DNA Isolation

Using Human Cheek Cells

Introduction

Learn how to isolate DNA from human cells. Yours!

Concepts

- Genetics
- DNA isolation

Materials

Ethyl alcohol, 95% denatured, CH_3CH_2OH, 6 mL
Sodium chloride solution, NaCl, 8.0%, 20 drops
Sodium lauryl sulfate solution, $C_{12}H_{25}NaO_4S$, 10%, 20 drops
Dropping bottles, 3
Glass stirring rod
Plastic drinking cup, 30-mL
Stopper, #2
Test tube, 12 × 75 mm
Test tube, 16 × 100 mm
Test tube rack
Water, tap, 10 mL

Safety Precautions

Ethyl alcohol is flammable and a dangerous fire risk; keep from flame and all sources of ignition. Use only clean drinking cups for this procedure. Wear chemical splash goggles, chemical-resistant gloves and a chemical-resistant apron. Please review currentSafety Data Sheets for additional safety, handling, and disposal information. Wash hands thoroughly with soap and water before leaving the laboratory.

Preparation

1. Prepare an 8.0% w/w sodium chloride solution: Dissolve 8.0 g of sodium chloride in 92 mL of distilled water. Place solution in dropper bottle and label.
2. Prepare a 10% sodium lauryl sulfate solution: Dissolve 10.0 g of sodium lauryl sulfate in 90 mL of distilled water. Place the solution in a dropper bottle and label it. If dishwashing detergent is used, make a 25% solution by mixing 25 mL of liquid detergent concentrate with 75 mL of distilled water. Place the solution in a dropper bottle and label it.
3. Place the ethyl alcohol solution in a dropper bottle and label it.

Procedure

1. Add 1 mL (20 drops) of the 8% sodium chloride solution to the larger test tube. Set the tube aside in a test tube rack.
2. Pour 10 mL of fresh tap water or bottled water into a clean 30-mL plastic drinking cup.
3. Put the 10 mL of water in your mouth and swirl the water around for at least 30 seconds. Spit the water back into the plastic cup. (The swirling of the water washes cells from the inside of your cheeks into the water.)
4. Pour several mL of the "cheek cell" water into the test tube containing the salt solution from Step 1.

Releasing the DNA from inside the cheek cells.

5. Add 1 mL (20 drops) of the 10% sodium lauryl sulfate solution *or* 1 mL (20 drops) of the 25% liquid dishwashing detergent solution to the "cheek" mixture in the test tube.
6. Stopper the test tube and mix the contents of the tube by gently inverting the test tube several times. *Do not shake the test tube.* (The detergent removes the cell membranes from the cheek cells, releasing the DNA into the salt solution.)

Publication No. 10196
061616

Precipitate the DNA.

7. Holding the test tube at a slight angle, carefully add 5 mL of 95% ethyl alcohol down the side of the test tube so that it forms a layer over the "cheek" mixture in the test tube.
8. Hold the test tube upright for one minute and observe what happens at the interface between the ethyl alcohol and the "cheek" solution. (The clouds of white strands are the DNA. The DNA is not soluble in ethyl alcohol, so it precipitates where the two liquids meet. Soap bubbles from the "cheek" solution will get trapped in the DNA strands.)

Collect the DNA.

9. Add 1 mL (20 drops) of 95% ethyl alcohol to the smaller test tube.
10. Place a clean glass stirring rod in the test tube containing the DNA. Collect the DNA by winding it on the rod by turning the rod in one direction.
11. Carefully, remove the rod and DNA from the solution and transfer it to the smaller test tube containing 1 mL of 95% ethyl alcohol. Observe the DNA strands floating in the alcohol.

Disposal

Please consult your current *Flinn Scientific Catalog/Reference Manual* for general guidelines and specific procedures, and review all federal, state and local regulations that may apply, before proceeding. The resulting mixtures can be flushed down the drain according to Flinn Suggested Disposal Method #26b.

Tips

- If your tap water has any unusual properties, use bottled water for this lab.
- If you do not have sodium laurel sulfate solution, you may substitute liquid dishwashing detergent. Make a 25% solution with 25 mL of detergent and 75 mL of distilled or deionized water.
- If the DNA yield is not sufficient for spooling, try the following:

1—Rinse your mouth more violently and for a longer period of time.

2—The action of the detergent in Step 5 can be enhanced by placing the test tube in a water bath at 55 °C. This enhances the action of the detergent and also denatures enzymes that might damage DNA.

3—The alcohol used in Step 7 might be more effective if it is made ice-cold in an ice bath.

Discussion

The steps in this laboratory procedure teach a great deal about the properties of cells, cell membranes, and deoxyribonucleic acid (DNA) itself.

The collection of cheek cells from the inside of the mouth highlights the nature of body tissue. Dead cells are continually being sloughed off on both the inside and outside of the body. Recently-sloughed cells still contain their nuclei and their DNA genetic material. This DNA can be collected and if in a forensics situation, analyzed and traced to a specific individual.

Detergents solubilize and break down the lipids and proteins that form the primary cell membrane and disrupt the bonds that hold the membrane together. The cell contents, including the nucleus, are thus released and become available for further treatment or isolation. Sodium lauryl sulfate is an active ingredient in detergents.

The final step requires the alcohol. The solubilized DNA comes in contact with the alcohol where the two liquid layers meet. The alcohol dehydrates and precipitates the DNA, as DNA is insoluble in the alcohol. If the procedure is done properly, fine, long strands of DNA will form at the interface and can be easily spooled onto the glass stir rod.

Connecting to the National Standards

This laboratory activity relates to the following National Science Education Standards (1996):

Unifying Concepts and Processes: Grades K–12
Systems, order, and organization
Evidence, models, and explanation
Content Standards: Grades 5–8
Content Standard C: Life Science, structure and function in living systems
Content Standard F: Science in Personal and Social Perspectives, personal health
Content Standards: Grades 9–12
Content Standard C: Life Science, the cell, molecular basis of heredity
Content Standard F: Science in Personal and Social Perspectives, personal and community health

Acknowledgment

Special thanks to David A. Katz, Cladwyne, PA for providing Flinn with this activity.

Materials for *DNA Isolation* are available from Flinn Scientific, Inc.

Catalog No.	Description
D0024	Dodecyl Sulfate, Sodium Salt 100 g
E0007	Ethyl Alcohol, 500 mL
GP5075	Glass Stirring Rod
S0063	Sodium Chloride, NaCl, 500 g

Consult your *Flinn Scientific Catalog/Reference Manual* for current prices.