The Effect of Antibiotics on Bacterial Growth

Pre-Lab:
Chemical substances that either kill bacteria or inhibit bacterial growth are called **ANTIMICROIAL AGENTS**. Antimicrobial agents are of three basic types: **ANTISEPTICS** which are chemicals used to inhibit the growth of or kill bacteria on living tissues, **DISINFECTANTS** are chemicals used to inhibit growth of or kill bacteria on non-living things and, **ANTIBIOTICS** which are chemical substances produced by living organisms which inhibit the growth of bacteria. The procedure used in this lab is similar to one used in hospital diagnostic laboratories to determine what chemical or antibiotic should be used to treat most effectively a particular bacterial disease.

**Purpose:**
To test the effectiveness of certain antiseptics, disinfectants, and antibiotics in inhibiting bacterial growth.

**Hypothesis:**
The _________________________ (antiseptic, disinfectants, anitbiotics) will have the greatest effect on bacterial growth.

**Materials:** 2 petri plates with agar, 10% bleach solution, marking pen, tape, alcohol burner, bacteria, disinfectants/antiseptics, test tube rack, filter paper, forceps, ruler

**Safety:** handle bacterial cultures safely by observing all aseptic transfer techniques

**Procedure:**
1) Obtain two petri plates.
2) Divide each plate into four equal quadrants. Use a marker on the agar side to mark and label it with your group names

**Preparing a Bacterial Lawn**
1) Choose one type of bacteria. Label each plate with the name of the bacterium you are going to use.
2) Pour approximately ______mL or bacteria onto the agar plate.
3) Swirl the plate so the liquid distributes evenly over the agar. Let it stand for 10-15 minutes.
4) Drain any excess liquid into the bleach container.
5) Repeat the same procedure using the same bacterial culture for the second plate.

**Plate 1: Disinfectants and Antiseptics**
1) You will place one disk in each of the four quadrants. In one of the quadrants you will place a disk that has been soaked in distilled water. Choose which three other disinfectants/antiseptics you wish to test.
2) Label each quadrant (on the agar side of the plate) with the disinfectant/antiseptic you have chosen to test.
3) Sterilize your forceps by flaming them and allowing them to cool.
4) Pick up a disk using the forceps.
5) Gently shake off any excess liquid
6) Position the disk in the center of the appropriate quadrant. Gently tap the disk with your forceps to make sure it sticks to the agar.
7) Repeat steps 3-6 for each quadrant.
8) When the four disks are in place, invert the plate and tape the edges with scotch tape.

**Plate 2: Antibiotics**
1) Using forceps, distribute evenly four different antibiotic disks using the same procedure as plate 1.

Place both plates, inverted (agar side-up), into the incubator. Observe the plates daily and record the presence of any clear areas associated with the disks on the data table in your lab report. Use a ruler to measure the DIAMETER of any clear areas present.

**Observations:**

<table>
<thead>
<tr>
<th>Plate Quadrant</th>
<th>Name of Disinfectant/Antiseptic</th>
<th>Zone of Inhibition for:</th>
</tr>
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<tbody>
<tr>
<td>1</td>
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<table>
<thead>
<tr>
<th>Plate Quadrant</th>
<th>Name of Antibiotic</th>
<th>Zone of Inhibition for:</th>
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Discussion Questions:
1. Explain the purpose of the disk with the distilled water.
2. How do you know that any inhibition you have observed is due to the disinfectants on the disks?
3. Which disinfectant was most effective in preventing the growth of your bacteria?
4. What is an antibiotic?
5. *Escherichia coli* is normally present in the human intestines. Does the sensitivity of this bacterium to the antibiotics suggest to you one of the reasons why doctors advise that antibiotics be used only when necessary?
6. Why did you have to flame the forceps?
7. Penicillin works by interfering with a bacterium’s ability to build a cell wall. Using this information, explain why Gram-positive bacteria are more susceptible to destruction by penicillin than Gram-negative bacteria are. (Hint: Review the structural difference between Gram-positive and Gram-negative bacteria in your text.)
8. ________ is a bacteria that lives in your large intestine. This is an example of ________ in which both organisms benefit. Explain how each organism benefits the other organism.
9. Why is using antibacterial soaps and detergents not necessarily beneficial?
10. Are all bacteria pathogens? Why or why not?
11. How are most bacterial infections transmitted?
12. Name 3 ways to prevent yourself from getting ill from bacteria.

Conclusion: The _________________________ was the most effective. This _________________________ my hypothesis.

(support/did not support)