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Engaging Undergraduate Students in Food Safety Study and Food Microbiology Research

ABSTRACT

Despite the importance of STEM (Science, Technology, Engineering, Mathematics) disciplines, the interest and test scores of students in this field seem to lag every year in the United States. Food science may be an ideal tool for enhancement of STEM education, because of its universality, cultural importance and scientific diversity. This study focused on the implementation of teaching tools to engage undergraduate students in learning about food safety and food microbiology. During the Food Microbiology class, three engagement strategies were used: agar art, outbreak case studies and a research group project. The agar art contest was conducted to learn and highlight bacterial morphological diversity; case studies were presented through short stories to teach microbiological and epidemiological principles and practices, and students were challenged with a research group project in which two plating alternatives were compared with regard to assessment of food preservation strategies. Quizzes, appraisal of laboratory notebooks and exams were used to evaluate learning outcomes. By the end of the semester, ~ 95% (55/58) of the undergraduate students had learned about foodborne pathogen characteristics through case studies, in-class discussion and a research project. Data were compared with data on the previous 2 years, in which no or minimal engagement strategies had been implemented. A pre- and post-questionnaire were used to assesses students' engagement. The results demonstrate that creative engagement strategies are beneficial for supporting and enhancing students' learning about food safety.

INTRODUCTION

The science of foods goes back to thousands of years ago, when humans discovered how to use fire *(3)*; since then, humankind has developed many methods and technologies to broaden food choices and to process and preserve foods. Today, food science is offered as a major with multiple career opportunities in many colleges and universities all over the United States and abroad. Currently, 40 institutions

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nationwide offer a food science program that meets the standards for degrees in Food Science set by the Institute of Food Technologists (IFT) Higher Education Review Board (HERB) *(6)*. The food science major is considered a STEM (Science, Technology, Engineering, Mathematics) discipline by the Department of Homeland Security (DHS) *(2)*. Since 2000, STEM-related majors have represented only one-third of all bachelor's degree awarded in the U.S., and the rate of transfer out of STEM into a non-STEM field by U.S. undergraduate students averaged around 20% in 2014 *(11)*. Coupled with a 7% decrease in enrollment at the undergraduate level between 2010 and 2017 in the U.S., a need exists to attract students to this important field of study *(10)*.

Many strategies have been implemented to attract students to STEM-related fields, and food science has proven to be a good mechanism, since it is a multidisciplinary field that includes microbiology, engineering, chemistry, and processing, among other areas. Researchers *(13)* have worked with students from elementary school through high school on a project that used foods to demonstrate the principles of phase change, sensory analysis, shear, formulation, energy and chemistry. Approaches of the activities varied according to the students' level of education. As an example, ice cream frozen with liquid nitrogen was the food model used to teach about phase change and heat transfer. Children from kindergarten to 4th grade learned that liquid nitrogen is colder than ice cream mix, whereas students from 4th to 8th grade observed the changes from liquid to gas for nitrogen and liquid to solid for ice cream, which highlighted the difference between heat requirement and heat release. Students in 9th to 12th grade focused on the phase change at the molecular level. Through these activities, researchers *(12)* showed that science can be taught at a young age and attract students into STEM at a very early stage. *"Food science is the study of the physical, biological, and chemical makeup of food and the concepts underlying food processing,"* according to the Institute of Food Technologists (IFT) *(7)*. Usually, the curriculum includes a food microbiology course as a core class, typically composed of lectures and practical laboratory sessions, during the junior or senior year. Engaging students on the importance of microbiology and safety of the food supply can be challenging for the instructor who relies on lectures alone. Several studies *(1, 4, 12)* have demonstrated that students are more successful when engaged in an active learning process rather than in a traditional lecture course. Basically, any activity that involves participation of the students (group problem solving, workshop course designs, or worksheets) rather than being in a traditional listening-only course contributes to engagement of the students *(4)*. Further, activities in which students participate in experiential learning, such as surveys, research group projects or outdoor work, promote more in-depth understanding of the subject *(1)*. Therefore, the overall goal of the present study was to evaluate strategies to engage undergraduate students with regard to the importance of food safety and microbiology. Three different approaches were used during the semester: agar art, outbreak case studies and a research group project. Quizzes, in-class discussions and appraisal of laboratory notebooks were used as means of assessing learning outcomes.

