Medical Microbiology Laboratory Case Studies

Resource Type: Curriculum: Laboratory

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Abstract

The purpose of this laboratory exercise is to give students the opportunity to use the basic knowledge and techniques (Gram stain, biochemical reactions, etc.) gained in a medical or allied health microbiology laboratory in a series of practical, "hands-on" simulated patient scenarios. Three case studies will test students' knowledge of the pathogens of the gastrointestinal, respiratory, and urogenital tracts.

Activity

Invitation for User Feedback. If you have used the activity and would like to provide feedback, please send an e-mail to MicrobeLibrary@asmusa.org. Feedback can include ideas which complement the activity and new approaches for implementing the activity. Your comments will be added to the activity under a separate section labeled "Feedback." Comments may be edited.

INTRODUCTION

Time Required.
When the exercise is used exactly as described in the procedure, students should be able to complete it in a single 2-hour laboratory period. However, if the instructor wishes to develop the exercise into an "unknown identification" lab as described in the possible modifications section, then two 2-hour laboratory periods are required.

Background.
This exercise is designed for use as an end-of-semester activity. Prior to participating in this exercise, students should have experience with basic aseptic technique, Gram staining, and antimicrobial susceptibility testing (Kirby-Bauer disk diffusion method). In addition, students should have an understanding of the pathogens of the gastrointestinal, respiratory, and urogenital tracts. Finally, students should have laboratory experience with the biochemistry of microbial metabolism and the various biochemical tests used to identify pathogenic microorganisms.

PROCEDURE

Materials.
Case Study 1

Case study 1 - "Spring Weekend" handout

Media for case study 1 (all should be inoculated with a pure culture of *Salmonella typhimurium*):

- Sulfide, Indole, Motility [SIM] (1)
- Methyl Red Voges Proskauer [MR-VP] (2)
- Simmons citrate plate (1)
- Urea broth (1)
- Phenol red carbohydrate broths with Durham tubes - lactose, sucrose, and dextrose (1 each)

Gram stain of *S. typhimurium*

Instructor's answer key - "Spring Weekend" answer key

Case Study 2

Case study 2 - "The Death of Kermit the Frog" handout

Media for case study 2 (all should be inoculated with a pure culture of *Streptococcus pyogenes*)

- Blood agar plates showing beta-hemolysis and susceptibility to bacitracin (1 each)
- Mueller-Hinton "giant" plate for antimicrobial susceptibility testing (1)

Antibiotic disks (12 different: i.e., ampicillin, bacitracin, chloramphenicol, erythromycin, kanamycin, nitrofurantoin, penicillin, streptomycin, sulfisoxazole, tetracycline, tobramycin, and vancomycin)

Hydrogen peroxide (for slide catalase test)

Gram stain of *S. pyogenes*

Instructor's answer key "The Death of Kermit the Frog" answer key

Case Study 3

Case study 3 - "The Seven-Year Itch" handout

Media for case study 3 (all should be inoculated with a pure culture of *Escherichia coli*)

- Sulfide, Indole, Motility [SIM] (1)
- Methyl Red Voges Proskauer [MR-VP] (2)
- Simmons citrate plate (1)
- Urea broth (1)
- Phenol red carbohydrate broths with Durham tubes - lactose, sucrose, and dextrose (1 each)
- Mueller-Hinton "giant" plate for antimicrobial susceptibility testing (1)

Antibiotic disks used in previous case study

Gram stain of *E. coli*

Reagents for indole, methyl red, and Voges-Proskauer tests (2 sets)

Inoculating loops and needles, sterile swabs, glass slides, and other standard microbiology equipment

Instructor's answer key - "The Seven-Year Itch" answer key

Instructor Version.
The instructor or other assigned laboratory technician should prepare all of the demonstrations 24 to 48 hours prior to the scheduled laboratory period. On the day of the exercise, groups of 3 to 5 students (depending on class size) should each be assigned one of the case studies. The students will work as a team to interpret the data presented and answer all questions on the handout. Each group should be given approximately 30 to 45 minutes to complete the exercise, at which point the instructor may choose to have the groups rotate to a different case study or have each group present their "findings" to the class in a brief informal presentation.

Safety Issues.
Prior to participating in this laboratory, students should have a clear understanding of aseptic technique and the general safety precautions of a microbiology lab.

