Enzymes in Action

By Heather Miller
Based on The Catalase Enzyme Lab
by Bryan Andriese at Yucaipa High School

Focus on Inquiry

The student will predict, investigate, observe, and report on the effects that pH, concentration, and temperature have on catalase enzyme reactions.

Lesson Content Overview

Students will conduct an experiment in which they will alter the pH, concentration, and temperature of the environment in which catalase enzyme reactions are taking place. Students will be able to describe how changes in these environmental conditions affect the action of the enzymes in living things.

Duration	Setting	Grouping	PTI Inquiry Subskills
100 minutes	classroom	2-4 per group	1.3, 2.1, 2.2, 2.5, 3.1, 3.2, 3.3, 3.5, 3.7, 4.3, 5.2, 5.7, 5.8, 5.9, 7.2

Lesson Components	Estimated Time	Inquiry Subskills Used	Technology Used	Level of Student Engagement	Brief Description
Engage	10 min	1.3, 5.2	none	3	Students will observe a demonstration of a catalase reaction between liver and hydrogen peroxide.
Explore	15 min	1.3, 4.3, 5.8, 5.9	none	3	Students will experiment with different enzymes and substrates to understand the structure and function of enzymes.
Explain	10 min	5.2, 5.7	none	2	Students will describe their understanding of how enzymes and substrates interact and how enzymes affect activation energy.
Expand/Elaborate	50 min	2.1, 2.2, 2.5, 3.1, 3.2, 3.3, 3.5, 3.7, 7.2	none	3	Students will experiment with altering the pH, concentration, and temperature of the catalase reaction. Students will explain their understanding of how each factor affected the reaction rate.
Evaluate	15 min	5.7	none	1	Students will complete a summative assessment on enzymes.

Level of Student Engagement

1	Low	Listen to lecture, observe the teacher, individual reading, teacher demonstration, teacher-centered instruction
2	Moderate	Raise questions, lecture with discussion, record data, make predictions, technology interaction with assistance
3	High	Hands-on activity or inquiry; critique others, draw conclusions, make connections, problem-solve, student-centered

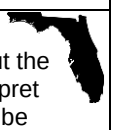
Next Generation Science Standards – Inquiry

NGSS Practice 1: Asking Questions and Defining Problems
NGSS Practice 2: Developing and Using Models
NGSS Practice 3: Planning and Carrying Out Investigations
NGSS Practice 4: Analyzing and Interpreting Data
NGSS Practice 6: Constructing Explanations
NGSS Practice 8: Obtaining, Evaluating and Communicating Information

Next Generation Science Standards – Life & Physical Science

HS-LS1-1.: Construct an explanation based on evidence for how the structure of DNA determines the structure of proteins which carry out the essential functions of life through systems of specialized cells.
HS-PS1-5.: Apply scientific principles and evidence to provide an explanation about the effects of changing the temperature or concentration of the reacting particles on the rate at which a reaction occurs.

Florida Science Standards – Nature of Science



SC.912.N.1.1: Define a problem based on a specific body of knowledge (biology)and pose questions about the natural world, conduct systematic observations, plan investigations, use tools to gather, analyze, and interpret data, pose answers, explanations, or descriptions of events, generate explanations that explicate or describe

natural phenomena (inferences), and communicate results of scientific investigations.

Florida Science Standards – Life Science

SC.912.L.18.11: Explain the role of enzymes as catalysts that lower the activation energy of biochemical reactions. Identify factors, such as pH and temperature, and their effect on enzyme activity.

Materials and Advance Preparation

Materials List

Class set:

- 500 ml of 1M Hydrochloric acid (HCl)
- 500 ml of 3% Hydrogen peroxide
- 500 ml of 1M Sodium hydroxide (NaOH)
- Graduated cylinders (10 ml)
- Hot Plate(s)
- Hot water bath *(directions in advanced prep)* in 250-500 ml beaker
- Ice water batch *(directions in advanced prep)* in 250-500 ml beaker
- Matches or lighter
- pH paper
- Potato cubes
- Purified water
- Small amount of chicken liver (raw, ground or cut into 1 cm cubes)
- Test tube cleaners
- Waste container
- Wooden splints

Group materials:

- 1 apron or lab coat for each student
- 1 mortar & pestle
- 1 pair of goggles for each student
- 1 stir rod
- 1 test tube clamp
- 1 test tube rack
- 1 tweezer
- 3 test tubes
- Access to hydrogen peroxide, sodium hydroxide, hydrochloric acid, pH indicator papers, graduated cylinders, hot water & ice water baths, and waste/cleaning area
- Demonstration Reflection Sheet (1 per student)
- Student Lab Sheets (1 per student)
- Enzyme Puzzle (1 per group)
- Enzyme Puzzle Reflection (1 per student)
- Enzyme Assessments (1 per student)

Blackline Masters

1. **Blackline Master #1:** Demonstration Reflection Sheet
2. **Blackline Master #2:** Enzymes Puzzle
3. **Blackline Master #3:** Enzymes Puzzle Reflection
4. **Blackline Master #4:** Enzyme Lab Directions
5. **Blackline Master #5:** Enzymes Student Lab Sheet
6. **Blackline Master #6:** Enzymes Check for Understanding

Advance Preparation

1. Print the Enzymes Puzzle (1 per group) preferably on cardstock but regular printer paper will also work. Cut out all of the enzyme pieces and place them in an envelope.
2. Obtain all materials and set up a station for students to have access to hydrogen peroxide, sodium hydroxide, hydrochloric acid, pH indicator papers, graduated cylinders, hot water bath, and waste/cleaning area.
3. Prepare the hot water bath by filling a beaker mostly full of water and bringing it to a boil. Once boiling has been reached, reduce heat to low-medium heat to maintain high water temperature.
4. Prepare the ice water bath by filling a beaker mostly full of water and adding ice to fill the beaker. Add ice as necessary throughout the day/period as the ice melts.
5. Set up lab group materials: 1 test tube rack, 3 test tubes, 1 mortar & pestle, 1 test tube clamp, 1 stir rod, 1 pair of goggles for each student, 1 apron or lab coat for each student
6. Chop up small pieces of chicken liver (1cm cubes). Obtain wood splints and matches or a lighter.
7. Set up a test tube rack, 3 test tubes labeled 1-3, hydrogen peroxide, and the wood splints with lighter or matches for the whole class demonstration.
8. Make copies of the student lab sheets (1 per student), the enzymes puzzle reflection (1 per student), the demonstration reflection sheet (1 per student) and the enzymes assessment (1 per student). The demonstration reflection sheet and the puzzle reflection sheet could be printed back to back to save paper.

