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Apical Meristems

DELIMITATION

The profuse and inconsistent terminology in the voluminous literature on apical meristems (Clowes, 1961a; Gifford, 1954; Guttenberg, 1960, 1961) reflects the complexity of the subject matter. Most commonly the term *apical meristem* is used in a wider sense than merely with reference to the initials and their immediate derivatives; the term also includes variable lengths of shoot or root proximal to the apex. Yet, when determinations of the dimensions of the apices of shoots are made, only the part above the youngest leaf primordium, or youngest node, is measured. Usually *shoot apex* and *root apex* are employed as synonyms of apical meristem.

The wide meaning of apical meristem is adopted in the discussions to follow, but when it is important to differentiate the most distal part of this meristem the term *protomeristem* is used in the sense indicated on page 71: it refers to the least determined part of the meristem and includes the initials and their most recent derivatives. The delimitation of this *protomeristem* is arbitrary, but the term is useful for referring to the distal part of the apical meristem, which is given much attention in the literature. The *promeristem* of Clowes (1961a) includes only the initials and thus does not coincide with the *protomeristem*. Johnson and Tolbert's (1960) *metrameristem*, on the other hand, refers to the same group of cells as the *protomeristem*.

Apical meristem and its synonyms are appropriate substitutions for the somewhat inaccurate term *growing point* (Foster, 1949). Growth in the sense of cell division, which is so characteristic of the meristematic state, is not restricted to the so-called *growing point* but occurs abun-

dantly—and is even more intense—at some distance from the apical meristem. Similarly, growth in the sense of increase in size of cells, tissues, and organs is most pronounced not in the apical meristem but in its derivatives.

INITIALS AND DERIVATIVES

An initial, or initiating cell (p. 69), is a cell that divides into two sister cells one of which remains in the meristem and the other is added to the meristematic tissues that eventually differentiate into the various tissues characteristic of the plant. The cell remaining in the apical meristem functions as an initial like its precursor. Investigators visualize the involvement of polarity, and a consequent cytologic differentiation, in the division into an initial and derivative; at the same time they agree that the status of a cell as an initial depends on its position in the protomeristem, and that the initial may be displaced by another cell and then differentiate into a body cell.

The inference about the existence of apical initials is generally based on microscopic views and on theoretical considerations, but there is also experimental evidence regarding this matter. By treatments with colchicine it is possible to change the number of chromosomes in individual cells. When cells occupying the position of initials in the shoot apex are thus affected, the change becomes detectable and is perpetuated developmentally in more or less extended parts of the plant body that develop after the treatment, and the alterations may be traced directly to the cells in the apical meristem. These cells thus fit the definition of initials. Changes in growth may cause a shift in the relative position of the modified cells in the apical meristem so that an initial ceases to act as such (Bain and Dermen, 1944). This observation supports the concept that a cell is an initial, not because of its inherent characteristics but only because of its particular position in the meristem.

The number of initials in root and shoot apices is variable. In many vascular cryptogams a single initial cell occurs at the apex (fig. 5.1); in other lower vascular plants, as well as in the higher, several initials are present. The single initial is morphologically rather distinct from its derivatives and is customarily spoken of as the *apical cell*. If the initials are more or less numerous, they are called *apical initials*, although considered semantically it would be appropriate to call them apical cells also. The recognition of apical initials under the microscope, in contrast to that of the single apical cells, is uncertain (pls. 16, 17).

The apical initials may occur in one or more tiers. If there is only one tier, all cells of a plant body are ultimately derived from it. In the

