Biotechnology Science for the New Millennium by Ellyn Daugherty

Cheese Production: The Evolution of Cheese-Making Technology

(Lab 1c)

(Cat. # BTNM-1C)

Developed in partnership with

Cheese Production: The Evolution of Cheese-Making Technology (Lab 1c)

Teacher's Guide

The following laboratory activity is adapted from "Laboratory 1c: Cheese Production: The Evolution of Cheese-Making Technology" from *Biotechnology: Laboratory Manua*l by Ellyn Daugherty. For more information about the program, please visit www.emcp.com/biotechnology.

EMC Publishing®

PARADIGM EDUCATION SOLUTIONS

This kit is produced under license from Paradigm Publishing, Inc., a division of New Mountain Learning.

About Ellyn Daugherty: Ellyn Daugherty is a veteran biotechnology educator and recipient of the Biotechnology Institute's National Biotechnology Teacher-Leader Award. She is the founder of the San Mateo Biotechnology Career Pathway (SMBCP). Started in 1993, SMBCP has instructed more than 10,000 high school and adult students. Annually, 30-40 SMBCP students complete internships with mentors at local biotechnology facilities.

G BIOSCIENCES®

About G-Biosciences: In addition to the Biotechnology by Ellyn Daugherty laboratory kit line and recognizing the significance and challenges of life sciences education, G-Biosciences has initiated the BioScience Excellence™ program. The program features hands-on teaching kits based on inquiry and curiosity that explore the fundamentals of life sciences and relate the techniques to the real world around us. The BioScience Excellence™ teaching tools will capture the imagination of young minds and deepen their understanding of various principles and techniques in biotechnology and improve their understanding of various social and ethical issues.

Cheese Production: The Evolution of Cheese-Making Technology (Lab 1c)

Teacher's Guide

Cheese Production: The Evolution of Cheese-Making Technology (Lab 1c)

Teacher's Guide

Upon receipt, store the materials as directed in the package literature.

MATERIALS INCLUDED

This kit has enough materials and reagents for 8 lab groups (32 students in groups of 4).

- 8 vials of 1mg/ml Chymosin (100µl)
- 8 vials of 1mg/ml Rennin (100µl)
- 8 vials of 1mg/ml Buttermilk (100µl)
- 8 vials of water (2ml)
- 32 pieces of filter paper
- 32 empty 6ml tubes
- 8 3ml transfer pipettes
- 40 small transfer pipettes

ADDITIONAL EQUIPMENT & MATERIALS REQUIRED

- Milk (>12ml/group) in paper cup or other vessel
- Graduated cylinders or other collection vessel
- Conical Funnels
- Balances (that measure to at least 0.01 grams)
- Water bath or incubator set at 37°C with test tube rack (Optional)

SPECIAL HANDLING INSTRUCTIONS

- Store the Chymosin, Rennin and Buttermilk frozen at -20°C.

GENERAL SAFETY PRECAUTIONS

- The reagents and components supplied in the *Biotechnology: Science for the New Millennium* ™ kits are considered non-toxic and are safe to handle (unless otherwise noted), however good laboratory procedures should be used at all times. This includes wearing lab coats, gloves and safety goggles.
- The teacher should 1) be familiar with safety practices and regulations in his/her school (district and state) and 2) know what needs to be treated as hazardous waste and how to properly dispose of non-hazardous chemicals or biological material.
- Students should know where all emergency equipment (safety shower, eyewash station, fire extinguisher, fire blanket, first aid kit etc.) is located and be versed in general lab safety.
- Remind students to read all instructions including Safety Data Sheets (SDSs) before starting the lab activities. A link for SDSs for chemicals in this kit is posted at www.gbiosciences.com
- At the end of the lab**,** all laboratory bench tops should be wiped down with a 10% bleach solution or disinfectant to ensure cleanliness.
- Remind students to wash their hands thoroughly with soap and water before leaving the laboratory.

