

Phylogenies I: Phylogenetic Inference

Prelab

Study this Phylogenetic Inference lab description, section 28.1 and BioSkills 7 in the textbook, and “The Tree-Thinking Challenge” in the Readings folder of the course website; then do the Lab 5 Prelab linked to the course website. The next week’s prelab link is posted each Thursday afternoon. See “Submitting Catalyst Exercises” in this course manual; prelabs are due by Tuesday at 8:00 AM. (2 pts)

Learning Objectives

This lab, the next lab, and the associated lectures, introduce what biologists call “tree thinking” – the ability to understand how evolutionary relationships are inferred and how to interpret them. At the end of this week’s exercise, you should be able to do the following:

- Distinguish between homologous and homoplasious traits.
- Explain why phylogenies can be estimated by analyzing shared, derived characters, and articulate the assumptions and logic involved in determining that a particular trait is shared and derived.
- Explain how knowledge of outgroup and descendant taxa and the principle of parsimony can be used to infer the ancestral states of traits.

The lab is also designed to help you review an array of concepts introduced in lecture, including outgroups, monophyletic groups, and the use of parsimony as a logical tool. The most important skill you’ll be practicing is how to use synapomorphies to construct evolutionary trees.

An Introduction to Phylogenetics

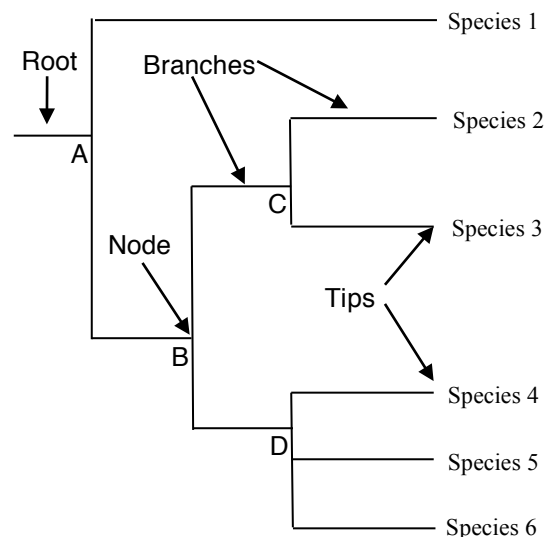
Phylogenetics is the branch of biology that focuses on reconstructing the evolutionary relationships among species. An evolutionary tree describes how populations or species of organisms are connected by descent from a common ancestor.

Reading an evolutionary tree

An evolutionary tree is a diagram that shows the evolutionary relationships among a group of organisms. You are probably familiar with how to read a family tree, but almost everyone needs help learning how to read a phylogenetic tree.

To begin, examine the tree at right. Note that it has four basic parts: a root, nodes, branches, and tips. The key to reading a tree is to realize that each branch represents a population or species through time.

To read a tree, start at the root and read toward the tips. Almost immediately, you’ll find a node marked A. It should make sense to you that the population at node A represents the common ancestor of species 1-6.



At node A, one population splits into two distinct populations. The split is represented by a vertical line; the two populations are represented by new branches (horizontal lines). One of these populations

(the upper one) continued to evolve into Species 1. The other (the lower one) evolved until it reached node B. At that point, it split into two distinct lineages.

What happened to these two descendant species? Keep reading to the right of node B, and you'll find that the upper one evolved until you reach node C. Then the population at node C split into distinct groups that eventually became Species 2 and Species 3. The other species that descended from node B continued to evolve until node D, when it split into three independent lineages. These three descendant populations went on to become Species 4, Species 5, and Species 6.

