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BIO 2312, Tuesday 2:30-5:00

# Chemical Breakdown of Food Stuff

**Introduction:** The main function of the digestive system is to break down food into smaller particles so that nutrients can be absorbed. To ensure that the body receives the nutrients it needs to maintain homeostasis, there are two processes involved in digestion: mechanical and chemical digestion. Mechanical digestion is the more physical part of digestion, consisting of chewing, churning, mixing, and segmentation. The second process of digestion is chemical digestion. It consists of catabolic processes that break down food particles using enzymes. This lab will focus on the chemically induced aspect of digestion.

Enzymes are protein molecules that react with substrates, which are organic food molecules. Each enzyme hydrolyzes only one or a small group of substrate molecules, and specific environmental conditions are necessary for it to function optimally (Marieb, 2011). This lab consists of three different enzymes and their substrates, The first chemical reaction that will be examined is the hydrolysis of starch by amylase. Amylase is an enzyme found in the saliva that initiates chemical break down as soon as the food has been ingested. The second chemical reaction includes the hydrolysis of proteins by trypsin. Trypsin is an enzyme synthesized by the pancreas to break down protein molecules. The third chemical reaction involves the hydrolysis of lipids such as litmus cream by lipase. Lipase is a protein that breaks down fats and oils into small molecules.

The objective of this lab activity was to investigate the interactions between digestive enzymes and their substrates. The substrate and enzymes pairing consisted of lipase on lipids; trypsin on proteins; and amylase on starch.

**Materials and Methods:** To test for the activities of amylase, 3gtts of distilled water were added to tubes labeled 1A, 2A, and 3A. 3gtts of starch were added to tubes 5A and 6A. 3gtts of amylase

were added to tube 4A. Tubes 1A, 2A, 3A, and 5A were placed on a rack in the 37 degrees Celsius water bath, which is the normal body temperature. Tube 4A is boiled for four minutes and Tube 6A will be remain at the temperature of 0 degrees Celsius. After incubating for an hour, a pipet was used to transfer the sample from each test tube to a numbered location on the spot plate. After the sample was placed, a drop of iodine solution was placed as well.

To test for the activities of trypsin, 3 gtts of distilled water were added to tubes 1T and 2T. 3gtts of trypsin were added to tubes 1T, 3T, 4T, and 5T. Additionally, 3 gtts of BAPNA were added to tubes 2T, 3T, 4T, and 5T. the 3gtt of trypsin in 3T should be boiled for four minutes and Tube 5T should remain at 0 degrees Celsius.

To test for the activities of lipase, 5 gtts of distilled water added to tubes 1L and 2L. 5 gtts of litmus cream were added to tubes 2L, 3L, 4L, 5L, 4B, and 5B. 5 gtts of pancreatin were added to tubes 3L, 4L,5L,4B, and 5B. Additionally bile salts were placed in tube 4B and 5B.

**Results**

**Table 1: Salivary Amylase Digestion of Starch**

| **Tube No.** | 1A | 2A | 3A | 4A | 5A | 6A |
|---|---|---|---|---|---|---|
| **Additives (3gtts each)** | Amylase, Water | Starch, Water | Maltose, Water | Boiled Amylase, Starch | Amylase, Starch | Amylase, Starcb |
| **Incubation Condition** | 37°C | 37°C | 37°C | 37°C | 37°C | 0°C |
| **IKI Test (color change)** | No color | Color change | No color | Color change | No color | Color change |
| **Result (+) or (-)** | Negative | Positive | Negative | Positive | Negative | Positive |
| **Benedict's Test (color Change)** | No color | No color | Color change | No color | Color Change | No color |
| **Results (+) or (-)** | Negative | Negative | Positive | Negative | Positive | Negative |

| Table 2: Trypsin Digestion of Proteins | | | | | |
|---|---|---|---|---|---|
| **Tube No.** | **1T** | **2T** | **3T** | **4T** | **5T** |
| **Additive's (3 gtts each)** | Trypsin, Water | BAPNA, Water | Boiled Trypsin, BAPNA | Trypsin, BAPNA | Trypsin, BAPNA |
| **Incubation condition** | 37 °C | 37°C | 37°C | 37°C | 0°C |
| **Color Change** | No color | No color | Color change | Color change | No color |
| **Results (+) or (-)** | Negative | Negative | Positive | Positive | Negative |

