## Lesson Title: It's All Negative: A Case Study in Antibiotic Resistance.

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<b>Major Sections</b>	Content
Lesson Overview	<b>Overall Purpose:</b> This will be an interrupted case study introducing the CRE (Carbapenem Resistant <i>Enterobacterales</i> ), a group of Gram negatives resistant to the carbapenem drugs and often other antibiotics as well. Students will use information learned from multiple units in the same course to solve a case study and to determine the best treatment for a patient.
	<b>Estimated Timeframe:</b> 1.5-2 hours, or possibly 2 lab periods if they do the work themselves.
	Courses for Implementation: Bio275, possibly Bio250 or Bio175
	Format: (Seated, Online, Hybrid): Starting seated, but want to eventually move to online as well
	<b>Key Terms:</b> (Quickly identify the major topics addressed in the lesson.) antimicrobials, antimicrobial resistance, bacterial cell structure, Antibiotic susceptibility testing (Kirby-Bauer), differential and selective media, Gram stain, Bacterial physiology, infectious disease, CRE, lab result analysis, biochemical tests
	<ul> <li>Standards/Skills Addressed:</li> <li>Academic: Bacterial physiology, Gram stain, antibiotic resistance, Kirby-Bauer (disc diffusion), infectious disease, CRE, lab result analysis, differential and selective media, biochemical tests</li> <li>Technical: Microsoft Word or Powerpoint to present information, possible use of bioinformatic programs such as BLAST (sequence analysis).</li> <li>21st Century/Employability: Applied academic skills, critical thinking skills, interpersonal skills, Communication (written and/or verbal)</li> <li>Industry (if applicable): N/A</li> </ul>
	<ul> <li>Learner Outcomes/Student Learning Objectives: <ul> <li>(Learners will be able to)</li> </ul> </li> <li>After completing this assignment, students will be able to <ul> <li>Interpret laboratory findings to diagnose a patient</li> <li>Interpret results from the Gram stain to determine appropriate differential and selective media to use</li> <li>Interpret results from MacConkey agar</li> <li>Determine cause of disease utilizing laboratory test results</li> <li>Use Kirby-Bauer results (zone of inhibition size and interpretation tables) to determine appropriate antibiotic</li> <li>Discuss the significance of the CRE (<i>carbapenem resistant Enterobacterales</i>) in healthcare.</li> </ul> </li> </ul>

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Equipment/Materials	List of Materials/Equipment/Texts:
	If done in lecture: handouts for the different parts of the case study, including images and tables with lab results
	Pencil/pen
	Microbiology textbook (online or not) which contains a section on antibiotic resistance
	Phone or computer with internet access.
	If done in lab you would also need: Gram stain supplies (crystal violet, Gram iodine, 95% ethanol, safranin) Microscope slides Compound microscope Immersion oil for microscope MacConkey plates and MRVP broth if desired Nutrient agar or Tryptic Soy Agar; nutrient broth or TSB Metal or plastic disposable loops Bunsen burner (if no sterile disposable loop) Biohazard bags
	<ul> <li>Autoclave</li> <li>Safety Precautions: If done in lecture, no real safety precautions as assignment will utilize images and tables for interpretation.</li> <li>If used in a lab setting, BSL I lab precautions (safety glasses, lab coats, gloves, appropriate disinfectants for cleaning lab benches, soap at sinks, autoclave).</li> <li>Cleanup Instructions: If done in lecture, no specific clean up instructions. If done in lab, microscope slides would be disinfected and thrown out. Agar plates with bacteria and any disposable loops which were used on bacteria would be autoclaved before</li> </ul>
	being thrown out. Lab benches would be disinfected before and after using.

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Discussion	<ul> <li>Industry/Real-world Scenario:         Antibiotics are drugs used to treat bacterial infections. The use of antibiotics has had a huge impact on human health in terms of infectious disease. Unfortunately, their overuse has led to the selection of bacteria that are resistant to the antibiotics that once controlled them. Antibiotic resistance is a major public health threat. The CDC states that in 2019, there were more than 2.8 million antibiotic resistant infections in the US, resulting in as many as 35,000 deaths or more. The problem continues to grow as antibiotics are overused in both human and animal settings, and as research into new antibiotics lags behind the emergence of antibiotic resistance. The CRE (Carbapenem Resistant <i>Enterobacterales</i>) are a group of antibiotic Gram-negative bacteria that are resistant to many drugs, including the carbapenem class of antibiotics. Once very rare, these bacteria have been identified in 42 states.     </li> <li>Sarah has not been feeling well and has made an appointment with her primary care physician. For the past two days, Sarah has been experiencing frequent urination and some pain and burning while urinating. Your group will help determine a diagnosis and a course of treatment for Sarah.</li> <li>Integrated Content - Possible Knowledge/Skills Overlap:         Interpersonal skills (group work), verbal communication, written communication, critical thinking skills, lab test skills, lab skills (if done in lab)     </li> </ul>
Instructional Strategies	<ul> <li>Proposed Teaching Strategies: Interrupted case study, presenting scenarios one at a time and giving students a chance to discuss and come up with answers. Students will analyze lab results to help diagnose an infection and come up with a treatment plan.</li> <li>Bloom's: This assignment falls into the apply, analyze, and evaluate categories in Bloom's. Students will apply information they have learned to answer questions; they will analyze data to make decisions; and provide evidence for their answers.</li> <li>REACT: Puts students in real life setting of diagnosing a disease using lab skills and information they have learned in lecture and lab. Students apply learned skills to a new situation. And this could be expanded by having a nursing instructor or other person come into the classroom.</li> </ul>
Activities/Lesson Procedure	Activity Preparation: Instructor: Handouts, images of results available as needed, If done in lab: Preparation of nutrient agar, nutrient broth, MacConkey agar, methyl red broth, culture of E. coli streaked the day before.