ASSESSMENT and OUTCOMES

Suggestions for Assessment.
During this exercise, students should be encouraged to rely on each other's critical thinking skills and mastery of the various basic microbiological techniques and concepts, rather than on their notes or other reference materials. Group discussion incorporates the concept of peer oral assessment into the activity. By limiting the group size to 3 to 5 students per case study, students are more likely to equally contribute to the exercise.

This exercise will work well as both a standard laboratory, in which point values can be assigned to each question, or as an extra-credit exercise to stimulate thinking and discussion the week before a final lab exam or practical. In addition, concepts from the exercise can be used on subsequent lecture or lab exams.

Field Testing.
Since this laboratory exercise was written relatively recently (April 2001), I have only used it once as an extra-credit exercise in a microbiology lab geared towards allied health majors. However, in an informal poll of my students, all of them indicated that this exercise was an enjoyable, interesting, and challenging way of learning microbiology and wished that this style of teaching could have been incorporated more often during the course of the semester. Further field testing and feedback will be greatly appreciated.

SUPPLEMENTARY MATERIALS

Possible Modifications.
Instructors who wish to develop this lab into a two week "unknown identification" can easily do so by giving each group of students a simulated clinical specimen (pure TSA slant or plate culture of the particular bacterium) on the first planned week of the exercise and having the students perform the Gram stain, antimicrobial susceptibility testing, and inoculation of the various media types. The second week can be used to carry out the exercise as detailed above.

In addition, although each exercise was designed to implicate a particular bacterium as the cause of each disease (C.S. 1 - *S. typhimurium*; C.S. 2 - *S. pyogenes*; and C.S. 3 - *E. coli*), with slight modifications to the "data" and available cultures, a number of gastrointestinal, respiratory, and urogenital infections can be demonstrated.

References.

1. **Bergey, D., et al.** 1993. Bergey's manual of determinative bacteriology, 9th ed. Williams & Wilkins, Baltimore, Md.
2. **Difco Laboratories.** 1998. Difco manual: dehydrated culture media and reagents for microbiology, 11th ed. Difco Laboratories, Division of Becton Dickinson & Co., Sparks, Md.
3. **Hart, T., and P. Shears.** 1996. Color atlas of medical microbiology. Times Mirror International Publishers Limited, London, England.
4. **Howard, B. J., et al.** 1994. Clinical and pathogenic microbiology, 2nd ed. The C.V. Mosby Co., St. Louis, Mo.

5. **Koneman, E. W., et al.** 1997. Color atlas and textbook of diagnostic microbiology, 5th ed. Lippincott-Raven Publishers, Philadelphia, Pa.
6. **Prescott, L. M., et al.** 1999. Microbiology, 4th ed. The McGraw-Hill Companies, Inc., Boston, Mass.

Appendices.

Bacterial cultures.
All bacterial cultures used in this exercise were obtained from Presque Isle Cultures, 3804 West Lake Rd., P.O. Box 8191, Erie, PA 16505. Phone: (814) 833-6262.

Media and reagents.
All dehydrated media, reagents, and antibiotic disks were obtained from BD Diagnostic Systems, 7 Loveton Circle, Sparks, MD 21152. Phone: (800) 638-8663.

Recipes for Media and Reagents.
All recipes for preparing the various types of media and reagents used in this exercise can be found in the Difco Manual.

Curriculum Resources

Case Study 1: Spring Weekend

Background Information:
Five hundred students attended the local college's Spring Weekend, an annual beach party the weekend before final exams. Approximately 25 of the students in attendance had recently returned from a Campus Ministry trip to a small village in Ecuador, where there had been rampant cases of inflammatory gastroenteritis caused by *Shigella* species in recent years. During their two-week trip, the students stayed in the villagers' homes, helped to build additional housing, and taught English to the students in the local schoolhouse.

In addition to the abundance of alcoholic beverages available at the party, there were a number of different types of food served that had either been bought or prepared by the students renting the beach houses. Two days after the party, 100 people developed severe diarrhea and the diagnosis of inflammatory gastroenteritis was made. Five of those affected were members of the Campus Ministry group while the other 95 were simply students who had attended Spring Weekend.

As in any outbreak of food-borne disease, the local health department first needed to determine the source of the bacteria. In this case, the possibilities were: 1) the bacteria came from the food, 2) the bacteria came from an infected student via the food, or 3) the bacteria were transmitted by direct contact with a member of the Campus Ministry group.

One way to determine the source of the bacteria is to question everyone about what food they ate during the 72 hours prior to the symptoms appearing. All guests were queried as to the food eaten while at Spring Weekend. The results appear in the tables below.

