Enzymes 2014***adapted from various labs – special credit to biologycorner.com*

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 the rate of reactions that would otherwise happen more slowly. The enzyme is not altered in the reaction. You have hundreds of different enzymes in your cells. Each of these is responsible for one particular reaction that occurs in the cell.

In this lab, you will study an enzyme that is found in 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 2 harmless substances: water & oxygen. The reaction is as follows:

$$2H_2O_2 \rightarrow 2H_2O + O_2$$

This reaction is important to cells because **hydrogen peroxide** (H_2O_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 & die.

In this lab, you will study the catalase found in liver cells. You will be using chicken or beef liver that your teacher purchased in the supermarket. It might seem strange to use dead cells to study the function of enzymes. This is possible because when a cell dies, the enzyme remains intact & active for several weeks, as long as the tissue is kept refrigerated. Recall that the substrate is the molecule that the enzyme acts on, and the products are the molecules produced by the reaction.

Answer the pre-lab questions before moving on to Part A:

Pre-lab Questions:

1. What is the name of the enzyme studied in this lab?_____
2. What chemical does the catalase break down? (In other words: what is the substrate?)_____
3. Catalase breaks the hydrogen peroxide into what two harmless substances? (In other words: what are the products?)

_____

Catalase Lab Question #1: Are enzymes reusable?

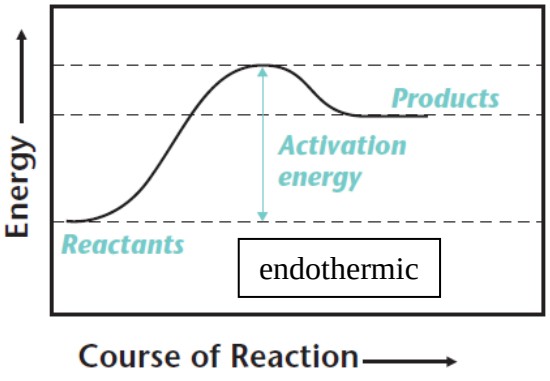
Procedure:

- Place 20 mL (about the height of your thumbnail) of 3% hydrogen peroxide solution into a clean test tube & add a small piece of liver.

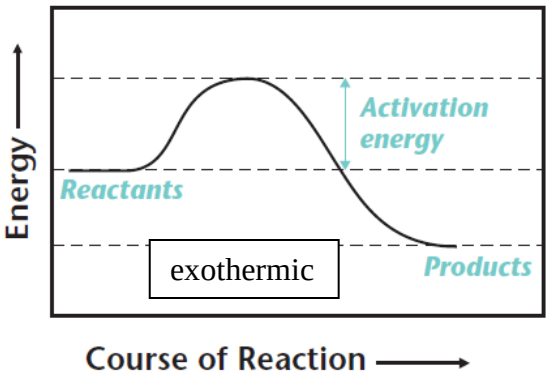
1. Observe the bubbles. What gas is being release? _____

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 that you just saw above proceeded at a rate of "4" .

Recall that a reaction that absorbs heat is endothermic (reactants have less energy; products have more which will feel hot); A reaction that gives off heat (reactants have more energy; products have less & feel cold) is exothermic.



Type of reaction: *energy-absorbing reaction*



Type of reaction: *energy-releasing reaction*

Now, feel the temperature of the test tube with your hand.

2. Has the tube gotten warmer or colder? _____ Is the reaction endothermic or exothermic? _____

- Pour off the liquid in the tube into the sink.

3. What was the liquid that you poured down the drain? _____

- Add another 20 mL of hydrogen peroxide to the liver remaining in the tube.

4. Was there another reaction? _____ What do you think would happen if you poured off this liquid & added more hydrogen peroxide to the remaining liver? _____
Why? _____

5. Are enzymes reusable? _____

Catalase Lab Questions #2: What Tissues Contain Catalase?

Catalase is present isn many kinds of living tissue. You will now test for the presence of catalase in tissues other than liver. Place 20 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.

Substance	Rate of Reaction (0-5)
Potato	
Apple or Carrot	
Chicken	

1. Which tissues contained catalase? _____

2. Do some contain more catalase than others? _____How can you tell? _____

Catalase Lab Question #3: How are enzymes in living tissue affected by temperature?

Under certain conditions enzymes are denatured. An enzyme is denatured when the protein molecule loses its proper shape & cannot function. Some things that can denature an enzyme are high temperatures, extremes of pH, heavy metals & alcohol.

➔ Put a piece of liver into the bottom of a clean test tube & cover it with a small amount of distilled water. Place this test tube in a boiling water bath (100^{o}C) for 5 minutes.

1. What will boiling do to an enzyme? _____

- Remove the test tube from the hot water bath, allow it to air cool, then pour out the water. Add 20 mL of hydrogen peroxide. CAUTION: use a test-tube holder when handling hot test tubes.

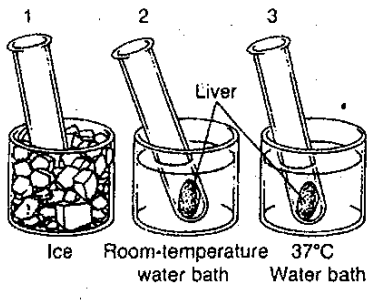
2. What is happening in the test tube? _____

- Record the reaction rate in the first column of the data chart (0 = no reaction → 5 = high reaction).

Data Table: Rates of Enzyme Reaction at Different Temperatures:

Temperature	100^{o}C (boiling)	37^{o}C (body temp)	22^{o}C (room temp)	0^{o}C (freezing)
Rate of Reaction (0→5)				

- Put equal quantities of liver into 3 clean test tubes and a 10 mL H_2O_2 into 3 other test tubes. Put one test tube of liver & one of H_2O_2 into each of the following water baths:
  - ✓ Ice bath (0^{o}C)
  - ✓ Room temperature bath (about 22^{o}C)
  - ✓ Warm water bath (37^{o}C) (human body temperature)
- After 3 minutes, pour each tube of H_2O_2 into the corresponding tube of liver & observe the reaction. Record the rates of reaction in the last three columns of the data chart (0→5).



3. What is the optimum temperature for catalase? (This is the temperature at which the reaction proceeds fastest). _____

4. Why did the reaction proceed slowly at 0^{o}C?_____

5. Why did the reaction not proceed at all at 100^{o}C? _____

6. Is rate of reaction of an enzyme affected by temperature? _____