## Adventure Description:

STEM CAREER

**ADVENTURES** 

In this adventure, students will think like a microbiologist and conduct an experiment to determine how varying concentrations of silver nanoparticles affect bacterial growth.

# Activity

Teacher Notes:

- Divide the silver into 50 mL quantities for each group of students.
- Reconstitute the E.Coli and have vials prepared for students to use. Students can share the vials.

### Step 1: Background Information on Microbiologists and Antimicrobial Research (5–10 minutes)

- Explain to students that a microbiologist is a scientist who conducts research on microorganisms, such as bacteria. Bacteria can be both good (like the bacteria in your intestines that help your digest your food) and bad (like bacteria that infect wounds).
- Discuss how microbiologists conduct research on microorganisms at a cellular level. By looking at organisms at a cellular level, microbiologists develop methods to kill or prevent the bacteria from harming other organisms, like humans and animals.
- One way that microbiologists prevent bacteria growth is with antibiotics. Antibiotics target bacteria by attacking cell parts that only bacteria have and human cells don't. Many antibiotics make it so that no cell can create a cell wall. Human cells don't have a cell wall, so this doesn't impact them, but many bacteria rely on a cell wall, so without it, they are easier to kill. Show Handout: Antimicrobial Agents. As a class, discuss how antimicrobial agents kill microbes and specifically how silver nanoparticles kill bacteria.
- Next, tell students that some microbiologists conduct research on the effectiveness of different concentrations of silver nanoparticles. Show Handout: Silver Nanoparticle Research.

### Step 2: Activity Set Up (5 minutes)

- Explain to students that they will design an experiment to test the effects of different concentrations of silver nanoparticles on bacteria growth.
- Show Handout: Overview of Experiment. As a class, read through the handout.
- Next, provide students with Handout: Designing an Experiment. Explain to students that you will tell them when to move on to each step. They should not move on to a new step until you tell them to do so.
- Divide students into groups or pairs. Students can also work individually.

#### Step 3: Experimental Design and Calculating Concentrations (20 minutes)

- Explain to students that they will first design their experiments and calculate the silver nanoparticle concentrations they will use.
- Instruct students to complete steps 1 and 2.
- Teacher note: Students should use information on the handouts provided in the first step to create their hypothesis. You can print these handouts for students or upload them to a Google Drive folder for them to view (if you want to upload them to a Google Drive folder, download the PDFs from the Rozzy portal and then upload them).

• As students are working, discuss the following:

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- Why do bacteria need to be placed on agar plates? (Bacteria are living organisms. They need a food source to live. The agar plates provide a food source).
- Do you think one extra drop of silver will affect the concentration of your solutions?
- Why will you have an agar plate with only bacteria and no silver nanoparticles? (The agar plate without the silver nanoparticles is the control. You will use this plate for comparison to the other agar plates).
- What could happen if you didn't have a control?

#### Step 4: Mixing Silver Nanoparticle Solutions (10 minutes)

- Explain to students that they will now mix the silver nanoparticle concentrations that they calculated in the last step.
- Provide students with the following materials:
  - Nitrile gloves
  - Safety goggles
  - 4 glass jars or beakers
  - Tape and a marker for labeling
  - 25 mL graduated cylinder
  - Plastic graduated syringe 50mL of colloidal silver (550 PPM)
  - Tweezers
  - 1 Bottle of distilled water (100mL minimum)
  - Small amount of isopropyl alcohol
- Have students complete step 3 on the handout.

#### Step 5: Inoculating Agar Plates (10 minutes)

- Explain to students that they will now inoculate the agar plates with bacteria. Tell students that inoculation refers to the placement of bacteria in a uniform pattern, making sure that bacteria are spread out on the surface of the agar.
- Provide students with the following materials:
  - 3 Sterile swabs
  - 12 Sterile paper disks
  - 4 Nutrient agar plates
  - Vial of E. Coli bacteria culture
- Have students complete step 4 on the handout.

#### Step 4: Mixing Silver Nanoparticle Solutions (10 minutes)

Explain to students that they will now add the silver nanoparticles to the agar plates.

- To do this, students will remove the small disks from glass jars where they mixed their silver concentrations. From the jar, the disks will be transferred to an agar plate. The agar plate and the glass jar should be labeled with the same concentration (i.e. if the concentration is 50,000 PPM, the disks from the 50,000 PPM jar should be placed on the 50,000 PPM agar plate).
- Have students complete Step 5 on the handout.

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- As students are working, discuss the following:
  - Why is it important to clean the tweezers between jars?
  - Why is it important that the disks be placed so that the edges are not touching?
  - How will we be able to tell if the silver nanoparticles are effective at inhibiting bacterial growth?
  - Do you think that the control will inhibit any bacterial growth?
  - Explain to students that they will now wait for at least 3 days to give the bacteria time to grow on the agar plate.
    - To expedite the bacteria growth, you can place the plates in an incubator, which would allow for observations within 24 hours.
    - Plates should be stored beneath a fume hood, or at least away from the general student area.

### Step 7: Analyze Results and Draw Conclusions (20+ minutes)

- Teacher note: this step will be completed 3 days later. If you do not see students after 3 days, take photographs of their plates for them so that they can analyze their results when you see them again. It is possible for the bacteria to grow for a week, depending on how thoroughly it was inoculated.
- Explain to students that they will look at their agar plates and determine which concentration of silver nanoparticles was most effective at preventing bacteria growth.
- Provide students with the following materials:
  - Handout: Experiment Observations
  - Ruler
- Explain to students that they will first determine whether there is a ring of inhibition around the disks in their agar plates. If the silver solution inhibits bacterial growth, a ring of empty space will form around the disks on the agar plates. These are the rings of inhibition. The larger the ring, the more antibacterial the silver solution.
- Students will be measuring the size of the rings using a ruler. They will measure from the edge of the disk to the outer rim of the ring.
- While students are working, be sure to collect data from each group. Use Handout: Class Data to gather student data in one data table. You can use the class data sheet provided or make your own. We suggest making a copy of the chart in your google drive and giving students access to edit the class sheet or printing the sheet, and putting in a central location (like under a document camera) for students to fill in their own data as they finish. This data will be used by to analyze class patterns and trends on step 6 of Handout: Experiment Observations.



- For clean up:
  - Contact your local high school hazardous waste disposal depository to dispose of the petri dishes.
  - Follow your state guidelines for disposal of the silver nanoparticle solutions
- Extra Time? Have students swab the back of their phone and inoculate an agar plate.

#### **Materials List**

#### **Provided online:**

- Handout: Antimicrobial Agents
- Show Handout: Silver Nanoparticle Research
- Handout: Overview of Experiment
- Handout: Designing an Experiment
- Handout: Experiment Observations
- Handout: Class Data

### Not provided (Each group needs):

- Nitrile gloves
- Safety goggles
- 3 Sterile swaps
- 12 Sterile paper disks
- 4 Nutrient agar plates
- Vial of E. Coli bacteria culture (1 vial per 2-3 groups)
- Tweezers
- About 20 mL isopropyl alcohol
- Small cup for isopropyl alcohol
- 4 Small glass jars or beakers
- 100 mL Distilled water
- 50 mL Colloidal silver (550 PPM)
- Graduated plastic syringe (25mL or smaller)
- 25mL graduated cylinder or beaker
- Ruler
- Permanent marker
- Clear tape
- 70/30 bleach solution
  - Separate solution into small spray bottles for each student to use on lab tables