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Anatomical aspects of *Asplenium adiantum-nigrum* L.

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The root of *Asplenium adiantum-nigrum* L. has a diarch structure, with modified layers of cortex around the stele and a monolayered pericycle. The rhizome (analyzed by serial transections) consists of an epidermis, a parenchymatous cortex and a centrally located dictyostele containing a variable number of meristeles. The leaf petiole and rachis exhibit a differential cortex (a sclerenchymatous hypodermis and an inner parenchyma). Close to the rachis tip, the sclerenchymatous hypodermis disappears. Remarkable are the variations in the organization of the vascular system. At the base of the leaf petiole, the vascular system is distelic, while up to the subterminal rachis, the monofascicular stele is X-shaped. The pinnulae have a homogenous mesophyll composed of branched and lobed cells. The veins have a collateral (midrib) and hadrocentric structure. The lower epidermis possesses many stomata of the polycytic and anomocytic types.

Key words: anatomy, root, rhizome, pinnulae, *Asplenium adiantum-nigrum, fern.*

Abbreviations used in figures: BS = bundle sheath, C = cortex, E = epidermis; Ed = endodermis; EC = epidermal cell, GT = ground tissue, H = hypodermis, IC = inner cortex, LE = lower epidermis, LVB = lateral vascular bundle, Mr = midrib, Mz = mesophyll, Mx = metaxylem, Pc = pericycle, Ph = phloem, Px = protoxylem, PT = parenchyma tissue, S = stoma, Sc = sclerenchyma, ScS = sclerenchyma sheath, SC = subsidiary cell, SS = starch sheath, UE = upper epidermis, X = xylem.

INTRODUCTION

Asplenium adiantum-nigrum L. (syn. *Asplenium andrewsii* A. Nels.) is a member of the Aspleniaceae family and derived from a cross between *A. onopteris* L. and *A. cuneifolium* Viv. It is known as black spleenwort. Black spleenwort is a short fern (10-14 cm high) with rather leathery and glossy leaves. The lowest pinnules of the middle pinnae are 6-10 mm long and the lowest pinnae 2-6 cm long. The midrib of the pinna has a characteristic winged appearance. The lowest pinnae are the longest. The stem is black at the base and green at the top. The spore cases are linear, lie on the veins and cover a large surface of the underside of the pinna. The knowledge on the variations of the vascular system organization of the fern vegetative organs is quite limited, and that of *Asplenium trichomanes* L. is almost lacking.

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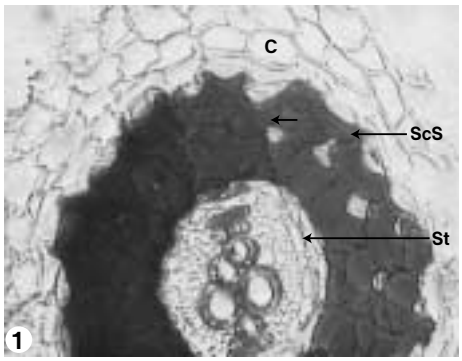
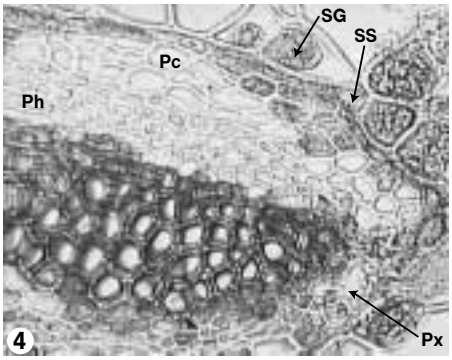
MATERIALS AND METHODS

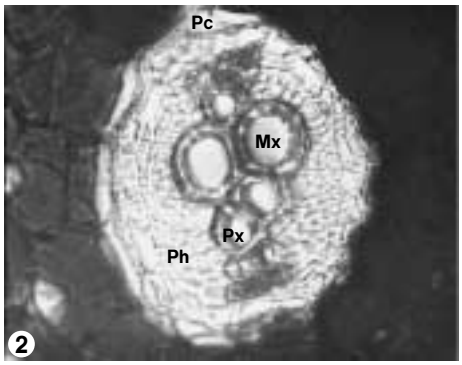
Cross sections of the root, rhizome and leaf were performed using a rotary microtome. The samples were stained with alum-carmine and iodine green and were embedded in glicerine-gelatine. The observations were made with a BIOROM-T bright field microscope, equipped with a TOPICA-1006A video camera. The microphotographs were obtained from the video camera through a computer.

