

## PERMINERALIZED FERNS FROM THE MIDDLE EOCENE PRINCETON CHERT. I. *MAKOPTERIS PRINCETONENSIS* GEN. ET SP. NOV. (ATHYRIACEAE)

Ruth A. Stockey, Harufumi Nishida, and Gar W. Rothwell<sup>1</sup>

Department of Biological Sciences, University of Alberta, Edmonton, Alberta, T6G 2E9 Canada; Laboratory of Biospherical Sciences, Chuo University, Kasuga, Bunkyo-ku, Tokyo, 112-8551, Japan; and Department of Environmental and Plant Biology, Ohio University, Athens, Ohio 45701, U.S.A.

The occurrence of numerous anatomically preserved specimens of an extinct filicalean fern in the middle Eocene Princeton chert from southern British Columbia, Canada, provides the basis for characterizing a new genus and species of the Athyriaceae. Fossils include narrow horizontal rhizomes with a radial amphiphloic dictyostele and internodes up to 1.5 cm long. Fronds are monomorphic. Adventitious roots are diarch and display a parenchymatous cortex. Stipes diverge in a radial fashion and display two hippocampiform bundles at the base. Distally, the bundles unite into a single trace, and pinnules are produced in an opposite to subopposite arrangement. The stipe, rachis, and pinnules display a continuous U-shaped groove on the adaxial surface. Sori of annulate sporangia are borne below veins on swollen receptacles. The sporangia are characterized by a vertical annulus and a narrow, elongated stalk. Spores are monolet and ellipsoidal, with a psilate exine and a distinctly spiny perispore. *Makopteris princetonensis* gen. et sp. nov. significantly increases our knowledge of the fossil record for the Athyriaceae and documents that essentially modern athyrioids were present in the flora of North America by the early Tertiary.

**Keywords:** paleobotany, Athyriaceae, dictyostele, Eocene, fossil fern, Tertiary.

### Introduction

The middle Eocene Princeton chert of southern British Columbia, Canada, contains one of the best preserved and most diverse assemblages of all permineralized Tertiary floras. Affinities with modern aquatic plants, anatomical modifications for the aquatic habit, and associated freshwater animal remains have allowed for the interpretation of many of the Princeton chert plants as aquatics (Cevallos-Ferriz et al. 1991; Pigg and Stockey 1996). However, organisms from other habitats are also represented in smaller numbers. In addition to conifers of the Pinaceae and Taxodiaceae, 19 dicots, six monocots, and nine fungal species have so far been described, with many others awaiting further description (Pigg and Stockey 1996).

Several filicalean ferns also occur within this diverse fossil biota (Basinger 1976, 1981; Cevallos-Ferriz et al. 1991; Pigg and Stockey 1996), and some species are represented by abundant material that includes all organs of the sporophyte (Rothwell et al. 1994). These species are particularly attractive candidates for the development of whole-plant concepts for extinct species because they display well-preserved internal anatomy. In this article, we reconstruct *Makopteris princetonensis* gen. et sp. nov. from material that includes organs of the vegetative sporophyte, sori, sporangia, and spores. Rachides of this fern were first recognized and briefly described by Basinger, who

suspected that they could represent the same species, as ferns A and D (Basinger 1976, 1981).

Because *Makopteris* is represented by anatomical sections of the rhizome, fronds, roots, sporangia, and spores, it is the most completely known extinct species of the Athyriaceae (*sensu* Pichi Sermolli 1977). It also documents the occurrence of *Diplazium*-like ferns in the early Tertiary of North America. Four additional filicalean ferns from the Princeton chert locality (Rothwell et al. 1994) will be the subjects of separate studies.

### Material and Methods

Chert layers containing material of *Makopteris princetonensis* and interbedded coal seams crop out on the east bank of the Similkameen River, ca. 8.4 km south of the town of Princeton, British Columbia, Canada. This deposit is located 630 m above the Princeton-Black coal seam and is part of the Princeton Group, Allenby Formation (Boneham 1968). The Princeton Group has been dated as middle Eocene based on studies of freshwater fishes (Wilson 1977, 1982) and mammals (Russell 1935; Gazin 1953) and by K-Ar dates (Hills and Baadsgaard 1967; H. Baadsgaard, personal communication). Chert blocks were cut into slabs and studied using a modified cellulose acetate peel technique and concentrated (48%) hydrofluoric acid (Basinger and Rothwell 1977; Basinger 1981). Peel sections were mounted in either Coverbond or Eukitt (Calibrated Instruments, Hawthorne, N.Y.), xylene-soluble mounting media for microscopic examination. Light photomicroscopy was conducted using a Zeiss Ultraphot IIIB, with image capture via a MicroLumina digital scanning camera

<sup>1</sup> Author for correspondence and reprints; e-mail rothwell@ohio.edu.

(Leaf Systems, Bedford, Mass.). Images were stored as tiff files, processed using Adobe Photoshop 4.0, and printed on a Yashika disublimation printer. Specimens for scanning electron microscopy were prepared following the methods of Rothwell and Stockey (1991). All chert slabs, peels, and slides of *Makopteris princetonensis* are housed in the University of Alberta Paleobotanical Collections (UAPC-ALTA).

### Systematics

#### Order—Filicales

Family—Athriaceae (sensu Pichi Sermolli 1977)

Genus—*Makopteris* gen. nov.

Species—*Makopteris princetonensis* sp. nov.

