Interpreting the *Pseudocot* Phylogeny

Students use physical characteristics, genetics, and a fossil record of hypothetical flowering plants to develop fundamental phylogenetic tree-building skills.

Grade Level: Post Secondary Courses
(vocabulary and activities can be modified for use in upper-level high school courses)
Subject: Biological Sciences- Evolution (phylogenetics and tree building)
Duration: Two class periods with homework between classes.

Description: In order for students to develop expertise in phylogenetic systematics and become efficient problem solvers, students must cultivate representational competency associated with tree-thinking. This lesson helps students build the fundamental skills for reading and building phylogenetic representations they are expected to develop upon completing a plant systematics or evolution course. This particular lesson uses hypothetical flowering plants to assess and instruct essential phylogenetic tree-building skills, specifically: appropriate use of evidence, identification of key tree features, understanding of significant patterns, transferability across representations, and ability to verbally describe depicted relationships.

Goals and Objectives: This lesson explicitly addresses and helps overcome the following challenges associated with phylogenetic tree building:

1. Lumping organisms based on single characteristics rather than looking holistically at the organisms.
2. Ignoring critical data & using uninformative evidence to construct phylogenetic trees.
3. Difficulties transferring empirical data into a visual representation illustrating evolutionary relationships.
4. Creating consensus nodes to address discrepancies.
5. Altering the format or orientation of the tree alters the relationships depicted.
6. Generating a comb-like branching structure without incorporating hierarchical lineages.
7. Using trees to reconstructing ancestral states.
8. Identifying the most likely tree by comparing representations

This two-class period *Pseudocots* lesson has been used with secondary and post-secondary students. Students engaged in tasks such as: selecting appropriate data sources to group taxa into monophyletic groups (clades), constructing a phylogenetic tree representing these clades, adjusting the representation to incorporate new taxa, comparing their tree to other hypotheses, and forming predictions about historic and future character states.

Objectives:

- Students will be able to identify informative data that can be used to build a phylogeny.
- Students will be able to generate a phylogenetic tree from hypothetical data.
- Students will be able to alter phylogenetic models to accommodate new data.
- Students will be able to compare phylogenetic trees and determine how different models provide support or dispute their phylogeny.

It helps if students are familiar with reading phylogenetic trees prior to this lesson. Thus, I suggest that the Pipe Cleaner Phylogeny lesson (Halverson, K.L. (2010). Using pipe cleaners to bring the tree of life to life. *The American Biology Teacher, 72(4),* 223-224. doi: http://dx.doi.org/10.1525/abt.2010.72.4.4) or similar tree-reading unit be implemented prior to this lesson. It is also helpful to make sure students are familiar with the following terms:

Apomorphy – a derived character state unique to one lineage
Branch – illustrates the different lineages after speciation events
Character - a specific heritable trait described in terms of its state
Consensus Tree (or node) - a phylogenetic tree derived by combining common features in multiple trees, often leading to undefined relationship hierarchies of sister taxa.
Internode - a shared lineage between speciation events
Lineage - a continuous line of decent
Monophyletic (Clade) - a grouping of a common ancestor and all of the descending lineages
Node - a hypothetical common ancestor or speciation event
Outgroup - a distantly related species that resides outside of the clade of interest
Pseudocot - hypothetical flowering plant clade
Phylogenetic tree - an evolutionary tree (shows relationships among organisms)

This lesson is very dependent upon the timing of when data is introduced to the students. Thus, it is important to be aware of when to hand out the different portions of supplementary data or unintended outcomes may result. To implement this lesson, you will need copies of provided student worksheets, supplementary data, scissors, and it is helpful to have colored pencils or markers.

Class 1:

Begin by engaging students with a scenario:

> You are part of a research team that made an amazing discovery of some unique-looking flowering plants. The lead investigator of your research team thinks that this new group of species is evolutionarily different that currently known plants and they are a new clade being called *Pseudocots*. It is your job to determine how these plant species are related to each other and the closest currently described flowering plant (a potential outgroup) by building a phylogenetic tree.

Once you have presented the scenario and engaged the students, allow them to talk about how they might want to proceed to create a phylogenetic tree. This is a good step to assess what students know about phylogenies before beginning the lesson. Most students may never have encountered this type of task previously and will need a lot of guidance. If you have some students with tree building experience, you may want to group them accordingly.

Have students form groups of 2-3 students per group (students can also complete this activity individually and discuss as a class). Hand out *Worksheet 1a, Scissors,* and the data for the *12 living Pseudocots and field notes.* For an optional adaptation, you can reduce the number of plants used in this activity to six plants: species 1, 2, (4 or 5), 6, 7, 11. This adaptation simplifies the activity and reduces the amount of time needed for the lesson.

