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Surface-Enhanced Fluorescence with Shell-Isolated Nanoparticles (SHINEF)**

*Ariel R. Guerrero and Ricardo F. Aroca**

Li et al. recently developed a new approach for surface-enhanced Raman spectroscopy (SERS), which they termed shell-isolated nanoparticle-enhanced Raman spectroscopy (SHINERS).[1] The plasmon-enhanced Raman signal is provided by Au nanoparticles with an ultrathin silica shell (2 to 4 nm). The enhancing coated Au nanoparticles can be spread as "smart dust" over the surface, and the coating separates them from direct contact with the probed material. The authors also pointed out that the SERS signal disappeared when changing the shell thickness from 2 to 20 nm. In SHINERS, working with an ultrathin shell for the Au/SiO_2 nanoparticles is essential to expose the adsorbate to the maximum electromagnetic field from the Au core (enhancement factor is 85 at 4 nm). However, for a fluorophore located on an enhancing nanostructure, an increase in thickness of the spacer layer (shell thickness) will make a continuous transition from fluorescence quenching to fluorescence enhancement apparent.[2] Therefore, shell-isolated nanoparticles (SHINs) with thicker coatings could be ideal substrates for surface-enhanced fluorescence.[2–4] Herein we demonstrate the application of the SHINs for surface-enhanced fluorescence (SEF; shell-isolated nanoparticle enhanced fluorescence, SHINEF) by using a single Langmuir–Blodgett (LB) monolayer that contains fluorescent probes. The enhanced fluorescence cross-section is one of the largest observed in molecular spectroscopy with values in the order of $10^{-16}\,cm^2\,molecule^{-1}$ and, correspondingly, there is a wide range of applications for this very strong spectroscopic signal.[3–5] A maximum fluorescence enhancement is achieved at a certain distance from the nanostructured surface, and this maximum has been demonstrated by Wokaun et al., using silver island films and SiO_x as spacer layers,[6] and by our group, by using the LB technique to separate the metal from the probe molecule.[7] Recently, the continuous transition from quenching to SEF for a single molecule on gold has been reported,[8] showing that for molecule–gold distances shorter than 5 nm, the fluorescence is quenched. Since it is possible to control the shell thickness of the shell-isolated nanoparticles,[9] it is evident that they are ideal SEF-enhancing nanostructures with a broad range of potential applications, thus expanding the versatility[10] of SEF with portable nanostructures. The dipole emission can be strongly modified by the coupling of an excited molecule with the surface states of a metal. This interaction has been discussed in an early paper by Philpot,[11] who explored the idea of using fluorescence to probe the surface plasmon polaritons in metals. Presently, plasmonics[12] provide the reference for plasmon-enhanced spectroscopies, and the surface plasmon polaritons observed in nanostructures or localized surface plasmon resonances (LSPR)[13] are central to SERS and SEF.[14] Gersten and Nitzan[2] provided the first complete electromagnetic (EM) study for a molecule–particle system in terms of a modified local electromagnetic field and plasmon resonance effects.

The plasmon absorption of the shell-isolated nanoparticles and the electronic absorption spectra of two dyes (octadecyl rhodamine B (R18) and bis(*n*-butylimido)perylene (nBPTCD)) in solution are shown in Figure 1. Notably, the absorption of the nanoparticles and the dyes is in resonance with the 514.5 nm laser line, and this line has been used to demonstrate the smart dust SEF. The scanning electron microscopy (SEM) and the TEM images of the SHINs in the smart dust are also shown in Figure 1, where the brightness is due to the gold core. The plasmon absorption of the SHINs selected in our work for a proof-of-concept, is relatively narrow (FWHM of 88 nm) with a gold core of approximately 45 nm and an approximate thickness of the coating of 20 nm; this thickness of the coating prevents the observation of SERS.[1] The reference fluorescence of a 10^{-4} M solution of