Cheese Production: The Evolution of Cheese-Making Technology (Lab 1c)

Teacher's Guide

TEACHER'S PRE EXPERIMENT SET UP

1. If available, place a test tube rack in a warmed incubator or water bath set to 37°C.
2. Aliquot 12ml milk into a suitable container for each group
3. Distribute the following items to each lab group:
    - 12ml milk
    - 1 vial of 1mg/ml Chymosin (100µl)
    - 1 vial of 1mg/ml Rennin (100µl)
    - 1 vial of 1mg/ml Buttermilk (100µl)
    - 1 vial of water (2ml)
    - 4 pieces of filter paper
    - 4 empty 6ml tubes
    - 1 3ml transfer pipet
    - 4 small transfer pipettes

TIME REQUIRED

- 30 minutes pre-lab (distribution of reagents)
- Two 1- hour lab periods (to prepare samples, run coagulation experiment, weigh dried whey and data analysis)
- 1 hour post-lab analysis

NEXT GENERATION SCIENCE STANDARDS ADDRESSED

- HS-LS1: From Molecules to Organisms: Structures and Processes
- HS-ETS1-2, 3, 6: Engineering Design

For more information about Next Generation Science Standards, visit: http://www.nextgenscience.org/

EXPECTED RESULTS

Results are dependent and variable based on several factors including temperature, agitation, and accuracy in measurement.

Data Table 1: The Characteristics of Cheese made by Different Curdling Agents

Curdling Agent	Curdling Time (min)	Weight of Cone & Curds (g)	Weight of Cone (g)	Weight of Curds (g)	Rate (mg/min)	Technician /Comments
Chymosin	15	2.0	1.0	1.0	66.67	
Rennin	30	2.0	1.0	1.0	33.34	
Buttermilk	240	2.0	1.0	1.0	4.17	
Water (-Control)	1440	2.0	1.0	1.0	0.70	

Cheese Production: The Evolution of Cheese-Making Technology (Lab 1c)

Teacher's Guide

ANSWERS TO ADDITIONAL QUESTIONS

1. In your notebook, create a chart similar to Data Table 1 and analyze the variable that impact the results of this experiment that need to be controlled if a technician wants to have confidence in the results. Include the variable to be controlled, the impact of variation, and how the impact might be minimized.

Answer: Tables will vary, but some factors to be analyzed include the following: temperature, timing, shaking, measurement of reagents, measurement of product, etc. For example:
- Shaking, so curds have time to form and are not broken up; keep still in a test-tube rack.
- Accurate measurement, so precise amount of curdling agent or milk (substrate) is added; practice pipeting and micropipeting.
- Filtering, paper absorption affects the amount of whey that filters through; use in the same way each time, and transfer curds and whey to filter more completely, possibly after centrifuging the samples.

2. In this experiment, each curdling agent is tested multiple times, and an average result is determined. Look at the class data. Does it appear that the number of replications for each curdling-agent experiment was sufficient? Yes or no? Explain your answer.

Answer: Some curdling groups contain replications with numbers that are very dissimilar. Without a formal statistical analysis, it is hard to determine exactly how many replicate samples are needed. Increasing the number of replications until the number of dissimilar values is very small will give more confidence in the averaged data.

Cheese Production: The Evolution of Cheese-Making Technology (Lab 1c)

Student's Guide

OBJECTIVES

Which curdling agent produces the cheese at the fastest rate (mass/time)?

BACKGROUND

Most commercially produced cheese (curds from milk) is made in one of the four ways:

1. Milk may simply be left to age, exposed to air and naturally occurring bacteria.
2. New batches of cheese are started with specific cultures of selected bacteria. These "known" bacteria make enzymes and acid that curdle milk. Buttermilk, yogurt and other fermented products have a good culture of *Lactobacillus* bacteria and can be used as a "starter."
3. Milk-curdling may be started adding purified enzymes (such as rennin) to milk. Rennin is a protein purified from the cells lining the stomachs of calves. Rennin is a protease, made in nursing calves, which cleave the casein protein in milk into small fragments that settle out as curds. To retrieve the calves' enzymes for commercial use, companies grind up the calf stomachs and purify the rennin enzyme from all of the other compounds made by the cells.
4. Scientists use genetic engineering techniques to transfer the cow's DNA code for rennin into fungus cells. Fungus cells then read the cow DNA and synthesize the rennin enzyme, which scientists call "chymosin." Cheese-makers can use the genetically engineered chymosin enzyme to curdle milk.

Modern-day cheese makers want to produce large amounts of high-quality cheese in the most economical way.

