CHAPTER 18

Gnetophyta

EPHEDRA, GNETUM, AND WELWITSCHIA

THE Gnetophyta is a small group of seed plants represented by three living genera, *Ephedra, Welwitschia,* and *Gnetum.* The distinctive organography and anatomy of the sporophyte and the many peculiar features of the reproductive cycle of these genera have attracted the attention of morphologists for more than a century and have resulted in a voluminous literature. Martens (1971) brought together in a single volume monographic treatments of each of the three genera of the gnetophytes and examined critically the many conflicting theories that have been advanced to explain their interrelationships and their affinities with other groups of living seed plants.

Throughout much of the literature on the gnetophytes, repeated efforts have been made to demonstrate that these plants form a "connecting link" between gymnosperms and angiosperms (Arber and Parkin, 1908). Among the "angiospermic" features usually mentioned in defense of this idea are (1) the compound nature of *both* the microsporangiate and megasporangiate strobili, which have been intepreted as "inflorescences" by some botanists, and (2) the presence of vessels in the xylem, a feature that has been regarded as a major divergence from the vessel-less wood characteristic of the cycads, *Ginkgo biloba,* and all members of the Coniferales.

The presumed isolation of the gnetophytes from all living and extinct gymnosperms is clearly emphasized in Pulle's (1938) classification of the major groups of seed plants, in which *Ephedra, Welwitschia,* and *Gnetum* are segregated under the subdivision "Chlamydospermae," which is placed *between* the "Gymnospermae" and "Angiospermae." The word Chlamydospermae (from the Greek words meaning "seeds with an envelope or cloak") refers to the so-called outer integument of the ovule, which has been interpreted as a pair of fused appendages or "bracteoles" (see Fig. 18-13, B, outer envelope). In this connection, Martens (1971) astutely observed that an ovule provided with an accessory envelope is not "naked" in the sense employed to characterize the ovules of gymnospermous plants. Some authors have even compared the external envelope of the ovule of the gnetophytes to the "ovary" of the angiosperms.

For many years the existence of gnetophytes in the fossil record was based entirely upon the presence of pollen grains resembling those of *Ephedra* and *Welwitschia.* Recently, Crane and Upchurch (1987) discovered megafossils (leafy shoots) from the Lower Cretaceous that have gnetalean-type organization. The fossils are associated with masses of the *Welwitschia*-type pollen. In the past, *Ephedra,* *Welwitschia,* and *Gnetum* were placed in the family Gnetaceae under the order *Gnetales.* The tendency now is to split this old order into three orders, namely, Ephedrales, Welwitschiales, and Gnetales, each consisting of a single family and a single genus (see Eames, 1952, and Martens, 1971). We have adopted this systematic treatment and have grouped the three orders, for convenience, under a single division, the Gnetophyta.

Geographical Distribution and Habit

The genus *Ephedra,* consisting of about thirty-five species, is confined to cool, usually arid regions in both the Eastern and Western Hemispheres (Fig. 18-1). In the New World, *Ephedra* is restricted to western North America, parts of Mexico, and a wide area in South America. According to Cutler's (1939) monograph, the sixteen species found in the United States occur in dry or desert areas of California, Nevada, Utah, Arizona, and New Mexico. Most species of *Ephedra* are profusely branched shrubs (Fig. 18-2), although a few are scandent, and one species, *Ephedra triandra,* native to Brazil, Uruguay, and Argentina, is a small tree.

In marked contrast, *Gnetum* inhabits tropical rain forests in parts of Asia, northern South America, Africa, and certain Pacific islands between Australia and Asia (Fig. 18-1). Most of the thirty or more described species of *Gnetum* are lianas that climb high into the crowns of various trees in the rain forest; one species, *Gnetum gnemon,* becomes a small tree (Maheshwari and Vasil, 1961).

The most bizarre and geographically restricted member of the Gnetophyta is the African genus *Welwitschia* consisting of the single species *Welwitschia mirabilis* (Fig. 18-3, A, B). The specific epithet "mirabilis" (Latin for "wonderful" or "marvelous") is quite appropriate because an adult plant, in habit and general organography, is unlike that of any known plant on earth. The exposed portion of an old plant consists of a short, woody, unbranched stem and a massive, woody, concave, "disc" or "crown" that bears only two huge strap-shaped leaves as the *permanent* photosynthetic organs of

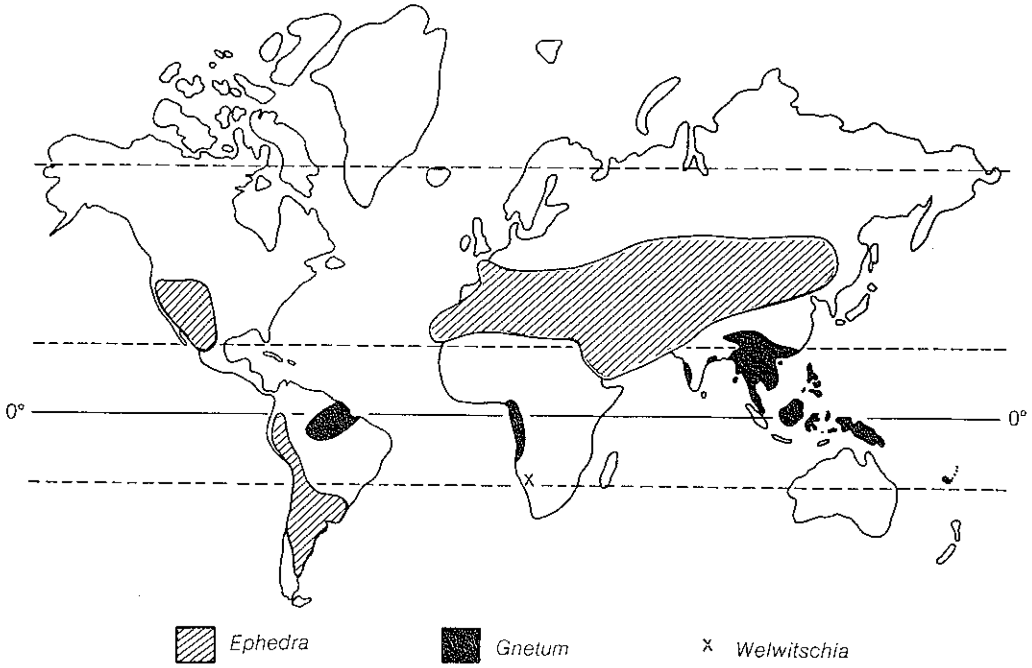

FIGURE 18-1 Present distribution of the three genera of the gnetophytes. See text for further explanation. [Based on Hutchinson, *Kew Bulletin of Miscellaneous Information,* No. 2, 1924. By permission of Her Majesty's Stationery Office.]

FIGURE 18-2 *Ephedra* sp. photographed near Monitor Pass, California. Note the large size of the basal stems of this shrubby species. [Courtesy of Dr. T. E. Weier.]

the plant (Fig. 18-7). These leaves become split and frayed in old plants, extending in a twisted and arching manner over the ground, and the tips become seared where they touch the ground. The leaves are perpetuated at the rate of 8 to 15 centimeters a year from a basal intercalary meristem. Bornman (1972) found that a leaf of one giant had an unbroken width of 1.8 meters and a length of 6.2 meters of which 3.7 meters were of living tissue. It is estimated that some plants may be 1,500 to 2,000 years old!

Welwitschia occurs in the Namib Desert near the coast of Angola and South-West Africa (Namibia) as well as inland to about 150 kilometers in a savannalike region. Rainfall of the Namib Desert varies from 0 to 100 millimeters per year whereas rainfall in the interior can exceed 200 millimeters. During the night for about nine months of the year, fog is blown inward from the coast to a distance of about 80 kilometers, but it dissipates by about 10:00 a.m. Moisture from the fog accumulates on the leaves. How does *Welwitschia* solve the problem of water economy and water uptake in a desert enviroment? *Welwitschia* has a large tap root that may extend downward for 1 to 1.5 meters before it divides into numerous thin roots. How far down the thin roots extend is uncertain. Some investigators believe that water from the fog enters the leaves, but the exact method, if it occurs, is unknown.

Many desert plants have a type of photosynthesis adapted for existence in a hot, dry desert environment. Stomata are open at night when the air is cool, and carbon dioxide is absorbed and fixed in organic acids. During the early daylight hours the carbon dioxide is released, not to the atmosphere because the stomata are now closed to prevent water loss by transpiration, but to chloroplasts of the mesophyll cells where it is transformed first into sugars and then into starch. *Welwitschia* has been described as having this type of photosynthesis — termed CAM (crassulacean acid metabolism) photosynthesis (Dittrich and Huber, 1975). In the more common type of photosynthesis, the carbon dioxide enters through stomata during the daylight hours and is fixed in the chloroplasts and then converted to sugars. The latter type of photosynthesis has been reported to occur in *Welwitschia* in the eastern part of its range where the climate is more moderate (Schulze and Schulze, 1976).

Recent physiological studies of plants from a variety of natural habitats support the belief that *Welwitschia* is not basically a CAM plant. Von Willert (1985) reported that carbon dioxide is taken up during the day through open stomata, leading to a tremendous loss of water by transpiration. A high transpiration rate prevents the occurrence of lethal leaf temperatures. These observations indicate that *Welwitschia* can tap water supplies in the soil not exploited by its associates. Von Willert (1985) sums up the enigma of *Welwitschia*: "If botanists should construct a plant best adapted to a desert environment, they would never come up with a monster like *Welwitschia mirabilis*."

For interesting and readable accounts of the morphology, anatomy, and physiology of *Welwitschia* see the publications of Bornman (1972, 1978).

Welwitschia is dioecious, and the microsporangiate and megasporangiate cones are borne terminally on ramified branch systems. As Martens (1971) emphasizes, these strobiliferous shoots represent the *only authentic branches* of this amazing plant (Figs. 18-22 A; 18-23). *Welwitschia* was discovered in 1860 in Angola by Dr. Frederic Welwitsch, in whose honor the plant was named by the

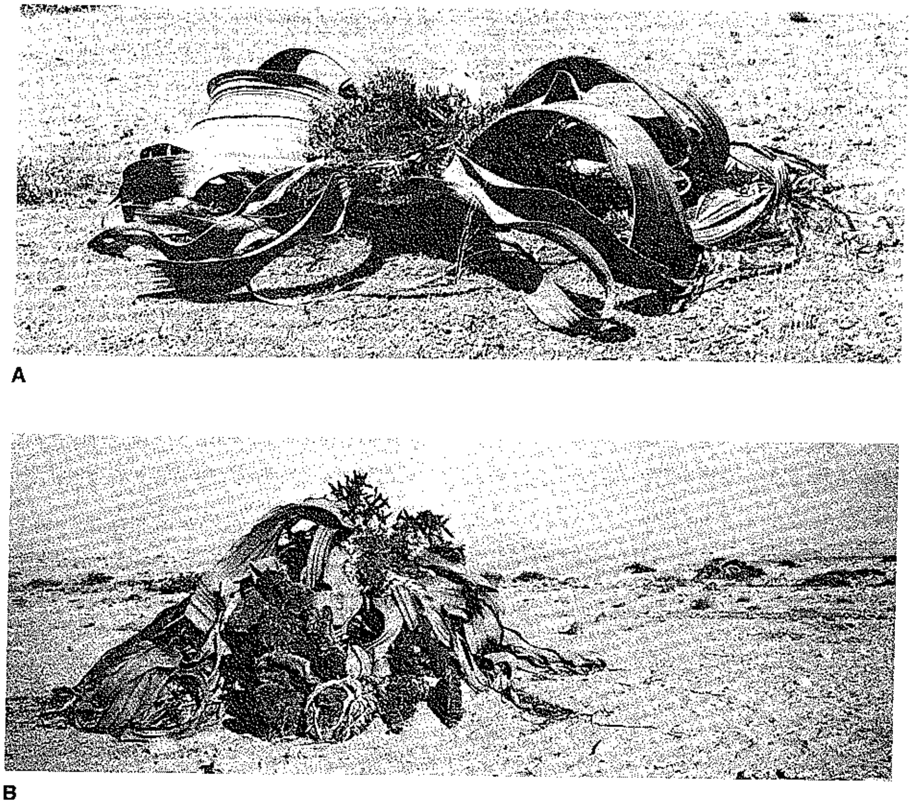

FIGURE 18-3 *Welwitschia mirabilis.* A, microsporangiate specimen growing in a desert area near Brandberg in South-West Africa (Namibia). Each of the two huge leaves has become split into a series of contorted and buckled "segments." Note the numerous clusters of strobili in the leaf axils at the periphery of the "woody" crown. B, extremely old plant growing on "*Welwitschia*-Flats," Namib Desert, South-West Africa; note fog in the background. [A, courtesy of Dr. R. J. Rodin; B, photograph by Dr. C. H. Bornman.]

British botanist, J. D. Hooker (1863). Dyer and Verdoorn (1972) claim that *Tumboa bainesii* is the valid name. However, the binominal *Welwitschia mirabilis* will undoubtedly continue to be used in descriptions of this bizarre plant; the name has been conserved in the International Code of Botanical Nomenclature, 1983.

Vegetative Organography and Anatomy

Despite the voluminous literature, which has been reviewed by Pearson (1929), Chamberlain (1935), and Martens (1971), much still remains to be done before an adequate treatment of the sporophytes of gnetalean plants as a whole can be accomplished. *Gnetum* and *Welwitschia* are both extremely complex anatomically and require further ontogenetic study.

The Leaf

The foliage leaves of *Ephedra, Gnetum,* and *Welwitschia* are strikingly different in form and venation and provide morphological characters that are definitive for each of the three genera.

