Diversity of Strand Cells and the Implications for Phylogeny of Liverworts

Rachel V. Murray and Barbara Crandall-Stotler
Southern Illinois University, Carbondale

Introduction:

It is well known that the gametophytes of most mosses and some simple thalloid liverworts possess strands of elongate, hydrolyzed cells that are hypothesized to function in water conduction and/or storage (Tansley & Chick 1901). Characterization of these cells has mostly utilized sectioning techniques, which have not completely resolved the differences observed among strand cell morphologies, particularly as regards wall architecture and pit anatomy.

Past studies of strand cells in the Pallaviciniaceae have been equivocal, especially with regard to pit structure. Smith (1966) and Hébant (1977, 1980) describe the pits as wall perforations, with pore dimensions of 1500-2500 nm. According to Frey et al. (1996) these pits comprise unthickened areas of the walls and are not actually perforated. Most recently, Ligrone et al. (2000) described the pits of *Symphyogyna* and *Pallavicinia* as being rarely perforated, except for a small plasmodesmata-sized pore. The debate over pit anatomy frames the larger question of whether or not the central strands of taxa in the Pallaviciniaceae evolved independently (Ligrone et al. 2002) or were derived from the more basal *Haplomitrium*.

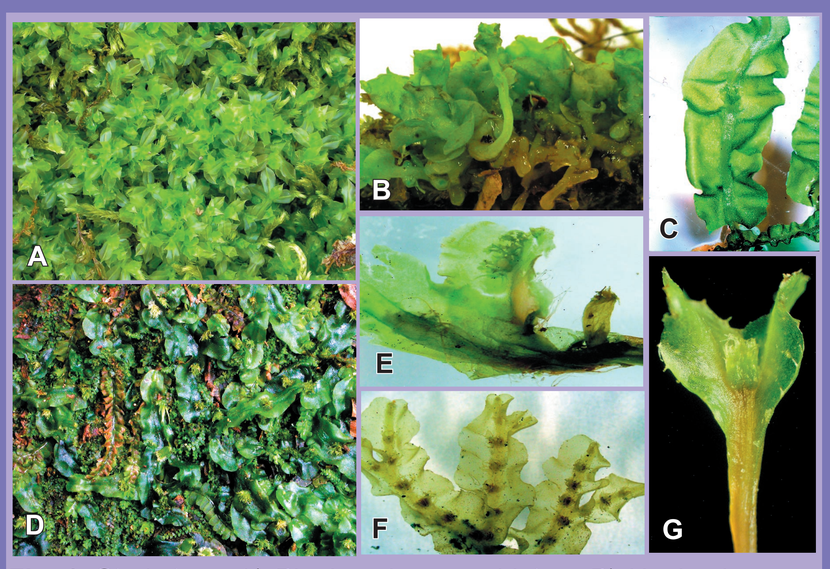


Fig. 1- Study taxa. A) *Plagiomnium cuspidatum;* B) *Haplomitrium blumii;* C) *Hattorianthus erimonus;* D) *Pallavicinia lyellii;* E) *Greeneothallus gemmiparus;* F) *Symphyogynopsis filicum* G) *Jensenia connivens*

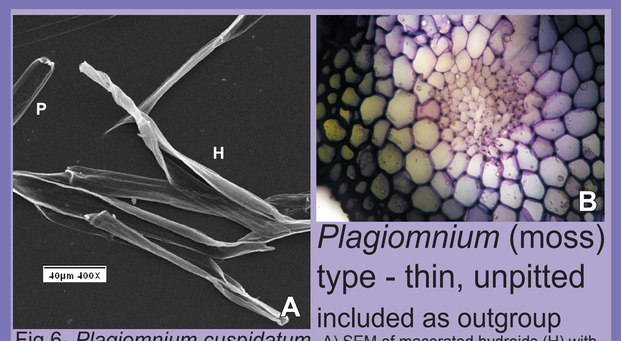
Materials:

