Biosystematic Studies of the Beech and Marsh Ferns¹

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Abstract

The taxonomic status of five species of ferns common to the Northeastern United States has been in a state of confusion for many years. The broad beech fern, the long beech fern, the marsh fern, the New York fern, and the bog fern have been placed in the genus *Thelypteris* by some authors. Others consider them members of the large cosmopolitan genus *Dryopteris*. In other interpretations the beech ferns are separated into the genus *Phegopteris*.

Evidences from morphological, cytological, and biochemical studies do not warrant the inclusion of these species in the genus *Dryopteris*. Significant differences were noted between the beech ferns and the marsh ferns which would appear to justify their separation into different genera.

This study was undertaken in an attempt to clarify the confusion regarding the taxonomic status of five species of ferns common to the northeastern United States. These ferns, and their counterparts from other regions, have been variously classified generically.

Some have considered the broad beech fern, the long beech fern, the bog fern, the marsh fern, and the New York fern as being of common generic rank. These ferns were treated as members of the large cosmopolitan genus *Dryopteris* by Christensen in his monograph (4, 5). This interpretation was followed by Fernald in his *Gray's Manual of Botany* (9) and by Deam in his *Flora of Indiana* (8) (Table 1).

Christensen, in a later work, set the genus *Thelypteris*, with the marsh fern, *Thelypteris palustris* as its type, apart from the genus *Dryopteris* (6). Morton (12) in his contributions to *Britton and Brown's Illustrated Flora* (10) recognized the differences between the "thelypteroid" and the "dryopteroid" ferns (Table 1).

On the basis that *Thelypteris* Schmidel was not validly published, Copeland (7) in his *Genera Filicum* used the name *Lastrea* Bory. The Nomenclature Committee at the International Botanical Congress in Stockholm decided, however, that the name *Thelypteris* is correct and voted down a proposal to conserve *Lastrea* (13, 14).

Cytologically the beech ferns differ from both the dryopteroids and the thelypteroids; further, their sori lack indusia in contrast with the latter. Therefore, some feel that these ferns should be placed in a separate genus, *Phegopteris* (11, 21) (Table 1).

Methods and Materials

Extensive collections of the five species were made throughout Indiana and Ohio. A total of 422 herbarium specimens from the Field Museum of Natural History and 432 specimens from the Missouri Botanical Garden was examined.

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			Gleason's Britton
	Fernals's Gray's	Wherry's Guide to	and Brown's New
	Manual of Botany	Eastern Ferns	Illustrated Flora
Broad Beech Fern	Dryopteris hexagonoptera	Phegopteris hexagonoptera	Thelypteris hexagonoptera
	(Michx.) Christens.	(Michx.) Fee	(Michx.) Weatherby
Long Beech Fern	Dryopteris Phegopteris	Phegopteris polypodioides	Thelypteris phegopteris
	(L.) Christens.	Fee	(L.) Slosson
Marsh Fern	Dryopteris Thelypteris	Thelypteris palustris	Thelypteris palustris
	(L.) Gray	Schott	Schott
Bog Fern	Dryopteris simulata Davenp.	Thelypteris simulata (Davenp.) Nieuwl.	Thelypteris simulata (Davenp.) Nieuwl.
New York Fern	Dryopteris noveboracensis	Thelypteris noveboracensis	Thelypteris noveboracensis
	(L.) Gray	(L.) Nieuwl.	(L.) Nieuwl.

PLANT TAXONOMY

The external morphology was carefully studied. Drawings and photographs were made to record distinguishing characteristics.

Rhizomes and stipes were cross sectioned at 10μ and stained with safranin-fast green.

Spore samples were obtained from both living and moribund specimens. The spores were mounted in Hoyer's medium for clearing and preservation.

For cytological study, meiotic materials were fixed in 3 parts ethyl alcohol : 1 part glacial acetic acid. Squashes were made in aceto-orcein.

Biochemical comparisons were made by means of paper chromatography. Extracts were prepared by powdering dried fronds and soaking the materials in 80% methanol for 48 hours. Fifty microliters of each sample were applied to Whatman #1 paper using the spot method. Both 1 and 2-dimensional chromatograms were run descendingly using butanol:acetic acid:water (12:3:5) as the solvent.

Dried chromatograms were examined in the presence of ultraviolet light. The chromatograms were sprayed sequentially with ninhydrin and Ehrlich reagent for the detection of free-amino acids and related substances. Some chromatograms were sprayed with Pauly's reagent or alkaline silver nitrate for the detection of phenolic compounds (16).

Morphological Observations

The frond of the beech ferns consists of a deltoid blade borne by a pilose stipe. The lanceolate apex of the blade is pinnatifid.

