Supplementary Material for Chemical Communications

Protein micropatterning based on electrochemically switched immobilization of bioligand on electropolymerized film of a dually electroactive monomer

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Experimental procedure

Tris(2-carboxyethyl)phosphine (TCEP), $HAuCl_4 \cdot 3H_2O$ and sodium citrate dehydrate, N-ethylmaleimide were obtained from Aldrich and were used as received without further purification. Tetrabutylammonium perchlorate (TBAP) and acetonitirle (ACN) were purchased from Merck. TBAP was dried in vacuum oven before use. Maleimide PEO_2-Biotin as a maleimide-conjugated biotin (MCB) was purchased from Pierce. Streptavidin-conjugated quantum dot was obtained from Quantum Dot Corp. Ultra-pure water (>18MΩ) from a Modulab water system (U.S. Filter Corp.) was used throughout this work. All glasswares were cleaned for 6h in Nochromix (Godax Lab., Inc.) cleaning solution, 6h in conc. HNO_3, and then rinsed with ultra-pure water. ACN and TBAP were dried before use. CV for the electropolymerization was performed in the ACN solution containing TBAP (0.1 M) using BAS 100B (Bioanalytical Systems, Inc.) potentiostat. A polymer film/ITO working electrode, Pt wire counter electrode, and 'no-leak' Ag/AgCl (Cypress Systems) reference electrode were used. For chemical reduction, the polymer film of hydroquinone monoester-conjugated disulfide (HMDS) was immersed into an aqueous solution containing 25 mM TCEP for over 4 h. For electrochemical reduction and oxidation, polymer-coated electrode was held at −500 and 300 mV vs Ag/AgCl in ACN with 0.1 M TBAP as supporting electrolyte for over 20 sec. AFM imaging was performed in the Tapping Mode on a Nanoscope IIIa Multimode Scanning Probe Microscope (Digital Instruments, Santa Barbara, CA) with Tapping Mode etched silicon probes. Scanning Electron Microscope (SEM) image was obtained by using Philips XL30SFEG. Reflectance FT-IR spectrum was acquired using IFS 66V FT-IR spectrometer (Brucker). To immobilize MCB, the film surface is

incubated in a solution of 1 mM MCB in pH 7.2 PBS with 1 mM EDTA at room temperature for over 5 h. And, the film surface is rinsed with buffer solution and then dried with nitrogen gas. For SA micropatterning, the biotin-modified array was immersed in pH 7.2 PBST (0.05% Tween 20) solution of streptavidin-conjugated quantum-dot (1:100) for 20 min. ITO on glass substrate was purchased from HOYA Co. The fabrication procedure of a band ITO microarray is as follows. We used image reversal of 1.4 μm PR layer (AZ5214) because pattern mask was fabricated mechanically using SUS-plate which has image-reversed masking characteristics. With the patterned PR layer upon ITO slides, we place slides in etchant solutions (HCl, H_2O, HNO_3 4:2:1 by volume, where HCl ~ 32% and HNO_3 70% concentration). After around 60 seconds at room temperature, ITO layer open to etchant was dissolved away almost. PR was removed by cleaning in acetone and isopropyl alcohol. Rinse in deionized water and dry immediately blowing with nitrogen. The resulting pattern was checked by optical microscope and finally confirmed the line pattern resistance and resistance between pattern and non-pattern area with multimeter.