Supplementary Information

Selenium Doped Graphene Quantum Dots as An Ultrasensitive Redox Fluorescent Switch

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Materials

All the chemicals were purchased from Aladdin (Shanghai, China) and used as received without further purification. The water used throughout all experiments was purified through a Millipore system. The NaHSe aqueous solution was prepared via a reduction reaction of Se and $NaBH_4$ in water under Ar.

The ·OH source used for reversible redox cycles was from Fenton's reaction system. 1.0 mL, 0.1 mM $FeSO_4$ aqueous solution was added into 1.0 mL, 1mM H_2O_2 aqueous solution, the mixture was kept at 0 °C for 2h, the concentration of ·OH in this mixture is 0.05 mM. [s1]

Synthesis of Se-GQD-2

The Se-GQD-2 were prepared as follows: typically, 1.0 mL 75 mM NaHSe aqueous solution was added to 9.0 mL GOQDs (1 mg mL^{-1}) aqueous solution, and then the mixture was transferred into a 15 mL Teflon-lined autoclave and heated and kept at 250 °C for 24 h. After that, the mixture was filtered using an alumina inorganic membrane with 20 nm pores. The resulting orange filtrate was dialysed in a 500 Da dialysis bag against deionised water for a week to remove excess salt. The Se-GQD-2 yield from GOQDs is approximately 81%.

Synthesis of Se-GQD-3

The Se-GQD-3 were prepared as follows: typically, 1.0 mL 10 mM NaHSe aqueous solution was added to 9.0 mL GOQDs (1 mg mL^{-1}) aqueous solution, and then the mixture was transferred into a 15 mL Teflon-lined autoclave and heated and kept at 250 °C for 24 h. After that, the mixture was filtered using an alumina inorganic membrane with 20 nm pores. The resulting orange filtrate was dialysed in a 500 Da dialysis bag against deionised water for a week to remove excess salt. The Se-GQD-3 yield from GOQDs is approximately 73%.

Synthesis of GQDs

The GQDs were prepared as follows: 10.0 mL GOQDs (1 mg mL^{-1}) aqueous solution was transferred into a 15 mL Teflon-lined autoclave and heated and kept at 250 °C for 24 h. After that, the mixture was filtered using an alumina inorganic membrane with 20 nm pores. The resulting orange filtrate was dialysed in a 500 Da dialysis bag against deionised water for a week to remove excess salt. The GQDs yield from GOQDs is approximately 95%.

Characterization methods

Transmission electron microscopy (TEM) measurements were carried out on a Hitachi H-8100 EM (Hitachi, Tokyo, Japan) with an accelerating voltage of 200 kV. Atomic force microscopy (AFM) measurements were carried out on Bruker Dimension Icon AFM microscope. X-ray photoelectron spectra (XPS) were carried out on a PHI Quantera II system (Ulvac-PHI, INC, Japan). Fluorescent emission and excitation spectra were recorded on a PerkinElmer LS55 luminescence spectrometer (PerkinElmer Instruments, U.K.) at room temperature (25 °C) in aqueous solution. The stability of these products was determined via contrast the fluorescent emission intensity of products aqueous solution under different conservative time at room temperature (25 °C). Quantum yield (Φ_f) was measured according to established procedure (Lakowicz, J. R. Principles of Fluorescence Spectroscopy, 2nd Ed., 1999, Kluwer Academic/Plenum Publishers, New York). The UV-vis spectra were obtained on a UV5800 Spectrophotometer. Rhodamine 6G solution (quantum yield 0.98 in EtOH) was chosen as standards. Absolute values are calculated using the standard reference sample that has a fixed and known fluorescence quantum yield value. In order to minimize re-absorption effects, absorbencies in the 10 mm fluorescence cuvette were kept under 0.1 at the excitation wavelength. Time-resolved fluorescence behavior was measured via the time-correlated single-photon counting (TCSPC) technique (HydraHarp 400, PicoQuant). The samples were excited by a frequency-doubled titanium: sapphire oscillator laser with approximately a pulse duration of 150 fs, and a repetition rate of 80 MHz (Chameleon, Coherent). Fluorescence emission was sent to a spectrometer (iHR550, Horiba Jobin Yvon) with 300/mm grating and then detected by a photomultiplier tube.

Cells experiments

The HELA cell line was obtained from the Cell Bank of Chinese Academy of Science and cultured in the standard medium at 37 °C in 5% CO_2. Cells were seeded in a 96-well plate for 24 h before Se-GQDs treatment. Serial dilutions of Se-GQDs with known concentrations were added into cells. After 24 h incubation, the relative viabilities of cell samples were determined by colorimetric 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl tetrazolium bromide (MTT) assays were performed to assess the metabolic activity of cells treated as described above. Cells were lysed with acidulated sodium dodecyl sulfate (SDS). Absorbance was measured at 570 nm using microplate reader (Bio-Rad 680, U.S.A.). All measure-ments were done in triplicate, and at least three independent experiments were carried out. The experiment of practical applications of switch system *in vivo* is as follows: HeLa cells loaded with 1 mL, 250.0 mg mL^{-1} Se-GQDs for 10 min. Then the Se-GQD-loaded cells treated with 1.0 μM H_2O_2 and 0.5 μM $FeSO_4$ for 10 min. Finally, Se-GQD-loaded cells exposed to a second dose of 5.0 μM GSH for an additional 10 min.

