Supporting Information for

A mobile precursor determines protein resistance on nanostructured surfaces

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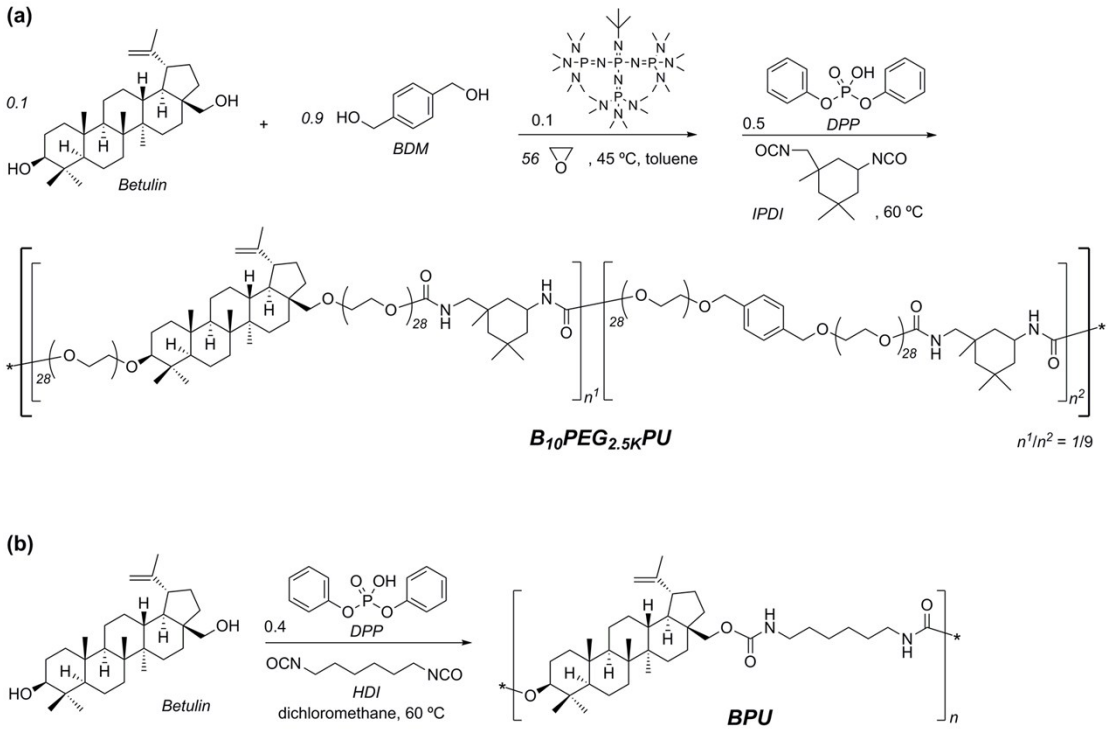


Scheme S1. Synthesis of (a) multi-segmented amphiphilic P(B-PEG-U) containing 10 mol% of the hydrophilic poly(ethylene glycol) (PEG) block, a hydrophobic betulinyl entity (B block) and urethane (U) segments in the center and (b) hydrophobic B-modified PU polymers.

Synthesis of multi-segmented amphiphilic P(B-PEG-U). Betulin (98%), 1,4-benzenedimethanol (BDM; 99%), isophorone diisocyanate (IPDI; 99%) and hexamethylene diisocyanate (HDI; 99%) were purchased from Aladdin. Betulin and BDM were dried in vacuum at 60 °C overnight and purified by azeotropic distillation of tetrahydrofuran (THF) prior to use. Diphenyl phosphate

(DPP; 99%), *t*-BuP_4 (0.8 M in *n*-hexane) and ethylene glycol (EG; 99.5%) were purchased from Aldrich. DPP was dried by azeotropic distillation of toluene, and then dissolved in purified toluene to prepare a 0.5 M solution. EG was condensed from a metal cylinder into a Schlenk flask and dried by stirring with sodium hydride in an ice-water bath for 4 h before cryo-distilled.

