

BioBits® Antibiotic Resistance Lab: Visualizing How Antibiotics Work and Mechanisms of Antibiotic Resistance

Ally Huang¹, Jessica C. Stark², Bruce C. Bryan¹, and Allison M. Nishitani¹

¹miniPCR bio, 1770 Massachusetts Ave, Cambridge, MA 02140, USA

²Stanford University, Department of Chemistry and Stanford ChEM-H, Street Address, Stanford, CA 94305, USA

(ally@minipcr.com; jcstark@stanford.edu; bruce@minipcr.com; allison@minipcr.com)

Antibiotic resistance is a pressing issue that is widely covered in college biology courses. Here, we present a hands-on laboratory exercise to experimentally demonstrate how antibiotics work as well as how cells can become resistant to antibiotics. The use of freeze-dried, cell-free protein synthesis reactions instead of live bacterial cultures makes this exercise highly accessible with minimal equipment requirements. Students add antibiotics that inhibit protein production to cell-free reactions and observe the effects using a fluorescent protein as a visual readout of translation. Additionally, students express an antibiotic resistance gene to test how resistance genes can rescue translation inhibition in an antibiotic-specific manner. The initial implementation of this exercise demonstrated that the activity is easy to implement, engages student interest in topics related to antibiotic resistance, and has the potential to be widely integrated into undergraduate courses.

Keywords: antibiotics, antibiotic resistance, biology, biotechnology, inquiry-based learning

Introduction

The rise of antibiotic resistance is one of the great public health challenges of the 21st century. While lab activities that demonstrate whether bacteria are susceptible to antibiotics are fairly common, lab activities that focus directly on mechanisms of antibiotic action and mechanisms of antibiotic resistance are limited.

The BioBits® Antibiotic Resistance lab is an easy-to-implement, hands-on activity that clearly demonstrates antibiotic function and antibiotic resistance using visual fluorescent readouts. In this activity, students use the BioBits® cell-free system to demonstrate 1) how specific antibiotics inhibit translation, and 2) how antibiotic resistance genes confer resistance to their target antibiotics.

The BioBits® cell-free protein expression system consists of freeze-dried pellets stored in 200 µl tubes. Each BioBits® pellet contains necessary reagents and cellular components required to perform

transcription and translation *in vitro*. By adding water and a plasmid containing a coding sequence with the correct promoter, proteins can be quickly produced without any of the difficulties of culturing live organisms. As this cell-free protein expression system was derived from *E. coli* bacteria, antibiotics that target protein expression in bacteria will also inhibit the BioBits® cell-free reactions. As such, BioBits® cell-free reactions are an excellent experimental model for antibiotic function.

Students use the BioBits® cell-free system to express red fluorescent protein (RFP) in the presence and absence of two different antibiotics: kanamycin and streptomycin. Both antibiotics inhibit translation by interrupting the function of the ribosome. In the absence of antibiotics, students will observe red fluorescence, indicating that RFP has been translated successfully. In the presence of either antibiotic, red fluorescence will be absent, indicating that the antibiotic inhibited translation. In this way, students can use a clear visual readout—the presence or

absence of red fluorescence—to assess if translation has been inhibited.

Students then test the function of an antibiotic resistance gene by first expressing the resistance gene (again using the BioBits® cell-free system) and then attempting to express the red fluorescent protein in the presence of the same two antibiotics. The addition of this particular antibiotic resistance gene will rescue translation in the presence of kanamycin, but not streptomycin, clearly demonstrating the specificity of antibiotic resistance mechanisms.

Experiment overview

To implement the lab in the classroom, we recommend that the class be divided into two test groups. One group will investigate kanamycin, while the other will test streptomycin.

Students will set up four reactions using the BioBits® cell-free system.

| Tube | Reagents added | Condition |
|------|---|--|
| 1 | H ₂ O only | Negative control |
| 2 | RFP plasmid | Positive control for successful translation and protein expression |
| 3 | RFP plasmid + antibiotic | Test for antibiotic mechanism |
| 4 | RFP plasmid + antibiotic + antibiotic resistance gene | Test for rescue by antibiotic resistance gene |

Students will set up a negative control where only water is added to the BioBits® pellet (tube 1), as well as a positive control where plasmid DNA encoding a red fluorescent protein (RFP DNA) is added (tube 2). Observing red fluorescence under blue light illumination will indicate that protein expression has occurred.