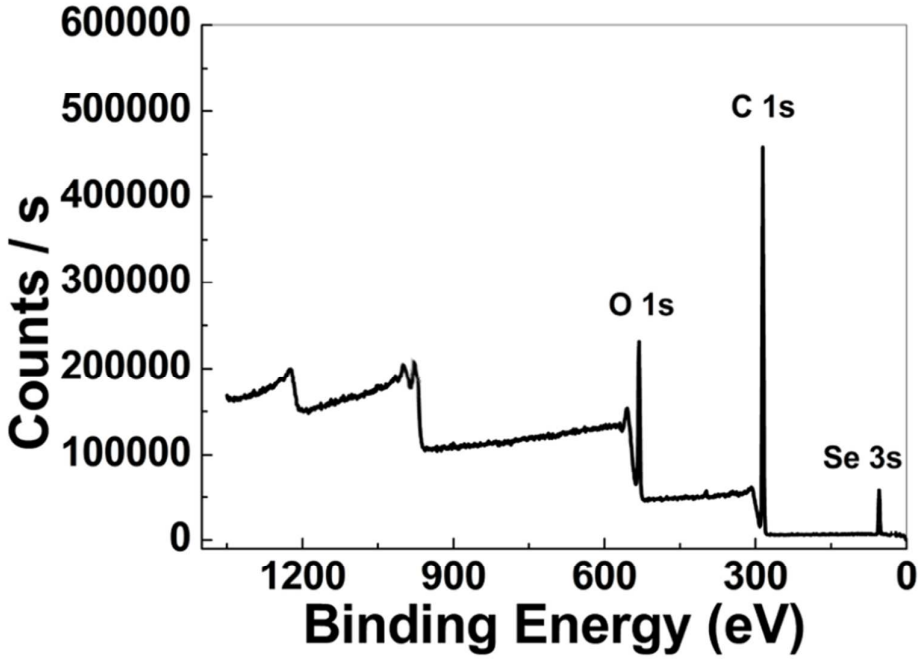

Fig. S1 XPS survey spectrum for Se-GQDs

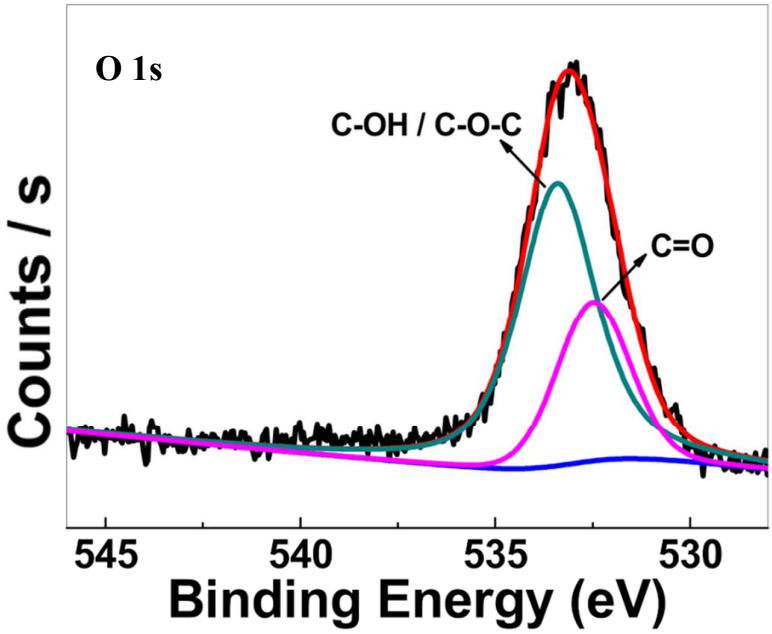

Fig. S2 O1s spectra of Se-GQDs.

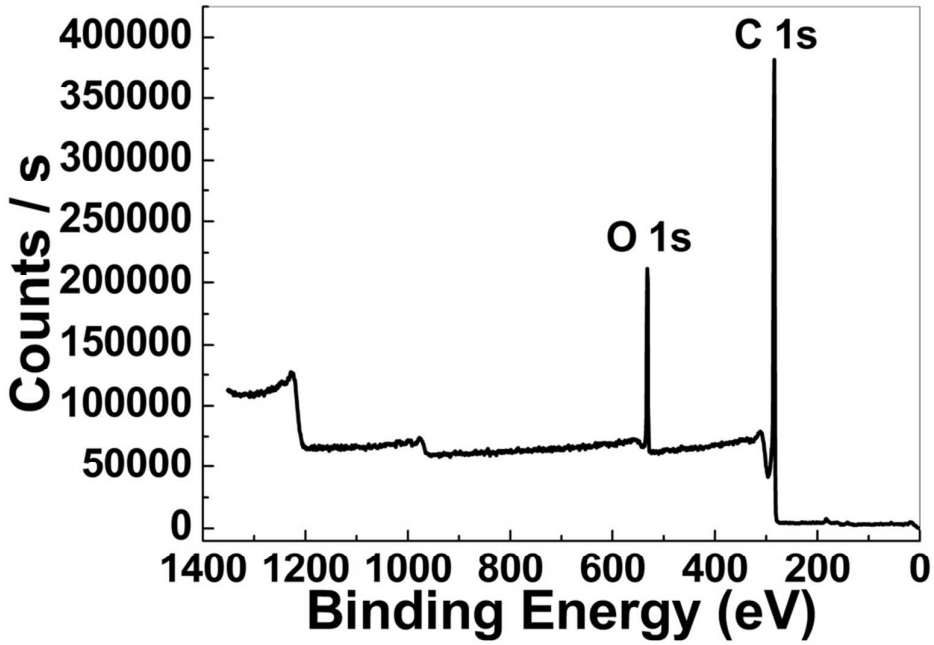

Fig. S3 XPS survey spectrum of GOQDs.

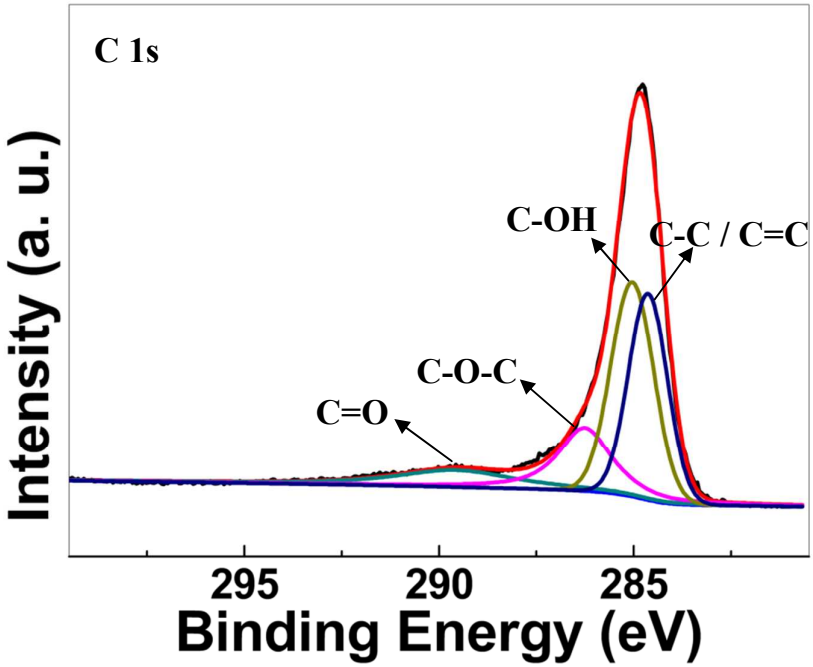

Fig. S4 C 1s spectrum of GOQDs.