MATERIALS AND METHODS

Classes: Food microbiology lecture (FDSCI 600) and food microbiology laboratory (FDSCI 601)

At Kansas State University, food microbiology is taught in two classes, FDSCI 600 (lecture component) and FDSCI 601 (laboratory component), in the 15-week fall semester. This upper level undergraduate course is also open to graduate students. Both classes are usually medium size in enrollment (60 students) and meet for two 50-min lectures and two 2-hour laboratory sessions per week. The classes have an instructional team composed of 1 full-time instructor, one 50%-time teaching assistant, and one 30%-time teaching assistant. The course fulfills a requirement for several majors (food science and industry, animal science and industry, and grain science) and colleges (Agriculture and Arts and Sciences). Thus, students arrive to this course with different backgrounds and experiences, but typically are in their junior and senior years. The classes address the role of microorganisms in foodborne illness and food quality, spoilage, and preservation, as well as the control and reduction of microorganisms in foods. The laboratory session (FDSCI 601) complements lecture materials (FDSCI 600) by providing hands-on experience with lecture concepts. Laboratory sections consist of a maximum of 30 students per section, and students perform activities in pairs.

Engaging strategies tested

Agar art. In 2015, the American Society of Microbiology (ASM) launched the ASM Agar Art contest to share the beautiful and diverse world of microorganisms with the public *(14)*. Inspired by the ASM initiative, students in FDSCI 601 had the opportunity to create their own art and to practice isolation and identification techniques on their own. Students were provided a sample of red meat (10 g) experimentally inoculated with 10^5 CFU/g of *E. coli* ATCC 12435. Instructors guided students on the principles of dilution and plating, demonstrating techniques. Samples were placed in 90 ml of 0.1% peptone water (BD Difco, Sparks, MD) and stomached for 1 minute. Appropriate dilutions, based on instructor suggestions, were performed, and diluted samples were plated on 3M™ Petrifilm™ *E. coli*/Coliform Count Plates (3M, Saint Paul, MN). A presumptive colony of *E. coli*, a blue colony with gas (based on manufacturer instruction), was picked and streaked for isolation on TSA (Tryptic Soy Agar, BD Difco, Sparks, MD), a non-selective medium. Students were provided the 3M™ Petrifilm™ *E. coli*/Coliform Count Plate

