

## Electronic Supporting Information (ESI)

### Quaternized Carbon Dots Modified Graphene Oxide for Selective Cell Labelling – Controlled Nucleus and Cytoplasm Imaging

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### Experimental section

**Synthesis of Graphene oxide.** Graphene oxide (GO) was obtained from graphite by modified Hummer's method.<sup>1</sup> In a typical procedure, 23 mL of concentrated H<sub>2</sub>SO<sub>4</sub> was mixed with 1 g of graphite and 0.5 g NaNO<sub>3</sub> in a 250 mL round bottom flask cooled in an ice bath at 0 °C. 3 g KMnO<sub>4</sub> was added slowly to the mixture in portions with vigorous stirring by keeping the reaction temperature below 20 °C. The reaction was warmed to room temperature and stirred for 30 min. To this 46 mL water was added slowly causing violent effervescence and rise of temperature to 98 °C. The resultant brown colored suspension was maintained at this temperature for 30 minutes. The suspension was then brought to room temperature and 140 mL water was added. The mixture was then treated with 1 mL of 30% H<sub>2</sub>O<sub>2</sub> to reduce any unreacted permanganate. The brown reaction mixture was then centrifuged at 6 000 rpm, washed several times with distilled water until the pH turned to neutral. The precipitate was dried at 80 °C.

## **Preparation of GO-QCD hybrids**

**Stock solution of GO.** 1 mg of GO was dispersed in 4 mL of distilled water and sonicated for one hour to obtain brown colored dispersion.

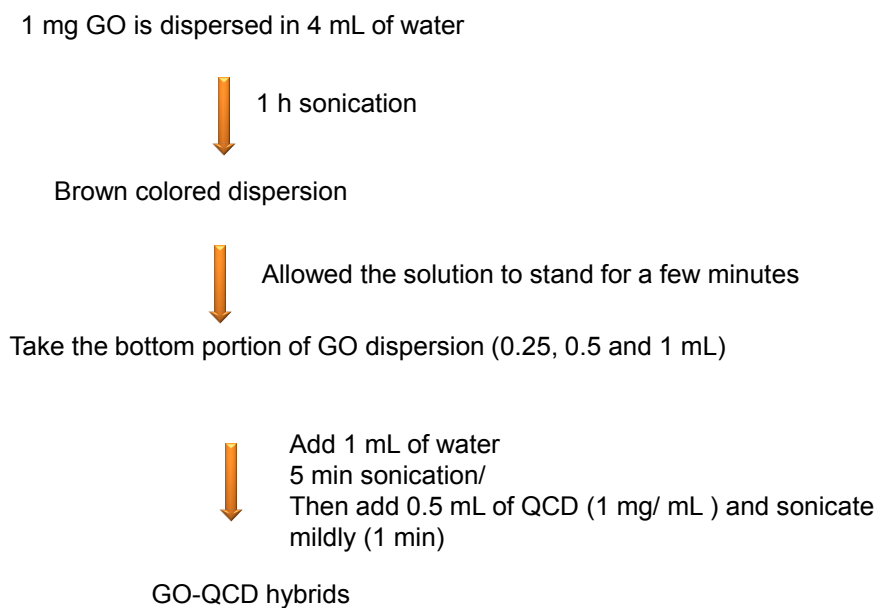
**Stock solution of QCDs.** 1 mg of quatnerized carbon dots (QCDs prepared according to previously reported synthetic strategy<sup>2</sup>) are dispersed in 1 mL of distilled water.

**GO-QCD hybrids.** 0.25 mL of GO stock solution was added to 1 mL of water and sonicated for 5 min. To this dispersion 0.5 mL of QCDs stock solution was added dropwise and the mixture was sonicated for 5 min. The obtained sample is referred as GO250QCD500. By fixing the amount of QCDs stock solution (0.5 mL), different amounts of GO were added (0.5 mL and 1 mL) to obtain GO500QCD500 and GO1000QCD500 hybrids, respectively.

