# **Electronic Supporting Information for:**

# Germanium nanocrystals as luminescent probes as fast, efficient and label-free detection of Fe<sup>3+</sup> ions

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# **Experimental Methods**

## Chemicals

Tetrahexadecylammonium bromide (THDAB) (98 %), 9,10-diphenylanthracene (97 %), fluorescein (for fluorescence), cyclohexane (CHROMASOLV Plus, for HPLC,  $\geq$  99.9 %), NaOH (BioUltra, for luminescence,  $\geq$  98.0 %), phosphate buffered saline (tablet), HCl ( $\geq$  37 %), sulfuric acid (95-97 %), hydrogen peroxide (30 % v/v), AlCl<sub>3</sub>•6H<sub>2</sub>O (99 %), Cd(ClO<sub>4</sub>)<sub>2</sub>•6H<sub>2</sub>O (99.999 % trace metal basis), CoCl<sub>2</sub> (97 %), CuCl<sub>2</sub> (97 %), CrK(SO<sub>4</sub>)<sub>2</sub>•12H<sub>2</sub>O ( $\geq$  98 %), FeCl<sub>2</sub> (98 %), Fe(NO<sub>3</sub>)<sub>3</sub>•9H<sub>2</sub>O (99.999 % trace metal basis), HgCl<sub>2</sub> ( $\geq$  99.5%), MnCl<sub>2</sub> (98 %), NiCl<sub>2</sub> (98 %), PbCl<sub>2</sub> (98 %), PdCl<sub>2</sub> ( $\geq$  99.9 %), H<sub>2</sub>PtCl<sub>6</sub>•H<sub>2</sub>O ( $\geq$  99.9 %), Zn(CH<sub>3</sub>COO)<sub>2</sub>•2H<sub>2</sub>O (99.999 % trace metal basis) were purchased from Sigma Aldrich Ltd. and stored in ambient atmosphere. Germanium tetrachloride (GeCl<sub>4</sub>) (99.99 %), lithium aluminium hydride (LiAlH<sub>4</sub>) (1M in THF), allylamine ( $\geq$  99 %), toluene (99.8 %, anhydrous), methanol (99.8 %, anhydrous) and propan-2-ol (99.5 %, anhydrous) were purchased from Sigma Aldrich Ltd. and solvents were used as received.

# Synthesis and purification

All glassware used was cleaned by soaking in a base bath overnight, followed by submersion in piranha solution (3:1 concentrated sulphuric acid : 30 % hydrogen peroxide) for 20 minutes. Caution: Piranha solution is a strong oxidising agent and should be handled with extreme care. All synthetic procedures were carried out in an inert atmosphere glove box with water and oxygen levels kept below 0.1 ppm in order to prevent simple hydrolysis of the Ge precursor. In a typical preparation, 2.74 mmol of the surfactant, tetrahexadecylammonium bromide (THDAB), was dissolved in 60 mL toluene in a 2-neck round bottomed flask with continuous stirring. 0.1 mL (0.876 mmol) GeCl<sub>4</sub> was then added to the solution and left to stir for 30 min. Ge NCs were formed by the drop wise addition of 2 mL of LiAlH<sub>4</sub> over a period of 2 minutes. Caution: germane gas, which is pyrophoric and highly toxic, could be evolved at this stage of the reaction and care should be taken to prevent exposure to air.<sup>1</sup> The solution was then left to stir for 2.5 h. The excess reducing agent was then quenched with the addition of 20 mL methanol. At this stage of the reaction the Ge NCs are terminated by hydrogen and encapsulated within the inverse micelle.

Chemically passivated nanocrystals were formed by modifying the germanium-hydrogen bonds at the surface *via* the addition of 0.5 mL of 0.1 M chloroplatinic acid in propan-2-ol as a catalyst, followed by 2 mL of allylamine. After stirring for 2.5 h, the amine–terminated Ge NCs were removed from the glove box and the organic solvent removed by rotary evaporation. The resulting brown dry powder (consisting of surfactant and Ge NCs) was then re-dispersed in 40 mL deionised water (18.2 M $\Omega$  cm) and sonicated for 20 min. The solution was first filtered using filter paper and then PVDF membrane filters (Acrodisc, 0.22  $\mu$ M) to remove the surfactant. The Ge NCs were further purified by chromatography. The solution was concentrated down to *ca*. 1.5 mL and loaded into the column ( $\emptyset$  = 1 cm, Length = 47.0 cm). Sephadex gel LH-20 was used as the stationary phase: fractions of *ca*. 3 mL were collected at a drop rate of approximately 1 drop every 25 seconds. The fractions were then combined and concentrated down to *ca*. 5 mL. The final product typically yielded 10 mL of 1  $\mu$ M solution of Ge NCs.