BDM (0.283 g, 2.05 mmol) and betulin (0.101 g, 0.23 mmol) was added in a Schlenk flask and dissolved in purified THF. After cryo-evaporation of THF the initiators were dried on the vacuum line with constant pumping for 1 h. Then purified toluene (20.0 mL) was condensed into the flask followed by addition of *t*-BuP_4 solution (0.28 mL, 0.05 equiv. with respect to hydroxyl) in an argon flow. Upon cooling at −20 °C, purified EG (5.7 mL, 103 mmol) was cryo-condensed in the flask, which was then immerged in a water bath and slowly heated to 45 °C. The suspended betulin powder dissolved in 30 min indicating the occurrence of ring-opening polymerization of EG from it. After 48 h, a small aliquot was withdrawn, quenched with acetic acid and used for SEC analysis. DPP solution (2.0 mL, 0.25 equiv. with respect to hydroxyl) was added to the reaction mixture in an argon flow to effect catalyst switch, after which IPDI (0.50 mL, 2.14 mmol) was injected in and temperature was raised to 60 °C. The step-growth polymerization was quenched after 3 days and the product was precipitated in a mixture of methanol and diethyl ether (1/1, v/v). $B_{10}PEG_{2.5K}$ precursor: $M_{n,SEC}$ = 11.3 kg mol^{-1}, $Đ_M$ = 1.04. P(B_{10}-$PEG_{2.5K}$-U): $M_{n,SEC}$ = 55.2 kg mol^{-1}, $Đ_M$ = 1.67; ^{1}H NMR (600 MHz, $CDCl_3$), δ/ppm = 7.25-7.19 (aromatic protons), 4.60-4.47 (betulin, =CH_2), 4.55-4.45 (BDM, −OCH_2C$_6$H$_4$CH_2O−), 4.30-4.08 (PEG, −CH$_2$CH_2OCONH−), 3.80-3.35 (PEG, −OCH_2 and IPDI, −C*H*NHCO−), 3.10-3.04 (betulin, −CH_2O−), 2.87-2.80 (IPDI, −CH_2NHCOO−), and 2.75-2.60 (betulin, −C*H*O− and IPDI, −CH_2NHCOO−).

Synthesis of hydrophobic B-modified polyurethane (PU). Betulin (1.00 g, 2.26 mmol) was added in a reaction flask, purified by azeotropic distillation of THF and dried at 60 °C under vacuum for 1 h. Dried dichloromethane (20 mL) was then cryo-condensed in the flask, followed by addition of HDI (0.36 mL, 2.26 mmol) and DPP solution (1.8 mL, 0.2 equiv. with respect to

hydroxyl). The flask was heated at 40 °C and soon after which the suspended betulin powder dissolved. The reaction was quenched after 3 days by addition of excess triethylamine and the product was precipitated in methanol. $M_{n,SEC}$ = 16.9 kg mol^{-1}, $Đ_M$ = 1.34. ^{1}H NMR (600 MHz, $CDCl_3$), δ/ppm = 4.60-4.47 (betulin, =CH_2), 4.39-4.30 and 3.9-3.78 (betulin, −CH_2OCONH−), 4.30-4.21 (betulin, −C*H*OCONH−) and 3.23-3.08 (HDI, −OCONHCH_2CH$_2$CH$_2$−).

Size exclusion chromatography (SEC) and NMR spectroscopy. SEC coupled with RI and UV detectors was conducted in THF at 35 °C using two identical PLgel columns (5 μm, MIXED-C) at a flow rate of 1.0 mL min^{-1}. Calibration was done with a series of narrowly dispersed polystyrene standards to obtain apparent number-average molar mass ($M_{n,SEC}$) and molar mass distribution ($Đ_M$) of the polymers. NMR spectra were recorded at room temperature on a Bruker AV600 NMR spectrometer using $CDCl_3$ as solvent and tetramethylsilane as the internal standard.

Table S1. Physiochemical parameters of fibrinogen (FBN), bovine serum albumin (BSA) and myoglobin (MYO) in the solution.[1-2] M_w: molecular weight of the protein. Size: approximate molecular dimensions of the protein in the solution. R_h: hydrodynamic radius of the protein in the solution. D: diffusion coefficients of the protein in the solution. pI: isoelectric point of the protein.

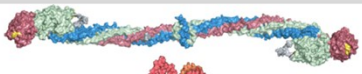
Protein	Fill model	M_w (kDa)	Size (nm^3)	R_h (nm)	D (μm^2/s)	pI
FBN		340	47 x 5 x 5	8.83	24.3	5.5
BSA		67	7 x 7 x 5	3.51	61.1	4.8
MYO		17	4 x 4 x 3	2.05	104.7	6.8

Table S2. Mean surface residence times fitted from the cumulative residence time distribution for fibrinogen (FBN), bovine serum albumin (BSA), and myoglobin (MYO).