- *Plagiomnium cuspidatum* - Illinois, USA
- *Haplomitrium blumii* - Genting Highlands, Malaysia
- *Hattorianthus erimonus* - Nara, Japan
- *Pallavicinia lyellii* - Louisiana, USA
- *Pallavicinia longispina*- Sri Lanka
- *Greeneothallus gemmiparus*- Tierra del Fuego, Chile
- *Jensenia connivens*- South Island, New Zealand

Methods:

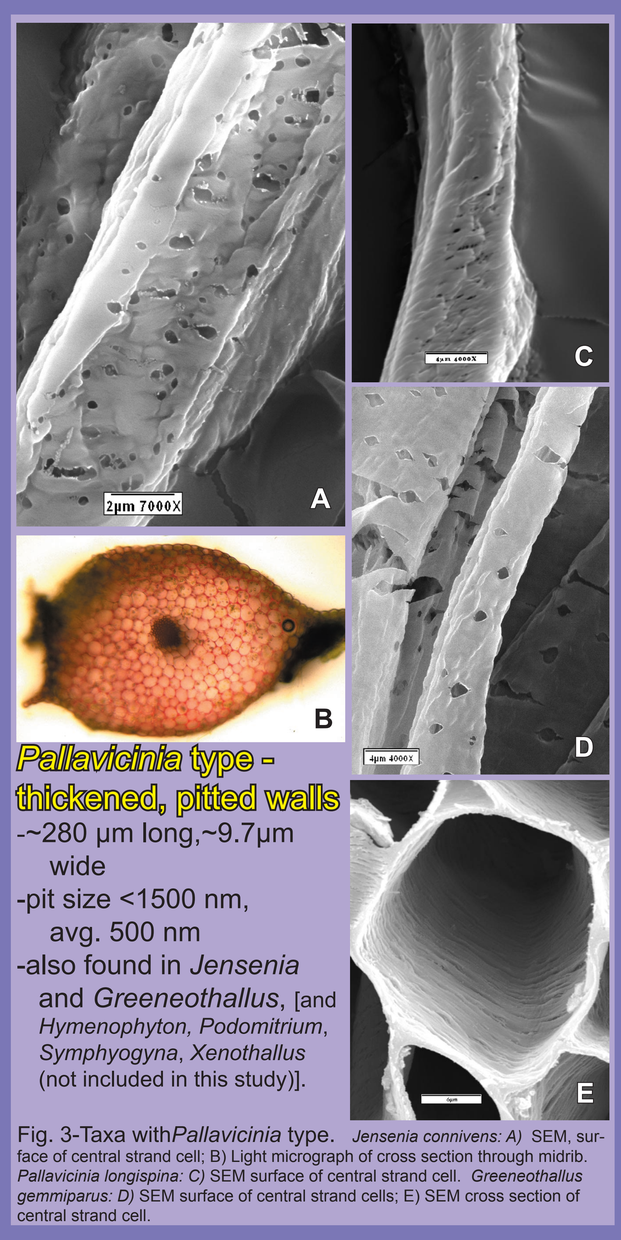
* Thallus midribs were sectioned transversely to locate central strand(s).
* Hand dissected central strands were macerated using Jeffrey's solution of 10%nitric/10%chromic acids for 2 1/2 hours.
* Some cells were stained using toluidine blue O or aqueous safranin for examination with the optical microscope; others were prepared for SEM microscopy as follows:
  - -Cells were teased apart onto a glass cover slip and air dried.
  - -After mounting on stubs, specimens were sputter- coated with 400Å gold-palladium in a Denton Desk II SC.
  - -Digital images were captured using a Hitachi S570 Scanning Electron Microscope.

Results: Three types of strand cell anatomy are found in the Pallaviciniaceae, including the *Haplomitrium-type*.