Reconstructing an evolutionary history

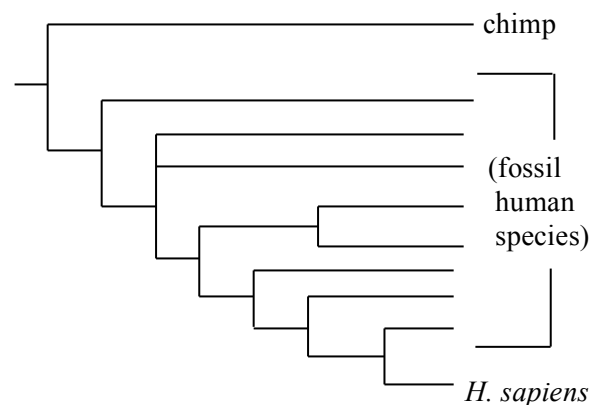
How can biologists reconstruct the evolutionary history of a group of species? To answer this question, biologists assume that species that are similar are probably closely related, while species that are less similar are less closely related. To assess the similarities and differences among species, biologists may analyze the composition of a particular molecule found inside cells or some aspect of morphology—for example, the size and shape of the body. The key is to group species on the basis of **synapomorphies**: similarities that are shared because they are derived from a common ancestor. By grouping the most similar species, then connecting them to less similar species, a tree of relationships can be estimated.

Although the logic of reconstructing an evolutionary tree sounds simple, in practice the analysis can be extremely complex. For example, suppose you wanted to reconstruct the evolutionary relationships of the human-like species in the fossil record, including *Homo sapiens*. Further, suppose you wanted to do this analysis based on similarities and differences in the morphology of teeth and skulls. To begin, you would need to document characteristics that are similar in some species and different in others. Then to decide which similarities occur because of descent from a common ancestor—meaning that they qualify as synapomorphies—you would need to decide which traits appeared early in evolution and which appeared later. Traits that appear early in the history of a lineage are referred to as **ancestral** or **basal**; traits that appear later are considered to be **more recent** or **derived**. How can biologists determine which traits are basal and which derived? There are several sources of evidence.

- 1) **Outgroup comparison.** An outgroup consists of one or more species that are closely related to the group in question, but not part of it. For example, various types of data suggest that chimpanzees are the closest living relatives of humans. Because chimps have relatively small braincases, most observers infer that the common ancestor of all humans also had a relatively small braincase, and that large braincases evolved later.

It's important to recognize, though, that traits rarely if ever change through time in a linear or “progressive” manner. For example, the species known as the Neandertals, which disappeared about 30,000 years ago, had *larger* braincases than contemporary humans.

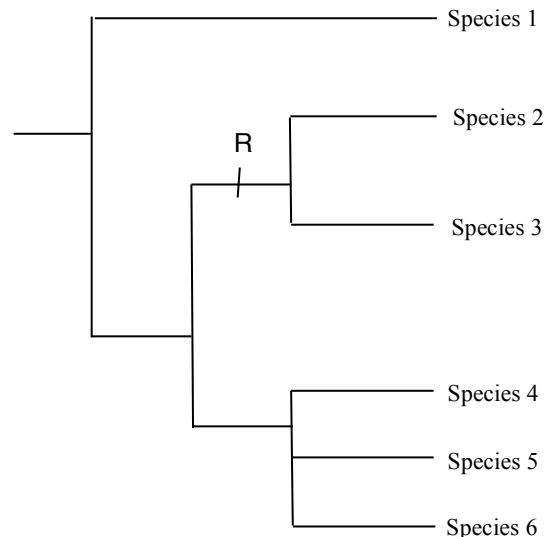
- 2) **Dating of fossils.** Most human fossils have been dated using radiometric techniques. Because the oldest fossils tend to come from species with relatively small braincases, biologists can argue that small braincases were an ancestral trait.



3) **Establishing trait polarity**, or the direction of evolutionary change. If a clear outgroup is lacking, logic can suggest which traits are derived from others. For example, in many cases it is logical to argue that derived traits are more complex or specialized or elaborated than basal traits. In the human lineage, for example, large brains and flattened faces appear to be a complex or specialized condition. Exceptions exist, however. Humans have a tiny, useless tailbone; snakes and whales have lost limbs that existed in their ancestors. These traits are derived, but are less complex or elaborate than their ancestors.

