

Basic Culture Media and Isolation Techniques Laboratory Exercises

After you have completed the Basic Culture Media and Isolation Techniques eLearning course, it is strongly recommended that you complete the following laboratory exercises to transfer the didactic content of the course to experiential knowledge gained through hands-on laboratory exercises with your equipment in your laboratory. Your supervisor/mentor should work with you to develop these laboratory skills as well as confirm that these exercises have been completed. The number and types of exercises you will complete will be at the discretion of your supervisor/mentor based on procedures followed within your laboratory. Included in the laboratory exercises portion of this course are the objectives of the exercises as well as the prepared exercises. After the laboratory exercises are completed and discussed with your supervisor/mentor, your supervisor/mentor should then follow-up the exercises with instruction related to your laboratory's specific procedures or guidelines.

Laboratory Exercise Objectives:

After completing the basic culture media and isolation techniques laboratory exercises, you will be able to:

- Explain the different types and forms of culture media.
- Correlate culture media with specimen type.
- Describe the critical steps in the inoculation of media to obtain isolated colonies.
- Identify commonly encountered problems with culture media and isolation techniques.

Note: Be sure to review the proper use of personal protective equipment (PPE) and laboratory equipment according to your laboratory's procedures and safety manual.

Supply List

- 1. Personal protective equipment (PPE) and laboratory equipment
- 2. Loops (sterile plastic or metal)
- 3. Incinerator or Bunsen burner (if using metal loops)
- 4. Broth culture containing isolated colonies
- 5. Labelling pen
- 6. Biohazard waste container: for personal protective equipment
- 7. Sharps container: For loops
- 8. Incubator

Culture Media List

- 1. Broth culture containing microorganisms to be plated
- 2. Culture media: BAP, CHOC, MAC, C-CNA/PEA, CAMPY, CHROMagar, HE/XLD, SMAC, AnaBAP
- 3. Previously inoculated media for observation



Laboratory Exercise I

Objectives:

After completing this laboratory exercise, you will be able to:

- Explain the different types and forms of culture media.
- Correlate culture media with specimen type.

Exercise: Choose Appropriate Culture Media

Obtain a broth culture containing microorganisms from your supervisor/mentor. Your supervisor/mentor should supply you with where the culture originated from (such as wound, respiratory, stool specimen, etc.) and any other pertinent information. For this exercise, you will determine the culture media to use from the specimen source type you received. Once you have chosen the media to use, label the plates with identification information such as name, source, and date.

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Laboratory Exercise II

Objectives:

After completing this laboratory exercise, you will be able to:

• Describe the steps of streaking a plate.

Exercise: Perform a Quadrant Streak

Perform quadrant streaking to obtain isolated colonies using the broth culture given to you in Exercise I. Use the labeled culture media you chose from that exercise to perform the quadrant streaking.

*Follow the Quadrant Streaking job aid (See Appendix) to perform isolate streaking.

Instructions

- 1. Label the bottom of the agar plate.
- 2. Use a sterile loop to remove a small amount of bacterial growth (from broth culture or a colony from solid media).

<u>Note:</u> If you are using a metal loop, sterilize it in the incinerator for 5 to 10 seconds before streaking each quadrant. Allow the loop to cool before streaking. You can make sure the loop is cool by touching an uninoculated area of the agar.

- 3. Inoculate the first quadrant by making a one-inch streak down, then streaking back and forth across this inoculum.
- 4. Turn the plate a quarter turn.
- 5. Use a sterile loop to streak the second quadrant by going through the edge of the first quadrant approximately four times and then continue streaking the second quadrant without going back into the first quadrant again.
- 6. Turn the plate another quarter turn.
- 7. Use a sterile loop and repeat the process above but only go into the second quadrant two or three times and complete the third quadrant streak.
- 8. Turn plate another quarter turn.
- 9. Use a sterile loop and streak the fourth quadrant the same as the third but let streaks get smaller and trail off.
- 10. After streaking the culture plates, you should invert the plates and incubate at the required temperature.

Notes:		
		Mentor/Supervisor /Date

Laboratory Exercises III

Objectives:

After completing this laboratory exercise, the participant will be able to:

• Identify commonly encountered problems with culture media and isolation techniques.

Exercise: Common Problems

You will examine several culture media plates given to you by your supervisor/mentor. Observe each culture plate for possible problems. Report the problem to your supervisor/mentor and discuss with him/her what should be done to fix the problem.

Notes:		
		Mentor/Supervisor /Date