PEER MENTORING IN BIOLOGY AT NEW MEXICO STATE UNIVERSITY: A PHYLOGENETIC TREE ACTIVITY

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Peer leaders are utilized in the first semester "biology for science majors" course at New Mexico State University. This course has two 50-minute lectures per week, plus one 75-minute mandatory peer-led workshop. The workshop activities address the materials covered in the lectures and are intended to give the students hands-on experience with the subject. This course is called "Natural History of Life," and, among other topics, covers reading phylogenetic trees. What follows is a review of the lecture material and the activity that are provided to the peer leaders to help students understand building and interpreting a phylogenetic tree.

The students have attended an introductory lecture on understanding phylogenetic trees before undertaking this activity. In this lecture the basic parts of a phylogenetic tree are introduced (taxa, branches, root, nodes, common ancestors, and evolutionary transitions). Students learn to read phylogenetic trees by tracing branches, relationships of taxa that trace closer to the root are less closely related than relationships nodes that are closer to the tips of the branches.

In this workshop we are working on how to build and interpret a phylogenetic tree. A phylogenetic tree is a hypothesis about the evolutionary relationships between groups (in our case these groups are species of animals). We are using a representation of the evolutionary relationships of animals called a cladogram. See an example of a cladogram in FIGURE 1. **The points of connection between the branches are called nodes.**
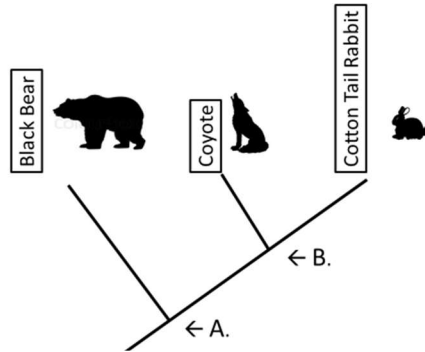
FIGURE 1: CLADOGRAM OF A BLACK BEAR, A COYOTE, AND A COTTONTAIL RABBIT. The nodes A and B are indicated.

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To interpret this cladogram, trace the branches of the tree from an animal species along the top of the diagram down towards the bottom of the tree. Trace into the tree until you join the branch that connects to the other animal species that you would like to estimate the evolutionary divergence.

When you are comparing different animal species evolutionary relationships, pairs of animals that join at nodes closer to the top of the tree are more closely related and pairs of animals that join at nodes further down in the tree are less closely related. The Cottontail Rabbit branch and the Black Bear branch meet at node A. The Cottontail Rabbit branch and the Coyote branch meet a node B. Since node B is closer to the top of the figure than node A, Cottontail Rabbit branch and the Coyote are more closely related than Cottontail Rabbit and the Black Bear.

An important corollary is that the Black Bear is equally related to the Coyote and the Cottontail Rabbit as to trace from the Black Bear to any other animal in the tree you must go through node A.

Nodes indicate evolutionary transitions that occurred in the ancestor to the species above the node. You can identify the evolutionary transition at the node, and all the species above that node will have that character. Also, each species above the node have unique characteristics that define that branch (see FIGURE 2).
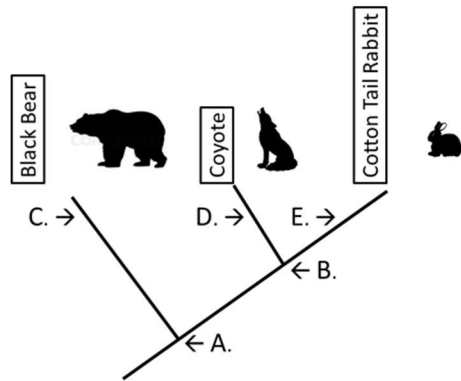
FIGURE 2: CLADOGRAM OF A BLACK BEAR, A COYOTE, AND A COTTONTAIL RABBIT. The evolutionary transitions at nodes A and B, and at branches C, D, and E are indicated. All animals above node A have four legs, all animals above node B have pointed ears. Animal that are on the branch labeled C have a large body size, on the branch labeled D have a non-retractable claws, and animal that are on the branch labeled E have obligatory nasal breathing.

