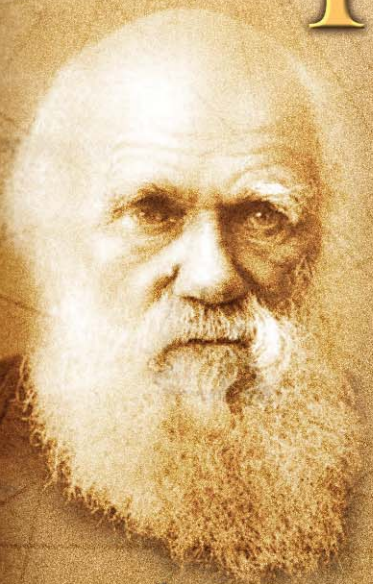


# From Finches to Fishes







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# KEY TERMS

**Abiotic** — the non-living part of the environment, such as air, water, and soil.

**Actin** — contractile protein found in muscle tissue, organized into thin filaments.

**Adaptive radiation** — relatively rapid evolution of many species from a single ancestor; generally occurs in environments where there are various unoccupied niches.

**Allele** — any of the alternate forms of nucleotide sequences forming genes that determine inherited traits. Organisms receive one allele from each parent; an organism inheriting two identical alleles is homozygous, and an organism inheriting two different alleles is heterozygous.

**Analogous** — structures that serve identical functions and look somewhat alike, but have different origins. An example would be the wings of a bird and the wings of an insect.

**Anode** — the positive electrode in an electrophoresis chamber toward which negatively charged particles are attracted.

**BME** —  **$\beta$ -mercaptoethanol** — reducing agent that may be added to an ionic detergent and heat to ensure complete breakage of disulfide bonds in a protein structure.

**Biogeography** — scientific study that uses geography to describe the distribution of plant and animal species, and is used to explain how unrelated species in different regions of the world look alike when found in similar environments.

**Biotic** — all the living organisms in the environment.

**Charge density** — ratio of charge to mass of a protein.

**Cladogram** — a dichotomous phylogenetic tree that branches repeatedly, suggesting a classification of organisms based on the time sequence in which evolutionary branches arise.

**Codon** — a set of three nucleotides (DNA bases) that code for an amino acid.

**Comparative anatomy** — scientific study that compares and describes two kinds of structures which may lead to the identification of evolutionary relationships among species.

**Dalton (Da)** — a unit of molecular weight equal to the mass of a hydrogen atom,  $1.66 \times 10^{-24}$  gm; also called atomic mass unit.

**Denature** — to disrupt or break apart a protein's three-dimensional or tertiary structure, usually with heat, alkali, or acid.

**Disulfide bridges (bond)** — the S-S bond between amino acids in a polypeptide chain which contributes to the tertiary and quaternary structure of proteins.

**Divergent evolution** — also known as adaptive radiation, by which organisms with a common ancestor that appear different externally but are similar internally.

**Embryology** — the scientific study which reveals similar stages in development (ontogeny) among related species. The similarities help establish evolutionary relationships (phylogeny). Early embryos of many different vertebrate species look remarkably similar.

**Evolution** — the process that has transformed life forms on Earth. One species can arise into another species through the gradual genetic changes over generations brought on by environmental influences or the more rapid change brought on by sudden environmental changes known as punctuated equilibrium. Evolution is supported by fossil record evidence, comparative anatomy and embryology, Gregor Mendel's research in genetics, and Charles Darwin's theory of natural selection.

**Evolutionary biology** — the scientific study which compares different living organisms to one another and to fossil forms to get an understanding of how individual species arose from earlier forms and the mechanisms that gave rise to the changes involved.

**Exon** — the region of a gene that is translated into amino acids.

**Fingerprint** — distinct pattern of bands on a protein gel formed during gel electrophoresis; useful as an identifying characteristic of a sample or species.

**Gel electrophoresis** — a technique used to separate molecules that carry electrical charges. The molecules separate from each other according to the different rates at which they migrate through an electrical field set up in a gel soaked in a chemical solution.

**Gene** — the basic unit of heredity found on a specific region of DNA, occupying a specific place on a chromosome, that encodes information for the synthesis of a single polypeptide.

**Genetic drift** — fluctuations in the frequency of a gene in a small isolated population that occurs strictly by random chance rather than natural selection.

**Genome** — the entire complement of genes in an organism.

**Genomics** — the study of all the nucleotide sequences in the chromosomes of an organism.

**Habitat** — the area in the environment where an organism lives.

**Homologous** — structures or similar features that originated in a shared ancestor. For example, the forelimbs of the penguin, alligator, bat, and human all derived from the same embryological structures.

**Intron** — region of a gene that is not translated into amino acids (compare to exon).

**Kilodalton (kD)** — 1,000 daltons.

**Molecular biology** — the scientific study of nucleotide and amino acid sequences of DNA and proteins from different species.

**Morphology** — form or structure of an organism.

**mRNA** — messenger ribonucleic acid that carries the message transcribed from DNA in the cell nucleus to the ribosomes found in the cytoplasm with information to carry out protein synthesis.

**Myosin** — a protein found in muscle tissue that is organized into thick filaments made up of an aggregate of similar proteins.

**Native** — the natural structure of a protein found within the organism.

**Niche** — the role of an organism in its environment; how an organism uses all the biotic and abiotic factors of its habitat.

**Ontogeny** — the development of an organism from the fertilized egg to maturity.

**PAGE** — polyacrylamide gel electrophoresis used in the laboratory analysis of protein and DNA structures.

**Paleontology** — the scientific study of life in the geologic past, including the study of fossils to determine the prehistoric existence of extinct species, changes in species, and the formation of new species.

**Phylogeny** — the evolutionary development of species based on lineage and history of descent.

**Posttranscriptional modification** — alterations that allow one gene to code for many proteins.

**Posttranslational modification** — alterations of proteins after they are synthesized by the cell.

**Protein** — one of the complex organic chemical compounds that form the basis of living tissues; consist of long chains of amino acids connected by peptide bonds to form one or more polypeptides. Examples are enzymes, hemoglobin, and antibodies.

**Protein folding** — the process by which a protein bends and twists to achieve its normal three-dimensional shape.

**Proteome (protein complement expressed by a genome)** — the complete protein profile found under given conditions in a biological sample.

**Proteomics** — the study of the proteome in specific cells, tissues, organs, organ systems, or organisms during a specific time period (e.g., during development).

**SDS** — sodium dodecyl sulfate, a strongly anionic detergent used to help denature a protein.

**SDS-PAGE** — sodium dodecyl sulfate — polyacrylamide gel electrophoresis, a form of electrophoresis that treats samples with SDS to denature proteins.

**Species** — organisms that have the ability to interbreed and produce fertile offspring.

**Speciation** — the formation of a new species which may occur as a result of isolation from a geographical barrier, or other kind of barriers, that prevents interbreeding between the two resulting populations.






**Transcription** — the process in a cell in which genetic information from DNA is copied to a complementary strand of RNA, specifically messenger RNA.

**Translation** — the process in the ribosomes in which a strand of messenger RNA directs the assembly of a protein from amino acids.

**tRNA** — transfer RNA delivers the amino acids necessary for protein synthesis to the ribosomes.



# The Key Components of the 5E Model

PHASE	WHAT THE TEACHER DOES THAT IS	
	Consistent with the 5E Model	Inconsistent with the 5E Model
<b>ENGAGE</b> 	<ul style="list-style-type: none"> <li>• Creates interest</li> <li>• Generates curiosity</li> <li>• Raises questions</li> <li>• Elicits responses that uncover what students know or think about the concept/subject</li> </ul>	<ul style="list-style-type: none"> <li>• Explains concepts</li> <li>• Provides definitions and answers</li> <li>• States conclusions</li> <li>• Provides premature answers to students' questions</li> <li>• Lectures</li> </ul>
<b>EXPLORE</b> 	<ul style="list-style-type: none"> <li>• Encourages students to work together without direct instruction from teacher</li> <li>• Observes and listens to students as they interact</li> <li>• Asks probing questions to redirect students' investigations when necessary</li> <li>• Provides time for students to puzzle through problems</li> <li>• Acts as a consultant for students</li> </ul>	<ul style="list-style-type: none"> <li>• Provides answers</li> <li>• Tells or explains how to work through the problem</li> <li>• Tells students they are wrong</li> <li>• Gives information or facts that solve the problem</li> <li>• Leads students step-by-step to a solution</li> </ul>
<b>EXPLAIN</b> 	<ul style="list-style-type: none"> <li>• Encourages students to explain concepts and definitions in their own words</li> <li>• Asks for justification (evidence) and clarification from students</li> <li>• Formally provides definitions, explanations, and new labels</li> <li>• Uses students' previous experiences as the basis for explaining concepts</li> </ul>	<ul style="list-style-type: none"> <li>• Accepts explanations that have no justification</li> <li>• Neglects to solicit students' explanations</li> <li>• Introduces unrelated concepts or skills</li> </ul>
<b>ELABORATE</b> 	<ul style="list-style-type: none"> <li>• Expects students to use formal labels, definitions, and explanations provided previously</li> <li>• Encourages students to apply or extend concepts and skills in new situations</li> <li>• Reminds students of alternative explanations</li> <li>• Refers students to existing data and evidence and asks "What do you already know?" "Why do you think . . .?"</li> </ul>	<ul style="list-style-type: none"> <li>• Provides definitive answers</li> <li>• Tells students they are wrong</li> <li>• Lectures</li> <li>• Leads students step-by-step to a solution</li> <li>• Explains how to work through the problem</li> </ul>
<b>EVALUATE</b> 	<ul style="list-style-type: none"> <li>• Observes students as they apply new concepts and skills</li> <li>• Assesses students' knowledge and/or skills</li> <li>• Looks for evidence that students have changed their thinking or behaviors</li> <li>• Allows students to assess their own learning and group process skills</li> <li>• Asks open-ended questions, such as "Why do you think?" "What evidence do you have?" "What do you know about x?" "How would you explain x?"</li> </ul>	<ul style="list-style-type: none"> <li>• Tests vocabulary words, terms, and isolated facts</li> <li>• Introduces new ideas or concepts</li> <li>• Creates ambiguity</li> <li>• Promotes open-ended discussion unrelated to concept or skill</li> </ul>

(Trowbridge & Bybee, 1990), adapted by Biological Sciences Curriculum Study  
 Available online at <http://science.education.nih.gov/supplements/nih1/diseases/guide/module3.htm>

# 2005-06 North Carolina Standard Course of Study for Biology — Grades 9-12

\*\*\* Highlighted sections are objectives addressed in the From Finches to Fishes module

**Strands:** Nature of Science, Science as Inquiry, Science and Technology, Science in Personal and Social Perspectives. The strands provide the context for teaching of the content Goals and Objectives.

**Competency Goal 1:**  
**The learner will develop abilities necessary to do and understand scientific inquiry.**

## Objectives

- 1.01 Identify biological questions and problems that can be answered through scientific investigations.
- 1.02 Design and conduct scientific investigations to answer biological questions.
  - Create testable hypotheses
  - Identify variables
  - Use a control or comparison group when appropriate
  - Select and use appropriate measurement tools
  - Collect and record data
  - Organize data into charts and graphs
  - Analyze and interpret data
  - Communicate findings
- 1.03 Formulate and revise scientific explanations and models of biological phenomena using logic and evidence to:
  - Explain observations
  - Make inferences and predictions
  - Explain the relationship between evidence and explanation
- 1.04 Apply safety procedures in the laboratory and in field studies:
  - Recognize and avoid potential hazards
  - Safely manipulate materials and equipment needed for scientific investigations
- 1.05 Analyze reports of scientific investigations from an informed, scientifically literate viewpoint including considerations of:
  - Appropriate sample
  - Adequacy of experimental controls
  - Replication of findings
  - Alternative interpretations of the data

**Competency Goal 2:**  
**The learner will develop an understanding of the physical, chemical and cellular basis of life.**

## Objectives

- 2.01 Compare and contrast the structure and functions of the following organic molecules:
  - Carbohydrates
  - Proteins
  - Lipids
  - Nucleic acids
- 2.02 Investigate and describe the structure and functions of cells including:
  - Cell organelles
  - Cell specialization
  - Communication among cells within an organism.
- 2.03 Investigate and analyze the cell as a living system including:
  - Maintenance of homeostasis
  - Movement of materials into and out of cells
  - Energy use and release in biochemical reactions

- 2.04 Investigate and describe the structure and function of enzymes and explain their importance in biological systems.
- 2.05 Investigate and analyze the bioenergetic reactions:
- Aerobic Respiration
  - Anaerobic Respiration
  - Photosynthesis

**Competency Goal 3:**

**The learner will develop an understanding of the continuity of life and the changes of organisms over time.**

**Objectives**

3.01 Analyze the molecular basis of heredity including:

- DNA replication
- Protein synthesis (transcription, translation)
- Gene regulation

3.02 Compare and contrast the characteristics of asexual and sexual reproduction.

3.03 Interpret and predict patterns of inheritance.

- Dominant, recessive and intermediate traits
- Multiple alleles
- Polygenic inheritance
- Sex-linked traits
- Independent assortment
- Test cross
- Pedigrees
- Punnett squares

3.04 Assess the impact of advances in genomics on individuals and society.

- Human genome project
- Applications of biotechnology

3.05 Examine the development of the theory of evolution by natural selection including:

- Development of the theory.
- The origin and history of life.
- Fossil and biochemical evidence
- Mechanisms of evolution.
- Applications (pesticide and antibiotic resistance)

**Competency Goal 4:**

**The learner will develop an understanding of the unity and diversity of life.**

**Objectives**

4.01 Analyze the classification of organisms according to their evolutionary relationships.

- The historical development and changing nature of classification systems
- Similarities and differences between eukaryotic and prokaryotic organisms
- Similarities and differences among the eukaryotic kingdoms: Protists, Fungi, Plants, Animals
- Classify organisms using keys

4.02 Analyze the processes by which organisms representative of the following groups accomplish essential life functions including:

- Unicellular protists, annelid worms, insects, amphibians, mammals, non vascular plants, gymnosperms and angiosperms
- Transport, excretion, respiration, regulation, nutrition, synthesis, reproduction, and growth and development

4.03 Assess, describe and explain adaptations affecting survival and reproductive success.

- Structural adaptations in plants and animals (form to function)
- Disease-causing viruses and microorganisms
- Co-evolution



4.04 Analyze and explain the interactive role of internal and external factors in health and disease:

- Genetics
- Immune response
- Nutrition
- Parasites
- Toxins

4.05 Analyze the broad patterns of animal behavior as adaptations to the environment.

- Innate behavior
- Learned behavior
- Social behavior

### **Competency Goal 5:**

**The learner will develop an understanding of the ecological relationships among organisms.**

#### **Objectives**

5.01 Investigate and analyze the interrelationships among organisms, populations, communities, and ecosystems.

- Techniques of field ecology
- Abiotic and biotic factors
- Carrying capacity

5.02 Analyze the flow of energy and the cycling of matter in the ecosystem.

- Relationship of the carbon cycle to photosynthesis and respiration
- Trophic levels — direction and efficiency of energy transfer

5.03 Assess human population and its impact on local ecosystems and global environments.

- Historic and potential changes in population
- Factors associated with those changes
- Climate change
- Resource use
- Sustainable practices/stewardship

# From Finches to Fishes

## Correlation to the National Science Education Standards

The Teaching Standards	
<b>From Finches to Fishes Correlation</b>	
<p>Each activity in the module provides short-term objectives for students. There is a conceptual flow of activities and a timeline for teaching the module and helping teachers plan.</p> <p>Use of this module helps teachers to update their curriculum in response to student interest in the topic.</p> <p>The module's focus is active, collaborative, and inquiry-based.</p>	<p><b>Standard A: Teachers of science plan an inquiry-based science program for their students. In doing this, teachers</b></p> <ul style="list-style-type: none"> <li>• develop a framework of yearlong and short-term goals for students.</li> <li>• select science content and adapt and design curriculum to meet the interests, knowledge, understanding, abilities, and experiences of students.</li> <li>• select teaching and assessment strategies that support the development of student understanding and nurture a community of science learners.</li> </ul>
<p>Student inquiry is encouraged by all activities in the module.</p> <p>The module promotes discourse among students, and challenges students to accept responsibility for their own learning by using hands-on, inquiry-based activities.</p> <p>The use of the 5E instructional model with collaborative learning is an effective way of responding to diversity in student backgrounds and learning styles.</p>	<p><b>Standard B: Teachers of science guide and facilitate learning. In doing this, teachers</b></p> <ul style="list-style-type: none"> <li>• focus and support inquiries while interacting with students.</li> <li>• orchestrate discourse among students about scientific ideas.</li> <li>• challenge students to accept and share responsibility for their own learning.</li> <li>• recognize and respond to student diversity and encourage all students to participate fully in science learning.</li> <li>• encourage and model the skills of scientific inquiry, as well as the curiosity, openness to new ideas and data, and skepticism that characterize science.</li> </ul>
<p>There are a variety of assessment components provided in the module, such as group discussion, data collection, and student writing.</p> <p>Answers are provided to help teachers analyze student feedback.</p>	<p><b>Standard C: Teachers of science engage in ongoing assessment of their teaching and of student learning. In doing this, teachers</b></p> <ul style="list-style-type: none"> <li>• use multiple methods and systematically gather data about student understanding and ability.</li> <li>• analyze assessment data to guide teaching.</li> </ul>
<p>The answers provided for teachers model respect for the diverse ideas, skills, and experiences of all students.</p> <p>Students work collaboratively in teams to complete activities in the module.</p> <p>Discussion activities in this module model the rules of scientific discourse.</p>	<p><b>Standard E: Teachers of science develop communities of science learners that reflect the intellectual rigor of scientific inquiry and the attitudes and social values conducive to science learning. In doing this, teachers</b></p> <ul style="list-style-type: none"> <li>• display and demand respect for the diverse ideas, skills, and experiences of all students.</li> <li>• structure and facilitate ongoing formal and informal discussion based on a shared understanding of rules of scientific discourse.</li> <li>• model and emphasize the skills, attitudes, and values of scientific inquiry.</li> </ul>

# From Finches to Fishes

## Correlation to the National Science Education Standards

The Content Standards	
<b>From Finches to Fishes activity</b>	
<b>Pre-lab Activities</b> <b>Wet-lab Activities</b>	<b>Standard A (Science as Inquiry) : As a result of activities in grades 9-12, all students should develop</b> <b>1. abilities necessary to do scientific inquiry.</b> <ul style="list-style-type: none"> <li>Identify questions and concepts that guide scientific investigations</li> <li>Use technology and mathematics to improve investigations and communications</li> <li>Formulate and revise scientific explanations and models using logic and evidence</li> <li>Recognize and analyze alternative explanations and models</li> <li>Communicate and defend a scientific argument</li> <li>Understand about scientific inquiry</li> </ul>
<b>Pre-lab Activities</b>	<b>Standard C (Life Science): As a result of their activities in grades 9-12, all students should develop understanding of</b> <b>1. the cell.</b> <ul style="list-style-type: none"> <li>Cells store and use information to guide their functions</li> <li>Cells can differentiate, and complex multicellular organisms are formed as a highly organized arrangement of differentiated cells</li> </ul> <b>2. molecular basis of heredity.</b> <ul style="list-style-type: none"> <li>In all organisms, DNA carries the instructions for specifying organism characteristics</li> <li>Changes in DNA occur spontaneously at low rates</li> </ul>
<b>Pre-lab Activities</b> <b>Wet-lab Activities</b> <b>Additional Activities</b>	<b>3. biological evolution.</b> <ul style="list-style-type: none"> <li>Species evolve over time</li> </ul>
<b>Pre-lab Activities</b> <b>Wet-lab Activities</b> <b>Post-lab Activities</b>	<b>Standard E (Science and Technology): As a result of activities in grades 9-12, all students should develop understanding of</b> <b>2. Understandings about science and technology.</b> <ul style="list-style-type: none"> <li>Scientists in different disciplines ask questions, use different methods of investigation, and accept different types of evidence to support these explanations</li> <li>Science often advances with the introduction of new technologies</li> <li>Creativity, imagination, and good knowledge base are all required in the work of science and engineering</li> <li>Science and technology are pursued for different purposes</li> </ul>
<b>Wet-lab Activities</b> <b>Post-lab Activities</b>	<b>Standard F (Science in Personal and Social Perspectives): As a result of activities in grades 9-12, all students should develop understanding of</b> <b>1. personal and community health.</b> <b>5. natural and human-induced hazards.</b> <b>6. science and technology in local, national, and global challenges.</b>



# INTRODUCTION

Teaching evolution as an inquiry process presents a challenge for educators. Traditionally students have used printed materials to learn about how organisms evolve. This module encourages students to explore the concepts of evolution using a more active, discovery-based approach.

Students are actively engaged in learning throughout this module. An interactive, large-group discussion helps to evoke and reinforce learning outcomes. During each phase of the lesson, the teacher assesses the knowledge gained by students by listening to the adequacy of their viewpoints. Enabling students to understand the practical implications of evolution is not an easy task. It necessitates overcoming apprehensions, misunderstandings, and incorrect scientific information that create barriers that keep students from being open to ideas.

## KEY CONCEPT

We often hear natural selection referred to as “survival of the fittest.” “Fittest” means that organisms must not only survive to adulthood, but they must actually reproduce.

To simulate a real trip experience to an undisclosed location (the Galapagos Islands), students are provided sunglasses, bandanas and travel information, including a map, pictures of flora and fauna found on the islands, and a departure checklist. Additional travel information is provided by a tour guide — the teacher — using a PowerPoint™ presentation.

Next on the agenda is a “fishing expedition,” since fishing is one of the islands’ major tourist attractions. To better understand the process of evolution, students use the bandanas as blindfolds, as they play the role of predatory fish whose favorite “food” is acetate “animals.”

The wet lab moves beyond DNA and utilizes one of the most widely used techniques in biotechnology research, protein gel electrophoresis. This provides a more conceptual approach for teaching biological evolution in an empirical fashion. Students can use protein electrophoresis to investigate molecular variations and examine evidence for biological evolution and common ancestry. In addition students are introduced to the science of proteomics, the study of the function, structure and interaction of proteins with each other and their environment. Additional



Water-color portrait of a young Charles Darwin, painted by George Richmond in the 1830s.

activities include computer-based bioinformatics activities specifically developed for this lab. These activities allow students to understand the power of databases and how they are used in scientific research.

## DARWIN'S THEORIES OF EVOLUTION

The observations made by Darwin on his trip to the Galapagos set the stage for the development of the theory of evolution by natural selection, which was set forth in his famous publication, *The Origin of Species*.

It is important to note that Darwin was but 26 years old when he visited the Galapagos Islands. He boarded the H.M.S. Beagle at the age of 22 as an unpaid observer. His credentials at that time were not very impressive. He was a medical school dropout from Edinburgh University and a noticeably uninspired divinity student. Prior to visiting the Galapagos, the Beagle spent three years surveying the shores of South America and spent only a brief five weeks (September-October, 1835) stay on the Galapagos Islands.

Charles Darwin developed the theories of evolution using information from several disciplines. He observed and was impressed by the amount of variation within a species, especially domestic animals. He spent a great deal of time studying cattle breeding programs. Darwin was familiar with artificial selection or choosing plants and animals with desirable characteristics.

### DID YOU KNOW?

Early embryos of many different vertebrate species look remarkably similar.

Darwin's first theory — descent with modification — states that newer forms appearing in the fossil records are actually the modified descendants of older species. Darwin's second theory — modification by natural selection — states how evolution occurs. The key factor in natural selection is the environment, which presents challenges that individuals with particular traits can better overcome. Thus the environment selects which organisms will survive and reproduce more often. These organisms are considered to be best adapted to their environment.

Darwin's theory of evolution by natural selection can be summarized in four statements:

1. Variations exist among individual species.
2. Organisms produce more offspring than the environment can support.
3. Competition exists among individuals.
4. The organisms whose variations best

adapt them to the environment are the ones who are most likely to survive, reproduce, and pass those desirable variations on to the next generation.

Evolution can also be defined as a change in the frequency of a trait (gene) within a population. Changes in gene frequency occur only when variations are passed along through succeeding generations. Evolution could not occur without genetic variation. The ultimate source of variation can now be understood as changes in the DNA caused by mutation or sexual recombination.

### SPECIATION

Over time mutations can lead to speciation. A standard definition of a species in animals is organisms that have the ability to interbreed and produce fertile offspring. Speciation or the development of a new species may occur in several ways:

- Isolation mechanisms may result from a geographical barrier, or other kinds of barriers, which prevent interbreeding between the two resulting populations. Genetic drift occurs when a resulting population is very small; some genes will increase or decrease strictly by chance. For example, one of the founding members of the small group of Germans that began the Amish community in Pennsylvania possessed an allele for polydactylism (more than five fingers or toes on a



Fossils of extinct species provide evidence for evolution.



The wings of birds and insects are an example of analogous structures: They serve the same functions and look similar, but developed from different origins.



limb). After 200 years of reproductive isolation, the number of cases of this trait among the 8,000 Amish exceeded the number of cases occurring in the remaining world's population. Other types of isolation mechanisms include:

- Habitat isolation, which occurs when species do not encounter one another because they live in different habitats.
- Temporal isolation, which occurs when species breed during different seasons, different times of the day, or even different years.
- Behavioral isolation, which exists when there is little or no sexual attraction between females and males because the potential mates do not perform the correct courtship rituals, display the proper visual signals, sing the correct mating songs, or release the proper chemicals (scents or pheromones).
- Mechanical isolation, which occurs when male and female sex organs are structurally incompatible.
- Gametic isolation, which occurs when a male and female mate but

Animals Being Compared	Number of Amino Acid Differences In Cytochrome C
Human and chimpanzees	0
Human and rhesus monkeys	1
Humans and dogs	13
Humans and rattlesnakes	20
Humans and tuna	31

the gametes do not form a zygote because the male gametes do not survive in the environment of the female gamete.

- Adaptive radiation is relatively rapid evolution of many species from a single ancestor. It occurs when the ancestral species colonize an area where diverse geographical or ecological conditions are available for colonization.

◦ Example: The fourteen species of Darwin's finches on the Galapagos Islands evolved from a single ancestor.



tral South American species. They diverged in response to the availability of different types of food in their different habitats.

## EVIDENCE FOR EVOLUTION

The following fields provide the evidence for evolution:

- **Paleontology** provides fossils revealing the prehistoric existence of extinct species and that organisms have evolved in a chronological sequence. As a result, changes in species and the formation of new species can be studied.
- **Biogeography** uses the geographic distribution of species to explain how unrelated species in different regions of the world look alike when found in similar environments.
- **Embryology** reveals similar stages in development (ontogeny) among related species. The similarities help establish evolutionary relationships (phylogeny). Early embryos of many different vertebrate species look remarkably similar.
- **Comparative anatomy** uses the comparison of body structures in different species as evidence for evolution. Homologous structures are anatomically similar features that originated in a shared ancestor. An example would be the forelimbs of the penguin, alligator, bat, and human, all of which derived from the same embryological structures. Analogous structures serve identical functions and look somewhat alike, but have different origins. An example would be the wings of a bird and the wings of an insect.
- **Molecular biology** examines nucleotide and amino acid sequences of DNA and proteins from different species.
- **Evolutionary biology** compares different living organisms to one another and to fossil forms to get an understanding of how individual species arose from earlier forms and what mechanisms gave rise to the changes involved.

## DNA AND DIVERGENCE

The tools of molecular biology have made it possible for scientists to compare DNA and protein of different organisms, in addition to their morphology. The rationale for using information about proteins to make inferences about evolution is based on the fact that DNA sequences, genes and their corresponding protein sequences are inherited. Species diverge through changes in their DNA sequences. These mutations can lead to changes in the amino acid sequences of proteins. The longer two species diverge from a common ancestor, the more differences in DNA and protein they will accumulate. Thus, the degree of relatedness of two species can be estimated from the degree of similarity or differences of their DNA or protein sequences.

In order to make a comparison of protein sequences, scientists assemble the amino acid sequences of a single protein or a group of proteins from the species in question. Next, they count the number of amino acid differences. The more differences, the longer ago the two species diverged and subsequently the degree of their relatedness is reduced. The degree of relatedness can be determined more precisely by comparing large groups of protein.

Evolutionary biologists use this comparative information along with data from the fossil records to estimate when lineages leading to various modern species diverged from a common ancestor. A common misconception is that humans descended from apes. In evolutionary terms, humans and apes each branched from a common ancestor about 5 million years ago. Humans are related to apes but did not descend from them.

## PRE-LAB

Students use a discovery-based approach to understanding the concepts of evolution by taking an imaginary journey to an undisclosed destination (the Galapagos Islands). They become totally involved in the discovery process by examining the plant and animal life on the islands as well as the physical characteristics of the islands. Students develop an understanding of the di-



Students will take an imaginary trip to the Galapagos Islands during the pre-lab, following in Darwin's footsteps.

iversity of plant and animal life, and discover species unique to this destination.

To simulate a real trip experience, students are given sunglasses, bandanas and travel information which includes maps, pictures of the flora and fauna found on the islands, and a departure checklist. Additional travel information is provided by a tour guide — the teacher — using a PowerPoint presentation which contains vivid pictures of the landscape, plants, and animals.

Next on the agenda is a “fishing expedition,” since fishing is one of the major tourist attractions of the area. To better understand the process of evolution, students are asked to become predatory fish whose favorite food is “acetate animals.” Not only are students able to develop an understanding of the predator-prey relationship, they also compile and examine the data from the class and determine the frequency of selection of a particular animal trait.

Students end their journey by returning home from their memorable trip with a figurative backpack full of information and many examples of lessons learned on their expedition. “Unpacking” from the trip leads to a discussion of vocabulary, evolutionary processes, and scientific concepts related to evolution by natural selection. During each

phase of the lesson, the teacher assesses the knowledge gained by students by listening to the adequacy of their viewpoints. One of the items students unpack upon returning is a small treasure chest that was found on their trip to the Galapagos. When students open the chest they find a treasure consisting of small strips of paper containing examples of findings from their expedition. The strips of paper in the treasure chest will provide the answers students need to complete an assignment the teacher gives them prior to taking the trip to the Galapagos.

## WET-LAB

Proteins are instrumental in almost everything cells do. They are used for structural support, storage, transport of substances, signaling from one part of the organism to another, and movement and defense against foreign substances. In addition, as enzymes, proteins selectively accelerate chemical reactions in cells. Proteins determine form and function and consequently serve as the raw materials of evolution.

The wet-lab provides students the opportunity to perform polyacrylamide gel electrophoresis to separate proteins and to compare the protein profiles of closely and distantly



Just a Theory,” from the PBS evolution series, Learning and Teaching Evolution, which combines storytelling with science content. Teachers can choose to show the video to provide closure to a class discussion or prior to an activity that involves students writing an essay in which they can apply vocabulary, evolutionary process, and scientific concepts. A rubric is even provided to help teachers score the essay.