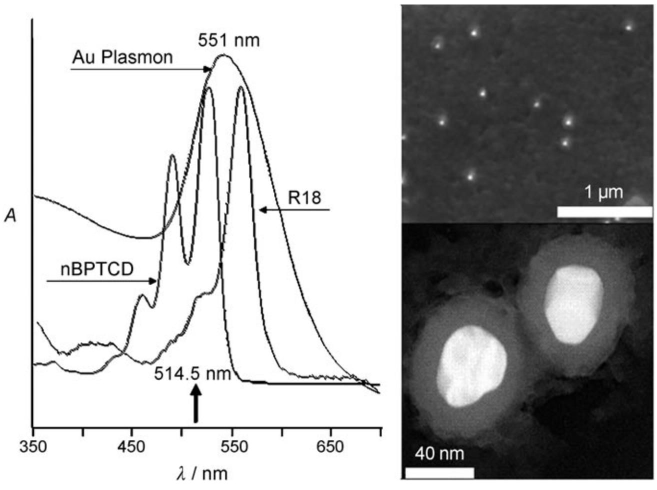

Figure 1. Left: Plasmon absorption spectrum of the Au-coated nanoparticles and absorption spectra of the R18 and nBPTCD dyes. Right: SEM (top) and TEM (bottom) images of the SHINs.

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[*] A. R. Guerrero, Prof. R. F. Aroca
Department of Chemistry and Biochemistry, Faculty of Science
University of Windsor, Windsor, Ontario N9B 3P4 (Canada)
E-mail: rarocal@cogeco.ca

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R18 and that of a mixed R18/arachidic acid 1:10 LB monolayer is shown in Figure 2. The solution emission of R18 shows a maximum at 589 nm and a shoulder at 618 nm. The LB fluorescence of R18 is quite similar and shows a maximum at 586 nm. The arachidic acid is used to facilitate the transfer of the monolayer to solid substrates.[15] The surface-enhanced fluorescence induced by the SHINs is shown in Figure 3 and corresponds to an average SEF enhancement (similar to the definition used for average SERS)[16] over the probed surface. This result means that there is a distribution of enhancement factors (EF) that contribute to the observed enhanced signal, and some localized spots may have a fairly large enhancement factor compared to the observed average.

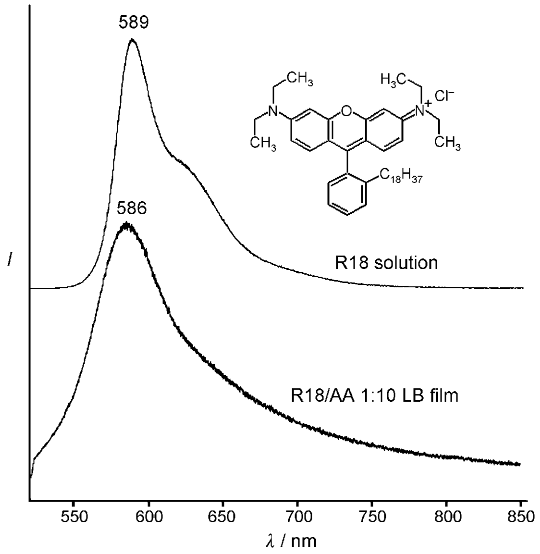

Figure 2. Reference fluorescence spectra of R18 in dichloromethane solution and of a LB monolayer on glass.

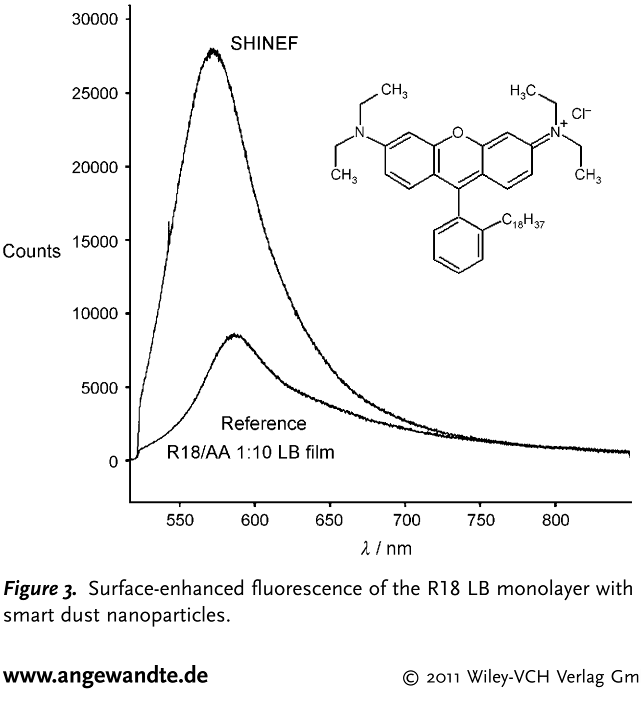

Figure 3. Surface-enhanced fluorescence of the R18 LB monolayer with smart dust nanoparticles.

