

Unique Down to Our Microbes—Assessment of an Inquiry-Based Metagenomics Activity †

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Metagenomics is an important method for studying microbial life. However, undergraduate exposure to metagenomics is hindered by associated software, computing demands, and dataset access. In this inquirybased activity designed for introductory life science majors and nonmajors, students perform an investigation of the bacterial communities inhabiting the human belly button and associated metagenomics data collected through a citizen science project and visualized using an open-access bioinformatics tool. The activity is designed for attainment of the following student learning outcomes: defining terms associated with metagenomics analyses, describing the biological impact of the microbiota on human health, formulating a hypothesis, analyzing and interpreting metagenomics data to compare microbiota, evaluating a specific hypothesis, and synthesizing a conceptual model as to why bacterial populations vary. This activity was implemented in six introductory biology and biotechnology courses across five institutions. Attainment of student learning outcomes was assessed through completion of a quiz and students' presentations of their findings. In presentations, students demonstrated their ability to develop novel hypotheses and analyze and interpret metagenomic data to evaluate their hypothesis. In quizzes, students demonstrated their ability to define key terms and describe the biological impact of the microbiota on human health. Student learning gains assessment also revealed that students perceived gains for all student learning outcomes. Collectively, our assessment demonstrates achievement of the learning outcomes and supports the utility of this inquiry-based activity to engage undergraduates in the scientific process via analyses of metagenomics datasets and associated exploration of a microbial community that lives on the human body.

INTRODUCTION

Microbes dominate global ecological processes, they account for most of the deep diversity of lineages, and they directly influence human health and well-being, both as beneficial organisms and through their ability to cause disease in plants, animals, and humans. Metagenomics, the study of the complexity of microbial communities using DNA sequence information, has become a powerful method for determining the taxonomic and genetic composition of our

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microbial world. It provides a first look at which taxa are present in a particular habitat (I). Facilitated by advances in metagenomics approaches, our understanding of the roles that microbes play in our daily lives is changing (2). Knowledge of metagenomics is becoming more important to undergraduate education, for both preparing future biologists and maintaining an informed society. Familiarity with the metagenomics tools we use to detect the presence of microbes is now akin, in its importance, to understanding how to use a microscope. This significance is emphasized by the National Microbiome Initiative (NMI), which will invest more than \$121 million in microbiome science in the 2016-2017 fiscal year, and national curriculum guidelines (e.g., Vision and Change and American Society for Microbiology Curriculum Guidelines) that include student understanding and incorporation of metagenomic concepts in undergraduate biology (3-5). Even more recently, CourseSource.org created a learning framework and bioinformatics learning outcomes to encourage the creation of bioinformatics

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activities appropriate for undergraduate students (6). By integrating metagenomics into introductory biology classrooms, we can prepare both students with future careers in biology and the larger group of students, who will not become biologists but nonetheless need to appreciate and understand complex data like those used in metagenomics studies and the science surrounding them. Such data are likely to play an ever larger role in informing environmental and public health policy decisions in the future, decisions in which each person has a stake.

Metagenomics studies generate large sequence data files (they have become a prototypical example of "big data"), and analysis of these data often requires sophisticated software and considerable computational resources. Despite growing relevance to undergraduate students, these computational demands present a barrier to the broad adoption of metagenomics-based activities in the classroom. Metagenomics research also requires molecular biology techniques, including isolation of sample DNA and preparation of libraries for sequencing. Activities and courses that introduce students to metagenomics in a laboratory environment exist (7-9). However, they are often too advanced, expensive, or dependent on specialized reagents and equipment to be suitable for the average student in an introductory classroom. While development of advanced data analysis and technical laboratory skills is valuable, neither is critical to understand the conceptual basis of metagenomics, which we view as the first step to using metagenomics as a tool.

