Experimental Design for Part B of the Enzymes lab (Lab 2)

Science is far more than following a recipe or procedure. The process of science involves exploration and discovery, testing ideas and gathering data, relies on community and benefits society. In Biol 107 we feel it is very important that students are given an opportunity to practice real science, beyond following a procedure in the lab manual. Part A of the lab walks you through how we can measure the rate of lactase activity in nmol/min using the equipment you learned how to use in Lab 1. Make sure you read the lab manual and watch the "Coming Prepared" video. For Part B, you will design an experiment to gather data about a real-life scenario applying the same technique as in Part A. There are 4 scenarios outlined in Part B of the Lab 2 lab manual. With your group, choose which scenario you want to base an experiment on, and with your group, fill out the following information, which is due **48 hours before** your on campus Lab 2. With 4 people in your group, each person should plan to do 3-4 reactions for Part B. This will allow you to create a robust experiment and also include replicates. Replicates are required as you will be using this data in your Unit 2 assignment. Although you are sharing data, the **Unit 2 assignment is independent work.**

Make sure you put your r**aw data and rate data** in this shared google doc so that all members of the group have access to the data. Students who do not enter their data within 48 hours of the lab may have a 5 mark deduction applied to their Unit 2 Assignment. It is also a good idea to take a picture of your teammates' data before you leave the lab.

In the space below, summarize how lactase enzyme activity is measured:

Lactase enzyme activity is measured through the reaction between glucose and galactose, but that is difficult to measure, so we use ONPG as a replacement.

Research question:

How does the level of pH in the stomach hinder the effectiveness of the enzyme lactaid in the small intestine?

Hypothesis:

It will be more effective in pH 8 after the enzyme has sat in pH 5 for 15 minutes because at pH 2 the enzyme may become denatured which will make it ineffective.

Independent variable:

The pH of the Lactaid (The enzyme at pH 2 and pH 5)

Dependent variable:

Effectiveness/condition of Lactaid at certain pH

Reaction rate is the dependent variable!

Control:

pH 2 and 5

Constants:

Time that enzyme sits in solution

Enzyme

pH 8

Details of procedure:

Each member of the group will carry out the same procedure therefore there are 4 replicates. This will ensure that we get the most accurate result using the standard deviation and mean. The procedure involves finding the concentration of the enzyme after crushing up the lactaid and letting it sit in ~1mL of pH 2 and ~1mL of pH 5 for 15 minutes. This will then be followed by finding the concentration of each in ~1mL of pH 8. The concentrations will be found by using a spectrophotometer.

Division of labour:

Below is an example of a division of labour chart. You can discuss with your team the pros and cons of one person doing all the replicates for one reaction versus having different people do the replicates. Fill out who is doing what for your experiment using the table below the example table. You can delete the example in your experimental design google doc.

	Kathy	Amy	Vikki	Monica
Reaction Tube 1	Enzyme kept at 65℃	Enzyme kept at 65℃	Enzyme kept at 65℃	Enzyme kept at 65℃

Reaction Tube 2	Enzyme kept at room temp	Enzyme kept at room temp	Enzyme kept at room temp	Enzyme kept at room temp
Reaction Tube 3	etc			
Reaction Tube 4				

Each person will measure absorbance every minute for 5 minutes then convert absorbance values into concentration. Then we will graph concentration over time so they can calculate the rate of reaction (nmol/min) using slope that includes the 0 reading (refer to the screencast for how to do this). We will put our raw data and rate data in our shared google doc so that all members of the group have access to the data.

Division of labour:

	Sehaj	Guntaas	Anisha	Purva (0 mins)
Lactaid in pH 2	8.89nM	-4.44 nM	22.2nM	3.3333nM
Lactaid in pH 5	2.22nM	1.11 nM	66.67nM	4.4444nM
Lactaid pH 2 in pH 8	17.78nM	10 nM	33.33nM	-6.6666nM
Lactaid pH 5 in pH 8	22.22nM	8.889 nM	11.11nM	26.667nM

Results: (Each member of the team must make a data table and enter their data within 48 hours after the lab)

Purva

Concentration = Absorbance/slope = (0.0009)/lactaid in pH(from above table)