Plagiomnium (moss) type - thin, unpitted included as outgroup

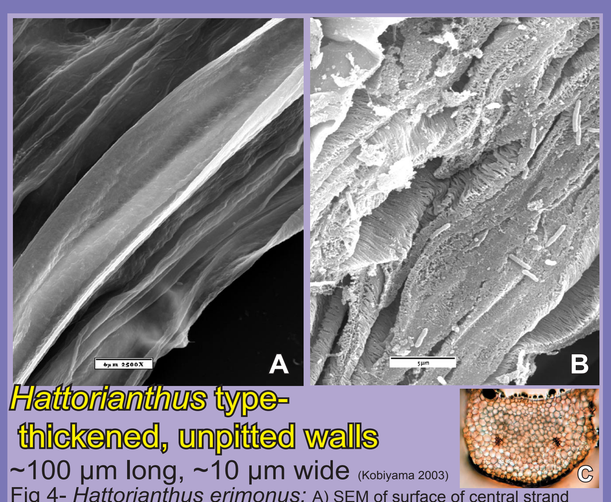
Fig.6- *Plagiomnium cuspidatum.* A) SEM of macerated hydroids (H) with a parenchyma cell (P); B) Light micrograph of cross section of midrib, stained with toluidine blue O



***Pallavicinia* type - thickened, pitted walls**

- -~280 µm long,~9.7µm wide
- -pit size <1500 nm, avg. 500 nm
- -also found in *Jensenia* and *Greeneothallus*, [and *Hymenophyton, Podomitrium*, *Symphyogyna*, *Xenothallus* (not included in this study)].

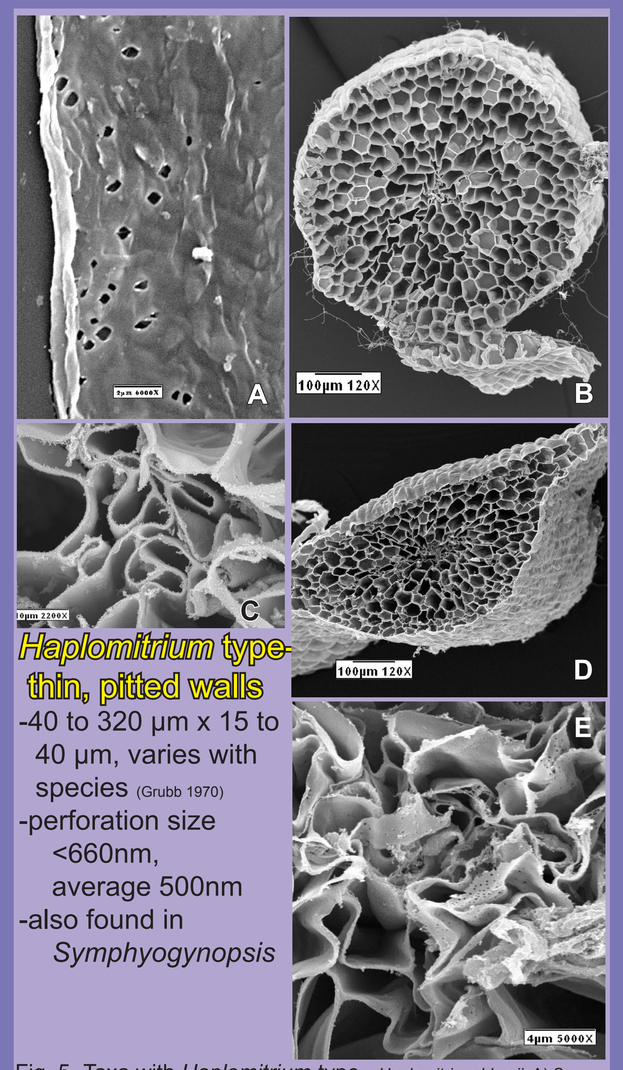
Fig. 3-Taxa with*Pallavicinia* type. *Jensenia connivens: A)* SEM, surface of central strand cell; B) Light micrograph of cross section through midrib. *Pallavicinia longispina: C)* SEM surface of central strand cell. *Greeneothallus gemmiparus: D)* SEM surface of central strand cells; E) SEM cross section of central strand cell.