Here we describe an activity, "Unique Down to Our Microbes...", that provides an opportunity for students to work with metagenomics data and explore the bacterial communities that live on our bodies, even if they do not have access to sophisticated software or wet lab resources. Students use an open-source, web browser-based bioinformatics tool, Phinch.org, to visualize and interact with microbiome data from a citizen-science project that sampled over 270 belly buttons to investigate their microbial biodiversity (10). While the students do not generate new data, they do work with data that have been generated in collaboration with the public and they do generate novel results given those data. By working through the case example, students become immediately aware of the extent to which metagenomics approaches bear on and engage daily, public life. In addition, the citizen science-collected belly button case example includes additional resources that students can engage by visiting the Your Wild Life website (http://navels.yourwildlife.org/). These resources include images of the microbes that grew from the same samples for which metagenomic data are available, the stories of the scientists, and the ways in which other scientists have built upon the initial results of the first belly button biodiversity study. Most importantly, this activity immerses students in the scientific process and introduces them to the concept of metagenomics through a relatable research project, where the process is repeatable, but where the results differ depending on the questions the students ask. Students design a hypothesis, analyze and interpret microbiome data to evaluate their hypothesis, and formulate a model to explain their experimental findings. Through this process they explore how a human host, as an environment, impacts the structure of one, associated microbial community, and they make predictions about how microbes impact human health and society. These are all competencies for life science students, as outlined in Vision and Change (3), and though it is not our focus here, they are also objectives that relate to many components of the high school common core. This activity is also inquirydriven, providing the opportunity for collaborative work on a classroom-based research project. Students demonstrate greater gains in learning, particularly in terms of development of critical thinking, conceptual understanding, retention of content knowledge, and research skills, when they work in collaborative environments and use inquiry-based pedagogies (II-I4).

"Unique Down to Our Microbes..." involves a short lesson and handouts that provide information on the importance of the human microbiota, key definitions, metagenomics techniques (e.g., 16S rRNA gene sequencing), and the scientific method. Students are asked to formulate a hypothesis about the human belly button microbiome and test whether it is supported by selecting relevant metadata for comparison. The purpose of this inquiry-based approach is to provide students with an authentic, yet manageable, metagenomics dataset for analysis with a user-friendly visualization tool. Students encounter the potential of metagenomics analyses, along with limitations and misconceptions. The activity is open-ended and encourages students to be creative and use their imagination. The final project, for example, challenges students to formulate a conceptual model to reflect on the human microbiome in light of whether their experimental hypothesis is supported or refuted. Additionally, this activity is modular by design. The requirements for instruction, in-class support, and assessment are such that it should be adaptable to a variety of introductory life science courses.

Intended audience

This activity is best suited for introductory courses covering microbiology, microbial communities, human health, and/or biotechnology, and is appropriate for life science majors or nonmajors. The activity could also be modified for higher-level life science courses for majors, such as microbiology or molecular biology.

Prerequisite knowledge

Basic computing skills, including use of internet browsers and presentation software (e.g., Microsoft PowerPoint or Google Docs) are required. Basic conceptual understanding of bacteria, differences between eukaryotes and prokaryotes, DNA, DNA isolation, PCR, and DNA sequencing is also helpful, but not required.

Learning time

This activity requires approximately six hours of in-class time and works best if distributed over two or three classes, with designated out-of-class time. An example workflow is presented in Figure 1. Adaptations can be made to use longer lab periods or multiple shorter sessions.

Learning outcomes

Upon completion of this activity, students will be able to:

- Define microbiota, microbiome, metagenomics, and metadata
- Describe the biological impact of the microbiota on human health
- Formulate a hypothesis to address a specific question
- Analyze and interpret metagenomics data to compare microbiota
- 5. Evaluate a specific hypothesis
- Synthesize a conceptual model as to why microbial populations vary

PROCEDURE

Materials

The activity is completed without a wet lab and requires only computational resources. Students will need a computer with internet access. The web browser Google Chrome is recommended for using Phinch.org, an open-access web resource that does not require registration or identifying user information. Because this resource is webbased, instructors should keep in mind that during periods of high use, website response may slow down. We did not experience any slowdown in response negatively impacting completion of the activity as described, with class sizes up to 20 students. However, if lag in responsiveness of the website

is encountered during class time, possible solutions would be to have students work in groups or for the instructor to perform a single demonstration for the class. Completion of the activity should still be possible with out-of-class access to the website. The form of the final project requested will also require access to presentation and/or word processing software. The Microsoft Office Suite (PowerPoint and Word) and Google Apps (Docs or Slides) software are both appropriate. Tutorial videos for use of Phinch.org are highly recommended and are available at https://vimeo.com/103012368 and https://vimeo.com/113492690. The belly button microbiome data file (BIOM format) associated with this activity can be downloaded from http://robdunnlab.com/projects/wild-life-of-our-homes/data-visualization/.

