## Chapter 3 - Growing cells

End of chapter review questions - Answer Key

## **Hands-on Exercise**

## 1. What is LB agar used for? What are the two important ingredients in LB agar?

LB agar is both the material you grow cells on and the nutrients/food that your cells consume. LB agar includes both LB broth, which includes sugars, minerals, vitamins that your cells use to grow, and agar which is a plant based carbohydrate that dissolves in water at high temperatures, but then gels at room temperature.

## 2. How do you know you have created your LB agar plates correctly?

Your plates solidify nicely at room temperature and they are transparent.

## 3. How can you confirm that you actually dipped your loop in cells before you start streaking?

Your loop should look slightly wet after dipping into the stab of cells.

### 4. What are the steps for inactivation?

Place your experimental waste that includes microorganisms, or other materials that have come into contact with microorganisms, into an inactivation bag. Make sure to open up tubes before placing them in the bag so that the bleach water can get into them. General paper waste like plastic bags or the paper/plastic packaging of loops can go into the normal garbage.

Once all of your waste has been added to the inactivation bag, add one bottle of bleach and then 4-6 bottles of warm tap water. The main outcome is to be able to submerge your waste in the bleach water. Zip it closed tightly.

Let your waste stand at room temperature for 24 hours or more so that the bleach inactivates the samples (kills microbes).

Over a toilet, clip a small hole in the corner of the inactivation bag so that the liquid can drain but the solid waste stays. Remember, there are some small items like tube lids! Once the liquid mostly drains you can flush the toilet and put the rest of the waste in the trash bin.

## 5. What is a plate?

A plate is a plastic container, also called a petri dish. Plates are often filled with LB agar and then cells are grown in them.

## **Fundamentals**

### 1. Where do K12 E. coli come from?

K12 E. coli are the most studied organism ever and were originally isolated from the feces of a human.

## 2. List three key deficiencies that K12 E. coli have compared to natural E. coli.

K12 E. coli have lost their O-Antigen in their slime layer, they no longer have a F-plasmid, and they no longer are infected with a phage. This deficiencies make K12 E. coli highly susceptible to dying in the environment, unable to communicate well with other bacteria, and do not get killed during experiments because of the phage.

# 3. What are the five key locations in an E. coli cell and how do they relate to locations in a factory?

Slime layer == Fenceline
Outer membrane == Outer brick wall
Intermembrane space = Lobby
Inner membrane = Inner walls
Cytoplasm = factory floor

#### 4. Describe what CHOPNS is.

Carbon, hydrogen, oxygen, phosphorus, nitrogen, and sulfur. This are organic elements (atoms) that are highly used in organisms. They are often called organic elements.

# 5. What three macromolecules were described in this chapter? What atoms are each made of?

Lipids (CHOPNS) Carbohydrates (CHO) Proteins (CHONS)