MORPHOLOGY OF THE TRUNK AND DEVELOPMENT OF THE MICROSPORANGIUM OF CYCADS

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(WITH PLATE X)

The original purpose of the investigation recorded in this paper was to secure as complete an account of the development of the microsporangium of cycads as possible, and especially to discover whether the archesporium is several-celled, as indicated by previous investigations. In securing staminate strobili at the earliest stage, attention was drawn to their successive appearance and the building up of the trunk, and the results of these observations have been included.

The staminate strobili of *Ceratozamia mexicana* Brogn. (*C. robusta* Miq.) were obtained from the Botanical Gardens of Jena and Berlin, and also from Fairmount Park, Philadelphia. A strobilus of *Encephalartos villosus* was obtained from the Botanical Garden at Bonn; and most of the strobili of *Zamia floridana* were sent from Miami, Florida, in 1902, 1905, and 1906. No material of Ceratozamia was younger than the division of sporogenous cells just preceding the mother cells; and the material of Encephalartos showed sporangia about to shed the pollen. In Zamia the series is much more complete than in the other forms. Collections of staminate cones were sent from Miami every two or three weeks, from June 1 to September 18, 1905; then less frequently up to the last of January, 1906, when pollination took place. In 1906 further collections of staminate and ovulate plants were made, at intervals of ten days, from June 5 to September 1. These gave stages in the development of the strobili as well as of the sporangia. Only in *Zamia floridana*, therefore, has there been any attempt to study the early development of the sporangium or strobilus. The other genera have been used for comparison in the later stages of sporangium-development.

I. Morphology of the trunk

The cycads have a columnar or tuberous trunk, thickly covered with scale leaves and old leaf bases. *Zamia floridana* illustrates a

somewhat intermediate type, in which the trunk is a little elongated, but still tuberous, the greater part of it being subterranean. The strobili of cycads apparently terminate the axis, and this position is so distinct from the laterally borne strobili of other gymnosperms that it assumes an importance demanding more than the brief statements given to it in the literature, all of which seem to be based upon observations of mature strobili.

Elongation of the stem by continued growth of the apical meristem is shown by all cycads until they begin to bear strobili, and continues to be the method of elongation of the ovulate plants of Cycas. In the staminate plants of Cycas, and in both staminate and ovulate plants of the other genera, however, a very different method of elongation sets in. This is evident from the fact that cycads live for many years after fruiting and produce a succession of crowns, so that the growth of the axis has not been stopped; and also from the fact that frequently more than one strobilus is produced at the summit of the stem, as in both ovulate and staminate plants of Zamia. The following brief historical sketch will serve to outline the views that have been held in reference to the relation of the strobili to the stem axis.

MIQUEL (1) does not commit himself to any theory as to the mode of branching. He finds the cones are elongated axes of the stem in Cycas, Macrozamia, and Encephalartos, but "perchance lateral axes, as in Zamia."

KARSTEN (2) states that "the flowers appear in *Zamia muricata* on the same stem, one after another," implying sympodial branching.

DEBARY (3) gives an instance of monopodial branching in connection with a staminate strobilus of *Cycas Rumphii*. At first the strobilus occupied a terminal position, but a vegetative point was found at its base. Later, when the strobilus had matured and had been cut off, the vegetative point assumed the terminal position; but because the strobilus showed a depression upon the side toward the vegetative point and the leaves of this point were stunted, DEBARY inferred that the staminate strobilus was really axillary, and that the vegetative point had been stunted and forced out of position. He gives the other possibility, namely, that the strobilus is really a terminal structure and is pushed to one side by the vegetative point; but he thinks that the facts stated above do not carry out this assump-

tion, although he says that it must be proved in either case by stages from young cones.

BRAUN (5) definitely suggests sympodial branching, regarding the staminate and ovulate strobili as terminal, and quoting KARSTEN as given above. ENGLER and PRANTL (14) in turn quote BRAUN.

WARMING (7), in his investigation of Ceratozamia, decides that the staminate strobilus is terminal, and in an excellent drawing of a transverse section through leaves and strobilus shows the bud appearing in the axil of one of the leaves and continuing the principal axis as a sympodium. The peduncle of the strobilus is indicated between two leaves.

SACHS (9) and GOEBEL (11) state in effect that if the strobilus is single it stands as a terminus of the stem; but if there are two or more strobili, as in *Zamia muricata* and *Macrozamia spiralis*, they may be the result of the forking of the stem.

COULTER and CHAMBERLAIN (19) state that the staminate strobilus in Cycas, at least, is terminal; but that in the other genera its true position, although apparently terminal, remains in doubt. Strobili may occur singly or several together, and in the latter case, at least, are considered lateral.