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Taxa can be added to cladograms: If you are interested in additional species relationships, they can be added to the cladogram. The characteristics of the new species is considered, and the new species is added. For example, we know that Jackrabbits share many characteristics with Cottontail Rabbits. In particular, both species of rabbit have four legs, pointed ears, and obligatory nasal breathing. Notice how the addition of one more rabbit species does not change the relationships of the other species on the graph.
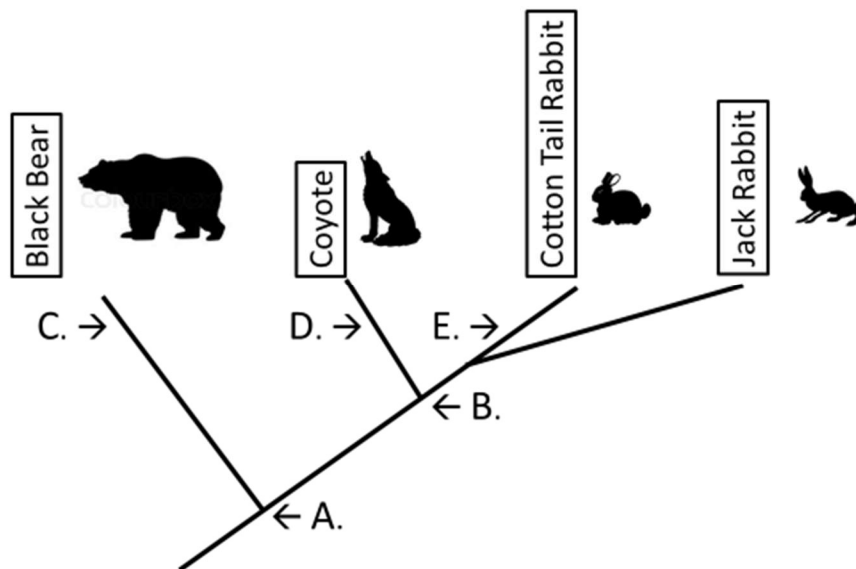
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FIGURE 3: CLADOGRAM OF A BLACK BEAR, A COYOTE, A COTTONTAIL RABBIT AND A JACK RABBIT.

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Data to build Cladogram:

In order to build a phylogenetic tree, on must tabulate the differences between the species of interest. A table of the relevant species and characters that may be similar or different is constructed.

We are going to consider the evolutionary transitions at nodes. In our Black Bear, Coyote and Cottontail Rabbit comparison we will consider the traits four legs and pointed ears. We can code each species with a "0" if the character is not present and a "1' if the character is present (TABLE 1). If there is no information about the trait, you may also enter a "?".

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TABLE 1, **Coded characteristics of bears, coyotes, or rabbits.**

	Black Bear	Coyote	Cottontail Rabbit
4 legs	1	1	1
pointed ears	0	1	1

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Considering the data from TABLE 1, a cladogram is built by grouping more similar species with shorter branches on the tree. Because Coyotes and Cottontail Rabbit share more traits (4 legs and pointed ears), they have shorter branch lengths that either to the Black Bear. Compare the data on TABLE 1 and images on FIGURES 1, 2, and 3.

1. Build your own Cladogram – collecting data:

Use the information provided in the species descriptions below to develop your own data set on the species and traits in TABLE 2.

Sharks: A shark is a vertebrate animal (an animal with a back-bone!), but that skeleton is made of cartilage rather than bone. When sharks produce eggs the eggs have no amnion or shell. Sharks have 8 fins rather than 4 limbs. Sharks do not have feathers or hair. Sharks can detect movement in water from great distances.

Trout: A trout is a species of fish with boney vertebrae. This animal has fins rather than limbs. Fish do not have hair or feathers. Females produce a large number of eggs that have no amnion or shell. Eggs are fertilized by males externally.

**Toad:** Toads are animals that have four limbs, a boney skeletal structure, and a backbone (vertebrae). Their reproduction is tied to an aquatic environment because their eggs are gelatinous rather than shelled, and there is no amnion. Their skin is porous and can exchange gasses, there is no hair or feathers.

Human: Humans have two arms and two legs (four limbs). The upright stature is supported by boney vertebrae. Humans produce an amniotic egg, but the eggs do not have shells. Humans have hair but not feathers.