In *Ephedra,* the phyllotaxis varies from *decussate* to *whorled,* and the leaves are basally joined by a membranous commissure to form a more-or-less conspicuous sheath at each node (Fig. 18-4, B). Throughout most of the literature on *Ephedra,* the leaves are characterized as "reduced" or "scalelike"

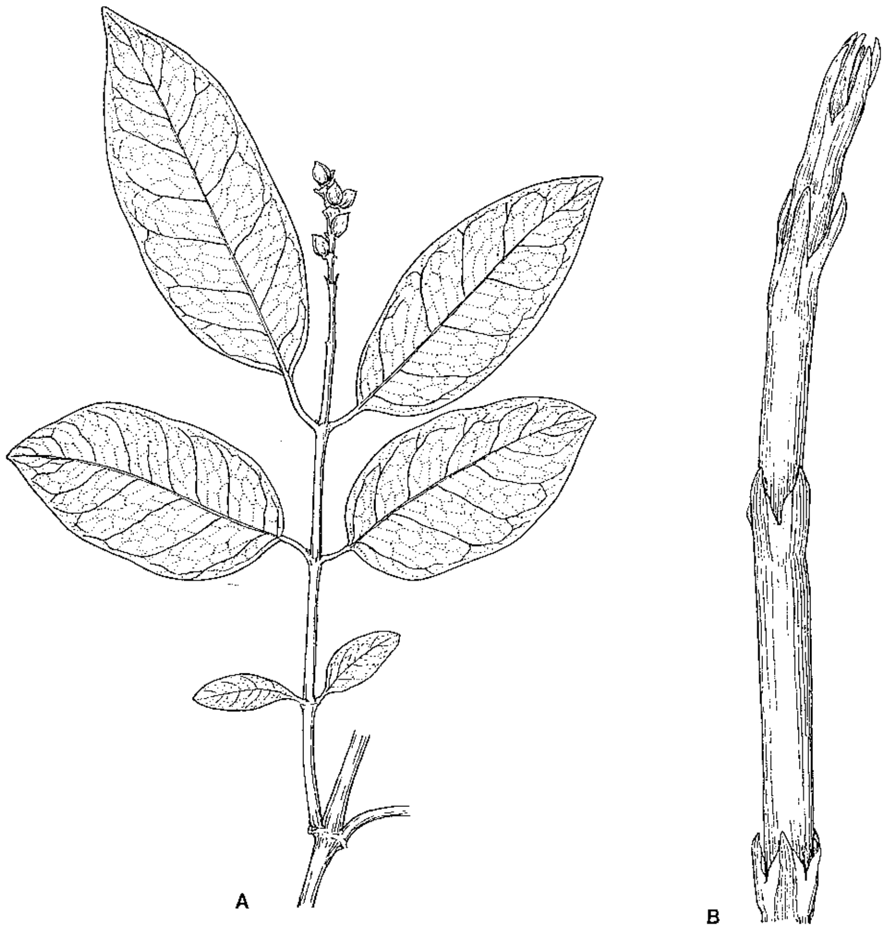

Figure 18-4 A, *Gnetum indicum.* Terminal portion of shoot showing seeds and three pairs of simple leaves with pinnate-reticulate venation (actual size); **B**, *Ephedra* sp. Tip of a vegetative shoot, illustrating the whorled arrangement of the reduced scalelike leaves (× 4).

in form. This generalization, however, needs to be qualified because in several species (e.g., *Ephedra foliata* and *Ephedra altissima)* the lamina is slender and needlelike, and, according to Stapf (1889), may attain a length of 3 centimeters and a width of 1 to 1.5 millimeters (Fig. 18-5, A). There also is considerable variation in the proportional development of the sheath and lamina in the successive leaves of a single shoot. In *Ephedra foliata,* for example, the leaves in the middle or upper part of a shoot may have well-developed laminae whereas the basal leaves of the same shoot tend to be much smaller, with laminae only 3 to 4 millimeters long (compare A and B in Fig. 18-5). In this and other species, there is obviously no sharp boundary between "well-developed" and "reduced" scalelike leaves. It is widely stated in the literature that the leaf in *Ephedra* is vascularized by a pair of traces that neither branch nor anastomose in their course through the sheath and lamina. The stem is eustelic, and each leaf is supplied by a trace from two adjacent sympodia (Marsden and Steeves, 1955). However, in *Ephedra chilensis* and *Ephedra fragilis* one of the leaves at a node may be three-veined. In a few instances, both members of a leaf pair are three-veined (Foster, 1972). Additional studies of the ontogeny and vascularization of three-veined leaves are needed.

The foliage leaf of *Gnetum* presents a complete morphological contrast to the leaf of *Ephedra.* In

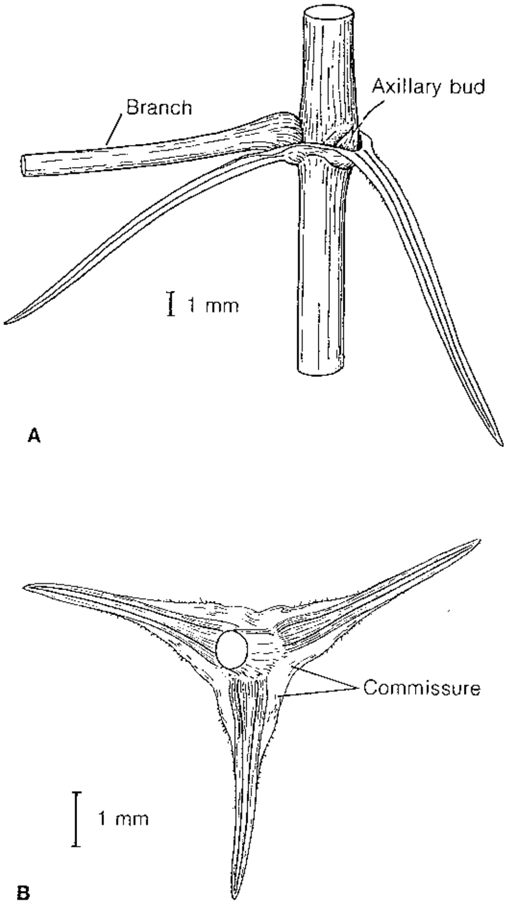

Figure 18-5 Variation in size and morphology of the leaves in *Ephedra foliata*. **A**, leaves with well-developed laminae and weakly connate sheaths; the leaf at the right is traversed by two freely terminating veins; **B**, whorl of three scale-leaves, detached from shoot and drawn from lower surface. Each leaf is vascularized by a pair of veins and is basally connected, by a prominent commissure, with adjacent leaves of the whorl. [From Foster, *Jour. Arnold Arboretum* 53:364, 1972.]

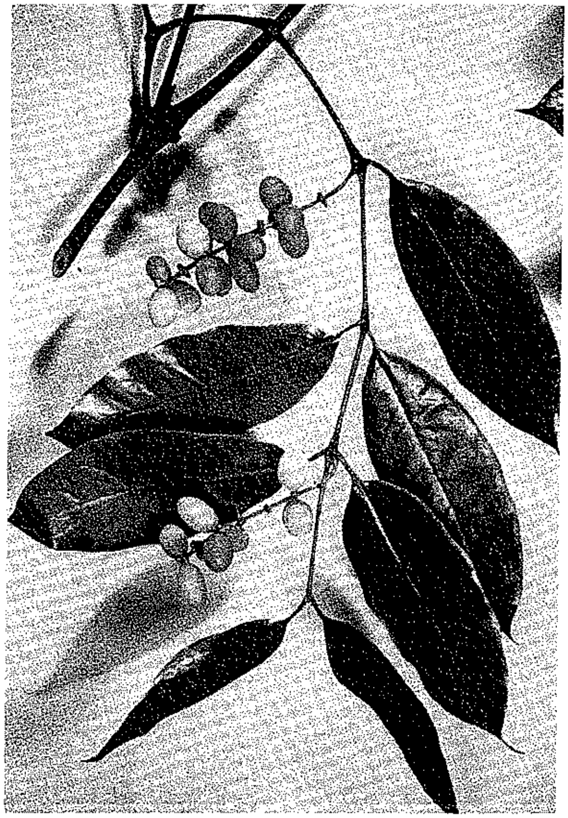

Figure 18-6 *Gnetum gnemon*, portion of branch system showing oppositely arranged leaves, and seeds. [General Biological Supply House, Inc., Chicago, Illinois.]

Gnetum the foliage leaves are arranged in pairs, and in their form and pinnate-reticulate venation bear a striking resemblance to the simple leaves of many dicotyledons (Figs. 18-4, A; 18-6). Very little comparative information is available regarding the number of leaf traces that vascularize the leaf in the various species of *Gnetum*. Rodin and Paliwal (1970) found that usually an *odd number of traces* (five to seven, or sometimes eight) extend into the leaf of *Gnetum ula*, but whether this type of nodal anatomy characterizes the genus as a whole is unknown. According to Rodin (1967), the five to seven leaf traces in *Gnetum gnemon* extend into the midrib of the young leaf as a series of longitudinal bundles that, at successive levels, dichotomize and form *pairs of secondary* veins that curve toward the margins of the lamina. In the submarginal region, each secondary vein bifurcates and the derivative branches unite, creating a series of coarse "meshes" at each side of the midrib. Within each mesh, a more delicate reticulum of veinlets is later differentiated. (For additional information and illustrations of venation patterns in several other species of *Gnetum*, see Rodin, 1966.)

The vegetative organography of *Welwitschia*, as we have already noted, is without parallel among all living vascular plants. Except for the two shortlived cotyledons, produced during the seedling phase, the *permanent* photosynthetic organs are represented by a single pair of enormous strap-shaped leaves, which continue to grow indefinitely in length through the activity of a meristem located at the base of each appendage (Fig. 18-7). According to Martens (1971), the young leaf of *Welwitschia* is at first vascularized by two and then four strands, and additional lateral bundles continue to differentiate as the leaf increases in width basally. A well-developed leaf in *Welwitschia* is traversed by numerous "parallel" longitudinal veins that become interconnected by smaller obliquely oriented veins. These may anastomose in various ways to form irregular areoles (i.e., meshes) or may fuse in pairs and terminate blindly in the mesophyll. This peculiar and highly distinctive type of venation also occurs in the blades of the cotyledons and is unique as compared with the venation of *Ephedra, Gnetum,* and other gymnosperms (see Rodin, 1953, 1958a, 1958b).

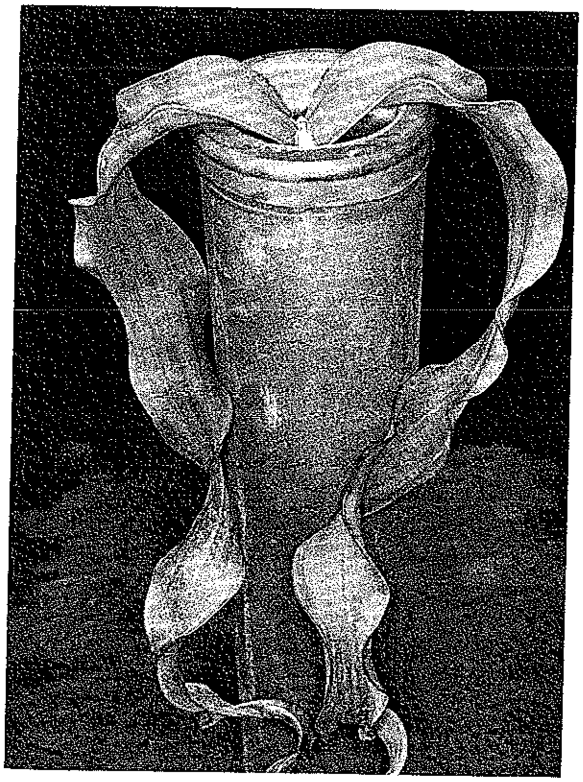

FIGURE 18-7 *Welwitschia mirabilis,* young sporophyte showing the two permanent foliage leaves. The specimen has been grown in a section of pipe to provide space for the development of the long tap root. [Courtesy of Dr. T. E. Weier.]

As described previously, stomata of the syndetocheilic type occur on both surfaces of a *Welwitschia* leaf and are somewhat sunken, although not as deeply as in most desert plants. Also, the cuticle is relatively thin in comparison with most xerophytes. Strands of thick-walled fibers of great tensile strength are present beneath the epidermis, and ramified sclereids occur in the mesophyll. The vascular bundles are collateral. The xylem consists of tracheids and vessels; the vessel members have large perforations in the end walls. The phloem consists of sieve cells and parenchyma; the sieve areas are much like those of conifers, and the nucleus of a sieve cell remains intact at maturity. The entire vascular bundle is surrounded by a layer of transfusion tissue (Evert et al., 1973a, b).

The Shoot Apex

Comparative studies have revealed a remarkable similarity in the *basic structure* of the shoot apex of the gnetophytes: in all three genera a more-or-less discrete surface layer, or *tunica,* is present, the con-

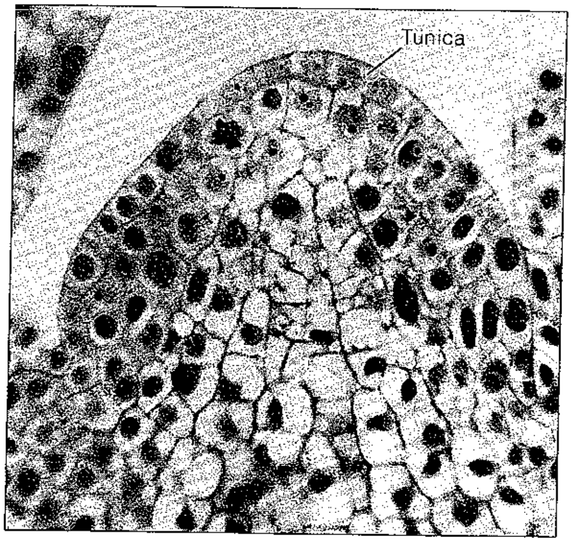

FIGURE 18-8 Median longisection of the shoot apex of *Ephedra altissima.* Note the single, clearly defined tunica.

tinuity of which is interrupted only by occasional periclinal divisions. The investigations of Gifford (1943) on *Ephedra altissima* and Seeliger's (1954) study on *Ephedra fragilis* var. *campylopoda* emphasized the infrequency of periclinal divisions in the surface layer of the apex and its tunicalike structure and growth (Fig. 18-8). Johnson (1950), in a study of eighty-five apices of *Gnetum gnemon*, found "no evidence of periclinal divisions in the tunica clothing the summit of the dome." The aberrant behavior of the shoot apex in *Welwitschia* presents a striking contrast to the "open system" of growth characteristic of *Ephedra* and *Gnetum*. According to the detailed study made by Martens and Waterkeyn (1963), the shoot apex of the seedling of *Welwitschia* first initiates a pair of primordia which