WORSDELL (25) follows the theory of the sympodium in part, stating that each strobilus is developed as a continuation of the main axis, and that growth in length is continued by means of a lateral vegetative shoot immediately below the strobilus, the latter being pushed to one side. He also suggests monopodial branching, where, "as is frequently the case, short lateral shoots are developed at intervals upon the main axis."

This historical sketch shows that the branching of the cycadean trunk has been regarded by various writers as dichotomous, as monopodial, and as sympodial.

In preparing the material so that the strobili could be severed from the stem for killing, the petioles of old leaves and the scale leaves had to be removed carefully from the top of the stem; for often in the younger stages the strobilus is almost concealed by the growing tissue around it, only the tip projecting above the firm tissue of the stem. This tip is completely covered by scale leaves, so that often the strobili reach the stage in *fig. 14* before the tip emerges from the scale leaves. In many cases more than one strobilus was

found at the summit of the stem; and in connection with the young strobili the petioles of old leaves and peduncles of old strobili were *in situ*, so that the relative positions of the organs could be ascertained.

A stem received July 5 showed the remnant of an old peduncle, a young ovulate strobilus, and about 5^{mm} from this a circle of leaves surrounding a growing point (*fig. 1*). Another ovulate plant of the same age had a young strobilus, but apparently no vegetative point; however, on sectioning this young strobilus and the tissue at its base, the vegetative point was disclosed very close to the strobilus, with a few leaves surrounding it. This growing point with its leaves was enclosed within the scale leaves, which in turn surrounded the strobilus (*fig. 2*).

Two other ovulate plants when sectioned showed each a smaller strobilus at the base of the larger one, and between them the vegetative point like a small protuberance surrounded by rudimentary leaves. This whole group was enclosed within a circle of leaves which formed a hollow cone (*fig. 3*).

One of the smallest staminate cones, received June 14, had a vegetative point at its base, with rudimentary leaves (*fig. 4*). In material received July 5 three staminate cones were found upon one plant (*fig. 5*). These were of different sizes and the vegetative point was situated at the base of the smallest cone. Each strobilus of the group had apparently been the terminal one, and had been pushed aside by the newer one in its development. The strobili would thus be in lateral positions at the conclusion of the year's production of cones. There was no stunting of any of the leaves of the vegetative point, and the point was small in every case and clearly meristematic, as shown by the staining. Later in the season, where there were several staminate cones, they were nearly of the same size and apparently of about the same stage in development, so that they might easily be supposed to be of the same age and to result from a forking of the stem, as SACHS thought. The axes of the sympodium in such cases are shortened, so that the branches stand almost upon a level (*fig. 6*). Good figures are shown by WIELAND (21).

The ovulate strobili are generally formed singly, one a year from the vegetative point. The staminate strobili develop more rapidly and from a succession of vegetative points, but they reach maturity at about the same time. The two or three cases in which more than one

ovulate strobilus was formed agreed in the details of development with those of the staminate strobilus, and like them the several strobili may mature the same year. This is not infrequent in *Zamia floridana*.

The trunk of the cycads, and especially of Zamia, is capable of forming new tissue readily upon wounding, as COULTER and CHRYSLER (22) have shown. Since there is this power of forming secondary meristem, it may be that some of the cases of lateral shoots, produced upon the lower part of the trunk, are the results of wounding.

Since other cycads are known to produce more than one strobilus upon the trunk in the same year, the manner of growth of the stem in these forms may be the same as in Zamia. The staminate cone mentioned by DEBARY (10) had the same position as those of Zamia, with the vegetative point at the base of the cone, so that this case may be explained as a sympodium, and there may have been no stunting of the point as DEBARY thought. In view of these facts it seems probable that in all the other genera the branching is sympodial, except in the case of the ovulate plants of Cycas.

The successive pushing aside of the terminal strobili in Zamia (*fig. 5*) suggests comparison with Bennettitales, as described by SCOTT (17) and WIELAND (19), in which the strobili arise terminally on short, lateral branches, wedged in between the bases of the leaves. In the modern cycads, as illustrated by Zamia, the strobili finally occupy positions in the axils of the leaves, and the vegetative point crowns the stem at the end of the year's production of strobili.

II. Microsporophylls and microsporangia

DEVELOPMENT OF SPOROPHYLLS

The youngest staminate strobilus of *Zamia floridana* was obtained from material received from Miami, June 1, 1905. The leaves were removed from the crown of the plant and the small cone exposed, less than 1^{mm} in length (*fig. 7*). The leaves in a vegetative point arise as small protuberances from the extreme base of the meristematic apex and incline toward each other, covering the point of growth (*fig. 8*). The strobilus, however, rises higher before showing any protuberances, and when these appear they rise acropetally and project at right angles to the cone (*fig. 9*). In the strobilus shown in *fig. 10* the sporophylls are not much farther advanced than in *fig. 7*, but the strobilus has lengthened and there are more primordia of sporophylls.

