Digestion of starch normally begins in the mouth of humans where an enzyme, salivary amylase, is secreted, catalyzing the break up of the starch by hydrolysis. Starch is a polymer, made from thousands of sugar molecules joined chemically into one giant chain-like molecule. Since the starch molecule is made from sugar molecules, we call it a polysaccharide.

Enzymes have specific shapes that enable them to only bind to certain molecules, or substrates. The enzyme and substrate fit together like a lock and key, which allows the reaction to take place. An enzyme can only work on a substrate that it can bind to. The area where an enzyme binds to a substrate is called an active site. An enzyme is not destroyed or used up during the reaction. After a reaction the enzyme is available to repeat the process again.

Salivary amylase is an enzyme which catalyzes the hydrolysis of the polysaccharide starch (substrate) to the disaccharide maltose. Salivary amylase is produced by the salivary glands. If amylase is added to a solution of starch, the starch will be digested to form the sugar, maltose.

<table>
<thead>
<tr>
<th>SUBSTRATE</th>
<th>ENZYME</th>
<th>PRODUCTS</th>
</tr>
</thead>
<tbody>
<tr>
<td>Starch</td>
<td>Amylase</td>
<td>maltose + maltose + maltose...</td>
</tr>
</tbody>
</table>

Enzymes perform best at their optimal temperatures and pH. Temperatures that are too high or too low, as well as a pH that is too acidic or basic can change the shape of the protein in the enzyme, and therefore, the enzyme’s effectiveness. For example, the rate of this reaction is best if it occurs at body temperature (37° C) and in a neutral pH. The progress of the reaction can be visualized by testing for the disappearance of the substrate (starch) using an iodine test.

Materials

- Salivary amylase
- distilled water
- 5 test tubes
- test tube rack
- tape
- marker
- 6 pipettes
- straw
- starch agar Petri dish
- incubator
- iodine solution – day 2
Procedure:

**DAY 1**

1. Obtain all materials. (keep Petri dish covered as much as possible)

2. Label the test tubes (F.S., 1/10, 1/100, 1/1000, and 1/10000)

3. Label 5 pipettes using the same codes as listed above. (the extra is for distilled water)

4. Have one person salivate 2 ml of saliva into the test tube labeled FS (full strength).

5. Perform SERIAL DILUTION

   1. Place 9 mL of distilled water in each remaining test tube. (except for the Full Strength test tube containing the saliva)
   2. Using the pipette labeled FS transfer 1 ml of full strength saliva to the test tube labeled 1/10.
   3. Swirl the test tube to mix the solution well.
   4. Using the pipette labeled 1/10, transfer 1ml of 1/10 solution to the test tube labeled 1/100.
   5. Swirl the test tube to mix well.
   6. Using the pipette labeled 1/100, transfer 1 ml of 1/100 solution to the test tube labeled 1/1000.
   7. Swirl the test tube to mix well.
   8. Using the pipette labeled 1/1000, transfer 1 ml of 1/1000 solution to the test tube labeled 1/10,000.
   9. Swirl the test tube to mix well.
6. Label bottom of the a Petri dishes and using the straw drill 6 wells in the agar of Petri dish (as seen in diagram below).

7. Using the non-labeled pipette, place ONE DROP of distilled water in the control well (C).

8. Using the pipette that matches each test tube, place ONE drop of each test tube dilution into the matching well.

9. DO NOT TILT plates; you will spill the solutions out of the wells.

10. Carefully place plates in an incubator set at body temperature (37° C)

DAY 2

1. Remove Petri dish from incubator.

2. Pour enough iodine onto the dish to coat the agar.

3. Let sit for a few minutes.

4. Pour excess off into the sink.

5. Using a metric ruler, measure the diameter of starch digestion for each well in centimeters, rounding to the nearest tenth.

(CREATE A DATA TABLE FOR THIS.)