Teacher's Guide

Spectrophotometry with Metal Nanoparticles

Grade Level: High school

Subject area(s): Chemistry; Physics

Time required: (2) 50 minute classes

Learning objectives: Through hands-on activity understand: 1. relationship between energy and frequency of light; 2. Matter may behave differently at the nanoscale.

Summary: This activity has students chemically synthesize gold and silver nanoparticles, measure the spectra of light absorption and transmission by these particles, compare these spectra to those of known-sized particles, and then estimate the size distribution of the particles they have made. By using the interaction of light with their nanoparticles, students will measure the size of particles much smaller than the smallest things they can see with an optical microscope. This lab is designed to help students gain an appreciation of the unique ways that matter may behave at the nanoscale. Students will also gain understanding of the relationship between energy and the frequency of light, and in the use of a spectrophotometer as a means of analyzing particle suspensions of different concentrations. The first class period should be used to introduce students to nanotechnology and quantum dots, and to synthesize the gold and silver nanoparticle samples. The second part of this activity will be used to conduct the spectrophotometry, collect data, and analyze the results. If the teacher does not have access to a spectrophotometer, one can be accessed at the Remotely Accessible Instruments for Nanotechnology (RAIN) that allows educators to freely and remotely access instruments including spectrophotometers (https://nano4me.org/remoteaccess).

Background Information: Nanoparticles are small collections of matter that range in size from 1 to 100 nm. For comparison, atomic radii are on the order of 0.1 nanometers and a water molecule is approximately 0.2 nanometers in width. Nanoparticles may be composed of metals, metal oxides, carbon, polymers, semiconductor material--virtually any solid element or compound may be made or used in nanoparticle form.

Certain types of nanoparticles are being used in a variety of practical applications. For example, nanoparticles of semiconductor materials, known as quantum dots, show interesting and useful optical properties when certain wavelengths of light are incident upon them. These nanoparticles emit light at a wavelength that depends on their size and shape. This property has led to their use in solar cells, light emitting diodes (LEDs), electronic displays, and medical imaging.

Nanoparticles made of the noble metals, particularly gold and silver, also exhibit a size-dependent interaction with light. Such size dependence can be harnessed as a tool to

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characterize nanoparticles made in the lab, allowing students to probe matter at the nanoscale using fairly simple lab tools.

Electromagnetic radiation refers to the energy released by an atom when one of its electrons returns to its ground state from an excited state. In the early 1900's it was found that this energy is directly related to the frequency of the radiation by

$$E = hf \qquad (1)$$

where E is the energy in Joules, h = 6.626 x 10^{-34} J·s (Planck's constant), and f is the frequency in Hertz. The frequency of this light is directly related to wavelength by

$$c = \lambda f \qquad (2)$$

where c is the speed of light (3.00 x 10^8 m/s) and λ is the wavelength in meters.

Electromagnetic radiation is arranged in order based on the frequency (or, inversely, the wavelength) of the radiation and is often shown in the **electromagnetic spectrum**.

