Electronic Supplementary Information

Self-assembled peptides on polymer surfaces: towards morphology-dependent surface functionalization

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Materials. PEI (Ultem 1000®, M_n = 12 000, M_w/M_n = 2.5, GE Plastic) was kindly provided by Fuji Electric Systems. PEI films were prepared by spin-coating (2000 rpm, 1 min) from a chloroform solution (1.7 mg ml^{-1}) onto the substrates. The obtained films were immediately used for the following experiments. BSA, streptavidin (SAv), and avidin (Av) were purchased from Sigma-Aldrich, Thermo Fisher Scientific, and Nacalai Tesque, respectively. They were dissolved in HBS-N (10 mM HEPES buffer containing 150 mM NaCl, pH 7.4, GE Healthcare) solution, and the solutions were stored at -20 °C until use. The peptides were synthesized by conventional solid-phase methods using the 9-fluorenylmethyloxycarbonyl (Fmoc) strategy, following our previous study[1]. The amounts of peptide immobilized on the PEI surfaces for AFM, ATR-IR, static contact angle, and ζ-potential measurements were adjusted by the peptide concentration and immobilization time, based on a binding analysis of the peptides on the PEI surfaces by SPR measurements.

SPR measurements. A Biacore X instrument (GE Healthcare) was used for the SPR analyses, following our previous study[1]. PEI films were prepared onto gold-coated glass slides (SIA Kit Au, GE Healthcare), and set on the SPR apparatus. HBS-N (10 mM HEPES buffer containing 150 mM NaCl, pH 7.4, GE Healthcare) was flowed at a rate of 20 μL min^{-1} at 25 °C during the experiment. After more than 3 h of HBS-N flow, freshly prepared peptide solutions were applied to the PEI films for 180 sec (association), and then the peptide solutions were exchanged to a peptide-free buffer for 900 sec (dissociation). The resulting sensorgrams at 4 concentrations were analyzed by global fitting using BIA evaluation software version 4.1. For the protein adsorption experiments, the peptides were immobilized onto the PEI films, and then after 5 min of HBS-N flow, the protein solution (1 μM in HBS-N) was applied for 5 min at a flow rate of 5 μL min^{-1} at 25 °C.

AFM measurements. The surface morphologies were visualized by non-contact mode AFM (SPM-9600, Shimadzu) in air using a silicon cantilever (PointProbe, NCH, resonance frequency 320 kHz, force constant 42 N m^{-1}, NanoWorld). All AFM images were flattened using software supplied by Shimadzu without further image processing.

ATR-IR absorption spectroscopy. The ATR-IR spectra of p1-EK immobilized onto PEI films were obtained using the refractive surface of gold-coated glass slides (SIA Kit Au, GE Healthcare) with a Spectrum One apparatus (Perkin-Elmer) in air at ambient temperature. The interferograms were co-added 50 times, and were Fourier transformed at a resolution of 4 cm^{-1}.

CD spectroscopy. The CD spectra were recorded on a CD spectropolarimeter (J-725, Jasco). The peptides were dissolved in 10 mM phosphate buffer (pH 7.4) to a concentration of 10 μM. The spectra were recorded in a 1 mm quartz cell over 190-240 nm with buffer baseline subtraction. Ten scans were averaged using a 1 nm band width at a scanning rate of 20 nm min^{-1}.

Static contact angle measurements. The static contact angle of PEI films (~10 nm thick) on Si wafers was measured in HBS-N with a commercial apparatus (CA-X, Kyowa Interface Science), following our previous paper[1]. After the PEI films were conditioned in water, 3 μL of sessile air bubbles were attached to the underside of the PEI films in water using a microsyringe. A monitor captured the bubble shapes, and the contact angles were calculated. The contact angles were measured 4 times at different locations.

Z-potential measurements. The ζ-potential of the PEI films was measured by an electrokinetic analyzer (SurPASS, Anton Paar) equipped with an adjustable gap cell (20 mm × 10 mm) based on the streaming potential method. PEI films with an area of 20 × 10 mm^2 prepared on polyimide films were placed in the measuring cell. The films were separated by a spacer that formed a streaming channel. The streaming potential was then detected by Ag/AgCl electrodes. A background electrolyte of 1 mM KCl solution was used, and the pH was adjusted with 0.1 M HCl and 0.1 M KOH.