The blade of the long beech fern has 10-12 pairs of separate pinnatifid pinnae below the apex. The length of the blade exceeds its breadth at the widest point. Nine to 18 pairs of surcurrent pinnae are noted in the broad beech fern. The lower 3-4 pairs are bipinnatifid and the upper ones are pinnatifid (Fig. 1, 2).

Both species bear rounded, supramedial sori which lack indusia.

The blades of the New York, bog, and marsh ferns are lanceolate in outline and are composed of pinnatifid segments below a pinnatifid apex. The dark stipe is glabrous in the marsh fern and scaly in the other two species.

The blade of the New York fern is composed of 18-40 pairs of oblong-lanceolate pinnae. The lower 4-5 pairs are greatly reduced in size (Fig. 3). The sori are located submarginally and have a kidney-shaped indusium.

The marsh fern possesses 13-25 pairs of linear-lanceolate pinnae which bear medial indusiate sori (Fig. 4).

The blade of the bog fern is composed of 10-38 pairs of oblonglanceolate pinnae. The lowermost are basioscopic (Fig. 5). The indusiate sori are borne submarginally.



FIGURES 1-5. Photographs of herbarium specimens. 1) Phegopteris hexagonoptera (Michx.) Fee, broad beech fern; 2) Phegopteris polypodioides Fee, long beech fern; 3) Thelypteris noveboracensis (L.) Nieuwl., New York fern; 4) Thelypteris palustris Schott, marsh fern; 5) Thelypteris simulata (Davenp.) Nieuwl., bog fern. FIGURES 6-10. Fern spores X 675. 6) Broad beech fern; 7) Long beech fern; 8) Bog fern; 9) Marsh fern, 10) New York fern.

Spore Morphology

The spores of all five species are bilateral in shape and bear an adherent perispore. Spores of the beech ferns are light greenish-brown in color. Their perispore does not form conspicuous ridges or wings. The spores of the broad beech fern are $20-27 \times 30-50\mu$. The surface is tuberculate in texture. The perispore is not winged (Fig. 6). Spores of the long beech fern are $28-30 \times 55-60\mu$. The spore surface is tuberculate. The perispore varies, some spores being wingless and ridgeless and others bearing short broken ridges and narrow discontinuous wings (Fig. 7).

The remaining species are yellow-brown in color and possess a tuberculate winged and ridged perispore. The spores of the marsh fern are $34-45 \ge 53-63\mu$. Ridges are few and broken (Fig. 9). The New York fern spores are highly sculptured with wide apical wings. The spores are $25-30 \ge 33-45\mu$ (Fig. 10). The spores of the bog fern are sculptured similarly to those of the New York fern, but approach those of the marsh fern in size (Fig. 8).

Cytological Observations

It is apparent from this and other studies that different chromosome numbers exist among the thelypteroid ferns.

Broad beech fern materials collected in Indiana showed a chromosome number of n = 30 (Voucher Specimen #0159 Oliver). Counts made of Virginia specimens by Wagner (18) and by Britton (1, 2, 3) of materials from Ontario confirm this number.

Studies of the long beech fern have been made by Manton (11) in Great Britain and by Britton (1, 2, 3). Both have reported numbers of n = 90 and 2n = 90. The long beech fern is apogamous. A doubling of the diploid number of 90 chromosomes occurs just prior to meiosis; thus the sporophytic and the gametophytic numbers are the same.

Counts of the marsh fern (Voucher #0172 Oliver) and the New York fern (Voucher #0195 Oliver) yielded numbers of n = 35 and n = 27, respectively. These numbers are in agreement with those reported by Wagner (17, 18). Britton reported a number of n = 35 for the marsh fern; however, his tentative studies of the New York fern indicated n = 29 (1).

Meiotic material of the bog fern was unavailable for this study. Wagner reported a number of n = 64 for specimens collected in Maryland (17).

Biochemical Observations

Chromatograms were prepared from dried specimens of ferns collected throughout the northeastern United States. Materials had been preserved for periods of 1 month to 25 years. Remarkable consistency was obtained from materials of different ages and localities. Examination of chromatograms in the presence of ultraviolet light revealed one compound with a rf value of 0.67 common to all species. Another substance, rf 0.80, was common to the marsh, the bog, and the New York fern. Several additional spots appeared to be species specific.

Subsequent spraying revealed all ninhydrin reacting compounds to be species specific. The compound common to all and that common to the three species were indicated by their positive reactions with Pauly's reagent and alkaline silver nitrate to be phenolic (Fig. 11).

Negative results were obtained with Ehrlich's reagent in all cases.



FIGURE 11. One-dimensional descending chromatograms. A. Marsh fern, B. Bog fern, C. Long beech fern, D. Broad beech fern, E. New York fern (n = ninhydrin positive substance p = phenolic compound).

Discussion and Summary

All aspects of this study indicate exclusion of the beech and marsh ferns from the genus *Dryopteris*.