Survey Results:

TABLE 1. Food eaten by guests who became ill

Food	Eaten	
	Yes	No
Hot dogs	45	55
Buffalo wings	100	0
Hamburgers	64	36
Soft-shell clams	30	70
Potato salad	52	48

TABLE 2. Food eaten by guests who remained healthy

Food	Eaten	
	Yes	No

Hot dogs	320	80
Buffalo wings	110	290
Hamburgers	265	135
Soft-shell clams	150	250
Potato salad	292	108

Question 1:
Based on this data, which food do you think was responsible for this outbreak of food-borne disease? Why?

Microbiology Lab Results:
When the local lab examined the suspected food, bacteria were isolated, Gram stained, and subjected to various biochemical tests for identification. Fill in the chart below based on your observations.

TABLE 3. Microbiology lab results

Microscopic morphology	IND	MR	VP	CIT	H_2S	URE	MOT	GAS	LAC	SUC	GLU

Based on the diagnosis of inflammatory gastroenteritis, several bacteria may be the culprit. The lab is able to narrow down the choices to the following six bacteria:

TABLE 4. Bacteria which cause inflammatory gastroenteritis

Bacteria	IND	MR	VP	CIT	H_2S	URE	MOT	GAS	LAC	SUC	GLU
Campylobacter jejuni	-	-	-	-	-	-	+	-	-	-	-
Escherichia coli	+	+	-	-	-	-	+	+	+	±	+
Salmonella typhimurium	-	+	-	+	+	-	+	+	-	-	+
Shigella sonnei	-	+	-	-	-	-	-	-	±	-	+
Vibrio parahaemolyticus	+	+	-	+	+	±	+	-	-	-	+
Yersinia enterocolitica	±	+	-	-	-	±	-	-	-	+	+

Question 2:
Based on your observations, which of the above bacteria is the most likely cause of the students' inflammatory gastroenteritis?

Question 3:

Why do you think that some people ate the infected food and did not get sick?

Question 4:
To a microbiologist, a quick look at the Spring Weekend menu reveals a number of items that, if not handled properly, could pose potential threats for food-borne diseases. Can you pick out the possibly hazardous foods and name the bacteria sometimes associated with them?

Curriculum Resources

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As in any outbreak of food-borne disease, the local health department first needed to determine the source of the bacteria. In this case, the possibilities were: 1) the bacteria came from the food, 2) the bacteria came from an infected student via the food, or 3) the bacteria were transmitted by direct contact with a member of the Campus Ministry group.

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Hot dogs	45	55
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Soft-shell clams	30	70
Potato salad	52	48

TABLE 2. Food eaten by guests who remained healthy

Food	Eaten

	Yes	No
Hot dogs	320	80
Buffalo wings	110	290
Hamburgers	265	135
Soft-shell clams	150	250
Potato salad	292	108

Question 1:
Based on this data, which food do you think was responsible for this outbreak of food-borne disease? Why?

-Buffalo wings; because everyone who developed inflammatory gastroenteritis ate this particular food.

Microbiology Lab Results:
When the local lab examined the suspected food, bacteria were isolated, Gram stained, and subjected to various biochemical tests for identification. Fill in the chart below based on your observations.

TABLE 3. Microbiology lab results

Microscopic morphology	IND	MR	VP	CIT	H_2S	URE	MOT	GAS	LAC	SUC	GLU
Gram-negative rod, 3.0 x 0.5µm, random arrangement	-	+	-	+	+	-	+	+	-	-	+

Based on the diagnosis of inflammatory gastroenteritis, several bacteria may be the culprit. The lab is able to narrow down the choices to the following six bacteria:

TABLE 4. Bacteria which cause inflammatory gastroenteritis

Bacteria	IND	MR	VP	CIT	H_2S	URE	MOT	GAS	LAC	SUC	GLU
Campylobacter jejuni	-	-	-	-	-	-	+	-	-	-	-
Escherichia coli	+	+	-	-	-	-	+	+	+	±	+
Salmonella typhimurium	-	+	-	+	+	-	+	+	-	-	+
Shigella sonnei	-	+	-	-	-	-	-	-	±	-	+
Vibrio parahaemolyticus	+	+	-	+	+	±	+	-	-	-	+

Yersinia enterocolitica	±	+	-	-	-	±	-	-	-	+	+

Question 2:
Based on your observations, which of the above bacteria is the most likely cause of the students' inflammatory gastroenteritis?

