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Laboratory Experiment

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Synthesis and Characterization of Silver Nanoparticles for an Undergraduate Laboratory

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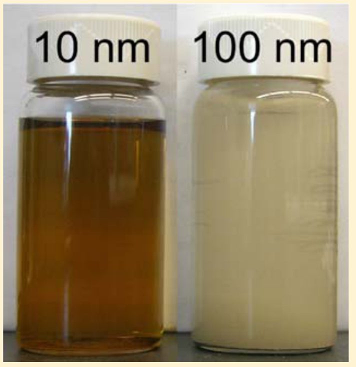
S *Supporting Information*

ABSTRACT: The aim of this simple, quick, and safe laboratory exercise is to provide undergraduate students an introduction to nanotechnology using nanoparticle (NP) synthesis. Students are provided two procedures that allow for the synthesis of different yet controlled sizes of silver NPs. After preparing the NPs, the students perform UV−visible spectroscopy and full-width-half-maximum (fwhm) analysis to determine the size of the NPs they have synthesized. In doing so, the students made nanoparticles of size ranges from 10−100 nm, thus spanning the range of nanotechnology. The experiments are designed to be accomplished in a single 90 min session and have been successfully conducted with over 1000 first-year-level students over a three-year period.



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■ INTRODUCTION

The inclusion of nanoscience and nanotechnology into introductory chemistry texts and courses has become the norm. Unfortunately, gaining a hands-on experience of nanoparticle (NP) synthesis and their size/property relationships is often difficult for typical Freshman Chemistry laboratory courses with >50 students per laboratory period. For example, the production of C_{60} or carbon nanotubes requires specialized lab equipment and complicated purification processes, making it suitable for an advanced laboratory with a smaller number of students. Furthermore, many of the chemicals needed to produce semiconductor quantum dots are either hazardous and/or toxic. There is a need to have a laboratory exercise that can safely and reproducibly produce NPs using standard chemistry equipment and techniques such as Erlenmeyer flasks, beakers, and stir plates. In this laboratory exercise, students are given the opportunity to conduct a lab that will enable them to enhance their chemistry skills while also applying principles taught in a freshman chemistry course as well as offering a first experience with nanomaterials. Very little chemistry experience is required for this lab to be successful and so it can be carried out with beginners and novices alike.

Silver has been known for use as a healing and antiseptic agent for centuries. For example, the inside of the wine chalice used for the Catholic congregation is lined with silver in order to prevent the spread of bacteria when sharing the wine,[1] and during the Second World War, silver powder was put on open wounds to keep them clean from infection.[2] Since the discovery of silver NPs, it has led to even greater use of silver in everyday products ranging from burn creams to food packaging. They are used in paint in hospitals to prevent the spread of disease, the inside lining of refrigerators to keep food fresher and longer lasting, and in socks and other clothing to kill odor-causing bacteria. Other uses of silver particles include: chemical sensors,[3] detection of disease,[4,5] and cancer treatment.[6,7] As such, Ag-NPs offer an excellent archetype of nanochemistry that students can appreciate their impact in society.[8]

The aims of this laboratory exercise include teaching synthetic and spectroscopic skills including using λ_{max} and full-width-half-maximum (fwhm) and how this related to NP size and size distribution, respectively. It is also designed to allow the teaching of concepts including how alteration of chemical conditions can control NP size and using the size of the resulting NP to assess the chemical reactivity of the reagents (i.e., the reducing agent in the present case). It may also be used in conjunction with either a pre- or postlaboratory session to teach the overlap with physical chemistry and surface plasmon resonance. By carrying out the reactions mentioned herein, students will be taught to calculate the fwhm and from this determine how the synthesis of silver NPs are affected by various experimental parameters.

Surface plasmon resonance (SPR) is an important effect associated with nanomaterials and their application in both research and industrial settings. Hence, due to the emergence of nanotechnology, it has become a phenomenon that is recently emerging within the undergraduate curriculum, this lesson plan is unique because it gives hand-on demonstration about the presence of the SPR, and because it creates such distinguished colors, it entices students to further their understanding about the chemistry at work.

