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Fluorescent Lifetime Quenching near $d = 1.5$ nm Gold Nanoparticles: Probing NSET Validity

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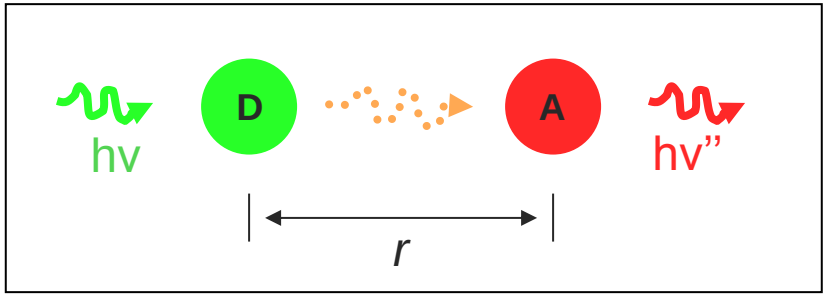
Abstract: The fluorescence behavior of molecular dyes at discrete distances from 1.5 nm diameter gold nanoparticles as a function of distance and energy is investigated. Photoluminescence and luminescence lifetime measurements both demonstrate quenching behavior consistent with $1/d^4$ separation distance from dye to the surface of the nanoparticle. In agreement with the model of Persson and Lang, all experimental data show that energy transfer to the metal surface is the dominant quenching mechanism, and the radiative rate is unchanged throughout the experiment.

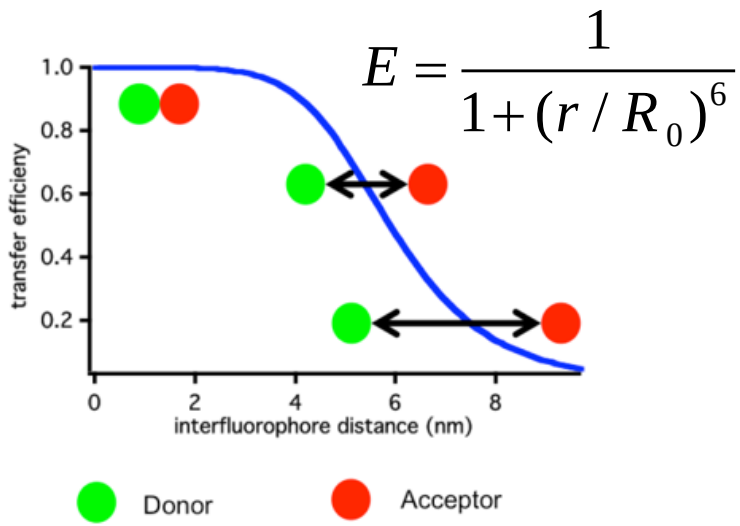
CONTENTS

FRET(Foster Resonance Energy Transfer)



