

*This protocol contains a list of the experiments that will be performed each lab period. The experiments are designed to provide students with the best possible opportunity to have a successful learning experience. The protocols may not follow the procedures outlined in the lab manual some substitutions for organisms may be made. That information will be provided by the lab instructor. Students will be responsible for understanding the **principles** set forth in each experiment. In addition text book readings from Microbiology A Human Perspective by Nester 4<sup>th</sup> edition are listed which will further explain and help you understand the principles in these experiments.*

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**June 25****Lab 1**

**Lab safety protocol**  
**Aseptic Technique**  
**Pure culture technique**  
**Bacterial Distribution**  
**Discuss: Genus and species; and morphology**

Textbook readings:

Pg 83-4 Section 4.1 Obtaining a Pure Culture read through The Streak-Plate Method

Pg 111-2 Microbiology Laboratories

Pg 250-1 Culture Characteristics

Pg 11 Nomenclature

Pg. 246-7 Taxonomic Hierarchies

Pg A-4 Appendix III Pronunciation Key

Pg G1-18 Glossary

**June 26****Lab 2**

**Complete aseptic technique; pure culture technique; bacterial distribution**  
**Bright-field Microscopy**  
**Making a Smear**  
**Simple stain**  
**Gram stain**

Textbook readings:

Pg 40-1 Section 3.1 Microscopic Techniques: The Instruments read through Contrast

Pg 282 The Genus *Corynebacterium*

Pg 45-7 Microscopic Techniques: Dyes and Staining read up to Acid-Fast Stain

Pg 49-51 Section 3.3 Morphology of Prokaryotic Cells

Pg 248-9 Microscopic Morphology read through special stains  
Pg 58-9 The Gram Positive Cell Wall and the Gram Negative Cell Wall read up to  
the lipopolysaccharide molecule  
Pg 61-2 Differences in Cell Wall Composition and the Gram Stain

**June 28**  
**Lab 3**

**Complete simple stain and gram stain**  
**Capsule stain**  
**Spore stain: Schaeffer-Fulton method**  
**Acid-fast stain: Ziehl-Neelsen method**  
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**streak plate and gram stain practice**

Textbook readings:

Pg 249 Special Stains  
Pg 63 Section 3.7 Surface Layers External to the Cell Wall  
Pg 47 Capsule Stain  
Pg 470 Avoiding Recognition and Attachment (Capsules)  
Pg 48 Endospore Stain  
Pg 67-9 Endospores read up through Microcheck 3.9  
Pg 283 Endospore-Formers  
Pg 698-703 Lockjaw and Gas Gangrene  
Pg 47 Acid-Fast Stain  
Pg 670-2 Hansen's Disease (Leprosy)

**July 2**  
**Lab 4**

**Quiz 1 (Labs 1-3)**  
**Complete capsule stain, spore stain, and acid-fast stain**  
**Effect of Temperature, pH, and Salt concentration on bacterial growth**  
**Effect of Ultraviolet light on cell viability**  
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**Streak plate for a grade**

Textbook readings:

Pg 86 Section 4.3 Environmental Factors that Influence Microbial Growth  
Pg 23-4 pH  
Pg 99 Measuring Biomass (Turbidity)  
Pg 54 Permeability of the Cytoplasmic Membrane  
Pg 293-4 Section 11.9 Archaea that Thrive in Extreme Conditions  
Pg 122-3 Section 5.6 Using Radiation to Destroy Microorganisms and Viruses  
Pg 196 Section 8.4 Repair of Damaged DNA

**July 3**

**Lab 5**

**Hand in Laboratory Reports (Labs 1-3)**  
**Complete temperature, pH, and salt concentration**  
**Complete ultraviolet light**  
**Atmospheric Growth Requirement testing**  
**Bacterial population counts (Spread and Pour Plates Methods)**

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**Gram stain for a grade**  
Textbook readings:  
Pg 98-9 Plate Counts  
Pg A-1 Appendix I Microbial Mathematics  
Pg 88-9 Oxygen (O<sub>2</sub>) Requirements  
Pg 94-5 Providing Appropriate Atmospheric Conditions  
Pg 268-276 Section 11.1 Anaerobic Chemotrophs  
Pg 281-2 Section 11.5 Aerobic Chemoorganotrophs

**July 5**

**Lab 6**

**Complete spread and pour plates; atmospheric growth**  
**Differential and selective media**  
**Staphylococci**  
**Streptococci**

Textbook readings:  
Pg 93-4 Section 4.5 Cultivating Prokaryotes in the Laboratory read up Providing Appropriate Atmospheric Conditions  
Pg 290 Section 11.8 Bacteria that Inhabit the Skin  
Pg 372-6 Section 15.1 Overview of the Innate Defenses and Section 15.2 First-Line Defenses  
Chapter 22 Skin Infections and Chapter 23 Respiratory System  
Pg 641-3 Staphylococcal Toxic Shock  
Pg 693-6 Section 27.2 Common Bacterial Wound Infections

**July 9**

**Lab 7**

**Quiz 2 (Labs 4-5)**  
**Complete Staphylococci; Streptococci**  
**Continue differential and selective media**  
**Dental Caries**  
**Preparation and care of stock cultures**  
**Gram Positive Cocci Unknowns**  
**Catalase production**  
**Coagulase production**  
**Novobiocin sensitivity**

Textbook readings:  
Pg 601-4 Section 24.3 Bacterial Diseases of the Upper Alimentary System  
Pg. 84 Maintaining Stock Cultures

**July 10**

**Lab 8**                    **Continue identification of Gram Positive Cocci Unknown**  
**Bacitracin, CAMP, SXT, Enterococcosel, 6.5% NaCl, and Optochin**  
**\*\*NOTE\*\***            **Any students that wants to bring in a water sample to test please collect**  
**sampling bottle from instructor.**

