has recently abscised from this node on a red maple
(*Acer rubrum*) stem. A single leaf scar shows where the petiole was attached,
and bundle scars mark the location of vascular bundles. The lateral (axillary)
bud will be the source of next year's leaf. The stem has multiple lenticels for
gas exchange. Scale bar = 2 mm (RR Wise)

12

12.11 **Chapter Review**

▪   **Concept Review**

12.1   *Leaves have a variety of shapes and functions.* Leaves arise
from the nodes of the stem, and while most leaves are the
plant's green, planar photosynthetic organs, other shapes
and functions have evolved. Gymnosperm leaves are
scalelike or needlelike. Some monocot leaves form a large
portion of the plant "stem." Other leaves serve as protective
spines or coverings, food or water storage organs, tendrils
for attachment, or carnivorous traps. Some plants lack
leaves entirely.

12.2   *The vasculature system spreads throughout the leaf.* Enations
are small outgrowths on the stems of primitive plants that
lack a vascular connection to the stele in the stem. Micro-
phylls are likewise small and primitive, but their vasculature
is connected to that of the stem. Subsequent evolution led to
the macrophyll with a branching system of veins seen in
most extant plant species. Ferns and *Ginkgo* have a simple,
dichotomously branched vascular system. Monocots have
parallel venation and eudicots have reticulate venation.

Vascular bundles have bundle sheaths that control the flux of water and photosynthate between the xylem, phloem, and leaf mesophyll. In heterobaric leaves, bundle sheath extensions extend to the leaf surface and divide the leaf into separate gaseous compartments. Homobaric leaves lack the extensions. Collenchyma and fibers surrounding the vasculature may provide structural support to the leaf.

12.3 *Leaf morphology is optimized for light absorption, gas exchange, and water conservation.* The prototypical eudicot leaf has a double-layer photosynthetic mesophyll divided into the palisade mesophyll and the spongy mesophyll, in an arrangement called bifacial. The leaf surfaces are bounded by two epidermises which contain stomata and trichomes. An isobilateral leaf only has palisade or spongy mesophyll. Unifacial and tubular leaves have one face, typically the adaxial, fused so that both faces are abaxial. Centric leaves are round and solid in the middle; tubular leaves are round and hollow in the middle.

12.4 *The light environment during development can modify leaf anatomy.* In plants with large foliage crowns, such as many trees, leaves that develop inside the canopy do so under shaded conditions and leaves that develop at the periphery of the canopy are exposed to high light during development. This leads to different leaf morphologies that take advantage of the availability of light. "Shade" leaves are thicker overall with a greater proportion of palisade than spongy mesophyll. "Sun" leaves are the opposite, being thinner and a lower ratio of palisade thickness to spongy thickness.

12.5 *Kranz leaf anatomy is a specialization of the C4 photosynthetic pathway.* C4 photosynthesis requires a spatial separation of the initial carbon fixation steps and the final carbon fixation steps. In many C4 plants, the initial fixation is performed in the mesophyll cells and the final in the large, conspicuous bundle sheath cells. This "halo" of large cells surrounding the vascular bundle is the source of the term "Kranz" anatomy. Warm season grasses have a single layer of bundle sheath cells. Cool season grasses have a double layer, the inner layer called the mestome.

12.6 *Xeromorphic angiosperm leaves conserve water in arid environments.* Xerophytes are plants adapted to water-limiting conditions and have a number of anatomical adaptations including a thick cuticle, dense covering of trichomes, sunken stomata, and the ability to roll their leaves via water loss from bulliform cells. The leaves are often sclerophyllous as a consequence of thick bundles of heavily sclerified fibers associated with the vasculature or scattered throughout the leaf.

12.7 *Succulent leaves are adapted for water storage.* Succulence is a morphology characterized by the storage of water in thick, fleshy stems or leaves. Many desert plants use this strategy, in addition to other xerophytic adaptations such as thick

cuticle and sunken stomata, to conserve water in extremely arid environments. Succulent leaves typically have their photosynthetic tissues close to the leaf surface and an interior of large, thin-walled, water-storing parenchyma. Other leaves develop one or more water-storing hypodermises lying just underneath the epidermis.

12.8 *Aquatic angiosperms have hydromorphic leaves.* Hydrophyte plants are adapted to an aquatic habitat. Their leaves may be submerged, floating, or emergent. Submerged leaves may be only 2–3 cell layers thick and have a thin cuticle with little to no vasculature. Floating leaves are epistomatous with stomata on the top (adaxial) surface only. A palisade parenchyma underlies the adaxial epidermis while large, air-filled aerenchyma tissue occupies the bottom portion of the leaf. Emergent leaves have both an aerenchyma as well as large open ducts called lacunae that provide a diffusional pathway for oxygen transport to the submerged roots. They may also have sclerenchyma and vasculature.

