Plasmon-enhanced fluorescence of dyes on silica-coated silver nanoparticles: A single-nanoparticle spectroscopy study

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ABSTRACT

Dye-doped silica-coated silver nanoparticles (SCNPs) with shell thicknesses ranging from 8.6 ± 1.4 nm to 75.4 ± 4.8 nm are examined at the single-nanoparticle level using correlated transmission electron microscopy (TEM), localized surface plasmon resonance (LSPR), and fluorescence microscopy. The average fluorescence enhancement of 128 individual RB/SCNPs is 2.4 ± 0.2, with individual values ranging from 1.0 to 12.8. For these hot-spot-containing SCNPs, the far-field Rayleigh scattering is not correlated to fluorescence enhancement. Instead, fluorescence enhancement is positively correlated to aggregation state (i.e., the number of hot spots) and dye-metal distance, with ~19 nm separation producing the largest fluorescence enhancement.

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1. Introduction

Understanding the optical properties of chromophores in proximity to metal nanostructures has the potential to impact the design of materials for photovoltaics [1–3], photocatalysis [4], and surface-enhanced spectroscopies [5–9]. For example, one promising strategy to improve the efficiency of dye-sensitized solar cells (DSSCs) is to reduce device thickness while maintaining optical absorption via the introduction of plasmonic nanostructures (e.g., Ag, Au, Cu) [3,10,11]. Chromophores in close proximity to metal nanostructures also exhibit surface-enhanced Raman scattering (SERS) [8] and plasmon-enhanced fluorescence [12], which have broad applications to chemical and biological sensing. In all of these cases, the collective oscillation of conduction band electrons in the metal nanostructure produces a surface plasmon resonance and a concomitant enhanced electromagnetic (EM) field at the metal surface [13].

Plasmon-enhanced fluorescence has been examined as a tool for biosensing [5] and single-molecule detection [14], though the reported fluorescence enhancements vary widely [5,12,15–22], due to the complex processes occurring at the dye/metal interface as well as a wide range of nanostructures under investigation. In particular, emitters in close proximity to metal nanoparticles are known to exhibit a variety of behavior including excitation enhancement (i.e., increased absorption) [9,20], emission enhancement (i.e., increased radiative decay) [21,23,24], and quenching (i.e., increased non-radiative decay) [25]. Moreover, molecular photophysics near metallic nanoparticles are sensitive to orientation [26,27], spectral overlap between emitters and the localized surface plasmon resonance (LSPR) [19,21], nanoparticle structure [28,29], and distance between the dye and metal substrate [21,23–25]. The latter represents a significant factor for EM enhancement, since the EM field decays exponentially away from the metal surface [7]. Although nanostructure is known to play a role in fluorescence enhancement, the impact of 'hot spots', regions of high EM enhancement at the junctions or crevices between adjoining nanoparticles [30,31], on plasmon-enhanced fluorescence is not thoroughly understood. Plasmon-enhanced fluorescence has been reported on several nanostructures including on roughened metal films [23,32], single silver and gold nanoparticles [14,16,18,19,21,26], and gap-based antennas [14,15,29,33]. Yet, these experiments use aggregated nanoparticles with relatively large gaps (i.e., ~20 nm) or study aggregates at the ensemble-averaged level, where the measured plasmon-enhanced fluorescence is averaged across many nanoparticle sizes, shapes, LSPRs, and aggregation states. The distance dependence of plasmon-enhanced fluorescence for molecules on Ag island films and Ag nanoparticles has been measured at long ranges (>10 nm) using multiple protein layers as spacers [23,24]. More recently, the distance and LSPR dependence of plasmon-enhanced fluorescence was reported for solutions of silica-coated Au nanorods [21]. Yet, understanding the short- and long-range distance dependence (i.e., from <10 nm to >20 nm) of plasmon-enhanced fluorescence from single nanoparticles, especially those

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containing hot spots, remains elusive. In order to understand the structure-activity relationships of chromophores near metal nanostructures, a deeper understanding of the effect of 'hot spot' formation, LSPR wavelength, and dye-metal distance on plasmon-enhanced fluorescence at the single-nanoparticle level is required.

Our approach is to perform single-nanoparticle spectroscopy studies of dye-doped silica-coated silver nanoparticles (SCNPs), which act as versatile nanocomposites with well-defined optical and structural properties as well as variable shell thicknesses for distance-dependent fluorescence studies. To investigate the LSPR-, structure-, and distant-dependent fluorescence enhancement of dyes in proximity to Ag nanoparticles (AgNPs), SCNPs of various shell thicknesses are synthesized using a modified Stöber method. A traditional Stöber method uses an ammonia catalyst to perform the hydrolysis and condensation of tetraethoxysilane (TEOS) [34]. However, this traditional approach cannot be used with AgNPs, since silver is oxidized by ammonia, which leads to etching or complete dissolution of the particles. Previous methods have used polymeric capping agents to protect AgNPs from degradation by ammonia, but these procedures are problematic if the particles are not completely coated [35]. Alternative amine catalysts (e.g., dimethylamine (DMA)) have demonstrated uniform silica shells of >30-nm thickness in a sol–gel type process, but not uniformly coated thin shells [36]. More recently, Xue et al. developed a procedure for silica-coating Ag nanoprisms in which the thickness of shells is controllable by changing the concentration of TEOS in the reaction mixture [37]. This approach uses surfactant-coated Ag nanoprisms, DMA, and an organic linker molecule, 16-mercaptohexadecanoic acid (MHA), as a substrate for the initiation of silica shell growth, though a minimum silica shell thickness of just ~15 nm was achieved on the nanoprisms. In this Letter, we adapt this procedure for use with citrate-reduced AgNPs to produce SCNPs of relatively uniform thickness, where the silica shell thickness is readily controlled from 8.6 ± 1.4 nm to 75.4 ± 4.8 nm by varying the concentration of TEOS in the reaction mixture.

