

Lab Report 3

Introduction:

Enzymes are biological catalysts that speed up chemical reactions during physiological conditions (in humans and most animals these conditions are relatively mild, including a temperature of 37°C and a pH close to 7.) Enzymes are not destroyed in the reaction and can be used again and again. For example, the enzyme sucrase will break down table sugar (sucrose) into glucose and fructose.

Salivary amylase in saliva + starch → ES complex → salivary amylase + glucose and fructose

An enzyme is a protein whose activity (ability to catalyze a specific reaction) is affected by exposure to certain environmental factors, including excessive hot or cold temperatures, pH, radiation, electricity, and certain chemicals. Enzymes function best within a specific chemical environment – known as its optimum environment.

The digestive enzyme in salivary secretions is called salivary amylase. This enzyme initiates the digestion of complex carbohydrates (starch). Salivary amylase breaks the chemical bonds between some of the monosaccharides in starch to reduce this long chain polysaccharide to maltose disaccharides.

Salivary amylase in saliva + starch → ES complex → salivary amylase + many maltoses

The first purpose of this exercise will be to investigate the optimum conditions of the enzyme, diastase, which is a type of amylase. You will be introduced to the concepts of a controlled scientific experiment which is part of the Scientific Method. (We do not do the second purpose which is a computer program which we do not have.)

Materials:

Prepared enzyme solution (1-2% diastase) and 1-2% boiled starch solution

Test tubes, test tube rack, masking tape, permanent ink pen

IKI solution (starch test), Benedict's solution (glucose or maltose test), 95°C water bath, water baths (ice, 37°C, 100°C)

Regular graph paper

- 1.) Amylase activity and effect of heat: Summarize the results of your enzyme experiments by using the following +/_ notation to complete the tables below:

+++ Strong positive
++ Moderately positive
+ Weak positive
- Negative

Control:	Starch (IKI)	Maltose (Benedict's)
#1 1 ml DW in tube 1a/equilibrate* in 37°C bath 5 ml DW in tube 1b/equilibrate* in 37°C bath quickly add DW to DW/swirl to mix incubate 60 min in 37°C bath	-	-
#2 1 ml DW in tube 2a/equilibrate* in 37°C bath 5 ml starch solution in tube 2b/equilibrate* in 37°C bath quickly add DW to starch solution/swirl to mix incubate 60 min. in 37°C bath	+++	-
#3 1 ml amylase solution in tube 4a/equilibrate* in ice bath 5 ml starch solution in tube 4b/equilibrate* in ice bath quickly add amylase to starch solution/swirl to mix incubate 60 min. in 37°C bath	-	+++
#4 1 ml amylase solution in tube 3a/equilibrate* in 37°C bath 5 ml starch solution in tube 3b/equilibrate* in 37°C bath quickly add amylase to starch solution/swirl to mix incubate 60 min. in 37°C bath	-	+++
#5 1 ml amylase solution in tube 5a/equilibrate* in 100°C bath 5 ml starch solution in tube 5b/equilibrate* in 100°C bath quickly add amylase to starch solution/swirl to mix incubate 60 min. in 100°C bath	+++	+++

2.) Tube 1b represents a control in this experiment. Explain what you learned by testing the distilled water in test tube 1b.

No reaction in the control.

3.) Test tube 2b is another control. Explain in your own words what you learned by testing the starch solution in test tube 2b.

+ for starch without sugar

4.) Test tubes 3b, 4b, and 5b represent experimental variables. Based on the results of these tubes, what is your conclusion concerning the effect of heat on the activity of the diastase enzyme?

0°C – no difference/starch/maltose (should have had just a slight positive)

37°C – no difference/starch/maltose (this is the ideal temp with a strong positive)

100°C – higher reaction/starch (should have been negative because the enzyme would have denatured)

- 5.) Most enzymes will not properly catalyze a reaction when they are either below or above their optimum temperature. Use proper terminology to explain what happens to an enzyme when a) it is too cold and b) it is too hot.
- a.) If it is too cold, the reaction decreases or may even stop. As the temperature decreases, particles (or atoms) of matter slow down in their movements, causing them to collide less frequently and with less force or not at all.
 - b.) If the temperature is too hot, the particles rate of movement would increase, causing more collisions at a faster rate, Also, the enzymes can denature, causing them to not react at all. It could unravel and/or lose its shape.

We were supposed to use graph paper to show Theoretical Amylase Activity. We were to copy what the instructor wrote on the board. We did this, but we were still marked wrong for our answers, even though we all had the same thing.