Supporting Information

For

Water Soluble Green Emitting Carbon Nanodots with Enhanced Thermal Stability for Biological Application

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Cytotoxicity Assay

Tongue squamous Cell (T-ca) cells were incubated with different concentration of CNDs at 37° C in 5% CO₂ for 24 hours. Then the cell viability was determined by cell counting kit-8 (CCK-8). In detail, cells after treatment were incubated with CCK-8 diluent for 1 hour, and then measured the optical densities (OD) at 450 nm. The experiments were performed 3 times.

Cell Imaging

CNDs (300 μ g/mL) were mixed with living T-ca cells in 24-well plates. After incubation for 4 h, the culture medium with free CNDs was then discarded and the T-ca cells were washed with phosphate buffered saline (PBS) for several times. Fluorescent images of cells were obtained on a Nikon Confocal laser scanning microscope A1R+Ti2E.

Quantum Yield (QY) measurement

Quantum yield (QY) of the obtained fluorescent CNDs was determined by a relative slope method.¹ Specially, quinine sulfat(QS) (QY=54%) was selected as a standard for the prepared CNDs. The aqueous solution of CNDs and QS ware diluted to keep the absorption intensity

below 0.1 at the excitation wavelength of 400. The QY of the prepared NCDs was calculated according to the following equation (1).

$$\varphi_{x} = \varphi_{st} (K_{x}/K_{st}) (\eta_{x}/\eta_{st})^{2}....(1)$$

Where ϕ is the quantum yield, K is the slope of the fitted line and η is the refractive index of the solvent. The subscript "x" refers to the testing sample and "st" refers to the standards (QS). The value of refractive index is 1.33 for water.



Figure S1. Raman spectra of the CNDs under 532 nm laser excitation, (1,2) representing different scanning position.



Figure S2.Optimized conditions of CNDs (a) ratio of starting materials (AA/p-PDA); (b) annealing temperature; (c) reaction time.



Figure S3. Quantum yield measurement of the CNDs using sulfate quinine as a reference.



Figure S4. pH dependent emission of CNDs in the range from 2-13.5.







Figure S6. Viability of T-ca cells after 24h treatment with CNDs as calculated from CCK8 assay.