The first protuberance of the sporophyll is brought about by a periclinal division of a hypodermal cell in the axial row (*fig. 11*), and later by the growth and division of the hypodermal cells and of layers beneath them (*fig. 12*). The outer layer divides anticlinally, but at the tip of the strobilus the divisions are also periclinal. Strobili of June 15 show further cell divisions in the young sporophyll and the appearance of rudimentary bundles (*fig. 13*). By July 8 the strobili have lengthened to 6 or 8^{mm}. The new sporophylls appear toward the apex (*fig. 9*), but there is as yet no differentiation of sori. The strobili of July 25, which are 10 to 12^{mm} long, exclusive of the peduncle, show fifteen or more sporophylls in a vertical row (*fig. 14*).

MATURE SPOROPHYLLS AND SPORANGIA

The mature microsporophylls of *Zamia floridana* are short-stalked and broaden outwards from their insertion, the sterile tip being thick and hairy. The microsporangia are borne upon the abaxial surface, and are grouped upon either flank, with the median region of the sporophyll bare. The sporangia extend to the margin but not beyond, so that there is no appearance of a "peltate expansion" of the sterile tip.

The microsporangia of *Ceratozamia mexicana* are borne upon obovate, cuneate sporophylls, whose sterile tips are produced into two horns spreading laterally. The sporangia cover the whole lower part of the sporophyll. The plates given by WARMING (7) show young stages of the sporophyll, when the sporangia are in two groups, one on either flank; but in the text he says that in further growth they become more numerous and spread toward the median region, so that at last this is covered.

In *Encephalartos villosus* the sporophyll has a very short stalk or is sessile; the tip is sterile and produced into a blunt point. The sporangia in the central sporophylls of the strobili cover the surface, but at the apex the outline of the sporangium-bearing area is notched in the center, showing a tendency toward a two-grouped arrangement. Single strobili of *E. Caffer* and of *E. Altensteinii* have been examined, and in both species throughout the median region of the cone the sporangia cover the sporophylls; but the lower sporophylls on the cone and those at the tip show fewer sporangia, and these are grouped into two lateral areas. The transition between these two conditions

can be traced in a series of sporophylls, which show the outline of the sporangium-bearing area becoming notched in the middle region above and below, and the notches gradually deepening until finally the middle region becomes bare. This series also occurs in *Dioon edule;* and in the one cone of *Macrozamia Miquelii* examined it was shown by the sporophylls at the base; but those of the central part and tip were entirely covered with sporangia.

The numbers of sporangia on a sporophyll range as follows: *Cycas circinalis* 700, *Encephalartos Caffer* 700, *Macrozamia Miquelii* 600, *Encephalartos villosus* 500, *Dioon edule* 200, *Zamia floridana* 24.

The sporangia on the outside of the sporophyll flank in *Zamia floridana* and *Ceratozamia mexicana*, also in Stangeria as recorded by LANG (16), are likely to have longer stalks than the sporangia on the central part, and are more rounded. In Ceratozamia they are so crowded that they become angular, often irregular in shape, and encroach upon one another (*fig. 15*); and, as in Stangeria, there are many hairs among the sporangia, arising from the sporophyll. In *Zamia floridana* and *Encephalartos villosus* there are no hairs in the region where the sporangia are borne.

The sporangia are definitely grouped into sori, arising from a cushion of meristematic tissue elevated slightly above the sporophyll (*fig. 16*). Two to five sporangia occur in the sori of Ceratozamia, Stangeria (according to LANG), *Macrozamia Miquelii, Encephalartos villosus*, and *E. Caffer;* two to four in *Cycas circinalis;* and seldom more than two in *Zamia floridana*. Into this soral cushion there is an extension of the vascular system, as in the synangia of Marattiaceae.

LANG (16) found the sporangia of Stangeria in every stage of development on the sporophyll, beginning at the center of each flank and extending toward the margin and middle region. He refers to WARMING's (7) figures of Ceratozamia to show that in that form the sporangia on a sporophyll are of the same age; but WARMING's text distinctly states that at first there are groups of twenty to thirty on each flank, and by further growth these spread so as to conceal the middle region. In my material of Ceratozamia, which was nearly all mature, the sporangia were about the same age; but a few on the edges and toward the center were a little younger.

In Zamia and Ceratozamia there is strong suggestion of the syn-

angial origin of the sorus. In the former, while the sporangia of a sorus are usually quite distinct, occasionally they are free for only about half their length (*fig. 17*); while in the latter this half-synangial character is more common (*figs. 18-20*).

One cone of *Zamia floridana* showed great variation in the number of sporangia on a sporophyll. The usual sporophyll (*fig. 21, a*) bore twelve on each flank, arranged in six pairs. Usually each flank had the same number of pairs, but this was not always the case. The number of sporangia ranged from two to forty-eight; and instead of being confined to the flanks, in several cases there were groups of sporangia on the middle region or even all the way across the sporophyll, approaching the condition in Ceratozamia (*figs. 21, b, c*). The number was likely to be less upon the sporophylls near the tip of the strobilus and upon those at the base. One sporophyll is shown in *fig. 21, d*, which suggests the megasporophyll in the position and number of sporangia; and it may be interesting to note that an ovulate cone furnished several sporophylls with two sporangia on each flank.

