

CHEMBIOCHEM

Supporting Information

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for

Immuno-Carbon Nanotubes and Recognition of Pathogens

Tara Elkin, Xiuping Jiang, Shelby Taylor, Yi Lin, Lingrong Gu, Hua Yang,
Jessica Brown, Susan Collins, and Ya-Ping Sun*

Experimental Details

Materials. The *E. coli* O157:H7 strain C7927 was kindly provided by Prof. Michael P. Doyle (University of Georgia). The affinity-purified goat anti-*E. coli* O157 antibody (Ab₁) and tetramethyl rhodamine isothiocyanate (TRITC)-labeled rabbit anti-goat IgG (H+L) (Ab₂) were supplied by Kirkegaard & Perry Laboratories, and the green fluorescence protein (plasmid vector pGFPuv) by ClonTech. The BSA and the Micro-Lowry Total Protein Kit (Onishi & Barr modification, Lot #043K6021) were purchased from Sigma-Aldrich, and 1-ethyl-3-(3-dimethylaminopropyl) carbodiimide hydrochloride (EDAC, 98+%) from Alfa Aesar.

The arc-discharge generated SWNT sample was supplied by Prof. A. M. Rao (Physics Department, Clemson University), and was purified via oxidative acid treatment by following a well-documented procedure.¹⁵

Measurements. AFM was conducted in the acoustic AC mode on a Molecular Imaging PicoPlus system equipped with a multi-purpose scanner for a maximum imaging area of

10×10 μm. SEM images were obtained on a Hitachi S4700 field-emission SEM system. Confocal microscopy analysis was carried out on a Carl Zeiss LSM 510 Laser Scanning Microscope. Optical absorption spectra were recorded on Shimadzu UV3100 and Thermo-Nicolet Nexus 670 FT-NIR spectrometers. TGA was performed on a Mettler-Toledo TGA/SDTA851e system.

***E. coli* O157:H7 Culture.** *E. coli* cells were grown in Tryptic soy broth supplemented with ampicillin (100 μg/mL) in a shaker incubator at 200 rpm for about 22 h. The as-cultured bacterial cells were washed thrice with a sterile NaCl solution (0.85%), and then suspended and diluted in the same NaCl solution to an optical density of 0.7 at 630 nm (corresponding to bacteria concentration ~ 10⁹ cfu/mL.²¹ The bacteria count was enumerated after spreading a series of dilutions (100 μL) of the bacterial suspension on Tryptic soy agar plates and allowing the bacteria to grow overnight in an incubator at 37 °C.

Specimens for SEM. In a typical experiment, a sample (immuno-SWNT - *E. coli*) was suspended in a gluteraldehyde solution (2.5%, 1 mL), allowed to fix at room temperature for 30 min, and passed through a filter membrane (0.2 μm, Whatman Nuclepore Polycarbonate). The collected specimen on the filter membrane was allowed to fix for an additional 30 min and then rinsed repeatedly with PBS buffer solution. The specimen was post-fixed by using freshly prepared 1% osmium oxide (enough to cover the filter) for 1 h and rinsed thoroughly with double-distilled water. The bacteria cells were dehydrated with ethanol solutions of gradient concentrations (50% for 30 min, 75% for 10 min, 85% for 10 min, 95% for 10 min, and twice with 100% for 10 min). After complete evaporation of ethanol, the specimen was mounted onto an aluminum stub and then coated with platinum (2 min) for the purpose of minimizing surface charging effects during imaging.