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EXERCIS

3

LABORATORY

Microscopy

INTRODUCTION

Originally the study of anatomy and physiology was based on macroscopic, or gross, observation. This study was limited by the **resolution** of the human eye, which is the ability to distinguish two objects as separate. With the invention and use of the compound microscope (microscopes increase the resolution), much greater detail was seen, and thus began the study of cells and tissues. **Light microscopy** involves the use of visible light and glass lenses to magnify and observe a specimen. These topics are covered in the Saladin text in chapter 1, "Major Themes of Anatomy and Physiology." This exercise involves the use of the compound microscope, how to examine prepared slides under the microscope, and how to make slides of fresh material for study.

OBJECTIVES

At the end of this exercise you should be able to

1. list the rules for proper microscope use;
2. name the parts of the microscope and their functions presented in this exercise;
3. demonstrate the proper use of the compound microscope;
4. place a slide on the microscope and observe the material, in focus, under all magnifications of the microscope;
5. calculate the total magnification of a microscope based on the eyepiece lenses used;
6. prepare a wet mount of an observation.

MATERIALS

Compound microscope  
Prepared slides with the following (not to be kept, as they are stained and contain acids)  
Glass microscope slides  
Coverslips  
Lens paper  
Kimwipes or other cleaning paper  
Lens cleaner  
Small dropper bottle of water  
1% methylene blue solution  
Toothpicks  
Histological slides of kidney, stomach, or liver  
Slide with silk threads

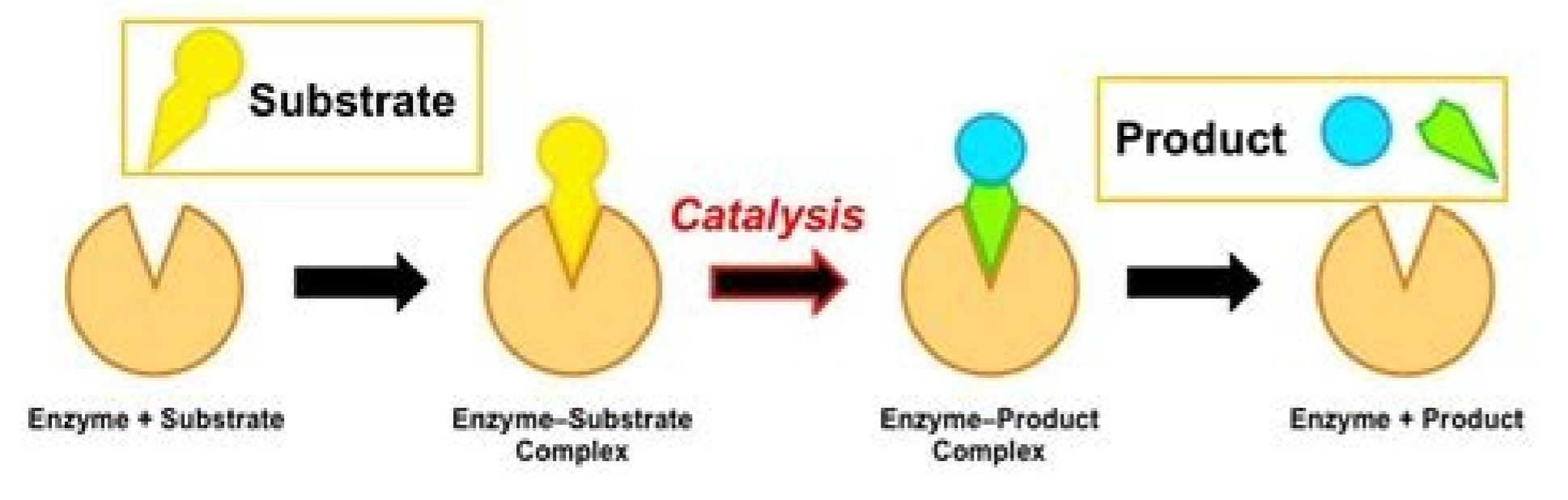
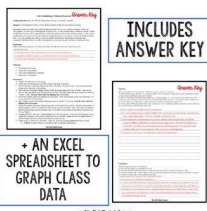
PROCEDURE

Microscopes are very expensive pieces of equipment, and you should always take great care handling them.

Care of the Microscope

There are a few rules concerning the care of a microscope.