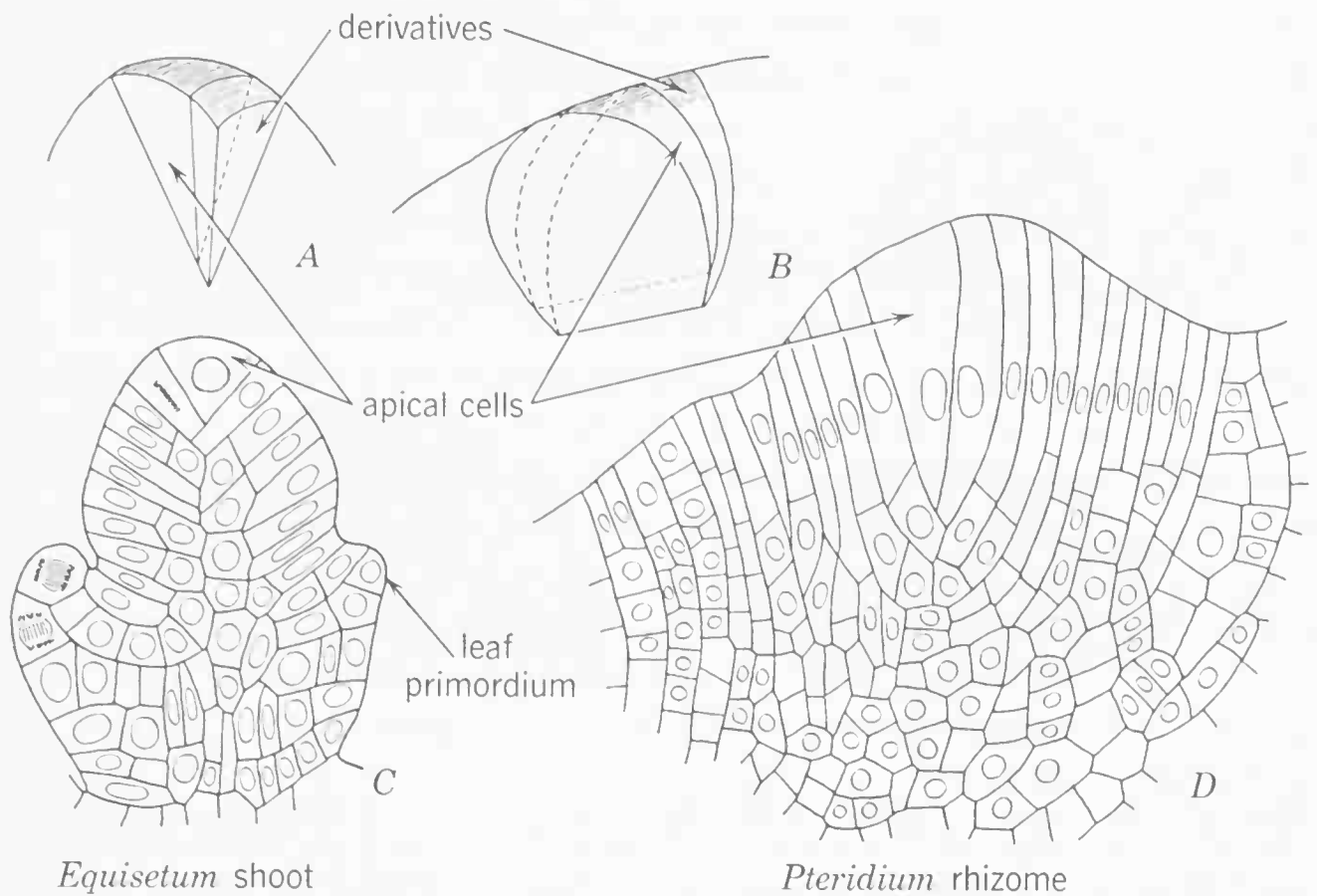


FIG. 5.1. Apical cells in shoots and rhizomes. A, B, two forms of apical cell, pyramidal (A) and lenticular (B). Cells are cut off from three faces in the pyramidal cell, from two in the lenticular. In each drawing a derivative cell is shown attached to right side of apical cell. C, D, apical cells in longitudinal sections of shoot (C) and rhizome (D). In C, apical cells in leaf primordia; one of these (left) is dividing. (A, B, adapted from Schüepp, *Handbuch der Pflanzenanatomie* 4, 1926; C, D, $\times 230$.)

alternative situation, different parts of a plant body are derived from different groups of initials. The existence of more than one independent layer of initials in certain plants has been clearly demonstrated in the previously mentioned experiments with colchicine. The treatment may induce polyploidy in one or more superficial layers of the apical meristem (fig. 5.2) and thus convert the plant into a cytochimera (Clowes, 1961a; Dermen, 1953, 1960). Induced and spontaneous chimeras showed that polyploidy could be perpetuated ontogenetically if any one of the three superficial layers in the apical meristem were polyploid, and that these three layers behaved independently in the transmission of their characteristic chromosome numbers. These plants obviously had three tiers of initials, that is, three self-propagating layers.

