

Biomanufacturing Module 2

Lesson 1 – Bacterial Inoculation

Lesson objectives:

Students will understand:

- The importance of aseptic technique.
- How to perform aseptic technique.

Essential Question

- Why do you need to use aseptic technique in biomanufacturing?

Materials:

- Aseptic Technique slide deck
- Testing the Sterility of LB SOP
- Sterile bacterial spreaders (1 pack/team)
- LB media (1 microfuge tube of media/student)
- P200 Micropipette and tips
- Sharpie markers
- LB agar plates (1/student)
- Bacterial plate incubator
- Aseptic Technique Check Document

What Students Will Do

- Listen to/watch the Aseptic Technique slide deck
- Prepare themselves and their lab benches to perform aseptic technique
- Follow the Testing the Sterility of LB SOP as a method of practicing aseptic technique
- Each team fills out their Aseptic Technique Check Document

Teacher Preparation

- Prior to class make copies of
 - Testing the Sterility of LB SOP (one per team)
 - Aseptic Technique Check Document (one per team)
- Prior to class turn on the bacterial incubator to a temperature of 37 degrees Celsius.
- Prior to class remove the aliquots of LB media from the refrigerator and let them warm to room temperature.
- Prior to class remove the LB agar plates from the refrigerator and let them warm to room temperature.
- Provide each team
 - One package of sterile bacterial spreaders
 - One p200 micropipette
 - One box of p200 micropipette tips
 - Sharpie marker for labeling plates
 - One microfuge tube of LB per student
 - Five LB agar plates
 - Spray bottle of 70% ethanol

- Paper towels
- Team file folders

Organizer

Time	Activity	Materials
10 minutes	Present the Aseptic Technique slide deck	Slide deck
5 minutes	Members of all teams put on PPE	Lab coats, gloves, safety goggles
5 minutes	Teams sanitize and prepare their bench space	70% ethanol, paper towels, sterile LB media, sterile LB agar plates, micropipette, tips, sterile spreaders
20 minutes	Each team member practices aseptic technique by spreading sterile LB media on an LB agar plate.	Testing the Sterility of LB SOP, sterile LB media, sterile LB agar plates, micropipette, tips, sterile spreaders
5 minutes	Each member of the team labels their plate and places it in the bacterial incubator. The plates incubate overnight and can then be stored in the refrigerator until the next class period.	Sharpie markers Bacterial incubator
5 minutes – NEXT CLASS	Each student puts on gloves and removes their plate from the incubator	
10 minutes	Each student counts the colonies on their plate. Teams fill out the Aseptic Technique Check Document and file it	Aseptic Technique Check Document, Team File Folder

Procedure

Introduction to Aseptic Technique

1. Present the Aseptic Technique slide deck

Preparation of the staff and lab benches

2. Members of each team put on PPE
3. Each team sanitizes and organizes their bench space
4. Each team member follows the Testing the Sterility of LB SOP to plate sterile LB onto an LB agar plate.

5. Each team member puts their plate in the bacterial incubator to incubate at 37 degrees Celsius for 24 hours. After 24 hours in the incubator, plates can be stored in the refrigerator until the next class period.

Data Analysis – Next class period

6. Each team member puts on gloves and retrieves their plate from the incubator.
7. Each team member counts the colonies on their plate
8. Each team fills out their section of the Aseptic Technique Check document.
9. The completed Aseptic Technique Check document is filed in the team file.