Lesson Information

Learning Objectives

The student will be able to:

1. Describe the relationship between enzymes and substrates.
2. Explain how enzymes affect the activation energy needed to carry out a chemical reaction.
3. Carry out an investigation exploring how pH, concentration, and temperature affect enzymatic reactions.
4. Describe how pH, concentration, and temperature affect enzymatic reactions.

Prior Knowledge Needed by the Students

- Students should have general background information about what pH and concentration are. A basic understanding of what an enzyme is would be helpful, but is not necessary. Students should also have a general understanding of how temperature can speed up or slow down molecule movement.

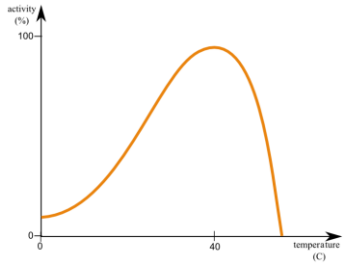
Background Information

Enzymes are proteins that act as catalysts and some help biochemical reactions occur at a rapid rate. Enzymes help us break down our food, for example. Enzymes are very specific catalysts and usually work to complete one task. For example, an enzyme that helps digest proteins like in meat will not be useful to break down carbohydrates like in potatoes. Also, you will not find all enzymes everywhere in the body. There are unique enzymes in all of the different areas of your body that carry out very specific tasks.

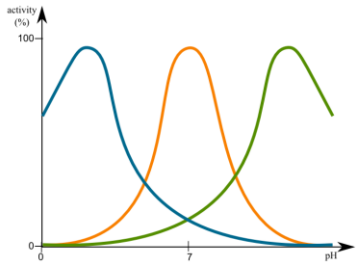
The 2 basic players in enzymatic reactions are enzymes and substrates. The substrates can only fit into the active sites of very specific enzymes, so not all enzymes can synthesize or decompose all substrates. The enzyme attaches to the substrate at an area called the active site. The combination is called the enzyme/substrate complex. When the enzyme releases the substrate, it returns to its original shape. It is then ready to synthesize or decompose another molecule of substrate.

There are several factors that can regulate enzyme activity, including temperature, concentration, and pH levels. Some enzymatic reactions are enhanced by higher temperatures while others are not. Generally speaking, increasing the temperature will speed up the reaction until the temperature gets to a certain point. At that point, the reaction will begin to slow down again and the enzyme will become “denatured” where by it will begin to lose its shape and function. The pH (acidity or basicity) of the environment changes the shape of proteins in the same way that temperature does. Certain enzymes work best at certain pH levels and each enzyme is different. Once again, if the pH gets too high or too low for the specific enzyme, the enzyme can become denatured. The concentration of enzyme or substrate

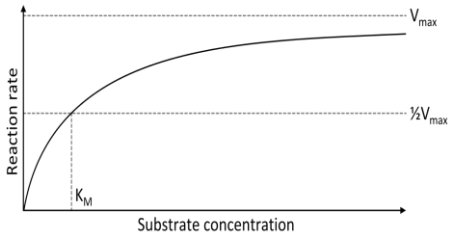
will also determine how fast or slow the reaction takes place. Up to a certain point when there are too many enzymes or substrates, you will see an increase in reaction rate as the concentration increases. Once there are the same number of enzymes and substrates, adding additional enzymes or substrates will not increase the speed of the reaction because they will have nothing available to connect with. The graphs below show the general trends of how temperature, pH, and concentration affect enzyme activity.



Temperature



pH



Concentration

Photo Credit: https://commons.wikimedia.org/wiki/File:Enzyme-temperature.png
Photo Credit: https://commons.wikimedia.org/wiki/File:Enzyme-ph.png
Photo Credit: https://commons.wikimedia.org/wiki/File:Michaelis_Menten_curve_2.svg

Lesson Procedure

Engage

1. **PLEASE READ THE DIRECTIONS FOR THIS ENGAGE ACTIVITY THOROUGHLY AND TRY IT BEFORE DEMONSTRATING IT IN FRONT OF YOUR STUDENTS.**
2. The teacher will complete a demonstration in which he/she will expose raw chicken liver to hydrogen peroxide to demonstrate enzyme action in the liver. The enzymatic reaction will break down the hydrogen peroxide to release oxygen. The presence of oxygen will be observed by the reigniting of a wooden splint.
   - First, place 3 test tubes in a test tube rack labeled 1, 2, and 3.
   - Next, using tweezers, place one 1cm^3 piece (does not have to be exact, just has to be small) of fresh chicken liver into test tubes 1 and 3 *(chicken liver should already be cubed as described in advanced preparation).*
   - Pour 5 ml of hydrogen peroxide into test tube 2.
   - At this time, pass out the demonstration Reflection Sheet (Blackline master #1) one to each student.
   - Test tube 1 and 2 will be your control test tubes. They will demonstrate to the students that there is no significant amount of oxygen being released from the chicken liver nor the hydrogen peroxide by themselves.
   - Show and explain to the students what is in test tubes 1, 2, and 3.
   - Ask students to predict what will happen when:
     - Hydrogen peroxide is added to the chicken liver in test tube 3.
     - Each test tube is tested for oxygen by introducing them to fire.
       - *Students should also explain **WHY** they think these things will happen.*
   - Give students 2-3 minutes to think and make their predictions. Have students write down their response on their Demonstration Reflection Sheet.
   - Next, have students share their ideas around their group by starting with one member of the group (to be determined by the teacher) and then moving around the group in clockwise fashion, each student sharing their two predictions.
   - When students are finished sharing, pour 5 ml of hydrogen peroxide into test tube 3 and give students time to record their observations on their Demonstration Reflection Sheet.
   - Test for the presence of oxygen by lighting the wooden splint, allowing it to burn for several seconds, and then shaking or blowing it out until the flame is extinguished with the end still glowing.

- Lower the splint into test tube 1 (be careful not to let it touch the sides of the test tube as they may be wet). Allow students to record their observations on their Demonstration Reflection Sheet. *Nothing should happen.*
- Relight the splint, extinguish the flame, and repeat the oxygen test with test tube 2. *Nothing should happen.*
- Relight the splint, extinguish the flame, and repeat the oxygen test with test tube 3. *You should experience a popping sound and the relighting of the wooden splint.*
- The splint should reignite when introduced into test tube 3 as there is a high oxygen concentration.
- Ask students to try to explain their observations.