References

1. T. Date, J. Sekine, H. Matsuno and T. Serizawa, *ACS Appl. Mater. Interfaces*, 2011, **3**, 351.

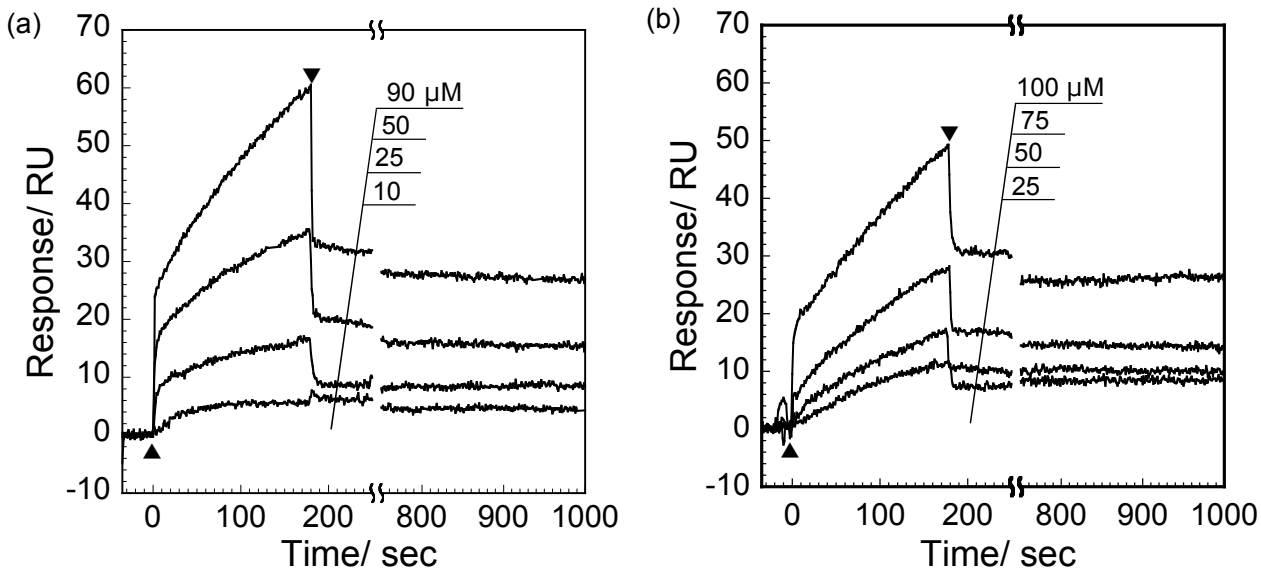

Fig. S1. SPR sensorgrams for the binding of (a) p1-EK and (b) EK to the PEI surface at four concentrations. The ▲ and ▼ indicate the points at which the injection of the peptide solution started and ended, respectively.