**Combined generic and specific diagnoses.** Rhizomes creeping, elongate, with internodes up to 1.5 cm; 1–3 mm in diameter; phyllotaxis helical. Amphiphloic dictyostele with two to four anastomosing cauline meristemes, producing paired hippocampiform stipe traces; one diarch adventitious root extending from base of each leaf gap. Protoxylem mesarch, metaxylem tracheids 20–55  $\mu\text{m}$  in diameter, with uni-biseriate scalariform wall thickenings. Ground tissues of tightly packed parenchyma with tanniferous cells; uniseriate endodermis of inconspicuous cells in rhizome and frond base, conspicuous endodermal cells containing dark contents at more distal levels. Sclerotic hypodermis immediately beneath uniseriate epidermis of closely spaced cells. Epidermal appendages unknown. Fronds monomorphic, pinnate, without swollen bases; primary pinnae terete at level of divergence; rachis pinnatifid or forming terminal pinnule toward apex. Stipe hemispherical at base, with blunt lateral wings; becoming adaxially concave and forming U-shaped groove distally; groove extending into pinnae. Pinnules with swollen veins and thin lamina, bearing adaxial sori below vein. Sporangia with vertical annulus of ca. 14 cells on spheroidal capsule 190–210  $\mu\text{m}$  in diameter, and elongate stalk approximately two cells wide diverging from swollen receptacle; indusium unknown. Spores ellipsoidal, monolet, 33–45  $\mu\text{m}$  long, with thin perispore bearing dense spines 2–3  $\mu\text{m}$  long.

**Holotype.** Rhizome with diverging stipes and roots in chert block P1411, figures 1.2, 2.1, 2.2; deposited in the University of Alberta Paleobotanical Collection.

**Paratypes.** Rhizomes with diverging stipes and roots in chert blocks P2345, figure 1.1; P1412, figure 1.3; P3585, figures 1.4, 1.5; P2712, figure 1.6; vegetative frond segments in chert blocks P2564, figure 2.3; P2314, figure 3.1; P2385, figure 4.1; P1179, figure 3.2; P2333, figures 3.3, 4.1; and fertile frond segments with sporangia and spores in chert block P1182, figures 4.2–4.6; deposited in the University of Alberta Paleobotanical Collection.

**Etymology.** The generic name *Makopteris* (*Makoto*, plus *pteris* = fern) is proposed in honor of significant advancements to our understanding of extinct vegetation by the late Professor Makoto Nishida, Chiba University. The specific epithet *princetonensis* refers to the collecting locality.

**Locality.** East bank of the Similkameen River, ca. 8.4 km south of Princeton, British Columbia (Princeton Map Sheet 92 H/7 1 : 50,000 U.T.M. Grid Ref. 783-724).

**Stratigraphic occurrence.** Princeton Group, Allenby Formation, 630 m above the Princeton-Black Coal seam in the Princeton Basin.

**Age.** Middle Eocene.

### Description of Material

Over 110 specimens of *Makopteris princetonensis* rhizomes and many more frond fragments are preserved in several layers of the Princeton chert but are most common in layers 5, 8, 14, 20, and 24 (sensu Stockey 1987). They are most often associated with stems, leaves, roots, ovulate cones, and pollen cones of *Pinus* but also occur with the aquatic dicot *Eorhiza* Robison and Person and with the filicalean fern *Dennstaedtiopsis aerenchymata* Arnold and Daugherty in some layers. Many specimens of *M. princetonensis* are represented by elongated horizontal rhizomes that bear stipes (fig. 1.1) and branch occasionally. Some specimens consist of sections of the stipe/rachis that grade from the base to near the tip (figs. 1.1, 2.3, 3.1–3.6). Others consist of pinnule fragments (fig. 4.1–4.3), a few of which have sori of annulate sporangia (fig. 4.2–4.5) that contain monolet spores (fig. 4.4–4.5).

