

# El Paso Community College

## Syllabus

### Part II

## Official Course Description

<b>SUBJECT AREA</b>	<u>Medical Laboratory Technology</u>
<b>COURSE RUBRIC AND NUMBER</b>	<u>MLAB 2434</u>
<b>COURSE TITLE</b>	<u>Microbiology</u>
<b>COURSE CREDIT HOURS</b>	<u>4                    3    :    4</u> Credits            Lec    Lab

### I. Catalog Description

Instructs in the theory, practical application, and pathogenesis of clinical microbiology, including collection, quality control, quality assurance, lab safety, setup, identification, susceptibility testing, and reporting results. A grade of "C" or better is required in this course to take the next course. **Corequisite:** MLAB 2360. (3:4). Lab fee.

### II. Course Objectives

- A. Unit I. Introduction to Medical Microbiology
1. State general safety rules for handling pathogenic cultures and potentially infective clinical specimens, to include:
    - a. the importance of personal cleanliness, frequent handwashing, and not putting pens and pencils in the mouth.
    - b. the importance of wearing a lab coat to protect clothing from contamination.
    - c. the importance of keeping personal articles such as books, coats, handbags, etc., off of bench areas.
    - d. reporting all spills of infectious materials and the proper method of cleaning up infectious spills.
    - e. reporting all spills of hazardous materials and the proper method of cleaning up hazardous spills.
    - f. the importance of not eating, drinking, smoking, or applying cosmetics in the laboratory.
    - g. the importance of not wearing open sandals in the laboratory.
    - h. the importance of proper trash disposal--separating paper trash from infectious materials and disposing of infectious materials only in specially marked biohazard bags and paper trash in the regular trash cans.
    - i. reporting any cut or accident involving infectious material, no matter how small.
    - j. the importance of disinfecting the bench area daily.
    - k. the importance of locating and interpreting MSDS.
    - l. the importance of Practice Infection Control.
  2. Demonstrate the proper use of common laboratory equipment, to include:
    - a. the microscope
    - b. the centrifuge
    - c. the autoclave
    - d. heating blocks and water baths
  3. State minimum quality control procedures necessary for the equipment listed above. 5.1
  4. Relate the requirements of bacteriologic isolation, to include,
    - a. basic isolation media and the purpose of each
    - b. proper incubation temperature and carbon dioxide levels for CO<sub>2</sub> incubators.

- c. special atmospheres required for some bacteria, such as anaerobes.
- 5. Describe basic microbial cellular structure, to include:
  - a. internal structures--the nucleus, mesosomes, ribosomes, vacuoles, and the function of each.
  - b. cellular wall structures--the cytoplasmic membrane, the cell wall, and capsule and the function of each.
  - c. cellular appendages--flagella and pili and the function of each.
- 6. Define the terms sepsis, asepsis, sterilization, disinfection, and pyogenic and be able to give examples of each state.
- 7. Explain the basic procedures for steam sterilization, dry heat sterilization, ethylene oxide gas sterilization, and filtration sterilization of liquids.

B. Unit II. Specimen Processing

- 1. State the three primary functions of the clinical microbiology laboratory, to include:
  - a. assisting the physician in the diagnosis of and treatment of infectious diseases.
  - b. maintaining excellence of patient care.
  - c. providing reliable results in as short a time as possible.
- 2. State the sequence of steps necessary for a physician to arrive at a clinical diagnosis.
- 3. Indicate which steps in the sequence the laboratory cannot control, which steps the laboratory can issue guidelines for, and which steps in the sequence the laboratory has no control over.
- 4. Define the term infectious disease.
- 5. List at least three manifestations of infectious disease.
- 6. List the four classic signs of infection, to include, redness, swelling, pain, and fever.
- 7. List at least three signs of subacute infection.
- 8. List three manifestations of infectious disease that may be found on X-ray.
- 9. List at least two common laboratory tests used to screen for infectious disease.
- 10. Explain the importance of proper specimen collection.
- 11. Briefly describe the basic concepts of specimen collection.
- 12. Explain how to properly collect specimens from the following body sites and specimen sources:
  - a. throat
  - b. nose
  - c. nasopharynx
  - d. sputum by expectoration, transtracheal aspiration, and bronchoscopy
  - e. urine by the clean catch method, catheterization, and suprapubic tap
  - f. cutaneous wounds and decubiti of the skin
  - g. feces submitted for bacterial culture
  - h. vaginal and urethral cultures
  - i. cultures for Neisseria gonorrhoeae from males and females
  - j. spinal fluid
  - k. eye cultures, conjunctiva and corneal
  - l. ear, external and middle ear
  - m. blood
  - n. surgically removed tissue
  - o. biopsy specimens
  - p. autopsy specimens

- 13. Recognize panic values, notify the doctor, and document the report of these values.

