

Bacterial Unknown Project in the COVID-19 Era: Transition from In-Person Lab Experience to Online Environment[‡]

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The eruption of the COVID-19 pandemic forced many universities to quickly transition traditional in-person laboratory courses to an online format for remote learning. Consequently, learning objectives focused on hands-on laboratory skills shifted to ones that assess skills that could be recapitulated in the online format. We have transitioned a staple experiment in most undergraduate microbiology labs, the Bacterial Unknown Project, for online delivery using the university Learning Management System. We maintained the learning objectives suited for online delivery, such as creating an experimental design for identifying an unknown bacterium and communicating scientific results, while replacing or modifying those which could not be replicated, such as demonstration of sterile techniques, with learning objectives that highlighted skills of collaboration, peer evaluation, and scientific communication. Assessment of these new and modified learning objectives demonstrated positive student learning. Additionally, an anonymous postproject survey asked students whether they perceived the online Bacterial Unknown Project had increased their skill level in the areas highlighted by the revised learning objectives. Results reflected that 80% of the students reported the Unknown Project had increased their skills in all areas evaluated.

INTRODUCTION

The Bacterial Unknown Identification project is a core experience for undergraduate microbiology laboratory programs. This project requires students to work independently to identify multiple unknown bacteria and communicate their results in a written lab report. The Student Learning Objectives (SLOs) for this project were designed to develop students' skills in aseptic transfer, media prep, staining and microscopy techniques, and knowledge of differential media. Therefore, the project is the fundamental assessment of the student's ability to perform the lab techniques and skills acquired throughout the course (1). While this project is a mainstay in many microbiology laboratory courses, instructors have adapted it in various ways, such as isolating bacteria from environmental samples (2), including molecular techniques (3), and transitioning the lab to a more inquiry-driven approach (4). However, these adaptations

were designed for in-person implementation and focused on wet lab technique-driven learning objectives.

Previously, students in the Bacterial Infectious Disease laboratory completed the Unknown Project with the goal of identifying bacterial pathogens from a mixed culture. The completion of this project in the in-person setting results in successful training of the students to: (i) demonstrate effective sterile techniques to isolate bacteria from a mixed culture, (ii) maintain accurate notes in a laboratory notebook, (iii) analyze clinical data, (iv) work independently to identify the bacterial pathogen(s) as the causative agent for a hypothetical disease in a patient, and (v) communicate results in a written laboratory report. Due to the limitations on in-person instruction imposed by the COVID-19 pandemic, we modified these SLOs and transitioned this project for online delivery using the campus Learning Management System (LMS) Blackboard.

One of the challenges of redesigning this wet lab project for online delivery was recapitulating the in-lab experiences using a digital delivery system. While we would be unable to replace certain aspects of this project (e.g., assessment of aseptic transfer and other hands-on microbiological techniques), we reimagined ways to emulate the overall experience. The learning objectives that could not be replicated online were replaced with SLOs that focused on developing skills in other important areas, such as scientific communication, teamwork, and peer evaluation. The SLOs for the online delivery were updated to include the ability to: (i) collaborate and implement an experimental design

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to identify the unknown bacteria, (ii) analyze clinical data and correlate findings with disease and clinical symptoms of patients, (iii) effectively communicate scientific findings in an oral format, and (iv) perform critical self- and peer-evaluation.

Below, we describe procedures that were implemented in this project to transfer to an online experience. In this project we addressed specific in-class experiences that cannot be reproduced in an online course by making peer learning, scientific communication, and peer evaluations center points in this project. Integration of these aspects into the Unknown Project emphasized the importance of peer review and collaboration in scientific discovery and publication in the scientific world. Additionally, emphasizing the skills of teamwork and of giving and receiving constructive criticism not only benefitted the students during this course but introduced them to processes which they likely will encounter in their future careers (5, 6). There is a critical need to develop online versions of such staples of microbiology lab courses, as this scenario may reoccur in the future, requiring a rapid conversion to online learning.