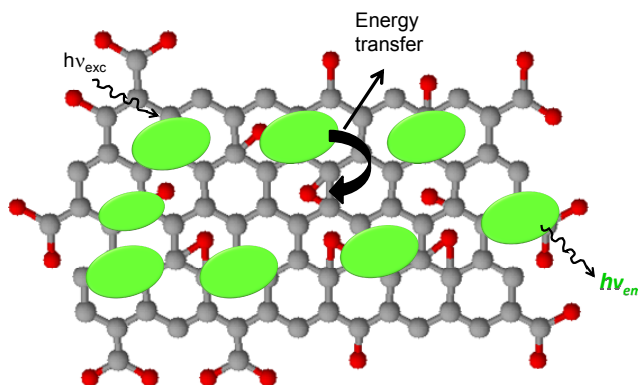
**Cytotoxic studies.** MTT assay has been used to the toxicity determination. Mouse fibroblasts NIH/3T3 were seeded to the 96-well plate and incubated with varying concentration (0-300  $\mu\text{g/mL}$ ) of samples for 24 h. Before measurement of MTT assay has been done microscopy analysis using an inverted fluorescence microscope (*Olympus IX 81*). Firstly, the cells were twice washed by phosphate buffer (pH 7.2) solution to remove the rest of nanomaterials and then observed under microscope. Subsequently, to the same cells was added 20  $\mu\text{l}$  tetrazolium salt for 4 h. Formed formazan crystals were solubilized with dimethyl sulfoxide and quantified by measuring of absorbance at 570 nm using microplate reader Synergy HT.

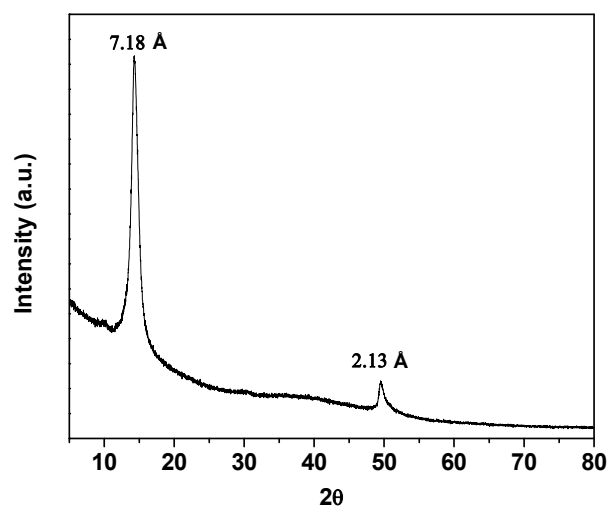
**Characterization techniques.** TEM were carried out on a JEOL 2010F microscope operated at 200 kV (LaB6 cathode, resolution 0.19 nm) using carbon coated copper grid. X-ray diffraction (XRD) patterns were recorded on an X'Pert PRO MPD diffractometer (PANalytical, Netherlands) using iron-filtered  $\text{CoK}\alpha$  radiation ( $\lambda = 0.178901$  nm, 40 kV, 30 mA). Spectroscopic studies were performed on an infrared spectrometer Nicolet iS5 (Thermo Scientific, USA), a UV-VIS diode array spectrophotometer Specord S 600 (Analytik Jena, Germany) and a QuantaMaster 40 Spectrofluorometer (PTI, USA), respectively. Zeta-potential was measured on a Zetasizer Nano particle analyser ZEN3600 (Malvern Instruments, UK). AFM images were obtained using an NTEGRA Aura (NT-MDT) microscope. A sample of the diluted dispersion was placed on a synthetic mica as an atomically smooth support and evaporated at room temperature. The measurements were performed in air at room temperature in noncontact mode, with Si tips of the 1650-00 type at resonance frequencies ranging from 180 to 240 kHz.

