

**Supplementary Information for**  
**Highly Emissive and Biocompatible Dopamine-derived**  
**Oligomers as Fluorescent Probes for Chemical Detection**  
**and Targeted Bioimaging**

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## **Experimental Section**

### **Materials**

The chemicals including dopamine hydrochloride, glycine, glutaraldehyde (25%), sodium borohydride, ethylenediaminetetraacetic acid disodium salt dehydrate (EDTA), fluorescein isothiocyanate (FITC), quinine sulfate, mercaptoethanol, polyethylene glycol (with number averaged molecule weight of 1000, 2000 and 6000), ethyl(dimethylaminopropyl) carbodiimide (EDC) and N-hydroxysulfosuccinimide sodium salt (sulfo-NHS) were obtained from Sigma-Aldrich Company. The aqueous ammonia (28%) and hydrochloric acid (36%) were purchased from Wako Pure Chemical Industries. Cyclic Gly-Phe-Lys-Ser-Arg-Gly-Asp (cRGD) peptides and G Protein-coupled receptor (GPR) 120 primary antibodies were obtained from ChinaPeptides Company and GeneTex Company, respectively.

### **Synthesis and Characterization of Dopamine-derived oligomers**

For the preparation of fluorescent dopamine-derived oligomers, 50 mg dopamine hydrochloride and 60 mg glycine were dissolved in 20 mL ultra-pure water. The pH of the solution was adjusted to 4 by adding hydrochloric acid solutions and then 200  $\mu$ L of 25% glutaraldehyde were added into the solution under stirring for a reaction of 12 h. The obtained Schiff bases were reduced by adding 1 mL of 0.1 M ice-cold sodium borohydride for 2 h. Then the polymerization reaction was initiated by introducing 400  $\mu$ L aqueous ammonia (28%) and terminated with excessive mercaptoethanol after 30 min. The obtained products were purified through dialysis (MWCO = 1000 Da) over water for removing unreacted reagents. The molecule weight distribution of the purified products was determined with Gel Permeation Chromatography (GPC) characterizations on a Waters HPLC system (Waters, Milford, USA) with a G1310A pump and a G1362A refractive index detector using  $\text{NaNO}_3$  (0.1 M) as the eluent with an elution rate of 1.0 mL/min. Commercial polyethylene glycol with molecule weight from 1000 to 6000 were used as standards for the determination of molecule weight. The structural characteristics of the obtained products after freeze-drying process was subjected to the Fourier Transform Infrared (FTIR) Spectroscopy measurements (Perkin-Elmer 16 PC). The optical properties of

the purified products were characterized with UV-visible spectroscopy (Perkin-Elmer Lambda 900) and fluorescence spectroscopy (Perkin-Elmer LS 55). The fluorescence lifetime of the oligomers dispersed in 0.2 M phosphate buffer saline solution (PBS, pH=7.4) was calculated from time-resolved fluorescence intensity decay on an Edinburgh Instruments FLS 920 (U.K.) The intensity decay curves were fitted with the equation:  $I(t) = \alpha \cdot \exp(-t/\tau)$ , where  $t$  is time,  $I(t)$  is the measured intensity decay,  $\alpha$  is the pre-exponential factor and  $\tau$  is the lifetime. For determining the quantum yield, the optical densities at 360 nm and fluorescence emissions of the diluted dopamine-derived oligomers and quinine sulfate in 0.1 M H<sub>2</sub>SO<sub>4</sub> were measured respectively. Then the quantum yield was calculated according to the

$$\varphi_x = \varphi_s \frac{I_x A_s n_x^2}{A_x I_s n_s^2}$$

following equation: where  $\varphi$  is the quantum yield,  $I$  is the integrated emission intensity, and  $A$  is the optical density, and  $n$  is the refractive index. The subscript 'x' and 's' represent the sample and the standard reference with known quantum yield. The stability was obtained by measuring the fluorescence intensity of the probes in 0.2 M PBS (pH=7.4) solution.

