

A response to charges of error in *Biology* by Miller & Levine

According to TEA, a citizen disputes two sentences on page 767 of our textbook, *Biology*, by Miller & Levine. These sentences are:

SE 767, par. 2, lines 8-9 – "Recent DNA analyses confirm that, among the great apes, chimpanzees are humans' closest relatives."
SE 767, par. 4, lines 1-2 – "The brains of chimpanzees, our closest living relatives,"

The changes suggested are these:

Revise SE 767, par. 2, lines 8-9, thus – "Recent DNA analyses ~~confirm that, among the great apes, chimpanzees are humans' closest relatives~~ suggest no consistent pattern of evolutionary relationships among these hominoids."
Revise SE 767, par. 4, lines 1-2, to read – "The brains of chimpanzees, ~~our closest living relatives,~~ one of the hominoids,"

In support of these suggested changes, copies of five scientific papers have been provided, along with seven objections to the existing wording.

We thank the unnamed citizen for his or her interest in our textbook, and are delighted to respond to these suggestions. We begin our response with a summary of each of the five papers ("technical monographs") provided to support the accusation of "errors" in our text.

1) Recent research strongly supports the conclusion that chimpanzees are humans' closest living relatives.

Significantly, none of the five technical monographs provides any evidence disputing the conclusion that chimpanzees are humans' closest living relatives. Taking these in order, with the oldest first, here is a summary of each paper.

Watanabe *et al* (2004) DNA sequence and comparative analysis of chimpanzee chromosome 22. *Nature* 429: 382-388.

This study compared the two smallest chromosomes of the two species (human #21 and chimpanzee #22) and appeared a year before the complete chimpanzee genome was sequenced. As such, it examined only a small section of the genome of both species, and therefore its conclusions are limited. The study compared corresponding genetic regions on the two chromosomes, and reported on the specific nature of differences between them in terms of insertions, deletions, and single-nucleotide substitutions. While the authors note that the biological consequences of these genetic differences "are more complicated than previously speculated," their studies did not dispute the uniquely close relationship between humans and chimpanzees at the genetic level.

Significantly, when the complete genome sequence was published a year later in the same journal (Mikkelsen *et al* [2005] Initial sequence of the chimpanzee genome and comparison with the human genome. *Nature* 437: 69-87) the results were very clear. As the authors wrote, “Modern molecular studies have spectacularly confirmed this prediction [*of common ancestry*] and have refined the relationships, showing that the common chimpanzee (*Pan troglodytes*) and bonobo (*Pan paniscus* or pygmy chimpanzee) are our closest living evolutionary relatives.”

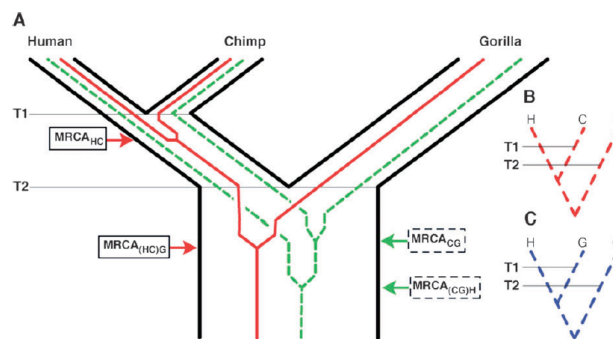
Ebersberger *et al* (2007) Mapping Human Genetic ancestry. *Mol. Biol. Evol.* 24: 2266-2276.

This study compared 23,210 DNA sequence alignments from five species (human, chimpanzee, gorilla, orangutan, and rhesus). Highlighted in this paper (by the citizen who alleges errors in our textbook) are passages noting that particular regions of the human genome, when taken in isolation, give conflicting results with respect to evolutionary relationships. As a result, according to the authors, “for about 23% of our genome, we share no immediate genetic ancestry with our closest living relative, the chimpanzee.”