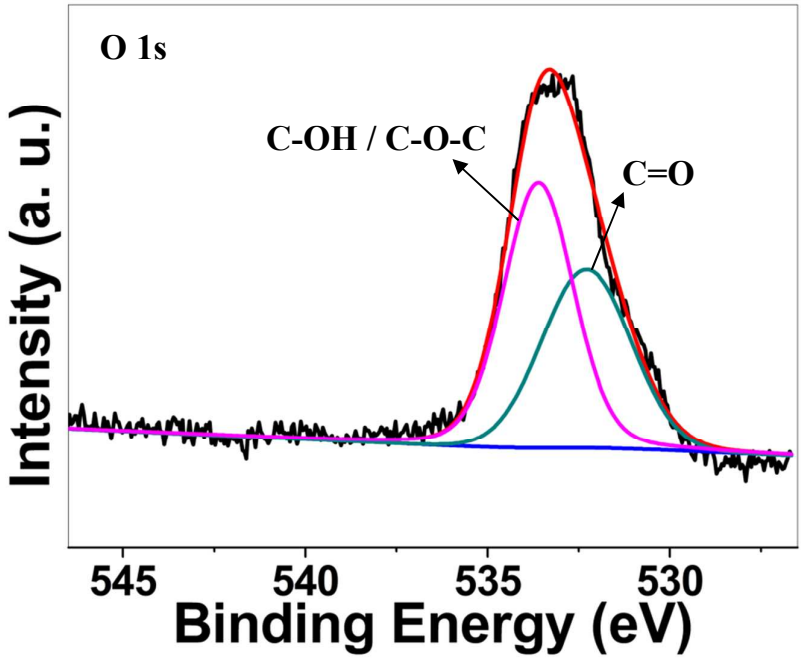

Fig. S5 O 1s spectrum of GOQDs.

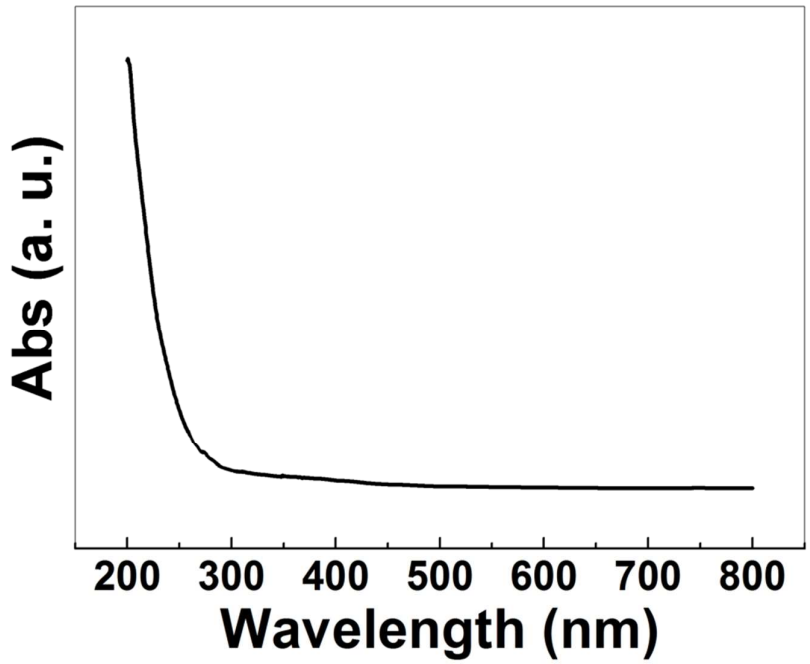

Fig. S6 UV-vis absorption spectrum of GOQDs aqueous solution.

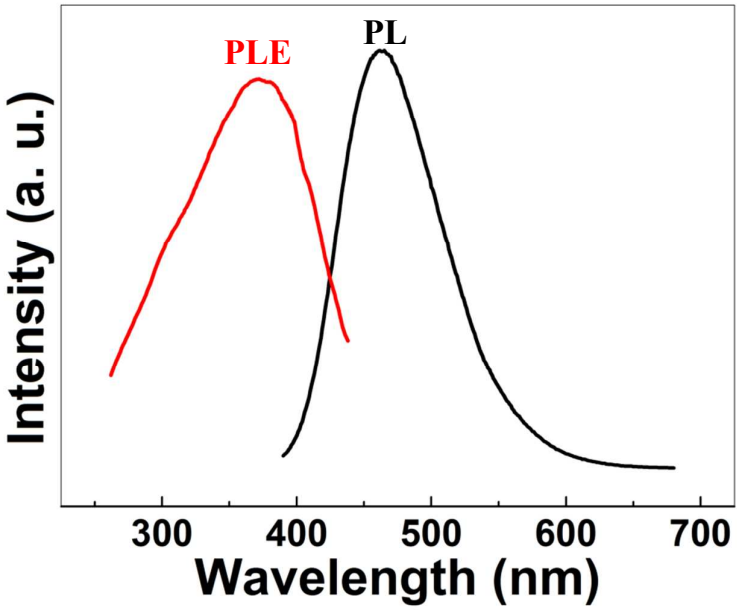

Fig. S7 Photoluminescence excitation (red curve) and PL (black curve) spectra of GOQDs aqueous solution.

Table S1. Elemental composition (at. %) of GOQDs and Se-GQDs

GQDs	C	O	Se
GOQDs	67.18	32.82	0
Se-GQDs	78.23	14.99	6.78

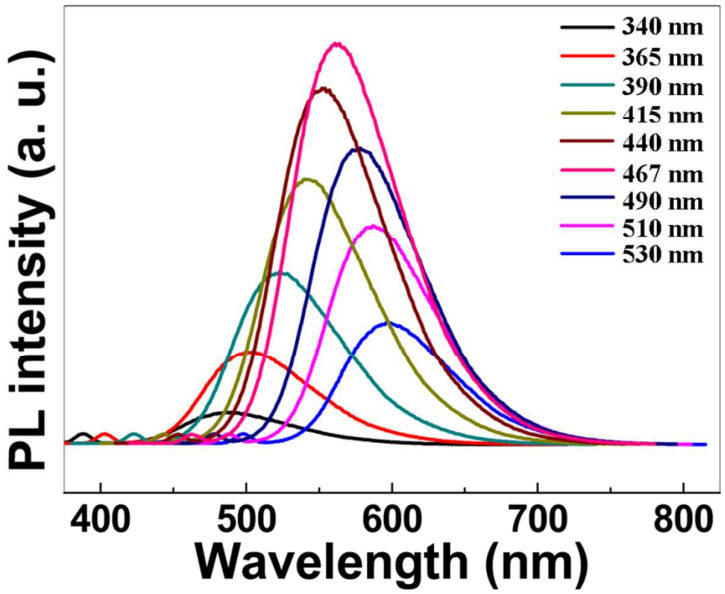

Fig. S8 PL emission spectra of Se-GQDs with different excitation wavelength

Table S2. Quantum yield (φ) Comparison of different GQDs

GQDs	φ	Ref.
Se-GQDs	0.28 [a]	This work
N-GQDs	0.16 [b]	S2
N-GQDs	0.24 [c]	S3
B&N-GQDs	0.49 [d]	S4
GQDs	0.11[e]	S5
GQDs	0.12[e]	S6
GQDs	0.28 [f]	S7
GQDs	0.088 [g]	S8
GQDs	0.18 [h]	S9

[a] Quantum yield was calculated using those of Rhodamine B (φ=0.68) in EtOH as a standard.

