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Molecular Explorations

Ocean Exploration / Grades 9-12 / Life Science, Chemistry

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Focus Question

What are some molecular biology techniques that scientists use to explore Earth's deep ocean?

Learning Objectives

- Students will be able to explain and carry out a simple process for separating DNA from tissue samples.
- Students will be able to explain and carry out a simple process for separating complex mixtures.
- Students will be able to explain the process of restriction enzyme analysis.

Links to Overview Essays and Resources Useful for Student Research

http://www.oceanservice.noaa.gov/topics/oceans/oceanex/ http://oceanexplorer.noaa.gov/explorations/03bio/background/molecular/molecular.html

Materials

- Electrophoresis chambers and power supplies (see "Learning Procedure")
- 1% agarose gel (Carolina Biological Supply No. WW-21-7075)
- TRIS/Borate/EDTA buffer (Carolina Biological Supply No. WW-21-9025)
- Gel casting trays (Carolina Biological Supply No. WW-21-3655)

- Four test tubes, each containing a pure food color, plus a fifth test tube containing a mixture of food colors; one set for each student group
- Five micropipettes (Carolina Biological Supply No. WW-21-1022)
- Liquid dish detergent, approximately 100 ml
- Sodium chloride, approximately 100 g
- Fresh meat tenderizer, approximately 50 g
- Distilled water, approximately 21
- 95% ethanol
- Ice, crushed, approximately 3 kg
- Paper towels
- Plastic sandwich bag, one for each student group
- Student instruction handout for each student (see "Learning Procedure")
- OPTIONAL: additional supplies for electrophoresis of student-prepared DNA extracts (see Learning Procedure, steps 5 and 6)

Audio/Visual Materials

None

Teaching Time

Three or four 45-minute class periods, depending upon the number of activities selected.

Seating Arrangement

Laboratory groups of two to three students.

Maximum Number of Students

30

Key Words

DNA Electrophoresis Restriction enzyme Molecular biology

Background Information

In the past twenty years, scientists have explored areas of the deep ocean that have never been visited before. These expeditions have discovered hundreds of new species, and even new ecosystems, but 95% of Earth's ocean remains unexplored.

Why is it important to explore the deep ocean? Consider this: Most drugs in use today were initially found in living organisms, and almost all of these organisms are terrestrial. Recent systematic searches for new drugs have shown that antibiotic, anti-cancer, and anti-inflammatory substances are much more common among marine invertebrates than among terrestrial organisms. Particularly promising invertebrate groups include sponges, tunicates, ascidians, bryozoans, octocorals, and some molluscs, annelids, and echinoderms (see the Ocean Exploration lesson "Benthic Drugstore" at

http://oceanexplorer.noaa.gov/explorations/03bio/background/edu/media/Meds_Drugstore.pdf for more information).

The National Oceanic and Atmospheric Administration (NOAA) created the Office of Ocean Exploration in 2001 to spearhead efforts to learn more about unexplored areas in the Earth's ocean. Recent expeditions have explored the Submarine Ring of Fire in the Mariana Arc, the New England seamount chain, the Gulf of Alaska seamounts, the Arctic Ocean, the Black Sea, the Galapagos rift, and the Northwestern Hawaiian Islands. Recent accomplishments include bioprospecting for new deep sea medicines, observation and monitoring of a new underwater volcano in Hawaii, testing non-invasive research tools to observe and collect deep-sea samples, and investigation of new species in previously unexplored deep-sea ecosystems.

Many of these accomplishments would have been impossible a few years ago, but innovations in equipment and techniques have given researchers powerful new tools for exploring Earth's ocean environments. Molecular biology techniques, in particular, are being used to answer questions about ecology, biodiversity, evolutionary genetics and systematics of marine organisms, as well as in prospecting for new natural products that can be used to treat disease. These techniques include DNA extraction, RNA extraction, the use of gel electrophoresis to visualize DNA and RNA, polymerase chain reaction (PCR), and DNA sequencing. This lesson is intended to introduce students to several of these techniques.

Learning Procedure

[NOTE: This lesson is based on activities designed by Ellen Averill, Karen Kyker, Sandy Collins, and Theresa Knapp while participating in the 1993 Woodrow Wilson Biology Institute. These activities are used with permission from the Woodrow Wilson National Fellowship Foundation. Visit <u>http://woodrow.org</u> for information on other activities and programs. The restriction enzyme activity is obtained from the Access Excellence Classic Collection: <u>http://www.accessexcellence.org</u>.]