$$E(r) = \frac{1}{1+(r/R_0)^6}$$

E : FRET efficiency
R_0 : where E become 50%

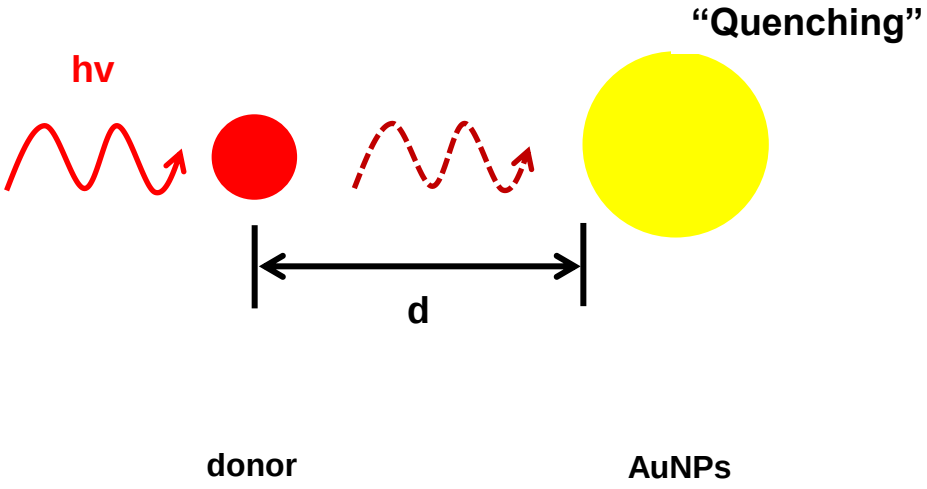


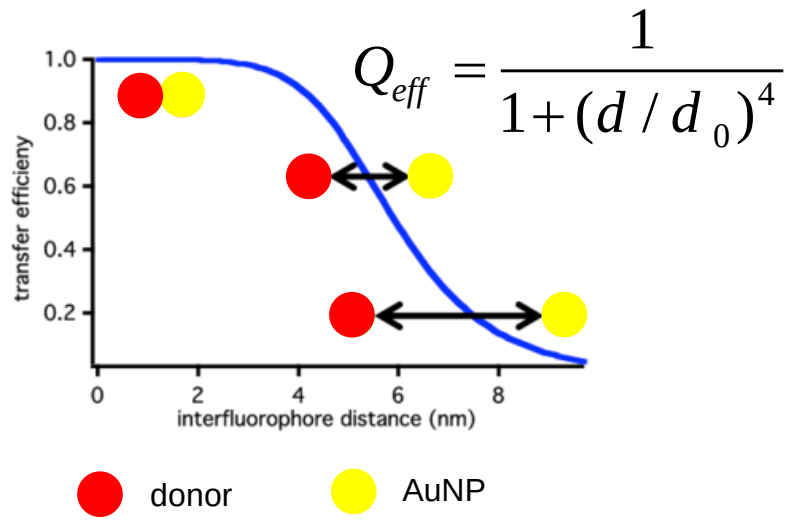
Sensitive in nanometer scale Real-time observation

FRET efficiency is change by distance change between donor and acceptor

This analysis of the dynamics of dye molecules
For example, we can analyze the Folding and unfolding process of DNA

NSET(Nanometal Surface Energy Transfer)





$$Q_{eff} = \frac{1}{1+(d/d_0)^4}$$

Q_{eff} : quenching efficiency
d_0 : where Q_{eff} become 50%

NSET is similar to Förster resonance energy transfer (FRET)

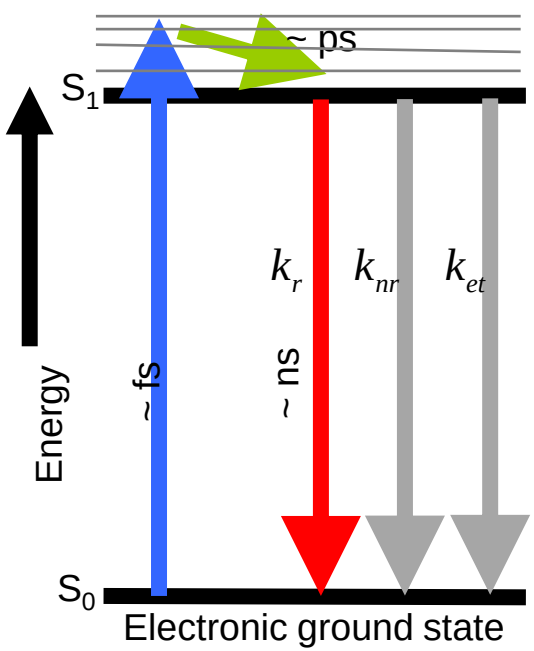
however, the measurable distances are extended nearly 2-fold for optical molecular rulers by following a $1/d^4$ distance dependence.

