Chapter 14 Project

DNA Fields Forever

Project Goal + Timeline

In keeping with the old adage "Seeing is believing," this project will show you how to extract a crude preparation of DNA from an organism. This laboratory activity requires a few household items and will take approximately two hours to complete. There are also several recommended variations on this experiment that you can try (each roughly taking the same time as the first). You can try these variations in parallel with the first by scaling up the ingredients and splitting the material.

Directions

Part 1: Extract DNA!

While this procedure works to extract DNA from many types of organisms, the recommended organisms are strawberries. Strawberries have seven unique chromosomes, but instead of two copies of each chromosome, they have eight copies of each. (They are octoploid!) Make sure you have all the materials and equipment prepared before beginning your DNA extraction. Some substitutions can be made; you don't need a particular brand of dish detergent, for example, and you can use glass cups instead of plastic, if needed.

At the end of the procedure, there are several optional variations to try. You should at least consider what might happen if you tried these. If you want to attempt these additional experiments, you can start each from scratch, or you can scale up the amount of material you use; for example, if you use eight strawberries and a proportionally greater amount of extraction solution, you can then split up your filtrates into separate cups for the different treatments.

First, you should weigh two strawberries and record their mass. Ideally, this mass should be in grams, since the metric system is the standard convention for scientific experiments. (If you weigh in ounces or pounds, you should convert this measurement.) Record this data in Table 1. If you are performing one of the variations of the experiment, use two strawberries per experiment and make sure to record the mass of your additional strawberries.

TABLE 1: Mass of Strawberries

Experiment	Mass of Strawberries (g)
DNA extraction	
Optional variation: Digestive enzymes	
Optional variation: Acid or base	

To isolate the DNA, we need to release it from the cells in the strawberry. In the native tissue, the cells are protected by cellulose walls, and the DNA is surrounded by both a plasmid membrane and a double-layered nuclear envelope (both consisting of hydrophobic lipids).

Place the strawberries in a plastic bag, seal it, and then mash the strawberries. You should make sure that the strawberries are completely crushed. Crushing breaks up the tissues and cells, mechanically disrupting the cell walls and helping the cells break open to release their DNA.

While you allow the crushed strawberries to sit, prepare an extraction solution. In a plastic cup, combine ½ cup water, 1 teaspoon salt, and 2 teaspoons dishwashing soap, and mix thoroughly. Then, add about 2 tablespoons of this solution to the bag of strawberries. Reseal the bag and gently crush it again. Take care not to introduce too many soap bubbles.

1. What was the purpose of adding the dishwashing soap to the strawberry mixture? How can soap help isolate or extract the DNA?
2. Why did you need to avoid excessive soap bubbles or mechanical disruption when adding the detergent?

Place the coffee filter over the other plastic cup. Gently pour the solubilized strawberry extract through the filter. The resulting solution or fraction is known as the filtrate.

3. What did you remove during the filtration step? What types of molecules do you have left in the filtrate?

Remove the coffee filter from the top of the plastic cup. Tilt the cup at an angle, and slowly pour approximately ½ cup of cold rubbing alcohol down the side of the cup so that it forms a layer on top of the filtrate. Place the cup back down and do not stir the solution. Watch for the development of a cloudy white substance in the top layer of this cup; that's the DNA precipitating!

4. Rubbing alcohol (isopropanol) is a largely nonpolar molecule. Why do you think the addition of rubbing alcohol caused the DNA to precipitate? Why might salt have been necessary or important for this step?

Finally, you can remove the DNA from this precipitate by spooling it around a wooden stick, skewer, coffee stirrer, or toothpick. Remove the spooled material and place it on the wax paper, foil, or plastic. Allow it to air dry for a minute and then record the mass of this material in Table 2. Congratulations—you have just extracted a crude preparation of DNA!

TABLE 2: Mass of DNA

Experiment	Mass of Precipitate (mg)
DNA extraction	
Optional variation: Digestive enzymes	
Optional variation: Acid or base	

5. What types of molecules would you expect to find in the precipitate? Why?
6. Why was it important to spool the precipitate? What else might be in the precipitate that will not be collected by spooling?

Optional Variation: Adding digestive enzymes to the filtrate

In this variation, you will follow the previous instructions to produce the filtrate. Then, you will add (in powder form) 1 serving of a digestive enzyme to the filtrate. If your enzyme comes in tablet form, you should first crush it up into a powder. Mix gently and let the mix sit at room temperature for 5 to 15 minutes before proceeding to the precipitation step.

Optional Variation: Adding acid or base to the filtrate

In this variation, you will follow the previous instructions to produce the filtrate. Then, you will add 1 teaspoon of a strong household acid or base (but not both!) to the filtrate. Mix gently for 1 to 2 minutes, then proceed to the precipitation step. **Note:** You have now slightly increased the volume of the solution, so you will need to add slightly more rubbing alcohol compared to the instructions.

Part 2: Analyze and Review

1. On average, there are approximately 4.0×10^{-9} grams of DNA per gram of strawberry. Multiply this value by the initial mass of strawberries recorded in Table 1 to find your expected DNA yield (the mass of DNA you could have expected to obtain from your strawberries). What was your expected DNA yield?
2. Compare your expected DNA yield from Question 1 to your actual mass of DNA extracted from Table 2. Propose an explanation for any differences between these values.
3. Now that you've isolated your DNA, imagine you wanted to determine the nucleotide sequence of a specific fragment. What technique could be used? How does this technique work?
4. If you completed the optional extraction with digestive enzymes, what results did you observe? If you didn't complete this extraction, what effect do you predict the digestive enzymes would have had on the precipitate?
5. If you completed the optional extraction with an acid or base, what results did you observe? If not, what would you predict to be different about the precipitate after treatment with acid? What about base? Why does changing the pH affect this procedure?

Project Materials

- 2 tables for recording data
- Pen or a pencil
- Fresh or frozen strawberries (remove any green leaves)
- A resealable plastic bag
- Liquid dishwashing soap
- Salt
- Water
- 2 plastic cups
- 1 coffee filter
- Cold rubbing alcohol (70% isopropanol)
- 1 coffee stirrer, wooden stick, or toothpick
- A balance or means of weighing material (together with a small piece of wax paper, plastic, or aluminum foil)
- *Optional: a meat tenderizer powder, bromelain, or digestive enzyme supplement (In all cases, these contain enzymes that digest proteins.)*
- *Optional: household acid (lemon juice, vinegar) or base (ammonia)*

Student Checklist

- ☐ Record the mass of strawberries (Table 1)
- ☐ Extract and record the mass of DNA (Table 2)
- ☐ Complete the Extract DNA! Questions
- ☐ Complete the Analyze and Review Questions