Intended audience and prerequisite student knowledge

This project was designed for upper-division undergraduates majoring in microbiology. The curriculum could be modified for introductory-level microbiology students or online courses intended for allied health majors who do not require the hands-on techniques for their future careers or education. Before the course is implemented, students should have an understanding of basic microbiological techniques, including bacterial morphology, differential and selective media, and commonly used staining techniques. The enrollment for this course was 49 students. The instructional team consisted of two graduate teaching assistants, one undergraduate teaching assistant, and one teaching faculty, but this project could be adjusted for a smaller teaching team.

Learning time and SLOs

The online Unknown Project was conducted over 2 weeks with an additional week allotted for preparation of oral presentations. For instructors with limited teaching assistance, this timeline can be adjusted so instructors have more time to monitor discussion boards and upload results. Evaluation of the success of this project was based on the correct identification of the pathogens presented, teamwork skills (as determined by both self- and peer evaluation), and presentation skills (as determined by both instructor and peer evaluations) (Appendix 2).

After completing the Unknown Project, students will be able to:

- 1) Collaborate to plan an experimental design to identify the unknown bacteria.

- 2) Identify possible disease origins and clinical symptoms of patients infected with the unknown pathogens.
- 3) Effectively communicate scientific findings in an oral presentation format.
- 4) Critically evaluate their own and their peers' contributions using rubrics provided.

PROCEDURE

An emphasis on teamwork and collaboration skills was integrated into the project in SLOs 1 and 2 for a few reasons. For the in-person delivery of this project, students worked independently, assessment of microbiological techniques being a key learning objective. However, with the shift to the online format, we were unable to assess these skills. Additionally, there was some concern students transitioning abruptly to the online format might feel isolated and overwhelmed with this project while adjusting to the new reality of virtual learning. Therefore, students were paired with a lab partner, and learning teams were assigned to a discussion board on the LMS in order to asynchronously collaborate. The discussion boards were monitored by graduate teaching assistants (GTAs) and served as the platform for delivering data to the learning team. The teams worked together to analyze the data and request additional "virtual tests" to be performed on their samples. This process continued until each bacterial sample was identified, and the learning teams then progressed to the next phase of the project, where they collaborated on the presentation of their results. Peer learning for in-person delivery of content has been shown by numerous studies to enhance student engagement, increase collaboration, and advance academic performance (7). We predicted a similar outcome for peer-learning on the online delivery of this project. A postproject survey was given to students to assess student perception of the effectiveness and delivery of peer-learning in the online format.

Integration of SLO 3, effective written and oral scientific communication, was achieved in two ways. First, the assigned discussion boards provided a forum for written communication between learning teams. Second, we transitioned away from the traditional written report, instead requiring students to present their findings in an oral presentation. The purposeful integration of communication skills as learning objectives for this project filled a recognized and consequential gap in current undergraduate training. The emphasis placed on these skills will help increase students' ability to effectively communicate scientific, content within both the public and scientific communities (8).

Inclusion of peer evaluation as a learning objective (SLO 4) was meant to develop the students' ability to (i) critically evaluate peers and (ii) respond to constructive criticism about themselves. Students were required to conduct two different forms of evaluations: an evaluation of their lab partner's performance, and an evaluation of two selected

peer oral presentations. Evaluation of their assigned lab partner (Appendix 2) accounted for 15% of the partner's grade and was meant to empower each partner to hold the other accountable to the project. Student evaluations of oral presentations were also rubric-based (Appendix 2) and accounted for 10% of the presenting student's project grade. The oral presentation evaluation served two purposes: first, students were assigned presentations about microbes not found in their own samples, so they were learning about different pathogens, and second, students were taught to critically evaluate their peers' work while offering constructive feedback. An example of the rubric used for the overall evaluation of student effort on this project is provided in Appendix 7.

Materials

Students require:

- A computer with access to the school's LMS (e.g., Blackboard or Canvas).
- Presentation software with the ability to voice record their presentation (e.g., PowerPoint or QuickTime Player).

