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Letter

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Quantum Yield Measurement in a Multicolor Chromophore Solution Using a Nanocavity

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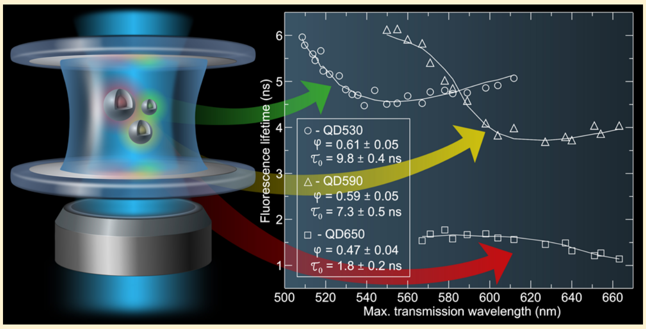
ABSTRACT: Using our nanocavity-based method for precise determination of the absolute quantum yield values, we measure the quantum yield of each of the three different chromophores types with strongly overlapping absorption spectra in a multicolor solution, which is impossible to do by any other existing technique. By measuring the modification of the radiative rate of the chromophores at different cavity lengths, we extract the quantum yield of CdSe/ZnS semiconductor nanocrystals emitting at three different wavelengths and excited by a single wavelength in the multicolor solution. Using several detection channels, we show that only one measurement is needed for determining the quantum yield values of all the types of chromophores in the sample. The obtained results show a very good agreement with the values commonly observed using other techniques. The nanocavity-based method opens up new opportunities of determining fluorescence quantum yields in complex systems and can be used for a big variety of fluorescence studies where multichromophore samples are involved.



KEYWORDS: *Quantum yield, CdSe/ZnS, quantum dots, nanocrystals, cavity, radiative rate*

Multicolor fluorescent complexes are widely used for biological labeling,[1] Förster resonance energy transfer spectroscopy,[2] and engineering of solar cells[3] and white-light sources.[4] The fluorescence quantum yield (QY), which is the ratio of the number of photons emitted to the number of photons absorbed, is one of the key photophysical properties of fluorescent species. It has recently been shown that the QY of a chromophore strongly depends on the local chemical environment.[5] Therefore, disentangling the QY of each type of chromophore within a multicolor sample could provide new insight into the complex photophysics of the sample. However, all of the existing techniques of QY determination will fail to simultaneously measure the different QY values in a multicolor sample, in particular when the absorption spectra of the different types of chromophores strongly overlap.[6−8]

We have recently introduced a nanocavity-based reference-free method of determining the fluorescence QY, which requires only few microliters of low-concentrated chromophore solution,[9,10] or even only a single emitter.[5] Placing a chromophore inside the cavity changes the coupling of its dipole transition to the electromagnetic field, thus leading to a modification of the radiative transition rate in a chromophore.[11−13] The measured fluorescence lifetime is then given by the inverse sum of radiative (k_{rad}) and nonradiative (k_{nr}) decay rates:

$$\tau = (k_{rad} + k_{nr})^{-1} \tag{1}$$

The QY, defined as the average chance to emit a photon upon return from the excited to the ground state, is quantitatively described by the ratio of radiative to nonradiative transition rates

$$\Phi = \frac{k_{rad}}{k_{rad} + k_{nr}} \tag{2}$$

As the cavity modifies only the radiative rate of the embedded emitter leaving the nonradiative rate unaffected, the QY can be determined by monitoring the modification of the fluorescence lifetime of a chromophore at different cavity lengths.

