Lactose Intolerance

Enzyme Digestion Lab Activity

Introduction

Intestinal gas, bloating, and stomach cramps—oh my! This can be a common concern for a majority of the world's population who lack the enzyme to digest certain foods. Milk and dairy products, for example, cause problems for many people who lack the enzyme required to digest lactose, the main carbohydrate found in milk. This lab activity illustrates the use of a commercial enzyme product called Lactaid™ as an aid in milk digestion.

Concepts

- Enzyme
- Disaccharide
- Metabolism

Materials

Galactose, 2 g
Lactaid™, ½ tablet,
Lactose, 4 g
Sucrose, 2 g
Yeast, suspension, 40 mL
Balloons, 4
Graduated cylinder, 10 mL
Mini soda bottles, 4
Resealable plastic bag
Water bath, 35–40 °C

Safety Precautions

Wear chemical splash goggles, chemical-resistant gloves, and a chemical-resistant apron. Wash hands thoroughly with soap and water before leaving the laboratory. Follow all laboratory safety guidelines. Please review current Safety Data Sheets for additional safety, handling, and disposal information.

Procedure

1. Obtain a warm water bath (35–40 °C) or an insulating block. Place the test tubes in the insulating block or test tube rack.
2. Weigh out the dry ingredients prior to the demonstration.
3. Review the summary diagram of the demonstration setup shown in Figure 1.
4. Clearly label each test tube as shown in Figure 1.
5. Place 2 g of the appropriate dry sugar into each test tube, as shown in Figure 1.
6. Add pre-ground Lactaid™ tablet to one flask containing 2 g lactose, as shown in Figure 1.
7. Add about 20 mL of warm (30–35 °C) tap water to the sugar in each test tube. Screw on the cap, ensuring no leaks, then shake gently until all the sugar has dissolved.
8. Add 10 mL of yeast suspension to each test tube. Swirl each test tube gently to mix the yeast and sugar together.
9. Place a balloon securely over the lip of each test tube. *Note:* Be sure each balloon is flexible and not stuck together. Inflate each balloon at least once before placing it on the test tube.

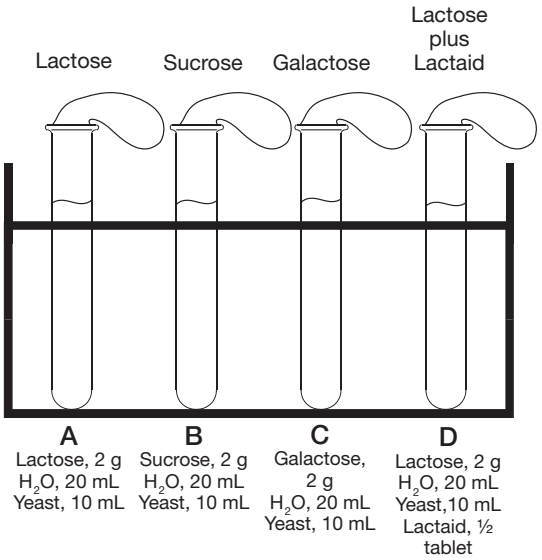

Figure 1.

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10. Place the test tube rack and test tubes in the water bath (35–40 °C).
11. Observe the flasks for 15–30 minutes, checking for the production of gas as observed in the balloons. Discuss the results and the effectiveness of Lactaid™ in the experiment.

Disposal

Please consult your current *Flinn Scientific Catalog/Reference Manual* for general guidelines and specific procedures, and review all federal, state and local regulations that may apply, before proceeding. The leftover solutions may be disposed of down the drain with excess water according the Flinn Suggested Disposal Method #26b.

Tips

- Yeast lacks the enzymes necessary to digest lactose or galactose, but it does contain the enzyme needed to digest sucrose. See Figure 2 for a summary of the demonstration results.
- The balloon in Flask B inflates rapidly due to the production of carbon dioxide from the breakdown of sucrose. Sucrose is a disaccharide consisting of a fructose and a glucose. The sucrase enzyme, also known as invertase, enzyme is present in yeast, so it is able to hydrolyze sucrose, and the glucose is immediately available for glycolysis and fermentation. It produces alcohol and carbon dioxide, and the balloon inflates.
- In Flask D, Lactaid™ effectively breaks down the lactose into glucose and galactose. The yeast can then use the glucose for fermentation, producing alcohol and carbon dioxide, which causes the balloon to inflate.
- Flasks A and C should reveal no activity, as the yeast cannot use these sugars. They lack the proper enzyme to facilitate the reaction needed to produce glucose from these sugars. The balloons should remain uninflated for the duration of the demonstration.

