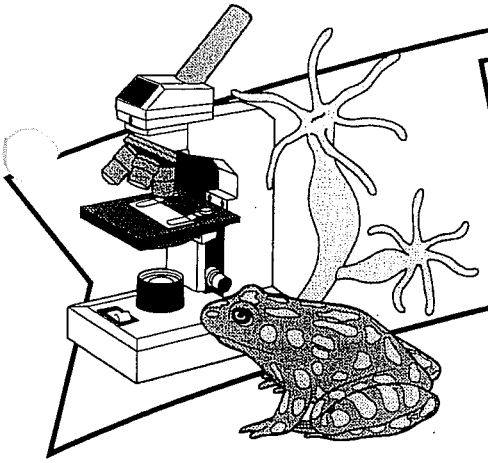

BIO-FAX!

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Wheat Germ DNA Extraction

Publication No. 10113

Introduction

This is a DNA extraction and isolation activity using common household chemicals. With dish soap, meat tenderizer, baking soda, and alcohol, students will isolate DNA from raw wheat germ.

Materials (for each lab group)

Raw (untoasted) wheat germ, 2 g

Liquid detergent (Palmolive®, Dawn®, Woolite®), 3 mL

Meat tenderizer (Adolph's® unseasoned original), 2 g

Alcohol, 95% (ethyl or isopropyl), 20 mL

Water bath at 55 °C

Sodium bicarbonate solution, $NaHCO_3$, 1 M, 5 mL

Paper clip (giant-sized)

Tap water

Thermometer

Beaker or clear plastic cup (200-mL/8 oz.)

Graduated cylinder, 50-mL

Ice bath

Serological pipet, 10-mL

Safety Precautions

Ethyl and isopropyl alcohol, 95%, are flammable and dangerous fire risks; keep away from flame and sources of ignition. Both alcohols are also toxic by ingestion. Chemical splash goggles are advised whenever chemicals, heat or glassware are used. Wash hands thoroughly with soap and water before leaving the lab.

Pre-Lab

The alcohol should be ice cold (approx. 0 °C) when used. Place it in an ice bath before class, or in a freezer overnight if possible. Prepare a 1 M sodium bicarbonate solution by dissolving 8.4 g of $NaHCO_3$ in 100 mL of tap, or distilled water. It is essential that the wheat germ be raw—toasted wheat germ will not work. Raw wheat germ can be found in health food stores and in some grocery stores.

Straighten the paper clip and form a small hook at one end. Roughen the hook portion with a file or steel wool—a roughened surface enhances adhesion of the DNA strands, and facilitates spooling.

Procedure

1. Measure 45 mL of tap water into a beaker and place it in a warm water bath. Allow it a few minutes to warm. The optimal temperature for the procedure is 55 °C—do not allow the temperature to exceed 60 °C.

2. Sprinkle the wheat germ into the beaker and gently stir in 3 mL of detergent. Allow this mixture to incubate in the 55 °C water bath for 5 minutes.
3. After 5 minutes, gently stir in 2 g of the meat tenderizer and 5 mL of the 1 M sodium bicarbonate solution. Incubate this mixture at 55 °C for an additional 15 to 20 minutes.
4. Transfer the beaker containing the wheat germ mixture to an ice bath for a few minutes to quickly cool it to room temperature. Stir gently during this period.
5. Using a serological pipet, carefully layer 20 mL of ice-cold alcohol over the wheat germ solution in the beaker. Allow the alcohol to flow from the pipet with the pipet tip held against the inside surface of the beaker just above the liquid level.
6. There will be a visible interface between the alcohol layer and the wheat germ mixture layer. A fibrous white precipitate should be evident at the interface. This is DNA. Immerse the paper clip hook into the wheat germ mixture below the interface. Use a slow, twirling motion to bring the DNA up into the alcohol where the strands will become visible and attached to the hook. DNA strands easily break apart so this step must be done carefully and without stirring!

Discussion

Wheat germ is the embryo (sprouting) section of the wheat kernel; the remainder being the endosperm (storage). The germ is extremely rich in vitamins and nutrients, and for the purposes of this experiment, an excellent source of DNA. The steps in this procedure can teach us a great deal about the properties of cells, cell membranes, and of deoxyribonucleic acid (DNA) itself. What was the purpose for each part of the procedure?

Heat is applied first to assist in softening the cell membranes and to denature enzymes that might otherwise damage the DNA. The temperature is kept below 60 °C—because higher temperatures denature the DNA and make spooling impossible. Detergents solubilize the lipids and proteins that form the cell membranes. This disrupts the bonds that hold the membranes together and causes them to break down. The contents of the cells, including the nuclei, are released into the mixture.

The sodium bicarbonate solution is added to maintain a near-neutral pH—at which the DNA is most stable and at which the enzyme present in the meat tenderizer is most effective. The meat tenderizer contains the proteolytic (protein breaking) enzyme papain—naturally present in papaya, pineapple, and other fruits. The papain completes the breakdown of the nuclear membrane, which puts the free DNA in solution after a 15–20 minute incubation period. [This time period is critical because even 55 °C will eventually break down DNA.] The mixture is quickly cooled to stop further reactions.

The final step requires the cold alcohol. The solubilized DNA contacts the alcohol where the two liquid layers meet. The alcohol dehydrates and precipitates the DNA, as DNA is insoluble in the alcohol (especially *cold* alcohol). If the procedure is carried out properly, fine, long strands of DNA will form at the interface—and can be readily spooled onto the paper clip.

Disposal

The resulting mixtures can be rinsed down the drain according to Flinn Suggested Disposal Method #26b. Please consult your current *Flinn Chemical Catalog/Reference Manual.*

Acknowledgment

Special thanks to Lu Ann Williams, El Reno High School, El Reno, Oklahoma, for providing us with the basis for this activity.