3. The teacher will now explain that there are special proteins at work in the chicken to make what they saw possible and they are now going to learn more about those special proteins called enzymes.

Explore

1. Pass out the puzzle piece envelopes to the groups and direct them to try to find which pieces fit together like a puzzle.
2. When a group appears as though they have successfully matched all the pieces, pass out the Enzyme Puzzle Reflection Sheet (unless students already have it on the back of their Demonstration Reflection Sheet) for students to complete.
3. Questions you might ask the students while they're working might include:
    a. Have you tried putting the small pieces into some of the other large pieces? What happened? *Student responses may vary but could include yes, no, the pieces only fit in certain spots, I was able to fit some other pieces but not well.*
    b. What do the different pieces represent? *Student responses should include that the larger pieces are the enzymes and the smaller pieces are the substrates.*
    c. What could you do to put this puzzle together faster? *Student responses may vary but could include having other people help, drinking a Red Bull, move really fast, make the pieces bigger, etc.*
    d. What do you think is happening to the substrate(s) when they are connected with the enzyme? *Student responses may vary but could include it's making something new, it's breaking something apart, and/or it's activating something.*

Explain

1. The questions asked on the Enzyme Puzzle Reflection Sheet include:
    - In the puzzle, the large piece of the puzzle represents the "**enzyme**," the area where the enzyme and the pieces meet is called the "**Active site**" and the little pieces that connect to the active site are called "**substrates**." Draw a labeled diagram that shows how the enzyme and substrate interact. *Students' diagrams should look something like this:*

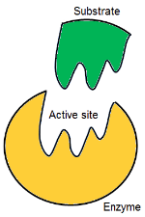


Photo credit: https://commons.wikimedia.org/wiki/File:Lock_and_key.png

    - Were you able to fit multiple different substrates into multiple different active sites? Why or why not? *Student responses will vary but should reflect that they were only able to make one specific substrate fit into the active site of one specific enzyme.*
    - Based on your response to question 2, what conclusion can be drawn about the relationship between enzymes and substrates? *Student responses may vary but should include something explaining that there is only one substrate that each enzyme is responsible for reacting with and that the enzyme-substrate relationship is very specific like a "lock & key" system.*

- How might this puzzle activity be related to the demonstration you saw with the liver and hydrogen peroxide? *Student responses may vary but should indicate that there are enzymes in the liver (catalase) that connect with the hydrogen peroxide (the substrate) to break it down into water and oxygen. The puzzle pieces could be the enzyme (catalase) from the liver and the substrate (hydrogen peroxide).*
- "Denaturing" of an enzyme occurs when the shape of the enzyme is altered due to changes in temperature and pH. How could you alter your puzzle pieces to show the denaturing of an enzyme? *Students responses may vary but could include that they could change the shape of one of their puzzle pieces by cutting it or ripping off a piece so that it no longer fits with its substrate puzzle piece.*
- The graph on the right shows the energy required to carry out a biological reaction with and without an enzyme present. Write an analysis of how the presence of the enzyme affects the energy requirements of the reaction. *The students should be able to interpret from the graph that enzymes lower the activation energy needed to carry out a reaction*.
- When you connect the substrate to the enzyme, this represents an "enzymatic reaction." What do you hypothesize might speed up or slow down an enzymatic reaction? Hypotheses will vary. *Students could hypothesize that increasing temperature, concentration, or surface area could result in a speed up of the reaction because they have learned this is true of chemical reactions. They could further hypothesize that the opposite would be true to slow down the reaction.*

Expand

1. Students will elaborate off the final question in their lab worksheet: What do you hypothesize might speed up or slow down an enzymatic reaction?
2. Students will explore how changes in temperature, pH, and concentration affect the rate of enzymatic reactions.
3. Students will use potatoes and hydrogen peroxide to induce an enzymatic reaction. They will then change the temperature (add and remove heat), change the pH (using 1M hydrochloric acid and sodium hydroxide (base)), and change the concentration (smashing up the potato to release the enzymes) and record their observations of how it impacted the reaction rate.
4. See the Enzyme Lab Directions (Blackline Master #3) for further directions on how to set up and conduct lab.
5. Student data collection, graphing, and reflection questions are included in the Enzymes Student Lab Sheet (Blackline Master #4).
6. Once students have gathered all of their data and have completed their reflection/conclusion questions, students will do a data share by having two members of their group go and share their data and conclusions with another group. Likewise, two members from another group will join their group.
7. New student groups will have 5 minutes to discuss their data, their analyses, and their conclusions and compare them to the new groups. There is a spot on their Enzymes Student Lab Sheet to reflect on their collaboration with the other group.
8. **NOTE**: Please make sure that you label the graduated cylinders that are used for measuring the hydrochloric acid, sodium hydroxide, and hydrogen peroxide so that there is no cross contamination of the liquids. Cross contamination will drastically affect the students' data and results.
9. **NOTE**: Please make sure that students are wearing appropriate safety equipment such as goggles, lab apron, and safety gloves as applicable during this portion of the lab. Give students advanced warning about the hazards of the materials they are using during this lab.

Evaluate

FORMAL EVALUTION

1. Enzymes Check for Understanding

INFORMAL or OPTIONAL EVALUTIONS

1. Student observation (do students seem to be participating in conversation, are they able to answer your questions when you ask about the activity).

2. Student demonstration Reflection & Enzyme Puzzle Reflection could be submitted as an informal or optional assessment.

WRAP UP.
Bring the lesson to a conclusion by watching the video (approx. 6:30 minutes), **Enzymes – A Fun Introduction,** by going to https://www.youtube.com/watch?v=XTUm-75-PL4. This video goes over the basics of enzymes, how enzymes work in the body, and how enzymes and substrates interact.

Make sure to fast forward past the advertisements and display the video in "full screen" mode so that the web page advertisements located on the right-hand side and underneath the video will not be seen. In addition, please be on the lookout for "pop-up" ads while the video is playing.

Supplementary Resources

Teachers
Johnson-Matthey Catalysts. (2016). Enzymes as catalysts. Retrieved from http://resources.schoolscience.co.uk/johnsonmatthey/page39.htm.

UCSB. (2015). How does catalase break down hydrogen peroxide? Retrieved from http://scienceline.ucsb.edu/getkey.php?key=166.