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	Learner: Students will have learned the lab skills required for this exercise. This will be done while learning about antibiotics and as a prelude into antibiotic resistance. Students should go back over the Gram stain and differential/selective media.
	<ul> <li>Activity Steps/Lesson Procedure: <ol> <li>Introduce case study to students: how it will run, what is expected of them</li> <li>This is an interrupted case study with five parts that build on each other. Each part will be handed out and student groups will have time to discuss and decide on a course of action.</li> <li>As students work their way through the case study, results will be handed to them for interpretation to determine next steps.</li> <li>Once all groups have completed the case study, there will be a class discussion on antibiotic resistance and the CRE. The discussion will include snippets of the documentary 'Hunting the Nightmare Bacteria'.</li> </ol> </li> </ul>
	<b>Expected Results/Learner Products:</b> Students will fill out the questions on each of the 5 parts of the case study with their group. They will explain correctly why they chose each test to help identify the bacterium. They will provide accurate assessments for each of the tests done. They will provide and explain all evidence used to identify the microbe. The worksheet for the case study could be turned in at the end.
	Students would also be responsible for participating in the class discussion on the case study. If there is enough time, student groups could be responsible for presenting their findings.
	<b>Extension Options:</b> As stated before, this lesson could be used in lab, with students doing the actual procedures. One could add to the complexity by expanding on the biochemical tests used to identify the bacterium. An enterotube product could be used, but these are expensive. An instructor could even have fake urine that they inoculate with the bacteria so students start from the beginning. There is a Carolina Biological kit (Enteric Biochemical Detective Kit, item 154717) that might be a lot of fun to use in conjunction with this assignment.
	Another extension that would make it appropriate for Genetics and Microbiology would be to use a molecular approach to identification, in addition to or instead of the biochemical tests. One could provide DNA sequences and use Bioinformatics to determine the cause.
	This could be used in an entire unit on antibiotic resistance. PCR could be used to identify the antibiotic resistance genes present in this microbe, and students could explore the genetic link to resistance. There is a MiniPCR kit that has a case study where students look for the NDM-1 antibiotic resistance gene in soil samples from farms. They also have a kit where students can actually look for antibiotic resistance genes (for tetracycline) in soil samples of their choice.

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Faculty Resources	<b>Background Material:</b> Instructors will need to understand how bacteria can be identified using different types of staining procedure, media or biochemical tests (Gram stain, MacConkey or EMB media, methyl red test). Instructors will also need to know about antibiotics (what they are and how they inhibit/kill bacteria) and the basis of antibiotic resistance. A microbiology textbook and lab manual will be very important here for background information. There are also very nice videos available in Youtube that cover these topics, as well as the lab skills required to do the procedures.
	Differential-Selective-Bacterial-Growth-Media-Microbiology-Lecture-Powerpoint-VMC
	Online text (Libre text) with information on the media used
	Video on MSA
	Video on MacConkey
	Video on Methyl red test (if you want to use it in assignment)
	Link to CDC information on the Carbapenem Resistant Enterobacterales
	Kirby-Bauer-Disk-Diffusion-Susceptibility-Test-Protocol(1).docx
	Video on reading Kirby bauer plates
	Kirby Bauer virtual lab
	Handouts and Supplemental Materials: <u>Table of bacteria</u> for unknown in practical 3 : This is a table they would have filled out in lab. They would use it to help narrow down their answer. You could expand this table and add more bacteria and tests.
	Case Study for Antibiotic Resistance in UTI patient (PDF): This is the actual case study.
	<b>Results for UTI Case study.pptx (PPT/PDF)</b> : This powerpoint/PDF contains images to pass out as students work through the case study.
	Answer Keys: Partial answer key with the included powerpoint/PDF.

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Assessment	How will students demonstrate what they have learned? Students will turn in the 5 different parts of the interrupted case study. Once I see how this goes, I could consider having them turn in a written report or having students present parts of the case study to the class. We will discuss the case study as a group once it is completed.
	Learner Products/Assessment Tools or Processes: I intend to use this as a participation grade, at least initially.