

Figure 1: Left: Duetta 2-in-1 absorbance and fluorescence spectrometer; Right: DeltaPro fluorescence and phosphorescence lifetime fluorometer

Introduction

Quantum dots (QDs) are semiconducting spheres in the size typically in the range of 1 to 10nm. The size of these small spheres give quantum dots the semiconducting properties and resulting photoluminescence that would not necessarily occur for the same material on larger scales. QDs absorb over a broad range and have photoluminescence emission over a narrow range which can be tuned depending on the material from which the quantum dot is made and the size of the quantum dot. The photoluminescence follows the quantum confinement effect, where smaller quantum dots emit at higher energy (lower wavelength) and larger quantum dots emit at lower energy (higher wavelength). The materials from which quantum dots are made range from CdS, CdSe, ZnS, ZnSe, PbS, and PbSe to combinations of these in core-shell compositions. (B.O. Dabbousi, 1997) (Murphy, 2002) The applications of quantum dots continues to be an expanding field. (Lin Cui, 2018) The materials that make up quantum dots typically have cytotoxicity so the nanocrystals can be coated in biocompatible polymer (PEG) or other materials to be used in applications such as cellular tracking. (Yun-Pei Chang, 2008). These materials have high extinction coefficients and high quantum yields, making them easily visible for spectroscopy applications and medical imaging. The small size makes QDs more soluble in some solutions so it is possible to print QDs into inks and coatings for films. The potential for use of QDs in photovoltaic devices is not new, but is on the rise, using the tunability of these nanoparticles for improvements in solar cells, LEDs, and other optoelectronic devices. (Lin Cui, 2018) (Duggan, 1990) (Zhinjun Ning, 2014)

There are many ways to analyze quantum dots, but fluorescence detection is probably one of the most common techniques as quantum dots are used for imaging applications in cell biology. In this application note, Qtracker® 655 quantum dots are analyzed using fluorescence spectroscopy, UV-Vis absorbance spectroscopy, and fluorescence lifetime measurements. These types of spectroscopy may analyze synthetic quality or differences in novel QD materials or the processes for synthesizing novel materials and even quality control for manufacturing QDs. The Duetta and DeltaPro benchtop instruments from HORIBA Scientific make it easy to analyze QDs spectroscopically.

Methods and Results

Qtracker® 655 quantum dots were obtained from Thermofisher Scientific¹. The stock solution was diluted 100x in PBS buffer and then sonicated for 5 minutes. The solution was then diluted 4x so that the total dilution was 400x. All samples were measured in 1-cm quartz cuvettes.

A-TEEM™ Fluorescence

The absorbance-transmittance excitation emission matrix (A-TEEM™) was collected for a 400x dilution of QTracker® 655 quantum dots in PBS buffer on a Duetta 2-in-1 absorbance and fluorescence spectrometer from HORIBA Scientific. A solution of clean water was used for the blank sample. The excitation wavelength was scanned from 300-700nm with 5 nm step increments, and the emission was collected from 300-800nm with 0.1 sec integration time on the CCD. The band pass was set at 3nm for both

¹<https://www.thermofisher.com/order/catalog/product/Q25021MP>

excitation and emission optical paths. The blank excitation emission matrix (EEM) EEM was collected using the water sample and this was subtracted from the Qtracker® EEM. Then, to achieve the A-TEEM, the resulting fluorescence intensities were automatically corrected for inner filter effects using the absorbance spectrum collected in the same acquisition.

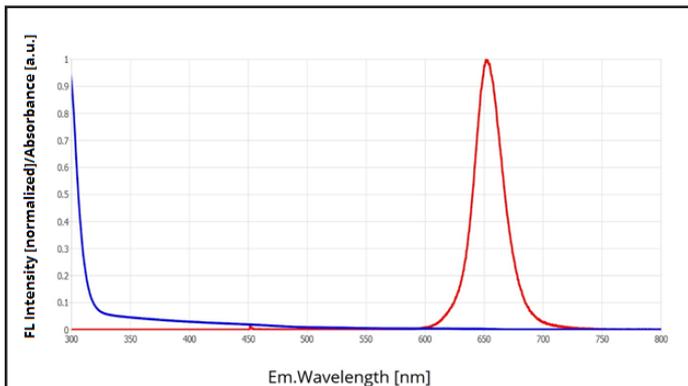


Figure 2: Absorbance (blue) and fluorescence emission (red) spectra for Qtracker 655 quantum dots. The fluorescence spectrum is normalized for easy viewing while the absorbance spectrum is not normalized.