[b] Quantum yield was calculated using those of Quinine sulfate (φ=0.577) in water as a standard.

[c] Quantumyield was calculated using Quinine sulfate (φ= 0.54) in 0.1 M H_2SO_4 as a standard.

[d] Quantumyield was calculated using Rhodamine B (φ=0.31) in water as astandard.

[e] Quantumyield was calculated using 9,10-Bis(phenylethynyl) anthracene (φ=1) in cyclohexane as a standard.

[f] Quantumyield was calculated using Rhodamine B (φ=0.31) in water as a standard.

[g] Quantumyield was calculated using Fluorescein (φ= 0.95) in water as astandard.

[h] Quantumyield was calculated using 9,10-Bis(phenylethynyl) anthracene(φ= 1) in cyclohexaneas a standard.

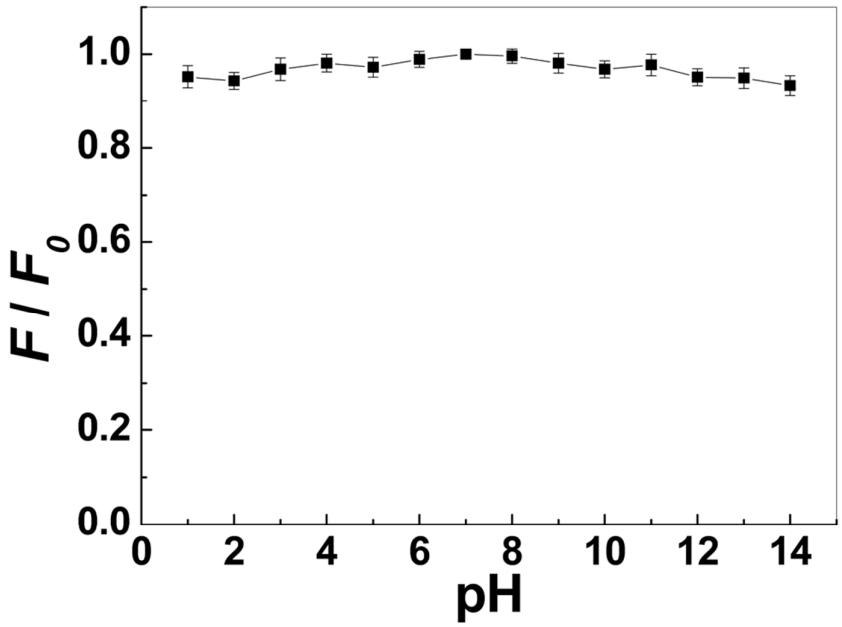

Fig. S9 PL intensity of Se-GQDs under different pH. The F and F_0 are PL intensity of Se-GQDs when pH=7 and other value, respectively. The concentration of Se-GQDs is 1 mg/L.

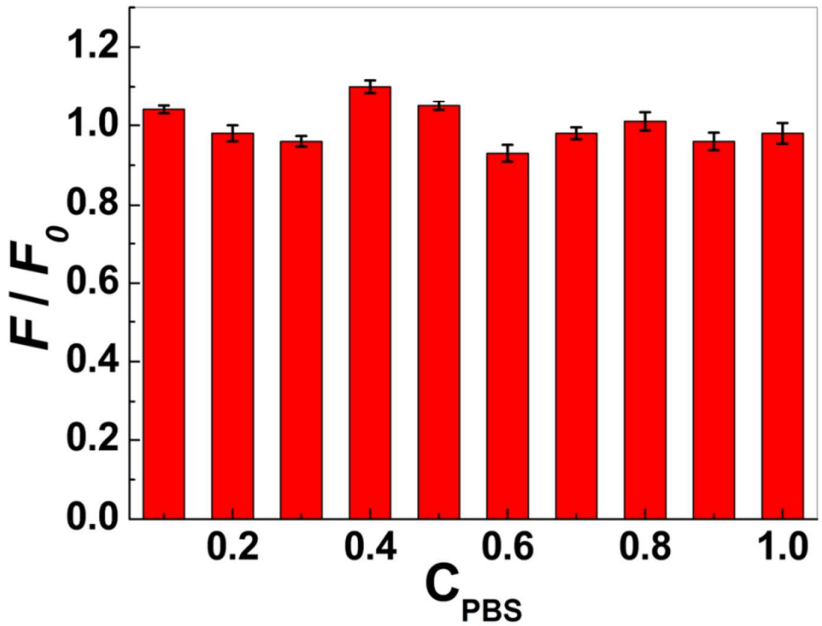

Fig. S10 The effect of ionic strength (PBS, pH=5.6) on Se-GQDs' PL intensity at room temperature. The F and F_0 are PL intensity of Se-GQDs with and without the presence of PBS, respectively. The concentration of Se-GQDs is 1 mg/L.

