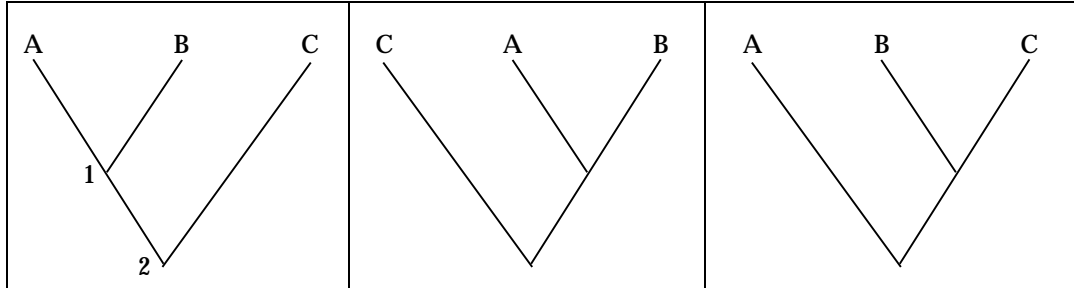


## Lab 06 Phylogenetics, part 1

A phylogeny is a visual representation of a hypothesis about the relationships among a set of organisms. Phylogenetics is the study of phylogenies and their development. This week and next week you will learn some of the tools we use to generate and evaluate phylogenies.

### Reading phylogenies as hypotheses

The figures below represent hypotheses about the relationship between three species, A, B, and C.



**Figure 1.**

In these phylogenies, related species are grouped together by lines connected at nodes. Nodes should always bifurcate. If there are three lineages rising from a single node, the relationships among the three species are considered unresolved.

Species sharing a node that they don't share with any other species (as in species A and B on the left, which share node 1) are considered each other's closest relative. Thus, A and B are more closely related to each other than either are to species C.

A clade is a group of species that share a node. A clade does not exclude species, for example, on the left, A&B are a clade, A, B, &C are a clade, but A&C are not a clade. Clades are sometimes given names (genus, family, order, class, and phylum are examples of clades).

Although there is a temporal order to events implied in a phylogeny, the vertical axis is not linearly related to time. The hypothesis in the phylogeny on the left can be interpreted as follows: The divergence between species C and the lineage which produced species A and B came before the divergence between species A and B.

Examine the center phylogeny and the right phylogeny. Are they both different from the left phylogeny? (To answer, state the hypothesis described in each.)

Based on your answer, what can you say about the horizontal ordering of species in a phylogeny?

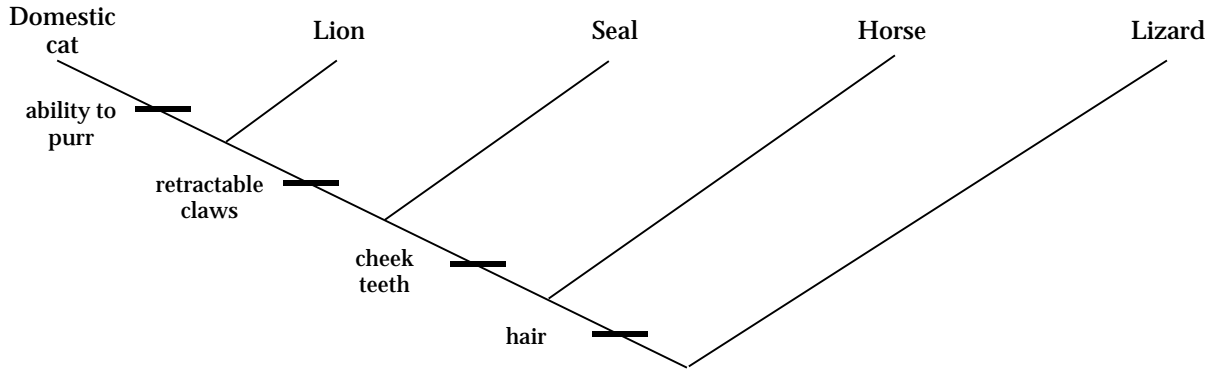
### Building phylogenies

In order to form a hypothesis about the relatedness between species A, B, and C, we need to have some measured basis for attributing relationships among them. You will learn two strategies, phenetics, and cladistics.

*Phenetics*

Phenetics is a method for organizing species based on shared similarities. Species that are similar are grouped together and species that are different are not grouped together. Similarities are measured by comparing phenotypes. Phenotypes are measured (morphology, DNA sequence, physiological processes, development pattern), then the differences among phenotypes in different species are compared. Pairs of species that have only small differences between them are grouped together.

Below is a figure that illustrates a phylogeny based on this phenetic method of attributing relationships among species.



**Figure 2.** Modified from Campbell, N. A. 1996. *Biology*. 4th ed. Benjamin/Cummings Publishing Co., Inc., Menlo Park, California. p. 476.