The reference fluorescence spectrum of nBPTCD in dilute solutions is the mirror image of the absorption spectrum shown in Figure 1, with maxima at 540, 578, and 623 nm. From several previous studies[16,17] it is known that the PTCD dyes, in addition to the monomer emission, produce a very strong red-shifted excimer emission in both the solid state and particularly also in "concentrated" LB films because of the formation of aggregates. In the nBPTCD/AA 1:10 mixed film, the broad (structureless) excimer emission prevails in the fluorescence spectrum, as can be seen in Figure 4 (bottom trace). The SHINEF at two points of the monolayer covered with SHINs is given in Figure 4. A mapping of the small surface area shows enhancement factors for the integrated intensity of the excimer emission in the range of 1–20. The presence of the monomer is occasionally seen in the mixed film. Experiments were also carried out with a dilute mixed monolayer (nBPTCD/AA 1:1000; Figure 5). The reference spectrum of the dilute mixed monolayer shows the typical bands of the monomer at 547 nm and 589 nm, and correspondingly the SHINEF shows similar structures. However, the enhancement may produce an overlap of the main peaks (Figure 5). It should be pointed out that when LB monolayers of PTCD derivatives are transferred directly onto silver or gold nanostructures and excited by the 514.5 nm laser line, the fluorescence is almost completely quenched and surface-enhanced resonance Raman scattering (SERRS) is observed.[15,17] In the work presented herein using thick coating of the SHINs, we do not observe SERRS. Synthesis of SHINs with a variable core (size and shape) and coating thickness for plasmonic manipulation is currently in progress. However, we are focusing on coating thicknesses between 10 and 20 nm for SHINEF applications.

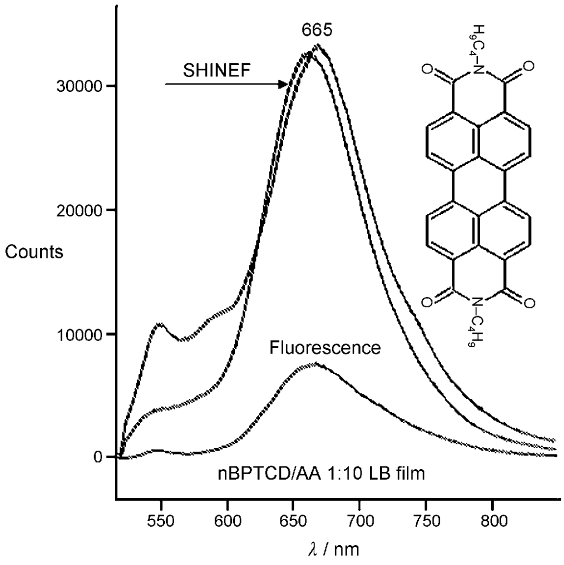

Figure 4. Reference fluorescence and SHINEF of a mixed LB monolayer of nBPTCD/arachidic acid 1:10.

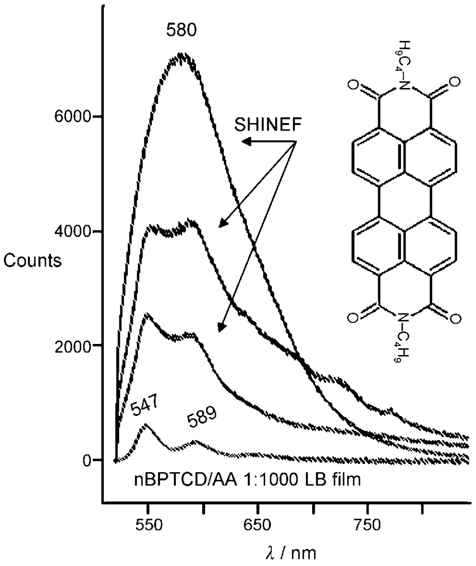

Figure 5. Reference fluorescence and SHINEF of a mixed LB monolayer of nBPTCD/arachidic acid 1:1000.

