



Makgeolli: Rapid Production of an Alcoholic Beverage from the Fermentation of Rice[†]

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INTRODUCTION

Fermentation has been used for thousands of years to naturally preserve food and drink; the types of fermented products vary regionally and culturally (1). Laboratory experiences highlighting the fermentation of foodstuffs allow students to develop an understanding of the biology behind this food preservation technique (2–4). Food fermentation exercises encourage students to consider other cultures and their food and beverage traditions, in addition to teaching students techniques that can be performed in their own kitchens.

In an interdepartmental collaboration, Mercer University chemistry majors enrolled in a junior-level laboratory capstone course have developed fermentation laboratories as a way to develop their technical writing skills. In previous semesters of an undergraduate microbiology course, we have taught a variety of fermentations through the production of yogurt, sauerkraut, kimchi, and cheese. One activity, the making of kimchi, was incorporated into the curriculum of a 300-level microbiology course housed in Mercer University's Biology Department (2). Student enthusiasm for these food labs led us to explore new fermentations, especially those from other cultures. To this end, we developed a laboratory procedure and worksheets for producing the Korean rice wine makgeolli.

Whereas kimchi is fermented using naturally occurring lactic acid bacteria, makgeolli requires the addition of yeast and amylase enzyme to produce ethanol through fermentation. While other laboratories and courses have been developed around beer production, these procedures often require costly, specialized equipment and can be time intensive (3, 5–11). The production of makgeolli can be completed in less than two weeks (three two-hour lab periods) with no specialized brewing equipment. Herein

we report a laboratory project based around the preparation of makgeolli, where students are introduced to yeast fermentations and the enzyme amylase.

HEALTH AND SAFETY

Fermented foods have been prepared safely for thousands of years. While there is a small chance of contracting a foodborne illness from homemade fermented foods, this risk is minimized by following ASM biosafety level 1 guidelines (12). Makgeolli is a carbonated beverage, and, as such, it should be handled with care during the bottling process and when opening the finished product. It is not uncommon for the effervescent final product to build up considerable pressure. Accordingly, bottle the makgeolli in a Grolsch-style bottle and use caution when opening the bottle. After priming sugar is added, store the bottles in a secondary container.

MAKGEOLLI PREPARATION AND LABORATORY PROCEDURE

Makgeolli is a popular South Korean alcoholic beverage, with the industry estimated at over one trillion won in 2013 (~US\$875 million) (http://www.koreatimes.co.kr/www/tech/2018/01/693_152634.html; <https://www.irs.gov/individuals/international-taxpayers/yearly-average-currency-exchange-rates>). As a result, there are numerous makgeolli recipes available online and in the primary literature (13–15). The procedure found in Appendix 1 varies from many traditional preparations in two ways. First, the rice used in this experiment is cooked in a rice cooker, and not steamed. Second, traditional preparations require *nuruk*, a mixture of ground wheat and/or rice containing many strains of airborne fungi, yeast, and bacteria. The *nuruk* serves to break down starch present in the ferment so it is accessible for fermentation via the added yeast. As *nuruk* is not commonly available in the United States, it was replaced by amylase enzyme.

The rice was washed and cooked before the lab period to provide students sufficient time to prepare the fermentation. Students worked in groups of three in a classroom separate from the microbiology laboratory. Approximate times for each step are included in the procedure (see Appendix 1), so

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instructors should modify the procedure to fit the available time. In small groups, many of these steps are conducted simultaneously. Prior to the fermentation, students rinse one-gallon glass jars with detergent and water, charge each empty jar with ~50 mL of 70% ethanol, and seal the jar. The jar is shaken to cover all surfaces with ethanol, drained, and inverted to dry.

Students then prepare a mixture of yeast, amylase, and sugar. While the yeast metabolizes the sugar over the next hour, the cooked rice is allowed to cool and students work in small groups to answer the questions on Worksheet A (Appendix 2). The worksheet addresses which sugar is fermented, the role of amylase, and why makgeolli is truly a rice beer and not a rice wine.

After the rice has cooled, the gallon jar is filled with rice and water, and the yeast mixture is added. Folded cheesecloth is placed over the mouth of the jar and held in place with a rubber band. In our experience, conventional fermentation vessels with water locks produced inconsistent batches of makgeolli; the use of cheesecloth as a breathable lid worked without fail. The ferment is stored at room temperature, and it is essential to avoid dramatic temperature changes and drafts. Students are then instructed on how to stir and monitor the pH of the fermentations over the next seven days. Each student is assigned a day to stir and to record the pH of the makgeolli.

After one week, students continue the procedure by straining out the rice from the liquid ferment. The filtered liquid, now called makgeolli, is bottled, and priming sugar is added. After adding priming sugar, students complete Worksheet B (Appendix 2), which asks them to consider broader questions about the makgeolli fermentation. One question requires students to consider lab techniques that could confirm the type of microbe at work, reinforcing concepts of microbial size and structural differences learned earlier in the semester. The final question prompts students to make connections across a variety of fermentations and to consider commonalities. If desired, this portion of the lab exercise could be expanded to a hypothesis-driven experiment on fermentation. For example, one might prompt students to consider why alcoholic fermentations are limited to ~15% alcohol content; students could then develop and test their hypotheses. The bottled makgeolli is left undisturbed for three to seven days, at which point it is cooled in a refrigerator. The entire procedure can be completed in 10 to 14 days, depending on the duration of the carbonation stage.

ASSESSMENT

Student worksheets were graded, and participation in the makgeolli preparation was also assessed. Our students' results confirmed the importance of cleanliness in handling fermentations and showed that the makgeolli pH decreases due to the increasing concentration of carbonic acid over time. Increased student engagement during the makgeolli lab suggests that it is an effective active learning activity

that connects ideas throughout our microbiology course and across disciplines.

CONCLUSION

This exercise involves concepts that meet American Society of Microbiology (ASM) curriculum guidelines (16), such as the different cellular properties of eukaryotic and prokaryotic microbes, the ability to chemically control the growth of microbes, and the ability of humans to utilize microorganisms. Furthermore, this exercise emphasizes ASM scientific thinking competencies, such as the ability to communicate and collaborate with other disciplines and the understanding of the relationship between science and society. If expanded, this exercise could include hypothesis formulation, experimental design, and hypothesis testing, which would allow the instructor to reinforce the ASM microbiology laboratory skills (16). Experiments should be performed in the microbiology laboratory following the appropriate ASM biosafety guidelines (12).

SUPPLEMENTAL MATERIALS

Appendix 1: Makgeolli instructions
Appendix 2: Worksheet A and Worksheet B

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The authors declare that there are no conflicts of interest.

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