Induced polyploidy has also served to demonstrate the presence of more than one initial cell in each tier. In addition to periclinal chimeras, sectorial polyploidy was observed in *Vaccinium* (Bain and Dermen, 1944). The restriction of polyploidy to individual sectors of the stem is possible only if the initials occur in groups, with each component cell capable of becoming polyploid independently of the others.

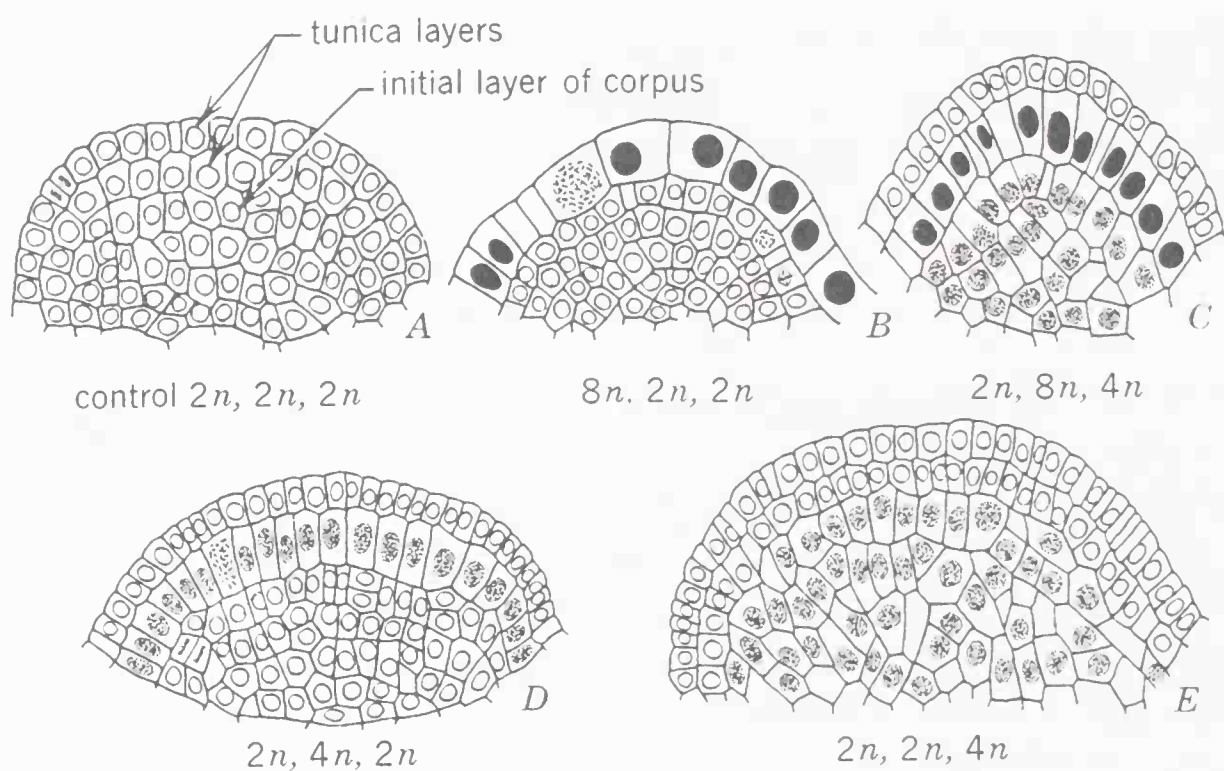


FIG. 5.2. Shoot apices of *Datura* from a diploid plant (A) and from several periclinal cytochimeras. Chromosomal combinations are indicated by values given below each drawing. The first figure of each group of three refers to first tunica layer; the second, to second tunica layer; the third, to initial layer of corpus. Octoploid cells are the largest, and their nuclei are shown in black for emphasis; tetraploid cells are somewhat smaller, and their nuclei are stippled; diploid cells are the smallest, and their nuclei are shown by circles. Chromosomal characteristics of tunica layers are perpetuated only in these layers and their derivatives (anticlinal divisions in tunica); those of the initial layer of the corpus are immediately transmitted to the subjacent layers (divisions in various planes). (Adapted from Satina et al., *Amer. Jour. Bot.* 27, 1940.)

