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FUTIs: an In-Person or Online Graphing, Bioinformatics, and Scientific Literacy Exercise That Explores the Presence of Antibiotic Resistance in Foodborne Urinary Tract Infections

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We developed a course-based undergraduate research experience (CURE) that gives students an opportunity to practice the process of science in a context that intersects with their everyday lives: purchasing grocery store chicken. Student mastery of concepts was assessed by pre- and postassessment questions and lab report worksheets that guided them through the process of writing a scientific paper. Learning to produce graphs from large data sets and comparing the results with published data emphasized quantitative reasoning, while working as a group and writing helped students practice scientific communication. Most students (>90%) met the learning objectives, and students in both groups reported feeling more confident producing graphs and figures; they also showed large gains in confidence and interest in bioinformatics. Lab protocols require biosafety level 2 safety guidelines; however, students in an online or dry lab setting can use the compiled data sets and whole-genome sequences to complete the objectives. Group discussions and essay prompts at the end encourage students to use evidence-based arguments to make decisions that impact the global issue of antimicrobial resistance.

KEYWORDS antibiotic resistance, One Health, bioinformatics, online lab, graphing, science literacy, urinary tract infection

INTRODUCTION

Background

Antimicrobial resistance (AMR) poses one of the biggest challenges to human and animal welfare today (1, 2). Biology students need to understand how AMR develops and how genes spread between humans, animals, and the environment, a concept emphasized by the One Health approach (https://www.cdc.gov/onehealth/basics/index.html). The NSF Vision and Change report (3, 4) and the National Research Council (NRC) (5) promote active learning, including research and collaborative projects that stimulate deeper understanding. In CURE programs, students make inquiries and discoveries of interest to outside stakeholders (6–8). Similar to "A CURE for Meat" (9), this project stemmed from the PBS Frontline special, "The Trouble with Antibiotics" (10), which presented evidence that antibiotic-resistant *Escherichia coli* from retail meat may cause urinary tract infections (UTIs) in some patients (11). The project was also informed by our own (T. J. Johnson) involvement in that project (12). In this exercise, students isolate antibiotic-resistant *E. coli* from retail chicken raised conventionally or without antibiotics (RWA). Isolates are characterized by Kirby-Bauer assays for resistance to common drugs used to treat UTIs (13), allowing students to investigate whether RWA practices result in a lower incidence of AMR. After students use bioinformatics tools to identify AMR and virulence genes in whole-genome sequences (WGS), they can predict whether an isolate is pathogenic and determine which drugs might be appropriate to treat an infection.

Conducting research requires scientific literacy skills. When students perform experiments similar to those found in the primary literature, it is easier for them to understand their relevance. Scientifically literate students also have the ability to graphically represent data and effectively communicate findings. This curriculum provides a student-generated data set of over 200 samples and instructional videos for graphing the data. It also provides WGS files and instructions for bioinformatic analysis. Writing an entire formal lab report that is similar to a scientific paper can be an overwhelming assignment that benefits from scaffolding (14–16). In this exercise, students complete worksheets guiding them through research and analysis required to write the traditional Introduction, Results, and Discussion sections of a scientific paper. Writing these reports

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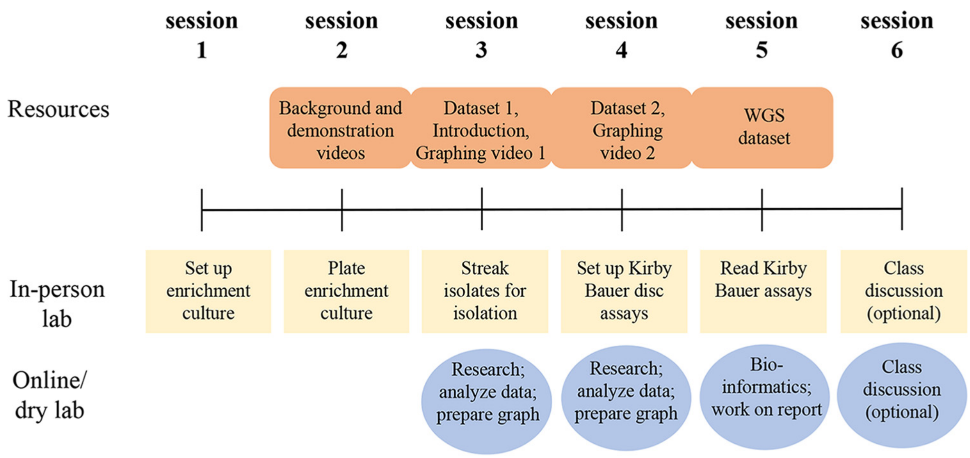