Here, we show that our technique allows for precise QY determination for each type of chromophore in a multicolor solution where the absorption spectra of different types of the chromophores strongly overlap. We measure the QY of CdSe/ZnS core−shell semiconductor quantum dots (QDs), emitting at three different wavelengths in the multicolor solution. The absorption spectra of the QDs are strongly overlapping, so that they can all be excited by a single wavelength in the blue spectral region. This makes their mixture a perfect test sample for our method. By measuring the modification of the radiative rate of the QDs at different cavity lengths, we extract both the QY and the free space fluorescence lifetime of each of the three types of chromophore. We demonstrate that by using several detection channels, only one measurement is needed for determining the QY values of all three types of chromophores in solution. The obtained results show a very good agreement

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with the QY values commonly observed using other methods. As the nanocavity-based method can be applied to any quantum emitter of interest, such as dye molecules, semiconductor nanoparticles, carbon nanotubes, etc., determination of chromophore QYs incorporated into multicolor samples opens up new perspectives for studying the photophysics of complex systems. Because a detailed description of the theoretical model and the measurement procedure can be found elsewhere,[10] we will skip these details in this letter and will focus on the obtained results.

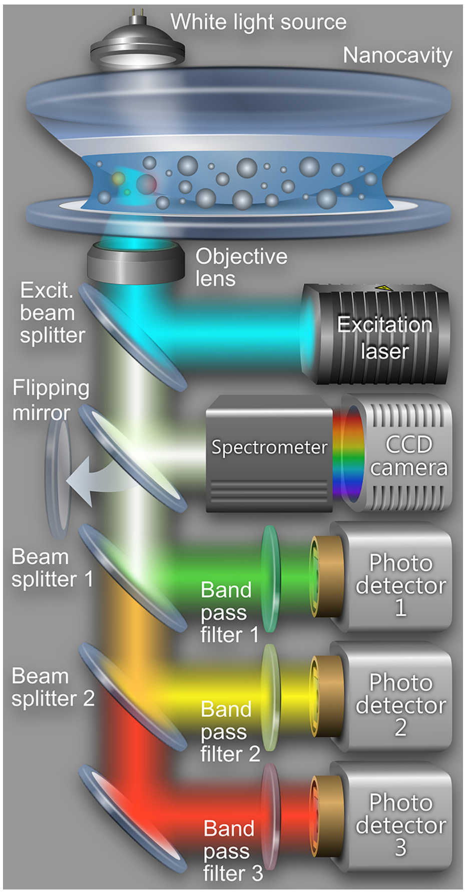

Figure 1. Scheme of the experimental setup. The nanocavity consists of the toluene solution of CdSe/ZnS quantum dots emitting at 530, 590, and 650 nm placed between two silver mirrors. The fluorescence is split between three detection channels. The distance between the cavity mirrors is determined by recording the white light transmission spectra.

Figure 1 shows a schematic of the home-built confocal microscope and the nanocavity. The nanocavity consists of two silver mirrors with subwavelength spacing. The bottom silver mirror (30 nm thick) was prepared by vapor deposition onto commercially available and cleaned microscope glass coverslides (thickness 170 μm) using an electron beam source Laybold Univex 350 under high-vacuum conditions ($\sim 10^{-6}$ mbar). The top silver layer (85 nm thick) was prepared by vapor deposition of silver onto the surface of a plano-convex lens (focal length of 150 mm) under the same conditions. Film thickness was monitored during vapor deposition using an oscillating quartz unit and verified by atomic force microscopy. The spherical shape of the upper mirror allowed us to reversibly tune the cavity length by moving the cavity laterally with respect to the excitation focus with a piezo nanopositioning stage (PI, P-562). Because of the very low curvature of the lens, the nanocavity can be considered to be a plane-parallel resonator across the focal spot of the microscope objective lens.[14] The cavity length was determined by measuring the white-light transmission spectrum[12,14] using a spectrograph (Andor SR 303i and a CCD camera Andor iXon DU897 BV) and by fitting the spectra with a standard Fresnel model of transmission through a stack of plano-parallel layers, where the cavity length (distance between silver mirrors) was the only free fit parameter.