Instructors require:

- Digital image files of results for experimental tests used in the project.
- A dichotomous key for bacteria included in the sample unknowns.

Student instructions

Unknown identification. Students log in to the LMS and enter their discussion board to find Gram stain images representing the results for the three bacterial unknowns. All subsequent results will be posted to the discussion board as image files (Appendix 1). Learning teams will analyze the results, create a running table of test results (Table I), and order additional "tests" to be performed on their samples. Learning teams can request a maximum of two tests per unknown each class period. This limit ensures students are thinking critically and are ordering tests most effective and efficient in specimen identification. Lab partners are required to confirm agreement on the requested tests before they are processed by the discussion board moderator.

Discussion board moderators will post the requested test result images within 24 to 48 hours, a timeframe that

TABLE I.
Sample of student-completed table of results.^a

	Sample 1	Sample 2	Sample 3
Gram Reaction	+	+	-
Morphology	Rod	Cocci	Rod
Tests Ordered 4/22	Blood Agar	Catalase Test	PR Lactose
	Endospore Stain	Coagulase Test	Oxidase Test
Results	Beta hemolytic	Catalase +	Lactose +
	Endospore -	Coagulase -	Oxidase -
Tests Ordered 4/24	Esculin Hydrolysis	TMPA	Methyl Red
	Tinsdale Agar	PR Trehalose	Lysine Decarboxylase
Results	Esculin Hydrolysis +	N/A	Negative
	N/A	N/A	Lysine Decarboxylase -
Tests Ordered 4/27	PR Sucrose	Mannitol Salt Agar	MIO
		PR Mannitol	
Results	Sucrose -	Negative	Motility + Indole - Ornithine Decarboxylase +
		Mannitol -	
CONFIRMATORY Tests 4/29	PR Glucose	Novobiocin sensitivity	Motility
Results	Glucose +	Novobiocin S	Motility +
Identification	<i>Listeria monocytogenes</i>	<i>Staphylococcus epidermidis</i>	<i>Enterobacter cloacae</i>

^a PR, phenol red broth; MIO, motility indole ornithine medium.

Table 2.
Unknown Project timeline of events.

	Moderator will:	Student teams will:
Session 1	<ul style="list-style-type: none"> • Provide initial Gram stain and morphology results 	<ul style="list-style-type: none"> • Discuss and analyze results • Create table of results (Table 1) • Order additional tests (max 2 per sample)
Session 2–4	<ul style="list-style-type: none"> • Provide picture file results of test requested in previous session 	<ul style="list-style-type: none"> • Discuss and analyze results • Upload updated table with results supplied at 8 a.m. • Order additional tests (max 2 per sample)
Session 5	<ul style="list-style-type: none"> • Provide picture file results of test requested in previous session 	<ul style="list-style-type: none"> • Discuss and analyze results • Upload updated table with results supplied at 8 a.m. • Request final confirmatory tests if not done previously
Session 6	<ul style="list-style-type: none"> • Provide picture file results of final confirmatory test 	<ul style="list-style-type: none"> • Student teams should upload a completed table of results to the discussion board including the names of all identified species (see Table 1) • Prepare presentation of results
Session 7	<ul style="list-style-type: none"> • Unknown Project presentations 	<ul style="list-style-type: none"> • Student teams will prepare 5 slide presentation of results • Individual team members will record oral presentation and upload to LMS media gallery

can be adjusted for instructors with limited teaching assistance. The process of discussing the results, agreeing on how to proceed, and requesting tests continues from class periods 1 to 7, or until the students are able to identify all bacterial unknowns. An additional positive confirmatory test is required for each species. Teams post a finalized table of results (Table 1) before preparing the oral presentation. A timeline of the identification of the unknown portion of this project is found in Table 2.