Students
Chem4Kids. (2016). Enzymes. Retrieved from http://www.chem4kids.com/files/bio_enzymes.html.

CITATION OF SOURCES.

Andriese, B. (2015). Catalase Enzyme Lab. Retrieved from http://biology-5-e-g.ycjusd.yhs.schoolfusion.us/modules/locker/files/get_group_file.phtml?gid=4994352&fid=28969571&sessionid=8a45ee78047079d6825efdaf59ad2ab1.

Chem4Kids. (2016). Enzymes. Retrieved from http://www.chem4kids.com/files/bio_enzymes.html.

Fvasconcellos. (2008). Enzyme Reaction Graphic. Retrived from https://commons.wikimedia.org/wiki/File:Carbonic_anhydrase_reaction_in_tissue.svg.

Gal M. (2007). Graph of Enzyme Activity against pH. Retrieved from https://commons.wikimedia.org/wiki/File:Enzyme-ph.png.

Gal M. (2007). Graph of Enzyme Activity against Temperature. Retrieved from https://commons.wikimedia.org/wiki/File:Enzyme-temperature.png.

Science Teachers' Association of Ontario. (2015). Liver Enzyme Action Demonstration. Retrieved from https://stao.ca/cms/stao2015-handouts?task=document.viewdoc&id=325.

Shafee, T. (2015). Saturation Curve for Enzymes. Retrieved from https://commons.wikimedia.org/wiki/File:Michaelis_Menten_curve_2.svg.

Hottuna080. (2013). Lock and Key Model. Retrieved from https://commons.wikimedia.org/wiki/File:Lock_and_key.png.

__✓__ Yes, I cited all materials and resources used in this lesson.

Heather Miller
Lesson author signature

Blackline Master #1

DEMONSTRATION REFLECTION SHEET

What is in the test tubes?

Test Tube 1	Test Tube 2	Test Tube 3

Your Predictions:
What will happen when hydrogen peroxide is added to the chicken liver?

What will happen when we introduce FIRE into each test tube?

	Test Tube 1	Test Tube 2	Test Tube 3
What will happen?			
Why will that happen?			

Your group's ideas:

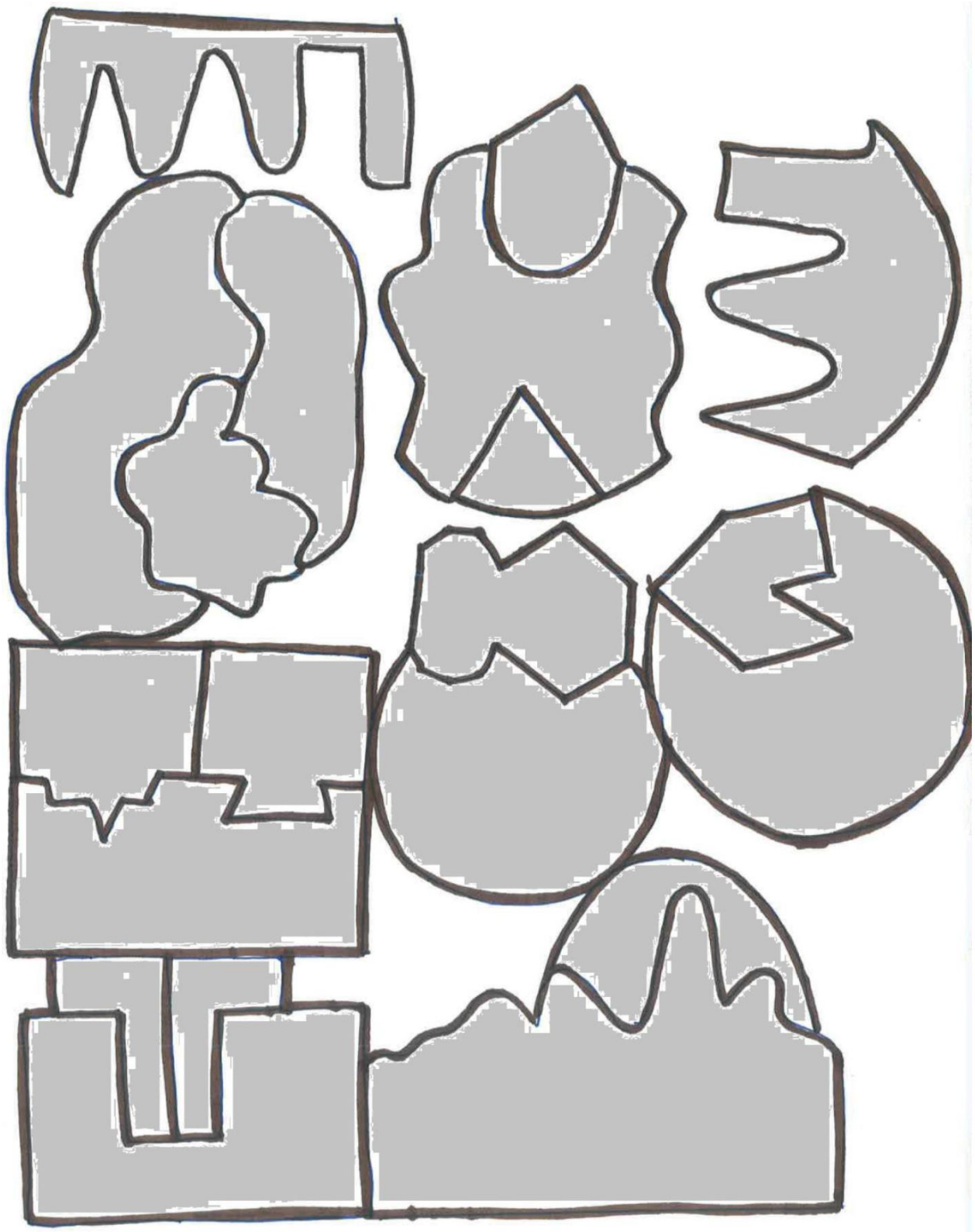
Your observations & explanations:

	Test Tube 1	Test Tube 2	Test Tube 3
What happened?			
Why did that happen?			

Blackline Master #2

Enzymes Puzzle

Cut out the pieces along the heavy black lines. *Credit: Hand drawn. Heather Miller*



Blackline Master #3

ENZYMES PUZZLE REFLECTION

1. In the puzzle, the large piece of the puzzle represents the "**enzyme**," the area where the enzyme and the pieces meet is called the "**Active site**" and the little pieces that connect to the active site are called "**substrates**." Draw a labeled diagram that shows how the enzyme and substrate interact.