- 1. Download the following activities:
- "Isolation of DNA from Onion" (<u>http://www.woodrow.org/teachers/bi/1993/isolation2.html</u>)
- "Electrophoresis Analogy" (<u>http://www.woodrow.org/teachers/bi/1993/electrophoresis.html</u>)
- "Rainbow Electrophoresis" (<u>http://www.woodrow.org/teachers/bi/1993/rainbow.html</u>)
- "Desktop Electrophoresis Lab Moving Molecules" (http://www.woodrow.org/teachers/bi/1993/moving.html)

 "How Restriction Enzyme, Probes and RFLP's Work" (<u>http://www.accessexcellence.org/AE/AEC/CC/word_activity.html</u>)

There are two options for obtaining the equipment needed for the electrophoresis activity. The first is to buy electrophoresis chambers and power supplies from a laboratory supply company (cost approximately \$400 and up). The second is to build your own chambers and power supplies as directed in the "Desktop Electrophoresis Lab" activity (cost approximately \$20 per system).

- 2. Prepare detergent/salt solution, meat tenderizer solution, sodium chloride solution, electrophoresis gels, TRIS/Borate/EDTA buffer, and student instruction sheets (from the downloaded activities) prior to the lab. If you want to use the (lower cost) "Desktop Electrophoresis" apparatus, prepare the chambers and power supplies as well, unless you plan to have students do this.
- 3. Briefly discuss the fact that much of the Earth's ocean is totally unexplored, particularly the deepest areas. Highlight some of the discoveries that have been made by recent expeditions to study the deep ocean. You may want to mention hydrothermal vent communities, cold seeps, methane ices, and deep sea medicines. The following Web sites have useful information for this discussion:

http://www.bio.psu.edu/cold_seeps for a virtual tour of a cold seep community; http://www.bio.psu.edu/hotvents for a virtual tour of a hydrothermal vent community; http://oceanexplorer.noaa.gov/explorations/deepeast01/background/fire/fire.html and http://oceanexplorer.noaa.gov/explorations/03windows/background/hydrates/hydrates.html for background on methane ices; and http://oceanexplorer.noaa.gov/explorations/03bio/ for background on deep sea medicines.

- 4. Tell students that recent advances in molecular biology techniques provide powerful tools for many kinds of scientific investigations, from marine research to crime scene investigations. Discuss the ways in which ocean exploration expeditions might use these techniques. Suggestions should include:
- distinguishing between different species and different growth forms or life cycle stages of the same species (see the lesson plan "Bad Algae" <u>http://oceanservice.noaa.gov/education/lessons/bad_algae.html</u> for an example of why this is important);
- investigating genetic similarities between species to obtain clues about evolutionary relationships and geographic dispersal; and
- determining the genes responsible for synthesizing natural products that can be used as disease treatments, such as anti-inflammatory or anti-tumor drugs (this information could allow these products to be synthesized, rather than endangering wild populations of species that produce these chemicals).

Tell students that the purpose of these activities is to introduce three of these techniques.

5. Have students complete the "Isolation of DNA from Onion" lab activity as directed in the student instructions.

If you want students to use their DNA isolates for the electrophoresis activity, have them transfer the DNA to a clean test tube, rinse with 70% ethanol to remove excess salts, then pour off the ethanol from the test tube. Add 0.5 ml distilled water to the test tube, cover, and refrigerate until the next day. Each student or student group should record their procedures and results in a lab notebook or written report.

6. Introduce the technique of electrophoresis using the "Electrophoresis Analogy." Have students complete the "Rainbow Electrophoresis" lab activity as directed in the student instructions. Each group should prepare written answers to the questions included in the activity, either in a lab notebook or separate written report.

If you are having students use the DNA extracts prepared in Step 5 for this activity, have them put 85 ml of their extract into a clean test tube, add 15 ml TRIS/Borate/EDTA buffer, and load the electrophoresis gels as directed in the student instruction sheets. Run the gels at 81 volts (use nine batteries in the power supply) for about one hour. Stain the gels by soaking overnight in a 0.02% solution of methylene blue in distilled water. Procedures and results should be recorded in a lab notebook or written report.

7. Introduce the technique of restriction enzyme cleavage using the word analogy activity described in "How Restriction Enzyme, Probes and RFLP's Work." If you want to do an actual restriction enzyme cleavage procedure, visit the University of Arizona's Biotech Project Web site (<u>http://biotech.biology.arizona.edu/labs/labs.html</u>) for a list of biotechnology laboratory experiments. The teacher guide and student guide for the Restriction Enzyme Analysis lab can be downloaded from <u>http://biotech.biology.arizona.edu/word/restenzy_tg.doc</u> and <u>http://biotech.biology.arizona.edu/word/restenzy_sg.doc</u>.