Throughout an analysis like this, it's important to realize that biologists reconstruct evolutionary relationships by comparing **homologous traits**—meaning, characteristics that are inherited from a common ancestor. To drive this point home, consider that no one would argue that penguins, which are birds, and dolphins, which are mammals, are closely related—even though they both have fins and streamlined bodies. Instead, the fins and streamlined bodies of penguins and dolphins evolved because these traits make swimming particularly efficient. The traits did not occur in a common ancestor of birds and mammals, but evolved independently in the two groups due to **convergent evolution**—the evolution of similar traits in distantly related organisms due to adaptation to similar environments. The fins and streamlined bodies of penguins and dolphins are examples of **homoplasy**, not homology.

When constructing a tree, it is helpful to map (the origin of) synapomorphies—that is, to label the points on the tree where the traits of interest first appeared. On the tree at right, if species 2 and 3 (and no other species) have trait "R," the origin of R would be indicated by a hashmark (short vertical bar) and label as shown.



Because it is often difficult to decide which traits are basal and which derived, and which are homologous or homoplasious, different researchers can reach different conclusions when interpreting traits and thus come up with different phylogenetic trees. Based on the information available at a certain time, biologists may be unable to decide which of their proposed trees most-accurately reflects the actual history of the group. As a result, phylogenetic trees must be considered hypotheses or estimates, and are continuously re-examined and updated as new fossils or other types of data come to light.

Exercise: Inferring the Phylogeny of Trilobites

To help you appreciate how biologists estimate evolutionary trees, you will reconstruct a phylogenetic history of a well-known group of arthropods called the trilobites. (Arthropods are a lineage that includes the spiders and mites, the insects, and the crustaceans—crabs, lobsters, shrimp.) Trilobites are extinct, but they existed for at least 325 million years and are extremely common in the fossil record. We will work with just a few of the more than 17,000 species that have been identified so far.

QUESTION: What is the phylogeny of the Trilobites?

Procedure:

- 1) Examine the trilobite fossils on loan from Dr. Liz Nesbitt at the Burke Museum. The fossils should give you a sense of the 3-dimensional shape of trilobites. Please handle these with care and respect.
- 2) Look through the 15 drawings of trilobite species. As you begin to study them, ask yourself some questions.
 - What traits do they all have in common?
 - What differences exist?
 - Which characteristics might be derived forms of simpler, more generalized traits?
 - Which traits seem most likely to reflect common ancestry, and which might be similar due to convergent evolution?
- 3) Your TA will hand out sheets that identify some of the more prominent structures on the dorsal (back) surfaces of trilobites. Using these traits and others that you've noticed on your own, begin to identify synapomorphies—derived traits that link certain species as members of the same lineage—in the 15 species drawn. Use the following criteria to decide how species relate to each other:
 - Complex or specialized characteristics do not arise all at once, but are derived from simpler precursors in ancestors.
 - If a complex structure is lost during evolution, it cannot be regained in a single step but must evolve independently through intermediates, if at all.
 - Once speciation occurs, the daughter species evolve independently. Changes will occur in one lineage without occurring in the other.
 - Similarity often reflects relatedness, but similar evolutionary pressures may cause similar features to evolve in separate lines. (Recall the dolphin and penguin example.) However, phylogenetic trees that minimize the independent development of such similar structures are more parsimonious than ones that require many trait gains or trait losses. By convention, biologists often consider the most parsimonious tree as the most likely. Parsimony assumes that convergence is much rarer than similarity due to modification from a common ancestor. This assumption has been supported by experimental and observational data.

Note: We have not provided information on average body size of the species you are studying because, in most lineages, it is common to find that overall body size changes rapidly during evolution. Aspects of body shape tend to be much more conservative, however—meaning that they change more slowly over evolutionary time. As you study trilobite morphology, consider quantifying aspects of body shape (relative proportions).

