Supporting Information

Electrochemical-TUNEL method for sensitive detection of apoptotic cells

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Experimental Section

Reagents. Simvastatin which used to induce apoptosis was bought from HEOWNS. TdT was acquired from Thermo scientific. Biotin-16-dUTP was purchased from PromoCell Gmbh Sickingenstrasse 63/65 69126 Heidelberg Germany. Multi-walled carbon nanotubes (c-CNTs) (1.0-2.0 μm in length and 20 -40 nm in diameter with purity level above 95%) were obtained from Shenzhen Nanotechnology Co. Ltd. 1-Ethyl-3-(3-dimethylaminopropyl) carbodiimide hydrochloride (EDC), N-hydroxysuccinimide (NHS), bovine serum albumin (BSA), dopamine (DA), folic acid (FA) and Calcein-AM were obtained from Sigma/Aldrich (St. Louis, MO).

Apparatus and Characterizations. Field emission scanning electron microscopy (FE-SEM) images were obtained using a Hitachi S-4800 field emission electron microscope. Fourier transform infrared (FT-IR) spectra were recorded on a Vector 22 FT-IR spectrometer (Bruker). Samples were thoroughly ground with exhaustively dried KBr. All electrochemical measurements were performed on a CHI 660D workstation (Chenhua, Shanghai, China) with a conventional three-electrode system composed of a platinum wire as the auxiliary, a saturated calomel electrode as the reference, and the modified glass carbon electrode (GCE) as the working electrode. Electrochemical impedance spectroscopy (EIS) was performed with an Autolab electrochemical analyzer (Eco Chemie, The Netherlands) in a 10 mM K3Fe(CN)6/K4Fe(CN)6 (1:1) mixture with 1.0 M KCl as the supporting electrolyte, using an alternating current voltage of 5.0 mV, within the frequency range of 0.01 Hz−100 kHz.

Preparation of c-CNT.

200 mg of CNTs were dispersed in a mixture of sulfuric acid and nitric acid (3:1) and sonicated for about 4 h to obtain carboxylic group-functionalized CNTs. After centrifugation from the mixture, the sediment was washed repeatedly with distilled water until the pH reached 7.0.

Preparation of the CNTs@PDA-FA nanocomposite. 10.0 mg of c-CNTs was first dispersed into 2.0 mL of Tris buffer (pH 8.5) and sonicated at room temperature for 10 min. After centrifugation of the mixture, the sediment was washed repeatedly with Tris buffer and dispersed into 2.0 mg mL^{-1} of dopamine Tris buffer (pH 8.5) and incubated in the dark at 37 ℃for 24 h. The obtained brown suspension was washed with Tris buffer and double distilled water for three times (3000rpm, 5min) to obtain the CNT@PDA nanocomposites. 5 mg of folic acid, 2.1 mg of EDC and 3.2 mg of NHS were mixed in 20 mL of DMSO/PBS (v/v=1:10) solution at room temperature for 2 h to modify the terminal carboxylate group. Then, 5.0 mg of CNT@PDA nanocomposite was added into the solution, stirred at room temperature for 12 h. After centrifugation (13000rpm, 10min), the collected CNT@PDA-FA nanocomposites were washed and redispersed in PBS (pH 7.4) to the concentration of 1.0 mg mL^{-1}.

Preparation of SA-SiO_2@CdTe probe. Quantum dot were synthesized as previous reported with small modification [1], Te powder was replaced by $NaTeO_3$ for avoiding oxidation. SiO_2 nanoparticles (about 30 nm) was selected to magnify the electrical signal by assisting streptavidin in adsorbing more QDs. Briefly, 10 mg of SiO_2 was dispersed into water to get a homogeneous solution under ultrasonic condition. Positive charged PDDA was covered on the silicon ball in terms of Si:PDDA=1:1,washed 2 times by centrifugation (12000 rpm, 6 min). Then, purified

QDs were mixed with the precipitation under ultra-sounding assistance and then centrifuged and redispersed into PBS to get SiO_2@QDs solution. After that, 3.2 mg of EDC and 0.1 mg of streptavidin (SA) were added to the solution and shaked for 2 hours at room temperature. After being cleaned with PBS, the final material called SA-SiO_2@QDs probe was put into 1.0 ml of PBS buffer contained 3% BSA before using.

Cell culture and apoptosis. K562 cells were cultured in a petri dish in RPMI 1640 medium (GIBCO, Grand Island, NY) containing 10 % fetal calf serum, penicillin (100 ug/ml), and streptomycin (100 ug/ml) in an incubator (5% CO_2, 37℃). The cells solution were collected and separated from the medium at the logarithmic growth phase by centrifugation at 1000 rpm for 5 min and then re-suspended in fresh culture medium to obtain a homogeneous cell suspension with the final concentration of 5×10^6.