DEVELOPMENT OF SPORANGIA

Two papers by WARMING (6, 7) give the earliest information concerning the development of the microsporangium of cycads, the species investigated being *Ceratozamia mexicana* and *Cycas circinalis*. Later, TREUB (8) investigated *Zamia muricata* and LANG (16) *Stangeria paradoxa*, both giving an account of the sporangium from its early stages to its maturity, but neither obtained stages earlier than a several-celled sporogenous tissue.

A tangential section through the strobilus of *Zamia floridana* gives the best view of the origin of the sporangia (*fig. 14*); about three showing upon each lobe of the sporophyll in such a section, the youngest near the margin. The cells in the region where the sporangia arise stain more deeply than the rest of the tissue, and this meristematic condition extends below the sporangia for some distance. The epidermis here divides only anticlinally. In such a section as *fig. 14*, certain cells may be distinguished by their larger size, more deeply staining contents, and larger nuclei in which the chromatin is more prominent. This is the region which will give rise to the sporangia. In such meristematic groups a single hypodermal cell with large nucleus and deeply staining chromatin can be distinguished,

being larger than the surrounding cells and in an axial position (*fig. 22*); this cell is the archesporium. In sections parallel to the surface of the sporophyll this same cell can be distinguished from the surrounding cells (*fig. 25*), so that it is evident that it is the single archesporial cell which GOEBEL (11) predicted in reviewing TREUB'S work. TREUB (8) failed to find in *Zamia muricata* less than four sporogenous cells, and LANG (16) concluded that the corresponding four cells in Stangeria represented the archesporium, which was "probably not a single cell." The archesporial cell divides usually by an anticlinal wall (*fig. 23*), and the two daughter cells lie side by side in a cross-section through the sporophyll; one cell is often larger than the other and divides earlier (*fig. 24*).

Occasionally the archesporial cell divides by a periclinal wall (*fig. 26*), suggesting the statement of BOWER (15) in reference to Angiopteris, in which the divisions are not always in the same direction in different sporangia. This exceptional division is shown also in a section parallel to the surface of the sporophyll (*fig. 27*).

The second division is anticlinal also, resulting in a hypodermal plate of four cells, only two of which are seen in cross-section.

The third division is periclinal, resulting in an outer and inner plate of four cells each (*fig. 24*), the outer plate being the primary wall cells, the inner plate the primary sporogenous cells. Further divisions of the sporogenous tissue are shown in *figs. 28–30.*

The wall is four to seven layers of cells in thickness, always thicker at the angles and in the region of the apex (*figs. 31, 32*); there is some small increase in the number of layers toward the center of the sorus also. The cells of the two layers adjacent to the tapetum are narrow and flattened (*fig. 33*), and later are crushed by the development of the sporogenous cells and the activity of the tapetum. The epidermal cells are thin-walled in Ceratozamia and Zamia up to the time of tetrad formation. At this time in Zamia they begin to show thickening along the crest of the sporangia, where the cells are deeper and narrower. Here the inner and vertical walls become thicker (*fig. 32*), so that by the time spores are formed these cells make a well-defined band along the crest of the sporangium; at the time the spores are ready to be shed this wall has become very thick. The new layers of the wall fill the cell until the lumen almost disappears. The

walls of the cells between the bands, *i.e.* those of the line of dehiscence, are at first very thin, but thicken up later, until they are almost as thick as those of the other cells.

The apex of the sporangium is composed of isodiametric cells (in cross-section) with uniformly thickened walls, beneath which is a group of cells which are thickened in the same way (*figs. 34-36*). From the apex, extending along either side of the line of dehiscence, the cells of the bands are longer, narrower, and have the inner and vertical walls thickened, the bands extending along the crest from the apex almost to the base of the sporangium.

The mature condition of the sporangium of Encephalartos, the only stage of this genus available, is much the same (*fig. 35*).

The older stages of the sporangium of Ceratozamia show the apical group of cells (*fig. 31*) projecting only slightly; and the band cells are little more thickened than the other cells of the sporangium, but are narrower. Cross-sections made from the younger stages (*fig. 37*) show in the wall a double line of cells running from the outside into the sporangial cavity, whose contents stain more deeply. These lie just beneath the line of dehiscence, and are therefore like a plate cutting into the wall from the surface. Just under the line of dehiscence in Marattia there are deeply colored mucilaginous cells which may aid in dehiscence; these slightly resemble those in Ceratozamia. Older stages of the wall do not show this plate; but there are occasionally several cells under the line of dehiscence, in which there is a great number of crystals. These are even more striking in Zamia, where early stages in their formation have not been seen (*fig. 38*).