## Centrifugation

This parent NC solution was then loaded into an Eppendorf Safe-lock tube and centrifuged on an Eppendorf 5415D centrifuge at 13,200 rpm for 15 minutes. The supernatant was decanted and the pellet which formed was re-dispersed in water by sonication.

## pH measurements

0.5 mL aliquots of a stock solution of Ge NCs were added to a series of PBS buffered solutions with different pH values (3-9). These solutions were formed by initially making up a 0.01 M phosphate buffer solution (1 tablet in 200 mL DI water), taking aliquots of this and adding either HCl or a 0.5 M NaOH solution to adjust the pH. The fluorescence intensity was measured at an excitation of 400 nm for both the blue and green NCs.

#### Metal ion sensing

Standard stock solutions (10 mM) of metal ions were prepared in DI water and different concentrations obtained by diluting these stock solutions. The detection of Fe<sup>3+</sup> was performed in DI water at room temperature. In a typical assay, 2.5 mL of a particular concentration of Fe<sup>3+</sup> was added to 0.5 mL Ge NCs, the solution was stirred and the PL spectra were subsequently recorded. The selectivity towards Fe<sup>3+</sup> was confirmed by adding other metal ion solutions (50  $\mu$ M) instead of Fe<sup>3+</sup> in the same way. Tapwater samples were obtained from the tap in our lab, riverwater from the river Lee in Cork and lakewater from the Lough in Cork. Prior to any fluorescence measurements, the riverwater and lakewater were centrifuged at 11,000 rpm for 10 minutes and filtered through 0.22  $\mu$ M PVDF membrane filters (Acrodisc). The tapwater was not purified. Fe<sup>3+</sup> with different concentrations was prepared in these real water samples and then analysed using the proposed method above.

# Characterisation

#### Structural characterisation

Transmission electron microscopy images and selective area electron diffraction patterns were acquired using a highresolution JEOL 2100 electron microscope, equipped with a LaB<sub>6</sub> thermionic emission filament and Gatan DualVision 600 Charge-Coupled Device (CCD), operating at an accelerating voltage of 200 keV. TEM samples were prepared by depositing a 40 µL aliquot of the Ge NC dispersion onto a holey carbon-coated copper grid (300 mesh, #S147-3, Agar Scientific), which was allowed to evaporate under ambient conditions. Data for size distribution histograms was acquired by analysis of TEM images of exactly 300 NCs located at different regions of the grid. NC diameter was determined by manual inspection of the digital images; in the case of anisotropic structures, the diameter was determined using the longest axis.

## **Optical characterisation**

UV-Vis absorption spectra were recorded using a Shimadzu UV PC-2401 spectrophotometer equipped with a 60 mm integrating sphere (ISR-240A, Shimadzu). Spectra were recorded at room temperature using a quartz cuvette (1 cm) and corrected for the solvent absorption. Photoluminescence spectra were recorded using an Agilent Cary Eclipse

spectrophotometer. Long-term PL stability measurements were carried out over 6 hours on the same spectrophotometer. Excitation wavelengths of 340 nm for the blue emitting Ge NCs and 400 nm for the green emitting sample were used. Spectra were integrated between 380 to 500 nm for the blue NCs and 500 to 600 nm for the green NCs. Quantum yields were measured using the comparative method described by Williams *et al.*<sup>2</sup> Dilute solutions of the Ge NCs in water were prepared with optical densities between 0.01-0.1 and compared against solutions of the reference emitters 9,10-diphenylanthracene in cyclohexane and fluorescein in 0.1 M NaOH with similar optical densities. PL spectra of Ge NCs and reference solutions were acquired using an appropriate excitation wavelength, and the total PL intensity integrated over a suitable range for each set of NCs and reference emitters.

