

Supplementary information

Materials: Silver nitrate ($\geq 99.8\%$, $M_w=169.87$ A.R.) and thioacetamide (TAA) ($\geq 99.0\%$, $M_w=75.13$ A.R.) were purchased from Tianjin Chemical Reagent Factory. Bovine serum albumin (purity $\geq 98\%$, $M_w=68\ 000$, BSA) was purchased from Xiamen Sanland Chemicals Company Limited, China.

Preparation of Ag₂S nanoparticles, nanorods and nanowires: Ag₂S nanoparticles were synthesized under ambient conditions. Briefly, 50 ml of 50 mM silver nitrate aqueous solution, and the mixed solution containing 100 ml of 1 mg/ml BSA and 50 ml of 50 mM TAA, used as the source of S²⁻, were added in flask at the same time. Then the solutions were mixed by stirring for a homogeneous reaction. Immediately, the solution changed to black. The mixed reaction solution was kept static under ambient conditions for 72 h. After being separated by high-speed centrifuging at 25 000 rpm, the collected black solid-state product was washed three times with double distilled water and ethanol, and then dried in a vacuum at room temperature for 24 h. Three replicas of the same experiment were run in parallel.

The synthesis of Ag₂S nanorods was performed by a two-step procedure. The first step was the generation of the silver (I) -BSA complex by mixing reaction of the AgNO₃ and BSA solutions for 6 h. The second step was the formation of Ag₂S nanorods by adding TAA into the above mixing solution and then it was kept static under ambient conditions for 72 h at ambient temperature. All the other parameters were the same with the synthesis of nanoparticles.

Ag₂S nanowires were synthesized using AAO tubular membranes as the physical environment. The AAO tubular membrane was prepared by a two-step aluminum anodic oxidation process in a 4 wt % oxalic acid solution as described previously.¹ The thickness of the membrane is about 20 μm , and the diameter of the tubes in the membrane is 50 nm. The AAO tubular membrane was mounted between the two halves of an H-tube cell comprising two reservoirs of solutions, each of which contains a part of the insoluble salt (Ag₂S). 50 ml of 50 mM silver nitrate aqueous

¹ H. Masuda and K. Fukuda. *Science*, 1995, 268, 1466-1468.

solution and 100 ml of 1 mg/ml BSA aqueous solution were mixed with vigorous stirring at room temperature. Then the BSA-Ag⁺ emulsion was added to one of the half-cells and kept static for 6 h. Then 150 ml of 50 mM TAA aqueous solution was added to the other half-cell. After immersion time of up to 72 h at room temperature (20 °C), the AAO tubular membrane embedded with Ag₂S nanowires was detached and thoroughly washed with deionized water. Then the membrane was ultrasonically dissolved in 2 M NaOH aqueous solution for 15 min, and separated by high-speed centrifuging at 20 000 rpm. The collected black solid-state product was washed three times with double distilled water and dried in vacuum for 24 h.

Characterization: To prepare the transmission electron microscope samples (TEM, FEI Tecnai G220, USA), a 5 µl droplet of dilute alcohol solution was dripped onto a holey carbon-coated formvar support. TEM equipped with selected-area electron diffraction (SAED) was used to confirm the samples size and determine the form of attachment. Scanning electron microscopy (SEM, JSM-5600LV, Japan) was used to observe the morphology of the samples after vacuum-coated with a gold layer.

Inhibition effect of nanoparticles, nanorods, and nanowires on tumor cells: C6 glioma cell line was used as the model to study the inhibition effect of nanoparticles, nanorods and nanowires on tumor cells. The C6 cells were routinely cultured in tissue culture flasks with High Glucose-Dulbecco's Modified Eagle's Medium (HG-DMEM), containing 10% fetal bovine serum and incubated at 37 °C in a humidified atmosphere with 95% air and 5% CO₂. The culture medium was refreshed every two days. When the cells became almost confluent after 5 days, they were released by treatment with 0.25% trypsin. The cells were counted to 10⁴ cells/cm² and 200 µl of cells suspension were pipetted into 96-well tissue culture plate. After 2 hours of culture, the medium was replaced with HG-DMEM medium containing 0.05 mg/ml of Ag₂S bulk crystals, or nanoparticles or nanorods or nanowires, and continued to culture another 48 hours. In addition, the group without any crystals was chosen as the control.

