Alcoholic Fermentation in Yeast

Adapted from "Alcoholic Fermentation in Yeast Investigation" in the School District of Philadelphia Biology Core Curriculum © 2011 by Drs. Jennifer Doherty and Ingrid Waldron, University of Pennsylvania Biology Department¹

All living cells, including the cells in your body and the cells in yeast, need energy for cellular processes such as pumping molecules into or out of the cell or synthesizing needed molecules. **ATP** is a special molecule which provides energy in a form that cells can use for cellular processes. Each cell in our body and each yeast cell can use the energy stored in organic molecules in food to make ATP.

<u>When O_2 is available</u>, cells use **aerobic cellular respiration** to transfer energy from the organic molecules in food to ATP. As shown in the figure, aerobic cellular respiration is a complex process that begins with **glycolysis**, followed by the **Krebs cycle** and the **electron transport chain**. Aerobic cellular respiration can make up to 29 molecules of ATP per molecule of glucose. Most of this ATP is produced by the electron transport chain which can only function if O_2 is available.



<u>When O_2 is not available</u>, cells can make ATP using glycolysis followed by **fermentation**. Glycolysis produces 2 ATP and fermentation restores molecules needed for glycolysis to continue. Glycolysis followed by fermentation produces much less ATP than aerobic cellular respiration, but fermentation is very useful when O_2 is not available.

In the figure, fermentation is referred to as **anaerobic** processes. The "an" in front of aerobic means "not aerobic". There are two types of anaerobic fermentation:

- lactate fermentation (e.g. in muscles when an animal exercises hard)
- alcoholic fermentation (e.g. in yeast, which can be used to make wine or beer)

★ Use the terms carbon dioxide and oxygen to complete the following equation to describe aerobic respiration.
Glucose + → + Water

¹ Teachers are encouraged to copy this student handout for classroom use. A Word file (which can be used to prepare a modified version if desired), Teacher Preparation Notes, comments, and other hands-on activities are available at http://serendip.brynmawr.edu/sci_edu/waldron/. Additional biology activities are available at http://serendip.brynmawr.edu/exchange/bioactivities

Experiment I - Effects of Sucrose Concentration on the Rate of Alcoholic Fermentation in Yeast

1. Humans use yeast every day to make bread, wine and beer. What is yeast?

If you want to make your own bread, you can buy yeast in the grocery store. This yeast consists of little brown grains. The little brown grains of yeast may not seem to be alive, but if you put them in water with sugar, the yeast will take up the sugar and use the energy stored in the sugar molecules to make ATP and carry out the processes of life.

2. What is **sucrose**?

Yeast can convert sucrose into glucose and use the glucose to provide the energy to make ATP.

3. In your experiment, you will grow yeast in a test tube filled with water and sealed with a balloon. Do you think these growth conditions are aerobic or anaerobic?

Under anaerobic conditions, yeast carries out glycolysis to produce ATP, followed by alcoholic fermentation which produces ______ and ______.

To measure the rate of alcoholic fermentation in yeast, you can measure the amount of CO_2 gas the yeast produces. CO_2 production can be measured by measuring the depth of the layer of bubbles trapped in foam on top of the yeast solution and also by observing the balloons, which get bigger as they catch the CO_2 produced by the yeast.

4. To test whether the concentration of sucrose affects the rate of alcoholic fermentation in yeast, you will measure the rate of CO_2 production for 4 different concentrations of sucrose. Complete the table to predict how much CO_2 production you expect in each case.

Sucrose Concentration	Predicted Amount of CO ₂ Production (e.g. a little, none, the most, less than, more than, the same as)		
0% (plain water)			
1% sucrose			
5% sucrose			
10% sucrose			

4. What will be the independent variable in your experiment?

What will be the dependent variable in your experiment?

5. What will be the control treatment in your experiment?

What is the purpose of this control treatment?

