

EDUCATION COMPANY®

LESSON PLAN: **Exploring DNA** Grade level: 6-12, College Subject area: Biotechnology

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General Overview:

LEARNING OBJECTIVES:

In this lesson, students will explore the structure and function of DNA. First, students will explore the structure of DNA by building a model. Next, they will explore DNA's physical characteristics by isolating it from cheek cells. These lessons will give students an opportunity to visualize many characteristics of DNA.

ESSENTIAL KEY QUESTIONS:

- What is DNA?
- What is the structure of DNA?
- How can we purify DNA from cells?

CONTENT STANDARDS:

NGSS Alignment:

<u>HS-LS1:</u> From Molecules to Organisms: Structures and Processes
<u>HS-LS3:</u> Heredity: Inheritance and Variation of Traits
LS3.A: Inheritance of Traits
LS3.B: Variation of Traits
<u>Science and Engineering Practices:</u>

Asking Questions
Developing and Using Models
Analyzing and Interpreting Data

<u>Crosscutting Concepts:</u>

Cause and Effect
Structure and Function

Common Core State Standards:

<u>ELA/Literacy:</u> RST.11-12.1 RST.11-12.9 WHST.9-12.9



Lesson Introduction:

The basic unit of all living organisms, from bacteria to humans, is the cell. Contained within the nucleus of these cells is a molecule called deoxyribonucleic acid (or DNA). Today, we know that DNA is the blueprint used to build an organism – our genetic makeup, or genotype, controls our phenotype (observable characteristics). The directions coded for by our genes controls everything from growth and development to cell specification, neuronal function, and metabolism.

The Swiss physician Friedrich Miescher discovered DNA in 1868, when he purified a novel substance from the nucleus of white blood cells. This molecule, which he called "nuclein", had chemical properties unlike any substance previously identified. By the end of the 19th century, scientists had described DNA as a polymer composed of building blocks known as nucleotides. Most scientists believed that DNA was too simple to comprise the genetic material, so biological importance of DNA was not realized until much later.

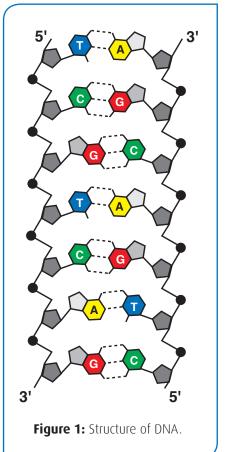
In 1928, Frederick Griffith observed that living cultures of a normally non-pathogenic strain of *S. pneumonia* were able to kill mice, but only after being mixed with a heat-killed pathogenic strain. Because the non-pathogenic strain had been "transformed" into a pathogenic strain, he named this transfer of virulence "transformation". In 1944, Oswald Avery purified DNA, RNA and protein from the virulent strain of *S. pneumonia* to determine which was responsible for transformation. Only those recipient cells exposed to DNA became pathogenic, leading to the

recognition of DNA as the genetic material. These experiments kicked off a worldwide race to unlock the secrets coded for in our DNA.

A strand of DNA is composed of building blocks known as nucleotides. Each nucleotide comprises three basic parts: a phosphate group, a deoxyribose sugar, and a nitrogen-containing base (adenine, cytosine, guanine, or thymine — abbreviated as A, C, G, or T). The sugar of one nucleotide forms a covalent bond with the phosphate group of its neighbor, making DNA a stable scaffold for genetic information. The order of these nucleotides gives rise to genes, each with a unique sequence.

In 1953, James Watson and Francis Crick determined that DNA forms a double helix structure, similar to a ladder that spirals around a central axis (Figure 1). The rungs of the ladder are formed by hydrogen bonds between bases (shown as dashed lines in Figure 1). Base-pairing interactions are highly specific – A will only pair with T, and G with C – but are weaker than covalent bonds, which allows paired DNA strands to be "unzipped" during DNA replication. For their work characterizing the molecular nature of DNA, Watson and Crick were awarded the Nobel Prize in Physiology or Medicine in 1962.

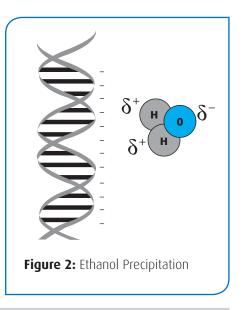
Before we can perform many molecular biology and biotechnology experiments, we must release DNA from cells by breaking down cellular and nuclear membranes. Then, ethanol precipitation is used to purify and concentrate the DNA. Since the negative charge of the sugar-phosphate backbone makes DNA a polar molecule, water molecules interact electrostatically with molecule. This allows DNA to be highly soluble in aqueous solution.





