

Molecular Biology and Primate Phylogenetics

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Target age or ability group: 9-14 biology, molecular biology, or interdisciplinary evolution courses

Class time: Two 50 minute class periods. Day 1: Students work on counting amino acid differences between each pair of organisms and complete table #1, compare gross anatomy of selected primates from reference materials supplied by the teacher, and then begin the discussion questions and identification of organisms on the graph. Students then take home the activity to complete the discussion questions and the graph. On Day 2 the students meet in teams to discuss/debate their responses and the labeling of the graph, and then participate in a class discussion guided by the teacher.

Materials and Equipment: 4 page student handout for each student and an overhead of pages 2 and 3 of the four pages of the activity for the teacher to use during review. It is also recommended that the teacher provide references comparing the anatomy of the primates mentioned in this activity (see page 3 of the student handout, question #3 for the list), a geologic time chart, and any phylogenetic tree that will serve as a good reminder to the students that they are discovering relationships that emphasize ancestry.

Summary or Overview of Activity: Working in groups of two, students begin by determining the number of amino acid differences in beta hemoglobin that occur between each of the pairs of organisms featured in the activity. The students do not know the names of the organisms studied until after they complete the amino acid comparisons and have begun the discussion. Students compare each possible pairing of organisms, looking for differences in the sequences, and discovering patterns in the degree of difference. The current view is that the number of differences for a particular molecule is an indicator of how long ago in the past any two organisms may have shared a common ancestor. By example, the relatively few differences between the beta hemoglobin of humans and gorillas is an indicator of a relatively recent common ancestor, whereas, a comparison of chimpanzee and lemur beta hemoglobin indicates a far more distant common ancestor. More striking, but less intuitive, is the observation that chimps, humans, and even monkeys are all approximately equidistant from lemurs. This is because the common ancestor of all anthropoidea diverged from the common ancestor of lemurs at the same time, a very long time ago. Students use the data in Table 1, summarized in Table #2, to infer relative points of divergence since a common ancestor and, upon comparing gross anatomy to amino acid sequences for each of the 7 species, infer where each of the seven species belongs on the phylogenetic tree.

Prior knowledge, Concepts, or Vocabulary Necessary to Complete Activity: Students must have a good working knowledge of the structure and function of DNA. They must also be aware of the growing body of evidence that supports the scientific conclusion that life has a long history and has diversified widely through time. Students should have some exposure to phylogenetic trees and sequences in the fossil record.

Teacher Instructions: Begin by showing several overheads of phylogenetic trees (Strickberger, Evolution is an excellent source). Students will be most interested in phylogenetic trees for dinosaurs and other prehistoric organisms that are now extinct. Ask students to offer an explanation as to how scientists establish such evolutionary relationships. Most of their responses will likely focus on anatomical comparisons and fossil sequences. The teacher should then point

out that living organisms also share common ancestry and that these ancestral relationships can be understood, in addition, by using molecular comparisons. The teacher might then put up an overhead (non-phylogenetic, merely comparative anatomy) which includes at least several of the primates mentioned in this activity and ask students why we group these organisms together. Most students will respond that these creatures share certain physical characteristics, especially a set of characteristics that they share with no other organisms classified outside of the order primates. Then ask the students why they share characteristics common to all vertebrates (ancestral characters - distant common ancestors) and characteristics common only to the order primates (derived characters - recent common ancestors). Here the teacher could talk about general patterns in the fossil record. The teacher should then pass out the 4 page student lab sheet along with a book or photocopies of the primates that are used in the activity. Working in 2 groups of 2, each lab station team will proceed through the activity. Each pair should work independently of the other pair, except when they hit a significant problem. The teacher should remind students that what they do not finish in class will need to be done by the student at home. On day 2 the teacher lets the students meet in their 4 person lab teams to discuss their answers. There will no doubt be loud debate and discussion. Allow 20 minutes. Students then present their findings in a discussion facilitated by the teacher (allow 30 minutes). The teacher should have overhead copies of pages 2 and 3 of the activity so that as answers are developed, all students can see the data and graph,

Answers to Table #1, Graph #1 and Discussion:

In Table #2: the row answers should read
 row A: 0,1,2,8,11,30 (19 in column H is actual),
 row B: 1,2,8,11,30 (20 in column H is actual)
 row C: 3,8,12,30 (20 in column H is actual)
 row D: 7,10,30 (18 in column H is actual)
 row E: 13, 28 (18 in column H is actual)
 row F: 30 (17 in column H is actual)
 row G: (27 in column H is actual)