Nanoparticles are especially intriguing; not only do they produce such interesting colors, but also they have been used in medicine and technology in a number of areas to date. For example, silver nanoparticles are often employed within sanitary materials such as burn creams and bandages as they exhibit antimicrobial and antifungal properties, thus making this exercise even more valuable as it gives an understanding to the students of the chemistry of materials that can be found on supermarket shelves.

■ EXPERIMENTAL OVERVIEW

There are several methods to synthesize silver NPs, but all follow, in one way or another, a similar pathway and similar functional reagents. The individual processes can be broken down into four conceptual stages: nucleation, growth, ripening, and termination of growth. In terms of the chemical reagents, for the sake of clarity and simplicity, the mechanism can be described by three reactants: the metal source, the reducing agent, and the capping ligand. Teachers should note that a capping ligand can also be referred to as a surfactant in the primary research literature. Both sets of these concepts can be presented as part of a prelaboratory exercise, a tutorial, or, depending on the students level, prelaboratory handouts. The students use two synthetic routes to prepare silver NP solutions: from a silver source, a reducing agent, and a surfactant. A detailed description of these factors is provided in Supporting Information.

Should further background reading be required it can be found in the teacher's notes as provided in the Supporting Information. In addition, a copy of a typical Power Point presentation that is used prior to the laboratory exercise is provided. Further to this, there is also provided an example of the questions that were given to the students to answer, along with the answer key.

■ EXPERIMENTAL PROCEDURE

Overview

We initiated our program in a manner similar to the proposed classroom demonstration by Mulfinger et al.[9] as well as the known syntheses of silver nanoparticles.[10−13] This plan is for a general chemistry curriculum. The majority of students were engineering students and premedical school students, but we have shown it to work for any student background, as demonstrated at the high school level also. This laboratory exercise was used to demonstrate the synthesis of nanoparticles and highlight how the average nanoparticle size changes over a period of time. Students were required to take spectral data using MicroLAB (or any other spectrophotometer that is available). Students can work in groups of two or more. Each group was told to prepare at least two separate solutions of silver nanoparticles; one using the Lee−Meisel method[11] (Lesson Plan 1) the other using the Turkevich method (Lesson Plan 2).[14] We suggest starting with Lesson Plan 1, as the nanoparticles are made rapidly and it does not require heating. While data is being acquired for this solution and students are determining full-width-half-maximum (fwhm) values, they can begin Lesson Plan 2. To assist in the process and ensure the work is carried out in a timely manner, solutions of appropriate concentrations were prepared in advance of the laboratory exercise. Through the combination of Lesson Plans 1 and 2, students are able to see how similar reactants can be used to make different products, namely NPs of varying sizes. This answers a fundamental question regarding how to make and shape objects at the nanoscale and how one can make materials from the bottom up.

The objective for the teacher is to ensure that students make nanoparticles of silver according to the Lesson Plans 1 and 2. In doing so, students should be directed to note a change in color of the solution during the formation of nanoparticles. The change in color is backed up by use of a spectrophotometer, and the data is used to determine λ_{max} and calculate the fwhm. Students will make two sets of particles, each with different sizes and thus with different colors. The size is affected by the strength of the reducing agent, and they will be able to determine which reducing agent of the two that are used is stronger. Stronger reducing agent makes smaller nanoparticles, and the size of the particles are determined according to the λ_{max}. Additionally, students should be guided toward additional observations, that of time for reaction completion and temperature. The stronger reducing agent takes less time for the reaction to come to completion and does not require additional energy, in the form of heat, to come to completion.

Lesson Plan 1: Synthesis of 10 nm Silver Nanoparticles

Students mixed 100 mL of a 1.2 mM solution of silver nitrate with 100 mL of a 3 mM solution of mercaptosuccinic acid using a magnetic stir bar to create agitation. To this mixture, students were told to slowly add 25 mL of 2 mM sodium borohydride solution, taking care to add $NaBH_4$ slowly, where the time frame was typically of the order 1−2 min. Should a large amount of $NaBH_4$ be added too rapidly, one can expect a large increase in pH to occur; this has been found to cause coinage metals to precipitate from solution[15] and will prevent the formation of nanoparticles from occurring. The solution color rapidly changed from a clear solution to a light yellow, then dark yellow, brown, and then black color (Supporting Information). Upon addition of the 25 mL of $NaBH_4$ solution, students then took aliquots from the reaction at regular intervals (every 5 min) for a period of 30 min. These aliquots were then characterized using a UV−visible spectrophotometer. The reaction solution should be diluted in order to carry out UV−visible spectroscopy; students carried this out by the addition of 5 drops of reaction solution into 20 mL of DI water. Each student determined the λ_{max} and calculated the fwhm for the peak after 30 min time frame. They noted how the peak changes during the first half hour of nanoparticle ripening.

