

Fermentation lab report

Business



tation lab report Lab Exercise 7 The Effect of Temperature on the Rate of Carbon Dioxide Production in *Saccharomyces* I. Student Objectives 1. The student will use this lab exercise as the basis for writing a scientific method report. 2.

The student will understand how the rates of chemical reactions are affected by temperature. 3. The student will understand the overall fermentation reaction by yeast, starting with glucose as an energy source. 4. The student will understand how to measure fermentation rate.

II. Introduction The student is to use this lab exercise as the foundation for writing a scientific method report. The instructions for writing the report are found in the addendum section of the lab manual. The purpose of the experiment is to test the effect of five different temperatures on the rate of carbon dioxide production in yeast. The experiment is an example of alcoholic fermentation that is characteristic of yeast.

The original energy source of glucose is in the form of molasses in the lab. The carbon dioxide that is measured is in the form of gas bubbles, seen in fermentation tubes. The overall chemical equation is the Gay-Lussac Equation, which states that: Glucose + water produces ethanol + carbon dioxide + ATP. This fermentation reaction is anaerobic, taking place without the presence of oxygen. It is an ancient method of alcoholic fermentation, using yeast, and produces a small amount of energy in the form of ATP.

A hypothesis is typically referred to as “ an educated guess”. The student is expected to generate a hypothesis for this lab experiment, test it, and then report if the hypothesis has been accepted or rejected, and why. Yeast is an

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example of a sac fungus, and is eukaryotic and unicellular. The rates of chemical reactions will increase with increasing temperatures, up to a certain point. When living organisms are used in chemical reactions, such as yeast, an added variable becomes important to consider.

That variable is the presence of enzymes in the yeast. Enzymes are proteins that function in optimal environmental conditions. The conditions include heat, pH and salinity. Different enzymes function best in different environments. Sometimes, the temperature of an environment becomes too hot, and the action of the enzymes becomes unsustainable.

As enzymes are dependent on their protein shape remaining unaltered, if anything should break chemical bonds to change the protein's conformation, that protein becomes denatured. A denatured protein is one with an altered shape, and that means it cannot function as it was originally designed. The fermentation rate in this experiment is measured in ml/min. It is the rate of carbon dioxide production that is measured over time spent in a water bath. Carbon dioxide is a gas, and the lab has no direct method of gas measure.

Thus, an indirect method must be used. Water will substitute for the gas measure's mark at the conclusion of the experiment, and the amount of water in milliliters will serve as an indirect method of fermentation rate. As this experiment is to be used for a scientific method report, the student must answer and cite the appropriate references for the following questions, as the Introduction of the report is written: 1. When one refers to the temperature of a system, what does this mean? 2. How are chemical

reactions, especially their rates, influenced by temperature? 3. Give various examples of the effect of temperature on biological systems.

4. What is yeast? 5. What is the overall fermentation reaction by yeast, starting with glucose as an energy source? 6. What is an enzyme? 7. How are enzymes and the reaction rates they control influenced by temperature? 8. How can fermentation rate be measured? 9.

What is the hypothesis of the effect of temperature on the rate of carbon dioxide production? III. Materials and Methods Students work in groups of four for the experiment, but each student writes his own scientific method report. The Title of the report is the title of this experiment. The student obtains the names of the lab partners, includes those names on the Title page of the report, and identifies the report as written by a specific student. Specific students identify their reports by putting their name in a different font, color, or in boldface type on the Title page. All lab materials are in the refrigerator, or on the carts set out in the lab.

The water baths are set up on the lab counters. They are labeled with the appropriate temperatures. Obtain 5 fermentation tubes and label them with the following temperatures: 25°C, 35°C, 45°C, 55°C, and 65°C. The first temperature is equivalent to room temperature. The remaining four temperatures are set up in four water baths in the lab.

Put group initials on each of the 5 tubes. Use the red wax pencils. Add 30 ml of yeast + sugar culture to each of the 5 tubes. Be sure and swirl the flask first to suspend any cells that may have settled out of solution over time. The yeast culture contains glucose, in the form of molasses.

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The brand of molasses used is called “Grandma’s Molasses”, a common retail product found in grocery stores. The yeast is common baker’s yeast, “Fleischmann’s rapid rise”, a common retail product found in grocery stores. It is important to note the expiration date of the yeast packets, found on the back. Three packets of yeast have been added to the glucose mixture. This is equivalent to a cell concentration of approximately 450, 000 cells/cm³.

This absolutely insures there will be enough yeast, and that the reaction will proceed rapidly. Place each tube in a water bath at the appropriate temperature, and record the START time. Allow to ferment (form gas bubbles) until 1/3 of the closed end of the tube has been filled with carbon dioxide. In the case of slow or non-reactions, keep in water bath as long as possible, leaving enough time to complete the following sections. When 1/3 of the closed end of the tube has been filled with gas, mark the level with a red marking pencil, and record the END time.