The Bridge Connection

The Bridge is a growing collection online marine education resources. It provides educators with a convenient source of useful information on global, national, and regional marine science topics. Educators and scientists review sites selected for the Bridge to insure that they are accurate and current.

<u>http://www.vims.edu/bridge</u> – Click on "Ocean Science Topics" in the navigation menu on the left, then "Human Activities," then "Technology"

The "Me" Connection

Have students write a brief essay describing three personal benefits that might result from molecular-level explorations of the deep ocean.

Extensions

- 1. Visit <u>http://oceanexplorer.noaa.gov/</u> for details of Ocean Exploration expeditions (many with detailed daily logs) and over 150 hands-on, standards-based lesson plans and a curriculum based on the explorations.
- 2. For a virtual restriction mapping activity, visit <u>http://www.geospiza.com/outreach/bio21/materials/restriction_mapping.pdf</u>
- 3. Biological supply companies have a variety of materials and kits suitable for other DNA research techniques.

Resources

<u>http://www.oceanservice.noaa.gov/topics/oceans/oceanex/</u> – Introductory page for the Ocean Exploration website

http://biotech.biology.arizona.edu/ – Web site for the University of Arizona's Biotech Project

<u>http://www.dnaftb.org/dnaftb/</u> – An animated primer on the basics of DNA, genes, and heredity from the DNA Learning Center at the Cold Spring Harbor Laboratory

<u>http://www.woodrow.org/teachers/bi/1993/</u> – Background and activities from the 1993 Woodrow Wilson Biology Institute on Biotechnology

National Science Education Standards

Content Standard A: Science as Inquiry

- Abilities necessary to do scientific inquiry
- Understandings about scientific inquiry

Content Standard B: Physical Science

- Abilities necessary to do scientific inquiry
- Understandings about scientific inquiry

Content Standard C: Life Science

- The cell
- Molecular basis of heredity
- Interdependence of organisms
- Biological evolution

Content Standard E: Science and Technology

• Understandings about science and technology

Content Standard F: Science in Personal and Social Perspectives

- Natural resources
- Science and technology in local, national, and global challenges

Ocean Literacy Essential Principles and Fundamental Concepts

Essential Principle 1. The Earth has one big ocean with many features.

- Fundamental Concept a. The ocean is the dominant physical feature on our planet Earth covering approximately 70% of the planet's surface. There is one ocean with many ocean basins, such as the North Pacific, South Pacific, North Atlantic, South Atlantic, Indian and Arctic.
- Fundamental Concept b. An ocean basin's size, shape and features (such as islands, trenches, mid-ocean ridges, rift valleys) vary due to the movement of Earth's lithospheric plates. Earth's highest peaks, deepest valleys and flattest vast plains are all in the ocean.
- Fundamental Concept c. Throughout the ocean there is one interconnected circulation system powered by wind, tides, the force of the Earth's rotation (Coriolis effect), the Sun, and water density differences. The shape of ocean basins and adjacent land masses influence the path of circulation.
- Fundamental Concept d. Sea level is the average height of the ocean relative to the land, taking into account the differences caused by tides. Sea level changes as plate tectonics cause the volume of ocean basins and the height of the land to change. It changes as ice caps on land melt or grow. It also changes as sea water expands and contracts when ocean water warms and cools.
- Fundamental Concept h. Although the ocean is large, it is finite and resources are limited.

Essential Principle 2. The ocean and life in the ocean shape the features of the Earth.

- Fundamental Concept a. Many earth materials and geochemical cycles originate in the ocean. Many of the sedimentary rocks now exposed on land were formed in the ocean. Ocean life laid down the vast volume of siliceous and carbonate rocks.
- Fundamental Concept b. Sea level changes over time have expanded and contracted continental shelves, created and destroyed inland seas, and shaped the surface of land.
- Fundamental Concept e. Tectonic activity, sea level changes, and force of waves influence the physical structure and landforms of the coast.

Essential Principle 3. The ocean is a major influence on weather and climate.

• Fundamental Concept a. The ocean controls weather and climate by dominating the Earth's energy, water and carbon systems.

- Fundamental Concept e. The ocean dominates the Earth's carbon cycle. Half the primary productivity on Earth takes place in the sunlit layers of the ocean and the ocean absorbs roughly half of all carbon dioxide added to the atmosphere.
- Fundamental Concept f. The ocean has had, and will continue to have, a significant influence on climate change by absorbing, storing, and moving heat, carbon and water.
- Fundamental Concept g. Changes in the ocean's circulation have produced large, abrupt changes in climate during the last 50,000 years.