To demonstrate the antibiotics' mechanism of action, each group will add either kanamycin or streptomycin in addition to the RFP DNA (tube 3). Both kanamycin and streptomycin inhibit protein synthesis by binding to the bacterial ribosome and inhibiting ribosomal function. Thus, the addition of either kanamycin and streptomycin results in a lack of red fluorescent protein expression, demonstrating that both of these antibiotics inhibit protein expression.

To demonstrate the specificity of antibiotic resistance genes, students add a plasmid that encodes an antibiotic resistance gene (tube 4). After a 10-minute incubation to give the cell-free system time to express the antibiotic resistance gene, the

RFP DNA and the assigned antibiotic are added to the tube. This particular antibiotic resistance gene

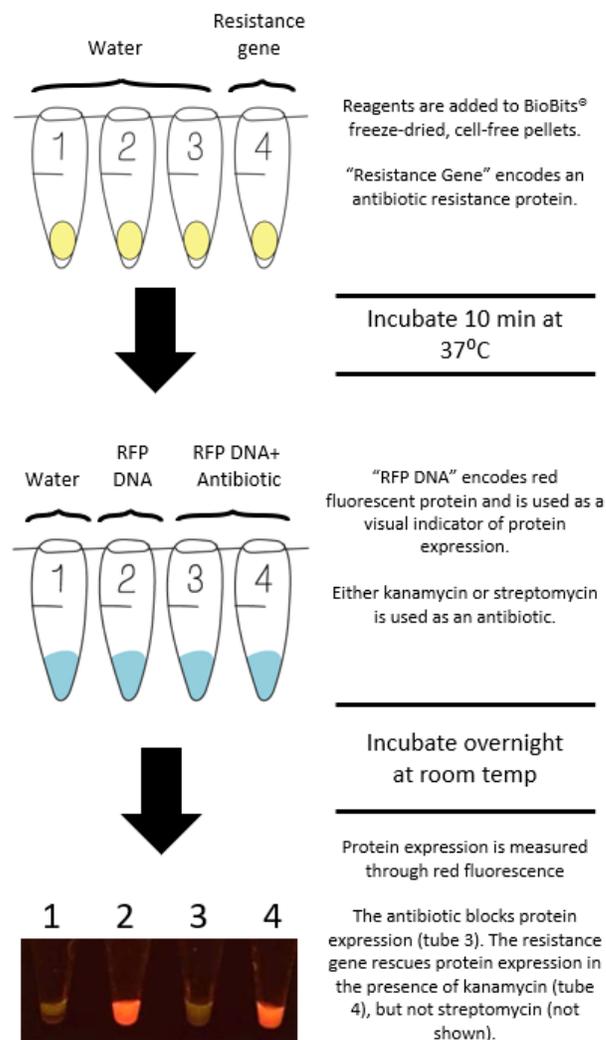


Figure 1: Overview of experimental protocol. Red fluorescent protein is expressed in cell-free reactions in the presence of different combinations of antibiotic and an antibiotic resistance gene. The presences or absence of red fluorescence serves as a readout for whether protein expression was successful.

encodes the enzyme kanamycin kinase. Kanamycin kinase phosphorylates kanamycin and disrupts its ability to bind to the ribosome. The addition of this resistance gene plasmid (Kinase DNA) only rescues red fluorescent protein expression in reactions inhibited by kanamycin but not in reactions inhibited by streptomycin.

Through this exercise, students will be able to observe the function of two antibiotics in a highly visual system, as well as demonstrate that antibiotic resistance can be conferred through the expression of specific antibiotic resistance genes.

The use of the BioBits® cell-free system allows for a simple protocol that can be implemented with minimal equipment requirements in virtually any laboratory or classroom setting. We are unaware of another activity that allows for direct experimentation

with antibiotic and resistance mechanisms that can be performed without the use of live organisms, with such few resources, or in as little time.

