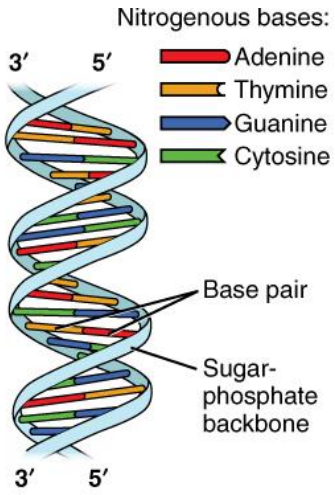
DNA EXTRACTION

Introduction

All living things contain cells, be it plants, animals or bacteria. The three main parts of the cell are the nucleus, the cell/plasma membrane and the cytoplasm. **Deoxyribonucleic acid** (DNA), is the control center of the cell, as it is the carrier of genetic information.

The double helix structure of the DNA consists of a sugar and the four **nucleotide bases**: adenine, thymine, guanine and cytosine. These bases pair-up via a covalent bond and form the "rungs" of the DNA ladder.



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Unbroken, a chain of DNA can contain millions of atoms, it is the largest known molecule. The extraction of deoxyribonucleic acid is the process where DNA is separated from proteins, membranes and other cellular materials

Grinding or crushing the cell will begin to break down the cell wall and nuclear membrane. During this step, negative ions will be released. Adding a solution containing salt will allow for an attraction between the positive salt ions and the negative ions from the sugar backbone of the double-helix. This way the fragments will stay separated and ready for extraction.

In order to obtain enough DNA to spool, a detergent must be added in order to fully **denature** (break down) the nucleus and cell membrane. As the enzymes break down the nucleus, DNA is released into the solution.

Seeing as DNA is **soluble** in water but not alcohol, there is the addition of ice-cold ethanol or isopropanol to the sample. This step allows for a **precipitate** to become visible and **spooled**. This precipitate, caught between the alcohol and solution is the DNA to be extracted.

Being able to extract DNA is a step of crucial importance in biotechnology. It allows for the study of genetics, from the causes of diseases to the development of diagnostics and pharmaceuticals. It is also used in forensic sciences, genome sequencing and paternity testing.

Purpose

The purpose of the experiment is for students to extract and visualize DNA using everyday materials.

Hypothesis

Write a hypothesis about the two samples you will be working with:

_____
_____
_____

Materials

- 1 or 2 fresh strawberries (if you use frozen ones, make sure they are room temperature for the experiment)
- 2 x 50mL beaker
- 150 mL beaker
- 250 mL beaker
- 250 mL graduated cylinder
- 2 x 10 mL graduated cylinder
- Alcohol (ice cold)
- Distilled Water
- Enzymes (meat tenderizer)
- Liquid detergent
- Filter Paper
- Funnel
- 2 x Glass stirring rod
- 2 x Test tube
- 2 x Pipette
- Re-sealable plastic bag
- Teaspoon
- Test tube + rack
- NaCl or non-iodized salt

Notes on Materials:
Other DNA sources can be used, you will find the best results from strawberries and split peas but raw broccoli, spinach and bananas will give results as well. Harder vegetables will need to go through a blender instead of simply crushing in the bag.

Either 95% ethanol or 70-95% isopropyl alcohol can be used to extract the DNA, provided it is cold.

Liquid detergent can range from dish soap to shampoo that does not have conditioner added.

If meat tenderizer (enzymes) can't be found, it can be replaced with 2-3 drops of pineapple juice provided the juice is pure and not a mixed drink.

Procedure

Part 1: Buffer Solution Preparation

1. Label 250 mL beaker Buffer Solution.
2. Add 220 mL of distilled water to beaker.
3. Dissolve 1 teaspoon of NaCl or non-iodized table salt into beaker.
4. Measure 25 mL of liquid detergent and slowly mix detergent into beaker with glass rod. Avoid as much foaming as possible.

Part 2: Control Sample Preparation

1. Label test tube Control Sample.
2. Pipette 3 mL of distilled water into test tube.
3. Measure 5 mL of buffer solution and slowly add to test tube.

Part 3: Experimental Sample Preparation

1. Remove stems from the strawberries.
2. Place strawberries into the re-sealable plastic bag, remove all air and seal well.
3. Crush the strawberries carefully for 5 minutes.
4. Measure 10 mL of buffer solution and add to bag.
5. Remove air and seal well.
6. Mash mixture for 1 minute.
7. Label 150 mL beaker with Experimental Sample.
8. Filter strawberry extract through a funnel lined with filter paper in 150 mL beaker.
9. Let filtrate rest.

Part 4: DNA Extraction

1. Label test tube with Experimental Sample.

2. Pipette 3 mL of filtrate into test tube.

3. Add a pinch of enzymes. Stir gently.

4. Measure 5 mL of alcohol in 10 mL graduated cylinder.

5. Hold test tube at a slight angle.

6. Carefully and slowly pipette alcohol down the side of the test tube to form a layer on top of the filtrate. DO NOT SHAKE THE TEST TUBE AT ANY POINT.

7. Watch the solution for precipitate.

8. Use glass rod to spool DNA.

9. Record Observations.

10. Repeat steps 3-7 with the control sample.

11. Record Observations.

Results

1. Did the sample contain DNA, circle your answer:

Control Sample	YES	NO
Experimental Sample	YES	NO

2. Was your hypothesis supported? Explain.

_____
_____

3. Describe your observations on the physical structure of DNA:

_____
_____

4. What was the purpose of filtering the experimental sample?

_____
_____