Laboratory Schedule for MCB 2010L

To be successful in microbiology lab, you must read the exercise in the lab manual and incorporate any changes posted in the schedule. Each week you must know the organisms you are studying and diseases they cause. The links below (blue) will connect with images, case studies, or articles pertaining to that particular lab.

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E-mail Dr. Yelena Rudayeva rudayevy@palmbeachstate.edu if you have questions.

WEEK	LAB EXERCISES
1.	**Introduction to Lab and Lab Policies--pages 1-6** 3-1 Introduction to the Light Microscope pages 70, 73 - 75 No activities from this module; use as needed to see Helminth slides. Parts to learn: oculars, objectives, diaphragm lever, condenser, coarse and fine adjustment
2	12-4 Helminth Parasites pages 416 - 424 Demo slides: Eggs: *Clonorchis sinensis, Taenia sp., Ascaris lumbricoides, Enterobius vermicularis , Schistosoma* Larva*: Strongyloides stercoralis* Scolex: *Taenia sp.* For each of these, you should be able to recognize the egg/scolex, know the name of the disease caused, symptoms, transmission and clinical specimen. **Case study:** Think wild salmon is the best? Think again. http://www.palmbeachstate.edu/faculty/duncombt/microlab/Salmon.html <> Website for reference: http://www.dpd.cdc.gov/DPDx/HTML/Para_Health.htm
2	12 - 3 Examination of Common Protozoans of Clinical Importance pages 409 - 415 Demo slides: Cysts: *Entamoeba histolytica, Giardia* Trophozoites: *Giardia, Plasmodium sp., Trichomonas, Trypanosoma* *Toxoplasma gondii* (there is no slide for this organism) For each of these, you should be able to recognize the trophozoites and/or cysts, know the name of the disease caused, symptoms, transmission and clinical specimen.

	Check out the local outbreak of malaria in Palm Beach County: http://www.cdc.gov/mmwr/preview/mmwrhtml/mm5238a3.htm Website for reference: http://www.dpd.cdc.gov/DPDx/HTML/Para_Health.htm
3	12-1 The Fungi---Common Yeasts and Molds pages 398 - 405 Demo slides and plates: Yeast: *Candida* Mold: *Aspergillus, Penicillium* Terms: dimorphic, hyphae, mycelium, mycosis 2-1 Ubiquity pages 34 - 35*** 4 nutrient agar (TSA) plates/table; swabs and saline tubes
4	2 - 1 Results All of the following links go to the same page. **3-5 Simple Stains pages 100 - 102** Use procedure on page 101 for making a smear of either *Escherichia coli* or *Staphylococcus epidermidis.* Once the smear is completed, follow instructions for Gram stain in Experiment 3-6, page 109. **3-8 Acid Fast Stain pages 110 - 114** Demo slide of *Mycobacterium tuberculosis* 3-9 Capsule Stain pages 115 - 116 Demo slide of *Klebsiella pneumoniae* **3-10 Endospore Stain pages 117 - 120** Demo slides of *Bacillus, Clostridium* **Case Study:** Read about a TB investigation aboard a naval ship http://www.cdc.gov/mmwr/preview/mmwrhtml/mm5551a3.htm?s_cid=mm5551a3_
5	**From this lab on, you will need proper attire for lab.** Both of the following modules are on the same webpage. **1-3 Aseptic Transfer of Microorganisms pages 17 - 24** Organisms: *Staph aureus, Escherichia coli* (slants) Each student transfers one organism (either *B. subtilis* or *E. coli*)into a broth and a slant. Read the Basics, pages 17-24. Follow instructions on page 21 for removing a sample from a slant; page 22, fish tail inoculation; page 22, inoculation of broth tubes. A practice run will be done by each student using blank tubes. 1-4 Streak Plate Method of Isolation pages 25 - 28 Your instructor will demonstrate a correct streaking technique. Each student will do a practice run on an empty Petri dish and then streak the following mixture onto an agar plate. Organisms: mixture of *Serratia, Enterobacter*