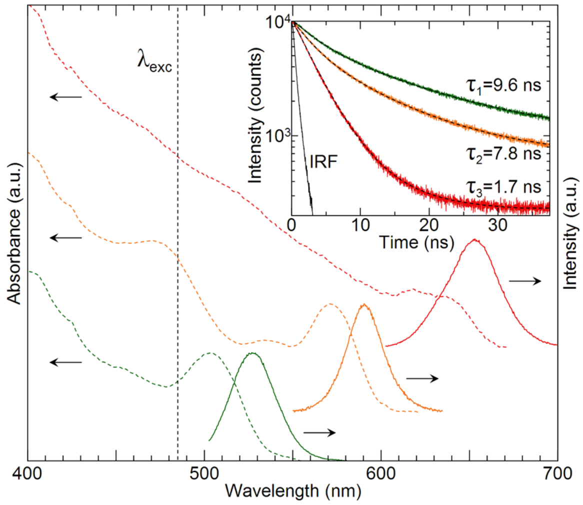

Figure 2. Room temperature fluorescence (solid curves) and absorption spectra (dashed curves) of CdSe/ZnS nanocrystals dispersed in toluene on a clean glass cover slide. The vertical dashed black line indicates the excitation wavelength of 485 nm. Inset: colored curves represent the fluorescence decays of the nanocrystals exhibiting emission maxima at 530 nm (τ_1), 590 nm (τ_2), and 650 nm (τ_3). Dashed lines show the best fit to the experimental data. Solid black curve shows the instrument response function (IRF).

We studied CdSe/ZnS core−shell quantum dots purchased from PlasmaChem, soluble in toluene and showing emission maxima at 530 nm (QD530), 590 nm (QD590), and 650 nm (QD650) (Figure 2). For the fluorescence measurements, a droplet of a micromolar solution of the mixture of the QD530, QD590, and QD650 quantum dots dispersed in toluene was placed between the cavity mirrors. The fluorescence emission was split between three photodetectors by Semrock BrightLine FF580-FDi01 and FF640-FDi01 dichroic mirrors. The back-scattered excitation light was blocked with band-pass filters (Semrock BrightLine FF01-525/45, FF01-593/40, and FF01-641/75) for separately detecting the fluorescence from the QD530, QD590, and QD650, respectively.

A white light laser system (Fianium SC400-4-20) with a tunable filter (AOTFnC-400.650-TN) served as excitation source (λ_{exc} = 485 nm). The light was reflected by a dichroic mirror (Semrock BrightLine FF484-FDi01) toward the objective lens (Apo N, 60×/1.49 NA oil immersion, Olympus). Collected fluorescence was focused onto the active area of avalanche photodiodes (PicoQuant τ-SPAD) for the QD530 and QD650 emission and on a single-photon detection module (MPD series) for the QD590 emission. Data acquisition was accomplished with a multichannel picosecond event timer (PicoQuant HydraHarp 400). Photon arrival times were histogramed (bin width of 50 ps) for obtaining fluorescence decay curves, and all curves were recorded until reaching 10^4 counts at the maximum. Finally, the fluorescence decay curves were fitted with a multiexponential decay model, from which the average excited-state lifetime was calculated:

$$\langle \tau \rangle = \int_0^\infty F(t)t \, \mathrm{d}t \Big/ \int_0^\infty F(t) \, \mathrm{d}t \qquad (3)$$

After placing a droplet of multicolor solution of quantum dots inside the nanocavity, we recorded the fluorescence decay curves of QD530, QD590, and QD650 as a function of cavity lengths. By using three detection channels, we recorded all three decay curves simultaneously for each cavity length, which reduced the total measurement time by a factor of 3. Measurements were performed in a range of white light transmission maxima between 500 and 670 nm, covering the fluorescence emission bands of the selected quantum dots (Figure 2). As all measurements were done at micromolar concentrations, one can neglect any modification of the QY due to interaction between the emitters.