Project presentation. Students create and upload recorded oral presentations of their results to the LMS media gallery. They can work on the slide presentation individually, or teams can collaborate using a shared document tool such as Google Slides. The recorded oral portion of the presentation should be completed individually using a screen recording tool such as QuickTime Player or PowerPoint. The presentation should be 5 to 7 minutes in length with a 10-minute maximum. Presentations should include:

1. A summary of the tests and data collected on each sample.
2. Identification of the bacterial species.
3. An overview of each pathogen.
4. Symptoms or disease a patient infected with the pathogen may be experiencing.

Peer evaluations. Students are each given a rubric instructing them to professionally and critically evaluate the presentation and communication skills of two of their peers' oral presentations (Appendix 2). Communication skills to be evaluated include pace, volume, use of filler words, and pronunciation. Presentations are critiqued on the length, slide design, and effective conveying of the information. The evaluations are included in the assessment of the presenting student and shared with the presenter but remain anonymous. In addition, students are asked to perform a

self-evaluation and an evaluation of their lab partner based on their overall efforts during the project (Appendix 2). All evaluations are rubric-based and considered in the final project grade.

Student postproject survey. At the conclusion of the online Unknown Project, students are asked to complete a survey in which they evaluate the perceived effectiveness of the project, advantages and disadvantages of the online format, and skills gained (Appendix 3). The survey also provides an opportunity for students to make comments and suggest modifications that may enhance the curriculum objectives.

Specific student-ready instructions can be found in Appendix 6.

Faculty instructions

Before assigning the Unknown Project, instructors provide online lecture material covering differential and selective clinical media, including demonstrations of useful identification tests and results. The instructors also provide relevant information regarding the bacterial species used. An example of a preproject learning timetable is shown in Appendix 5.

Prior to beginning, instructors group the students into teams of two or three. Lab partner pairings should consider student input to create more balanced experiences and increase student enthusiasm for the project (9). Instructors must create discussion boards for each team to use as their digital platform for the project. A tutorial for the students on how to access and use these sites is advisable.

To facilitate uploading data images of test results to the discussion boards, we recommend creating a file of digital images prior to initiating the project. Positive and negative results of each virtual test used for this project should be

rial Infectious Disease Laboratory) designed for students with a prerequisite introductory microbiology course at the University of Kansas. Field testing occurred during the 2020 spring semester after 7 weeks of in-person instruction when in-person instruction was suspended due to the COVID-19 outbreak. A total of 49 students participated. The completion of the project was followed by a 15-question postproject survey in which students evaluated the project. Although the majority of students had primarily positive experiences (82%), the remaining students reported both positive and negative aspects of their experience or remained neutral in their assessment (18%). Many students reported growth in communication skills and the acquisition of those skills outlined in the SLOs (Fig. 2). This field test showed that an online delivery of the Bacterial Unknown Project was possible and serves as a method for evaluating student learning of microbiological techniques and clinically relevant bacteria, as well as providing a forum for student growth in collaborative and oral presentation skills.

Evidence of student learning

The primary goal of the in-person delivery of the Bacterial Unknown Project is to provide a comprehensive assessment of clinical knowledge of bacteria, understanding of the application of clinical media, aseptic techniques and microscopy skills, experimental design and implementation, and written communication of scientific findings. The objective in transitioning this project to online delivery was to maintain these learning objectives as much as possible and to substitute meaningful experiences for learning objectives incompatible with the online experience. The first learning

objective of this project focused on the student's ability to collaborate and design an experimental approach to identify the unknown bacteria. This enhanced student knowledge of clinical media, abilities in experimental design, and teamwork skills. This was first implemented by assigning lab partners for the project and was then assessed through a rubric-based evaluation (Appendix 2) of discussion boards and the correct identification of the assigned bacterial species. Through the discussion boards, instructors evaluated students' professionalism, accuracy, participation, and timeliness. Professionalism was defined by their ability to articulate ideas and effectively communicate with their teammate(s) in a considerate, workplace-appropriate manner. Accuracy reflected their ability to logically deduce an appropriate next step, correctly interpret the test results, and maintain an organized results table. Finally, participation and timeliness were assessed based on how communicative each partner was and whether responses occurred within the prescribed time limits. Student scores averaged 27.3 of 30, or 91.0%, and many students received full marks for their insightful, respectful, and cooperative communications (Table 3).