Student instructions

Students should begin the activity by reviewing the Student Handout (Appendix I), the PLOS ONE paper that describes the belly button study (10), and the Your Wild Life website (http://navels.yourwildlife.org/). This is followed by an in-class lesson (Appendix 2), in which the concepts of microbiota, the human microbiome, and metagenomic analysis are introduced. At the end of this lesson, students should load the belly button data file in Phinch and familiarize themselves with the user interface. Viewing the video tutorials described above is advised. Once familiar with the data set, students should complete the Hypothesis Development Worksheet found on pages 6 and 7 of the Student Handout (Appendix I). This worksheet guides identification of a broad question about the human belly button microbiome that interests the student and refines that question into a testable hypothesis. Once a specific hypothesis has been defined and approved by the instructor, students use the Phinch web tool to identify relevant data points and perform a basic analysis to evaluate whether their hypothesis is supported or refuted. For introductory students, analysis may be a simple comparison of the taxonomic diversity between data points. For example, the hypothesis that bacterial composition of

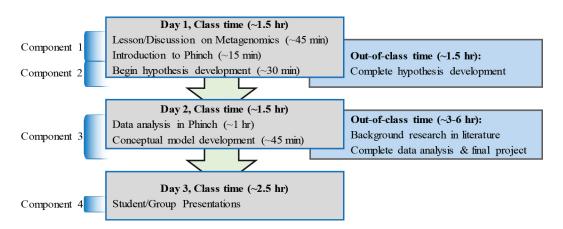


FIGURE 1. Proposed timeline for implementation of the activity. The recommended allotment of time for each component of the activity is indicated. Activity components are divided based on which are suggested for in-class or out-of-class time.

"innie" versus "outie" belly buttons differ may be addressed by comparison of the number of different classes of bacteria in these data sets. Students must identify the data points relevant to their hypothesis and choose a visualization output from Phinch that best addresses this comparison.

This activity is completed with the preparation and presentation of a final project, either a short oral presentation or lab report (refer to page 8 of the Student Handout, Appendix I). Students should include several specific elements in these projects: background information about metagenomics and the human microbiota, description of the specific hypothesis tested, experimental data they chose to analyze, and a clear conclusion as to whether the hypothesis was supported or refuted. Students should also create and present a conceptual model describing how they interpret their findings. This conceptual model should focus on the relationship between the human microbiota and personal health. These elements are detailed in the assessment rubric for final projects (Appendix 3), which students should reference in preparing their final project.

Faculty instructions

Preparation should begin by adapting the activity based on individual course constraints. First, decide what form of the final project (oral presentation or written report) can be accomplished in the available class time. We had success implementing final oral presentations with students working individually and in groups of two to three. Alternatively, written reports do not require class time and may be more suitable for time-restricted courses. Second, define expectations for depth and rigor of analysis. With introductory and advanced students, clear benchmarks will need to be set for what constitutes an acceptable hypothesis, the level of data analysis necessary to evaluate a hypothesis, and quality and depth for the conceptual model. For example, upper-level students might be expected to perform statistical analyses that compare relative abundance or perhaps investigate the metabolism of specific microbes. Introductory students, however, may be expected to only provide basic comparisons of taxonomic diversity.

Once a structure for the class has been established, the materials for the activity should be prepared. Each student or group will need a computer with access to the Internet, a copy of the Student Handout (Appendix I), and access to the belly button data file (i.e., BIOM file). The file can be downloaded via the "belly button microbiome download" link at: http://robdunnlab.com/projects/wild-life-of-our-homes/data-visualization/. We highly recommend downloading a copy of the file locally and distributing it to computers or students through a course website, email, or Learning Management System to avoid any issues with availability the day of the activity. We also recommend that instructors conduct the activity and work with the data set, metadata, and the Phinch.org web tool prior to classroom

implementation. We found that instructor familiarity with the interface and tools at Phinch.org was important to helping students complete the activity.

