DNA Extraction from Bacteria

Lab Safety
Safety goggles should be worn during the lab
Carefully pour all reagents during this lab
Clean work area thoroughly after lab completion
Return all materials to appropriate location as directed

Overview
This 90 minute lesson (two class periods of 45 minutes) is for students in grades 5 through 9 to explore the extraction of DNA from bacteria. Through the Lab Center website that provides visualizations, students will learn a basic procedure for extracting DNA from prokaryotic cells (bacteria). They will define key concepts from the lesson, use the scientific inquiry method to conduct an experiment, interact with the media via specially designed activity sheets that support their learning, compare and contrast prokaryotic and eukaryotic cells, make a decision on the most efficient method of DNA extraction from prokaryotic cells, and examine real DNA.

Learning Outcomes
Students will be able to:
- use a basic procedure to extract DNA from bacteria cells.
- compare and contrast eukaryotic and prokaryotic cells.
- describe the significance of understanding cell composition in a DNA extraction.
- discuss cell structures to determine the most efficient method for DNA extraction from prokaryotic cells.
- examine real DNA without a microscope.

Prerequisite Knowledge & Skills
Knowledge
Students should:
- be familiar with the structure and function of the DNA molecule.
- be familiar with the structure and function of cell structures, including the cell membrane.
- understand the relationship between chromosomes and DNA.

Skills
Students should be able to:
- read the temperature from a thermometer in degrees Celsius.
- use a transfer pipette.
- measure with a graduated cylinder.
- conduct careful observations.
- record, analyze, and report data from observations.

Misconceptions
Students sometimes believe that all bacterial strains are infectious. They also expect to see the double helix at the conclusion of the lab.

Materials and Equipment
- Student lab note book
- Sentence strips
- LCD projector and a computer with internet access or copy projectable information to transparencies and project using a overhead projector
- 5 ml bacterial culture in a plastic test tube with a screw cap-1 per student pair
- Transfer pipettes-1 per student pair
- Water bath- with temperature set between 65-75°C - 1 per class
- Small racks or large cups to hold materials vertically
- Clear plastic inoculating loops-1 per student pair
- Permanent markers-1 per student pair
- Photocopy the corresponding printable student worksheets
- See "Bowl" icon on Teacher's desk on Lab Center website for lab materials

Lesson Structure

Pre-lab: Day 1 (1 class period of 45 minutes).

Teacher Prep
- Print and copy "Background Reading" from the **Student Lab Notebook** on website
- Review the history of the DNA molecule on the DNA Interactive Internet site at: www.dnai.org > Code
- Read the "Background Information" and "Teaching Tips" in the **Teacher's Guide**
- Copy the comparison chart from the **Student Lab Notebook** on a transparency
- Prepare materials: sentences strips, student worksheets
- Make sure computers with internet access are available

Before Class
Students will receive "Background Reading" to read for homework the night before starting the lab. They will write 2-3 questions they have about the information.
Students will also highlight any unfamiliar terms and write the meaning of one of the terms that they have highlighted according to the context in which the word is used in the "Background Reading."

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During Class:
Pair –Square (2 pairs) - Using the questions they developed from the homework assignment, each group will choose the question that they find most interesting. Each group will record this question on a sentence strip to be collected by the teacher and posted in the classroom. The teacher will also post one or two of his/her own questions.

Mini-lesson – This activity will help the students to understand the similarities and differences between prokaryotic and eukaryotic cells and to review the

Hand out a copy of the Venn diagram sheet (**Print from Student Lab Notebook**) to each student.
Discuss the similarities and differences between prokaryotic and eukaryotic cells. Instruct the students to use the Venn diagram to compare and contrast.

Review the cellular parts of animal and bacteria cells with the class from the following websites:
http://cache.eb.com/eb/image?id=63515&rendTypeId=4 (animal cell) and
http://morayeel.louisiana.edu/SeaweedsLab/Gavio/bacterial%20cell%20copy (Bacterium) or copy the pictures of the animal and bacteria cell on to a transparency.

Discuss some of the basic characteristics of bacteria such as prokaryotic, unicellular, and rapid cell division with the class. Instruct them to write what they have learned about the characteristics of bacteria on the student's work sheet entitled "Characteristics of Bacteria" from the **Student Lab Notebook.**

As a pre-lab exit strategy, ask each group to respond to one of the questions that they may now be able to answer and hand it to the teacher as they leave the class.

Lab Activity: Day 2 (This lab activity will take 1 class period of 45 Minutes)

Teacher Prep

- Prepare bacterial cultures as described in the **Recipes** section, several days prior to the lab.
- Photocopy "Video Quest" worksheets from **Student Lab Notebook** for each set of students.
- Prepare water bath. Temperature should range from 65-75°C.
- Set up stations with appropriate material for each pair of students. This should include a rack, a permanent marker, soap, ethanol, bacterial culture, a dropper and a plastic rod.

Part 1

- Use the Dolan DNA Learning Center website (http://www.dnalc.org/labcenter/dnaextraction) as a tool for introducing this lab. Use LCD projector along with a computer to project the introduction to the lab. The introduction has some illustrations that are used to explain what the students will do in this lab. Instruct students to read the questions on the "Video Quest" sheet and complete the questions as they watch the video clip.
- Introduce the bacteria (*E.coli)* being used for the experiment to the class. Discuss the numerous strains that exist, such as pathogenic strains, strains that live in the human body and the harmless strains that are used for research (such as the mm294 *E.coli* bacteria used for this experiment.)
- Ask students how they might extract DNA from prokaryotic cells. They should at this point realize that the outer membrane is the only barrier between a bacterial cell's DNA and the outside environment.
- Point out that breaking through a cell membrane can be compared to breaking through a wall. If a builder needed to knock down a wall, how would he or she determine which tools might work best? What specifically would the builder need to know about the wall itself?