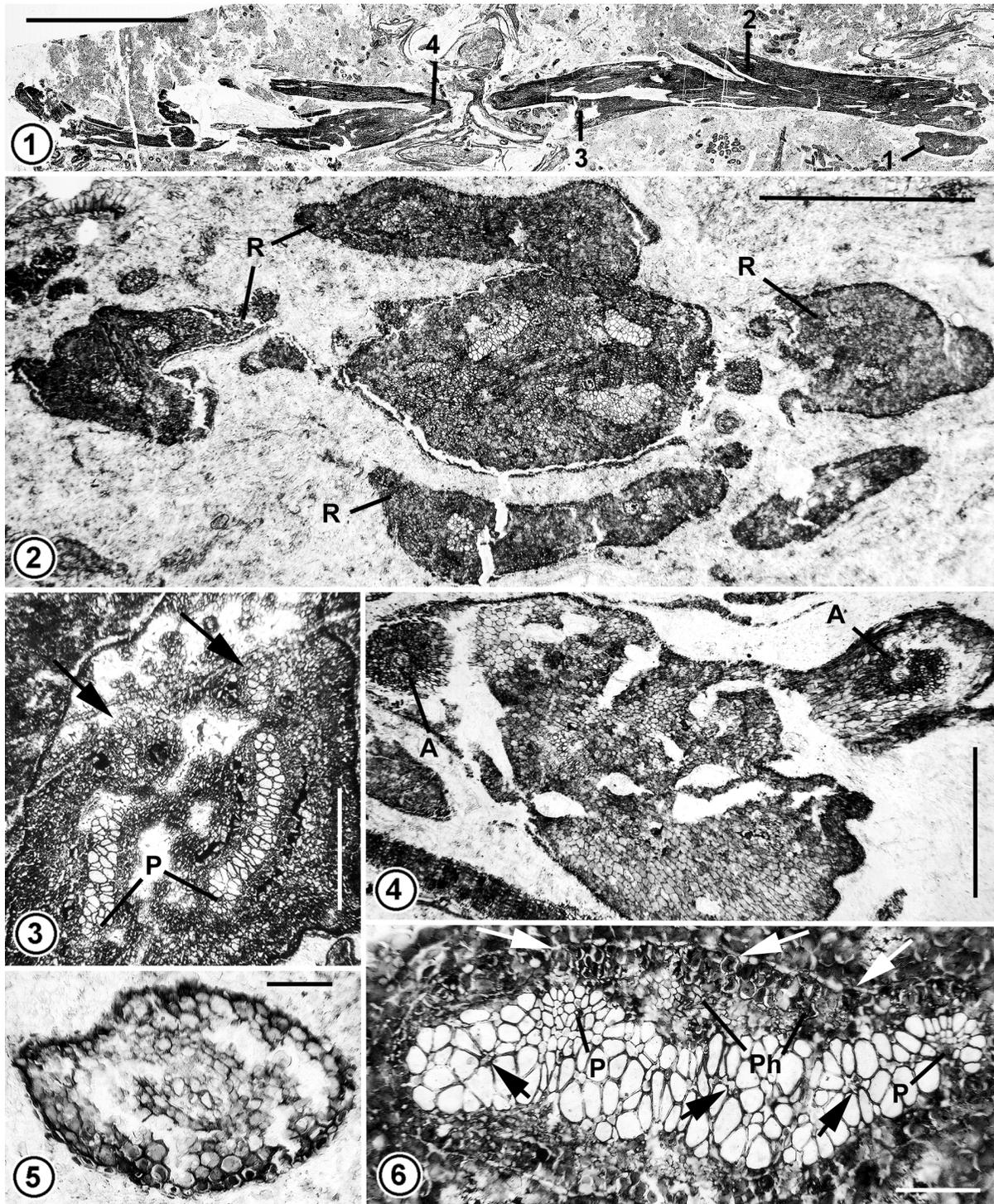
#### Rhizome

Rhizomes are radial and range from 0.8 × 0.6 mm to 3.2 × 1.7 mm in diameter. They produce stipe traces all around the circumference (fig. 1.2), but the fronds often appear to extend away from the rhizome primarily to one side, as if the rhizomes were oriented horizontally in life. Specimens show from one to four frond bases in a single cross section, indicating that there is a range of variation in internodal length. Stipes diverge at intervals of up to 1.5 cm (fig. 1.1). Root traces diverge from the stele, extend radially, and separate from the rhizome (fig. 1.4).

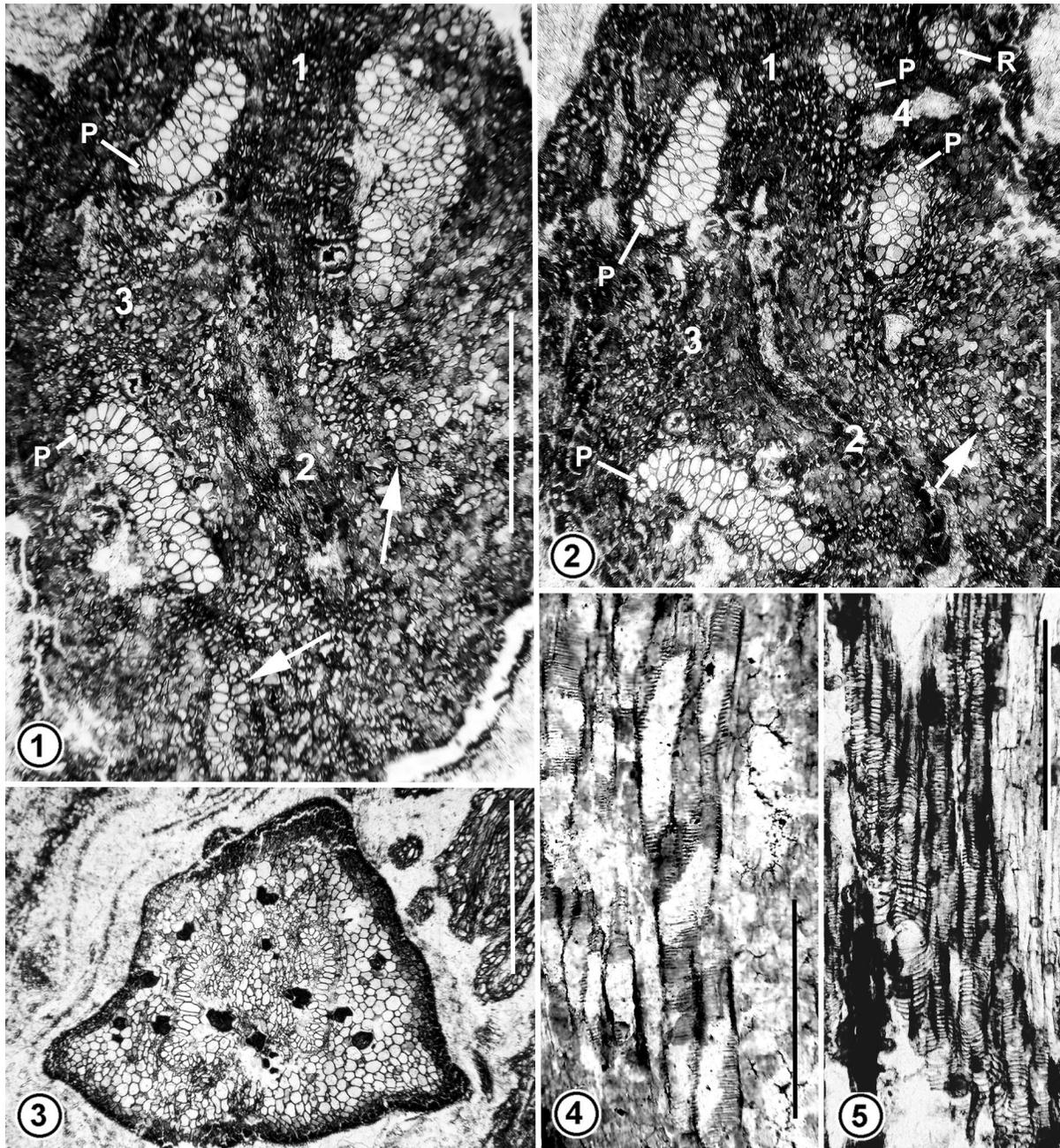
In cross sections, the rhizomes show a broad parenchymatous pith, cauline bundles, stipe traces, root traces, cortex, and epidermis (figs. 1.2–1.4, 2.1, 2.2). The stele consists of a ring of amphiphloic mesarch (fig. 1.6) meristemes that are usually tangentially elongated and of unequal size (figs. 1.2–1.4, 2.1, 2.2), and frond traces are represented by two hippocampiform bundles (fig. 2.1, arrows). Some sections also display one or two diverging root traces (fig. 1.4). Roots are diarch, with poorly preserved phloem, parenchymatous cortex, and epidermis (fig. 1.5).

