

Cornell Institute for Biology Teachers

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Title: Molecular Evolving Trees

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Based on Evolving Trees by:

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Appropriate Grades 9-12 Living Environment, Advanced Biology

Level:

An understanding of transcription and translation is required for this lab.

Abstract: This lab activity is intended to be an add on to the Evolving Trees activity. It

demonstrates how changes in a genetic sequence can be used to make a cladogram. It also shows students that mutations in the DNA of a protein-coding gene can have

no, a small, or a large effect on the encoded protein.

Time Required: 30-45 minuets in addition to the Evolving Trees activity.

Special Needs: Lab handout. The original Evolving Trees Activity

Teacher Information

This activity is designed as a extension of the Evolving Trees lab. Thus, most of the background information in that lab is applicable to this one.

Answers to the student section:

Differences in DNA sequence can be used just like differences in physical characteristics to make a cladogram. Below is a collection of DNA sequence from an non-coading region of DNA from the hypothetical fly species that you just built a cladogram for. Following the steps below, build a cladogram using only the information in the DNA sequences.

1. Carefully compare the aligned sequences for mutations that have occurred since the species have diverged. It is often useful to use a marker or highlighter to mark positions where mutations have occurred.

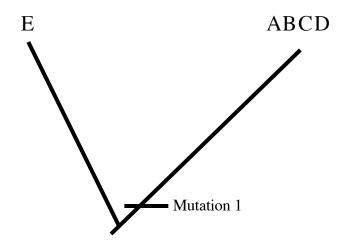
Position:		1					2			3							4					
Species A:	Α	С	С	G	С	\mathbf{T}	Α	С	G	Α	Т	Т	С	G	G	С	Т	Α	G	С	Α	\mathbf{T}
Species B:	Α	С	С	G	С	\mathbf{T}	G	С	G	Α	Т	Т	С	G	G	С	С	Α	G	С	Α	\mathbf{T}
Species C:	Α	С	С	G	С	\mathbf{T}	Α	С	G	Т	Т	Т	С	G	G	С	Т	Α	G	С	Α	\mathbf{T}
Species D:	Α	С	С	G	С	\mathbf{T}	G	С	G	Α	Т	Т	С	G	G	С	Т	Α	G	С	Α	\mathbf{T}
Species E:	Α	G	С	G	С	\mathbf{T}	G	С	G	Α	Т	Т	С	G	G	С	Т	Α	G	С	Α	\mathbf{T}

2. Fill out the chart below to arrange the data (With a + indicating that the mutation occurred).

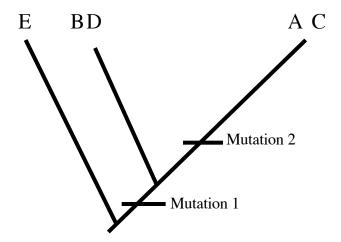
	Species A	Species B	Species C	Species D	Species E
Position 1: G to C	+	+	+	+	-
Position 2: G to A	+		+	-	-
Position 3: A to T	-	-	+	-	-
Position 4: T to C	-	+	-	-	-

Design a cladogram for using these animals to postulate relationships between the groups.

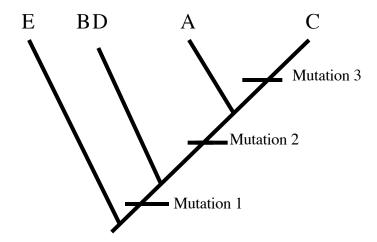
1. Mutation 1



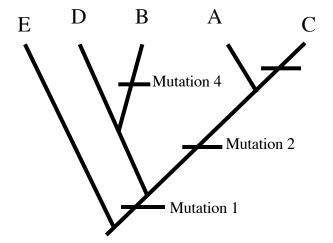
2. Mutation 2



3. Mutation 3



4. Mutation 4



How does this tree compare to the tree you made based on physical traits?
The two trees should be identical.
Which tree would you trust more, one based on physical characteristics or one based on mutations in DNA?
Ideally, both pieces of evidence should agree. If there is a disagreement between them, you should make the tree that is most parsimonious. If there's a tie, it's a good idea to look for extra pieces of information (such as additional DNA sequences or an extra physical trait).
Do all DNA mutations lead to a change in the physical characteristics of an organism?
No. Changes in non-coding sequences usually won't change the phenotype of the organism. Exceptions include mutations in promoter regions, centromeres, and other non-protein coding regions that are structurally important in DNA.