Student Outline

Objectives

- Experimentally model one mechanism for how antibiotics specifically target cellular functions and cite examples of other mechanisms
- Experimentally model mechanisms for how cells can defend against antibiotics and cite examples of other mechanisms
- Experimentally model one mechanism for how cells can acquire antibiotic resistance and cite examples of other mechanisms

Introduction

Antibiotics and human health

Since the widespread distribution penicillin in the 1940s, people have taken for granted that bacterial infections can be quickly and reliably treated with antibiotics. Today, there are over 100 different antibiotics that can be used to treat bacterial infections, and together, they have prevented countless patient deaths.

With the widespread use of antibiotics however, a new problem is emerging: the decreasing efficacy of these antibiotics due to the rise of bacteria that have become resistant to them. Today, the spread of antibiotic resistance in bacteria represents one of the great public health challenges of our time. At least 700,000 deaths a year are due to “superbugs” that are resistant to all known antibiotics (World Health Organization 2018). Experts estimate that if no action is taken to counteract antibiotic resistance, this number could rise to 10 million deaths every year by 2050 (World Bank 2017).

How antibiotics work

An antibiotic is a chemical that specifically targets and inhibits bacterial function or growth. Each antibiotic has a specific mechanism that targets a particular cellular function in bacteria. Antibiotics are often grouped into classes according to their mechanism of action (Werth 2020). Some of the major classes are listed below:

- Targeting bacterial cell wall or membrane synthesis: A common example of this class of antibiotics is amoxicillin, which inhibits cell wall formation. Another example includes polymyxin B, which disrupts the cell membrane. Improper cell wall or membrane formation leads to bacterial cell death.
- Targeting protein synthesis: Antibiotics in this class can target different steps during transcription or translation, though blocking translation is more common. For example, tetracycline prevents the attachment of tRNAs to the ribosome during translation and thus prevents protein expression in bacteria.
- Targeting other essential enzymes: Antibiotics in this class can target different enzymes that are critical for proper bacteria function and survival. A common example is ciprofloxacin, which targets an enzyme involved in unwinding DNA for replication. This prevents bacteria from dividing and growing.

Resistance to antibiotics

Antibiotics have been used widely in both clinical and agricultural settings. This widespread use has placed a strong and ongoing selective pressure on many bacteria to survive in the presence of antibiotics. As bacteria that exhibit resistance are able to survive and reproduce in the presence of an antibiotic, the genes that conferred resistance will become more common in bacterial populations.

Antibiotic resistance can be grouped into different classes of mechanisms (Munita and Arias 2016). Some common mechanisms and an example of each is listed below:

- Modifying/inactivating the antibiotic: Kanamycin is an antibiotic that functions by binding to the ribosome and disrupting translation. Kanamycin kinase enzyme provides resistance by phosphorylating kanamycin and disrupting the ability of the antibiotic to bind to the bacterial ribosome.
- Altering the target binding site: Tetracycline is another antibiotic that functions by binding to the ribosome and disrupting translation. Certain mutations to the 16S ribosomal RNA provide resistance to tetracycline by altering ribosomal structure enough to prevent tetracycline from binding.
- Altering/bypassing the targeted pathway: Certain sulfonamide antibiotics inhibit enzymes involved in the synthesis of folic acid, which is required to synthesize nucleotides and amino acids. Some bacteria can develop certain mutations that lead to the overproduction of these targeted enzymes, essentially drowning out the ability of sulfonamide from targeting the folic acid synthesis pathway.
- Pumping the antibiotic out: Efflux pumps are able to pump specific antibiotics out of the cell faster than they can enter and prevents the antibiotics from targeting cellular function in bacteria. Efflux pumps are a common mechanism of antibiotic resistance to many different types of antibiotic drugs.

