STUDIES IN PTERIDOPHYTES

IX. The Root Apex Organization in some Pteridophytes

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The paper describes the organization and structure of the root apex in 10 species of pteridophytes. Three types of root apical organization have been noted : (*i*) with a conspicuous apical cell, (*ii*) with a plate of conspicuous apical initials, and (*iii*) with a group of inconspicuous apical initials (fern allies). *Osmunda* shows two types of apical organization, *i.e.* some roots with a conspicuous cell and others with a group of cells. A region resembling the quiescent centre has been observed in the ferns presently studied.
Two prominent features of the present study are (i) the apparent inactivity and prismatic appearance of the pyramidal cell in *Actiniopteris, Lygodium,* etc., and the unusual columnar cells of *Angiopteris,* and (ii) the types of roots studied can be arranged in the ascending order—*Lygodium,* etc., *Angiopteris,* and *Isoetes,* viewing from the complexities of root apex organization.

Introduction

Root apex organization in vascular plants is interesting and has attracted the attention of botanists for a long time (*see* Eames and MacDaniels 1951; Esau 1965; Clowes 1965; Wardlaw 1968). It was, however, once considered to be very simple in pteridophytes. The occurrence of a single cell with 3- or 4-cutting faces was believed to be common. Lately, it has been shown to be complex, with considerable variation and specialization (Bower 1923, 1935; Sifton 1944; Bhambie 1963). The present communication presents data on the root apex structure in 10 species of pteridophytes.

Materials and Methods

The following materials were collected from places mentioned below :

	Name of the material	Place	Date of collection
I.	Leptosporangiate ferns :		
	Actiniopteris radiata (Swartz) Link.	Kanouj (U.P.)	August 1965
	Lygodium flexuosum (L.) Swartz	Calcutta (W. Bengal)	January 1964

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	Microsorium alternifolium (Willd.) Copel.	Ranikhet (U.P.)	October 1964
	Nephrolepis exaltata (L.) Schrott.	Pachmarhi (M.P)	October 1969
	Osmunda regalis L.	Pachmarhi (M.P.)	October 1969
II.	Eusporangiate ferns:		
	Angiopteris evecta Hoffm.	Near Pathanamthitta (Kerala)	December 1968
	Ophioglossum costatum R. Br.	Pachmarhi (M.P.)	October 1969
	O. nudicaula L.	Pachmarhi (M.P.)	October 1969
III.	Fern allies :		
	Isoetes coromandelina L.	Meerut (U.P.)	August 1956
	I. panchananii Pant and Sriv.	Pachmarhi (M.P.)	October 1963

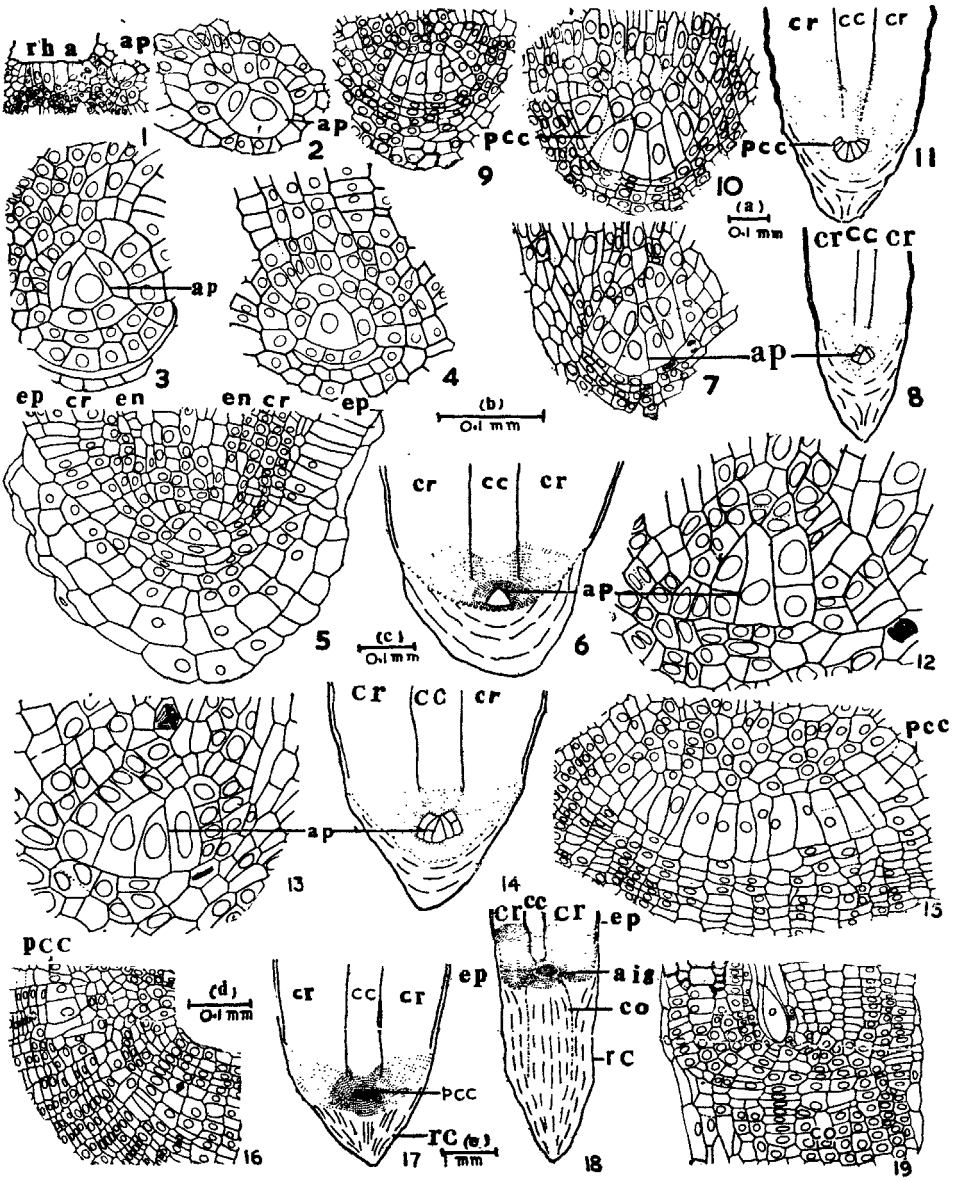
The materials were fixed in Formalin-Acetic-Alcohol and Acetic-Alcohol, washed with water and preserved in 70 per cent alcohol for study. Customary methods of dehydration and embedding were followed. Heidenhain's haematoxylin-fast green and safranin-light green combinations were used for staining.

