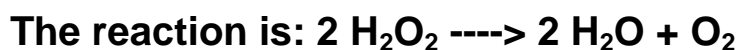


Enzyme Lab

INTRODUCTION:

What would happen to your cells if they made a poisonous chemical? You might think that they would die. However, your cells are always making poisonous chemicals as a waste product. Luckily, 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 enzyme in your body 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 2 harmless substances--water and oxygen.



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.

MATERIALS:

4 Test tubes with rack
10-ml Graduated cylinder
Test tube holders

Scissors and Forceps (tweezers)
Fresh liver, Apple, and Potato
3% Hydrogen peroxide solution
(found in stores)

Ice bath
Warm water bath
Boiling water bath

PART 1 - Observe Normal Catalase Reaction

(make sure to answer all ► questions)

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.

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? (consider the equation above) _____*

3. Recall that a reaction that absorbs heat is endothermic; a 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?*

4. Pour off the liquid into a second test tube.

Name: _____

Per: ____ TOC: _____

▶ Assuming the reaction is complete. What is this liquid composed of? _____

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

▶ Test this and record the reaction rate. Reaction Rate _____ (1 – 5)

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

▶ What is the reaction rate? _____

▶ Is the *catalase* reusable? Explain how you know.

Part 2 - What Tissues Contain Catalase

You will now test for the presence of catalase in tissues other than liver.

1. 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.
2. As you add each test substance, record the reaction rate (0-5) for each tube.

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

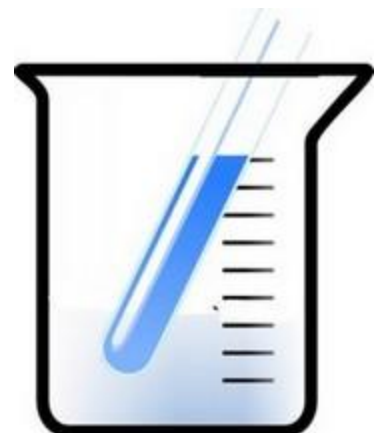
▶ Based on your observations, which tissues contained catalase?

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

PART 3 - What is the Effect of Temperature on Catalase Activity?

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

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.



Name: _____

Per: ____ TOC: ____

CAUTION: Use a test-tube holder for hot test tubes.

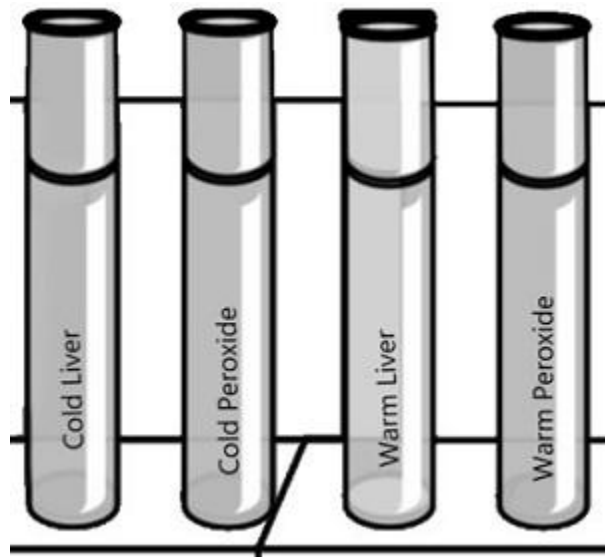
▶ What is the reaction rate for the boiled liver and peroxide? _____

3. Put equal quantities of liver into 2 clean test tubes and 1 ml H_2O_2 into 2 other test tubes. Put one test tube of liver and one of H_2O_2 into an ice bath. Place the other set in a warm water bath (not boiling).

After 3 minutes, pour each tube of H_2O_2 into the corresponding tube of liver and observe the reaction.

▶ What is the reaction rate for the cold liver/peroxide?

▶ What is the reaction rate for the warm liver/peroxide?



▶ Compare and contrast the reactions rates of temperature to the room temperature test you did in part 1.

Name: _____

Per: _____ TOC: _____

Part 4 - Design an Experiment

Using the techniques you learned in this lab, design a **new** experiment to test the properties of enzymes and substrates. Consider tests that may look at different properties or environments of enzymes (i.e. we looked at temperature, what else could you change?)

DATA ANALYSIS

1. Describe the relationship between catalase and hydrogen peroxide. Indicate which is the enzyme, which is the substrate and what occurs during the reaction. It may be helpful to write the equation.

2. Is catalase reusable? Use your **data** to support your answer.

3. Amylase is an enzyme that can be used to break down carbohydrates, like those found in bread and crackers to individual subunits of sugars. This is why bread begins to taste sweet as we chew it, amylase is found in saliva. Benedict's solution is a chemical that changes color in the presence of sugar. In water, Benedict's is a blue color, but it will turn orange in the presence of glucose.

How would you design an experiment to test the reaction rate of amylase in breaking down starch into sugars?