When we add alcohol to our aqueous DNA solution, it disrupts the electrostatic interactions between the water molecules and the sugar-phosphate backbone, forcing the DNA out of solution as sticky white fibers. The addition of salts like sodium acetate enhances DNA precipitation because the positively charged sodium ions further disrupt the interactions between the DNA molecules and water. The DNA precipitates out of the cellular lysate as sticky white fibers.

For this DNA isolation, we will carefully overlay our cellular lysate with alcohol, which, due to its lower density, "floats" on top of the lysate. The alcohol causes the DNA to aggregate at the interface of the two liquids as a viscous precipitate that can be collected from the interface using a stirrer or glass rod. The amount of DNA extracted from a sample will vary depending upon the amount and integrity of the starting material.



Pre-Lab Assessment

Before performing the exercise, be sure that students are comfortable with the following topics:

- 1. Function of DNA in an organism
 - a. DNA contains genetic information as genes
 - b. Each gene contains the instructions to build a protein
 - c. The information contained within the DNA is responsible for carrying out all of the processes in an organism
- 2. Structure of DNA
 - a. DNA is built of four different nucleotides
 - b. The nucleotides are linked by phosphodiester bonds between the sugar of one nucleotide and the phosphate of the second nucleotide
 - i. The backbone has a strong negative charge, making it a polar molecule
 - ii. The backbone interacts electrostatically with water, making DNA soluble in water
 - c. DNA bases hydrogen bond to one another in an antiparallel orientation, holding the strands together
 - i. Base-pairing interactions are highly specific A will only pair with T, and G with C.
- 3. Purification of DNA
 - a. DNA is released from the cell by breaking down cellular and nuclear membranes in a process called lysis.
 - b. DNA is precipitated from cellular lysate as sticky white fibers.
 - i. Ice cold alcohol (ethanol or isopropanol) disrupts the interactions between the water and the DNA, making the DNA insoluble

ii. The addition of salts like sodium acetate enhances precipitation because the positively charged sodium ions further disrupt the interactions between DNA and water.

- c. Precipitated DNA is removed from solution
 - i. DNA is spooled using a glass rod.
 - ii. Centrifugation moves the DNA to the bottom of the tube, creating a pellet.



Activity #1: Creating a 3D Model of DNA

Students should work in groups of 2-4.

REQUIRED MATERIALS:

- Toothpicks (wood or plastic)
- Bag of multicolored soft candy (Gumdrops, gummy bears, or marshmallows work well. Must have four colors)
- Bag of licorice sticks
- Digital Camera or Cell Phone (optional)

PROCEDURE:

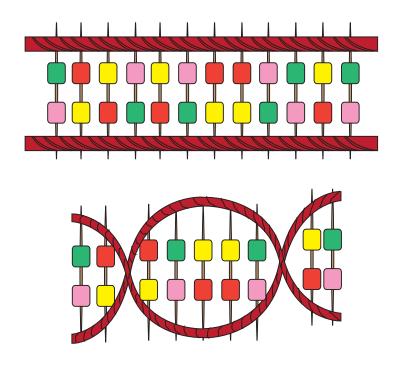
- 1. If students intend to eat the candy after the experiment, be sure they thoroughly clean their hands before creating the model. Plastic wrap or aluminum foil can be placed over desktops to create a clean surface.
- 2. Each group will receive the following items in a plastic bag:
 - a. Four colors of gummy candy represent the nucleotide bases. Each group should receive at least six candies of each color. Assign a nucleotide to each of the four colors of gummy candy.
 - i. Adenine =_____
 - ii. Thymine =_____
 - iii. Cytosine =_____
 - iv. Guanine =_____
 - b. Licorice sticks represent the sugar-phosphate backbone. Each group should receive two.
 - c. Toothpicks represent the base pairing interactions between nucleotides. Each group should receive at least toothpicks.
- 3. Using a toothpick, skewer two of the gummy candies. Be sure to follow base pairing rules (A=T, G=C). Repeat until all of the gummy candy is used.
- 4. Attach one piece of licorice to each side of the toothpick. The base pair toothpicks should be added to the licorice in random order. The resulting DNA duplex should look like a ladder, with the licorice as the rails and the nucleotides as the rungs.
- 5. Pick up the ladder at each end. Carefully twist the DNA model so that it forms a double helix.