Pathogen: *Salmonella*	Pathogen: *E. coli*	Pathogen: *Listeria*	Pathogen: *Campylobacter*	Pathogen: *Bacillus*
Case study #1 **Purpose:** To familiarize students with microbiological testing associated with non-meat-based animal product and PCR (Appendix 60). **Introduction:** In 2015, Ted Grant was training for a boxing match at his local gym. In order to bulk up, he followed a similar training regimen from the 1976 cinematic masterpiece *Rocky*. He ran several miles a day, only wore gray sweats, and ate a similar diet. Grant would crack 3 raw eggs in a blender, then add dried milk powder, and would put in asparagus for flavor and color. Grant ate these three times a day for the next three days. After the third day, Grant had uncontrollable diarrhea, cramping, vomiting, and chills. Grant then went to the doctor and was told to stay in bed and drink plenty of water in order to start feeling better. Needless to say, Grant lost the boxing match. **Preliminary Questions:** **What food or foods caused Grant to miss the boxing match?** **What biochemical or media testing should be conducted in order to provide conclusive results for your assumption?** **Is enrichment needed for this case? Why or why not?**	**Case Study #2** **Purpose:** The purpose of this laboratory exercise is to give students a better idea on pathogens found in red meat. **Case:** Rachel White is a 26-year-old female who lives in southeast Oklahoma. She is an active member in her community, frequently volunteering her time at the church cooking casseroles for the less fortunate. Rachel had finished making a wonderful Meat Lasagna for the soup kitchen, when she got a call from her work, asking her to come in and assist in a discrepancy. Rachel covered the lasagna with tin foil and left her house around 3:00 PM for work. She returned home at 8:00 PM and put the lasagna in the fridge. The next day she dropped the lasagna off at the church for the weekly soup kitchen service. 5 days later, Rachel received a call from the church saying that several of the attendees went to the hospital after experiencing bloody diarrhea, stomach cramping and several severe cases of bloody urine, denoting kidney failure. All the attendees reported eating the lasagna she dropped off. However, Rachel felt no symptoms the church was describing. **Preliminary Questions:** **At what point was the food "contaminated?" How do you know?** **Based on symptoms described, which microorganism is of concern?** **In order to confirm your hypothesis, which biochemical testing and media should be used?**	**Case study #3** **Purpose:** To familiarize students with microbiological testing of produce. **Introduction:** In 2012, Edward and Marry were a happily married couple living in Colorado. They had recently found that Marry was pregnant. With much delight, Marry began doing research online on what foods she should be eating. She stumbled upon a blog by "Healthy Momma," and read that she should be eating more fruits than she was before. So, for the next 6 weeks, Marry began eating nothing but fruit salads with produce purchased from her local supermarket chains. The fruit salads contained pineapple, raspberries, cantaloupe, pears, and cherry tomatoes. Marry kept all of the fruits in the fridge after they were purchased and rinsed all fruits that would have their flesh eaten. After the 6th week, Marry began feeling ill and went to the doctor. After some blood testing was done, it was found she had some form of bacteria in her blood. Concerned for her child, further testing was performed. Unfortunately, her child had passed. The doctor urged her to make a list of everything she had eaten the past week and contact the local supermarkets and restaurants about what had happened. The Centers for Disease Control and Prevention urged markets across the nation to test their produce using ELISA testing after receiving several other reports of similar cases (Appendix 60). **Preliminary Questions:** **Which is the food and microorganism of concern? What information lead you to this conclusion?** **How would you conduct a microbiological analysis on the food suspected of contamination?**	**Case study #4** **Purpose:** The purpose of the laboratory exercise is to allow the students to learn how to analyze pet food. **Case Study:** In 2017, the Jones family wanted to expand their family by purchasing a puppy. The dog was purchased from a chain pet store. The salesperson informed the family of the type of food the dog was fed while in the store. Not wanting to change the diet of the puppy, they purchased the same food the store was feeding the dog. The next several days, the family took turns feeding the dog and playing with it. After the fifth day of having the dog, the family began showing flu like symptoms of fever, vomiting, with occurrences of bloody diarrhea. The dog seemed fine and showed no symptoms of illness however. **Preliminary Questions:** **Based on the information above, what could be the cause of the illness?** **Do some research regarding cases like this one from 2018. What seems to be the microorganism of concern? Is it always the same biological hazard?**	**Case Study #5** **Purpose:** To familiarize students with microbiological analyses of starch-based products. **Introduction:** Joseph Romano flew to New Jersey to visit his family for Thanksgiving. Upon arriving he was greeted by his grandmother, Sofia. Excited to have Joseph home for the holiday, she invited all the extended family; 56 members of the Romano family attended Thanksgiving. The meal itself consisted of turkey, mashed potatoes, gravy, cranberry sauce and the Romano's traditional rice stuffing. To make all the food in time, Grandma Sofia made everything the day before and kept it in the refrigerator. The turkey was made the day of, cranberry sauce was put in several containers, gravy and mashed potatoes were put in small containers, however all the stuffing was placed in one container. All food items were reheated at 145 degrees F and then consumed. After 30 minutes, all 56 members of the family felt ill and began throwing up. Grandma Sofia, being from the old country, believes that it is because everyone ate too much. However, symptoms of vomiting and cramping lasted for 24 hours after the meal. **Preliminary Questions:** **Which food item is responsible for the illnesses and at what point was the food "contaminated?" How could it have been prevented?** **Based on the information provided, what microorganism is responsible for the Romano family illness? What information supports your hypothesis?** **What media and biochemical testing should be conducted to provide conclusive results with your hypothesis?**

Figure 1. The five case studies extrapolated from the laboratory manual are presented by featured foodborne pathogen. Clues such as onset, symptoms and potential food vehicle of transmission are highlighted in yellow for easier reading.