At the time of spore-formation in Zamia there is also just opposite the cells of dehiscence, on the inner surface of the sporangium, a projection into the wall of the inner layers and of the tapetum (*fig. 38*). The cells between are flattened and out of shape, as if degeneration had begun. This has been seen in a number of sections, and may or may not be a normal occurrence. Enough sections have not been seen to make a definite statement as to the function of these cells, but it may be suggested that there is a breaking down of the walls at the time of dehiscence, which aids in the breaking open of the sporangium. This may be seen in Danaea, where of course there is no mechanical arrangement for dehiscence; and possibly

this breaking down of cells is combined in Angiopteris with the regular mechanical arrangement for dehiscence.

In Zamia the stomata are situated in about the same region of the sporangium as in Stangeria. They are especially numerous among the thinner cells of the lower part of the sporangium, and the guard cells in cross-section are only about one-half as high as the other cells. The walls of the guard cells and of the subsidiary cells are strongly thickened, and the whole apparatus resembles that found among xerophytic angiosperms. The stomata of Ceratozamia are not so deeply sunken; but in Encephalartos they are more deeply placed than in Zamia (*figs. 39, 41*).

In Zamia and Ceratozamia the tapetum (some of it at least) is derived from the sporogenous tissue. The blocking out of the cells and the irregular width of the layer, especially toward the base of the sporangium, suggests this, and in many cases can only be explained by the fact that there is a distinct transformation of sporogenous cells into tapetum. In many sections it is hard to distinguish the tapetum, but in the stage shown in *fig. 42* the nuclei are smaller and a little more elongated, and the chromatin stains more deeply. At the base of the sporangium the irregularity in the width of the tapetum is more striking. Usually there are one or two layers all around the sporogenous mass; but *fig. 43* shows four or five layers, dipping into the sporogenous tissue. The nuclei of the tapetal cells become divided and spindles (*fig. 42*) are occasionally seen, so that although there may be fragmentation, as LANG (16) states, there is also regular mitosis. The tapetum is a distinct layer up to the tetrad stage, when the walls disappear, and the mass of nutritive substance lines the sporangial cavity.

There is some sterilization of sporogenous tissue, for occasionally regions of tissue in the center of the sporangium or at the base, with extensions into the center, become sterilized, and, as has been mentioned, the cells near the base often appear to function as tapetal cells.

The division of the spore mother cell was observed in *Ceratozamia mexicana* (*fig. 44*). The spirem in the nucleus of the mother cell is large and clear, the chromosomes are thick and short, so that the chance for counting is unusually good. A count was made in 50 nuclei, resulting in 12 chromosomes in 46 cases, 11 in 3 cases, and 13

in one case. In most of these cases the 12 chromosomes were visible in one section 3 to 5 μ thick (*fig. 45*), but in a few cases the sections on either side had to be examined and drawings made to determine the actual number of chromosomes. This in every case changed what would have been a count of 6 or 8 to 12. GUIGNARD (12) found 8 chromosomes in the pollen mother cells of Ceratozamia; and OVER-TON (13) found the same number in a count of the chromosomes in the endosperm cells of the same species. In the 25 nuclei of *Zamia floridana* counted, 12 chromosomes were found; and CHAM-BERLAIN (27) gives the same number for *Dioon edule*.

Most of the stages in the development of the tetrad were seen in Zamia, and they followed closely the figures of JURANYI (4) and TREUB (8). In most of the tetrads the ring of cellulose was not seen all the way across the cell until the four daughter nuclei were clearly outlined and the cell plate between them had commenced to grow.

In *Zamia floridana* the output of spores per sporangium is about 500 or 600; in *Ceratozamia mexicana* 8,000; and in *Encephalartos villosus* 26,000. This makes the output per sporangium in the few species examined increase according to the number of sporangia to the sporophyll. The number of observations, however, is too small to furnish a safe basis for conclusions. Among the Marattiaceae the largest output is for Kaulfussia (7,850); and in Isoetes SMITH (18) estimates several hundred thousand spores in a single microsporangium.

THE MALE GAMETOPHYTE

The strobili of Zamia received from Florida the first of February were about ready to shed the pollen, which was in the three-celled stage. At the first observed division of the spore the prothallial cell is cut off, lying against the wall of the spore, with its inner face arching almost into the center of the spore (*fig. 46*). At the next division the generative cell is cut off (*fig. 47*), its nucleus being somewhat flattened and deeply staining. The tube nucleus is very large, stains less deeply, and lies generally in line with the other two.