**Scheme 1.** Flow chart showing the synthesis of GO-QCD hybrids.

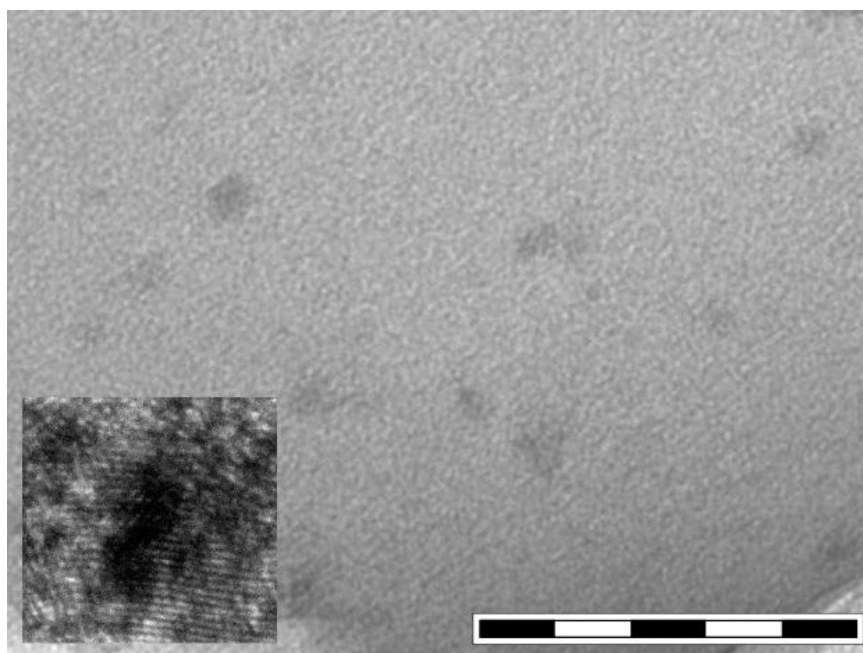


**Scheme 2.** Schematic (not to scale) showing the energy transfer process from photoexcited QCDs to graphene oxide lattice. QCDs are represented by green circles, carbon and oxygen atoms of graphene oxide are represented by grey and red colors respectively.