1. When carrying the microscope, hold it securely with two hands—one hand under the base and one on the arm.
2. Keep the microscope upright at all times.
3. Keep microscope lenses clean with lens cleaner and softened lens paper. Do not use paper towels or your clothing.
4. Use only the fine-focus knob when using the high-power objective lens.
5. Remove slides from the microscope before putting it away.
6. Secure the cord with a rubber band or wrap the cord carefully around the base of the microscope.
7. Store the microscope with the low-power (scanning) objective lens in place.
8. Put the microscope away in its proper location.



Enzyme Activity

Background: Catalysts are substances that speed up and control chemical reactions (Crierie & Greig, 1999). An enzyme is an organic catalyst that whilst completing its function is not used up or changed.

The speed at which an enzyme-catalyzed reaction can be affected by a variety of factors such as temperature, pH level, substrate concentration, enzyme concentration or the presence of inhibitors (Worthington Biochemical Corporation, 2014) and co-factors (Crierie & Greig, 1999). Substrate concentration can increase the reaction rate of the enzymes until it reaches the saturation point, where all enzymes are busy and the reaction rate can therefore no longer increase. After this point, the rate of reaction plateaus (Macromolecules and Cells Themes Workbook, 2014). Substrates attach to the active site of an enzyme, enabling it to function. The more substrate that is available the quicker it will attach to the active site.

H<sub>2</sub>O<sub>2</sub>, referred to as hydrogen peroxide, acts as a substrate for the enzyme catalase, found in the liver (Macromolecules and Cells Themes Workbook, 2014). In this practical, the substrate concentration (hydrogen peroxide) is varied, and will therefore increase the reaction rate of the enzyme catalase (found in the liver). During this reaction, hydrogen peroxide will decompose into water and oxygen. The oxygen can be measured by the volume of foam produced, indicating the reaction rate of the enzyme (Macromolecules and Cells Themes Workbook, 2014). This reaction is easily explained by the following equation:

$$2\text{H}_2\text{O}_2 \xrightarrow{\text{catalase}} 2\text{H}_2\text{O} + \text{O}_2$$

Aim: To investigate the effect of the substrate concentration on the reaction rate of the enzyme catalase.

Hypothesis: If the concentration of the substrate H<sub>2</sub>O<sub>2</sub> is increased, then the reaction rate of the catalase will also increase until the saturation point is reached.

Independent variable: The concentration of the hydrogen peroxide.  
Dependent variable: The volume of foam produced after 2 minutes which is an indicator of enzyme activity.

Control variables:

- Amount of substrate (9mL)
- Size of the liver pieces
- Amount of detergent (2 drops)
- Use of same enzyme (catalase)
- Use of same animal's liver
- Same measuring cylinders, beakers and pipettes used for each sample
- Same room, therefore same temperature/climate, light and air pressure
- Completed all at once, at the same time
- Same people conducting the experiment

Safety assessment: This experiment required the use of the corrosive substance hydrogen peroxide, as well as breakable glassware. Subsequently, gloves, laboratory coats, enclosed shoes and safety glasses were worn. Care was taken when disposing of the chemical and water was provided for

