# Student Guide

## Creating and Testing of Silver Nanoparticle Socks

Name: ____________________________ Date: _______________Class:__________________

**Introduction:**

Nanoparticles are microscopic particles that have a width of about a few billionths of a meter ($10^{-9}$m). A nanometer is one billionth of a meter. These small particles often have different properties than their macro-size composition material. For example the color of elemental silver is gray, while silver nanoparticles are yellow/orange. In this lesson you will be taking advantage of the antibacterial properties of nanosilver to create your own antibacterial socks! Socks such as these are currently on the market and can be found in stores where you probably shop. You can also buy a silver nanoparticle spray to treat the socks you already own. After creating your own silver nanoparticles you will treat a pair of socks and compare their effectiveness against bacteria growing in petri dish. You will also be testing control socks that contain no nanosilver, socks treated with a commercial spray (Mesosilver), and commercially available nanosilver socks that were bought in a store.

**Purpose:**

- To study the characteristics and uses of nanoparticles
- To produce a sample of silver nanoparticles
- To examine the antibacterial properties of nanosilver particles
- To compare the effectiveness of the particles with commercial sock treatments

**Key Terms:**

**Nanoparticle:**

**Nanotechnology**:

**Synthesis**:

**Antibiotic**:

**Culture**:

**Zone of Inhibition:**



*Developed by Nathaniel Eugene, Douglas Schunk, and Stephen Stilianos*
*Development and distribution partially funded by the* ***National Science Foundation***

NNIN Document: NNIN-1281
Rev: 07/2012

**Environment**:

**Materials for Day 1:** (for groups of 2- 4)

- Goggles and nitrile gloves (lab aprons are optional)
- Glassware: 50 mL Erlenmeyer flask, 10 mL graduate cylinder, small vial to store colloid solution
- Distilled water
- 20 mL 0.001 M Silver nitrate solution
- 2 mL 0.003 M Sodium citrate solution
- Hotplate with stirrer
- Magnetic stir bar
- Two 5 cm X 5 cm pieces of normal socks
- Mesosilver Spray – 1 bottle shared amongst class
- One small vial (10 ml) for silver particle solution
- Oven safety mitt
- Aluminum foil

**Day 1 Procedure:**

1) Retrieve an Erlenmeyer flask, a 10 mL graduated cylinder, a magnetic stir bar, and a hot plate and bring them to your lab station.

2) Carefully place the magnetic stir bar in the flask. Measure out 20 mL of silver nitrate ($AgNO_3$) into your Erlenmeyer flask. ***Silver nitrate will stain your skin. Wear gloves and clean all spills immediately.**

3) Heat the silver nitrate solution on the hot plate on medium heat. The magnetic stirrer will mix the solution while it heats.

4) While the silver nitrate is heating, clean your graduated cylinder with soap and distilled water.

5) Measure out 2 mL of sodium citrate using the graduated cylinder.

6) Once the silver nitrate is at a rolling boil, add the 2 mL of sodium citrate solution.

7) Observe and record any changes that take place in the solution. Why do you believe these changes are occurring?

8) Five minutes after adding the sodium citrate, carefully remove the flask from the heat source with an oven-safe mitt and wait for the solution to cool.

9) Continue to watch for 2-3 minutes and record any changes that occur to the solution as it cools.

10) Once your solution is cooled, transfer the contents to your vial. Wrap the vials in aluminum foil to protect the solution from light.



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**Cleanup:** Ask your teacher about the disposal of chemicals. Solutions will be collected in a labeled container designated by your teacher.

**Analysis Questions:**

1. How many nanometers are in one meter?

2. Why did the colloid solution begin to change colors once the sodium citrate was added?

3. What are the potential benefits of wearing socks with silver nanoparticles in them?

4. Based on your scientific knowledge, make a prediction as to whether you think bacteria will grow in the presence of these four types of socks (*this will be done later in the lab).* Explain your reasons for your predictions below the table.

| TYPE OF SOCKS | PREDICTION |
|---|---|
| ***Regular Cotton Sock*** | |
| ***Mesosilver Sock*** | |
| ***Nanoparticle Sock*** | |
| ***Store Bought Silver Sock*** | |



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**<u>Materials for Day 2</u>:** (for groups of 2- 4)

- Goggles and nitrile gloves (lab aprons are optional)
- Four prepoured nutrient agar plates
- Bacteria (*Micrococcus* or *Staphylococcus epidermidis*)
- Ethanol
- L-shaped glass stirring rod (to spread the bacteria)
- 5 cm X 5 cm piece of nanosilver sock
- 5 cm X 5 cm piece of normal sock (NOT TREATED WITH SILVER)
- 5 cm X 5 cm piece of Mesosilver treated sock
- 5 cm X 5 cm piece of Silver particle sock
- Forceps / Tweezers
- Pipette for bacteria
- Biohazard waste container
- Bunsen burner
- Oven safety mitts
- Artificial sweat - optional

**<u>Day 2 Procedure</u>:**

1) Put on safety goggles, gloves, and tie any lose hair back. Set up a bunsen burner, and then obtain four agar plates and the bacteria. Label the plates as control, mesosilver, nanosilver, and store bought silver sock.

2) Use a pipette to pour 0.1 mL of bacteria into the center of each agar plate. Dip the glass stirring rod into a beaker of ethanol, and then pass it through the flame of the bunsen burner to sterilize. ***(Ethanol is highly flammable. Keep the part that was dipped in ethanol facing down)***

3) Allow the stirring rod to cool on the inside of the agar plate. Then swipe gently along the surface to spread the bacteria onto the entire plate. When the plate is covered, use the same stirring rod to coat the other three plates. When all four plates are covered, leave the stirring rod in a small beaker of ethanol to sterilize.