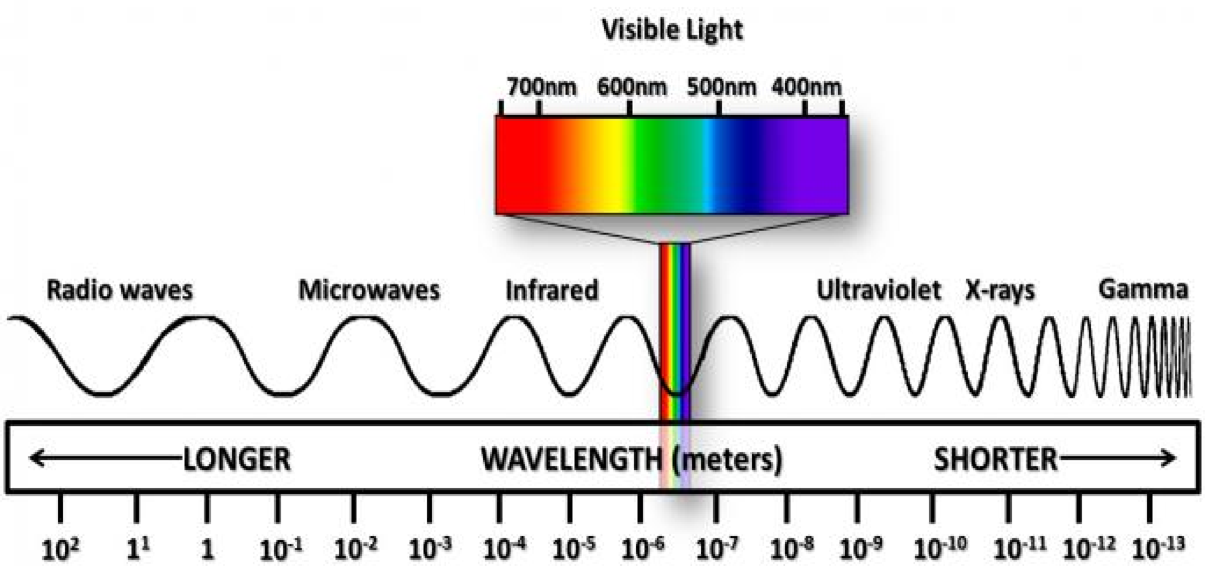

Figure 1. The electromagnetic spectrum is shown with higher frequency waves on the right. Image from Climate Science Investigations, http://www.ces.fau.edu/nasa/module-2/radiation-sun.php

Using Equation 1 with Fig. 1, it can be seen that the short wavelength radiation on the right side of the spectrum (i.e., x-rays and gamma rays) has more energy than the longer wavelength radio waves on the left side. Also from Figure 1, in a very small section of the electromagnetic spectrum is visible light, often called the **visible spectrum**. It can be seen in this section that blue and violet light has greater energy than does red light.

Quantum dots are nanoparticles (usually less than 8-10 nm in diameter) made of semiconductor compounds, such as cadmium selenide, cadmium sulfide, and zinc sulfide. These

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nanoparticles have the unique property of emitting light at a frequency that is directly related to particle size[1,2]. For a given incident light, quantum dots will emit light at a very specific frequency, or color. Figure 2 below shows blue light incident on varying sizes of quantum dots and the color of light emitted. By controlling the size of quantum dots in a specific application, the emitted color is accurately obtained.

An interesting article on the use of quantum dots in solar cells can be found at http://nanooze.org/solar-cells-and-quantum-dots/ [3]. In addition, the NNCI.net education materials have several other lessons focused on QDs.

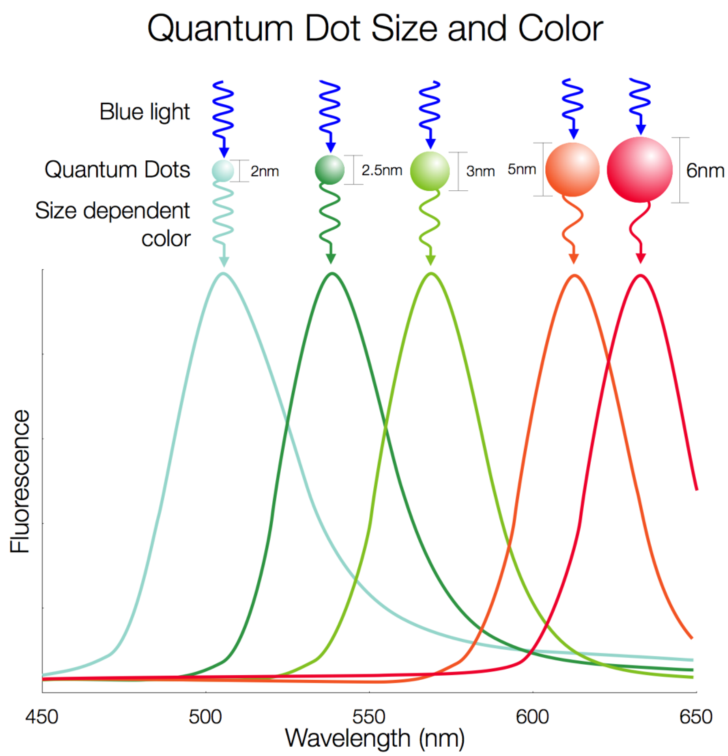

Figure 2. The color of light emitted from quantum dots is dependent on its size. Image courtesy of www.nanosync.com

While the size-dependent light absorption and emission is most pronounced in true quantum dots, other nanoparticles also exhibit such phenomena. In particular, the light absorbed by nanoparticles of silver and gold absorb will depend on the size of the nanoparticles. Since gold and silver nanoparticles are much easier to synthesize in the lab than quantum dots (or, alternatively, are cheaper to buy), they are used in this lab to demonstrate the light absorbing properties of nanoscale materials.