The paper summarizes its results like this:

“Thus, in two-thirds of the cases [i.e., phylogenies], a genealogy results in which humans and chimpanzees are not each other's closest genetic relatives. The corresponding genealogies are incongruent with the species tree. In concordance with the experimental evidences, this implies that there is no such thing as a unique evolutionary history of the human genome. Rather, it resembles a patchwork of individual regions following their own genealogy”

However, this does not mean that the relationship of humans to chimpanzees has been called into question. The authors themselves explain this in a chart of gene lineage in Figure 1 of the paper:

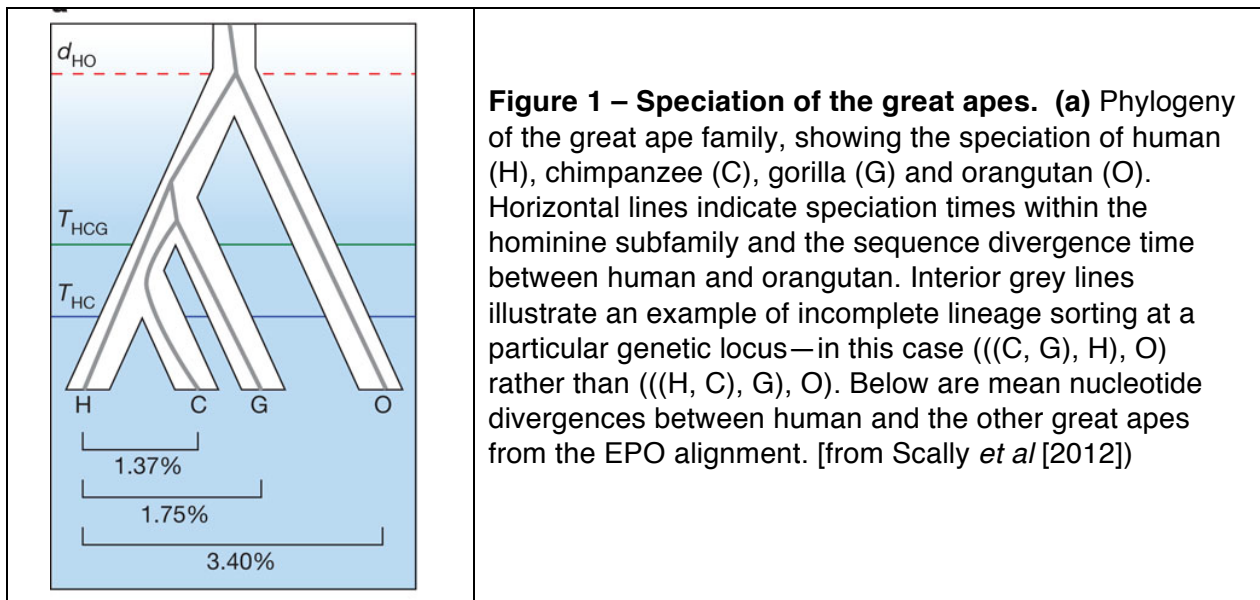


What this figure shows is how the *evolutionary histories* of certain genes can differ from the *actual histories* of the species themselves. Specifically, the authors identify three possible pathways that two genes might take, and show that for two of them, the human gene would not be most closely related to the chimpanzee gene. This is the point of this study, and it confirms

the closest relationship among the five species studied is between humans and chimpanzees. It is worth noting that the authors of the Ebersberger *et al* (2007) consistently maintain that the evidence shows that the most recent split in this lineage was the human-chimpanzee split, just as our textbook notes. In fact, even the passage highlighted in the paper’s abstract notes that our “closest living relative” is the chimpanzee. Therefore, this paper supports and confirms the language in our textbook in every respect.

It is important to note that the Ebersberger *et al* (2007) study was done before a complete sequence of the gorilla genome was completed. They point this out on page 2274 of their paper: “From this perspective, an extensive sequencing of the gorilla genome will be required to make full use of the chimpanzee genome sequence on the way toward a map of our genetic ancestry.” That more complete sequence is now available, and was analyzed in a study reported last year (Sally *et al*, “Insights into hominid evolution from the gorilla genome sequence,” *Nature* 483: 169-175).

What was their conclusion? Sally *et al* (2012) noted “In 30% of the genome, gorilla is closer to human or chimpanzee than the latter are to each other.” This, of course, is the directly result of ILS. However, when the much larger database used for this study is used to compare humans, chimpanzees, and gorillas, the data are very clear, as summarized in Figure 1 of the paper and its associated caption:



Given these data and their exhaustive analysis by an international team of scientists, it would be highly misleading to tell students that there is “no consistent pattern” or relationships among these organisms. The statement in our textbook, that “recent DNA analyses confirm that, among the great apes, chimpanzees are humans’ closest relatives” is correct and consistent with the most extensive genetic study of the three species to date.

