


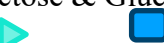



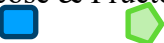
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Looking Into Lactase

Understanding Enzyme Specificity and Activity

In our bodies we have many processes to help us digest, or break down, our foods. Our teeth help to break the food into small pieces and our stomach acid can break down the food even further.

When it comes to foods that have sugar, our bodies need to break the food down to the simplest sugars. One example is fructose, which is found in many fruits. But not all foods are made solely of simple sugars. Many foods are made of larger sugar molecules that contain combinations of simple sugars. For example, sucrose is a sugar molecule that is made of two sugars, glucose and fructose. This complex sugar is actually the same as the table sugar you have at home.

| TABLE 1: Carbohydrate Breakdown in Cow's and Soy Milk | | |
|---|---|--|
| Milk | Sugar in the Milk | Simplest Sugar |
| Cow | Lactose  | Galactose & Glucose  |
| Rice | Glucose  | Glucose  |
| Soy | Sucrose  | Glucose & Fructose  |

To break down sugars into their simplest sugars, our bodies use enzymes. Enzymes are proteins that are made by the body specifically to break down molecules very quickly. Each enzyme is specific to a certain molecule. For example, the complex sugar lactose, which is found in dairy products, can only be broken down by the enzyme lactase to break the molecule into galactose and glucose, the simplest sugars.

If you think of your body like a big kitchen, the enzymes are like the chefs; they take components and turn them into something else. And to keep the kitchen efficient, every chef has a specific job. One person might be in charge of chopping vegetables, one person might make sauces, and another person might cook meats on a grill. Just as each chef has a specific job, so do the enzymes.

Today you will be working in the quality control department of a local bioscience company that produces a lactase enzyme product used to treat lactose intolerance. You will test to see that the enzyme is specific to lactose by testing it with cow's, rice, and soy milks. You will also determine the best pH conditions for the lactase enzyme product.



QUICK CHECK: What is an enzyme?

MATERIALS

- | | | | |
|---------------------|---------------------|--------------------------------|------------|
| Glucose test strips | Micropipette & tips | pH buffer solutions | Cow's milk |
| Lactase solution | Test tubes (x5) | Unidentified milk samples (x3) | |

PART I – Identify the cow's milk

1. Locate the three test tubes that contain the unidentified milk samples, and predict which milk is in each tube using qualitative observations.

| TABLE 2: Qualitative Milk Analysis | |
|------------------------------------|--|
| A | |
| B | |
| C | |

2. Locate the glucose test strips. Glucose test strips are used to test for glucose levels and will change color in the presence of glucose.
3. Label the three glucose test strips A, B, and C. Dip each strip into its corresponding test tube and immediately place each strip on your paper towel. After ten seconds, record any color change and the relative amount of glucose present in the table below.

| TABLE 3: Quantitative Milk Analysis | | |
|-------------------------------------|------------------|------------------------------------|
| | Test strip color | Relative amount of Glucose (mg/dL) |
| A | | |
| B | | |
| C | | |

4. Mix the lactase enzyme by inversion. Add 250µL to each test tube.
5. Locate your mini-vortex and set the dial to eight. Vortex each tube for two seconds.



QUICK CHECK: Predict which type(s) of milk will test positive for glucose after adding the enzyme.

6. Label three new glucose test strips A, B, and C. Dip each strip into its corresponding test tube and immediately place each strip on your paper towel. After 10 seconds, record any color change, the relative amount of glucose present, and whether or not the enzyme was active in the sample.

| TABLE 4: Quantitative Milk Analysis – Enzyme Activity | | | |
|---|------------------|------------------------------------|-----------------|
| | | Post-Lactase Glucose Test Strip | |
| | Test strip color | Relative amount of Glucose (mg/dL) | Enzyme Activity |
| A | | | 0 |
| B | | | 0 |
| C | | | 0 |



QUICK CHECK: Based upon your analysis, what type(s) of milk did the enzyme effect and why is it used to treat lactose intolerance?

PART II – Determine the optimal pH condition for the lactase enzyme.

7. Locate the five (5) empty test tubes. Label the test tubes **2, 4, 7, 10, and 12**.
8. Locate the pH buffer solutions labeled 2, 4, 7, 10, and 12. Add 500µL of each pH buffer to the corresponding test tube.
9. Locate the lactase enzyme and mix by inversion.
10. Add 250µL of the lactase enzyme to each test tube.
11. Add 500µL of cow’s milk to each test tube. Vortex each tube for two (2) seconds.
12. Label five new glucose strips 2, 4, 7, 10, and 12 and dip the labeled strips into the appropriate test tubes. Immediately lay the test strips on the paper towel. After ten seconds, record your observations in Table 5.

| TABLE 5: Enzyme Activity at Different pH Levels | | |
|---|---------------------|---|
| | Color of Test Strip | Relative amount of Glucose (mg/dL) after adding lactase |
| pH 2 | | |
| pH 4 | | |
| pH 7 | | |
| pH 10 | | |
| pH 12 | | |

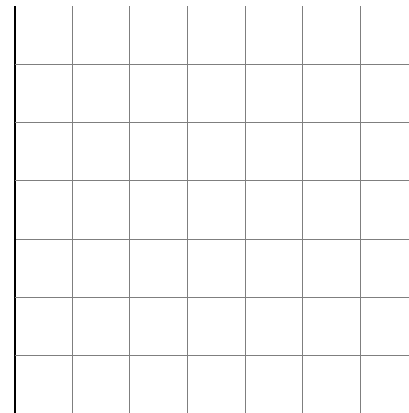
PART III – Data analysis

Graph the relative amount of glucose to the corresponding pH on the graph. Be sure to give your graph a title and label the axes.

What is the independent variable?

What is the dependent variable?

What effect (if any) does pH have on lactase activity?



PART IV - Conclusion

Label each section of the digestive tract with its specific pH. Compare the results found in PART III to the pHs found in the digestive tract. In what part of the digestive system would the enzyme work best?