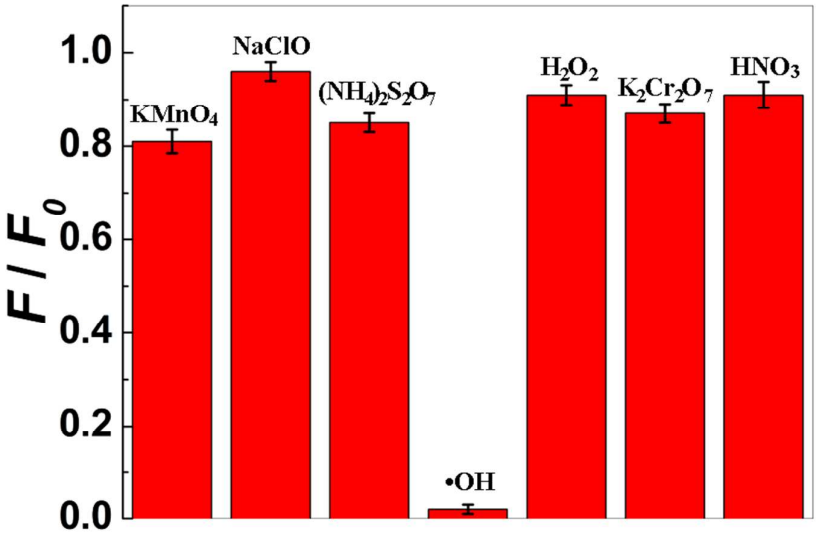

Fig. S11 the change of PL intensity of Se-GQDs with the presence of different oxidizer. The F and F_0 are PL intensity of Se-GQDs with and without the presence of oxidizer, respectively.

Table S3. Detection limit for ·OH of different measurgin method

Method	Detection limit	Ref.
Se-GQDs	3.0×10^{-10} M	This work
1-propanol	2×10^{-4} M	S10
Cupric oxide nanoparticles	3.4×10^{-7} M	S11
BMPTA-Tb^{3+}	5.0×10^{-6} M	S12

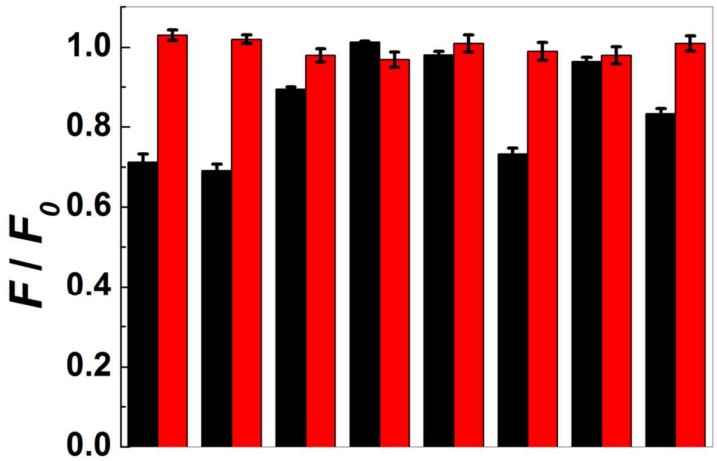

Fig. S12 PL intensity change of the switch system in "off" state. The black is Se-GQD–metal ion system, and the Se-GQD–metal ion system with EDTA, respectively. F_0 is PL intensitie of switch system in "off" state in the absence of ions. The concentrations of ions and EDTA are all 0.5 M.

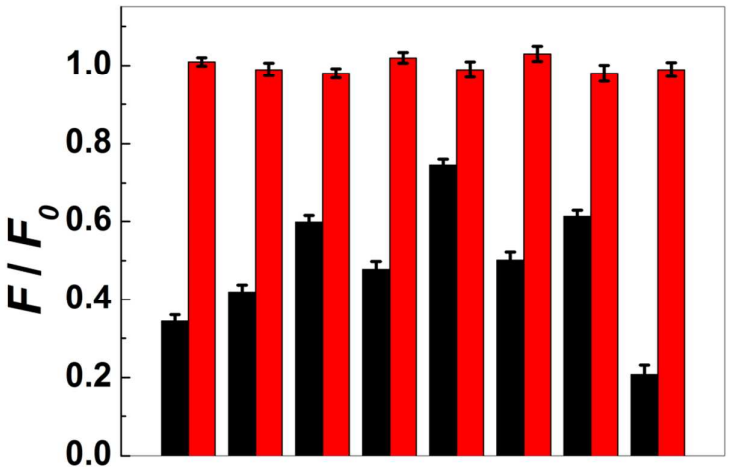

Fig. S13 PL intensity change of the switch system in "on" state. The black is Se-GQD–metal ion system, and the Se-GQD–metal ion system with EDTA, respectively. F_0 is PL intensitie of switch system in "on" state in the absence of ions. The concentrations of ions and EDTA are all 0.5 M.

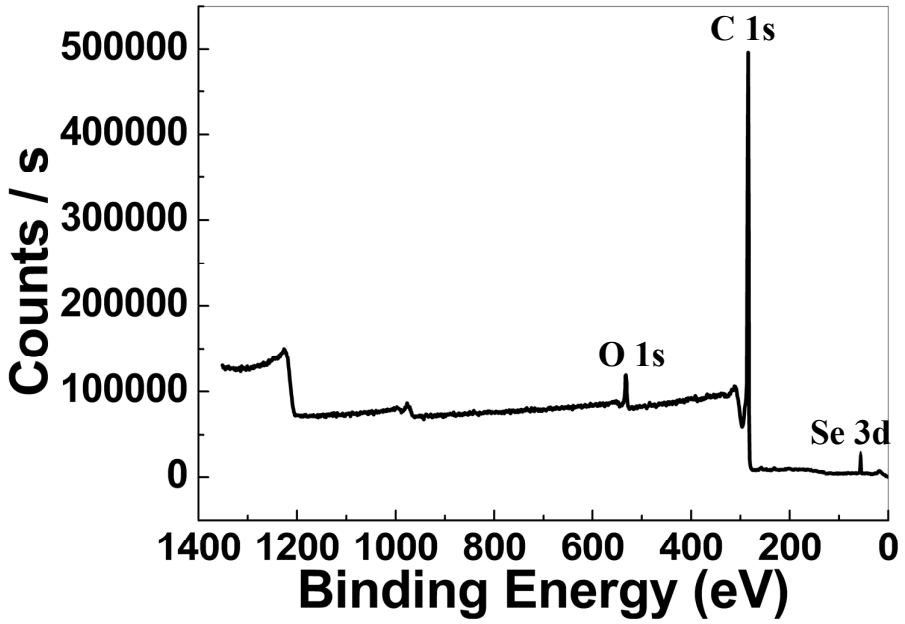

Fig. S14 XPS survey spectrum for Se-GQD-2.