Calculate the difference between start and end times, noting how much time each fermentation tube remained in the water baths. Pour out the yeast solution, fill the closed end of the tube with water to the mark, pour into a graduated cylinder, and record the volume in milliliters. The rationale is that gas cannot be measured directly in the lab; water, however can be measured to the mark, and can serve as an indirect measure of gas production. Calculate the rate of carbon dioxide production by dividing the volume of the gas produced by the time. Dimensions are ml/min.

There are 5 values, one for each of the 5 water bath temperatures. This is the data that should appear on a graph, marked Figure 1. Calculate the

temperature co-efficient value of Q10. This is done for four temperature intervals. Start with the two lowest temperatures, 250 and 350 C. The temperature co-efficient is a numerical value that refers to the relative rate of change in carbon dioxide production, over a 100 C change in temperature.

Four temperature intervals are used for calculation purposes because the lowest temperature, corresponding to room temperature, does NOT have a ten degree lower temperature for comparison. The refrigerator is not used in this experiment for any of the temperature variables. The calculation is straight forward: $Q_{10} = \frac{\text{Rate at } T}{\text{Rate at } T - 100 \text{ C}}$ The value of T is the temperature of the individual water bath used for comparison. Thus, T - 100 C represents the cooler temperature by 100. An example is: If the carbon dioxide production was 5 ml/min at 250 C, and 10 ml/min at 350 C, then $Q_{10} = \frac{10 \text{ ml/min}}{5 \text{ ml/min}} = 2$ The value of 2 is interpreted to mean that the rate of carbon dioxide production was twice as fast (it had doubled) as the temperature rose by 100 C, when these two particular temperatures were compared.

The values of the temperature co-efficient can be graphed, or presented in histogram format, as the student prefers. IV. Results and Interpretation A continuous graph summarizing the effect of temperature on carbon dioxide production should be presented in the report as Figure 1. The graph can be roughly drawn in the lab exercise, and used later for reference. Temperature of water bath is the independent variable, and should be presented along the x-axis.

The carbon dioxide rate of production is the dependent variable, and is graphed along the y-axis. A graph for Figure 1 is presented. Graph the five variables, and write out the conclusions based on the data in the graph.

Sarah, Please insert one of the Kendall/hunt graphs here for the students.

Thank you. Please label it Figure 1.

I guess we had better give the students an entire page for this graph + their conclusions, under it. So the size of the graph should take up, say ? of the page size, with enough room under the graph for them to write down their conclusions. The second graph should be used to display the Q10 data. The data can be presented in graph form, or the student may choose to display it in histogram form. Either way is acceptable.

This is Figure 2. Write out the conclusions of the data below the graph. The water bath temperature intervals (in 100 C differences) represent the independent variables, and will be graphed along the x-axis. Remember there are four of them. The numerical values of Q10 represent the dependent variables, and are graphed along the y-axis.

Sarah, please insert the 2nd graph at this point, labeled Figure 2, same instructions as above. V. Application and Conclusions 1. What is the hypothesis of this experiment? 2. Which water bath represented the control in this experiment and why? 3.

Was the hypothesis accepted or rejected? 4. Why or why not? VI. Instructor's Guide Have the students arrange into groups of three or four to conduct this experiment. Five students in a group are too many. Two is really too few.

The lab should already have the yeast available in packet form – 3 packets per lab. This is a lot of yeast – the reactions will proceed rapidly, as long as the water bath temperatures are constantly monitored. Usually, the gas production will be completed within 45 minutes of the start time. The sugar (molasses) solution should already be made up and kept in the refrigerator until the lab begins. At least 1 liter per lab of a 2.5% glucose solution is used.

Stir bars are highly desirable to keep the molasses from settling to the bottom of the beaker, and to swirl the yeast, once it is added. The lab instructor should add the yeast five to 10 minutes before the students are instructed to add the solution to the fermentation tubes. Each student group should use 5 fermentation tubes, and at least 1 100 ml. graduated cylinder. The groups should also have 1 10ml. graduated cylinder to measure very small amounts of gas production.

Red wax marking pencils should be provided. Five water baths are needed for this lab to work. One of them does not even need to be turned on, as it represents the control, room temperature. The other four need to be watched carefully. Once the temperatures are set properly, it is important to keep them that way.

If the temperature of any one of them starts shifting too much, it will skew the results. Answers to the questions: 1. the hypothesis should be something along these lines: If temperature of water bath rises, the rate of carbon dioxide production in yeast will also rise, up to a point. Yeast is a living eukaryotic cell, and contains enzymes, as the chemical reaction of alcoholic

fermentation is characteristic of it. It is possible that at the highest temperature, the chemical bonds of the enzymes might start breaking, and the enzymes would not work as well. Thus, at the highest temp.

, the rate may fall. Their graph should reflect that. The water bath, especially at the highest temp. , has to be maintained constantly. If that temp drops, even a little bit, for even as much as 5 minutes, it will skew these results.

2. room temp - no heat added 3. the hypothesis should be accepted. 4. b/c of effect of rising temperature on chem. Rxns, and presence of enzymes in yeast.