Measuring Catalase Action with Sensors

Introduction

Measure the rate of decomposition of hydrogen peroxide by catalase using an O_2 Gas Sensor or Gas Pressure Sensor.

Concepts

- Homeostasis
- Enzyme catalyst
- Reaction rate

Materials

Catalase solution

3% Hydrogen peroxide solutions, H_2O_2, 3%

Buret clamp

Disposable graduated pipet

Graduated cylinder

Magnetic stirrer and stir bar

Support stand

Vernier interface

Vernier O_2 Gas Sensor with gas sampling chamber or Gas Pressure Sensor fitted with rubber tubing and #5 two-hole stopper with 125-mL Erlenmeyer flask

Water, distilled

Safety Precautions

The hydrogen peroxide can cause skin and eye irritation upon contact. Wear chemical splash goggles, chemical-resistant gloves, and a chemical-resistant apron. Wash hands thoroughly with soap and water before leaving the laboratory. Please review current Safety Data Sheets for additional safety, handling, and disposal information.

Preparation

1. Prepare the catalase solution by adding 10 milligrams (0.01 grams) of catalase to 50 milliliters of distilled water.
2. Stir on a magnetic stirrer until catalase is dissolved. Catalase is only slightly soluble in water and dissolving it completely takes several minutes.
3. Determine the concentration of the catalase solution. Catalase activity is measured for each lot and printed on each bottle. Determine the concentration of the catalase solution using Formula 1, where x = the number of units printed on the bottle of catalase.

$$\frac{10\ mg}{50\ mL} \times \frac{X\ units}{1\ mg} = \text{Concentration of catalase} \qquad \textit{Formula 1}$$

$$\frac{10\ mg}{50\ mL} \times \frac{3000\ units}{1\ mg} = 600\ \frac{units}{mL} \qquad \textit{Example}$$

Procedure

1. Connect the O_2 Gas Sensor or Gas Pressure Sensor to the data-collection interface and start the data collection program.
2. Set the duration of the experiment to 5 minutes or 300 seconds.
3. Use a buret clamp to fasten the sensor to a support stand.
4. Consult Table 1 to determine the appropriate volume of 3% H_2O_2 and catalase solution to use based on your sensor choice.
5. Using a graduated cylinder, add H_2O_2 to the gas collecting chamber.
6. Using a graduated pipet, add catalase solution and swirl to mix.

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Sensor	Vol. of H_2O_2	Vol. of catalase	Type of gas collecting chamber
O_2 Gas Sensor	10 mL	1 mL	Included gas sampling chamber
Gas Pressure Sensor connected to plastic tubing with Luer-lock connectors	50 mL	2 mL	125 mL Erlenmeyer flask fitted with #5 two-hole rubber stopper with closed valve in second hole.

Table 1.

7. Quickly insert and gently twist the stopper or O_2 gas sensor into the gas collection chamber to form an airtight seal.
8. Start data collection. Data collection will stop after five minutes.
9. Using the data-collection software, perform a linear fit on the area of the graph that is most linear, generally between 50 and 200 seconds. The slope of the line is the rate of the reaction.
10. Rinse the gas sampling chamber and dry the inside.

Disposal

Please consult your current *Flinn Scientific Catalog/Reference Manual* for general guidelines and specific procedures, and review all federal, state and local regulations that may apply, before proceeding. The hydrogen peroxide soluton may be disposed of according to Flinn Suggested Disposal Method #26b.

NGSS Alignment

This laboratory activity relates to the following Next Generation Science Standards (2013):

Disciplinary Core Ideas: High School

HS-LS1 From Molecules to Organisms: Structures and Processes

LS1.A: Structure and Function

LS1.C: Organization and Matter and Energy Flow in Organisms

Science and Engineering Practices

Using mathematics and computational thinking

Analyzing and interpreting data

Constructing explanations and designing solutions

Crosscutting Concepts

Stability and Change

Structure and function

Tips

- The O_2 Gas Sensor measures change in the percent of oxygen above the sample, while the Gas Pressure Sensor measures a change in air pressure within the chamber. Never submerge either sensor in solutions.
- Substituting yeast for purified catalase is a cost-effective alternative if this activity is used as a demonstration of enzyme activity.
- For quantitative studies, purified catalase allows for precise control of the concentration of the enzyme.
- Catalase solution should be made fresh each day. 50 mL is enough for one class to complete one trial. Store powdered catalase in the refrigerator. Enzyme activity will decrease over time.
- Prior to conducting this lab, become familiar with using the Vernier interface and data-collection software. Visit www.vernier.com for detailed information on how to use Vernier data collection software and sensors.

Discussion

Hydrogen peroxide (H_2O_2) is a toxin that readily forms as a byproduct of metabolic reactions in most organisms. In order to survive, organisms have enzymes that catalyze the redox reaction that breaks H_2O_2 into non-toxic molecules. Enzymes are biochemical catalysts. A catalyst is a substance that accelerates the reaction rate but is not consumed during the reaction. The class of enzymes that break down hydrogen peroxide are peroxidases. The group of peroxidases that does not require a reducing agent are catalases.

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

Acknowledgment

Special thanks to Vernier Software & Technology for sharing this activity with Flinn Scientific.

Materials for *Measuring Catalase Action with Sensors* are available from Flinn Scientific, Inc.

Catalog No.	Description
C0359	Catalase, 1 g
H0009	Hydrogen Peroxide 3%, Lab Grade, 473 mL
AP1721	Disposable Graduated Pipets, 20
TC1515	Oxygen Gas Sensor
TC1561	LabQuest 2 Interface
TC1559	LabQuest Mini Interface

Consult your *Flinn Scientific Catalog/Reference Manual* for current prices.