Evidence of Evolution: Homologous Gene Expression in Four Mammal Species
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Introduction

A bat, a pig, and a mouse enter a classroom...

The bat flies, the pig walks in on two digits (fingers or toes) per limb, the mouse on five. But despite the differences, the limbs of all these animals are much more alike than they might seem. That's because they are homologous, meaning that their origin can be traced back to a common ancestor. You have probably learned that the common ancestor of the bat, pig, and mouse had limbs with a certain arrangement of bones and passed the trait on to its descendants, but have you ever thought about just how the limb trait was passed from one generation to the next? In the form of genes! So not only are the limbs of these animals homologous, but the genes underlie limb development in each animal are homologous, too. In this lesson, you will learn that changes in a small number of genes are actually responsible for the diversity and differences of mammalian forelimbs observed in nature.

What To Do

Complete the analysis questions, reading the Bite when instructed.

Analysis Questions

The forearms of a bat, pig, mouse, and opossum are shown in Figure 1.

![Figure 1. Forearms of Four Mammals.](image)

1. Complete the table below by counting the number of bones in each species.

<table>
<thead>
<tr>
<th></th>
<th>Bat</th>
<th>Pig</th>
<th>Mouse</th>
<th>Opossum</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number of bones</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>between shoulder and</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>elbow (dark purple)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Number of bones</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>between elbow and wrist</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>(light purple and yellow)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Approximate number of</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>bones in wrist and</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>hand (orange)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
A phylogenetic tree is shown in Figure 2. It models the evolutionary relationships among four mammals: a bat, pig, mouse, and opossum. Nodes, or branching points (shown as small circles) represent common ancestors. For example, the common ancestor of bats and pigs is represented by the node labeled C.

Figure 2. Phylogeny and Limbs of Four Mammals.

2. Use Figure 2 to answer the following questions.
   a. Which node in Figure 2 (A, B, or C) represents the common ancestor of bats, pigs, and mice?

   b. Make a claim about how many upper arm bones (dark purple) and forearm bones (yellow and light purple) the ancestor represented at node A had.

   c. Explain, using evolutionary concepts, why you made the claim you did in Part b.

   d. Make a claim about when the gene, or genes, that underlie the general forearm structure you described in part b evolved. To make your claim, place an “X” on the diagram in Figure 2 to represent when the gene(s) evolved.
Two claims about homologous traits are below.

**Claim 1:** The genes underlying forearm structure in mammals are homologous.

**Claim 2:** The genes underlying forearm structure in mammals are not homologous.

e. Which claim, claim 1 or claim 2, do you support? Explain your choice with evidence from Figure 2 and your answer part d.

3. Although they clearly share a general overall pattern, the forearms of these animals are not identical.

a. Propose a hypothesis for how the differences you see among the structures of the different bones in each mammal could have arisen.

b. Make a claim about which of the four species (bat, mouse, pig, opossum), if any, is the most advanced. Defend your claim using evolutionary reasoning.

STOP & read Science Bite:

**What Do a Bat, a Pig, a Mouse, and an Opossum Have In Common?**

Genes are transcribed into mRNA. Looking at mRNA can tell us something about the function of a gene because we assume that the mRNA will be translated into a protein. Scientists have ways of actually seeing where a particular mRNA is located in a living embryo. The researchers you read about were interested in where and when a specific “toolkit” gene, *hoxd13*, was being expressed (turned into mRNA and eventually into a protein) in a developing limb. They added a dye to the embryos that would show up as purple anywhere the *hoxd13* mRNA was found. The result is shown in Figure 4.
4. Use Figure 4 to answer the following questions.

a. How does the expression of Hoxd13 change from the ridge stage to the paddle stage in the mouse? Is it a similar pattern compared to the bat and the opossum?

b. Which developmental stage (ridge, bud, or paddle) is the expression of Hoxd13 most similar across species? Which is the most different?

c. Look at the forelimb structure of the bat, mouse, and opossum shown in Figure 1 in the Bite.

- Which part of the forelimb is most similar between species?
- Which is the least similar?
- How does this pattern of gene expression across species relate to the bone structure across species?
d. Do you think that the gene expression patterns (Figure 4) and anatomical structure of the limb (Figure 1) are two different lines of evidence for evolution, or are they both part of a single line of evidence? Explain your reasoning.

5. Connect to the Big Question. Scientists rely on all kinds of evidence to help them reconstruct evolutionary relationships. Before the discovery of DNA and molecular tools, scientists like Charles Darwin heavily relied on comparing and analyzing anatomical structures. Do you think one form of evidence is more convincing than the other? Why?