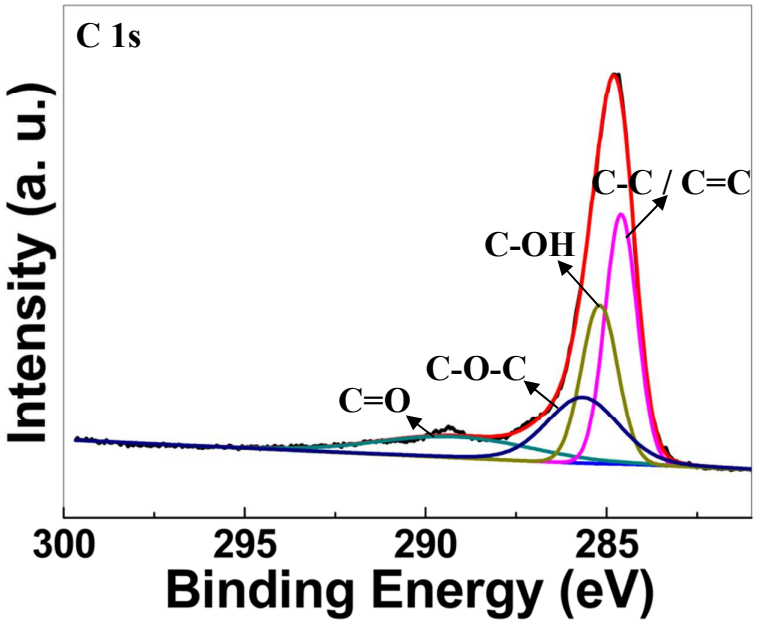

Fig. S15 C 1s spectrum of Se-GQD-2.

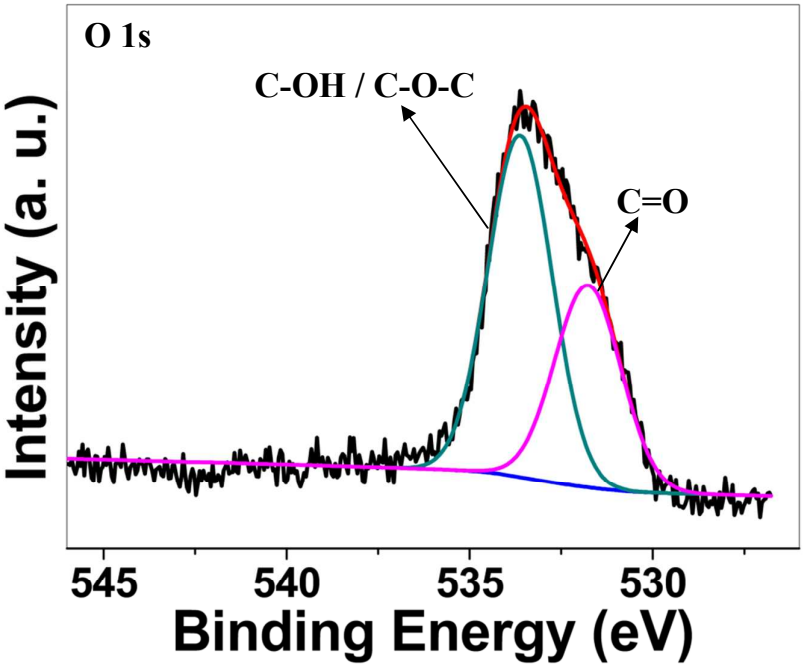

Fig. S16 O 1s spectrum of Se-GQD-2.

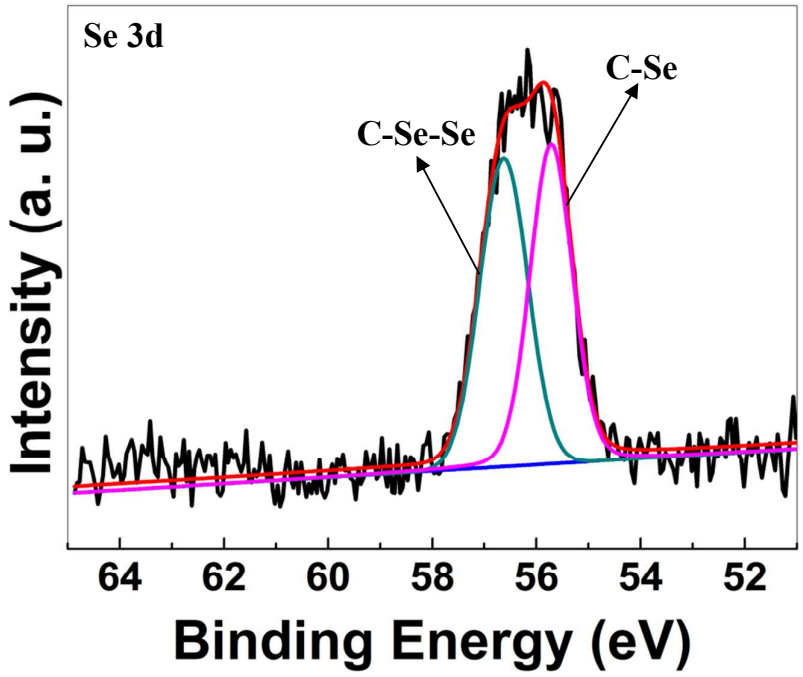

Fig. S17 Se 3d spectrum of Se-GQD-2.

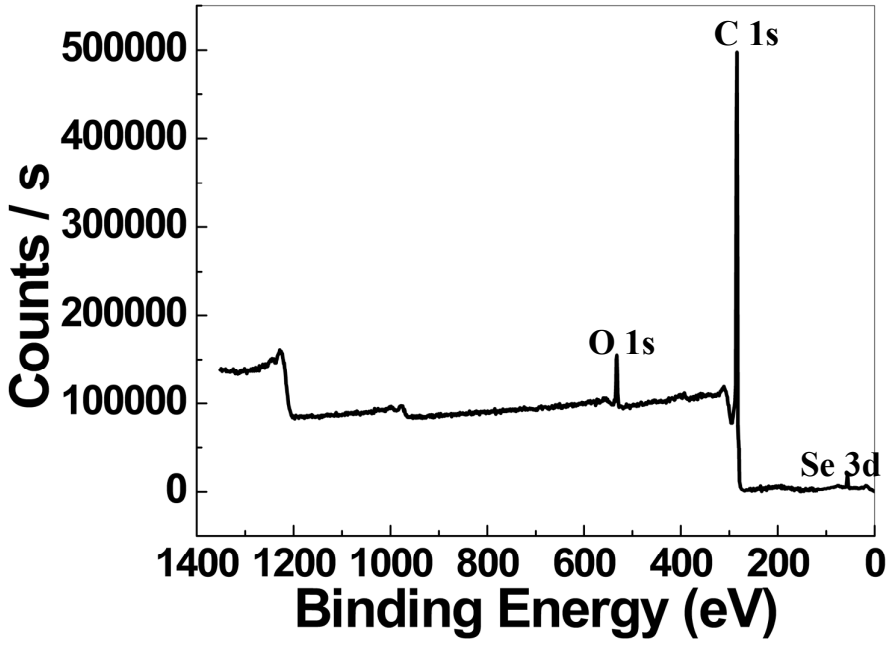

Fig. S18 XPS survey spectrum for Se-GQD-3.