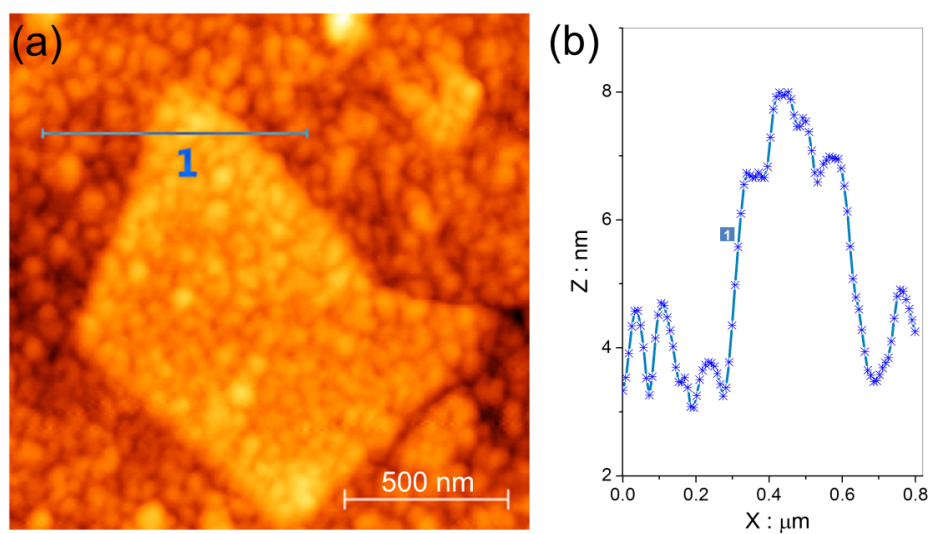




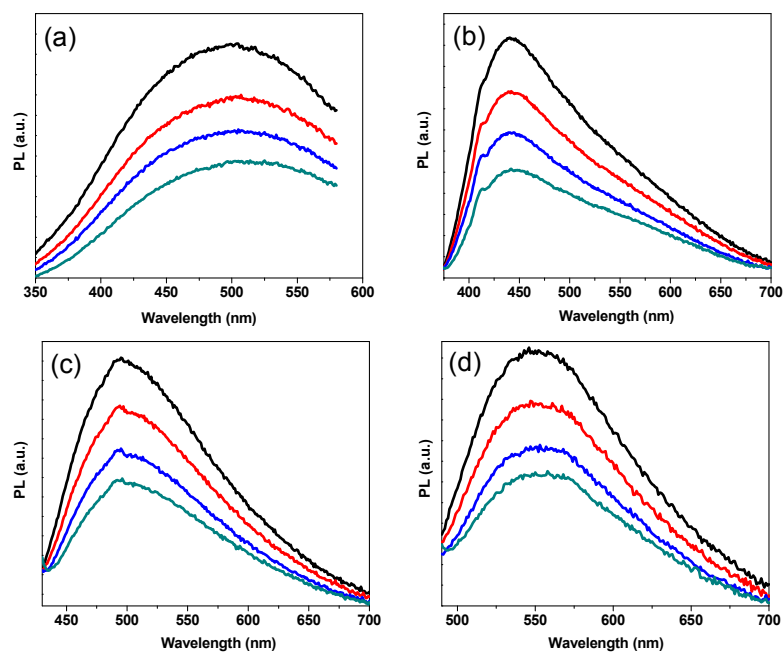
**Fig. S1.** X-ray diffraction pattern of the as-prepared graphene oxide. The peak at  $14.33^\circ$  corresponds to the (002) plane of the GO with an interlayer spacing of 0.72 nm.



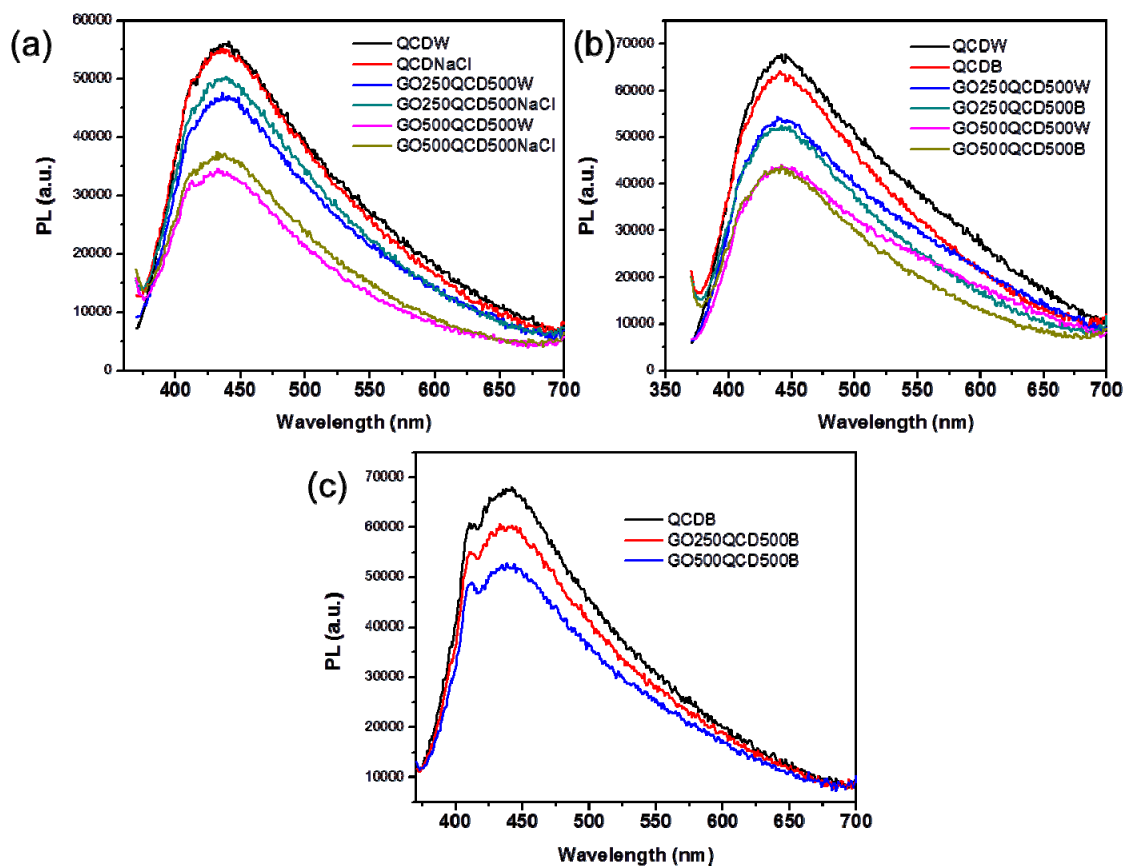
**Fig. S2.** TEM of GO250QCD500 hybrid, scale bar corresponds to 50 nm, inset showing HRTEM image of individual QCD.