The additional activities included in this module were designed to augment the pre-, wet-, and post-labs. The exercise entitled Using Databases to Obtain Real Amino Acid Sequence Data to Create Cladograms provides teachers with an activity for more advanced students. There are also activities that allow for interactive learning from various websites. The Peppered Moth Game, Chromosome Connections, Building Bodies, and others are all exciting ways for students to use the Internet and receive immediate

feedback on issues relating to evolution. Students can also access online lessons and an interactive website from PBS that can be used in conjunction with the video.

Interdisciplinary activities and additional resources are also provided. Dr. Amber Vogel’s Picture This is a wonderful writing and listening exercise for science and non-science classrooms that allows students to explore their feelings about evolution. This activity is recommended as a starting point for the evolution unit in a biology class and for science and non-science teachers to work cooperatively to incorporate evolution-themed work into other assignments.

The use of any or all of these post-lab and additional activities will provide teachers with a variety of ways to evaluate student understanding of concepts covered in the module and as a link to real world applications.

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## BACKGROUND

### From Bio-Rad’s Comparative Proteomics Kit I: Protein Profiler Module

#### EVOLUTION

The term evolution probably brings to mind Charles Darwin and the theory of natural selection and common descent. The observation that every species on the planet, including humans, produces far more offspring in each generation than nature can support and that the pressure of so much excess population is a harsh but efficient test of the value of accidental variations in any species was the central observation that underlies Darwin’s theory. As such, all species change gradually over time through natural selection. Each of these individuals is different — even among the same species. The environment selects organisms best suited to survive and reproduce based on those differences. Adaptations are the differences that make one organism more suited to the environment than another individual. These adaptations are phenotypic (physical) characteristics such as the variations in finch beaks that are determined by a genetic component. The genetic component is inherited from the parent in the form of genes.

The discovery of the chemical structure of DNA gave us an understanding of how the triplet code of nitrogen bases allows the synthesis of proteins (which is the phenotypic expression) and how phenotypic adaptations are the result of changes in the DNA code (mutations).

However, current research in the field of proteomics is leading some scientists to question whether or not DNA is the final determining factor in the synthesis of proteins and thus the determining factor in evolution.

The central dogma of molecular biology (DNA → RNA → protein) has given us a comfortable explanation of how the information encoded by our DNA is translated and used to make an organism. It describes how a gene made of DNA is transcribed by messenger RNA and is then translated into a protein by transfer RNA in a complex series of events utilizing ribosomal RNA and amino acids. New discoveries about alternative roles for RNA, multiple forms of proteins being encoded by single genes in our cells,



## ON THE WEB

The Human Genome Project

[www.ornl.gov/sci/techresources/Human\\_Genome/home.shtml](http://www.ornl.gov/sci/techresources/Human_Genome/home.shtml)

and changes to proteins after translation are changing this comfortable scenario and we are finding that things (as ever in biology) are not so simple. Although in essence the central dogma remains true, investigations into genomics and proteomics are revealing a complexity that many had never imagined.

In 1990, a massive research effort took place to sequence what was estimated to be the 100,000 genes that coded for each protein synthesized by humans (the human genome).

This study, the Human Genome Project, took 13 years to complete (Jasny and Kennedy, 2001). When the study began, scientists estimated that there were over 100,000 human genes. Now, years after the genome has been sequenced, there is still no consensus on the actual number of human genes, but the current estimate is down to around 22,000 human genes; this is only a few thousand more genes than encodes the genome of a much simpler organism, *C. elegans*, a nematode worm that has around 19,000 genes.

So why are a similar number of genes required to make a worm and a person? Importantly, a human has a much larger total genome (3 billion base pairs) than a worm (100 million base pairs), suggesting that the total amount of DNA rather than the actual number of genes may be what gives rise to complexity. In addition, recent developments have shown it is quite common in complex organisms for a single gene to encode multiple proteins.

Moreover, changing when, to what level and where a protein is expressed or changing a protein after it has been translated (posttranslational modification) can result in proteins with very different functions. This realization of the importance and diversity of proteins started a whole new field termed proteomics.

Proteomics was initially defined as the effort to catalog all the proteins expressed in all cells at all stages of development. That definition has now been expanded to include the study of protein functions, protein-protein interactions, cellular locations, expression levels, and posttranslational modifications of all proteins within all cells and tissues at all stages of development. Thus, it is hypoth-

esized that a large amount of the exogenous noncoding DNA in the human genome functions to highly regulate protein production, expression levels, posttranslational modifications, etc. and it is this regulation of our complex proteomes, rather than our genes, that makes us different from worms.

Researchers in the proteomics field have discovered a number of modification systems that allow one gene to code for many proteins and mechanisms that finely regulate the sub- and extracellular locations and expression levels of proteins. These include alternative splicing of exons, use of different promoters, posttranscriptional modification, translational frameshifting and posttranslational modification. Let's examine some of these systems.

## POSTTRANSCRIPTIONAL MODIFICATIONS

### • RNA Editing

A newly discovered form of posttranscriptional modification is RNA editing. Higher eukaryotes can change the sequence of their messenger RNAs (mRNAs) by substituting bases in their primary mRNA transcripts at specific positions, while lower eukaryotes insert and delete specific bases. These types of changes to the codons of mRNA can change the amino acid sequence of a protein, create a new open reading frame where one did not originally exist, or introduce a new stop codon to create a truncated protein. These changes can be regulated by the location of their expression, developmentally and by the organism's environment, and the resulting proteins can have very different functions. For example, fasting rats produce 50% less of an edited form of apolipoprotein B (a protein involved in cholesterol metabolism) than rats on a high carbohydrate diet (Maas and Rich, 2000). This happens when a cytidine is substituted with uridine. These base substitutions happen at specific locations, indicating they are involved in the regulation of metabolism.

RNA editing is not limited to messenger RNA. In a number of yeast and higher eukaryotes, transfer RNA is also edited at the wobble positions of its anticodons (Maas and Rich, 2000).

- **Alternative Splicing**

Messenger RNA (mRNA) of higher eukaryotes has two types of sequence segments — introns and exons. Introns are portions of the code that are removed or edited from the sequence to be transcribed into protein. The remaining base segments, exons, move to the ribosome for translation. Exons can be differentially included or excluded in a process called alternative splicing to produce different mature mRNAs, which in turn generate distinct proteins. This process can be used at different stages in development or simply when a cell is signaled to alter its protein production.

- **mRNA Synthesis and Degradation**

Since the level of mRNA within a cell in part determines the level of protein expression, the rate at which mRNA is synthesized and the rate at which mRNA is degraded are important factors in protein expression levels. Some mRNAs are inherently more unstable than others and there are active processes within cells that regulate mRNA's degradation as well as its synthesis. Modifications to mRNAs can also change their stability and thus their levels of protein expression. For example, capping an mRNA with a 7-methyl guanosine nucleotide and the presence of a polyadenosine tail increases its stability. As such, the regulation of mRNA by capping and polyadenylation increase protein expression.

## POSTTRANSLATIONAL MODIFICATIONS

- **Proteolytic Cleavage**

Most proteins undergo cleavage after translation: the simplest form of this is the removal of the initiation methionine. In addition, many enzymes are regulated by proteolytic cleavage and are either activated or inactivated (or both) by cleavage of specific domains within the protein. Examples of this are the procollagenases that digest extracellular matrix collagens only after the removal of their “pro” domain. Protein cleavage results in different proteins of different sizes derived from the same gene.

- **Protein Degradation**

Similar to mRNA synthesis and degradation, proteins are actively regulated by degradation mechanisms. Proteins have specific half-lives and these can be regulated by posttranslational modifications. In one mechanism, that won its elucidators the 2004 Nobel Prize in Chemistry, proteins are tagged with ubiquitin proteins and degraded in highly regulated cellular degradation machinery known as proteasomes. By regulating the degradation of its proteins, a cell can quickly change the levels, as well as the presence or absence of a protein in response to outside factors.

- **Protein-Protein Interaction**

Many proteins exist in complexes and the presence or absence of other proteins from these complexes can determine their function. Thus proteins are dependent on other proteins for their activity. Although a specific protein may be present in abundance, if its binding partner is in short supply, then the protein will have little, if any, activity.

- **Carbohydrate Modification (Glycosylation)**

Many proteins, especially those associated with the plasma membrane and those that are secreted, are covalently bound to carbohydrates — usually sugars. Carbohydrate modification drastically changes the way proteins behave and interact with other proteins or structures and act as targeting molecules to direct proteins to specific locations within the organism or cell. Lymphocytes (white blood cells) have carbohydrate groups on their outer membranes which are vital in determining how lymphocytes infiltrate sites of infection. Carbohydrate modifications can result in proteins with different molecular weights and different physio-chemical properties derived from the same gene.

- **Phosphorylation**

Many proteins are regulated by phosphorylation. Most phosphorylation reactions change the activity of enzymes, often by causing conformational changes

within the protein. This is the case with myosin light chain 2 or myosin regulatory light chain, which contracts smooth muscle when phosphorylated. There is a special class of enzymes that phosphorylate proteins called kinases, and their counterparts, which dephosphorylate proteins are called phosphatases. Many proteins are phosphorylated on multiple sites, and each phosphorylation site has a specific role in the function or activity level of the protein. Thus a protein may exist in many different states of activity depending on which sites have been phosphorylated.

Other forms of posttranslational modification include methylation, sulfation, prenylation and acetylation.

Based on this new research, proteomic researchers can feasibly argue that it becomes increasingly important to examine differences in the proteins being expressed in different species just as it is important to examine differences in DNA code. Phenotypic diversity is achieved with little cost. Life shows amazing economy when one gene can encode many proteins and these proteins can be subsequently modified to suit a particular environment.

Imagine the flexibility of an organism with an extensive RNA editing system! We previously thought protein sequence could only be changed at the level of DNA mutations, which were rare and occurred randomly, an adaptation mechanism inherent with high risk. We are now beginning to understand that there are indirect forces, in addition to gene mutation, that can drive evolution. Modifying an RNA editing system to change a protein complement would be far less risky than irreversibly changing the genes encoding proteins themselves since edits are optional and reversible. Thus an organism would have much more flexibility to adapt to new and different environments. Mounting research suggests that the number of RNA editing systems is great and that the similarity in these systems may be of evolutionary significance.

In the Comparative Proteomics Kit I: Protein Profiler Module we focus on comparing the proteomes of muscle cells and the differences in these proteomes between distantly and closely related species. The differences found can then be used to determine, or infer, evolutionary relationships between different species.

## CONNECTION TO OTHER MODULES

The wet-lab presented here, along with the three additional wet-lab activities mentioned below, provides students with first-hand experience of common techniques used by molecular biologists. Molecular biology examines nucleotide and amino acid sequences of DNA and proteins from different species. Because the genetic information in DNA provides the instructions eventually expressed as proteins, alterations in DNA can alter proteins so that they function differently.

### EXPLORING NEW ENVIRONMENTS

Exploring New Environments was developed by DESTINY to enable students to investigate ecosystems and

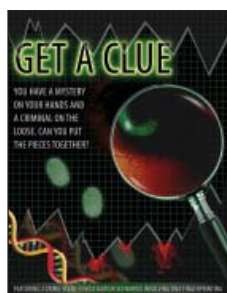


explore the complex relationships between organisms and their physical environment. In Finches to Fishes, students investigate the ecosystems of the Galapagos Islands.

They identify the flora and fauna of these enchanting islands as they contemplate interaction between living organisms and their physical environment.

### GET A CLUE

In DESTINY's module Get a Clue, students use agarose gels and horizontal gel chambers

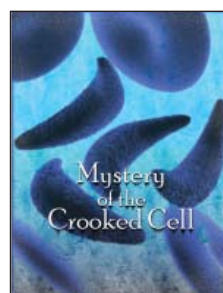


to separate DNA that has been cut with a restriction enzyme. The wet lab in Finches to Fishes provides an opportunity for students to increase their knowledge of molecular biology and refine their techniques of

electrophoresis by separating proteins using precast polyacrylimide gels and vertical gel chambers.

### MYSTERY OF THE CROOKED CELL

The module Finches to Fishes examines the structure and function of proteins as it relates to the DNA code. To prepare students for this content, we strongly recommend Mystery



of the Crooked Cell, a module developed by Boston University School of Medicine's CityLab. In Mystery of the Crooked Cell, students examine the genetic basis for sickle cell anemia. Students observe functional

differences in the normal hemoglobin and sickle cell hemoglobin which result from a point mutation that changes the DNA. In both modules students perform an electrophoresis of proteins and examine the changes that result in the protein as a result of changes in the DNA.

### SEQUENCE OF MODULES

A sequence relating the four modules is summarized below:

#### 1. EXPLORING NEW ENVIRONMENTS

Students investigate complex relationships between organisms and the physical and biological environment as well as the movement of materials and energy through an ecosystem.

#### 2. GET A CLUE

Students learn the structure and function of DNA. Students learn basic techniques of DNA gel electrophoresis.

#### 3. MYSTERY OF THE CROOKED CELL

Students learn how changes in DNA structure lead to changes in protein structure. Students learn basic techniques of protein electrophoresis.

#### 4. FROM FINCHES TO FISHES

Students compare protein from the muscle cells of fish to determine which fish are most closely related. Since DNA determines protein, those fish which have the most similar proteins would be expected to have similar DNA and similar origins. This is a more advanced type of electrophoresis lab using polyacrylimide vertical gels to separate proteins.





## FROM FINCHES TO FISHES IMPLEMENTATION PLAN — PRE-LAB

Activity	Estimated Time	Materials/Equipment	Purpose/Objectives/ Essential Question
Engagement: Imaginary trip to the Galapagos	20 minutes	Travel Package: maps, pictures of flora and fauna, departure checklist, bandanas, sunglasses, etc. Group report sheet CD — PowerPoint presentation/tour guide script	<p><b>Purpose:</b> To examine the process of evolution by natural selection. To discover the features, wildlife, and scientific significance of the Galapagos Islands in relationship to the study of natural selection.</p> <p><b>Objectives:</b></p> <ul style="list-style-type: none"> <li>• To define evolution</li> <li>• To discuss predator-prey relationships</li> <li>• To understand the mechanisms that bring about change</li> <li>• To examine the process of speciation and geographic isolation</li> <li>• To provide evidence that supports the theory of evolution by means of natural selection</li> </ul> <p><b>Essential Question:</b> How does natural selection work to create new species?</p>
Exploration: Fishing Trip — Go Fish	20 minutes	Blindfolds, acetate sheets (two colors) Go Fish activity sheet Tally sheet Calculators	
Explanation	20 minutes	Data table Discussion questions	
Elaboration: Discussion of trip — “Backpack of Memorabilia”	20 minutes	“Memorabilia backpack” consisting of travel packet, CD PowerPoint (2nd part) with discussion questions, and group report sheet Paper people models	
Evaluation	10 minutes	Small treasure chest Answer strips — placed in the treasure chest Evaluation questions sheet  NOTE: For higher-level classes, consider using the essay and rubric from the Additional Activities section instead.	

## Alignment with NC Competency Goals

### Biology

Goal 1  
Objectives 1.01, 1.02, 1.03  
Goal 2  
Objectives 2.01, 2.02, 2.03

Goal 3  
Objectives 3.01, 3.04, 3.05  
Goal 4  
Objectives 4.01, 4.02, 4.03  
Goal 5  
Objective 5.01

# FROM FINCHES TO FISHES PRE-LAB ACTIVITIES: UTILIZING THE 5E INSTRUCTIONAL MODEL

## ENGAGEMENT

Working in teams, students will become explorers as they take an imaginary trip to an undisclosed destination (only the teacher knows it's the Galapagos Islands).



Each student will receive a bandana and a pair of sunglasses for the trip. Teachers are encouraged to take a group photo, representative of an actual group trip to an exotic location. Each student group will be presented with a travel package that includes maps, pictures of the flora and fauna found on the island, and a departure checklist. These items will provide clues to assist students in naming their final destination. A Group Report Sheet is provided for possible explanations or questions about features, location, wildlife, and the scientific significance of the destination to be visited.

A PowerPoint presentation consisting of pictures from the Galapagos and a tour guide script is available on the module CD for teacher use. In addition Questions from Group Report Sheet; Imaginary Trip to the Galapagos is provided for additional information on the Galapagos.

## EXPLORATION

One of the key tourist attractions of the islands is fishing. To enable students to better understand the process of evolution, they are asked to become predatory fish whose favorite "food" is acetate "animals." Students are blindfolded and led into an adjacent lab room where they become predatory fish that have a limited period of daylight to search for their prey (blindfolds are removed for 10 seconds); they must catch as many as possible before the sun goes down. After sundown, predators leave the adjacent lab room, take off the blindfolds and determine the number of each type of "acetate animal" remaining.



## EXPLANATION

Upon completion of the Go Fish activity, the class will complete the data table together. Each group will be asked to present an oral summary of its findings. Their presentations will provide a launching pad to present scientific explanations and terminology. Questions to be addressed are as follows:

1. Did one color of acetate "animal" have an advantageous variation?

2. What happened to the frequency of the clear color? Why?
3. How might Darwin have explained the change in the acetate "animal" population?
4. How would you define evolution?

## ELABORATION



Students return home from a memorable trip with a backpack filled with memories of their journey. As they unpack their bag they pull out of the backpack (pictures and descriptions) many examples of lessons learned on their expedition.

During this elaboration phase, students have an opportunity to correctly use technical vocabulary and apply scientific reasoning. The instructor will lead a discussion related to the class's findings. Some of the concepts to be discussed are as follows:

- Environment and conditions for survival
- Relatedness vs. common ancestor
- Descent with modification
- Adaptive radiation
- Modification by natural selection
- Isolating mechanisms
- Evidence for evolution
- Genetic drift
- Adaptations, fitness
- Extinction
- Variations, sources of variation
- Species, subspecies and speciation process

## EVALUATION

During each phase of the lesson, the teacher will assess the knowledge gained by students by listening to the adequacy of their viewpoints. One of the items students unpack from their bag is a small treasure chest which was found on their trip to the Galapagos. When students open up the chest they find treasure that consists of small strips of paper containing examples of findings from their expedition.



The strips of paper in the treasure chest will provide the answers students need to complete an assignment given them by their teacher prior to taking the trip to the Galapagos.

## ENGAGEMENT ACTIVITY: THE TRIP — PREPARE FOR DEPARTURE

### MATERIALS NEEDED

- 1 bandana and 1 pair of inexpensive sunglasses for each student
- (optional) Polaroid camera and film to take pictures of each group
- 1 travel packet for each group of four

To create travel packets, place into a pocket folder color printouts of pages 29-34 of this module, which contain: a map with a checklist titled Prepare for Departure, pictures of the flora and fauna found on the island (place these sheets in plastic sheet protectors to prolong use), and a Group Report Sheet. All items are available on the CD accompanying the module.

- PowerPoint presentation part one (available on module CD)
- Narrative for PowerPoint Presentation script

### INSTRUCTIONS FOR TEACHERS

- Working in groups, the students will become explorers as they take an imaginary trip to an undisclosed destination. At the beginning of the class, distribute inexpensive sunglasses and bandanas to each student in preparation for the trip activity. You may choose to build upon the excitement of traveling to an enchanting island by taking a picture of the students in each group wearing

sunglasses and bandanas tied around their necks for their scrapbook.

- Divide the class into groups of four and provide each group a travel packet.
- Each group of students should use its Group Report Sheet to record possible explanations or questions about physical features, location, wildlife, and the scientific significance of the destination to be visited.
- The CD that accompanies this module includes a three-part PowerPoint presentation; the first part consists of pictures from the Galapagos. Act as a tour guide, showing part one of the PowerPoint presentation to the class while reading the script called Narrative for PowerPoint Presentation, Imaginary Trip to the Galapagos. The script provides interesting additional information about each picture.
- After finishing the “tour,” give groups a few minutes to complete their group report using the information they’ve learned during the presentation and the materials in their travel packets. Once the groups have made their preliminary report, discuss their findings and questions as a class. Use the Questions From Group Report Sheet: Imaginary Trip to the Galapagos for additional information on the Galapagos and to make sure all the questions the class has generated have been addressed.

### NOTE FOR TEACHERS

Do not reveal to the students the destination (the Galapagos Islands) since they will make observations and notes from the information given to them.



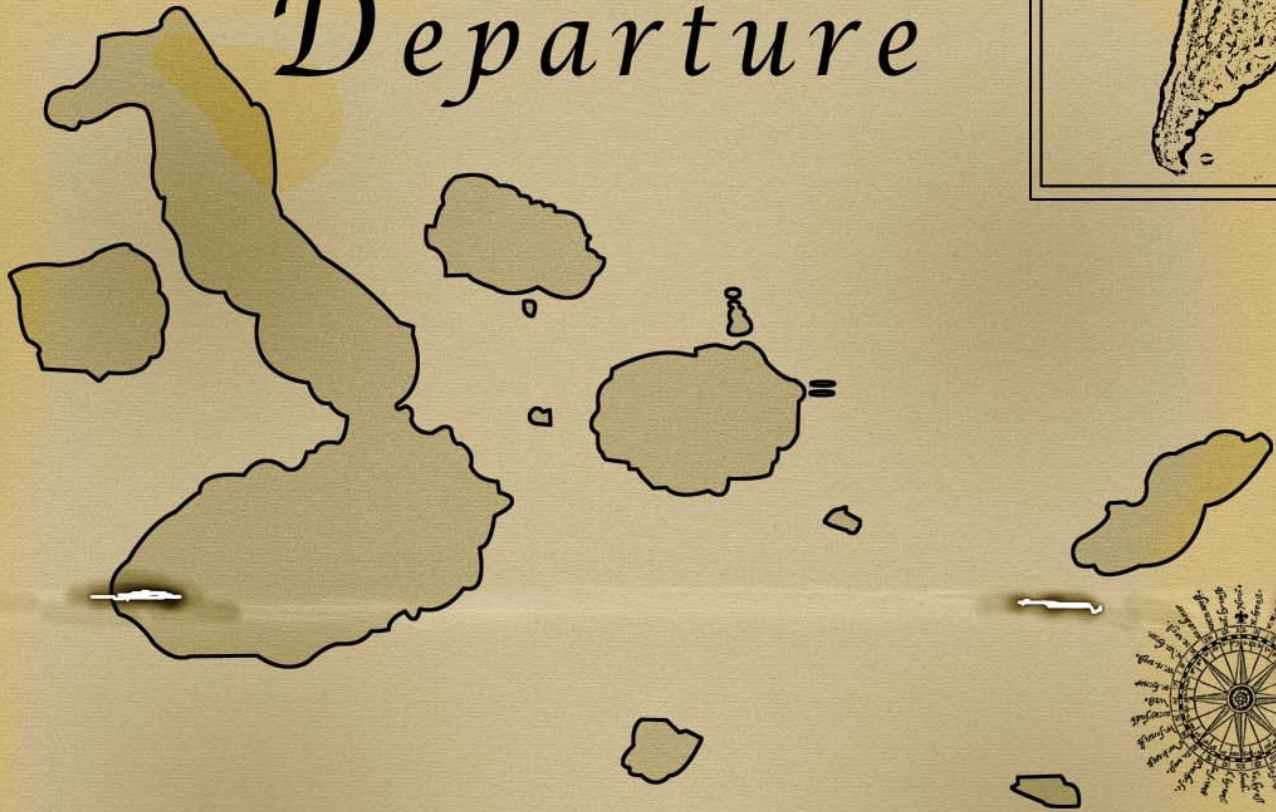


Pre-lab Materials				
Item	Suggested Vendor	Model or Catalog #	Unit	Price
Acetate sheets (clear and gray)*	Office Depot	741361	1 pack	\$4.79
Treasure Chests	Oriental Trading**	IN-3/570	dozen	\$3.95
Plastic sheet protectors	Staples	489131	100 box	\$11.89
Pocket portfolios	Staples	907701	10 pk	\$3.49
Optional Pre-lab Materials				
<i>A Traveler's Guide to the Galapagos Islands.</i> Barry Boyce. 4th ed. Aptos CA; Galapagos Travel, 2004.	www.amazon.com		1	\$12.34

\* The acetate sheets are actually poly construction folders that come in packs of 10. Use only the clear and gray folders. Cut each folder into two 8½" x 11" sheets. Then cut each sheet into 200 squares.

\*\* Also a good source for bandanas and sunglasses.

# Prepare For Departure



The islands are isolated (by several hundred miles of ocean) so the climate is determined almost entirely by the ocean currents, which in turn are influenced by the trade winds, which push the currents. There are two distinct seasons—one short season with rainfall and another longer season with very little rainfall. Even though the islands are located directly under the equator, the climate is far from being excessively hot, caused singularly by the low temperature of the surrounding water, brought by the great southern Polar current.

## TRAVEL NECESSITIES

- Travel documentation — valid passport
- Money — \$350.00 (cash and travelers checks)
- Credit cards
- Telephone calling card
- Medications

## CLOTHING

- Shorts and bathing suits (2 or 3 of each)
- Bed clothes (pajamas, etc.)
- T-shirts (1 week supply)
- Socks
- Underwear

- Sweatshirt, jacket, or sweater
- One pair of pants
- 2-3 shirts or blouses
- Athletic shoes (sneakers) or hiking boots
- Hat that fits rather snug or one with an adjustable chin-strap, sweatband
- Toiletries

## UTILITY ITEMS

- Binoculars
- Business cards
- Camera and film
- Daypack (water bottle, bandages, first aid cream, guide-

- book, film camera), fanny pack
- Earplugs
- Plastic bags
- Flashlight/book light
- Rain gear/poncho
- First aid kit
- Sunglasses
- Extra pair of eyeglasses (or contact lenses)
- Sunscreen lotion
- Vitamins
- Water resistant watch
- Water bottle
- Snorkeling equipment/wet suit





Photos from the National Oceanic and Atmospheric Administration/Department of Commerce  
Watercolor painting by Owen Stanley





Iguana photos courtesy of Dr. Bob Rothman (<http://www.rit.edu/~rhrsbi/HomePage/homepage.html>)  
Other photos from the National Oceanic and Atmospheric Administration/Department of Commerce





Lower left photos courtesy of Dr. Bob Rothman (<http://www.rit.edu/~rhrsbi/HomePage/homepage.html>)  
Other photos from the National Oceanic and Atmospheric Administration/Department of Commerce





Photos courtesy of Dr. Bob Rothman  
(<http://www.rit.edu/~rhrsbi/HomePage/homepage.html>)

## GROUP REPORT

Students should take notes from the travel package and from the “tour guide” presentation.

Possible destination \_\_\_\_\_

Your observations during the presentation	Questions you have about the presentation
Physical Factors of the Environment	Physical Factors of the Environment
Plants	Plants
Animals	Animals

What do you think is the possible scientific significance of this trip?

## NARRATIVE FOR POWERPOINT PRESENTATION

### IMAGINARY TRIP TO THE GALAPAGOS

**Slide 4:** Today we are taking an imaginary journey to an undisclosed destination four hours from Miami. We will visit a remote paradise 600 miles west of Ecuador, to a small group of 15 islands located on the equator. Formed 3-5 million years ago, these islands are volcanic in origin and are sometimes referred to as the Enchanted Islands.

**Slide 5:** To get there, we first have to fly to Ecuador, then hop on a boat and sail to the set of mysterious islands. The cruise ship we're taking holds 34-90 passengers, and we will visit two islands per day.

**Slide 6:** Welcome ashore! It's been a long journey and I'm sure you're all tired, but we have a lot of exploring to do. Notice the landscape; it's rocky, and it appears to be pretty dry and arid. There are black, porous volcanic rocks, jagged cliff formations, lava fields, pit craters, and columns of gas-driven steam and seascapes. The landscape could best be described as a moonscape.

**Slide 7:** As we begin to explore, we are able to see some of the flora of the island. Here we see some prickly pears; there are 14 different varieties of prickly pears found on these islands.

**Slide 8:** Again, as we take a look at the landscape, it appears to be rocky and mountainous, and we see a small cactus growing out of rock formed from a lava flow.

**Slide 9:** Back at the water's edge, we see a fur seal. This seems oddly out of place since we are on the equator. They have large eyes, probably to help hunt for fish and squid.

**Slide 10:** Also at the water is a sea lion, which also seems to be out of place. Colonies of sea lions are found here on the gently-sloping rocky shore where they can hunt for fish. The males are larger than the females.

**Slide 11:** Take a look at the marine iguanas. They are frozen in a pose atop the lava rocks and are sun bathing to get their body temperatures up. The marine iguana is a very competent diver and swimmer; it is the only lizard to achieve this ability. It can also sneeze brine out of the efficient salt-excreting glands located in its nostrils. This frequent salt-sneezing gives the marine iguana its white-crested forehead. It looks a lot like other iguanas, but it behaves much differently. The marine iguana is an avid swimmer, tucking its legs and whipping its body and tail to propel itself through the water. It can stay underwater between 5 to 10 minutes, and dive as deep as 2 meters. It also has very sharp claws, which it uses to hold onto rocks in the surf while feeding on algae. When a male wants to attract a female, its black skin turns bright red.

**Slide 12:** Here is another iguana, but this is a land iguana. Confined to the interior of the island, land iguanas can grow to over three feet in length, and can live more than 60 years. They actually eat prickly pear cactus, standing on their hind legs to reach the plant. Their tongue is very leathery, and they don't even remove the spines from the cactus before eating.

**Slide 13:** Look at the giant tortoise. They look like gentle giants. They are very social creatures. Each island in this group has its own race of giant tortoise. The giant tortoise can have 3 differently shaped shells. They eat large quantities of grass, leaves, and cactus pads. They are very fond of water, drinking large quantities and wallowing in the mud. Giant tortoises can grow to up to 500 pounds and may live up to 150 years.

**Slide 14:** Recognize this face? The face of the giant tortoise was used to model ET.

**Slide 15:** This brightly colored Sally Lightfoot crab shows up pretty clearly on the black lava rock. The Sally light-foot juveniles, however, are dark in color, camouflaging them against the dark rocks.

**Slide 16:** This male Magnificent Frigatebird has inflated his throat pouch to about the size of a football to attract a mate. He also makes a warbling sound to get her attention. These birds are large and black with long wings, long

hooked beaks, and forked tails. The Great Frigatebird is another species found in this same place.

**Slide 17:** Penguins? Is this a little unusual? One of the main problems for this penguin is keeping cool living so close to the equator. The temperature can get over 100 degrees F (38 degrees Celsius) during the daytime. The penguins hold their flippers out to help the heat escape their bodies. They protect their feet from getting sunburned by holding their flippers over their feet when on land. They keep cool by swimming and hunting for food in the cold water of the ocean currents during the day. During the cool nights they sleep and nest on the land. These penguins eat mostly small fish such as mullet and sardines, which are brought by ocean currents to their feeding grounds.

**Slide 18:** These blue-footed boobies are some of the most common birds on the islands. The booby in the middle is “pointing” into the air to attract a mate. The boobies will also “parade” to get a mate. They have a very elaborate mating dance. Blue-footed boobies will gather nesting materials, only to ignore them later to lay and incubate their eggs directly on the ground. They have really silly behavior and blue feet.