**Phenetic exercise 1**

Examine five leaves from each of six species of trees. Choose three phenotypes that can be measured on each leaf, and that can be identified as present or absent. Use only phenotypes that are either always present or always absent within a species.

Record your measures here. Indicate a "+" for present and a "-" for absent.

Species (with sketch of leaf)	Character		
A			
B			
C			
D			
E			
F			

<-- write character names here

Estimate the number of shared similarities (shared "+" or shared "-"). Do not fill in the cells on or above the main diagonal.

	A	B	C	D	E	F
A						
B						
C						
D						
E						
F						

Create a phylogeny that groups the species with the most similarities together. If your phylogeny has unresolved nodes, repeat the exercise adding a fourth phenotype. Note that you may be able to create more than one phylogeny that is consistent with your data, but for now, draw only one.

### Phenetic exercise 2

Identifying characters by presence or absence prevents you from including characters that vary quantitatively from one species to another. Yet much variation, as you well know, is quantitative. In this exercise you will use a method for assessing quantitative similarities and differences among your six species.

Reexamine your five leaves from each of six species of trees. Choose three phenotypes that are present on all species and that can be quantified. Measure those phenotypes, and calculate an average phenotype for each species.

Species (with sketch of leaf)	Character					
	1		2		3	
	indiv.	avg.	indiv.	avg.	indiv.	avg.
A						
B						
C						
D						
E						
F						

Calculate the proportion of difference between species pairs for each character separately, then add up the total difference between species pairs by adding the proportional differences for all three characters. As before, do not fill in the cells on or above the main diagonal.

Character 1: \_\_\_\_\_

	A	B	C	D	E	F
A						
B						
C						
D						
E						
F						

Character 2: \_\_\_\_\_

	A	B	C	D	E	F
A						
B						
C						
D						
E						
F						

Character 3: \_\_\_\_\_

	A	B	C	D	E	F
A						
B						
C						
D						
E						
F						

Total difference

	A	B	C	D	E	F
A						
B						
C						
D						
E						
F						

Create a phylogeny that groups the species with the most similarities together. If your phylogeny has unresolved nodes, repeat the exercise adding a fourth phenotype.

Is this phylogeny the same as the one you drew in exercise 1? If not, how is it different? Why?

#### Cladistics

The use of phenetics was widespread for determining the taxonomical relationships among organisms until researchers recognized that two species can share a trait even if they are not closely related. Similarities between unrelated species can occur for at least two broad classes of reasons. First, two unrelated species may have the

same character due to *convergent evolution*. Birds, bats, and insects all have wings, but they are not each others' closest relatives in a phylogeny that includes crocodiles and squirrels. They have converged on the phenotype by independent evolution, though the selection pressures may have been similar.

A second reason that two unrelated species may have the same character is because the character is *unchanged in those two species since they diverged* from a common ancestor. For example, humans have eyes and salmon have eyes, but blind snakes do not. Yet in a phylogeny that includes humans, fish, and blind snakes, humans and snakes are each others' closest relative. The similarity between fish and humans is due to sharing an *ancestral character*, one that existed in the common ancestor to fish and humans. In the snake lineage, blindness is a *derived character*.

As a result of these two spurious sources of similarity between species, researchers have refined the method with which they build phylogenies. They now restrict the characters that can be used to infer similarity to *shared, derived characters*. In other words, the only characters that can be used are the characters that two species have in common because they evolved that characters to a state different from the state in the common ancestor. For example, the ancestor to fish, snakes, and humans was aquatic, but snakes and humans are terrestrial. Terrestriality is a shared character between snakes and humans that is in a state that is *derived* relative to the ancestral state.

The key to being able to determine whether shared similarities between pairs of species are derived or not depends on knowing what the ancestral state is. To do this, we use an outgroup. An outgroup is a species that we know is more distantly related to all the species we are studying than they are to each other. For example, if we are building a phylogeny of apes, an old-world monkey would serve well as an outgroup.

The outgroup is helpful because it identifies characters that are similar among species due to common ancestry. If two species that we are studying have the same character, and our outgroup species also has a character, that we can identify that character as ancestral, and not derived. The only characters that are usable for inferring similarity among species are those that are similar in two species of interest but different from the outgroup. These are shared derived characters.

### Cladistics exercise 1

When you are ready for this exercise, come to me to find out which of your six tree species should be the outgroup.

Transcribe your data from page 2 here. Identify the outgroup. For the remaining four species, circle the presences or absences of characters that are derived relative to the outgroup.

Species (with sketch of leaf)	Character		
A			
B			
C			
D			
E			
F			

Use thonly the shared derived characters to calculate the number of shared similarities among the species.

	A	B	C	D	E	F
A						
B						
C						
D						
E						
F						

Create a new phylogeny that groups the species with the most shared derived similarities together. Make the outgroup species more distantly related to all the other species than they are to each other. If your phylogeny has unresolved nodes, repeat the exercise adding a fourth phenotype.

Is this phylogeny the same as your first phylogeny? If so how? If not, why not?