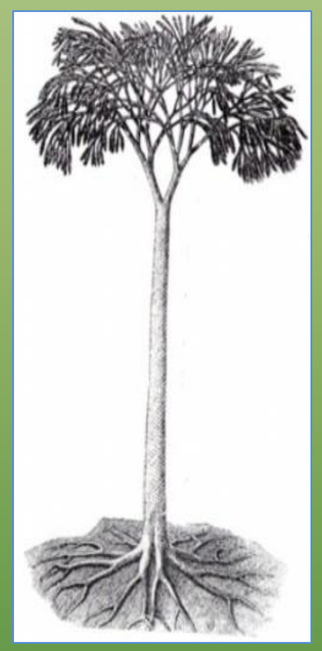
Determining evolutionary relationship among lycopsids using spore wall ultrastructure

James Taylor, Jason Ryba, and Courtney Lynne, Faculty Adviser Dr. Wilson Taylor
University of Wisconsin-Eau Claire, Dept. of Biology, Spring 2009

Background

Lycopsids are recognized as ancient seedless vascular plants that arose during the Early Devonian Period (Figure 1). Lycopsids demonstrate a variety of growth types, existing as both free-sporing herbs and 40 meter high trees that dominated many swamp forest landscapes (Figure 2). Fossil remains of these 300 million year old arborescent (tree-sized) lycopsids exhibit more diversity and ecological and economic significance than the living lycopsids of today (impressively, these lycopsid fossils account for a substantial portion of the Pennsylvanian coals of Euramerica).



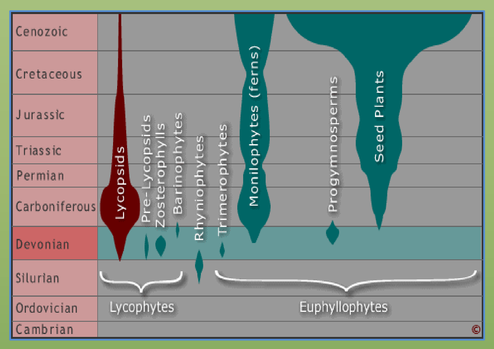


Figure 1. (above) Lycopsids, including the lycophytes, diverged from the earliest vascular plants during the Early Devonian Period. [1]
Figure 2. (left) Depiction of an early Lepidodendron, existing as a 40 meter high tree. [1]

Objectives

We aim to determine evolutionary relationships among early lycopsids via a multilamellated area of spore walls that is present in the modern lycopsids *Isoetes* and *Selaginella*, as well as basal lycopsid fossils as old as 400,000,000 years (Figure 3). Identifying this feature in other fossil lycopsids provides one means of deciphering evolutionary relationships. One major group in which this structure has yet to be identified is the Lepidodendrales. The purpose of this study was to find this structure in isolated microspores of the genus *Lycospora*, using electron microscopy.

Methodology

Specimen preparation:
- Fixation: Preserves suspended specimen in static state within plastic epoxy resin.
- Embedment: Epoxy resin permeates the specimen, replacing all water within the specimen.

Trimming & Sectioning:
- Specimen is centered in the resin and sectioned via an ultramicrotone into 100nm thick sections.
- Sections are transferred to grids.

Staining: Grids are stained using heavy metal salts to increase the density of organelles and macromolecules which ultimately increase contrast within the electron microscope.

TEM: Grids are inserted into the electron microscope. Electromagnetic lenses control imaging electrons with magnification and resolution capabilities over 1000x beyond a light microscope.

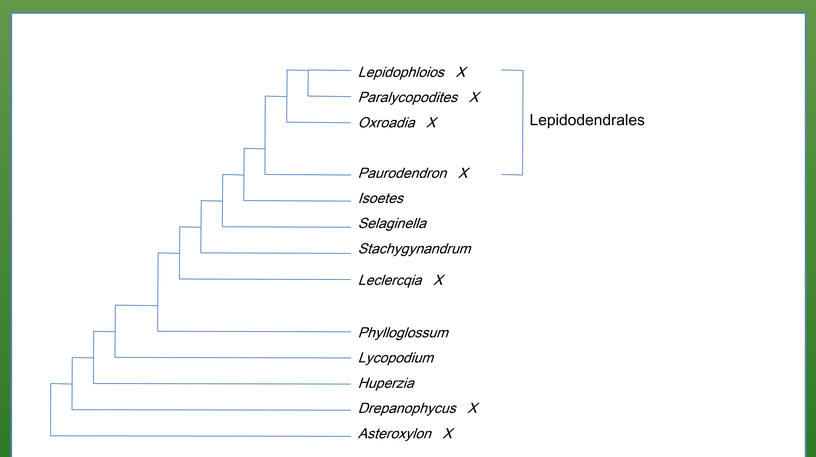


Figure 3. Numerical cladistic hypothesis of relationships in lycopsids (1992). X=extinct. [2]

Results

Although the mulilaminated structure has a very localized placement, we possibly identified one in a spore of *Lycospora* (Figure 4). In addition, the complex two-layered nature of the walls of these spores is similar to that seen in other spores with the multilaminated structure (Figure 5). These similarities between members of the lycopsid group support the hypothesized phylogeny and additionally help determine evolutionary relationships.

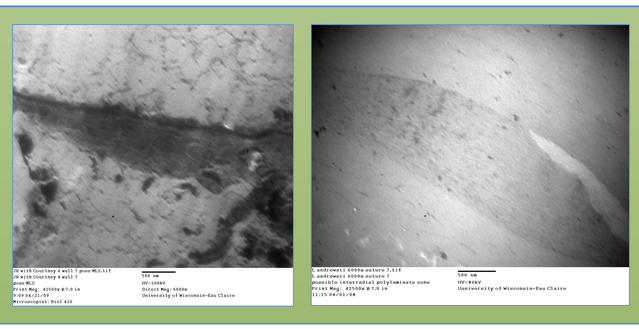


Figure 4. (Above, left) Possible spindle-shaped multilaminated structure of *Lycospora*. (Above, right) Similar multilaminated structure of *Leclercqia*.

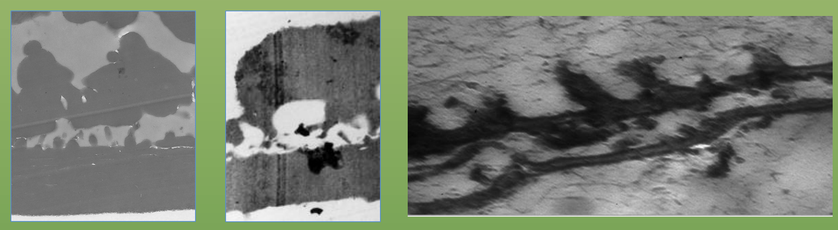


Figure 5. (Above, left) Wall cross section of *Leclercqia*. (Above, center) Wall cross section of *Paurodendron*. (Above, right) Wall cross section of *Lycospora*. Note the similarities of the complex two-layered walls with the enclosed central space within the different groups of lycopsids.

References

[1]Murphy, Dennis. 2006. "More About Lycopsids." *The Devonian Times*.
[2] Kenrick, Paul and Peter R. Crane. 1997. The Origin and Early Diversification fo Land Plants: A Cladistic Study. Washington and London: Smithsonian Institution Press.

Acknowledgements

▪UW-Eau Claire Center of Excellence for Faculty and Undergraduate Student Research Collaboration
▪UW-Eau Claire Department of Biology