	Case Study: This article illustrates the damage done by not following aseptic procedures. Read "Multiple Misdiagnoses of Tuberculosis Resulting from Laboratory Error-- Wisconsin, 1996" in CDC's, MMWR, (page 797) http://www.cdc.gov/mmwr/PDF/wk/mm4634.pdf Read Incidents 2 and 3, then scroll to and read the Editorial Note.
6	1-3 Results 1-4 Results 3-7 Gram Stain pages105 - 109 Use procedure on page 101 for making a smear of either *Escherichia coli* or *Staphylococcus aureus.* Once the smear is completed, follow instructions for Gram stain in Experiment 3-6, page 109. Each student does a Gram stain on the smear they made in 3-5.
7	**MID TERM PRACTICAL** 1. A 5. GIARDIASIS 9. D 13. C 2. D 6. B 10. D 14. NO 3. C 7. STOOL 11. C 15. GRAM(-) BACILLI 4. C 8. C 12. SPUTUM 16. C
8	2-7 Fluid Thioglycollate Medium*** Organisms: *E. coli, Clostridium perfringens, Micrococcus luteus* <>**2-8 Anaerobic Jar pages 52 - 53** 2-14 Disinfectants*** page 66-67 Organisms: *Staphylococcus aureus* and *Escherichia coli* Each table will prepare a set of plates covering the 8 disinfectants. One pair will test disinfectants 1-4 against *S. aureus* and *E. coli;* the other pair will test disinfectants 5-8 against the two organisms. Note: You will be swabbing the plate to create a "lawn" rather than streaking for isolation in this experiment as well as 2-14. See page 27. 7-3 Antimicrobial Susceptibility Test pages 268 - 271 Omit steps 1-3 under Test protocol. Nutrient agar/TSA plates will be substituted for Mueller-Hinton.
9	2-7 Results 2-8 Results 2-14 Results 7-3 Results **DIFFERENTIATION OF GRAM NEGATIVE ORGANISMS pages 142 - 146** **ISOLATION OF ENTERICS*****Read "Selective Media.." on page 139 Organisms *E. coli, Enterobacter aerogenes, Proteus vulgaris, Salmonella spp., Shigella spp.* Using resources in your lab/text books and others provided by your instructor, know

	the color of the colonies formed by the above organisms on EMB and Hektoen (4-6, 4-7) It is helpful to prepare a chart listing the above organisms on the left side of the chart and the 3 agars across the top. Fill in the colors. For example, *E. coli* will turn a metallic green on EMB. Each pair will streak an unknown for isolation on the 2 agars
10	4-6, 4-7 Results 7-6 Results **5-13 Urea Hydrolysis pages 187 – 189** **5-22 Lysine Iron Agar pages 209-210** Organisms: *Proteus spp., Salmonella spp., Shigella spp.* Materials: Per table – 1 Urea broth tube, 1 Lysine broth tube Procedure: 1) One pair at each table will inoculate a urea broth tube with an enteric unknown. 2) The other pair at the table will inoculate a lysine broth tube with a different enteric unknown. 3) Record the reaction results after incubation. **10-5 Ultraviolet Radiation Damage and Repair pages 64 - 67, 364 - 366** http://www.palmbeachstate.edu/faculty/duncombt/UVRadiationLab.pdf Organism: *E.Coli* (fresh broth) Radiation equipment: PCR chamber with UV length 254 nm Materials: Per pair – 3 TSA plates, 3 cotton swabs Procedure: 1) Label each of the TSA plates: #1 control no exposure, #2 exposure 30 seconds, #3 exposure 60 seconds 2) Spread the 3 TSA plates with *E. Coli* 3) After all plates have been inoculated, remove the lids from the 30 second plates and place into the PCR chamber. Replace the lids after the exposure and seal with parafilm. Repeat the process with the 60 second plates. **Case study:** Follow the detectives as they trace a disease outbreak to alfalfa sprouts! http://www.cdc.gov/mmwr/preview/mmwrhtml/00048994.htm **5-30 Enterotube II pgs.232 – 235** Materials: Enteric unknown plates Procedure: Inoculate Enterotube from your Enteric Plates
11	5-13 Results 5-22 Results 10-5 Results **DIFFERENTIATION OF GRAM POSITIVE ORGANISMS** **4-1 Mannitol Salt Agar pages 137 - 138** Read "Selective Media for Isolation of Gram Positive Cocci", page 108 Organisms: *Staphylococcus aureus, Staphylococcus epidermidis*