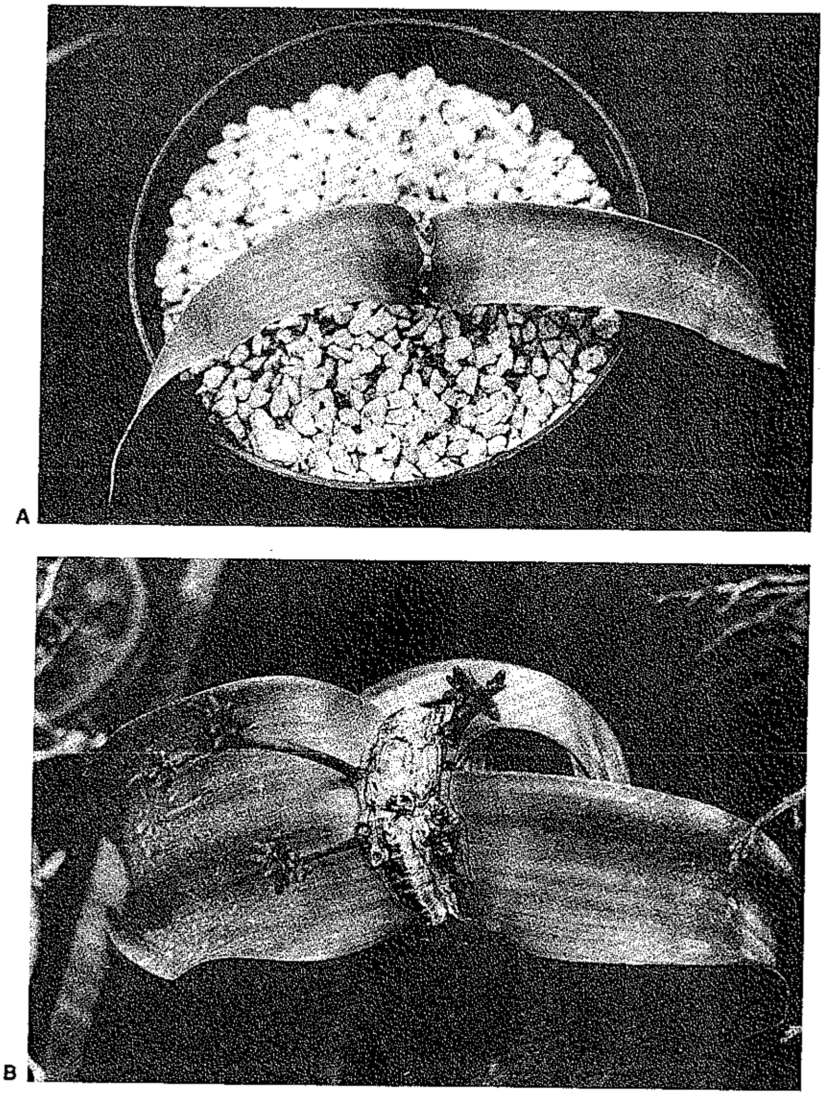

FIGURE 18-9 *Welwitschia mirabilis.* A, young plant showing the two permanent leaves and the two opposite scaly bodies at the bases of the leaves. B, older plant showing the two leaves and strobiliferous branches that arise from the basal or intercalary meristem; note that the scaly bodies have become much larger than in A, and the leaf to the right is split down to the basal meristem.

later develop into the two permanent leaves of the sporophyte. But contrary to the classical interpretation, an *additional pair* of appendages — the so-called scaly bodies — are formed by the apex in a plane at right angles to the plane of the foliage-leaf primordia (Fig. 18-9, A, B). Martens and Waterkeyn reject the prevalent view that the scaly bodies are cotyledonary buds, and interpret them instead as a *third pair of foliar appendages.* Through the activity of a basal meristem the scaly bodies increase in size, and become conspicuous structures on plants several years old or more (Fig. 18-9, B). Soon after the formation of scaly bodies, the shoot apex loses its meristematic character and finally degenerates. Meristematic activity is now shifted to quite another region — to that of the leaf bases of the two permanent photosynthetic leaves. This activity results in the bilobed condition of the crown which, in very old plants, may become a concave disc surmounted by a band of meristematic tissue that contributes new tissue to the two large foliage leaves and provides concentric crests of tissue in which reproductive structures are initiated. The precocious death of the terminal meristem of the *young sporophyte* of *Welwitschia* is unique among vascular plants and is an additional example of the bizarre morphology of this extraordinary plant. Martens (1977) considers *Welwitschia* to be a plant that has lost its head!

The phylogenetic significance of the angiospermic organization of the shoot apex of the gnetophytes is, of course, problematic and very possibly the result of evolutionary convergence. Martens (1971) believes that the structural similarity of the apices of the three genera of gnetophytes supports the evolutionary position of this group at a level intermediate between those of the conifers and angiosperms.

Vessels

The presence of vessels in the Gnetophyta distinguishes them from other living gymnosperms and has often been used as an argument for their presumed evolutionary relationship to the angiosperms. However, it is generally believed that the *method* of origin of the perforations in the vessel members of the gnetophytes differs from that of all other vascular plants including the angiosperms. In the angiosperms, as well as in certain species of *Pteridium* and *Selaginella,* the intial step in vessel evolution was the loss of the primary cell walls from the *transversely elongated* bordered pits situated at each sloping end of a tracheidlike cell. Further elaboration of these slitlike perforations led to the development of vessel members with well-defined scalariform perforation plates. The most advanced vessel members in angiosperms possess large circular or oval simple perforations which originated by the elimination (phylogenetically and ontogenetically) of the bars between the slitlike openings. In contrast, as Thompson (1918) has shown, the initial step in vessel evolution in the Gnetophyta began with the loss of primary cell walls from a series of *circular* bordered pits located near the ends of long tracheidlike cells. In *Ephedra,* there are transitional conditions between intact bordered pits and *bordered foraminate perforations,* that is, there are clear transition forms between typical tracheids and vessel members (Fig. 18-10). The vessel members of *Gnetum* commonly possess large circular or elliptical simple perforations and thus markedly resemble the specialized vessel members of many angiosperms. Thompson (1918) was of the opinion, however, that the *Gnetum* type of vessel perforation resulted from the further enlargement of a series of circular perforations and the elimination of the portions of the end wall between them.

Thus in the initial steps of their origin as well as in their subsequent specialization, the vessels of the gnetophytes are thought to have evolved differently and independently from those in the angiosperms. As Bailey (1953) remarked, "although the highly evolved vessels of *Gnetum* resemble those of comparably specialized vessels of angiosperms, the similarity cannot be used as an indication of close relationship, but provides a very significant illustration of convergent evolution in plants." However, on the basis of a detailed study of the xylem of two species of *Gnetum,* Muhammad and Sattler (1982) have suggested that tracheary element morphology does not rule out the possibility that *Gnetum* may have been close to the ancestral stock of at least some angiosperms. In the two species, scalariform as well as circular pits occur. Scalariform perforation plates are present as well as the large simple perforations, and these compare well with those of selected angiosperms.

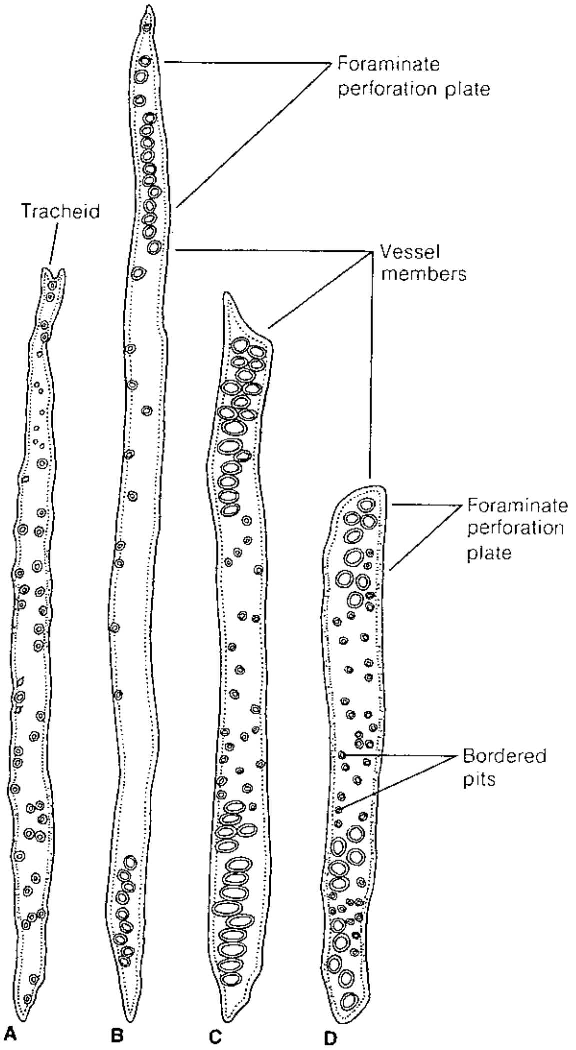

FIGURE 18-10 Tracheary elements from the secondary xylem of *Ephedra californica.* **A,** tracheid with numerous circular bordered pits; **B–D,** vessel members with foraminate perforation plates on end walls. [From *Plant Anatomy* by K. Esau. Wiley, New York. 1953.]

The Reproductive Cycle in *Ephedra*

By way of introduction we must comment briefly on the confusing "angiosperm-centered" terminology commonly used in describing the reproductive structures of the gnetophytes. To many writers the strobili of *Ephedra, Gnetum,* and *Welwitschia* are directly comparable with angiospermic inflorescences, and the parts of the gnetalean "flower" are very frequently designated by such terms as "perianth," "stamen," "anther," and "column." This kind of nomenclature is particularly confusing and misleading for *Ephedra,* which is notable for a morphology similar to conifers in its gametophytes and embryo; these structures are commonly described in the literature using "gymnosperm-centered" terminology. To eliminate the implication of homology between sporogenous structures of the Gnetophyta and those of angiosperms, the terms and general interpretations proposed by Eames (1952) will be adopted in the following resume of the life cycle of *Ephedra.*

The Strobili

Most species of *Ephedra* are strictly dioecious, and both the microsporangiate and ovulate cones are compound in structure; i.e., the cone axis bears pairs of bracts which subtend either microsporangiate or ovuliferous structures (Fig. 18-11). In certain monoecious species (e.g., *Ephedra campylopoda)* some of the strobili are *bisporangiate* with the microsporangiate structures developed in the axils of the lower bracts and the ovules located in the terminal part of the same cone.

The microsporangiate strobilus consists of a number of pairs of bracts; the lowest pairs are usually sterile, whereas each of the other pairs subtend microsporangiate shoots (Fig. 18-11; 18-12, A). The microsporangiate shoot consists of an axis bearing a pair of bracteoles, which enclose a stalked microsporophyll (or "microsporangiophore") with a number of sporangia (Fig. 18-12, B). The number of sporangia varies with the species. According to Eames (1952) the evidence from ontogeny and vascular anatomy indicates that the microsporangiophore of *Ephedra* is the result of the phylogenetic fusion of a pair of microsporophylls; in certain species, recognized as primitive by taxonomists, the two sporophylls are free and each bears a terminal cluster of four microsporangia.

The ovulate cone of *Ephedra* also consists of an axis bearing decussately arranged pairs of bracts. However, most of the bracts are sterile, and the cones of many species contain only two ovules, one

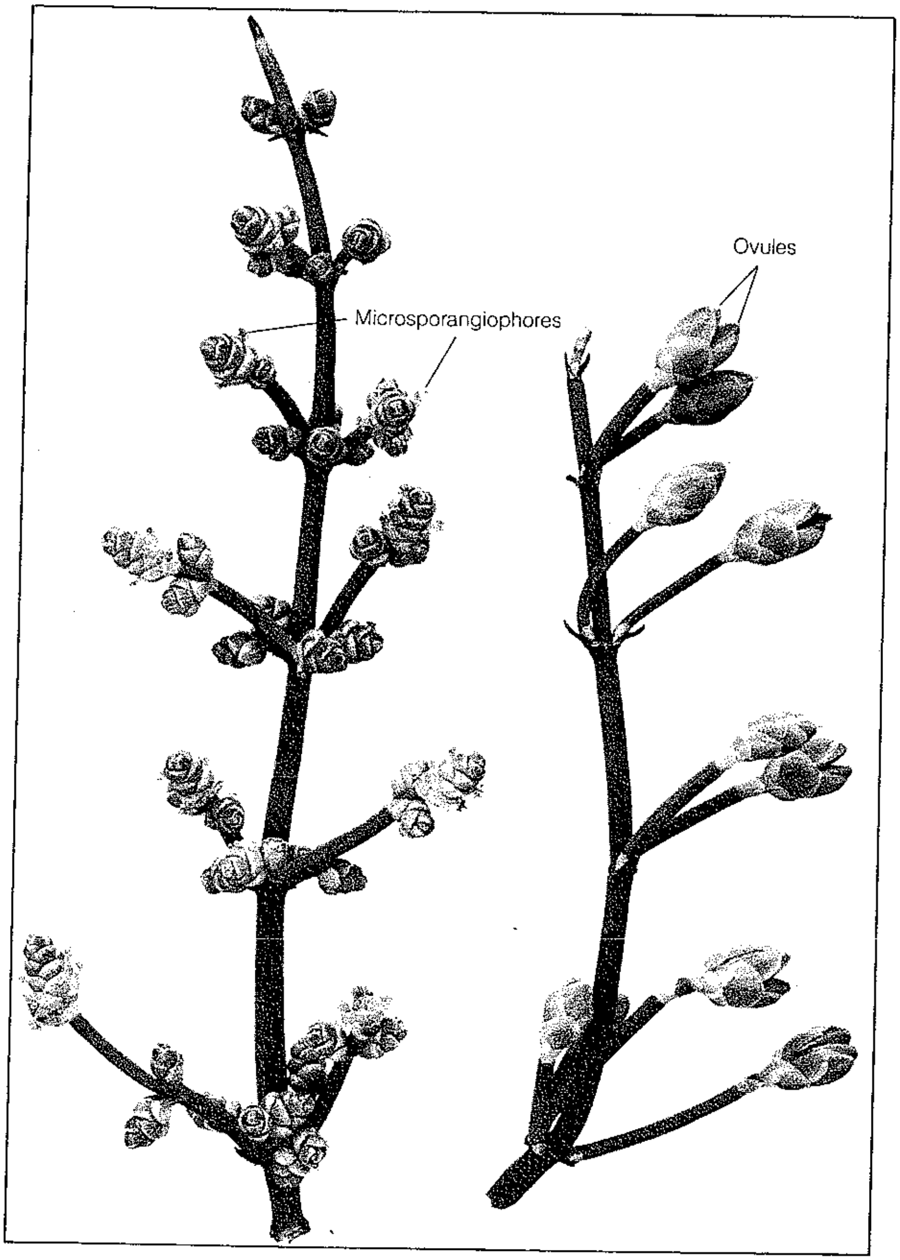

FIGURE 18-11 *Ephedra chilensis.* Microsporangiate (left) and megasporangiate (right) strobili. Note protrusion of tubular integument of the ovules of the megasporangiate strobili.