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ASSESSMENT:

The instructor should evaluate each DNA model before the students consume the candy. Alternatively, students photograph their final products for submission. The photo is printed, the component parts of DNA are labeled, and the photo is entered into the student's lab notebook.



Activity #1: Discussion Questions

1. Record the base pair sequence of your DNA molecule. Why is the order of the nucleotides important?

Sequences will vary with each student group. The order of these nucleotides gives rise to genes, each with a unique sequence.



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Activity #2: Extracting DNA

Students should work in groups of 2-4.

REQUIRED MATERIALS:

- EDVOTEK[®] Kit 119 Genes in a Tube[™]
- Ice cold ethanol or isopropyl alcohol
- Water bath
- Test tube racks
- Ice and ice buckets
- Personal protective equipment (lab gloves and goggles)
- Centrifuge

PROCEDURE:

In this experiment, students will extract DNA from their cheek cells. The DNA is visualized and stored in a Genes in a Tube[™] necklace.

Before performing the experiment, be sure to emphasize good laboratory practices (proper personal protective equipment, exercise caution when using equipment, hand washing, etc.) Perform experiment as outlined in the lab packet (Available online at http://www.edvotek.com/site/pdf/119.pdf). This experiment should take about thirty minutes, so plan accordingly.

Before starting the experiment, students should:

- Carefully read the introduction and the protocol. Use this information to form a hypothesis for this experiment.
- Predict the results of the experiment.

During the experiment, students should:

Record any observations in a lab notebook.

After the experiment, students should ask the following questions:

- Interpret the results does the data support or contradict the hypothesis?
- If the experiment was repeated, what should change? Revise the hypothesis to reflect this change.

Be sure to have your students reflect upon these points in their lab notebooks. Students should also submit a formal lab report upon completion of the exercise.



Activity #2: Discussion Questions

1. How do alcohol and salt precipitate DNA?

Ice-cold alcohol (ethanol or isopropanol) disrupts the interactions between the water and the DNA, making the DNA insoluble. The addition of salts like sodium acetate enhances precipitation because the positively charged sodium ions further disrupt the interactions between DNA and water.

2. What did the DNA look like?

The spooled DNA looked like long, white, sticky fibers. The fibers can clump up, forming thick strands.

3. What were the main parts of the DNA extraction? What did each part do?

- a) Collect cheek cells this is the material from which we will extract DNA
- Add lysis buffer and protease to cheek cells and incubate this breaks down the cellular and nuclear membranes, releasing the DNA into solution.
- c) Add salt and alcohol this solution disrupts electrostatic interactions between water and the DNA molecules, forcing the DNA out of solution as long stringy fibers.
- d) Remove the precipitated DNA the DNA is now ready to be further manipulated!

4. What other samples contain DNA that we could isolate?

Student answers may vary. Most prokaryotic and eukaryotic cells contain DNA. Notable exceptions include red blood cells and mature hair cells. These cells destroy their nucleus before fully maturing.





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Related Resources



An Introduction to Biotechnology

Biotechnology represents the use of cellular, molecular, and biochemical technologies to improve society and the environment. Basic biotechnology techniques have been used for centuries for the production and preservation of food, selective breeding of livestock, and to improve human health. Modern laboratories that discover many of these biotechnological innovations use simple but powerful techniques to visualize and manipulate DNA and proteins.

Edvotek® offers multiple biotech products to outfit your classroom. From gel electrophoresis and PCR, to bacterial transformation and exciting forensics experiments, you can now bring a wide variety of inquiry-based activities into your classroom!

Electrophoresis

Electrophoresis is a technique that allows scientists to separate DNA, RNA, or proteins according to their size. The molecules are pulled by an electrical field through a defined gel, forcing them to move through microscopic pores. This process separates the molecules based on size, with smaller fragments moving more easily through the gel. Because of this, electrophoresis experiments can be used to detect the presence and size of DNA from a variety of sources. We offer a broad range of exciting electrophoresis experiments for the classroom, including both dye and DNA-based gel electrophoresis.

Cat. 5062

Classroom DNA Electrophoresis LabStation[™] Supports up to 24 students

Includes:

- 1 Cat. #515 M36 HexaGel™ Electrophoresis Apparatus
- 1 Cat. #509
- DuoSource™ 150
- (75/150 V for 1 or 2 units) 2 Cat. #588
- Fixed Volume MiniPipet (40 µl)
- 1 Cat. #636 Yellow Micropipet Tips
- (1 200 μl / 2 Racks of 96) 1 Cat. #130
- DNA Fingerprinting Classroom Experiment





cat. 101 Principles & Practice of Agarose Gel Electrophoresis

In this safe, colorful and fun experiment, students learn the basics of agarose gel electrophoresis. Groups of students will cast gels, load samples, and observe the separation of visible bands. This experiment arrives Ready-to-Load and does not require DNA or toxic chemicals.