As part of the activity, students will use a **spectrophotometer** to analyze the light absorbed and transmitted by a suspension of nanoparticles. A typical spectrophotometer is shown in Figure 3. This device passes white light through a prism to break the light into its component parts. It can then adjust the prism in very small increments so that only a specific wavelength of light passes through an aperture which then strikes the sample to be analyzed. A detector then measures the amount of the light passing through the sample. Spectrophotometers allow different

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concentrations of solutions or suspensions to be analyzed. This is possible since, prior to any analysis, the spectrophotometer is calibrated between a sample allowing no light to pass and a sample allowing 100% of the incident light to pass[4,5]. If the class does not have access to a spectrophotometer, you may contact RAIN (https://nano4me.org/remoteaccess) which has three available for remote access. Each site will explain how to access and how to send samples for analysis.

In this activity, students synthesize samples of gold and silver nanoparticles suspended in an aqueous mixture, and then use a spectrophotometer to determine the absorption spectrum of each sample. They will then compare their measured spectrum to a set of absorption spectra collected from nanoparticles of the same material and known sizes. Students are challenged to determine the size of the nanoparticles they have made, based solely on the light absorption curves.

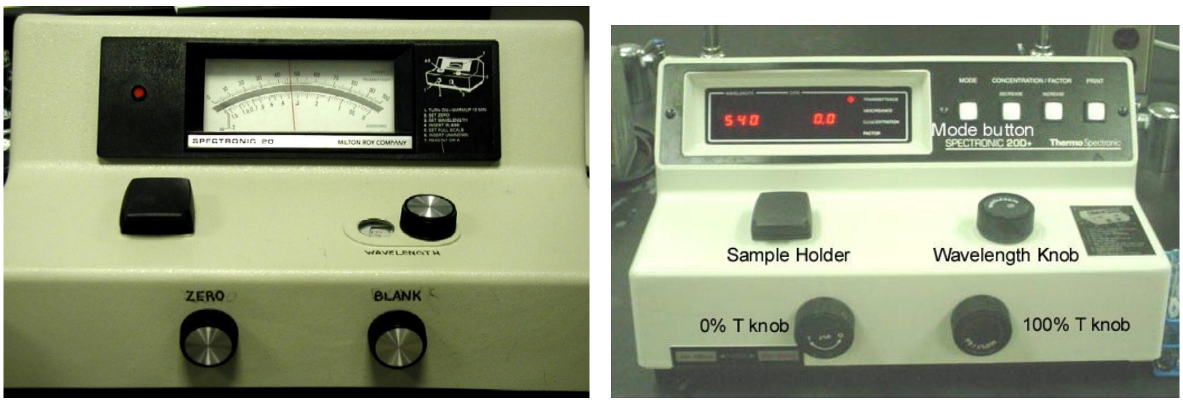

Figure 3. Typical spectrophotometers are shown. An analog version is shown on the left while a digital spectrophotometer is shown on the right.

Sources and resources:

1. Trusted Reviews, "Quantum Dots Explained: What are quantum dots and why are they so awesome?", Andy Vandervel, 2014, http://www.trustedreviews.com/opinions/quantum-dots-explained-what-they-are-and-why-they-re-awesome
2. Parker, A., "Mighty Small Dots." *Science and Technology Review,* July/August, 2000
3. Nanooze, "Solar Cells and Quantum", Lynn Charles Rathbun, November 2013, http://nanooze.org/solar-cells-and-quantum-dots/
4. "Spectrophotometer Use", David Fankhauser, University of Cincinnati Clermont College, 2007, http://biology.clc.uc.edu/fankhauser/labs/microbiology/Growth_Curve/Spectrophotometer.htm
5. "ChemPages, Spectronic 20", University of Wisconsin, http://chem.wisc.edu/deptfiles/genchem/lab/labdocs/modules/spec20/spec20desc.htm
6. Mutavdzic, D, et al., "Determination of the size of quantum dots by fluorescence spectroscopy, *Analyst*, April, 2011.