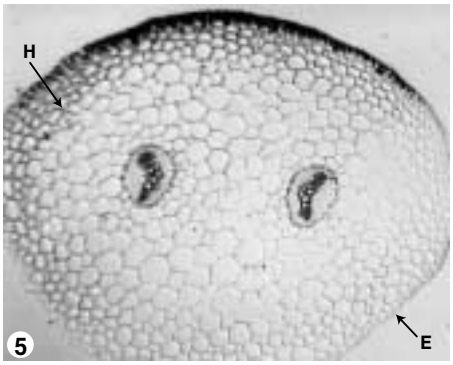
RESULTS AND DISCUSSION

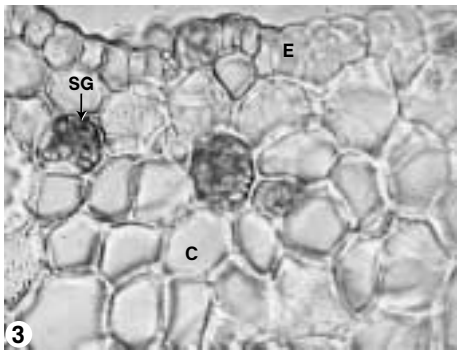
Cross sections of the root revealed that the cortex is composed of 3-4 layers of large parenchyma cells (Fig. 1). The inner cells of the cortex are modified and have thick walls. Kroemer (1903) has suggested that this wall thickening is the result of cutinized blade superpositions. A long time ago, some authors have noticed the presence of such "curious cells"

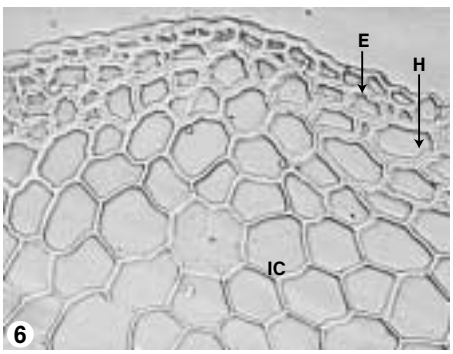












FIGS 1-2. Cross sections of the root.

FIGS 3-4. Cross sections of the rhizome.

FIGS 5-7. Cross sections of the petiole base.

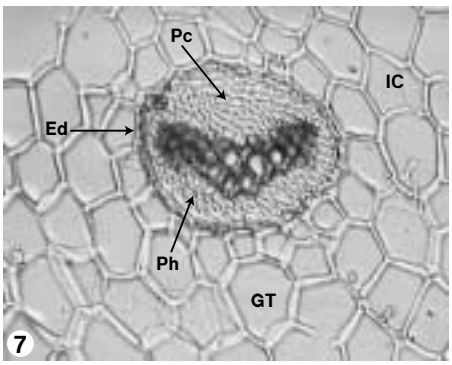
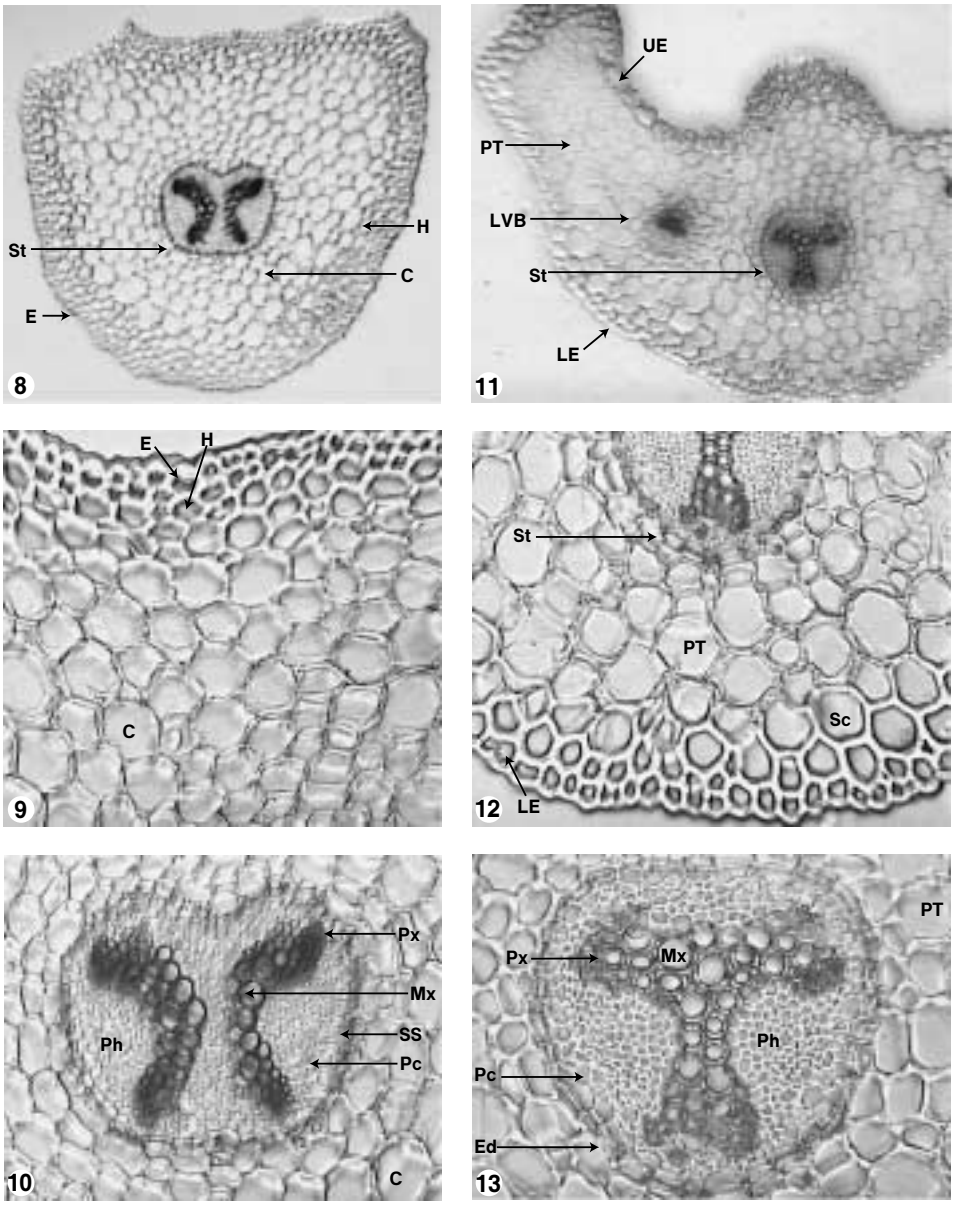


