DNA Spooling
Extraction of DNA from Strawberries

(Submitted by Katy Korsmeyer, Santa Clara County Biotechnology Education Partnership)

Objective: To extract and isolate DNA from strawberries.

Principle: DNA Spooling would be used as an introductory biology hands-on laboratory experiment. The teacher would first explain what DNA is, why it's important and where it is located. With middle school students, it is important to stress that DNA is in plants AND animals. The emphasis is that DNA is a blueprint for life.

Materials:

Ziploc sandwich bags (1 per person)
Strawberries (1 per person) 25 ml graduated cylinder (for measuring) Small funnels (for filtering) Cheesecloth (2 ply thickness) Ice cubes Test tube racks Test tubes (glass, 13 x100 or longer, 1 per person) Table Salt Scissors Liquid detergent (Dawn or Ivory) Isopropyl alcohol (rubbing alcohol, 91%) Bamboo skewers or glass rods (1 per person)
100 ml of an "Extraction Buffer Solution" for strawberry spooling made with:
- 90 ml water
- 10 ml of liquid detergent 2 grams of salt

Preparations:

1. Cut squares of the cheesecloth into sizes large enough to fit the funnels

2. Prepare an ice bath of ice cubes and water to chill some isopropanol on ice.

Procedure:

1. Spread a cheesecloth square into a funnel.

2. Set the funnel with cheesecloth over a test tube.

3. Place one strawberry in a Ziploc sandwich bag and zip it closed. Try to get out as much air as you can.

4. Crush the strawberry with your fingers only for 1 minute. If you didn't get out most of the air, the bag will pop at this point. If the bag breaks, put the entire broken bag and crushed strawberry into another bag and continue.

5. Open the bag and add 10 ml of the Extraction Buffer Solution to the bag and zip it closed again. Try to get out as much air as you can.

6. Crush the strawberry pulp-buffer solution mixture again with your fingers for about 1 minute.

7. Use the scissors to make a small cut across a lower corner of the bag. Squeeze the extract (strawberry mixture) from the bag onto the cheesecloth in the funnel and let it drip into the test tube.

8. Filter the strawberry mixture into a test tube so that it is **ONLY 1/4 to 1/3 full**. **Do not fill to the top!**

9. **Slowly** pour the **cold isopropyl alcohol** down the **side** of the test tube until the total volume is doubled. You should now have a red/pink bottom layer and a clear top layer.

10. Dip the bamboo skewer or glass rod into the test tube where the alcohol and strawberry extract layers meet and "spool" or pull out the precipitated DNA!

Observations and Questions:

1. What does salt do to a cell?

2. What does soap do to a cell?

3. Why would you crush the pulp for 1 minute instead of 15 minutes?

4. What is happening in the top layer after the alcohol is added?

5. What is happening at the interface between the two layers?

6. Can you see the DNA? What does it look like?

7. Why do you think the DNA appears?

8. What is the color of your strawberry DNA?

9. Is it pure DNA? What else could be attached to it?

10. How does it compare with another team's results in terms of amount and color?

11. Why did some people pull out white DNA and some got pink DNA?

12. Why did some people get more DNA than others?

Answers to Questions:

1. What does salt do to a cell?

   Salt breaks the isotonic balance of the cell, causing it to burst and provides the sodium ions that neutralizes the negative DNA charge making them clump easier.

2. What does soap do to a cell?

   Soap detergents mimic the lipid bilayer structure of a cell. It pokes holes in the cell membrane and nuclear membrane, releasing the DNA.

3. Why would you crush the pulp for 1 minute instead of 15 minutes?

   The longer you crush, the warmer the mixture gets.. Enzymes that break up DNA also mix with the DNA when the membranes are all broken. The warmer your mixture gets, the better those enzymes are at breaking up your DNA, making it harder to clump and spool.

4. What is happening in the top layer after the alcohol is added?

   Bubbles of oxygen form and rise, helping to carry DNA from the interface through the alcohol where it is not soluble and, thus, precipitates out of solution.

5. What is happening at the interface between the two layers?

   The interface is where DNA precipitates out of solution.

6. Can you see the DNA? What does it look like?

   Normally, you can't "see" the DNA strands if they are pure. The clear, colorless, single strand is too small for the eye to see. What you should see are clumps of DNA that appear clear to opaque or slightly pink due to colored proteins that clump up with it.

7. Why do you think the DNA appears?

   The DNA is not soluble in alcohol.

8. What is the color of your strawberry DNA?

It should be clear to opaque or slightly pink .

9. Is it pure DNA? What else could be attached to it?

Pure DNA is clear, but, most likely it is not 100% pure DNA. Lost of proteins that normally bind to DNA to form chromosomes that may precipitate with the DNA.

10. How does it compare with another team's results in terms of amount and color?

This activity should result in lots of variety in terms of amount and appearance of DNA. There are a number of variables that can affect this including the size of the strawberry, the time used to crush it, and the time of extraction.

11. Why did some people pull out white DNA and some got pink DNA?

Varying amounts of colored proteins in the strawberries will give these color changes. .

12. Why did some people get more DNA than others?

The sample size and the degree to which the fruit is crushed will give different results.