Although SHINEF has been demonstrated for specific SHINs with a coating thickness of about 20 nm, SHINEF can also be attained with thinner coatings. SHINs with coating thicknesses of about 10 nm produce SHINEF (see Figure S1 in the Supporting Information). For practical applications, a coating thickness of approximately 10 nm or more is recommended to maximize SEF and avoid any SERS signal. For instance, when a section of the LB monolayer is covered with the stock nanoparticles, the fluorescence is partially quenched and SERRS is observed (Figure S2). However, the section of the LB surface covered with SHINs gives only SHINEF.

In conclusion, average surface-enhanced fluorescence is demonstrated with silica-coated gold nanoparticles (SHINEF) acting as smart dust on the surface of single Langmuir–Blodgett monolayers. Coating gold and silver nanoparticles of different sizes and shapes (different plasmon absorptions) opens a wide scope of applications for SEF, where the shape of the core and the thickness of the coating can be tuned for specific tasks. The technique of spreading the enhancing nanostructure provides a new approach to experimental SEF.

Experimental Section

All chemicals were purchased from Sigma–Aldrich and used without further purification. Solutions were prepared by mixing octadecyl rhodamine B (R18) with *n*-eicosanoic acid ($C_{20}H_{40}O_2$) or arachidic acid (AA) 1:10, and bis(*n*-butylimido)perylene (nBPTCD) with arachidic acid 1:10 and 1:100. The concentration of the dye was 10^{-4} M and that of the arachidic acid was 10^{-3} M, both in dichloromethane. To improve solubility of nBPTCD, three drops of trifluoroacetic acid were added and then the solution was sonicated for 5 min. The solutions were covered in aluminum foil to protect them from light. UV/Vis absorption spectra for the solutions were recorded with a Varian Cary 50 spectrophotometer.

SHINs were synthesized by adapting the method described by Li et al.[1] The gold core was synthesized by gold citrate reduction of tetrachloroauric acid ($HAuCl_4$) based on existing protocols.[18,19] A solution of 0.01 % $HAuCl_4$ (50 mL) in water was heated to reflux. A solution of 1 % sodium citrate in water (667 μL) was added. Heating was continued for 10 min, then the mixture was removed from heat, while stirring was continued for 15 min. Then a 1 mM aqueous solution of 3-aminopropyltrimethoxysilane (3 mL; APTMS) was added under vigorous stirring; the resulting mixture was allowed to stand at room temperature. The resulting solution was heated in a water bath to a temperature between 90–95 °C; when reaching this range, 0.54 % aqueous sodium silicate solution (9 mL) was added to coat the silica, then the solution was allowed to stand at this temperature for 2 h, followed by cooling to room temperature. The samples were filtered using 0.20 μm pore polyethersulfone filters (Sarstedt), and then were centrifuged at 14000 rpm for 7 min, thus recovering the sediment. SEM images were taken with a FEI Quanta 200 Environmental scanning electron microscope equipped with an Everhart–Thornley secondary electron detector and a solid state backscatter detector. TEM images were obtained with a high-resolution TEM instrument FEI Titan 80–300 (at the Canadian Centre for Electron Microscopy at McMaster University). Langmuir monolayers of R18 and nBPTCD mixtures were prepared at the air–water interface of a Nima film balance (model 302M) with dimensions 414×70 mm^2, in order to obtain the surface pressure area isotherms and perform the depositions on glass slides. The solutions were spread on the aqueous surface by using a Hamilton microsyringe held very close to the aqueous surface; then the solvent was allowed to evaporate completely over a period of at least 25 min. The subphase was ultrapure Milli-Q water (18.2 MΩ cm) at a constant temperature of 23 °C. The monolayer was then compressed at a fixed barrier speed of 10 cm^2 min^{-1} to record the surface pressure area (π–A) isotherm. The Langmuir–Blodgett monolayers on Corning glass slides were fabricated by z-deposition at a constant surface pressure of 25 mN m^{-1}. To perform the SHINEF experiments, a drop of the SHIN particles (5 μL) was deposited on the LB monolayer (on the glass slide) and allowed to dry. Fluorescence spectra were acquired by point-by-point mapping of a section of the LB surface with the 50 × objective (1 μm^2 spatial resolution), with the Renishaw InVia micro-Raman system using the 514.5 nm laser line.

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