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Tips and Tools

A case study to engage students in evolutionary thinking around antibiotic resistance using the MEGA-plate experiment

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ABSTRACT In this article, we describe curricular materials developed to engage undergraduate students in evolutionary thinking around antibiotic resistance using the MEGA-plate experiment (Microbial Evolution and Growth Arena). This elegant and visual experiment, developed by the Kishony Lab, shows the development of antibiotic resistance on the timescale of hours and days. It not only provides important biological insights but also captures students' attention, making it a very useful tool for education. While a short video describing the method and major results has already been widely used in the classroom setting, our case study connects details of the methods and results of the MEGA-plate experiment and antibiotic resistance to core biological concepts. The interrupted case study consists of four major parts: 1) an opening hook activity to capture students' attention and introduce the antibiotic crisis, 2) a jigsaw activity to research different classes of antibiotic targets and the resistance mechanisms that can arise, 3) a discussion of antibiotic resistance in real-time using the MEGA-plate experiment video, and 4) three different options for students to dive deeper into the experimental data from the MEGA-plate research article. These components are modular and can be used in many different combinations to reach different audiences or connect to other topics related to microbiology, evolution, or genetics.

KEYWORDS antibiotics, antibiotic resistance, bacteria, evolution, genomic analysis, MEGA-plate, microbiology, natural selection

While antibiotics are one of the most significant advances in medicine, the accelerating development of resistance is outpacing the discovery and development of new antibiotics (1, 2). This increasing ineffectiveness of drugs against infectious diseases is referred to as an "antibiotic crisis" (2). Education about the crisis and potential solutions are critical for students pursuing careers in science and health care as well as the general public.

In the article "*Spatiotemporal microbial evolution on antibiotic landscapes,*" the laboratory of Dr. Roy Kishony described an elegant experimental method to study and visualize the development of antibiotic resistance in real-time (3, 4). Their setup, called the MEGA-plate (Microbial Evolution and Growth Arena), is a giant Petri dish divided into sections containing either none or increasing concentrations of an antibiotic. As bacteria inoculated into the plate progressed into sections containing higher concentrations of antibiotics, their movement was tracked, showing the development of resistance on the timescale of hours and days. The article describing their method also demonstrates many important results and insights gained from the technique, making it a powerful resource in the understanding of, and fight against, antibiotic resistance (3). When a short video explaining their technique and the major results was released, it went viral with millions of views on YouTube, capturing the attention of not only scientists but also of many news outlets and the general public (5).

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To further communicate the antibiotic crisis and leverage the MEGA-plate experiment, we have designed curricular materials to engage undergraduate students in evolutionary thinking. We used the case study method (6) as our curriculum model to actively engage students, make science content relevant, and improve critical thinking (2, 7–9). Our case study, "Survive and Conquer: Dissecting the MEGA-plate Experiment," consists of four major parts: 1) an opening hook activity to capture students' attention and introduce the antibiotic crisis, 2) an assignment to research different classes of antibiotic targets and the common mechanisms of antibiotic resistance, 3) a discussion of antibiotic resistance in real-time by using the MEGA-plate experiment video, and 4) an analysis activity to further dissect the MEGA-plate experiment (Fig. 1). These activities are modular and can be used in many different combinations, or connected to other activities, to reach a variety of audiences. While the modules have been implemented in lower-level microbiology courses for allied health majors and upper-level microbiology courses for biology majors, parts of the case could also be useful in other courses related to microbiology, genetics, genomics, or evolution. This aligns with the ASM curriculum guideline statements 3 and 7 and the Vision and Change core concepts of Evolution and Structure and Function (10, 11). The learning objectives of the case study are for students to be able to

1. Define antibiotics and antibiotic resistance.
2. Determine the causes, impacts, and potential solutions of the antibiotic resistance crisis.
3. Describe mechanisms of action and targets of antibiotics. Categorize the spectrum of action, selective toxicity, and mechanisms of resistance to specific antibiotics.
4. Describe the methods used and interpret data from the MEGA-plate experiment.
5. Identify the biological processes that occur in the development of antibiotic resistance (mutation, reproduction, selection, and evolution) and the concept that evolution can occur in a short time span.