Resources:
7. Spectrophotometry at Libre Texts: https://chem.libretexts.org/Bookshelves/Physical_and_Theoretical_Chemistry_Textbook_Maps/Supplemental_Modules_(Physical_and_Theoretical_Chemistry)/Kinetics/Reaction_Rates/Experimental_Determination_of_Kinetcs/Spectrophotometry
8. Spectrophotometry at Wikipedia: https://en.wikipedia.org/wiki/Spectrophotometry
9. YouTube – various videos available on spectrophotometry: https://www.youtube.com/results?search_query=spectrophotometry
10. What are quantum dots? At NanoWerk: https://www.nanowerk.com/what_are_quantum_dots.php

Pre-requisite Knowledge: Students should understand atomic structure, electrical conduction, frequency of light, and fluorescence**.**

Materials: (per lab group)

- Stirring hot plate
- Stir bar I or 2 inch
- 50 and 100 mL Erlenmeyer flasks
- 25 mL and 50mL graduated cylinders
- 2mL dropper
- Laser pointer
- Spectrophotometer (typical specifications: wavelength range 340 – 950 nm at 5 nm increments, 0.100 %T, 110 VAC, 50/60 Hz) or RAIN remote instrument
- Cuvette for spectrophotometer
- Test tube rack
- Thin films of silver and gold nanoparticles (can make by trapping nanoparticles of silver and gold between small squares of acetate sheets or two glass slides)
- LED lights – UV, red, blue, green and white (Photon Light.com and Educational Innovations are good sources for these)

For production of silver nanoparticles:

- bowl with crushed or cubed ice
- 30 mL of 0.002M sodium borohydride ($NaBH_4$)
- 2 mL of 0.001M silver nitrate ($AgNO_3$)

For production of gold nanoparticles:

- 20 mL of 1.0M $HAuCl_4$
- 2 mL of a 1% solution of trisodium citrate dihydrate, $Na_3C_6H_5O_7 \bullet 2H_2O$
- 2 oz amber glass dropping bottle for storage

Safety Information: Read and review with the students the Safety Data Sheets for all the reagents used in the nanoparticle synthesis. Safety glasses and gloves should be worn during the nanoparticle synthesis.

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Vocabulary and Definitions:

- *Nanometer*: 10^{-9} meters or a billionth of a meter
- *Nanoscale:* objects typically within the range of 1 to 100nm but nanotechnology research is often with 100s of nanometers.
- *Electromagnetic energy:* a form of energy that is reflected or emitted from objects in the form of electrical and magnetic waves that can travel through space.
- *Frequency:* the number of occurrences of a repeating event per a unit of time; number of waves that pass a fixed point in a given amount of time is called a wave frequency.
- *Wavelength:* the spatial period of a periodic wave or the distance over which the wave's shape repeats. For example, it is the distance between consecutive corresponding points of the same phase on the wave such as the peak or trough.
- *Visible spectrum:* the portion of the electromagnetic spectrum that is visible to the human eye. Electromagnetic radiation in this range of wavelengths is called visible light or simply light. A typical human eye will respond to wavelengths from about 380 to 740 nanometers in terms of frequency, this corresponds to a band in the vicinity of 405–790 THz. (definition from Wikipedia: https://en.wikipedia.org/wiki/Visible_spectrum*Absorption*
- *Transmittance:* the ratio of the light energy falling on a body to that transmitted through it; it is how much light passes through a sample unchanged or light that is not absorbed, scattered, or reflected.
- *Spectrophotometer:* instrument that measures the amount of photons (intensity of light) absorbed after it passes through sample solution; it measures the intensity of electromagnetic energy at each wavelength of light in a specified region.

Advance Preparation:

1. Prepare stock solutions as directed in the MRSEC procedures in the links below. Store these solutions in a dark, cool place.

- https://education.mrsec.wisc.edu/synthesis-of-silver-nanoparticles-nabh4/
- https://education.mrsec.wisc.edu/citrate-synthesis-of-gold-nanoparticles/

Other sites with synthesis procedures:

- Gold: https://chemistry.beloit.edu/edetc/nanolab/gold/index3.html
- Silver: https://pubs.acs.org/doi/abs/10.1021/ed500036b

2. Make copies of the absorption spectra for silver and gold nanoparticles. A master is attached as Appendix.

3. Make thin films of gold and silver nanoparticles as noted in the materials section above. This will require the teacher to make the nanoparticles prior to the students synthesize their own.