C. Unit III. Direct Examination of Clinical Specimens

- 1. Assess the quality of a specimen received for culture using these criteria:
  - a. sufficient amount received for tests ordered
  - b. appropriate container
  - c. proper label
  - d. slip correctly filled out

- e. specimen received in lab within two hours of collection
  2. Evaluate the specimen by appearance using the following criteria:
    - a. odor
    - b. purulence
    - c. presence of sulfur granules
  3. Perform the following direct microscopic assessments of the specimen and report direct evidence of bacterial, fungal, or parasitic infection.
    - a. wet mounts for motility of amoeba and for the presence of bacteria, yeasts, or trichomonads.
    - b. darkfield microscopy for treponemes.
    - c. Gram's stain on a portion of the specimen to find evidence of bacteria, yeast, or mycelial elements.
    - d. iodine and saline wet mounts for parasites.
    - e. 10% potassium hydroxide (KOH) wet mounts for the detection of fungal elements.
    - f. India ink preparations for the detection of Cryptococcus neoformans in spinal fluid or other body fluid.
  4. Discuss the importance, principle, and purpose of the Gram's stain.
  5. Discuss the importance of reporting direct Gram's stain reports to the physician or to the floor as rapidly as possible when significant organisms are found.
  6. Explain the value of assessing the quality of sputum specimens using the Gram's stain to determine the amount of contamination with saliva by determining the ratio of neutrophils to epithelial cells.
- D. Unit IV. Selection of Initial Plating Media
1. Discuss the various types of media used for primary plating of clinical specimens and state the purpose of each.
  2. Choose the proper selection of media for a variety of clinical specimens according to anatomical site, expected normal flora for that site, and the potential pathogens which might be expected at that site.
  3. Discuss the differences in non-selective media, selective media, differential media, and enrichment media and give examples of each type of media.
  4. Discuss the purpose of using solid agar media and the importance of the four quadrant streaking procedure for obtaining isolated colonies.
  5. Explain the use of a calibrated loop when plating urine specimens and explain why the colony count is important when evaluating urine cultures.
- E. Unit V. Interpretation of Culture Growth
1. Utilize colonial morphology, the Gram's stain reaction, and hemolysis (if present) to begin an identification process.
  2. Determine whether or not an organism is significant by considering the body site the specimen is from, the normal flora expected at that site, and the potentially infectious organisms expected at that site.
  3. Utilize all of the following tests to help confirm or change a preliminary identification:
    - a. Gram's morphology
    - b. catalase test
    - c. slide coagulase test
    - d. bile solubility
    - e. spot indole test
    - f. cytochrome oxidase
  4. Utilize flow charts to plot a series of tests to identify a particular bacterial organism.
- F. Unit VI. Identification of Medically Significant Bacteria
1. Differentiate and evaluate the clinical significance of members of the Staphylococci and Micrococci from members of the Streptococci and Aerococci.

- a. Briefly explain how the differences in the cell wall composition of Gram-positive cocci affect the action of antibiotics, germicidal agents, surface active detergents, and the survival time when they are exposed to drying, heat, sunlight, or chemicals.
  - b. Cite a brief description of the Gram-positive cocci's distribution in the environment and explain their role in normal human flora.
  - c. State the modes of transmission of Gram-positive cocci from one person to another and relate a brief description of the types of infections caused by the Staphylococci and the Streptococci.
  - d. Describe the colonial morphology of the Staphylococci as compared to the Streptococci.
  - e. Cite a brief description of the Gram's stain morphology of the Staphylococci and the Streptococci.
  - f. Describe the hemolytic actions of the Staphylococci and of the Streptococci.
  - g. State the principles, purposes, and brief procedures for the slide coagulase test and the tube coagulase test.
  - h. Explain how the DNase test and the mannitol reactions of Staphylococci can be used to help differentiate the coagulase positive and the coagulase negative Staphylococci.
  - i. Name two types of commercial isolation media available to isolate the Staphylococci from mixed microbial flora.
  - j. Explain the hemolytic action of Streptococci on blood media and discuss the classification of Streptococci by their hemolytic reaction.
  - k. List the four main groups of Streptococci which are important to human pathogens.
  - l. Explain Lancefield typing by the classic method and Streptococcal typing by the co-agglutination method, comparing and contrasting the two methods.
  - m. State the principle, purpose, and a brief procedure for the bacitracin test, or "A" disc method, of identifying Group A Streptococci.
  - n. Relate the principle, purpose, and a brief procedure for the CAMP test.
  - o. State the principle, purpose, and a brief procedure for the sodium hippurate hydrolysis test.
  - p. State how the alpha-hemolytic Streptococci are grouped and explain which tests are used to differentiate each group.
  - q. State the principle, purpose, and a brief procedure for the Lancefield Typing of Streptococci.
  - s. State the principle, purpose, and a brief procedure for the sodium chloride tolerance test.
  - t. Explain the types of diseases caused by pneumococci, their occurrence as normal flora, and the tests used for their differentiation.
  - u. State the principle, purpose, and a brief procedure for the optochin, or "P" disc test.
  - v. State the principle, purpose, and a brief procedure for the bile solubility test.
  - w. Draw a flowchart for the identification of Gram-positive cocci, showing how to differentiate these organisms using the tests discussed above.
2. Differentiate members of the Enterobacteriaceae from other Gram-negative bacilli and differentiate individual members of the Enterobacteriaceae from one another.
    - a. Define the family Enterobacteriaceae by listing reactions of the family, to include:
      - (1) fermentative use of glucose
      - (2) negative cytochrome oxidase activity
      - (3) generally positive nitrate reduction positive
    - b. List the following common methods used to determine the tests required above:
      - (1) Triple Sugar Iron (TSI) reactions
      - (2) Spot cytochrome oxidase test
      - (3) 4-hour nitrate reduction strip or 48-hour nitrate broth test

