

## Enzyme Lab

### 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 which **breaks down hydrogen peroxide**, a toxic chemical, into **water and oxygen**- 2 harmless substances.

The reaction is as follows:  $2\text{H}_2\text{O}_2 \rightarrow 2\text{H}_2\text{O} + \text{O}_2$  (fix)

This reaction is important to cells because hydrogen peroxide ( $\text{H}_2\text{O}_2$ ) 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 potato cells. 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.

### OBJECTIVE:

To measure the effects of changes in temperature, pH, and enzyme concentration on reaction rates of an enzyme catalyzed reaction in a controlled experiment.

### MATERIALS:

Distilled water  
6 Test tubes and Test tube holder  
10-ml Graduated cylinder  
40 ml-3% Hydrogen peroxide solution (found in stores)  
Scissors and Forceps (tweezers)  
Thermometer  
Stirring rod  
pH paper  
Potato

## PART A - Observe Normal Catalase Reaction

1. Place 2 ml of the 3% hydrogen peroxide solution into a clean test tube.
2. Take a small piece of potato and add it to the test tube. Push it into the hydrogen peroxide with a stirring rod. Observe the bubbles;

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"

What gas is being released? \_\_\_\_\_

What liquid is formed from this reaction? \_\_\_\_\_

Write the reaction rate and your observations in the DATA TABLE for the normal reaction.

## Is Catalase Reusable?

1. Pour the used peroxide from the first test tube into a second test tube.
2. Add a piece of potato to this used peroxide.

What's the reaction rate? What did you observe? Record it in the DATA TABLE.

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

What is the reaction rate? What did you observe? Record it in the DATA TABLE

Is catalase reusable? \_\_\_\_\_ Explain your results

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## Part B - What is the Effect of Temperature on Catalase Activity?

1. Put a piece of potato into the bottom of a clean test tube and cover it with a small amount of distilled water. Place this test tube in a boiling water bath for 5 minutes.

What will boiling do to an enzyme? \_\_\_\_\_

2. Remove the test tube from the hot water bath, allow it to air cool, then pour out the water. Add 2 ml of hydrogen peroxide. **CAUTION:** Use a test-tube holder when handling the hot test tubes. Record the reaction rate (0-5) in DATA TABLE

3. Put a piece of potato into a clean test tube and 1 ml of H<sub>2</sub>O<sub>2</sub> into another test tube. Place both of these test tubes into the ice bath for 3 minutes.

4. After 3 minutes, pour the test tube of H<sub>2</sub>O<sub>2</sub> into the test tube of potato and observe. Record the reaction rate (0-5) in the DATA TABLE

5. Put 1 ml H<sub>2</sub>O<sub>2</sub> into a clean test tube. Put a piece of potato into another clean test tube. Place both these test tubes into warm water bath (not boiling) for 3 minutes.

4. After 3 minutes, pour the test tube of H<sub>2</sub>O<sub>2</sub> into the test tube of potato and observe the reaction. Record the reaction rates (0-5) in DATA TABLE

You recorded the reaction rate for room temperature earlier in the normal reaction in Part A.

What is the **optimum temperature** for catalase? (This is the temperature at which the reaction proceeds fastest.) \_\_\_\_\_

Why did the reaction proceed slowly at 0 degree C? \_\_\_\_\_

Why did the reaction not proceed at all at 100 deg. C? \_\_\_\_\_

### PART C - What is the Effect of pH on Catalase Activity

1. Add 2 ml hydrogen peroxide with an adjusted pH of 7 to test tube 1. Add 2 ml of hydrogen peroxide with an adjusted pH of 3 in test tube 2. Add 2 ml of hydrogen peroxide with an adjusted pH of 10 in test tube 3.

2. With each test tube add a small piece of potato and record the reaction rates.

**CAUTION:** Do not let acids or bases contact your skin or clothing.

Does there appear to be an **optimum pH** - at what pH? \_\_\_\_\_

What is the effect of low or high pH on enzyme activity? \_\_\_\_\_

**DATA TABLE**

		Rate of Reaction (1-5)	Observations & Conclusions (answer section questions)
PART A	Normal potato		
	Potato added to Used Peroxide		
	Reused Catalase		
PART B	Boiled potato		
	Ice Bath potato		
	Room temp potato		
PART C	Basic Solution (pH 10)		
	Acidic Solution (pH 3)		
	Neutral Solution (pH 7)		