Hughes *et al* (2012) Chimpanzee and human Y chromosomes are remarkably divergent in structure and gene content. *Nature* 463: 536-539.

Like the Watanabe *et al* (2004) study, this paper reports on a detailed investigation of one portion (the MSY region) of the smallest chromosomes, the Y chromosome, in humans and chimpanzees. It does not compare the complete genomes of the two species, as the Mikkelsen *et al* (2005) study did. In fact, the actual size of the region studied was just 25.8 megabases (25.8 million DNA bases). This is less than one percent of the total genome of either species.

The researchers found substantial differences in the MSY regions of the two species, which they attribute to a variety of evolutionary forces operating since the separation of the two lineages. They also indicate that the results in this small region are not representative of the state of the two genomes in their entirety: “We conclude that, since the separation of the chimpanzee and human lineages, sequence gain and loss have been far more concentrated in the MSY than in the balance of the genome.”

This study provides absolutely no evidence to dispute the conclusions in our textbook, and actually refers to the chimpanzee as “our closest living relative.” Therefore, it strongly supports our textbook.

Cabili *et al* (2011) Integrative annotation of human large intergenic noncoding RNAs reveals global properties and specific subclasses. *Genes & Development* 25: 1915-1927.

This interesting study reports on an analysis of 8,195 long, non-coding RNAs (lincRNAs) generated from a variety of human cell and tissue types. The individual lincRNAs were characterized as to expression level, tissue specificity, and genomic location. While the authors speculate that these non-coding RNAs may play important roles in gene expression, they did not identify a single functional role for any of these sequences: “Future work will be necessary to identify RNA sequence domains that relate to function..” In addition, none of the data presented in this paper relate to differences or similarities between human and chimpanzee sequences.

Therefore, this study is simply not relevant to the relationship between the human and chimpanzee genomes.

Hangauer, Vaughn, & McManus (2013) Pervasive transcription of the human genome produces thousands of previously unidentified long intergenic noncoding RNAs. *PLOS Genetics* 9: e1003569.

Like the Cabili *et al* (2011) paper, this study reports on the pervasive transcription of the human genome and the generation of large numbers of lincRNA sequences. The key conclusion of the researchers is that “the majority of the genome is indeed transcribed,” which

is interesting and potentially important. Like the previous study, these authors have characterized the lincRNAs with respect to size, tissue-specificity, and association with known genes. However, like the previous study, no direct connection with any function has been found. The authors admit this, writing: “many of these new lincRNAs are low expressed and definitive proof of functionality for a lincRNA requires functional experiments.” They hope that such experiments will be conducted in the future, but at the present time they can refer to these regions only as the “dark matter of the human genome.”

Like the Cabili *et al* (2011) study, none of the data presented in this paper relate to differences or similarities between human and chimpanzee sequences. Therefore, this study is not relevant to the relationship between the human and chimpanzee genomes.

2) The seven specific charges leveled against our textbook are unreferenced and incorrect.

A list of seven charges appears in the document sent us from the TEA. Our response to each of these is shown below:

(1) RANK CHERRY-PICKING In comparing chimp and human genomes, commonly-used "low-complexity sequence masking" excludes many non-aligning DNA segments, understating disparities between the two genomes.

No reference or example is given of a paper or study in which “low-complexity sequence masking” was used to compare genomes, and therefore it is impossible to respond directly to this charge. In addition, when studies are carried out in ways that avoid this problem, chimpanzees are still shown to be humans’ closest living relatives. We have already cited an exhaustive study of three primate species (human, chimpanzee, and gorilla) that did not involve any sort of “cherry-picking,” namely, Scally *et al* (2012). The conclusion in this study was that chimpanzees are indeed our closest relatives.

(2) STATISTICAL SLANT Focusing only on the most similar portions of chimp and human genomes exaggerates their total actual harmony.

This would be a fair criticism if it were accurate. However, as sequencing tools and methods have improved it has become increasingly possible to examine entire genomes rather than certain segments. Once again, when this is done, chimpanzees still emerge as our closest relatives.