Note: In making the gold and silver nanoparticles, the last section (adding NaCl to the nanoparticle suspension) may be omitted so as not to form aggregates of the NPs.

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Suggested Instructional Procedure:

Time	Activity	Goal
	The day before the lab	
25 min	Introduce students to the topic of light and the relationship between light and energy (E = hf).	Students understand there is a simple and direct relationship between energy and the frequency/wavelength/color of light.
Day 1	**The first day of the student lab**	
5 min	Students answer warm-up questions to review from yesterday (E = hf and c = λf)	To ensure students understand the relationship between energy, frequency, and wavelength
15 min	Divide the class in two groups. One will make silver nanoparticles, the other gold. Introduce the chemical reaction that each group will be using. Cover proper safety gear and how to work safely. Explain the fundamentals of and procedures for using a spectrophotometer.	
30 min	Students run chemical reactions to form nanoparticles.	
5 min	Students clean up, label their samples, and store for analysis.	
Day 2	**The second day of the student lab**	
Prior to class	Turn on spectrophotometers or connect to RAIN.	So that they will be ready to go by the time students are prepared to operate them.
30 min	Have students take out *Student Worksheets* to students. Students follow procedures to measure the absorption of their particle samples and compare their spectra with the standard references.	To allow students to collect absorption data at varying wavelengths for their samples of metal nanoparticles.
5 min	Clean up.	To prepare workspace for next class.
20 min	Read, *Chemmatters*, Nanotechnology's Big Impact, by Nadia Halim, Oct. 2009; or Nanotechnology 101 https://www.nano.gov/nanotech-101	Learn how nanoparticles are used in real-world applications.

Directions for the Activity: In Student Guide with answers below.
Begin with a discussion using these questions:4
Ask students questions to provoke thought and review what they already know:

1. Why do we see colors?

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2. What is happening within the atom when colors are produced?
3. What happens if we shine, for example, red light on a blue object?
4. How can scientists use colors analytically?

Next, cover the vocabulary to ensure students understand the terms correctly. Finally, have students examine the thin films with the LED lights.

Cleanup: Collect the suspensions of silver and gold nanoparticles separately. The gold nanoparticles can be disposed of down the drain. The silver nanoparticles on the other hand are a powerful antimicrobial, and should not be released into municipal sewers or septic tanks, since they can destroy beneficial bacteria needed for sewage treatment. Instead, collect all the waste silver nanoparticle suspension in a beaker and add table salt (NaCl) until the suspension becomes visibly cloudy. The addition of the salt causes the nanoparticle to agglomerate into larger particles that can be captured in a paper filter. Using a paper lab filter (or a coffee filter) and a funnel, pour the suspension into the funnel and allow it to pass through the filter. The liquid passing through the filter should be clear; if not, add more NaCl to it and re-filter. When filtration is complete, the clear liquid may be discarded down the drain. Allow the paper filter containing the silver to air dry, or gently dry it in a lab oven at low temperature. When dry, the filter paper may be disposed of in the trash.

Post Lab Follow-up:
Review the findings with students:
1. Ask random students for their observations on part 1, where different colors of light were applied to the quantum dot film.
2. Ask for students to explain their results.
3. Ask students, "What new questions does this activity lead to?"
4. For Part 2, have students share their absorption graphs. Were the results identical? Why or why not?
5. Did the absorption graphs obtained by the students match up with any of the references? If there was not an exact match, were there perhaps multiple sizes (or a broad size distribution) of nanoparticle in their sample? Work with the class to identify evidence of multiple sizes or wide size ranges.

Assessment: Upon completion of this activity, students should be able to:
- Calculate the energy for different wavelengths of light.
- Use a spectrophotometer for analytical purposes.
- Explain an absorption curve.
- Estimate the size distribution (peal size) of nanoparticles, using a spectrophotometer.
- Explain how nanoparticles can be used in modern technology.

The criteria for grading should be based on logical reasoning and creative thoughts, not necessarily on the accuracy of their responses. However, data obtained from the spectrophotometry should be consistent and clear. Grade deductions should occur for improper use of the spectrophotometer and poor quality graphs.

