Name _____ Date _____

Effects of Temperature on Enzymes

Purpose

The determine the effects of temperature and pH on enzymes

Background

Animal cells produce hydrogen peroxide (H_2O_2) as a byproduct of cellular respiration. Hydrogen peroxide is a toxic to the cells. Fortunately, cells also have organelles called peroxisomes that contain an enzyme called catalase that is able to break down hydrogen peroxide into water (H_2O) and oxygen (O_2).

Enzymes are special proteins that help speed up all biological reactions. There are hundreds of different types of enzymes and each enzyme is very specific as to what reactions they work with. The reason they are so specific is their shape. Only certain substrates (reactants in an enzyme reaction) can fit into the active site of an enzyme to complete the enzyme-substrate complex.

Temperature and pH can alter the conditions in the cell which causes enzymes to change their shape. When the shape of an enzyme is changed, it is said to be denatured. Denatured proteins no longer fit their specific substrates and so that reaction can no longer take place. This is why maintaining a stable body temperature and pH level is so important to our health.

Water boils at 100^0C. Water freezes at 0^0C. Room temperature is about 25^0C.

Problem

How do different temperatures affect the rate at which the catalase enzyme breaks down hydrogen peroxide?

Materials

6 Test Tubes	Test Tube Rack	Test Tube Holder	1 Plastic Cup
2 Styrofoam cups	Funnel	Graduated Cylinder	Ice
Hydrogen Peroxide	Beef Liver	Water	Hot Plates

Procedure

Preparation for Testing the Effects of Temperature on Enzyme Activity

Water Baths

1. (Hot Bath) Fill the large glass beaker up about half-way and place it on the hot plate. Turn the hot plate on and let the water come to a boil.
2. (Ice bath) Place ice in one of the Styrofoam cups.
3. The room temperature one doesn't need a water bath.

Hydrogen Peroxide

4. Place the funnel inside the graduated cylinder.
5. Pour exactly 10 ml of hydrogen peroxide into the graduated cylinder.
6. Move the funnel to one of the test tubes. Pour the hydrogen peroxide into the test tube.
7. Repeat this step two more times, being sure that at least one of the numbered test tubes contains hydrogen peroxide.

Liver

8. Rinse the funnel really well, in the sink.
9. Place the funnel inside one of the empty test tubes.
10. Pour the liver homogenate into the test until it is about 1/3 full.
11. Repeat this step two more times being sure that the amount of liver in each test tube is as close the same amount as possible.
12. Rinse the funnel really well, in the sink.

Performing the Experiment

1. Place one of the liver test tubes and one of the hydrogen peroxide test tubes into the ice cup and place it in the freezer, record the time so you can remove them after 10 minutes.
2. Place the numbered liver test tube and the numbered hydrogen peroxide into the hot water bath, record the time so you can remove them after 10 minutes.
3. While you are waiting, have someone in your group start a timer then pour the room temperature hydrogen peroxide into the test tube with the room temperature liver. Stop the timer after the bubbles have stopped forming. Record the amount of time it took for the reaction to take place in the data table.
4. Rate the amount of bubbles on a scale form 0 – 5 with 5 being a whole lot of bubbles to 0 being no bubbles. (You may need to adjust your scale as you see how reaction occurs at different temperatures.)
5. When the 10 minutes are up, remove the hydrogen peroxide and liver from the ice bath and set them in the test tube rack. Have someone start the timer then pour the hydrogen peroxide into the liver and rate the bubbles. Record the time it took for the reaction to take place in the data table.
6. When the 10 minutes are up, remove the hydrogen peroxide from the hot water bath and set them in the Styrofoam cup, in order to carry them back to your table. Set both test tubes in the rack. Have some start the timer then pour the hydrogen peroxide into the liver and rate the bubbles. Record the time it took for the reaction to take place in the data table.

Are Enzymes Reusable?

1. Pour 10ml of hydrogen peroxide into an empty test tube that previously held hydrogen peroxide.
2. Decide which of your earlier test tubes had the greatest reaction. (the liver and hydrogen peroxide from the earlier reaction should still be in the test tube)
3. Have someone start the timer then pour the hydrogen peroxide into the test tube that had the greatest reaction. Rate the bubbles and record the time it took for the reaction to take place.
4. Decide if the enzyme was still able to carry out the reaction and record your results in the data table.

Clean Up

1. Use the wooden stick to remove all the liver homogenate from the test tubes into the trash cans.
2. Carefully use the test tube brush to clean each test tube, including the hydrogen peroxides ones, in soapy water.
3. Rinse each test tube three times and place upside down in your rack to dry.
4. Wash and rinse the cups and all other materials and set them out to dry.

Data Table

Temperature	Amount of Reaction	Time
O^0C (Freezing)		
25^0C (Room Temp)		
100^0C (Boiling)		
Reusable Test		

Graph Your Data

1. Use the graph paper to create a bar graph for the tests with the different temperatures.
2. Be sure to include a title for your graph and a label for each axis.

Analysis and Conclusion

1. Explain what enzymes are, such as the type of molecules they are, and what enzymes do.

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_____
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_____

2. Explain what the basic reaction was in the lab using the terms: *enzyme, substrate, products, hydrogen peroxide, catalase, water, and oxygen.*

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3. Discuss the effects of temperature on enzymes using the results of your lab to support your claims.

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4. What is the optimal temperature for the enzyme catalase?

_____

5. Which had a greater denaturing effect on the enzyme catalase, hot or cold temperatures?

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6. Keeping the lab in mind, how do you think our body increasing its temperature, otherwise known as a fever, helps us fight off bacterial infections?

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7. Use the information gained in this lab to explain why Jack, in the movie *Titanic*, died.

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