EVOLUTION OF THE CONCEPT OF APICAL ORGANIZATION

As has been discussed by several writers (Foster, 1939, 1941; Romberg, 1963; Schüepp, 1926; Sifton, 1944; Wardlaw, 1945), the view concerning the number, the arrangement, and the activity of the initial cells and their recent derivatives in the apical meristems has undergone profound changes since the shoot apex was first recognized by Wolff (1759) as an undeveloped region from which growth of the plant proceeded.

The discovery of the apical cell in cryptogams led to the concept that such cells exist in phanerogams as well. The apical cell was interpreted as a constant structural and functional unit of apical meristems governing the whole process of growth. Subsequent researches refuted the assumption of a universal occurrence of single apical cells and replaced it by a concept of independent origin of different parts of the plant body. The *apical-cell theory* was superseded by the *histogen theory*.

The histogen theory was developed by Hanstein (1868, 1870) on the basis of extensive studies of angiosperm shoot apices and embryos. Its basic theses are, first, that the main body of the plant arises, not from superficial cells but from a mass of meristem of considerable depth, and, second, that this mass consists of three parts, the *histogens*, which may be differentiated by their origin and course of development. The outermost, the *dermatogen* (from the Greek words meaning skin and to bring forth), is the primordial epidermis; the second, the *periblem* (from the Greek, clothing), gives rise to the cortex; the third, the *plerome* (from the Greek, that which fills), constitutes the entire inner mass of the axis. The dermatogen and the periblem form mantle-like layers covering the plerome. The dermatogen, each layer of the periblem, and the plerome begin with one or several initials distributed in superposed tiers in the most distal part of the apical meristem.

Hanstein's "dermatogen" is not equivalent to Haberlandt's (1914) "protoderm." The protoderm refers to the outermost layer of the apical meristem, regardless of whether this layer arises from independent initials or not, and regardless of whether it gives rise to the epidermis only or to some subepidermal tissue also. In many apices the epidermis does originate from an independent layer in the apical meristem; in such apices the protoderm and dermatogen may coincide. The plerome and periblem in the sense of Hanstein are discernible in many roots but are seldom delimited in shoots. Thus the subdivision into dermatogen, plerome, and periblem has no universal application. But Hanstein's histogen theory is criticized chiefly because it contains an assumption that the destinies of the different regions of the plant body are determined by the discrete origin of these regions in the apical meristem. The prevalent view is that histogenesis and organogenesis have no obligate relationship to the segmentation and layering of cells in the apical meristems.

A modified use of histogen, meaning an already determined but still meristematic tissue, is being advocated by Guttenberg (1960). He places the histogen initials into lower levels of the apical meristem than Hanstein does and visualizes separate initials for procambium, pith, and cortex. Actually, in the shoot the ground meristem of the cortex adds cells to the procambium down to levels where vascular elements begin to differentiate. The delimitation between vascular and nonvascular tissues is not established in the apical meristem (Esau, 1943).

The apical-cell and the histogen theories have been developed with reference to both the root apex and the shoot apex. The third theory of apical structure, the *tunica-corporis theory* of Schmidt (1924), was an outcome of observations on angiosperm shoot apices. According to this

theory, two tissue zones occur in the apical meristem: the *tunica*, consisting of one or more peripheral layers of cells, and the *corpus*, a mass of cells overarched by the tunica (fig. 5.6, pl. 16A-C). The demarcation between these two zones results from the contrasting modes of cell division in the tunica and the corpus. The layers of the tunica show anticlinal divisions; that is, they are undergoing surface growth. The corpus cells divide in various planes, and the whole mass grows in volume. Each layer of the tunica arises from a small group of separate initials, and the corpus has its own initials located beneath those of the tunica. In other words, the number of tiers of initials is equal to the number of tunica layers plus one, the tier of corpus initials. In contrast to the histogen theory, the tunica-corporis theory does not imply any relation between the configuration of the cells at the apex and histogenesis below the apex. Although the epidermis usually arises from the outermost tunica layer, which thus coincides with Hanstein's dermatogen, the underlying tissues may have their origin in the tunica or the corpus or both, depending on plant species and the number of tunica layers.