Cheese Production: The Evolution of Cheese-Making Technology (Lab 1c)

Student's Guide

MATERIALS FOR EACH GROUP

Supply each group with the following components.

- 12ml milk
- 1 vial of 1mg/ml Chymosin (100µl)
- 1 vial of 1mg/ml Rennin (100µl)
- 1 vial of 1mg/ml Buttermilk (100µl)
- 1 vial of water (2ml)
- 4 pieces of filter paper
- 4 empty 6ml tubes
- 1 3ml transfer pipet
- 4 small transfer pipettes

Some components will be shared by the whole class and should be kept on a communal table.

- Balance
- 37°C water bath (optional)

ADDITIONAL MATERIALS FOR EACH GROUP

The following standard lab equipment should be available for each group.

- Beakers, flasks, cylinders, or tubes (to catch filtrate)
- Lab scoops or plastic knifes

Cheese Production: The Evolution of Cheese-Making Technology (Lab 1c)

Student's Guide

PROCEDURE

1. Label four 6ml tubes with the type of curdling agent and group number.
2. Use a large pipet to transfer 3ml of milk to each of the 6ml tubes.
3. Use a small pipet and transfer the entire contents of the tubes of rennin, chymosin or buttermilk to the labeled tube containing the milk. For water, fill the small transfer pipet to the bottom of the bulb and add to the labeled tube containing the milk. Use a different pipet for each transfer to avoid cross contamination.
4. Cap the tubes and invert the tubes three times and then transfer to 37°C water bath or place at body temperature (i.e. armpit) for incubation.
5. Set a timer and check for curdling every 5 minutes, by gently inverting the tube and examining for curds.
6. Record the time (in minutes) when the milk begins to curdle (small or large lumps) or solidified.
7. If the milk has not curdled in 30 minutes, check for curdling every hour.
8. In a data table similar to the Data Table 1, record the time (in minutes) when the milk begins to curdle (small or large lumps) or solidify.
9. Upon return to the lab, during the next work period (next day in most lab classes), determine the amount of curds produced by each treatment.
10. For each treatment, weigh a paper cone and record the empty cone weight.
11. Transfer the entire contents of a tube into a labeled filter paper cone over a suitable collection vessel. Once all liquid has drained through, dry the filter paper with curds overnight.
12. Weight the dry cone with dry curds. Subtract the dry cone weight. Record the weight of the curds in mg by multiplying the mass in grams by 1000.
13. Repeat with each treatment.
14. Create a data table that reports the Rate of Curd Production (weight/time) by each Curdling Agent.
15. Create a bar graph that shows the Rate of Curd Production (weight/time) by each Curdling Agent.

Cheese Production: The Evolution of Cheese-Making Technology (Lab 1c)

Student's Guide

Data Table 1: The Characteristics of Cheese made by Different Curdling Agents

Curdling Agent	Curdling Time (min)	Weight of Cone & Curds (g)	Weight of Cone (g)	Weight of Curds (g)	Rate (mg/min)	Technician /Comments
Chymosin						
Rennin						
Buttermilk						
Water (-Control)						

DATA ANALYSIS AND CONCLUSION:

Imagine you are an employee at a cheese (curdling) company and you must summarize the results of your experiments and give your supervisor the best answer to the scientific questions asked. Write a conclusion that reports the Results of the experiment (answer to the purpose question) including Evidence and Explanations for your findings. Discuss how well the results support what you expected might happen (the hypothesis). Identify sources of Possible Errors in the technique that may lead to variations in results. Think about the Practical Applications of the results of the experiment. Make a recommendation to the cheese company supervisor about which curdling agent should continue to be the focus of production. Include any variations in the procedures that you think may improve the cheese production.

ADDITIONAL QUESTIONS

1. In your notebook, create a chart similar to Data Table 1 and analyze the variable that impact the results of this experiment that need to be controlled if a technician wants to have confidence in the results. Include the variable to be controlled, the impact of variation, and how the impact might be minimized.

2. In this experiment, each curdling agent is tested multiple times, and an average result is determined. Look at the class data. Does it appear that the number of replications for each curdling-agent experiment was sufficient? Yes or no? Explain your answer.

Last saved: 11/28/2016 CMH

www.GBiosciences.com

EMC Publishing®

PARADIGM
EDUCATION SOLUTIONS