### **Fluorescence detection of Ferric Ions**

Typically, 500  $\mu$ L of 150  $\mu$ g/L fluorescent dopamine-derived oligomers dissolved in 10 mM PBS (pH 7.4) were mixed with 500  $\mu$ L of ferric ions solutions with final concentrations from 0 to 100  $\mu$ M, respectively. After a reaction of 5 min at room temperature, the fluorescence spectra was recorded under the excitation wavelength of 397 nm, the slit width for both the excitation and emission were set as 15 nm. The selectivity were evaluated by measuring the quenching spectra induced by different metal ions including K<sup>+</sup>, Na<sup>+</sup>, Ag<sup>+</sup>, Ca<sup>2+</sup>, Mg<sup>2+</sup>, Zn<sup>2+</sup>, Cu<sup>2+</sup>, Fe<sup>2+</sup>, Hg<sup>2+</sup>, Pb<sup>2+</sup>, Ni<sup>2+</sup>, Fe<sup>3+</sup>, Cr<sup>3+</sup>, Y<sup>3+</sup> and all the concentrations of metal ions were 20  $\mu$ M.

### **Bioconjugation of Oligomers**

For the bioconjugation with cRGD peptides, the mixture of EDC (0.8 M) and Sulfo-NHS (0.2 M) in 0.1 M phosphate buffer (pH = 7.4) with a final volume of 1 mL was injected in to 4 mL of 5 mg/mL purified oligomers under sonication for 30 min at 4 °C. Then 1 mL of 15 mg/mL cRGD peptide solution was added for another 1 h

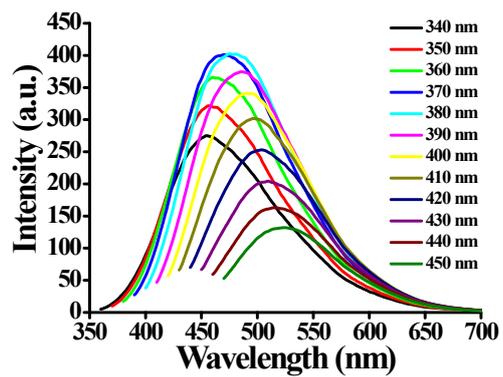
sonication. The cRGD-conjugated oligomers were purified with dialysis (MWCO = 1000 Da) and the volume of conjugated oligomer solution was concentrated to 2 mL under a drying flow of nitrogen gas. During the bioconjugation with GPR 120 primary antibodies, 10  $\mu$ L of 1 mg/mL antibody solution was firstly added into the mixture of 800  $\mu$ L 0.4 M EDC and 200  $\mu$ L 0.1 M Sulfo-NHS in 0.1 M  $\text{NaH}_2\text{PO}_4$  for a sonication of 30 min at 4  $^\circ\text{C}$ . Then 1 ml of 5 mg/L oligomer solution was added and the sonication was lasted for another 1 h. Residual reactants were removed by dialysis (MWCO = 7000 Da) and also the solution volume containing antibody-conjugated oligomers was concentrated to 1 mL before use.