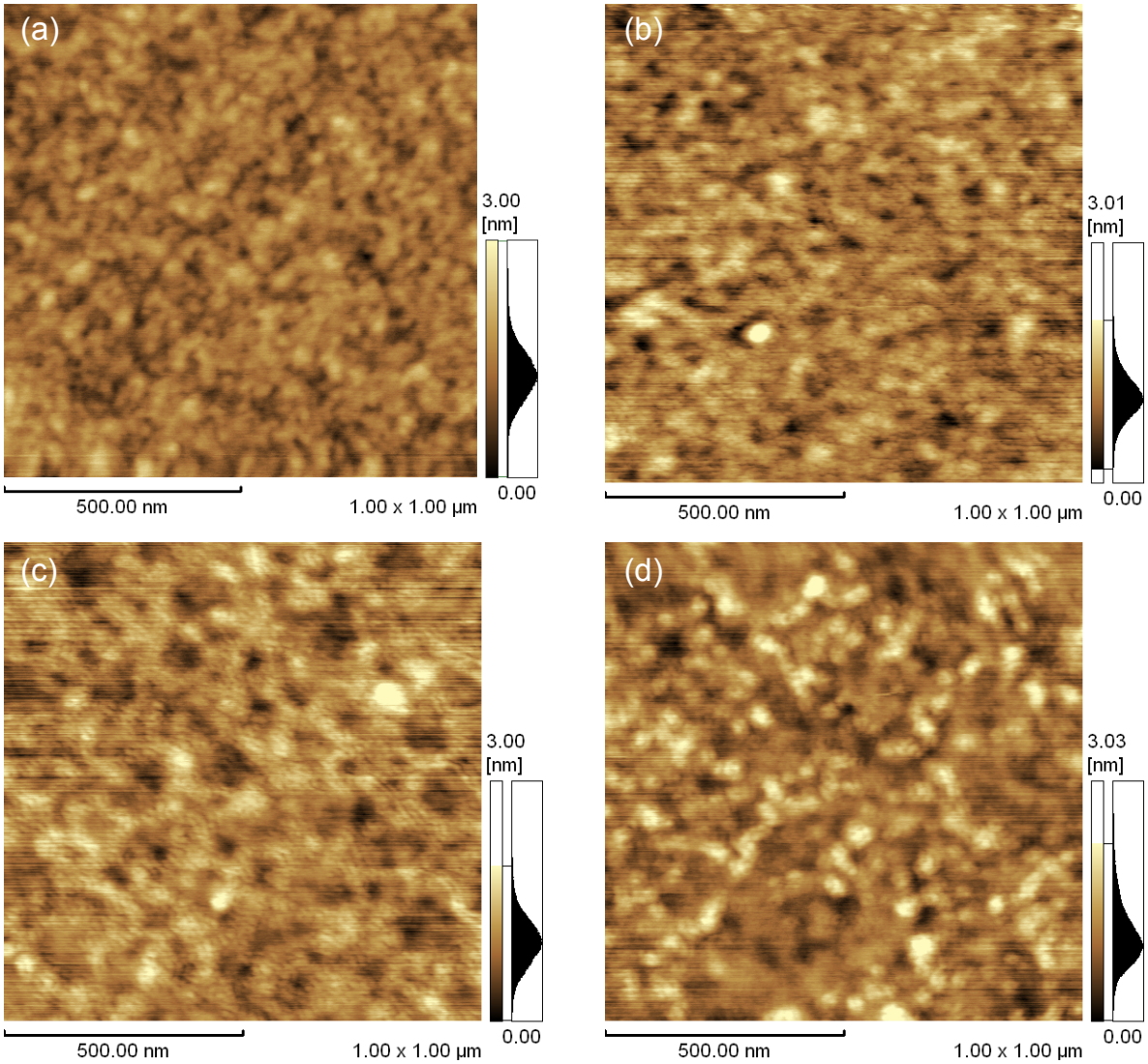

Fig. S2. AFM height images of (a) the bare PEI surface, the peptide-immobilized PEI surfaces with a peptide amount of (b) 200, (c) 400, and (d) 500 pmol cm^{-2}.

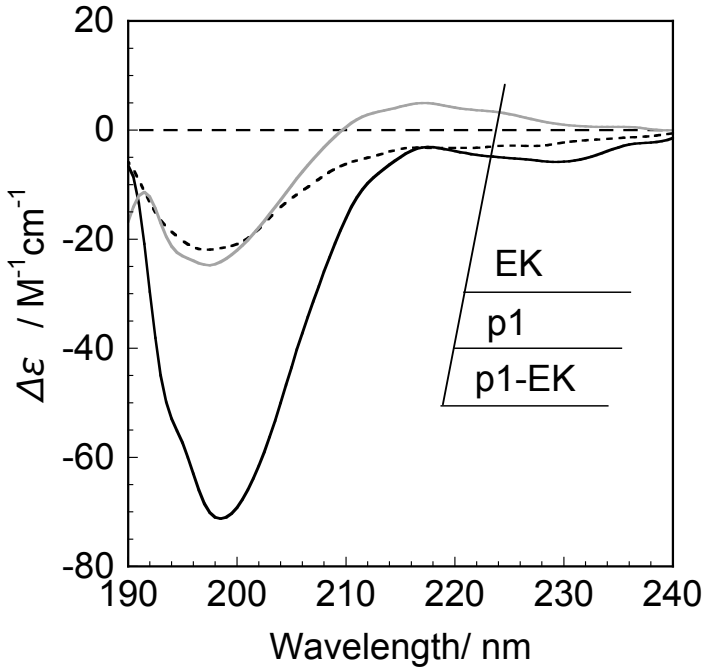

Fig. S3. CD spectra of p1-EK, p1, and the EK sequence in their aqueous solutions. The negative peaks at 190-200 nm indicate that all peptides have random coil conformations.