Metaxylem tracheids of the cauline bundles are in bands two to six cells wide (fig. 1.6) and range 20–55  $\mu\text{m}$  in diameter. Most bundles have a mesarch protoxylem strand near one end (figs. 1.3, 1.6, 2.1–2.2), but in some bundles there is a more centrally located protoxylem (fig. 1.6), and in others protoxylem cannot be identified. A few xylem parenchyma cells are also evident in cross sections (fig. 1.6, black arrows), but in all but the most well-preserved specimens, xylem parenchyma appears as dark areas between adjacent metaxylem tracheids. The best-preserved bundles display a zone of thin-walled phloem cells and a uniseriate endodermis surrounding the tracheids (fig. 1.6, white arrows).

Pith and cortex are constructed of tightly packed, thin-walled parenchyma cells 26–65  $\mu\text{m}$  in diameter with interspersed dark areas (fig. 1.2–1.4). Some of these areas are iso-



**Fig. 1** *Makotopteris princetonensis* gen. et sp. nov. Fig. 1.1, Longitudinal section of rhizome showing lengths of internodes between divergence of fronds (numbers 1–4). The most distal division of the rhizome (at left) may be either a rachis or branch. P2345 A #0,  $\times 2.8$ ; bar = 1 cm. Fig. 1.2, Cross section of dictyostelic rhizome surrounded by detached stipes with both paired hippocampiform bundles and roots. Frond arrangement appears opposite, but vascular bundles indicate transition to helical phyllotaxis. R = rachis. Holotype P1411 H<sub>2</sub> top #4a  $\times 3.1$ ; bar = 1 mm. Fig. 1.3, Cross section of rhizome with apparently distichous phyllotaxis and two cauline meristemes. Note paired hippocampiform traces to diverging stipe (top, at arrows) and protoxylem strands at bottom of each meristeme. P = protoxylem. P1412 E<sub>1</sub> top #0  $\times 32$ ; bar = 0.5 mm. Fig. 1.4, Cross section of rhizome showing the divergence of two adventitious roots. A = adventitious root. P3585 E top #0  $\times 35$ ; bar = 1 mm. Fig. 1.5, Cross section of adventitious root showing diarch protostele, endodermis, parenchymatous cortex, and epidermis. P3585 E top #0  $\times 84$ ; bar = 0.1 mm. Fig. 1.6, Cross section of cauline meristeme below level where frond trace will diverge (from right end) and where divergence of a root trace (protoxylem strand to the left of center) will open new gap. Note xylem parenchyma (at black arrows), phloem tissue, and endodermis (at white arrows). P = protoxylem, Ph = phloem. P2712 F bot #9  $\times 150$ ; bar = 0.5 mm.



**Fig. 2** *Makotopteris princetonsensis* gen. et sp. nov. Fig. 2.1, Cross section of rhizome showing both three cauline meristeles surrounding pith and cortex of specimen with conspicuous amber cell contents. Specimen shows three leaf gaps (numbers) at this level, and inconspicuous paired traces to frond (arrows). Gaps are numbered in the order in which they were produced. Note that there are no protoxylem strands adjacent to gaps above level where frond traces have diverged (1 and 2) and protoxylem strands characterize positions from which frond trace xylem will diverge distally (3, at left). P = protoxylem. Holotype P1411 H<sub>2</sub> top #4b × 60; bar = 0.5 mm. Fig. 2.2, Cross section of rhizome in fig. 2.1 at more distal level. Note that cauline bundle at upper right has divided to produce three bundles, a central adventitious root trace, and two cauline bundles that flank newly formed gap (4). R = root trace. Other labeling as in fig. 2.1. Holotype P1411 H<sub>2</sub> top #23b × 60; bar = 0.5 mm. Fig. 2.3, Cross section of slightly distorted stipe near level of divergence from rhizome. Note tightly packed parenchyma with tanniferous cells and cell clusters, conspicuous hypodermis, and inconspicuous endodermis. Adaxial surface (at bottom) with lateral ribs. P2564 H top #0 × 70; bar = 0.5 mm. Fig. 2.4, Longitudinal section of cauline bundle showing pitting of metaxylem tracheids. P2345 C top #0 × 145; bar = 0.2 mm. Fig. 2.5, Longitudinal section of stipe trace showing wall-thickening patterns of metaxylem tracheids. P2333 G top #0 × 160; bar = 0.2 mm.

lated cells (fig. 2.3), while others are small groups of cells (fig. 2.3) that are reminiscent of the tanniferous cells that characterize the ground tissues of *Woodwardia radicans* (L.) J. E. Smith (Lucansky 1981). Ground tissues typically display a distinct amber color that obscures features of the cell walls in some specimens (figs. 1.1–1.3). In other specimens, the pith and cortical cells are clearly thin walled, polygonal, and tightly packed (figs. 2.1, 2.2). At the periphery of the cortex there are up to three layers of smaller, thicker-walled cells that form a

sclerotic hypodermis in some specimens. Hypodermis is more extensively developed in the stipe, where the cell walls are not obscured by amber cellular contents. Epidermis is often incompletely preserved or absent, but where present it consists of isodiametric cells that measure 14–22  $\mu\text{m}$  in diameter (figs. 1.2–1.4). No scales have been identified, but this may be because of incomplete preservation of the epidermal zone or the loss of such structures as the organs matured.

