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Antibiotic Effects Kit

Student Worksheet

The use of antibiotics has revolutionized clinical medicine. Antibiotics allow modern doctors to treat a far wider range of diseases more effectively and economically. By definition, an antibiotic is a biochemical produced by a microorganism that inhibits the growth of, or kills, another microorganism. One way to test the effectiveness of an antibiotic against a specific microorganism is the Bauer-Kirby test which measures the degree of inhibition produced by antibiotic disks (disks which contain a known amount of antibiotic) when placed on an agar dish swabbed with the desired microorganism. The antibiotic disks produce zones of inhibition (clear areas of no growth) which are measured in order to determine the susceptibility of the microorganism to the different antibiotics used in the test. You are using a newly developed substitute for the outdated Agar dish that is a product invented and developed by Dr. Jonathan Roth of Roth Bioscience, LLC, that is trademarked R-CARD®.

Day 1 of the experiment:

1. Your group will be provided with 3 R-CARD®, 3 bottles of bacterial cultures (A=Enterococcus, B=Bacillus, C= E.coli), and 3 sterile 1 mL droppers.

2. Write your name or initials, and the date on the bottom lines of the R-CARDs. Then write the name of the culture with which each dish will be inoculated (R-CARD® #1 = Enterococcus, R-CARD® #2 = Bacillus, R-CARD® #3 = E.coli).

3. As instructed by your teacher, add 1 mL Swirl each bottle of bacteria/water and add 1 mL of the mix from bottle #1 onto the R-CARD® #1 and lower the top piece so that the fluid spreads to cover the antibiotic containing disks. Repeat with bottle and R-CARD®sets #2 and #3.

4. Once the R-CARD® are inoculated do not pick them off the counter until instructed to do so by your teacher. If the R-CARD® need to be moved, they can be gently slid to a new location.

5.When instructed pick up the R-CARD® and move them to an Incubator or a warm, dark place. Try to incubate the R-CARD® at 25-35º Celsius for 24 hours.

6. When finished, Wash your hands using soap and warm water to assure that they have not become contaminated with any bacteria during your work

R-CARD® Diagram chart day 1

Day 2 of the experiment:

Measure the diameter of the zones of inhibition in millimeters by placing the ruler on the top of the RCARD®. Record results in Chart I. The tops of the R-CARD® should not be opened under any circumstances except with your Instructor's permission.

Study Questions:

1. Which antibiotic seems to be most effective in inhibiting E.coli? Which antibiotic seems to be least effective? Justify your choices.

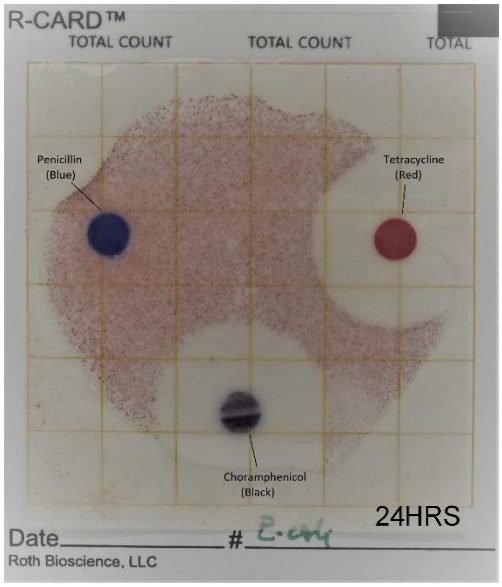
2. Which organism was penicillin most effective against? Least effective against? How can you explain this difference?

3. If the zones of inhibition of two antibiotic disks (A and B) on an Enterococcus dish measure 17 and 18 mm respectively, which antibiotic is more effective against the Enterococcus? Why?

4. Do the antibiotics kill the bacteria or only inhibit the growth? With your Instructor's help, design a method to determine whether the antibiotics are bacteriostatic (inhibit) or bactericidal (kill).

5. If the antibiotic concentration is doubled, will the growth zone be twice as large? Explain.

6. A doctor is prescribing medicine for a person with a systemic E. coli infection (a systemic infection is disseminated throughout the body by the circulatory system). Which antibiotic might a doctor choose?



R-CARD® Diagram chart day 2: Zones of inhibition (diameter in mm)

Organism	Chloramphenicol	Penicillin	Tetracycline
Escherichia coli			
Bacillus cereus			
Enterococcus faecalis			