

Lab essay example



Introduction

The activities that take place in a living organism are mainly controlled through the use of enzymes. Enzyme refers to a molecule that is protein in nature and functions as a catalyst to speed up the rate of a given reaction. An enzyme as a protein molecule has three main characteristics. First, an enzyme has its basic function is as to increase the reaction rate and this makes most of the cellular reactions occur more than million times that they would have if the enzyme was absent. Second, almost all enzymes act in a specific manner with only one substrate in order to produce the products. Thirdly, the regulation of enzymes is from a state of low activity to that of high activity or from high activity to low activity. The uniqueness of different cells in the body is mainly due to the unique enzymes that the cells are programmed to produce. Lack of a single enzyme in such cells can be disastrous to the cell and the whole system (Ophardt, 2003).

The huge information that is available concerning enzymes has been enabled by the fact that enzymes can be isolated from the cells where they are located. The isolated enzymes are then made to work in an external environment in a test tube. Additional work has also been done in the determination of the three-dimensional structure of various enzymes (Ophardt, 2003).

Enzyme activity is dependent on various components with the main one being the specific protein chain. Enzyme mainly consists of the protein chain in combination with other parts that are known as cofactors, making an enzyme complex. The protein or polypeptide part of the enzyme is referred to as apoenzyme. The apoenzyme may be inactive when they are

synthesized, and the inactive form is referred to as zymogen or proenzyme. In most cases, the proenzyme may include additional amino acids that are removed in order to enable the final tertiary structure to be formed and then activated to form an apoenzyme. The other component that makes up an enzyme is a cofactor. This is a non-protein substance that may be organic in nature and thus be referred to as a coenzyme. The other type of cofactor is the inorganic metal ion also known as a metal ion activator and is mainly bound through covalent bonds (Ophardt, 2003).

The associations that take place between the apoenzymes and the cofactors vary and in most cases the bonds that join them are loose. The two components only come together when the reaction is about to take place. In other cases, the components are tightly held together through covalent bonds. The cofactors are involved in the activation of the apoenzymes either by changing the geometric shape of the apoenzymes or by participating in the reaction (Ophardt, 2003). The enzyme has a specific geometric shape that is referred to as an active site, and it is in this site that the reaction occurs (Ophardt, 2003).

The understanding of the basic theory of enzyme kinetic is an essential in the understanding of the mechanisms involved in the activity of an enzyme, as well as in selecting the best method of analyzing the enzyme. There are several factors that have an effect on the rate of an enzymatic reaction. These factors include temperature, pH, enzyme concentration, as well as the concentration of the substrate (Worthington Biochemical Corporation, 2013). In terms of enzyme concentration, a change in the concentration of the enzyme may affect the reaction rate of a reaction that is catalyzed by the enzyme. When the level of enzyme concentration is increased, there is an

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increase in the rate of the reaction as there will be more enzymes that will be colliding with the substrate molecules. This takes place only up to a given concentration when an increase in the enzyme concentration does not result into an increase in the rate of reaction as at such a point enzyme concentration is not a limiting factor. Reducing the concentration of the enzyme will result in a reduction in the rate of reaction as long as it is the limiting factor (Adam-Day, 2012).

Substrate concentration is the other factor that affects the rate of a reaction. When the amount of the substrate is increased in a reaction, the rate of the reaction also increases. This is because there will be more substrates that will be colliding with the enzymes and this results in the formation of more products. However, after a given amount of substrate has been added, any more addition of the substrate will not result in an increase in the rate of reaction as the substrate concentration is no longer the limiting factors in the reaction. The enzymes at this point are saturated with substrate and are working at the maximum possible rate (Adam-Day, 2012).

Temperature is the other factor that affects the rate of a given reaction. When the temperature is increased, the kinetic energy that is possessed by the molecules is also increased. This means that the random collisions that take place between the molecules are increased especially in a fluid media. Since the enzymes that catalyze the reactions are in a random collision with the substrate molecules, the increased collision as a result of the increased temperature results in an increase in the rate of reaction and hence increase in product formation. However, when the temperature is increased, there is an increase in the vibrational energy of the molecules. This strains the bonds that hold the molecules together and may result in the breakdown of the

bonds as the temperature increase. This mainly occurs mainly to the hydrogen and ionic bonds that are weaker, and when the breakdown of the bonds takes place in the enzyme molecules there may be changes in the shape of the active sites (Adam-Day, 2012).