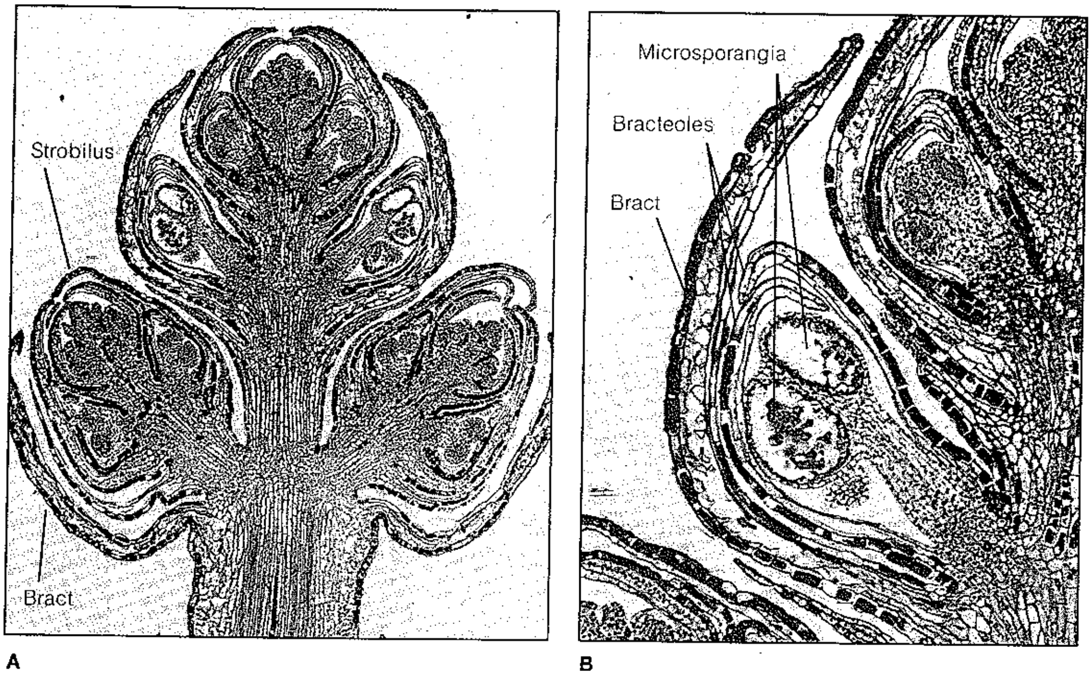

FIGURE 18-12 Structure of microsporangiate strobili in *Ephedra chilensis.* **A**, longisection of tip of reproductive shoot (each of the microsporangiate strobili is situated in the axil of a bract); **B**, enlargement of portion of A showing a microsporangiate shoot which consists of an axis with apical microsporangia enclosed by a pair of fused bracteoles.

in the axil of each of the upper bracts (Fig. 18-13, A). In some species the cones are *uniovulate,* and this condition is commonly the result of the abortion of one ovule and the crowding of the other into a false "terminal" position (see Eames, 1952).

The nucellus of the ovule of *Ephedra* is enclosed by two envelopes, *each* of which has been regarded as an ovular integument. Particularly notable is the marked elongation of the upper region of the integument, which protrudes from the tip of the ovule as a delicate open tube (Fig. 18-13, B). This *micropylar tube,* as it is frequently called, functions as a receptive organ for the pollen and represents one of the salient characters shared by the ovules of *all* gnetophytes. Description of the morphology of the outer envelope of the ovule of *Ephedra,* however, is highly controversial; Martens (1971) has summarized the conflicting opinions. We have adopted the view that the so-called outer integument of the ovule of *Ephedra* represents a pair of connate bracteoles, comparable to the pair of bracteoles of the microsporangiate shoot, whereas the "inner envelope" represents the only true integument.

Microsporogenesis and the Male Gametophyte

Microsporogenesis has been carefully investigated in several species of *Ephedra* (Land, 1904; Maheshwari, 1935; Singh and Maheshwari, 1962). The sporangial initials are hypodermal and divide periclinally, forming an outer layer of *primary wall cells.* The periclinal division of the layer of primary wall cells yields a single wall layer, which eventually becomes crushed, and the tapetum. During the meiotic division of the microsporocytes, the tapetal cells become multinucleate and finally degenerate. At maturity, the microsporangium contains tetrads of microspores enclosed within a thick-walled epidermis.

The early steps in the ontogeny of the endosporic male gametophyte of *Ephedra* closely paral-

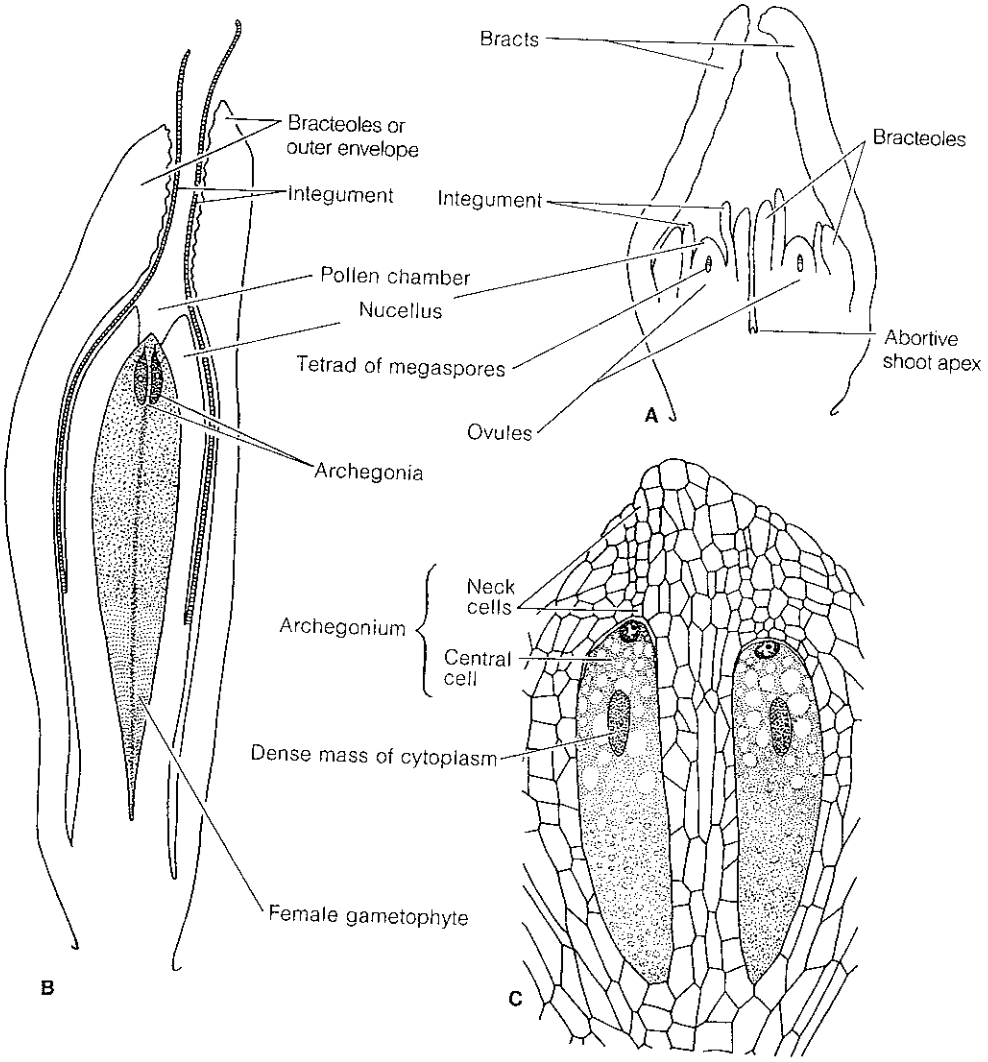

FIGURE 18-13 The ovule and mature female gametophyte of *Ephedra foliata*. A, longisection of megasporangiate strobilus showing two young ovules, each with a linear tetrad of megaspores; B, median longisection of an ovule showing the female gametophyte, the conspicuous pollen chamber, and elongated integument; C, details of micropylar region of female gametophyte showing structure of archegonia. [From Maheshwari, *Proc. Indian Acad. Sci.* 1:586, 1935.]

lel the mode of development of the male gametophyte of *Pinus* (see Chapter 17). The first two mitotic divisions yield two lens-shaped *prothallial cells* which begin to degenerate soon after their formation. Then the nucleus of the meristematic, or antheridial, initial divides again, forming the generative and tube cell (Fig. 18-14, A-E). The division of the generative cell produces a sterile cell and a spermatogenous cell (Fig. 18-14, E, F). In this five-celled stage in development, the pollen grain is shed from the microsporangium. There appears to be some species variation in the formation of persistent cellulose walls that delimit cells in the pollen grains (Singh and Maheshwari, 1962; Martens, 1971).

The mature pollen grains of *Ephedra* are ellipsoidal in form, and the exine is characteristically

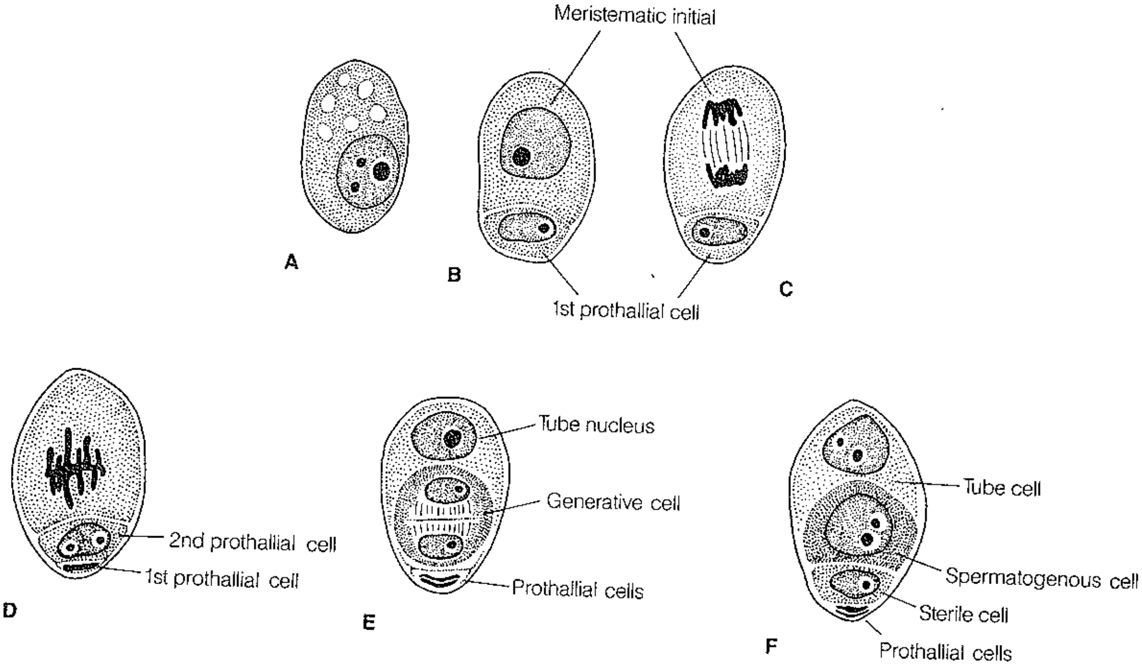

FIGURE 18-14 *Ephedra gerardiana.* Development of the male gametophyte. A, uninucleate microspore; B, microspore has divided resulting in the formation of the first prothallial cell and continuing meristematic initial; C, division leading to formation of second prothallial cell; D, metaphase, leading to formation of the generative cell and tube nucleus; E, division of generative cell to produce the spermatogenous cell and sterile cell as shown in F. [Redrawn from Singh and Maheshwari, *Phytomorphology* 12:361–372, 1962.]

sculptured into a series of ridges—extending from pole to pole—separated by longitudinal furrows (Fig. 18-15). Because of these definitive structural characters, it has been possible to identify *Ephedra* pollen in the fossil record (from the Cretaceous and Tertiary periods) and to draw structural comparisons between it and the pollen of living species (see the detailed study by Steeves and Barghoorn, 1959).

Megasporogenesis and the Female Gametophyte

The megasporocyte, prior to meiosis, is a large, conspicuous cell located rather deeply within the nucellus of the ovule (Fig. 18-13, A). According to some investigators (Maheshwari, 1935; Seeliger, 1954; Singh and Maheshwari, 1962), the sunken position of the megasporocyte is due (1) to the formation above it of several layers of parietal cells, derived from the early division of a hypodermal cell, and (2) the very active periclinal division of the cells of the nucellar epidermis. The meiotic divisions of the megasporocyte result in the formation of four megaspores (Fig. 18-13, A), although in *Ephedra distachya* wall formation between the megaspores is delayed which may give the illusion of a tetrasporic type of development found in angiosperms (Lehman-Baerts, 1967). The megaspore furthest from the micropyle enlarges, and a series of free-nuclear divisions occur in the peripheral cytoplasm that surrounds the large central vacuole. Land (1907) found 256 free nuclei in the coenocytic female gametophyte of *Ephedra trifurca* but higher numbers have been reported for other species—500 in *Ephedra foliata* and about 1,000 in *Ephedra distachya*.

Following the free nuclear phase, the female gametophyte passes through the usual *alveolation* stage as in most other gymnosperms. Anticlinal walls begin to form centripetally, forming long, tubular uninucleate alveoli which later become subdivided by periclinally oriented walls (Lehman-

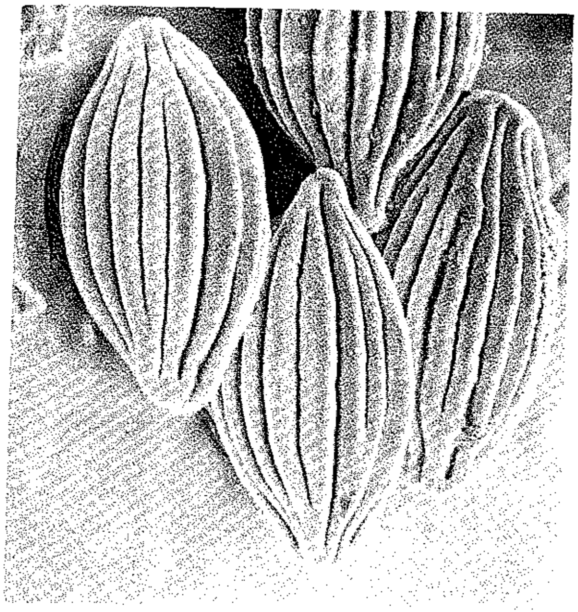

Figure 18-15 Scanning electron micrograph of a group of pollen grains of *Ephedra* sp. Note the prominent longitudinal (meridional) ridges characteristic of the pollen of this genus (X 700).

Baerts, 1967). Subsequently, active cell divisions continue throughout the young gametophyte which soon becomes histologically differentiated into two regions: a lower zone (at the chalazal end) of small, compact, frequently dividing cells, and an upper zone (at the micropylar end) of longer, thinner-walled cells; certain of the superficial cells of the upper zone later function as *archegonial* initials. The number of archegonia formed varies from two to eleven, according to Martens (1971).

The archegonial initial divides periclinally into an outer *primary neck cell* and an inner *central cell.* The most distinctive feature of archegonial development in *Ephedra* is the formation of a massive neck which is produced by the repeated periclinal and anticlinal divisions of the derivatives of the primary neck cell. At first the divisions are so regular that three to five tiers of cells are produced, each tier composed of a quartet of cells. Subsequent divisions are less regular, and the cells of the archegonial neck merge with the adjacent cells of the gametophyte (Fig. 18-13, C). The mature archegonial neck may consist of about thirty to forty cells; Land (1904) remarks: "of all gymnosperms, *Ephedra* has the longest-necked archegonium." The enlargement of the central cell is followed by the division of its nucleus to form the *ventral canal nucleus* and the *egg nucleus*. Sometimes the ventral canal nucleus appears to degenerate soon after its formation, but in some species it persists and remains intact near the upper part of the archegonium (Lehman-Baerts, 1967).

Following the first division of the archegonial initial, the contiguous cells of the gametophyte divide, producing a jacket around the entire developing archegonium.

Pollination

Prior to pollination, the nucellar tissue lying directly above the archegonia of the gametophyte begins to break down, creating a very conspicuous funnel-shaped *pollen chamber* (Fig. 18-13, B). During pollen-chamber formation, the disintegrating cells, at the summit of the nucellus, produce a liquid, rich in sugar, which fills the micropylar canal of the ovule and, at the time of pollination, exudes as a "pollination drop" from the open end of the exserted integument (Fig. 18-16).