FIG. 1. Partial view of the cortex and the stele (× 140).
FIG. 2. Detail of the stele (× 400).
FIG. 3. Portion of the epidermis and cortex (× 170).
FIG. 4. Portion of a meristele (× 170).
FIG. 5. General view (× 60).
FIG. 6. Portion of the epidermis and cortex (× 240).
FIG. 7. One meristele of the bifascicular stele (× 240).

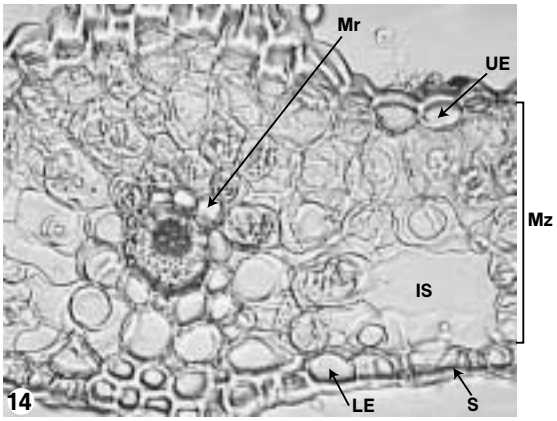


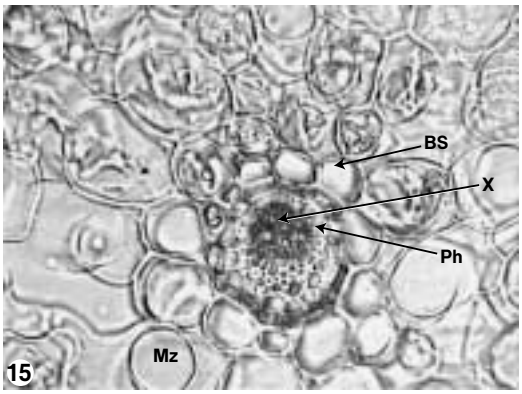
FIGS 8-10. Cross sections below the subterminal rachis.

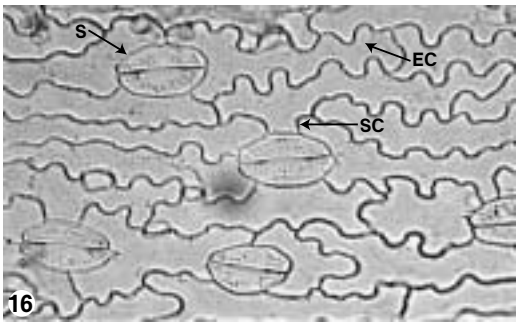
FIG. 8. General view (× 80).
FIG. 9. Portion of the cortex (× 200).
FIG. 10. Detail of the stele (× 200).