2. Were you able to fit multiple different substrates into multiple different active sites? Why or why not?

3. Based on your response to question 2, what conclusion can be drawn about the relationship between enzymes and substrates?

4. How might this puzzle activity be related to the demonstration you saw with the liver and hydrogen peroxide?

5. "Denaturing" of an enzyme occurs when the shape of the enzyme is altered due to extreme temperatures or changes in pH. How could you alter your puzzle pieces to show the denaturing of an enzyme?

6. The graph on the right shows the energy required to carry out a biological reaction with and without an enzyme present. Write an analysis of how the presence of the enzyme affects the energy requirements of the reaction.

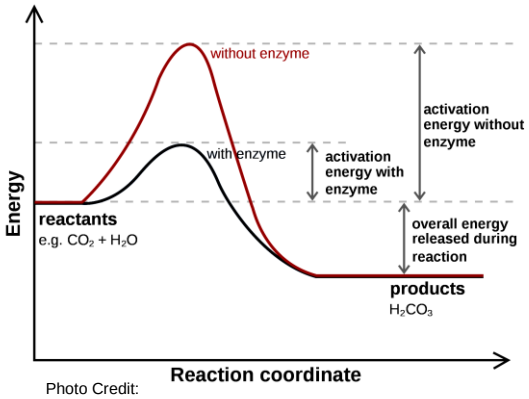


Photo Credit: https://commons.wikimedia.org/wiki/File:Carbonic_anhydrase_reaction_in_tissue.svg

7. When you connect the substrate to the enzyme, this represents an "enzymatic reaction." What do you hypothesize might speed up or slow down an enzymatic reaction?

Blackline Master #4

ENZYME LAB DIRECTIONS

Introduction:

Catalase is an enzyme that speeds up the breakdown of **hydrogen peroxide** (H_2O_2) into **water** (H_2O) and **oxygen gas** (O_2). Hydrogen peroxide is a poisonous by-product of cell metabolism. Without catalase, the cells would be damaged by the hydrogen peroxide they produce. In this activity, you will use the catalase in potato cells to see how various factors such as concentration, temperature, and pH level affect enzyme activity.

Materials:

Test tube rack
3 test tubes
3% Hydrogen peroxide
Potato cubes
Mortar and pestle
Hot water bath
Ice water bath
Test tube holder
Graduated cylinders
Test tube cleaners
Waste container
Purified Water
Hydrochloric acid (HCl)
Sodium hydroxide (NaOH)

Estimating the Reaction: For each test, allow the reaction to fully occur for ALL test tubes, and then describe the pattern of reaction and rank the test tubes in order of reactivity.

0 = no reaction (no foam or bubbling)
1 = minor reaction (very few bubbles or very slow to bubble, delayed reaction)
2 = moderate reaction (medium amount of bubbles, medium speed, delayed reaction)
3 = major reaction (a lot of bubbles, bubbles quickly and immediately)

Procedures:

Test 1: Enzyme Concentration (High, Medium, & Low Concentration)

1. Set up a test tube rack with 3 test tubes.
2. Place an **unground** piece of potato (low enzyme concentration) in tube 1.
3. Cut a second piece of potato into 4 pieces (medium enzyme concentration) and add the pieces into tube 2.
4. Grind one pea-sized piece of potato in a mortar and pestle. (Grinding releases the enzyme from the cells, which will **increase** the concentration of the enzyme.) Add the ground potato to tube 3.
5. Pour **10ml** of hydrogen peroxide into tubes 1, 2, and 3.
6. Estimate the rate of reaction (based on the scale) and record your results in the data table.
7. **Clean the test tubes** and move on to the next section.

Test 2: Temperature *(Freezing 0°C, Room 25°C, Boiling 100°C)*

1. To tube 1, 2, and 3, add one piece of potato.
2. Place tube 3 in a hot water bath for 5 minutes.
    a. Caution: Follow instructions exactly as demonstrated when using the hot water bath!
    b. Use a test tube holder to remove the tube from the hot water.
3. At the same time, place tube 1 in an ice water bath for 5 minutes.
4. **Test tube 2 will remain at room temperature in the test tube rack.**
5. **After** the five minutes have passed, put test tubes 1 and 3 in the test tube rack and **add** 10ml of hydrogen peroxide to all 3 test tubes.
6. Estimate the rate of reaction (based on the scale) and record your results in the data table.
7. **Clean test tubes** and move to the next section.

Test 3: pH level (Neutral pH, acidic pH, or basic pH)

Test Tube #1

1. Add **5 ml of hydrochloric acid** to test tube 1.
2. Add 5 ml of hydrogen peroxide to test tube 1.
3. Using the pH indicator paper, measure the pH of the solution in the test tube.

Test Tube #2

1. Add **5 ml of water** to test tube 2.
2. Add 5 ml of hydrogen peroxide to test tube 2.
3. Using the pH indicator paper, measure the pH of the solution in the test tube

Test Tube #3

1. Add **5 ml of sodium hydroxide** to test tube 3.
2. Add 5 ml of hydrogen peroxide to test tube 3.
3. Using the pH indicator paper, measure the pH of the solution in the test tube.

All Three Test Tubes

1. Add one piece of potato to each test tube.
2. Estimate and compare the rate of reaction (based on the scale) and record your results in the data table.
3. **Thoroughly clean all test tubes, mortars and pestles, and table area. Return all equipment to its assigned location**.

Blackline Master #5

ENZYMES STUDENT LAB SHEET

Question: How does a change in temperature, concentration, or pH affect enzyme activity?

Hypotheses:

Make a hypothesis in which you predict under which condition the enzyme will work **best**.
Follow the example shown.

a. **Concentration** (High, Moderate, Low concentration)
Example Hypothesis: If the enzyme catalase concentration is LOW, then it will react faster/better than if the concentration was high.