Below is a collection of DNA sequence from an protein coding region of DNA (a gene) from the hypothetical fly species that you just built a cladogram for. The sequence starts at the beginning of the gene. First, use a marker or highlighter to mark positions where mutations have occurred

```
Position:

1 2 3 4

Species A: A T G G C T G C A A T T T A C C T A G C A

Species B: A T G G C T G C G A T T T A G C T A G C A

Species C: A T G G C T G C A A T T T A C C T A G C A

Species D: A T G G C T G C G T T T T A C C T A G C A

Species E: A T G A C T G C G A T T T A C C T A G C A
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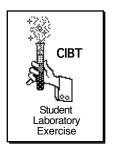
Next, use the genetic code (from the chart on the last page) to determine the protein sequence that these genes encode.

Protein Sequence:

Species	A:	A	Т	G	G	С	Т	G	С	A	A	Т	Т	Т	A	С	С	Т	A	G	С	A
Species	A:	Me	t		Al	a		A]	La		IJ	Le		Т	ŗ		L€	eu		A]	La	
Species	B:	A	Т	G	G	С	Т	G	С	G	A	Т	Т	Т	A	G	С	Т	A	G	С	Α
Species	B:	Me	t		Al	a		A]	La		IJ	Le		St	op)						
Species	C:	A	Т	G	G	С	Т	G	С	A	A	Т	Т	Т	A	С	С	Т	A	G	С	Α
Species	C:	Me	t		Al	a		A]	La		IJ	Le		ТJ	ŗ		Le	eu		A]	La	
Species	D:	A	Т	G	G	С	Т	G	С	G	Т	Т	Т	Т	A	С	С	Т	A	G	С	A
Species	D:	Me	t		Al	a		A]	La		Pł	ne		ТJ	ŗ		Le	eu		A]	La	
Species	E:	A	т	G	Α	С	Т	G	С	G	Α	Т	Т	Т	Α	С	С	Т	Α	G	С	Α
Species	E:	Me	t		Тh	ır		A]	La		IJ	Le		ТŊ	/r		Le	eu		A]	La	

How many different positions of the DNA had mutations between the five different insects?
There are four different mutations at the DNA level.
How many different positions of the Protein had had changes because of the mutations in DNA between the five different insects?
There are three changes in the amino acid sequences.
Changes in DNA sequences that don't result in a change in the protein sequence are called silent mutations. Were any of the mutations above silent mutations ? Which ones were, if any?
Mutation number 2 was a silent mutation.
Did any of the mutations cause changes in the protein sequence that are different than just changing one
amino acid?
Mutation 4 introduces a stop codon. This mutation prevents the rest of the protein from being synthesized. Mutations that introduce stop codons are called nonsense mutations .

Molecular Evolving Trees



Differences in DNA sequence can be used just like differences in physical characteristics to make a cladogram. Below is a collection of DNA sequence from an non-coading region of DNA from the hypothetical fly species that you just built a cladogram for. Following the steps below, build a cladogram using only the information in the DNA sequences.

3. Carefully compare the aligned sequences for mutations that have occurred since the species have diverged. It is often useful to use a marker or highlighter to mark positions where mutations have occurred.

Position:		1					2			3							4					
Species A:	Α	С	С	G	С	Т	Α	С	G	Α	Т	\mathbf{T}	С	G	G	С	Т	Α	G	С	Α	Т
Species B:	Α	С	С	G	С	Т	G	С	G	Α	Т	Т	С	G	G	С	С	Α	G	С	Α	Т
Species C:	Α	С	С	G	С	Т	Α	С	G	Т	Т	Т	С	G	G	С	Т	Α	G	С	Α	Т
Species D:	Α	С	С	G	С	Т	G	С	G	Α	Т	Т	С	G	G	С	Т	Α	G	С	Α	Т
Species E:	Α	G	С	G	С	Т	G	С	G	Α	Т	Т	С	G	G	С	Т	Α	G	С	Α	Т

4. Fill out the chart below to arrange the data (With a + indicating that the mutation occurred).