**Slide 19:** The red-footed booby is just as colorful as the blue-footed booby. The red-footed booby prefers to lay eggs and hang out in and around trees, making it one of the few seabirds that perch in vegetation.

**Slide 20:** The masked booby got its name for obvious reasons. This is the biggest species of booby found on the islands, with a wingspan of over 5 feet.

**Slide 21:** Another bird we see is the enormous albatross, whose wingspan can be over 10 ½ feet. They also have a long lifespan and can live about 50 years.

**Slide 22:** This is a flightless cormorant. It is flightless because it traded in its “flying wings” to make it better suited for swimming and diving. Flightless cormorants are large, dark-colored, and have a long hooked end on their beak, which helps them hunt for eels, octopus, and fish.

**Slide 23:** We also have smaller birds called finches. These islands are home to 13 endemic species of finches, which are believed to have come from one common ancestor. One of the most prominent features that help us distinguish finch species is the size and shape of beaks. The unique beak is specifically shaped for the foods eaten, such as insects, flowers, leaves, seeds, and even cactus.

**Slide 24:** The small tree finch feeds on insects, while the large cactus finch feeds on seeds. Notice the different shape of their beaks.

**Slide 25:** It’s been a very busy trip, and we’ve seen a lot. We saw vast numbers of birds and reptiles, but only a few mammals. If we had gone snorkeling or diving, we would have encountered an undersea world rich with tropical fish, corals, sharks, eels, rays, dolphins, and more. Now, it’s time for us to return home to reflect on what we saw on our trip. We have lots of memories that we will want to share with others.

## POSSIBLE QUESTIONS THAT MAY ARISE FROM GROUP REPORT SHEET: IMAGINARY TRIP TO THE GALAPAGOS

These are questions that may arise following the “tour.” Discuss these questions with your students so that they can add to the group report sheet.

1. How did the seals get here (so far south)?
2. Do seals migrate?
3. Are any of the volcanoes active?
4. What are the temperature extremes?
5. How far are the islands off the coast of Ecuador?
6. Are the islands still growing?
7. Are the pictures we have of extinct or living animals?
8. Do we have the same or different species from island to island?
9. How did the animals get to the different islands?
10. Is the water safe? Clean? Drinkable?
11. What is the elevation of the islands?
12. What is the human population?
13. Would inhabitants need weapons?
14. What is the weather like?
15. What did Charles Darwin find interesting about the Galapagos?
16. What do currents have to do with the dry climate? Why isn't it hot?
17. Why hasn't the wildlife evolved past birds and reptiles?
18. What is the volcanic life?
19. What types of mineral deposits exist?
20. What is the impact of human involvement on the Galapagos?





## POSSIBLE QUESTIONS THAT MAY ARISE FROM GROUP REPORT SHEET: IMAGINARY TRIP TO THE GALAPAGOS

1. How did the seals get here (so far south)?

These animals could have crossed the ocean on their own power.

2. Do seals migrate?

Yes, seals do migrate.

3. Are any of the volcanoes active?

Today, the Galapagos are among the world's most active volcanic areas. There have been 50 eruptions in the last 200 years, some quite recently. The most recent was in 1995 on Fernandina Island.

4. What are the temperature extremes?

“Considering that these islands are placed directly under the equator, the climate is far from being excessively hot; this seems chiefly caused by the singularly low temperature of the surrounding water, brought here by the southern Polar current. Excepting during one short season, very little rain falls, and even then it is irregular; but the clouds generally hang low. Hence, whilst the lower parts of the islands are very sterile, the upper parts, at a height of a thousand feet and upwards, possess a damp climate and a tolerable luxuriant vegetation. This is especially the case on the windward sides of the islands, which receive the moisture from the atmosphere....”

— Charles Darwin, *Voyage of the Beagle*, 1839

One of the keys to understanding the Galapagos is the climate. The islands are isolated by several hundred miles of ocean, so the climate is determined by the ocean currents, which in turn are influenced by the trade winds which push the currents. The Galapagos are at a major intersection of several currents, which vary in intensity during the year as their driving trade winds blow and then weaken in a cycle that gives two distinct seasons to the island.

May-November water temperature 70-75° F.

Dec-April water temperature 75-80° F.

5. How far are the islands off the coast of Ecuador?

600 miles from Ecuador/1000 miles from Central America

6. Are the islands still growing?

Yes, the Galapagos are quite young and are still in early stages of formation. The western islands (Fernandina and Isabela) are 100,000-200,000 years old. The Galapagos Islands first broke through the ocean floor some 3-5 million years ago. (The Galapagos consist of a chain of 15 islands.)

7. Are the pictures we have of extinct or living animals?

The pictures are of animals that are currently living on the Galapagos Islands.

8. Do we have the same or different species from island to island?

Darwin saw many examples of one species eventually becoming another — for example, the thirteen species of finches. They obviously were of the same parent population and yet they were distinct in a way that was arguably adaptive. There are also several different species of iguanas (land and marine) and prickly pears found on different islands.

9. How did the animals get to the different islands?

Ocean passengers (reptiles aboard rafts of tangled vegetation).

No amphibians or land mammals with the exception of rats aboard ships.

Some animals could cross the ocean on their own power (examples: fish, sea turtles, penguins, and marine mammals — the sea lions, fur seals, and dolphins).

Sea birds and shore birds had the ability to arrive by air. Land birds would either have to get caught up in a gust and be borne along by an exceptionally strong tail wind or perched on a branch in a tree hollow.



10. Is the water safe? Clean? Drinkable?

Don't drink the water unless it is bottled. It is not safe to drink.

11. What is the elevation of the islands?

The highest elevation occurs on Santiago Island: 900 meters above sea level.

12. What is the human population?

Four of the islands have human settlements. The total human population in the Galapagos is 18,000.

13. Would inhabitants need weapons?

No weapons will be needed. The animals are not aggressive for the most part.

14. What is the weather like?

(Refer back to question #4) "Except during one short season, very little rain falls, and even then it is irregular; but the clouds generally hang low. Hence, whilst the lower parts of the islands are very sterile, the upper parts, at a height of a thousand feet and upwards, possess a damp climate and a tolerable luxuriant vegetation. This is especially the case on the windward sides of the islands, which receive the moisture from the atmosphere...." (Charles Darwin, *Voyage of the Beagle*, 1839)

15. What did Charles Darwin find interesting about the Galapagos?

Darwin found the Galapagos to be a "living laboratory" for observing evolution.

16. What do currents have to do with the dry climate? Why isn't it hot?

(Refer back to question #4) Considering that these islands are placed directly under the equator, the climate is far from being excessively hot; this seems chiefly caused by the singularly low temperature of the surrounding water, brought here by the southern Polar current. (Charles Darwin, *Voyage of the Beagle*, 1839)

One of the keys to understanding the Galapagos is the climate. The islands are isolated (by several hundred miles of ocean), so the climate is determined by the ocean currents, which in turn are influenced by the trade winds which push the currents. The Galapagos are at a major intersection of several currents, which vary in intensity during the year as their diving trade winds blow and then weaken in a cycle that gives two distinct seasons to the island.

17. Why hasn't the wildlife evolved past birds and reptiles?

Mammals are generally not suited to the rigors of the long ocean voyage to the archipelago. Although understandable, it is still amazing to note that there are only seven species of mammals that are indigenous to the Galapagos. It is even more amazing that, of these, there are only three land mammal species — all rats! The balance of this under-represented class were either carried through the air by storm-force winds (two bat species) or by long-distance swimming (the sea lions and fur seals).

18. What is the volcanic life?

Pioneers after volcanic activity include bacteria, algae, and fungi, which provide the beginnings of an organic base for higher forms of life.

19. What types of mineral deposits exist?

Sulfur vents, pockets of magna, pit craters, molten rocks

20. What is the impact of human involvement on the Galapagos?

The influence of humans has brought several wild species to the point of extinction. These include the giant tortoise, sperm whale, and the fur seal.

# EXPLORATION ACTIVITY: GO FISH

## MATERIALS NEEDED

- 1 clear and 1 gray 8½" x 11" acetate sheet (see Pre-lab Materials for ordering information)
- Bandanas for each student (from Engagement activity)
- Go Fish handout for each student (with frequency data table)
- 1 Go Fish tally sheet

## INSTRUCTIONS FOR TEACHERS

- In the activity, students will pretend to be predatory animals whose favorite food is “acetate animals.”
- To prepare the “food,” cut the gray and clear acetate sheets into squares. You will need 200 squares of gray and 200 squares of clear. (Other colors may be substituted depending on the coloration and type of flooring.)
- In the adjacent lab room (which represents the habitat for the fish population), place all the squares randomly on the floor, equally distributing the gray and clear squares. Make sure the lights are off or dimmed so when the students enter they do not notice the squares on the floor prior to the beginning of the activity.
- Each student has received a bandana for the Galapagos “trip” and will use this bandana as a blindfold. After students have been blindfolded, they need to be led into the adjacent room. Depending on the size of the class and the size of the other room, the students may need to be taken into the room in small groups. Position the students around the room so that each student has an individual area, or “niche,” in which to “fish.”
- Once the students are in place, have them remove their blindfolds, turn on the light (which represents the daylight in which they search for food), and instruct the students to gather as many pieces of acetate as possible within 10 seconds. After 10 seconds, dim the light again and instruct the students that the “sun has gone down” and they can no longer search for food.
- The students will then return to the classroom and count the color of each acetate animal they captured and then subtract that amount from the number of organisms before selection. Record the data for each individual student on the tally sheet. You can have students come forward to record their data or take the data orally and record it. You may choose to put the tally sheet on an overhead for convenience. Total the data for the entire class on the tally sheet and have students record the data in the frequency data tables on their handouts. This information will be used to complete the rest of the table to determine the frequency of the trait after the first selection and to answer the questions that follow. (You will need to provide to students the number of organisms before selection.)
- Point out to the students that they will determine the number of organisms left after the first selection. Also discuss with your students the variations in predator strategy used by the students, such as selective (picking up pieces individually) or nonselective (scooping up all the pieces possible). Repeat the activity in order to compare the results from no prior knowledge of the “feeding” (1st selection) to an understanding of the “fishing” objective (2nd selection).

## TALLY SHEET — GO FISH

Record the data for the individual students on the tally sheet. Then use this information to fill in the frequency data table.

Results after first selection				Results after second selection				
Student	# Smoky	# Clear	Total	Student	# Smoky	# Clear	Total	Final total
1.				1.				
2.				2.				
3.				3.				
4.				4.				
5.				5.				
6.				6.				
7.				7.				
8.				8.				
9.				9.				
10.				10.				
11.				11.				
12.				12.				
13.				13.				
14.				14.				
15.				15.				
16.				16.				
17.				17.				
18.				18.				
19.				19.				
20.				20.				
21.				21.				
22.				22.				
23.				23.				
24.				24.				
<b>Total</b>				<b>Total</b>				

# EXPLORATION ACTIVITY: GO FISH

## INSTRUCTIONS FOR STUDENTS

You will be blindfolded and led into an adjacent lab room, which is the habitat for a population of acetate “animals.” These animals are all the same species. The variations in color of these animals are due to genetic inheritance. The allele for smoky (gray) is dominant over the recessive allele for clear (no coloration) in these animals.

You are a predator and your favorite food is acetate “animals.” You will have a limited period of daylight to search for your prey, so catch as many as you can before the sun goes down. After sundown, you and the other predators in your class will determine the number of each type of animal left after selection.

Directions: First, count each color of the acetate “animals” you selected and then subtract that number from the total number of organisms before selection (200) for each color. Record all the data for each student on the tally sheet. Using the totals for the entire class from the tally sheet, record the data in the table below and complete the rest of the table using the formulas to determine the frequency of the trait after the first selection. Repeat the process for a second selection and compare your results.

Remember that evolution is defined as a change in the gene frequency over a period of time.

**Formula to keep in mind:**  $p + q = 1$   
**p = Frequency of smoky organisms**      **q = Frequency of clear organisms**

FREQUENCY DATA TABLE

Color	Number of organisms before selection	Frequency of trait before selection	Number of organisms left after the 1st selection	Frequency of trait after 1st selection	Number of organisms left after the 2nd selection	Frequency of trait after 2nd selection
Smoky (gray) p						
Clear q						
Total						

*\*To find the frequency of each trait, divide the total number for each column into the number of smoky or clear in that column. Answers will vary depending on the number of organisms left after each selection.*

1. What happened to the frequency of the smoky color? Why?
2. What happened to the frequency of the clear color? Why?
3. Which color of acetate “animal” had the advantage?
4. How would Darwin explain the results of this activity?
5. What type of predator strategy did you use in the activity for the first selection?
6. How did your strategy change with a second selection?
7. Define evolution in your own words.





KEY

## EXPLORATION ACTIVITY: GO FISH

Color	Number of organisms before selection	Frequency of trait before selection	Number of organisms left after the 1st selection	Frequency of trait after 1st selection	Number of organisms left after the 2nd selection	Frequency of trait after 2nd selection
Smoky (gray) p	200	0.5	varies	# of smoky left/total left = s	varies	Same formula as first selection
Clear q	200	0.5	varies	# of clear left/total left = c	varies	Same formula as first selection
Total	400	1	varies	s + c = 1	varies	Same formula as first selection

*\*To find the frequency of each trait, divide the total number for each column into the number of smoky or clear in that column. Answers will vary depending on the number of organisms left after each selection.*

1. What happened to the frequency of the smoky color? Why?

Decreased. Because the smoky-colored pieces of acetate were easier to see and therefore “catch” (if the floor tile is light colored), so there were fewer smoky animals left after the selection.
2. What happened to the frequency of the clear color? Why?

Increased. Because the clear acetate pieces were more difficult to see (if on a light-colored floor tile), there were more clear animals left after the selection.
3. Which color of acetate “animal” had the advantage?

The clear color had the advantage because they were more difficult to see on a light-colored floor and, therefore, they blended into the surroundings.
4. How would Darwin explain the results of this activity?

Variation and natural selection.
5. What type of predator strategy did you use in the activity for the first selection?

Student answers will vary, depending on whether they picked up the pieces of acetate individually (selective predator strategy) or whether they simply scooped up the pieces (nonselective predator strategy).
6. How did your strategy change with a second selection?

Student answers will vary.
7. Define evolution in your own words.

(Student answers will vary.) Evolution is defined as a change in the gene frequency over a period of time.

## EXPLANATION ACTIVITY

### INSTRUCTIONS FOR TEACHERS

Upon completion of the Go Fish activity, the class will complete the data table together. Ask each group to present an oral summary of its findings. Their presentations will provide a launching pad to present scientific explanations and terminology. Questions to be addressed are as follows:

1. Did one color of acetate “animal” have an advantageous variation?
2. What happened to the frequency of the clear color? Why?
3. How might Darwin have explained the change in the acetate “animal” population?
4. How would you define evolution?

## ELABORATION ACTIVITY

### MATERIALS NEEDED

- PowerPoint presentation part 2
- Discussion questions for teacher reference
- (Optional) Discussion question handout for each student
- (Optional) Inexpensive backpack or suitcase
- 1 set “Family Figures”
- Assorted items to pull from backpack (possible examples include picture of young Darwin, trip journal, family figures for family trip, images or models of mentioned animals)

### INSTRUCTIONS FOR TEACHERS

All travelers return home from a trip with memorabilia of their journey, from photographs and journals to souvenirs and the information they learned during their visit. As students look back on their trip (the information from travel packet materials, their “trip journal” in the form of the completed group report sheet, and information from the Elaboration activity presentation) they will pull out many examples of lessons learned on their expedition.

During this elaboration phase, you will help students sift through their trip “memorabilia.” In the class discussion that ensues, they will have the opportunity to correctly use technical vocabulary and apply scientific reasoning.

Use the second part of the PowerPoint presentation on the module CD to guide the discussion along (you may also print overheads or copies of the slides to distribute if a presentation is not feasible). Refer to the Discussion Questions for Elaboration for prompts to ensure student involvement. (You may distribute copies of the Discussion Questions

before beginning so that students can write responses as you go or use the questions verbally as prompts.) In keeping with the theme of unpacking from a trip, you may wish to pack a small backpack with some materials to pull out at key moments that will facilitate the class discussion. Depending on the item, you may find it useful to pass around the room or post on a wall or black/white board. Examples are provided with the Discussion Questions but use your imagination for additional possibilities.

Some of the concepts to be discussed are as follows:

### Darwin’s Theories of Evolution

- Descent with modification (Examples: marine iguana, once a land dweller; flightless cormorant)
- Natural selection
  - Survival of the fittest/ adaptations, fitness/ extinction
  - Can occur quickly (peppered moth, coat color of mice, multi-drug resistant TB)



## What Causes Natural Selection

- Variations; sources of variation; reproductive potential; limited resources
- Struggle for resources

## Speciation

- Adaptive radiation examples: finches, giant tortoises, prickly pear
- Geographical barrier
- examples: Kaibab and Abert Squirrels
- Genetic drift
- Isolating mechanisms—behavioral (example: boobies); habitat; temporal; mechanical



## Evidences of Evolution

- Paleontology, biogeography, embryology, comparative anatomy, molecular biology

## Relatedness vs. common ancestor

- Example: the family figures



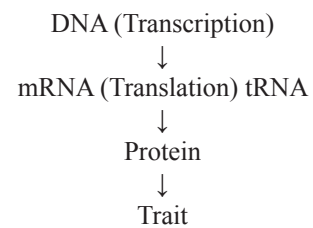
## Biochemical Similarities

- Chimpanzees and humans have 2% difference in their DNA sequence
- In primates, DNA sequences change about 1% every 3 million years

## Similarities in Body Structure

- Homologous (different function/same structure — common ancestor) and analogous structures (same function/different structure — different ancestor)

## The flow of genetic information in the cell



## DISCUSSION QUESTIONS FOR ELABORATION

1. Explain what is meant by Darwin's Theory of Descent with Modification.

- Give examples
- What are the implications of Darwin's theory of natural selection?
- Can you think of examples that illustrate this theory?
- What did Darwin mean when he described certain organisms as being more fit?
- What kinds of evidence did Darwin use to support his theory?

2. What causes natural selection? Give examples of adaptations.

3. What does natural selection equal?

4. Give examples of findings by other biologists that support Darwin's hypothesis.

5. What causes speciation? Give examples.

6. Discuss the evolution of Darwin's finches.

7. What kinds of evidences support evolution?

8. What are homologous structures, analogous structures, and vestigial organs? Provide examples and discuss the relevance of each to evolution.

9. How can two species that look very different from one another be closely related?

10. Distinguish between related and descended.

11. How is DNA used to show evolutionary similarities in species?





## DISCUSSION QUESTIONS FOR ELABORATION

1. (Pull a picture of young Darwin from the backpack or refer to slide 28. This reminds students of Darwin's theories of evolution.)

Explain what is meant by Darwin's Theory of Descent with Modification. (slide 29)

**New species are modified descendants of older species.**

- Give examples

**Marine iguanas, giant tortoise, flightless cormorants, finches.**

- What are the implications of Darwin's theory of natural selection?

**Those best adapted to the environment will survive and reproduce.**

- Can you think of examples that illustrate this theory?

**Finches, marine iguanas, prickly pears.**

- What did Darwin mean when he described certain organisms as being more fit?

**Fit organisms are those that are better suited to survive in their environment and pass on their traits to the next generation.**

- What kinds of evidence did Darwin use to support his theory?

**Darwin used observations of species and the fact that the earth has changed over time. He also argued that fossil records, the geographical distribution of species, homologous structures of living species, and similarities in early development, provided evidence for his theories.**

2. (Pull from the backpack a well-worn trip journal. Read a "journal entry" about what causes natural selection; for example: "From my observations, it seems to me that four main factors influence or cause this phenomenon of natural selection.")

- What causes natural selection? (slide 30)
- There are variations among individuals within a population that are inherited.
- Populations possess enormous reproductive potential.
- Resources are limited.
- There is a struggle for resources; only the fit individuals survive.
- Give examples of adaptations.

**Adaptations are inherited characteristics that increase an organism's chance of survival. Example-Variations in the beaks and feet of the finches.**

3. What does natural selection equal? (slide 31)

**Natural selection = Populations + Environment**

**Populations exhibit variations and possess enormous reproductive potential.**

**The environment has limited resources and there is competition for these resources.**

4. Give examples of findings by other biologists that support Darwin's hypothesis.

- Natural selection in the peppered moth before and during the industrial revolution. (slide 32 or figure 1)
- Experiment conducted at the University of Michigan studying barn owls' success at catching buff-colored mice and gray mice on differing colors of soil. (slide 33 or figure 1)
- Antibiotic resistant bacteria.

5. What causes speciation? Give examples. (slide 34)

**Isolating Mechanisms** (slides 35-36 or figure 2)

- **Behavioral Isolation** — two populations are capable of interbreeding but have different courtship rituals or other strategies that involve behavior. Example: boobies.
- **Geographical Isolation** — two populations separated by geographical barriers. Example: Kaibab and Abert Squirrels. (slide 37 or figure 2)
- **Temporal Isolation** — two species reproduce at different times. Example: boobies; three different but similar species of orchids.
- **Mechanical Isolation** — occurs when male and female genitalia are structurally incompatible. Example: flower structures select for different pollinators.

**Adaptive Radiation** — process by which a single species or small group of species evolves into several different forms that live in different ways. (slide 38 or figure 3)

**Genetic Drift** — in a small population, individuals that carry a particular allele may leave more descendants than other individuals, simply by chance. Over time, this allele may become more prevalent in the population.

6. Discuss the evolution of Darwin's finches. (slides 38-39 or figure 3)

Speciation in the finches occurred by a new population which became geographically isolated; this resulted in changes in the new population's gene pool, reproductive isolation, and competition for resources.

7. What kinds of evidences support evolution? (slide 40)

- Paleontology — study of fossils reveals the prehistoric existence of extinct species.
- Biogeography — study of the distribution of organisms; similar selection pressures, different animals end up evolving certain features in common.
- Embryology — similar stages in development (ontogeny) among related species helps establish evolutionary relationships (phylogeny).
- Comparative anatomy — describes two kinds of structures that contribute to the identification of evolutionary relationships among species (homologous structures, analogous structures).
- Molecular biology — examines the amino acid sequences of DNA and proteins to determine evolutionary relationships between species; even small changes in nucleotides can tell us how organisms are related.

8. What are homologous structures, analogous structures, and vestigial organs? Provide examples and discuss the relevance of each to evolution. (slide 41)

**Homologous Structures** — structures that have different mature forms but develop from the same embryonic tissue; provide strong evidence for common ancestry.  
Example: forelimbs of vertebrates.

**Analogous Structures** — structures that have similar functions but different origins; do not indicate common ancestry.  
Example: wings of a bird and wings of an insect.

**Vestigial Structures** — organs that show no useful function in an organism provide strong evidence for common ancestry; suggest that an organism's ancestor once used that organ.  
Example: miniature legs, tails, or other structures.

9. How can two species that look very different from one another be closely related?

Two species are closely related if they share a common ancestry. If one species experiences natural selection in a particular kind of environment, such as an aquatic environment, it may look different from its related species in a different environment. Example: marine iguana and the land iguana.

10. Distinguish between related and descended. (figure 4, figure 5 or slide 42, and family figures. Cut out the images of the eight family members found in the notebook or on the module CD. For added durability, laminate each figure or mount it on foam board. Use figure 4: How Did We Evolve, as a guide to place each figure on the wall, black or white board as you describe how this family member relates to the others.) *NOTE: Students often make the mistake of thinking that humans are descended from apes. After the family tree exercise, figure 5 or slide 42 can help to demonstrate humans' and apes' descent from a common ancestor.*

Use the example of the family tree.

11. How is DNA used to show evolutionary similarities in species? (slide 43, figure 5 or slide 42)

DNA is passed directly from common ancestors to their descendants and controls development of traits. Primate DNA sequencing changes about 1% every three million years. By looking at protein sequences we can tell that humans and chimpanzees have a DNA sequence that is 98% identical; thus we can tell that humans and chimpanzees shared a common ancestor until between five and six million years ago.

## FIGURE 1: PEPPERED MOTHS — A CASE OF RAPID NATURAL SELECTION

The peppered moth is a famous example of evolution through natural selection. Before the industrial revolution, the common variety of this moth was white with black speckles (called *typica*), which serves as a good camouflage on lichen-covered tree barks. Starting around 1850, scientists began to notice a dark morph of the peppered moth (called *carbonaria*) and a range of intermediately shaded variant (*insularia*). Over the next 50 years, the dark variety gradually became the most prevalent in regions downwind from large industrial centers, where soot and other air pollutants from factories darkened the bark of trees and killed most lichens.

In 1896, amateur moth collector J. W. Tutt proposed that the dark morph of the moth might be better camouflaged on polluted surfaces than the white morph, making it more difficult for birds to prey on the dark moths and therefore leading to an increase in the frequency of *carbonaria* — a case of natural selection. Subsequent studies, most notably one by Bernard Kettlewell starting in 1953, provided hard evidence to support this theory.



Before the industrial revolution, the light (*typica*) peppered moth had a better camouflage than the dark (*carbonaria*) variety and was the more common variety.



Coal burning during the industrial revolution darkened the bark of trees and killed the lichens, making the light moth more easily detectable to predators. The result was that the dark moths survived predation more frequently and became the more common variety.



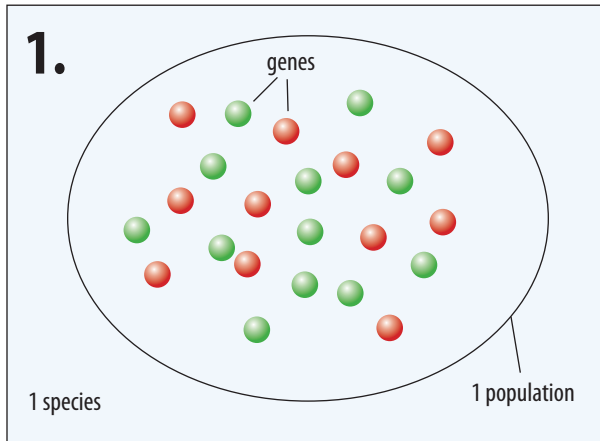
### ANOTHER EXAMPLE

A biologist at the University of Michigan conducted an experiment in which he released buff-colored mice and gray mice into an enclosed room with barn owls. The floor alternated between buff-colored soil and gray soil each day and had an ample supply of sticks for the mice to hide under. Each day, four mice of each color were released for 15 minutes. In 44 trials with each soil type, almost twice as many of the more visible color of mice were caught by the owls.

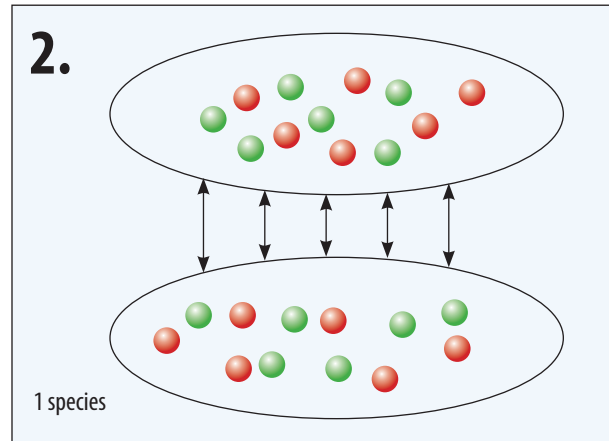


## FIGURE 2: SPECIATION

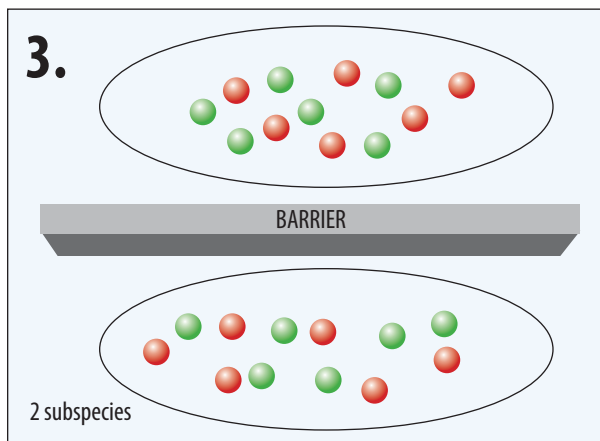
A model of how two populations of a species may become isolated and evolve to produce two species.



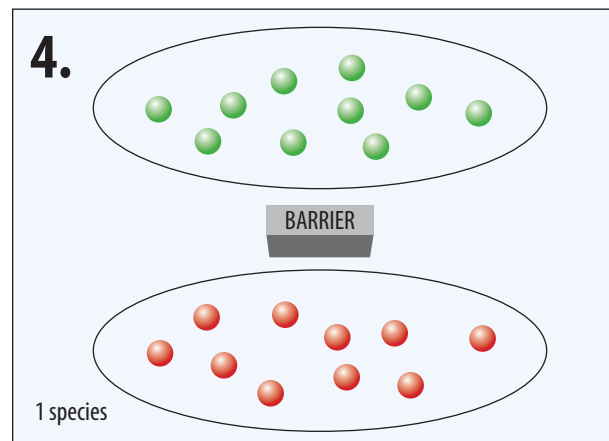
A species made up of one interbreeding population.



In time, the species may expand its range and divide into two or more populations that have little gene exchange.



If a barrier to interbreeding arises, the gene pools become isolated. Over a very long period of time, the different populations become genetically distinct subspecies through mutation and selection.



The populations may be considered separate species when they become so different that they can no longer interbreed even though little or no physical barrier remains.

### SPECIATION IN ACTION

The Kaibab (left) and Abert (right) squirrels are related. When they became geographically isolated on opposite rims of the Grand Canyon, they began to develop differences.