FIG 1. Timeline for activities in the in-person lab or the online or dry lab setting (possible modification). For students completing the full set of experiments, the overall goals are to (i) describe how small numbers of bacteria can be cultured and detected with the enrichment technique, (ii) define the terms coliform and fecal coliform, (iii) describe ways that antibiotic-resistant bacteria can be spread between people, animals, and the environment, and (iv) explain four steps to food safety. Session 2 goals are to (i) describe the basic ways antibiotics have been used in the poultry industry and how some practices have changed in response to the 2017 Veterinary Feed Directive and (ii) select for Gram-negative bacteria from poultry resistant to the antibiotics ampicillin, gentamicin, tetracycline, or ceftriaxone. Session 3 goals are to (i) remember which types of bacteria cause the most foodborne illnesses each year in the United States, (ii) explain why *E. coli* is used as a marker for food safety, and (iii) describe why antibiotic resistance found in *E. coli* might also be present in other pathogenic bacteria. Session 4 goals are to (i) explain how MALDI-TOF MS identifies microbial unknowns, (ii) set up Kirby-Bauer disk diffusion assays to assess resistance to several antibiotics commonly used to treat UTIs, (iii) prepare a streak plate for MALDI-TOF MS (optional), and (iv) analyze a genomic sequence to identify species, serotype, sequence type, virulence, and antibiotic resistance genes. Session 5 goals are to (i) explain how a Kirby-Bauer disk diffusion assay works, (ii) determine antibiotic sensitivity of isolate, and (iii) prepare a graph(s) depicting multidrug resistance from a large data set. Session 6 is an optional session where students examine each other's graphs and discuss the value of choosing RWA poultry as a class. The online or dry lab modification sessions are described in the text. Resources provided here include five videos (linked in text and in appendices), two data sets for graphing, and a set of *E. coli* whole-genome sequences (see Appendix S1 in the supplemental material).

requires students to engage with primary literature, to generate figures similar to those used in published literature, and to critically evaluate several sources of data in order to write an argument that affects their daily lives.

The 2019 coronavirus disease pandemic accelerated the trend toward online pedagogy (17). The graphing, bioinformatics, and data analysis exercises provided in this report can be used as a stand-alone module in an online course. These activities meet the ASM Curricular Guidelines for Introductory Microbiology (18) for scientific thinking, including the ability to apply the process of science, ability to use quantitative reasoning, ability to communicate and collaborate with other disciplines, and the ability to understand the relationship between science and society.

Intended audience

This unit was designed for students in a 2000-level mixed-major allied health microbiology course but could be used in a biology majors' microbiology, genetics, or animal science course.

Learning time

Figure 1 depicts the timeline for experiments and analysis. Completing the unit requires five to six 1.5- to 2-h lab sessions. An online modification (see "Possible modifications") requires three 2-h or two 3-h lab sessions.

Prerequisite student knowledge

Students should have had at least one semester of college biology that included study of genetic principles. Wet lab experiments should be conducted near the end of the term, after students have mastered serial dilutions and aseptic techniques.

Learning objectives

This unit has five learning objectives.

- Explain the concept of One Health and how it relates to AMR.
- Describe conventional, organic, and raised without antibiotics systems of poultry production and formulate a hypothesis regarding the prevalence of AMR among chicken raised in each system.
- Graphically represent and interpret research data from large spreadsheets.
- Use databases to identify virulence and AMR genes in WGS data; predict pathogenicity and antibiotic susceptibility.

- Explain principles of food safety related to poultry; evaluate the merits of "raised without antibiotics" (RWA) and "conventionally raised" (CONV) chicken.

PROCEDURE

Materials

Appendix S1 in the supplemental material contains a list of media and other supplies needed for *E. coli* enrichment cultures from chicken and characterization of antibiotic-resistant isolates. It also contains links to whole-genome sequence FASTA files for bioinformatics activities.

Student instructions

Appendix S2 is a student lab manual. It contains links to videos with background for the project (https://youtu.be/fAJYZTTIIGI), graphing (https://youtu.be/qYBq0wmQIBA, https://youtu.be/uuQQu4fH8dQ), and bioinformatics instructions, as well as lab protocols. It also contains a video (https://youtu.be/47cgtltAaxs) that demonstrates how chicken and cultures are handled safely in a biosafety cabinet (BSC). Worksheets (Appendices S3 and S4) provide instructions for completing two group lab reports and a bioinformatic analysis of WGS data using Center for Genomic Epidemiology (CGE) tools (https://www.genomicepidemiology.org/).

Faculty instructions

This CURE is designed to be carried out in either an in-person laboratory (6 sessions) or in an online or dry lab (4 sessions). Fig. 1 depicts the timeline and activities for each session. Appendix S1 also contains important safety instructions.

(i) Session 1. Students place chicken drumsticks in a bag containing MacConkey (MAC) broth for incubation. After the experiment is set up, students can read or watch suggested articles and videos and start answering questions for Report A, "Antibiotic resistance in *E. coli* isolated from grocery store chicken" (Appendix S3).