Student contact time during "Unique Down to Our Microbes..." consists of four modular components outlined in Figure I. Students should review the Student Handout (Appendix I) and the PLOS ONE paper describing the belly button study prior to the activity (10). The first component of the activity is an introductory lesson. We recommend allotting ~45 minutes for this lesson and class discussion. Example slides to deliver the intended content can be found in Appendix 2. Following this lesson, students should be given the chance to work with the data file via the visualization tools at Phinch.org. Instructions for loading the data file are provided on page 5 of the Student Handout (Appendix I).

The second component of this activity requires students to develop a hypothesis to test with the given data set and available metadata. Instructors should allot approximately 30 minutes of class time to helping students begin the Hypothesis Development Worksheet (pages 6–7 of Appendix I), which can be completed outside of class. We recommend that instructors present clear expectations for, and provide guidance through the initial stages of, hypothesis development.

In the third component of this activity, students use Phinch.org to select the data points most relevant to their hypothesis. This is the step that allows instructors to adapt the challenge of the assignment to an appropriate level for their class. Students should be instructed to choose a form of visualization that effectively communicates the comparison they are striving to make. Instructions on exporting/ capturing charts, graphs, or figures from Phinch.org are provided on page 5 of the Student Handout (Appendix 1). We also recommend that students be encouraged to generate their own representations of the data (e.g., charts or graphs) if they are not satisfied with those available in Phinch.org. As part of this activity, students are expected to create a visual model that reflects their tested hypothesis and how the human microbiome impacts personal health or how the human microbiome might be impacted by external factors. Students should be creative and use various artistic and/or multimedia platforms to create their conceptual model. We observed that creativity in the data analysis correlated with development of interesting and unique conceptual models.

The last component of this activity is submitting the final project (oral presentation or written report). This final project should represent the culmination of what the students have learned from this activity and be used to assess the student learning outcomes. Recommended expectations for the content of the final project are outlined in the assessment rubric (Appendix 3). We highly recommend making the rubric available to students during preparation of final projects and referring to it in the lesson when communicating expectations for this activity.

Suggestions for determining student learning

All of the activity student learning outcomes (SLOs) can be assessed by completion of student final projects. The evaluation rubric (Appendix 3) assesses all of the activity learning outcomes. SLOs I and 2 can also be assessed by questions on a post-activity quiz or test. Questions used for this purpose are available in Appendix 4.

Sample data

Example student final projects, oral presentation format, can be found in Appendix 5. Example A received a score of 98/100 and Example B received a score of 93/100.

Safety issues

There are no safety risks associated with this activity.

DISCUSSION

Field testing

This activity was field tested with six different cohorts across five different institutions during the 2015–2016 academic year. Participating campuses included North Carolina State University (NCSU), University of Maryland, Baltimore County (UMBC), North Carolina Central University (NCCU), Washington and Lee University (WLU), and Kalamazoo College (KC). A different instructor conducted the activity at each institution, and Institutional Review Boards at all participating institutions approved or exempted student assessment. A total of 83 students participated in the assessment of this activity.

Classroom composition spanned introductory to upper level students. The UMBC cohort (n = 8) consisted of firstyear students planning to pursue STEM degrees in the biomedical and behavioral sciences. This cohort participated in a Summer Bridge Program and was supported by the NIH-funded STEM BUILD at UMBC Initiative. NCSU contributed two cohorts (fall 2015, n = 8, and spring 2016, n = 12) of students enrolled in an introductory, laboratory-intensive biotechnology course that typically fills either a general education (natural science) or interdisciplinary perspectives requirement. These cohorts consisted of first- and second-year students, with an even mix of STEM and non-STEM degree plans. The NCCU cohort (n = 20) was from an introductory biology course for students pursuing life science degree plans. This cohort consisted of a relatively equal mix of lower- (first or second year) and upper-level (third or fourth year) undergraduate students. The WLU cohort (n = 18) was from a sophomore-level genetics lab course consisting of students pursuing life science degree plans. The KC cohort (n = 17) was from a general and medical microbiology course and consisted of upper-level (third and fourth year) students pursuing biology and chemistry degree plans.