Have the students cut out the images of the *Pseudocots* (be sure to keep the species number attached to each image) so they can physically arrange how they think the plants might group together. Give students time to group the species and think about how they might be related. As you observe the students have them consider what types of evidence they are using to organize their plant species and why that evidence is informative (e.g., are they using the field notes and/or morphological data – why?). It is important to have students indicate the evidence they used for their classification method to ensure they are not basing the relationships on uninformative ecological data.

Students often think that the ancestral character states are more primitive regardless of the evidence presented. Thus, once students have settled upon a classification scheme, have them predict what the most ancestral *Pseudocot* might look like and draw this prediction on their worksheets. Images will vary quite a bit at this point.

Once they have committed to their prediction, hand out *Worksheet 1b and the Fossil Record.* Allow students time to compare and discuss the fossil record and work through the two problems on this worksheet. Have students discuss differences/similarities between their previous predictions and their current ideas. This is an optimal point to discuss issues with ancestral state reconstruction as a class. It is good to note that the oldest fossil in the record may or may not be indicative of the ancestral states for multiple reasons. For example, we might not have a complete fossil record and there could be older fossils not yet found, there could be ancestral states that have been lost prior to the fossilized species.

By the end of the first class period, students should have completed Worksheets 1a and 1b. You can collect these or let students keep them for reference as they work on the homework portion of this lesson. Regardless, all students should make note of their original species grouping and should keep the species images to assist with the homework and bring them to class the next period as well. These images are extremely helpful with all portions of the lesson.

For homework (or completed after Day 1 or in class at the beginning of Day 2 if time allows), give students (or post online) the *Homework* worksheet with *Genetic Data*. Instruct the students to fill out the data matrix with 0's and 1's (and 2's if necessary when there are three variations). The 0's represent traits present in the outgroup or "ancestral" traits and 1's represent differing traits than what can be found in the outgroup. Note that if you you are only using six species then reduce the genetic data to the following codons: A, D, F, G, H, I, J, K.

Class 2: **Constructing a Phylogenetic Tree**

This is the most challenging portion of this activity. Students have already organized morphological character states and genetic data of the *Pseudocots* in a data matrix. This step helps students translate the matrix information into a phylogenetic tree and should be done as a class. You may find that students may have differing matrices, so I suggest you begin by creating a single consensus matrix as a class and use this to complete *Worksheet 2a*.

There are five steps that you can use to help provide a scaffold for students to be able to translate raw evidence into a phylogenetic tree:

Step 1: Begin with a Data Matrix that organizes raw evidence (completed as homework)

Step 2: Reorder Matrix so that it is easier to see patterns of apomorphies (optional)
Step 3: Create Venn Diagram of Species
Step 4: Add Branches
Step 5: Connect Lineages and Root Tree

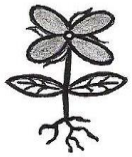
You may want to encourage students to record these steps on their worksheets as a reference for later tree-building problems. Please see the Instructor's 5 Step resource for a key to the lesson activities. As an optional adaptation, to further assess students' tree-building abilities, you may have them build a tree from their personal matrix if they are different than the one created in class. Or you could have them build a tree from the full or remaining species if you elected to only include the selected six species as a class on the original tree.

After the class has constructed a *Pseudocot* phylogenic tree, hand out *Worksheet 2b*. The clades organized as a class can be tested against other phylogenetic trees. Have students circle the monophyletic groupings on each tree and compare clades, or "collapse branches to the nodes." If only the species of interest are included, then there is support, if a different species is included within the clade, then there is no support.

You can opt to have students compare their tree to all or just a subset of the trees provided. Or, if you have students develop trees individually after the example completed by the entire class, have students compare their trees with a neighbor. How these trees compare will vary based upon how the students grouped the species. For another example of how to compare phylogenies see: BioQUEST Curriculum Consortium. (2008, October 4). BEDROCK: Whippo Problem Space. Retrieved October 15, 2006 from http://www.bioquest.org/bedrock/problem_spaces/whippo/

For a follow-up activity, you can assess students' tree-building skills is by having them apply their knowledge to an example that relies upon a conceptual understanding of phylogenies. This follow-up example presents a problem similar to those faced by scientists on a regular basis - new data is introduced that they have to accommodate in their understanding. You can share this scenario with the students:

> Late breaking news: Two scientists were on a collecting exhibition in the Amazon (where there is limited sunlight and poor soil available) and found a new *Pseudocot* species (#24). Upon genetic analysis, we have found the coding sequence to be: ATG, ATA, CGT, TAA, AAC, TGC, TAA, ACC, AGT, ATA, CAA, ATT, CTG. Accommodate this new data and incorporate this new species into your phylogenetic tree.



New Species (#24)

Please note that students may have to readjust the current relationships they had predicted in order to fit this species into the phylogeny. Have them build a tree and turn it in for evaluation.