

My Favorite DNA Extraction



Purpose

The purpose of this activity is become familiar with lab procedures for extracting DNA and observe the physical characteristics of DNA.

Background

DNA is stored in the nuclei of your cells and a great source is your saliva. So, you will collect a sample of your epithelial cells from your saliva, break the cells open, and remove the DNA from the cells you have collected. The epithelial cells that line the inside of your mouth divide frequently, coming off easily as new cells continuously replace the old ones. In fact, these cells are coming off and being replaced every time you chew and eat food.

The process of breaking open the cells is called **lysis** and you will use a lysis buffer to accomplish this task. A lysis buffer is a solution containing salt and dishwashing liquid. The dishwashing liquid will dissolve the membrane of the cell, just like dishwashing detergent dissolves fats and proteins from a greasy pan, because cell and nuclear membranes are composed of fats and proteins. Dissolving the membranes results in the release of the DNA.

DNA is packed tightly around proteins. Like spools for thread, these proteins keep the DNA tightly wound and organized so that it doesn't get tangled inside the nucleus. In order to collect the DNA, it helps to remove the proteins so that the DNA can first loosen and expand, then collect into a mass with the DNA from all the other cells.

You will incubate your cheek cell collection with **protease**, which breaks down proteins so that they can no longer bind the DNA. Protease is an **enzyme**, or protein machine, that works best at 50°C, which is the temperature of slightly hot water. The protease chews up the proteins associated with the DNA and also helps digest any remaining cell or nuclear membrane proteins.

Strands of DNA are so thin that it is not possible to see them when they are dissolved in solution. Think of the long, thin strands of DNA as fine white threads. If one long piece of thread were stretched across the room, it would be difficult to see. But if you collect several threads and put them together they would become more visible. Well in this laboratory exercise, you will use cold ethanol to bring several strands of DNA out of solution. DNA is insoluble in ethanol and will precipitate out of solution and become a visible solid mass.

Much like sugar, DNA is colorless when it is dissolved in liquid, but is white when it precipitates in enough quantity to see. As it precipitates, it appears as very fine white strands suspended in liquid. The strands are somewhat fragile — like very thin noodles, they can break if handled roughly. Also, if a mass of precipitated DNA is pulled out of its surrounding liquid, it will clump together, much like cooked noodles will clump together when they are pulled out of their liquid.

Once your DNA is collected you can store it in alcohol for quite some time as long as you don't add water or shake the container.

Materials

- 1 - 6oz. paper cup
- 1 - bottle of water
- 10 ml lysis buffer
- warm water bath
- meat tenderizer
- Ice-cold ethanol (90% or higher concentration)
- test tubes
- small spoon (sample type)
- coffee stirrer
- Safety goggles

Safety Precautions

Dispose of all used paper cups and stirrers properly. Make sure you wash your hands after the lab.

Pre-lab

Describe the following terms.

lysis – _____

enzyme – _____

protease – _____

precipitate – _____

Name: _____ Date: _____

Procedure

1. Obtain one 6 oz. paper cup and a bottle of water. Fill the cup about $\frac{1}{4}$ with water and put the water in your mouth. Swish the water in your mouth for about 1 minute and then spit the water back into the cup.

DO NOT SWALLOW THE WATER.

2. Mix 10 ml of lysis buffer with your cell solution. Mix well with coffee stir.

3. Fill your collection tube about $\frac{1}{2}$ full with your cell solution.

4. Add 1 small spoon of protease, then cap and shake well. Now place the tube in a warm water bath for about 10 minutes.

5. After 10 minutes remove tube from water bath and put it in a test tube rack and let it settle for a couple of minutes.

6. Next **slowly** drizzle **ice-cold** ethanol **along the side of the test tube** until DNA begins to appear.

7. Keep the test tube at eye level so you can see what is happening! Pay attention to the characteristics of the DNA as it precipitates.

Note: You may have either the strawberry DNA or your DNA. Please pick it up after school.

Analysis

1. What did the DNA look like?

2. What was different about this lab from the strawberry DNA lab?

“To be trusted is a greater compliment than being loved.”
-- George MacDonald