The second and third learning objectives included the ability to identify the possible disease origin and clinical symptoms associated with a given pathogen and to then effectively communicate these findings. These SLOs were evaluated based on the effectiveness of each student's oral presentation. This presentation encouraged creativity in presenting patient histories, with the expectation of a concise summary of the identification process and characterization of the pathogens. The instructor rubric for scoring the presentation (Appendix 2) evaluated speaking skills, presentation organization, use of effective graphics,

"The online Unknown project has increased...."

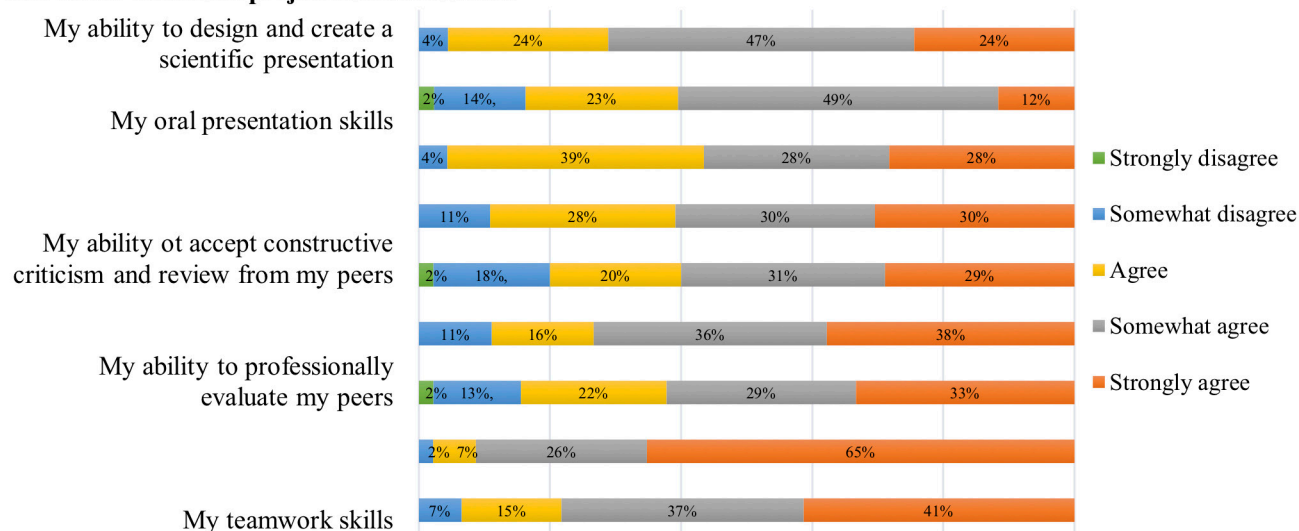


FIGURE 2. Student satisfaction survey responses. Forty-eight participating students rated their perceived gains in skills and abilities over the course of the online unknown project duration. A total of 80% of students reported that the Unknown Project increased their skills in every area evaluated. The highest perceived acquired skill, with a positive response rate of 98%, was the ability to develop and execute a plan to identify unknown microbial samples. The lowest perceived acquired skill, with a positive response rate of 80%, was the acquisition of increased teamwork skills. The complete, student-ready survey can be found in Appendix 3.