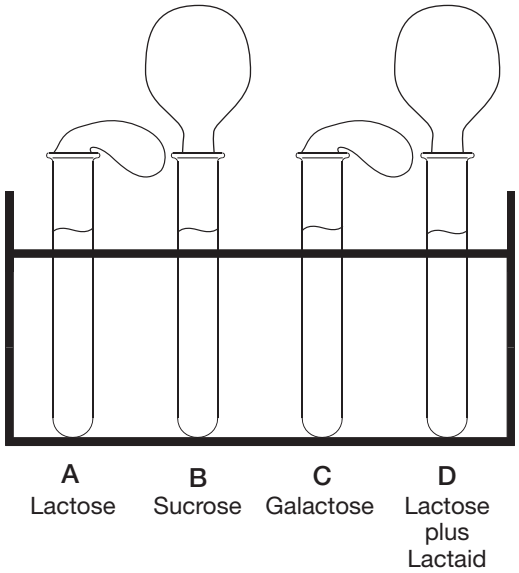

Figure 2.

- This demonstration can serve as a springboard for lab project extensions. The following research questions are easily tested using this protocol. How much lactose can a Lactaid tablet digest? How long does the Lactaid remain active? What factors influence the rate of the reaction? Are yeast able to metabolize other sugars, such as glucose and maltose? How does the rate of reaction change if the sugar is a monosaccharide rather than a disaccharide?
- This demonstration can also be used as an introduction to topics of evolution. Why is it that some continue to produce lactase after infancy while others do not? What environmental factors could have led to this evolutionary event for humans?

Discussion

Lactose (also called milk sugar) is the principal carbohydrate in milk. It is a *disaccharide*, which means that is it composed of two simple sugars or monosaccharides—glucose and galactose. Some individuals produce insufficient quantities of lactase, the enzyme required to break the bond between the two monosaccharide units in lactose. The condition in which lactase is not produced so that lactose cannot be broken down into the two simpler sugars is referred to as *lactose intolerance*. With this condition, the lactose from milk and various other milk products remains undigested and causes an increase in the osmotic pressure in the intestinal contents. Consequently, water is "drawn" from the tissues into the intestine. At the same time, intestinal bacteria may act upon the undigested lactose and produce organic acids and gases. As a result, the person may feel bloated and suffer from intestinal cramps, diarrhea, and gas.

Researchers at Lactaid, Inc. discovered a way to mass produce the enzyme lactase. Lactase converts lactose into glucose and galactose, both easily digestible monosaccharides. The mass-produced lactase is formulated into the tablet product—Lactaid™. The enzyme units in Lactaid are eaten by the lactose-intolerant person before or simultaneous to ingesting lactose. The Lactaid breaks down the lactose allowing the person's system to successfully utilize the resulting glucose and galactose.

An external test for the effectiveness of Lactaid is to find an organism that will only digest the simple sugars and not lactose. In this experiment, yeast is used as a test organism.

Yeast does not produce lactase and therefore cannot digest lactose. (It is lactose intolerant!) Yeast does, however, digest glucose very efficiently. When it digests glucose, the yeast breaks down the glucose and produces carbon dioxide gas as a waste product (Equation 1). This production of gas can easily be monitored. The absence or presence of gas production is used as evidence of digestion by the yeast.

$$C_6H_{12}O_6(aq) \rightarrow 2CH_3CH_2OH(aq) + 2CO_2(g) + \text{energy} \qquad \textit{Equation 1}$$

Reference

This activity was adapted from *Flinn ChemTopic™ Labs, Vol. 20, Biochemistry—The Molecules of Life.*; Cesa, I., Editor; Flinn Scientific, Inc.; Batavia, IL (2002).

Materials for *Lactose Intolerance* are available from Flinn Scientific, Inc.

Catalog No.	Description
FB1570	Lactose Intolerance Demonstration Kit
G0018	Galactose, 100 g
S0134	Sucrose, 500 g
L0002	Lactose, 500 g
Y0008	Baker's Yeast, Pkg. of 3 Packets (7 g each)
AP7518	Mini Soda Bottle Tubes and Rack
AP6996	Caps for Mini Soda Bottles
AP1900	Balloons, Latex, 12″, Pkg. of 20

Consult your *Flinn Scientific Catalog/Reference Manual* for current prices.