#### *Phyllotaxis and Vascular Architecture*

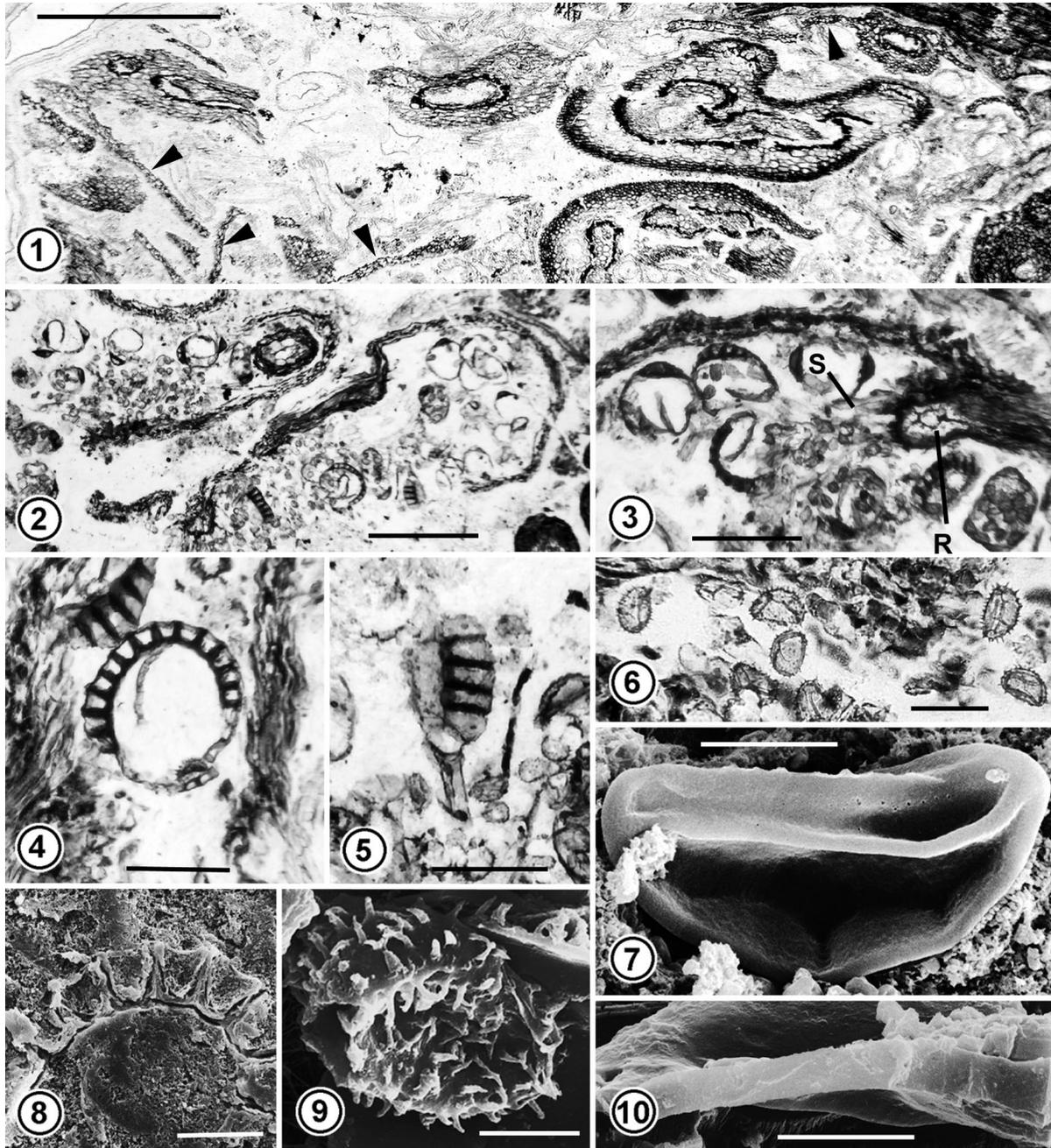
There is some variation in leaf arrangement among the rhizomes. Most rhizomes display helical phyllotaxis with three or more orthostichies (fig. 1.2), but a few relatively small specimens have an apparently distichous frond arrangement (fig. 1.3). Specimens with distichous fronds have two tangentially elongated cauline meristemes (fig. 1.3). At each node in such specimens, a vascular bundle diverges from one end of each cauline meristeme to produce the two-stranded stipe trace (fig. 1.3, top).

All of the other rhizomes have from three to five bundles, as well as one or more paired frond traces, in each cross section (fig. 2.1, 2.2). At about the level of frond trace divergence in a specimen that shows three cauline bundles (fig. 2.1), one of the bundles that lies adjacent to the gap of the diverging frond trace (fig. 2.1, upper right) divides into three (fig. 2.2). The dividing bundle first becomes broadly V-shaped, with the base of the V pointed outward (fig. 2.1) and then divides (fig. 2.2). Distal to this division, the central strand extends toward the periphery of the rhizome as a diverging root trace (fig. 1.4). The two remaining meristemes are stem bundles that flank a newly opened gap. This architecture is similar to many “derived” living ferns, where leaf gaps are typically produced by the divergence of one or more root traces (Ogura 1927, 1972; Lucansky 1981; White and Weidlich 1995; Serbet 1997) and then enlarged by the divergence of two or more frond traces. At this level the rhizome of *M. princetonsensis* displays five bundles (i.e., four cauline meristemes and a root trace; fig. 2.2) plus frond traces.

Progressing distally, adjacent ends of the newly divided cauline bundles elongate (fig. 1.3, bottom; fig. 2.2, left), and then paired bundles separate to produce frond traces that are hippocampiform by the level of stipe divergence (fig. 2.3). At this



**Fig. 3** *Makotopteris princetonsensis* gen. et sp. nov. Cross sections of frond segments. Fig. 3.1, Stipe near base, showing straight adaxial surface, blunt lateral ribs, and histology intermediate between rhizome and distal frond members. P2314 C<sub>2</sub> bot #0  $\times 40$ ; bar = 1 mm. Fig. 3.2, Stipe showing slightly concave adaxial surface and histology of ground tissues slightly distal to level of fig. 3.1. P1179 D top #1  $\times 40$ ; bar = 1 mm. Fig. 3.3, Several frond members showing U-shaped groove in abaxial surface. Proximal section of rachis at bottom, more distal section showing divergence of opposite pinnule traces at left, and base of pinnule at upper right (arrow). P2333 J bot #0  $\times 25$ ; bar = 1 mm. Fig. 3.4, Stipe or rachis showing trace at level where paired strands have united to form U-shaped bundle. P2569 C top #0  $\times 50$ ; bar = 0.2 mm. Fig. 3.5, Rachis in distal region, showing deep abaxial groove, and single bundle. P2333 K top #0  $\times 33$ ; bar = 0.5 mm. Fig. 3.6, Most distal sections showing diverging pinnule trace (at left) and lamina extending from midrib (at center). P2333 G top #0  $\times 65$ ; bar = 0.5 mm.



