MILK—HOW SWEET IS IT?

INTRODUCTION

In the film, *The Making of the Fittest: Got Lactase? The Co-Evolution of Genes and Culture*, Dr. Dallas Swallow determines that Dr. Spencer Wells is able to digest lactose, the main sugar in milk, by having him rapidly drink a liter of milk and then measuring glucose levels in his blood. Why would measuring blood glucose levels tell you whether someone can digest lactose?

In the intestine, the lactose in milk is broken down into glucose and galactose by the enzyme lactase. These two simpler sugars are absorbed from the intestine into the bloodstream. As a result, if the lactase enzyme is present, blood glucose levels increase within 20 to 60 minutes of drinking milk.

Not all human adults, however, produce lactase—in fact, most people worldwide don’t. They are referred to as lactose intolerant or lactase nonpersistent. When lactose-intolerant individuals drink milk, the lactose passes undigested through the small intestine into the large intestine and may cause gas, bloating, and sometimes diarrhea. A significant minority of adults worldwide can digest milk without any problems; they are lactose tolerant or lactase persistent.

In this activity, you will test glucose levels in four “patient” samples to determine whether they are lactase persistent or nonpersistent. Instead of using blood samples, as Dr. Swallow did in the film, you will measure glucose in samples of milk to which you will add fluids that represent the contents of the small intestines of the four patients.

MATERIALS

- 12 30-mL plastic medicine cups
- 1 permanent marker
- 18 glucose reagent test strips
- 1 glucose color chart
- 6 plastic stirring rods or coffee stir sticks
- 1 graduated cylinder that measures up to 5 mL (optional)
- 1 timer (optional)
- 1 clock or watch
- paper towels
- colored pencils

You will need to measure the following solutions:

- 4 5-mL samples of patients’ intestinal fluid
- 1 5-mL sample of water
- 1 5-mL sample of lactase solution
- 6 5-mL samples of milk
PROCEDURE

Setting up your experiment.

1. Obtain all of the materials listed above.

2. To set up the solutions in the medicine cups, follow these steps:
   
   a. Label one of the medicine cups Patient #1. Use a graduated cylinder to measure 5 mL of Patient #1 intestinal fluid and pour it into the Patient #1 cup.
   
   c. Label another medicine cup Patient #2. Use a clean (i.e., rinsed in water) graduated cylinder to measure 5 mL of Patient #2 intestinal fluid and pour it into the Patient #2 cup.
   
   d. Repeat for patients #3 and #4, each time using a clean graduated cylinder.
   
   e. Label a medicine cup “Negative Control” and add to it 5 mL of water.
   
   f. Label a medicine cup “Positive Control” and add to it 5 mL of lactase solution.

   h. Label six medicine cups “Milk” and add 5 mL of milk to each one.

   3. Remove the two pages at the end of this document and place them on your desk. The Glucose Data Table is where you will record your data. The Glucose Test Strips Chart is where you will place the wet glucose reagent test strips after you use them.

   The photo to the right shows an example of the setup.
Determining baseline (time 0) glucose levels.
5. Use six glucose test strips to test the baseline (i.e., before the experiment) glucose levels of the intestinal fluids of the four patients and the two controls. To do so follow these instructions:
   (a) Dip the test area of the glucose test strip into the Patient #1 cup and remove it immediately.
   (b) Lay the strip on the Glucose Test Strips Chart on the section labeled Patient #1, 0 min.
   (c) Wait 30 seconds. (This time may vary depending on the instructions for the particular brand of glucose test strips you are using.)
   (d) Match the color of the test area on the strip to the glucose color chart and record the glucose concentration in the Lactose Data Table, in the column labeled “Glucose levels” for time 0 min., in the row for Patient #1. (Do not wait longer than 30 seconds to record your data. The test is time-sensitive and waiting longer will affect the accuracy of your measurement.)
   (e) Repeat these steps for the remaining three patient samples, the negative control, and the positive control. Each time write your results in the “time 0” column. This is your baseline reading for each reaction.

Determining whether lactase is present in your samples.
Read all the steps below before starting the experiment. If you are working with a partner, decide who will do what. To complete your measurements in a class period, you will need to work efficiently. It will take too long to do all the steps for one sample before moving to the next one. If you are working with a partner, you can divide the samples in half and have each person be responsible for testing three. Alternatively, you can do all the samples simultaneously and one person dips in the glucose test strips at the appropriate times and the other person records the results after 30 seconds.