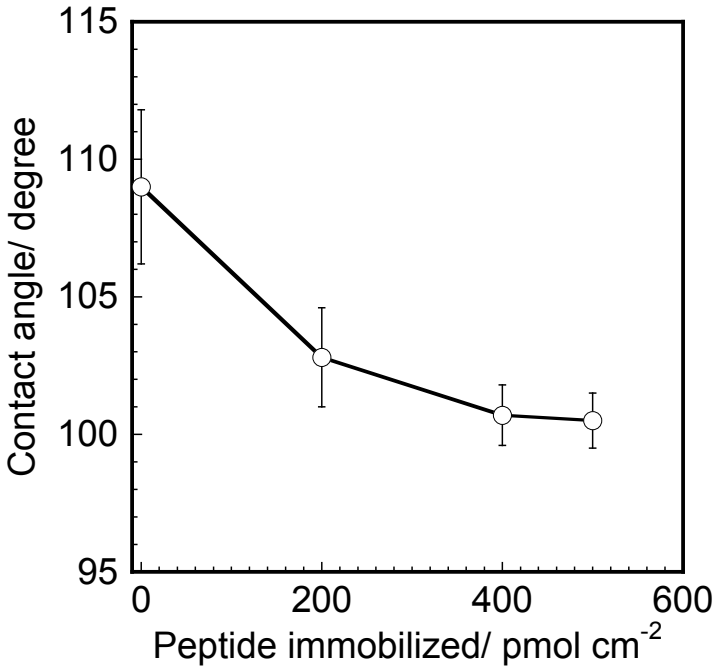

Fig. S4. The static contact angles of air bubbles for the PEI surfaces against the amounts of immobilized p1-EK.

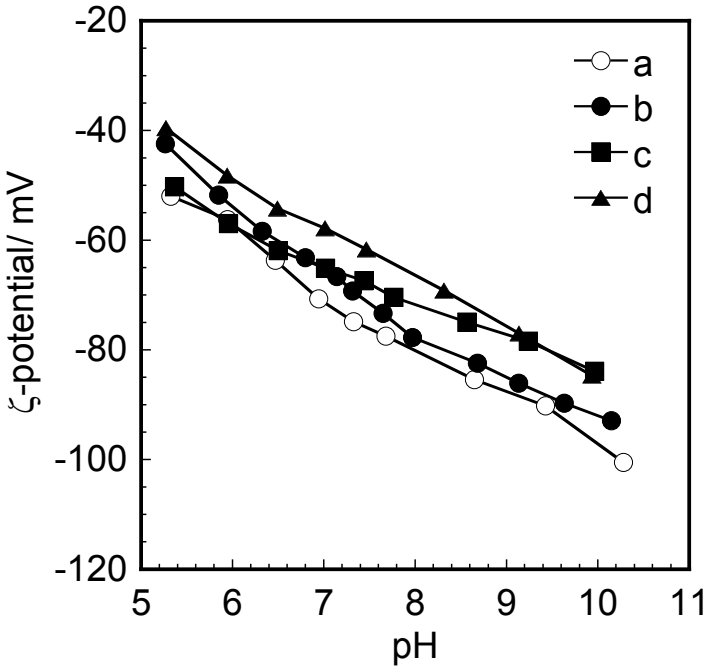

Fig. S5. ζ-potentials of (a) the bare PEI surface, the peptide-immobilized PEI surfaces with a peptide amount of (b) 200, (c) 400, and (d) 500 pmol cm^{-2}.

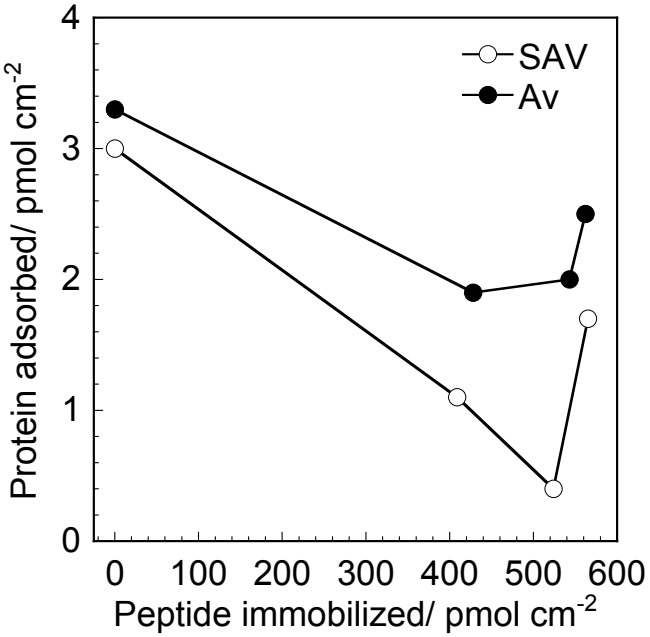

Fig. S6. Profiles of the adsorption amounts of SAv and Av on p1-EK-immobilized PEI surfaces against the amount of peptide immobilized peptides.