**July 12**

**Lab 9**                    **Quiz 3 (Labs 6-8)**  
**Hand in Laboratory Reports (Labs 4-6)**  
**Continue identification of Gram Positive Cocci Unknown**  
**Detecting Coliform Bacteria in water: Multiple tube method**  
**Bacterial Analysis of food**  
**Motility**

Textbook readings:

Pg 791 Section 31.2 Drinking Water Treatment and Testing

Pg 99 Most Probable Number (MPN)

Pg 112 Foods and Food Production Facilities

Pg 114-5 Read Pasteurization through The Commercial Canning Process

Pg 123-4 Section 5.7 Preservation of Perishable Products

Pg 474 Superantigens

Pg 610-621 Section 24.5 Bacterial Diseases of the Lower Alimentary System up to  
Hepatitis A

Pg 672-4 Botulism

Pg Chapter 32 Food Microbiology

Pg 63-6 Section 3.8 Filamentous Protein Appendages

Pg 74 Flagella and Cilia

**July 16**

**Lab 10**                    **Complete motility experiment**  
**Complete identification of Gram Positive Cocci Unknown**  
**Continue water experiment**  
**Complete food experiment**  
**Biochemical tests**  
**Identification of Gram negative unknown**  
**\*\*NOTE\*\***            **Hand out vials for students to bring in mouth wash for next period**

Textbook readings:

Pg. 251 Biochemical Tests

Pg 29-31 Section 2.5 Carbohydrates

Pg 25 Section 2.4 Proteins and Their Functions

The media in these experiments have been pre-inoculated to provide both a positive and negative test result. Students are to observe and record the reactions and where appropriate will perform certain tests

with the reagents provided. Tests will be available for observation over the next lab period. 1 set-up/3 students. Also: See Biochemical charts 1-8 in laboratory manual and the color pictures in the illustration binders.

**Carbohydrate fermentation:**

phenol red lactose blank  
*E. coli* in phenol red lactose  
*P. vulgaris* in phenol red lactose

**Methyl red test (Mixed acid fermentation):**  
**students will perform test with methyl red reagent**

MRVP broth blank  
*E. coli* in MRVP broth  
*E. aerogenes* in MRVP broth

**Voges-Proskauer test (Butanediol fermentation):**  
**students will perform test with Voges-Proskauer reagents**

MRVP broth blank  
*E. aerogenes* in MRVP broth  
*E. coli* in MRVP broth

**Indole (Tryptophan hydrolysis):**

tryptone broth blank  
*E. coli* in tryptone broth  
*E. aerogenes* in tryptone broth  
**students will perform test with Kovac's reagent**

**Urea hydrolysis:**

urea broth blank  
*P. vulgaris* in urea broth  
*E. coli* in urea broth

**Triple Sugar Iron Agar:**

Triple sugar iron agar slant  
*P. vulgaris* on Triple sugar iron agar slant  
*E. aerogenes* on Triple sugar iron agar slant

**Citrate utilization:**

Simmon's citrate slant  
*E. aerogenes* on Simmon's citrate slant  
*E. coli* on Simmon's citrate slant

**Phenylalanine deamination:**

*P. vulgaris* on phenylalanine slant  
*E. coli* on phenylalanine slant  
**dropping bottle of 10% ferric chloride**

**Decarboxylase**

lysine decarboxylase blank  
*E. aerogenes* in lysine broth  
*P. vulgaris* in lysine broth

**July 17**  
**Lab 11**

**Quiz 4 (Labs 9-10)**

**Hand in Laboratory Reports (Labs 7-10)**  
**Gram Positive Cocci unknown report due**  
**Continue identification of Gram negative unknowns**  
**Complete water experiment**  
**Antiseptics Chemical Agents of Control: The Filter Paper Disk Method**

Textbook readings:  
Pg 110 Disinfection  
Pg 112-3 Section 5.2 Selection of an Antimicrobial Procedure  
Pg 116-121 Section 5.4 Using Chemicals to Destroy Microorganisms and Viruses

**July 19**  
**Lab 12**

**Continue identification of Gram negative unknowns**  
**Complete antiseptics experiment**  
**Isolation and Enumeration of Bacteriophages**

Textbook readings:  
Pg 323-9 Viruses of Bacteria Section 13.1 General Characteristics of Viruses and  
Section 13.2 Virus Interactions with Host Cells

**July 23**  
**Lab 13**

**Continue identification of Gram negative unknowns**  
**Complete virus experiment**  
**Antibiotic Sensitivity Testing: A. Kirby-Bauer test    B. Synergistic effects**  
**Determination of penicillin sensitivity**

Textbook readings:  
Chapter 21 Antimicrobial Medications

**July 24**  
**Lab 14**

**Quiz 5 (Labs 11-12)**  
**Continue identification of Gram negative unknowns**  
**Complete Antibiotics experiment; penicillin experiment**  
**Slide agglutination test: Sure-Vue Select**  
**Synthetic epidemic**

Textbook readings:  
Pg 372 3<sup>rd</sup> paragraph In addition.. read up to Section 15.1 Overview of Innate Defenses  
Pg 397-6 Section 16.3 and 16.4 The Nature of Antigens and The Nature of Antibodies  
Pg 432 Section 17.5 Agglutination Reactions  
Pg 6-8 Medical Microbiology  
Chapter 20 Epidemiology

**July 26**  
**Lab 15**

**Lab Exam (Labs 13, 14, and comprehensive)**  
**Gram negative unknown report due**  
**Hand in Laboratory Reports (Labs 11-14)**