***Hattorianthus* type- thickened, unpitted walls**

~100 µm long, ~10 µm wide (Kobiyama 2003)

Fig 4- *Hattorianthus erimonus:* A) SEM of surface of central strand cells; B) SEM of longitudinal section of central strand; C) Light micrograph of cross section of midrib



***Haplomitrium* type- thin, pitted walls**

- -40 to 320 µm x 15 to 40 µm, varies with species (Grubb 1970)
- -perforation size <660nm, average 500nm
- -also found in *Symphyogynopsis*

Fig. 5- Taxa with *Haplomitrium*-type. *Haplomitrium blumii:* A) Surface of central strand cell; B) Cross section of stem; *Symphyogynopsis gottscheana:* C) Cross section showing collapsed central strand cells; D) Cross section of thallus midrib; E) Cross section of central strand cells, showing pitted walls

Conclusions:

These data confirm that *Pallavicinia* type strand cells resemble those of the *Haplomitrium* type in having pits that are clearly perforated, with an average perforation size of 500 nm in both. The perforations likely are formed by the dissolution of the plasmodesmata during cell hydrolysis, and not by dissolution of wall material. Thus, the only major difference between these two types of central strand cells is the presence or absence of secondary wall thickenings.

Our findings further suggest that the *Pallavicinia* type is derived from the *Haplomitrium* type, as also supported by molecular phylogenetic reconstructions (Fig. 7). The *Hattorianthus* type is unique within the Pallaviciniaceae, but supports the placement of *Hattorianthus* with *Moerckia*, a taxon that also has strands that lack pits (Hébant 1977).

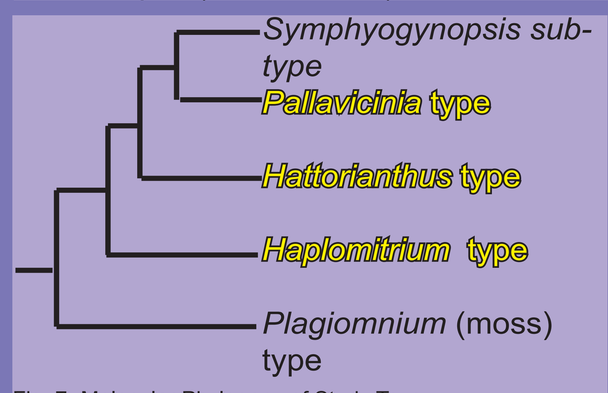


Fig. 7- Molecular Phylogeny of Study Taxa

References:

Frey, W, H. H. Hilger & M. Hofmann. 1996. Nova Hedwigia 63: 471-481; **Grubb, P. J**. 1970. New Phytologist 69: 303-326; **Hébant, C**. 1977. The Conducting Tissues of Bryophytes. Lehre, J. Cramer. **Hébant, C**. 1980. J. Hattori Bot. Lab. 47: 63-74; **Kobiyama, Y**. 2003. Comparative Development and Ultrastructure of the Specialized Parenchyma Cells and/or Hydrolyzed Cells in Select Liverworts and Hornworts. PhD Dissertation, Southern Illinois University; **Ligrone, R., J. G. Duckett & K. S. Renzaglia**. 2000. Phil. Trans. R. Soc. Lond. (B) 355: 795-813; **Ligrone, R. K. C. Vaughn, K. S. Renzaglia, J. P. Knox & J. G. Duckett.** 2002. New Phytologist 156: 491-508; **Smith, J. L.** 1966. The Liverworts *Pallavicinia* and *Symphyogyna* and their Conducting System. U. California Press, Berkeley; **Tansley, A & E. Chick**. 1901. Ann. Bot. 15: 1-38. pls. I-II.

Acknowledgements: **Funding for this project was provided by an REU supplement to DEB-9977961, a grant of the NSF PEET program. Thanks to Ray Stotler, Li Zhang, and Dylan Kosma for valuable input.**