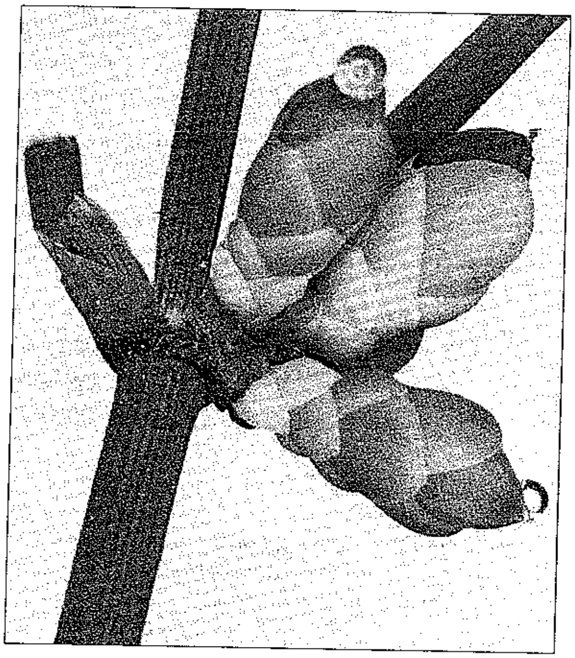

Figure 18-16 *Ephedra* sp. Basal cluster of three megasporangiate strobili. Note conspicuous "pollination drop" at tip of two of the strobili. See text for further discussion. [Courtesy of Dr. E. G. Cutter.]

Wind pollination is undoubtedly the important process in *Ephedra*. However, earlier accounts of insect pollination have been substantiated (Bino, Dafni, and Meeuse, 1984; Bino, Devente, and Meeuse, 1984). The pollination drops on ovules as well as drops of nectar on other parts of both pollen and ovulate cones contain about 10 percent sucrose; in contrast, pollination drops in pine contain only about 1.25 percent. The flying insects visit the drops of nectar and in so doing transfer pollen grains from their bodies to the ovules. By whichever pollination method, the pollen grains adhere to and float on the sticky surface of the pollination drop. As the water begins to evaporate, the column of liquid in the micropylar canal shortens and the pollen grains are pulled inward into the pollen chamber.

Fertilization

According to Land (1907), the interval between pollination and fertilization in *Ephedra trifurca* may be as short as ten hours, which is a remarkable contrast with the more extended interval typical of *Pinus* and a number of other conifers. A unique morphological feature of *Ephedra* is the fact that at the time of pollination, the archegonial end of the female gametophyte is freely exposed at the base of the deep pollen chamber (Fig. 18-13, B). As a result, when the pollen grain germinates, the pollen tube penetrates *gametophytic tissue,* i.e., the tissue of the archegonial neck. This represents a striking contrast to the growth of the pollen tube through the nucellar — i.e., *sporophytic* — tissue of the ovule, characteristic of the conifers.

Following the emergence of the pollen tube, the exine of the pollen grain is shed and the spermatogenous cell divides, forming two male gametes. After reaching the egg, the tip of the pollen tube ruptures and the two gametes, together with the sterile cell and the tube nucleus, are discharged into the egg cytoplasm.

Although one of the male gametes unites with the egg nucleus and forms a diploid zygote, the behavior of the other male gamete is extraordinary in certain species. Khan (1943), working on *Ephedra foliata,* reported that the second male gamete may fuse with the ventral canal nucleus. Although an embryo does not result from this fusion, Khan observed in one specimen, two nuclei that he considered to have been formed by the division of the fertilized ventral canal nucleus. Likewise, Moussel (1978) reported one instance in *Ephedra distachya* in which the ventral canal nucleus, which otherwise degenerates, probably fused with the second male gamete. No nutritive tissue was formed from this union as in the case of "double fertilization" in angiosperms (see Chapter 20). Moussel speculated that additional proembryonic nuclei may be formed from divisions of the second diploid nucleus.

Embryogeny

In *Ephedra* there is a process of free nuclear division, beginning with the first mitosis of the zygotic nucleus. Each of the eight (or more) free diploid nuclei that are produced becomes surrounded by a densely staining sheath of cytoplasm; later a cellulosic wall is formed around each nucleus (Fig. 18-17, A). As each of these proembryonic cells may develop *independently* into an embryo (Fig. 18-17, A–C), *Ephedra* exhibits a distinctive and precocious type of cleavage polyembryony. From an ontogenetic standpoint, polyembryony in *Ephedra* has been "pushed back" to the free nulear stage in embryogeny.

Each proembryonal cell puts out a tubular outgrowth. The nucleus may divide before the formation of the outgrowth or it may move into the tube and divide. A transverse wall is then formed, giving rise to a terminal embryonal cell and an embryonal suspensor which elongates (Fig. 18-17, C, D). As an embryo grows through the archegonium and into the female gametophyte, the terminal embryonal group of cells increases in number; some of the proximal cells elongate, forming embryonal tubes (Fig. 18-17, E), a feature characteristic of all other gymnosperms (Chapter 17). With further development, two cotyledons and the shoot apex are formed at the distal end of the embryo, and a root apical meristem becomes organized at the proximal end (Fig. 18-17, F).

Of the several embryos which competitively develop in a single ovule, only one normally reaches a fully developed stage in the seed. Kahn (1943) observed in one ovule eighteen to nineteen separate developing embryos. He interpreted this large num-

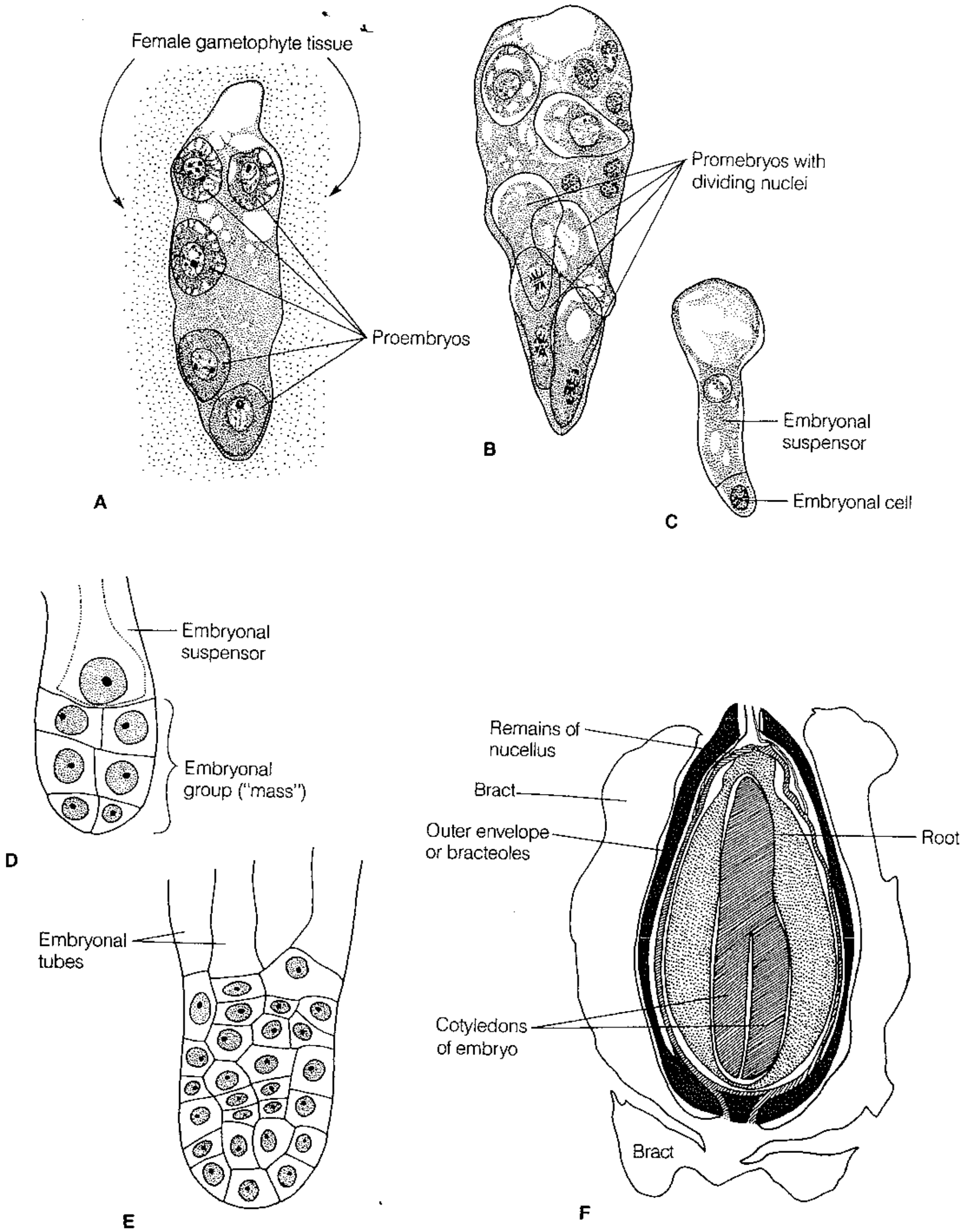

FIGURE 18-17 Embryogeny and seed structure in *Ephedra.* **A**, longitudinal section of zygotic cell showing five proembryos; **B**, reconstruction from several sections showing six proembryos in various stages of development (note mitoses and origin of embryonal suspensors in the lower proembryos); **C**, embryo consisting of embryonal cell and suspensor cell; **D**, group of embryonal cells derived from derivatives of embryonal cell in C; **E**, older developing embryo with embryonal tubes derived from the embryonal group ("mass"); **F**, longisection of mature seed. [**A–C, F** redrawn from Khan, *Proc. Nat. Acad. Sci. India* 13:357–375, 1943; **D, E** redrawn from Lehmann-Baerts, *Cellule* 67:51–87, 1967.]

ber to be the result of the combination of simple and cleavage polyembryony.

The general structure of a ripe seed in *Ephedra foliata* is shown in Fig. 18-17, F. The conspicuous embryo, with its two large cotyledons, is embedded within the tissue of the female gametophyte, and the remains of the nucellus are evident as a disorganized sheath of cells. At the micropylar end of the seed the remains of the "true" integument are evident, and the entire seed is externally enclosed by the fused bracteoles (= outer envelope), many cells of which develop thick hard walls. As the seed matures in *Ephedra foliata,* as well as many other species, the adjacent subtending bracts of the ovulate strobilus become thick and fleshy, forming an additional investment (Fig. 18-18). The bracts may become ivory, red, or orange in color. According to Land's (1907) study on *Ephedra trifurca,* there appears to be no resting, or "dormant," period for the seed, which may even begin to germinate within the parent strobilus.

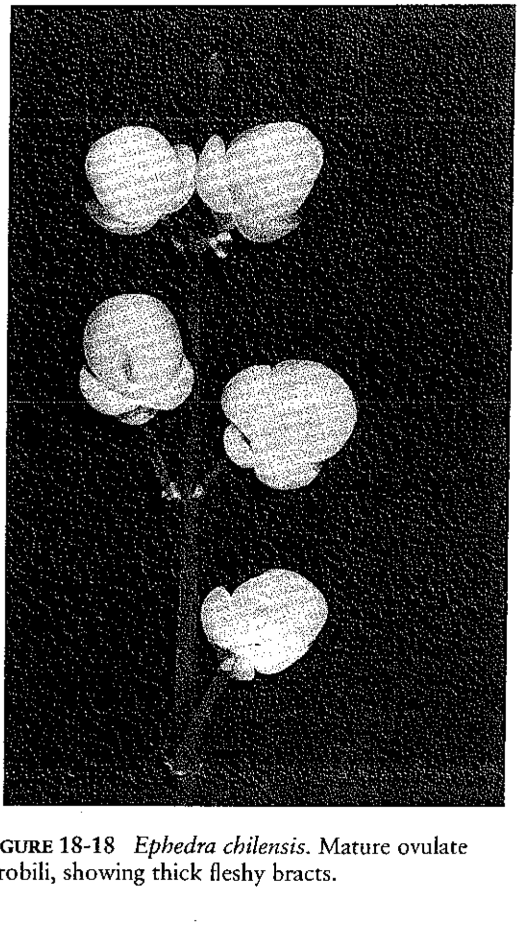

FIGURE 18-18 *Ephedra chilensis.* Mature ovulate strobili, showing thick fleshy bracts.

In addition to the formation of diploid embryos through the normal processes of fertilization and embryogeny, Konar and Singh (1979) demonstrated that plantlets, consisting of shoots and roots, can be formed from the haploid female gametophyte of *Ephedra.* Callus was initiated in vitro on a basal nutrient medium plus 2 ppm 2,4-D (2,4-dichlorophenoxyacetic acid). The callus when subcultured on a medium containing 2 ppm kinetin led to the formation of plantlets.

Morphological Comparisons Between *Ephedra, Gnetum* and *Welwitschia*

The three genera of gnetophytes share the following morphological characters: compound microsporangiate and megasporangiate strobili, an extended micropylar tube formed by the integument of the ovule, vessels in the secondary xylem, shoot apices having a well-defined surface layer (tunica), decussate phyllotaxis, and embryos with two cotyledons. The phylogenetic and taxonomic significance of these points of resemblance, however, must be judged in the light of equally impressive *differences* among the genera with reference to (1) the organization of strobili, (2) the development and structure of both the male and female gametophytes, (3) the methods of fertilization, and (4) the types of embryogenesis. The details of reproduction in *Ephedra* already have been described and will not be repeated here. The reproductive features of *Gnetum* and *Welwitschia* will be emphasized in the following discussions.

The Reproductive Cycle in *Gnetum*

The strobili in *Gnetum* are compact or elongate axes with conspicuous nodes and internodes. In a microsporangiate strobilus there are two fused bracts at a node forming a cupulelike structure that partially surrounds numerous fertile shoots (often referred to as "flowers"). Each fertile shoot consists of two fused bracteoles, enclosing a microsporophyll (or microsporangiophore). Generally there are two separate microsporangia at the tip of the sporophyll. In *Gnetum gnemon,* the top whorl consists of abortive ovules (Fig. 18-19, A, B).

At each node of a megasporangiate strobilus, the "cupule" or "collar" subtends a whorl of eight to ten or fewer ovules (Fig. 18-20, A). An ovule is surrounded by three concentric insheathing structures—the outer and inner "envelopes" and the integument (Fig. 18-20, B). Many authors do not consider the outer and inner envelopes as true integuments, but Rodin and Kapil (1969) have shown that the inner envelope can become sclerified, and may become fused with the outer envelope to form a hard seed coat.