This is an biology lab report example about enzyme concentration and the activity of catalase. This biology lab report example can be used in order to figure out how to write a lab report for biology courses. The headings of the biology lab report example are given below. Aim To investigate the effect of enzyme concentration on the activity of catalase. Research question How does the concentration of catalase where Saccharomyces cerevisiae is the source of enzyme affects the rate of the process of the break down of hydrogen peroxide will be measured by Vernier oxygen gas sensor where the values of temperature, pH, medium, substrate concentration are kept constant? As the enzyme concentration is increased, the activity of catalase will increase until a point and then it will stay the same. Introduction Chemical reactions can be speeded up by a few ways, one of which is the use of catalysts. A catalyst is a substance that speeds up a reaction by decreasing the activation energy without being affected by the reaction or being used up. Enzymes are biological catalysts that speed up reactions. A Substrate is a substance on which Enzymes act. Enzymes are specific for particular substrates. Enzymes work best when some conditions such as pH, temperature and ion concentration are at optimum values. Every enzyme has a specific optimum value that it works. If the optimum values are not provided, the reaction will proceed in a low rate or there will be no reaction at all. Hydrogen Peroxide (H2O2) is a reactive chemical that is formed as a by-product in cellular reactions. It must be removed to prevent it from disturbing chemical reactions in the cell. Catalase is an enzyme that breaks down Hydrogen Peroxide to water and oxygen, which are harmless, and thus Hydrogen Peroxide is a substrate for catalase enzyme. Hydrogen Peroxide is broken down by catalase as in the following equation: 2(H2O2) → 2H2O + O2 The enzyme, Catalase can be obtained from assaccharomyces cerevisiae and in this experiment Catalase will be obtained from assaccharomyces cerevisiae. Oxygen is a product of the break down of Hydrogen Peroxide and in this experiment, the rate of reaction will be calculated by measuring the Oxygen given off with a Vernier Oxygen Probe. 0.5, 0.75, 1.00, 1.25, 1.50, 1.75, 2.00, 2.25, 2.50 grams of yeast are measured with a digital balance. Each of the different amounts of yeast are poured into different beakers and the beakers are named from 1 to 9 with respect to the amount of yeast it contains. Take a syringe and fill the syringe with a 10% Hydrogen Peroxide solution. Yeast is put into the special flask of the Vernier gas sensor, which has a neck that fits with the gas sensor so that there is no gas escaping from the flask when the reaction occurs. Add the Hydrogen Peroxide solution into the flask that contains yeast. After adding the Hydrogen Peroxide solution, place the vernier gas sensor so that no oxygen gas escapes. Repeat the experiment with all the different amounts yeast and take three trials for each amount of yeast. Record your data in a table. Calculate the rate of Oxygen production for each trial and plot the graph of your data. By making use of the amount of O2 gas produced with every amount of yeast used, it is possible to deduce the affect of enzyme concentration on the activity of catalase. Increasing amount of oxygen gas given off means increasing activity of catalase. Three trials are taken for each sample of yeast. Amount of yeast used (g) (±0.01 g) Amount of oxygen gas given off (mL) (± 0.10 mL) Trial 1 Trial 2 Trial 3 0.50 12.50 12.55 12.55 0.75 14.20 14.10 14.20 1.00 15.50 15.55 15.55 1.25 17.00 17.20 16.80 1.50 17.50 17.40 18.60 1.75 18.70 18.90 19.10 2.00 19.20 19.70 20.20 2.25 20.20 20.60 19.40 2.50 21.40 20.60 21.40 Table 1: Shows the relationship between the amount of yeast used and the amount of oxygen gas given off. As a step to make a conclusion about the affect of enzyme concentration on the activity of catalase an interrelation should be found between the enzyme concentration and the amount of oxygen gas given off. The way that will be used in this experiment to find the interrelation is plotting a graph. For each sample of yeast, three trials are taken to make the experiment more accurate. To plot the graph of amount of yeast used versus the amount of oxygen gas given off, the arithmetic mean values for each sample should be calculated. Arithmetic mean is the central tendency of a collection of numbers taken as the sum of the numbers divided by the size of the collection. 1. Assuming a set of n values from 1 to n, shown mathematically as A = {a 1, a 2, a 3, ..., a n} , the general formula for arithmetic mean is: Formula 1: The general formula of arithmetic mean: ((Value of 1<sup>st</sup> element) + (Value of 2<sup>nd</sup> element) + ... + (Value of n<sup>th</sup> element))/n For the experiment, there are three trials and thus 3 values. To find the Arithmetic mean of these 3 values the following formula can be generated and used. Formula 2: The general formula for the arithmetic mean of the trials for each yeast sample used in this experiment. ((Value of 1<sup>st</sup> trial) + (Value of 2<sup>nd</sup> trial) + (Value of 3<sup>rd</sup> trial))/3 Finding the arithmetic mean of the first sample of yeast: The general formula is generated, as in Formula 2, so the arithmetic mean of the three trials in the first sample can be found by inserting the values into the formula. (12.50 + 12.55 + 12.55)/3 = 12.53 The arithmetic mean of the oxygen gas given off for each sample can be calculated by following the same way. Amount of yeast (g) (±0.01 g) Mean volume of Oxygen gas given off (mL) (± 0.10 mL) 0.50 12.53 0.75 14.17 1.00 15.53 1.25 17.00 1.50 17.83 1.75 18.90 2.00 19.70 2.25 20.07 2.50 21.13 Table 2: Shows the mean volume of Oxygen gas given off for each sample amount of yeast used. Calculating average percentage uncertainty To find the average percentage uncertainty, first of all, it is needed to find the percentage uncertainties for the mean volume of Oxygen gas given off for each sample. The general formula for percentage uncertainty is: Formula 3: General formula of percentage uncertainty Percentage uncertainty = Uncertainty/(value (mean)) × 100 Formula 3 can be specialized for this