In Graph #1: the "letter" order is G,C,B,A,D,E,F, and the "name of organism" sequence is lemur, gorilla, chimpanzee, human, gibbon, rhesus monkey, squirrel monkey, and horse (for H). To scale the lower x axis as required in discussion question #9 merely draw a dotted line down from the first vertical split on the graph (at about 29 amino acid dissimilarities) to the lower axis and label that "43"(43 million years ago). Then mark the spot on the lower axis directly below the "0 dissimilarities" from the upper axis as "6"(6 million years ago). $43 - 6 = 37$ million years from left to right boundary so each 10 units of amino acid dissimilarity is approximately equivalent to 12 million years. Merely subdivide so that 1 amino acid dissimilarity is equal to about 1.2 million years of elapsed time. Obviously your scale can be extended to the left beyond the 43 million year mark.

For the Discussion: 1. A and B 2. A and B 3. See above under Graph #1

4. All organisms A-F share a common ancestor with G at the same point in time. Using comparative anatomy to develop phylogenetic trees can be misleading, especially when we have a tendency to want to create hierarchical relationships (like a ladder, with the "best" or "most advanced" at the top). This is a typical and difficult error to overcome. If we place these organisms hierarchically, then we are inferring, in fact, that one type of creature evolved into the next which is a significant error. These organisms are all alive today. The difficulty is in determining when they diverged from a common ancestor. Remember: No scientist has ever said that humans evolved from modern day monkeys. What trained scientists have been saying is that humans and monkeys

(and all other life on earth in fact) share a common ancestor somewhere in the distant past. By analogy: You are related to your cousin Donald or Sally, but you are not descended from them.

5. Typically, students will see that horses are more distantly related (they are mammals, but not primates) and predict that the last column in Table #2 be about high-30s or low-40s. The line of descent would be drawn from the right side of the graph all the way to the left and connect somewhere on the single line of descent that enters from the left.

6. It suggests that humans and chimpanzees share a more recent common ancestor than do horses and zebras. Even though horses and zebras "look" more similar, looks can be deceiving. Check a phylogenetic tree for the "horse" in any good college evolution text. The reason that they look so similar even though a longer period of time has passed since they (as opposed to humans and chimpanzees) shared a common ancestor is that selective pressure has favored a similar body plan for a nearly identical feeding pattern. Both are swift, herding herbivores of dry grasslands.

7. Typically, students will say they were too high in their estimate. They will be confused that organisms A-F are apparently more closely related to a horse than to another primate, the lemur. They should question the validity of the data, but the teacher should comment that the data is accurate. They should then be questioned if this single piece of aberrant data is enough to shake confidence in all of the molecular analysis that has been done. Clearly, one piece of unusual data begs explanation, not total rejection of a fruitful technique of great value.

8. This is perhaps the most difficult of the 10 questions. Students will be expected to draw on their understanding of the structure and function of DNA, RNA, and proteins. The teacher is needed here to help guide the students to a logical plan of investigation that involves examining DNA and RNA nucleotides to investigate whether or not a back mutation has occurred in a triplet that would cause the amino acid product to be the same as a primate in its sequence. This can be best understood by showing the table of amino acid triplet codes. Some amino acids have 1 triplet (methionine which is a start codon) and several that have 6 different triplet codes. The giveaway would be if the organism had the same amino acid (when you might predict it should be different) but has a distinctly different triplet. Here it could be inferred that the organism has more similarities because of one or more chance mutations that changed the amino acid to the same as a long distant evolutionary cousin.

9. The rate is about 1 amino acid mutation per 1.2 million years according to this graph. This rate has little to do with experimentally determined rates. For one, there is a difference between a meaningful rate (changes in DNA that produce a noticeable affect such as a change in the type of amino acid encoded) and there is also a silent rate in which changes in the DNA may have no effect on the final amino acid because it has multiple triplet codons. Both silent and meaningful base changes are being studied. You might want to refer to research being done by Li, Nei, Roychoudhury, or the late Motoo Kimura for additional information.

10. No they do not. Chimpanzees and squirrel monkeys last shared a common ancestor approximately 23 mya according to this graph.