Antibiotic resistance is passed through genetic mechanisms. There are two primary ways that bacteria can acquire these genetic changes that lead to antibiotic resistance:

- *Mutation*: Mutations to genes in the bacterial genome can change the existing bacterial proteins expressed from the mutated genes. These changes could lead to antibiotic resistance by disrupting antibiotic function. For example, as mentioned above, mutations to the bacterial ribosome can alter its structure enough to prevent tetracycline from binding and disrupting translation.
- *Horizontal gene transfer*: Bacteria regularly incorporate genetic elements from the environment or through exchange with other bacteria, usually in the forms of transposable elements or plasmids. Horizontal gene transfer will typically result in the introduction of a new antibiotic resistance gene to the cell; for example, a gene that codes for an efflux pump or an enzyme that inhibits a specific antibiotic.

Once bacteria have either acquired resistance gene through either of these mechanisms, these genetic elements can be passed vertically through cell division or other means.

Today's lab

In this lab, you will use the BioBits® cell-free protein expression system to test the function of two different antibiotics: kanamycin and streptomycin. You will also investigate how an antibiotic resistance gene is able to protect cellular function in the presence of an antibiotic.

The BioBits® cell-free system contains cellular components derived from *E. coli* bacteria, freeze dried into a pellet. By adding water and a plasmid containing a gene with the correct promoter, the BioBits® cell-free system will begin expressing protein. Antibiotics that target protein expression in bacteria will therefore also interrupt protein expression in the BioBits® system.

NOTE: For more information on the BioBits® protein synthesis system and transcription/translation in general, please see our video tutorial here: <https://www.youtube.com/watch?v=FCwJyRvpDOU>.

Testing antibiotic function:

In this activity, you will use BioBits® reactions to test whether translation has been interrupted by an antibiotic. To do this, you will add a plasmid (named *RFP DNA*) that encodes a *red fluorescent protein (RFP)* to the BioBits® pellets. The production of RFP, visible as red fluorescence under blue light, will serve as a visual reporter that translation has occurred. In some reactions you will also add an antibiotic, either *kanamycin* or *streptomycin*. Both kanamycin and streptomycin function by binding to the ribosome and inhibiting protein synthesis. If the ribosomes are inhibited by the antibiotics, then translation of the RFP cannot take place and no red fluorescence will be seen.

Testing antibiotic resistance:

Antibiotic resistance is passed genetically and in one of your reactions you will also add a plasmid (named *Kinase DNA*) with an *antibiotic resistance gene* to the reaction. This particular resistance gene codes for

a kinase enzyme that will chemically modify one of the antibiotics and inactivate it. If the antibiotic is inactivated, then it cannot block the ribosomes, meaning the translation of the RFP can take place and red fluorescence will be seen. You will use your results and other lab groups' results to determine the target antibiotic to which this gene confers resistance.

Before you start the experiment, you will predict what you will see in the following reactions. Use the information from the background to fill out the table below:

| | Tube 1 | Tube 2 | Tube 3 | Tube 4 |
|---|----------------|----------------|--|---|
| Reaction conditions | Water | RFP DNA | RFP DNA + kanamycin OR streptomycin | RFP DNA + kanamycin OR streptomycin + Kinase DNA |
| Red fluorescent protein expression expected? | Yes/No/Unknown | Yes/No/Unknown | Yes/No/Unknown | Yes/No/Unknown |
| Justify your prediction | | | | |

Methods and Data Collection

You will use the BioBits® cell-free system to express red fluorescent protein. You will also use two different antibiotics to disrupt regular protein expression. Finally, you will use an antibiotic resistance gene to test how resistance mechanisms are able to rescue protein expression. You will perform a total of four reactions, including a negative control and a positive control.

Part A: Selecting the antibiotic for your group

Your instructor will assign your group an antibiotic to test. Circle the antibiotic you have been assigned:

Kanamycin

Streptomycin

Part B: Setting up BioBits® reactions

- Label and number each tube in your strip of four BioBits® pellets, 1-4
- Add 4 µl of either water or Kinase DNA sample to each BioBits® pellet in the strip according to the table below.

| | Tube 1 | Tube 2 | Tube 3 | Tube 4 |
|----------------|---------------|---------------|---------------|-----------------|
| Reagent | 4 µl water | 4 µl water | 4 µl water | 4 µl Kinase DNA |

- Place tubes at 37°C for 10 minutes
 - If you don't have a 37°C heat source, you can use body heat (i.e., your hands, under the arm, in your pocket) to warm the tubes (try to use a very warm body heat – the closer the incubation temperature is to 37°C, the better your results will be).