Quenching : coupling of the oscillating electronic dipole of a dye to a metal surface with loss of energy

Introduction

The relationship between photoluminescence (PL) intensity and lifetimes is easily shown by considering that

$$\tau_{obs} = \frac{1}{k_{obs}} = \frac{1}{k_r + k_{nr} + k_{et}} \qquad \Phi_{em} = \frac{k_r}{k_r + k_{nr} + k_{et}} = k_r \tau_{obs}$$



τ_{obs} : inverse of all rates of decay
k_r : radiative rates
k_r : nonradiative rates
k_{et} : the rate of energy transfer
Φ_{em} : quantum yield

The radiative and nonradiative rates are normally considered constants for a dye under defined conditions, leaving k_{et} as the major contributor to the shortening of an observed lifetime.

$$k_{et} = \frac{1}{\tau'_{obs}} - \frac{1}{\tau_{obs}}$$

Setup

Absorption measurements

Varian Cary 50 UV-vis spectrophotometer

using 50uL quartz cuvettes

Photoluminescence measurements

Varian Cary Eclipse Fluorescence spectrophotometer

Lifetime Measurements

Sample

Result

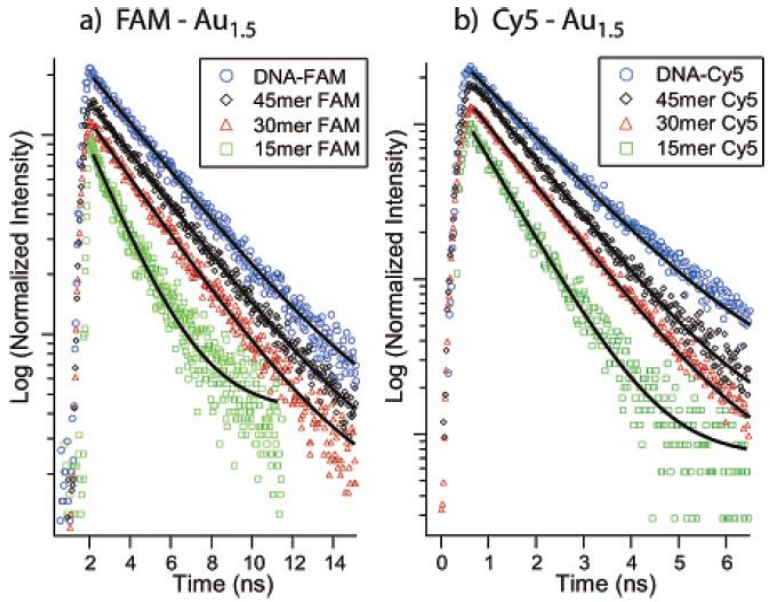


Figure 2. Lifetimes of the 15 bp (green), 30 bp (red), and 45 bp (black) NP−dsDNA−dye assemblies for FAM (a) and for Cy5 (b) relative to dsDNA−dye controls (blue, top). The data have been normalized and offset vertically for viewing. Single-exponential fits through the data are shown (—).

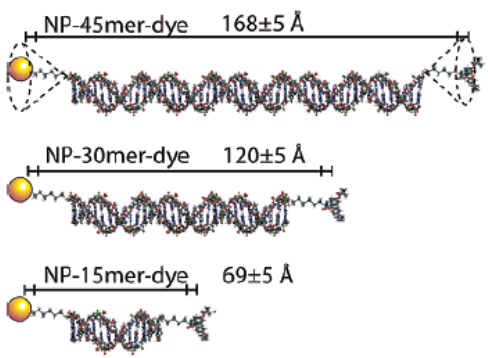


Figure 1. Scheme of DNA binding to a 1.5 nm Au NP. By varying the length of the DNA strand, the terminal dye fluorophore is separated from the Au NP by discrete distances (168, 120, and 69 Å.)

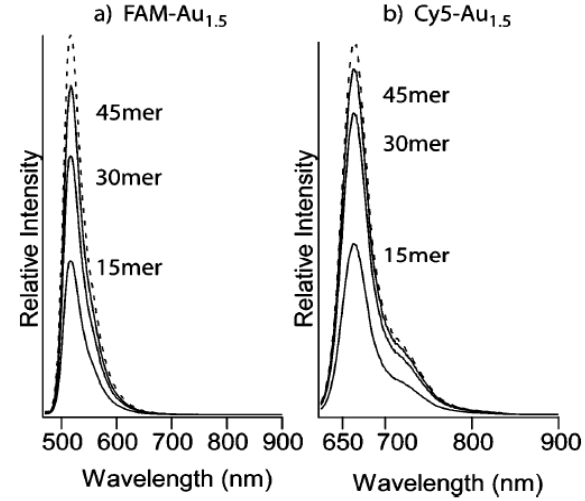


Figure 3. (a) Photoluminescence quenching of FAM PL intensity as a function of dsDNA spacer length. (b) Photoluminescence quenching of Cy5 dye as a function of spacer length. The top curve in both (a) and (b) (- - -) is a normalized control intensity.