- c. List the most common selective and/or differential media used to isolate members of the Enterobacteriaceae from routine clinical specimens:
- (1) MacConkey's agar
  - (2) Eosin Methylene Blue agar (EMB)
  - (3) Hektoen Enteric agar
  - (4) XLD agar
  - (5) Salmonella-Shigella agar
- d. List the most common identification tests for the family Enterobacteriaceae and explain the principle and a brief procedure for each test:
- (1) Carbohydrate utilization test ("Sugar Fermentations")
  - (2) ONPG test
  - (3) Oxidase
  - (4) Nitrate
  - (5) Indole
  - (6) Citrate
  - (7) Voges-Proskauer
  - (8) Urea utilization
  - (9) Decarboxylation of lysine, ornithine, and arginine
  - (10) Phenylalanine deamination
  - (11) H<sub>2</sub>S production
  - (12) Determination of motility
- e. Explain the use of a checkerboard matrix chart and flow-chart diagram, including the advantages and disadvantages of each.
- f. Briefly describe the colonial morphology of the Enterobacteriaceae as they appear on blood agar and on MacConkey's agar.
- g. List and evaluate the clinical significance of medically important Genera of the Enterobacteriaceae, to include:
- (1) Citrobacter
  - (2) Enterobacter
  - (3) Escherichia
  - (4) Hafnia
  - (5) Klebsiella
  - (6) Morganella
  - (7) Proteus
  - (8) Providencia
  - (9) Salmonella
  - (10) Serratia
  - (11) Shigella
  - (12) Yersinia
- h. Explain the role of Escherichia coli as normal enteric flora and as a pathogen.
- i. Explain the past history of the Entero-pathogenic Escherichia coli as agents of bacterial diarrhea in infants and explain why serologic typing for these E. coli should be limited to outbreaks of diarrheal disease in newborn nurseries.
- j. Compare and contrast the enterotoxic E. coli and the invasive E. coli as causative agents of diarrheal disease and compare and contrast the methods for their isolation and identification.
- k. Explain the significance of the group of E. coli designated the alcalescens-dispar group and explain how these organisms are differentiated from the Shigellae.
- l. List the four species of Shigellae which cause human disease: Shigella dysenteriae, Shigella flexneri, Shigella boydii, and Shigella sonnei.
- m. Explain the disease caused by Shigellae, bacillary dysentery, and state the most common means of transmission of this disease.
- n. Explain the necessity of serologic typing as the basis for identification of Shigellae as opposed to identification by biochemical means alone.

- o. List the serogroups and their corresponding species: Group A - Shigella dysenteriae, Group B - Shigella flexneri, Group C - Shigella boydii, and Group D - Shigella sonnei.
- p. Outline the media required for the isolation of Shigellae and name the most common types of isolation media used in the clinical laboratory, to include MacConkey's agar, Hektoen Enteric Agar, Xylose Lysine Desoxy- cholate, and Hajna's Gram Negative Broth or Selenite Broth as enrichment media.
- q. Briefly discuss Edwardsiella tarda and give its defining biochemical features, an E. coli-like organism which is lactose negative and H<sub>2</sub>S positive.
- r. Discuss the Salmonellae as causative agents of disease, with attention to the role the Salmonellae play in outbreaks of food poisoning and as agents of enteric fevers, with special attention to Salmonellae typhi and the historical importance of typhoid.
- s. Discuss the serologic complexity of Salmonellae, explaining why clinical laboratories type isolates and why an isolate which does not type in commercial polyvalent A-I + Vi antisera may still be a Salmonella species.
- t. Discuss the necessary isolation media and enrichment broths required for the isolation of Salmonellae.
- u. Compare and contrast the Arizonae to the Salmonellae according to biochemical reactions, pathogenicity, and prevalence.
- v. Discuss the Genus Citrobacter, explaining the role of Citrobacters as members of the normal flora and as potential pathogens, covering C. freundii, C. diversus, and C. amaloniticus.
- w. Discuss the Genus Klebsiella, naming the four species (Kleb. pneumoniae, Kleb. oxytoca, Kleb. ozaenae, and Kleb. Rhinoscleromatis) and explaining the role the Klebiellae play as members of the normal flora and as potential pathogens.
- x. Discuss the Genus Enterobacter, naming the seven currently recognized species (E. aerogenes, E. cloacae, E. agglomerans, E. sakazakii, E. gergoviae, E. amaloniticus, and Hafnia alvei) and explaining the role the Enterobacters play as members of the normal flora and as potential pathogens.
- y. Discuss the Genus Serratia, naming the five currently recognized species (S. marcescens, S. liquefaciens, S. rubidaea, S. odifera 1, and S. odifera 2) and explaining the role the Serratias play as members of the normal flora and as potential pathogens.
- z. Discuss the Genus Proteus, naming the two currently recognized species, P. mirabilis and P. vulgaris, and explaining the role these organisms play as members of the normal flora and as potential pathogens.
- aa. Discuss the Genus Providencia, naming the four currently recognized species (Prov. alcalifaciens, Prov. stuartii, Prov. stuartii, urea positive, and Prov. Rettgeri) and explaining the role these organisms play as members of the normal flora and as potential pathogens.
- bb. Discuss the Genus Morganella, naming the one currently recognized species, Morganella morganii, and explaining the role this organism plays as a member of the normal flora and as a potential pathogen.
- cc. Discuss the Genus Yersinia, naming the three currently recognized species (Y. pestis, Y. enterocolitica, and Y. pseudotuberculosis) and explaining the role these organisms play as animal pathogens and how occasionally man is infected.
- dd. Explain the historical importance of the plague bacillus as the causative agent of the European plagues and as an edemic organism of rodents in the southwest United States.
- ee. Explain the manifestations of disease caused by Y. enterocolitica as an enteric pathogen and of Y. pseudotuberculosis as an occasional pathogen of man.
- ff. Discuss the Genus Erwinia and the Genus Pectobacterium, their roles as plant pathogens, and their rare involvement in human infection.