### SPECIATION MAY BE A GRADUAL PROCESS

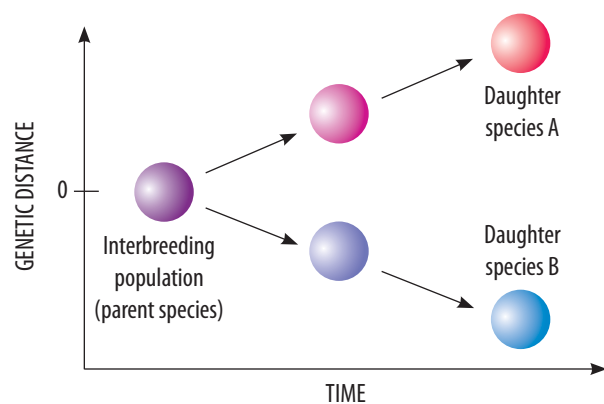
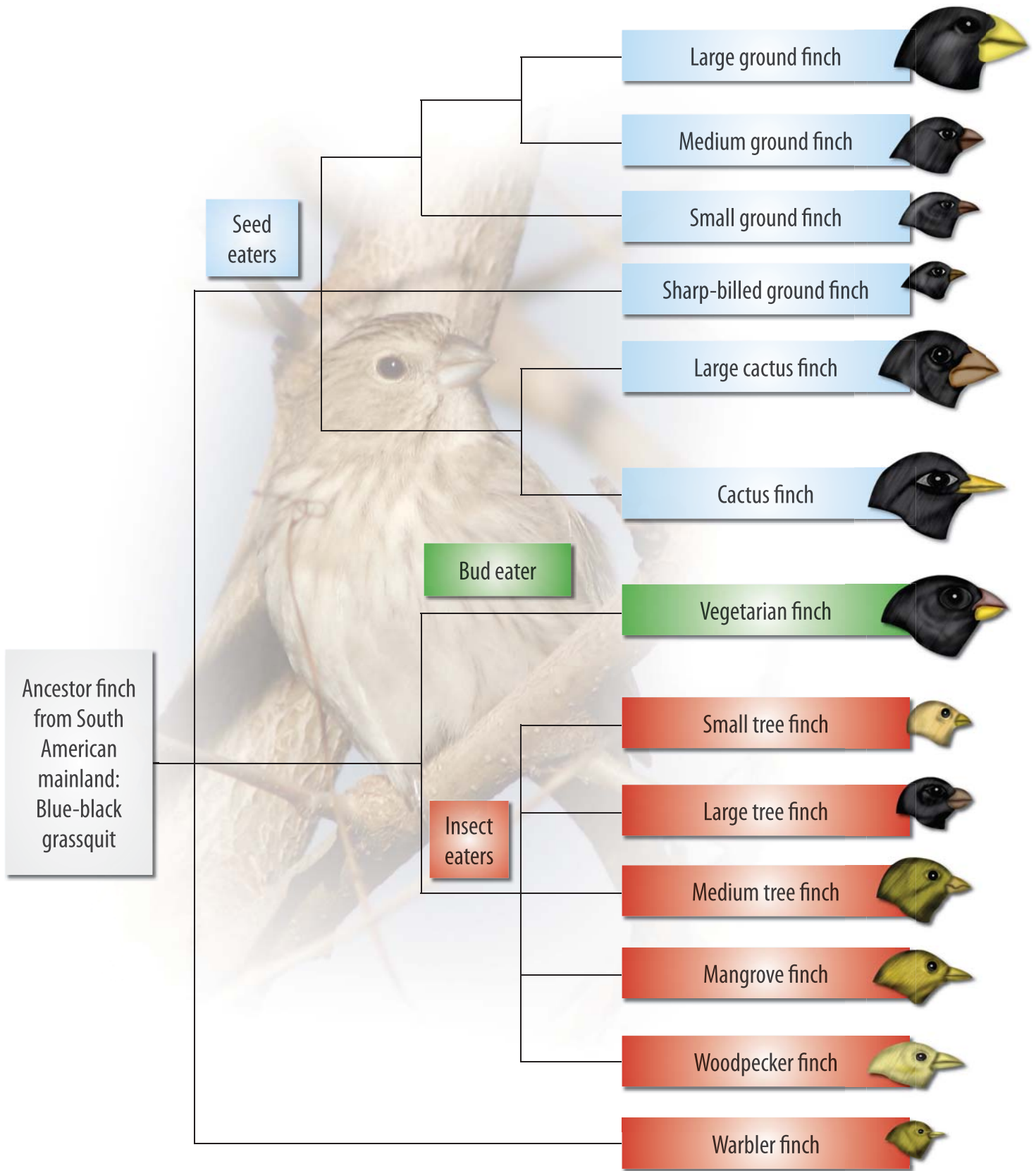




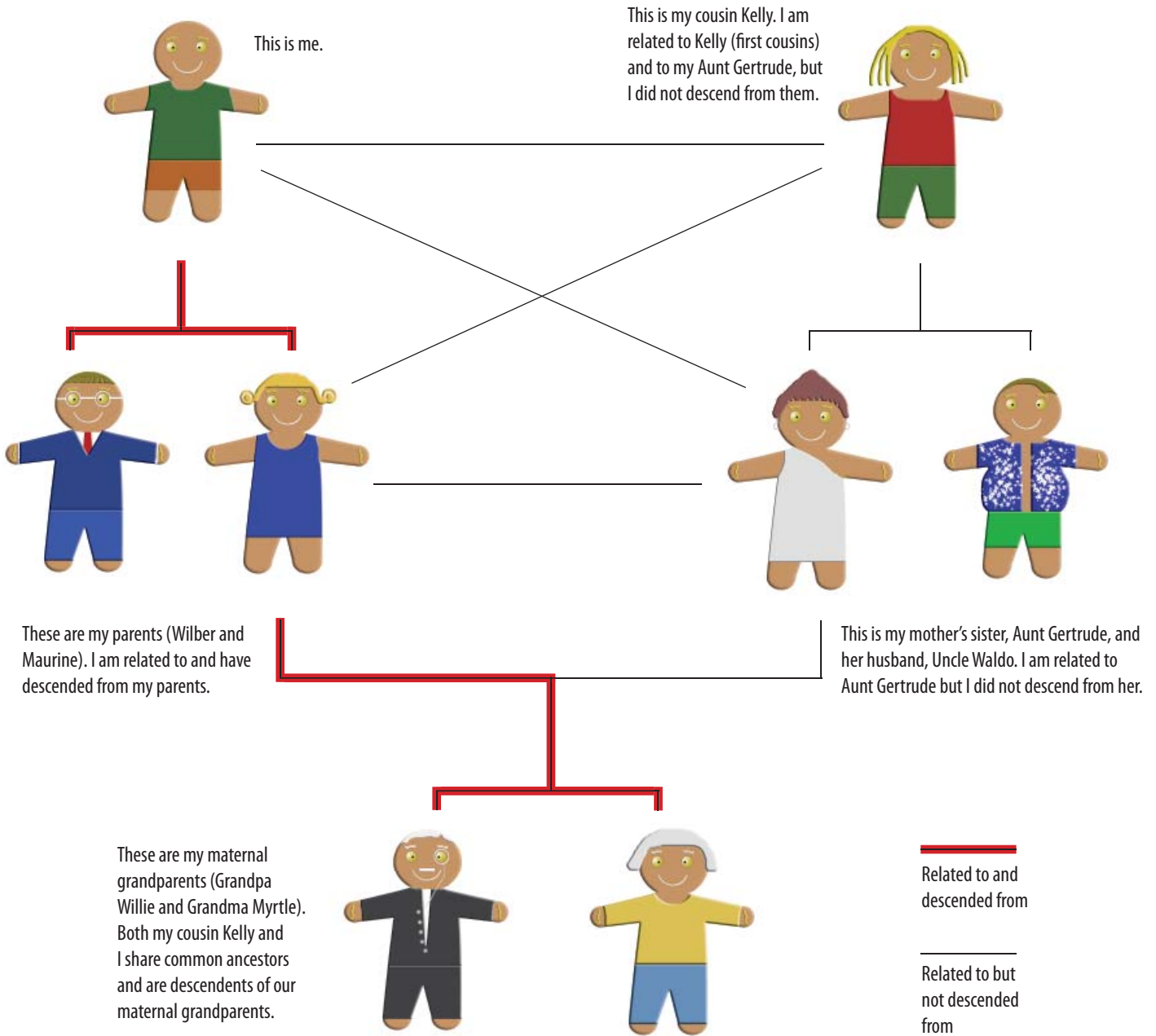
FIGURE 3: EVOLUTION AMONG GALAPAGOS FINCHES

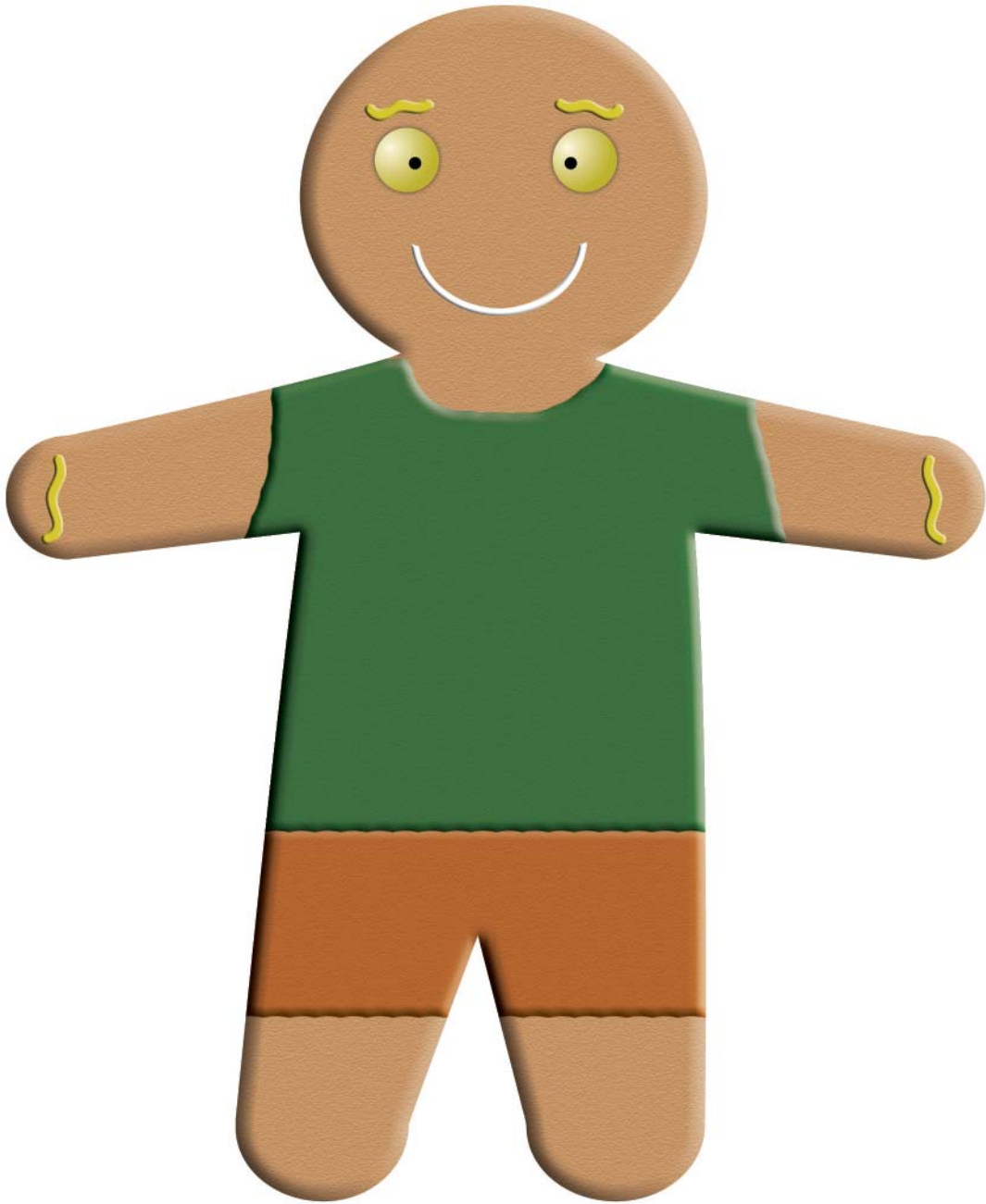


## FIGURE 4: DESCENDED OR RELATED?

To better understand the difference between being descended or related, let us consider my own family.

Put Scotch tape or glue-on magnetic strips on the back of each picture to attach the family figures to the white/chalk board when you introduce them.





ME

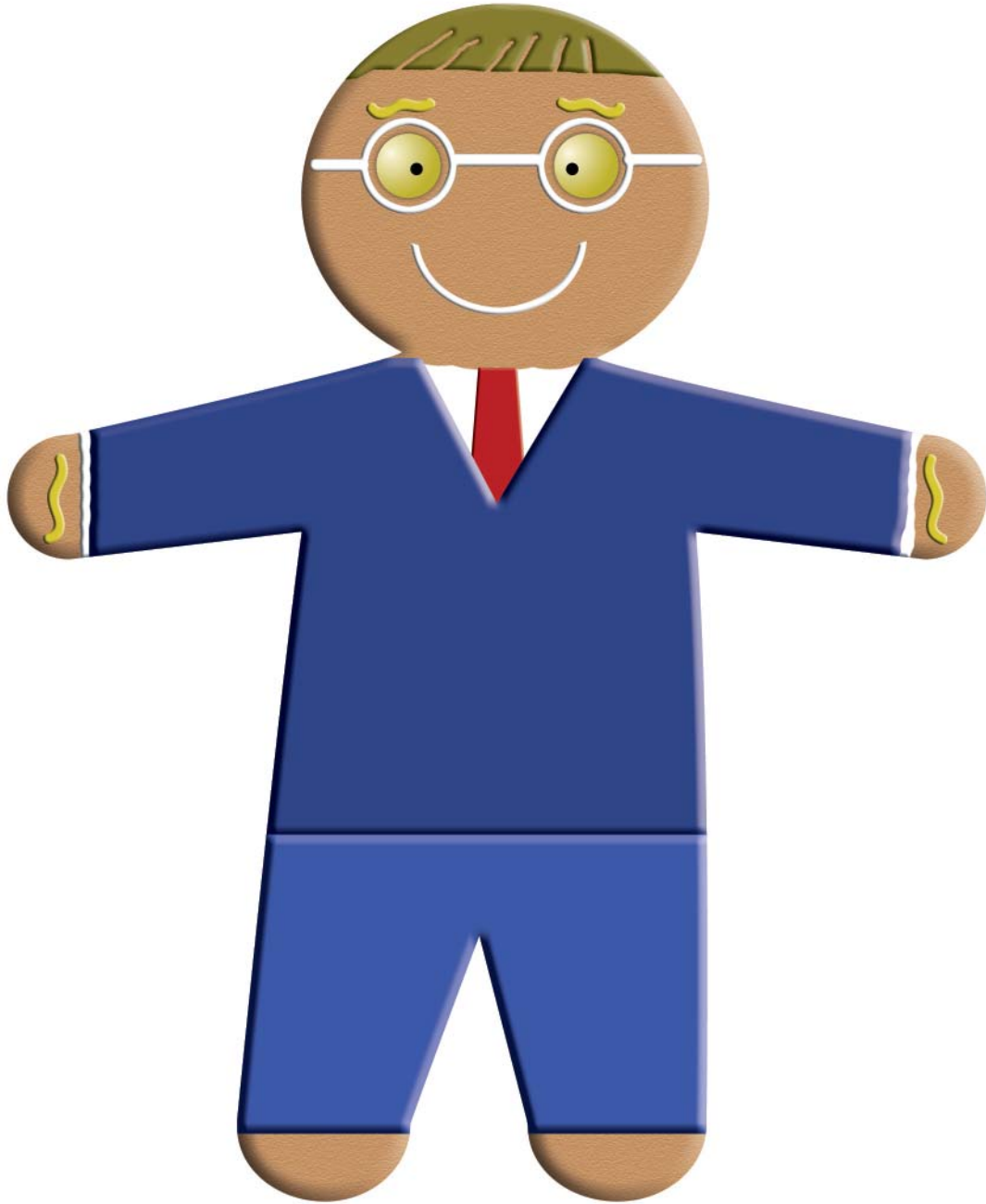






KELLY





WILBER







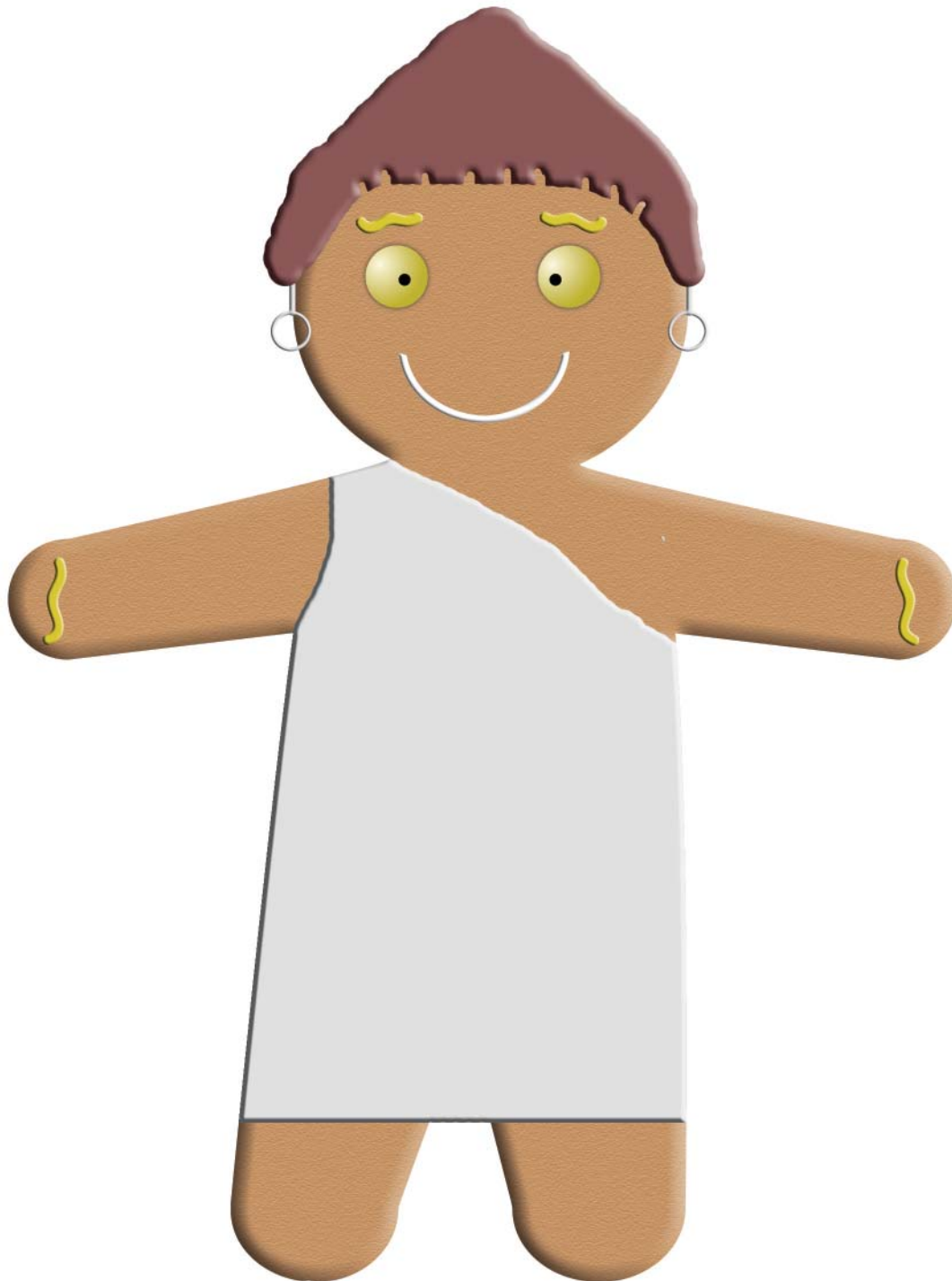
**MAURINE**





WALDO





**GERTRUDE**







WILLIE



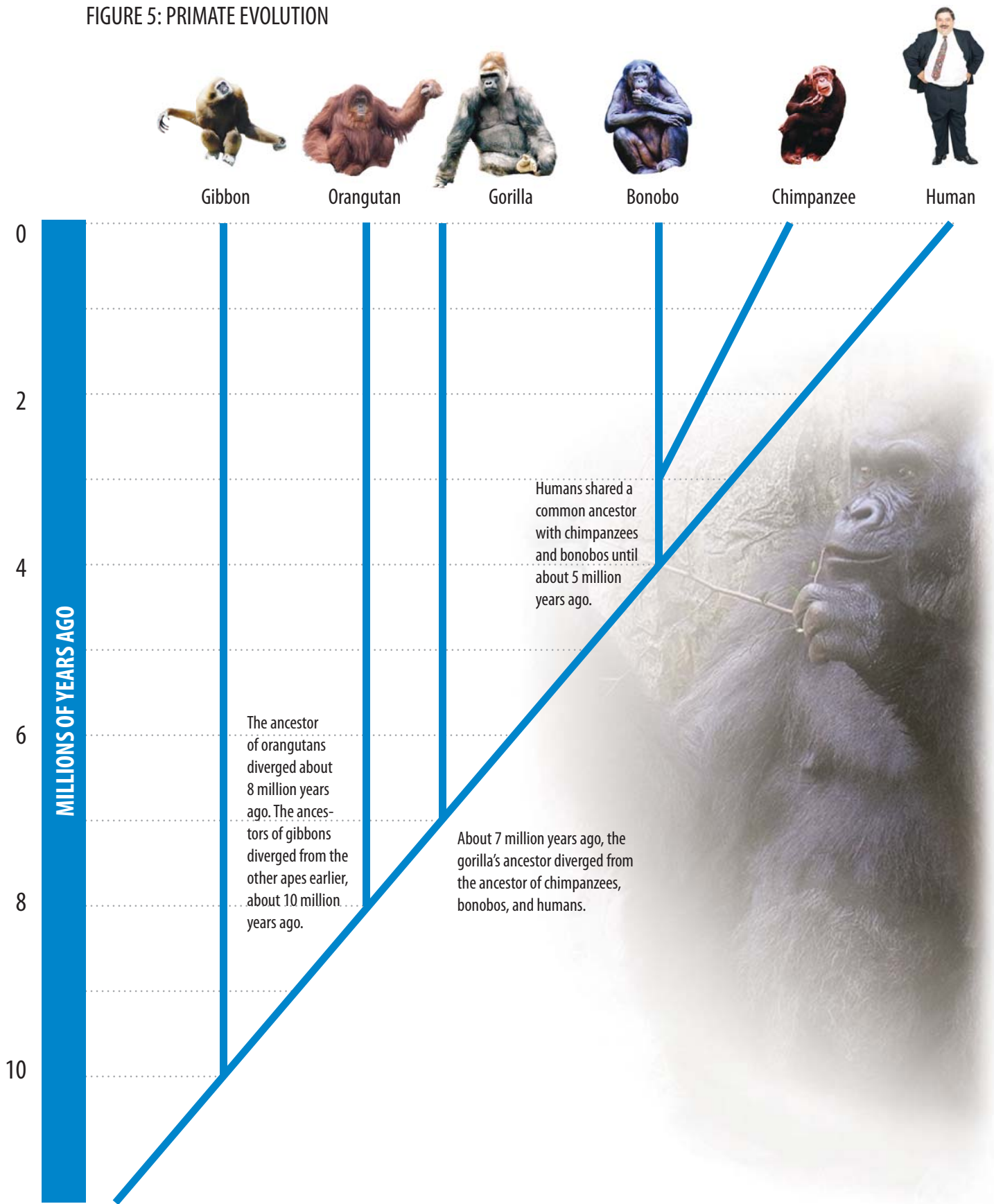


MYRTLE





FIGURE 5: PRIMATE EVOLUTION

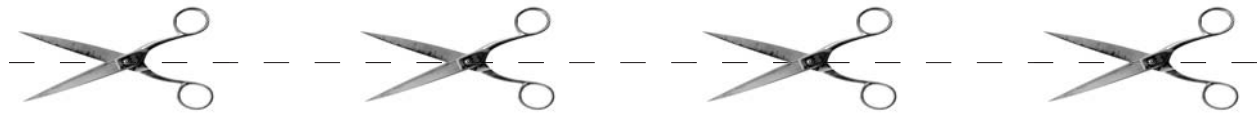


# EVALUATION ACTIVITY

## INSTRUCTIONS FOR TEACHERS

Assemble enough “treasure chests” for the class to have one treasure chest per pair of students (see Pre-lab Materials list for ordering instructions or use small boxes or envelopes). Photocopy the treasure answer strips below or print them from the module CD; make enough copies to place one set of strips in each chest. Cut out each answer, resulting in eight strips of paper per set. Place all eight strips into each treasure chest.

Have the students work in pairs. Give each pair an Evaluation Activity handout and a treasure chest containing a complete set of eight answer strips. (If you are not using the treasure chests, you can simply give a photocopy of the answers sheet to students and have them cut out each strip). Ask them to answer each question by matching it to the appropriate answer strip. Students can paste down strips and turn in for grade. After the class has finished, review the responses with the class.



Sexual recombination and mutations.

New species are modified descendents of older species (examples found on the Galapagos include the marine iguana, once a land dweller, and the flightless cormorant).

Kaibab and Albert squirrels in the Grand Cannon; the different dances performed by the boobies.

Adaptive radiation and isolating mechanisms.

Survival of light- vs. dark-colored peppered moth; survival of buff-colored vs. gray mice; antibiotic resistant bacteria.

Darwin’s finches, land and marine iguanas, different species of prickly pears.

Those organisms best adapted to the environment will survive, reproduce, and pass on their traits to the next generation.

Paleontology, biogeography, embryology, comparative anatomy, and molecular biology.

Name \_\_\_\_\_

Date \_\_\_\_\_

## EVALUATION ACTIVITY

Complete the following questions using the strips from the treasure chest:

1. Explain what Darwin meant by his theory of descent with modification.

2. Explain the meaning of modification by natural selection.

3. Name some experiments that have demonstrated evolution by natural selection.

4. What are the sources of variation that lead to evolution?

5. Evidences for evolution come from the following:

6. What processes can result in speciation?

7. Isolating mechanisms which prevent mating include behavioral, habitat, temporal, mechanical, and geographical barriers. What are examples of behavioral and geographical isolation?

8. Give examples of adaptive radiation found on the Galapagos.

Name \_\_\_\_\_

Date \_\_\_\_\_



KEY

## EVALUATION ACTIVITY

Complete the following questions using the strips from the treasure chest:

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Survival of light- vs. dark-colored peppered moth; survival of buff-colored vs. gray mice; antibiotic resistant bacteria.

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Paleontology, biogeography, embryology, comparative anatomy, and molecular biology.

6. What processes can result in speciation?

Adaptive radiation and isolating mechanisms.

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Kaibab and Albert squirrels in the Grand Cannon; the different dances performed by the boobies.

8. Give examples of adaptive radiation found on the Galapagos.

Darwin's finches, land and marine iguanas, different species of prickly pears.

## FROM FINCHES TO FISHES IMPLEMENTATION PLAN — WET-LAB

Activity	Estimated Time	Materials/Equipment	Purpose/Objectives/ Essential Question
Engagement Activities:  Fishy Family Tree Activity  Protein Electrophoresis Role Play	20 minutes	Fish with scales handout  Fishy Family Tree handout  Three cards representing three different proteins	<p><b>Purpose:</b> To compare proteins from five different fish species to determine evolutionary relatedness.</p> <p><b>Objectives:</b></p> <ul style="list-style-type: none"> <li>• To describe the structure and function of proteins in relation to gel electrophoresis</li> <li>• To identify proteins present in each of five samples extracted from fish using gel electrophoresis</li> <li>• To interpret the results of gel electrophoresis</li> <li>• To make inferences about the relatedness of fish species based on the similarities of their protein “fingerprints”</li> <li>• To describe the process of protein electrophoresis</li> </ul> <p><b>Essential Question:</b> How are protein “fingerprints” used to determine the evolutionary relationships among different species?</p>
Wet-lab Activity  Practice Loading Wells  Load Fish Samples  Run Gel	60 minutes	Comparative Proteomics Kit I: Protein Profiler module (Bio-Rad Kit #166-2700EDU)  p20 Micropipettes  Equipment for vertical gel electrophoresis	
Staining and Destaining of gel	1 hour staining  Overnight destaining	Staining Tray  Bio-Safe™ Coomassie stain  Water  Rocking Platform (optional)	

### Alignment with NC Competency Goals

Biology	
Goal 1 Objectives 1.01, 1.02, 1.03, 1.04, 1.05 Goal 2 Objectives 2.01, 2.02, 2.03	Goal 3 Objectives 3.01, 3.04, 3.05 Goal 4 Objectives 4.01, 4.02, 4.03



## WET-LAB ENGAGEMENT: A FISHY FAMILY TREE

### MATERIALS NEEDED

- Six color printouts of fish and scales (available on module CD) — one for each student group (you may wish to place these in plastic sheet protectors or laminate for added durability)
- One Fishy Family Tree handout for each student (copied from notebook or printed from module CD)
- PowerPoint presentation part 3 (slide 46) or overhead of figure 7 from module notebook or CD

### TEACHER INSTRUCTIONS

Divide the class into six groups and provide each group with a Fish with Scales handout. Distribute a Fishy Family Tree handout to each student. Have each group examine the images of the fish and their scales and work together to fill out the data charts on their individual handouts. Each student should then answer questions 1-3 individually. When students have finished, review the fish evolutionary tree (slide 46 of PowerPoint presentation part 3 or figure 7).

## A FISHY FAMILY TREE

We know that catfish, tuna, and swordfish are all fish and that all three share an ancestor. But which two of these fish share a more recent ancestor? In other words, which two of these fish are most closely related?

To find out, examine the illustrations of these three fish and of their scales. Then fill out the data chart below.

Kind of Fish	Tail Fin Shape (fan shaped vs. lobed)	Presence of Barbels ("whiskers" present vs. absent)	Scale Type (cycloid, ctenoid, etc.)
Catfish			
Tuna			
Swordfish			

1. Which two fish do you think are most closely related?

2. Please explain your reasoning.

3. Label the evolutionary tree below to show your hypothesis.  
(Write the name of one kind of fish in each of the three boxes.)

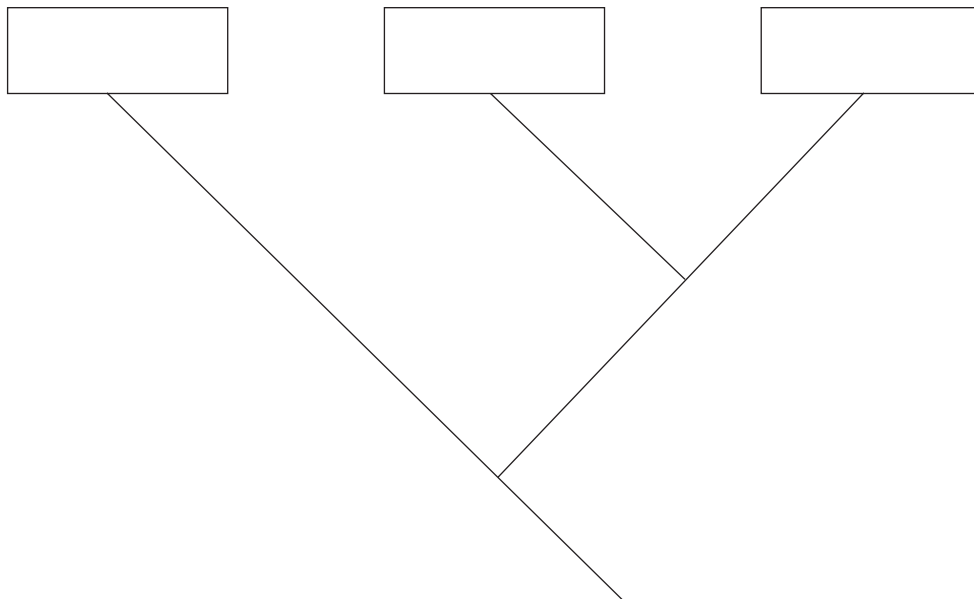
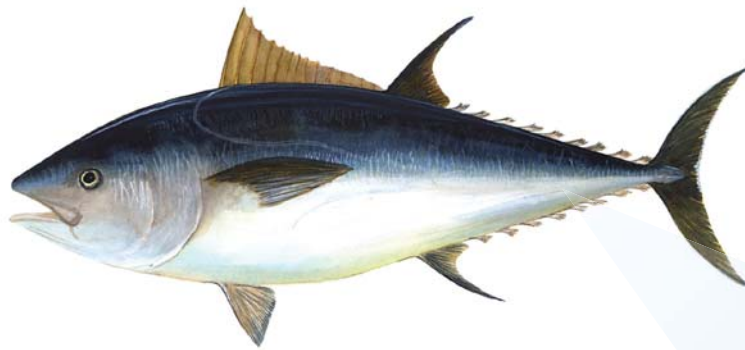


FIGURE 6: FISH WITH SCALES



Channel catfish  
*Ictalurus albidus*

Scaleless



Bluefin tuna  
*Thunnus thynnus*

Ctenoid scales  
Have a toothed edge



Swordfish  
*Xiphias gladius*

Ctenoid scales  
Have a toothed edge





KEY

## A FISHY FAMILY TREE

We know that catfish, tuna, and swordfish are all fish and that all three share an ancestor. But which two of these fish share a more recent ancestor? In other words, which two of these fish are most closely related?