Correlated fluorescence, transmission electron microscopy (TEM), and LSPR measurements of individual SCNPs coated in the probe molecule rhodamine B (RB) are used to investigate structure-activity relationships in monomers, dimers, trimers, and higher-order nanoaggregates. RB is chosen as the fluorophore since it absorbs at the 532-nm laser excitation wavelength and somewhat overlaps with the LSPR spectrum of SCNPs. Moreover, rhodamines have been used as sensitizers in DSSCs [38,39], such that RB-doped SCNPs (RB/SCNPs) are a reasonable model system for understanding dye–metal interactions in plasmon-enhanced photovoltaics. Although plasmon-enhanced fluorescence is most significant for weak absorbers and poor emitters, high quantum yield molecules can also exhibit substantial fluorescence enhancements [33]. We find that plasmon-enhanced fluorescence is not correlated to LSPR wavelength, consistent with recent SERS studies that demonstrate the far-field Rayleigh scattering of nanoaggregates does not directly relate to their near-field EM enhancement effects [30,40]. However, adding more hot spots to the SCNPs results in a modest increase in fluorescence enhancement. Furthermore, the highest fluorescence enhancement is observed for RB/SCNPs with ~19-nm thick shells. The combination of optical and structural measurements on individual SCNPs reveals the full distribution of structure-activity relationships in these hot-spot-containing nanostructures.

2. Experimental

2.1. Materials

Ultrapure water (Barnstead Easypure II System, Thermo Scientific, 18.2 MΩ/cm) is used for all experiments. Concentrated HCl, HNO_3, sodium citrate, and ethanol were purchased from Fisher Scientific. Silver nitrate ($AgNO_3$, 99.99%) and 16-mercaptohexadecanoic acid (MHA) were obtained from Sigma–Aldrich. A 40-wt% aqueous solution of dimethylamine (DMA) and rhodamine B (RB, 99+%) were purchased from Acros Organics. Tetraethoxysilane (TEOS, 99.9%) was obtained from Alfa Aesar.

2.2. Synthesis of SCNPs

All glassware was cleaned with aqua regia and rinsed thoroughly with deionized water. Citrate-reduced AgNPs were synthesized using the Lee and Meisel method [41]. Briefly, 500 mL deionized water was added 90 mg $AgNO_3$ with heating and stirring. Once the solution came to a rapid boil, 10 mL of a 1% sodium citrate solution was added, and the solution was boiled for 30 min. The resulting silver colloidal suspension was allowed to cool, diluted with deionized water to a final volume of ~1 L, and stored in the dark. All AgNPs were used within two weeks of synthesis for optimal results during silica encapsulation.

The reaction scheme for silica coating the citrate-reduced AgNPs is shown in the Supplementary material. AgNPs (~1.3 mL) were sonicated in sterile 1.5 mL microcentrifuge tubes (Fisher Scientific) for 30 min. Ethanolic solutions of MHA were added to a final concentration of 20 μM. Solutions incubated in MHA overnight and then centrifuged for 10 min at ~2000 rpm to concentrate nanoparticles and remove excess ligand. After removal of the supernatant, the nanoparticles were re-suspended in ~14 mL ethanol and sonicated for 30 min. Next, various amounts of TEOS in ethanol (i.e., 3.35, 2.51, 1.50, 1.32, 1.02, 0.95 or 0.84 μL) were added to the stirring nanoparticle solutions to obtain final TEOS concentrations of 1.0, 0.75, 0.45, 0.40, 0.30, 0.28 and 0.25 mM, respectively. A 1.139 mL aliquot of aqueous DMA was rapidly injected into the colloid solution (final [DMA] = 0.6 M), and the solution was allowed to stir for approximately 3 h. Finally, the solution was sonicated for 30 min and centrifuged for 10 minutes at ~2000 rpm to concentrate and purify the resulting SCNPs. Ensemble-averaged spectroscopy of SCNPs and RB were performed on UV/vis (Perkin Elmer, Lamda 35) and fluorescence (Perkin Elmer, LS55) spectrophotometers.

2.3. Sample preparation

The centrifuged SCNPs were dispersed in deionized water (~1 mL), sonicated for 30 min, and 6 μL of the resulting solution was drop coated onto a TEM grid (Ted Pella, Copper Formvar 300-mesh, Carbon Type B). After drying, the TEM grid is immobilized on a clean glass coverslip (Fisher Scientific, 25 × 25 mm) for spin coating by placing the grid edges on 2 mm sized pieces of mounting putty (Loctite) and lightly pressing down with tweezers (Ted Pella, PELCO Reverse Fine 5376-NM) to ensure attachment. Next, 50 μL of 10^{-4} M RB in ethanol was spun coat (WS-400-6NPP-LITE, Laurell Technologies) onto the grid using a custom spin-coating program: 15 s at 500 rpm, 45 s at 1500 rpm, and 15 s at 3000 rpm. Grids were allowed to dry overnight before fluorescence measurements and subsequent TEM analysis.

2.4. Single-nanoparticle LSPR and fluorescence measurements

TEM grids containing SCNPs were placed on clean glass coverslips atop a nano-positioning piezoelectric stage (Queensgate, NPS-XY-100B) on an inverted microscope (Nikon Eclipse, Ti-U) equipped with a dark-field condenser (Nikon, Dry NA = 0.95–0.80) and a variable numerical aperture (NA) 100 × oil-immersion objective (Nikon Plan Fluor, NA = 0.5–1.3). The objective was set to NA = 0.5 to collect the Rayleigh scattering from individual SCNPs following white-light illumination. The dark-field Rayleigh scattering,