- 6. The procedure to measure alcoholic fermentation is:
 - 1) Label each test tube, 0%, 1%, 5%, or 10%.
 - 2) Add 10 mL of the appropriate water or sucrose solution to each tube.
 - 3) For each tube, add 0.5 mL or 1/8 tsp of yeast and put a balloon firmly over the top.
 - 4) With your thumb sealing the top, shake each tube until the yeast is dissolved.
 - 5) Measure the depth of bubbles produced and observe the balloons as soon as the test tubes are prepared and after 10 minutes and 20 minutes.
- 7. Record your observations in these data tables.

Sucrose Concentration	Depth of CO ₂ bubbles at:		
	0 minutes	10 minutes	20 minutes
0% (plain water)			
1% sucrose			
5% sucrose			
10% sucrose			

Sucrose Concentration	Balloon Description				
	0 minutes	10 minutes	20 minutes		
0% (plain water)					
1% sucrose					
5% sucrose					
10% sucrose					

8. <u>While you're waiting</u> for the 10 minute and 20 minute data collections, complete the question below and the two questions on the top of the next page.

 \star Use the information from page 1 to complete the figures below. Fill in the ovals with the appropriate molecule. On the blank lines write the name of the appropriate process. In the boxes at the bottom of the figure write how much ATP is made in each pathway.

With Oxygen (Aerobic)



Without Oxygen (Anaerobic)



★ What is the main advantage of aerobic respiration?

 \star What is the main advantage of anaerobic fermentation?

9. Compare your results in question 7 with your predictions in question 4. Did the amounts of CO_2 produced at different sucrose concentrations match your predictions? If not, how did the results differ from your expectations?

10. Discuss your results with your group. What conclusions concerning the relationship between sucrose concentration and the rate of alcoholic fermentation are supported by your results?

11. Compare your results with the class results or with the results of the group next to you. Are your results generally similar? If there are any significant differences in results, what could be the reason for these differences?

Experiment II - Yeast and Other Ingredients in Bread

When you make bread, if you just mix flour, sugar and water, the dough does not rise, and the bread will be flat and hard. If you include yeast in the bread dough, then the dough rises and the bread is bigger and fluffier.

1. Explain how yeast helps the bread dough to rise.

2. Consider the results of your last experiment with yeast and sucrose. If you added flour, which treatment would have made the fluffiest bread?

Today you will design and carry out an experiment to investigate other variables, besides the concentration of sugar, which may affect the fluffiness of bread. Bread dough usually has other ingredients besides yeast, sugar, water and flour. Some other common ingredients in bread dough are salt, fats (e.g. oil or butter), eggs, and flavorings such as cinnamon and raisins. Any of these ingredients could affect the rate of alcoholic fermentation of the yeast and thus affect the fluffiness of the bread. The temperature at which the bread dough is kept to rise might also affect the fluffiness of bread.

You will not actually test how one of these ingredients or temperature affects the fluffiness of bread. Instead, you will use the same experimental setup as before (that is, test tubes with yeast mixture, a ruler and balloons) to test the effect of one of these variables on the rate of CO_2 production.

3. What question will you investigate?

4. Write a hypothesis that you will test to help you answer this question.

5. Plan an experiment to test your hypothesis. What is the independent variable in your experiment?

What is the dependent variable in your experiment?

6. What is the control treatment in your experiment? (Hint: You are not testing how sugar affects alcoholic fermentation, so you will want to use 10% sucrose in each of your treatments)

7. Describe your procedures.

8. Create a data table or tables.

9. Perform your experiment and record your data.

10. Did the yeast produce different amounts of CO_2 with your different treatments? Do the results match your hypothesis?

11. What do your results mean for people who make bread using the ingredient or temperatures you investigated?

If you have used clean test tubes, ingredients and mixing procedures, you can use the yeast solution in one of your treatment test tubes to produce a small loaf of bread. Your teacher will give you instructions on how to add flour and water to your yeast solution and knead a bread ball. Make sure you label your baking cup with your name so you know which is yours when it comes out of the oven.