Cells proliferation was measured by 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl tetrazolium bromide assay (MTT) based on succinic dehydrogenase activity at OD490 nm (n = 6), 630 nm was chosen as the reference wavelength. And the inhibition rates

(IR) of nanoparticles, nanorods, nanowires, and bulk crystals on C6 cells were calculated as follows:

$$IR = (ODc - ODs) / ODc \times 100\%$$

ODc and ODs are the OD490 nm values of control group and sample group (containing nanoparticles, or nanorods, or nanowires, or bulk Ag_2S), respectively. For each sample of MTT Measurements was performed three times independently.

Results: Fig. 1 shows the SEM image of Ag_2S bulk crystals, nanoparticles, nanorods, and nanowires. The morphologies of Ag_2S bulk crystals were ruleless, and the size was very inhomogeneous varying from several microns to over 100 micron as shown in Fig. 1a. The nanoparticles exhibited a sharp particle distribution with an average particle diameter of 65 nm as shown in Fig. 1b. The nanorods and nanowires were with the mean sizes 40, 50 nm in diameter and 220 nm, over 1000 nm in length, respectively (Fig. 1c and 1d). All of these nanocrystals were obviously well dispersed and uniform.

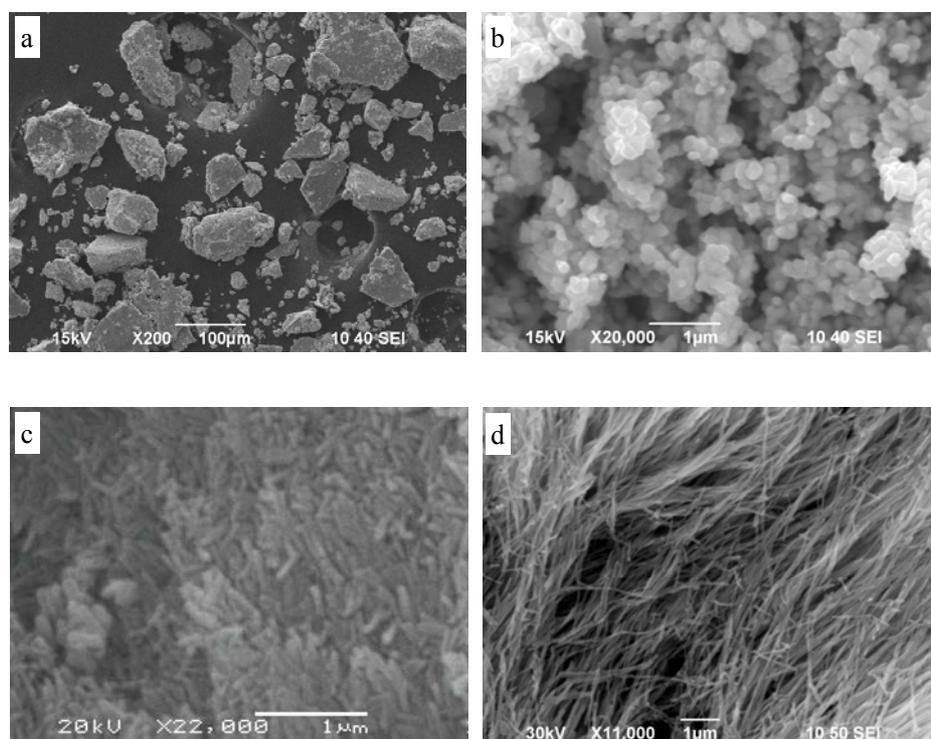


Fig. 1 SEM images of Ag_2S bulk crystals (a), nanoparticles (b), nanorods (c), and nanowires (d)