Interest in the tunica-corporis theory was strongly stimulated by the work of Foster and his students (Foster, 1939, 1941; Gifford, 1954) and has dominated studies on shoot meristems for two decades. As more plants came to be examined, the concept underwent some modifications, especially with regard to the strictness of definition of the tunica. According to one view, tunica should include only those layers that never show any periclinal divisions in the median position, that is, above the level of origin of leaf primordia (Jentsch, 1957). If the apex contains additional parallel layers that periodically divide periclinally, these layers are assigned to the corpus and the latter is characterized as being stratified. Other workers treat the tunica more loosely and describe it as fluctuating in number of layers: one or more of the inner layers of tunica may divide periclinally and thus become part of the corpus (Clowes, 1961a). The term *mantle* has been proposed for tunica in the loose sense; it overarches a body of cells called the *core* (Popham and Chan, 1950). Still others reject the tunica-corporis concept entirely because it does not relate the apical activity to the origin of tissues (Guttenberg, 1960). Nevertheless, the tunica-corporis theory remains useful for characterizing growth in the shoot apex of angiosperms. It is used in this book with the assumption that during the vegetative growth the tunica has a characteristic number of layers, which may be attained in steps during the development of the plant and may change during the transition to the reproductive stage; and that the corpus may vary between stratified and nonstratified configurations.

As was mentioned before, the tunica-corporis concept was developed

with reference to the angiosperms; it proved to be largely unsuitable for the characterization of the apical meristem of gymnosperms (Foster, 1941, 1949; Johnson, 1951). Shoot apices of only a few gymnosperms have an independently propagating layer that could be interpreted as tunica; in others, the outermost layer divides periclinally and thus is ontogenetically related to the subjacent tissue. Studies of gymnosperm apices, stimulated by Foster (1941), led to the recognition of a zonation based not only on planes of division but also on cytologic and histologic differentiation and degree of meristematic activity of component cell complexes (figs. 5.3, 5.4; pl. 17A). Similar cyto-histologic zonation has since been observed in many angiosperms (Clowes, 1961*a*). The concept of zonation in Foster's sense has considerably advanced the understanding of growth in shoot apices. It has also related the apical organization to that of the underlying derivative shoot parts without reintroducing a formalized concept of histogen initials. But efforts to bring about such reintroduction are not lacking (Bartels, 1960, 1961; Guttenberg, 1960; Kalbe, 1962).

The cytologic zones that may be recognized in apical meristems vary in degree of differentiation and in details of grouping of cells. As a