(ii) Session 2. This exercise involves unknown bacteria with potential AMR and should be conducted using biosafety level 2 (BSL-2) guidelines (see below). After dilutions and plating are completed in the BSC, students can read about the 2017 Veterinary Feed Directive (19), which has helped change the way antibiotics are given to production animals. They can also learn how AMR data are collected by the National Antibiotic Resistance Monitoring Service (NARMS) (20). They will compare the compiled student results (Appendix S5) with data in this repository when they work on Report A (Appendix S3). In our course, students also use lab time to observe and discuss "good food bacteria" by making yogurt and examining Gram stains of yogurt and kefir (protocol not shown).

(iii) Session 3. Students examine plates for growth. They can watch the Frontline video, "The Trouble with Antibiotics" (10) and prepare graphs using the data in Appendix S5 or by using their class data. In our experience, many students struggle to manipulate a fairly large spreadsheet. We prepared a step-by-step video on using Google sheets to analyze and graph data, which our students found helpful (https://youtu.be/qYBq0wmQIBA).

(iv) Session 4. If students isolate a strong lactose fermenter (pink) on MAC plates using this enrichment technique (21), it is almost always *E. coli*. Our students prepare a second isolation streak plate on blood agar that is sent to our campus diagnostic lab for matrix-assisted laser desorption ionization–time of flight mass spectrometry analysis (MALDI-TOF MS), similar to the process in clinical labs (22). Alternatively, students could confirm isolate identities using biochemical tests (e.g., indole, oxidase). In the final lab experiment, students set up Kirby-Bauer disk diffusion assays (23). Students then work on bioinformatics exercises. Each group is given a set of four WGS FASTA files obtained from previous student isolates. In the Genomics Analysis Worksheet (the first portion of Appendix S4), there are step-by-step instructions for using CGE tools to confirm the species, find the serotype and sequence type, and identify antibiotic resistance and virulence genes. Using this information, groups can predict which isolate(s) could cause a UTI and which isolate(s) may have come from chicken (as opposed to contamination from humans). To help instructors choose which sequences to use, Table 1 gives a brief summary of results for all 10 sequences provided in Appendix S1.

(v) Session 5. Students analyze their Kirby-Bauer disk diffusion data. Students can then prepare graphs using the supplied student multidrug resistance data (Appendix S6) or their class data. As in session 3, students will compare their results with published data. Advanced students may propose why there might be differences between the data sets and/or perform statistical analysis.

(vi) Session 6 (optional). For the final section of Report B, students are asked, "How important do you think it is for consumers to choose poultry that has been raised without antibiotics?" In this session, students share their results and arguments.

Suggestions for determining student learning

At the end of each session, students took short quizzes (individual or group) consisting of multiple choice or true/false (T/F) questions; they also completed five pre-/postassessment questions (Appendix S1). Groups of students completed Reports A and B (Appendices S4 and S6). These contained students' literature research and a hypothesis, two graphs, and comparative analysis using data from one published paper and a national database, NARMS (https://www.fda.gov/animal-veterinary/national-antimicrobial-resistance-monitoring-system/narms-now-integrated-data). Using whole-genome sequences and bioinformatic analysis, students predicted whether *E. coli* isolates could cause an extraintestinal infection (extraintestinal pathogenic *E. coli* [ExPEC]), a urinary infection (urinary pathogenic *E. coli* [UPEC]), or likely came from poultry (avian pathogenic *E. coli* [APEC]).

TABLE 1
Genomic characteristics of *E. coli* isolates from retail poultry

Isolate[a]	Virulence gene(s) present[b]			Identified as an ExPEC, UPEC, or APEC strain[c]	Antibiotic resistance class(s)[d]
	ExPEC	UPEC	APEC		
GCC3	*iutA*		*iutA*, *iss*	No	β-Lactam, tetracycline, polymyxin
GCC4	*iutA*		*iutA*	No	Aminocyclitol, aminoglycoside, β-lactam, polymyxin, tetracycline
GCC10			*hlyF*, *iroN*, *iss*, *ompTp*	APEC	Aminoglycoside, β-lactam, tetracycline
GCC14	*kpsMII*	*chuA*	iss	No	Aminoglycoside, folate pathway antagonist
RS249		*chuA*	*iroN*, *iss*, *ompTp*	No	β-Lactam
RS254	*iutA*	*fyuA*	*iutA*, *iroN*, *iss*, *ompTp*	APEC	Aminoglycoside, folate pathway antagonist, tetracycline
RS260	*iutA*		*hylF*, *iroN*, *iss*, *iutA*, *ompTp*	APEC	None
RS271	*iutA*		*iroN*, *iss*, *iutA*, *ompTp*	APEC	β-Lactam
RS275	*iutA*		*hlyF*, *iroN*, *iss*, *iutA*, *ompTp*	APEC	Aminocyclitol, aminoglycoside, tetracycline
RS297	*iutA*	*chuA*, *fyuA*	*iutA*	No	Aminocyclitol, aminoglycoside, β-lactam, folate pathway antagonist, tetracycline

[a]A link to the FASTA sequence file for each of the isolates is present in Appendix S1 in the supplemental material.
[b]The presence of the *ompTp* gene was analyzed through BLAST analysis, separate from the CGE database analysis (instructions can be found at the end of Appendix S4).
[c]ExPEC (extraintestinal pathogenic *E. coli*), UPEC (urinary pathogenic *E. coli*), and APEC (avian pathogenic *E. coli*) strain designations are further explained in Appendix S4.
[d]Only the antibiotic resistance classes are shown. Resistance to disinfectants and other chemicals was also present, but these data are not shown in this table.