Your Hypothesis:

b. **Temperature** (Freezing 0ºC, Room 25ºC, Boiling 100ºC)
Your Hypothesis:

c. **pH level** (Neutral pH (7), acidic pH (0-6.9), or a basic pH (7.1-14))
Your Hypothesis:

Data Table:

Test 1: Concentration

	Contents & Observations	Reaction rate
Test tube 1		
Test tube 2		
Test tube 3		

Test 2: Temperature

	Contents & Observations	Reaction rate
Test tube 1		
Test tube 2		
Test Tube 3		

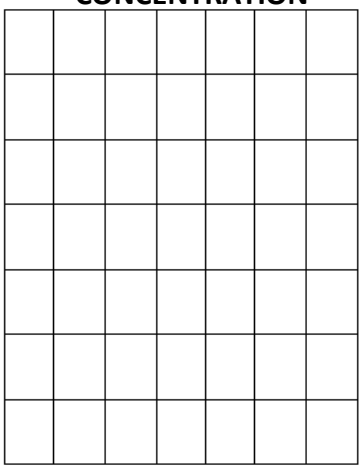
Test 3: pH

	Contents & Observations	Reaction rate
Test tube 1		
Test tube 2		
Test tube 3		

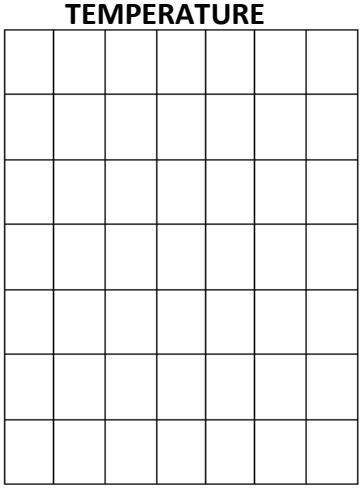
Data Analysis:

Graph the results of your three tests: concentration, temperature, and pH vs reaction rate. Don't forget your axis labels for each graph.

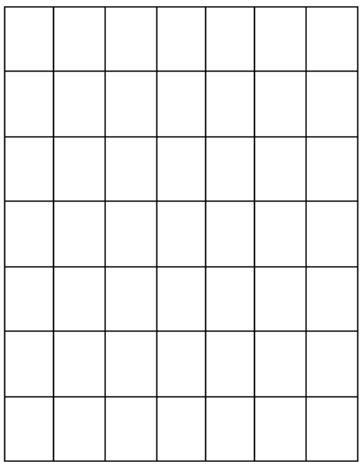
CONCENTRATION



TEMPERATURE



pH



Conclusions:

1. How did reaction rate change with the change in concentration? Explain why you think this happened. Cite your data in your explanation.

2. How did reaction rate change with the change in temperature? Explain why you think this happened. Cite your data in your explanation.

3. How did reaction rate change with the change in pH? Explain why you think this happened. Cite your data in your explanation.

4. Where might we see changes in pH, temperature, OR concentration in nature and how could these changes affect living organisms?

5. How might the reactions you observed be different if enzymes were not present?

6. Based on your data, predict what the "optimum" pH, temperature, and concentration is for the catalase enzyme reactions that you observed.

7. How and why might the "optimum" temperature for the enzymes in liver be different that the "optimum" temperature for the enzymes in potatoes?

8. Based on your data, at what pH and temperature do you predict that denaturing would occur in the catalase enzyme?

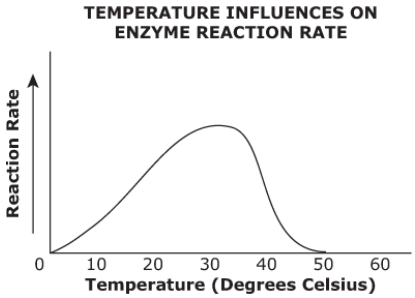
WAIT TO COMPLETE THIS QUESTION. We will do this after we rearrange our groups.

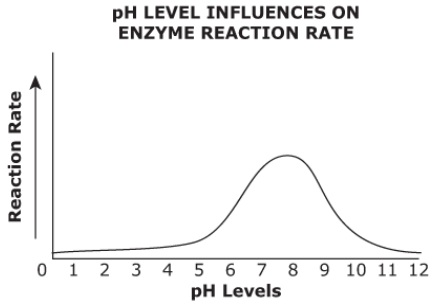
9. How was your data, your analysis, and your conclusions similar or different than your collaborative group?

Blackline Master #5

Enzymes Check for Understanding

1. Researchers know that the reaction rate of an enzyme is affected by temperature and pH level. Some of these effects are shown on the graphs below.





Images created using Excel: Heather MIller

If the enzyme was placed in an environment where the temperature is 50°C with a pH 5.0, what would be expected to happen to the rate of reaction?

A. There would be an increased rate of reaction.
B. There would be no change to the rate of reaction.
C. There would be a reduced rate of reaction and possible denaturing of the enzyme.
D. There would first be an increased rate of reaction and then a decreased rate of reaction.

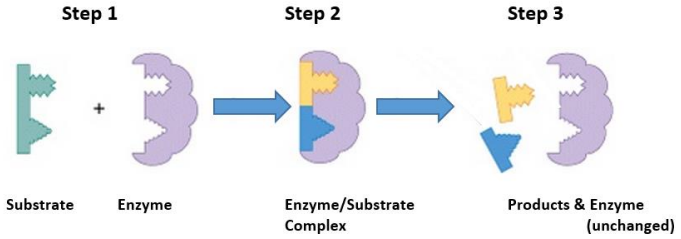
2. Enzymes are catalysts that are able to speed up the rate of chemical reactions. How are enzymes able to do this?

A. Enzymes make the particles interacts more frequently.
B. Enzymes are able to increase the temperature of the reaction.
C. Enzymes lower the activation energy needed to carry out the reaction.
D. Enzymes increase the activation energy needed to carry out the reaction.

3. Which of the following best explains why enzymes are necessary for many cellular reactions?

A. Enzymes supply the oxygen necessary for the reactions.
B. Enzymes change reactants from solid to liquids during the reactions.
C. The reactions take up too much space in the cell if the enzymes are missing.
D. The reactions are too slow to meet the needs of the cell if enzymes are missing.

4. The picture below shows the lock-and-key model of enzyme function.



Picture created and edited in Word: Heather Miller

What is taking place in the second step of this process?

A. The catalyzed reaction is releasing a product.
B. The active sites are restructuring the enzyme.
C. The substrates are beginning to alter the enzyme.
D. The enzyme is causing bonds to break within the substrate.

5. A certain enzyme will react with protein but not fats. Which statement best explains this observation?

A. Proteins act as a coenzyme for reactions.
B. Fat molecules are too large to be reacted with.
C. Enzyme molecules are specific in their actions.
D. Fats are composed of lipids which cannot be reacted.

Blackline Master #6: ANSWER KEYS

DEMONSTRATION REFLECTION SHEET

What is in the test tubes?