Tiger: Tigers are large carnivorous animals. They are hairy animals that produce amniotic eggs without shells. Their strong bony vertebrae give them structural support. They run quickly on 4 limbs. Tigers do not have feathers. Tigers are an endangered species, there are probably less than 6,000 animals left in the world.
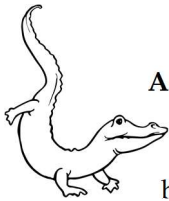
Alligator: Alligators are predatory reptiles. Their amniotic eggs have leathery shells. Their bony skeleton includes vertebrae and four limbs that are good at fast speeds in bursts. Alligators do not have feathers or hair.

Bald Eagle: The Bald Eagle is a national symbol of the United States. These birds became an endangered species in the 1900s because pesticides accumulated in ecosystems and poisoned animals at all levels of the food web. These poisons compromised the shells of the amniotic eggs, and many offspring did not survive. Birds have hind-legs and feathered fore-wings (thus 4 limbs), but do not have hair. Birds have many bones that are hollow, which makes them light and able to fly. Birds have more vertebrae that most animals, which allows for a flexible neck.

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**TABLE 2: CODED TRAITS OF VERTEBRATE ANIMALS.** Code traits with a "1" if present and a "0" if absent.

	Human	Trout	Bald Eagle	Alligator	Toad	Shark	Tiger
vertebrae							
boney skeleton							
four limbs							
amniotic egg							
eggs with shells							
hair							
feathers							

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2. Build your own Cladogram – tracing evolutionary transitions:

Consider the data from TABLE 2 to fill in the species on the cladogram in FIGURE 4. The taxa humans and the trait feathers have been identified on this tree to help you. Follow these directions to build your tree.

2.1 Find the *species* that share the most common traits with humans. Label that species on the tree that represents the placement of the closest related species.

2.2 Identify the *trait* that is unique to humans and the species that is closest related to humans and place that *trait* on the branch leading to those two taxa.

2.3 Identify the two *species* that are left and share the most common traits with the human group. Place them in the boxes for taxa at the top of the tree.

2.4 Transcribe the evolutionary transitions that represent those branches on the horizontal lines beside branch.

2.5 Work your way down the major branch of the tree, identifying less closely related taxa and transitionary traits as you go.

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FIGURE 4: **CLADOGRAM OF SOME ANIMAL TAXA.**
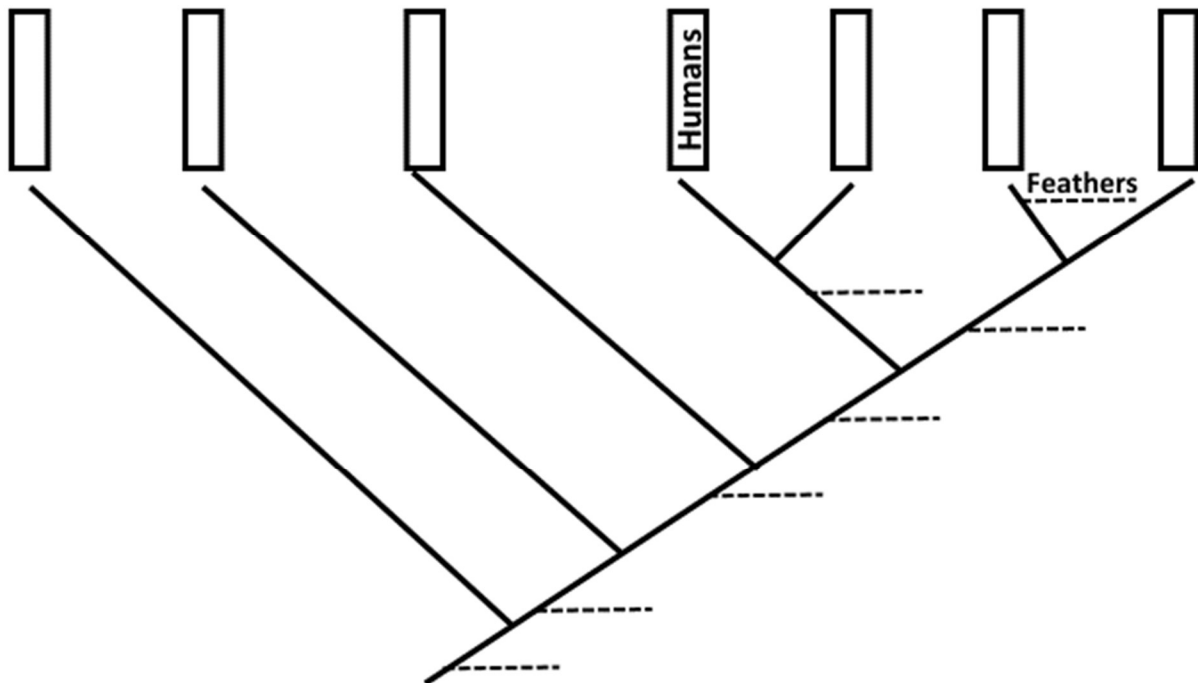
When you are finished, be certain that each taxa box along the top of the tree and each evolutionary transition along the branched are filled in. (6 points, 0.5 point each)