Evidence of student learning

Student learning was assessed in multiple forms over the course of the activity. First, student final projects (oral presentation format) were implemented in four of six cohorts (UMBC, NCSU fall 2015, NCSU spring 2016, and KC) and assessed each student learning outcome (Table I), as evaluated with the rubric provided in Appendix 3. The average total grade for all participating students/groups (n = 37) was 87.9% ± 7.3%. Two example presentations are included in Appendix 5. Separating scores by rubric item allowed for assessment of individual student learning outcomes. Students/groups scored greater than 80% on rubric items associated with all learning outcomes except SLO I. The highest scores (>92% average) were achieved with SLOs 3, 4, and 5. Given that student performance was highest on final project components associated with SLOs 3 to 6, it can be concluded that this activity results in the attainment of higher-order Bloom's taxonomy cognitive domain learning outcomes. SLOs 3 to 6 also relate to core competencies outlined in Vision and Change (3), such as the ability to apply the process of science, use modeling and simulation, and understand the relationship between science and society.

Pre- and post-activity quizzes were administered before students received any materials/lessons relating to the activity and after the activity was completed, respectively (Appendix 4). Students were assessed for their ability to define key terms (SLO I) and identify the importance of the microbiota to human health (SLO 2) in nine questions. A significant increase (p < 0.001, t = 4.156) was observed in aggregate average quiz scores (post-versus pre-activity), as measured by paired Student's t-test (two-tailed) (Fig. 2). Analyzed individually, three cohorts demonstrated significant increases in quiz scores: UMBC (p = 0.036, t = 2.593), NCSU fall 2015 (p = 0.040, t = 2.510), and KC (p = 0.005, t = 0.0053.225). Three cohorts did not demonstrate significant gains in quiz scores: NCCU (p = 0.751, t = -0.322), NCSU spring 2016 (p = 0.097, t = 1.817), and WLU (p = 0.064, t = 1.982). Additionally, a significant increase (p < 0.05) was observed in some, but not all questions (Appendix 4). Though we did observe statistically significant increases in average quiz grades in aggregate, the disparity in improvement in select cohorts and in individual questions suggest SLOs I and 2 require greater coverage. To ensure gains in these SLOs, we recommend that instructors emphasize and revisit SLOs I and 2 throughout the activity. This could be accomplished by repeating a short lesson of what was learned at the conclusion of the activity and/or reviewing the terminology and importance of the microbiota to human health on multiple occasions throughout this activity via group discussion.

Perceived gains in student learning outcomes were assessed at the conclusion of the activity in a survey included with the post-activity quiz. Using a Likert-type scale (I = strongly disagree, 5 = strongly agree), students reported perceived gains in all of the activity learning outcomes (Table 2). The greatest gains were reported for SLOs 3

TABLE I. Assessment of final projects for student learning outcomes.

Rubric Item – Knowledge or Skill		Total			
	UMBC (n = 7)	NCSU Fall '15 (n = 7)	NCSU Spr '16 (n = 14)	KC (n = 9 ^a)	— (n = 37)
SLO I – Terminology Definitions /5 points	57.1%±13.8%	63.3%±26.6%	90.0%±15.2%	71.1%±14.5%	74.1%±21.0%
SLO 2 – Human Health Impact /5 points	65.7%±25.1%	60.0%±40.0%	91.4%±17.0%	100.0%±0.0%	83.2%±26.0%
SLO 3 – Hypothesis Development /20 points	98.6%±3.8%	84.2%±8.6%	93.6%±6.3%	91.1%±7.4%	92.4%±7.7%
SLO 4 – Analyze and Interpret Data /15 points	92.4%±13.0%	90.0%±11.7%	96.7%±7.3%	89.6%±8.9%	93.2%±9.7%
SLO 5 – Evaluate Hypothesis /20 points	95.7%±11.3%	93.3%±11.7%	91.8%±11.0%	95.0%±5.6%	93.4%±9.8%
SLO 6 – Synthesize Conceptual Model /20 points	81.0%±17.0%	92.4%±9.8%	82.4%±16.5%	79.3%±13.5%	83.2%±15.0%
Total Grade ^b /100 points	89.9%±4.2%	84.1%±9.8%	88.6%±8.9%	88.4%±3.3%	87.9%±7.3%

^aKC cohort completed the final project in groups of two students.