	Materials: 1 MSA plate per pair Procedure: Fishtail streak each organism on one half of the MSA plate 5-25 Blood Agar & Hemolysis pages 217 - 219 **You will not do throat cultures.** Streak using the loop. Organisms: *Streptococcus pyogenes, Staphylococcus aureus, Staphylococcus epidermidis* Materials: two BA plates per pair Procedure: 1) On one BA plate, streak *S. pyogenes* for isolation. 2) Divide the other BA plate in half. 3) Fishtail streak one half of that BA plate with *S.aureus* 4) Fishtail streak the other half of the BA plate with *S. epidermidis.* **5-24 Bacitracin Susceptibility Test*** pgs. 214 - 216** Read the information. Materials: The BA plate that was streaked with *S. pyogenes.* One bacitracin antibiotic disk from disk clip Procedure: Put a bacitracin disk on the first streak of the *S. pyogenes* BA streak plate.

	4 - 1 Results 5-25 Results 5-24 Results **5-5 Catalase Test*** pgs. 165 - 167** Materials: MSA plate from 4-1 and BA plate from 5-25 1 dropper bottle with hydrogen peroxide, H_2O_2 Procedure: 1) Using the growth on plates from 4-1, MSA and 5-25, BA, perform the catalase test. 2) Place one drop of hydrogen peroxide directly on the growth on each plate. **5-27 BBL™ Staphyloslide™ Latex Test for *Staphylococcus aureus* pgs. 222 - 223** Materials: MSA plate from 4-1 and BA plate from 5-25 Procedure: 1) Transfer a loopful of material on the slide 2) Mix with one drop of coagulase plasma 3) Observe agglutination in 2 min You must register at http://www.medscape.com to read the following article. **Case Study:** Read about MRSA cases found in the community at http://www.medscape.com/viewarticle/450901_print **11 - 4 Slide Agglutination Test** pages 382 - 383: (read for theory only)** This will be demonstrated using a rapid diagnostic test kit for *Staphylococcus aureus* antigen. **DEMO** Look at demo plates of *Streptococcus pneumoniae* to observe alpha hemolysis and reaction to optochin. **6-2 Urine Streak*** SemiQuantitative Method pages 248 - 249** Unknown cultures will be used rather than fresh urine specimens. Agars: EMB, Blood, SAB Organisms: *Escherichia coli, Enterobacter aerogenes, Staphylococcus epidermidis, Streptococcus pyogenes, Candida albicans* Procedure: Each pair will streak their unknown on 1 of each of the agar plates.
13	6-2 Results 7-6 Epidemic Simulation ***pages 276 – 277
14	**ELISA*** Test for HIV/ Rapid Diagnostic testing (Immune System applications)** View an ELISA animation prior to coming to lab.

15	Review for Final Lab Practical
16	**FINAL LAB PRACTICAL** **Answers to Final Lab Practice Practical:** 1) microaerophilic 2) c. Micrococcus luteus 3) If an anaerobic organism is suspected of being the cause of an infection, the plates would be incubated in this jar 4) a and b both will grow 5) 3 6) non-fermenter 7) a, b, or e either *Salmonella*, *Shigella* or *Proteus* 8) a 9) yes 10) *Staphylococcus aureus* 11) *Streptococcus pyogenes* 12) antibody 13) If the person were recently infected, their antibody levels may not yet be detectable 14) blood 15) *Streptococcus pneumoniae* 16) c gram positive cocci 17) c 18) *Escherichia coli* 19) a 20) d
	*****Procedure different than in the lab manual *****