Lesson Plan 2: Synthesis of 100 nm Silver Nanoparticles

Students placed 75 mL of 2.4 mM silver nitrate solution in a glass vessel along with a magnetic stir bar, thus creating a moderate vortex. Upon heating the solution to 90 °C, students then added 2 mL of a 38.8 mM trisodium citrate solution dropwise, allowing a second or two between each drop; this consisted of approximately 50 drops (using a plastic Pasteur pipet) over a 2 min period. Keeping the solution at 90 °C, the students removed aliquots at regular intervals (every 5 min) for spectral analysis; typically, 10 drops of reactant can be added to

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20 mL of DI water. During the course of the reaction, a laser pointer can be used to identify when the colloids grow greater than 40 nm. The color should change to a light yellow color and, within 30 min, change to a dark brown. Finally, it will end as a gray milky white color (Supporting Information).

■ HAZARDS

As with all undergraduate chemistry laboratories, safety glasses or goggles, lab coats, gloves, and closed toe shoes are required.[16] Skin protection is necessary when using silver nitrate solutions because skin contact will result in purple, brown, or black stains; with constant exposure to high concentrations, burns can result. Due to its highly oxidizing property, silver nitrate should be properly stored away from organic compounds. Sodium borohydride and trisodium citrate can both stain clothing, so the appropriate lab protective clothing should be worn at all times. Mercaptosuccinic acid can form an unpleasant odor if spilled on clothing. Leftover solutions should be disposed of correctly, and silver solutions can be collected for reuse.

■ EXPERIMENTAL DATA AND ANALYSIS

For each solution students gathered UV−visible data over a time period lasting approximately 30 min for each solution. From the UV−visible data, students plotted the spectra according to time, which was submitted for grading. They answered a series of questions regarding the spectra and from the data infer which of the two methods incorporates the stronger reducing agent. Students completed a written report after the laboratory exercise was completed. In their report, they were to submit a UV−visible spectra of silver nanoparticles from both Lesson Plan 1 and 2 each. Using the UV−visible spectra, they could then determine, based on the fwhm, which of the two utilized the faster acting reducing agent. The details of this lesson can be found in the instructor's notes as Supporting Information. The written report is based on the following three attributes: size ripening of nanoparticles with respect to time, understanding the fwhm and using it to determine the approximate size (and size range) of the nanoparticles, and using the fwhm to determine which reaction uses the faster acting reducing agent.

Size Ripening with Time

The condition of surface plasmon resonance will cause an absorbance peak to occur in the UV−vis spectra. The wavelength of maximum peak absorbance can be used to determine the approximate size range of the nanoparticles. In the case of Lesson Plan 1, a successful reaction will create nanoparticles with λ_{max} = 400 nm and for Lesson Plan 2 λ_{max} = 440 nm. The shape of the peak can be used to qualitatively ascertain the size range of the particles in solution. For samples of monodisperse nanoparticle sizes, one can expect a narrow peak centered at λ_{max}. By acquiring UV−visible spectroscopic data at 5 min intervals and plotting a figure similar to that of Figure 1, students can observe how the peaks width changes with time up to a certain time frame, after which the fwhm does not fluctuate further. These changes in the peak width are indicative of the ripening of the nanoparticle sizes, and as the peak narrows down this indicates a more homogeneous size distribution. This can all help to ascertain a qualitative understanding of the size and size distributions of the nanoparticles in solution. It should be noted that the spectrophotometer utilized in this class acquired wavelength