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Optional: Students who have a good grasp of the content of the lab can be further challenged with these questions:

1. What, if any, is the significance of the *width* of the peak absorption graph?
2. How does this activity relate to the photoelectric effect?
3. Suppose a scientist were to transmit blue light through a sample of quantum dots that emitted both red and green light. What would be produced? *Combining blue, red and green produces white light.*
4. Given the (correct) results of the previous question and your knowledge of light, what are some creative uses of quantum dots, gold and silver nanoparticles? Research these. *Creative uses include imaging, electronic displays (TV's, tablets, phones, computer monitors), solar cells, etc.*

Next Generation Science Standards:
HS-PS1.A: Structure and Properties of Matter
HS- PS1.B: Chemical Reactions
HS -PS2-1: Analyzing and Interpreting Data
HS- PS4.A: Wave Properties
HS- PS3.D: Energy in Chemical Processes

Contributors: Chris Kaus (NNIN RET) and James Marti, University of Minnesota

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Student worksheet with answers

Student Worksheet
(with answers in red)

Spectrophotometry with Metal Nanoparticles

Safety
Students should exercise care when handling the glass cuvettes, when working around hotplates and using chemicals.

Introduction

You have a summer job as an intern at a company that is leading the research for creative uses of new materials. Your boss gives you a small jar of something she calls 'metal nanoparticles' suspended in water. After providing a little background on what they are, she asks you to use your creativity and investigate some properties of these particles.

You decide to investigate the optical properties of the nanoparticles. First, you will synthesis a sample of either gold or silver nanoparticles. You will follow this up with an investigation of the optical absorption properties of the different nanoparticle samples using a spectrophotometer.

Materials:

- Two transparencies with thin film of either gold or silver nanoparticles between them, or 2 microscope slides with thin film of nanoparticles between them
- LED Flashlights of red, green, blue, and UV
- Spectrophotometer
- Absorption curves of a range of both gold and silver nanoparticles
- Calculator
- Chemicals and associated materials to synthesize gold or silver nanoparticles. Your teacher will provide a list of what you need depending on which metal youwill be working with.

Procedure:

Part 1: Thin Film of Gold and Silver Nanoparticles - Teacher Demonstration with Student Observation & Response. Put all responses in Table 1.

1. Your teacher will show you a thin film of nanoparticles. Make observations about this film, including size, shape, color, etc.
2. Shine white light on top of the film at an angle. Note any observations.
3. Shine white light through the thin film (transmission). Note observations.
4. Repeat steps 2 & 3 for lights of various colors, including the UV light.
5. Explain your observations with your classmates.

Part 2: Synthesis of Gold and Silver Nanoparticles

Silver Nanoparticles (from https://education.mrsec.wisc.edu/synthesis-of-silver-nanoparticles-nabh4/

1. Put on goggles and wear gloves.
2. Add 30 mL of 0.002M sodium borohydride ($NaBH_4$) to an Erlenmeyer flask. Add a magnetic stir bar and place the flask in an ice bath on a stir plate. Stir and cool the liquid for about 20 minutes.
3. Carefully pour 2 mL of 0.001M silver nitrate ($AgNO_3$) into a graduated cylinder. Then, squeeze the bulb of the plastic pipette and insert the tip into the graduated cylinder. Slowly release the bulb to draw the silver nitrate into the pipette. Slowly drip this solution into the stirring $NaBH_4$ solution **at approximately 1 drop per second**. Stop stirring as soon as all of the $AgNO_3$ is added.
4. Test the presence of a colloidal suspension (silver nanoparticles) by shining a laser beam into the solution. A reflected beam indicates the presence of the nanoparticles.

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5. Carefully pour your nanoparticle solution into the provided container. Label your container with: Silver Nanoparticles, your names, and the date.
6. Rinse out your glassware.

- Gold Nanoparticles (from https://education.mrsec.wisc.edu/citrate-synthesis-of-gold-nanoparticles/)

1. Put on goggles and gloves.
2. Add 20 mL of 1.0mM $HAuCl_4$ to a 50 mL beaker or Erlenmeyer flask on a stirring hot plate. Add a magnetic stir bar and bring the solution just to a boil.
3. To the boiling solution, add 2 mL of a 1% solution of trisodium citrate dihydrate, $Na_3C_6H_5O_7 \bullet 2H_2O$. The gold solution gradually forms as the citrate reduces the gold (III). Use forceps to remove from heat when the solution has turned deep red (about 10 minutes).
4. Test the presence of a colloidal suspension (gold nanoparticles) by shining a laser beam into the solution. A reflected beam indicates the presence of the nanoparticles.
5. Carefully pour your nanoparticle solution into the provided container. Label your container with: Gold Nanoparticles, your names, and the date.
6. Rinse out your glassware.