-*Salmonella typhimurium*

Question 3:
Why do you think that some people ate the infected food and did not get sick?

-Individuals who ate the infected food and did not get sick may simply have consumed less or, less likely, may have had better immunity to the infectious agent.

Question 4:
To a microbiologist, a quick look at the Spring Weekend menu reveals a number of items that, if not handled properly, could pose potential threats for food-borne diseases. Can you pick out the possibly hazardous foods and name the bacteria sometimes associated with them?

- *Hot dogs – Listeria*
- *Buffalo wings – Salmonella*
- *Hamburgers – E. coli*
- *Soft-shell clams – Vibrio*
- *Potato salad – S. aureus, Salmonella, etc.*

Curriculum Resources

Case Study 2: The Death of Kermit the Frog

Background Information:

Jim Henson, the Muppets creator, was taken to the emergency room of New York Hospital late Monday night. As the attending nurse on duty, you assisted the doctor in taking the patient's history and vital signs. During your interview with Mr. Henson's daughter, she told you that over the past weekend her father had seemed to be fighting a cold, and today he missed a day of work, something he rarely did, saying that he felt tired. Finally, around six o'clock in the evening, he began coughing up blood. Mr. Henson also displayed a sudden onset of symptoms which included shaking chills with a high fever (>104.9°F), chest pain, respiratory distress, and a bluish tint to the lips and nailbeds. The attending physician immediately ordered a chest x-ray and asked you to take samples of Mr. Henson's sputum, nasopharyngeal secretions, and blood. After taking Mr. Henson's samples and escorting him back to his room from radiology, you noticed that he was no longer as responsive as he had been earlier. A few minutes after you got him back into bed, he lost consciousness, went into multiple organ failure, and slipped into a coma. A mere 24 hours after being admitted to the hospital, Mr. Henson, a generally healthy 53-year-old man with no history of immunodeficiency or somatic disease, was dead.

A few hours after Mr. Henson's death, the first results began to come back from the radiology department and the microbiology lab.

Radiology Department Results:

Since you are not an expert in radiology, you ask for a consult when reading Mr. Henson's chest x-ray. You are informed by the radiologist that the patient's x-ray shows disseminated interstitial infiltration (bronchopneumonia) with large pleural effusion (accumulation of fluid in the lungs).

Question 1a:
What illness do the radiology results indicate Mr. Henson was battling?

Question 1b:
Is it common for this condition to cause such a rapid demise in a person with a competent immune system?

Microbiology Laboratory Results:
When the lab cultured Mr. Henson's sputum, nasopharyngeal secretions, and blood, bacteria were isolated from all three samples.

Question 2:
Is it normal for bacteria to be isolated from all three of these bodily fluids? If not, then what does this indicate?

The isolated bacteria were cultured on blood agar, Gram stained, and subjected to a catalase test for identification. Fill in the table below based on your observations.

TABLE 1. Laboratory observations of isolated bacteria

Microscopic morphology	Colonial morphology

	Form	Elevation	Margin	Color	Surface	Density	Consistency

Catalase reaction	Type of hemolysis	Reaction to bacitracin

Question 3:
Based on your observations, which bacteria was responsible for Mr. Henson's death? (Presumptive identification.)

While not commonly known as a cause of bacterial pneumonia, this organism is classified as one of the new "superbugs," virulent strains of bacteria that can cause massive, rapid, and disseminated infection. If Mr. Henson's infection had been diagnosed earlier, it may have been treatable with antibiotics. The lab performed antimicrobial susceptibility testing on the bacteria isolated from Mr. Henson's samples. Fill in the table below based on your observations.

TABLE 2. Results of antimicrobial susceptibility testing

Code	Antibiotic	Type of bacteria most effective against	Zone of inhibition size (mm)	Is bacteria susceptible, resistant, or intermediate
AM10	Ampicillin	G+		
B10	Bacitracin	G+ (topical use only)		
C30	Chloramphenicol	G+ & G-		
E15	Erythromycin	G+		
K30	Kanamycin	G-		
F/M300	Nitrofurantoin	G-		
P10	Penicillin	G+		
S10	Streptomycin	G-		
G.25	Sulfisoxazole	G+		
Te30	Tetracycline	G-		
NN10	Tobramycin	G-		
Va30	Vancomycin	G+		

Question 4:
Based on your observations, and keeping in mind the type of bacteria each antibiotic is effective against, which antibiotic(s) would probably have been most effective in treating Mr. Henson's infection?