12.9 *Xeromorphic gymnosperm leaves conserve water during winter.* Evergreen conifer trees experience drought conditions during their overwintering phase. Therefore, their leaves have anatomical adaptations similar to those found on other xerophytes.

12.10 *Leaf abscission is a timed and genetically controlled process.* Plants may shed their leaves upon the imposition of seasonal drought or winter freezing in a process called abscission. The formation of an abscission zone in the leaf petiole is preceded and accompanied by a reallocation of the leaf protein and recovery and export of those products from the leaf to the stem. Cells in the abscission zone form three layers, a middle, thin-walled weak layer called the separation layer which is bounded by two thicker, corky layers, one on either side. Eventually, the separation layer will fail and the leaf will abscise. The scar on the stem left by the leaf will be sealed by the corky layer, which will prevent desiccation and block microbial invasion.

12

■ **Concept Connections**

❓ 1.   Match the leaf type to the function and the example given in the text.

Leaf type	Function	Example given in text
a. Foliage	i. Support	a. Eudicotyledon seed
b. Leaf sheath	ii. Photosynthesis	b. Cacti
c. Cataphyll	iii. Antiherbivory	c. Many grasses
d. Frond	iv. Photosynthesis	d. Monocotyledon seed
e. Pseudostem	v. Protection	e. Flowering plants
f. Scutellum	vi. Photosynthesis	f. *Rosa* sp.

Leaf type	Function	Example given in text
g. Needle	vii. Food storage	g. Ferns and duckweeds
h. Spine	viii. Floral structures	h. Bud scales
i. Cotyledon	ix. Support	i. Conifers
j. Hypsophyll	x. Attachment	j. *Clematis* sp.
k. Tendril	xi. Absorption	k. *Musa* sp.

■ **Concept Assessment**

2. Hypostomatous leaves have
   a. no stomata.
   b. paracytic stomata.
   c. stomata on abaxial epidermis.
   d. stomata always open as a part of a hydathode.
   e. a lack of stomatal chambers.

3. An areole is
   a. a leaf mesophyll region bounded by vascular tissues.
   b. an air space within the mesophyll of a leaf.
   c. the opening of a stoma.
   d. an enlargement of a petiole.
   e. a leaf separated into different regions by bundle sheath extensions.

4. Mestome sheath cells
   a. possess large chloroplasts with starch.
   b. have thickened walls.
   c. function as bulliform cells.
   d. encircle only the phloem of leaf veins.
   e. are commonly found in C3 plants.

5. Leaf abscission is early recognized by the appearance of
   a. separation of the vascular tissues.
   b. an accumulation of starch at the site of abscission.
   c. meristematic activity of the protoderm.
   d. cell division parallel to the axis of the petiole.
   e. tabular cells in the abscission zone.

6. What plant regulator growth regularly helps promote the formation of leaf abscission?
   a. gibberellin.
   b. abscisic acid.
   c. ethylene.
   d. auxin.
   e. florigen.

? 7. When the two sides of mesophyll of a leaf are similar in
      structure, the leaf is said to be
      a. hypostomatous.
      b. mesophytic.
      c. dorsiventral.
      d. unifacial.
      e. tubular.

? 8. Commissural veins are found
      a. in eudicots.
      b. connecting parallel veins in monocots.
      c. as leaf midveins.
      d. only in petioles.
      e. where there is dichotomous vein branching.

? 9. "Closed" minor veins lack
      a. secondary walls.
      b. a bundle sheath.
      c. companion cells.
      d. plasmodesmata.
      e. protoplasmic structure.

? 10. What is missing in leaves with Kranz anatomy?
      a. bundle sheath.
      b. palisade mesophyll.
      c. spongy mesophyll.
      d. stomata.
      e. vascularization.

? 11. Bulliform cells
      a. enable the leaf to fold.
      b. are found in the endodermis.
      c. are highly photosynthetic.
      d. are characteristic of hydromorphic plants.
      e. are derived from procambium.

■   **Concept Applications**

? 12. Compile and discuss the xeromorphic adaptations found in
      the *epidermis* of the oleander (*Nerium oleander*) leaf.

? 13. Few gymnosperms occupy desert or arid ecological niches.
      However, most gymnosperms have leaves with multiple
      xeromorphic anatomical adaptations. Why is this so?

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