***Supplementary Information* for**

Potentiometric Nanosensor for Real-Time Measurement of Hydrogen Sulfide in Single Cell

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Experimental Section

1 Regents and Materials. $Na_2S·9H_2O$, $AgNO_3$, KNO_3, L-cystine (Cys), glutathione (GSH), ascorbic acid (AA), dopamine (DA), $CaCl_2$, calcein-AM, Propidium iodide (PI) and $Ru(NH_3)_6Cl_3$ were purchased from Sigma-Aldrich (shanghai, China). 3,4-Dihydroxyphenylacetic acid (DOPAC), uric acid (UA), and serotonin hydrochloride (5-HT) were purchased from Alfa Aesar. HEPES buffer solution contained NaCl (150 mM), KCl (5 mM), $CaCl_2·2H_2O$ (2 mM), $MgCl_2·6H_2O$ (1.2 mM), H-HEPES (10 mM), D (+)-Glucose (5 mM). High concentration K^+ solution contains NaCl (55 mM), KCl (100 mM), $CaCl_2·2H_2O$ (2 mM), $MgCl_2·6H_2O$ (1,2 mM), H-HEPES (10 mM), D (+)-Glucose (5 mM). Aqueous solutions were prepared with Milli-Q water (18.2 MΩ·cm^{-1}). Purification was performed with 0.22 μm filter membrane before using for the cells. RPMI 1640 culture medium and other related reagents for cell culture were bought from GIBCO (U.S.A.).

2 Apparatus and Measurements. The electrochemical potential measurements were conducted in two-electrode system using a computer-controlled CHI 760e electrochemical analyzer (Shanghai Chenghua, China). Ag_2S/AgNPs/CFNE was used as a working electrode and microsized Ag/AgCl which is manufactured with a micropipette was used as a reference electrode. Scanning electron microscopy (SEM) images were recorded with a microscope (Nikon Japan). SEM analysis was implemented for visual characterization of nanoelectrode and determination of nanoelectrode width. X-Ray photoelectron spectra (XPS) were collected on VG Scientific ESCALab220i-XL X-Ray photoelectron spectrometer, using Al Ka radiation as the excitation sources. The generating S^0 electrochemical reaction was carried out in PBS buffer containing 1 mM $Ru(NH_3)_6^{3+}$ and 1 mM Na_2S at a voltage of 0.0 V for 1000 s.

3 Fabrication of Ag_2S/Ag NPs/CFNE. Carbon fiber microelectrodes were fabricated firstly and then carbon fiber nanoelectrodes (CFNE) was prepared by flame etching until the electrode tip was controlled within 500 nm. The prepared electrodes were activated in 0.5 M H_2SO_4 and were electrodeposited Ag nanoparticles (AgNPs) on the surface of CFNE in 0.1 M KNO_3 and 2.5 mM $AgNO_3$ with three-electrode system at – 0.2 V for 300 s (Pt wire and Ag/AgCl were used as counter and reference electrodes, respectively). Finally, the AgNPs/CFNE were treated with 5 μM Na_2S until the potential balanced at Open Circuit Potential (OCP) for 300 s, forming Ag_2S/Ag NPs/CFNE.

4 Cell Culture and Cell Experiments. SH-SY5Y (Human neuroblastoma cells) were purchased from Natural Infrastructure of Cell Line of China. SH-SY5Y were maintained in Dulbecco's modified Eagle medium (RIMP 1640, high glucose, Gibco) and were cultured in a petri dish 37 °C with 5% CO_2 in a 95% humidified atmosphere. The fresh culture medium RIMP 1640 contain 10% of fetal bovine serum (FBS) and 100 U·mL^{-1} penicillin, streptomycin. In order to identify electrode with good biocompatibility, Calcein-AM (live/green cytoplasmic stain) and propidium iodide (PI) (dead/red nucleic stain) were added into the medium to stain these cells after being inserted for about 80 s. The dish with the cells was fixed on a microscope (Nikon, Japan) table, and the electrodes were moved near the target cells by a micromanipulator MPC-200 (Canon, America) for in situ detection. Besides, a micropipette with an open tip filled with solution was connected with micro-injection pump (2 μL/s for 10 s) as a stimulant. When the potential arrived to the steady state, acetylcysteine, HEPES solution and K^+ solution was injected to the cell suspension through the prepared stimulant, which can motivate cells generation of H_2S.