Curriculum Resources

Case Study 2: The Death of Kermit the Frog (Answer Key)

Background Information:
Jim Henson, the Muppets creator, was taken to the emergency room of New York Hospital late Monday night. As the attending nurse on duty, you assisted the doctor in taking the patient's history and vital signs. During your interview with Mr. Henson's daughter, she told you that over the past weekend her father had seemed to be fighting a cold, and today he missed a day of work (something he rarely did), saying that he felt tired. Finally, around six o'clock in the evening, he began coughing up blood. Mr. Henson also displayed a sudden onset of symptoms which included shaking chills with a high fever (>104.9°F), chest pain, respiratory distress, and a bluish tint to the lips and nailbeds. The attending physician immediately ordered a chest x-ray and asked you to take samples of Mr. Henson's sputum, nasopharyngeal secretions, and blood. After taking Mr. Henson's samples and escorting him back to his room from radiology, you noticed that he was no longer as responsive as he had been earlier. A few minutes after you got him back into bed, he lost consciousness, went into multiple organ failure, and slipped into a coma. A mere 24 hours after being admitted to the hospital, Mr. Henson, a generally healthy 53-year-old man with no history of immunodeficiency or somatic disease, was dead.

A few hours after Mr. Henson's death, the first results began to come back from the radiology department and the microbiology lab.

Radiology Department Results:
Since you are not an expert in radiology, you ask for a consult when reading Mr. Henson's chest x-ray. You are informed by the radiologist that the patient's x-ray shows disseminated interstitial infiltration (bronchopneumonia) with large pleural effusion (accumulation of fluid in the lungs).

Question 1a:
What illness do the radiology results indicate Mr. Henson was battling?

- *Bacterial pneumonia*

Question 1b:
Is it common for this condition to cause such a rapid demise in a person with a competent immune system?

- *No, this condition does not usually cause such a rapid death in an individual with a competent immune system.*

Microbiology Laboratory Results:
When the lab cultured Mr. Henson's sputum, nasopharyngeal secretions, and blood, bacteria were isolated from all three samples.

Question 2:
Is it normal for bacteria to be isolated from all three of these bodily fluids? If not, then what does this indicate?

- *No, bacteria are not found in the blood of healthy individuals. Since bacteria were isolated from Mr. Henson's blood, this indicates that the patient was also suffering from sepsis.*

The isolated bacteria were cultured on blood agar, Gram stained, and subjected to a catalase test for identification. Fill in the table below based on your observations.

TABLE 1. Laboratory observations of isolated bacteria

Microscopic morphology	Colonial morphology						
	Form	Elevation	Margin	Color	Surface	Density	Consistency
Gram-positive cocci, 0.5μm, in chains	Punctiform	Convex	Entire	Clear	Smooth	Translucent	Gummy

Catalase reaction	Type of hemolysis	Reaction to bacitracin
Negative	*Beta*	*Susceptible*

Question #3:
Based on your observations, which bacteria was responsible for Mr. Henson's death? (Presumptive identification.)

- *Streptococcus pyogenes*

While not commonly known as a cause of bacterial pneumonia, this organism is classified as one of the new "superbugs," virulent strains of bacteria that can cause massive, rapid, and disseminated infection. If Mr. Henson's infection had been diagnosed earlier, it may have been treatable with antibiotics. The lab performed antimicrobial susceptibility testing on the bacteria isolated from Mr. Henson's samples. Fill in the table below based on your observations.

TABLE 2. Results of antimicrobial susceptibility testing

Code	Antibiotic	Type of bacteria most effective against	Zone of inhibition size (mm)	Is bacteria susceptible, resistant, or intermediate
AM10	Ampicillin	G+		
B10	Bacitracin	G+ (topical use only)		
C30	Chloramphenicol	G+ & G-		
E15	Erythromycin	G+		
K30	Kanamycin	G-		
F/M300	Nitrofurantoin	G-		

P10	Penicillin	G+		
S10	Streptomycin	G-		
G.25	Sulfisoxazole	G+		
Te30	Tetracycline	G-		
NN10	Tobramycin	G-		
Va30	Vancomycin	G+		

Question #4:
Based on your observations, and keeping in mind the type of bacteria each antibiotic is effective against, which antibiotic(s) would probably have been most effective in treating Mr. Henson's infection?