(3) OVERRATED SYNC The longer the DNA sequence segments compared, the lower the percentage of match-up between portions of chimp and human genomes.

This may be correct, since longer sequences typically include more opportunities for transversion, inversions, and other indels. However, no study has emerged of “longer” sequences in which any organism other than the chimpanzee has been shown to be our closest

relative. Therefore this charge does not actually address the point in question, which is the relationship between human and chimpanzee genomes.

(4) HIGH DISCREPANCY 23% of chimp and human DNA sequences show no similarity. Chimp and human Y-chromosome DNA sequences differ by over 30%, or about as much as human and chicken autosomes differ. 83% of amino acid sequences in chimp chromosome 22 differ from those in its human chromosome 21 counterpart.

The claim of 23% of DNA sequences showing “no similarity” is false. This claim seems to be based on the Ebersberger *et al* (2007) paper, but these authors made no such statement. Rather, they noted that 23% of the DNA sequences they studied “support a closer relationship of gorilla to either humans or chimpanzees, although they recover the monophyly of the 3 species.” In plain language, this means that 23% of these sequences do show a very high degree of similarity between humans and chimpanzees, although their similarity to gorilla sequences is somewhat higher. This phenomenon is explained by incomplete lineage sorting, as the authors of several studies have pointed out. The use of the term “monophyly” is especially important. It indicates that these sequences are more similar within this group of three species than they are to any other organism. The claim that they show “no similarity” is therefore completely incorrect.

The differences between chimp and human Y-chromosomes related to less than 1% of the genome (as noted earlier) and are not representative of differences in the rest of the genome, as the authors of the Y-chromosome study noted. Finally, nearly all of the “88% of amino acid sequences” differ by only one or two amino acids, making the differences insignificant for most proteins.

(5) CONFLICTING DESCENTS Biochemical phylogenies of chimps, humans, gorillas, and orangutans contradict their standard anatomical phylogeny 40% of the time.

No reference is given for this charge, so it is impossible to learn who might have determined the “standard anatomical phylogeny” in question or by what methods such phylogenies were constructed. Also, it is not clear what is meant by a “biochemical phylogeny.” Is this a comparison of protein structure, amino acid sequences, or DNA base sequences? Without such details we cannot respond to this comment.

(6) BLOATED PERCENTAGE Counting the gaps between closely aligning strands of chimp and human DNA sequences deflates the overall identity ratio between their respective genomes to 70-87%.

Once again, no reference is given to any study, research finding, or published paper. Furthermore, even if these figures are correct, they do not challenge the conclusion that chimpanzees are humans’ closest living relatives.

(7) RISKY SNAP JUDGMENT Any assured correspondence between the two

genomes is premature and arbitrary. Unacknowledged functions of now-omitted, non-aligning DNA sequence sections may revolutionize comparisons.

This charge, presumably, is why the two most recent papers were included (Cabili *et al*, 2011, and Hangauer, Vaughn, & McManus, 2013). It is certainly true, of course, that new knowledge may change our understanding of the evolutionary relationships between the great apes. However, that does not and should not prevent us from communicating the current scientific consensus on such relationships to our students. In this respect it is worth noting that the authors of both of these papers pointed out that their studies did not identify *any functions* associated with lincRNAs, and that neither paper addressed the relationship between human and chimpanzee genomes. Therefore, it is neither “premature” nor “arbitrary” to present students with the current state of scientific understanding on this relationship. Our textbook is current and accurate on this score.

3) No changes are necessary in wording, because the sentences in question are not mistaken.

As we have shown here, the current scientific evidence clearly shows that chimpanzees are humans’ closest living relatives. The first change proposed for page 767 of our textbook would mislead students by telling them that there is “no consistent pattern of evolutionary relationships among these hominoids.” There is, in fact, a consistent pattern of relationships, as increasingly detailed molecular studies have shown, and that pattern indicates the close genetic relationship between humans and chimpanzees. The second proposed change would imply that chimpanzees are just “one of the hominoids,” when in fact they are our closest relatives among these organisms.

Because we are committed to scientific accuracy and integrity, we dispute the charge of “errors” on page 767, and insist on keeping the fair, accurate, and complete descriptions current in our textbook.