MiSP Enzyme Action Worksheet 1 L3

Name _____ Date_____

"TEMPERATURE AND ENZYME ACTIVITY"

Introduction:
Hydrogen peroxide (H_2O_2) is a poisonous substance that can be made in a living thing. It can damage cells if it is not removed. 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 with catalase----------> water + oxygen

REMEMBER: CATALASE is an enzyme. An enzyme is an organic CATALYST that increases the rate of a reaction without being used up in the process. Certain plants and animal organs contain high concentrations of catalase. Potatoes and liver are two commonly used sources of catalase. Your teacher has prepared an **extract** of potato by chopping it up into tiny pieces, mixing it in cold water and removing large chunks. The potato extract contains 100 units of catalase/ml.

SAFETY:
YOU WILL BE WORKING WITH HOT WATER
WEAR GOGGLES AT ALL TIMES!!!
Use caution when handling hydrogen peroxide

Problem:
How does temperature affect the rate of enzyme action?
How the lab works:
A filter paper disc will be coated with the potato extract (enzyme source) and then dropped into a vial of the substrate (hydrogen peroxide). As the enzyme breaks down the hydrogen peroxide into water and oxygen gas, the bubbles of oxygen will collect underneath the filter and make it rise to the surface of the hydrogen peroxide. The time it takes for the filter to rise is an indication of the rate of enzyme activity. The LONGER (more seconds) the disk takes to rise to the surface, the SLOWER the reaction. The SHORTER (less seconds) the time the disk takes to rise to the surface, the FASTER the reaction.

MATERIALS:
catalase – extracted from potatoes
3% hydrogen peroxide
forceps
filter paper discs (holes punched out of filter paper)
water

ice
water baths – ice, warm (approximately 37° C), and boiling
vials, little beakers, and/or test tubes
test tube clamp
test tube rack if test tubes are being used
marking pencils
stopwatch or timer
5 or 10 mL graduated cylinder (or pipette)
50 or 100 mL graduated cylinder

PROCEDURES:

___ 1.Your teacher will set up water baths and tell you the temperatures. Record the temperatures in your data chart: cold (ice) bath, room temp water bath, warm water bath and boiling water bath.

___ 2. Place 5.0 ml of potato extract containing catalase at 100 units/ml in each of 4 test tubes. Label the test tubes with your name and the water bath temperature. Place 1 test tube in each of the water baths.

___ 3. Place 5 ml (or a different volume directed by your teacher) of 3% H_2O_2 in each of four vials or four additional test tubes. Label. Place 1 vial or test tube in each of the water baths.

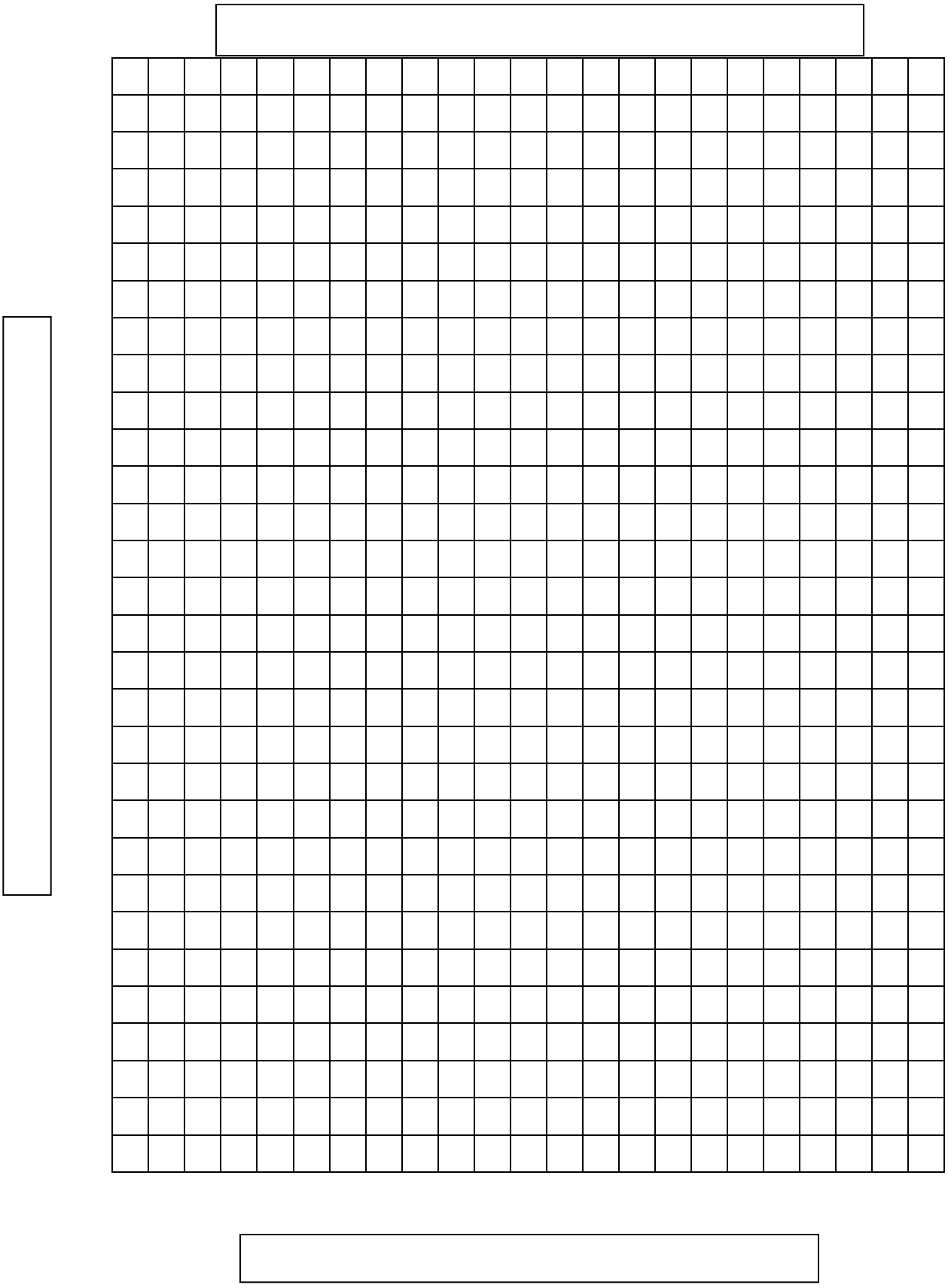
___ 4. Allow the catalase and substrate to incubate at each temperature for about 5 minutes. After five minutes, test the reaction time at each temperature by dipping a filter paper disk into potato extract (enzyme) at that temperature, draining it, and then dropping it into substrate at the same temp. Time how long it takes the filter to rise to the surface at each temperature. Record your results in the data chart. If the reaction does not occur after 3 minutes, mark the chart for that temperature "n.r." (no reaction).