TABLE 3.
Comparison of discussion board grades from instructors, partners, and self-evaluations.^a

Student	Instructor Grade	Lab Partner Evaluation	Partner Evaluation Compared with Instructor	Self-Evaluation	Self-Evaluation Compared with Instructor
1	28	30	+2	27	-1
2	25	28	+3	25	0
3	29	29	0	30	+1
4	30	30	0	30	0
5	14	15	+1	21	+7
6	25	30	+5	30	+5
7	25	27	+2	27	+2
8	24	27	+3	25	+1
9	28	29	+1	24	-4
10	26	28	+2	27	+1
11	26	26	0	30	+4
12	25	29	+4	30	+5
13	28	30	+2	29	+1
14	26	30	+4	29	+3
15	27	29	+2	30	+3
16	28	30	+2	29	+1
17	28	28	0	30	+2
18	28	27	-1	29	+1
19	30	30	0	30	0
20	28	30	+2	29	+1
21	28	26	-2	28	0
22	28	29	+1	27	-1
23	30	29	-1	28	-2
24	27	28	+1	27	0
25	26	30	+4	28	+2
26	30	30	0	30	0
27	30	30	0	29	-1
28	30	30	0	30	0
29	21	30	+9	29	+8
30	30	30	0	30	0
31	29	30	+1	27	-2
32	30	30	0	30	0
33	28	29	+1	27	-1
34	28	30	+2	30	+2
35	30	30	0	29	-1
36	23	29	+6	24	+1
37	30	30	0	30	0
38	28	30	+2	28	0
39	27	24	-3	26	-1
40	28	26	-2	28	0
41	28	30	+2	30	+2
42	30	28	-2	30	0
43	26	26	0	24	-2
44	28	28	0	30	+2
45	25	28	+3	20	-5
46	26	30	+4	30	+4
47	30	30	0	28	-2
48	30	26	-4	22	-8
49	26	30	+4	29	+3
Average	27.3	28.5	+1.2	27.9	+0.6

^a Possible total of 30.

and pacing. Student scores averaged 41.3 of 45, or 91.7% (Table 4), which mirrored the percentages of the discussion board scores (Table 3). These scores indicate that students who had professional communication skills in the discussion boards translated those skills to their final presentations. The relationship here highlights the increased need to use teamwork within the lab environment to strengthen scientific communication.

The final learning objective, the ability to critically evaluate their own and their peers' contributions, used the provided rubrics (Appendix 2). One mechanism used to demonstrate effective student learning of peer-evaluation techniques was to compare peer-given scores with instructor-assigned grades (Tables 3 and 4). In each case, student scores were similar to instructor grades awarded to each student. In the discussion board partner evaluations, students on average earned 1.2 points higher from their peers than from the instructors. When comparing self-evaluations with instructor scores, self-assessed scores were on average 0.6 points higher than instructor-awarded grades. There were exceptions; for example, Student #29 gave themselves and their partner, 8 and 9 points more than the instructor grades, respectively (Table 3). One primary concern with including student evaluations in the final grade was that students would attempt to artificially inflate their final grades by increasing the scores on their self-evaluations and lab-partner evaluations, as potentially happened with this student. However, for most evaluations, the student peer evaluations accurately reflected the performance of the students, as determined by the instructors. In cases where it appears grade inflation may be occurring; it is up to the instructor's discretion to determine how this may impact the grade. In our course, this did not appear to be a prevalent issue and students seemed to include thoughtful evaluations that accurately represented the quality of the work and the effort provided. This trend carried through for the oral presentation peer evaluations as well. Combined peer evaluations resulted in percentage grades similar to those given by the instructors, and students were able to identify issues with a presentation that were noted by the instructor as well (Table 4). Because the peer evaluation rubrics were on a different scale than the instructor rubric, direct comparisons are not exact. Percentage scores from peer evaluations and instructor evaluations average 93.5% and 91.7%, respectively. The ability to accurately assess a peer's performance is an important skill in science, wherein scientists are constantly evaluating each other's work, a basic tenet of the scientific approach. Overall, these evaluations and the scores of the students suggests that this project is effective in addressing the new learning goals.