PROCEDURE

The full set of case study activities and materials, including the case study handouts to be used with students (S1); thorough teaching notes containing implementation tips, timing, and additional resources (S2); a detailed answer key (S3); a set of presentation slides for guiding students through activities and facilitating discussion (S4); and a final assessment (S5), are included in the Supplemental Materials. Completing the full case study requires two or three class periods (75- and 50-minute periods, respectively), but specific activities can be used individually, requiring reduced time (Fig. 2). Basic bacterial cell structure and bacterial replication is fundamental knowledge for this case study.

Briefly, this interrupted case study is broken into four parts that are given to students, one at a time, and worked through in succession. Part 1 uses a data interpretation activity and a video to capture students' attention, help define the "antibiotic crisis" (and key terms: antibiotic and resistance), and identify its causes, impacts, and potential solutions. In Part 2, students perform out-of-class research and an in-class jigsaw activity to identify the main categories, targets, and features of antibiotics, including the spectrum of action, selective toxicity, and mechanisms of resistance. Part 3 helps students visualize the evolution of antibiotic resistance in real-time using the video of the MEGA-plate experiment (5). Students review concepts and make predictions before watching the video and then answer follow-up discussion questions to evaluate their understanding. The final section (Part 4) takes students deeper into the MEGA-plate experiment. It includes three options so that instructors can choose the one that best matches their audience and/or goals. Option 4A focuses on genomic methods, experimental design, and applying knowledge of antibiotic resistance mechanisms. Option 4B focuses on analyzing data figures from the MEGA-plate paper, similar to a data nugget (6). Option 4C focuses on scientific literacy as students read the MEGA-plate article (3) and answer discussion questions. Together, these diverse activities allow for engagement