Figure 3 shows the results of the cavity-controlled fluorescence lifetime measurements for QD530 (circles), QD590 (triangles), and QD650 (rectangles) as a function of the cavity's maximum transmission wavelength, which is linearly proportional to the cavity length. The curves show a chromophore-specific decrease of the lifetime values with increasing cavity length. The excitation and emission transition dipole moments of CdSe/ZnS quantum dots are randomly distributed in three[15,16] and two dimensions,[16−18] respectively, and are uncorrelated to each other. This allows for fitting the cavity-modulated lifetime using our previously developed model for linear dipole emitters in the limit of infinitely fast rotational diffusion.[10] The solid lines in Figure 3 show fits of the theoretical model to the experimental data, where the only free fit parameters were the free space lifetime τ_0, and the fluorescence QY value Φ of the chromophore.

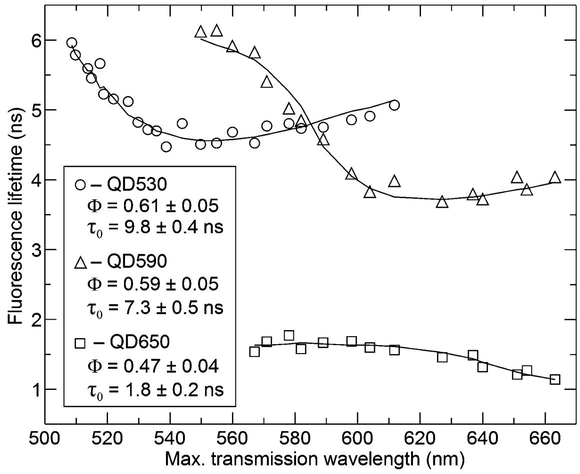

Figure 3. Fluorescence lifetime of CdSe/ZnS quantum dots with maximum emission wavelengths of 530 nm (circles), 590 nm (triangles), and 650 nm (rectangles) as a function of the cavity's maximum transmission wavelength. Solid lines show the best fit to the experimental data, giving the quantum yield (Φ) and free space lifetime (τ_0) values as indicated in the inset.

The inset in Figure 3 displays the determined values of the free space fluorescence lifetime and QY of the quantum dots QD530, QD590, and QD650 in the mixture. For determining the errors of the QY and free space lifetime values, we employed Efron's bootstrap algorithm.[19] The calculated values of the free-space lifetime are in good agreement with the lifetime values of the quantum dots measured in free space under identical conditions (see inset in Figure 2). The obtained QY values vary from 0.47 (QD650) to 0.61 (QD530), which is in good agreement with commonly observed values of 0.5−0.85 for the commercially available CdSe/ZnS quantum dots emitting in the visible spectrum.[20−22] The big deviation of values obtained in other studies is caused by the differences in synthesis and surface passivation of the quantum dots as well as the precision of the methods used for determining the QY. The error of our QY measurement does not exceed 0.05 and can be as small as 0.02 (inset in Figure 3). Whereas the nanocavity-based method requires only few microliters of low-concentrated chromophore solution, its precision exceeds most of the existing methods of QY determination.[22] Moreover, this experiment demonstrates that our technique can be used for the QY measurements of the samples with intrinsically multiexponential fluorescence decay. To unravel the inherent complexity of the photophysics of various multichoromophoric systems, the dedicated studies will be carried out in the near future. It should be also noted that although in this work the emission dipoles exhibited an infinitely fast rotational diffusion, the nanocavity-based method can be used in case of slow rotational diffusion[9] or even fixed dipoles,[5] which allows to determine the QY of the solid state samples, e.g., powders or thin films for characterization of optoelectronic devices.

In summary, we have shown that our nanocavity-based technique allows one to simultaneously measure the fluorescence QY of several types of chromophores with strongly overlapping absorption spectra within a mixture, which is impossible to be done by any other existing method. Our method is reference-free and requires only minute amounts of sample. The obtained values are in a good agreement with commonly reported values for commercially available quantum dots emitting in the visible spectrum. Moreover, we demonstrated that using several detection channels, only one measurement is needed for determining the QY values of all the different chromophores in solution. Thus, the nanocavity-based method opens up new opportunities of determining fluorescence QYs in complex systems and can be used for a large variety of fluorescence studies where multichromophore samples are involved.