The change in the shape of the active site may result in the loss of the complimentary of the shape of the active site with that of the substrate. This results in reduced catalytic activity of the enzyme. Eventually, as the temperature is increased, the enzyme will be denatured and thus will not be able to catalyze the reaction and thus a reduction in the rate of reaction. The rate of reaction, therefore, increase initially as the temperature increases because of the increased kinetic energy, but the rate reduces as the shape of the active site is deformed and finally denatured by the increased temperature. The temperature range at which there is a maximum rate of reaction is referred to as enzyme's optimum temperature. The optimum temperatures for different enzymes vary with that of the enzymes in the body being around 37°C (Adam-Day, 2012).

The other factor that affects the rate of enzyme activity is the level of pH of the medium where the enzyme activity is taking place. The term pH refers to either acidity or basicity of a solution. It measures the concentration of the hydrogen ions in the solution and usually ranges from pH1 (most acidic) and pH 14 (most basic). Low pH values are an indication of high hydrogen and low hydroxyl ions concentration. On the other hand, high pH values are an indication of low hydrogen ions and high hydroxyl ions concentration (Adam-Day, 2012).

The presence of hydrogen and hydroxyl ions in a reaction may interfere with the ionic bonds that hold the enzyme together. This is due to the attraction

and repulsion experience that is felt by the charges that are created by the bonds. This kind of interference may result, in a change in the shape of the active site, or the enzyme as a whole. Different enzymes show variations on the pH levels at which the bonds joining them are in perfect order to provide the best shape of the active site to be most complementary to that of their substrate. This pH level offers the optimum rate of reaction and thus the pH level is known as the Optimum pH level. For instance, the enzymes such as the pepsin that are found in the stomach have an optimum pH of about pH2 due to the acid nature of the stomach caused by the presence of hydrochloric acid (Adam-Day, 2012).

An increase or decrease in the pH level from the optimum pH may result in a decrease, in the rate of the reaction as the shape of the active site will in most cases be at a state that is not complementary to that of the substrate that they catalyze. Small changes that may take place in the level of the pH do not result in a permanent change to the shape of the enzyme as the affected bonds are reformed once the optimum pH level is retained.

However, extreme pH changes may result in the enzyme being denatured causing a permanent loss of the enzyme activity (Florida State University, 2013).

One of the organs in the body that has most of the enzymatic activities taking place is the liver. The numerous enzymes found in the liver help in facilitating the different kinds of biological activities to work at an optimum rate. Extremely low or high levels of pH result in total loss of enzyme activity. The level of pH also determines the stability of enzymes and the optimum pH is necessary for an enzyme to work effectively. This optimum pH value varies in a great way from one enzyme to another. The catalase enzyme has an

optimum pH level of 7.0. It thus requires the environment to have a pH of about 7 for it to work effectively. The vinegar that is mostly used in medical laboratories and for cleaning purposes such as baking, cooking, as well as meat preservation, has a pH level 2.4.

The liver employs huge amount of specialized enzymes in breaking down toxic substances in order to make them harmless for the body to process. One of the enzymes in the liver is the catalase enzyme, which is involved in breaking down the toxic hydrogen peroxide into harmless water and oxygen. Catalase enzyme enables the breakdown of hydrogen peroxide to oxygen and water efficient and thus reduces the harm that would be caused by the accumulation of hydrogen peroxide. The enzyme is, therefore, useful in breaking the hydrogen peroxide down to harmless products. Occurrence of this reaction is confirmed by the presence of bubbles that escape creating foam (Scientific American, 2013)

This experiment was aimed at determining the effect of acidic fluid on the enzymatic activity of the catalase enzyme. The experiment used the acidic fluid, which was provided for by vinegar as the independent variable while the enzymatic activity of catalase enzyme was taken to be the dependent variable.

Research Question

The experiment was done in order to find out whether acidic fluids such as vinegar have an effect on the activity of catalase enzyme, catalase.

Research Hypothesis

The hypothesis that was to be tested in the experiment was that acidic fluids have an effect on the activity of catalase enzyme.