**Fig. 4** *Makotopteris princetonensis* gen. et sp. nov. Fig. 4.1, Oblique cross sections of several frond members at various levels. Note laminar tissue (arrowheads) attached to tip of rachis (upper right), and fragments of lamina, lower left. P2385 B bot #0  $\times 26$ ; bar = 1 mm. Fig. 4.2, Cross section of fertile pinnules showing swollen lateral vein and several sporangia in section views. P1182 D top #3  $\times 44$ ; bar = 0.5 mm. Fig. 4.3, Pinnule with swollen receptacle that displays abaxially attached sporangial stalks and scattered sporangia. R = receptacle, S = sporangial stalk. P1182 D bot #0  $\times 43$ ; bar = 0.5 mm. Fig. 4.4, Longitudinal section of sporangial capsule in plane of annulus. P1182 C<sub>2</sub> bot #0  $\times 155$ ; bar = 100  $\mu\text{m}$ . Fig. 4.5, Distal region of sporangium showing orientation of annulus with respect to sporangial stalk. P1182 D top #3  $\times 175$ ; bar = 100  $\mu\text{m}$ . Fig. 4.6, Ellipsoidal monoete spores with spiny perispore. P1182 D top #0  $\times 240$ ; bar = 50  $\mu\text{m}$ . Fig. 4.7, SEM of longitudinally folded monoete spore with smooth exine. P1080 D top  $\times 2000$ ; bar = 10  $\mu\text{m}$ . Fig. 4.8, SEM of annulus. Note thickened inner and lateral walls of cells. P1080 D top  $\times 330$ ; bar = 50  $\mu\text{m}$ . Fig. 4.9, SEM of fragment of spiny perispore. P1080 D top  $\times 2900$ ; bar = 5  $\mu\text{m}$ . Fig. 4.10, SEM showing fractured surface of spore wall. Note solid, homogeneous appearance. P1080 D top  $\times 4000$ ; bar = 5  $\mu\text{m}$ .

level there are four cauline bundles in the rhizome, but, somewhat more distally, two cauline bundles fuse to return the number to three. When mapped over a length of rhizome, this pattern of cauline bundle divisions, root trace production and divergence, frond trace production and divergence, and cauline bundle fusion can be recognized as forming a repeating pattern in which there are anastomosing cauline bundles. If every third stipe were to diverge from the same orthostichy, phyllotaxis of these specimens would conform to a one-third helix, but this is usually not the case. Even from the short lengths of distorted rhizomes that are available for study, traces to the fourth stipe in the genetic helix appear to be consistently offset from traces to the first (fig. 2.2). This indicates that phyllotaxis approaches a higher order fraction, probably two-fifths or three-eighths (Church 1968; Beck et al. 1982).

### *Frond*

There are many small frond segments in the same chert layers that yield the rhizomes (figs. 2.3, 3.1–3.6, 4.1–4.3). Some are attached to the rhizomes, and others have distinctive features that help identify them as all belonging to the same species. Sections of foliar segments intergrade in size and structure of the frond trace and display a wide range of characters that allow for reconstruction of the general frond architecture. The largest specimens have two bundles (figs. 2.3, 3.1), but in smaller specimens the bundles are united into a single U-shaped strand (fig. 3.4), as is characteristic of the rachides in living athyroid ferns (Ogura 1972). In the smallest specimens the strand is broadly V-shaped (figs. 3.5, 4.1). Some specimens of the rachis show the divergence of opposite or subopposite primary pinna traces (fig. 3.3). These produce primary pinnae that are terete at the level of divergence (fig. 3.3, upper right), but distally they expand laterally to form laminar pinnules with prominent veins (fig. 4.2). Other distal sections of the rachis display laminar tissue at the sides of the rachis (figs. 3.6, 4.1, 4.2) and represent either a pinnatifid region or a terminal pinnule.

Specimens of the stipe/rachis range in size from 3.0 to 0.8 mm wide and 2.0 to 0.5 mm thick. In cross sections near the level of divergence from the rhizome, the stipe is roughly hemispherical, with a convex adaxial surface, a more or less straight or slightly concave adaxial surface, and two small, blunt lateral flanges (fig. 3.1). Cross sections of stipes that are somewhat smaller typically display a concave adaxial surface (fig. 3.2), forming a U-shaped channel that in successively smaller specimens becomes progressively deeper and narrower (figs. 3.3–3.6, 4.1).

Stipes that are attached to or closely associated with rhizomes have histological features like those of the rhizomes (figs. 1.2, 2.3). Specimens of this type intergrade to considerably smaller stipes/rachides that have distinctly different histological features (figs. 3.1–3.6) and a single vascular bundle (fig. 3.4). Some rachides with a single bundle show the divergence of opposite or subopposite terete primary pinnae (fig. 3.3, left). There is a small number of specimens in the same size range as the diverging primary pinna bases that are elliptical in cross section with an oval trace (fig. 3.3, arrow). The latter specimens are interpreted as basal sections of pinnules. Still other frond fragments consist of laminar pinnules,

with thicker areas that represent the positions of veins (figs. 4.1, 4.2). Pinnules appear to be three to four cells thick between veins and display small amber globules (figs. 4.1–4.3). Otherwise, cell preservation is inadequate to determine histological features of the mesophyll and epidermis.