Part 3: Spectrophotometry of Gold and Silver Nanoparticles

1. Turn on your spectrophotometer at least 20 minutes before beginning this experiment.
2. On the top line of Data Table 2, indicate which type of nanoparticles you are analyzing.
3. If you are analyzing SILVER nanoparticles, set your first wavelength to 350 nm using the wavelength control knob. If you are analyzing GOLD nanoparticles, set your first wavelength to 450 nm using the wavelength control knob. Adjust the amplifier control knob to produce 0% transmittance.
4. In a CLEAN test tube (no fingerprints!) add about 3 mL of distilled water. Place the test tube in the sample holder and close the cover. Adjust the light control knob until the spectrophotometer reads 100 %T. Remove this test tube from the holder.
5. In another CLEAN test tube, add about 3 mL of your nanoparticle solution. Insert this test tube into the holder and record the %T. Remove this test tube from the holder.
6. Adjust the wavelength to 20 nm greater than your previous one. Use the amplifier control knob to adjust the transmittance to 0%T.
7. Place the water-filled test tube into the holder and adjust the light control knob to produce 100%T. Remove this test tube from the holder.
8. Place your nanoparticle sample into the holder and record the transmittance. Remove this test tube from the holder.

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9. Repeat steps 6 – 8 (set the new wavelength, adjust to 0%T with nothing in the holder, place the water in the holder and adjust to 100%T, remove the water and place the nanoparticle sample in the holder and record the %T) until you have collected a total of 16 values.
10. When finished, return your nanoparticles to your storage bottle.

Cleanup:

There should be minimal cleanup for this activity. All test tubes containing silver and gold nanoparticles should be emptied into the appropriate storage container provided by your instructor. In the event of a spill or breakage of a cuvette containing gold/silver nanoparticles, refer to the MSDS for cleanup and disposal.

Record Your Observations: *[Possible student responses are in red italics.]*

Table 1. Observations of Thin Film Nanoparticles

Observations of the Thin Film	*Students may state: various colors are seen as film is rotated. Some may not note any colors observed.*	
Color of Light	**Light shined on top of film**	**Light shined from behind film (transmission)**
White	*Nothing is observed, no changes*	*Nothing is observed, no changes*
UV	*Red and green colors at different locations*	*Red and green colors at different locations*
Red	*Some red light is reflected.*	*Red light shines through.*
Green	*Some green light is reflected.*	*Red light shines through.*
Blue	*Many colors are reflected.*	*Many colors shine through.*

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Table 2. Spectrophotometry of Gold/Silver Nanoparticles

	Quantum Dot 1		Quantum Dot 2		Mixed Quantum Dots	
Wavelength, nm	**A**	**%T**	**A**	**%T**	**A**	**%T**
450						
470						
490						
510						
530						
550						
570						
590						
610						
630						
650						
670						

Analyze the Results:

1. Create a <u>nice</u> graph of %T vs. wavelength. Use Excel or Google Sheets. Be sure to put a title and appropriate labels on your graphs.
2. Use the relationship below to create a graph of absorbance vs. wavelength.

   $$A = 2 - \log(\%T)$$

3. At what wavelength is your peak %T?___________________

   At what wavelength is your peak A?____________________
4. Absorption spectrum graphs from three silver standards of silver and three gold standards are provided on the next page. Each of these represents a distinct size of nanoparticle. Compare your graph to these standards to estimate the size of nanoparticles you made. You may have to interpolate or extrapolate.
5. Record your estimate here: _____________________
6. Perform some research on the Internet to find three applications of the nanoparticles you created. Be sure to explain how they are used in these applications.

Optional: Your teacher may request that you answer the extension questions:

1. What, if any, is the significance of the *width* of the peak absorption graph?
2. How does this activity relate to the photoelectric effect?
3. Suppose a scientist were to transmit blue light through a sample of quantum dots that emitted both red and green light. What would be produced? *Combining blue, red and green produces white light.*
4. Given the (correct) results of the previous question and your knowledge of light, what are some creative uses of quantum dots, gold and silver nanoparticles? Research these. *Creative uses include imaging, electronic displays (TV's, tablets, phones, computer monitors), solar cells, etc.*

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