Interpretation Guide available from the manufacturer website and were "challenged" to pick the right colony. Subsequently, students performed the citrate, motility, and sugar fermentation biochemical tests to confirm the identity of the microorganism and used the isolated colonies as "paint" to create art that would fluoresce if *E. coli* had been correctly isolated *(5)*. EMB (Eosin Methylene Blue, Hardy Diagnostic, Santa Maria, CA), a selective medium for *Enterobacteriaceae* and a differential medium for *E. coli*, was used for this exercise.

Case study. During lectures and laboratory sessions, students were presented five case studies involving different foodborne pathogens (*Salmonella* spp., *E. coli*, *Listeria* spp., *Campylobacter* spp., and *Bacillus* spp.), in which patient histories, symptoms and situations in which foods had been consumed recently were detailed. The five case studies, taken from the laboratory manual and shown in *Fig. 1*, were designed to introduce students to the methods involved in the investigation of foodborne disease and were highly relatable to real-life experiences. The illness episodes were provided in the laboratory manual, and students had one week to read the case, form a hypothesis about the causative agents, and identify the vehicle of transmission (food item). During the same week, the lecture (FDSCI 600) emphasized the characteristics, symptoms and control strategies for each foodborne pathogen. A discussion preceded the laboratory section, after which the students were provided the food identified in the case study and challenged to isolate and enumerate the pathogen of concern. As an example, for case study #3 *(Fig. 1)*, cantaloupes were used as the food implicated in a *Listeria* outbreak. Students made appropriate dilutions with Modified Oxford Media (MOX, BD Difco, Sparks, MD), identified presumptive *Listeria* colonies and confirmed their isolates by using tests such as tests of motility and the litmus milk test to confirm specific metabolic activities of microbes *(13)*.

Research group project. Throughout the semester, students were taught how to conduct research by developing a research hypothesis and designing an experimental protocol. They were asked to select a food from a list of items historically linked to *E. coli* outbreaks and choose among three preservation technologies: (UV radiation, high temperatures and commonly used antimicrobials found in consumers' kitchens, such as spices, garlic and vinegar.) Students justified their choices, formulated a hypothesis (which included the expected log reduction on the inoculated item after treatment with the selected technology) and described the methods they intended to use. Finally, the class was randomly divided into two groups, with four teams in each group. Group 1 was asked to perform dilutions and plate on Plate Count Agar (PCA, Millipore Co., Billerica, MA) media, while Group 2 used a PCA ready-made plate containing a cold-water-soluble gelling agent. Thus teams worked with the same food items and technology, but the enumeration technique differed. The research project occurred during weeks 11 to 15 of the semester. Students inoculated the food matrices, applied the identified interventions and evaluated total aerobic bacteria in the food samples. Students calculated the microbial reduction brought about by the intervention. Subsequently, the instructor helped students estimate the correlation between the two enumeration techniques, and the students provided an oral group presentation to their peers, instructor and guests.

Assessment of learning outcome

To measure the effect of the three engagement strategies used in class, quizzes, appraisal of laboratory notebooks and in-class discussions were used throughout the semester. A

Figure 2. Examples of agar art "paint" of *E. coli* ATCC 12435 on EMB (Eosin Methylene Blue) agar.

questionnaire pre- and post-semester was used to evaluate students' engagement based on scores of 1 or 2 (strongly disagree or disagree) or of 5 or 6 (agree or strongly agree) on a Likert scale. Comparisons with the two previous years (2016 and 2017) were made to measure quantitative reasoning and critical thinking as well as to estimate the results of the format changes in these classes. During 2016 and 2017, little or no engagement activities had been implemented in this course because of a change of instructors. Feedback, in the form of a detailed rubric, a one-on-one meeting, and a model example for each activity were also provided to the students so that they could understand their strengths and weaknesses and thus enhance their knowledge and their desire to be life-long learners.

Statistical analysis

Descriptive statistical analysis was performed on the compiled results in order to assess students' improved knowledge and engagement, compared with the previous years (2016 and 2017). A statistical comparison was used to determine accuracy of results with the two enumeration techniques used in the research project by computing the coefficient of variation and the concordance correlation coefficient. All analyses were conducted with Stata/SE 12.0 (StataCorp LP, College Station, TX).