The blades of these ferns are light green, thin-membranous, and nonevergreen; whereas, those of true *Dryopteris* are deep green, subcoriaceous and evergreen. The vascular bundles reach the margins of the pinnae in contrast to those of the dryopteroids which terminate in hydathodes. The thelypteroids bear slender rhizomes; the stipes contain two vascular bundles which unite at the base of the blade. Massive rhizomes and stipes bearing 3-7 bundles are characteristic of the dryopteroids.

The spores are distinguished with some difficulty. Basically, the spores of the thelypteroids have a tuberculate surface; perispore ridges, if present, are quite discontinuous. Dryopteroid spores tend to be cristate with closed, anastomosing ridges (15).

Extensive cytological studies have been made of the American species of *Dryopteris sensu stricto* (19, 20). A base chromosome number of n = 41, accompanied by various levels of ploidy, was noted. This number has not been seen in any thelypteroid fern.

There appears to be much basis for maintaining the beech ferns as a separate genus *Phegopteris*. Some special characters common to the beech ferns as opposed to the marsh ferns are as follow: deltoid blades; exindusiate sori; spores wingless or bearing much reduced broken wings; chromosomes extremely small. Chromatographically, fewer common phenolic substances were detected in the beech than in the marsh ferns (Fig. 11).

In conclusion, the results of this study indicate the exclusion of the thelypteroid ferns from *Dryopteris;* further basic differences appear to justify the delimitation of the beech ferns as a distinct genus. A number of outstanding problems are inherent in these ferns. The relationship of the oak-fern to the beech ferns is one such problem. Another is the relationships of the thelypteroids of the western United States to those of the east. Extensive studies of Western North American, Asian, and European counterparts are needed before the taxonomy of the group can be understood fully.

Literature Cited

- 1. BRITTON, D. M. 1953. Chromosome studies on ferns. Amer. J. Bot. 40:575-583.
- BRITTON, D. M. 1961. The problems of variation in North American Dryopteris. Amer. Fern. J. 51:23-30.
- 3. BRITTON, D. M. 1965. The cytology and distribution of *Dryopteris* species in Ontario. Can. J. Bot. 44:63-78.
- CHRISTENSEN, CARL. 1913. A monograph of the genus Dryopteris Part I. The tropical American pinnatifid-bipinnatifid species. Kgl. Dansk. Vid. Selsk. Skr., VII. 10:55-582.
- CHRISTENSEN, CARL. 1920. A monograph of the genus *Dryopteris* Part II. The tropical American bipinnate-decompound species. Kgl. Dansk. Vid. Selsk. Skr., VIII. 6:1-123.
- 6. CHRISTENSEN, CARL. 1938. Manual of Pteridology. Chronica Botanica Press. 550 p.
- 7. COPELAND, E. B. 1947. Genera Filicum. Chronica Botanica Press. 247 p.
- DEAM, C. C. 1940. Flora of Indiana. Department of Conservation, Indianapolis. 1236 p.
- 9. FERNALD, M. L. 1950. Gray's Manual of Botany. American Book Co., New York. 1632 p.

- GLEASON, H. A. 1952. Illustrated Flora of the Northeastern United States and Adjacent Canada. New York Botanical Garden, New York. Vol. 1. 482 p.
- 11. MANTON, I. 1950. Problems of cytology and evolution in the Pteridophyta. University Press, Cambridge, Eng. 316 p.
- MORTON, C. V. 1950. Notes on the ferns of the eastern United States. Amer. Fern J. 40:213-225.
- MORTON, C. V. 1958. The Californian species of *Thelypteris*. Amer. Fern J. 48:136-162.
- 14. MORTON, C. V. 1963. The classification of Thelypteris. Amer. Fern J. 53:149-154.
- OLIVER, J. C. 1968. A study of spore characteristics of the ferns of Indiana. Amer. Fern J. 58:5-12.
- 16. SMITH, I. 1958. Chromatographic techniques: Clinical and biochemical application. Heinemann Medical Books, Ltd. London. 419 p.
- WAGNER, W. H. JR. 1963. A biosystematic survey of United States ferns-preliminary abstract. Amer. Fern J. 53:1-16.
- WAGNER, W. H. JR. 1966. Pteridophytes of the Mountain Lake area, Giles Co., Virginia: Biosystematic studies, 1964-5. Castanea 31:121-140.
- WALKER, S. 1961. Cytogenetic studies in the Dryopteris spinulosa complex II. Amer. J. Bot. 48:607-614.
- WALKER, S. 1962. Further studies in the genus Dryopteris; the origin of D. elintoniana, D. celsa and related taxa. Amer. J. Bot. 49:497-503.
- 21. WHERRY, E. T. 1937. Guide to eastern ferns. Science Press, Lancaster, Pa. 252 p.