4. After 10 minutes, remove tubes from heat source, uncap the tubes, and add additional reagents according to the chart below
- Use the antibiotic assigned by your instructor (the one circled above in part A).

| | Tube 1 | Tube 2 | Tube 3 | Tube 4 |
|-------------------|-----------------|--------------------------------------|---|---|
| Reagent(s) | 5 μ l water | 3 μ l RFP DNA 2 μ l water | 3 μ l RFP DNA 2 μ l antibiotic | 3 μ l RFP DNA 2 μ l antibiotic |

5. Store tubes at room temperature
- Leave the reactions to proceed overnight at room temperature.
 - Tubes may be left in a rack or simply lying flat on a lab bench or table.
 - Avoid storing your tubes under direct sunlight (ambient indoor light is fine).

Part C: Observing your final results

Final observations are best taken between 8 and 72 hours later, although results tend to be optimal at approximately 24 hours. After 24 hours of room temperature incubation, results may be stored in the refrigerator for at least a week.

- Observe your tubes under blue light in the P51™ viewer or other 400 nm blue light illuminator
- Record your observations in the table below

Data Analysis

For your observations of each tube, circle “Yes” or “No” if the red fluorescent protein has been expressed. If there are other observations you notice (i.e., how bright or dim the red fluorescence is compared to other tubes), note that under “Other observations”.

Antibiotic used: _____

| | Tube 1 | Tube 2 | Tube 3 | Tube 4 |
|--|---------------|---------------|-----------------------|-------------------------------------|
| Reaction conditions | No reagents | RFP DNA | RFP DNA Antibiotic | RFP DNA Antibiotic Kinase DNA |
| Red fluorescent protein expression? | Yes / No | Yes / No | Yes / No | Yes / No |
| Other observations? | | | | |

Compare your results with another lab group that used a different antibiotic than you. Fill out the table again for the other antibiotic.

Antibiotic used: _____

| | Tube 1 | Tube 2 | Tube 3 | Tube 4 |
|----------------------------|---------------|---------------|-----------------------|-------------------------------------|
| Reaction conditions | No reagents | RFP DNA | RFP DNA Antibiotic | RFP DNA Antibiotic Kinase DNA |

| | | | | |
|--|----------|----------|----------|----------|
| Red fluorescent protein expression? | Yes / No | Yes / No | Yes / No | Yes / No |
| Other observations? | | | | |

Discussion Questions

1. In this experiment, tube 1 was a negative control. Why is having a negative control important for your experiment? If tube 1 was not included in this lab, what incorrect conclusions could a person make?
2. In this experiment, tube 2 was a positive control. Why is having a positive control important for your experiment? If tube 2 was not included in this lab, what incorrect conclusions could a person make?
3. The antibiotics used in this lab both interfere with ribosome function. Explain why such interference would inhibit bacterial growth.
4. Did the antibiotic that you added to your reaction inhibit protein expression? Cite your experimental data in your explanation.
5. Which antibiotic was inhibited by the antibiotic resistance gene? Cite your experimental data in your explanation.
6. How does your experimental evidence support the fact that antibiotic resistance genes confer resistance to specific antibiotics?
7. Previously, you were asked if protein expression had occurred by indicating whether or not you observed fluorescence. But you may also have observed differences in the brightness of fluorescence in different tubes. Did all the tubes in which you observed fluorescence have the same level of brightness? Based on your experimental set-up, can you think of an explanation for this difference?
8. Why did you think the plasmid with the antibiotic resistance gene (Kinase DNA) was added in advance with an additional incubation step before adding the antibiotics?
9. Which mechanism of acquiring antibiotic resistance did adding the plasmid with the antibiotic resistance gene (Kinase DNA) model most closely: random genetic mutation or horizontal gene transfer? Justify your answer.