#### Lifetimes

Photoluminescence lifetime measurements were recorded on a scanning confocal fluorescence microscope (MicroTime 200, PicoQuant GmbH) equipped with a TimeHarp 200 TCSPC board. NC samples were excited in solution using a 402 nm pulsed diode laser (10 MHz; 70 ps pulse duration, LDH-P-C-400) that was spectrally filtered using a 405 nm band-pass filter (Z405/10x, Chroma Technology Corp.). A 50X objective (0.5 NA; LM Plan FL, Olympus Corp.) was used for focusing the excitation light onto the NC dispersion and collecting the resultant fluorescence, which was directed onto an avalanche photodiode (APD; SPCM-AQR-14, Perkin-Elmer, Inc.). Backscattered excitation light was blocked with a 410 nm long-pass filter placed in the collection path (3RD410LP, Omega Optical). The excitation power was adjusted to maintain a count rate of < 10<sup>4</sup> counts/s at the APD in order to preserve single photon counting statistics. All emission lifetimes were fitted to a weighted multi-exponential model on FluoFit 4.2 software (PicoQuant GmbH). All lifetimes were fitted with a  $\chi^2$  value of less than 1.1.

# Figures



Fig. ESI1. HRTEM image of an individual Ge NC with indicated lattice spacing.







Fig. ESI3. Normalised PL intensity of 3.9 nm and 6.8 nm Ge NCs as a function of pH.







Fig. ESI6.  $I_0/I$  vs concentration for the linear range 0 to 1  $\mu$ M.



Fig. ESI7. Normalised UV-Vis spectra for the Ge NCs before, and after the addition of increasing concentrations of  $Fe^{3+}$ 



Fig. ESI8. UV-Vis spectra for the investigation of the interaction between Ge NCs and Fe<sup>3+</sup>.



**Fig. ESI9**. Photoluminescence spectra (left) and  $I_0/I$  *vs* concentration (right) of Ge NCs measured for Fe<sup>3+</sup> concentrations from 1 to 200  $\mu$ M in tapwater, (a) and (b), riverwater, (c) and (d) and lakewater (e) and (f). The excitation was fixed at 340 nm for all spectra. I and  $I_0$  are the PL intensities in the presence and absence of Fe<sup>3+</sup> ions, respectively.

Table ESI1. Determination of Fe <sup>3+</sup> in real water samples				
Sample	Added amount (µM)	Measured (µM)	Recovery (%)	
	1	1 ± 0.01	<b>99.2</b> ± 1.4	
	10	$10 \pm 0.1$	<b>99.9</b> ± 1.4	
	20	$19.9\pm0.3$	99.5 ± 1.4	
Tapwater	30	<b>30.6 ± 0.4</b>	102.1 ± 1.2	
	40	<b>41.7 ± 0.6</b>	$104.2 \pm 1.5$	
	50	50.5 ± 1.3	101.1 ± 2.5	
	100	111.5 ± 1	$111.5 \pm 1$	
Riverwater	1	$1.1 \pm 0.01$	105.1 ± 0.9	
	10	$10.4 \pm 0.1$	$103.8 \pm 0.8$	
	20	$20.5\pm0.2$	$102.7 \pm 0.9$	
	30	<b>30.6 ± 0.3</b>	101.9 ± 1.1	
	40	41.4 ± 0.5	103.6 ± 1.3	
	50	51.9 ± 0.6	103.8 ± 1.3	
	100	110 ± 1.1	110 ± 1.2	

Lakewater	1	$1 \pm 0.01$	$104.7 \pm 1.1$
	10	$10.3 \pm 0.2$	$102.8 \pm 1.7$
	20	$20.1 \pm 0.2$	$100.7 \pm 1$
	30	$30.6 \pm 0.3$	$102.1 \pm 1.2$
	40	$40 \pm 0.8$	$100 \pm 2$
	50	$48.7 \pm 0.9$	97.3 ± 1.7
	100	$107.3 \pm 1.1$	$107.3 \pm 1.1$

# References

- J. P. Wilcoxon, P. P. Provencio and G. A. Samara, Phys. Rev. B, 2007, 76, 199904.
  A. T. R. Williams, S. A. Winfield and J. N. Miller, Analyst, 1983, 108, 1067.