	Species A	Species B	Species C	Species D	Species E
Position 1: G					
to C					
Position 2:					
G to A					
Position 3:					
A to T					
Position 4:	_				
T to C					

How does this tree compare to the tree you made based on physical traits?
Which tree would you trust more, one based on physical characteristics or one based on mutations in DNA?
Do all DNA mutations lead to a change in the physical characteristics of an organism?

Below is a collection of DNA sequence from an protein coding region of DNA (a gene) from the hypothetical fly species that you just built a cladogram for. The sequence starts at the beginning of the gene. First, use a marker or highlighter to mark positions where mutations have occurred

Position:			1					2	3					4						
Species A:	Αſ	ГG	G	С	Т	G	С	Α	Α	Т	Т	Т	Α	С	С	Т	Α	G	С	Α
Species B:	Αſ	ГG	G	С	Т	G	С	G	Α	Т	Т	Т	Α	G	С	Т	Α	G	С	Α
Species C:	Αſ	ГG	G	С	Т	G	С	Α	Α	Т	Т	Т	Α	С	С	Т	Α	G	С	Α
Species D:	Αſ	ГG	G	С	Т	G	С	G	Т	Т	Т	Т	Α	С	С	Т	Α	G	С	Α
Species E:	Αſ	ГG	Α	С	Т	G	С	G	Α	Т	Т	Т	Α	С	С	Т	Α	G	С	Α

Next, use the genetic code (from the chart on the last page) to determine the protein sequence that these genes code for.

Protein																						
Species	A:	A	Т	G	G	С	Т	G	С	A	Α	Т	Т	Т	A	С	С	Т	A	G	С	Α
Species	A:	_			_			_			_			_			_			_		
Species	В:	A	Т	G	G	С	Т	G	С	G	Α	Т	Т	Т	A	G	С	Т	A	G	С	Α
Species	B:	_			_			_			_			_			_			_		
Species	C:	A	Т	G	G	С	Т	G	С	A	A	Т	Т	Т	A	С	С	Т	A	G	С	Α
Species	C:	_			_			_			_			_			_			_		
Species	D:	A	Т	G	G	С	Т	G	С	G	Т	Т	Т	Т	A	С	С	Т	A	G	С	Α
Species	D:	_			_			_			_			_			_			_		
Species	E:	A	Т	G	A	С	Т	G	С	G	A	Т	Т	Т	A	С	С	Т	A	G	С	A
Species	E:	_			_			_			_			_			_			_		

How many different positions of the DNA had mutations between the five different insects?
How many different positions of the Protein had had changes because of the mutations in DNA between the five different insects?
Changes in DNA sequences that don't result in a change in the protein sequence are called silent mutations. Were any of the mutations above silent mutations ? Which ones were, if any?
Did any of the mutations cause changes in the protein sequence that were different than just changing one amino acid?

First Nucleotide in Codon

Second Nucleotide in Codon

Third Nucleotide in Codon

	III Codoii				
4					. 🖶
	Т	С	Α	G	
	TTT Phenylalanine (Phe)	TCT Serine (Ser)	TAT Tyrosine (Tyr)	TGT Cysteine (Cys)	Т
Т	TTC Phe	TCC Ser	TAC Tyr	TGC Cys	С
	TTA Leucine (Leu)	TCA Ser	TAA STOP	TGA STOP	Α
	TTG Leu	TCG Ser	TAG STOP	TGG Tryptophan (Trp)	G
	CTT Leucine (Leu)	CCT Proline (Pro)	CAT Histidine (His)	CGT Arginine (Arg)	Т
С	CTC Leu	CCC Pro	CAC His	CGC Arg	С
	CTA Leu	CCA Pro	CAA Glutamine (Gln)	CGA Arg	Α
	CTG Leu	CCG Pro	CAG Gln	CGG Arg	G
	ATT Isoleucine (Ile)	ACT Threonine (Thr)	AAT Asparagine (Asn)	AGT Serine (Ser)	Т
Α	ATC Ile	ACC Thr	AAC Asn	AGC Ser	С
	ATA Ile	ACA Thr	AAA Lysine (Lys)	AGA Arginine (Arg)	Α
	ATG Methionine (Met) or START	ACG Thr	AAG Lys	AGG Arg	G
	GTT Valine (Val)	GCT Alanine (Ala)	GAT Aspartic acid (Asp)	GGT Glycine (Gly)	Т
G	GTC Val	GCC Ala	GAC Asp	GGC Gly	С
	GTA Val	GCA Ala	GAA Glutamic acid (Glu)	GGA Gly	Α
	GTG Val	GCG Ala	GAG Glu	GGG Gly	G