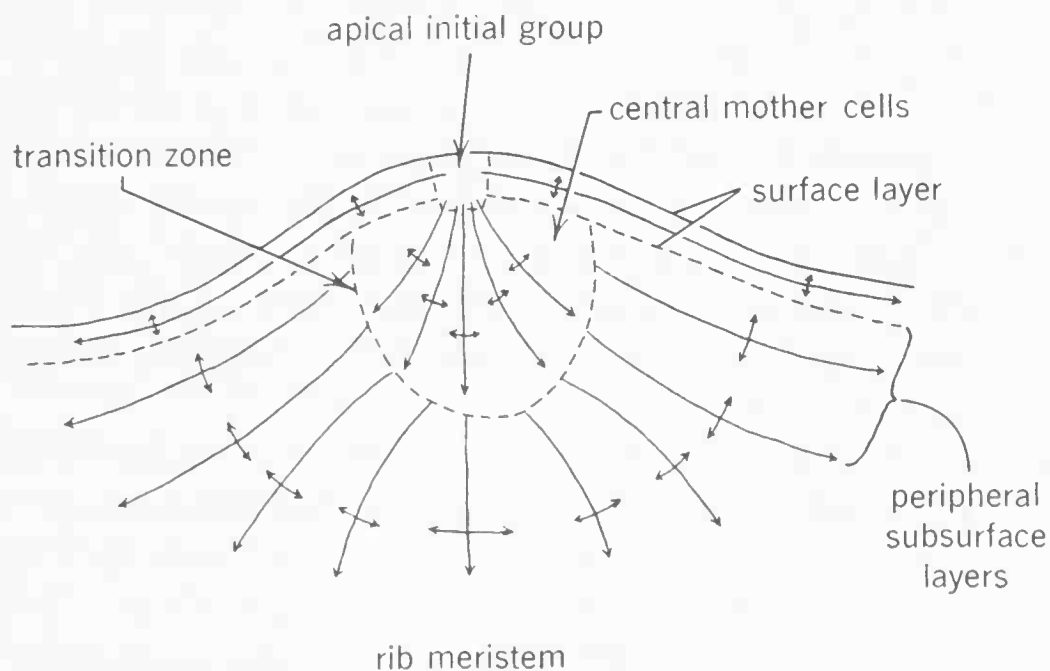


FIG. 5.3. Zones and their mode of growth in shoot tip of *Ginkgo biloba* as seen in longitudinal section. Arrows indicate prevailing direction of growth. Apical initial group contributes to surface layer by anticlinal divisions. It also adds cells by periclinal divisions to central mother-cell group. Growth in volume by cell enlargement and occasional divisions in various planes characterize the central mother-cell zone. Outermost products of divisions in this zone become displaced toward transition zone where they divide by walls periclinal with reference to mother-cell zone. Derivatives of these divisions form peripheral subsurface layers and prospective pith, the rib-meristem zone. (After Foster, *Torrey Bot. Club Bul.* 65, 1938.)

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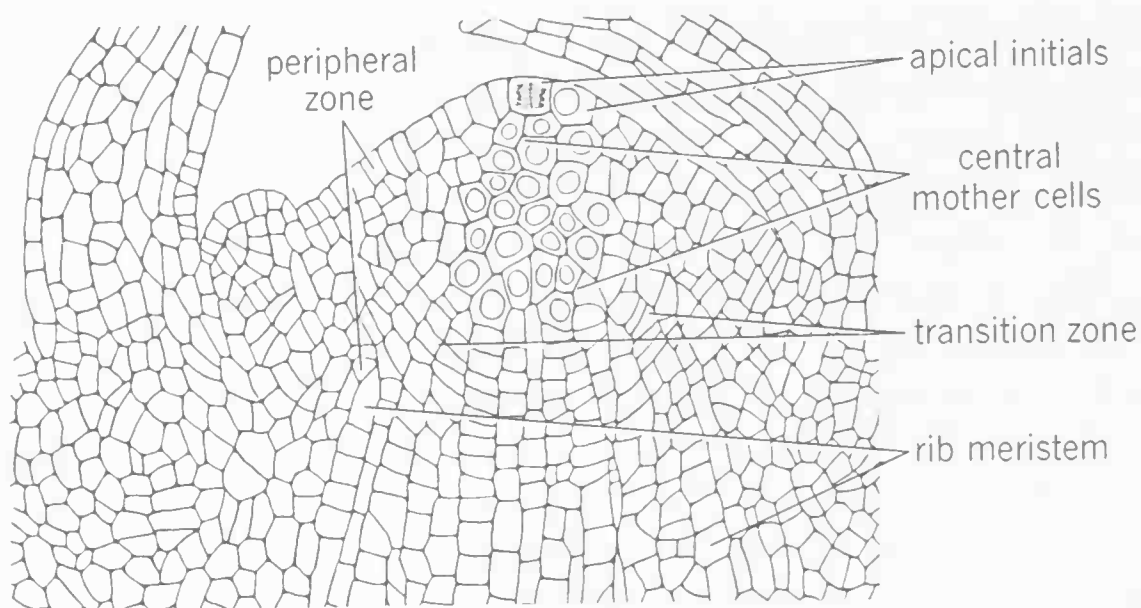


FIG. 5.4. Shoot tip of *Pinus strobus* in longitudinal view. Apical initials contribute cells to surface layer by anticlinal divisions and to central mother-cell zone by periclinal divisions. The mother-cell zone (cells with nuclei) contributes cells to the transition zone composed of actively dividing cells arranged in series radiating from the mother-cell zone. Products of these divisions form the rib meristem and the subsurface layers of peripheral zone. ($\times 150$. Slide by A. R. Spurr.)

result, the pertinent terminology is constantly growing and changing. Succinctly, the zonation may be characterized by dividing the apical meristem into a *distal axial zone* terminating the axis and two zones derived from it. One of these, the *proximal axial zone*, or the *inner zone*, appears directly below the distal zone, is centrally located in the apex, and usually becomes the pith after additional meristematic activity has occurred. The other, the *peripheral zone*, or *outer zone*, encircles the other zones. The peripheral zone is also called flank meristem in the literature because of a common tendency to describe structures as seen in sections in two dimensions.