RESULTS AND DISCUSSION

Demographic information

Throughout 2016 (number of students = 52), 2017 (number of students = 63) and 2018 (number of students = 58), more than 50% (91/173) of the students enrolled in food microbiology classes (FDSCI 600 and FDSCI 601) were pursuing a Food Science and Industry degree, followed by 32% in Grain Science (55/173), 9% in Microbiology (16/173) and 6% in Animal Sciences and Industry (11/173). The majority of the students were in their senior year, followed by juniors; only a few were sophomores. It is recommended that courses such as FDSCI 600 and 601 be taken after completion of basic courses in biology, microbiology, and introduction to food science, so that students can relate the importance of food safety and microbiology to already familiar concepts of food processing and quality.

Agar art

Overall, 95% (164/173) of the students were able to isolate, identify and use *E. coli* colonies from an inoculated sample of red meat to "paint" on petri dishes. *Figure 2* displays some examples of agar art on EMB agar (Hardy Diagnostic, Santa Maria, CA). During this experience, students learned the importance of differential media to

TABLE 1. Three examples of specific assesment outcomes collected in 2016 and 2017 (when no or few engagement strategies were used) and 2018 (when all three engagement strategies were implemented). The number of students in each performance category is indicated

Asssesment of learning outcome (examples)	Year	Exceeds Expectations	Meets Expectations	Needs Improvement	Below Expectations
1. A quiz question tested a pre-required concept from general microbiology, briefly mentioned in class, but necessary to understand yeast fermentation.	2016	21% (11/52)	32% (17/52)	29% (15/52)	18% (9/52)
	2017	31% (20/63)	35% (22/63)	24% (15/63)	10% (6/63)
	2018	35% (20/58)	50% (29/58)	10% (6/58)	5% (3/58)
2. A case study question asked about the advantage of using an API test for pathogen identification (a method recently used in the laboratory class).	2016	14% (7/52)	27% (14/52)	29% (15/52)	30% (16/52)
	2017	29% (18/63)	32% (21/63)	26% (16/63)	13% (8/63)
	2018	38% (22/58)	41% (24/58)	17% (10/58)	4% (2/58)
3. An assigment challenged students with a specific food item and tasked them with identifying and describing the intrinsic and extrinsic parameters affecting microbial growth.	2016	13% (7/52)	35% (18/52)	19% (10/52)	33% (17/52)
	2017	35% (22/63)	46% (29/63)	11% (7/63)	8% (5/63)
	2018	55% (32/58)	37% (21/58)	6% (35/58)	2% (12/58)

microbial isolation and confirmation; if they did not correctly isolate *E. coli* colonies, their art would not exhibit the green sheen. The University of Alabama at Birmingham has also used agar art in teaching students about microbe-microbe interactions in isolates obtained from environmental samples; students studied changes in their agar artwork over time and then formulated hypothesis about the isolated bacteria *(9)*.

Case studies

Students were presented with five case studies for studying various foodborne pathogens during the semester *(Fig. 1)*. During in-class discussions, students expressed their thoughts about possible causative pathogens and vehicles of contamination. The majority of students correctly identified the responsible microorganism and understood the use of specific identification techniques *(Table 1)*, although some expressed doubt and asked the instructor to clarify and highlight the key points for recognizing a specific microorganism (on-site time, symptoms, handling practices described, etc.). Throughout the three years, we observed an improvement in students' performance when all three engagement strategies were implemented *(Table 1)*. Students appreciated the challenge (according to final feedback, as indicated in *Table 2*); they were able to engage in laboratory experiences correctly and to describe and correlate biochemical confirmation results with their initial hypothesis. Some researchers *(9)* state that solving case studies is a process in which students engage themselves with the situation, initially using their intuition to identify the problem and then analyzing the data to identify possible causes. Once they have stated the problem clearly, they can start to provide creative possible solutions, which they evaluate and from which they finally select the best ones. The process of solving a case study offers an opportunity to use logical reasoning skills, to be intuitive and creative in real-life situations and, in this class, to enhance their STEM skillset *(8)*.