A 10 per cent. sugar solution, sterilized and kept in sterilized dishes, was favorable for the growth of the pollen tubes. Some of the solutions containing pollen grains were kept in an oven at a temperature of 28° C., while others were kept at 21° C., the temperature of the room. In forty-two hours the tubes in the oven were two or

three times longer than those in the room (*figs. 48, 49*). These cultures gave an opportunity to study the breaking of the exine and the extrusion of the tube (*fig. 50*). In the unstained specimens the portion of the wall just touching the prothallial cell and passing down either side of it was more refractive than the rest. In stained material (*figs. 48, 49*) this had the appearance of a lighter wall just within the intine, and agrees with the "third wall" described by Miss FERGUSON (24) in Pinus. The starch in the growing tube is very abundant, massing about the tube nucleus and the generative cell (*fig. 49*); in longer tubes there is also a large quantity near the tip. The tube nucleus slowly migrates toward the tip, keeping just behind it. In Zamia there is no sign of the second prothallial cell mentioned by WEBBER (20), though many hundred microspores were examined.

Microspores of Encephalartos, which are very large, were especially favorable for a study of the divisions to the three-celled stage (*figs. 51, 52*), which are like those described for Zamia. Only one mitotic figure was seen, but it shows the size and shape of the chromosomes fairly well (*fig. 52*).

Summary

1. The stem of *Zamia floridana* is a sympodium, a vegetative point lying at the base of each strobilus. The staminate strobili develop one after the other from the successive vegetative points, each strobilus with a small circle of leaves, and all enclosed within the larger scale leaves of the first strobilus.

2. The youngest staminate strobili received June 1 showed the sporophylls arising in acropetal succession, by a periclinal division of a hypodermal cell, and later by the divisions of other hypodermal cells and layers beneath them.

3. The microsporangia cover the abaxial face of the sporophylls in *Ceratozamia mexicana*, and are grouped upon each flank in Zamia. In Dioon, *Encephalartos villosus, E. Caffer*, and *Macrozamia Miquelii* there are intergrades, where the sporangia cover the sporophylls of the central part of the strobilus, but are in two groups upon the sporophylls of the tip and base.

4. The microsporangia are grouped in sori (two to six sporangia in each sorus), which are raised on a cushion of tissue into which a vascular bundle passes.

5. In Zamia and Ceratozamia the sporangia of a sorus are sometimes free only half their length.

6. One strobilus of *Zamia floridana* had two to forty-eight sporangia on its different sporophylls, and some sporophylls in which the middle region was entirely covered.

7. Strobili of *Zamia floridana* received July 25 showed in tangential sections three or four developing sporangia upon either flank of the sporophyll, the youngest upon the margin.

8. The archesporium is a single hypodermal cell which usually divides first anticlinally, followed by periclinal divisions; the outer plate of four cells developing the wall layers, the inner plate the sporogenous tissue.

9. The wall of the mature microsporangium is composed of four to seven layers of cells, the cells of the two layers next the tapetum being tabular. The apex of the sporangium consists of thick-walled cells and beneath it there are isodiametric, thick-walled cells. Extending from the apex on either side of the line of dehiscence there is a band of thicker-walled cells, which suggests the structure of the sporangium of Angiopteris.

10. Beneath the line of dehiscence there is a plate of cells extending toward the center of the sporangium, which contain crystals; and in Zamia these cells seem to degenerate as the sporangium breaks.

11. Stomata occur on the microsporangium, the guard cells being deeply sunken and thick-walled.

12. The tapetum is derived from sporogenous tissue, at least in part, and the sporogenous cells at the base of the sporangium function as tapetal cells, which may project into the sporangial chamber. Sometimes, at least, the tapetal nuclei divide mitotically, for spindles were seen; and there are often two nuclei to a cell. The tapetum remains a distinct layer up to the tetrad stage.

13. There is some additional sterilization of sporogenous tissue in the sporangium.

14. The divisions of the spore mother cells conform to the descriptions of JURANYI and TREUB. The reduced number of chromosomes in Ceratozamia and Zamia is twelve.

15. The output of spores per sporangium for *Zamia floridana* is 500–600, for *Ceratozamia mexicana* 8,000, and for *Encephalartos*

villosus 26,000. Therefore the output per sporangium in these species increases according to the number of sporangia upon the sporophyll.

16. The pollen grains are three-celled at the time of shedding, the cells being the prothallial, the generative, and the tube.

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EXPLANATION OF PLATE X

(Unless otherwise stated, the figures are from Zamia.)

FIG. 1. Diagram of the crown; *a*, peduncle of ovulate strobilus of last year; *b*, young ovulate strobilus; *v*, position of the vegetative point. ×1.

FIG. 2. Longitudinal section of two ovulate strobili; between them is the vegetative point (*v*) and the leaves (*l*) surrounding it; the upper portion of the strobilus is cut away. ×13.

FIG. 3. Longitudinal section of ovulate strobilus; *v*, vegetative point; *l*, rudimentary leaves; two vascular bundles enter the strobilus. ×23.

