

Background

Microorganisms are plentiful and widespread in the environment. Like any other environment, many microorganisms can be found in the laboratory either floating in the air or on different surfaces. Armed with this information, microbiologists must practice techniques to prevent **contamination**. To contaminate is to accidentally grow an unwanted organism. So, the technique used to prevent contamination is called the **aseptic technique**.

The purpose of this laboratory exercise is to purposely contaminate to show that microorganisms are found everywhere. We will also learn how to describe colonial morphology of colonies growing on nutrient agar plates and to describe growth in nutrient broths.

Protocol

First Lab Period

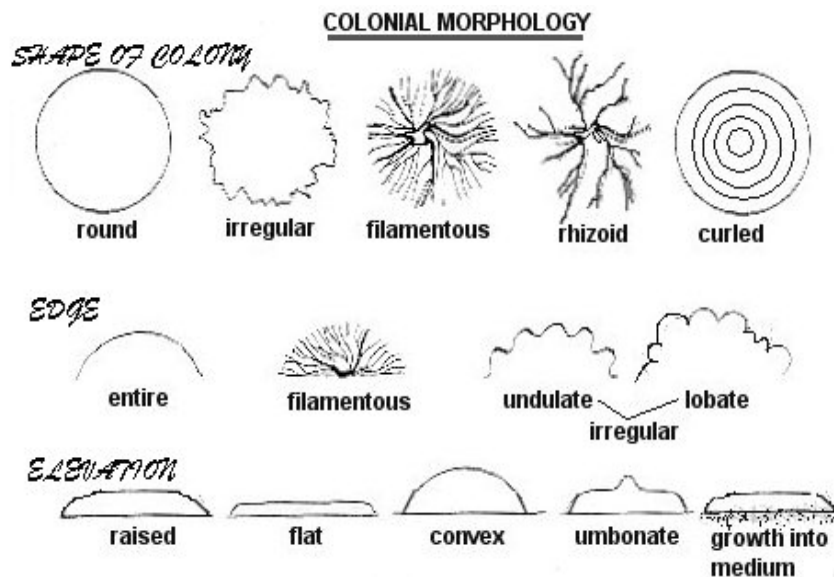
1. Break up into groups of two to three students. Each group will be designing their own experiments.
2. Each group should retrieve three nutrient agar plates, two nutrient broths, two sterile saline, and four cotton swabs. If working in groups of threes then retrieve four nutrient agar plates, three nutrient broths, three sterile saline and six cotton swabs.
3. Each group will label a nutrient agar plate “AIR” on the bottom (NOT THE LID). Be sure to also put group initials, today’s date, and instructor’s last name (since there are multiple sections). Remove the lid from the nutrient agar plate and leave it open to the air for 30 to 60 minutes. Upon exposure of air to a sterile nutrient-agar plate or **inoculation**, colonies of microorganisms should appear along the surface of the agar.
4. Each group member should have one plate, one broth and two cotton swabs. The nutrient agar plate will be used to sample the environment (ex. Coke machine, washroom, or any area in the Business and Health Sciences Building). The nutrient broth will be used to sample your body or personal belongings. Label the plate and broth with location you sampled, your initials, today’s date, and instructor’s last name.
5. Using the cotton swab, inoculate the plate with a sample from the environment by swabbing back and forth all over the plate along the surface of the agar gently. Do not cut into the agar medium surface.
6. Using the cotton swab, inoculate the broth with location you sampled by placing the cotton swab into the broth and leaving it there during incubation. After incubation, the liquid media will appear **turbid** or cloudy.
7. Place the plates upside down and place them in your lab drawer at room temperature or in the incubator along with the broths. Placing plates

BIOL 2250L – Microbiology for the Health Sciences
Lab Report – Microorganisms in the Environment

upside down will prevent condensation from falling back onto the plates as they incubate.

Second Lab Period

1. Retrieve plates and broths from incubator.
2. Sketch and describe the resulting growth on the plates using the figure below.
3. Describe the appearance of the nutrient broths. Is it turbid or cloudy? Is it **flocculant** (clumps of cells), **pellicle** (film across the surface of the broth) or **sediment** (settled cells at bottom of tube)?
4. Record your results in the lab report.
5. Pick ONE of the plates or broths you would like to stain and view under the microscope. Once you choose, please give to instructor to store for later use. Discard the remaining plates and broths properly by placing them into the biohazard.
6. Observe the fungal and bacterial colonial morphology plates on display. Record the results in the lab report so you can identify the different types of fungi and distinguish bacterial colonial morphology from fungal colonial morphology.



BIOL 2250L – Microbiology for the Health Sciences
 Lab Report – Microorganisms in the Environment

Name: _____ Lab Section: _____

Part A – Observing Agar Plates:

1. Examine the plates and count the number of colonies on each. In the chart provided, record the number of colonies observed.
2. In the chart, describe the appearance of these colonies according to the descriptive terms for colonial morphology that were given at the end of the handout.
3. Draw sketches of colonies from each plate to illustrate colonial types.

| Number and Appearance of Colonies on Nutrient Agar | | | |
|--|-----------------------------|------------------------------|-------------------------|
| Specimen | Number of colonies on plate | Word description of colonies | Sketch of some colonies |
| Air | | | |
| Environment | | | |
| Temperature of incubation: | | Days of incubation: | |

Part B – Observing Nutrient Broths

| Inoculated Broth | Turbidity | Appearance | Color |
|----------------------------|-----------|---------------------|-------|
| Area sampled: | | | |
| Temperature of incubation: | | Days of incubation: | |

Part C – Colonial Morphology of Fungi and Bacteria

| Specimen | Color of colonies | Word description of colonies | Sketch of some colonies |
|----------------------|-------------------|------------------------------|-------------------------|
| Penicillium | | | |
| Rhizopus | | | |
| Bacteria #1 Name: | | | |
| Bacteria #2 Name: | | | |

Part D – Questions

1. Why are Petri plates inverted during incubation?
2. What are some steps that could be taken to keep down contamination in the laboratory?
3. What are two ways to distinguish mold from bacteria?