Possible modifications

The inclusion of a 16S bacterial rRNA sequencing experience has been previously reported in the literature for wet lab environments (2). Introducing students to the

more modern approach of sequence analysis to identify bacterial species could also be applied to the online version of the project. Students could identify their pathogen using the traditional clinical methods and then compare those results with the mock molecular analysis of their samples.

Perhaps the most significant modification to this project would be a blending of the online and wet lab experiences. As a teaching team, we were pleased with the implementation of the new SLOs and plan for future offerings of this project to be a hybrid of both old and new SLOs in order to more completely assess the students on the many skills needed to be successful in the scientific arena, not just the ability to identify a bacterial unknown.

CONCLUSION

The COVID-19 pandemic unexpectedly and rapidly altered education at all levels, forcing many to implement online learning platforms. Transitioning of laboratory courses, where hands-on learning has always been a critical aspect of the learning goals, was a challenge facing university lab instructors. Many educators who previously relied exclusively on wet-lab experiences had no choice but to transition their courses to an online format. Here, we offer an updated online version of the Bacterial Unknown Project for our upper-level undergraduate microbiology course. Through this project, students were able to synthesize technical knowledge of clinical media and bacteria with important career-building skills of collaboration, scientific communication, and peer assessment. Comparisons of student evaluations with instructor evaluations demonstrated that students acquired skills to critically evaluate both themselves and their peers. Student satisfaction surveys indicated that students responded positively to the online transition of this project and reported they had successfully learned many of these desired SLOs. While not a direct substitute for learning hands-on techniques, the online Bacterial Unknown Project can address these skills often overlooked in scientific curriculum.

While the COVID crisis forced many instructors to embrace a virtual lab experience, some within the education field have been encouraging a blend of wet and dry lab experiences for a number of years, highlighting the need to incorporate dry labs as a means to reduce budgetary and staffing constraints faced by laboratory classes (10). In fact, the future of many laboratory courses will likely be a blend of these two experiences, depending on the needs of their students. For example, allied health students, whose future careers will likely not use the technical skills gained from an in-person lab, may benefit from an online offering of the Unknown Project to the same degree as if they had the wet-lab experience (10). However, students such as microbiology majors, who require transferrable technical skills such as aseptic transfer, bacterial isolation, and microscopy, would have a gap in their fundamental training if unable to comple-

TABLE 4.
Comparison of Unknown Presentation grades from instructors, partners, and self-evaluations.

Student	Instructor Grade (out of 45)	Combined Peer Evaluation (out of 20)
1	41	19
2	40	18
3	38	18
4	43	20
5	22	19
6	43	19
7	41	16
8	44	19
9	43	18
10	41.5	18
11	43	20
12	40	19
13	45	20
14	45	20
15	42	19
16	45	20
17	41	20
18	43	20
19	41	20
20	40	18
21	38	20
22	43	19
23	40	17
24	42	15
25	43	20
26	45	20
27	43.5	18
28	40	19
29	35	18
30	38	18
31	41	16
32	38	19
33	45	20
34	41.5	17
35	40	20
36	42	20
37	42	19
38	40	17
39	42.5	18
40	41	18
41	44	18
42	43	18
43	39	18
44	44.5	20
45	39	17
46	43	19
47	44	19
48	42	19
49	41	20
Average	41.3	18.7

ment their experience with wet labs that addressed these skills. We were fortunate this semester that our students, who were mostly upper-division microbiology majors, were able to complete the first part of the semester at the lab

bench, and we relied on these experiences when transitioning to the online format. Here, we provide the tools to perform this project in future virtual classrooms and suggest that this template could also be used in fields outside of microbiology.

SUPPLEMENTAL MATERIALS

- Appendix 1: Sample images and sources for *Staphylococcus epidermidis*
- Appendix 2: Various rubrics
- Appendix 3: Student post-Unknown Project survey questions
- Appendix 4: Lab partners sample discussion board
- Appendix 5: Suggested outline, including preproject learning
- Appendix 6: Student instructions for Unknown Project
- Appendix 7: Summary rubric used for overall evaluation of effort on Unknown Project

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