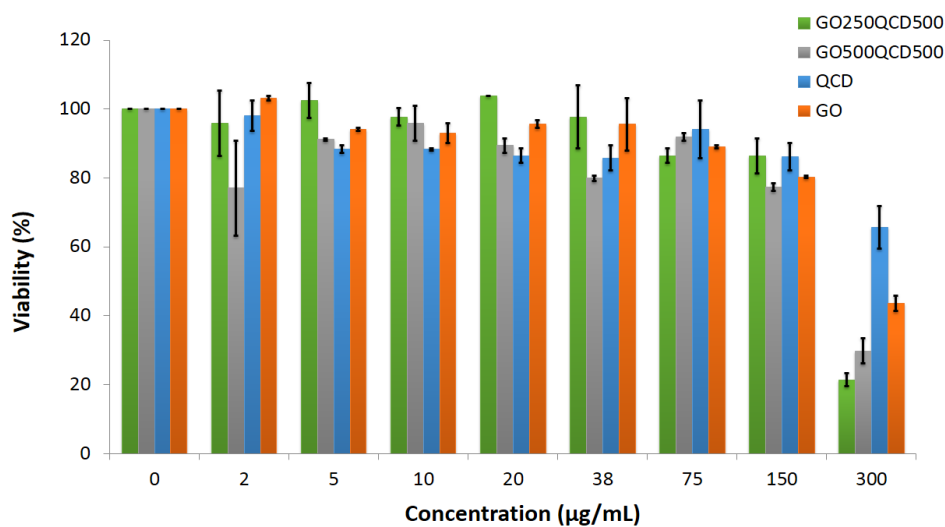
**Fig. S3.** (a) Semi-contact topographic AFM image of GO250QCD500 deposited on mica; the hybrid is overloaded with QCDs; (b) correspondent height profile ( $z$  vs  $x$ ).