As indicated above, the most basal segments of stipe have histological features like those of the rhizomes (fig. 2.3). Other frond fragments have parenchyma cells with much lighter-colored cell lumens (fig. 3.2). In the latter specimens, endodermal cells are extremely prominent because they either have amber or black contents that line the inner and lateral walls or are filled with dark contents. Hypodermal sclerenchyma shows up much more clearly as a dark outer zone in these specimens (figs. 3.5, 4.1), and isolated resinous cells may be absent from the ground tissue. In preliminary studies of the Princeton chert, specimens with these divergent types of ground tissue histology were suspected to represent different entities (Basinger 1981). However, other characters intergrade completely, and there are a few specimens with intermediate histology (fig. 3.1) that demonstrate that all of these frond sections represent the same species.

### *Sori, Sporangia, and Spores*

A few pinnule sections show thickened areas on the abaxial surface that represent the receptacle of a sorus (fig. 4.3). Each receptacle protrudes from the pinnule surface beneath a vein and shows several diverging sporangial stalks (fig. 4.3). We have been unable to identify an indusium with confidence. Whether this is because a delicate indusium has not been preserved or cannot be recognized among the debris that surrounds the sori or because the species is exindusiate cannot be determined.

Sporangial capsules range 190–210  $\mu\text{m}$  in diameter, and the stalk is ca. 18–20  $\mu\text{m}$  wide. There are about 14 cells in the annulus (fig. 4.4). As is typical of filicalean ferns in general, cells of the annulus are thickened on the inner and lateral walls (figs. 4.4, 4.5, 4.8). Sporangia are usually broken or senescent, but many contain spores, and additional spores with identical features are concentrated in the matrix around the sporangia (figs. 4.2–4.6). The spores are bilateral and monolete, distinctly spiny (fig. 4.6), and measure 33–45  $\mu\text{m}$  long (mean = 38.3  $\mu\text{m}$ ). When macerated for scanning electron microscopy, the spiny outer layer is removed as a thin perispore with spines that are 2–3  $\mu\text{m}$  long (fig. 4.9). The exine is psilate (figs. 4.7, 4.10), ca. 1.7  $\mu\text{m}$  thick, and broken spores show a uniformly dense, solid wall under scanning electron microscopy (fig. 4.10).

### **Discussion**

A combination of several characters that are distinctive among filicalean ferns allows us to recognize *Makotopteris princetonensis* as a member of the Athyriaceae *sensu* Pichi Sermolli (1977). Included are abaxial sori of leptosporangia with a vertical annulus and long narrow stalk, dictyostelic rhizome, diarch roots with a parenchymatous cortex, and monolete spores with a psilate exine and ornamented perispore (Kato 1977; Tryon and Tryon 1982; Kramer et al. 1990; Tryon and Lugardon 1990; Schneider 1996). Although stelar architecture has been characterized in only a very small percentage

of living aspidioid ferns (*sensu* Ogura 1972), including the athyrioids (*sensu* Pichi Sermolli 1977), species are relatively uniform in some characters. Particularly distinctive among these is the formation of gaps that usually are not the result of leaf-trace divergence, as they are in most solenostelic ferns and the dictyostelic tree ferns (Ogura 1972). Rather, gaps in the dictyosteles of these ferns typically open as the result of root-trace divergence, and leaf traces diverge tangentially from meristemes in a fashion that is reminiscent of the stelar architecture in seed plants with closed sympodial systems (Ogura 1927, 1972; White and Weidlich 1995; Serbet 1997).

Among athyrioid species placed in the family Dryopteridaceae, tribe Physematieae, subfamily Athyrioideae by Kramer (1990), *Athyrium* Roth, *Diplazium* Swartz, and several segregate genera (Kato 1977; Tryon and Tryon 1982; Kramer et al. 1990; Smith 1993) are characterized by species that have monomorphic fronds and two stipe bundles that unite distally (Ogura 1972). However, a combination of two factors make more precise relationships of *Makotopteris* difficult to determine. On the one hand, several of the most generically diagnostic morphological features of athyrioid ferns are not available from the anatomical sections of *Makotopteris*. These include exact pinnule shape and venation pattern, shape of the sorus, nature of the indusium (if present), and features of epidermal appendages (i.e., scales or trichomes, if present). On the other hand, the distinctive anatomical features of the fossils cannot be used with confidence to assess precise systematic relationships because comparable features of living ferns have been studied in only a small minority of living species. Moreover, several of these characters are either highly variable and overlapping among genera or are correlated with plant size. Species with creeping rhizomes, radial dictyosteles, and fronds that are once pinnate like those of *Makotopteris* occur in *Athyrium*, *Diplazium*, *Dictyodroma* Ching, *Deparia* Hooker and Greville, and *Gymnocarpium* Newman (Kramer et al. 1990).

One character that may suggest a close relationship of *M. princetonensis* to the genus *Diplazium* is the morphology of the adaxial groove on the rachis and pinnules. In a survey of several athyrioid species, Kato (1977) documented that the adaxial groove is V-shaped in some species of *Athyrium* and *Cornopteris* Nakai, U-shaped with a flat bottom in some species of *Diplazium*, and U-shaped with a rounded bottom in other species of *Diplazium* and *Deparia*. *Makotopteris* is clearly most similar to the latter group of species. The groove

is continuous from the rachis to the lateral frond members in species of *Athyrium*, *Cornopteris*, and *Diplazium* but discontinuous from the rachis to the primary pinnae in the species of *Deparia* studied (Kato 1977). The continuous U-shaped groove with a rounded bottom in *Makotopteris* is, therefore, most reminiscent of species such as *Diplazium wichurae* and *Diplazium pin-faense* (Kato 1977).

Bean-shaped spores with a psilate exine and ornamented perispore are common among athyrioid ferns (Tryon and Lugardon 1990). Among the living taxa that show morphological and anatomical similarities to *Makotopteris*, *Deparia fenziiana* (Luers.) M. Kato, *Gymnocarpium oyamense* (Bak.) Ching, and *Diplazium cognatum* (Hieron.) Sledge have spores with spiny perispore sculpturing (Tryon and Lugardon 1990). The perispore spines of *D. cognatum* are long and narrow and appear to be most similar to those of *Makotopteris*.