To find out, examine the illustrations of these three fish and of their scales. Then fill out the data chart below.

Kind of Fish	Tail Fin Shape (fan shaped vs. lobed)	Presence of Barbels ("whiskers" present vs. absent)	Scale Type (cycloid, ctenoid, etc.)
Catfish	Lobed	Present	Scaleless
Tuna	Fan shaped	Absent	Ctenoid
Swordfish	Fan shaped	Absent	Ctenoid

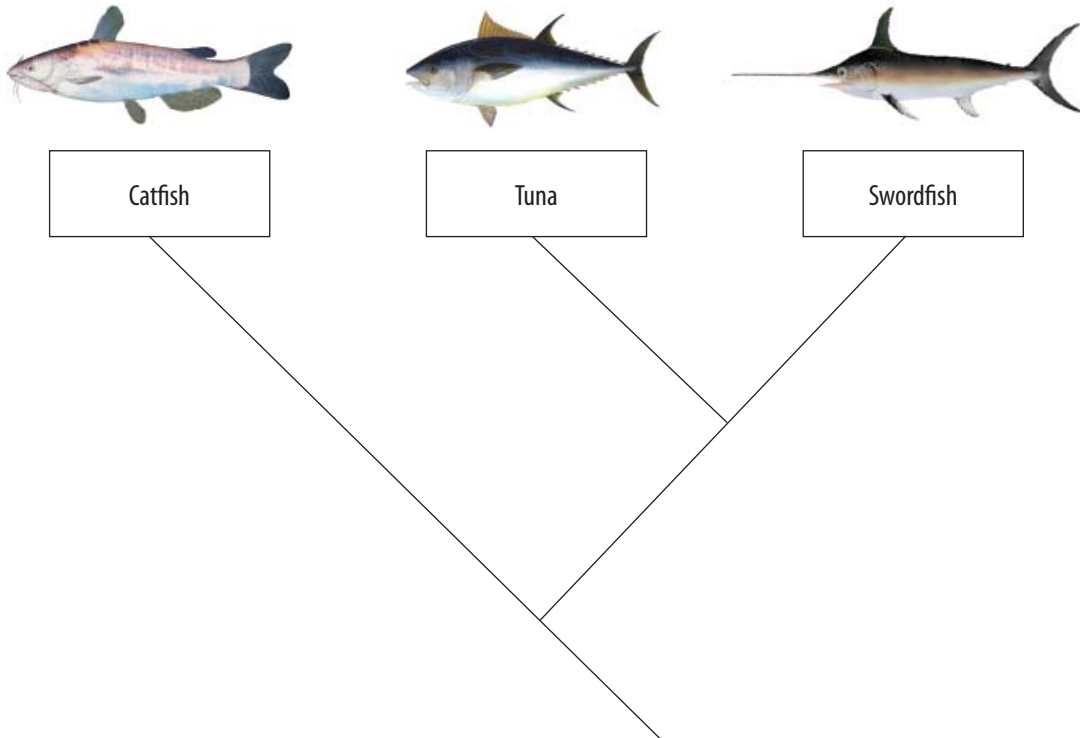
1. Which two fish do you think are most closely related?

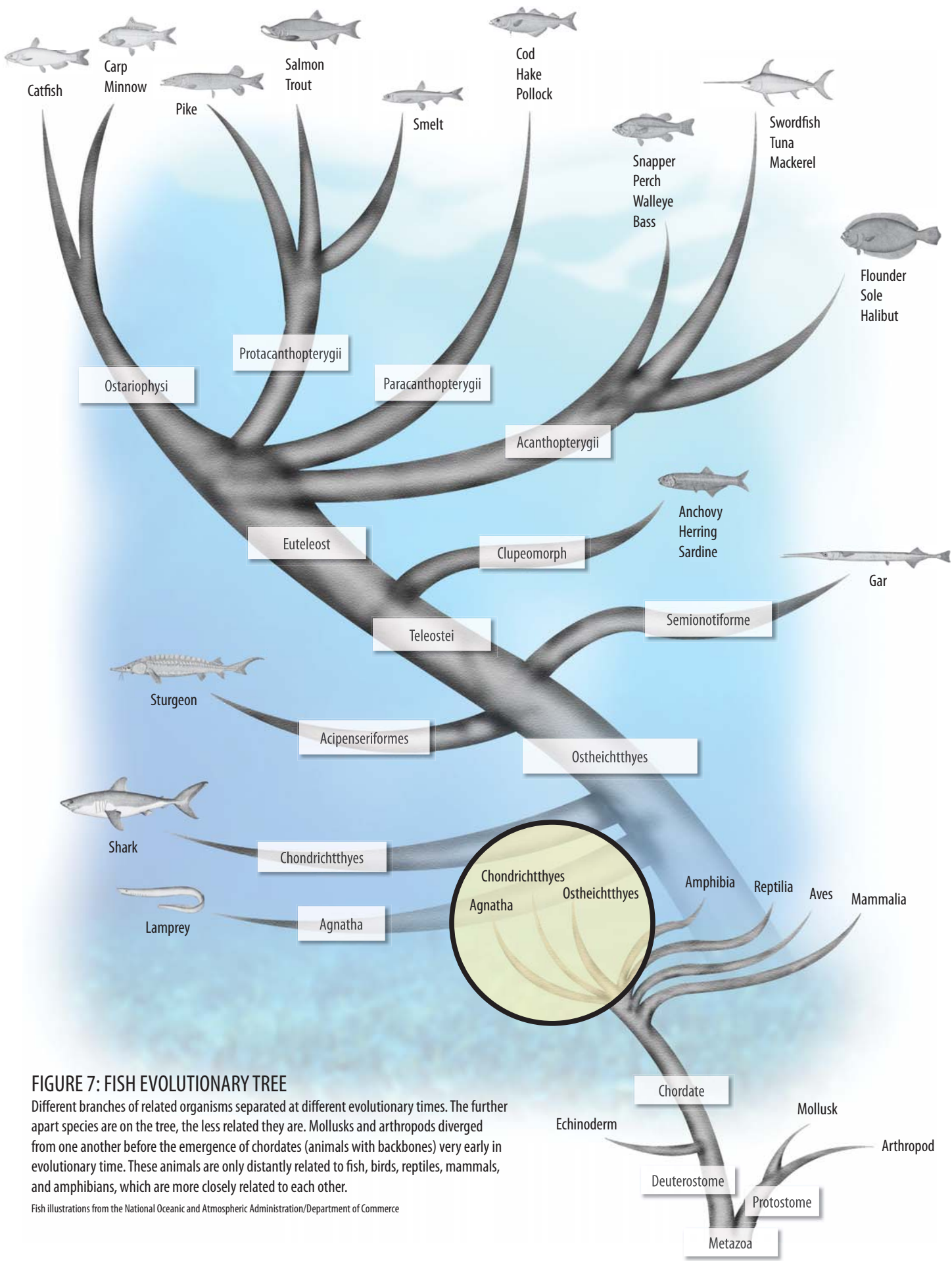
Tuna and swordfish.

2. Please explain your reasoning.

Similar traits.

3. Label the evolutionary tree below to show your hypothesis.  
(Write the name of one kind of fish in each of the three boxes.)





**FIGURE 7: FISH EVOLUTIONARY TREE**

Different branches of related organisms separated at different evolutionary times. The further apart species are on the tree, the less related they are. Mollusks and arthropods diverged from one another before the emergence of chordates (animals with backbones) very early in evolutionary time. These animals are only distantly related to fish, birds, reptiles, mammals, and amphibians, which are more closely related to each other.

Fish illustrations from the National Oceanic and Atmospheric Administration/Department of Commerce



## BACKGROUND FOR THE WET-LAB

Proteins account for more than 50% of the dry weight of most cells, and they are instrumental in almost everything cells do. Proteins are used for structural support, storage, transport of substances, signaling from one part of the organism to another, and movement and defense against foreign substances. In addition, as enzymes, proteins selectively accelerate chemical reactions in cells.

### KEY CONCEPT

Gel electrophoresis is a group of techniques used to separate molecules based on physical characteristics such as size, shape, or isoelectric point.

In gel electrophoresis, separation of charged molecules is achieved by subjecting the charged molecules to an electric current that forces them to migrate through a matrix (the gel). The two types of gel matrices used in molecular biology applications are agarose and polyacrylamide. Agarose is commonly used to separate large fragments of DNA. Polyacrylamide has a greater resolving power and is commonly used for separating proteins.

The behavior of molecules during gel electrophoresis depends on their size, shape, and net charge. Linear DNA molecules have uniformly negatively charged backbones and a shape that normally varies only in its length, so that migration is directly dependent on the size of the DNA fragment. With proteins, the story is different. The net charge of a protein is dependent on its amino acid content; proteins can carry a positive net charge, negative net charge, or they may be neutral. Similarly, the shapes of proteins vary widely. Furthermore, a protein may consist of several polypeptide sub-units held together by hydrogen bonds, hydrophobic interactions, and/or disulfide bridges. Therefore if proteins in their native configurations are electrophoresed, they will not all necessarily migrate in the same direction, and the distances migrated will not be solely a function of their sizes. Thus gel electrophoresis of native proteins cannot be used to determine molecular weights of proteins, but it can provide other information on characteristics of the protein in a mixture.

To make protein migration rates a function of molecular weight, it is necessary to impose a uniform shape and charge on all proteins in a mixture. This goal can be largely achieved by treating the protein mixture with the detergent sodium dodecyl sulfate (SDS). If a

sample mixture is treated with hot SDS, this disrupts all the hydrogen bonds maintaining the protein's three-dimensional shape. If the sample is simultaneously treated with a reducing agent, such as beta-mercaptoethanol, disulfide bridges will also be broken, leaving the protein a linear chain of amino acids. The SDS binds to the protein backbone without regard to amino acid sequences, imparting a uniform negative charge to the molecules. Under these conditions, all the proteins in a mixture assume the same shape and charge. During electrophoresis, they migrate toward the positive pole at a rate proportional to the log 10 of their molecular weights.

Gel electrophoresis of proteins does not provide any direct information about amino acid sequences, and so cannot be used in a precise way to reconstruct evolutionary history. The overall protein fingerprints obtained from different species are more similar when closely related species are compared. The evolution of different groups of fish and the varying degrees to which they are related are a topic of ongoing study.

## WET-LAB ENGAGEMENT



### ENGAGE

To demonstrate the concept of protein electrophoresis, have three students come to the front of the room. Present each student with one of the three cards following this section (also on the module CD). Each card represents a protein molecule with a different molecular weight. Have students move as their proteins would move through the polyacrylamide gel based on their molecular weight. The smaller the mass of the protein molecule, the farther it moves towards the opposite end of the gel.

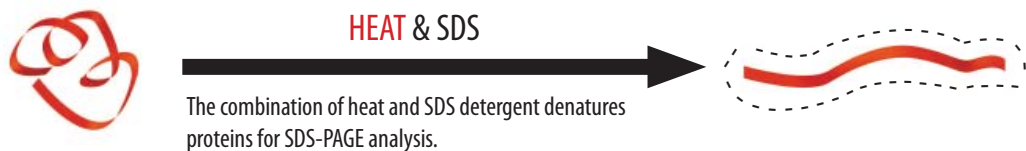
Three protein cards:

<u>Protein</u>	<u>Molecular weight, Dalton, D</u>
1. Myosin (H-chain)	200,000
2. Ovalbumin	43,000
3. $\beta$ -lactoglobulin	18,400 (smaller mass moves farther toward the positive end)

Three factors affect the movement of molecules through a gel during electrophoresis:

- The size of protein molecules: The smaller the protein, the farther down the polyacrylamide gel it will move. The size of a protein molecule is expressed by its molecular weight (in Daltons). Most proteins have masses on the order of thousands of daltons, so we measure them in kilodaltons (kD).
- Charge density of the protein: A ratio of a protein's electrical charge and mass. Charge density affects a protein's mobility through a gel during electrophoresis. Since each protein is made of a unique combination of amino acids, each of which may have a positive, negative, or neutral charge, the net charge of each protein is naturally different.
- Protein structure: Secondary, tertiary (which result from protein folding), and quaternary structure must be disrupted to separate proteins by size. The combination of heat and the detergent SDS denatures the protein's structure. The intrinsic charges of proteins are obscured by placing a strong anionic detergent SDA in both the sample buffer and the gel-running buffer. SDS binds to and coats the proteins and keeps them as denatured linear chains (see Figure 8). In this form, proteins migrate in polyacrylamide gels as if they have equivalent negative charge densities, and mass becomes the only variable affecting the migration rate of the protein. This technique is called SDS-PAGE (sodium dodecyl sulfate-polyacrylamide gel electrophoresis).

FIGURE 8: Sodium Dodecyl Sulfate



### Discussion Questions:

What is a protein?

Proteins are charged molecules; one of the complex organic chemical compounds that form the basis of living tissues; consist of long chains of amino acids connected by peptide bonds to form one or more polypeptides. Examples are enzymes, hemoglobin, and antibodies.

How do proteins function in organisms?

Proteins determine how an organism functions, what it eats, how it looks, and where it lives. Proteins form the basis of living tissues and carry out the thousands of chemical reactions necessary to maintain life.

Do humans and fish have the same proteins?

Actin and Myosin make up the structure and function of muscle found in all animals.

## WET-LAB EXPLORATION



### EXPLORE

(Depending on class time and ability, you may prepare samples in advance or guide students through the preparation.) Before entering the lab, review safety procedures. Give each student a copy of the lab procedure quick guide. Remind them to keep accurate records. In the lab, students will use the technique of protein electrophoresis to discover the relatedness of five types of fish. Using demonstration equipment, briefly describe the process used in the lab.

The purpose of the gel is to act as a medium to slow the rate of movement of the protein molecules, and to provide a lane for each sample to move in a straight line, much like a track. The sample loading buffer contains SDS and dithiothreitol (DTT), which will disrupt all the hydrogen bonds and disulfide bridges leaving the proteins linear with an overall uniform negative charge.

Add samples of fish proteins to designated wells.

Close the lid of the electrophoresis chamber and turn on the electricity.

After 30 minutes, remove the gel from the electrophoresis chamber and place it in a weigh boat for staining. (10 minutes)

Return the stain to the original container.

Flood the gel with destain solution and let it sit overnight until the bands are clearly visible.

## WET-LAB EXPLANATION



### EXPLAIN

After the electrophoresis process, students will analyze the gels. The protein profile for each fish will vary. To facilitate discussion, choose a representative gel. Place the results on an overhead projector. Highlight the bands with a marker. Some sample questions for discussion are as follows:

How would you identify the five proteins based on their movement through the gel?

<u>Protein</u>	<u>Molecular weight, Dalton, D</u>
1. Myosin (H-chain)	200,000 (largest mass travels least)
2. Bovine serum albumin	68,000
3. Ovalbumin	43,000
4. Carbonic anhydrase	29,000
5. $\beta$ -lactoglobulin	18,400 (smallest mass travels farthest)

Which fish seem to have similar protein profiles? How many proteins do they have in common?

Which fish do not have similar protein profiles? What does this tell you about the relatedness of the fish?

## WET-LAB EVALUATION



### EVALUATE

Based upon their findings, ask students to place the names of the five fish on a hypothetical evolutionary tree. Lead a discussion with the class regarding their decisions for placement of fish on the hypothetical evolutionary tree. Remind students that gel electrophoresis of proteins does not provide any direct information about amino acid sequences, and so cannot be used in any precise way to reconstruct evolutionary history. The evolution of different groups of fish and the varying degrees to which they are related are topics of ongoing study.



# Myosin

# 200,000 D





# Ovalbumin

# 43,000 D



**$\beta$ -lactoglobulin**

**18,400 D**





# FROM FINCHES TO FISHES WET-LAB: FISH PROTEIN FINGERPRINTING BY GEL ELECTROPHORESIS

## PURPOSE

To compare proteins from five different fish to determine phylogenic relationships.

- To make inferences about the relatedness of fish based on the similarities of their proteins profiles.
- To demonstrate the process and concept of protein electrophoresis.

## OBJECTIVES

- To perform gel electrophoresis to identify proteins present in each of five samples extracted from fish.
- To interpret the results of electrophoresis.

## TEACHER PREPARATION

Obtain fish samples and distilled water—trip to grocery store.  
Set up student and teacher workstations.

### Student Workstations

1.5 ml micro test tube	6 each	<input type="checkbox"/>
1.5 ml screw-cap micro test tube	7 each	<input type="checkbox"/>
Micropipettes and tips or disposable pipettes	1 each	<input type="checkbox"/>
Fish samples (5 types)	1 gram each	<input type="checkbox"/>
Indelible marking pen, fine point	1	<input type="checkbox"/>
Laemmli sample buffer	1.5 ml	<input type="checkbox"/>
Knife or scissors to cut fish samples	1	<input type="checkbox"/>

### Teacher's (Common) Workstation

Water bath or hot plate set to 95° C	1	<input type="checkbox"/>
Laemmli sample buffer — 30 ml	1	<input type="checkbox"/>
Actin and myosin standards	1 vial	<input type="checkbox"/>
Precision Plus Protein Kaleidoscope prestained standards — 500µl	1 vial	<input type="checkbox"/>

## ACTIN AND MYOSIN STANDARD

A control sample containing actin and myosin proteins is provided in a lyophilized form that is stable at room temperature. Store these proteins, along with the Precision Plus Protein Kaleidoscope standards, in the freezer for long-term safe-keeping. To rehydrate the sample, add 500 µl of Bio-Rad Laemmli sample buffer

to the vial and incubate it at room temperature for five minutes. Transfer the rehydrated actin and myosin sample to a labeled screw cap tube using a disposable plastic transfer pipet. Like the fish protein samples, the actin and myosin samples must be heated for five minutes at 95°C before loading on gels.

## LABORATORY DAY 1: SAMPLE PREPARATION – MUSCLE PROTEIN EXTRACTION

### PROCEDURE

1. Label (with indelible pen) 1.5 ml flip-top microtubes with the names of the fish samples to be analyzed. There should be one labeled tube for each fish sample being prepared for electrophoresis.
2. Add 250  $\mu$ l of Laemmli sample buffer to each labeled tube.
3. For each sample, obtain a piece of fish muscle (avoid skin, fat, and bones) approximately  $0.25 \times 0.25 \times 0.25 \text{ cm}^3$ , and transfer it to the appropriately labeled microtube. Close the lid.
4. Gently flick the microtube 15 times with your finger to agitate the tissue in the sample buffer.
5. Incubate the samples for 5 minutes at room temperature to extract and solubilize the proteins.
6. Pour the buffer containing the extracted proteins, but not the solid fish piece, to a labeled 1.5 ml screwcap tube. Note: It's not necessary to transfer all of the fluid to the screwcap tube, since only a small volume ( $< 20 \mu$ l) is actually needed for gel loading.
7. Obtain aliquots of the Kaleidoscope (KS) and actin and myosin (AM) standards from your teacher.
8. Heat the fish samples and the actin and myosin (AM) sample in their screwcap tubes for 5 minutes at  $95^\circ\text{C}$  to denature the proteins in preparation for electrophoresis.
9. Store the samples at room temperature if they are to be loaded onto gels within 3-4 hours, or store them at  $-20^\circ\text{C}$  for up to several weeks.

## LABORATORY DAY 2: ELECTROPHORESIS

So far you have extracted, denatured, and given the proteins from fish muscle tissue a negative charge. Now they can be separated according to their molecular weights using gel electrophoresis.

**Electrophoresis:** gel loading, running, and staining

### PURPOSE OF THIS LABORATORY

Generate profiles for various fish species via electrophoresis of extracted protein samples.

Procedure Overview:

1. Reheat fish and actin and myosin standard controls
2. Set up electrophoresis gel boxes
3. Load and run gels
4. Stain gels to visualize protein bands

---

### Laboratory checklist

#### Student Workstations

Fish protein extracts prepared on laboratory day 1	5 each	<input type="checkbox"/>
Prot/Elec pipet tips for gel loading	7 tips	<input type="checkbox"/>
Mini-PROTEAN® 3 electrophoresis module (gel box — runs one or two gels)	1	<input type="checkbox"/>
Buffer dam (if running only one gel/box)		<input type="checkbox"/>
Power supply (200 V constant)	1	<input type="checkbox"/>
2-20 µl micropipette	1	<input type="checkbox"/>
Ready Gel precast gel, 15% – 10 wells	1	<input type="checkbox"/>
Sample loading guides – for 10 well comb	1	<input type="checkbox"/>
Thin metal weighing spatula	1	<input type="checkbox"/>

#### Teacher's (Common) Workstation

**Actin and myosin standard sample, rehydrated	1 vial	<input type="checkbox"/>
Precision Plus Protein Kaleidoscope prestained standards	1 vial	
1X Tris-glycine-SDS (TGS) electrophoresis buffer	As needed	<input type="checkbox"/>
Bio-Safe Coomassie stain for proteins	As needed	<input type="checkbox"/>
Staining trays		<input type="checkbox"/>

\*\*You may already have aliquots at student stations

## LABORATORY DAY 2

### STEP 1: PREPARE SAMPLES, ELECTROPHORESIS GELS, AND GEL BOXES

**Note:** Teachers may have already assembled the gel boxes. If not, follow these instructions.

1. Reheat frozen samples at 80-95°C for 2-5 minutes to redissolve any precipitated detergent.
2. Make sure the comb and the tape along the bottom of the Ready Gel cassette have been removed. If two gels are to be run in one electrophoresis box, place a Ready Gel cassette on each side of the electrode assembly, with the short plates facing the inside of the assembly. If you are running only one gel in the box, place a Ready Gel cassette on one side of the electrode assembly and a buffer dam on the other side. Be sure to place the side of the buffer dam that says “BUFFER DAM” toward the electrode assembly.
3. Open the gates (cams) on the front of the clamping frame. Hold the two Ready Gel cassettes, or one Ready Gel cassette and buffer dam, against the electrode assembly and slide the electrode assembly into the clamping frame.
4. Press down on the outer edge of the electrode assembly, not the gels, while closing the cams of the clamping frame to ensure a seal on the bottom edge of each cassette.
5. Place the assembled clamping frame containing the gel(s) into the gel box tank. Fill the upper buffer chamber, the space between the two gels, with ~150 ml 1X TGS electrophoresis buffer, so the buffer level is above the inner short plates. Check for leaks. If the assembly is leaking, remove the assembled clamping frame, pour off the buffer, reopen the cams, and push down on the electrode assembly again while closing the cams.
6. Pour ~200 ml of 1X TGS electrophoresis buffer into the lower buffer chamber, or tank. Double-check the buffer fill level within the upper buffer chamber.

**Note:** If leakage of the upper buffer cannot be corrected by reassembling the clamping frame in Step 4, the outer chamber can be filled to above the inner small plates, to equalize the buffer levels in both reservoirs. This requires approximately 900 ml of 1X TGS electrophoresis buffer.

### STEP 2: LOAD AND RUN GELS

Place a yellow sample loading guide on the top of the electrode assembly. The guide will direct the pipet tip to the correct position for loading each sample in a well.

Assign samples to wells, loading samples in middle of the gel, where separation is best with the standards on each side. For example, for five fish samples on a 10 well gel, you may choose to follow this guide:

Lane	Volume	Sample
1	Empty	None
2	Empty	None
3	5 µl	Precision Plus Protein Kaleidoscope prestained standard
4	10 µl	Fish sample 1
5	10 µl	Fish sample 2
6	10 µl	Fish sample 3
7	10 µl	Fish sample 4
8	10 µl	Fish sample 5
9	10 µl	Actin and myosin standard
10	Empty	None

To load each sample, use a thin, gel loading micropipette tip to withdraw 10 µl of each protein sample from its tube and gently transfer it into the designated gel well. After loading all samples, remove the sample loading guide, place the lid on the tank, and insert the leads into the power supply, matching red to red and black to black. Set the voltage to 200V and run the gels for 30 minutes.

Record your samples here:

**Lane Sample**

1. Precision Plus Protein Kaleidoscope prestained standard
2. Precision Plus Protein Kaleidoscope prestained standard
3. Precision Plus Protein Kaleidoscope prestained standard
4. \_\_\_\_\_
5. \_\_\_\_\_
6. \_\_\_\_\_
7. \_\_\_\_\_
8. \_\_\_\_\_
9. Actin and myosin standard
10. Precision Plus Protein Kaleidoscope prestained standard

**STEP 3: STAIN AND VISUALIZE THE PROTEINS**

**Gel staining**

1. When gels are finished running, turn off

the power supply and disconnect the leads. Remove the lid and lift out the electrode assembly and clamping frame.

2. Pour out the running buffer from the electrode assembly. Open the cams and remove the gel cassettes.

3. To keep the gel free of contamination from your fingertips, wear gloves to handle the gels from this point on. Lay a gel cassette flat on the bench with the short plate facing up. Cut the tape along the sides of the gel cassette.

Carefully pry apart the gel plates, using a spatula or your fingertips. The gel will usually adhere to one of the plates. Transfer the plate with the gel adhering to it to a tray containing Bio-Safe Coomassie stain, allowing the liquid to detach the gel from the plate. The gel may also be lifted directly (and gently!) from the plate and placed into the stain.

4. Allow the gels to stain for one hour, with shaking if available.

5. Your teacher will discard the stain and replace it with a large volume of water to destain the gel overnight.



Name \_\_\_\_\_

Weigh Boat number \_\_\_\_\_

## FROM FINCHES TO FISHES: DATA/OBSERVATION SHEET

1	2	3	4	5	6	7	8	9	10
Precision Plus Protein Kaleidoscope standard	Precision Plus Protein Kaleidoscope standard	Precision Plus Protein Kaleidoscope standard						Actin and myosin standard	Precision Plus Protein Kaleidoscope standard

### FOCUS QUESTIONS

1. What are some important roles that proteins play in organisms?
2. Why did we apply heat and SDS buffer to the protein samples?
3. Why did we use a polyacrylamide gel instead of an agarose gel?
4. Which protein will travel further, a smaller protein or a larger protein?
5. What will the resulting protein bands help us to determine?



## FROM FINCHES TO FISHES: DATA/OBSERVATION SHEET

1	2	3	4	5	6	7	8	9	10
Precision Plus Protein Kaleidoscope standard	Precision Plus Protein Kaleidoscope standard	Precision Plus Protein Kaleidoscope standard	Fish sample #1	Fish sample #2	Fish sample #3	Fish sample #4	Fish sample #5	Actin and myosin standard	Precision Plus Protein Kaleidoscope standard

### FOCUS QUESTIONS

1. What are some important roles that proteins play in organisms?

Proteins such as enzymes acts as catalysts in biochemical reactions. Proteins also have structural and mechanical functions. Structural proteins include the keratin and collagen found in hair, skin, and fingernails. Actin and myosin are the principal fibrous proteins of muscle tissue; interaction of the actin and myosin brings about muscle contraction and results in movement.

2. Why did we apply heat and SDS buffer to the protein samples?

The combination of heat and the addition of an ionic detergent, SDS (sodium dodecyl sulfate), act to denature the proteins. The detergent coats dissolved proteins and polypeptides with negative charges. Heating causes the proteins to lose their three dimensional structure and change to a linear structure. All of the net negatively-charged proteins move through the gel toward the positive electrode according to their size.

3. Why did we use a polyacrylamide gel instead of an agarose gel?

The polyacrylamide gels are used to separate smaller proteins. DNA and hemoglobin are much larger proteins and are separated using agarose gels.

4. Which protein will travel further, a smaller protein or a larger protein?

Smaller proteins will travel further in the gel.

5. What will the resulting protein bands help us to determine?

Proteins from fish samples that are more closely related will have more bands in common. The size of the bands can be used to determine the amounts of that particular sized protein. Proteins are synthesized according to the genes of an organism's DNA and similar proteins reflect similar DNA.

# PROTEIN PROFILER MODULE — QUICK GUIDE

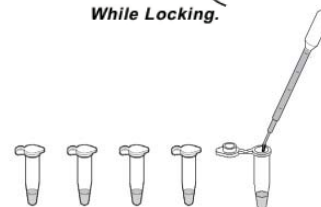
## From Bio-Rad's Comparative Proteomics Kit I: Protein Profiler Module

1. Setup Mini-PROTEAN 3 gel box and add 1x TGS electrophoresis buffer to the chamber.



2. Label one 1.5 ml flip-top micro tube for each of five fish samples. Also label one screwcap microtube for each fish sample.

3. Add 250 µl of Bio-Rad Laemmli sample buffer to each labeled flip-top micro tube.



4. Cut a piece of each fish muscle about 0.25 x 0.25 x 0.25 cm<sup>3</sup> and transfer each piece into a labeled flip-top micro test tube. Close the lids.

5. Flick the microtubes 15 times to agitate the tissue in the sample buffer.

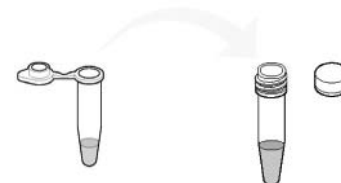


6. Incubate for five minutes at room temperature.

7. Carefully transfer the buffer by pouring from each flip-top micro-tube into a labeled screwcap microtube. Do not transfer the fish!

8. Obtain the Kaleidoscope prestained standards (KS) and the actin and myosin standard from your teacher.

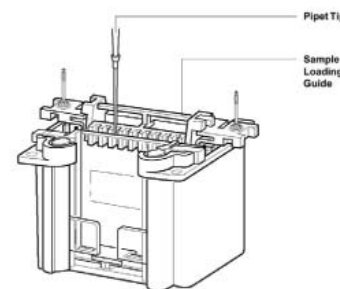
9. Heat the fish samples and the actin and myosin standard (AM) in screwcap microtubes for five minutes at 95°C.