- gg. Discuss the Genus *Edwardsiella*, naming the one currently recognized species, *Edwardsiella tarda*, and explaining the role this organism plays a potential pathogen.
3. Differentiate, evaluate the clinical significance of, and identify the *Haemophilus* species from other small Gram-negative bacilli. 1.19
- a. Explain the origin of the name *Haemophilus*, meaning "blood loving," in relation to the nutritional requirements of the *Haemophilus*.
  - b. List the growth factors required by the *Haemophilus* and list sources of both factors:
    - (1) Factor V, or nicotinamide adenine dinucleotide (NAD), is also known as Co-enzyme I and is destroyed by heat. It can be isolated from potatoes and yeast extract and is also made in excess by some bacteria, notably *Staphylococci*.
    - (2) Factor X is a heat-stable iron protoporphyrin derivative of heme, present in blood (lysed red cells), or peptic digest of blood, known as Fildes enrichment.
  - c. Describe the morphology of the *Haemophilus* on artificial media and describe their morphology on Gram's stain.
  - d. Discuss the *Haemophilus* species in relation to which members of the group are commensals and which members of the group are pathogens.
  - e. List the pathogenic species and the diseases they cause and discuss the collection, transport techniques, and media required for isolation.
  - f. Discuss the role of serologic typing of capsule material in identification of *Haemophilus influenzae* species, especially in relation to Type B.
  - g. Discuss the disease chancroid caused by *Haemophilus ducreyi*, the methods for its isolation from clinical specimens, and its prevalence in the United States.
  - h. Discuss the condition called "pinkeye," or epidemic conjunctivitis, naming the causative agent(s) and explaining how the organism(s) can be isolated and identified.
  - i. Discuss presumptive methods of identification of the *Haemophilus* species by determination of growth factor requirements and state what tests are needed to completely identify an isolate.
  - j. List the specialty agars available to isolate *Haemophilus* species, such as Casman's Blood agar, Levinthal's enriched agar, and Fildes enrichment agar.
4. Differentiate, evaluate the clinical significance of, and identify the *Neisseria* species isolated from human clinical specimens.
- a. Describe the colonial and Gram's morphology of the *Neisseria*, explaining the similarities between the *Neisseria*, *Acinetobacter*, and *Moraxella* species, including similarities on Gram's stain.
  - b. List the nutritional and atmospheric conditions which must be maintained to isolate pathogenic *Neisseria* species.
  - c. List the members of the *Neisseria* and differentiate the pathogenic species from the commensal species.
  - d. Discuss the disease states caused by *Neisseria gonorrhoeae*, to include symptomatic gonorrhea, asymptomatic gonorrhea, pelvic inflammatory disease (PID), and disseminated *Neisseria gonorrhoeae* infections with involvement of the blood, joints (septic arthritis), and deep organ systems.
  - e. Discuss the types of meningitis caused by *Neisseria meningitidis*, including both epidemic outbreaks and sporadic cases.
  - f. Discuss the proper collection and transport of specimens submitted to the laboratory for the isolation of either *Neisseria gonorrhoeae* or *Neisseria meningitidis*, emphasizing the collection of a specimen which will yield the most organisms and the fewest contaminants.
    - (1) Specimens for *Neisseria gonorrhoeae* in the male should be collected from the anterior urethra and rectum, when indicated.

- (2) Specimens for Neisseria gonorrhoeae in the female should be collected from the endocervical canal and rectum, when indicated.
- (3) Specimens for Neisseria meningitidis are usually spinal fluid and should be collected before administration of any antibiotic.
- g. Explain why a Gram's stain is of little value for diagnosing gonorrhea when no intracellular Gram-negative diplococci can be found.
- h. Discuss the various types of transport media available for collection of N. gonorrhoeae and why it is important to choose a system which will insure a 5 to 10 percent CO<sub>2</sub> atmosphere, for example the Jembec system, a Transgrow bottle, or a Marion Biobag Type C and Thayer Martin plate.
- i. Explain why Thayer Martin enriched chocolate agar is preferable to plain chocolate agar for isolating N. gonorrhoeae from specimens containing mixed flora, but why a back-up plate of plain enriched chocolate should be set up when possible.
- j. List the tests that should be performed on an isolate which is growing on Thayer Martin and which resembles N. gonorrhoeae, to include an oxidase test and a Gram's stain to determine a presumptive identity.
- k. Discuss carbohydrate fermentations by the CTA method and enzymatic carbohydrate degradations as a faster alternative to determining the identity of the isolate of Neisseria.
- k. List the expected carbohydrate utilization reactions for both Neisseria meningitidis and Neisseria gonorrhoeae.
- l. Explain why a beta-lactamase reaction should be performed on all isolates of Neisseria gonorrhoeae and explain the current status of the penicillinase-producing Neisseria gonorrhoeae (PPNG) in the United States.
- m. List at least two methods which allow for identification of Neisseria meningitidis and Neisseria gonorrhoeae from growth at 24 hours, to include fluorescent conjugated antibody stains, hadebact co-agglutination studies, or counter-immunoelectrophoresis.
- n. Explain the serologic typing of Neisseria meningitidis isolates and state which serologic groups are associated with the epidemic form of meningitis and which groups are most often associated with sporadic outbreaks of meningitis.
- 5. Differentiate, evaluate the clinical significance of, and identify the Gram-positive bacilli of importance in human specimens.
  - a. Differentiate the Bacillus species as a group from other Gram-positive bacilli on the basis of colonial morphology and Gram's stain morphology.
  - b. Explain the role Bacilli play as human pathogens, naming Bacillus anthracis as the major invasive pathogen and Bacillus cereus as a cause of outbreaks of food poisoning.
  - c. List the identifying characteristics of Bacillus anthracis and explain how a clinical isolate of Bacillus can be screened to confirm or exclude it as an anthracis species by its colonial appearance, hemolysis reaction on blood agar, and motility reaction.
  - d. Explain why non-hemolytic, non-motile species of Bacillus must be handled with extreme care and be sent to a state health department or reference laboratory for identification.
  - e. Differentiate the Corynebacterium species as a group from other Gram-positive bacilli on the basis of colonial morphology and Gram's morphology.
  - f. List the identifying characteristics of Corynebacterium diphtheria and discuss the historical significance of diphtheria and how development of vaccine has made diphtheria a seldom-seen disease in this country.
  - g. Discuss the pathogenicity of Corynebacterium diphtheria, explaining the role of the beta-phage in toxin production and discussing the Elek test for determining toxin production of isolates.



