DNA EXTRACTION FROM KIWI

Prepared by the Office of Biotechnology, Iowa State University

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. It is based on the use of household equipment and supplies.

MATERIALS

For teacher preparation

- two 4-cup measuring cups (1000 ml) with ml markings
- one 1-cup measuring cup (250 ml) with ml markings
- measuring spoons
- knife for cutting kiwi
- large spoon for mixing and mashing kiwi
- thermometer that will measure 60° C (140° F), such as a candy thermometer
- strainer or funnel that will fit in a 4-cup measuring cup
- #6 coffee filter or cheese cloth
- hot tap water bath (60° C) (a 3-quart saucepan works well to hold the water)
- ice water bath (a large mixing bowl works well)
- distilled water
- clear shampoo, such as Suave Daily Clarifying Shampoo
- 2 to 3 kiwi
- table salt, either iodized or non-iodized

Supplies provided to the class

- 1 test tube for each student, preferably with a cap, that contains the kiwi solution. (A narrow glass container or clear bud vase can substitute for the test tube.)
- pasteur pipettes or medicine droppers
- 95% ethanol (grain alcohol)
- laboratory instructions
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TEACHER PREPARATION

1. Set up hot water bath at 55-60° C and an ice water bath.

2. For 2 to 3 kiwi, make a solution consisting of one tablespoon (10 ml) of liquid dishwashing detergent or shampoo and one level 1/4 teaspoon (1.5 g) of table salt. Put in a 1-cup measuring cup (250 ml beaker). Add distilled water to make a final volume of 100 ml. Dissolve the salt by stirring slowly to avoid foaming.

3. Peel the kiwi, cut them into about 12 pieces, and put the pieces into a 4-cup measuring cup (1000 ml).

4. Cover the kiwi with the 100 ml of solution from step 2. The detergent dissolves the fatty molecules that hold the cell membranes together, which releases the DNA into the solution. The detergent, combined with the heat treatment used in step 5, causes lipids (fatty molecules) and proteins to precipitate out of the solution, leaving the DNA. The salt enables the DNA strands to come together.

5. Put the measuring cup in a hot water bath at 55-60° C for 10-12 minutes. During this time, press the kiwi mixture against the side of the measuring cup with the back of the spoon. Do not keep the mixture in the hot water bath for more than 15 minutes because the DNA will begin to break down.

6. Cool the mixture in an ice water bath for 5 minutes. During this time, press the kiwi mixture against the side of the measuring cup with the back of the spoon.

7. Filter the mixture through a #6 coffee filter placed in a strainer over a 4-cup measuring cup. When pouring the mixture into the strainer, avoid letting foam get into the measuring cup. It can take more than an hour to recover most of the liquid. The filtering can be done in a refrigerator overnight.

8. Dispense the kiwi solution into test tubes, one for each student. The test tube should contain about 1 teaspoon of solution or be about 1/3 full, whichever is less. For most uniform results among test tubes, stir the solution frequently when dispensing it into the tubes. The solution can be stored in a refrigerator for about a day before it is poured into the test tubes. When the solution is removed from the refrigerator, it should be gently mixed before the test tubes are filled.
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STUDENT INSTRUCTIONS

The process of extracting DNA from a cell is the first step for many laboratory procedures in biotechnology. The scientist must be able to separate DNA from the unwanted substances of the cell gently enough so that the DNA is not broken up.

Your teacher has already prepared a solution for you, made of kiwi treated with salt, distilled water and dishwashing detergent or shampoo. The salt allows the DNA to precipitate out of a cold alcohol solution. The detergent causes the cell membrane to break down by dissolving the lipids and proteins of the cell and disrupting the bonds that hold the cell membrane together. The detergent then forms complexes with these lipids and proteins, causing them to precipitate out of solution.

PROCEDURE

1. Add cold alcohol to the test tube to create an alcohol layer on top of about 1 cm. For best results, the alcohol should be as cold as possible. The alcohol can be added to the solution in at least three ways. (a) Put about 1 cm of alcohol into the bottom of a test tube and add the kiwi solution. (b) Fill a pasteur pipette with alcohol, put it to bottom of the test tube, and release the alcohol. (c) Slowly pour the alcohol down the inside of the test tube with a pasteur pipette or medicine dropper. DNA is not soluble in alcohol. When alcohol is added to the mixture, the components of the mixture, except for DNA, stay in solution while the DNA precipitates out into the alcohol layer.

2. Let the solution sit for 2 to 3 minutes without disturbing it. It is important not to shake the test tube. You can watch the white DNA precipitate out into the alcohol layer. When good results are obtained, there will be enough DNA to spool on to a glass rod, a pasteur pipette that has been heated at the tip to form a hook, or similar device. DNA has the appearance of white mucus.

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