Natural and catalyzed decomposition of hydrogen peroxide

ASSIGN BUSTER

Enzymes are proteins that catalyze biochemical reactions by lowering activation energy. Enzymes are specific to one reaction due to their structure; they bind with a specific substrate at an active site to induce a conformational change that allows chemical bonds to be broken easier-thus reducing the amount of energy needed to initiate a reaction. Catalysts, and therein enzymes, are neither reactants nor products in chemical reactions. Therefore, the same enzyme can be used to catalyze numerous reactions. 1 Several conditions affect the activity of an enzyme: salt concentration, temperature, pH, and other activators/inhibitors. Enzyme action is characterized by peak conditions, so a great increase or decrease in certain conditions could cause the enzymes to denature and lose the ability to increase the rate of reactions. For example, enzyme activity typically increases in correlation to an increase in temperature due to more kinetic energy, but when temperature reaches a certain point (about 40-50ËšC for most enzymes) the protein is denatured and can no longer function correctly. 2

Natural decomposition of hydrogen peroxide experiences variation due to changes in temperature and concentration. 3 These variables are eliminated in this experiment by the constant room temperature and constant concentration of hydrogen peroxide. This allows the only variable in the experiment to be the amount of time that the catalase reacts with the hydrogen peroxide.

This experiment tests the effect of an enzyme, catalase, on the breakdown of hydrogen peroxide (H2O2). The decomposition of H2O2 into water and oxygen occurs spontaneously, but at an extremely slow rate. A base line will

be established to measure the initial amount of H_2O_2 in the solution. After the base line is established, the breakdown of hydrogen peroxide will be catalyzed by catalase. After a certain amount of time, the addition of sulfuric acid (H_2SO_4) will stop the activity of the enzyme, halting the decomposition of hydrogen peroxide. The remaining amount of H_2O_2 will be titrated with assistance of potassium permanganate ($KMnO_4$). The potassium permanganate will react with the excess hydrogen peroxide and the sulfuric acid. Once all the hydrogen peroxide has been consumed, the addition of potassium permanganate will permanently dye the solution a pinkish or brownish color. This full reaction is shown in the equation: . 2 It is probable that the catalase will facilitate the breakdown of increasing amounts of H_2O_2 as more time elapses during the experiment. Because of the slow rate of natural breakdown of hydrogen peroxide, it is probable that an un-catalyzed decomposition reaction that sits out for 24 hours will contain about the same concentration of H_2O_2 as the base line assay.

Materials and Methods:

Experiment 1: Establishing a Base Line2

10 mL of 1. 5% solution of H_2O_2

1 mL of H_2O

10 mL of H_2SO_4

5 mL of $KMnO_4$

10 mL syringe (2)

5 mL syringe

2 beakers

1 mL Pipette

White paper

10 mL of 1. 5% H_2O_2 was put into a clean beaker. The pipette was used to add 1 mL of water to the solution. A clean 10 mL syringe was used to add 10 mL of H_2SO_4 and the entire solution was mixed well. A 5 mL sample of the solution was removed using the 5 mL syringe; it was placed into another clean beaker. A clean 5 mL syringe was used to measure 5 mL of $KMnO_4$. The $KMnO_4$ was added to the 5 mL sample one drop at a time, and the solution was mixed well after each drop. The solution was compared to the white paper so that it would be clearly apparent when a permanent color change occurred. When the solution was permanently dyed a pinkish or brownish color, the level of solution left in the syringe was measured. The amount of $KMnO_4$ used was measured by subtracting the final reading of the syringe from the initial 5 mL. This base line value was used in calculations later in the experiment. 2

Experiment 2: The Uncatalyzed Rate of H_2O_2 Decomposition2

10 mL of 1. 5% solution of H_2O_2

1 mL of H_2O

10 mL of H_2SO_4

5 mL of $KMnO_4$

10 mL syringe (2)

5 mL syringe (2)

2 beakers

1 mL Pipette

White paper

The same procedure for Experiment 1 was followed, except the beaker containing only hydrogen peroxide was stored at room temperature for 24 hours before the anything was added to it. The observations and calculations of this experiment were used to calculate the natural rate of decomposition of H_2O_2. 2

Experiment 3: Enzyme Catalyzed Rate of H_2O_2 Decomposition2

100 mL of 1. 5% solution of H_2O_2

10 mL of catalase

ice

100 mL of H_2SO_4

50 mL of $KMnO_4$

10 mL syringe (2)

5 mL syringe (2)

14 beakers

1 mL Pipette

White paper

Stopwatch

Catalase was kept on ice until needed in the experiment. 10 mL of H_2O_2 were added to a clean beaker. The 1 mL pipette was used to add 1 mL of catalase to the H_2O_2 and the solution was mixed for 10 seconds. After 10 seconds, a clean 10 mL syringe was used to add 10 mL of H_2SO_4. After the solution was mixed, a 5 mL sample was removed and placed into a clean beaker. A clean 5 mL syringe was used to measure 5 mL of $KMnO_4$. The $KMnO_4$ was added to the 5 mL sample one drop at a time, and the solution was mixed well after each drop. The solution was compared to the white paper so that it would be clearly apparent when a permanent color change occurred. When the solution was permanently dyed a pinkish or brownish color, the level of solution left in the syringe was measured. The initial and final readings of the syringe containing the $KMnO_4$ was observed and recorded. These steps were repeated at time intervals of 30, 60, 90, 120, 180, and 360 seconds (measured from when the catalase was added to when the H_2SO_4 was added). 2

Results:

Table 1: Base Line Measurements2

Initial Reading of Syringe with $KMnO_4$ (in mL)

5. 0

Final Reading of Syringe with $KMnO_4$ (in mL)

1. 3

Table 2: Uncatalyzed Reaction Measurements (after 24 hours) 2

Initial Reading of Syringe with $KMnO_4$ (in mL)

5. 0

Final Reading of Syringe with $KMnO_4$ (in mL)

1. 3

Table 3: Catalyzed Reaction Results2

$KMnO_4$ (in mL)

Time (seconds)

10 30 60 90 120 180 360

Final Reading

2. 2

3. 2

3. 4

3. 7

2. 8

2. 4

2. 0

Initial Reading

5. 0

5. 0

5. 0

5. 0

5. 0

5. 0

5. 0