LABORATORY3–PROPERTIES OF ENZYME ACTION

Purpose

Understand some aspects of the action of pancreatic lipase and bile salts on lipids.

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. Preincubatethe litmus cream and the pancreatin separately in a 37C 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 37C 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.

Result

4. Summarize the results in the following table:

Tube	Color	pH	Time to change color
#1 pan	Ombre blue to purple	7	20 min
#2 H2O	GogurtPurple/Dark Bottom	8.5	10 min
#3 pan+bile	GogurtPurple/Dark Bottom	5.5	20 min
#4 H2O+bile	GogurtPurple/Dark Bottom	8	20 min

Discussion

Litimus is an agent used to identify acidic or basic pH turning blue in basic solutions and red in acidic solutions. For Tube #1, the digestion of fat from pancreatin caused the pH to go to 7 which is neutral explaining the lack of color change. The ombre blue to purple color indicates lipase beginning to hydrolyze, but not completely because this isn't an optimal pH to do so. Tube #2 contained only water and cream leaving a purple color, the 8.5 pH is too basic to be optimal for fat digestion. Tube #3 with pancreatin and bile salt had a pH of 5.5 after incubation, the color was similar to the other tubes which caused confusion as to if the litmus powder was giving valid results, but the pH indicates an

acidic solution which is optimal for digesting fats. The last tube #4 contained water and bile salt, and the pH was 8, while the color looked the same as the previous 3.

Conclusion

The digestion of fats causes the pH to drop making an acidic solution.