UMBC = University of Maryland, Baltimore County; NCSU = North Carolina State University; KC = Kalamazoo College; SLO = student learning outcome.

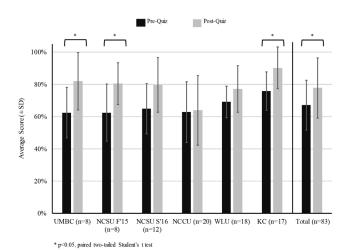


FIGURE 2. Pre- and post-quiz scores assessing attainment of SLOs I and 2. Students were administered a quiz assessing SLOs I and 2 in a pre-/post-activity manner, with the average (\pm SD) scores for each cohort displayed.* p < 0.05 using paired, two-tailed Student's t-test. SLO = student learning outcome; UMBC = University of Maryland, Baltimore County; NCSU F'15 = North Carolina State University, Fall 2015; NCSU S'16 = North Carolina State University, Spring 2016; NCCU = North Carolina Central University; WLU = Washington and Lee University; KC = Kalamazoo College.

and 5, formulating and evaluating a hypothesis to address a specific question. The lowest gain was reported in SLO 6. This is an interesting result, given that students performed well on the SLO 6 component of the final project (Table 1).

Student perception of gains in SLO 6 may reflect a lack of confidence in their ability to create a model to explain their experimental findings. Thus, the emphasis on developing this advanced cognitive skill may be a strength of this activity.

In addition to evaluating learning gains, we collected student opinions of the activity in the post-activity survey. Students were asked to indicate level of enjoyment and submit frustrations. Most students reported enjoyment of the activity (Fig. 3). However, the most commonly reported student frustrations related to a desire to be able to sort and visualize the data differently than Phinch.org allowed, and to perform mathematical operations (Appendix 6). Many of these responses indicate a desire to analyze the data to greater depth than we had envisioned for the activity. To address these responses, we recommend that instructors clearly define their expectations for data analysis and familiarize themselves with Phinch.org before attempting the activity with students. We also recommend that instructors: I) perform an example analysis for students, 2) explicitly state that this program has constraints on the operations it can perform, and 3) point out that these constraints reduce computational demand on the software and make it possible to work with such a large data set.

Overall, we observed achievement of the activity SLOs in our student population. We purposefully sought a broad pool of instructors and students in order to evaluate how well this activity can be incorporated in different classes. Though differences were observed in the performance of individual cohorts, our student population as a whole made noticeable gains in the activity's learning outcomes.

^bTotal grade includes rubric items not listed in this table.

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TABLE 2. Average Likert-type score of student-perceived gains in knowledge and skills associated with this activity.

By participating in this activity, I gained the ability to	Cohort Average Likert-Type Score ^a						Total
	UMBC (n = 8)	NCSU Fall '15 (n = 8)	NCSU Spr'16 (n = 14)	NCCU (n = 20)	WLU (n = 18)	KC (n = 18)	(n = 86)
Define terms microbiota, microbiome, metagenomics, and metadata (SLO I)	4.50±0.76	3.63±1.30	4.50±0.52	4.05±0.51	3.47±1.07	4.22±0.73	4.05±0.87
Describe the biological impact of microbiota on human health (SLO 2)	4.13±0.83	3.75±1.16	4.29±0.47	4.25±0.64	3.71±1.05	3.83±0.71	4.00±0.82
Formulate a hypothesis to address a specific question (SLO 3)	4.50±0.76	4.25±1.39	4.21±0.58	4.35±0.59	4.00±1.17	4.11±0.76	4.21±0.86
Analyze metagenomics data (SLO 4)	4.50±0.53	3.88±1.25	4.14±0.66	3.55±0.83	4.06±1.03	4.00±0.97	3.96±0.92
Interpret metagenomics data to compare microbiota (SLO 4)	4.38±0.52	3.88±1.25	3.93±0.73	3.55±0.89	4.00±1.00	3.89±0.90	3.88±0.91
Evaluate a specific hypothesis (SLO 5)	4.50±0.53	4.00±1.31	4.21±0.58	4.10±0.72	4.00±1.00	4.06±0.73	4.12±0.81
Synthesize a model on why microbial populations vary (SLO 6)	4.13±1.13	3.63±1.19	4.00±0.68	3.74±0.99	3.82±1.13	3.56±0.92	3.79±0.98

a Likert scale: I = strongly disagree, 2 = disagree, 3 = neither agree nor disagree, 4 = agree, 5 = strongly agree.