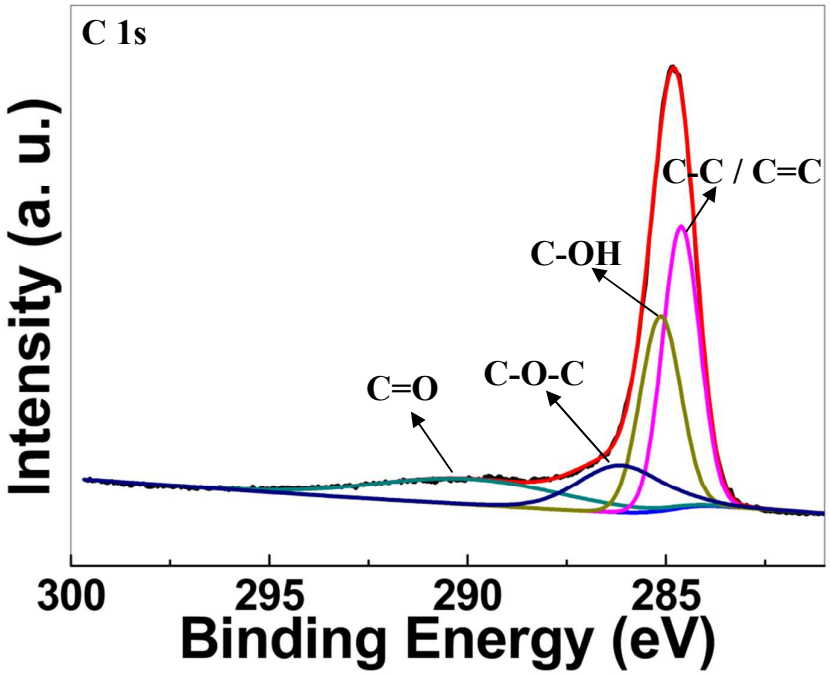

Fig. S19 C 1s spectrum of Se-GQD-3.

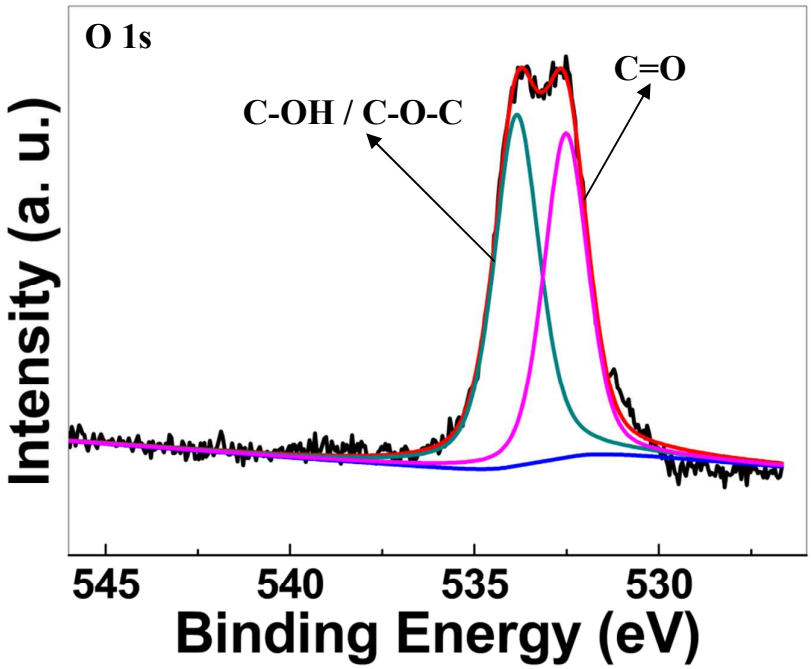

Fig. S20 O 1s spectrum of Se-GQD-3.

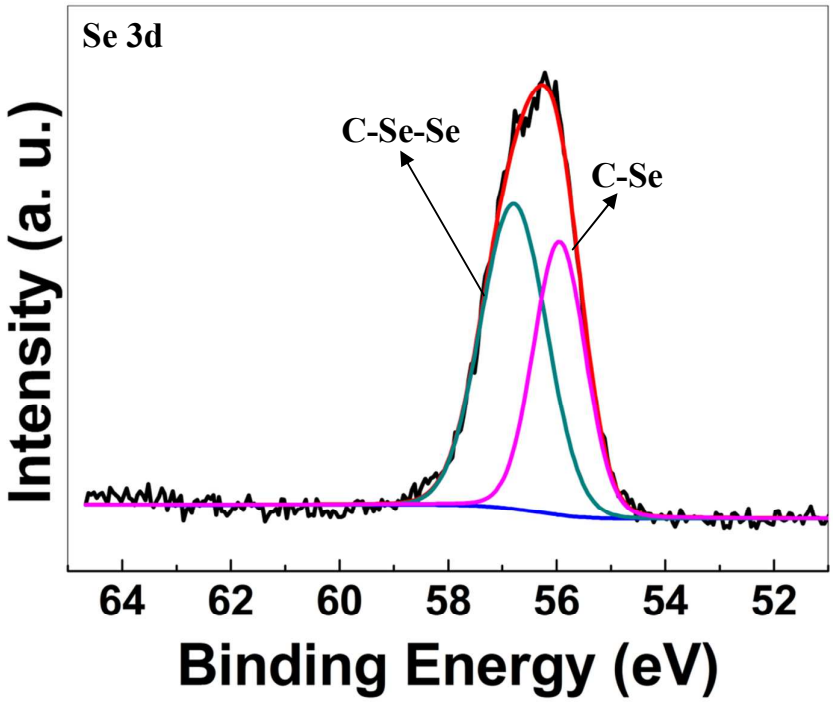

Fig. S21 Se 3d spectrum of Se-GQD-3.