Data

Water Temperature	Reaction Time (seconds)
Cold: ____ °C	
Room Temperature ____ °C	
Warm: ____ °C	
Boiling: ____ °C	

___ 5. Graph the data on the next page.

- Label the X axis
- Label the Y axis
- If there is no observable reaction, that data point (temperature) cannot be graphed.
- Draw a best fit line from the cold to warm ordered pairs.



Discussion L1-3

1. Which temperature had the fastest reaction (the paper disk came to the top in the shortest time)? Which temperature had the slowest reaction? Was there a temperature when there was no reaction?

Fastest: _____

Slowest: _____

No reaction: _____

2. Generally, when temperature increases the rate of a reaction increases. Is that true for all temperatures in this experiment? Explain.

_____

_____

3. Explain the results with the substrate and enzyme kept in the boiling water bath. (Hint – an enzyme is a protein. The white of an egg is protein. What happens to an egg when it is put in boiling water?)

_____

_____

_____

4. Use the graph to predict the number of seconds for the enzyme coated disk to rise to the top of the vial or test tube at

- 10° C _____
- 25° C _____

5. Review your data and write a conclusion statement by completing this sentence (*Remember that shorter time indicates faster rate of reaction*):

As the temperature of an enzyme reaction increases, the

rate of the reaction _____.

Discussion L2-3

6. Determine the change in reaction time as temperature increased by calculating the unit rate of change (slope) of the best fit line

$$\text{Unit Rate of Change} = \frac{\Delta \text{ Reaction Time (Seconds)}}{\Delta \text{ Temperature } °C}$$

(When using a best fit line, the ordered pairs to determine unit rate of change (slope) must be from the best fit line, not from your data chart.)

$$\text{Unit Rate of Change} = \frac{\Delta \text{ Reaction Time (Seconds))}}{\Delta \text{ Temperature } °C} = \frac{\Delta y}{\Delta x} = \frac{(y_2 - y_1)}{(x_2 - x_1)}$$

Ordered Pair used for calculation (x_1, y_1) (x_2, y_2)	Δ Reaction Time (Seconds) Δ y	Δ Temperature °C Δ x	**Unit Rate of Change (slope)** Δ y/Δ x

7. What is the sign (-/negative or +/positive) of the unit rate of change? What does that sign tell you about the changes in reaction time (seconds) as the temperature increases?

_____

_____

_____

Discussion L3

8. Find the Y intercept for the best fit line on the graph. Use the equation for a line to calculate the y-intercept. Use the best fit line you used in #6. The equation for a line is

y = mx + b
where m is the unit rate of change (slope) and
b is the y-intercept

Y Intercept
m = Ordered pair (x, y) = (___ , ___) y = mx +b Solve for b:

9. Based on the unit rate of change (slope) that you calculated above and the y intercept, write an equation for the best fit line on the graph. Remember that the equation for a line is **y = mx + b** and m is the unit rate of change (slope) and b is the y intercept.

Equation – Best fit line Temperature and Enzyme Activity

10. Using the equation, above, calculate the reaction time (in seconds) for the indicated temperature

X =8° C y = _____ seconds	X = 31° C y = _____ seconds