Based on Kirby-Bauer data, students determined which antibiotics would be available to treat a potential infection. Finally, students used their analysis to answer the question, "Should consumers purchase chicken raised without antibiotics?"

Sample data

In the first of two graphing assignments (antibiotic prevalence), groups received a spreadsheet containing compiled student data (Appendix S5). Enrichment cultures from six brands of chicken representing two production types were plated on MacConkey plates containing different antibiotics. The spreadsheet details whether coliforms (dark pink) colonies grew on each of the plates. (The coliforms were later confirmed to be *E. coli*.) The spreadsheet contains data from over 200 samples. Students were instructed to create a graph that displayed the percentage of samples from each brand in which resistant coliforms were found. Appendix S1 contains grading rubrics.

Figure 2 contains two examples of student-produced figures with legends. In Fig. 2A, students received full points for the graph but lost one point for the legend because it was missing critical details of the experiment. The legend could have been improved by leaving out obvious details from the graph. In Fig. 2B, students received half of the possible points because it was not clear what the data in this graph represented. In the figure legend, they left out critical experimental details, did not clearly define abbreviations, and did not mention sample numbers for each brand of chicken. As discussed below, most students were able to produce fairly high-quality graphs.

In the second graphing assignment (multidrug resistance), students received a spreadsheet (Appendix S6) containing resistance data for over 200 *E. coli* isolates tested with nine different antibiotics. Instructions were to create a graph depicting multidrug resistance prevalence in isolates from the different brands and production types of chicken. Since this was a challenging assignment for many students during the pilot semesters, we produced a video with detailed instructions (https://youtu.be/uuQQu4fH8dQ).

Figure 3 contains examples of student-produced figures with legends. In Fig. 3A, students received full points. The legend could have been improved by mentioning where data were obtained and by adding a list of the antibiotics used. In Fig. 3B, students received 44% of the possible points because they did not depict the percentage (or ratio) of ⩾0 to ⩾8 antibiotics as instructed, they put the title on the graph instead of in the figure legend, the *y* axis was mislabeled, and the production method was not clearly marked. The figure legend also contained numerous errors.

Safety issues

Our labs adhere to ASM Laboratory Safety Teaching Guidelines (24). Since the laboratory portion of this exercise

FIG 2. Examples of student-generated graphs depicting the incidence of antibiotic-resistant *E. coli* in grocery store chicken for part A of the lab report worksheet. (A) A good example that received 7 out of 8 points using a scoring rubric (Appendix S10). (B) A poor example that received 4 out of 8 points.

involves unknown bacteria with potential antibiotic resistance, it is conducted using BSL-2 guidelines and protocols approved by the University Biosafety Committee. Our students wear disposable gowns and gloves and work under close supervision in BSC; in addition, goggles are used when observing results on lab benches. Data collection is done in a separate area by students who do not handle cultures. Students performed these experiments late in the semester after they had many weeks of practice using serial dilutions, plating, and aseptic technique. One alternative would be for an instructor to perform some or all of the steps so that students do not work with liquid cultures. Alternatively, students can watch a demonstration video of this exercise performed in BSC (https://youtu.be/47cgtltAaxs) and use the data provided here as a dry lab or online exercise.

RESULTS AND DISCUSSION

Field testing

This unit was designed for students in a 2000-level mixed-major allied health microbiology course at an R1 university and used for six semesters ($n = 694$); most students were juniors and seniors, although in some semesters, almost half the class were sophomore nursing students. There were 414 students who completed the in-person laboratory bioinformatics exercises; 280 completed exercises online using data similar to that provided here. Online student groups met once a week via Zoom with an instructor and other students in the section. A portion of the online activities was also used during three semesters for a 3000-level biology majors' microbiology course at an M3 public university ($n = 50$) (see "Possible modifications," below).

Evidence of student learning

Table 2 contains the learning objectives and the types of assessments used along with a student performance summary for individuals and groups enrolled in the Fall 2022 (in-person) section ($n = 67$); these students completed the latest revisions of the exercise and worksheets, similar to those presented here. Individual learning gains were measured in two ways: (i) pre- and postassessment questions were given before the lab unit and again on the last exam (Appendix S1), and (ii) a Genomics worksheet (Appendix S4). Groups were assessed through graphs and their answers to questions submitted in the reports.