Materials

Reagents supplied in kit:

| <u>Reagent</u> | <u>Amount needed per lab group</u> | <u>Storage</u> |
|---|------------------------------------|----------------|
| BioBits® in PCR strip tubes | 1 strip of four tubes | Freezer |
| Nuclease-free water | 25 µL | Freezer |
| RFP DNA samples for protein expression of RFP | 15 µL | Freezer |
| Kinase DNA samples for antibiotic resistance gene | 10µL | Freezer |
| Antibiotics <ul style="list-style-type: none"> • Kanamycin • Streptomycin | 10 µL of either | Freezer |

Equipment needed (supplied by teacher):

| <u>Supply</u> | <u>Amount needed per lab group</u> |
|---|---|
| 37°C heat source <ul style="list-style-type: none"> • e.g., miniPCR™ machine, incubator, water bath • Body heat works as well | 1 (can also be shared between groups) |
| Micropipettes and tips <ul style="list-style-type: none"> • 2-20 µL: one per lab group • 20-200 µL: one for the teacher to dispense reagents | 1 |
| P51™ molecular fluorescence viewer or other 400 nm blue light illuminator | 1 (can also be shared between groups) |

Other supplies:

- Disposable laboratory gloves
- Protective eyewear
- Permanent marker (ideally fine-tipped)
- Cup to dispose of tips

Notes for the Instructor

Expected results:

Listed below are the expected results for each reaction accompanied by a brief explanation. Note that exact timing of results may differ based on variability in incubation temperature/timing or other factors, but the overall trends should not change. You may also observe small differences in brightness across different groups which can arise from variability in micropipetting and sample handling.

| <u>Tube</u> | <u>Expected observations (kanamycin)</u> | <u>Expected observations (streptomycin)</u> |
|-------------|--|---|
| 1 | No fluorescence | No fluorescence |
| 2 | Red fluorescence | Red fluorescence |
| 3 | No fluorescence | No fluorescence |
| 4 | Red fluorescence | No fluorescence |

Tube 1 – Negative control (H₂O only):

Students added only water with no DNA or antibiotic to tube 1, so no fluorescence is expected. Students may detect low levels of autofluorescence, as the components in BioBits® pellets will show low levels of fluorescence on their own. This low level of autofluorescence can be used to demonstrate the importance of including negative controls in experiments.

Tube 2 – Positive control (RFP DNA):

Students added RFP DNA, which encodes a red fluorescent protein. In the BioBits® system, the RFP DNA is transcribed and translated, leading to red fluorescence protein. This red fluorescence will be observable under blue light within 8 hours, and can be used as a readout of successful protein expression.

Tube 3 – Test antibiotic function (RFP DNA + antibiotic):

Students added RFP DNA, along with either kanamycin or streptomycin. Both of these antibiotics bind to the ribosome and interfere with translation. Because translation is inhibited by kanamycin and streptomycin, in the presence of either antibiotic RFP is not produced and red fluorescence is not observed.

Tube 4 – Test antibiotic resistance (RFP DNA + antibiotic + antibiotic resistance gene):

Students first added Kinase DNA which encodes an antibiotic resistance gene expressing kanamycin kinase, an enzyme that chemically modifies kanamycin. The tubes were incubated for 10 minutes to give the cell-free system time to express the kanamycin kinase protein without antibiotic interference. Then, students added RFP DNA, along

with either kanamycin or streptomycin. Because kanamycin kinase inhibits kanamycin action, translation is expected to be rescued in the presence of kanamycin and students will observe red fluorescent protein. The red fluorescence likely will not be as bright as the red fluorescence in tube 2, as there will still be some protein synthesis inhibition by the antibiotic. For groups that are working with streptomycin, translation will not be rescued, RFP is not produced, and red fluorescence is not observed.

Tube 4 models horizontal gene transfer because a plasmid containing a resistance gene was added to the system to provide resistance to an antibiotic.

Fluorescent proteins will remain visible for at least 1 week at room temperature (longer if stored in the fridge).