OBSERVATIONS

In leptosporangiate ferns, e.g. *Lygodium flexuosum, Actiniopteris radiata, Microsorium alternifolium* and *Nephrolepis exaltata,* the roots are adventitious and originate endogenously in the rhizome at the base of very young leaf primordia. The initiation and further development of the root primordium has been studied in *Lygodium flexuosum.* A cell in the growing region of the rhizome apex enlarges showing pyramidal appearance (Figs. 1, 2) and produces cells laterally on all its faces (Figs. 3,4). It appears that the surrounding cells in the expanding region of the rhizome apex arrange themselves around the pyramidal cell and its derivatives, and divide to form the developing root primordium almost next to the rhizome apex (Fig. 1). Further development of young root primordium is shown in Figs. 2-5. A central pyramidal cell with four well-defined faces and a number of surrounding cells usually in two tiers are seen (Fig. 5). The procambial tissue differentiates between this group and the differentiating vascular tissue of the rhizome.

In all the leptosporangiate ferns studied presently, a mature root apex is somewhat obtuse and is protected by a conical cap. It has a characteristic large, inverted pyramidal apical cell having four cutting faces — three directed towards the body, while the fourth one faces the root cap (Figs. 5, 6). The cells derived on the fourth surface form the root cap while others in lateral position organise the tissues of the root. In these cells the stain absorption and cell division are variable. At its inception the apical cell is densely stained and shows prismatic outline subsequently. It divides less frequently unlike the densely cytoplasmic cells adjacent to it (Fig. 6).

In *Osmunda regalis,* a variation from the above condition is observed; the roots have broad or conical apices. In the root with conical apex a single pyramidal



FIGS. 1-19 *Lygodium flexuosum*. 1, L S. of the rhizome apex showing initiation of root primordium; 2-5, L. S. through young root tips showing successive stages in the development of the root; 6, same, semidiagrammatic; 7-11, *Osmunda regalis*. 7, 9 and 10, a part of the central region of three different root apices in L. S. (note a conspicuous single cell in Fig. 7 and a group of cells in Fig. 9); 8 and 11, same, semidiagrammatic; 12-14. *Ophioglossum nudicaule*. 12 and 13, a part of central region of two different root apices in L. S.; 14, same, semidiagrammatic; 15-17. *Angiopteris evecta*. 15, a part of the central region of a root in L. S; 16, same, a part of thelateral side (note activity in the surrounding region); 17, L. S. root, semidiagrammatic; 18-19, *Isoetes coromandelina*. 18, L. S. root; 19, same, semidiagrammatic.

