Effects of Disinfectants

**Materials:**
- 2- Tryptic Soy Agar plates (or Nutrient Agar, whichever is available)
- Sterile Filter Paper disks
- 2- Sterile Cotton Swabs
- Forceps
- Broth Cultures Of E. Coli, S. Aureus
- 10% Bleach
- 10% Lysol
- Bleach, Full Strength
- Lysol, Full Strength
- Listerine Mouthwash
- Isopropyl Alcohol
- Other disinfectants as provided
- Ethanol for sterilization
- Sharpie
- Bunsen burner
- Striker

**Procedures:**

1. Disinfect your work area using the 10% Lysol in the spray bottle on your table. Obtain your materials, and light your Bunsen burner.

2. Using a sterile swab, inoculate one of your plates for confluent growth with E. coli. Using a fresh plate and swab, repeat the inoculation with the S. aureus. Remember to label the bottom of each plate with the name of the organism you used to inoculate that plate.

3. Using your Sharpie, divide the bottom of the plates into quadrants. Choose four disinfectants to test. **Use the same four disinfectants on both plates.** Label each quadrant of each plate in such a way that you will know which disinfectant you are testing in that quadrant.

4. Dip the tips of your forceps into the ethanol and allow them to sit for 30 seconds. Then, leaving them pointed tip-down, pass them through the flame of the Bunsen burner. This will sterilize them. Once they have been sterilized, do not touch the forceps’ tips to anything. If the tips touch anything, you will need to resterilize them.

5. Using your sterile forceps, pick up a sterile filter paper disk.

6. Without touching anything except the disinfectant, dip the disk into the disinfectant. If the paper or the forceps’ tips touch anything except the disinfectant solution, you will need to discard the paper and resterilize your forceps.

7. Allow the excess disinfectant to drain off the paper disk. You can do this by holding the disk against the side of the jar in which the disinfectant is stored.

8. Carefully place the disk in the center of the appropriate quadrant of the plate inoculated with E. coli. Do not drag the disk around the quadrant, and avoid dripping excess disinfectant on the plate. Gently press the disk onto the agar.

9. Using the same disinfectant, repeat steps 4-8 on the plate inoculated with S. aureus.

10. Repeat steps 4-9 using three other disinfectants.

11. When you have placed disinfectant disks on all four quadrants of both plates, tape the plates together, invert them, label them, and place them on the tray your instructor has provided. The plates will be incubated at 37°C for 48 hours.