*Makotopteris princetonensis* is the most completely reconstructed extinct species of athyrioid ferns described thus far. It has small, procumbent rhizomes with an amphiphloic dictyostele, pinnate fronds with an adaxial groove and paired traces that unite distally, diarch roots with parenchymatous cortex, adaxial sori of sporangia with a vertical annulus and elongated stalk, and monolete bean-shaped spores with an ornamented perispore. These document that essentially modern species of the Athyriaceae were present in the flora of North America by at least middle Eocene time. The most common association of *Makotopteris* with plant parts of the conifer *Pinus*, and the absence of aerenchyma from the ground tissues of *Makotopteris*, further suggest that this species grew on the forest floor adjacent to the aquatic environments from which most of the Princeton chert plants are derived. In this regard, *M. princetonensis* not only provides an early benchmark for assessing the mode and tempo of athyrioid evolution, but also contributes valuable data for beginning to characterize the diversity of habitats that are represented in the remarkable biota of the Princeton chert.

### Acknowledgments

This work was supported in part by grants from the Natural Sciences and Engineering Research Council of Canada (grant A-6908 to R. A. Stockey) and the National Science Foundation (grant DEB-9527920 to G. W. Rothwell).

### Literature Cited

- Basinger JF 1976 Permineralized plants from the Eocene, Allenby Formation of southern British Columbia. MS thesis. University of Alberta, Edmonton.
- 1981 The vegetative body of *Metasequoia milleri* from the Middle Eocene of southern British Columbia. *Can J Bot* 59: 2379–2410.
- Basinger JF, GW Rothwell 1977 Anatomically preserved plants from the Middle Eocene (Allenby Formation) of British Columbia. *Can J Bot* 55:1984–1990.
- Beck CB, R Schmid, GW Rothwell 1982 Stelar morphology and the primary vascular system of seed plants. *Bot Rev* 48:691–815.
- Boneham RFG 1968 Palynology of three Tertiary coal basins in south central British Columbia. PhD diss. University of Michigan, Ann Arbor.
- Cevallos-Ferriz S, RA Stockey, KB Pigg 1991 The Princeton chert: evidence for in situ aquatic plants. *Rev Palaeobot Palynol* 70: 173–185.
- Church AH 1968 On the interpretation of the phenomena of phylotaxis. Hafner, New York.
- Gazin CL 1953 The Tillodontia: an early Tertiary order of mammals. *Smithson Misc Collect* 1221:1–110.
- Hills LV, H Baadsgaard 1967 Potassium-argon dating of some lower Tertiary strata in British Columbia. *Can Petr Geol* 15:138–149.
- Kato M 1977 Classification of *Athyrium* and allied genera of Japan. *Bot Mag Tokyo* 90:23–40.

- Kramer KU 1990 Subfamily Athyrioideae. Pages 130–142 in KU Kramer, PS Green, eds. The families and genera of vascular plants. Vol I. Pteridophytes and gymnosperms. Springer, Berlin.
- Kramer KU, RE Holttum, RC Moran, AR Smith 1990 Dryopteridaceae. Pages 101–144 in KU Kramer, PS Green, eds. The families and genera of vascular plants. Vol I. Pteridophytes and gymnosperms. Springer, Berlin.
- Lucansky TW 1981 Chain ferns of Florida. *Am Fern J* 71:101–108.
- Ogura Y 1927 On the structure of *Diplazium esculentum*, (Retz.) Sw. *Bot Mag Tokyo* 41:172–180.
- 1972 Comparative anatomy of vegetative organs of the pteridophytes. 2d ed. Borntraeger, Berlin.
- Pichi Sermolli REG 1977 Tentamen Pteridophytorum genera in taxonomicum ordinem redigendi. *Webbia* 31:481–500.
- Pigg KB, RA Stockey 1996 The significance of the Princeton chert permineralized flora to the middle Eocene upland biota of the Okanogan Highlands. *Wash Geol* 24:32–36.
- Rothwell GW, RA Stockey 1991 *Onoclea sensibilis* in the Paleocene of North America, a dramatic example of structural and ecological stasis. *Rev Palaeobot Palynol* 70:113–124.
- Rothwell GW, RA Stockey, H Nishida 1994 Filicaleans of the middle Eocene Princeton chert. I. A dryopteroid species. *Am J Bot* 81:101–102.
- Russell LS 1935 A Middle Eocene mammal from British Columbia. *Am J Sci* 229:54–55.
- Schneider H 1996 The root anatomy of ferns: a comparative study. Pages 271–283 in JM Camus, M Gibby, RJ Johns, eds. Pteridology in perspective. Royal Botanic Gardens, Kew.
- Serbet R 1997 Morphologically and anatomically preserved fossil plants from Alberta Canada: a flora that supported the dinosaur fauna during the Upper Cretaceous (Maastrichtian). PhD diss. Ohio University, Athens.
- Smith AR 1993 Dryopteridaceae Herter. Pages 246–249 in Flora of North America north of Mexico. Vol 2. Pteridophytes and gymnosperms. Oxford University Press, New York.
- Stockey RA 1987 A permineralized flower from the middle Eocene of British Columbia. *Am J Bot* 74:1878–1887.
- Tryon AF, B Lugardon 1990 Spores of the Pteridophyta. Springer, New York.
- Tryon RM, AF Tryon 1982 Ferns and allied plants with special reference to tropical America. Springer, New York.
- White RA, WH Weidlich 1995 Organization of the vascular system in the stems of *Diplazium* and *Blechnum* (Filicales). *Am J Bot* 82:982–991.
- Wilson MVH 1977 Middle Eocene freshwater fishes from British Columbia. *R Ont Mus Life Sci Contrib* 113:1–61.
- 1982 A new species of the fish *Amia* from the Middle Eocene of British Columbia. *Palaeontology* 25:413–424.