### **Cell Culture and Imaging**

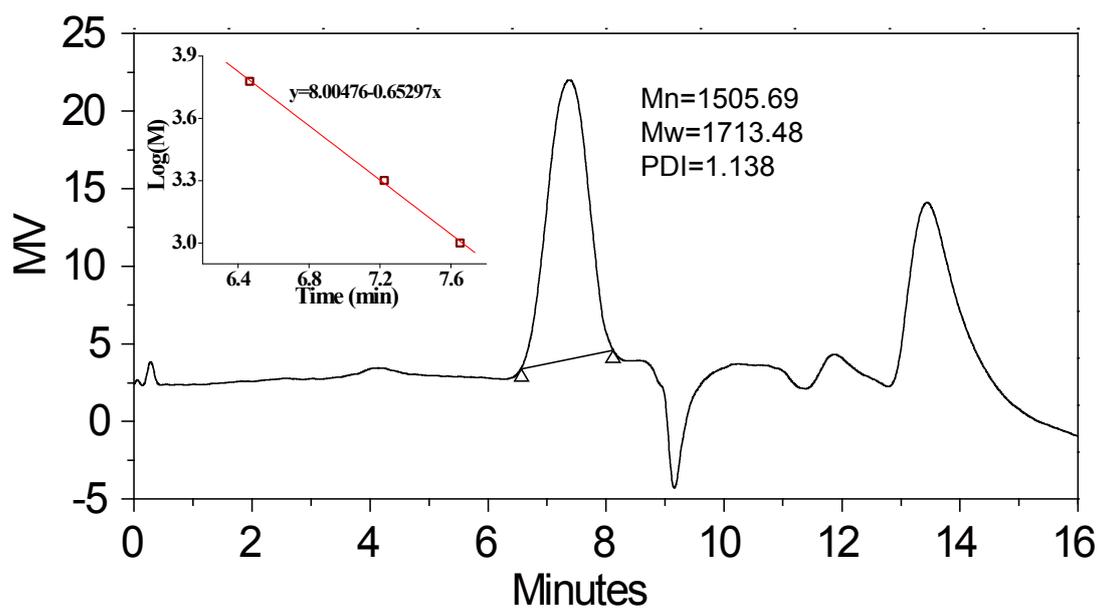
Human hepatoma (HepG2) cells and human cervical cancer (HeLa) cells were obtained from the American Type Culture Collection (ATCC, USA). The cells were maintained in Dulbecco's modified Eagle's medium (Gibco) supplemented with 10% fetal bovine serum (Gibco) at 37  $^\circ\text{C}$ , 5%  $\text{CO}_2$  in a humidified atmosphere. The cells were cultured in a plastic cell culture dish. The cell cytotoxicity of the prepared oligomers for both HepG2 and HeLa cells were tested by using standard MTT assays, where the incubation time was 24 h and the final concentrations of the probes ranged from 0 to 10 mg/mL. Also the effect of the probes on cell proliferation was studied by adding 10 mg/mL probes into the cells and then counting the cell numbers every 12 h. For labeling the HepG2 cells, 500  $\mu$ L obtained cRGD-conjugated oligomers and 500  $\mu$ L pure oligomers (at a concentration of 10 mg/mL) were respectively added into the culture medium for 4 h. Then the culture medium was refreshed and the cells were observed under fluorescence microscope. For targeted imaging of G protein-coupled receptor 120 (GPR120) on HeLa cell membrane, the fluorescent oligomers and GPR120 primary antibodies conjugated fluorescent oligomers were added into the medium for an incubation of 1 h respectively. After refreshing the culture medium, the labeled membrane proteins were imaged under fluorescence microscope. The bandpass of excitation and emission filters used for imaging are 379-401 nm and 460-500 nm, respectively.

Table S1. Quantum yield measurements by using quinine sulfate as standard reference

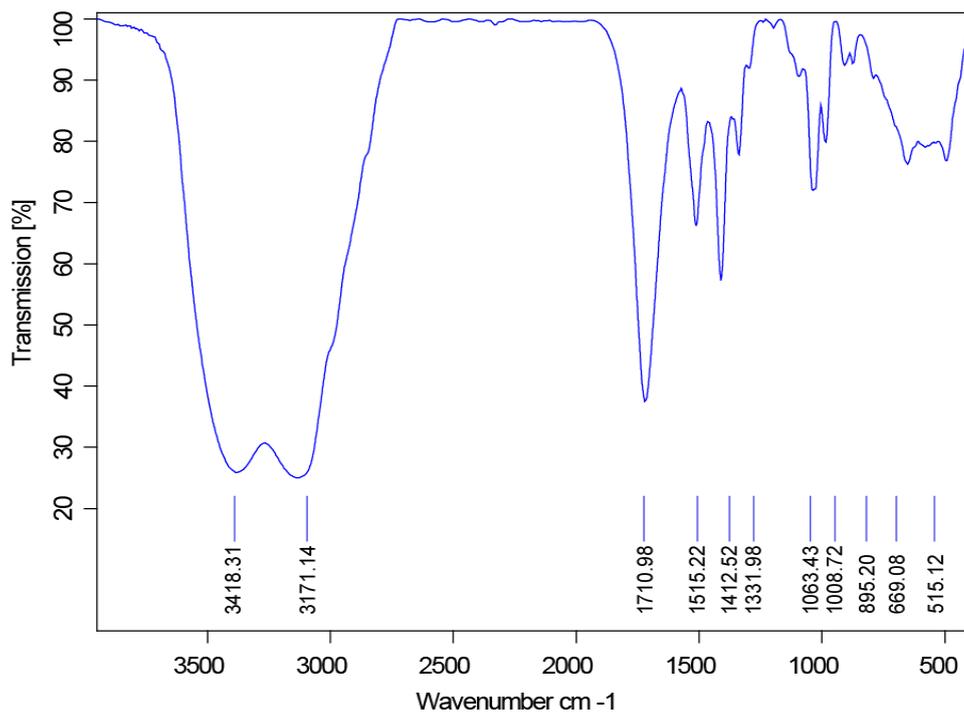
	Absorption at 360 nm	Emission intensity	Refractive index	Quantum yields
Quinine Sulfate	0.120	22141.321	1.33	54%
Oligomer	0.242	20050.460	1.33	<b>16.2%</b>