The peripheral zone typically is the most meristematic of all three zones, has the densest protoplasts and smallest cell dimensions. It may be described as a eumeristem (p. 75). Leaf primordia and the procambium arise here, as well as the cortical ground tissue. The inner zone early shows its destiny—differentiation into the vacuolated pith—by being cytologically less dense than the outer zone. Depending on the manner of growth of the shoot, especially the degree of elongation of future internodes, the inner zone assumes more or less definitely the characteristics of a rib meristem. The distal zone is somewhat variable in appearance. All of it, or only its proximal part, may be considerably vacuolated. The term protomeristem is applicable to the distal zone in the sense that it contains the initials and their most recent derivatives.

The derivatives of the distal zone, the outer zone and the inner zone,

may merge imperceptibly with the distal zone or they may be delimited from the distal zone by an additional, *transitional zone*, often compared to the cambium because of orderly seriation of cells resulting from divisions periclinal with reference to the distal zone. The transitional zone is composed of particularly actively dividing derivatives of the distal zone. The presence of the transition zone apparently depends on the rate of growth in the shoot apex, and the zone shows fluctuations in its distinctness in the same kind of apex (Philipson, 1954).

The next development in the interpretation of apical meristem resulted from the efforts of Buvat and his students to obtain a unified concept of growth of this meristem (Buvat, 1955*a*; Clowes, 1961*a*). Meristematic activity drew the chief attention in this work. Counts of mitoses, and cytological, histochemical, and ultrastructural studies served to formulate the theory that the distal zone of the apical meristem is relatively inert during vegetative growth and that the real initial zone is the peripheral one, where leaf primordia arise. The distal zone received the appellation of waiting meristem (*méristème d'attente*), because it was said to be waiting for the change from vegetative to reproductive stage before taking up meristematic activity. The peripheral zone became the initiating ring (*anneau initial*) and the inner zone the medullary (pith) meristem (*méristème medullaire*). The concept of the inactive distal zone in the apical meristem was extended from the shoots of the angiosperms to those of the gymnosperms (Camefort, 1956; who called the distal zone *zone apicale*) and the lower vascular plants (Buvat, 1955*b*), and to the roots (Buvat and Genève, 1951; Buvat and Liard, 1953). The concept was later somewhat modified in that variations in degree of inactivity of the distal zone in relation to the size of the apex and its stage of development came to be recognized (Catesson, 1953; Lance, 1957; Loiseau, 1959).

The revision of the concept of apical initials by the French workers stimulated a considerable amount of research in other countries and led to refinement of techniques for determining the degree of meristematic activity in the apical meristem (Clowes, 1961*a*). Extensive counts of mitotic figures (Edgar, 1961; Hagemann, 1956; Hara, 1962; Jacobs and Morrow, 1961; Popham, 1958); studies of cell patterns in fixed (Paolillo and Gifford, 1961) and living apices (Ball, 1960; Newman, 1956); histochemical studies (Gifford and Tepper, 1962*b*); use of labeled compounds to determine the location of synthesis of DNA, RNA, and protein (Clowes, 1961*a*; Gifford, 1960); experimental injury of the meristem (Clowes, 1961*b*; Davidson, 1961; Wardlaw, 1957); and theoretical discussions (Cutter, 1959) served to evaluate the concept of the inactive distal zone in the apical meristem. Most of the investigators outside

France consider that the apparent scarcity of divisions in the distal cells of the shoot does not justify regarding these cells as being inconsequential in the construction of the shoot; these cells are the ultimate source of all other cells of the shoot and hence are the initials. This interpretation is used in the description of the shoot apices in the subsequent sections of the present chapter.