(i) Objective 1. A multiple choice pre- and postassessment question was, "One Health is an approach emphasizing . . .(Appendix S1)". Before the unit, 62% of students chose the correct response, ". . . that human health is connected to animal health and our shared environments"; on the last exam, essentially the same number (66%) chose this response. Most other students chose ". . . the importance of global antimicrobial resistance surveillance," perhaps because the unit emphasized surveillance and/or ambiguous wording in the question. Groups answered the essay question, "In your own words, what is the concept of One Health and how does it relate to antibiotic resistance?," on Report A (Appendix S3); the average group earned 86% of the points.

(ii) Objective 2. Individuals answered the T/F question, "The label 'Raised Without Antibiotics' (RWA) refers to poultry raised from hatch to slaughter without antibiotics" (Appendix S1). This was designed to discriminate between RWA and organic production methods; organic allows antibiotics to be administered up to 2 days after hatch. Most students answered correctly before the unit (false, 78%), although the fraction was larger (89%) after the unit. Groups were assessed through two questions on Report A (Appendix S3), "Define the following terms as they relate to poultry production/processing, including antibiotic exposure and cite your sources (Raised Without Antibiotics/No Antibiotics Ever, USDA Certified Organic, Kosher)" and "State your groups' hypothesis regarding the prevalence of antibiotic resistance in different types of chicken (chicken raised without antibiotics and chicken raised conventionally). Explain your reasoning"; students scored 83% and 85% on the questions, respectively.