6. For each sample, you will carry out the following steps:
   (a) Add 5 mL of milk to the Patient #1 cup and gently stir with a plastic rod. (If you don’t have a plastic rod, you can gently swirl the medicine cup with the two solutions.)
   (b) Start your timer immediately – you need to take more measurements at 2 minutes and at 7 minutes after adding milk to your sample!
   (c) At time 2 minutes dip the glucose test strip in the mixture in the Patient #1 cup, lay the strip down on the Glucose Test Strips Chart in the Patient #1 2 min. slot, wait 30 seconds (or the time specified by your test strip manufacturer), and determine the glucose concentration using the glucose test strip. Record the glucose concentration on the Lactase Data Table in the column labeled “Glucose levels (mg/dL),” 2 min., Patient #1.
   (d) Repeat the step above at the 7 minute time point.
   (e) Follow the same procedure for all four patient samples and the two control samples. Be sure to use a different stirring rod for each sample.

The photo to the right shows an example of a setup at the end of the experiment. The colors of the test strips will vary.
ANALYSIS

1. When you were measuring patients’ samples, why did you have to use a clean graduated cylinder each time?

________________________________________________________________________________________________________________
________________________________________________________________________________________________________________

2. The chemical reaction that takes place in the positive control sample is shown below.

Lactose $\rightarrow$ Glucose + Galactose

enzyme

a. What is the source of lactose? ________________________________
b. What is the name of the enzyme that digests lactose into glucose and galactose? ____________________________
c. Which component of the chemical reaction did you measure using the test strip? ____________________________
d. What compound in the chemical reaction above is missing from the negative control sample? ________________

3. Use colored pencils to construct a bar graph based on your data. The graph should show the results of the 0, 2-, and 7-minute glucose test for all your samples, including the controls. Be sure to provide an appropriate title, labels for the x- and y-axes, and legend.

Title: ___________________________________________________________
4. a. Which patient(s) appear to be lactase persistent (lactose tolerant)? ______________________
   b. Describe the evidence supporting this claim.
      _____________________________________________________________
      _____________________________________________________________

5. a. Which patient(s) appear to be lactase nonpersistent (lactose intolerant)? ______________________
   b. Describe the evidence supporting this claim.
      _____________________________________________________________
      _____________________________________________________________

6. a. Explain why it was necessary to measure glucose levels in a sample of milk to which 5 mL of water was added (the negative control).
      _____________________________________________________________
      _____________________________________________________________
      _____________________________________________________________

   b. Explain why it was necessary to measure glucose levels in a sample of milk to which 5 mL of lactase solution was added (the positive control).
      _____________________________________________________________
      _____________________________________________________________
      _____________________________________________________________
      _____________________________________________________________

   c. Why do you need both negative and positive controls?
      _____________________________________________________________
      _____________________________________________________________
      _____________________________________________________________

   d. If the negative control had resulted in the data shown in the graph below, what would you conclude? Provide two possible explanations to account for these results.
      _____________________________________________________________
      _____________________________________________________________
      _____________________________________________________________

![Graph showing glucose levels over time](image)
7. Instead of measuring glucose levels in the milk, identify another compound you could have measured the levels of to determine whether lactase was active.

8. The test you performed using simulated intestinal fluids required 5 mL of milk and 2 to 7 minutes to obtain results. Explain why in the film Dr. Wells had to consume a liter of milk and blood glucose levels were measured periodically for 40 minutes, a much longer period of time.

9. a. Read the nutrition label on a container of milk. In addition to carbohydrates, which include lactose, list the other nutrients present.

   b. If lactose intolerance did not cause any symptoms, lactose-intolerant individuals would be able to use milk as a source of protein. Explain why it is possible for them to digest the proteins in milk even though they lack the lactase enzyme.

   c. Sucrose is a disaccharide present in many cow milk substitutes such as soy, rice, and almond milk. It is composed of glucose and fructose. Explain why lactose-intolerant individuals may be able to digest sucrose without any problems.

10. A milk allergy is the result of an immune reaction to one or more of the components of milk, such as the protein casein. Explain how an allergy to milk is different from lactose intolerance.
LACTASE DATA TABLE

<table>
<thead>
<tr>
<th>Samples</th>
<th>0 minutes (baseline)</th>
<th>2 minutes</th>
<th>7 minutes</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Glucose levels (mg/dL)</td>
<td>Glucose levels (mg/dL)</td>
<td>Glucose levels (mg/dL)</td>
</tr>
<tr>
<td>Patient #1</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Patient #2</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Patient #3</td>
<td></td>
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<tr>
<td>Patient #4</td>
<td></td>
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<td></td>
</tr>
<tr>
<td>Negative Control</td>
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<td></td>
<td></td>
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<tr>
<td>Positive Control</td>
<td></td>
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<tr>
<td></td>
<td>Positive Control</td>
<td>Patient #4</td>
<td>7 min.</td>
</tr>
<tr>
<td>------------------</td>
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</tr>
<tr>
<td>Negative Control</td>
<td></td>
<td>Patient #3</td>
<td>7 min.</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Patient #2</td>
<td>7 min.</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Patient #1</td>
<td>0 min.</td>
</tr>
</tbody>
</table>