FIG. 4. Longitudinal section of staminate cone (June 14); *v*, vegetative point; *l*, rudimentary leaves. ×23.

FIG. 5. Longitudinal section of three staminate strobili (July 15); *v*, vegetative point; youngest strobilus (*st*) is cut at an angle. ×11.

FIG. 6. Diagrams: *a*, sympodium; *b*, sympodium with shortened internodes; *c*, sympodium with internodes shortened until strobili are on a level, as in Zamia.

FIG. 7. Longitudinal section of young staminate strobilus (June 1), showing primordia of sporophylls (*sp*) and leaves surrounding strobilus. ×27.

FIG. 8. Longitudinal section of typical vegetative point and rudimentary leaves. ×32.

FIG. 9. Longitudinal section of staminate strobilus (July 8), showing sporophylls. ×23.

FIG. 10. Longitudinal section of staminate strobilus a little further advanced than in *fig. 7.* ×32.

FIG. 11. Section of staminate strobilus of *fig. 7*, showing the periclinal division of a hypodermal cell which is the first stage in the development of a sporophyll. ×650.

FIG. 12. Section of staminate strobilus, showing further divisions of hypodermal cells and layers beneath them to form sporophyll. ×650.

FIG. 13. Longitudinal section of strobilus, showing elevation of sporophylls and rudimentary bundles passing into them. ×80.

FIG. 14. Tangential section of staminate cone of July 25, showing sporophylls and position of young sporangia. ×27.

FIG. 15. Cross-section of sporophyll of Ceratozamia, showing sporangia. ×37.

FIG. 16. Cross-section of sporophyll of Ceratozamia, showing grouping of sporangia into sori and extension of bundles into them. ×27.

FIG. 17. Cross-section of sporophyll of Zamia. ×39.

FIGS. 18, 19. Three sporangia of Ceratozamia "united" at base. ×95.

FIG. 20. Detail of *fig. 19*, showing union of sporangia at base. ×500.

FIG. 21. Sporophylls from one cone of *Zamia floridana:* a, usual arrangement of sporangia; b, sporangia extending to middle region; c, sporangia extending across middle region; d, reduced number of sporangia. ×2.

FIG. 22. Sporophyll of *fig. 14*, showing hypodermal archesporial cell. ×920.

FIG. 23. Division of archesporium by an anticlinal wall. ×920.

FIG. 24. The periclinal divisions resulting in inner and outer plates of cells. ×920.

FIG. 25. Archesporial cell in a section parallel to the surface of the sporophyll. ×920.

FIG. 26. Periclinal division of archesporial cell (exceptional). ×920.

FIG. 27. First (anticlinal) division of archesporial cell in a section parallel to the surface of the sporophyll. ×920.

FIG. 28. Further periclinal divisions succeeding the division shown in *fig. 26*. ×920.

FIG. 29. Usual divisions following the stage shown in *fig. 24*; lower four cells of section are sporogenous. ×920.

FIG. 30. Longitudinal section of sporangium in more advanced stage. ×920.

FIG. 31. Longitudinal section of sporangium of Ceratozamia; tapetal cells marked by +. ×180.

FIG. 32. Longitudinal section of sporangium of Zamia, showing thickened cells at apex and band of dehiscence. ×95.

FIG. 33. Detailed drawing of part of sporangium: w, outer wall layers; y, two layers of tabular cells, which are crushed by the activity of the tapetum; t, tapetum; s, sporogenous cells. ×800.

FIG. 34. Apex of sporangium of Zamia, showing thick-walled cells; isodiametric cells beneath them. ×580.

FIG. 35. Diagram of sporangium of Encephalartos.

FIG. 36. Tangential section through sporangium of Zamia near apex, showing thick-walled cells and line of dehiscence (d). ×180.

FIG. 37. Cross-section of sporangium of Ceratozamia, with plate of cells extending from beneath line of dehiscence to the sporangial cavity. ×500.

FIG. 38. Cross-section of mature sporangium wall; *c*, cell containing crystals, just beneath the line of dehiscence; cells of inner layers of sporangial wall projecting into outer layers; *t*, tapetum broken down; *m*, microspore. ×800.

FIG. 39. Longitudinal section of two sporangia of Encephalartos. ×70.

FIG 40. Surface section of stoma in the sporangial wall of Encephalartos; *g*, guard cells; *su*, subsidiary cells. ×650.

FIG. 41. Cross-section of stoma of Encephalartos. ×650.

FIG. 42. Detail of sporangium of Ceratozamia: *t*, tapetum; *sp*, sporogenous cells. ×120.

FIG. 43. Longitudinal section of sporangium of Ceratozamia at the base; tapetal cells becoming more numerous by sterilization of sporogenous tissue. ×180.

FIG. 44. First division of pollen mother cell. ×1500.