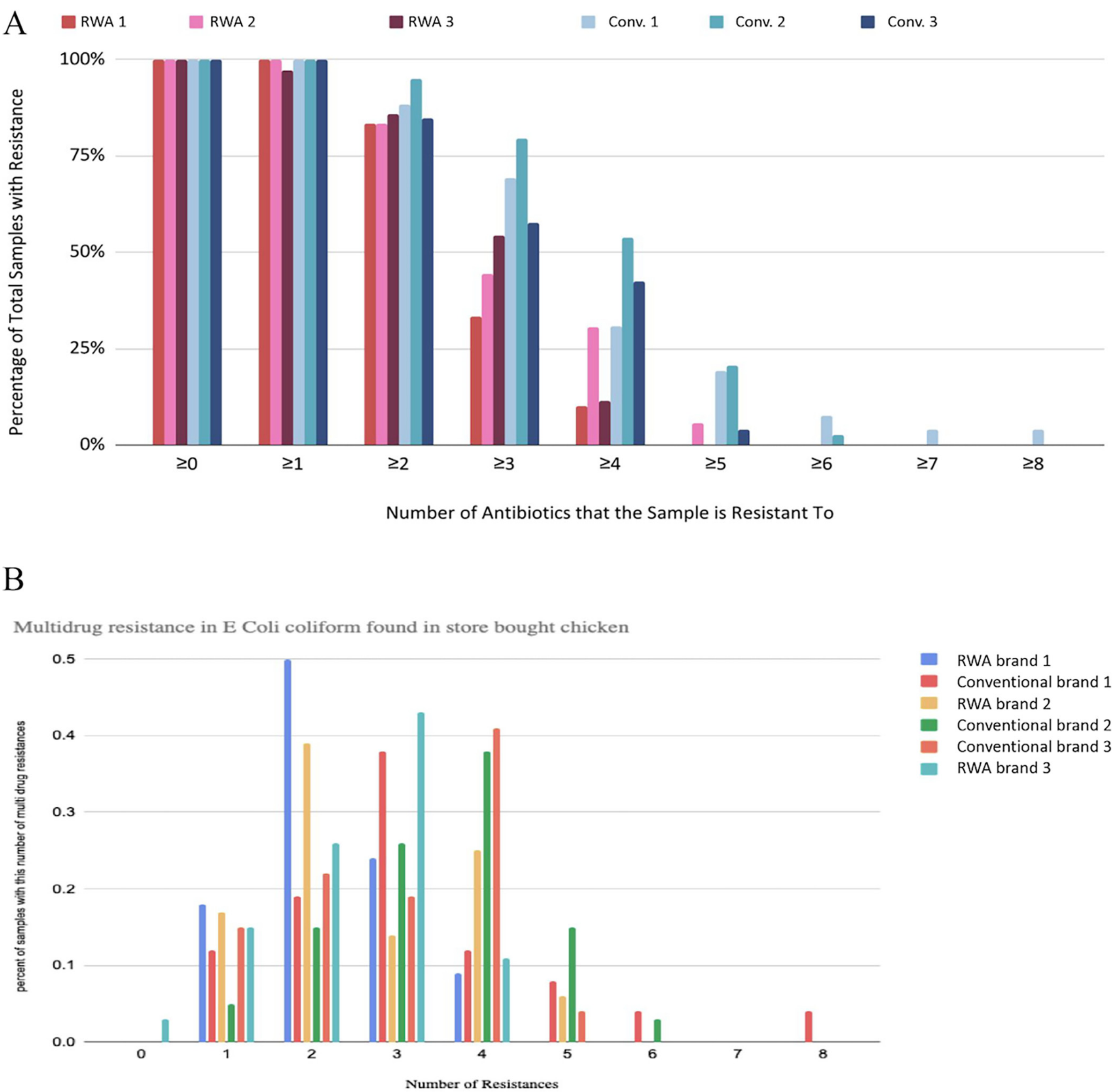

FIG 3. Examples of student-generated graphs depicting the incidence of multidrug-resistant *E. coli* in grocery store chicken for Part A of the lab report worksheet. (A) A good example that received 9 out of 9 points using a scoring rubric (Appendix S10). (B) A poor example that received 4 out of 9 points.

(iii) Objective 3. Groups scored 88% and 91% on graphing exercises in parts 2 and 3 of Report A (Appendix S3), respectively. The quality of graphs increased over previous semesters after two instructional videos were introduced (https://youtu.be/qYBq0wmQIBA, https://youtu.be/uuQQu4fH8dQ). Student examples are shown in Fig. 2; grading rubrics are in Appendix S1.

(iv) Objective 4. Individuals completed the Genomic Analysis Worksheet (Appendix S4) by using a FASTA sequence and several search engines on the CGE website; students scored between 73 and 100% on seven prompts. Individuals in groups compiled their results and completed virulence and resistance gene analysis together, scoring 95% and 86%, respectively.

(v) Objective 5. A T/F pre- and postassessment question was, "Foodborne illness caused by antibiotic-resistant bacteria present on poultry can largely be prevented by cooking meat to an internal temperature of 165°F" (Appendix S1). Before the unit, 86% of students chose the correct answer (true); 97% chose this answer after the unit. We saw a larger gain for the second T/F question: "Raw poultry should be washed before cooking." A minority of students (43%) selected the correct answer (false) before the unit, but by the end, this number

TABLE 2
Learning objectives and assessments used in the study

Learning objective	Assessment(s)	Student performance
i. Explain the concept of One Health and how it relates to antibiotic resistance	Pre- and postassessment quiz question; antibiotic resistance in *E. coli* from grocery store chicken Report A (Appendix S3, Part 1 Q1)	Individuals gained 4% on a pre- and postassessment question; groups scored 86% of points on an essay question
ii. Describe conventional, organic, and raised without antibiotics systems of poultry production and formulate a hypothesis regarding the prevalence of antibiotic resistance on chicken raised in each system	Two pre- and postassessment quiz questions; Report A (Appendix S3, Part 1 Q 3 and 8)	Individuals gained 11% on pre- and postassessment questions; groups scored 83% on essay question 3; 85% of groups received full points for their hypotheses (Q8)
iii. Graphically represent and interpret research data from large spreadsheets	Report A (Appendix S3, Parts 2 and 3)	Groups scored 88% and 91% on graphing exercises in Parts 2 and 3, respectively
iv. Use databases to identify virulence and AMR genes in WGS data; predict pathogenicity and antibiotic susceptibility	Genomic analysis worksheet (Appendix S4), Report B Appendix S4, Part 4B and Part 4C, Q6	Individuals scored 73–100% on 7 prompts; groups scored 95% and 86% on Part 4B and Part 4C Q6, respectively
v. Explain principles of food safety related to poultry; evaluate the merits of "raised without antibiotics" (RWA) and "conventionally raised" (CONV) chicken	Report B (Appendix 4, Part 5)	Individuals gained 9% and 57% points on pre- and postassessment questions; 76% of groups wrote "good" answers to an essay question; groups scored an avg of 96% on a final essay question

rose to 100%. Groups of students answered the prompt, "Is chicken safe to eat? Review the food safety tips outlined by the CDC (https://www.cdc.gov/foodsafety/chicken.html). Write a paragraph intended for a friend or family member to give them advice. In your own words, what should they know?" (Appendix S4). The CDC site contains 10 bullet points of advice to prevent food poisoning. "Good" answers described at least five; 76% of reports contained good answers. The final essay question was, "Should consumers buy RWA poultry? Review your answers in both reports A and B. Discuss this question as a group. How important do you think it is for consumers to choose poultry that has been raised without antibiotics? Use at least three sources of data to support your answer (cite your sources)" (Appendix S4). In earlier versions of the assignment that lacked the requirement for three sources, we noticed a wide range in the quality of evidence-based arguments. With this specification, the average group provided 3.6 arguments to support their position, earning an average of 96% of the points. A little more than one-third (38%) of groups recommended that consumers buy RWA poultry, 19% recommended against it, and 43% were neutral.

