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Metal-Enhanced Fluorescence from CdTe Nanocrystals: A Single-Molecule Fluorescence Study

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Signal detection is a challenging task in chemistry and biology. Most often, background noise hinders the detection of the signal of interest. This can be prevailed if one has a signal enhancing and/or directing technique. In this regard, Metal-Enhanced Fluorescence (MEF) is a newly recognized technology, where we study the interactions of fluorophores with metallic colloids or surfaces, which provides fluorescence enhancement.[1] Along with signal enhancement, MEF also provides several other important spectral changes, such as increased photostability, decreased lifetime due to increased rates of radiative decay, and increased distance for fluorescence resonance energy transfer (RET).[1c] Because of this exceptional improvement in signal detectability provided by the MEF, we were able to increase the intrinsic DNA fluorescence, which is otherwise impractical to perceive.[1d] Accordingly, we have made considerable progress with MEF studies, where we used wide varieties of fluorophores that can be potentially used in different applications.[2]

Single-molecule fluorescence spectroscopy provides several advantages over ensemble measurements.[3] For instance, it eliminates averaging of the spectral properties over all members of the ensemble and can reveal fundamental features otherwise masked in ensemble experiments. Accordingly, using single-molecule fluorescence studies, we anticipate that the metal−fluorophore interactions in the MEF studies can be better revealed, especially at the single-molecule level. For this, we choose CdTe nanocrystals (QDots) as fluorophores. QDots having few nanometer sizes, tunable absorption, and fluorescence spectral properties are widely used as probes in various fields, including biological labeling and imaging.[4] Potential applications of QDots are rapidly increasing. Single QDot fluorescence spectroscopy has been accomplished successfully for several colloidal semiconductor materials, including CdSe,[5a] CdS,[5b] CdTe,[5c] Si,[5d] and InP.[5e] Additionally, quantum dot fluorescence spectral properties are reported in the presence of metallic nanoparticles at the ensemble level with little details.[6,7]

In this communication, we describe the ensemble and single-molecule fluorescence spectral properties of CdTe nanocrystals spin coated in polyvinyl alcohol (PVA) matrix on glass and silver island films (SIFs). CdTe nanocrystals were prepared using a modified Weller method (for further details, see Supporting Information).[8] Figure 1 shows the fluorescence emission spectra and corresponding intensity decays of CdTe nanocrystals on glass and SIF surfaces. Interestingly, the emission spectra from metalized and that from nonmetalized areas are completely overlapped on each other with a band maxima at about 660 nm (Figure 1A inset). As seen from the figure, a significant enhancement (∼5-fold) in fluorescence intensity is observed from the CdTe nanocrystals on the SIF surface as compared to that on the glass. On the other hand, the fluorescence intensity decay of CdTe nanocrystals on SIFs show a faster decay with ∼3-fold decrease (from 4.52 ns on glass to 1.45 ns on SIFs) in average fluorescence lifetime. These anomalous fluorescence properties, such as increase in fluorescence intensity and decrease in lifetime of CdTe nanocrystals near metallic surface, can be reasonably explained as below.

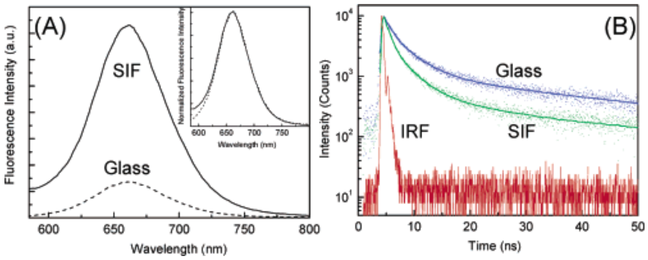

Figure 1. (A) Fluorescence spectra of CdTe on glass and SIF. Inset shows the normalized emission spectra. (B) Intensity time decay of CdTe on glass and SIF. Instrument response function (IRF) is also included.

The emission of fluorophores near the silver nanostructures is dependent on at least two factors: an enhanced local field and an increase in the intrinsic decay rate of the fluorophore, both due to MEF. The first factor provides stronger excitation rates, but does not modify the fluorescence lifetime of the molecules. The second factor changes the quantum yield and lifetime of the fluorophore.[1] Results shown in Figure 1 are in accordance with these arguments. The radiative decay rate modification of the CdTe nanocrystals in close proximity to the silver nanostructures is consistent with the previous reports on the MEF phenomenon and is due to the proximity of the fluorophores to the silver nanostructures.

Single CdTe nanocrystal fluorescence measurements were obtained using a scanning confocal microscope (Picoquant MicroTime 200). The excitation laser was reflected by a dichroic mirror to a high numerical aperture (NA) oil objective (100x, NA 1.3) and focused to a diffraction limited spot (∼300 nm) on the sample surface. CdTe fluorescence was collected by an avalanche photodiode through the dichroic beam splitter and a band-pass (650−720 nm, Chroma) filter. Integration times of 3 ms per pixel were used to obtain 512 × 512 pixel raster scanned 20 × 20 μm images. The samples were excited with a typical power density of ∼55 W/cm^2 from a 470 nm solid state laser. Intensity time trajectories and intensity time decays were obtained by positioning the excitation beam above the individual CdTe.