FIG. 45. Nuclear plate in spore mother cell, showing 12 chromosomes. ×1500.

FIG. 46. Male gametophyte of Encephalartos in 2-celled stage; *p*, prothallial cell; *a*, antheridial initial. ×1500.

FIG. 47. Male gametophyte of Encephalartos in 3-celled stage; *p*, prothallial cell; *g*, generative cell; *t*, tube cell. ×1500.

FIG. 48. Male gametophyte of Zamia, showing beginning of tube; lettering as before; *z*, "third wall," showing within intine. ×1500.
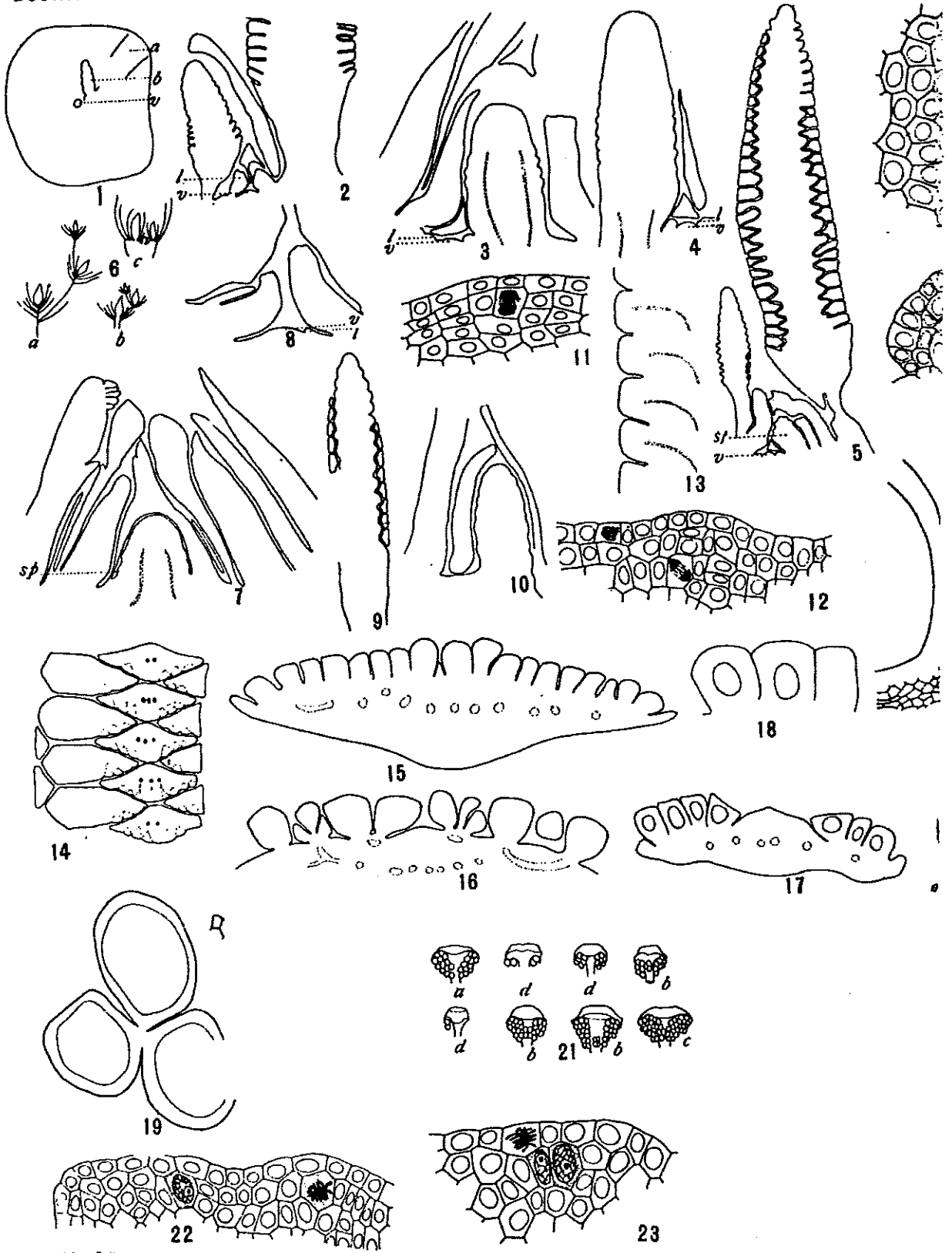
FIG. 49. Further development of pollen tube; tube nucleus passing into the tube; much starch present. ×1500.

FIG. 50. Culture from Zamia, showing extrusion of tube and breaking of exine. ×1500.

FIG. 51. Microspore of Encephalartos in resting stage. ×1500.

FIG. 52. Microspore of Encephalartos, showing some of the chromosomes. ×1500.

The magnifications given in the foregoing explanations are those of the original drawings, which have been reduced three-fifths in reproduction.

PLATE X