We gathered student feedback through an optional survey with 13 multiple choice and 2 open-ended questions during two semesters taught in-person, Fall 2021 ($n = 114$, 97% participation rate) and Spring 2022 ($n = 217$, 95% participation rate) and two semesters taught online, Spring 2021 ($n = 219$, 96% participation rate) and Summer 2021 ($n = 47$, 92% participation rate). The study was submitted to the university IRB and did not qualify as human subject research. The survey was given after reports were turned in; completing the survey allowed students to drop an additional quiz score. Responses from in-person semesters indicated a high level of interest and increased perceived learning gains (Fig. 4 and 5). A minority of students (37%) did not carefully read the labels of grocery store chicken before the project, but the majority (77%) reported they would afterwards. Likewise, a few students (21%) could explain the difference between production types, but nearly all (94%) could after the project. Many students (72%) had previous experience graphing and interpreting research data from large spreadsheets, but 81% reported feeling more confident producing graphs and figures after completing the project. The largest gain was from the bioinformatics segment. Few (11%) had previous experience analyzing whole-genome bacterial sequences, but after the unit, most (76%) felt more confident analyzing genomic sequences. In fact, 55% reported an increased interest in bioinformatics (Fig. 5). Many students commented that they enjoyed the worksheet format of the lab reports. While none of the students rated the project as "very difficult," 15% rated it as "difficult," and 73% found it "moderately difficult." However, 93% agreed that the project should be kept in the course curriculum (Fig. 5). In fact, despite the perceived difficulty, of those who rated the project as "moderately difficult," 49% found it at least "somewhat interesting" and 51% found it "very interesting" (data not shown). Survey results from students in the online sections were similar for the most part (see below).

In summary, the majority of students met the project learning objectives, scoring 90% (in-person) and 91% (online) on Report A and 91% (in-person) and 94% (online) on Report B. (Those completing the latest versions scored 92% on both Reports A and B.) Students in both in-person and online learning modalities reported feeling more confident producing graphs and figures and showed large gains in confidence and interest in bioinformatics. Students in the online sections found the project somewhat more difficult, and surveys indicated that their

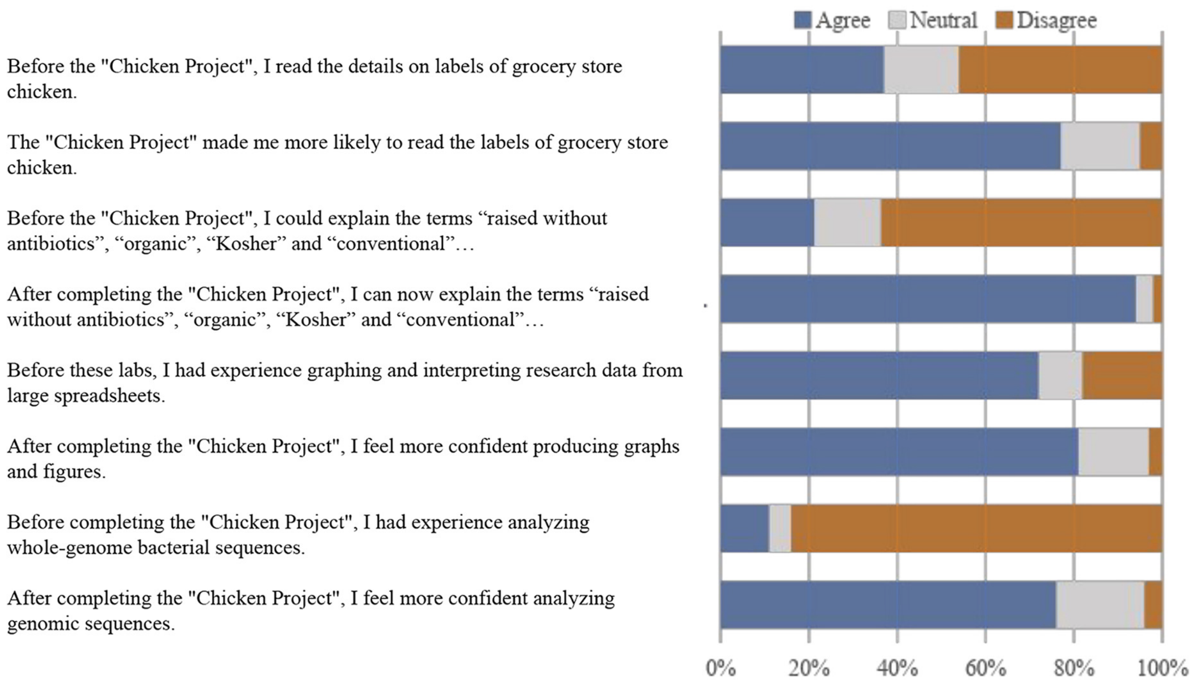

FIG 4. Student perceptions of learning gains. The optional survey was completed after both lab reports, "Antibiotic resistance in *E. coli* isolated from grocery store chicken," parts A and B (Appendices S4 and S6), were turned in. Data are compiled from two semesters that were taught in person ($n = 331$). The answers "Agree" (blue) and "Disagree" (orange) here are a combination of Strongly Agree/Agree and Disagree/StronglyDisagree answers from the original 5-point Likert scale.

perceived learning gains were slightly lower (data not shown). However, 78% recommended keeping it in the curriculum. When answering the question, "Please tell us something you liked about the 'Chicken Project,'" about a third of all students wrote that they liked the real-life relevance of the project; 16% wrote that this project helped them apply knowledge to their daily lives and/or would influence their behavior.