UMBC = University of Maryland, Baltimore County; NCSU = North Carolina State University; NCCU = North Carolina Central University; WLU = Washington and Lee University; KC = Kalamazoo College; SLO = student learning outcome.

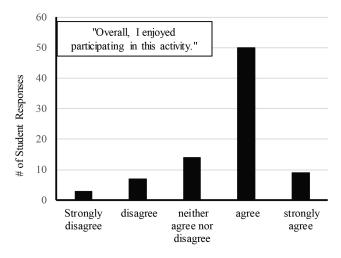


FIGURE 3. Post-activity assessment of student enjoyment. Students (n = 83) were asked to respond to the indicated statement in the post-activity quiz. Bars indicate the total number of students who provided each response.

Possible modifications

Refer to Appendix 7 for a list of possible modifications to this activity.

SUPPLEMENTAL MATERIALS

Appendix 1: Student handout Appendix 2: Sample lesson slides

Appendix 3: Final project rubric

Appendix 4: Pre- and post-activity surveys

Appendix 5: Examples of student final projects

Appendix 6: Example student comments about the activity

Appendix 7: Possible modification to activity

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REFERENCES

- Torsten TJG, Folker MA. 2015. A 123 of metagenomics, p 1–9. In Nelson KE (ed), Genes, genomes and metagenomics: basics, methods, databases and tools. Springer, New York, NY.
- Blaser MJ, Cardon ZG, Cho MK, Dangl JL, Donohue TJ, Green JL, Knight R, Maxon ME, Northen TR, Pollard KS, Brodie EL. 2016. Toward a predictive understanding of

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- Earth's microbiomes to address 21st century challenges. mBio 7:e00714–16.
- American Association for the Advancement of Science. 2011.
 Vision and Change in Undergraduate Biology Education: A Call to Action: a summary of recommendations made at a national conference organized by the American Association for the Advancement of Science, July 15–17, 2009. Washington, DC.
- American Society for Microbiology. 2012. Recommended curriculum guidelines for undergraduate microbiology education. American Society for Microbiology, Washington, DC.
- Handelsman J. 2016. Announcing the national microbiome initiative. Available at: https://www.whitehouse.gov/blog/2016/ 05/13/announcing-national-microbiome-initiative.
- Rosenwald AG, Pauley MA, Welch L, Elgin SCR, Wright R, Blum J. 2016. The CourseSource bioinformatics learning framework. CBE Life Sci Educ 15:le2.
- Gibbens BB, Scott CL, Hoff CD, Schottel JL. 2015. Exploring metagenomics in the laboratory of an introductory biology course. J Microbiol Biol Educ 16:34.
- 8. Muth TR, McEntee CM. 2014. Undergraduate urban metagenomics research module. J Microbiol Biol Educ 15:38.

- Robertson-Albertyn S, Hardee E, Stanley-Wall NR. 2016. Microbe motels: an interactive method to introduce the human microbiome. I Microbiol Biol Educ 17:282.
- Hulcr J, Latimer AM, Henley JB, Rountree NR, Fierer N, Lucky A, Lowman MD, Dunn RR. 2012. A jungle in there: bacteria in belly buttons are highly diverse, but predictable. PLOS One 7:e47712.
- Cracolice M. 2009. Guided inquiry and the learning cycle, p 20–34.
 In Pienta N, Cooper M, Greenbowe T (ed), Chemists' guide to effective teaching. Prentice Hall, Upper Saddle River, NJ.
- 12. Gormally C, Brickman P, Hallar B, Armstrong N. 2009. Effects of inquiry-based learning on students' science. Int J Scholarsh Teach Learn 3:1–22.
- 13. Kober N. 2015. Designing instruction, p 89–120. *In* Reaching students: what research says about effective instruction in undergraduate science and engineering. The National Academies Press, Washington, DC.
- National Research Council. 2015. Reaching students: what research says about effective instruction in undergraduate science and engineering. The National Academies Press, Washington, DC.