Optional extensions:

The standard experimental design for this lab allows for students to investigate antibiotic function and resistance in a way that is highly accessible but also appropriate for college classes. Here we suggest a few ways that instructors may alter this approach:

- Individual lab groups may investigate both antibiotics.
- Students may research and hypothesize what would happen if other types of antibiotics are added. If these antibiotics are available on hand, they may be experimentally tested to confirm student hypotheses. For example:
 - Neomycin, streptomycin, and kanamycin are all aminoglycoside antibiotics. Kanamycin kinase, however, will rescue translation in the presence neomycin and kanamycin, but not streptomycin. Students can propose what structure of the antibiotic the kanamycin kinase must act upon based on structural similarities.
 - Carbenicillin is a cell wall biosynthesis inhibitor, but because the BioBits® reactions are made from cell extract (and therefore lack cell walls), carbenicillin will have no impact on the protein synthesis reaction.
- Further protocol modifications using additional antibiotics, antibiotic concentrations, and enzymes can be found in the publication describing the original

prototype of this activity (Stark, Huang, et al, 2019).

- Students may quantify their visual results using image analysis software. [A guide to do so](#) was included in our publication of an initial prototype of this lab (Stark, Huang, et al, 2019).
- Students may focus on the BioBits® cell-free protein synthesis aspect for this lab and discuss more generally how a synthetic biology system can be applied.

Cited References

- Stark, JC, Huang, A, et al. 2019. BioBits Health: Classroom Activities Exploring Engineering, Biology, and Human Health with Fluorescent Readouts. ACS Synth. Biol. 8(5):1001-1009.
- World Health Organization. Global tuberculosis report 2018 (2017 data).
- World Bank. Drug-resistant infections: A threat to our economic future. March 2017
- Werth, BJ, et al. 2020. Overview of Antibiotics. Merck Manuals Consumer Version. <https://www.merckmanuals.com/home/infections/antibiotics/overview-of-antibiotics>
- Munita, JM, Arias, CA. 2016. Mechanisms of Antibiotic Resistance. Microbiol Spectr. 4(2).

Acknowledgments

Thank you very much to all of the students and teachers at Waukegan High School, Round Lake Senior High School, Chicago Math and Science Academy, Mather High School, and Glenbard East High School who have helped pilot test this laboratory exercise (Stark et al. 2019), to the laboratories of Jim Collins at the Massachusetts Institute of Technology and Mike Jewett at Northwestern for supporting and funding the development of this laboratory exercise, to the curriculum team at miniPCR bio for translating this from prototype to product, and to ABLE for the opportunity to present and publish on this lab activity.

About the Authors

Ally Huang received her B.S in Biomedical Engineering from Johns Hopkins University and her Ph.D. in Biological Engineering from MIT, with a

thesis focused on making molecular and synthetic biology accessible to classrooms and other educational settings. She believes that everyone should have access to quality hands-on science education and is now at miniPCR bio, dedicated to the mission of making science accessible to everyone, everywhere.

Jessica Stark is currently an American Cancer Society Postdoctoral Fellow at Stanford University. She received her B.S. in Chemical and Biomolecular Engineering from Cornell University. As an NSF Graduate Research Fellow at Northwestern University, Jessica developed new, portable technologies for glycoprotein therapeutic and vaccine biomanufacturing. Her postdoctoral work focuses on identifying and targeting glyco-immune checkpoints for cancer immunotherapy. Jessica is committed to

enhancing diversity, equity, and inclusion in STEM through mentoring, outreach, and service activities.

Bruce Bryan, M.S., is the Director of Curriculum at miniPCR bio. Bruce studied evolutionary genetics at Brown University before becoming a high school teacher. He has taught all levels of biology including Advanced Placement. Bruce believes that science education should empower students to take part in authentic discovery and works to make miniPCR bio curriculum meet that standard.

Allison Nishitani earned her Ph.D. in neuroscience at Harvard, where she studied neurodevelopment. Allison is passionate about sharing her love of science with others and spent several years teaching high school biology before joining the miniPCR bio team.

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Citing This Article

Ally Huang and Jessica C. Stark 2022. BioBits® Antibiotic Resistance Lab: Visualizing how Antibiotics Work and Mechanisms of Antibiotic Resistance. Article 8 In: Boone E and Thuecks S, eds. *Advances in biology laboratory education*. Volume 42. Publication of the 42nd Conference of the Association for Biology Laboratory Education (ABLE). <https://doi.org/10.37590/able.v42.art8>

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