

DNA Extraction from Kiwifruit

Student Instructions

Introduction

DNA is present in the cells of all living organisms. This procedure is designed to extract DNA from kiwi in sufficient quantity to be seen and spooled. . Because DNA exists inside of cell and ultimately inside of the nucleus proteins must be denatured to locate the DNA. There are different proteins that form the membranes of the organelles and that allow DNA to coil so that it can fit inside of the nucleus. This activity is ideal for students to work in pairs, but each student will have a tube of DNA at the end.

Some questions to get you thinking about today's lab:

1. One way to purify a molecule is to get rid of everything but that molecule. If we want to isolate DNA from kiwifruit, what do we have to get rid of?
2. What materials would you use to do that?
3. What can we do with the DNA once we've purified it?

Materials

- ziplock bags
- jar or beaker that fits strainer or funnel strainer or funnel
- cheese cloth (cut to cover the funnel)
- ice water bath (a large mixing bowl works well)
- extraction solution
- kiwifruit
- cold 95% ethanol or isopropanol
- small test tubes (1 per student)

Protocol

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. Mash the kiwi thoroughly but carefully so the bag doesn't break, for about 5 minutes. What does mashing the kiwi do?
3. What do you think the extraction solution is? What does it do to the kiwi?
4. Cool the kiwi mixture in the ice bath for a minute. Then mash the kiwi more. Cool, then mash. Repeat this several times. Why do we cool the mixture?
5. Filter the mixture through the cheesecloth. All the groups can combine their mixtures at this point, to filter together. What is being filtered out? What is going through the filter?
6. Dispense 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. What do you think the ethanol does? Why do we want it cold?
8. Take a look at your tube. What do you see in the top portion of the liquid?

Teacher Guide: What is DNA? DNA Extraction from Kiwifruit

Introduction

DNA is present in the cells of all living organisms. This procedure is designed to extract DNA from kiwi in sufficient quantity to be seen and spooled. Because DNA exists inside of cell and ultimately inside of the nucleus proteins must be denatured to locate the DNA. There are different proteins that form the membranes of the organelles and that allow DNA to coil so that it can fit inside of the nucleus. This activity is ideal for students to work in pairs, but each student will have a tube of DNA at the end.

Management & Grouping Procedures

1. Students will work in predetermined groups of 3.
2. Students will be assigned specific jobs within the group
Example: time keeper and organizer
Collect materials and clean up
*All students will be responsible for making and recording observations

Safety

Students will be reminded of the following safety rules

1. Do not perform any experiment unless you are given permission by your teacher.
2. Do not touch, taste, or smell any of the lab materials.
3. No eating or drinking in the lab.

Phase of Inquiry: Explore and Explain

Some questions to get you thinking about the lab:

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

All parts of the cell besides the DNA, i.e. cell wall (kiwi is a plant, after all), cell membrane, mitochondria, Golgi apparatus, endoplasmic reticulum, vacuoles, lysosomes, nuclear membrane, etc.

What materials would you use to do that?

Something to mush the cells (blender or your hands), something to destroy membranes (soap dissolves them), something to get rid of proteins and carbohydrates (salt causes them to precipitate), something to separate insoluble cell stuff from soluble DNA, and something to help get the DNA (alcohol precipitates it).

- What can we do with the DNA once we've purified it?

Use it in DNA fingerprinting (solve a crime, see a genetic defect), put it into another organism to give it specific traits (this is called transformation or genetic engineering), other?

Motivational Opening:

When students enter the classroom, they will answer the following question.

Why do people use conditioners after washing their hair?

Teacher will review the structure and function of protein.

Teacher and students will discuss the answer to the question listed above.

Content: (These areas will be covered throughout a period of 5-7 days.)

1. Structure and function of proteins.
2. Properties of protein.
3. Denaturing proteins
4. Structure and function of DNA.
5. Comparison between the structure and DNA and the structure of protein.
6. Relationship between the function of DNA and the function of protein.

Materials

1. ziplock bags (1 per student pair)
2. jar or beaker that fits strainer or funnel (1 per 10 students)
3. strainer or funnel (1 per 10 students)
4. cheese cloth (cut to cover the funnel)
5. ice water bath (a large mixing bowl works well) (1 per 4-6 students)
6. extraction solution (1 tube of 20 mls per student pair)
7. kiwifruit (cut into 12 pieces, each student pair needs 6 pieces)
8. cold 95% ethanol or isopropanol (2-3 mls per student)
9. small test tubes (1 per student)

Extraction solution recipe:

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.

Protocol - These are the directions for each pair. Students will complete the critical thinking questions upon their completion of the lab.

- 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. Mash the kiwi thoroughly but carefully so the bag doesn't break, for about 5 minutes. What does mashing the kiwi do?**
Breaks the cell wall
- 3. What do you think the extraction solution is? What does it do to the kiwi?**
Soap will cause the solution to bubble so students should be able to guess what's in here. The soap 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 mash the kiwi more. Cool, and then mash. Repeat this several times. Why do we cool the mixture?**
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. What is being filtered out? What is going through the filter?**
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. Dispense 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. What do you think the ethanol does? Why do we want it cold?**
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. What do you see in the top portion of the liquid?**
You can actually pick up the DNA at this point, using a toothpick, wood pencil, or glass stirring rod.

Closure:

Students will observe each other's test tube.

Students will answer the following questions on an exit card.

Why do people use meat tenderizer?