Test Tube 1	Test Tube 2	Test Tube 3
Chicken Liver	*Hydrogen Peroxide*	*Chicken Liver & Hydrogen Peroxide*

Your Predictions:
What will happen when hydrogen peroxide is added to the chicken liver?
Student responses will vary as this is a prediction.

What will happen when we introduce FIRE into each test tube?

	Test Tube 1	Test Tube 2	Test Tube 3
What will happen?	*Student responses will vary as this is a prediction.*	*Student responses will vary as this is a prediction.*	*Student responses will vary as this is a prediction.*
Why will that happen?	*Student responses will vary as this is a prediction.*	*Student responses will vary as this is a prediction.*	*Student responses will vary as this is a prediction.*

Your group's ideas:
Student responses will vary as this is a prediction.

Your observations & explanations:

	Test Tube 1	Test Tube 2	Test Tube 3
What happened?	*Nothing happened*	*Nothing happened*	*There was a loud "POP" and then the wooden stick lit on fire.*
Why did that happen?	*There was no oxygen present*	*There was no oxygen present*	*There was oxygen present and oxygen is flammable so it lit on fire causing the wood to catch on fire.*

ENZYMES PUZZLE REFLECTION

1. In the puzzle, the large piece of the puzzle represents the "**enzyme**," the area where the enzyme and the pieces meet is called the "**Active site**" and the little pieces that connect to the active site are called "**substrates**." Draw a labeled diagram that shows how the enzyme and substrate interact.

Students' diagrams should look something like this:

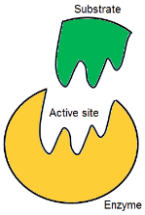


Photo credit: https://commons.wikimedia.org/wiki/File:Lock_and_key.png

2. Were you able to fit multiple different substrates into multiple different active sites? Why or why not?
Student responses will vary but should reflect that they were only able to make one specific substrate fit into the active site of one specific enzyme.

3. Based on your response to question 2, what conclusion can be drawn about the relationship between enzymes and substrates?
Student responses may vary but should include something explaining that there is only one substrate that each enzyme is responsible for reacting with and that the enzyme-substrate relationship is very specific like a "lock & key" system.

4. How might this puzzle activity be related to the demonstration you saw with the liver and hydrogen peroxide?
Student responses may vary but should indicate that there are enzymes in the liver (catalase) that connect with the hydrogen peroxide (the substrate) to break it down into water and oxygen. The puzzle pieces could be the enzyme (catalase) from the liver and the substrate (hydrogen peroxide).

5. "Denaturing" of an enzyme occurs when the shape of the enzyme is altered due to extreme temperatures or changes in pH. How could you alter your puzzle pieces to show the denaturing of an enzyme?
Students responses may vary but could include that they could change the shape of one of their puzzle pieces by cutting it or ripping off a piece so that it no longer fits with its substrate puzzle piece.

6. The graph on the right shows the energy required to carry out a biological reaction with and without an enzyme present. Write an analysis of how the presence of the enzyme affects the energy requirements of the reaction.
The students should be able to interpret from the graph that enzymes lower the activation energy needed to carry out a reaction.

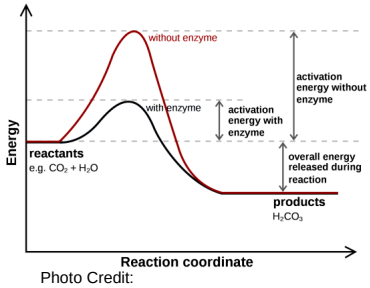


Photo Credit: https://commons.wikimedia.org/wiki/File:Ca

7. When you connect the substrate to the enzyme, this represents an "enzymatic reaction." What do you hypothesize might speed up or slow down an enzymatic reaction?
Students could hypothesize that increasing temperature, concentration, or surface area could result in a speed up of the reaction because they have learned this is true of chemical reactions. They could further hypothesize that the opposite would be true to slow down the reaction.

ENZYMES STUDENT LAB SHEET

Question: How does a change in temperature, concentration, or pH affect enzyme activity?

Hypotheses:

Make a hypothesis in which you predict under which condition the enzyme will work **best**.
Follow the example shown.

d. **Concentration** (High, Moderate, Low concentration)
Example Hypothesis: If the enzyme catalase concentration is LOW, then it will react faster/better than if the concentration was high.

Your Hypothesis: *Students responses may vary*

e. **Temperature** (Freezing 0ºC, Room 25ºC, Boiling 100ºC)
Your Hypothesis: *Students responses may vary*

f. **pH level** (Neutral pH (7), acidic pH (0-6.9), or a basic pH (7.1-14))
Your Hypothesis: *Students responses may vary*

Data Table:

Test 1: Concentration

Contents & Observations		Reaction rate
Test tube 1	*whole potato* *a small amount of bubbles and foam*	*1*
Test tube 2	*quartered potato* *a medium amount of bubbles and foam*	*2*
Test tube 3	*ground up potato* *a lot of bubbles and foam* *potato could be hard to distinguish from foam*	*3*

Test 2: Temperature

	Contents & Observations	Reaction rate
Test tube 1	*cold water, potato may sink, longer reaction, slow to start, delayed reaction, small amount of bubbles*	*1-2*
Test tube 2	*room temperature water, potato may float, fast reaction then slows down, a lot of bubbling*	*2-3*
Test Tube 3	*hot water, potato may sink, slow to start, delayed reaction, very few bubbles*	*0-1*

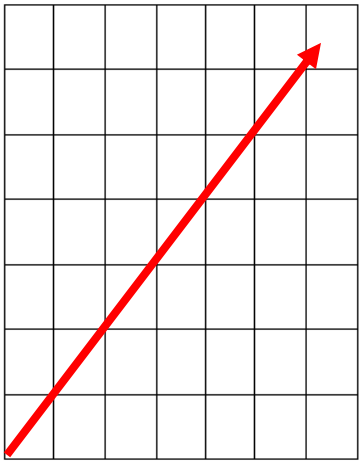
Test 3: pH

	Contents & Observations	Reaction rate
Test tube 1	*hydrochloric acid, pH 1, little to no reaction, very few if any bubbles*	*1*
Test tube 2	*water, pH of 7, bubbled a lot and continuously bubbles*	*3*
Test tube 3	*sodium hydroxide, pH 12+, moderate bubbling that lasted a long time*	*2*

Data Analysis:

Graph the results of your three tests: concentration, temperature, and pH vs reaction rate. Don't forget your axis labels for each graph.