- h. Discuss the other members of the genus Corynebacterium, their potential pathogenicity, and their role as members of the normal flora of the skin and mucous membranes.
  - i. List the identifying characteristics of Listeria monocytogenes and discuss the diseases caused by Listeria infection.
  - j. List the identifying characteristics of Erysipelothrix rhusiopathiae and discuss the diseases caused by Erysipelothrix infection.
  - k. List the identifying characteristics of the Lactobacilli and discuss their role as members of the normal flora of mucous membranes and their rare involvement in human disease.
6. Differentiate, evaluate the clinical significance of, and identify members of the non-fermentative gram-negative bacilli from members of the Enterobacteriaceae and from members of the fastidious and unusual gram-negative bacilli.
- a. Explain how the oxidase test, carbohydrate utilization tests, ability to grow on MacConkey's, and the growth rate can be used to identify a gram-negative bacillus as belonging to the non-fermenter group.
  - b. List the identifying characteristics of the most common members of the non-fermenter group, to include:
    - (1) Pseudomonas aeruginosa
    - (2) other Pseudomonads, particularly Ps. fluorescens, Xanthomona (S.) maltophilia,
    - (3) Burkholderia (P.), cepacia B. mallei, and B. pseudomallei
    - (4) Acinetobacter calcoaceticus variants Lwoffii and anitratus
    - (5) Flavobacterium species
    - (6) Moraxella species
    - (7) Alkaligenes species
  - c. Compare and contrast the various methods of identification of non-fermenting gram-negative bacilli, to include conventional media and the Elizabeth King charts, micro-method biochemical systems with computer-assisted identification designed for the Enterobacteriaceae, and specialized systems designed to identify non-fermenters.
  - d. Briefly describe the involvement of non-fermentative gram-negative bacilli in human disease.
7. Differentiate, evaluate the clinical significance of, and identify the members of the fastidious and unusual gram-negative bacilli from the Enterobacteriaceae and the non-fermentative gram-negative bacilli.
- a. Explain how slow growth rate, failure to grow on blood agar and MacConkey's agar in twenty four hours, and fermentative use of glucose can be used to separate the fastidious and unusual gram-negative bacilli from other gram-negative bacilli.
  - b. List the members of this group which are important pathogens in human disease but which occur infrequently in this country, to include:
    - (1) Brucella species
    - (2) Francisella tularensis
    - (3) Bordetella pertussis
    - (4) Pasteurella multocida
    - (5) Vibrio cholera
    - (6) Yersinia pestis
    - (7) Legionella pneumophila
    - (8) Chlamydia
    - (9) Rickettsia
    - (10) Leptospira
    - (11) Borrelia
    - (12) Treponema
    - (13) Campylobacter
    - (14) Mycoplasma
    - (15) Helicobacter

- (16) Kingella
- c. Briefly discuss the diseases caused by each of the above organisms and pertinent information required for isolation of the organism, such as special media or special culture techniques.
    - a. Discuss the advantages of serologic tests and direct fluorescent antibody tests for the identification of members of the unusual and fastidious gram-negative bacilli group.
- G. Unit VII. Asepsis and Disinfection
1. Define the terms septic and aseptic.
  2. Define the terms antiseptic and disinfectant and give examples of both types of chemicals.
  3. Explain methods available for determining the effectiveness of specific antiseptics and disinfectants.
- H. Unit VIII. Methods of Sterilization
- State the proper procedures for steam sterilization, to include:
- a. Proper loading of the autoclave to allow good steam flow around materials to be sterilized.
  - b. Selection of the proper cycle and time period.
  - c. State the standard of steam sterilization at 15 minutes at 121° C at 15 pounds of pressure per square inch at sea level.
  - d. Explain the proper procedures for sterilizing glassware using dry heat.
  - e. Explain the proper procedures for sterilizing rubber, plastic, or other materials which cannot withstand steam sterilization using ethylene oxide gas sterilization.
  - f. Explain how liquid suspensions that contain biochemical compounds that break down at high temperature are sterilized using membrane filtration.
- I. Unit IX. Antimicrobial Compounds
1. Briefly discuss the history of antimicrobial compounds.
  2. Discuss the effect antimicrobial compounds have had on the quality of life since their introduction.
  3. List the major groups of antimicrobials and discuss the basic mode of action for each group, to include:
    - a. sulfonamides
    - b. penicillins
    - c. cephalosporins
    - d. erythromycin
    - e. tetracycline
    - f. chloramphenicol
    - g. aminoglycosides
    - h. clindamycin
    - i. vancomycin
- J. Unit X. Selection of Bacterial Isolates for Susceptibility Testing
1. Explain how bacterial isolates are chosen to be tested for antimicrobial susceptibility testing and list the main groups of bacteria which are known to have variable antimicrobial resistances and which must routinely be tested, to include:
    - a. staphylococci
    - b. streptococci
    - c. enteric gram negative bacilli
    - a. non-fermenting gram negative bacilli
  2. Explain why it is not necessary to test all bacterial isolates for antimicrobial susceptibilities.
- K. Unit XI. Antibiotic Susceptibility Testing Method
1. Briefly discuss the history of antimicrobial susceptibility testing.