experiment as below: Percentage uncertainty=0.10/(Average volume of O<sub>2</sub> given off)×100 Formula 4 Percentage uncertainty of the average volume of O<sub>2</sub> given off with the first sample can be found by using Formula 4: Following the same path, the percentage uncertainties for the other samples can be found. When Formula 4 is applied to all the samples, the values in the following table can be found as the uncertainties of the samples. Amount of yeast (g)±0.01 g Percentage Uncertainty (%) 0.50 0.80 0.75 0.71 1.00 0.64 1.25 0.59 1.50 0.56 1.75 0.53 2.00 0.51 2.25 0.50 2.50 0.47 The percentage uncertainties for all samples are found, thus the average percentage uncertainty can be found by using Formula 1. There nine values and according the Arithmetic mean and thus Formula 1, the sum of the all values of percentage uncertainties should be divided by 9. As found in the above equation, the average percentage uncertainty for this experiment is 0.59%. Aspect 3: Presenting Processed Data To find the interrelation between the amount of yeast used and the volume oxygen produced, it is a good way to draw a graph. The graph can be plotted by using technology. By using a computer software called Graphical Analysis, the graph can be plotted. For the graph, the data in Table 2, which contains the mass of yeast used and volume of oxygen produced, should be used. As found in the earlier steps, the average percentage uncertainty is 0.59%. By using percentage uncertainty, error bars can be added to the graph. Graph 1: Graph of the relation between the amount of yeast and the volume of oxygen gas produced. Conclusion And Evaluation Aspect 1: Concluding By checking over Graph 1, it can be seen that the slope of the graph decreases as the mass of the yeast used increases. Thus, the results and the graph supports my hypothesis. Aspect 2: Evaluating Procedure The percentage uncertainty of the experiment is 0.59% and the results support my hypothesis but there can be some errors that affect the accuracy of the experiment. After doing the experiment, I realized some points that can make my results inaccurate. In total, I made 27 trials but in the laboratory, there were two different packets of yeasts of different brands and I used yeasts from both of the packets. There can be differences between the yeasts in the two different packs. The second mistake I did is that, when starting the reaction I couldn't immediately place the vernier gas sensor on the flask so there can be some gas escaped from the flask while trying to place the gas sensor. In the research question, I stated that temperature is kept constant but as I was doing the experiment at the same time with my friends in the same laboratory so the temperature values could be different while taking different trials so my results can be inaccurate in this way. Aspect 3: Improving The Investigation To improve the investigation, some steps can be taken. First of all, the observer should be careful about using the same species of yeast. Secondly, the observer should use a different design to reduce the oxygen gas loss while placing the vernier oxygen gas sensor or pouring the yeast or H2O2 into the flask. If those steps are taken, the experiment will give more accurate results. References < . 30.10.2012 Abstract The purpose of this experiment was to record catalase enzyme activity with different temperatures and substrate concentrations. It was hypothesized that, until all active sites were bound, as the substrate concentration increased, the reaction rate would increase. The first experiment consisted of five different substrate concentrations, 0.8%, 0.4%, 0.2%, 0.1%, and 0% H2O2. The second experiment was completed using 0.8% substrate concentration and four different temperatures of enzymes ranging from cold to boiled. It was hypothesized that as the temperature increased, the reaction rate would increase. This would occur until the enzyme was denatured. The results from the two experiments show that the more substrate concentration,...show more content...The null hypothesis for the first experiment was that substrate concentration would have no effect on the reaction rate. It was hypothesized that the reaction rate would increase with rising substrate concentrations, until all active sites were bound. The null hypothesis for the second experiment was that temperature would not have an effect on reaction rates. It was hypothesized that until the enzyme is denatured, as temperature increased, so would the reaction rate. Methods and materials The first experiment begun by filling a 600-ml beaker, almost to the top, with water. Next, a 10-ml graduated cylinder was filled to the top with water. Once water was added to the beaker and graduated cylinder, a thumb was placed over the top of the graduated cylinder. This would ensure that no water was let out and no bubbles were let into the graduated cylinder. Next, it was turned upside down and fully submerged into the beaker. Then, a U-shaped glass tube was attained. The short end of the glass tube was placed into the beaker with the tip inside of the graduated cylinder. Next, a 50-ml Erlenmeyer flask was received. After, 10-ml of substrate concentration and 10-ml of catalase/buffer solution were placed into the flask. A rubber stopper was then placed on the opening of the flask. After adding these, the flask was held at the neck and spun softly

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