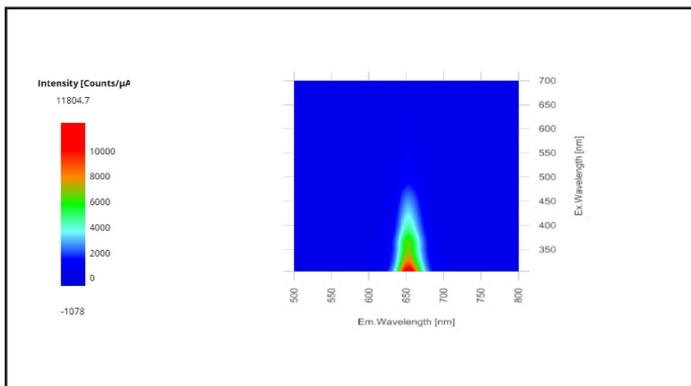


Figure 3: A-TEEM™ for Qtracker® 655 quantum dots shows that the emission peak wavelength does not change based on excitation wavelength

Fluorescence Lifetimes

The lifetime decay for the same solution of Qtracker® 655 quantum dots was collected on a DeltaPro instrument from HORIBA Scientific with PPD-850 PMT and DeltaDiode-485L (481nm actual wavelength) pulsed laser source. The measurement time range was set at 800ns. DeltaDiode repetition rate was 1 MHz and the emission wavelength was selected using a 500nm long pass filter on the emission optical path. The acquired decay and fit are shown in Figure 4.

The decay was fit to a 5-exponential decay equation and so that the error residuals became randomized and the reduced chi-squared value of the fit was close to 1 (1.16 actual) to show goodness of fit. As shown in Figure 5, the average lifetime was calculated to be 3.22ns and the individual decay rates ranged from 407ps to 114ns.

*Qtracker® is a registered trademark of ThermoFisher Scientific.

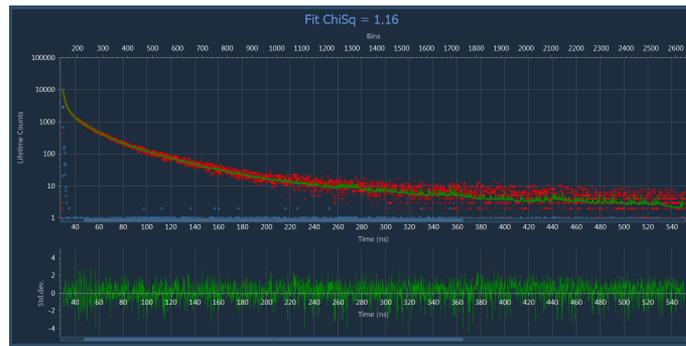


Figure 4: Lifetime decay (red) and fit (green) to a 5-exponential decay equation. The lower graph shows the standard deviation error residuals from the fit to the fitted data.

Decay	1			
		Value	Relative Amplitude	Normalised pre-exponential
T1	=	2.53126 ns	15.84%	0.2
T2	=	12.0648 ns	32.46%	0.09
T3	=	114.873 ns	5.43%	0.0
T4	=	35.7012 ns	37.73%	0.03
T5	=	0.407507 ns	8.54%	0.68
A	=	1.30559		
Average LifeTime	=	3.22377 ns		
Chi sq.	=	1.15622653750221		

Figure 5: Fit results for lifetime decay of 400x dilution of QTracker 655 quantum dot solution

Summary

Quantum dots may be useful in tracking cell culture populations, and in other biological measurements on the nanoscale. The fluorescence spectrum of the Qtracker® 655 nanocrystals (QDs) emits at 655nm as expected, and the absorbance covers a broad range so that the QDs can be excited at any wavelength below 655nm. Absorbance and fluorescence spectra, including A-TEEM profiles, can be useful for qualifying homogeneity in QD solutions as well as identifying contaminants. Inner filter effect corrections to the fluorescence spectra and profiles were necessary to correct for high absorbance values – especially in the UV excitation wavelength range. On the DeltaPro, fluorescence lifetimes measured for the Qtracker® 655 solution are complex, yielding 5 time constants and an average lifetime of 3.22ns. Complex lifetime decays are common for core-shell quantum dots and can be useful for quantifying interactions of quantum dots with other molecules due to energy transfer. Duetta and DeltaPro benchtop fluorescence instruments are easy to use and ideal for measurement of quantum dot absorbance and photoluminescence spectra and lifetimes.

References

B.O. Dabbousi, e. (1997). (CdSe)ZnS Core-Shell Quantum Dots: Synthesis and Characterization of a Size Series of Highly Luminescent Nanocrystallites. The Journal of Physical Chemistry B, 101(46), 9463-9475.

Duggan, K. B. (1990). A new approach to high efficiency multi-bandgap solar cells. Journal of Applied Physics, 67(7), 3490-3493.

Lin Cui, C.-c. L.-y. (2018). Advances in the integration of quantum dots with various nanomaterials for biomedical and environmental applications. Analyst, 143, 2469-2478.

Murphy, C. J. (2002, October 1). Optical Sensing with Quantum Dots. Analytical Chemistry, pp. 520A-526A.

Yun-Pei Chang, F. P. (2008). Tracking bio-molecules in live cells using quantum dots. Journal of Biophotonics, 1(4), 287-298.

Zhinjun Ning, e. (2014). Air-stable n-type colloidal quantum dot solids. Nature Materials, 13, 822-828.

info.sci@horiba.com

www.horiba.com/scientific

USA: +1 732 494 8660
 UK: +44 (0)1604 542 500
 China: +86 (0)21 6289 6060

France: +33 (0)1 69 74 72 00
 Italy: +39 06 51 59 22 1
 Brazil: +55 (0)11 2923 5400

Germany: +49 (0) 6251 8475 20
 Japan: +81 (0)3 6206 4721
 Other: +1 732 494 8660

HORIBA
Scientific