Simvastatin was used to induce K562 cell apoptosis. 4 ml of medium containing 100 μl of cell solution and 27 μl of apoptotic inducer (50 nmol/ml) was added in 6-well-plate. Then we took all of them into the incubator (5% CO_2, 37℃) for different time.

Electrochemistry detection of apoptotic cells. Before modification, glassy carbon electrode (GCE, 3 mm in diameter) should be cleaned with 0.05 μm and 0.3 μm alumina slurry. Next, GCE was rinsed with water, alcohol in turns to obtain the smooth and bright surface, stored in the vacuum drier for further using.

5μL of the prepared CNTs@PDA-FA solution was dropped to the GCE and dried in a silica gel desiccator to fabricate a functional film (CNTs@PDA-FA/GCE). The modified electrode was subsequently immersed into 10.0 mg mL^{-1} BSA solution for 1 h at 37℃ to prevent the nonspecific adsorption followed by being carefully rinsed

with 0.1 M PBS (pH 7.4) to remove physically absorbed protein. After that, the electrode was immersed in cells solution and incubated at 37℃ for 1 h to capture cells; and then washed by PBS thoroughly to take off the unbound cells. 10 μl of fresh prepared TUNEL reaction buffer was dropped on the electrode for another 1 h of incubation at 37℃.

After the capture of apoptotic cells, the electrode was immediately incubated with 10 μL of SA-SiO_2@QDs nanoprobe at 37 ℃ for 1 h. Then the electrode was washed thoroughly with incubation buffer. Afterwards, the electrode was immersed in 0.4 ml of 0.1 M HNO_3 solution for 2 h to dissolve the captured QDs nanoprobe, and the resulting solution was mixed with 4.5 mL of 0.2 M HAc-NaAc buffer (pH 4.6) to perform anodic stripping voltammetric (ASV) detection with a mercury film modified GCE.

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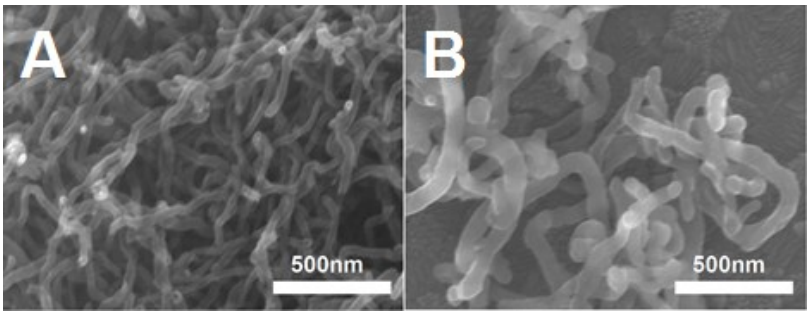

Fig. S1. SEM images of (A) CNTs (B) CNTs@PDA.

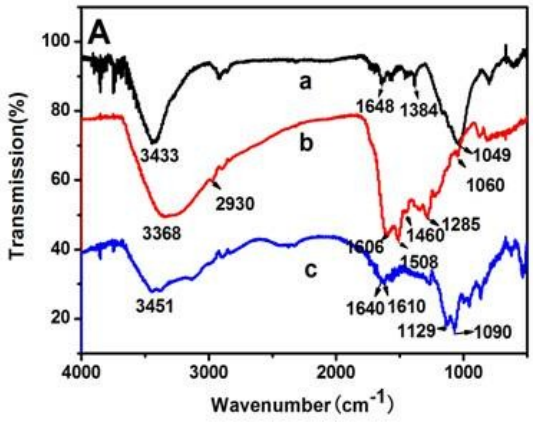

Fig. S2. FI-IR spectra of (a) CNTs, (b) CNTs@PDA and (c) CNTs@PDA-FA

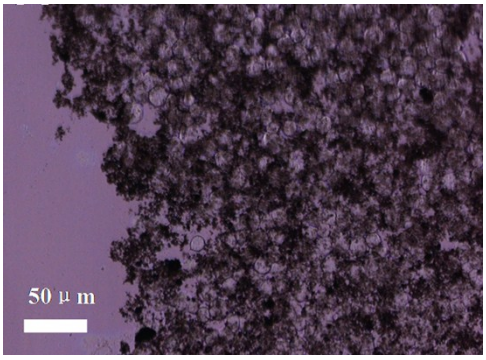

Fig. S3. The microscope images of cells covered on the ITO.

1. L. Zou, Z. Gu, N. Zhang, Y. Zhang, Z. Fang, W. Zhu and X. Zhong, *Journal of Materials Chemistry*, 2008, **18**, 2807-2815.