(*aig*, apical group of initials ; *ap*, apical cell ; *cc*, central cylinder ; *co*, columella ; *cr*, cortex; *en*, endodermis ; *ep*, epidermis, *pcc*, plate of columnar cells; *rc*, root cap; *rha*, rhizome apex.)

cell has been observed like in other leptosporangiate ferns (Fig. 8). Its derivatives do not divide frequently. In some cases a plate of five cells, sometimes more, can be observed. All these cells are prismatic in appearance in comparison to the cells surrounding them which are densely cytoplasmic and show division figures (Fig. 7). In roots with broad apices a plate of columnar prismatic cells is present in the central region of the apex (Fig. 9). The cells surrounding this region are densely cytoplasmic and meristematic (Fig. 11). In some broad apices a somewhat pyramidal cell is seen in the centre of this group of cells (Fig. 10). A study of a number of root apices shows that there is a single apical cell in the beginning and subsequently in the same region a group of equally prominent columnar cells appear, which presumably are derived from the former. The cell division activity is transferred to the surrounding cells of this group.

In the eusporangiate ferns, mature root apices of *Ophioglossum costatum, O. nudicaule* and *Angiopteris evecta* have been studied. In both the species of *Ophioglossum,* a single pyramidal cell, centrally placed, with four cutting faces has been observed. The cells formed by its activity on its lateral sides may divide by a transverse division or this division may be delayed (Figs. 12–14). Thus in *Ophioglossum* the central cell is replaced by a group of cells. The surrounding smaller cells show division figures in various planes.

In *Angiopteris evecta,* the root apices show centrally placed, elongated columnar cells of almost equal size. Their number, however, varies considerably depending upon the thickness of the root (Fig. 15). These cells are distinguishable from the neighbouring cells by their larger size and rectangular shape as seen in longitudinal sections. They form a sort of concave plate, when observed in transverse section. In young roots these cells are fewer in number, densely cytoplasmic and appear to be active. In mature roots these cells are bigger, prismatic and vacuolated. The surrounding cells of the plate are meristematic, arranged in regular rows, and show division figures (Figs. 15–17). It appears, therefore, that the meristematic activity might have been shifted from the central cells to the surrounding cells. The well known 'T' or 'Y' divisions found in angiospermous roots have been observed frequently in the body of the root as well as root cap.

In *Isoetes* (*I. coromandelina* and *I. panchananii*), the stele of the axis has two well-demarcated regions, *i.e.* the stem stele which sends off leaf traces and a rhizomorphic stele which gives off root traces. The rhizomorphic stele has a thick multilayered, self-perpetuating basal meristem, which is also the place of origin of the roots. The first sign of root formation is the appearance of a group of cells. To begin with they divide irregularly but later on transverse divisions predominate resulting into two regions—a central cylinder and an outer inverted cup-shaped structure (for details *see* Bhambie 1963).

A mature root apex shows a common group of centrally placed cells (Fig. 19), which appears to be responsible for the formation of the root cap as well as the body. A more or less inverted cupe-like structure can be seen in the mature root apices, the base of which is occupied by the initials. The lateral sides of this structure form the epidermis and outer cortex while the base contributes to the inner cortex and the stele (Figs. 18, 19). In mature root cap, a central well-developed columella of stratified cells and lateral peripheral parts with elongated cells can be distinguished.

In the species mentioned above the tissue differentiation and gross root anatomy have also been studied. To begin with, the central cylinder is recognized from the surrounding tissue of the cortex by the small size of its cells. A layer of prominent cells differentiates between the procambial region and the cortex to form the endodermis. The small cells of the central cylinder show marked elongation and form the vascular tissue and pericycle. In *Isoetes* and *Actiniopteris*, the pericycle is not recognisable. In *Isoetes, Lygodium, Actiniopteris, Osmunda* and *Nephrolepis*, four to six central cells, and in *Angiopteris*, a number of small cell patches, slightly away from the centre, increase in number and cells become vacuolated. Cells very near to these vacuolated ones and towards the periphery give rise to protoxylem. The metaxylem elements are differentiated later from the vacuolated cells. The groups of sieve elements vary in different genera; one (*Isoetes, Ophioglossum*), two or three (*Lygodium, Actiniopteris, Osmunda* and *Nephrolepis*), or more (*Angiopteris*) groups forming small arches at regular intervals. The first phloem elements mature before the differentiation of protoxylem elements. The mature roots exhibit considerable variation. In *Isoetes* the xylem arrangement is monarch, in *Ophioglossum* monarch or diarch, in *Actiniopteris* diarch, in *Lygodium, Osmunda* and *Nephrolepis* di- or triarch, and in *Angiopteris* polyarch, with 12 or more xylem strands. In *Ophioglossum*, the endodermal cells are big, comparable with the cortical cells, and in *Lygodium* endodermis is double layered. No variations are seen in others.