Possible modifications (optional)

We used a modified online version of this exercise for three semesters at a smaller (M3) public university in a microbiology course for biology majors. Student groups of three to four met in a synchronous Zoom setting with an instructor present for two 3-h sessions. Prior to meeting, students were

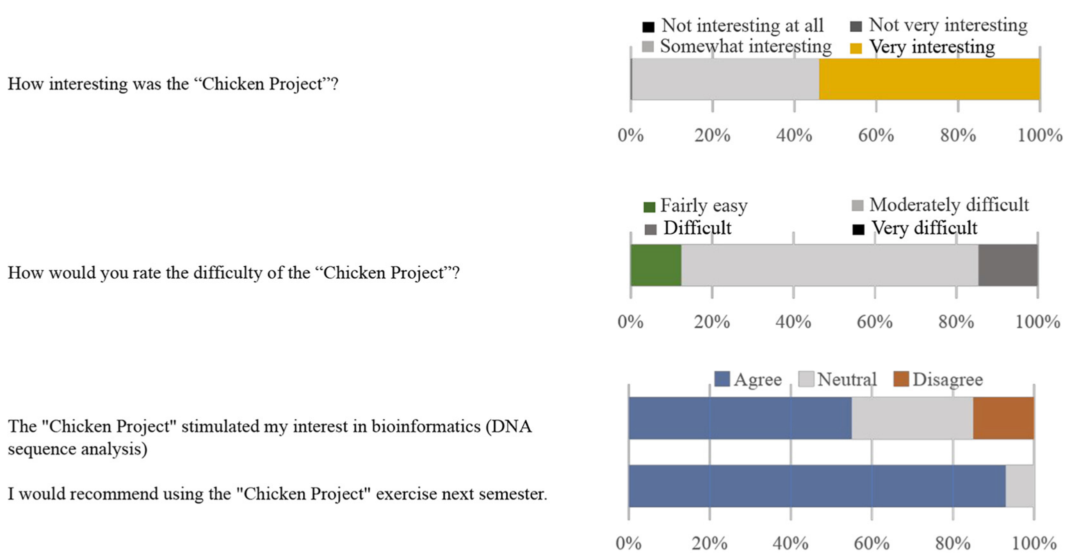

FIG 5. Student's interest levels and perceptions of difficulty. The optional survey was completed after both lab reports, "Antibiotic resistance in *E. coli* isolated from grocery store chicken," parts A and B (Appendices S4 and S6), were turned in. Data are compiled from two semesters that were taught in person ($n = 331$). Answers for "Agree" (blue) and "Disagree" (orange) are a combination of Strongly Agree/Agree and Disagree/Strongly Disagree answers from the original 5-point Likert scale.

given the lab background and graphing videos (Appendix S2). During the lab, they were given Appendix S5 and modified worksheets (Appendix S8), which reduced the amount of work to fit the allotted time. Students took the same survey (described above) at the end of the exercise. Students' general performance was comparable to those at the R1 university who completed the entire exercise; however, scores on graphing figures and figure legends were about two points lower. This was supported by the survey in which 59% of the M3 (data not shown) compared with 81% of the R1 students agreed with the statement, "After completing the 'Chicken Project,' I feel more confident producing graphs and figures" (Fig. 4). One possible reason was that the modified worksheet contained only one graphing exercise instead of two. Also, students at the R1 prepared a modified lab report earlier in the semester in which they were given extensive feedback. On the other hand, the M3 students reported having more confidence in bioinformatics prior to this lab, perhaps because they completed two bioinformatics exercises before this project. In general, the M3 students reported feeling more confident in graphing and bioinformatics; they agreed that the lab increased their understanding of commercial poultry labels and the likelihood of paying attention to them when buying poultry (data not shown). Most of the students rated the lab as moderately difficult but recommended it for future semesters. Suggestions would be to have a debriefing session at the end of the exercise (Fig. 1) and include both graphing exercises.

We did not have students perform statistical analysis because of time limitations and because it was outside the scope of the course. However, this could be a valuable addition. For the same reasons, students were given DNA sequences that were ready to upload to https://www.genomicepidemiology.org/; they did not pre-pare DNA for sequencing or participate in generating FASTA files.

SUPPLEMENTAL MATERIAL

Supplemental material is available online only.

SUPPLEMENTAL FILE 1, PDF file, 1.2 MB.
SUPPLEMENTAL FILE 2, XLSX file, 0.01 MB.
SUPPLEMENTAL FILE 3, XLSX file, 0.01 MB.

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