| Table 3: Pancreatic Lipase Digestion of Fats | | | | | | | |
|---|---|---|---|---|---|---|---|
| **Tube No.** | **1L** | **2L** | **3L** | **4L** | **5L** | **4B** | **5B** |
| **Additives (5 gtts of each)** | Pancreatin, Water | Litmus Cream, Water | Boiled Pancreatin, Litmus Cream | Pancreatin, Litmus Cream | Pancreatin, Litmus Cream | Pancreatin, Litmus Cream, Bile Salts | Pancreatin, Litmus Cream, Bile Salts |
| **Incubation Condition** | 37°C | 37°C | 37°C | 37°C | 0°C | 37°C | 0°C |
| **Color Change** | No color | No color | No color | Color change | No color | Color change | Color change |
| **Results (+) or (-)** | Negative | Negative | Negative | Positive | Negative | Positive | Positive |

**Discussion:** The amylase digestion test showed that amylase is only able to digest starch at normal body temperature of 37°C. When the amylase is boiled, as we saw in test tube 4A, it becomes denatured and therefore does not digest starch. The enzyme is becoming inactive when it is incubated at 0°C. These findings indicate that amylase requires specific temperatures to be able to digest starch. The IKI test performed in activity 1, is aimed to identify the presence of starch. In solutions with starch and amylase interacting at normal body temperature, the solution will be negative and lack a color change because the starch would have been completely digested. On the other hand, when starch is paired with any other additive such as, water,

maltose, or amylase at temperatures other than 37°C, the final solution will be positive and experience a color change because starch has not been digested by an active enzyme. Benedict's test was also performed in activity 1 with a goal of identifying if sugar is in any of the samples. Test tube 3A tested positive and experienced a light blue color change because its sample included maltose, which is a sugar. Additionally, test tube 5A tested positive for the Benedict test because amylase hydrolyzes starch, breaking it down into sugar.

In activity 2, we see enzyme trypsin interacting with substrate BAPNA. In tubes 3T and 4T there has been a color change from clear to yellow. The color change signified the hydrolysis of BAPNA within the sample. Tube 2T remained colorless because it lacked an enzyme. Without the addition of an enzyme, hydrolysis of the substrate BAPNA could not occur. In tube 3T, since the trypsin has been boiled it has been denatured so it will not actively hydrolyze the substrate. Lastly in tube 5T, there was light tint of yellow indicating that some BAPNA has been hydrolyzed but since it was incubated at a very low temperature, the enzyme is not as active as it they usually are at normal body temperature.

In activity 3, the pancreatic lipase digestion test shows that lipase can digest lipids such as litmus cream under specific temperatures. When the pancreatin is boiled, it does not hydrolyze its substrate because the temperature has caused it to denature. Pancreatin is also inactive when it is incubated at 0° C, preventing the digestion of the fat. However, when the pancreatin is incubated at 37°C, it actively digest litmus cream. Also, bile salts assists the enzyme in the digestion of fats into smaller molecules. In tubes 4B and 5B, although they were incubated at different temperatures, digestion still occurred due to the presence of the bile salts. Overall, throughout this experiment enzymes depend on certain temperatures to be able to hydrolyze its

substrates. When the temperature is different than normal body temperatures, the enzyme may become denatured or inactive, causing digestion to be inhibited.

Works Cited

Marieb, Elaine N., and Elaine N. Marieb. “Human Anatomy and Physiology Laboratory Manual, Fetal Pig Version 12th Edition (9780134019949).” *Human Anatomy and Physiology Laboratory Manual, Fetal Pig Version 12th Edition (9780134019949) - Textbooks.com*, Vitalsource Technologies, Inc., 1 Jan. 1970, https://www.textbooks.com/Human-Anatomy-and-Physiology-Laboratory-Manual-Fetal-Pig-Version-12th-Edition/9780134019949/Elaine-N-Marieb.php.