Part 2

- Have students examine the diagram of the cell membrane (diagram found in **Student Lab Notebook**) to review the components of the cell membrane. Make sure that the students understand that lipids are molecules of fat. Ask the students if a membrane is made primarily of fat molecules, what kind of tool could be used to break it down, or dissolve it?
- Discuss the fact that household soaps/detergents are used all the time to dissolve fat from greasy dishes, skin and laundry. In this experiment, dishwashing detergent dissolves the prokaryotic cell membrane in the same way, thus releasing DNA from the cell.
- Ask a student to read steps 1-4 of the procedures aloud and demonstrate each step then give students the opportunity to begin their experiments in small groups of 4.
- During the 15-minute incubation, discuss what is happening inside all of the tubes, and what will need to be done next with the class. Once the DNA is released from the cells, it needs to be precipitated (separated from the solution) from the soapy solution where it floats..
- Have another student read step 5 from the procedure, while the rest of the class listens. Instruct students to draw a diagram of what will happen when DNA precipitation with ethanol is completed in step 5. Make sure students understand that ethanol is a type of alcohol, similar to rubbing alcohol. Because of its chemical properties, DNA is not soluble in ethanol,

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and it can therefore be used to separate the DNA from the soapy solution in the tube.

- Demonstrate how to use the transfer pipette to add ethanol. Slowly pour ethanol down the side of a slanted tube with bacteria and emphasize the importance of holding the tube on an angle.
- Show students how to spool the DNA from the ethanol layer using a plastic rod without breaking through the soap layer. It is essential that the two layers remain separated.
- If students would like to keep their DNA samples, they can be stored in small tubes of ethanol indefinitely.

Post Lab-Day 3 (This post-lab will take 1 class period of 45 minutes)

Analysis & Discussion

- Revisit the pre-lab questions. Which can you answer? If you couldn't what additional information do you need, and can you obtain the information?
- Ask students how it was possible to see real DNA without the help of a microscope, and how James Watson and Francis Crick were able to determine the double helical shape of the DNA molecule.
- Discuss the fact that the DNA extraction is an experiment that is not only used by scientists, but also by detectives (fingerprinting) and doctors (disease diagnosis.)

Students will discuss the following questions in groups and then students will write each question with individual answers to the questions in their **lab note books**.

1. Why was it important to shake the tube with the bacterial cells and soap?
2. Explain the importance of placing the tube with the soap and bacterial cells in the hot water bath for 15 minutes.
3. Describe what happens after the ethanol is added to the soap solution.
4. Explain the benefits of using a prokaryotic organism for a DNA extraction.

Applications

Instruct students to work with a partner. Each Group of 4 students (at each lab table) will select a reporter for each lab table. Have students read the questions on their own, discuss the questions with a partner then write their responses to the questions in their lab note books.

1. How is DNA extraction important to forensic scientists?
2. How was the anthrax DNA used to track the source of anthrax?

Use this website to get background information on Anthrax: http://www.msnbc.msn.com/id/25991925/

Further Exploration

Write a one (1) page **essay** about the ethical, legal, and social implications or issues of recombining DNA into bacterial cells. Use this website for background information: http://www.genome.gov/25019880

Write a **recipe** for DNA extraction as if you were going to put it in a cookbook. You may include diagrams.

Use a different protocol to extract DNA from eukaryotic cells, such as fruit. Compare and contrast the results.

Explore the *Recovering the Romanovs* activity on DNAi, at: www.dnai.org > Applications > Recovering the Romanovs. Students can learn how DNA extracted from skeletal remains can be used to identify individuals form the Romanov family of Russia, and solve the mystery of the missing Princess Anastasia themselves by analyzing mitochondrial DNA sequences online!

Use use the following vocabulary words to create a poem entitled "DNA Extraction" : Bacteria, extraction, cell membrane, eukaryotic cell, prokaryotic cell, DNA (Deoxyribonucleic Acid), water bath and ethanol.

Vocabulary (see glossary for definitions)

Extract
Bacteria
Pathogenic strain
Deoxyribonucleic Acid (DNA)
Chromosome
Eukaryotic
Prokaryotic
Animal cell
Bacterial culture
Cell membrane
Molecules
Incubation

Resources

Web Sites:
http://www.bacteriamuseum.org/main1.shtml
The Virtual Museum of Bacteria

http://www.dnaftb.org
The Dolan DNA Learning Center's Internet site.
Use this site to explore various genetics concepts from inheritance to genetic engineering.

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http://www.dnai.org
The Dolan DNA Learning Center's Internet site.
Use this site to learn about the past, present and future of DNA science.

http://www.ygyh.org
The Dolan DNA Learning Center's Internet site.
Use this site to learn about the connection between genes and health.

http://www.msnbc.msn.com/id/25991925/
Scientist: DNA led agents to anthrax suspect

http://www.genome.gov/25019880
This website has vignettes and a video on the ethical, legal, and social implications of genetic engineering.

Books:

Balkwill, F. (2002). *Enjoy your cells*. MN: Carolrhoda Books, Inc.

Balkwill, F. (1990). *Cells are us*. MN: Carolrhoda Books, Inc.

Balkwill, F. (2002). *Have a nice DNA.* MN: Carolrhoda Books, Inc.

Hoagland, M., & Dodson, B. (1995). *The way life works*. MN: Carolrhoda Books, Inc.

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