Cat. 112 Restriction Enzyme Analysis of DNA

Restriction enzyme analysis of DNA Introduce your students to the concept of DNA digestion by restriction enzymes, followed by electrophoresis to examine the results. This Ready-to-load experiment examines digestion of lambda DNA at specific nucleotide sequences. You can also Save money with one of our convenient Classroom DNA Electrophoresis LabStations, designed to provide everything you need to perform many of our electrophoresis experiments!



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Staining Agarose Gels

InstaStain® Ethidium Bromide



SYBR® Safe DNA Stain

InstaStain® Blue and FlashBlue™



After electrophoresis, DNA fragments will have separated according to their size. However, DNA bands are colorless, and it necessary to first stain the DNA before it can be seen. EDVOTEK® offers several methods for visualizing DNA within agarose gels.

Fluorescent DNA Stains:

The most frequently used method for visualizing DNA is through the use of fluorescent DNA stains. These stains are extremely sensitive and rapid, making them perfect for use in the classroom. Fluorescent stains must visualized using an ultraviolet (UV) light source. We offer two options for fluorescent stains: InstaStain® Ethidium Bromide and SYBR® Safe DNA Stain.

Visible Dye-based DNA Stains:

Dye-based DNA stains are an excellent alternative to fluorescent stains, requiring no special equipment or waste-disposal. Although the are less sensitive to DNA concentrations, our visible stains provide excellent results and dye DNA with and intense blue color. EDVOTEK® offers two visible dye-based DNA stains: InstaStain® Blue and FlashBlue™ stain.

Stain	Advantages	Disadvantages
InstaStain® EtBr	Very sensitive Very fast	Requires UV transilluminator Potentially mutagenic
SYBR® Safe	Very sensitive Non-mutagenic	Requires UV transilluminator More expensive
InstaStain® Blue	Easy to use Generates minimal waste	Less sensitive More time
FlashBlue™	Simple and fast Reusable, inexpensive	Less sensitive Disposal of liquid



^{cat. 558} Midrange UV Transilluminator

The EDVOTEK® Midrange UV Transilluminator is designed to visualize DNA stained with Ethidium Bromide or SYBR® Safe. The UV filter is 7 x 14 cm and is optimal for visualizing all of our gel sizes. Safety features include a UV-blocking cover and a power cut-off switch when the cover is opened.

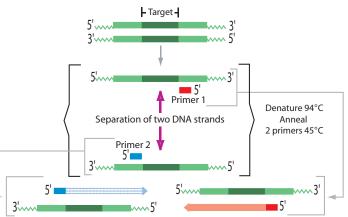
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Polymerase Chain Reaction

Polymerase Chain Reaction (PCR) is a technique that allows researchers to rapidly create many copies of a desired stretch of DNA. PCR is currently used in disease screening, forensic testing, and biological research, and represents a valuable platform for students to explore STEM concepts. Edvotek® has developed a wide variety of PCR experiments and equipment, including the EdvoCycler[™] and MegaCycler[™] PCR machines.





cat. 541 & 542 EdvoCycler™ and MegaCycler™

The Edvocycler[™] and MegaCycler[™] are affordable, stand alone PCR machines. Each machine arrives ready-to-use, and comes pre-programmed with all EDVOTEK PCR protocols, with space for you to make your own!

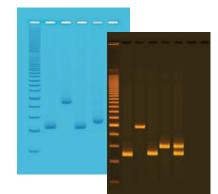


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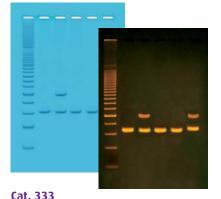
Cat. S-48 What is PCR?

Students will be introduced to PCR with this easy to use kit. This experiment introduces the fundamental principles of PCR without the need for a thermocycler or staining!



cat. 953 Multiplex PCR-based Testing of Water Contaminants

PCR is commonly used to test drinking water for contamination. In this classroomsafe experiment, students will test for the presence of three organisms in a water sample using PCR.



Alu-Human DNA Typing

Students use with their own DNA to look for a 300 base pair Alu insertion in chromosome 16, allowing them to determine their genotype!



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