Names:\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_ Hour:\_\_\_

**Enzyme Lab**

**DEFINITIONS: BACKGROUND INFORMATION**

*Many living tissues contain the enzyme* ***catalase****. This enzyme breaks down hydrogen peroxide (H2O2), which is a harmful by-product of the process of cellular respiration if it builds up in concentration in the cells. If we use tissue-containing catalase, we can measure the relative influence of varying several different factors on the activity of enzymes in living tissue.*

*In order to obtain energy and building blocks from food, the digestive system must break down proteins, fats and carbohydrates. In this process, specific enzymes catalyze hydrolysis reactions in which foods are broken down into monomers. In this lab, you will perform reactions involved in digestion of carbohydrates, lipids, and proteins and observe the results of these reactions.*

**OBJECTIVES:**

**1. Measure the effects of changes in temperature, pH, and enzyme concentration on reaction rates of an enzyme-catalyzed reaction in a controlled experiment.
2. Explain how environmental factors affect the rate of enzyme-catalyzed reactions.**

**INTRODUCTION:**

What would happen to your cells if they made a poisonous chemical? You might think that they would die. In fact, your cells are always making poisonous chemicals. They do not die because your cells use enzymes to break down these poisonous chemicals into harmless substances. Enzymes are proteins that speed up the rate of reactions that would otherwise happen more slowly. The enzyme is not altered by the reaction. You have hundreds of different enzymes in each of your cells.

Each of these enzymes is responsible for one particular reaction that occurs in the cell. In this lab, you will study an enzyme that is found in the cells of many living tissues. The name of the enzyme is **catalase** (KAT- uh-LAYSS); **it speeds up a reaction that breaks down hydrogen peroxide**, a toxic chemical, into 2 harmless substances--water and oxygen.
 **The reaction is: 2 H2O2 ----> 2 H2O + O2**

This reaction is important to cells because hydrogen peroxide (H2O2) is produced as a byproduct of many normal cellular reactions. If the cells did not break down the hydrogen peroxide, they would be poisoned and die. In this lab, you will study the catalase found in liver cells. You will be using chicken or beef liver. It might seem strange to use dead cells to study the function of enzymes. This is possible because when a cell dies, the enzymes remain intact and active for several weeks, as long as the tissue is kept refrigerated.

**🡪What are the substrate(s), product(s) and enzyme we are using in this lab (be sure to pick out all three!)?**

**MATERIALS:**

* 6 Test tubes and Test tube holder
* 10-ml Graduated cylinder
* 40 ml 3% Hydrogen peroxide solution (found in stores)
* Straight-edged razor blade
* Scissors and Forceps (tweezers)
* Thermometer
* Stirring rod
* pH paper
* Fresh liver, chicken meat, Apple, and Potato

 **Note: It is very important to record your observations as you go!**

 **PART A - Observe Normal Catalase Reaction**

1. Place 2 ml of the 3% hydrogen peroxide solution into a clean test tube.
2. Using forceps and scissors, cut a small piece of liver and add it to the test tube. Push it into the hydrogen peroxide with a stirring rod. Observe the bubbles.

 **🡪What gas is being released?**

Throughout this investigation you will estimate the rate of the reaction (how rapidly the solution bubbles) on a scale of 0-5 (0=no reaction, 1=slow...5= very fast). *Assume that the reaction in step 2 proceeded at a rate of "4"*.

A reaction that absorbs heat is **endothermic**; reaction that gives off heat is **exothermic**.

**🡪Now, feel the temperature of the test tube with your hand. Has it gotten warmer or colder?**

**🡪Is the reaction endothermic or exothermic?**

**Is Catalase Reusable?**

1. Pour off **just** the liquid into a second test tube. Assume the reaction is complete.

**🡪What is this liquid composed of?**

**🡪What do you think would happen if you added more liver to this liquid?**

1. Test this and record the reaction rate.

**🡪Explain your results (what is the liquid composed of?)**

2. Add another 2 ml of hydrogen peroxide to the liver remaining in the first test tube.

**🡪Based on your observations is catalase reusable? Why or why not?**

**Part B - What Tissues Contain Catalase?**

You will now test for the presence of catalase in tissues other than liver. Place 2 ml of hydrogen peroxide in each of 3 clean test tubes and then add each of the three test substances to the tubes.  As you add each test substance, record the reaction rate (0-5) for each tube. Remember 0=no reaction, 1=slow...5= very fast.

|  |  |  |
| --- | --- | --- |
| **Substance** | **Rate of Reaction (0-5)** | **Observation (What does it look like)** |
| **Potato** |  |  |
| **Apple** |  |  |
| **Chicken** |  |  |

**🡪Based on your observations, which tissues contained catalase?**

**🡪Do some contain more catalase than others? How can you tell?**

**PART C - What is the Effect of Temperature on Catalase Activity?**

1. Now you will follow the same basic procedure from part A (a piece of liver in about 4mL of peroxide. This time however you will be observing liver that has been boiled and frozen. Record your observations below.

|  |  |  |
| --- | --- | --- |
| **Substance** | **Rate of Reaction (0-5)** | **Observation (What does it look like)** |
| **Fresh Liver (From Part A)**  |  |  |
| **Boiled Liver** |  |  |
| **Frozen Liver** |  |  |

* **How was your observations different for the boiled and frozen liver?**
* **Why do you think you go the results you did? What happened to the enzymes?**