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Thursday Lab

September 12, 2023

Laboratory 3 – Properties of Enzyme Action

Purpose:

The purpose of this lab is to identify that the pancreatic lipase plays a significant part in the breakdown of fat, lipase on its own is unsuccessful because it attempts to act on big lipid droplets that are water insoluble. Bile salts function as emulsifying agents, breaking the fat into smaller droplets so that lipase has a wider surface area for its hydrolysis of lipids. This helps to solve the issue. Additionally, the pancreas facilitates lipid digestion by secreting sodium bicarbonate (NaHCO_3). This substance maintains the small intestine's pH at roughly 7.8, which is ideal for the pancreatic enzymes to function. You will investigate a few facets of how pancreatic lipase and bile salts affect lipids in the exercise that follows.

Procedure:

1. Add just enough litmus powder to a container of dairy cream to produce a medium blue color. Pour 3 ml of the litmus cream into 4 separate test tubes. Into two additional test tubes pour 3 ml of 2% pancreatin. Preincubate the litmus cream and the pancreatin separately in a 37° C water bath for 5 minutes. Then prepare four test tubes as follows:

Tube #1:	3 ml cream	+	3 ml pancreatin	
Tube #2:	3 ml cream	+	3 ml distilled water	
Tube #3:	3 ml cream + 3 ml pancreatin + pinch of bile salts			
Tube #4:	3 ml cream + 3 ml distilled water + pinch bile salts			

2. Gently shake each tube for 30 seconds to mix in the bile salts. Incubate all four tubes in a 37° C water bath for 1 hour, checking every minute for the first 5 minutes or until the first tube changes color, then every 15 minutes for the rest of the hour. Record the time and number of the tube. Continue checking for the remainder of the hour.

3. Remove the tubes from the water bath. Test the pH of each tube using pH paper and note the odor and color of each tube.
NOTE: Blue litmus will turn pink in an acid environment.

4. Summarize the results in the following table: Tube Color pH Odor #1#2#3 #4

Time to change color

5. Explain how the digestion of fat affects the pH of the solution and how bile affects the rate of digestion.

Results:

Tube	Color	pH	Odor	Time Change
#1	Lavender/Pink	7	Sweet	30 Mins
#2	Purple	8	Shaved Metal	30 mins
#3	Light Purple	7	Stinky	30 mins
#4	Dark Purple	8	Charcoal	30 mins

Discussions:

The lipids begin converting into fatty acids when the pancreatin and bile salts are added to the solution. Since the breakdown will cause a low pH, we tested to track the pH shift.

Conclusions:

In conclusion, the bile salts help overcome the problem by acting as the emulsifying agent, which breaks down fat into smaller droplets. So that the lipase has a large surface area for its hydrolysis of fats.