2. Explain the procedure for performing a Bauer-Kirby Disk Diffusion susceptibility test.
  3. Explain the procedure for performing a Minimal Inhibitory Concentration test (MIC) by both the standard tube method and the microwell method.
  4. Explain how a Minimal Bactericidal Concentration determination is performed from the tubes or wells of a MIC.
  5. Explain the rationale and procedures for performing Serum Bactericidal Concentration titers on peak and through serums from patients undergoing antimicrobial therapy for life-threatening infections.
- L. Unit XII. Quality Control in Microbiology
1. Briefly discuss the required quality control methods for common microbiologic laboratory equipment.
  2. Explain why microbiologic media must be tested with stock cultures prior to use for each lot of media received.
  3. Discuss the quality control methods required for reagents used in the microbiology laboratory.
  4. Discuss the quality control methods used to monitor both Bauer-Kirby disk diffusion susceptibility tests and minimal inhibitory concentration tests.
  5. Explain the rationale for testing microbiology personnel on an individual basis by subscribing to a survey system such as the surveys offered through the College of American Pathologists (CAP).
- M. Unit XIII. Definition of Anaerobic Microbiology
1. Define the term "anaerobic microbiology" and explain the differences between facultative anaerobic organisms and obligate anaerobic organisms.
  2. Explain the difference between microaerophilic organisms and aerotolerant obligate anaerobes.
  3. State conditions that predispose patients to anaerobic infection, to include:
    - a. infections occurring in close proximity to a mucosal membrane
    - b. infections occurring after a human or animal bite
    - c. previous therapy with aminoglycoside antibiotics
    - d. infections in patients who have chronic underlying disease
    - e. patients who develop aspiration pneumonia
- N. Unit XIV. Methods of Anaerobic Culture
1. List the most common methods of providing anaerobic atmosphere for anaerobic culture, to include:
    - a. anaerobic chamber
    - b. anaerobic jar method
    - c. anaerobic bags used for individual cultures
  2. Explain how anaerobic conditions are achieved and maintained in the systems listed above.
  3. Explain how anaerobic media differs from conventional bacterial media and list the required supplements of Vitamin K and heme which must be added to anaerobic formulations.
  4. Explain the term "pre-reduced, anaerobically sterilized" (PRAS) and compare PRAS media to conventional media.
  5. List the selective and differential media that allow isolation of anaerobes from polymicrobial infections.
- O. Unit XV. Identification of Anaerobes
1. Discuss Gram's morphology of anaerobic specimens and list the shapes of the following organisms:
    - a. Anaerobic cocci
    - b. Bacteroides
    - c. Fusobacteria
    - d. Clostridia

- e. Propionibacteria
  - f. Eubacteria and Bifidobacterium
  - g. Actinomyces
2. Compare and contrast the following methods of anaerobic identification:
    - a. Conventional PRAS tubed media
    - b. Gas-liquid chromatography
    - c. Bile susceptibility, esculin hydrolysis, egg yolk agar, and antibiotic disks
    - d. API 20A micromethod
  3. Draw a brief flowchart using the tests mentioned above in section 2 c. showing identification to genus for each of the following groups:
    - a. Anaerobic cocci
    - b. Bacteroides and Fusobacteria
    - c. Clostridia
    - d. Propionibacteria, Eubacteria, and Arachnia
    - e. Actinomyces

P. Unit XVI. Antibiotic Susceptibility Testing of Anaerobes

1. Discuss the most commonly used method of anaerobic susceptibility test.
  - a. Explain how the volume of broth and number of disks is adjusted to simulate the achievable blood level of the antibiotic to be tested
  - b. Explain how the inoculum is added to each tube and how the test is interpreted
2. Briefly explain the Wilkens-Chalgren standardized method of anaerobic susceptibility testing.
3. Compare and contrast the commercial anaerobic MIC tray methods with the both-disk tube method and the standardized Wilkens-Chalgren method.

Q. Unit XVII. Laboratory Behavioral Objectives

The student shall perform the following procedures within the competency limits stated by the instructor:

**Week 1**

Perform ten (10) Gram's stains and streak four (4) agar plates to obtain isolated colonies. Examine six (6) cultures and classify the colonies by size, form, elevation, margin, color, surface, density, and consistency.

**Week 2**

Plate four (4) urine specimens using calibrated loops to obtain colony counts.

Calculate the colony counts of the four (4) plates streaked in the previous lab. Examine five (5) patient urine plates, calculate the colony counts, and assess the significance of the type of growth on each plate.

Perform 10 readings of Gram Staining Sputums and define the criteria of acceptance and nonacceptance.

**Week 3**

Perform Gram's stains on the three (3) Staph-Micro isolates provided. Perform three (3) catalase tests on the organisms provided. Perform two (2) benzidine tests, two (2) slide coagulase tests, and two (2) tube coagulase tests on the Staphylococcus aureus and Staphylococcus epidermidis cultures provided. Streak both Staphylococcus isolates to Mannitol Salt Agar and to DNase agar. Inoculate two tubes of Glucose Fermentation medium, one with Staphylococcus and one with Micrococcus. Stain and interpret the two clinical Gram's stain specimens provided. Assignment of unknowns.