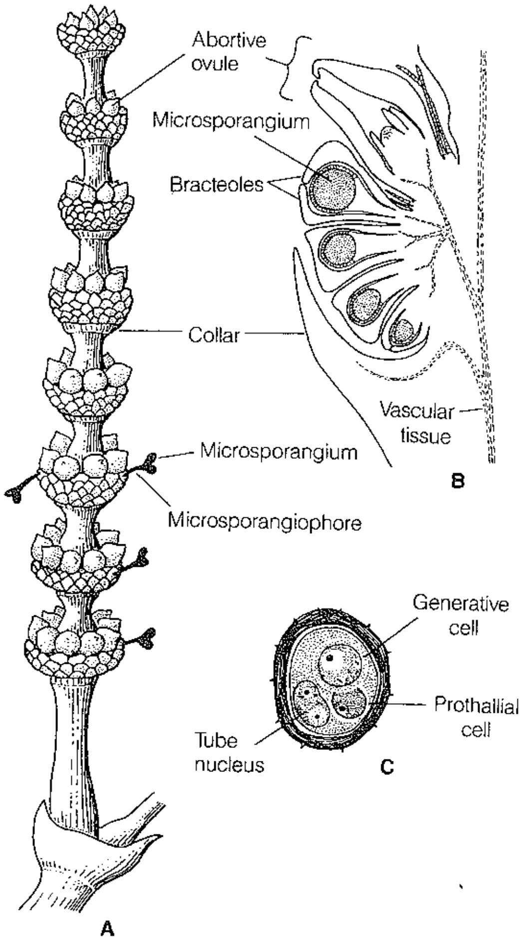

Figure 18-19 A, microsporangiate strobilus of *Gnetum gnemon;* B, Longitudinal section through a node, showing four developing microsporangiate fertile shoots and one abortive ovule; C, *Gnetum ula:* pollen grain at time of shedding from microsporangium. [A, B redrawn from Sanwal, *Phytomorphology* 12:243–264, 1962; C redrawn from Vasil, *Phytomorphology* 9:167–215, 1959.]

There are several accounts of the reproductive cycle in *Gnetum.* However, investigators differ in their descriptions, even for the same species. The following discussion is more or less representative for the genus. Terminology is that of Singh (1978).

A pollen grain at the time of shedding consists of a tube nucleus, a prothallial cell, and a generative cell (Fig. 18-19, C). When the pollen grain germinates in *Gnetum,* the generative cell in the pollen tube produces two male gametes just prior to fertilization.

In *Gnetum,* walls do not form between the four megaspore nuclei; the result is a *tetrasporic* type of female gametophyte development found in certain angiosperms (Chapter 20). The four nuclei undergo a period of free nuclear divisions resulting in 250 to 1,000 nuclei. Wall formation begins at the lower (chalazal) end of the female gametophyte, and may be initiated before or after fertilization. At the upper (micropylar) end single nuclei or groups of two or more nuclei become surrounded by membranes. These are the egg cells (Fig. 18-21, A). One or more pollen tubes push into the female gametophyte and come to lie next to the egg cells (Fig. 18-21, A). Only one male gamete in a pollen tube may fertilize an egg, but there are reports that both male gametes may be functional and two eggs could become fertilized. Following fertilization the micropylar end of the female gametophyte becomes cellular. A zygote undergoes cell division, resulting in two cells that branch repeatedly, each branch having one nucleus at its tip (Fig. 18-21, B, C). The cells elongate as they grow deep into the cellular female gametophyte; the greatly elongated cells are termed suspensor tubes (Fig. 18-21, C, D). A nucleus at the tip of a tube undergoes mitosis, followed by wall formation. By repeated cell divisions of the embryonal cell, an embryonal group or mass is formed. Some of the proximal cells elongate, forming embryonal tubes as in other gymnosperms (Fig. 18-21, E–L). Generally only one embryo develops to maturity in the seed. This type of embryogeny is another example of cleavage polyembryony. The embryo becomes dicotyledonous and has a conspicuous structure, the "feeder," that arises from the hypocotyl region and develops into

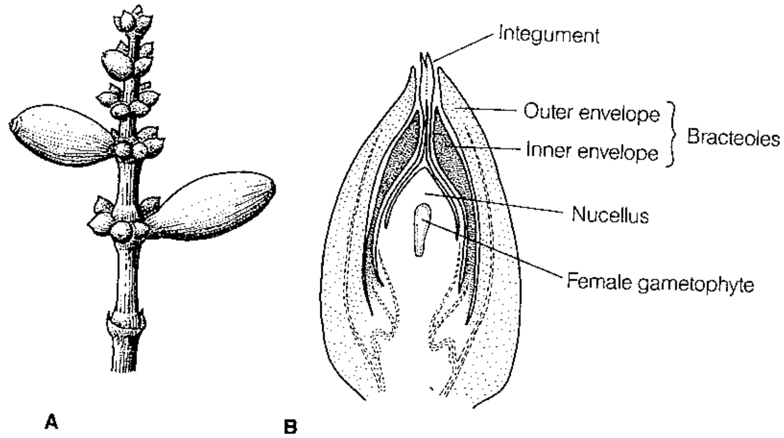

Figure 18-20 *Gnetum gnemon.* A, megasporangiate strobilus, showing ovules and partially developed seeds; B, longitudinal section of young ovule. [Redrawn from Sanwal, *Phytomorphology* 12:243–264, 1962.]

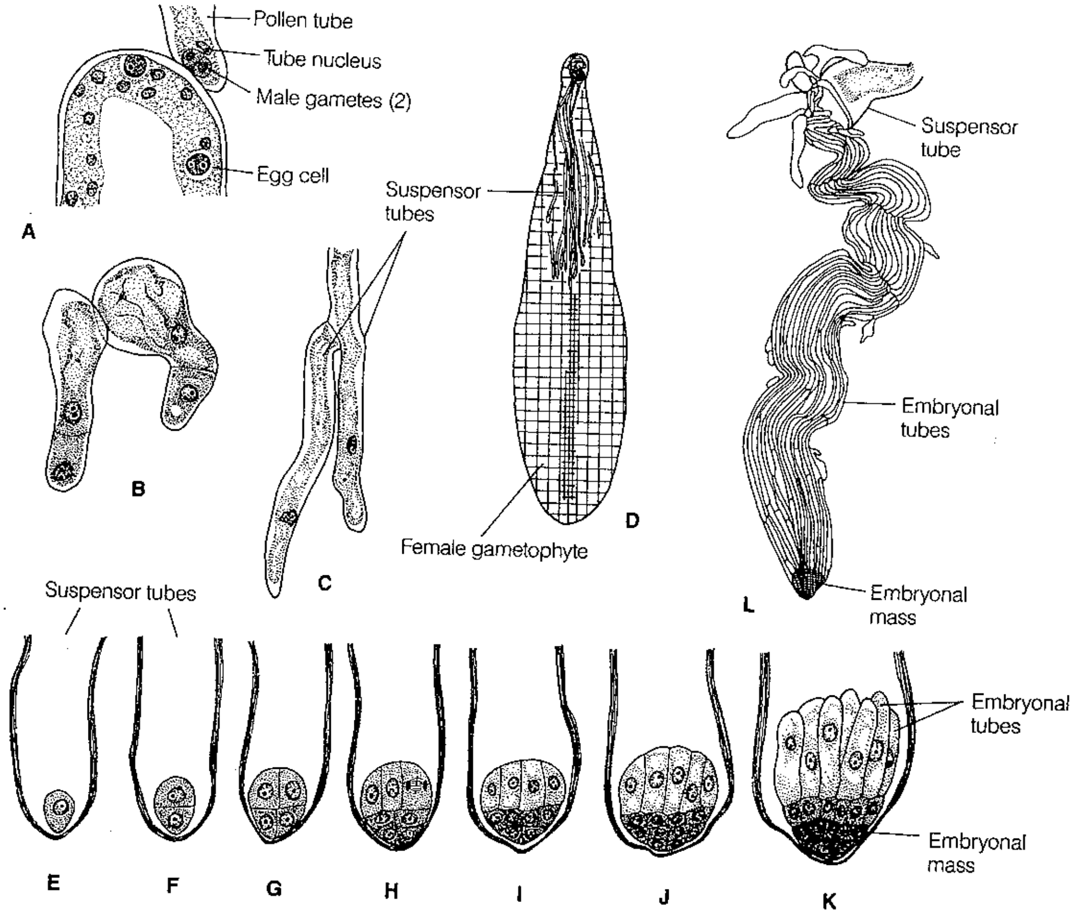

Figure 18-21 Early embryogeny in *Gnetum.* A, pollen tube appressed to female gametophyte with two differentiated egg cells; B, zygotes have divided forming suspensor tubes; C, branching suspensor tubes; D, longitudinal section of cellular female gametophyte with numerous suspensor tubes; E–K, tips of suspensor tubes showing embryonal cell and its derivatives; L, older developing embryo. [A redrawn from Thompson, *Amer. J. Bot.* 3:135–184, 1916; B–D, L redrawn from Vasil, *Phytomorphology* 9:167–215, 1959; E–K redrawn from Swamy, *Phytomorphology* 23:176–182, 1973.]

a large absorptive structure. Seeds are reported to be shed during early embryogeny (suspensor-tube stage); further development occurs on the ground, as in *Ginkgo*. [For additional reading on the subject, see the following: Waterkeyn (1954, 1959), Vasil (1959), Sanwal (1962), Martens (1971), Swamy (1973), and Singh (1978).]

The Reproductive Cycle in *Welwitschia*

The branched, reproductive shoot systems arise as buds from tissues of the "crests" at the bases of the two permanent leaves. Both microsporangiate and megasporangiate cones are compound in organization (Figs. 18-22, A; 18-23, A, B).

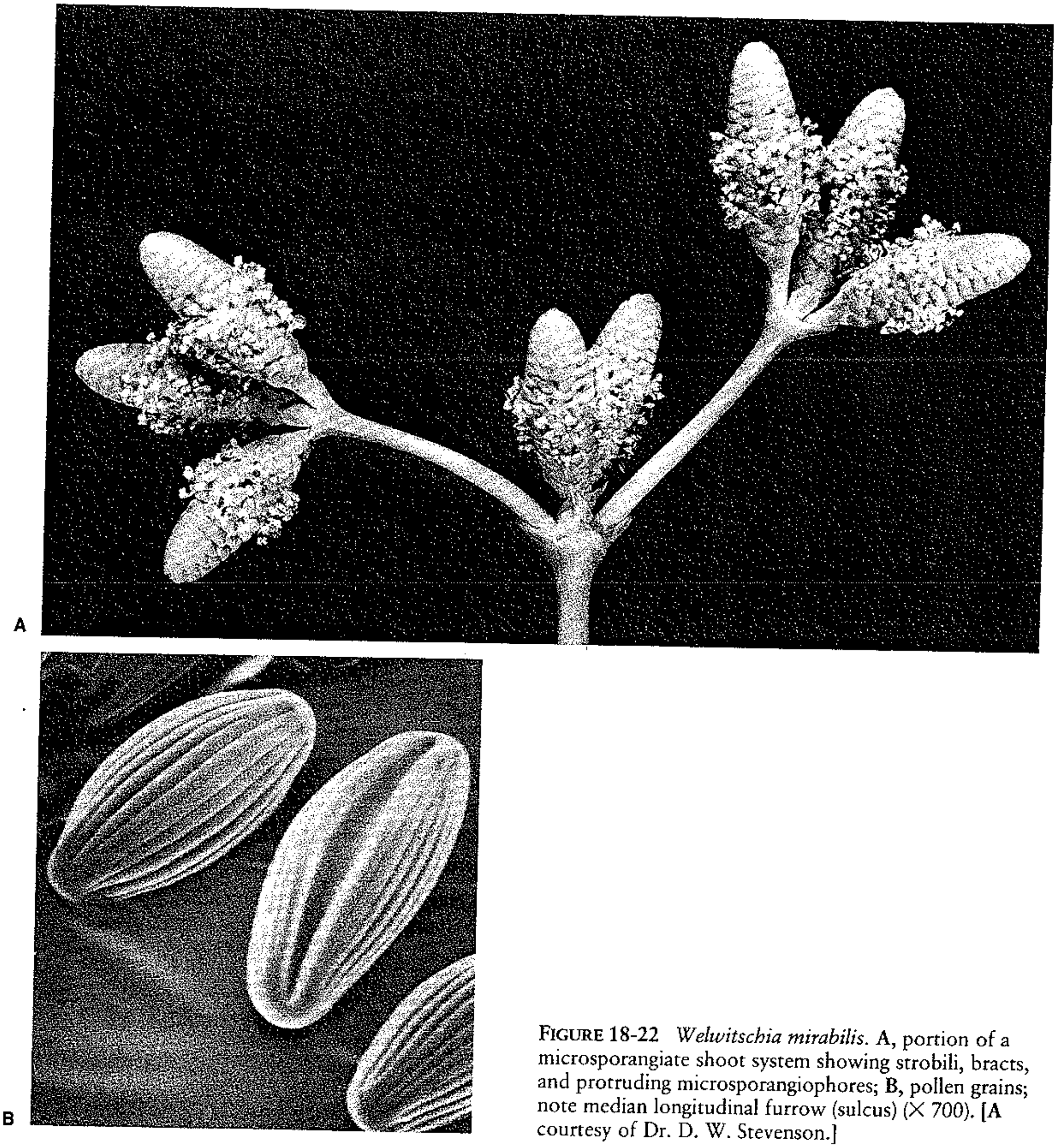

FIGURE 18-22 *Welwitschia mirabilis*. A, portion of a microsporangiate shoot system showing strobili, bracts, and protruding microsporangiophores; B, pollen grains; note median longitudinal furrow (sulcus) (× 700). [A courtesy of Dr. D. W. Stevenson.]