Figure 2 shows representative scanning confocal images of CdTe nanocrystals spin coated on glass and SIF surfaces. Well separated bright spots represent fluorescence emission from the single CdTe nanocrystals. The significant differences in the peak intensities of the two images are immediately evident from the figure (see the scale bar). For CdTe nanocrystals on the glass and SIF surfaces, the average values of peak intensities are ∼25 and 125 counts/bin, indicating single CdTe nanocrystals on the SIF surface are ∼5-fold brighter than that on the glass. This brightness of the spots can be ascribed to our so-called MEF.[1] Also, it is pertinent to note the heterogeneity in the spots' brightness. On glass surface, the

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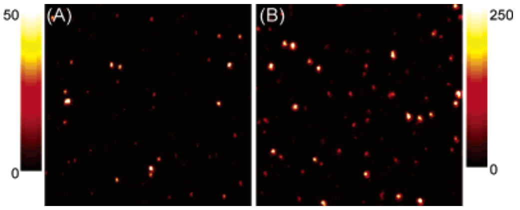

Figure 2. Scanning confocal images (20 × 20 μm) of CdTe on (A) glass and (B) SIF. Scale bar shows the intensity counts in 3 ms bin.

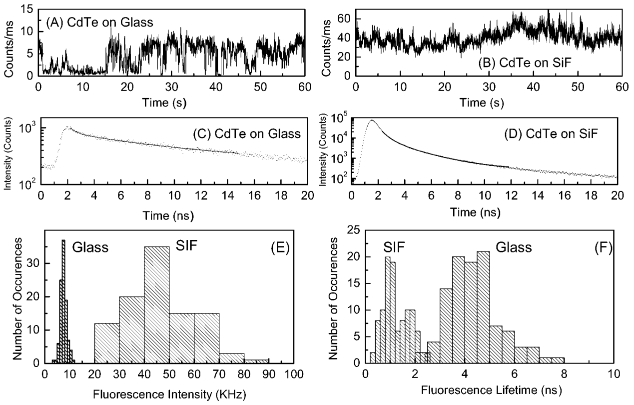

Figure 3. Intensity time trajectories (A and B), intensity decays (C and D) of individual CdTe on glass and SIFs. Fluorescence intensity (count rate) (E) and average fluorescence lifetime (F) histograms of CdTe on glass and SIF surfaces.

observed heterogeneity in the brightness could be ascribed simply to the size-dependent emission efficiencies of CdTe nanocrystals, which is an inherent property of QDots.[5c] However, explaining this on the SIFs' surface is complex, where the metal fluorophore interactions also play a dominant role (vide infra).

To more quantitatively compare the collected single-molecule count rates and explore the changes in underlying photophysics of individual nanocrystals on glass and SIF surfaces, we monitored the fluorescence intensity of individual nanocrystals as a function of time, while under continuous excitation. Accordingly, we monitored fluorescence intensities of over 100 CdTe nanocrystals on both glass and SIF surfaces. Representative intensity time trajectories collected for CdTe nanocrystals on glass and SIF surfaces are shown in Figure 3, top panel. From these intensity time trajectories, one can deduce various spectral features. At first, the fluorescence intensity of individual CdTe nanocrystals on SIFs is several-fold higher than that on the glass. Second, the CdTe nanocrystals show characteristic blinking phenomenon on the glass surface (Figure 3A).[4,5c] On the other hand, the blinking is almost eliminated on SIF surfaces, which results in a more constant brightness of the CdTe nanocrystals on SIF surfaces. These results suggest that the CdTe nanocrystals show reduced blinking on SIF surfaces. Figure 3 (middle panel) shows the fluorescence intensity decays of single CdTe nanocrystals on both surfaces studied. Similar to that observed in ensemble measurements, the CdTe nanocrystals show longer decay times on glass surface compared to that on the SIF surfaces.

Figure 3E and F shows the CdTe fluorescence intensity and lifetime histograms collected on glass and SIF surfaces. Within the same experimental conditions, the fluorescence intensities of single CdTe nanocrystals on glass are ~8 kHz and that on SIF surfaces are ~45 kHz, which imply ~5-fold increase in fluorescence intensity on SIFs that is similar to that observed in ensemble measurements (Figure 1). From these histograms, it is clear that the fluorescence intensities as well as lifetimes of individual CdTe nanocrystals on SIF surfaces are more heterogeneously distributed than that on glass surfaces. Such a distribution phenomenon is impossible to observe with ensemble measurements. CdTe fluorescence intensity distributions on glass surfaces show relatively low standard deviation of 16%, where as the corresponding value on SIFs is ~30%. This is because the heterogeneity in the fluorescence intensity on the glass surface largely depends on deviation in the CdTe nanocrystals' size, whereas for SIFs, additional complexity arises from the varied metal−fluorophore interactions that may be due to the heterogeneous surface of SIFs. A similar heterogeneous distribution for the CdTe lifetimes is also observed (Figure 3F) on SIFs (SD ~ 43%) compared to that on glass (SD ~ 24%).

In conclusion, in this communication, we reported, for the first time, the metal-enhanced fluorescence from the CdTe nanocrystals spin coated on SIF surfaces. CdTe nanocrystals show ~5-fold increase in fluorescence intensity, 3-fold decrease in lifetimes, and reduction in blinking on SIF surfaces that can be observed by ensemble and single-molecule fluorescence studies. The single-molecule study also provides further insight on the heterogeneity in the fluorescence enhancement and lifetimes of the CdTe nanocrystals on both glass and SIF surfaces, which is otherwise not possible to observe using ensemble measurements. We believe single-molecule study can reveal further information on the metal−fluorophore interactions to better understand the MEF when we use more defined sample and surface conditions. Further work in this direction is under consideration.

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Supporting Information Available: Synthetic procedure and spectral properties of CdTe nanocrystals in water. This material is available free of charge via the Internet at http://pubs.acs.org.

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