**Lab Exam 3 Objectives**

Note: These learning objectives are intended as a study guide. This is not intended to be the sole source of your studies. **This is not necessarily a complete guide, use of the lab handouts and lab questions too!!**

Discuss the relationship of capsules with virulence.
Recognize steps of a capsule stain and compare with a negative stain.
Perform and list all the steps of a spore stain. Note the results of a positive and negative stain.
What genera are endospore formers, and you will need to do at least one spore stain without a lab book.
Perform and list all the steps of an acid fast stain. Note the results of a positive and negative stain. What genera are acid fast positive, and what makes them stain this way. You will need to do at least one acid fast stain without a lab book.
Use the spectrophotometer to estimate growth of broth cultures, note how to use a blank to zero, and read OD or absorbance.

Compare the growth curve by plate count and spectrophotometry.
Be able to make bacteria counts from plates with known amounts of solution.
Be able to explain how to make a serial dilution of a sample and also read of plate with growth from a diluted sample to determine the concentration of the original sample.
see web site  
http://faculty.sdmiramar.edu/dtrubovitz/micro/dilutions/dilutionproblems.htm

Compare the effectiveness of hand sanitizer, soap and the alcohol wipes. Explain and be able to interpret the raw data, see lab report.

Compare the effectiveness of the antiseptics we used with the different organisms we tested. Note the methods used to evaluate the antiseptics.

Define optimum, minimum and maximum temperatures for growth.

Explain how to grow obligate anaerobes.

Recognize oxygen requirements from FTM tubes.

Compare the growth of *Serratia marcescens* and *E. coli* at different temperatures.

Define mesophiles, psychrophiles, and thermophiles and give examples.

Compare TDP and TDT as terms and for the organisms used in lab.

Compare the lethal effects of temperature on *Staphylococcus aureus*, *Bacillus species* and *Escherichia coli* and discuss why their is a difference.

Compare the effects of pH on the growth of the organisms that we used in lab.

Graph the growth using percent transmittance against pH or temperature, see worksheet for these lab.

Describe how UV did effect our test organisms, and how that differed with what we expected. Explain why.

Define Transformation and briefly summarize the procedure

Compare the results and the purpose of the plates:
- LB DNA-
- LB/amp DNA -
- LB/amp DNA +
- LB/amp/ara DNA+

Explain what pTOM (red version of pGLO) is and what RFP (GFP) is. See web site:
http://faculty.sdmiramar.edu/dtrubovitz/micro/pglo
or
http://faculty.sdmiramar.edu/dtrubovitz/micro/pglo.pdf
Explain the function of the following in transformation:

- Transformation solution
- LB
- Ampicillin
- Arabinose

Explain how to use the filter paper disk method of antiseptic evaluation.

Compare effectiveness of different antiseptics on the different organisms used in class.

Define zone of inhibition, and what causes it as well as why it may differ with diffusion.

Speculate on how an antimicrobial may have a large zone of inhibition but in reality is not very effective.

Give examples of effective and ineffective antibiotics against the organisms we used in lab:

- *S. aureus*
- *E. coli*
- *P. vulgaris*
- *P. aeruginosa*

Explain how gel electrophoresis works.

Compare the speed of DNA by size of fragment.

Explain how size of the DNA piece is related to the distance traveled in agarose.

Estimate DNA fragment size by comparison to a DNA ladder.

Compare RFLP (and define it) with PCR for DNA fingerprinting.

What is the function of the following:

- agarose
- TAE or TBE buffer
- loading dye
- UV light box