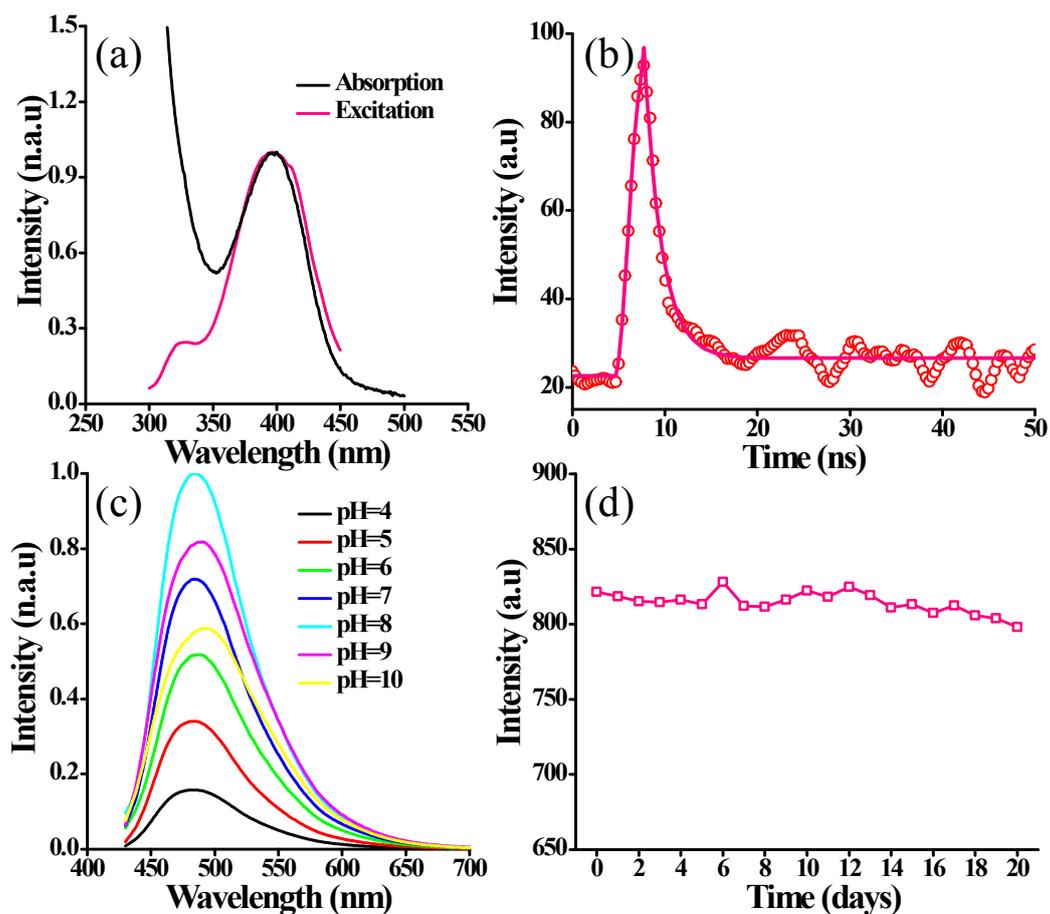
**Figure S1.** Fluorescence spectra of fluorescent polydopamine at different excitation wavelength, where the polymerization of dopamine was terminated with mercaptoethanol after 30 min.