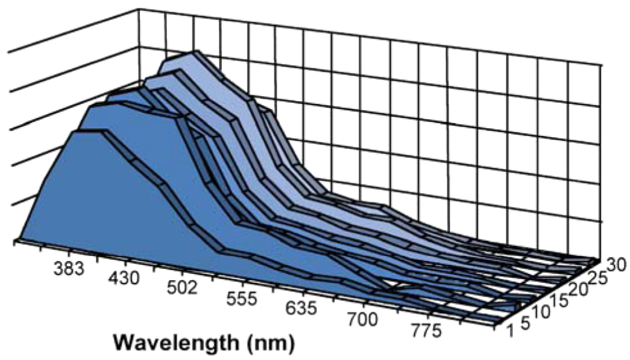

Figure 1. Typical UV−visible spectra from the formation of silver nanoparticle by Lesson Plan 1.

data at nonlinear and irregular wavelengths, as can be seen in Figure 1. This is an attribute of the hardware and cannot be avoided as such; if possible, more appropriate time intervals of consistent regularity would be preferred.

Full-Width-Half-Maximum and Diameter Distribution of Nanoparticles

Following from the observation of changing peak width, the concept of the full-width-half-maximum is introduced. The students are provided with an example of how to determine the fwhm of a UV−visible spectrum (Figure 2). In a typical

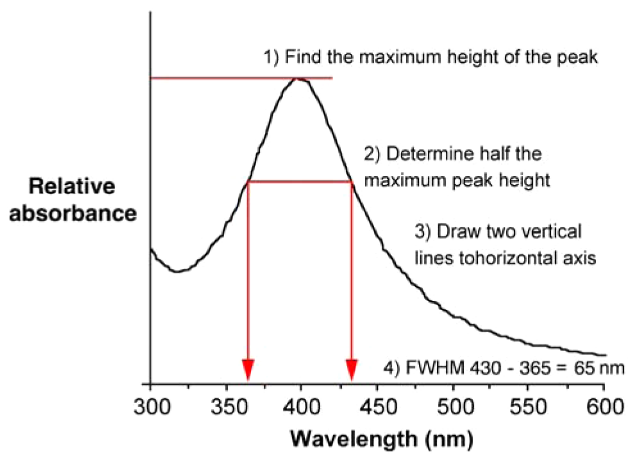

Figure 2. A four-step process used to determine the full-width-half-maximum for an absorbance peak in UV−visible spectra.

example and using spectral data from each of Lesson Plans, based on a quick comparison of the fwhm, one can see in Figure 3 the fwhm is found to be 87 nm (Lesson Plan 1), and from Figure 4 the fwhm can be found to be 252 nm (Lesson Plan 2). The students are asked to comment on the implication of this result: Lesson Plan 1 created NPs that are more homogeneous in scale as compared with those from Lesson Plan 2.

Strongest Reducing Agent

The students are provided with the information that stronger reducing agents tend to make smaller nanoparticles with a more homogeneous size distribution. The students are asked to indicate which is a stronger reducing agent: sodium borohydride or trisodium citrate. Further, to this it should be noted that the strength of the reducing agent is not the only determinant factor influencing size of resultant nanoparticles.

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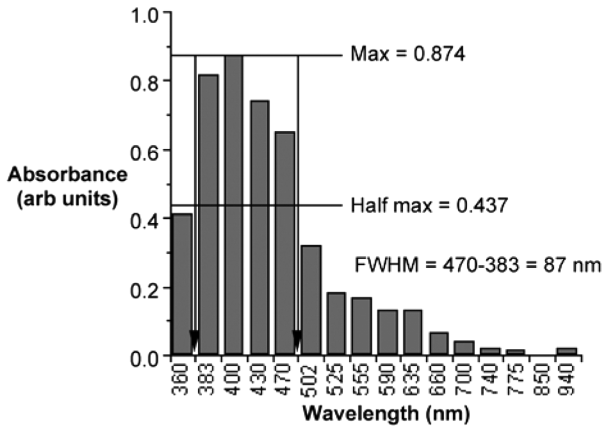

Figure 3. Full-width-half-maximum (fwhm) calculations of the UV−visible spectra acquired using MicroLab for a solution of 10 nm silver nanoparticles made using Lesson Plan 1.

