



# The Cycad Apical Meristem: An Investigation of the Development and Lineage of Cycads



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**Abstract** The hundreds of species of cycads are seed bearing plants found native to many parts of the world. They are characterized by spiny leaves protruding from a stout trunk that is fortified by tough scales. The question my research poses is, where do these species fit in with other modern plants? Cycads are currently classified as gymnosperms do to the shared quality of bearing naked seeds. However, my practicum advisor Todd J. Cooke submits these plants are a lineage of seed ferns that are believed to have gone extinct approximately 100 million years ago. I have spent the last several months dissecting down to the apical meristem of cycads and photographing them using Scanning Electron Microscopy (SEM). The hope of this research is to build a body of evidence to support the hypothesis of a cycad-seed fern relationship.

**Introduction** Cycads have been classified as gymnosperms for the reasons that they both bear naked seeds and produce wood. An intrusive observation of cycads reveals this "wood" is more stiff, vascular tissue and hardly wood at all. Going further, we assert that the ability to bare naked seeds was a trait independently evolved in cycads and gymnosperms, not an ancestral trait. This would sever the evidence defining cycads as gymnosperms. Our research begins with looking at the cycad meristem for geometric patterns different than the gymnosperm meristem.

**Methods** My partner and I began by cutting away 95% the tissue on each plant. The meristem is a bundle of a few hundred cells located in the center of the plant. Most of the tissue was removable by a razor blade. Finer tools were used as the meristem was approached. At a certain point the tissue became too indistinguishable and the portion of the plant containing the meristem was cut away

The heavy hitting technology behind my research was the scanning electron microscope. This instrument allowed me to look at plant cells under more than a thousand times magnification.



The first stage of dissection did not require magnification. As more tissue was cut away, a specialized dissection light microscope was utilized to cut away as much excess plant material as possible without harming the meristem.

On average this piece of plant flesh was about the size of an apple seed. It was left to fix in formaldehyde for at least 24 hours. At this step I proceeded to fix the samples for scanning electron microscopy using a series of washes. Once washed, the samples were dehydrated completely via an ethanol wash series and critical point drying. The tissue was now easily removable, allowing further dissection that completely exposed the meristem.

I coated the samples with tungsten in a vacuum instrument. This is the final step before SEM observation. This lays down a conductive surface allowing the electron beam to bounce off the sample and be read by the machine.

The fruits of our work is to look at the apical meristem, the youngest part of the plant.

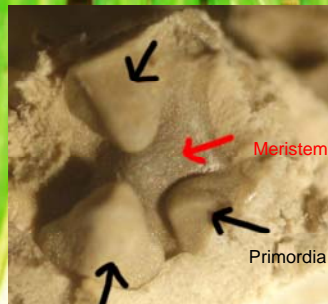
**Results and Conclusions** The research I have been doing is ongoing. My lab partner, site advisor, and I are compiling a collection of high definition cycad apical meristems via SEM to properly support reclassification of these plants. We are still in the research stage and have yet to adequately interpret the data. Images with a lack of definition in the critical region of interest have been setbacks. We are experimenting to see if this problem is developing during the fixation process or elsewhere.

Future work involves obtaining clear documentation of apical meristem geometry across several cycad species. Contrasting these data with images of gymnosperm meristems will be the definitive argument for this hypothesis.

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1010X magnification with SEM. The cell's of this sample are desiccated, accentuating the geometry of each cell's wall. The youngest cell's of this meristem are highlighted.

40 X magnification under a light microscope.



156X magnification with SEM. Each bulge is a cell of the meristem.

