

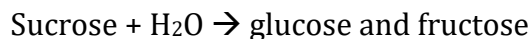
Activity Guide for Students: Fracking for Fructose

Purpose: To learn more about the enzymatic modification of sugars through hydrolyzing sucrose (breaking down sucrose with water) and to build molecular models of fructose, glucose, sucrose and a common artificial sweetener, sucralose.

Materials:

- Molecular modeling set for each lab group
- Sheet of relevant chemical structures
- URS-1G-100 Teco glucose assay strips
- Scissors (to cut glucose strips in half lengthwise, so one strip will become two)
- Invertase enzyme
- Bread yeast packets
- Sucrose (table sugar)
- Test tubes (6 per student group)
- Test tube rack
- Wax pencils or lab markers for labeling test tubes
- Disposable graduated plastic pipettes (2 per student group)
- 250-ml beakers or cups (2 per student group)
- Thermometers
- Balances
- Weigh paper or weigh boats
- Stop watches, timers or timers on a cell phone
- Gloves
- Water
- Ice (may be needed to make a cold water bath)

Background: Sucrose, or table sugar, is a disaccharide composed of two chemically bonded monosaccharides — glucose and fructose. The enzyme invertase, or sucrase, catalyzes the hydrolysis reaction that breaks sucrose into glucose and fructose (see reaction below).



Fructose is sometimes preferred over sucrose in the food industry because it tastes sweeter and does not crystalize as easily. Purified invertase can be added to candy recipes to break up sucrose and keep sugars liquid (like the fillings inside some chocolates). Yeast uses lots of enzymes to digest nutrients, but among others it has the invertase enzyme. Glucose test strips measure glucose (not sucrose or fructose) and can be used to monitor the enzymatic hydrolysis reaction of sucrose to glucose and fructose.

Procedure for the hydrolysis of sucrose:

1. Cut the glucose test strips in half lengthwise so you can perform two tests with one strip.
2. Label six test tubes with letters or numbers to distinguish the tubes from each other.
3. Fill those six test tubes until they are half full of room temperature water.
4. Use the thermometer to measure the temperature of the water. Record your result.
5. Briefly dip a glucose test strip into a tube of water, then let the strip dry on the table for a minute or two.
6. Compare the color of the strip to the color code on the side of the bottle of test strips. How much glucose is in the water? Record your data on the accompanying sheets.
7. Using the weigh boat or paper, measure about 0.5 gram of sucrose. Record actual mass on your data table. Transfer the sucrose to a test tube. (If possible, measure the sucrose directly into the test tube by zeroing out the mass of the tube and water first.) Repeat this process for each of the remaining test tubes.
8. Shake or swirl each of the test tubes to help the sucrose dissolve in the water.
9. Briefly dip a glucose test strip into a tube of sucrose water, then let the strip dry on the table for a minute or two. Compare the color of the strip to the color code on the side of the bottle of test strips. How much glucose is in the water? Record your data on the accompanying sheets.
10. Use the scale and a clean sheet of weigh paper or a weigh boat to measure out 0.5 grams of yeast. Add 0.5 grams of yeast to three of the test tubes. Record which test tubes the yeast is added to. Shake or swirl the test tubes.

11. Shake or swirl the bottle of invertase enzyme to make sure it is well mixed. Use a disposable graduated pipette to add 0.5 ml of invertase to the other three tubes, and label the tubes. Shake or swirl the test tubes.

12. Fill a beaker most of the way with hot water. What is the water temperature? Record your results.

13. Fill a beaker most of the way with cold water (add ice, if needed). What is the water temperature? Record your results.

14. Set or hold one yeast tube and one invertase tube in the beaker of hot water, one yeast tube and one invertase tube in the beaker of cold water, and one yeast tube and one invertase tube at room temperature (in no water bath).

15. After 1 minute, test the solution in each tube with a new glucose test strip and put the tubes back into their appropriate water bath. How much glucose is present? Also, take the temperature of your water baths. Record your data on the accompanying sheets.

16. After 2 minutes, test each tube with new glucose test strips and take the temperature of your water baths. Record your data.

17. Repeat the testing and recording procedure on each minute until 7 minutes.

18. Graph your data for the cold temperature on the accompanying sheets (add appropriate units to the axes).

19. Graph your data for room temperature on the accompanying sheets (add appropriate units to the axes).

21. Graph your data for the hot temperature on the accompanying sheets (add appropriate units to the axes).

22. Analyze your data by answering the questions that follow at the end of this procedure.

23. If time allows, you can add the following procedures. Based on your results in the first activity, write a hypothesis to accompany each change in variable. Then, using the given procedure as a guide, write your own procedure for testing each hypothesis. Using the given data tables, record your data. Then, summarize your results.

A. Test and graph the results for different amounts of sucrose per tube. Time, temperature and invertase or yeast amount should be the same for all tubes.

B. Test and graph the results for different amounts of invertase or yeast per tube, with the time, temperature and sucrose amount the same for all tubes.

Analysis questions:

1. How did the temperature affect the enzymatic cleavage of sucrose to glucose and fructose? Did the temperature of your water bath change over the 7 minutes? If so, how might the change in temperature have affected your results?

2. What hydrolyzed sucrose to glucose and fructose more rapidly — yeast or invertase?

3. Pick one of the measured time points and graph the results for one of the different water baths on the same graph. Graph other temperatures and record your results. What temperature gives the most glucose?

Procedure for molecular modeling:

1. Use the molecular model kit and the sheet of chemical structures to build a model of glucose. Show the model to your teacher or take a photo with a cell phone if one is available to you.

2. Use the molecular model kit and the sheet of chemical structures to build a model of fructose, preferably without dismantling your glucose (if you have enough parts). Show the model to your teacher or take a cell phone photo.
3. Hook your glucose and fructose models together to make a sucrose model (refer to chemical structure sheet). Show the model to your teacher or take a cell phone photo. Notice what changes must occur when enzymes convert sucrose into glucose and fructose.
4. Use the molecular model kit to build a model of sucralose (Splenda artificial sweetener). If sucralose were hydrolyzed by invertase, what molecules would result? Build a model of the resulting molecules and show them to your teacher or take a cell phone photo.
5. Based on your knowledge of enzymes, could invertase hydrolyze sucralose? Why or why not?
6. What did you learn about the structure of sugars?
7. What did you learn about the structure of artificial sweeteners?
8. What did you learn about enzymes that operate on sugars?