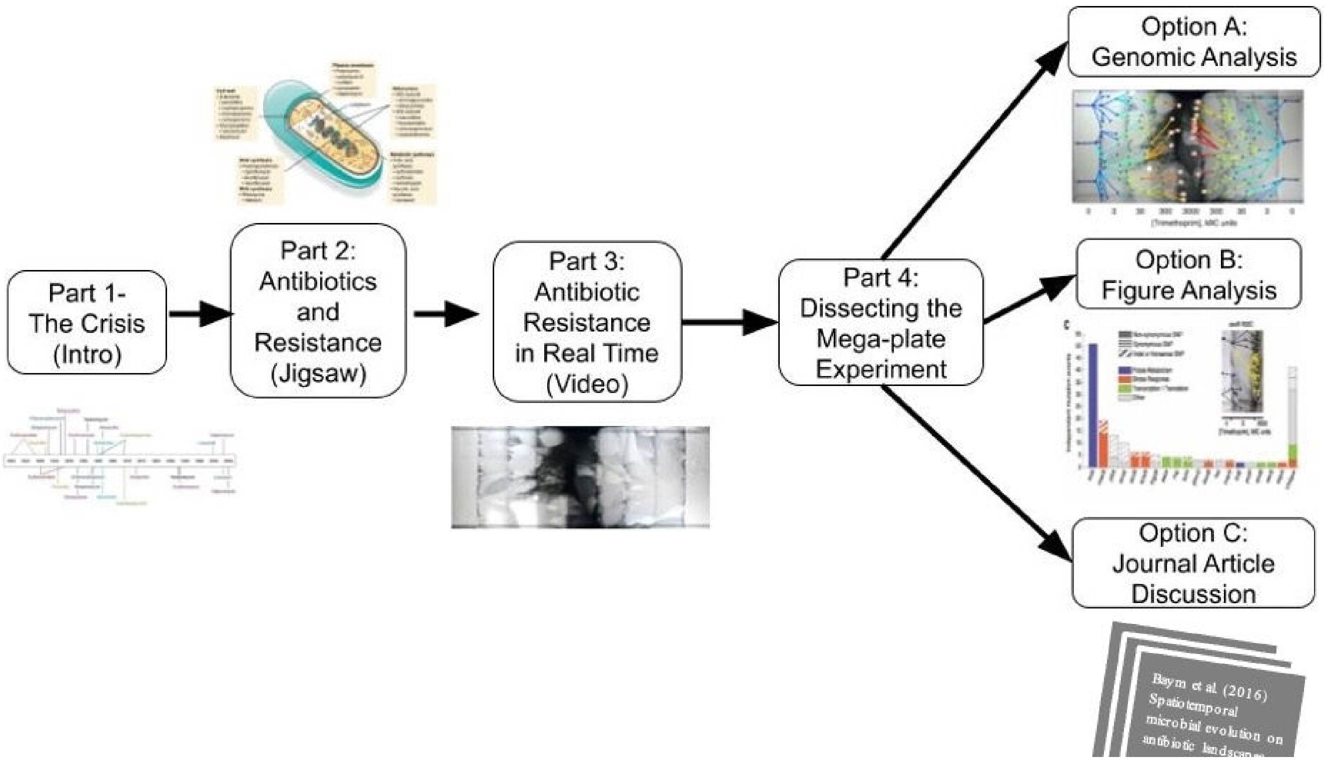

FIG 1 Flow chart of the recommended progression of the presented case study activities. Images are adapted from Clatworthy et al. (12) with permission of Springer Nature (part 1), Parker et al. (13) (under a Creative Commons license) (part 2), and Baym et al. (3) with permission of AAAS (parts 3 and 4).

with the material in a variety of ways to achieve the learning objectives. There are no laboratory safety issues to consider for these lecture-based activities.

CONCLUSION

The case study activities have been successfully implemented multiple times at both primary undergraduate institutions and large public research institutions with different audiences (lower-level allied health and upper-level majors). The case generates strong engagement among students, which can be seen in their attentiveness, interest in

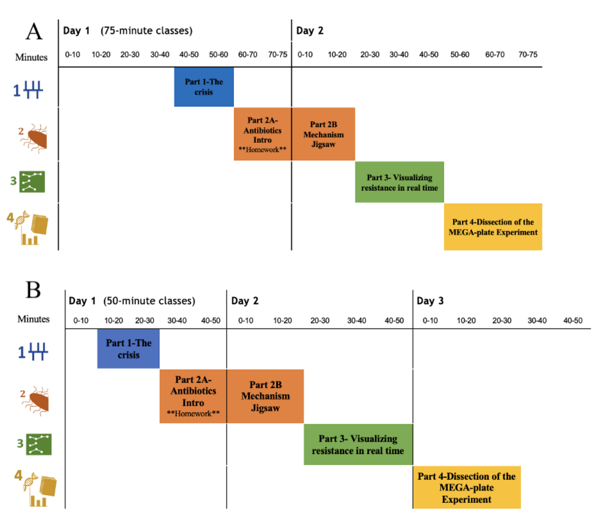

FIG 2 Case study implementation timeline for 75-minute (A) and 50-minute (B) classes.

the topic, the questions they ask, and the interactions they have with other students and the instructor. In particular, the fact that it only takes 11 days in the MEGA-plate experiment to result in growth of bacteria at extremely high antibiotic concentrations leads to surprise among students, promoting curiosity that drives them through the activities for a deeper understanding. Preliminary results of examining student products from the case and their responses on exams following the activities suggest they are achieving the desired objectives. In one implementation, over 90% of the students were able to identify the molecular basis of antibiotic resistance (Learning Objective 5). After the case study, students are increasingly aware of antimicrobial stewardship, which is broadly applicable to anyone using antimicrobials but particularly impactful for future antibiotic prescribers. Overall, the case study helps students understand some fundamental concepts of a typical microbiology course, while engaging them in a variety of active learning exercises and allowing them to go beyond the basic course material to connect the concepts to real-world applications and cutting-edge research.

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ADDITIONAL FILES

The following material is available online.

Supplemental Material

Supplemental Material S1 (jmbe00039-24-S0001.docx). Case study for students.
Supplemental Material S2 (jmbe00039-24-S0002.docx). Teaching notes for instructors.
Supplemental Material S3 (jmbe00039-24-S0003.docx). Answer key for case study document.
Supplemental Material S4 (jmbe00039-24-S0004.pptx). Presentation slides used to present the case study.
Supplemental Material S5 (jmbe00039-24-S0005.docx). Assessment.

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