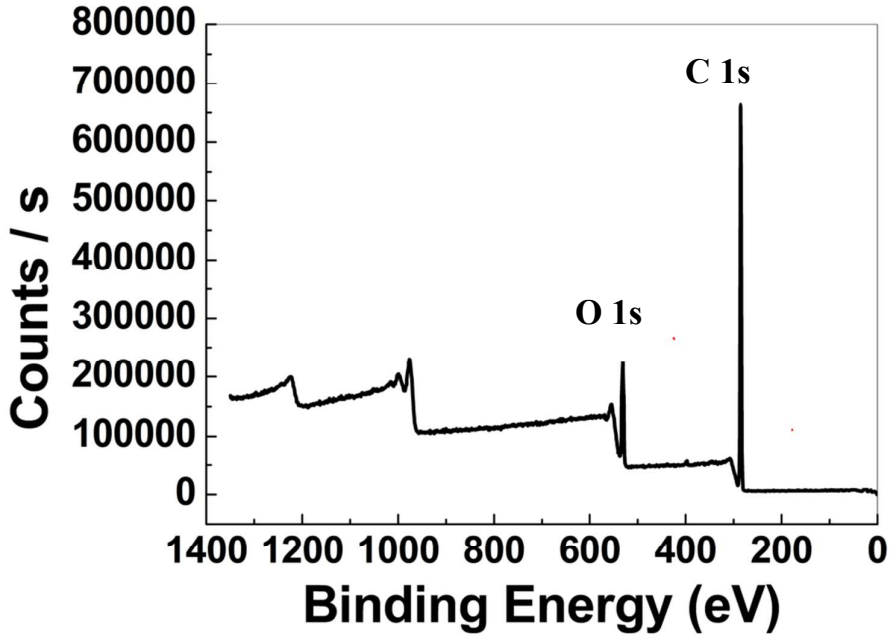

Fig. S22 XPS survey spectrum for GQD.

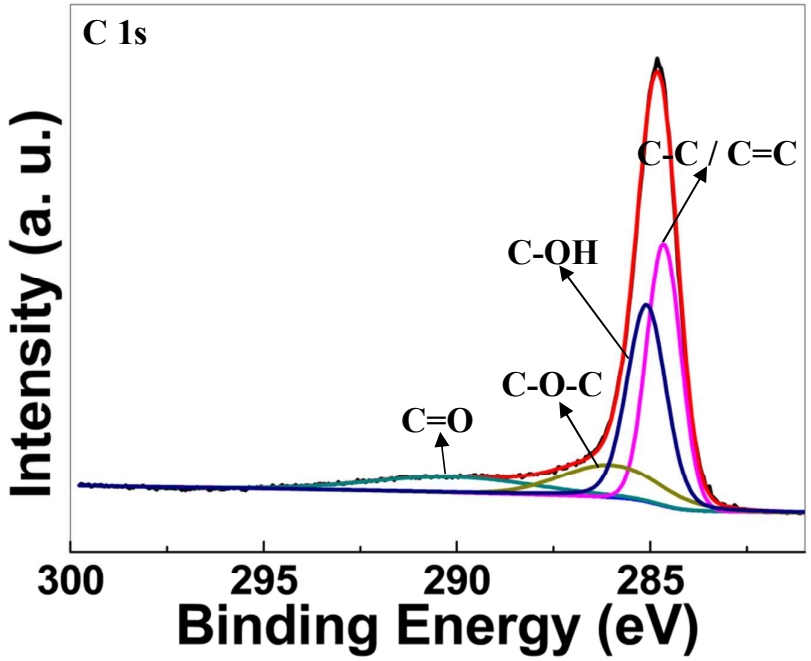

Fig. S23 C 1s spectrum of GQD.

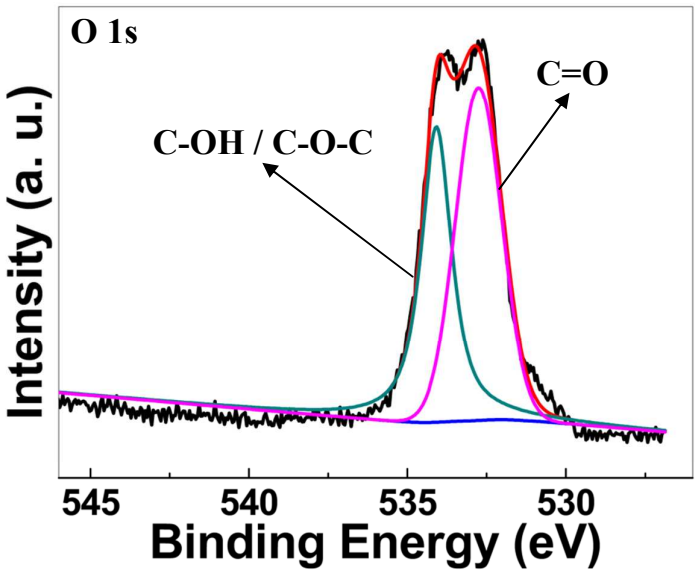

Fig. S24 O 1s spectrum of GQD.

Table S4. Elemental composition (at. %) of Se-GQDs with different selenium content

GQDs	C	O	C/O	Se
Se-GQDs	78.23	14.99	5.21	6.78
Se-GQD-2	81.91	14.85	5.51	3.24
Se-GQD-3	84.40	14.57	5.79	1.03
GQDs	84.72	15.28	5.54	0

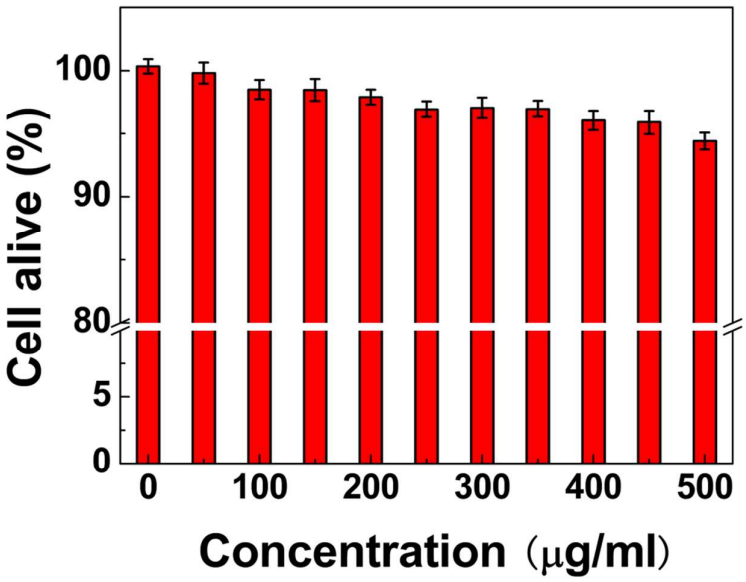

Fig. S25 Metabolic activity of HeLa cells treated with different concentrations of N-GQDs.

References

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