CONCENTRATION



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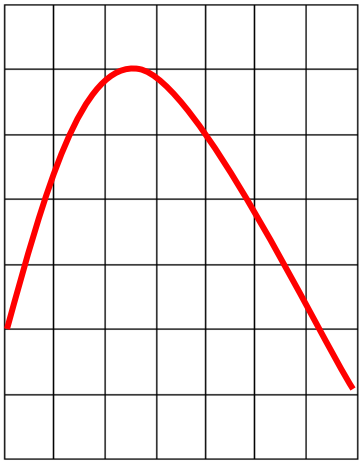
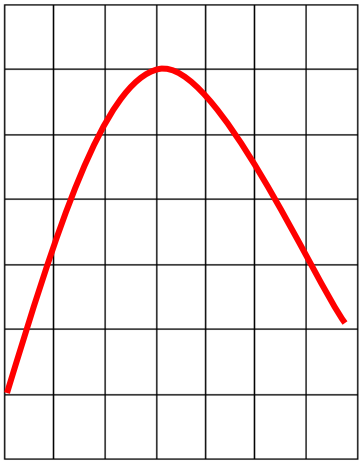


pH



Conclusions:

1. How did reaction rate change with the change in concentration? Explain why you think this happened. Cite your data in your explanation.
   As the concentration increases, the reaction rate increases. Student data will vary but should be cited specifically in their response, for example, "With low concentration we had a reaction rate of only 1 but with a high concentration we had a reaction rate of 3." Possible explanations for this occurrence could include that when there are more enzymes available to react (increased concentration) then the reaction speeds up and produces more oxygen gas as a byproduct (bubbles).

2. How did reaction rate change with the change in temperature? Explain why you think this happened. Cite your data in your explanation.
   Enzyme reaction should be the low for the cold temperature water and even lower for the high temperature water as the enzymes become denatured. The highest reaction should be for the room temperature water. Student data will vary but should be cited specifically in their response, for example, "With our ice water bath we had a reaction rate of 1-2, with room temperature it was more like a 3, and with hot water is was 0-1." Possible explanation for this occurrence could include that when the enzymes are cold, it slows their reaction because the kinetic energy of the reaction slows. In hot water, the reaction increases to such a speed that the enzymes become denatured and can no longer react with the substrate. Moderate temperatures may be the temperature of their natural environment.

3. How did reaction rate change with the change in pH? Explain why you think this happened. Cite your data in your explanation.
   Enzyme reaction should be the lowest for the low pH and low for the high pH with the best reaction occurring when the pH is near neutral. Student data will vary but should be cited specifically in their response, for example, "With the low pH we had a reaction rate of 1, with neutral pH it was more like a 3, and with high pH it is was 1-2." Possible explanation for this occurrence could include that when the enzymes react best when they are in a neutral pH environment and that this might be the pH of their natural environment.

4. Where might we see changes in pH, temperature, OR concentration in nature and how could these changes affect living organisms?
   Changes in pH in nature could occur from acid rain, a change in environment where the soil or water has a different pH, or from illness in the organism. Change in temperature could occur from changes in the environment such as climate or weather change, change in location, the cooking process, or illness in the organism. Changes in concentration would occur if medications were taken to provide the body with extra enzymes or reduce enzyme activity. Another reason might be from eating specific foods that need a specific enzyme to be digested or metabolized (example: lactase). Changes in any of these conditions would heighten or diminish an organisms' ability to carry out these reactions.

5. How might the reactions you observed be different if enzymes were not present?
   If the enzymes were not present in the reactions observed, the reaction would take much longer and no reaction would likely be observed.

6. Based on your data, predict what the "optimum" pH, temperature, and concentration is for the catalase enzyme reactions that you observed.
   Student responses will correlate with their data, but should follow the general pattern of optimum pH is around neutral, optimum temperature is around room temperature, and optimum concentration is high.

7. How and why might the "optimum" temperature for the enzymes in liver be different that the "optimum" temperature for the enzymes in potatoes?
   The optimum temperature for liver from an ANIMAL would likely be higher than the optimum temperature for a PLANT like a potato as the internal temperature of most birds and mammals is somewhere between 98°F and 109°F (37°C to 43°C). The internal temperature of plants depends on their exterior surroundings with most plants growing in the spring and summer where temperatures are between 60°F and 90°F (16°C to 32°C).

8. Based on your data, at what pH and temperature do you predict that denaturing would occur in the catalase enzyme?
   Students responses will vary based on their data, but denaturing of this enzyme likely occurs in a pH lower than 6 and higher than 8, and a temperature higher than 70-80°F (21°C to 27°C).

WAIT TO COMPLETE THIS QUESTION. We will do this after we rearrange our groups.

9. How was your data, your analysis, and your conclusions similar or different than your collaborative group?
   Student responses will vary.

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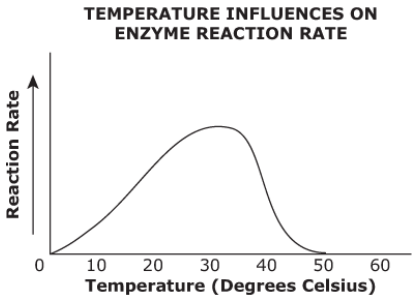
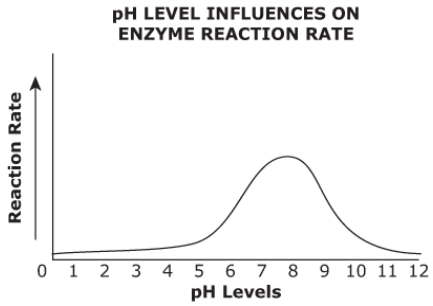
1. Researchers know that the reaction rate of an enzyme is affected by temperature and pH level. Some of these effects are shown on the graphs below.





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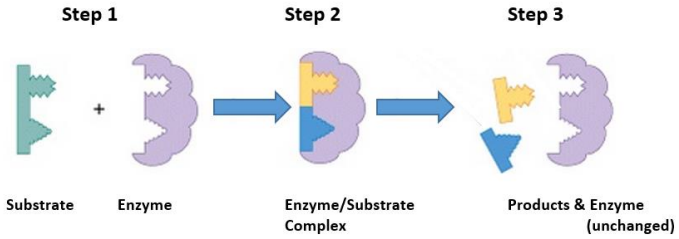

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