Biotechnology Workshop B: Strawberry DNA Extraction

LEARNING GOALS: After the completion of this workshop, students will understand:

1. Students will learn what DNA is
2. Students will learn how DNA is structured and how it is contained within cells, particularly plant cells
3. Students will learn how DNA can be removed from a cell

CONCEIVE – What do I wish to accomplish through this project?

This stage involves guiding students in defining the goals of the project, then helping them develop conceptual, technical and action plans to meet those goals while considering the technology, knowledge, and skills that apply. This guidance is provided in the form of Essential Questions that use student’s preconceptions, and misperceptions then move them toward a deeper and more realistic understanding of the process and skills needed to complete the project.

ESSENTIAL QUESTIONS:

1. What is DNA?
2. Where is DNA housed in the cell?
3. How can we break through a plant cell wall/membrane to access the DNA?
4. How can we extract DNA from strawberry cells?

NOTES: Some of this may be redundant from biotechnology workshop A

DESIGN - How will I accomplish the project?

1. An introductory lecture will provide students with basic understanding of DNA. Students should understand the structural differences between plant and animal cells. Some cell biology terms should be introduced, such as double helix, nucleus, membrane, and cell wall.

Copyright University of Colorado Colorado Springs, 2011. For permission to use these materials, contact the Director at the Center for STEM Education, (719) 255-3595. Curriculum format design adapted from CDIO developed by MIT.
2. During the hands-on activity, the instructor should provide further information on the mechanisms of DNA extraction and how each step contributes to the product (isolated plant DNA). Each reagent’s purpose should also be mentioned.

NOTES: See http://www.carolina.com/category/teacher+resources/instructions+and+buying+guides/biotech+kit+instruction+manuals/strawberry+dna+extraction+kit.do for sufficient background information for this experiment.

IMPLEMENT - From an idea to a product!

Procedure: This procedure will extract and collect DNA from a plant cell.

1. Obtain one fresh or one frozen and thawed strawberry. If you are using a fresh strawberry, remove the green sepals (tops) from the berry.
2. Place the strawberry in a resealable plastic bag.
3. Close the bag slowly, pushing all of the air out of the bag as you seal it.
4. Being careful not to break the bag, thoroughly mash the strawberry with your hands for two minutes.
5. Pour the 10-mL aliquot of extraction buffer into the bag with the mashed strawberry. Reseal the bag.
6. Mash the strawberry for one additional minute.
7. Place a funnel into a 50-mL centrifuge tube. Fold the cheesecloth in half along the longer side and place it in the funnel to create a filter. The cheesecloth will overlap the edge of the funnel.
8. Pour the strawberry mixture into the funnel, filtering the contents through the cheesecloth and into the 50-mL centrifuge tube.
9. Carefully pour 2 mL of the filtered contents from the 50-mL tube into a clean 15-mL tube. Use the lines on the side of the 15-mL tube to help measure the amount added.
10. Hold the 15-mL tube at an angle. Using a transfer pipet, carefully add 5 mL of cold 95% ethanol by running it down the inside of the tube. Add the 95% ethanol until the total volume is 7 mL (use the lines on the side of the tube to help you measure). You should have two distinct layers.
   Caution: Do not mix the strawberry extract and the ethanol!
11. Watch closely as translucent strands of DNA begin to clump together where the ethanol layer meets the strawberry extract layer. Tiny bubbles in the ethanol layer will appear where the DNA precipitates.
12. Slowly and carefully rotate the wooden stick in the ethanol directly above the extract layer to wind (or “spool”) the DNA. Remove the wooden stick from the tube and observe the DNA.
NOTES: Distribute information sheets, supplies, and instructions before students arrive. It may be beneficial to also cover pipetting and homogenizing techniques with students before they begin.

OPERATE – Does it work the way I planned?

Note the cautionary warning listed after step 10. Students should not mix the strawberry extract with ethanol.

RESOURCES NEEDED – What equipment and supplies do I need?

Supplies listed are for 32 students working in pairs. Adjustment made be made as needed for different class sizes.

Included in the Carolina kit:
- 17 50-mL tubes with lids and bases
- 33 15-mL tubes with lids
- 16 resealable plastic bags
- 16 wooden sticks
- 16 funnels
- 1 pack of cheesecloth
- 16 transfer pipets
- 1 bottle of ethanol, 95% (100 mL)
- 1 bottle of liquid detergent
- 10 g salt (sodium chloride, NaCl)

Needed, but not supplied:
- scissors for cutting cheesecloth
- container (400-mL capacity or more) for mixing extraction buffer
- 16 transfer vessels (5-mL capacity or more) for 95% ethanol
- 16 ripe, whole strawberries (fresh or frozen)
- 380 mL of water
- graduated cylinder or other device for measuring water
- freezer, refrigerator, or bucket of ice for chilling 95% ethanol
- spool of thread for demonstration

SET-UP
Before the Lesson

1. Chill the 95% ethanol by storing it in a freezer (preferred), refrigerator, or on ice until use.
2. If using frozen strawberries, allow the berries to thaw at room temperature before use. Do not heat the strawberries to hasten thawing, as this will hinder the DNA extraction process.
3. Prepare 400 mL of DNA extraction buffer. Use one of the 50-mL tubes (included) to measure 20 mL of detergent (included).
4. Pour the detergent into a container along with 380 mL of water and the entire contents (10 g) of the salt pack (included).
5. Stir to mix.
6. Aliquot 10 mL of extraction buffer into 16 15-mL tubes (the tubes are calibrated for measuring).
7. Prepare 16 5-mL aliquots of 95% ethanol in transfer vessels. An extra 15-mL tube is included for measuring out the 5-mL amounts.
8. Unroll cheesecloth from the spool and cut 16 pieces approximately 6” long. (The spool is about 8” wide, so each piece should measure about 6 /8”.)
9. Reproduce the *Strawberry DNA Extraction Student Instructions* for each student or pair of students.
10. For each pair of students, distribute the following materials:
   - 10-mL aliquot of DNA extraction buffer in 15-mL tube
   - 5-mL aliquot of 95% ethanol in transfer vessel
   - 1 resealable plastic bag
   - 1 strawberry
   - 1 50-mL tube
   - 1 15-mL tube
   - 1 piece of cheesecloth
   - 1 funnel
   - 1 transfer pipet
   - 1 wooden stick
<table>
<thead>
<tr>
<th>Colorado Academic Standards – High School</th>
<th>21st Century Skills</th>
</tr>
</thead>
<tbody>
<tr>
<td>2. Life Science</td>
<td><strong>Concept and skills students master:</strong></td>
</tr>
<tr>
<td></td>
<td>7. Physical and behavioral characteristics of an organism are influenced to varying degrees by heritable genes, many of which encode instructions for the production of proteins</td>
</tr>
</tbody>
</table>