Discussion

Studies on the architecture of root apical meristem have often been made because of the variation in the tissue pattern and arrangement (Sifton 1944; Eames and MacDaniels 1951; Guttenberg 1964; Clowes 1961, 1965; Esau 1965). It was Nägeli (1845) who proposed the "apical cell theory" which referred to a single prominent cell as the structural and functional unit. This view has become obsolete for higher cryptogams, although it is still applicable to the leptosporangiate ferns. In the present study also a conspicuous tetrahedral cell is observed at the central region of the apex in *Lygodium, Actiniopteris, Microsorium, Nephrolepis,* and in some apices of *Osmunda* as well as in *Ophioglossum*—a eusporangiate fern. Its developmental behaviour is similar to that of other ferns. In *Lygodium* the apical cell divides during early development of the root but no divisions were seen subsequently. The apical cell in *Osmunda* is replaced by a group of columnar cells but they seldom divide unlike the surrounding tissue. Such apices of *Osmunda*, therefore, simulate a condition observed in a eusporangiate fern.

The apical cell theory was replaced by the "Histogen theory" of Hanstein (for details, *see* Eames and MacDeniels 1951; Guttenberg 1964). On the basis of the plane of division demarcation of the body and root cap, Schüepp put forward the "Körper-kappe" concept (for details, *see* Esau 1965). He also drew some displacement curves either from a single cell or from a multicellular promeristem, considering it as the cytogenerative centre and explained different root apical configurations. Clowes (1961) proposed another concept known as "Quiescent centre concept" which is said to be a modification of the histogen theory though it replaces the initial of the tissue region outside the distal region (Esau 1965, p. 117). Recently, Clowes (1965) emphasized that most roots have quiescent centre though its size and degree of

quiescence may vary. Guttenberg (1964), however, considered it as an intermittent centre, as the cells of the central region of a root apex may also divide at long or short intervals. He has mentioned the occurrence of a quiescent centre in very old roots only. Clowes (1965) derived fern roots from a single tetrahedral apical cell which cuts off a daughter cell from each of its faces on a regular sequence in the beginning. Later on it does not divide and remains there as a structural unit. The adjacent cells, however, show division figures indicating that the growth activity is shifted from the apical cell to these adjacent cells. Our observations on four leptosporangiate ferns (*Lygodium, Microsorium, Actiniopteris* and *Nephrolepis*) and one eusporangiate fern (*Ophioglossum*) are in agreement with the views of Clowes (1965). D'Amato and Avanzi (1965) have also proposed a 'unicellular quiescent centre' in *Marsilea strigosa* roots. The apical cell in the leptosporangiate ferns divides very rarely. In *Angiopteris*, several initials are arranged in one plate. They not only increase in size and number with the age of the root (Wardlaw 1968) but also become prismatic in appearance. Division figures are usually absent in the central cells of the plate though division figures commonly occur in the cells around the plate. A somewhat similar condition has been noticed in *Osmunda*. On the contrary, a single conspicuous cell like that of leptosporangiate ferns, is noticed in *Ophioglossum* as observed by Joshi (1940), Nozu (1956) and Peterson (1968). The derivatives of this cell sometimes remain undivided and form a group of three or more conspicuous cells at the centre.

In *Isoetes* roots no quiescent zone has been observed. However, a thorough study reveals that all parts of the root originate from a common group of initials (c.f. Bhambie 1963). This can be compared with that in the Ranales and certain gymnosperms (Esau 1965; Pillai 1966).

Janczewski (1874), Haberlandt (1914), Schüepp (1926) and Guttenberg (1964) recognized five, six, seven and four types of root apices respectively in vascular plants on the basis of structural complexities without any mention of evolutionary lines. Voronin (1956), however, considers root apices with common initials as phylogenetically primitive—a viewpoint supported by Pillai and his school on the basis of extensive studies on a number of angiosperms and gymnosperms (Pillai 1963, 1966 Pillai *et al.* 1961, 1965). In the present study a conspicuous apical cell in most of the leptosporangiate ferns, a plate of conspicuous apical initials in the eusporangiate ferns and a group of conspicuous initials in the fern allies have been observed, which indicate that there is no uniformity in the organization of the root apices. *Actiniopteris, Angiopteris* and *Isoetes* appear to form an ascending series so far as the complexities of the root apex organization are concerned, considering the condition found in *Osmunda* and *Ophioglossum* as an intermediate stage between the forms with a single conspicuous cell and a group of cells. It is not suggested that this series constitutes a phylogenetic trend since ontogenetic studies may have very little to contribute to phylogenetic speculations (Bhambie and Puri 1963).

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*originals not seen