

Yeast Respiration Rate

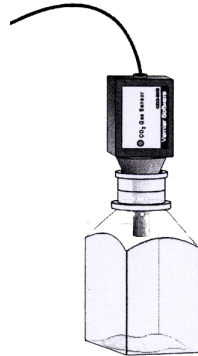
Most organisms can utilize a variety of fuel molecules for cell respiration, but not all potential fuel molecules are used equally well. Yeast, like all fungi, must be able to transport a potential fuel molecule into its body and have the enzymes that can oxidize the potential fuel molecule. Yeasts are able to metabolize some, but not all, sugars in their respiratory pathways. Recall that yeasts are facultative organisms. When oxygen is available to them, they will do aerobic respiration; in the absence of oxygen, they do alcohol fermentation. We can investigate the rate of aerobic respiration of different fuel molecules in yeast by measuring the rate of CO₂ production. CO₂ is a product of aerobic respiration. In this exercise we will use the Vernier CO₂ Sensor to determine the rate of respiration of a variety of sugars by yeast.

Materials Needed


Data Recorder (Computer)	Pipette
Vernier LabPro Box and Power Supply	5% Solutions of:
Vernier CO ₂ Sensor	Sucrose
250-ml Respiration Chamber (See diagram below)	Glucose
Distilled water	Lactose
Thermometer	Fructose
6 10X100 test tubes	Maltose
	600 ml beaker water bath 35 – 40°C
	Yeast suspension

Procedure

1. Remove the data recorder from the data recorder cubicles. You may have to ask your instructor to unlock the cubicles.
2. Assemble your CO₂ Sensor as shown in the diagram below. Connect the CO₂ Sensor to the Vernier LabPro box and plug the Vernier LabPro box into the appropriate data recorder port. Start your data recorder.



Respiration Chamber with CO₂ Sensor Attached

3. Open the Biology 211 folder that is located on the computer desktop screen, and open "Exp 12A – Respiration (Fermentation) in Yeast" from the *Biology with Computers* experiment files of *Logger Pro*.
4. When you have the experiment open, the vertical axis has CO₂ concentration scaled from 0 – 5000ppm, and the horizontal axis has time on a scale from 0 – 4 minutes. Data will be sampled at a rate of 4 samples per minute. Be sure that you have the collect button  displayed at the top of your screen "graph". If you do not, ask your instructor for assistance in configuring the computer.



5. Prepare your hot water bath for the yeast by filling your 600ml beaker with a combination of hot and cold tap water to reach 35 – 40° C. You will need about 300 – 400 ml of tap water. Leave your thermometer in the beaker for the duration of the experiment so you can monitor the temperature.
6. Label your 6 test tubes G (Glucose), S (Sucrose), F (Fructose), L (Lactose), M (Maltose) and W (Water).
7. Add 2ml of each of the sugar solutions to the appropriate test tube. Add 2 ml of distilled water to the tube labeled W.
8. Gently swirl your yeast suspension to suspend the yeast. Add 2ml of yeast into each of the 6 test tubes and agitate the test tubes to mix the yeast suspension with the sugar solution (or water in tube 5).
9. Set the 6 tubes in your water bath and incubate for 10 minutes. Keep the temperature constant by adding more hot or cold water as needed. Use a pipette to remove amounts of water equal the amount being added.
10. After 10 minutes, use a pipette to put 1 ml of solution G (Glucose) into the 250ml respiration chamber. (If you have attached the CO₂ Gas Pressure Sensor to the respiration chamber, remove it before adding the Glucose solution.) Record the temperature of the water bath in Table 1.
11. Quickly place the CO₂ Sensor into the opening of the respiration chamber. Gently twist the **stopper** on the shaft of the CO₂ Sensor into the respiration chamber opening. Do not twist the shaft of the CO₂ Sensor; it may get damaged if you do.
12. You are now ready to collect data! Click the collect button . Data will be collected for 4 minutes.
13. When the data collection is completed (4 minutes), remove the CO₂ Sensor from the respiration chamber. Use a notebook or notepad to fan air across the openings in the probe shaft of the CO₂ Sensor for 1 minute.
14. Thoroughly clean and dry the respiration chamber.
15. Determine the respiration rate(ppm/min) for the curve of respiration rate vs. time by performing a linear regression to calculate the rate of change. To do this:
 - Move the cursor to the point on your screen where the data values begin to change. Hold down the trackpad button and drag the cursor (using the finger on the trackpad) to the end of the data and release the button.
 - Click the Regression button  to perform a linear regression. A floating box will appear with the formula for a best fit line.
 - Record the slope of the line, m , as the respiration rate in Table 1. Note: You will record the "m" number displayed.
 - Close the linear regression floating box, and choose "Store Latest Run" from the Data menu.
16. Repeat steps 10 – 15 for each of the other 4 sugar solutions and your water sample.
17. Record your data on the board so we can obtain averages for the entire class.

Table 1

Sugar Tested	Temperature (°C)	Respiration Rate (ppm/min)
Glucose		
Sucrose		
Fructose		
Lactose		
Maltose		
Water		

When all data has been collected, calculate the class averages and record them in Table 2.

Table 2: Class Averages

Sugar Tested	Respiration Rate (ppm/min)
Glucose	
Sucrose	
Fructose	
Lactose	
Maltose	
Water	

Discussion Questions

1. Make a bar graph of sugar type vs. rate of fermentation. The rate values should be plotted on the y-axis and the sugar type on the x-axis. Use the class average data.
2. Based on your results, do yeasts utilize all sugars equally? If not, why do you think some sugars were metabolized better than others?
3. Why did you incubate the yeast prior to collecting data in the experiment?

* Materials for this laboratory were modified from *Biology with Computers*, by Holman and Masterman © Vernier Software and Technology.