**Week 4**

Perform Gram's stains on the two slides of Streptococci provided. Examine the three blood plates provided and write descriptions for the hemolysis patterns of Alpha-, Beta-, and Non-hemolytic

Streptococci. Perform two (2) bacitracin disk tests, two (2) CAMP tests, two (2) optochin disk tests, two (2) Bile Esculin Azide tests, two (2) Salt Tolerance tests, two (2) benzidine tests, and two (2) bile solubility tests. Using the results of these tests, identify the eight (8) organisms provided. Assignment of unknowns.

**Week 5**

Discussion study cases and first laboratory Practical Exam.

**Week 6**

Perform Gram's stains on Escherichia coli and Acinetobacter calcoaceticus. Perform oxidase tests on Escherichia coli and Pseudomonas aeruginosa. Inoculate nitrate broths with Escherichia coli and Acinetobacter calcoaceticus. Inoculate three (3) 5-tube biosets of Triple Sugar Iron, Lysine Iron Agar, Motility Indole Ornithine, Simmon's Citrate, and Christensen's Urea. The nitrate tubes and biosets will be read and interpreted after 24 hours incubation.

**Week 7**

Inoculate two (2) API-20E enteric gram-negative bacillus identification strips and one (1) Microscan frozen Minimal Inhibitory Concentration/ Identification tray. Examine the plates of Salmonella and Shigella provided and record the results of the demonstration biosets for Salmonella and Shigella. After 24 hours of incubation, the API-20E strips and the micro-scan tray will be determined using the appropriate computer code books. The first set of gram-negative unknowns will be assigned and the first case history presentation will be demonstrated.

**Week 8**

Two (2) Bauer-Kirby tests will be set up and interpreted. A frozen Microscan MIC/Id tray will be set up and interpreted. The first case history with written questions will be presented.

**Week 9**

Perform Gram's stains on Escherichia coli and on Haemophilus influenzae. A blood plate will be inoculated with Haemophilus influenzae to demonstrate the satellite phenomena. Mueller-Hinton plates will be set up with Haemophilus influenzae and Haemophilus parainfluenzae to demonstrate the use of X, V, and XV factor determination strips. A Microscan HNID (Haemophilus-Neisseria Identification tray) will be demonstrated. Beta-lactamase tests will be performed using both a pH detection system and a chromogenic cephalothin system. Case History #3 will be presented with written questions answered.

Perform Gram's stains on the isolates of Neisseria and Acinetobacter. Examine cultures of Neisseria gonorrhoeae, Neisseria meningitidis, Branhamella catarrhalis, and Neisseria lactamica. Transfer these isolates to Thayer-Martin plates and note growth or inhibition.

Inoculate two (2) sets of conventional CTA carbohydrates (CTA Dex, CTA Mal, CTA Lac, and CTA Suc). Set up DNase test on Branhamella and inoculate a Nitrate broth with Branhamella. Minitek Neisseria Identification System and the RIM rapid Neisseria Identification system will be demonstrated for interpretation. The fourth Case History with written questions will be presented.

**Week 10**

Gram's stains will be performed on the following organisms: Bacillus, Clostridium, Corynebacterium, Listeria, Erysipelothrix, and Nocardia. Catalase tests will be performed on all of the listed organisms except for Nocardia. CAMP tests will be performed on Listeria and Beta-hemolytic Strep, Group B. Motility tests at 37° C and at 25° C will be set up on Listeria. TSI slants will be inoculated with Listeria and Erysipelothrix. Biosets of Nitrate, Urea, CTA Dex,

CTA Mal, and CTA Suc will be set up on two isolates of Corynebacterium. Plates of Nocardia will be provided for examination, and the modified acid-fast stain for Nocardia will be performed. Case History #5 will be presented with written questions.

### **Week 11**

Gram's stains will be performed on Acinetobacter, Pseudomonas, and Escherichia. Plates of the following organisms will be presented for study: Pseudomonas aeruginosa, Pseudomonas fluorescens, Acinetobacter calcoaceticus anitratus, Pseudomonas maltophilia, and Pseudomonas stutzeri. Oxidase tests will be performed on each of these organisms. Glucose fermentation media will be inoculated with Escherichia and Pseudomonas. Nitrate testing will be reviewed using Pseudomonas and Acinetobacter isolates. Each student will perform two (2) API-20E gram-negative identification strips on non-fermenters. Case History #6 will be presented with written questions.

Unusual and fastidious gram-negative bacilli will be presented for study, to include Eikenella, Cytocapnophaga, Flavobacterium, and Pasteurella multocida. Gram's stains, oxidase tests, and API-20E strips will be performed. Case History #7 will be presented with written questions.

### **Week 12**

Aerotolerance test will be performed on strict anaerobe, aerotolerant, and facultative anaerobes. Two anaerobic systems type will be used. Identification procedures for anaerobic cocci will be set up on Veillonella, Peptostreptococcus anaerobius and P. asaccharolyticus. Indol, SPS, Gram stain and aerotolerance test will be used in order to facilitate identification of isolates.

### **Week 13**

Second Laboratory Practical Exam.

### **Week 14**

Gram's stains will be performed on the following organisms: Bacteroides fragilis, Fusobacterium necrophorum, F. nucleatum, P. acnes, P. loeschii, and C. sordelli. Special Potency discs for the identification of anaerobes will be used. Perform esculine hydrolysis, indol, lipase, catalase, and lecithinase in known and unknowns strains. Inoculate two Rapid ANA II System for the biochemical identification of anaerobic unknowns.

