Microbiology Lab Experiment Changes

Experiment #: 3-7

Title: Acid-Fast Stain

Live Organisms: *Mycobacterium phlei, E. coli*

Changes:

Modified procedure:

1. Make E. coli smear slightly cloudy as usual. For the M. phlei smear, just start mixing right away. Break up clumps as best as you can. Smear will be clumpy.
2. Put **heat fixed** smear onto "staining hot plate" in fume hood.
3. Place a filter paper over smear. Saturate filter paper with carbol fuchsin stain.
4. Allow stain to sit approximately 5 minutes.
5. Get a paper towel to carry slide to sink. Throw filter paper into trash. Rinse slide well.
6. Decolorize with Acid-Alcohol: 1-2 drops for M. phlei and five+ drops for E. coli.
7. Counterstain with Methylene Blue for 1-2 minutes.
8. Rinse with water. Blot dry. You may wipe **BOTTOM** of slide with lens cleaning fluid and tissue.

Each student will make one slide of each organism.

Take Home Lesson: You need to know the reagents and their function in the staining procedure. Why do we use the acid-fast stain? On what organisms is it used? Be able to recognize that an organism is acid-fast or non-acid-fast based on its appearance under the microscope.