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2.3 Every workshop group should draw their hypothesis of the relationships on the whiteboards. Walk around and look at the other trees. You should take your worksheet papers with you and discuss any differences. Everybody should have the same hypothesis before the workshop continues. Have your Peer Mentor confirm your participation. *(note, if you do not get this right, you may not get the rest of the answers in this workshop correct).*

	2 points
Participation	

3. Interpretation: Please discuss the phylogenetic trees at your table to get these concepts correct before you leave workshop today. If in doubt, ask your Peer Mentor. (1 point each)

3.1 What species is more closely related to Humans?

      A. Alligators

      B. Toads

3.2 What trait do Toads and Bald Eagles have in common, but are not shared by Trout?

3.3 Name all the species that have a common ancestor that had the trait hair.

3.4 Name all the species that have a common ancestor that had the trait 4 limbs.

3.5 List the species that do not have four limbs.

3.6 Which is most closely related- Tigers and Alligators or Tigers and Toads?

      A. Tigers and Alligators

      B. Tigers and Toads

**3.7** What trait do all species on the cladogram have in common?

3.8 Is there a species that is more advanced on this tree? Explain your answer.

PHYLOGENETIC TREE ACTIVITY KEY.

TABLE 2: CODED TRAITS OF VERTEBRATE ANIMALS. Code traits with a "1" if present and a "0" if absent.

	Human	Trout	Bald Eagle	Alligator	Toad	Shark	Tiger
vertebrae	1	1	1	1	1	1	1
boney skeleton	1	1	1	1	1	0	1
four limbs	1	0	1	1	1	0	1
amniotic egg	1	0	1	1	0	0	0
eggs with shells	0	0	1	1	0	0	0
hair	1	0	0	0	0	0	1
feathers	0	0	1	0	0	0	0

(4 points)
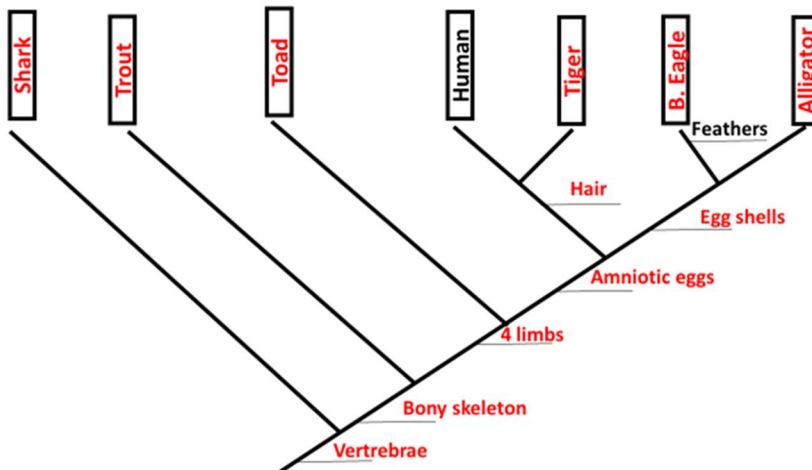
(6 points, 0.5 point each)

Participation in drawing trees on whiteboards (2 points).

3.1 *A. Alligators* (1 point)

3.2 *4 limbs* (1 point)

3.3 *humans and tigers* (1 point)

3.4 *frog, human, tiger, alligator, eagle* (1 point)

3.5 *shark, trout* (1 point)

3.6   *A. Tigers and Alligators* (1 point)

3.7 *vertebrae* (1 point)

3.8 *NO! There is no direction to evolution. One species is not better than another.* (1 point)

Note: All illustrations were either drawn by the first author or downloaded from openclipart.com (open access).