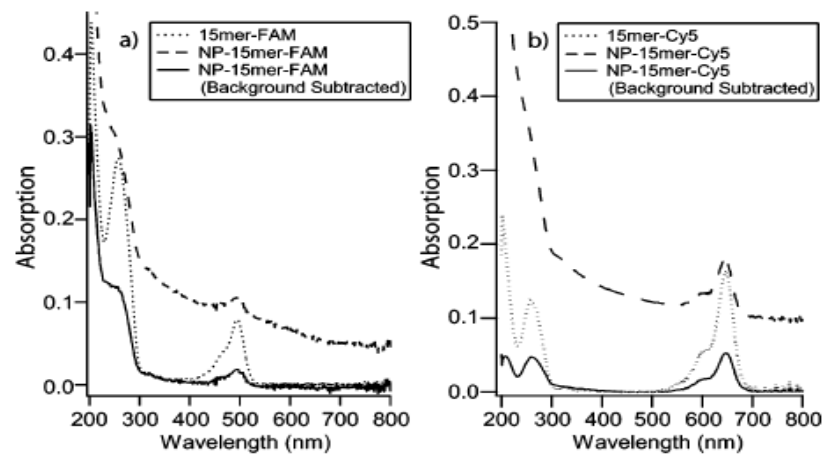


Figure 4. (a) Absorption and corrected spectra for the FAM−dsDNA−NP system and (b) absorption spectra and corrections for Cy5−dsDNA−NP. These spectra compare purified dye−dsDNA−NP (black, offset 0.05 au−FAM: 0.10 au−Cy5), dye−dsDNA in buffer (· · ·), and the background subtracted absorption spectrum of the dye−dsDNA−NP to correct for NP absorption (—).

Result

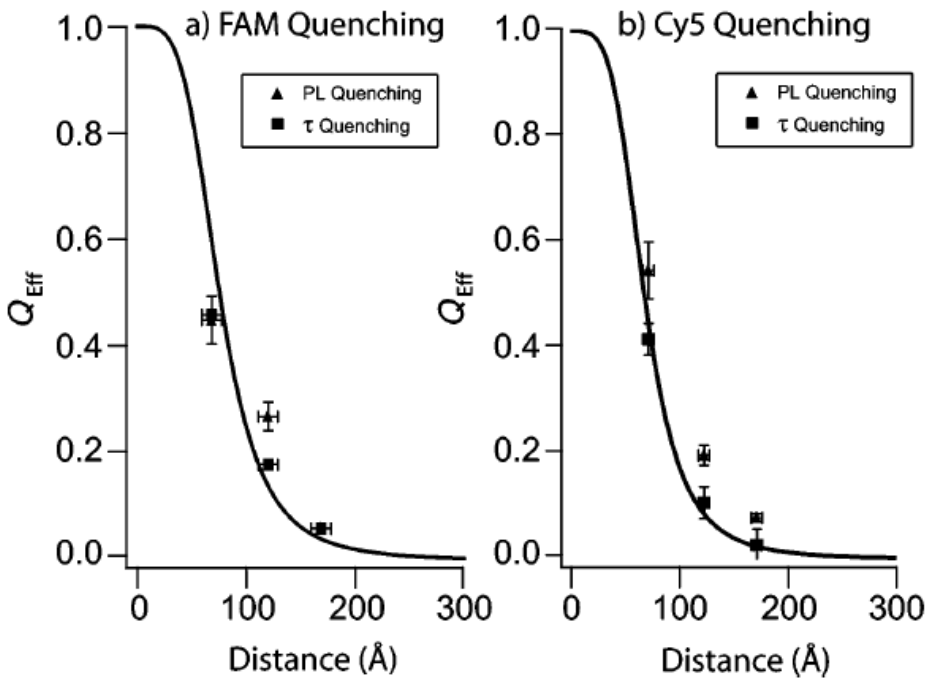


Figure 6. Quenching data for FAM (a) and Cy5 (b) based upon photoluminescence (▲) and lifetimes (■) overlaid on top of a theoretical curve generated using eq 9.