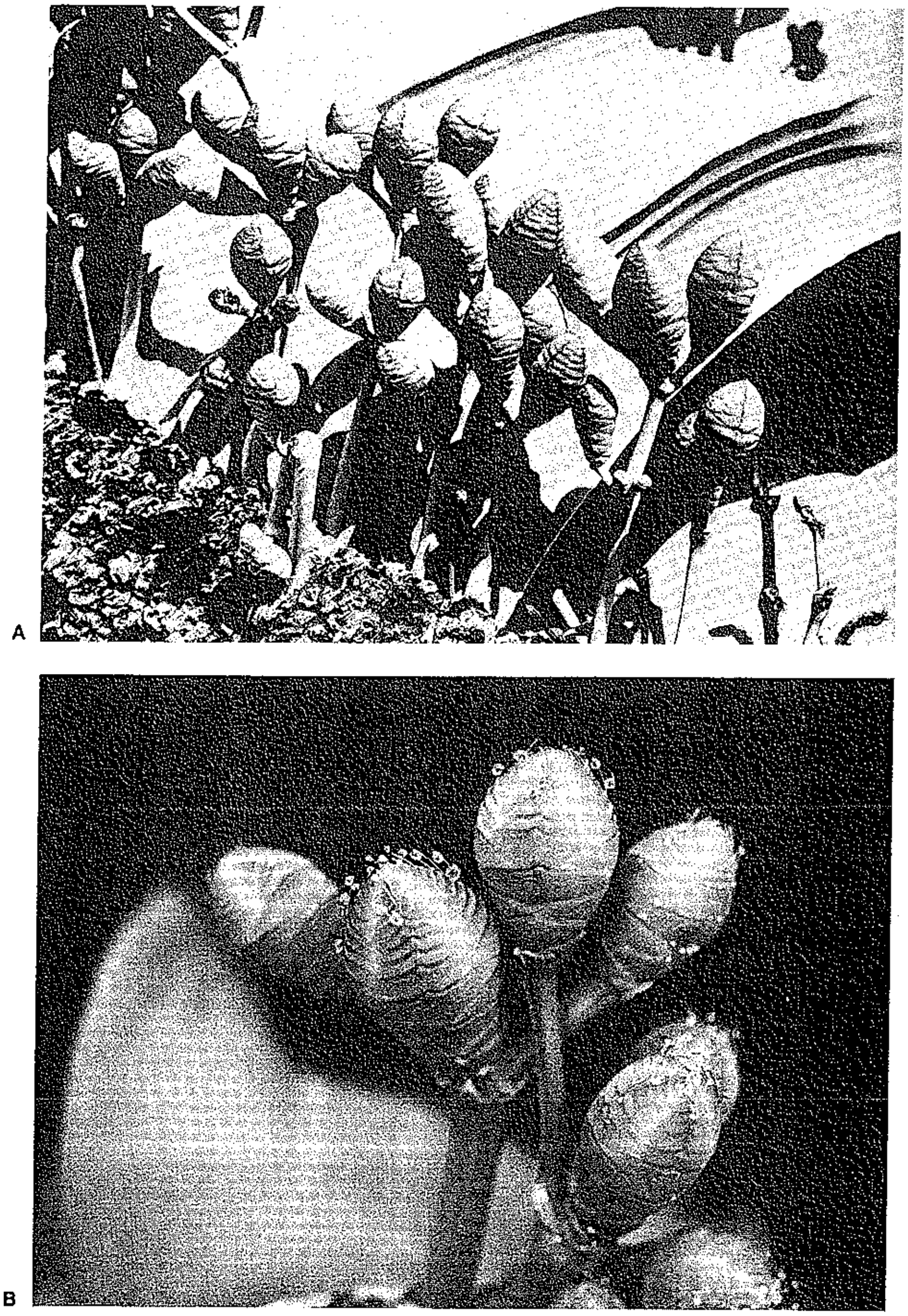

FIGURE 18-23 *Welwitschia mirabilis.* **A**, plant from Namib Desert showing megasporangiate (ovulate) branch systems at the base of a leaf; scars of previous branches can be seen at left; **B**, ovulate strobili showing bracts and exserted integuments with pollination droplets; from greenhouse grown plant. [**A** from Lindsay, *Pacific Discovery*. California Academy of Sciences, Vol. 35, No. 5, 1982; photograph courtesy of Mr. Edward S. Ross]

A microsporangiate strobilus (pollen cone) consists of four rows of bracts (decussate phyllotaxy). In the axil of a bract is a fertile shoot consisting of (1) two lateral unfused bracteoles, (2) two fused bracteoles forming an envelope, (3) six microsporangiophores, fused at their bases into a sheath or cup, and (4) an abortive (sterile) ovule that terminates the axis of the fertile shoot. The integument of the ovule forms a flange of tissue at the apex (Fig. 18-24). At the tip of each microsporangiophore are three fused sporangia with radial lines of dehiscence across the top. The external morphology of the pollen grain is similar to that of *Ephedra* (compare Figs. 18-15 and 18-22, B). At the time of dehiscence the pollen grain of *Welwitschia* consists of an ephemeral prothallial cell, a generative cell, and a tube cell (Martens and Waterkeyn, 1974). As with *Gnetum* some authors apply angiosperm terminology to a fertile shoot. Hence, the bracteoles constitute the perianth, the microsporangiophores are the stamens (androecium), and the abortive ovule occupies the position of the gynoecium. The entire fertile shoot is referred to as a flower. However, gymnosperm terminology will be used in this book (or, at least, noncommital terms) because the phylogeny of the gnetophytes is still to be resolved.

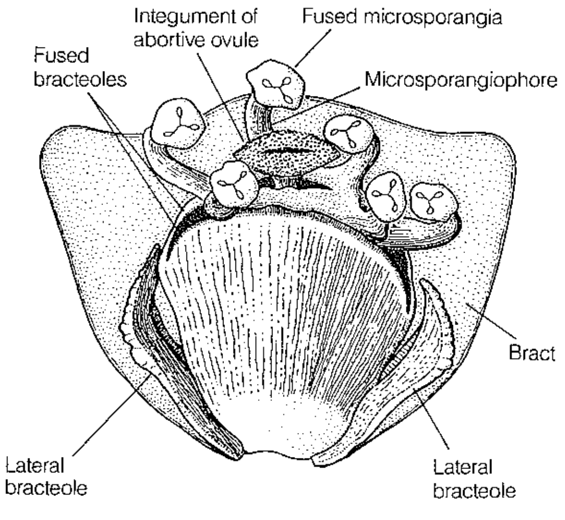

FIGURE 18-24 Fertile shoot and subtending bract from microsporangiate strobilus of *Welwitschia*. [Based on Les Gnétophytes by P. Martens. In *Handbuch der Pflanzenanatomie,* Band XII, Teil 2. Gebrüder Borntraeger, Berlin. 1971.]

The organography of a fertile shoot in a megasporangiate strobilis (ovulate cone) is very similar to that of a microsporangiate fertile shoot. There are two pairs of oppositely arranged bracteoles. Those of the lower pair are relatively small and unfused. Bracteoles of the second pair are elongate in the plane of the bract and are fused, forming an envelope around the ovule that has an exserted integument (Fig. 18-25). In the mature seed, the fused bracteoles surrounding the ovule produce winglike extensions that aid in seed dispersal.

Megasporogenesis in *Welwitschia* is similar to *Gnetum,* and in constrast to *Ephedra,* since walls are not formed between the four nuclei resulting from meiosis. The four haploid nuclei then enter a period of free nuclear divisions. The formation of alveoli does not occur as in the majority of gymnosperms. Rather, wall formation results in the enclosure of two to eight nuclei per cell in the micropylar end and six to twelve nuclei in cells at the basal or chalazal end of the female gametophyte. At the basal end the nuclei fuse to form polyploid cells that undergo cell divisions. Subsequent development is without parallel in other gymnosperms or angiosperms. Figure 18-26 should be consulted in following the sequence of developmental events. The multinucleate cells at the micropylar end of the female gametophyte grow up into the nucellus, forming female gametophytic tubes (FGTs) into which the nuclei migrate. Pollen tubes growing downward in the nucellus meet and fuse with the upward growing multinucleate FGTs. Fertilization

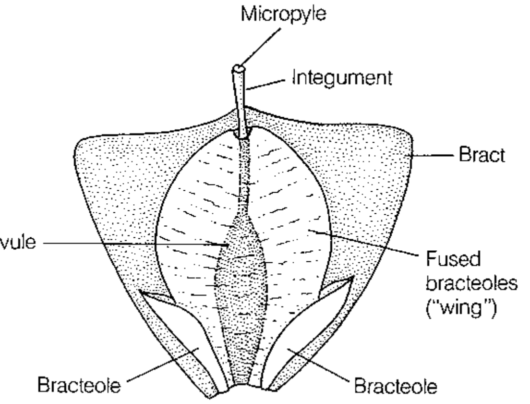

FIGURE 18-25 Fertile shoot and subtending bract from megasporangiate strobilus of *Welwitschia.*

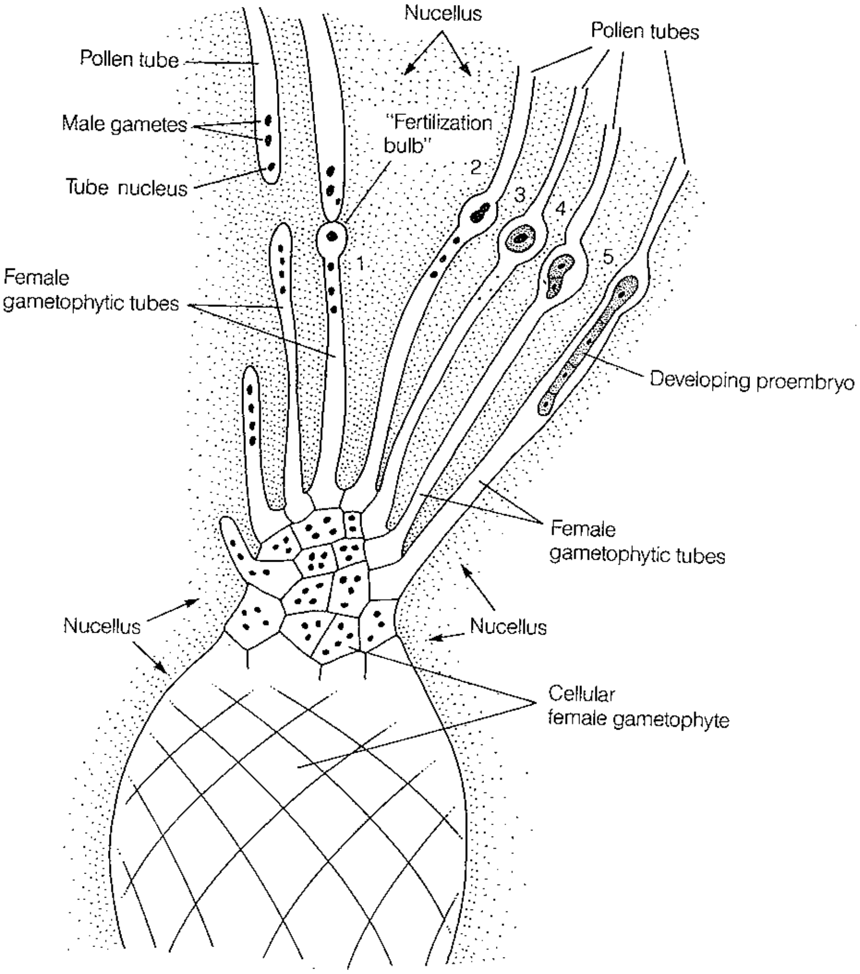

FIGURE 18-26 Schematic representation of the developmental events leading up to fertilization and proembryo development in *Welwitschia*. Starting from the left, certain multinucleate female gametophyte cells form tubes that grow up into the nucellus. Pollen tubes growing down in the nucellus meet the upward growing tubes of the female gametophyte (1); wall dissolution occurs and fertilization occurs in the "fertilization bulb" (2) to form a zygote (3); zygote divides, forming a two-celled proembryo (4); the proembryo grows down inside the female gametophytic tube toward the nutritive tissue below (5).

of one of the nuclei by a male gamete occurs in a bulbous tip ("fertilization bulb") of a FGT. A zygote is formed which, upon division, gives rise to an embryonal suspensor cell and an embryonal cell. The latter cell divides repeatedly, giving rise to additional embryonal suspensor cells that elongate, and the entire proembryo grows toward the female gametophyte *inside* the FGT. During the growth of the embryo, a terminal group of embryonal cells (embryonal "mass") is produced from which numerous embryonal tubes are formed as they are in other gymnosperms (Fig. 18-27). Many embryos begin development but normally only one completes its development.

Later stages of development occur within the female gametophyte from which the developing

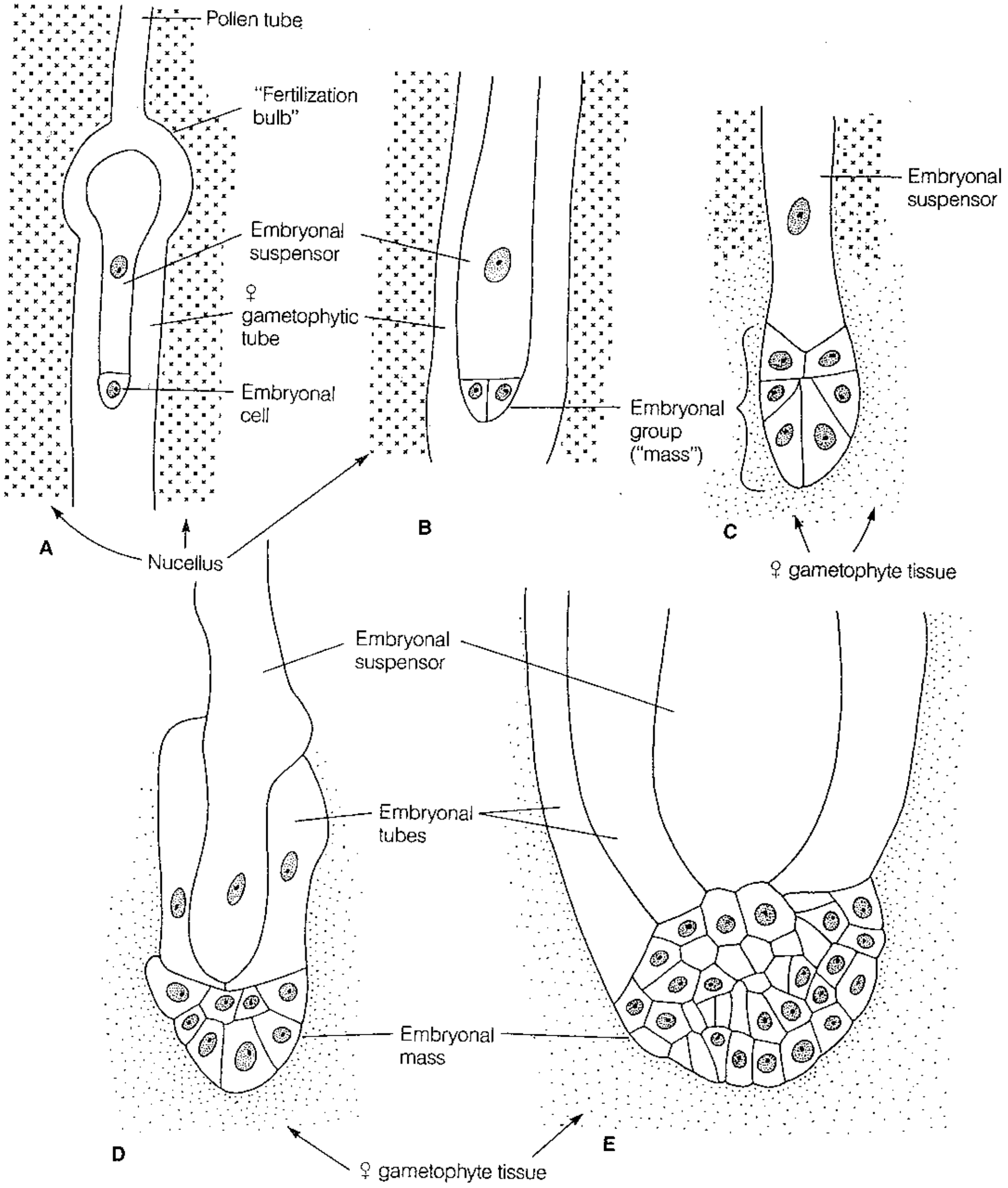

FIGURE 18-27 Stages in the early embryogeny of *Welwitschia*. **A**, the zygote has divided forming a two-celled proembryo growing inside the female gametophytic tube; **B**, embryonal cell has divided to form two embryonal cells; **C**, developing young embryo has grown into female gametophyte tissue; **D**, **E**, later stages in which elongate embryonal tubes have been produced by cells of the embryonal group (mass). [Based in part on Martens and Waterkeyn, *Cellule* 70:163–258, 1974.]