- The results for Table 2 will depend on the particular culture used. When determining which antibiotics to use in treatment they must be: 1) effective against infections caused by gram-positive bacteria, and 2) drugs to which the clinical isolate is susceptible.

Curriculum Resources

Case Study 3: The Seven-Year Itch

Background Information:
Melissa is a 23-year-old woman who has been sexually active from the age of 16. In that span of time, Melissa has had about 40 partners. Melissa and her partners rarely, if ever, practiced safe sex. She was first diagnosed with herpes at 17. Then at 18, she received treatment for syphilis. Shortly thereafter, she contracted gonorrhea; however, she was asymptomatic and did not seek treatment for over a year. As a result, she developed pelvic inflammatory disease (PID), a condition that has left Melissa sterile. Finally about a year ago, Melissa tested HIV-positive and is currently on a strict "AIDS cocktail" regimen.

She has come to the Southside Clinic, where you are a nurse, presenting a number of symptoms including: purulent (pus-filled) discharge from the urethra, pain during urination, urge to urinate even when bladder is not full, pain in the lower back, fever, and chills. The clinic physician asks the patient to provide a "clean-catch" urine sample that is sent to the lab for a quantitative urine culture. Twenty-four hours later, the lab reports "significant bacteriuria" with greater than 100,000 colony-forming units (CFU) of bacteria present per milliliter of the patient's urine.

Question 1:
Based on this laboratory result, what do you conclude the patient is currently suffering from?

Microbiology Lab Results:
The lab also Gram stained the isolated bacteria and ran a number of biochemical tests to aid in identification. Fill in the table below based on your observations.

TABLE 1. Microbiology laboratory results

Microscopic morphology	IND	MR	VP	CIT	H_2S	URE	MOT	GAS	LAC	SUC	GLU

Based on your earlier diagnosis, several bacteria may be causing the patient's current condition. The lab is able to narrow down the choices to the following six bacteria:

TABLE 2. Bacteria which may be responsible for the patient's symptoms

Bacteria	IND	MR	VP	CIT	H_2S	URE	MOT	GAS	LAC	SUC	GLU
Enterobacter aerogenes	-	-	+	+	-	-	+	+	+	+	+
Escherichia coli	+	+	-	-	-	-	+	+	+	±	+
Klebsiella pneumoniae	-	-	+	+	-	+	-	+	+	+	+
Proteus mirabilis	-	+	±	±	+	+	+	+	-	±	+

Providencia stuartii	+	+	-	+	-	±	+	-	-	±	+
Serratia marcescens	-	-	+	+	-	-	+	±	-	+	+

Question 2:
Based upon your observations, which of the above bacteria is the most likely cause of the patient's current condition?

In order to determine the most effective antibiotic to treat the patient's infection, the lab performed antimicrobial susceptibility testing on the bacterium isolated from the patient's urine sample. Fill in the table below based on your observations.

TABLE 3. Results from antimicrobial susceptibility testing

Code	Antibiotic	Type of bacteria most effective against	Zone of inhibition size (mm)	Is bacteria susceptible, resistant, or intermediate
AM10	Ampicillin	G+		
B10	Bacitracin	G+ (topical use only)		
C30	Chloramphenicol	G+ & G-		
E15	Erythromycin	G+		
K30	Kanamycin	G-		
F/M300	Nitrofurantoin	G-		
P10	Penicillin	G+		
S10	Streptomycin	G-		
G.25	Sulfisoxazole	G+		
Te30	Tetracycline	G-		
NN10	Tobramycin	G-		
Va30	Vancomycin	G+		

Question 3:
Based on your observations, and keeping in mind the type of bacteria each antibiotic is effective against, which antibiotic(s) would be most effective in treating Melissa's infection?

Question 4:
In addition to opportunistic, gram-negative members of the fecal flora, this condition is also commonly caused by another type of bacteria in sexually active young women. Name that bacteria.

Question 5:

Is this disease directly due to the patient's sexual activity? Explain.

Curriculum Resources

Case Study 3: The Seven-Year Itch

Background Information:
Melissa is a 23-year-old woman who has been sexually active from the age of 16. In that span of time, Melissa has had about 40 partners. Melissa and her partners rarely, if ever, practiced safe sex. She was first diagnosed with herpes at 17. Then at 18, she received treatment for syphilis. Shortly thereafter, she contracted gonorrhea; however, she was asymptomatic and did not seek treatment for over a year. As a result, she developed pelvic inflammatory disease (PID), a condition that has left Melissa sterile. Finally about a year ago, Melissa tested HIV-positive and is currently on a strict "AIDS cocktail" regimen.