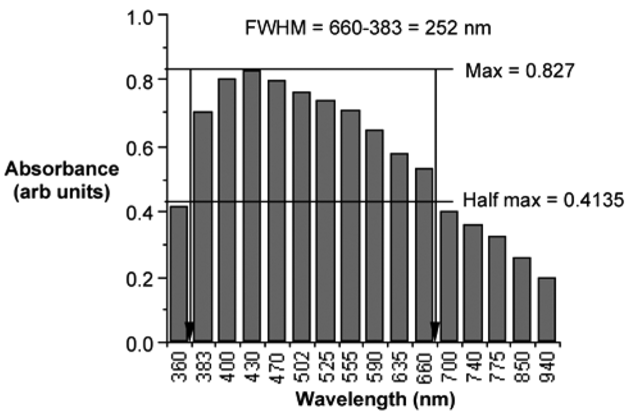

Figure 4. Full-width-half-maximum of the UV−visible spectra acquired using MicroLab for a solution of 100 nm silver nanoparticles made using Lesson Plan 2.

During the laboratory exercise, students were also asked to report on other factors that would influence the decision as to which is the stronger of the two reducing agents; to do this, they were asked to list two observations in their reports. The two factors to consider are also the length of time that it took for nanoparticle formation to occur; in the case of sodium borohydride, it was within moments upon addition, as opposed to 15−30 min upon addition of sodium citrate. Additionally, the sodium borohydride did not require heating to produce nanoparticles, suggesting it is more energetic and therefore a stronger reducing agent than sodium citrate.

■ CONCEPTS RELATED TO CHEMISTRY LECTURES

A wide variety of topics can be covered by this laboratory exercise, but in the interest of brevity here is a brief list of those topics that have been used in the course of this work. The concepts discussed include: scale, scanning electron microscopy and transmission electron microscopy, chemical reduction, the Beer−Lambert law, plasmons, surface area to volume−effective surface area, full-width-half-maximum (fwhm), reducing agents and the Tollens (silver mirror) reaction, and colloidal stability.

With regard to size, the question arose about the relationship in size between a nanoparticle and an atom. Explaining that silver has an atomic radius of 1.4 Å (i.e., 0.14 nm) and that the nanoparticles of silver made by the students are approximately 20 nm in diameter allows the students to calculate that their nanoparticles are on the order of 140 silver atoms across. In this way, the students are able to put into the perspective both the size of the atom and the size of nanoparticles. Special emphasis was placed on the fact that, at the nanoscale, a number of interesting properties are enhanced, the most relevant of which deals with the surface plasmon resonance. In particular, it was presented that at nanoscale dimensions materials have properties that are described by a mixture of classical continuum physics and quantum mechanics. In essence, at the nanoscale, there is a blurred distinction between the types of physics that apply. In effect, many of the properties of nanoscale materials are greatly enhanced because of this, and the example used most predominantly in the class room stems from Mie theory on the scattering of light, as this pertains directly to the understanding of the surface plasmon resonance (SPR) effect of nanoparticles.

Because optical microscopy is insufficient to observe individual nanoparticles, the techniques of scanning electron microscopy (SEM) and transmission electron microscopy (TEM) are introduced to the students. The working principles behind both methods were described in the introductory lecture associated with the laboratory. Example of SEM and TEM micrographs of silver nanoparticles, previously prepared by the same methodology, were provided as introductory course material (Supporting Information).

The reaction mechanism for making the silver nanoparticles involves the reduction of the silver ions.[17] The chemical process of reduction is discussed and compared to other known reduction reactions. As prepared, the silver nanoparticle solution is too concentrated to allow for UV−visible measurement. The concept of the Beer−Lambert law in relating adsorption to concentration is used to justify the necessary dilution of the samples.[18] A useful approach to the discussion of SPR is as an analogue to pushing someone on a swing because you slowly increase the momentum of the swing by adding a force to the person sitting on it. As the person gains momentum, it takes less and less energy to keep that person going higher and higher. Furthermore, the person only needs to be pushed at certain times; this can be likened to the frequency of the light. Thus, in pushing a person, your energy couples with the energy of the person on the swing, which is analogous to the incident light energy coupling to the electrons. Gradually, less and less force is required to make someone swing faster and faster, when you push at the right frequency, because you are pushing in resonance with the swinging person. This is exactly the same as what happens to the silver nanoparticles. The incident light resonates with the surface electrons, and they oscillate back and forth. This whole process is known as surface plasmon resonance, or SPR, and there is a lot of research being carried out with different metals and various sizes and shapes of nanoparticles to use this optical effect.[19] Because the SPR of a nanoparticle is very discrete, it can be deliberately created in one part of the spectrum or another.