FIGS 11-13. Cross sections at the subterminal rachis.

FIG. 11. General view (× 100).
FIG. 12. Portion with epidermis and ground tissue (× 200).
FIG. 13. Detail of the stele (× 280).







FIGS 14-15. Cross sections of the leaf blade.

FIG. 14. Portion with mesophyll and midrid (× 270).

FIG. 15. The midrib (× 400).

FIG. 16. Paradermal section of the lower epidermis (× 270).

around the stele and have suggested that this tissue beloned to the stele naming it "sclerenchymatous mass" (Russow, 1872; Bierhorst, 1971) or "stereomic sheath" (de Bary, 1877; Ogura, 1938; Bercu, 1998; Bercu *et al*., 2000). This configuration has led Schneider (1996) to include this type of root to that of *Asplenium*.

The stele consists of xylem and phloem and is surrouded by the pericycle (Fig. 2). The xylem elements are joined together towards the center by their metaxylem vessels (two for each bundle). The protoxylem vessels (five for each bundle) are in an exarch position and face the pericycle. The phloem sieve cells lack companion cells and are located on either side of the xylem string (Fig. 2). This attributes to the adventitious root a diarch structure.

Cross sections of the rhizome disclosed that the epidermis consists of a single layer of cells uncovered by cuticle (Fig. 3). Below epidermis is the cortex, which consists of large parenchyma cells. Remarkable is the abundance of starch grains in the cortical cells (Fig. 3). As Ogura (1938) and Bir (1957) have reported for Aspleniaceae species, the stele is a dictyostele composed of a variable number of meristeles (in accordance with the number of foliar traces), each surrounded by its own endodermis (starch sheath) and pericycle. The latter is composed of parenchyma cells regularly arranged in one row, (locally in two or even three rows). Each meristele is hadrocentric with a binary structure (metaxylem vessels towards the center, and protoxylem elements facing the pericycle) (Fig. 4). The pith occupies the central area of the rhizome.

Transections of the petiole and rachis cut from the base to the tip of the leaf revealed almost the same succession of tissues, that is a monolayered epidermis covered by cuticle, a cortex and a centrally located stele (Fig. 5). Under epidermis is the hypodermis, which consists of a few layers of compactly arranged sclerenchyma cells. The hypodermis is internally followed by a region of ground tissue (Fig. 6). The vascular system of the leaf petiole base is composed of two meristeles, unequal in size (distelic) (Fig 7). Each meristele is surrounded by an endodermis bearing a Casparian strip and a "special pericycle" (Andrei, 1978). The meristeles are composed of centrally located xylem elements surrounded by phloem. Metaxylem vessels are in a central position and protoxylem elements have an exarch arrangement. This attributes to the vascular bundles a hadrocentric structure (Fig. 7). Cross sections cut from the leaf petiole base up to the subterminal rachis disclosed a unistratose epidermis composed of cutinized-walled cells covered by a thick cuticle, a cortex and a stele (Figs 8-10). The stele is monofascicular, composed of metaxylem vessels arranged first in an X-shape, characteristic of the Aspleniaceae (Fig. 10) (Bir, 1957; Ogura, 1972; Bercu, 1997/98; Bercu *et al*., 2000). The phloem surrounds the xylem elements.

Transections of the subterminal leaf rachis exhibited a T-shaped arrangement of the xylem elements (Figs 11, 13). Remarkable is the reduced

number of vascular elements because of the marginal formation of the pinnulae veins. Under the lower and upper epidermises of the rachis, sclerenchyma cells are present (Fig. 12). They provide mechanical support to the weak and delicate rachis tip. A typical hypodermis is absent.

The pinnulae appear in cross section to consist of a single layer of irregularly arranged epidermal cells, covered by cuticle (Fig. 14). On the lower epidermis, numerous polycytic and anomocytic stomata occur (Cothem van, 1970; Dilcher, 1974) (Fig. 16). The epidermal cells possess wrinkled anticlinal walls. The mesophyll is composed of cells with large intercellular spaces and of vascular bundles (Fig. 14) (Poirault, 1893). The poorly developed vascular system of the midrib is represented by a collateral bundle with xylem in an excentric position and hadrocentric for the other small veins embedded in the mesophyll. The bundle is surrounded by a parenchymatous sheath (Fig. 15).

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