With regard to the root apices, the occurrence of an inactive center in the meristem found confirmation in many studies, which resulted in the development of the concept of *quiescent center* by Clowes (1961a). This center is described as a nonmeristematic group of cells roughly hemispherical in shape and surrounded by actively dividing cells, the initials, or the promeristem. The center becomes quiescent during the development of the root, either the main root (taproot) or the lateral root, after the architectural pattern of the apex is established, and it remains capable of resuming meristematic activity. There is apparently a range of development of the quiescent center. The center may be larger in large roots and smaller, or even absent, in small roots.

The origin of the architectural pattern in roots and shoots beginning with the embryo has been studied in a number of species. The subject is reviewed by Guttenberg (1960, 1961). The pattern is organized gradually in terminal apices of epicotyls, in lateral shoots, in radicles of embryos or seedlings, and in lateral and adventitious roots. Moreover, the distribution of the meristematic activity in the apical meristem changes with the development of shoot or root.

Apical meristems receive much attention in connection with studies of causal relations in morphogenesis. Many efforts have been directed toward determining the role of the apical meristem in the development of form and internal organization of plant organs (Clowes, 1961a; Cutter, 1959; Gifford, 1954). Some studies have been concerned with the determination of the arrangement of leaves (phyllotaxy, chapter 15) and their bilateral symmetry (chapter 16), others with the determination of the vascular patterns in roots (chapter 17) and shoots (chapter 15). Workers also consider the question whether the apex is a self-determining and dominant center of development controlling the growth of the parts derived from it or whether it is a plastic region operating under the control of stimuli sent to it from the mature subjacent tissues.

Results of experimental studies involving cultures of isolated shoot and root tips and partial isolation of apical meristems and leaf primordia by operations on growing plants have been interpreted as indicating a high degree of independence of the apical meristem. The culture studies have shown that apical meristems of roots are capable of forming vascularized roots and that the tissue pattern in the root is a prod-

uct of apical activity (Torrey, 1955). Apical meristems of shoots including the youngest leaf primordia may grow into entire plants, whereas the subjacent regions form only vascularized masses of cells (Ball, 1946). Operations on shoot apices show that the apex may continue to grow and form primordia after its procambial connection with the subjacent region is severed (Ball, 1948; Snow and Snow, 1947; Wardlaw, 1947). Some experimental work indicates a considerable degree of resistance of the apical meristem to disturbances that may be caused by environmental conditions, such as variations in light, temperature, and nutrient conditions (Thomson and Miller, 1962).

VEGETATIVE SHOOT APEX

Vegetative shoot apices vary in shape, size, cytologic zonation, and meristematic activity. The shoot apices of conifers are commonly relatively narrow and conical in form (fig. 5.4); in *Ginkgo* (fig. 5.3, pl. 17A) and in the cycads they are rather broad and flat. The apical meristem of some monocotyledons (grasses, *Elodea*) and dicotyledons (*Hippuris*) is narrow and elongated, with the distal zone much elevated above the youngest node (pl. 17B). In many dicotyledons the distal zone barely rises above the leaf primordia (fig. 5.6), or even appears sunken (pl. 18A; Gifford, 1950). In some plants the axis increases in width close to the apex and the peripheral region bearing the leaf primordia becomes elevated above the apical meristem leaving the latter in a pit-like depression (pl. 18B; Ball, 1941; rosette type of dicotyledons, Rauh and Rappert, 1954). Examples of widths of apices measured in microns at insertion of the youngest leaf primordia are: 280, *Equisetum hiemale*; 1,000, *Dryopteris dilatata*; 2,000–3,300, *Cycas revoluta*; 280, *Pinus mugo*; 140, *Taxus baccata*; 400, *Ginkgo biloba*; 288, *Washingtonia filifera*; 130, *Zea mays*; 500, *Nuphar lutea* (Clowes, 1961a). The shape and size of the apex change during the development of a plant from embryo to reproduction, between initiation of successive leaves, and in relation to seasonal changes. An example of change in width during growth is available for *Phoenix canariensis* (Ball, 1941). The diameter in microns was found to be 80 in the embryo, 140 in the seedling, and 528 in the adult plant.

Attempts to classify apical structure of shoots have resulted in the assigning of shoot apices to several types (Johnson, 1951; Popham, 1951), but these classifications are subject to criticism on the grounds that they do not reflect fundamental differences in structure and are not helpful in making the behavior of meristems better understood (Clowes, 1961a; Newman, 1961). The simple classification into three types