

Name: _____ Date: _____ Class: _____

Student Lab Sheet Part 1

Question: How does the presence or absence of oxygen affect the growth of *Escherichia coli*?

Your group will grow two different bacterial cultures of *E. coli*, but all bacteria will come from the same petri dish.

Both colonies will be grown in the same nutrient-rich medium for microorganisms (LB Broth) that bacteria need to grow. One colony will be grown aerobically (in the presence of oxygen) and the other anaerobically (without any oxygen). After a day, you will analyze the cultures to check for differences in size.



Write down your hypothesis below detailing what you think will happen to the bacteria grown aerobically versus the bacteria grown anaerobically.

Hypothesis:

PART 1 – CELL PRE-GROWTH SET UP

Materials:

Each group needs:

- One 1000 μL micropipette
- One 1000 μL micropipette tip (sterile)
- Two 15 mL culture tubes (sterile)
- Two 1 μL tip inoculating loops (sterile)
- One test tube rack
- 1.5 ml of LB Broth
- Fine-tip labeling marker

pay attention to the units of measure! 1000 μL = 1 mL

To share with the entire class:

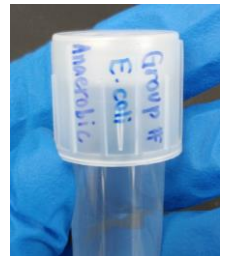
- One petri dish of *Escherichia coli*
- 1000 ml re-sealable jar with lid
- One test tube rack
- One tabletop shaker
- One 150 ml beaker
- Electric tape
- Tea candle
- One long-reach lighter

Safety Reminders:

- Long pants and close-toed shoes are required.
- Goggles, gloves, and aprons must be worn at all times.
- When working with bacteria, be careful not to touch sterile materials to prevent contamination of the experiment.
- Be sure to dispose of all materials as directed.
- Be sure to remove your gloves correctly.

Procedure:

1. Put on protective equipment and retrieve necessary materials.
2. **Discuss in a group:** What do bacteria need to survive and thrive? Share your group's ideas with your neighbors.
3. Label the two tubes: Group # E. coli Aerobic and Group # E. coli Anaerobic.
4. Using one sterile pipette tip on the micropipette, transfer 1000 μ L of the LB broth into the **aerobic** culture tube.
5. Without touching the inside of the tube with anything other than the pipette tip, transfer 1000 μ L of the LB broth into the **anaerobic** culture tube using the same tip. *If you touch a different surface, dispose of the tip and ask your teacher for another one.*
6. Use one sterile inoculating loop to transfer a single colony from the petri dish to the **aerobic** tube.
7. Swirl the tip around in the liquid to transfer the colony to the solution and dispose of the tip when finished.
8. Repeat previous step with the other inoculating loop, the **anaerobic** tube, and another colony.
9. Swirl both cultures briskly to mix all contents. Be careful to avoid spills.
10. Take both culture tubes to your teacher. Place the labeled aerobic tube on the test tube rack that is secured on the tabletop shaker and the anaerobic tube in the anaerobic jar.
11. Once all groups are done, your teacher will light the tea candle and seal the jar using lid and electrical tape and turn on the shaker. Leave the bacteria cultures to grow for one day.



Data:

In the space below, write detailed observations of the culture color, smell, etc. and draw your set up.

Culture observations

Aerobic - _____

Anaerobic - _____

Set Up