Student Data Page

Isolation of DNA

Some questions to get you thinking about the lab:

1. One way to purify a molecule is to get rid of everything but that molecule. If we want to isolate DNA from kiwi fruit, what do we have to get rid of?

2. What does DNA actually look like?

Results

1. Describe your observations from the isolation of DNA.

2. How does the DNA feel?

3. Have you ever eaten DNA?

Instructor's Notes

Isolation of DNA

Resources and Other Procedures

1. A kit for isolating DNA from an onion can be purchased for $11.95 at:

   http://www.hometrainingtools.com/catalog/special-categories/science-kits/life-science-kits/p_be-dnalab.html

2. Other procedures for isolating DNA from an onion can be found at:

   http://www.cmri.com.au/forms/DNAExtraction.pdf
   or
   http://library.thinkquest.org/18617/data/funstuff/funstuff.html

Materials

- knife for cutting kiwi
- one small ziplock bag per group of students
- jar or beaker that fits strainer or funnel
- strainer or funnel
- cheese cloth (or a #6 coffee filter)
- ice water bath (a large mixing bowl works well)
- water
- clear-colored shampoo, such as Suave Daily Clarifying Shampoo
- kiwifruit, half a kiwi per group of students
- table salt, either iodized or non-iodized
- 1 large test tube (holds 20 ml) per group, preferably with a cap
- 1 small test tube (holds 10 ml) for each student, preferably with a cap
- cold 95% ethanol (grain alcohol)

Preparation

1. Prepare Extraction Solution:
   For one liter of the extraction solution, mix 100 ml of shampoo (eg Suave Daily Clarifying Shampoo, many shampoos will work, but do not use shampoos with conditioner or baby shampoo) and 15 g of table salt (iodized or non-iodized both will work). Add water to make a final volume of 1 liter. Dissolve the salt by stirring slowly to avoid foaming. Measure 20 ml of solution for each pair of students
2. Set up an ice water bath.
3. Each group will use half a kiwi and 20 ml of the following shampoo solution:
   For one liter of the shampoo solution, mix 100 ml of shampoo and 15 g of table salt. Add water to make a final volume of 1 liter. Dissolve the salt stirring slowly to avoid foaming. Measure 20 ml of solution for each group of students.
4. Peel the kiwi, cut them into about 12 pieces each.

Explanation of what's happening in each step

1. **Get 6 pieces of kiwi and put them in a ziplock bag.**
2. **Add 20 ml of extraction solution to the ziplock bag. Make sure the bag is closed without much extra air.**
3. **Mush the kiwi thoroughly but carefully so the bag doesn't break, for about 5 minutes. <u>What does mushing the kiwi do?</u>**

   Breaks the cell wall. The soap in the extraction solution destroys the cell and nuclear membranes, allowing the DNA to get out. There is also salt in the extraction solution, which causes the proteins and carbohydrates to precipitate, while the DNA remains in solution.

4. **Cool the kiwi mixture in the ice bath for a minute. Then mush the kiwi more. Cool, then mush. Repeat this several times. <u>Why do we cool the mixture?</u>**

   Cooling protects the DNA. There are DNases (enzymes that destroy DNA) in the cell's cytoplasm. The DNA is usually protected from DNases by the nuclear membrane, but that is destroyed by the soap. Cooling slows down the DNases, just like it would any enzymatic reaction. DNases are in our cells to protect us from foreign DNA (like viruses).

5. **Filter the mixture through the cheesecloth. All the groups can combine their mixtures at this point, to filter together. <u>What is being filtered out?</u> <u>What is going through the filter?</u>**

   Students can usually see the seeds being filtered out. Most of the cell parts and the precipitated protein and carbohydrate are also being filtered out at this point.

6. **Add approximately 2 ml of kiwi solution into each test tube, one for each student.**
7. **Being careful not to shake the tubes, add approximately 2 ml of cold 95% ethanol to each tube. <u>What do you think the ethanol does?</u> <u>Why do we want it cold?</u>**

   We don't have to worry about the DNases at this point, because hopefully they've mostly been filtered out. What we are most concerned about is precipitating (or solidifying) the DNA. The colder something is, the more likely it will precipitate or solidify. Cooling the alcohol just increases the amount of DNA that precipitates.

8. **Take a look at your tube. <u>What do you see in the top portion of the liquid?</u>**

   That's the DNA!