$$Q_{eff} = \frac{1}{1+(d/d_0)^4}$$

the process of quenching is an energy transfer event and that it follows a $1/d^4$ distance dependence.

Result

Monitoring K_r

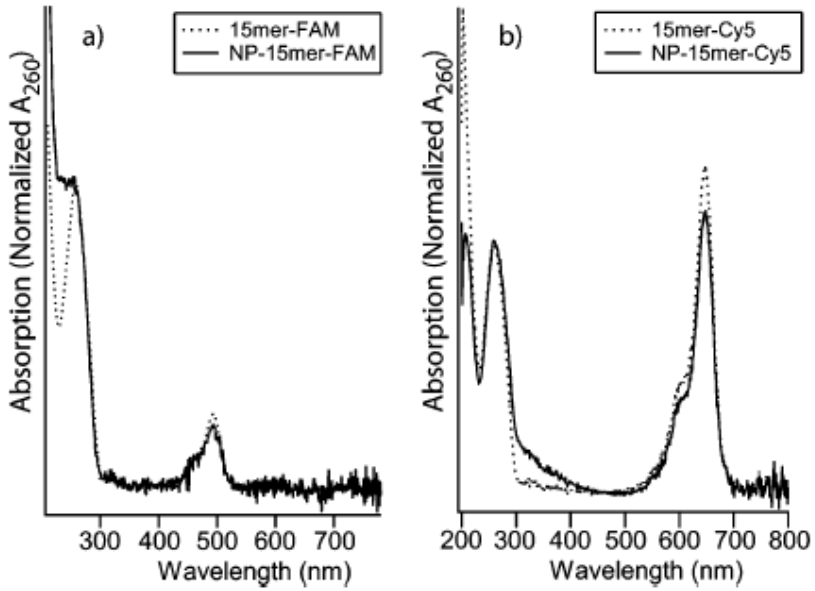


Figure 7. Absorption comparison of the dye−15mer (w/o NP, · · ·) to the dye−15mer−NP absorption, after subtracting gold absorption and correcting for scattering (—). All absorbances have been normalized at the DNA absorption wavelength, 260 nm. A small scattering correction has been applied to the NP−15mer−FAM absorption to correct the baseline from the 200−450 nm range. Absorption comparison for both FAM−15mer−NP (a) and Cy5−15mer−NP (b) allows direct monitoring of the oscillator strength for the dye at the closest proximity to the gold NP measured here.

possibility of radiative rate (k_r) changes is garnered by considering the changes in the absorption intensity or oscillator strength (*f*) for the dye molecule upon binding the gold NP

Oscillator strength, f, is directly related to the radiative rate, k_r

$$k_r^0 = 3\times10^{-9}\bar{\nu}_0^{-2}\int \varepsilon d\bar{\nu} \cong \bar{\nu}_0^{2} f$$

$\bar{\nu}_0$: energy in wavenumbers corresponding to the maximum absorption

ε : experimental extinction coefficient

changes in the oscillator strength were calculated to be 5-10%, which does not account for the observed 50-70% drop in PL intensity

Conclusion

three different lengths of dsDNA-dye, using two dyes of different energies, were appended to 1.5 nm gold NPs as a means of measuring quenching efficiency of the fluorophore at discrete distances

Absorption data on the fluorophore are forthcoming in proving a mechanism which does not rely upon a changing radiative rate for this system.

The quenching was measured by cw-PL and by picosecond lifetime spectroscopy to determine that the process of quenching is an energy transfer event and that it follows a $1/d^4$ distance dependence.