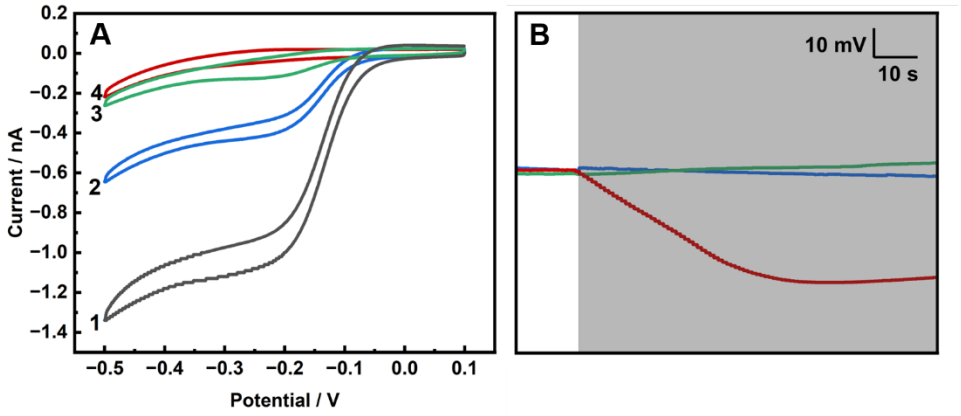

Figure S1. (A) CVs at CFNE inserted into a SH-SY5Y cell for different depths in HEPES containing 1 mM $Ru(NH_3)_6^{3+}$. Curve 1: before insertion, curve 2: 60% insertion, curve 3: 80% insertion, curve 4: nearly complete insertion. (B) OCPs at CFNE inserted into a SH-SY5Y cell for different depths (bule line: 60% insertion, green line: 80% insertion, red line: nearly completely insertion) in HEPES cell bath containing 1 mM $Ru(NH_3)_6^{3+}$.

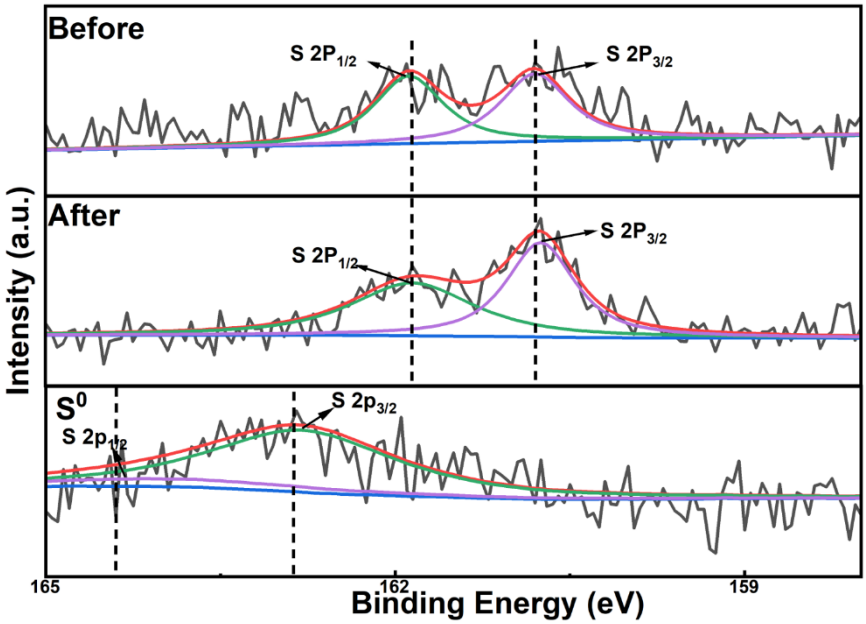

Figure S2. S2p XPS spectra of Ag_2S/AgNPs/ITO before and after continuous detection of H_2S for 1000 s, and S^0/ITO.