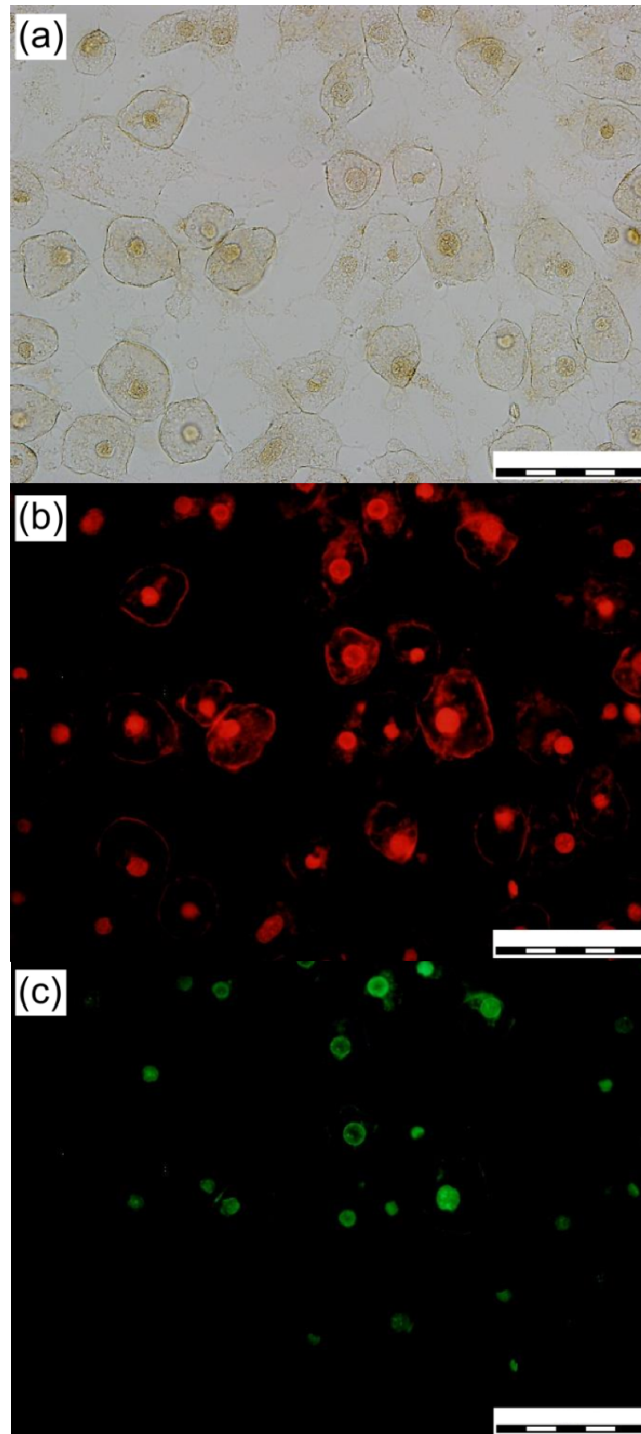
**Fig. S4.** Photoluminescence spectra of QCDs (black) and GO250QCD500 (red), GO500QCD500 (blue), GO1000QCD500 (green) hybrids excited at a) 300 nm, b) 360 nm, c) 420 nm and d) 480 nm, respectively.



**Fig. S5.** Effect of optical properties of QCD and GOQCD hybrids with respect to NaCl and various biologically relevant pH values, all the samples are excited at 360 nm (a) PL properties QCDs and GOQCD hybrids measured in the presence of NaCl (0.15 M) to assess the impact of higher ionic strength on their optical properties. It was found that higher ionic strength of the medium neither alter the shape of spectra nor decrease the PL intensity. QCDs in water (black line) and NaCl solution (red line). GO250QCD500 in water (blue line) and NaCl solution (green line). GO500QCD500 in water (pink line) and NaCl solution (brown line). (b) PL properties of QCDs and GOQCD hybrids in potassium phthalate buffer (pH = 5.4). QCDs in water (black line) and phthalate buffer (red line). GO250QCD500 in water (blue line) and phthalate buffer (green line). GO500QCD500 in water (pink line) and phthalate buffer (brown line). (c) PL properties of QCDs (black line) and GOQCD hybrids (red and blue lines) in tris buffer (pH = 7). There are no significant changes in the PL intensity or peak positions suggesting the material stability in biological conditions.



**Fig. S6.** Plot showing a cell viability for various concentrations of QCDs, GO and GO-QCD hybrids using MTT assay. Type of cells used: mouse fibroblasts (NIH/3T3) cells.



**Fig. S7.** Phase-contrast (a) and fluorescence images (b, c) of mouse fibroblast NIH/3T3 cells labelled with QCDs (100  $\mu\text{g}/\text{mL}$ ) from optical microscopy; scale bars: 100  $\mu\text{m}$ . The slight aggregation of QCDs after nuclei internalization is witnessed.



**References:**

1. D. C. Marcano, D. V. Kosynkin, J. M. Berlin, A. Sinitskii, Z. Z. Sun, A. Slesarev, L. B. Alemany, W. Lu, and J. M. Tour, *ACS Nano*, 2010, **4**, 4806.
2. A. B. Bourlinos, R. Zboril, J. Petr, A. Bakandritsos, M. Krysmann, and E. P. Giannelis, *Chem. Mater.*, 2012, **24**, 6.