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Notes

The authors declare no competing financial interest.

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■ REFERENCES

(1) Gao, X.; Cui, Y.; Levenson, R. M.; Chung, L. W. K.; Nie, S. *Nat. Biotechnol.* **2004**, *22*, 969−976.
(2) Wang, L.; Tan, W. *Nano Lett.* **2006**, *6*, 84−88.
(3) Huynh, W. U.; Dittmer, J. J.; Alivisatos, A. P. *Science* **2002**, *295*, 2425−2427.

(4) Qian, F.; Gradečak, S.; Li, Y.; Wen, C.-Y.; Lieber, C. M. *Nano Lett.* **2005**, *5*, 2287–2291.

(5) Chizhik, A. I.; Chizhik, A. M.; Khoptyar, D.; Bär, S.; Meixner, A. J.; Enderlein, J. *Nano Lett.* **2011**, *11*, 1700–1703.

(6) Demas, J. N. *Measurement of Photon Yields*; Academic Press: New York, 1982.

(7) Würth, C.; Grabolle, M.; Pauli, J.; Spieles, M.; Resch-Genger, U. *Anal. Chem.* **2011**, *83*, 3431–3439.

(8) Resch-Genger, U.; DeRose, P. C. *Pure Appl. Chem.* **2012**, *84*, 1815–1835.

(9) Chizhik, A. I.; Gregor, I.; Schleifenbaum, F.; Müller, C. B.; Röling, C.; Meixner, A. J.; Enderlein, J. *Phys. Rev. Lett.* **2012**, *108*, 163002.

(10) Chizhik, A. I.; Gregor, I.; Ernst, B.; Enderlein, J. *ChemPhysChem* **2013**, *14*, 505–513.

(11) Purcell, E. M. *Phys. Rev.* **1946**, *69*, 681.

(12) Chizhik, A.; Schleifenbaum, F.; Gutbrod, R.; Chizhik, A.; Khoptyar, D.; Meixner, A. J.; Enderlein, J. *Phys. Rev. Lett.* **2009**, *102*, 073002.

(13) Chizhik, A. I.; Chizhik, A. M.; Kern, A. M.; Schmidt, T.; Potrick, K.; Huisken, F.; Meixner, A. J. *Phys. Rev. Lett.* **2012**, *109*, 223902.

(14) Steiner, M.; Schleifenbaum, F.; Stupperich, C.; Failla, A. V.; Hartschuh, A.; Meixner, A. J. *ChemPhysChem* **2005**, *6*, 2190–2196.

(15) Chizhik, A. I.; Chizhik, A. M.; Khoptyar, D.; Bär, S.; Meixner, A. J. *Nano Lett.* **2011**, *11*, 1131–1135.

(16) Empedocles, S. A.; Neuhauser, R.; Bawendi, M. G. *Nature* **1999**, *339*, 126–130.

(17) Patra, D.; Gregor, I.; Enderlein, J.; Sauer, M. *Appl. Phys. Lett.* **2005**, *87*, 101103.

(18) Brokmann, X.; Coolen, L.; Dahan, M.; Hermier, J. P. *Phys. Rev. Lett.* **2004**, *93*, 107403.

(19) Efron, B. *Ann. Stat.* **1979**, *7*, 1–26.

(20) Kumar, C. *Semiconductor Nanomaterials*; Wiley-VCH Verlag GmbH & Co. KGaA: Weinheim, 2010.

(21) Hines, M. A.; Guyot-Sionnest, P. *J. Phys. Chem.* **1996**, *100*, 468–471.

(22) Grabolle, M.; Spieles, M.; Lesnyak, V.; Gaponik, N.; Eychmüller, A.; Resch-Genger, U. *Anal. Chem.* **2009**, *81*, 6285–6294.