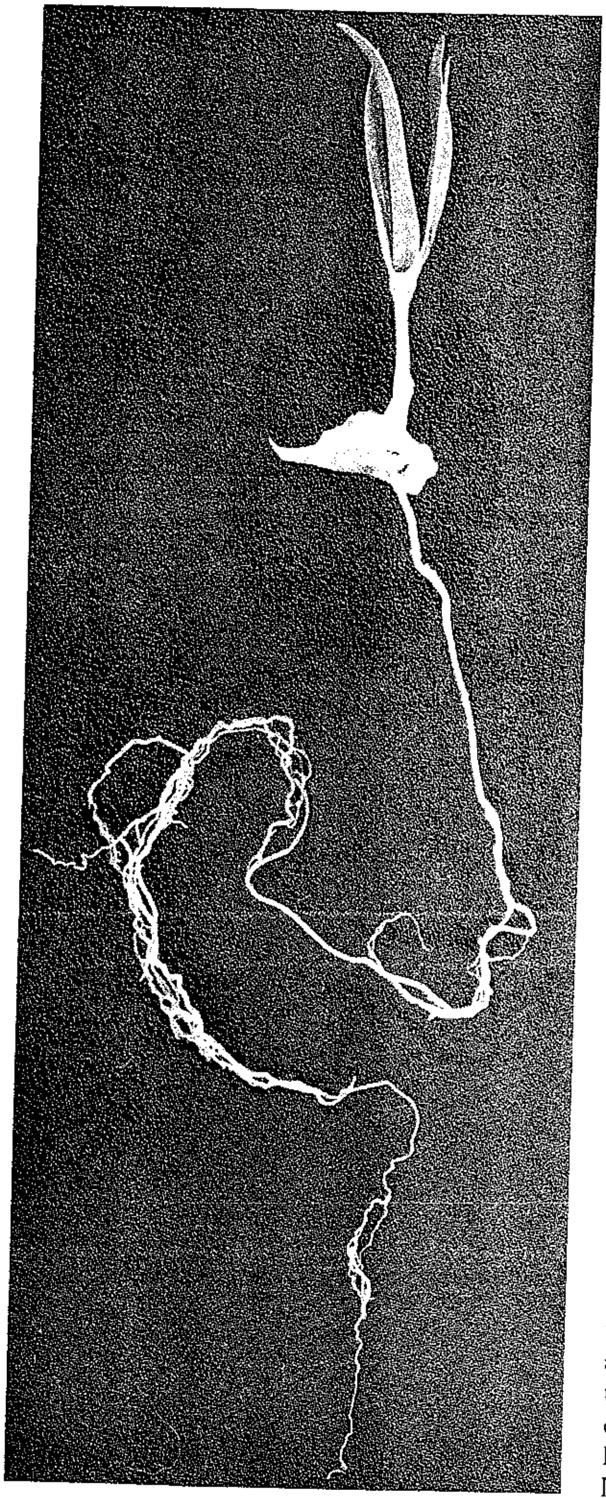

FIGURE 18-28 Seedling of *Welwitschia* showing two cotyledons and the two young permanent leaves; note that a prominent tap root is formed. [Courtesy of Dr. B. Dehgan.]

embryo receives its nutrition. In a ripe seed the embryo consists of a shoot apex, two cotyledons, hypocotyl, primary root (radicle), and a lateral bulge that develops into a "feeder" when the seed germinates (Fig. 18-28). Cells of the female gametophyte contain numerous lipid and protein bodies. When the seed germinates, enzymes are released that break down the food reserves, making them available to the growing embryo (Butler et al., 1979).

Gnetophytes, a Heterogeneous Group

We must emphasize that the gnetophytes represent, morphologically and phylogenetically, a paradoxical group of seed plants. On the one had, as Chamberlain (1935) maintained, the *combination* of certain characters — such as the compound nature of strobili and the presence of vessels in the xylem — is sufficient to keep the three genera together. On the other hand, Eames (1952) considered *Ephedra* to be closer to the conifers than to *Gnetum* or *Welwitschia* based upon differences in the organization of strobili. Details of the reproductive cycles also tend to support this belief (Table 18-1).

As can be seen in Table 18-1, *Gnetum* and *Welwitschia* share certain reproductive characters, but the methods of fertilization and embryo development are quite different and specialized in the two. The differences among all three genera suggest that the gnetophytes are a very heterogeneous assemblage of plants. This was the view of Martens (1971), who remarked that he was more struck by the contrasts among the three genera than by the traits they have in common. *Gnetum,* at various times, has been proposed as a possible progenitor of angiosperms, but the gnetophytes as well as all extant gymnosperms lack "pollenkitt" (a sticky lipoidal substance on the surface of pollen grains). Pollenkitt is apparently restricted to angiosperms, leading one scientist to rule out the gnetophytes as progenitors of angiosperms (Hesse, 1984). However, the gnetophytes for many years have been implicated in angiosperm phylogeny; more will be said on the subject in later chapters.

Table 18-1 Comparison of certain reproductive characteristics in *Ephedra, Gnetum,* and *Welwitschia*

	Ephedra	Gnetum	Welwitschia
Sterile (stalk) cell in the pollen grain	Present	Absent	Absent
Female gametophyte monosporic in origin	Present	Absent (Tetrasporic)	Absent (Tetrasporic)
Alveolation in development of female gametophyte	Present	Absent	Absent
Archegonia	Present	Absent	Absent
A free nuclear period in embryogenesis	Present	Absent	Absent

REFERENCES

Arber, E. A., and J. Parkin

1908. Studies on the evolution of the angiosperms. The relationship of the angiosperms to the Gnetales. *Ann. Bot.* 22: 489-515.

Bailey, I. W.

1953. Evolution of the tracheary tissue of land plants. *Amer. Jour. Bot.* 40:4-8.

Bino, R. J., A. Dafni, and A. D. J. Meeuse

1984. Entomophily in the dioecious gymnosperm *Ephedra aphylla* Forsk. (=*E. alte* C. A. Mey.), with some notes on *E. campylopoda* C. A. Mey. I. Aspects of the entomophilous syndrome. *Proc. Kon. Ned. Akad. Wetensch.* (Amsterdam), Ser. C, 87:1-13.

Bino, R. J., N. Devente, and A. D. J. Meeuse

1984. Entomophily in the dioecious gymnosperm *Ephedra aphylla* Forsk. (=*E. alte* C. A. Mey.), with some notes on *E. campylopoda* C. A. Mey. II. Pollination droplets, nectaries, and nectarial secretion in *Ephedra. Proc. Kon. Ned. Akad. Wetensch.* (Amsterdam), Ser. C, 87:15-24.

Bornman, C. H.

1972. *Welwitschia mirabilis:* Paradox of the Namib Desert. *Endeavor* 31(113):95-99.

1978. *Welwitschia: Paradox of a Parched Paradise.* C. Struik Publishers, Cape Town.

Butler, V., C. H. Bornman, and W. A. Jensen

1979. *Welwitschia mirabilis:* Fine structure of the germinating seed. II. Cytochemistry of gametophyte and feeder, *Z. Pflanzenphysiol.* 91:197-210.

Chamberlain, C. J.

1935. *Gymnosperms. Structure and Evolution.* University of Chicago Press, Chicago.

Crane, P. R., and G. R. Upchurch, Jr.

1987. *Drewria potomacensis* gen. et sp. nov., an early Cretaceous member of Gnetales from the Potomic group of Virginia. *Amer. J. Bot.* 74:1722-1736.

Cutler, H. C.

1939. Monograph of the North American species of the genus *Ephedra. Ann. Missouri Bot. Gard.* 26:373-428.

Dittrich, P., and W. Huber

1975. Carbon dioxide metabolism in members of the Chlamydospermae. In M. Avron (ed.), *Proc. Third Intl. Congress on Photosynthesis,* pp. 1573-1578. Elsevier, Amsterdam.

Dyer, R. A., and I. C. Verdoorn

1972. Science or sentiment: the *Welwitschia* problem. *Taxon* 21:485-489.

Eames, A. J.

1952. Relationships of the Ephedrales. *Phytomorphology* 2:79-100.

Evert, R. F., C. H. Bornman, V. Butler, and M. G. Gilliland

1973a. Structure and development of the sieve-cell protoplast in leaf veins of *Welwitschia. Protoplasma* 76:1-21.

1973b. Structure and development of sieve areas in leaf veins of *Welwitschia. Protoplasma* 76:23-34.

Foster, A. S.

1972. Venation patterns in the leaves of *Ephedra. Jour. Arnold Arboretum* 53:364-378.

Gifford, E. M., Jr.

1943. The structure and development of the shoot apex of *Ephedra altissima* Desf. *Bull. Torrey Bot. Club* 70:15-25.

Hesse, M.

1984. Pollenkitt is lacking in Gnetatae: *Ephedra* and

Welwitschia; further proof for its restriction to the angiosperms. *Pl. Syst. Evol.* 144:9–16.

Hooker, J. D.
1863. On *Welwitschia,* a new genus of Gnetaceae. *Trans. Linn. Soc. London* 24:1–48.

Johnson, M. A.
1950. Growth and development of the shoot of *Gnetum gnemon* L. I. The shoot apex and pith. *Bull. Torrey Bot. Club* 77:354–367.

Khan, R.
1943. Contributions to the morphology of *Ephedra foliata* Boiss. II. Fertilization and embryogeny. *Proc. Nat. Acad. Sci. India* 13:357–375.

Konar, R. N., and M. N. Singh
1979. Production of plantlets from the female gametophytes of *Ephedra foliata* Boiss. Z. Pflanzenphysiol. 95:87–90.

Land, W. J. G.
1904. Spermatogenesis and öogenesis in *Ephedra trifurca. Bot. Gaz.* 38:1–18.
1907. Fertilization and embryogeny in *Ephedra trifurca. Bot. Gaz.* 44:273–292.

Lehmann-Baerts, M.
1967. Études sur les Gnétales. XII. Ovule, gamètophyte femelle et embryogenèse chez *Ephedra distachya* L. *Cellule* 67:51–87.

Maheshwari, K.
1935. Contributions to the morphology of *Ephedra foliata* Boiss. I. The development of the male and female gametophytes. *Proc. Indian Acad. Sci.* 1:586–606.

Maheshwari, P., and V. Vasil
1961. *Gnetum.* Council of Scientific and Industrial Research, New Delhi.

Marsden, M. P. F., and T. A. Steeves
1955. On the primary vascular system and the nodal anatomy of *Ephedra. Jour. Arnold Arboretum* 36:241–258.

Martens, P.
1971. *Les Gnétophytes. (Handbuch der Pflanzenanatomie,* Band 12, Teil 2.) Gerbrüder Borntraeger, Berlin.
1977. *Welwitschia mirabilis* and neoteny. *Amer. J. Bot.* 64:916–920.

Martens, P., and L. Waterkeyn
1963. The shoot apical meristem of *Welwitschia mirabilis* Hooker. *Phytomorphology* 13:359–363.
1974. Études sur les Gnétales. XIII. Recherches sur *Welwitschia mirabilis.* V. Évolution ovulaire et embryogenèse. *Cellule* 70:163–258.

Moussel, B.
1978. Double fertilization in the genus *Ephedra Phytomorphology* 28:336–345.

Muhammad, A. F., and R. Sattler
1982. Vessel structure of *Gnetum* and the origin of angiosperms. *Amer. J. Bot.* 69:1004–1021.

Pearson, H. H. W.
1929. *Gnetales.* Cambridge University Press.

Pulle, A.
1938. The classification of the spermatophytes. *Chron. Bot.* 4:109–113.

Rodin, R. J.
1953. Seedling morphology of *Welwitschia Amer. Jour. Bot.* 40:371–378.
1958a. Leaf anatomy of *Welwitschia.* I. Early development of the leaf. *Amer. Jour. Bot.* 45:90–95.
1958b. Leaf anatomy of *Welwitschia.* II. A study of mature leaves. *Amer. Jour. Bot.* 45:96–103.
1966. Leaf structure and evolution in American species of *Gnetum. Phytomorphology* 16:56–68.
1967. Ontogeny of foliage leaves in *Gnetum. Phytomorphology* 17:118–128.

Rodin, R. J., and R. N. Kapil
1969. Comparative anatomy of the seed coats of *Gnetum* and their probable evolution. *Amer. J. Bot.* 56:420–431.

Rodin, R. J., and G. S. Paliwal
1970. Nodal anatomy of *Gnetum ula. Phytomorphology* 20:103–111.

Sanwal, M.
1962. Morphology and embryology of *Gnetum gnemon* L. *Phytomorphology* 12:243–264.

Schulze, E.-D., and I. Schulze
1976. Distribution and control of photosynthetic pathways in plants growing in the Namib Desert, with special regard to *Welwitschia mirabilis* Hook. *fil. Madoqua* 9(3):5–13.

Seeliger, I.
1954. Studien am Sprossvegetationskegel von *Ephedra fragilis var. campylopoda* (C. A. Mey.) Stapf. *Flora* 141:114–162.

Singh, H.
1978. *Embryology of Gymnosperms. (Handbuch der Pflanzenanatomie,* Band X, Teil 2.) Gebrüder Borntraeger, Berlin.

Singh, H., and P. Maheshwari
1962. A contribution to the embryology of *Ephedra gerardiana. Phytomorphology* 12:361–372.

Stapf, I.
1889. Die Arten der Gattung *Ephedra. Denkschr. Akad. Wien. Math.-Naturw.* Cl. II. 56:1–112.

Steeves, M. W., and E. S. Barghoorn
1959. The pollen of *Ephedra. Jour. Arnold Arboretum* 40:221–255.

Swamy, B. G. L.
1973. Contributions to the monograph on *Gnetum.* I. Fertilization and proembryo. *Phytomorphology* 23:176–182.

Thompson, W. P.
1918. Independent evolution of vessels in Gnetales and angiosperms. *Bot. Gaz.* 65:83–90.

Vasil, V.
1959. Morphology and embryology of *Gnetum ula* Brongn. *Phytomorphology* 9:167–215.

von Willert, D. J.
1985. *Welwitschia mirabilis*—new aspects in the biology of an old plant. *Adv. Bot. Res.* 11:157–191.

Waterkeyn, L.
1954. Études sur les Gnétales. I. Le strobile femelle, l'ovule et la graine de *Gnetum africanum* Welw. *Cellule* 56:103–146.
1959. Études sur les Gnétales. II. Le strobile mâle, la microsporogenèse et le gamétophyte mâle de *Gnetum africanum* Welw. *Cellule* 60:5–78.