She has come to the Southside Clinic, where you are a nurse, presenting a number of symptoms including: purulent (pus-filled) discharge from the urethra, pain during urination, urge to urinate even when bladder is not full, pain in the lower back, fever, and chills. The clinic physician asks the patient to provide a "clean-catch" urine sample that is sent to the lab for a quantitative urine culture. Twenty-four hours later, the lab reports "significant bacteriuria" with greater than 100,000 colony-forming units (CFU) of bacteria present per milliliter of the patient's urine.

Question 1:
Based on this laboratory result, what do you conclude the patient is currently suffering from?

- *Urinary tract infection*

Microbiology Lab Results:
The lab also Gram stained the isolated bacteria and ran a number of biochemical tests to aid in identification. Fill in the table below based on your observations.

TABLE 1. Microbiology laboratory results

Microscopic morphology	IND	MR	VP	CIT	H_2S	URE	MOT	GAS	LAC	SUC	GLU
Gram-negative rod, 2.0 x 0.5µm, random arrangement	+	+	-	-	-	-	+	+	+	+	+

Based on your earlier diagnosis, several bacteria may be causing the patient's current condition. The lab is able to narrow down the choices to the following six bacteria:

TABLE 2. Bacteria which may be responsible for the patient's symptoms

Bacteria	IND	MR	VP	CIT	H_2S	URE	MOT	GAS	LAC	SUC	GLU
Enterobacter aerogenes	-	-	+	+	-	-	+	+	+	+	+
Escherichia coli	+	+	-	-	-	-	+	+	+	±	+

Klebsiella pneumoniae	-	-	+	+	-	+	-	+	+	+	+
Proteus mirabilis	-	+	±	±	+	+	+	+	-	±	+
Providencia stuartii	+	+	-	+	-	±	+	-	-	±	+
Serratia marcescens	-	-	+	+	-	-	+	±	-	+	+

Question 2:
Based upon your observations, which of the above bacteria is the most likely cause of the patient's current condition?

- *Escherichia coli*

In order to determine the most effective antibiotic to treat the patient's infection, the lab performed antimicrobial susceptibility testing on the bacterium isolated from the patient's urine sample. Fill in the table below based on your observations.

TABLE 3. Results from antimicrobial susceptibility testing

Code	Antibiotic	Type of bacteria most effective against	Zone of inhibition size (mm)	Is bacteria susceptible, resistant, or intermediate
AM10	Ampicillin	G+		
B10	Bacitracin	G+ (topical use only)		
C30	Chloramphenicol	G+ & G-		
E15	Erythromycin	G+		
K30	Kanamycin	G-		
F/M300	Nitrofurantoin	G-		
P10	Penicillin	G+		
S10	Streptomycin	G-		
G.25	Sulfisoxazole	G+		
Te30	Tetracycline	G-		
NN10	Tobramycin	G-		
Va30	Vancomycin	G+		

Question 3:

Based on your observations, and keeping in mind the type of bacteria each antibiotic is effective against, which antibiotic(s) would be most effective in treating Melissa's infection?

- *The results for Table 3 will depend on the particular culture used. When determining which antibiotics to use in treatment they must be: 1) effective against infections caused by gram-negative bacteria, and 2) drugs to which the clinical isolate is susceptible.*

Question 4:
In addition to opportunistic, gram-negative members of the fecal flora, this condition is also commonly caused by another type of bacteria in sexually active young women. Name that bacteria.

- *Staphylococcus saprophyticus*

Question 5:
Is this disease directly due to the patient's sexual activity? Explain.

-*This patient's urinary tract infection (UTI) is probably, but not necessarily, related to her sexual activity, although it is not actually a sexually transmitted disease like the infections she has received from her partners in the past. Females in general are more prone to UTI's than males because of the shortness of their ureter and the nearness of the urethral opening to the source of most UTI agents (i.e., fecal bacteria from the woman's own gastrointestinal tract). Females who are sexually active and/or engage in certain other vigorous physical activities suffer the transmission of these bacteria from the gut into the urinary tract more commonly than females who do not engage in these activities. Although the patient's positive HIV status puts her at greater risk for some life-threatening infections, it probably did not contribute to her UTI.*