Nanoparticles are defined as objects with components that measure in length from 0.1−100 nm[20] that specifically exhibit a unique property, different from the bulk material, that is caused by that surface area to volume. In a simple model, one can imagine a tennis ball (7 cm in diameter) that is shrunk in size ten million times ($\times 10^7$) to a nanoparticle (7 nm diameter). When you shrink something that much, you end up having most of the atoms that make up the particle sit on the surface, i.e., it has a very large aspect ratio because the surface area compared to the volume is large. This is very important in defining the properties of the nanoparticle because chemistry happens on surfaces, so the more surface you create, the more reactions you can get. The surface area of a solution of silver nanoparticles can be so high that a small teaspoon of nanoparticles can have the same surface area as a football field, but the mass of nanoparticles in this teaspoon of solution may only be a few hundred milligrams, whereas silver metal covering an entire football field may weigh a few hundred kilograms. The impact of a high surface area to the reactivity of nanoparticles is discussed. In addition, the concepts of surface energy and Ostwald ripening are readily introduced here using the time-dependent UV−visible spectra.[21]

The measurement of full-width-half-maximum (fwhm) is a standard used to determine the properties of the peaks in our acquired UV−visible spectroscopic data. Specifically, it helps the students understand something about the size of peaks when the shape is not always regular. As an example, the students find it easy to measure the size of a rectangle because the edges are flat, but for a Lorentzian peak, as seen in the UV−visible spectrum of the silver nanoparticles, this is more difficult. Introduction of the fwhm concept allows them to obtain a numerical value that can then be used to compare with other sets of data.

With respect to reducing agents, discussion can be made that making silver nanoparticles can be compared with the classical silver Tollens reaction. An additional topic for discussion includes the properties of colloids, what they are, and how they are made, and what can affect their stability. In a brief demonstration, adding brine solution to the silver nanoparticle product can cause the nanoparticles to precipitate. Further to this, the concept of solubility of metal ions and pH can be explored, as there is reference given to the fact that sodium borohydride was once touted as a reactant that may be able to separate coinage metals based on pH.[15]

■ SUMMARY

A laboratory protocol is described that was carried out by freshman undergraduates during a typical time frame for laboratory exercises. In the protocol the procedure of making nanoparticles of various sizes and how this size is related to color was explored as a means to inquiry-based learning. This laboratory experiment was carried out with over 1000 students during a three-year period. To measure the success of the reaction, students were asked to submit what the value was for λ_{max} for each Lesson Plan. As an example of a typical class of 60 students, Figure 5 shows that for Lesson Plan 1 ca. 80% of students reported the λ_{max} to be 400 nm. Minor deviations were found to usually result from students not giving sufficient time for the reaction to come to equilibrium, as determined from the observation of their UV−visible plot, cf., Figure 1. Significant deviation has been attributed to the inability of some students to follow protocol correctly and, hence, can be used as part of the laboratory evaluation.

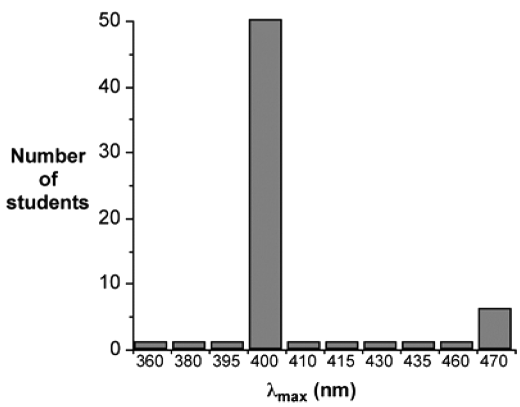

Figure 5. Chart showing the values of SPR peak (λ_{max}) acquired by students making silver NPs using Lesson Plan 1, reduced by SBH. NB: It should be noted that the high values were determined to be due to the students incorrectly following the laboratory protocol.

■ ASSOCIATED CONTENT

Supporting Information

Instructor notes with sample questions and report key for students. This material is available via the Internet at http://pubs.acs.org.

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Notes

The authors declare no competing financial interest.

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