4) Sterilize your forceps by soaking it the ethanol and carefully heat it in the Bunsen burner. Be sure to repeat when using a new sock sample to avoid contaminating your samples.

5) Optional Sweat Procedure-Before placing each of the socks on the bacteria plate, dip them in an artificial sweat solution. This may be done at your teacher's discretion.

6) Soak a normal sock piece in distilled water and place on the bacteria plate labeled control.

7) Soak a piece of normal cotton sock in the mesosilver solution. Then place the sock on the bacteria plate labeled "mesosilver."

8) Soak a piece of normal cotton sock in the nanosilver solution you prepared. Then place the sock on the bacteria plate labeled "nanosilver."

9) Soak a piece of your store bought sock in distilled water and place it on the bacteria plate labeled "store bought silver sock."



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10) The samples should be placed in an oven at 37 degrees Celsius for 24 hours. During the next class you will examine the growth on the plates and determine which, if any, of the socks inhibited bacterial growth.

**Clean up:** Ethanol can be poured down the drain with excess water. All bacteria waste should be disposed of in a biohazard container and not in a regular garbage can.

**Analysis Questions:**

1. Why is it important to use a normal cotton sock as your control?

2. Why did you need to place the L-shaped glass stirring rod into ethanol and heat it prior to spreading the bacteria?

3. Why were *Micrococcus* or *Staphylococcus epidermidis* used as the bacteria for this testing?

**If you did the artificial sweat portion of the lab answer the following question**

4. Why would it be more appropriate to use artificial sweat instead of distilled water?



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**Materials for Day 3:** (for groups of 2- 4)

- Goggles and nitrile gloves (lab aprons are optional)
- Four agar plate samples with bacteria and socks
- Biohazard waste container

**Procedure for Day 3:**

1) Put on safety goggles and gloves. Retrieve your four agar plates and record any observations. Where was there bacteria growth? Did the bacteria cover the entire plate? Were there any sections that did not have bacteria growth? For the following table if there was a zone of inhibition write in "yes" and if there was no noticeable zone of inhibition write in "no". The zone can be very thin.

| TYPE OF SOCKS | RESULTS |
|---|---|
| ***Regular Cotton Sock*** | |
| ***Mesosilver Sock*** | |
| ***Nanoparticle Sock*** | |
| ***Store Bought Silver Sock*** | |

2) Record your results on the blackboard including your group name, and what was observed on each of the four agar plates. The plates should be labeled control, Mesosilver, nanosilver, and store bought silver sock.

3) When you are finished with your plates, they should be disposed of in a biohazard container (NOT a regular garbage can). Clean up your lab area and then return to your seats for a discussion of the results

4) As a class, discuss and answer the following questions:

Which pair of socks was most effective in preventing bacterial growth? Why do you believe there was a difference if they all contained silver nanoparticles?

Was there any growth that looked different from the rest of the bacteria? If it was different, where did it come from?



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NNIN Document: NNIN-1281
Rev: 07/2012

5) Your teacher will lead a short discussion on the potential damage nanoparticles can cause in the environment. Think about whether you feel nanotechnology research should be banned, regulated, or left alone. Are the potential rewards worth the risk? After the discussion, read and work on the following prompt:

| PROS OF NANOTECHNOLOGY | CONS OF NANOTECHNOLOGY |
|---|---|
| | |
| | |
| | |
| | |

**Analysis Questions and Activities:**

1) According to the makers of Mesosilver, you should reapply the spray after each wash. Why do you need to reapply? What do you believe happened to the silver nanoparticles? Do you believe store bought nanosocks also need silver nanoparticles to be reapplied for them to continue to work? Explain.

**2) Just when you thought it was safe to go back in the water...**

As a responsible chemist who knows that nanosilver can be harmful for the environment, it is your responsibility to find a way to test how much silver is removed from your nanosocks when washed. For exact measurements there are precise detectors, but they are also very expensive. Your group's task is to design an experiment to test whether or not nanosilver is removed from your socks when washed. The measurements can be qualitative, so complicated calculations are not necessary. You may use resources, including the Internet, but your activity must be original. If the procedure is detailed and has potential for success, your teacher may allow you to perform the experiment with the class.



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## Extension Activity (optional for the teacher to assign)

**3) Show me the money!**

"Many a small thing has been made large by the right kind of advertising."
Mark Twain

You have successfully created silver nanoparticles, and used them to inhibit bacterial growth on socks. Now you must find a way to market your socks to the public so that you can pay back your investors. For this project you can create a magazine, radio, or television advertisement or infomercial. Other advertising methods are valid as well, but make sure your teacher checks your project idea first. This is your chance to really be creative, so make it count. You should be aware of the following:

Who is your target audience? Are you trying to sell the socks to teenagers such as yourselves, or adults? What kind of balance do you need between the science of how they are made and how they work versus how awesome and cool they are? Would they be great for sports, hunting, or just lounging around the house? What price would you sell them at and why? Use the internet to research prices of nanosocks that are on the market.. Are there any other product lines you might consider and why (use your imagination)? Do you think you will be facing a very competitive market in the long run? Also think about medical conditions where the socks may be beneficial. You do not need to market your socks to everyone, but you can.

[1]This activity is an extension of the lab "Study of Silver Nanoparticles" created by a 2009 MRSEC RET group at Penn State University. During that lab, students created silver nanoparticles and used sugar and salt to formulate an understanding of the interactions between the particles.



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