### **Week 15**

Discussion of study cases and third Laboratory Practical Exam.

### **Week 16**

FINALS

## **III. THECB Learning Outcomes (WECM)**

1. Apply principles of safety, quality assurance and quality control in Clinical Microbiology.
2. Evaluate specimen acceptability.
3. Describe morphology and physiology of microbes.
4. Identify and classify microorganisms.
5. Demonstrate sterile technique.
6. Perform and interpret antimicrobial susceptibility testing.
7. Select additional procedures based on preliminary results.
8. Correlate test results with patient condition(s).

#### IV. Evaluation

##### A. Preassessment

Official MLT Challenge exams have as yet not been structured. Students wishing to challenge a certain course will be given a series of written examinations to display proficiency of lecture material and a series of procedures to display proficiency of laboratory material. The student must score a minimum grade of 70% in order to successfully complete each examination and must meet the minimum competency limits set for individual laboratory procedures.

##### B. Postassessment

Written quizzes will be administered during each unit of lecture material. Written forms for each laboratory procedure are given to the student at the beginning of each laboratory session and are to be completed and presented to the instructor at the next class meeting. Three one-hour exams and a final exam are scheduled for the course.

##### C. Final Examination

A comprehensive Final Exam is scheduled for this course.

##### D. Evaluation

To evaluate students' achievement of course objectives, student grades are tabulated using a final grade break down sheet. To successfully complete MLAB2434 Microbiology, the student must achieve at least a 70% in course components and 80% in lab components. The students overall grade must be no less than a "C," to be allowed to progress to the next program level.

##### E. Remediation

No retakes will be offered for quizzes or exams, including the final exam. Students will receive a grade of zero on all quizzes missed. Make-up exams may be given for excused absences on major exams only at the discretion of the instructor.

THE STUDENT MUST PASS THE FINAL LECTURE EXAM AND THE FINAL LABORATORY EXAM TO SUCCESSFULLY COMPLETE THIS COURSE. A SCORE OF AT LEAST 70% MUST BE SCORED ON THE LECTURE FINAL, AND A SCORE OF AT LEAST 80% MUST BE SCORED ON THE LABORATORY FINAL GRADE.

##### F. Grading

Grading will follow current EPCC Catalog standards. The assignments of letter grades to percentage scores and the final grade determination will be as follows:

90 - 100 = A  
 80 - 89 = B  
 70 - 79 = C  
 60 - 69 = D  
 59 and below = F

Lecture and Laboratory Grades will be evaluated as follows:

<u>Lecture Grades</u>		<u>Laboratory Grades</u>	
Quiz Average	= 20%	Quiz Average	= 22.5%
First hour exam	= 20%	Practical Exam #1	= 22.5%
Second hour exam	= 20%	Practical Exam #2	= 22.5%
Third hour exam	= 20%	Practical Exam #3	= 22.5%
Final hour exam	= <u>20%</u>	Practical Reports	= <u>10.0%</u>
	100%		100%

Laboratories will be graded on a Pass/Fail system based on the competency limits set by the program for each individual procedure.

#### G. **Health Occupations Division Criteria for Course Pursuit**

In order to establish guidelines for determining when a student has ceased to pursue the course objectives, the Health Occupations Division has set the following standards.

1. The student must adhere to the attendance requirements of course MLAB 2434. In order to pursue the course, the student must attend a minimum of 42 lecture hours and 42 laboratory hours of instruction. No more than 6 hours of lecture or 6 hours of laboratory may be missed if the student intends to pursue the course.
2. The student will be able to make up none of the missed lecture hours and up to 3 hours of missed laboratory hours by appointment with the instructor.
3. Tardiness will be defined as being ten (10) minutes or less late to a lecture or laboratory session. Students tardy in excess of the above will be considered absent and this will be recorded as an unexcused absence. Students will be allowed three (3) events or less of tardiness, after which the tardiness will be considered an absence.
4. If required by instructor/coordinator, student also must follow the standards established in the Allied Health & Nursing Program Student Handbook and/or program addendum. The student is bound by standards in the Allied Health/ Nursing Handbook as evidenced by the return of a signed/dated acknowledgment sheet.
5. Where the student continues to pursue the course objectives but is receiving failing grades, he/she will remain eligible to complete the course, except in instances where unsafe practice occurs.
6. The student must appear for examinations, presentations, or other required class activities and submit required papers, projects and/or reports as identified in the course syllabus/calendar.

Failure of the student to follow the above guidelines will indicate that the student is no longer pursuing the objectives of the course and will result in faculty initiated withdrawal.

#### V. **Disability Statement (American with/Disabilities Act [ADA])**

EPCC offers a variety of services to persons with documented sensory, mental, physical, or temporary disabling conditions to promote success in classes. If you have a disability and believe you may need services, you are encouraged to contact the Center for Students with Disabilities to discuss your needs with a counselor. All discussions and documentation are kept confidential. Offices located: VV Rm C-112 (831-2426); TM Rm 1400 (831-5808); RG Rm B-201 (831-4198); NWC Rm M-54 (831-8815); and MDP Rm A-125 (831-7024).

#### VI. **6 Drop Rule**

Students who began attending Texas public institutions of higher education for the first time during the Fall 2007 semester or later are subject to a 6-Drop limit for all undergraduate classes. Developmental, ESL, Dual Credit and Early College High School classes are exempt from this rule. All students should consult with their instructor before dropping a class. Academic assistance is available. Students are encouraged to see Counseling Services if dropping because exemptions may apply. Refer to the EPCC catalog and website for additional information.



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