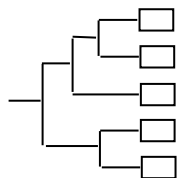
- 4) Arrange the 15 trilobite species into a phylogeny. There are many ways to proceed. You can use the following suggestions or devise your own scheme.
 - Based on the above rules, you might sort the species into piles that probably represent different lines of descent from a common ancestor.
 - Which species or group of species appears to have the fewest derived characteristics? Start from there and choose one pile.
 - Order the species in that pile in a progression that connects the most basal traits to most derived, according to the trend (or trends) that seem most significant to you.
 - Do all of the characteristics of each species fit these trends, or are there conflicting changes that must be reconciled?
 - What changes do not fit with your proposed scheme?
 - Does another arrangement of the species give more consistent trends among traits?
 - Does a straight progression seem to accommodate all the changes you observe or is branching at some point better able to explain what you see?

When the first pile is arranged to your satisfaction, repeat the process with a second pile. Which species in the first series that you laid out has features most similar to the species with the most basal characteristics in your second pile? It may be helpful to write down brief descriptions of the hypothesized appearance of the organisms in the ancestral populations at each node on your tree.

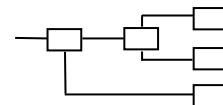
Note that many different phylogenetic trees are possible, depending on how you analyze characteristics and on what assumptions you make. Decide on your “best” tree. We will compare trees, so be prepared to state your assumptions, defend why you placed each species as you did, and explain any problems you see with your scheme.

- 5) Use your sorted trilobites to build a tree on a section of the whiteboard. Assume that none of the species is the direct ancestor of any of the others. Your tree should resemble the one on the left below, not the one on the right:

Like this ...



not this ...



- 6) Map the origin of synapomorphies with labeled bars across branches; see the textbook for examples. Add the numbers of the trilobites (large enough to be seen easily). Write your names and lab section next to your tree.
- 7) Draw your tree or have your TA photograph it. It's important to do this **now**, before you make any changes!
- 8) Your TA will provide a sheet with additional information about the 15 trilobite species. Examine these data carefully, and determine whether they suggest changes in your tree.

- 9) Construct a character matrix (a table that records the state of each character in each species in your study) below.
- Note:** In constructing the character matrix, use a 1 to indicate present and a 0 to indicate absent (for presence-absence data). If the traits you've coded are more complex than present-absent, then create numerical codes for each of the character states you've identified (e.g. if you scored the trait "relative size of cephalon" you might code the states as 1 = < 10% of body length, 2 = 10-25% of body length, 3 = > 25% of body length). At the bottom of the matrix, write notes that specify the codings for each trait.
- 10) Spend some time examining the trees estimated by other student groups. With the other members of your lab group, take turns explaining your tree to other students.
- 11) As a group, provide brief answers to the questions on the Phylogeny Inference Questions sheet. Hand in your group's answers and character matrix before you leave lab.

Student names: _____

Trilobites character matrix

NOTE: If you coded more traits than the columns given here, hand-write additional columns and staple them to this sheet.

[illegible]

Character codings:

Student names: _____

Phylogenetic Inference questions

1. On your phylogenetic tree, which lineage is the sister group to all of the other species on this tree? Why did you place this lineage where you did?
2. According to your phylogenetic tree:
What is one basal characteristic?
What is one derived characteristic?
Are there any traits that were lost but then evolved again independently? If so, what are they and where do they occur?
3. According to your phylogenetic tree, are the paired rear spines observed in *Albertella helena* homologous or homoplasious with the paired rear spines of *Crepicephalus towensis*? Explain.
4. Describe one important difference between your phylogenetic tree and a tree estimated by a different lab group. Upon reflection, do you stand by your original tree? Why or why not?

Who constructed the comparison tree? (list the student names):

5. Did you revise your initial tree based on the additional data provided? Explain why or why not.

Give this sheet and character matrix to your TA, and be sure your trilobite tree was photographed.