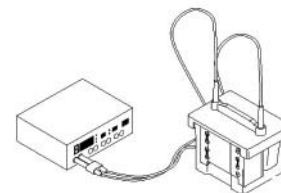
10. Load your gel:

Lane	Volume*	Sample
1 & 2	empty	empty
3	5 µl	Precision Plus Protein Kaleidoscope standards (KS)
4	10 µl	fish sample 1
5	10 µl	fish sample 2
6	10 µl	fish sample 3
7	10 µl	fish sample 4
8	10 µl	fish sample 5
9	10 µl	Actin and myosin standard (AM)
10	empty	empty

\* — DESTINY has found that doubling the volume to 20 µl helps students get enough protein out of the pipettes.

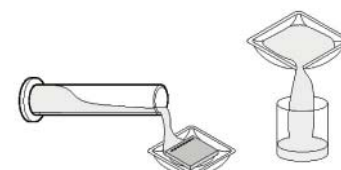


11. Electrophorese for 30 minutes at 200 V in 1x TGS electrophoresis buffer.



12. After electrophoresis, remove gel from cassette and transfer gel to a container with 25 ml Bio-Safe Coomassie stain per gel and stain gel for 1 hour, with gentle shaking for best results.

13. Discard stain and destain gels in a large volume of water for at least 30 minutes to overnight, changing the water at least once. Blue-stained bands will be visible on a clear gel after destaining.



14. Dry gels using GelAir™ cellophane.

## From Finches to Fishes Equipment Needed for Wet-lab

Vendor	Catalog Number	Item	Unit	Price	Minimum Purchase	TOTAL
<b>REQUIRED EQUIPMENT</b>						
Bio-Rad	164-5050EDU	PowerPac™ Basic Power Supply	1	\$325.00	2	\$650.00
Bio-Rad	165-3302EDU	Mini-PROTEAN 3 cell	1	\$238.00	8	\$1,904.00
Bio-Rad	166-0506EDU	2-20 Digital micropipette	1	\$159.00	8	\$1,272.00
Bio-Rad	166-0507EDU	20-200 Digital micropipette (Instructor)	1	\$159.00	1	\$159.00
Bio-Rad	165-3146EDU	10-well Sample Loading Guides	1	\$8.80	8	\$70.40
<b>EQUIPMENT TOTAL</b>						<b>\$4,055.40</b>
<b>REQUIRED CONSUMABLES</b>						
Bio-Rad	166-2700EDU	Comparative Proteomics Kit I: Protein Profiler module*	1	\$125.00	1	\$125.00
Bio-Rad	161-1103EDU	Ready Gel precast gels 15%	1	\$6.75	8	\$54.00
<b>CONSUMABLES TOTAL</b>						<b>\$179.00</b>
* — Protein Profiler module provides enough material for 8 workstations up to 32 students						
<b>OPTIONAL EQUIPMENT</b>						
Bio-Rad	166-0709EDU	Rocking Platform	1	\$575.00		
Bio-Rad	166-0504EDU	Water Bath	1	\$505.00		
Fisher	11-718	Dry Bath Incubator	1	\$251.88		
Fisher	11-718-9	Heating block for 1.5mL microtubes	1	\$84.85		
<b>OPTIONAL CONSUMABLES &amp; REFILLS</b>						
Bio-Rad	166-0010EDU	Actin & Myosin Standards	500µg	\$17.00		
Bio-Rad	161-0737EDU	Laemmli sample buffer	30mL	\$9.60		
Bio-Rad	161-0375EDU	Precision Plus Protein Kaleidoscope prestained standards	500µL	\$66.00		
Bio-Rad	161-0732EDU	Electrophoresis Buffer, 10X	1L	\$17.60		
Bio-Rad	161-0786EDU	Bio-Safe Coomassie stain	1L	\$52.00		



## FROM FINCHES TO FISHES IMPLEMENTATION PLAN — POST-LAB

Activity	Estimated Time	Materials/Equipment	Purpose/Objectives/ Essential Question
<p>Analyze polyacrylamide gels: Use inquiry methods to encourage students to analyze and discuss their electrophoresis results.</p> <ul style="list-style-type: none"> <li>• How many bands do similar looking fish have in common?</li> <li>• Which fish have no bands in common?</li> <li>• Which fish appear to be related based on the number of corresponding bands or proteins?</li> </ul> <p>Remind students that gel electrophoresis of proteins does not provide any direct information about amino acid sequences, and so cannot be used in any precise way to reconstruct evolutionary history.</p>	30 minutes	<p>Polyacrylamide gels</p> <p>Handout: Protein Fingerprinting: Analyzing Results of Gel Electrophoresis</p> <p>Handout: What Do the Bands Reveal? What Is the Significance of Your Data?</p> <p>Rulers/magnifiers</p>	<p><b>Purpose:</b> To compare the protein profile from five different fish to determine phylogenetic relationships.</p> <p><b>Objectives:</b></p> <ul style="list-style-type: none"> <li>• To interpret the results of electrophoresis</li> <li>• To make inferences about the relatedness of fish based on the similarities of their protein profiles.</li> </ul> <p><b>Essential Question:</b> Which fish are most closely related based upon their protein profile?</p>
Qualitative analysis of data	20 minutes	Worksheet: Interpreting the Bands	
Detailed gel analysis	60 minutes	Worksheet: Analysis and interpretation of results	
From Finches to Fishes Quiz Game	20 minutes	CD provided	
Isn't Evolution Just a Theory? Video 1	20 minutes	<p><i>PBS Evolution Series Learning and Teaching Evolution</i>, Video 1 only Video Discussion questions</p> <p>Optional: Show video prior to Additional Activity essay assignment to incorporate into writing activity.</p>	

### Alignment with NC Competency Goals

#### Biology

Goal 1  
Objectives 1.01, 1.02, 1.03, 1.05

Goal 2  
Objectives 2.01, 2.02, 2.03

Goal 3  
Objectives 3.01, 3.04, 3.05

Goal 4  
Objectives 4.01, 4.02, 4.03

Goal 5  
Objectives 5.03



# ANALYZING RESULTS OF GEL ELECTROPHORESIS AND DETERMINING SIGNIFICANCE OF DATA

## MATERIALS NEEDED

- 1 Protein Fingerprinting: Analyzing Results of Gel Electrophoresis handout for each student
- 1 What Do the Bands Reveal? What Is the Significance of Your Data? handout for each student
- Students' gels from wet-lab
- 1 ruler per student
- 1 magnifying glass per student

## BACKGROUND INFORMATION FOR THE TEACHER

Similarities and differences between protein fingerprints are easily spotted. By comparing the banding pattern of the shark with other fish it is clear that this fish is dissimilar from all of the others. This is consistent with the proposed fish evolutionary tree, since the shark belongs to the Class Chondrichthyes — cartilaginous fishes — as compared to the other species, which belong to the Class Osteichthyes — bony fishes. Salmon and trout, which are on the same branch, have many similar bands. Swordfish and tuna, located on the same branch, show some similarities in their protein bands. By contrast, salmon and catfish, located on different branches, reveal significant differences in

their banding patterns. The actin and myosin standard is included as a reference to help identify the major, conserved muscle proteins and to serve as a positive control for gel analysis. This protein consists of myofibrils isolated from rabbit skeletal muscle. The Precision Plus Protein Kaleidoscope prestained standards are included to provide a means of practice for loading the samples into the polyacrylamide ready gels and to help create a standard curve when graphing the molecular weights of proteins.

## INSTRUCTIONS FOR TEACHERS

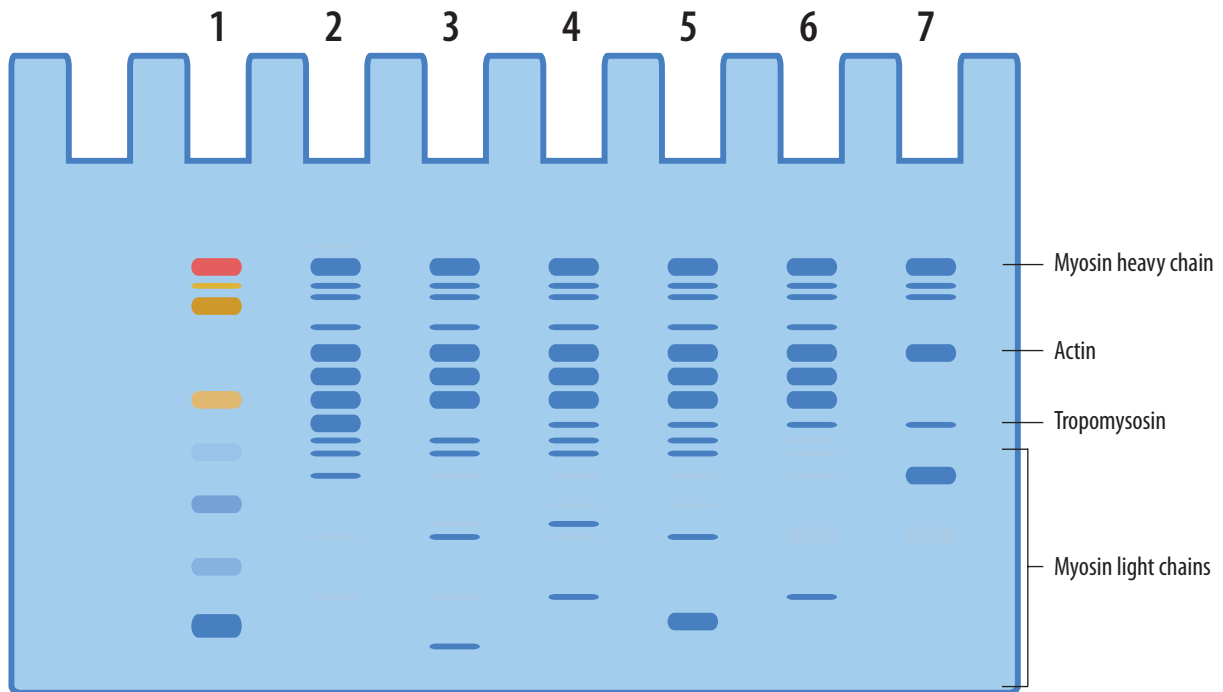
Distribute the two handouts to the class (copy from notebook or print from module CD). Use the Protein Fingerprinting handout to review the process for analyzing results of gel electrophoresis. Because the diagram on this handout is clear and easy to read, it provides a good opportunity to practice the technique of counting bands.

After practicing, students should count the bands on their own gels, filling out the chart on the What Do the Bands Reveal? handout. After they have completed the chart and read the additional information on the significance of the data, have them answer the three questions and then review the results together as a class.

# PROTEIN FINGERPRINTING: ANALYZING RESULTS OF GEL ELECTROPHORESIS

Evolutionary biologists make hypotheses about relationships among different groups of organisms based on how similar they are, in terms of both morphological and molecular traits. Below are the results of a gel electrophoresis procedure, used to create protein “fingerprints” of several species of fish. By comparing the number of protein bands that each kind of fish has in common with one another, you can make your own hypothesis about which of these fish are most closely related.

## SAMPLE PROTEIN FINGERPRINTS



Lane: 1. Precision Plus Protein  
Kaleidoscope standards  
2. Shark

3. Salmon  
4. Trout  
5. Catfish

6. Swordfish  
7. Actin and myosin standard

## WHAT DO THE BANDS REVEAL?

Using a ruler or straight edge, carefully count the number of bands that each kind of fish has in common with the others. Do not include any of the bands in the actin and myosin standards in your counts, since every kind of fish has these bands. Then fill out the table below with your results. Which of these fish do you think are most closely related?

COMPARISON OF NUMBER OF SIMILAR PROTEIN BANDS FOUND IN FISH

	Shark	Salmon	Trout	Catfish	Swordfish
Shark					
Salmon					
Trout					
Catfish					
Swordfish					

## WHAT IS THE SIGNIFICANCE OF YOUR DATA?

To make meaningful inferences about evolutionary relationships among the sample fish species, use a ruler and a magnifying glass to compare the banding patterns.

The actin and myosin standard is included as a reference to help identify the major, conserved muscle proteins and to serve as a positive control for gel analysis. The Precision Plus Protein Kaleidoscope prestained standards are included to provide a means of practice for loading the samples into the polyacrylamide ready gels and to help create a standard curve when graphing the molecular weights of protein.

Gel electrophoresis is a powerful tool for separating and visualizing the individual proteins in complex samples like muscle tissue. The two types of molecules most often analyzed by electrophoresis are nucleic acids, like DNA, and proteins. Electrophoresis not only lets you determine how many distinct types of molecules are in the sample, it can also tell you their sizes, which can be a clue to its identity.

The procedure that you have completed displays a profile of protein composition in the muscle tissue of different fish. Since proteins are a reflection of an organism's DNA, variations in these composition profiles indicates variations in DNA sequences. Evolutionary relationships among species are inferred

from the degree of genetic (DNA) similarity among them. The protein fingerprints you've generated, indirectly representing the genetic compositions of your chosen fish species, are molecular-level indicators of evolutionary relationships.

Similarities and differences between protein fingerprints are easily spotted.

1. Compare the banding pattern of the shark with other fish samples. How might you explain any differences?
2. How does the salmon and catfish compare? Are they located on the same branch?
3. Do the swordfish and tuna, located on the same branch, show similarities in their protein bands?

## INTERPRETING THE BANDS COLLECTIVELY: QUALITATIVE COMPARISONS OF PROTEIN PROFILES

From Bio-Rad's Comparative Proteomics Kit I: Protein Profiler Module

1. Is there any variation among the protein profiles of your samples?
2. How would you distinguish the protein profiles of different species from each other?
3. What are possible explanations for this variation?
4. Which samples are most alike?

Tape your gel or a photocopy of it in the box. The bands are the data upon which your analysis will be based.



# ANALYSIS AND INTERPRETATION OF RESULTS: DETAILED GEL ANALYSIS

## From Bio-Rad's Comparative Proteomics Kit I: Protein Profiler Module

### Does your molecular evidence support or refute your predictions?

Each protein band that a fish has in common with another fish is considered a shared characteristic. A fish family tree, or cladogram, can be constructed based on proteins bands that the fish have in common. Cladistic analysis assumes that when two organisms share a common characteristic that they also share a common ancestor with that same characteristic.

Create a cladogram using your results to find out.

From your gel you can create a cladogram based on proteins that the fish have in common. You can then determine whether your cladogram supports your predictions and/or matches the evolutionary relatedness of the fish species determined by morphological analysis in the evolutionary tree provided. Each protein band that a fish has in common with another fish is a shared characteristic. Cladistic analysis assumes that when two organisms share a characteristic, they had a common ancestor that had that characteristic, and this can be represented as a node on a cladogram with two branches coming from that node representing the descendent organisms.

In this exercise you will define the shared characteristics (i.e., make a list of all the different proteins in fish muscle), find which proteins (characteristics) are shared between fish, and construct a cladogram based on your data.

## PROCEDURES

### Generate a standard curve to calculate protein molecular weights

(Optional) Although it is not strictly necessary for this exercise, you may want to create a standard curve from your gel and determine the actual size of each protein band.

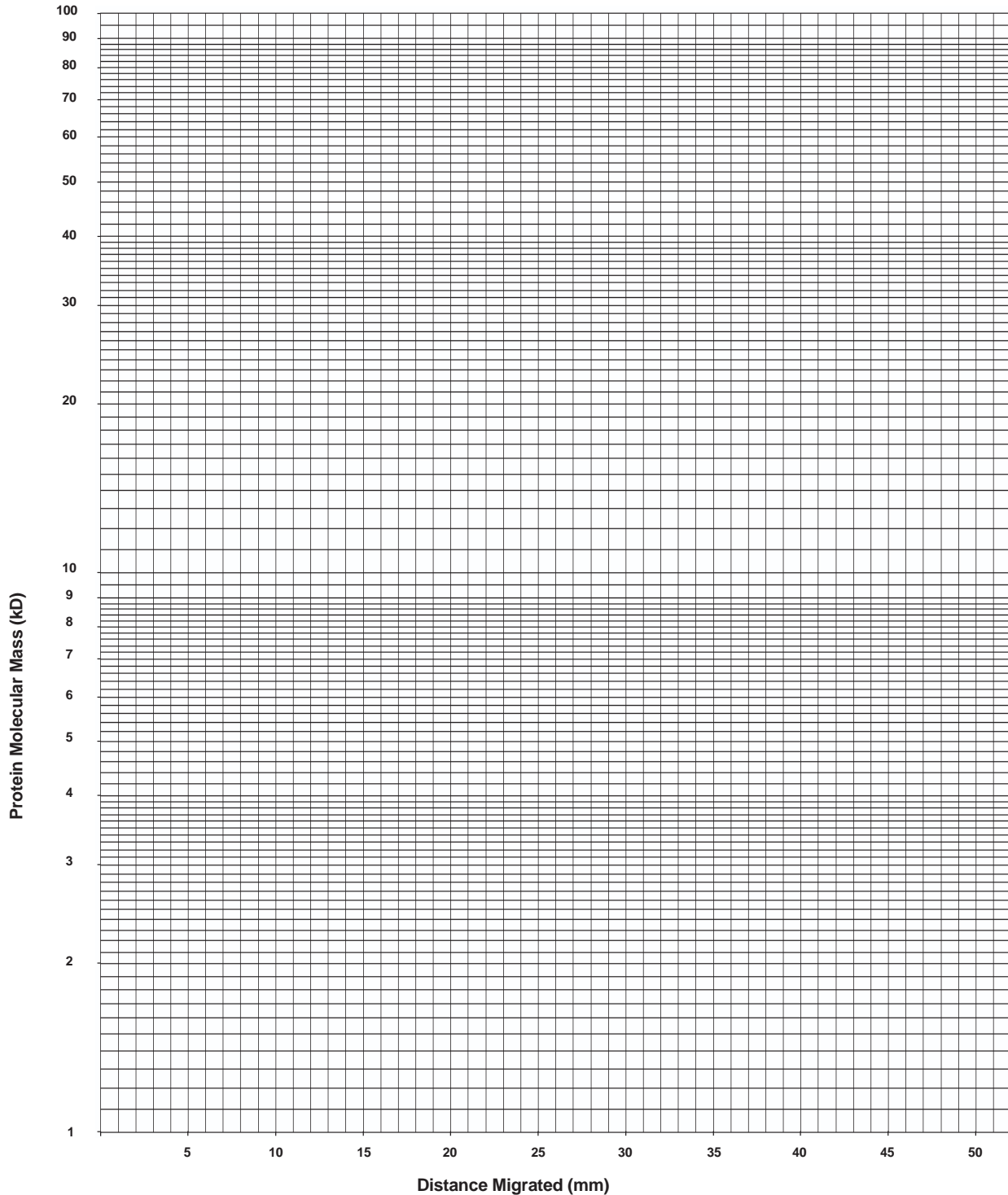
Alternatively, the cladogram can be generated just using the distance in millimeters the different protein bands have migrated from the wells of the gel.

To create the standard curve measure and record the distances the five visible protein bands contained in the Precision Plus Protein Kaleidoscope prestained standards. Start from the green 37 kD band down to the yellow 10 kD band that has migrated from the wells. Accuracy to 0.5 mm is required.

Precision Plus Protein Kaleidoscope Prestained Standards Molecular Weight (kD)	Distance Migrated (mm)
37	
25	
20	
15	
10	

On the graph paper provided, plot the distances migrated in mm on the x-axis against the molecular weight of the bands in kD on the y-axis as a scatter plot.

Draw a line through the points. On a logarithmic scale, plotting the molecular weights against the distances migrated for each protein in the standard should result in a linear (straight line) curve. Alternatively, you can use a graphing computer program to generate the chart and make a line of best fit (or a trend-line) through these points and to formulate an equation to calculate the MW of the unknown proteins on the gel.





### Define the characteristics (proteins) of the different fish

Make a horizontal line on the dried gel (or gel image) between the 37 kD (green) and 25 kD (pink) markers below the fat bands that occur at around 30 kD (see gel above). Then, for each band below the line for each fish sample, measure the distance the protein band has migrated from the wells (and, if required, determine its size in kD using the standard curve or the formula generated from the standard curve) and record this data (see example below):

	Species A	Species B	Species C	Species D	Species E
Distance protein bands migrated (mm)	25, 26.5, 29, 36, 36.5, 39, 44, 52	26, 27.5, 29, 32, 34.5, 36.5, 37.5, 40.5, 42, 45	26, 27.5, 29, 29.5, 32, 34.5, 36.5, 37.5, 40.5, 42, 45, 46.5, 51.5	26, 27.5, 29, 32, 36.5, 38, 38.5, 41, 46, 47.5, 44, 47	26, 27.5, 30, 30.5, 33, 35.5, 37, 39, 39.5, 42

Record your data in the table below:

Distance protein bands migrated (mm)					

### Determine which fish have each characteristic (protein)

In the blank table provided on page 108, use one row for every band size you have recorded and one column for each type of fish on your gel. Then make a mark in each cell of the table where the fish has that size band (see example on next page).

Example:

CHARACTERISTIC	Distance Migrated (mm)	Protein Molecular Mass (kDa)	Species A	Species B	Species C	Species D	Species E
	25	32.5	X				
	26	31.5		X	X	X	X
	26.5	31.0	X				
	27.5	30.0		X	X	X	X
	28.5	29.1					
	29	28.6	X	X	X	X	
	30	27.6			X		X
	30.5	27.1					X
	32	25.6		X	X	X	
	33	24.7					X
	34.5	23.2		X	X		
	35.5	22.2					X
	36	21.7	X				
	36.5	21.2	X	X	X	X	
	37	20.7					X
	37.5	20.2		X	X		
	38	19.7				X	
	38.5	19.3				X	
	39	18.8	X				X
	39.5	18.3					X
	40.5	17.3		X	X		
	41	16.8				X	
	41.5	16.3					
42	15.8		X	X		X	
43	14.8						
44	13.9	X				X	
45	12.9		X	X			
46	11.9				X		
46.5	11.4			X			
47	10.9					X	
47.5	10.4				X		
51.5	6.5			X			
52	6.0	X					
	<b>COUNT</b>	<b>8</b>	<b>10</b>	<b>13</b>	<b>10</b>	<b>12</b>	



### Find the number of characteristics shared by each of the fish

In the table below both the row and column headings are the types of fish. From the table on page 105, separately compare the number of bands (X's) in common with every other fish sample from your gel and put those numbers into the table below, such that each fish is individually compared with every other fish. In this example, species A and B have just two bands in common while species B and C have 10 bands in common. The table below will be the basis for drawing your cladogram.

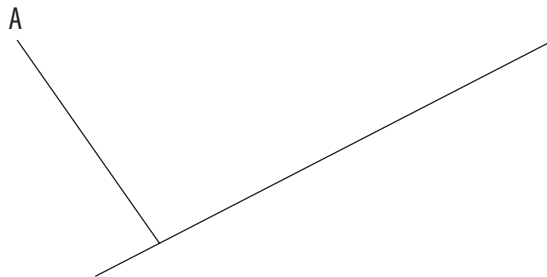
	Species A	Species B	Species C	Species D	Species E
Species A	8	2	2	2	2
Species B		10	10	5	3
Species C			13	5	4
Species D				10	2
Species E					12

Record your data in the table below:

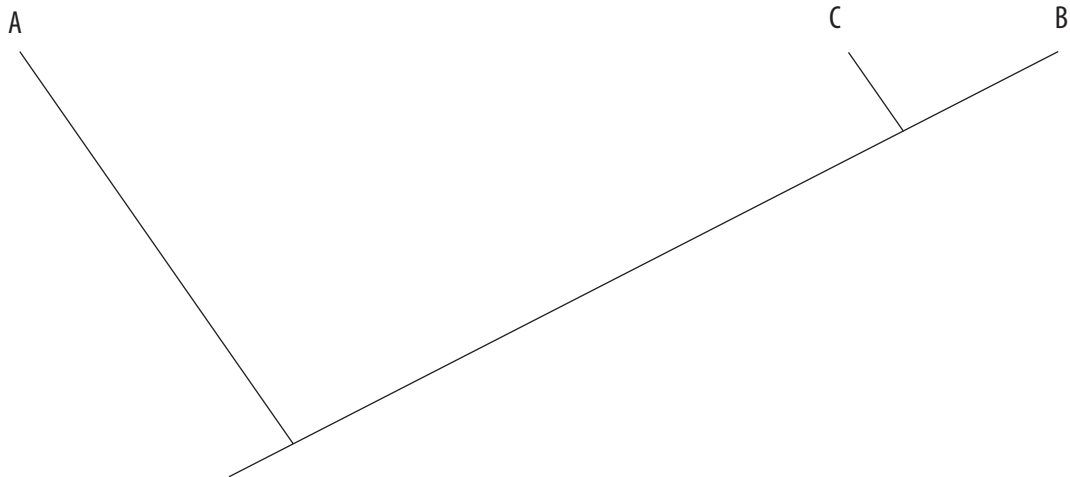
	Species A	Species B	Species C	Species D	Species E

### Construct your cladogram

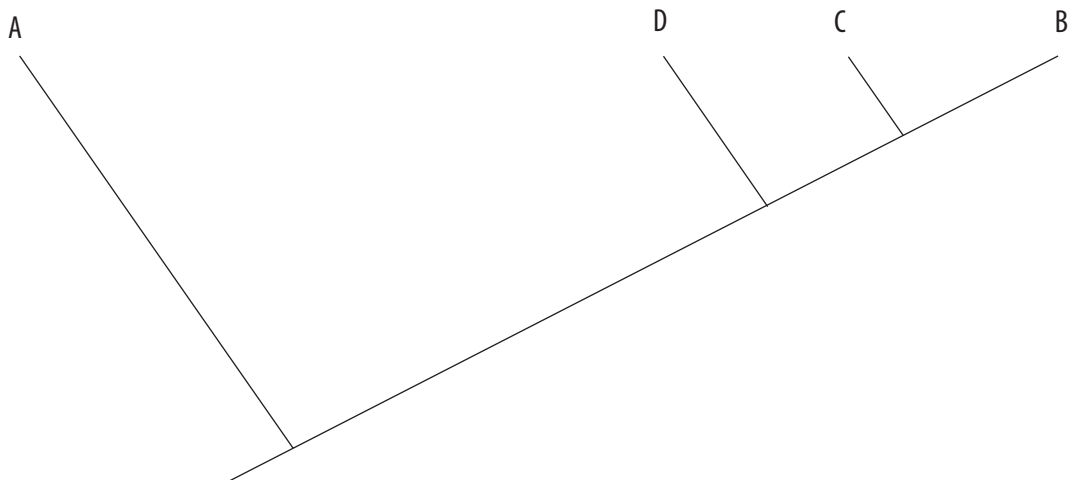
Now you are ready to construct your cladogram. First draw a line to form the trunk of your cladogram. Find the fish with the least bands in common. In the example above it is species A, which has only 2 bands in common with any of the other fish. Then draw a side branch off the line near the bottom of the trunk and label that branch with the fish's name, in this case, species A. This fish is the outlier, i.e., it is the least similar to any of the others. The node (where the side branch meets the trunk) represents an ancestor that is common to all the fish in this analysis.



Find the two fish with the most bands in common (in this example it is species B and C, which have 10 bands in common). Draw a side branch off the trunk near the top and label the two ends with the fishes' names, in this case, species B and species C (it doesn't matter which branch gets which label). The node represents a common ancestor of species B and species C that had all the same characteristics (proteins).

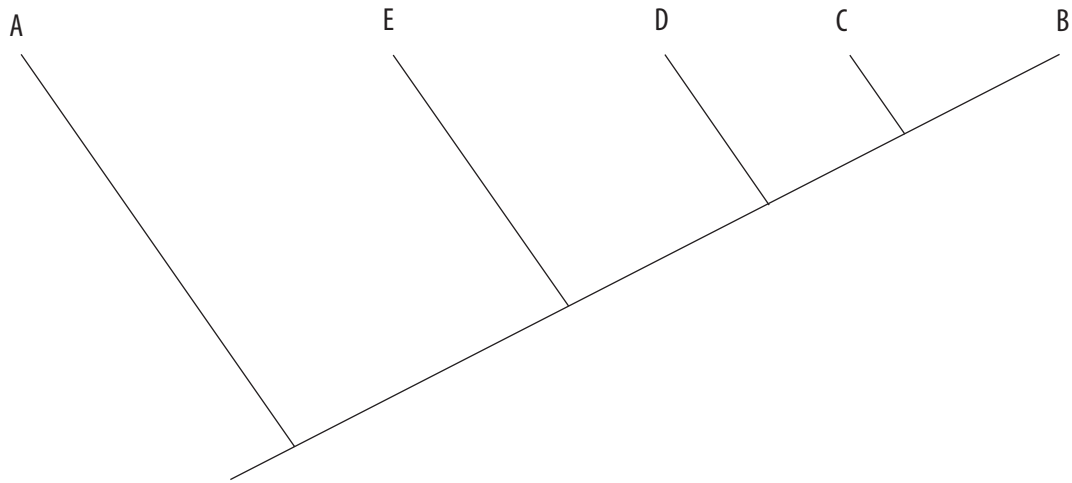


Identify those fish species with the next most bands in common. In this example, species D has five bands in common with species B and species C, which indicates species D is the same cladistic distance from B and C (i.e. species D is not more closely related to either B or C). Draw a branch further down the trunk. This node represents an ancestor that is common to species B, C, and D that had these 5 characteristic proteins.



The last fish to add to the cladogram in this example is species E, which shares four bands with species C, three bands with species B, and only two bands with species A and D. This fish may seem trickier to place than the others because it shares more characteristics with species B and C than it does with D, but D shares more characteristics with B and C than E does. So, to place this fish you might ask: Does species E share the five proteins that the common ancestor of species B, C, and D had? Answer (no).

Does species E share more proteins with B, C, and D than A? Answer (yes). Therefore, species E gets its own branch in between the D and A branches to indicate that it has more shared characteristics with B, C, and D than A, but fewer shared characteristics with B and C than D.



Using your own data, draw a cladogram below.

Compare your cladogram with your original predictions. Write your deductions below.