Research group project

Students understood the different phases of a research project: media and supplies preparation before starting the experiments (materials and methods), scheduling (experimental design) and interpretation and reporting of results. Some groups were successful with the selected interventions in reducing or controlling microbial growth, whereas other groups did not observe any statistically significant difference between control and treatment ($P > 0.05$). Nevertheless, they all learned some of the fundamental processes of research, including comparing results and presenting the information in a summarized and clear manner. Each group compared the two enumeration techniques for microbial counts. Correlation between the

TABLE 2. Summary of pre- and post-semester questionnaires with evaluation scale and general course feedback (data are from 2018, when all 3 engagement strategies were implemented)

	Evaluation scale				
	Strongly Agree	Agree	Undecided	Disagree	Strongly Disagree
Pre-semester questionnaire summary					
I look forward to this class.	38% (22/58)	25% (15/58)	9% (5/58)	10% (6/58)	18% (10/58)
In food microbiology course, I will learn mainly about microoganisms that spoil food.	53% (31/58)	37% (21/58)	6% (4/58)	4% (2/58)	0% (0/58)
Post-semester questionnaire summary					
I learned about food safety and microbiology.	95% (55/58)	2% (1/58)	3% (2/58)	0% (0/58)	0% (0/58)
The laboratory manual was a useful tool to understand experiments and methodology.	3% (2/58)	78% (45/58)	15% (9/58)	2% (1/58)	2% (1/58)
I enjoyed coming to class.	14% (8/58)	72% (42/58)	10% (6/58)	3% (2/58)	0% (0/58)
General feedback about the course					

- I thought there were good case studies that stimulated students to assess what they had learned in the class.
- The instructor was always there to guide us and improve our laboratory techniques.
- I appreciate the opportunity to do undergraduate research because it allows students to experience directly all the process it takes for a research project. It allows students to see if they are interested in research or not.
- This research project makes students feel like they are applying a variety of concepts they learn during class. It was the first time for the majority of students to be part of a research project.

TABLE 3. Correlation coefficient, F-test of equality of means between colony counts and agreement by enumeration techniques used by the students (Group 1 used PCA medium, while Group 2 used a ready-made plate PCA containing a cold-water-soluble gelling agent)

	Group 1	Group 2
Students' count correlation (# obs)	0.84 (8)	0.78 (8)
Correlation coefficients	-0.876	-0.307
F-test of equality of means	0.118	0.592
Kappa	0.43	0.38
Agreement (%)	50.0	50.0

colony counts using the two techniques was high and positive ($P < 0.05$). Although the agreement beyond that due to chance (kappa) between colony count reads between the two types of enumeration techniques was slight, concordance was fairly high, as shown by the large concordance correlation coefficients and non-significant P-values for the F-test of equality of means and variances *(Table 3)*. Overall, there was no statistical difference between the two enumeration methods ($P < 0.05$). During the final presentation, students offered several creative solutions to food safety challenges; one group suggested the use of hot sauce for microwaved chicken wings stored over time, and another group proposed the application of a garlic spread on bread. This experience stimulated their creativity in thinking of practical solutions for everyday problems. Students had the opportunity to use logical reasoning skills and creativity, expanding their appreciation for STEM disciplines.

Final course feedback

Near the end of the course, students provided feedback to questions regarding the activities during the semester, and learning outcomes were assessed. By the end of the semester, 79% (46/58) of undergraduate students had learned about foodborne pathogen characteristics, identified possible contamination vehicles through case studies and in-class discussion, and conducted a research project *(Table 1)*. In comparing learning outcome #2, we observed that 40% (21/52) and 61% (39/63) met or exceeded expectations in 2016 and 2017, respectively, when few or no engagement activities were used. However, in 2018, when all three engagement activities were incorporated into the class, we observed an increase to 79% (46/58). Similar improvements were also observed by Freeman and collaborators *(4)*, in whose study active learning activities helped students improve their performance, especially in small classes. Examples of pre- and post-semester questionnaires and general course feedback are given in Table 2. At the end of the 3 years of study (2018), in the year in which all three engagement strategies were implemented, 95% (55/58) of the respondents answered that in this class they had learned about food safety and microbiology, and 72% (42/58) responded that they usually enjoyed coming to class. Further, the laboratory manual was judged to be a useful tool to help students understand and follow experiment protocol and methodology by 78% (45/58) of students. The data collected in this study suggest that creative engagement strategies are beneficial for supporting students' learning in food microbiology and food safety.

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