Methodology

Materials

The requirements for the experiment were collected and set ready for the experiment. These requirements were unprocessed and uncooked liver, beakers, a pen, a knife, distilled water, hydrogen peroxide solution and vinegar. Two beakers were labeled using a pen as Distilled Water and Vinegar respectively. The unprocessed and uncooked liver was to be used as the source of catalase enzyme. The beakers were used to setup the two experiments and a pen was used to label the beakers. A knife was to be used for cutting the liver. Distilled water was used to offer the neutral environment to the enzyme while the vinegar was used to offer an acid environment to the enzyme. Hydrogen peroxide solution was used to provide the substrate for the catalase enzyme.

Procedure

Using the knife, the liver was cut into small pieces and divided equally into the two beakers. In each beaker, 40 ml of distilled water and blended and the extract kept frozen. In the beaker labeled distilled water, 0.5 ml of hydrogen peroxide added, and the appearance of bubbles observed. In the beaker labeled vinegar, 5 ml of vinegar was added followed by 0.5 ml hydrogen peroxide.

Data Collection and Analysis

Data collection was done by monitoring the number of bubbles produced in each of the two containers. This was done by observing whether there were any bubbles that were produced after hydrogen peroxide was introduced

into each of the two samples. The data was analyzed by comparing the presence of bubbling in the two experiments.

Results

The appearance of bubbles after the addition of hydrogen peroxide to each of the beakers was monitored and the presence or absence of the bubbles recorded in Table 1.

Discussion

The liver samples that were used contains catalase enzyme that works in degrading hydrogen peroxide to form water and oxygen. When hydrogen peroxide is introduced in the beaker containing the liver sample, a degradation process is supposed to take place as the hydrogen peroxide is degraded to produce water and oxygen molecules. When the oxygen molecules are produced, they escape into the air and this result in the bubbles that are observed. The presence of bubbles in the sample with distilled water indicated production of oxygen from the hydrogen peroxide breakdown. The neutral pH of distilled water is similar to the optimum pH of catalase enzyme, which is pH 7 and, therefore, favorable for the activity of the enzyme. Distilled water did not affect the stability of the catalase enzyme and thus the structural configuration of the active site was favorable for substrate binding.

In the beaker that had vinegar, there were no bubbles that were observed. This is due to the acidic property of vinegar which does not provide the necessary environment for the activity of catalase enzyme. The acidic nature of the vinegar may have inactivated the enzyme by destroying the stability of the enzyme (Worthington Biochemical Corporation, 2013). The wrong pH

also destroys the right structural configuration for the active site of the enzyme (Adam-Day, 2012). This affects the binding of the substrate on the active site of the enzyme and hence no enzymatic activity.

Introduction of vinegar into the sample introduced hydrogen ions into the reaction. This resulted in the interference of the ionic bonds that hold the catalase enzyme together. This is due to the attraction and repulsion experience that is felt by the charges that are created by the bonds. This kind of interference resulted in a change in the shape of the active site of the catalase enzyme, or to the catalase enzyme as a whole. This pH changes finally result in the catalase enzyme being denatured causing a permanent loss of the enzyme activity (Adam-Day, 2012). This, therefore, resulted in the lack of bubbling in the beaker that has the vinegar as the degradation of the hydrogen peroxide, which is the substrate for the catalase enzyme, did not take place.

Different enzymes show variations on the pH levels at which the bonds joining them are in perfect order to provide the best shape of the active site to be most complementary to that of their substrate. This pH level offers the optimum rate of reaction and thus the pH level is known as the Optimum pH level. For instance, the enzymes such as the pepsin that are found in the stomach have an optimum pH of about pH2 due to the acid nature of the stomach caused by the presence of hydrochloric acid (Adam-Day, 2012).

Conclusion

Enzymes being proteins in nature work best at optimum condition in terms of temperature, substrate concentration and pH. When there is an increase or decrease from this optimum range, the enzyme activity is decreased. From

the results of this experiment, it can be concluded that, acidic fluids have an effect on the activity of the catalase enzymes. It can, therefore, be said that the experiment supported the hypothesis that acidic fluids have an effect on the activity of the catalase enzymes. This was due to the reduced pH level that was caused by vinegar beyond the optimum pH level for the catalase enzyme. It is thus necessary to have the optimum pH level for an enzyme in order to realize the optimum enzymatic activity.

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