## From Finches to Fishes Quiz Game Questions

	<b>Evolution</b>	<b>Proteins and Such</b>	<b>Wet-Lab</b>	<b>Protein Finger-printing</b>	<b>Dictionary</b>	<b>Hodge Podge</b>
<b>200</b>	Who first developed the theory of evolution through natural selection?	Fact or fiction? Proteins account for more than 95 percent of the dry weight of most cells.	Fill in the blank: The process used in the wet-lab to obtain a protein fingerprint is called gel _____.	Fill in the blank: Gel electrophoresis is used to separate proteins according to _____.	Define evolution.	How many friction stops were on the plunger of the micropipette used in the wet lab?
<b>400</b>	What was the name of the ship that Darwin sailed on during the voyage that led to his development of his theory of evolution through natural selection?	What are proteins?	In the wet-lab we used digital micropipettes, which measure very small quantities of liquid. What units do the micropipettes use to measure?	In protein fingerprinting, which proteins traveled further on the polyacrylamide gel, small ones or large ones?	What is the term for a sequence of DNA that codes for a protein and determines a trait?	Convert 2.7 milliliters to microliters.
<b>600</b>	What is the term for the study of evolutionary relationships among organisms?	How does DNA code for a specific trait such as hair color? (Hint: creation of proteins is one step)	Heat and SDS buffer were applied to our fish protein samples for what two reasons?	What piece of equipment was used to place samples of protein into the wells of the polyacrylamide gel?	What is genetic drift?	During the wet-lab, one well contained actin and myosin; what are actin and myosin?
<b>800</b>	Name three adaptations in birds that may give them an evolutionary advantage over other organisms.	How can proteins be used to determine evolutionary relationships among organisms?	Why were polyacrylamide gels used in the wet-lab instead of agarose gels?	Why is it necessary that polyacrylamide gels be stained and destained after protein electrophoresis?	Define speciation.	The "fishy family tree" you looked at before the wet-lab used morphological traits to show relationships between organisms, what is morphology?
<b>1000</b>	Explain Darwin's theory of evolution through natural selection.	Name four ways in which proteins are used in our bodies.	According to the results of the wet-lab, which fish appear to be the most closely related? Explain your answer.	Why is the process used in the wet-lab called protein fingerprinting?	Define adaptive radiation.	The field of comparative anatomy uses occurrences of homologous and analogous structures in different species as evidence that those species evolved from similar or different ancestors. What is the difference between homologous and analogous structures?



KEY

## Finches to Fishes Quiz Game Answers

	Evolution	Proteins and Such	Wet-Lab	Protein Finger-printing	Dictionary	Hodge Podge
200	Charles Darwin.	Fiction. Proteins account for 50% of the dry weight of a cell.	Electrophoresis.	Size.	A change in the frequency of a trait (gene) within a population.	2.
400	The HMS Beagle.	Organic molecules that are made of amino acids held together by peptide bonds.	Microliters.	Small proteins.	Gene.	2.7 ml = 2700 µl.
600	Phylogeny.	DNA is used to create mRNA. mRNA codes for a specific combination of amino acids. Those amino acids form proteins. Proteins act by themselves or in combination with other proteins to give an organism a trait (for instance hair color).	The heat and SDS buffer made the proteins linear and negatively charged.	Micropipette.	Changes in the gene pool of a small population due to chance.	Actin and myosin are proteins found in the muscle tissue of many organisms.
800	Answers may vary. Examples: Wings, feathers, hollow bones for flight; beaks, claws for digging for food; talons for hunting; camouflage coloring.	One of 2 answers would be correct. 1) The number of shared proteins between organisms can illustrate relative relatedness. 2) The number of amino acid differences in a single protein between 2 species can tell you how long ago the 2 species diverged.	Polyacrylamide gels were used because they are more effective for smaller molecules, such as proteins.	So that the proteins can be seen as bands on the gel.	The origin of new species in evolution.	The branch of biology that deals with the form and structure of organisms without consideration of function. An example of a morph trait is a fish's mouth shape.
1000	Darwin's theory can be summarized in 4 points: 1. Variations exist among individuals 2. Organisms produce more offspring than the environment can support 3. Competition exists among individuals 4. The organisms whose variations best adapt them to the environment are the ones who are most likely to survive, reproduce, and pass on desirable traits to the next generation.	Answers may vary, but may include: proteins as part of hair, muscle, fingernails, or as enzymes, neurotransmitters, hormones, etc. In cells proteins are used for structural support, storage, transport of substances, movement and defense against foreign substances.	The answers to this question will vary depending on the fish proteins used. The students should justify their answer by stating that the percentage of proteins shared by those two fish is greater than any other combination of fish.	It's called protein fingerprinting because the technique leaves a banding pattern on the gel where the proteins were separated by size. This banding pattern is unique to each species of fish, just like a fingerprint is unique to each human.	A relatively rapid evolution of many species from a single ancestor.	Homologous structures are similar features on different species that don't have the same use (forelimbs on alligator and human). Analogous structures are similar structures with the same purpose (wings on insects and birds).

## DISCUSSION QUESTIONS: ISN'T EVOLUTION JUST A THEORY?

From *PBS Evolution Series: Learning and Teaching Evolution*

1. How does the scientific meaning of a term like “theory” differ from the way it is used in everyday life?

2. Can the “facts” of science change over time? If so, how?

## FROM FINCHES TO FISHES IMPLEMENTATION PLAN — ADDITIONAL ACTIVITIES & RESOURCES

Activity	Est. Time	Materials/Equipment	Subjects Covered	Suggestions
<i>Essay applying learned vocabulary, evolutionary process, and scientific concepts</i>	50 minutes	Rubric for essay, Text Book, Internet Access	Evolution, writing skills, computer skills	Additional summative assessment  Optional: Show Isn't Evolution Just a Theory? video prior to assigning essay
<i>Creating cladograms</i> Students use taxonomic information from the BLAST databases to create a phylogenetic tree.	60 minutes	Handouts: Using Databases to Obtain Real Amino Acid Sequence Data to Create Cladograms	Technology, bioinformatics	
<i>Peppered Moth Game</i>	20 minutes	<a href="http://www3.district125.k12.il.us/faculty/nfischer/Moth/default.htm">www3.district125.k12.il.us/faculty/nfischer/Moth/default.htm</a>	Evolution, molecular biology	
<i>Chromosome Connections</i> How closely related are we to other primates? Students will match up chromosomes to determine which primate is our closest living relative.	30 minutes	Internet access <a href="http://www.becominghuman.org">www.becominghuman.org</a> (Click on "learning center" and then on "lesson plans.")	Molecular biology, genetics, process of science, computer skills	
<i>Building Bodies</i> Humans and other hominids evolved unique skeletal adaptations. Students will "build" two skeletons — one of an ape and one of a modern human.	30 minutes	Internet access <a href="http://www.becominghuman.org">www.becominghuman.org</a> (Click on "learning center" and then on "lesson plans.")	Human anatomy and physiology, evolution, computer skills	
<i>Calculating Cousins</i> Although fossils can be accurately dated, the lines of descent between species are widely debated. How a hominid family tree is drawn depends on the current fossil evidence. Students will evaluate fossil evidence and map out the history of the hominid family tree.	30 minutes	Internet access <a href="http://www.becominghuman.org">www.becominghuman.org</a> (Click on "learning center" and then on "lesson plans.")	Evolution, genetics, pedigrees, human biology, genealogy, taxonomy, computer skills	
<i>Evidence for Evolution WebQuest</i> Students will investigate a variety of evidence for evolution. Each team will be responsible for learning about fossil evidence, structural evidence, and genetic evidence and presenting the information to the class.	50 minutes	Internet access <a href="http://www.pbs.org/evolution">www.pbs.org/evolution</a> Large paper (24-46) Markers, rulers	Evolution, earth science, anatomy, presentation skills, computer skills	
<i>Evolution and Time</i> Students act as paleontologists investigating a particular period of time in Earth's history. Students visit their chosen time period and make a detailed journal entry of what they "see." Students also create a "Geologic Birthday Card."	50 minutes	Calculator, Paper (8½"X11") Markers, crayons, scissors, magazines, glue	Evolution, earth science, math, art, writing skills	
<i>How Much Variation? Doing the Math</i> Students examine variation in a 1,691-base segment of the beta globin gene to consider the extent of human genetic variation at the molecular level and the relationship between genetic variation and evolution.	20 minutes	Student Activity Sheet from NIH Curriculum Supplement Series: Human Genetic Variation	Genetics, molecular biology, math skills	
<i>Alike, But Not the Same</i>	20 minutes	Student Activity Sheet from NIH Curriculum Supplement Series: Human Genetic Variation	Genetics	
Comparative Proteomics Kit II: Western Blot Module		Comparative Proteomics Kit II from Bio-Rad; Cat # 166-2800EDU		
<p>Additional Resources:  <i>Evolution Web</i> features, articles images, and more . . . all easily accessible by going to: <a href="http://www.pbs.org/evolution">http://www.pbs.org/evolution</a>. Then go to <i>Evolution Teachers Guide</i>: where you will find the Teacher's Guide Web Resources organized by unit.                      Human Evolution website: <a href="http://www.becominghuman.org">www.becominghuman.org</a>                      Bio-Rad's website: <a href="http://explorer.bio-rad.com">explorer.bio-rad.com</a></p>				

# FROM FINCHES TO FISHES

A general rubric used for scoring the evolution essay

CONCEPTS/VOCABULARY TO BE INCLUDED	MAXIMUM 5 POINTS EACH
Environment and conditions for survival	
Descent with modification	
Adaptations, fitness	
Isolating mechanisms	
Definition of evolution	
Evidence for evolution	
Modification by natural selection	
Relatedness vs. common ancestor	
Species, subspecies, speciation	
Adaptive radiation	
Extinction	
Genetic drift	
<b>PRESENTATION FORMAT</b>	
Overall use of scientific terms and scientific thinking	
Use of observations to support statements	
Spelling and punctuation	
Sentence structure	
Grammar	
Organization of topics and subtopics	
Use of additional evidence to support statements	
Overall presentation and writing format	
<b>Total Points</b>	

# USING DATABASES TO OBTAIN REAL AMINO ACID SEQUENCE DATA TO CREATE CLADOGRAMS

## From Bio-Rad's Comparative Proteomics Kit I: Protein Profiler Module

In order to determine how closely related species are, scientists often will study amino acid sequences of essential proteins. Any difference in the amino acid sequence is noted and a phylogenetic tree is constructed based on the number of differences. More closely related species have fewer differences (i.e., they have more amino acid sequence in common) than more distantly related species.

There are many tools scientists can use to compare amino acid sequences of muscle protein. One such tool is the National Center for Biotechnology Information protein databases (<http://www.ncbi.nlm.nih.gov/>). By entering the amino acid sequence of a protein you are interested in, the BLAST search tool compares that sequence to all others in its database. The data generated provides enough information to construct cladograms.

The purpose of this activity is to use data obtained from NCBI to construct an evolutionary tree based on the amino acid sequences of the myosin heavy chain. In this example we have input a 60 amino acid sequence from myosin heavy chain of rainbow trout and then pulled out matching sequences using BLAST, which include chum salmon, zebra fish, common carp, and bluefin tuna, and then compared each of these sequences with each other.

You may either use the data provided below or have your class go online and obtain their data directly by performing BLAST searches. A quick guide to performing BLAST searches is given at the end of this activity.

The data below was obtained by entering a 60 amino acid sequence from the heavy myosin chain of rainbow trout. The database search tool returned all sequences that were a close match. The results are formatted as such:

```
gi|755771|emb|CAA88724.1 myosin heavy chain [Oncorhynchus mykiss]
Length=698
Score = 119 bits (299), Expect = 2e-26
Identities = 60/60 (100%), Positives = 60/60 (100%), Gaps = 0/60 (0%)

Query 1
VAKAKGNLEKMCRTLEDQLSELKTKNDENVRQVNDISGQRARLLTENGEGFRQLEEKEAL 60
VAKAKGNLEKMCRTLEDQLSELKTKNDENVRQVNDISGQRARLLTENGEGFRQLEEKEAL

Sbjct 1
VAKAKGNLEKMCRTLEDQLSELKTKNDENVRQVNDISGQRARLLTENGEGFRQLEEKEAL 60
```

The value for “identities” is the number of amino acids exactly in common, the value for “positives” is the number of amino acids that are similar to each other (such as serine and threonine), and the value for ‘gaps’ is the number of amino acid positions that are absent one of the sequences. “Query” is the original trout sequence, “Sbjct” is the aligned sequence, and the middle sequence shows the mismatches: a “+” indicates a positive and a space indicates a mismatch that is not a positive. There are resources on the NCBI website to help you understand more about the information a BLAST search generates.

The data on the following pages compares rainbow trout to salmon, zebra fish, carp, and tuna, and then compares salmon to zebra fish, carp, and tuna, then zebra fish to carp and tuna, and finally carp to tuna.

Use the data provided to determine how many amino acid differences exist between the organisms. Organize your data in charts.



## Rainbow trout compared to chum salmon

[gi|21623523|dbj|BAC00871.1](#) myosin heavy chain [Oncorhynchus keta]  
Length=1937

Score = 119 bits (299), Expect = 2e-26  
Identities = 60/60 (100%), Positives = 60/60 (100%), Gaps = 0/60 (0%)

Query 1  
AKAKGNLEKMCRTLEDQSELKTKNDENVQVNDISGQRARLLTENGEFGRQLEEKEAL 60  
VAKAKGNLEKMCRTLEDQSELKTKNDENVQVNDISGQRARLLTENGEFGRQLEEKEAL

Sbjct 1240  
VAKAKGNLEKMCRTLEDQSELKTKNDENVQVNDISGQRARLLTENGEFGRQLEEKEAL 1299

## Rainbow trout compared to zebra fish

[gi|68360600|ref|XP\\_708916.1](#) PREDICTED: myosin, heavy polypeptide 1, skeletal muscle [Danio rerio]  
Length=2505

Score = 108 bits (269), Expect = 6e-23  
Identities = 52/60 (86%), Positives = 57/60 (95%), Gaps = 0/60 (0%)

Query 1  
VAKAKGNLEKMCRTLEDQSELKTKNDENVQVNDISGQRARLLTENGEFGRQLEEKEAL 60  
VAKAK NLEKMCRTLEDQSE+K+KNDEN+RQ+ND+S QRARL TENGEFGRQLEEKEAL

Sbjct 1240  
VAKAKANLEKMCRTLEDQSEIKSKNDENLRQINDLSAQRARLQTENGEFGRQLEEKEAL 1299

## Rainbow trout compared to common carp

[gi|806515|dbj|BAA09069.1](#) myosin heavy chain [Cyprinus carpio]  
Length=955

Score = 104 bits (259), Expect = 8e-22  
Identities = 51/60 (85%), Positives = 56/60 (93%), Gaps = 0/60 (0%)

Query 1  
VAKAKGNLEKMCRTLEDQSELKTKNDENVQVNDISGQRARLLTENGEFGRQLEEKEAL 60  
VAKAK NLEKMCRTLEDQSE+KTK+DENVRQ+ND++ QRARL TENGEF RQLEEKEAL

Sbjct 259  
VAKAKANLEKMCRTLEDQSEIKTKSDENVRLQNDMNAQRARLQTENGEFSRQLEEKEAL 318

## Rainbow trout compared to bluefin tuna

[gi|1339977|dbj|BAA12730.1](#) skeletal myosin heavy chain [Thunnus thynnus]  
Length=786

Score = 104 bits (259), Expect = 8e-22  
Identities = 49/60 (81%), Positives = 57/60 (95%), Gaps = 0/60 (0%)

Query 1  
VAKAKGNLEKMCRTLEDQSELKTKNDENVQVNDISGQRARLLTENGEFGRQLEEKEAL 60  
VAK+KGNLEKMCRT+EDQSELK KNDE+VRQ+ND++QQRARL TENGEF RQ+EEK+AL

Sbjct 88  
VAKSKGNLEKMCRTIEDQSELKAKNDEHVRQLNDLNGQRARLQTENGEFSRQIEEKDAL 147

## Chum salmon compared to common carp

[gi|806515|dbj|BAA09069.1](#) myosin heavy chain [Cyprinus carpio]  
Length=955

Score = 104 bits (259), Expect = 8e-22  
Identities = 51/60 (85%), Positives = 56/60 (93%), Gaps = 0/60 (0%)

Query 1  
VAKAKGNLEKMCRTLEDQSELKTKNDENVQVNDISGQRARLLTENGEFGRQLEEKEAL 60  
VAKAK NLEKMCRTLEDQSE+KTK+DENVRQ+ND++ QRARL TENGEF RQLEEKEAL

Sbjct 259  
VAKAKANLEKMCRTLEDQSEIKTKSDENVRLQNDMNAQRARLQTENGEFSRQLEEKEAL 318

## Chum salmon compared to zebra fish

[gi|68360600|ref|XP\\_708916.1](#) PREDICTED: myosin, heavy polypeptide 1, skeletal muscle [Danio rerio]  
Length=2505

Score = 108 bits (269), Expect = 6e-23  
Identities = 52/60 (86%), Positives = 57/60 (95%), Gaps = 0/60 (0%)

Query 1  
VAKAKGNLEKMCRTLEDQSELKTKNDENVQVNDISGQRARLLTENGEFGRQLEEKEAL 60  
VAKAK NLEKMCRTLEDQSE+K+KNDEN+RQ+ND+S QRARL TENGEFGRQLEEKEAL

Sbjct 1240  
VAKAKANLEKMCRTLEDQSEIKSKNDENLRQINDLSAQRARLQTENGEFGRQLEEKEAL 1299

## Chum salmon compared to bluefin tuna

[gi|1339977|dbj|BAA12730.1](#) skeletal myosin heavy chain [Thunnus thynnus]  
Length=786

Score = 104 bits (259), Expect = 8e-22  
Identities = 49/60 (81%), Positives = 57/60 (95%), Gaps = 0/60 (0%)

Query 1  
VAKAKGNLEKMCRTLEDQSELKTKNDENVQVNDISGQRARLLTENGEFGRQLEEKEAL 60  
VAK+KGNLEKMCRT+EDQSELK KNDE+VRQ+ND++QQRARL TENGEF RQ+EEK+AL

Sbjct 88  
VAKSKGNLEKMCRTIEDQSELKAKNDEHVRQLNDLNGQRARLQTENGEFSRQIEEKDAL 147

## Zebra fish compared to common carp

[gi|806515|dbj|BAA09069.1](#) myosin heavy chain [Cyprinus carpio]  
Length=955

Score = 108 bits (271), Expect = 4e-23  
Identities = 53/60 (88%), Positives = 59/60 (98%), Gaps = 0/60 (0%)

Query 1  
VAKAKANLEKMCRTLEDQSEIKSKNDENLRQINDLSAQRARLQTENGEFGRQLEEKEAL 60  
VAKAKANLEKMCRTLEDQSEIK+K+DEN+RQ+ND++AQRARLQTENGEF RQLEEKEAL

Sbjct 259  
VAKAKANLEKMCRTLEDQSEIKTKSDENVRLQNDMNAQRARLQTENGEFSRQLEEKEAL 318

### Zebra fish compared to bluefin tuna

```

gi|1339977|dbj|BAA12730.1| skeletal myosin heavy chain [Thunnus thynnus]
Length=786

Score = 102 bits (253), Expect = 4e-21
Identities = 47/60 (78%), Positives = 57/60 (95%), Gaps = 0/60 (0%)

Query 1
VAKAKANLEKMCRTLEDQLSEIKSKNDENLRQINDLSAQRARLQTENGEGFRQLEEKEAL 60
VAK+K NLEKMCRT+EDQLSE+K+KNDE++RQ+NDL+ QRARLQTENGEGFRQ+EEK+AL

Sbjct 88
VAKSKGNLEKMCRTIEDQLSELKAKNDEHVRQLNDLNGQRARLQTENGEGFRQIEEKDAL 147
    
```

### Common carp compared to bluefin tuna

```

gi|1339977|dbj|BAA12730.1| skeletal myosin heavy chain [Thunnus thynnus]
Length=786

Score = 104 bits (259), Expect = 9e-22
Identities = 49/60 (81%), Positives = 57/60 (95%), Gaps = 0/60 (0%)

Query 1
VAKAKANLEKMCRTLEDQLSEIKTKSDENVRQLNDMNAQRARLQTENGEGFRQLEEKEAL 60
VAK+K NLEKMCRT+EDQLSE+K K+DE+VRQLND+N QRARLQTENGEGFRQ+EEK+AL

Sbjct 88
VAKSKGNLEKMCRTIEDQLSELKAKNDEHVRQLNDLNGQRARLQTENGEGFRQIEEKDAL 147
    
```

Construct a table of your data containing the number of amino acid differences between each of the different fish.

	Rainbow trout	Chum salmon	Zebra fish	Common carp	Bluefin tuna
Rainbow trout					
Chum salmon					
Zebra fish					
Common carp					
Bluefin tuna					

Which two fish share the most amino acids in their myosin heavy chains based on your data?

Which two fish share the fewest amino acids?

Are there any fish that share more amino acids with each other than each does with the two fish in question one? If yes, which fish?

Construct a cladogram based on this data:

The myosin heavy chain of white croaker (*Pennahia argentata*) (BAB12571) has the following amino acid differences with the five fish above.

	<b>Rainbow trout</b>	<b>Chum salmon</b>	<b>Zebra fish</b>	<b>Common carp</b>	<b>Bluefin tuna</b>
<b>White croaker</b>	4	4	11	9	11

Add this fish to your cladogram and explain why you placed it where you did.



KEY

Construct a table of your data containing the number of amino acid differences between each of the different fish.

	Rainbow trout	Chum salmon	Zebra fish	Common carp	Bluefin tuna
Rainbow trout	0	0	8	9	11
Chum salmon		0	8	9	11
Zebra fish			0	7	13
Common carp				0	11
Bluefin tuna					0

Which two fish share the most amino acids in their myosin heavy chains based on your data?

Trout and salmon

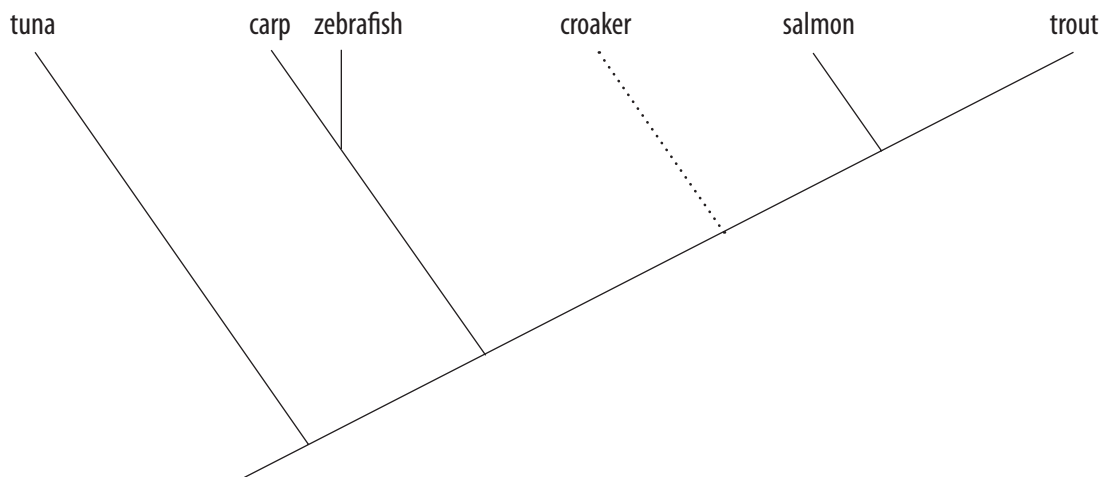
Which two fish share the fewest amino acids?

Tuna and zebra fish

Are there any fish that share more amino acids with each other than each does with the two fish in question one? If yes, which fish?

Yes, carp and zebra fish

Construct a cladogram based on this data:



Taxonomic data can be derived from many sources: DNA sequences, protein sequences, morphology, and paleontology. Classification of organisms derives from these sources. Inconsistencies in the phylogenetic trees generated between molecular and taxonomic data emphasize why data from different sources is required to generate phylogenetic trees and why there is still much dispute in the field of phylogenetics on the correct placement of organisms within phylogenetic trees. The amount of work required to process the small amount of data provided here also emphasizes the need for skilled bioinformaticists to process and analyze the vast amount of data generated by genomic and proteomic research.

Examine the taxonomic classification of the fishes below and construct a phylogenetic tree based on that data. The large phylogenetic tree figure will be useful for this exercise.

Rainbow Trout (*Oncorhynchus mykiss*) Vertebrata; Euteleostomi; Actinopterygii; Neopterygii; Teleostei; Euteleostei; Protacanthopterygii; Salmoniformes; Salmonidae; Oncorhynchus.

Chum Salmon (*Oncorhynchus keta*) Vertebrata; Euteleostomi; Actinopterygii; Neopterygii; Teleostei; Euteleostei; Protacanthopterygii; Salmoniformes; Salmonidae; Oncorhynchus.

Zebra Fish (*Danio rerio*) Vertebrata; Euteleostomi; Actinopterygii; Neopterygii; Teleostei; Ostariophysii; Cypriniformes; Cyprinidae; Danio.

Carp (*Cyprinus carpio*) Vertebrata; Euteleostomi; Actinopterygii; Neopterygii; Teleostei; Ostariophysii; Cypriniformes; Cyprinidae; Cyprinus.

Bluefin Tuna (*Thunnus thynnus*) Vertebrata; Euteleostomi; Actinopterygii; Neopterygii; Teleostei; Euteleostei; Neoteleostei; Acanthomorpha; Acanthopterygii; Perciformes; Perciformes; Scombroidei; Scombridae; Thunnus.

White Croaker (*Pennahia argentata*) Vertebrata; Euteleostomi; Actinopterygii; Neopterygii; Teleostei; Euteleostei; Neoteleostei; Acanthomorpha; Acanthopterygii; Perciformes; Perciformes; Perciformes; Perciformes; Sciaenidae; Pennahia.

Does the taxonomic classification support the molecular data?

Why do scientists need to examine multiple data sets before determining evolutionary relatedness?

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## QUICK GUIDE TO BLAST SEARCHING

Please note, this is a quick guide to obtain a list of fish myosin sequences, there are many refinements you can make to your search and many different ways to use BLAST searches.

Further information can be found on the NCBI website.

- 1) Go to <http://www.ncbi.nlm.nih.gov/> and choose BLAST.
- 2) Choose Protein-Protein BLAST.
- 3) Enter your myosin sequence into the search box.  
Rainbow Trout Myosin Heavy Chain Protein Sequence (CAA88724):  
  
VAKAKGNLEKMCRTLEDQLSELKTKNDEN-  
VRQVNDISGQRARLLTENGEGFRQLEEKEAL
- 4) Leave the other fields as found and hit the BLAST button.
- 5) A new window should pop up. Hit the Format button.

6) After a short wait the BLAST results window will come up and may well be hundreds of pages long — don't worry. There should be a long list of sequences that produced significant alignments. Although the search may pick up hundreds of sequences, they are in order of homology, so the ones you are interested in should be in the first 25 or so.

7) Further down the BLAST results page, after the list of sequences, each sequence will be aligned with the original trout sequence (as shown in the example) so that you can see how the two compare.

8) To compare your second fish, say bluefin tuna, with the other fish, you must perform a second BLAST search with the tuna sequence to obtain the protein alignments of tuna with the other fish. Alternatively, you can align 5 protein sequences yourself from your original search in a word processing document (use Courier font, this aligns sequences because all the letters are the same width) and have your students manually compare them.

## BLAST-SEARCHING QUESTIONS

Construct a simple phylogenetic tree based on the taxonomic data.

Does the taxonomic data support the molecular data? Please explain your answer.

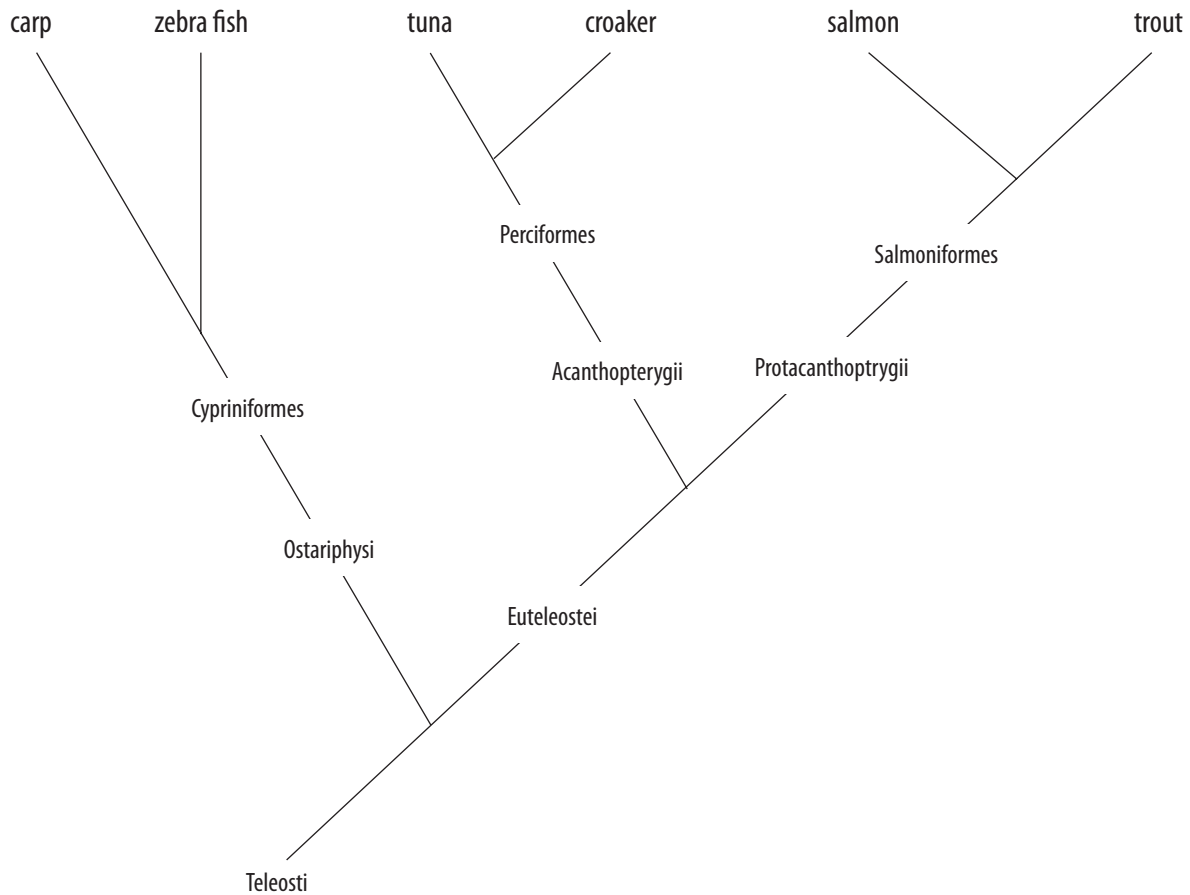
Why do scientists need to examine multiple data sets before determining evolutionary relatedness?



KEY

# ANSWERS TO BLAST-SEARCHING QUESTIONS

Construct a simple phylogenetic tree based on the taxonomic data (the large phylogenetic tree figure will be useful here).



Does the taxonomic data support the molecular data? Please explain your answer.

The trees do not entirely match. Both trees show a close relationship between salmon and trout and zebra fish and carp. However, tuna is in the same sub-phylum (Euteleostei) as salmon and trout, yet this does not concur with the molecular data and croaker is in the same order as tuna (Perciformes) and yet the amino acid sequence of croaker's myosin is much closer to salmon than tuna.

Why do scientists need to examine multiple data sets before determining evolutionary relatedness?