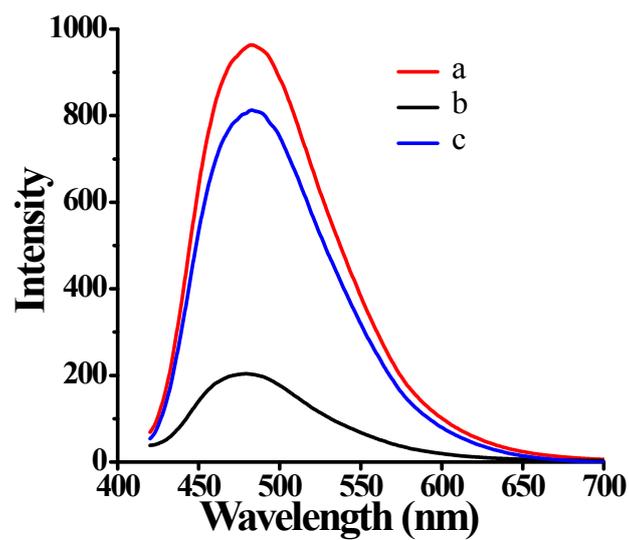
**Figure S2.** GPC characterization of the prepared dopamine-derived oligomers. The measured number averaged molecule weight and weight averaged molecule weight are 1505.69 and 1713.48, respectively, according to the measured calibration curve (inserted) with polyethylene glycol as standards.



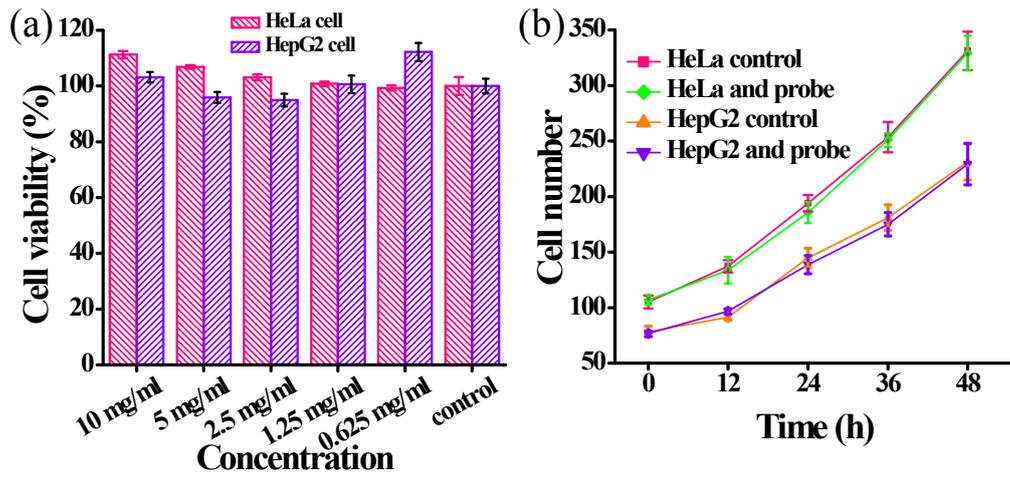
**Figure S3.** FTIR spectroscopy of dopamine-derived oligomers. The strong peak at 1710 cm<sup>-1</sup> and broad peak at 3171 cm<sup>-1</sup> can be assigned as the absorption bands of carboxylic acid groups. The broad peak at 3418 cm<sup>-1</sup>, and multiple peaks at 1515 and 1412.52 cm<sup>-1</sup> are associated with the characteristic absorption of phenol groups.



**Figure S4.** (a) Normalized absorption and excitation of the obtained dopamine-derived oligomers. (b) Measured (dot) and fitted (curve) fluorescence decay of the probes, indicating a lifetime of around 2.35 ns. (c) Normalized fluorescence spectra of dopamine-derived oligomers in 0.2 M PBS with different pH values. (d) Fluorescence stability measurements for 20 days.



**Figure S5.** Fluorescence spectra of the probes (a), mixture of solution a and  $\text{Fe}^{3+}$  ions with final concentration of  $50 \mu\text{M}$  (b), mixture of b and EDTA with final concentration of  $1 \text{ mM}$  (c), indicating the detection mechanism in our experiment is based on the coordination interaction between  $\text{Fe}^{3+}$  ions and the probes.



**Figure S6.** (a) Viability of cells when treated with dopamine-derived oligomers at different concentrations from 0 to 10 mg/mL. (b) Cell proliferation with or without the treatment of oligomer probes at a final concentration of 10 mg/mL.