Essential Principle 4. The ocean makes Earth habitable.

- Fundamental Concept a. Most of the oxygen in the atmosphere originally came from the activities of photosynthetic organisms in the ocean.
- Fundamental Concept b. The first life is thought to have started in the ocean. The earliest evidence of life is found in the ocean.

Essential Principle 5. The ocean supports a great diversity of life and ecosystems.

- Fundamental Concept a. Ocean life ranges in size from the smallest virus to the largest animal that has lived on Earth, the blue whale.
- Fundamental Concept b. Most life in the ocean exists as microbes. Microbes are the most important primary producers in the ocean. Not only are they the most abundant life form in the ocean, they have extremely fast growth rates and life cycles.
- Fundamental Concept c. Some major groups are found exclusively in the ocean. The diversity of major groups of organisms is much greater in the ocean than on land.
- Fundamental Concept d. Ocean biology provides many unique examples of life cycles, adaptations and important relationships among organisms (such as symbiosis, predator-prey dynamics and energy transfer) that do not occur on land.
- Fundamental Concept e. The ocean is three-dimensional, offering vast living space and diverse habitats from the surface through the water column to the seafloor. Most of the living space on Earth is in the ocean.
- Fundamental Concept f. Ocean habitats are defined by environmental factors. Due to interactions of abiotic factors such as salinity, temperature, oxygen, pH, light, nutrients, pressure, substrate and circulation, ocean life is not evenly distributed temporally or spatially, i.e., it is "patchy". Some regions of the ocean support more diverse and abundant life than anywhere on Earth, while much of the ocean is considered a desert.
- Fundamental Concept g. There are deep ocean ecosystems that are independent of energy from sunlight and photosynthetic organisms. Hydrothermal vents, submarine hot springs, and methane cold seeps rely only on chemical energy and chemosynthetic organisms to support life.

Essential Principle 6. The ocean and humans are inextricably interconnected.

• Fundamental Concept a. The ocean affects every human life. It supplies freshwater (most rain comes from the ocean) and nearly all Earth's oxygen. It moderates the Earth's climate, influences our weather, and affects human health.

- Fundamental Concept b. From the ocean we get foods, medicines, and mineral and energy resources. In addition, it provides jobs, supports our nation's economy, serves as a highway for transportation of goods and people, and plays a role in national security.
- Fundamental Concept c. The ocean is a source of inspiration, recreation, rejuvenation and discovery. It is also an important element in the heritage of many cultures.
- Fundamental Concept e. Humans affect the ocean in a variety of ways. Laws, regulations and resource management affect what is taken out and put into the ocean. Human development and activity leads to pollution (such as point source, non-point source, and noise pollution) and physical modifications (such as changes to beaches, shores and rivers). In addition, humans have removed most of the large vertebrates from the ocean.
- Fundamental Concept f. Coastal regions are susceptible to natural hazards (such as tsunamis, hurricanes, cyclones, sea level change, and storm surges).
- Fundamental Concept g. Everyone is responsible for caring for the ocean. The ocean sustains life on Earth and humans must live in ways that sustain the ocean. Individual and collective actions are needed to effectively manage ocean resources for all.

Essential Principle 7. The ocean is largely unexplored.

- Fundamental Concept a. The ocean is the last and largest unexplored place on Earth—less than 5% of it has been explored. This is the great frontier for the next generation's explorers and researchers, where they will find great opportunities for inquiry and investigation.
- Fundamental Concept b. Understanding the ocean is more than a matter of curiosity. Exploration, inquiry and study are required to better understand ocean systems and processes.
- Fundamental Concept c. Over the last 40 years, use of ocean resources has increased significantly, therefore the future sustainability of ocean resources depends on our understanding of those resources and their potential and limitations.
- Fundamental Concept d. New technologies, sensors and tools are expanding our ability to explore the ocean. Ocean scientists are relying more and more on satellites, drifters, buoys, subsea observatories and unmanned submersibles.
- Fundamental Concept e. Use of mathematical models is now an essential part of ocean sciences. Models help us understand the complexity of the ocean and of its interaction with Earth's climate. They process observations and help describe the interactions among systems.
- Fundamental Concept f. Ocean exploration is truly interdisciplinary. It requires close collaboration among biologists, chemists, climatologists, computer programmers, engineers, geologists, meteorologists, and physicists, and new ways of thinking.