The statistical relevance of data grows as the size of the data set increases. The 60 amino acid segment of myosin heavy chain constitutes just 3% of the myosin heavy chain molecule, which is around 1,900 amino acids long. Performing a BLAST search with a larger portion of the molecule generates a cladogram with different relationships, demonstrating that the 60 amino acid piece is not large enough to provide a full picture of relatedness. However, even if the full-length myosin were compared, that is just a single protein out of the thousands generated by the organism. The data would be much stronger if the sequences of multiple proteins were compared and stronger still if molecular data were used with other types of classification data such as morphological data.



**FROM FINCHES TO FISHES IMPLEMENTATION PLAN — INTERDISCIPLINARY BRIDGES**

Activity	Arts	English	Health	Math	Science	Social Studies	Provided materials
Picture This: A Writing and Listening Exercise for Science and Non-Science Classrooms	X	X			X		Picture This handout
Darwin, the Writer		X			X	X	Darwin, the Writer handout

# PICTURE THIS

## A Writing and Listening Exercise for Science and Non-Science Classrooms



**Subjects: English, Biology**

**B**iological evolution — or, indeed, any complicated or controversial topic — can present an opportunity for teachers to help students develop skills as critical thinkers, thoughtful writers, and good listeners. Perhaps this last set of skills is the most vital of the three — as throughout their lives your students will benefit from an ability to listen respectfully and mindfully when other people express views and ideas that may seem to differ from their own.

Picture This serves several functions. This activity calls upon students to stretch linguistically and imaginatively—to use a range of vocabulary that typical class discussion might not require of them, and to make creative connections between what they already feel, think, and know and what they are about to learn. As its name suggests, Picture This enables students who are more visually oriented to relate images to ideas. And, like other activities recommended by DESTINY, it encourages active participation by all the students in your class.

We recommend this activity as a starting point for your evolution unit in a biology class. If you are a non-science teacher, you may find Picture This to be useful at the beginning of a unit involving an evolution-themed literary work (such as *Inherit the Wind*, *The Time Machine*, or *Cosmicomics*) or argumentative communication (such as an editorial, speech, or letter). However, Picture This can be adapted for a number of classroom uses. Though biological evolution is the topic described here, this writing and listening exercise will work well to engage students in many other topics and assignments.

## RESOURCES

- A large selection of photographs you have cut from magazines or catalogues. Almost any weekly or monthly magazine (*Time*, *People*, *Sports Illustrated*, *Smithsonian*, etc.) and many catalogues (particularly those that are related to travel or gardening) will yield images that are useful for Picture This.



So that your students will have a number of images from which to choose, provide two or three photographs per student (e.g., fifty or sixty photographs for a class of twenty-five students). The photographs should offer a range of images: landscapes, cityscapes, animals, machinery, objects, abstractions, and ordinary people (not celebrities or other people your students will recognize) in interesting situations or against interesting backdrops. These should be images your students can invest with their own thoughts and feelings.

- Picture This handout: enough copies so that each student has one to write on. (If you are unable to make copies of the Picture This handout, you can write the questions on your blackboard or on a transparency. Students can write their answers and glue their pictures on loose sheets of blank paper or in their journals.)
- Glue sticks: five or six for the class to share, so that your students can affix the photographs they select to their Picture This handouts.

## ACTIVITIES/PROCEDURES

### WHAT YOU DO

Display all the photographs on a large surface (a counter, several empty student desks, or even the floor will

work). Invite your students to come forward and survey the photographs. Ask each student to select the photograph that best illustrates or symbolizes her feelings or ideas about evolution. Give your students time to look at the pictures and to give some thought to their selection process. Each student selects one photograph.

**Variation: Group Work.** Divide your class into groups of five or six students. Give each group ten or twelve photographs. Ask the group to choose a photograph that represents the group's views. Have each group report to the class on its answers to the Picture This handout.

## WHAT YOUR STUDENTS DO

This activity is divided into Writing and Listening phases that enable all students to participate equally and simultaneously.

**Writing (10-15 minutes).** Give your students the following instructions: "From the photographs on display, select the one that best illustrates or symbolizes your feelings or ideas about evolution. Look at the pictures and give some thought to their selection process. After selecting a photograph, return to your desk and write down your answers to the following questions (listed on the Picture This handout). Use a glue stick to attach the photograph to your handout.

### Questions from Picture This Handout

1. Every picture deserves a title. Select a title, or a caption, for the picture you've chosen.

2. There were many pictures from which to choose. Why did this picture appeal to you? (Please write at least two sentences.)

3. How does the picture you've chosen reflect or symbolize your thoughts about evolution? (Please write at least three sentences.)

**Listening.** Now comes the opportunity for everyone's voice to be heard—and for everyone to listen to their classmates' ideas and opinions. Move around the class, asking every student to briefly describe the picture they selected and to read one of their answers aloud—any answer they feel most comfortable reading. Acknowledge each student positively—with a smile, or a "Thank you," or "Good work." A simple, friendly, and non-judgmental acknowledgement of each student's effort to express herself is what you will aim for.

Remember that this is a chance for your students to articulate ideas that may be rather difficult to express. Most students will be at the beginning of the process of learning about this complicated scientific concept; they are engaging the topic and readying themselves to learn about it. At the end, you may need to gently correct any of your students' misunderstandings about the science and its history that were revealed during the exercise; but do this in a general way, without pinpointing a particular student's error. At the very end, praise all your students for their good listening.

# PICTURE THIS: EVOLUTION

Paste or tape your picture here.

If your picture is large, you can  
place it on the back of this paper.

1. Every picture deserves a title. Select a title, or a caption, for the picture you've chosen.
2. There were many pictures from which to choose. Why did this picture appeal to you? (Please write at least two sentences.)
3. How does the picture you've chosen reflect or symbolize your thoughts about evolution? (Please write at least three sentences.)

# Darwin, the Writer



We cannot overestimate the importance of the written word to the development of the science of evolution and to its dissemination among a wide public of scientists and non-scientists that continues to grow. The first edition — 1,250 copies — of *On the Origin of Species* sold out entirely in 1859. New editions quickly followed. In the years since, the rhetorical style in which Darwin made his groundbreaking arguments has been an important reason for this book's continuing interest and influence.

That Darwin's work as a writer is entwined with his work as a scientist is important to know. The long list of his works published in his lifetime, beginning with his account of the five-year voyage that took him to the Galapagos Islands off South America (*Voyage of the Beagle*, 1839), attests to his productivity as a writer. The nature of many of his publications suggests his wish to convey news of his findings and to describe his life as a scientist in language that could be understood and appreciated by both expert and lay audiences.

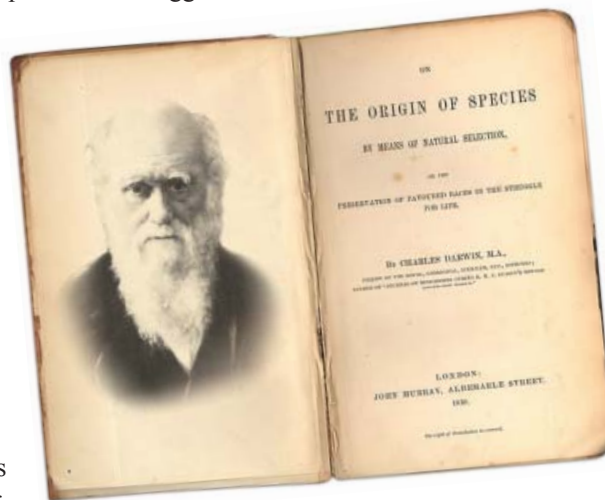
A letter to his publisher indicates the breadth of the audience Darwin envisioned for *On the Origin of Species*. He wrote: "My volume cannot be mere light reading, & some parts must be dry & some rather abstruse; yet as far I can judge perhaps very falsely, it will be interesting to all (& they are many) who care for the curious problem of the origin of all animate forms" (Darwin, 1859, April 2). While some parts may indeed be heavy going, Darwin's book as a whole is written to engage and inform a fairly wide audience (who might be interested and knowledgeable, but not necessarily expert in the field). *On the Origin of Species* was, in some senses, in its time, a work of popular science not unlike those we may find at Amazon.com or frequently on best-seller lists today.

In his autobiography, Darwin describes in some detail the creation and reception of a number of his publications, including his magnum opus:

*In September 1858 I set to work by the strong advice of [Charles] Lyell and [Joseph Dalton] Hooker to prepare a volume on the transmutation of species, but was often interrupted by ill-health. [...] It cost me thirteen months and ten days' hard labour. It was published under the title of the Origin of Species, in November 1859. Though considerably added to and corrected in the later editions, it has remained substantially the same book.*

*It is no doubt the chief work of my life. It was from the first highly successful. The first small edition of 1250 copies was sold on the day of publication, and a second edition of 3000 copies soon afterwards. Sixteen thousand copies have now (1876) been sold in England and considering how stiff a book it is, this is a large sale. It has been translated into almost every European tongue, even into such languages as Spanish, Bohemian,*

*Polish, and Russian. [...] Even an essay in Hebrew has appeared on it, showing that the theory is contained in the Old Testament! The reviews were very numerous; for a time I collected all that appeared on the Origin and on my related books, and these amount (excluding newspaper reviews) to 265; but after a time I gave up the attempt in despair. (Darwin, 1993, pp. 122-123)*



Though Darwin was modest in his assessment of his facility as a writer, he nonetheless cared to do his best. It is

clear that he worked hard to be a good writer. English teachers in particular will appreciate Darwin's methods: his use of outlines at the "pre-writing" stage, his quick roughing in of early drafts, and his subsequent work to pare, correct, and polish.

*I have as much difficulty as ever in expressing myself clearly and concisely; and this difficulty has caused me a very great loss of time; but it has had the compensating advantage of forcing me to think long and intently about every sentence, and thus I have been often led to see errors in reasoning and in my own observations or those of others.*

*There seems to be a sort of fatality in my mind leading*

*me to put at first my statement and proposition in a wrong or awkward form. Formerly I used to think about my sentences before writing them down; but for several years I have found that it saves time to scribble in a vile hand whole pages as quickly as I possibly can, contracting half the words; and then correct deliberately. Sentences thus scribbled down are often better ones than I could have written deliberately.*

*Having said this much about my manner of writing, I will add that with my larger books I spend a good deal of time over the general arrangement of the matter. I first make the rudest outline in two or three pages, and then a larger one in several pages, a few words or on word standing for a whole discussion or series or facts. Each of these headings is again enlarged and often transformed before I begin to write in extenso [“at full length”]. (Darwin, 1993, p. 137)*

The result of Darwin’s effort is prose that is typically lucid and sometimes beautiful. In *A Short History of English Literature*, Ifor Evans writes, “Charles Darwin would have disclaimed any right to be considered as a literary artist, yet the clarity of his style, and the very quietness with which he presents his profound conclusions, give to much of his work the qualities of art” (Evans, 1940, 1961, p. 220).

Because Darwin took pains with his writing, even the paragraph that ends the first edition of *On the Origin of Species* — a passage considered by many a reader to be both graceful and effective — did not escape modification. Darwin continued to tweak the text in subsequent editions: “an entangled bank” thus became “a tangled bank,” for instance, and “external” was deleted before “conditions of life.” He also made the significant addition of the phrase “by the Creator” in the second edition.

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## TEXTS OF THE LAST PARAGRAPHS IN THE FIRST AND SECOND EDITIONS

### First edition (published on November 24th, 1859)

It is interesting to contemplate an entangled bank, clothed with many plants of many kinds, with birds singing on the bushes, with various insects flitting about, and with worms crawling through the damp earth, and to reflect that these elaborately constructed forms, so different from each other, and dependent upon each other in so complex a manner, have all been produced by laws acting around us. These laws, taken in the largest sense, being Growth with Reproduction; Inheritance, which is almost implied by reproduction; Variability from the indirect and direct action of the external conditions of life, and from use and disuse; a Ratio of Increase so high as to lead to a Struggle for Life, and as a consequence to Natural Selection, entailing Divergence of Character and the Extinction of less-improved forms. Thus, from the war of nature, from famine and death, the most exalted object which we are capable of conceiving, namely, the production of the higher animals, directly follows. There is grandeur in this view of life, with its several powers, having been originally breathed into a few forms or into one; and that, whilst this planet has gone cycling on according to the fixed laws of gravity, from so simple a beginning endless forms most beautiful and most wonderful have been, and are being, evolved.

### Second edition (published on January 7th, 1860)

It is interesting to contemplate an entangled bank, clothed with many plants of many kinds, with birds singing on the bushes, with various insects flitting about, and with worms crawling through the damp earth, and to reflect that these elaborately constructed forms, so different from each other, and dependent upon each other in so complex a manner, have all been produced by laws acting around us. These laws, taken in the largest sense, being Growth with Reproduction; Inheritance, which is almost implied by reproduction; Variability from the indirect and direct action of the external conditions of life, and from use and disuse; a Ratio of Increase so high as to lead to a Struggle for Life, and as a consequence to Natural Selection, entailing Divergence of Character and the Extinction of less-improved forms. Thus, from the war of nature, from famine and death, the most exalted object which we are capable of conceiving, namely, the production of the higher animals, directly follows. There is grandeur in this view of life, with its several powers, having been originally breathed by the Creator into a few forms or into one; and that, whilst this planet has gone cycling on according to the fixed laws of gravity, from so simple a beginning endless forms most beautiful and most wonderful have been, and are being, evolved.



# DISCUSSION QUESTIONS, GUIDED READING, AND ACTIVITIES FOR CHARLES DARWIN'S *ON THE ORIGIN OF SPECIES*

**Subjects:** English, Science

## OVERVIEW

Teachers want their students to develop the patience and focus needed to carry out tasks that take place in more than one class period, such as following all the steps in a lengthy experimental process or attentively reading a whole novel. But asking students to pay very close attention while reading a short piece of writing—a poem, a newspaper article, or a passage from a longer work—can in a manageable timeframe help them to develop their analytical skills, their understanding of how language works, and their appreciation for what makes writing compelling and useful. If the passage they are asked to read with close attention is written well, is rich in ideas, and has historical or literary significance, so much the better!

Depending upon the teacher's guiding questions, the well-known last paragraph of Charles Darwin's *On the Origin of Species* can provide a focus for analytical reading in a biology class or an English class.

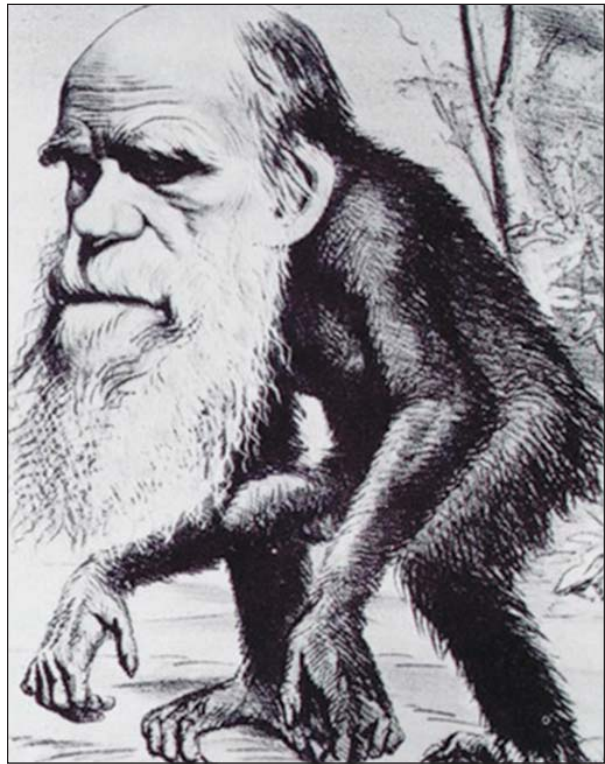
## RESOURCES

- Handout (attached) — The last paragraph of Charles Darwin's *On the Origin of Species*
- Optional — *On the Origin of Species* by Charles Darwin

## CLASS DISCUSSION

Listed below are a number of questions that you can use to guide your students' reading and analysis of this passage. Some questions may be more suitable for use in your lesson plan than others. Choose the questions that will meet the needs of your class.

- If you are a biology teacher, you may wish to ask questions that will encourage your students to draw on knowledge they have gained in your class or in their previous science courses. While introducing them to a significant work in the scientific literature, such a discussion will also help your students see the centrality of biological evolution to any understanding of modern biology.
- If you are an English teacher, you may wish to focus on questions that call on your students to think of the style of the passage, its vocabulary, the rhetorical techniques employed, and the literary period in which it



An 1871 caricature in *Hornet* magazine portraying Darwin as an ape.

was written. Discussion of this passage would fit into a unit on Victorian literature; as an introduction to a study of an evolution-themed literary work (see “Additional Activities for English Classrooms”); or as a model for analysis of rhetorical strategies in other works. If your students can also connect the passage with what they have learned in their science classes, you will have helped them build a useful bridge from discipline to discipline.

## GUIDED READING

You can lead the whole class in a discussion of the questions you have selected. Or you can divide your class into four groups. Assign one sentence to each group. Ask each group to answer all or some of the questions about the sentence assigned to them. Each group then reports their answers to the class as a whole.

### 1. The First Sentence

*It is interesting to contemplate an entangled bank, clothed with many plants of many kinds, with birds singing on the bushes, with various insects flitting about, and with worms crawling through the*



*damp earth, and to reflect that these elaborately constructed forms, so different from each other, and dependent upon each other in so complex a manner, have all been produced by laws acting around us.*

- Darwin begins his concluding paragraph with a very long sentence in which he describes “an entangled bank.” Have you been on the bank of a river or beside a creek? What did you see there? What would you expect to see there?

- List the organisms Darwin describes as inhabiting the entangled bank. (He mentions “plants” and “bushes,” “birds,” and “insects” and “worms.” Perhaps we can also include the people contemplating the bank — the “us” at the end of the sentence — in the list of organisms.)

- What verbs does Darwin use to describe the organisms on the bank? (The plants “clothe” the bank, the birds “sing,” the insects “flit,” and the worms “crawl.”) What would the sentence be like if you removed those verbs? Would the sentence be a good one? What would the sentence be like if you removed the organisms? Why does Darwin include all of these organisms?

- Can you envision the environment or ecosystem that Darwin describes in this sentence?

- Can you draw (or find) a picture that matches Darwin’s description of this bank?

- Why does Darwin include these words in this sentence: “entangled,” “elaborately,” “complex”? (Perhaps they reflect the complexities of the world around us, the interconnections among organisms and their environments, or the complexity of the ideas Darwin’s book has discussed.)

- What have you learned in biology that helps you understand what Darwin is referring to when he uses these phrases:

- a. “elaborately constructed forms” (biological development);
- b. “dependent upon each other” (interrelationships among organisms and their environments);
- c. “produced by laws” (genetics, biological evolution; also see the second sentence of the paragraph)?

- Why does Darwin include these words in the opening sentence of this paragraph: “interesting,” “contemplate,” and “reflect”? What is he asking his reader to do? Is Darwin saying something about his own thought processes?

- Why do you suppose Darwin chose to begin this important paragraph—the final impression he is leaving with his readers—with a somewhat detailed description of the “entangled bank”?

- Why does this sentence end with the words “around us”?

## 2. The Second Sentence

*These laws, taken in the largest sense, being Growth with Reproduction; Inheritance, which is almost implied by reproduction; Variability from the indirect and direct action of the external conditions of life, and from use and disuse; a Ratio of Increase so high as to lead to a Struggle for Life, and as a consequence to Natural Selection, entailing Divergence of Character and the Extinction of less-improved forms.*

- What does Darwin do in this long second sentence? Is he writing a summary of his book’s main points?

- How does Darwin punctuate the sentence? (There are three semi-colons, which serve to connect four independent clauses. A semi-colon is also used in the fourth sentence. You might ask your students to try to use this punctuation mark in their next writing assignment, or to look for it in their next reading assignment.)

- What have you learned in biology that helps you know something about the processes that Darwin briefly mentions in this sentence:

- a. “Growth with Reproduction” (biological development);
- b. “Inheritance, which is almost implied by reproduction” (genetic inheritance);
- c. “Variability from the indirect and direct action of the external conditions of life, and from use and disuse.” (biological adaptation);
- d. “A Ratio of Increase so high as to lead to a Struggle for Life, and as a consequence to Natural Selection, entailing Divergence of Character and the Extinction of less-improved forms.” (biological adaptation)?

## 3. The Third Sentence

*Thus, from the war of nature, from famine and death, the most exalted object which we are capable of conceiving, namely, the production of the higher animals, directly follows.*

- This is the shortest sentence in the paragraph, and yet it is very powerful. What do you think of Darwin’s use of these dramatic words: “war,” “famine,” “death,” and “exalted”? When you read or hear these words, what specific things do you personally think about? What meaning do these words have for you?

- To what is Darwin referring when he uses the phrases “war of nature” and “famine and death”? (A brief description of the inspiration Darwin drew from Malthus’s writing on population may be useful here. Thomas Robert Malthus [1766-1834] posited that, through calamitous events such as famine or outbreak of disease, populations were stabilized so that they would not outpace available resources.)

- To what is Darwin referring when he uses the phrases “exalted object” and “higher animals”? Could he be referring to his readers—to us?

- What have you learned in biology that helps you understand this sentence?

#### 4. The Fourth Sentence of the Paragraph — and the Last Sentence of the Book

*There is grandeur in this view of life, with its several powers, having been originally breathed by the Creator into a few forms or into one; and that, whilst this planet has gone cycling on according to the fixed laws of gravity, from so simple a beginning endless forms most beautiful and most wonderful have been, and are being, evolved.*

- Which three words did not appear in the first edition, but were added to the second edition? (“by the Creator”). Why might Darwin have included these words?

- Did references to a “Creator” appear in other parts of the first edition? (Yes. For example, in Chapter 6, in the section entitled “Organs of the extreme perfection and complication,” Darwin writes about the development of the eye:

*Have we any right to assume that the Creator works by intellectual powers like those of man? If we must compare the eye to an optical instrument, we ought in imagination to take a thick layer of transparent tissue, with a nerve sensitive to light beneath, and then suppose every part of this layer to be continually changing slowly in density, so as to separate into layers of different densities and thicknesses, placed at different distances from each other, and with the surfaces of each layer slowly*

*changing in form. Further we must suppose that there is a power always intently watching each slight accidental alteration in the transparent layers; and carefully selecting each alteration which, under varied circumstances, may in any way, or in any degree, tend to produce a distincter image. We must suppose each new state of the instrument to be multiplied by the million; and each to be preserved till a better be produced, and then the old ones to be destroyed. In living bodies, variation will cause the slight alterations, generation will multiply them almost infinitely, and natural selection will pick out with unerring skill each improvement. Let this process go on for millions on millions of years; and during each year on millions of individuals of many kinds; and may we not believe that a living optical instrument might thus be formed as superior to one of glass, as the works of the Creator are to those of man?*

A search of the many on-line texts of *The Origin of Species* will reveal other instances of Darwin’s use of the word “Creator.”)

- What contrast is Darwin drawing in the first and last sentences of this paragraph? In the last sentence he mentions “so simple a beginning.” How does this relate to the first sentence of the paragraph, in which he used the words “entangled,” “elaborately,” and “complex”? Is he referring to changes occurring in nature over time? Is Darwin contrasting the entangled bank in the present with a primordial scene in the long-distant past?

- Why does Darwin include these words in the final sentence of his book: “grandeur,” “beautiful,” and “wonderful”? What is he asking his reader to think about? Is Darwin saying something about his own thought processes?

- What does Darwin mean by the phrase “this view of life”? Can you put his view of life into your own words?

- What is your opinion of the last sentence of *On the Origin of Species*?

- What is your opinion of the final paragraph of *On the Origin of Species*? What did you learn from it that you did not know before? (Answers might include opinions about Darwin himself, about the book, or about the science of biological evolution.)

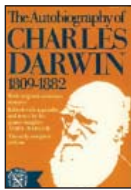
## ADDITIONAL ACTIVITIES FOR ENGLISH CLASSROOMS

- After your class analyzes the last paragraph of *On the Origin of Species*, ask each student to find another short passage of non-fiction writing that has also had an important impact on humankind (e.g., the Preamble to the United States Constitution or a portion of Martin Luther King Jr.'s "I Have a Dream" speech). Ask each student to consider where the passage's power lies: the beauty of the language, the strength of the argument, the historical moment it addressed, or other factors. You might wish to limit the assignment by genre, time frame, or nationality. Ask your students to explain their choices.

- Alternatively, you may wish to ask your students to find and analyze a short passage of writing that has had a powerful or important impact on them personally (e.g., the passage from a book they like, part of a letter or message they may have received, the lyrics of a song). Ask your students to explain their choices. Why has the author of each piece of writing succeeded in reaching his or her audience?

- Discussion of the last paragraph of *On the Origin of Species* would serve as a useful introduction to a unit on other evolution-themed literary works. We suggest several texts that we know work well in secondary classrooms:

- a. **Charles Darwin's *Autobiography*.** Darwin's *Autobiography*, prepared by one of his sons for publication several years after Darwin's death, was written primarily for a readership of family members — his children and grandchildren. It is an accessible book that with surprising openness and charm provides insights into Darwin's youth and gradual development from unfocused student (more interested in outdoor



sport, he was considered "a very ordinary boy, rather below the common standard in intellect") to influential scientist (Darwin, 1958, 1993, p. 28). Readers seeking to understand more about Darwin's childhood, family, and marriage, his life aboard the *Beagle*, his religious beliefs, and his career will find information about these topics in this short book. While the whole book has been assigned successfully to secondary students, you may prefer to make selections among the chapters, or perhaps assign chapters to groups of students. Selections from this book may be assigned as readings in an upper-level biology class, or they may be assigned during a unit on autobiography or memoir in an upper-level English class.

- b. ***The Time Machine*.** H. G. Wells, a pioneer in the science fiction, called this, his first book, a "scientific romance." Having been a student of T. H. Huxley (a vocal defender of evolution,



Huxley became known as "Darwin's Bulldog") and also a teacher of biology, Wells was well able to explore the "what ifs?" suggested by the swirl of ideas around evolutionary biology in the Victorian Age. *The Time Machine* (1895)

follows the adventures of a scientist and inventor (called "The Time Traveller") who builds a device that hurtles him forward along evolution's timeline. Arriving in the year 802,701, he encounters the strange life ways of the Eloi and the Morlocks, species that are humans' evolutionary descendents. *The Time Machine* is a novella, and therefore short enough to become a manageable reading assignment. In addition, a number of movies have been based on *The Time Machine*. We recommend the most recent, which starred Guy Pearce and Samantha Mumba in 2002. Though some liberties are taken with the plot, there is a useful emphasis on science, including evolution. There is even a futuristic librarian who discusses time travel as theme in literature. The special effects in this version will probably be more plausible to high school audiences today than those of George Pal's generally well-regarded effort of 1960.

- c. ***Inherit the Wind*.** Though it takes some liberties with the facts (adding a love interest where there was none, for instance), this drama by Jerome Lawrence and Robert E. Lee is recognizably

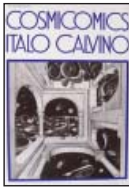


based on the Scopes "Monkey Trial." The play is set in a small Southern town patterned on Dayton, Tennessee, where John T. Scopes (Bertram T. Cates in the play), was charged with breaking a law recently enacted to prohibit

the teaching of evolution in the state's public schools. Though the Scopes trial was short-lived (lasting July 10-21, 1925), it generated national interest at the time, in large part because of the famous men representing each side in the dispute. Clarence Darrow (Henry Drummond in the play) was a famed criminal attorney who stepped in for the defense; William Jennings Bryan (Matthew Harrison Brady), a leading politician,

represented the prosecution. Both men were outstanding orators, and their dramatized rhetorical tangle ranges over science's impact on many facets of society: education, law, religion, politics, the press. *Inherit the Wind* was first produced in 1955 on Broadway. Several film versions have since been made, with the best considered to be the 1961 production, which features Spencer Tracy as Drummond.

- d. **Cosmicomics.** Italo Calvino's *Cosmicomics* (1965) is rather more abstract than the other works mentioned here. First published in Italian, this collection of short stories follows the adventures of beings in deep time — well before language, before dinosaurs, before vertebrae, before colors. While the whole book has been assigned successfully to secondary students, selected stories may serve as more accessible assignments. In particular, we recommend “The Aquatic Uncle,” which tells of the coelacanth, who prefers to stay in the water while his fellows move to the land. This story can be accompanied by information about the surprising discovery of living coelacanth off the coast of southern Africa in 1938, though this fish had been assumed to have become extinct tens of millions of years before. Students can even go online to view photographs and video clips of living coelacanths—large (reaching five feet in length), blue fish that existed in the time of the dinosaurs and also exist in our time, too.



- e. **Evolution in the news.** Because biological evolution continues to spur discussion in public forums, any number of news stories, opinion-editorials, letters to editors, and web-site content related to this topic may serve as current, useful for texts for analysis. A comparison of such a text with what your students have discovered in reading the last paragraph of *On the Origin of Species* may be a valuable exercise.



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## THE LAST PARAGRAPH OF CHARLES DARWIN'S *ON THE ORIGIN OF SPECIES*

### First edition (published on November 24th, 1859)

It is interesting to contemplate an entangled bank, clothed with many plants of many kinds, with birds singing on the bushes, with various insects flitting about, and with worms crawling through the damp earth, and to reflect that these elaborately constructed forms, so different from each other, and dependent upon each other in so complex a manner, have all been produced by laws acting around us. These laws, taken in the largest sense, being Growth with Reproduction; Inheritance, which is almost implied by reproduction; Variability from the indirect and direct action of the external conditions of life, and from use and disuse; a Ratio of Increase so high as to lead to a Struggle for Life, and as a consequence to Natural Selection, entailing Divergence of Character and the Extinction of less-improved forms. Thus, from the war of nature, from famine and death, the most exalted object which we are capable of conceiving, namely, the production of the higher animals, directly follows. There is grandeur in this view of life, with its several powers, having been originally breathed into a few forms or into one; and that, whilst this planet has gone cycling on according to the fixed laws of gravity, from so simple a beginning endless forms most beautiful and most wonderful have been, and are being, evolved.

### Second edition (published on January 7th, 1860)

It is interesting to contemplate an entangled bank, clothed with many plants of many kinds, with birds singing on the bushes, with various insects flitting about, and with worms crawling through the damp earth, and to reflect that these elaborately constructed forms, so different from each other, and dependent upon each other in so complex a manner, have all been produced by laws acting around us. These laws, taken in the largest sense, being Growth with Reproduction; Inheritance, which is almost implied by reproduction; Variability from the indirect and direct action of the external conditions of life, and from use and disuse; a Ratio of Increase so high as to lead to a Struggle for Life, and as a consequence to Natural Selection, entailing Divergence of Character and the Extinction of less-improved forms. Thus, from the war of nature, from famine and death, the most exalted object which we are capable of conceiving, namely, the production of the higher animals, directly follows. There is grandeur in this view of life, with its several powers, having been originally breathed by the Creator into a few forms or into one; and that, whilst this planet has gone cycling on according to the fixed laws of gravity, from so simple a beginning endless forms most beautiful and most wonderful have been, and are being, evolved.

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