Surface homo-PS coverage (%)	FBN			BSA			MYO		
	τ_1	τ_2	τ_3	τ_1	τ_2	τ_3	τ_1	τ_2	τ_3
0	0.20 ± 0.05	N/A	N/A	0.28 ± 0.10	N/A	N/A	0.25 ± 0.03	N/A	N/A
2	0.14 ± 0.02	0.50 ± 0.05	N/A	0.43 ± 0.10	N/A	N/A	0.12 ± 0.05	0.50 ± 0.10	N/A
7	0.13 ± 0.07	0.55 ± 0.02	N/A	0.21 ± 0.06	0.83 ± 0.10	N/A	0.16 ± 0.04	0.52 ± 0.10	2.40 ± 0.50
11	0.30 ± 0.10	1.25 ± 0.05	N/A	0.15 ± 0.03	0.60 ± 0.05	2.40 ± 0.50	0.20 ± 0.05	0.80 ± 0.20	3.70 ± 0.50
19	0.20 ± 0.05	0.80 ± 0.05	3.80 ± 0.12	0.18 ± 0.05	0.56 ± 0.10	2.30 ± 0.55	0.13 ± 0.01	0.70 ± 0.10	2.12 ± 0.13
100	0.25 ± 0.08	0.95 ± 0.02	4.40 ± 0.09	0.15 ± 0.05	0.50 ± 0.08	2.10 ± 0.10	0.15 ± 0.01	0.62 ± 0.03	2.89 ± 0.20

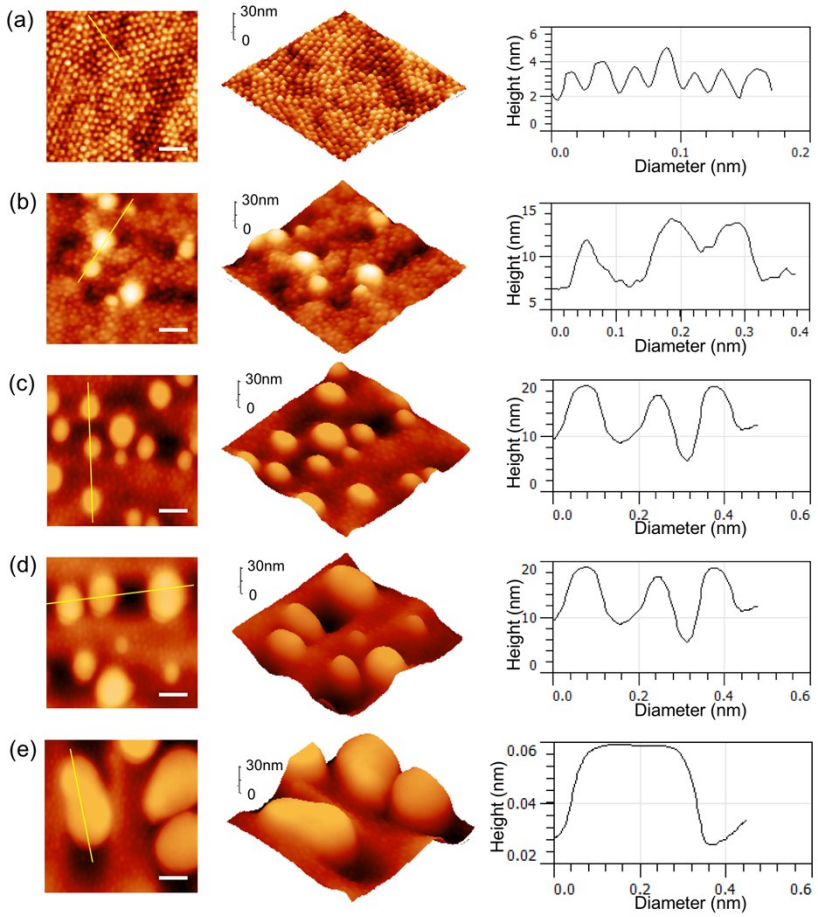


Figure S1. The cross-section analysis of AFM images taken under water for thin films of (a) PS_{60}-*b*-$PHEMA_{150}$ and (b–e) PS_{60}-*b*-$PHEMA_{150}$/homo-PS mixture thin films with different contents of PS homopolymer (homo-PS): (b) 1 wt%, (c) 5 wt%, (d) 10 wt%, and (e) 20 wt%. The 3D AFM images for each case are also attached.

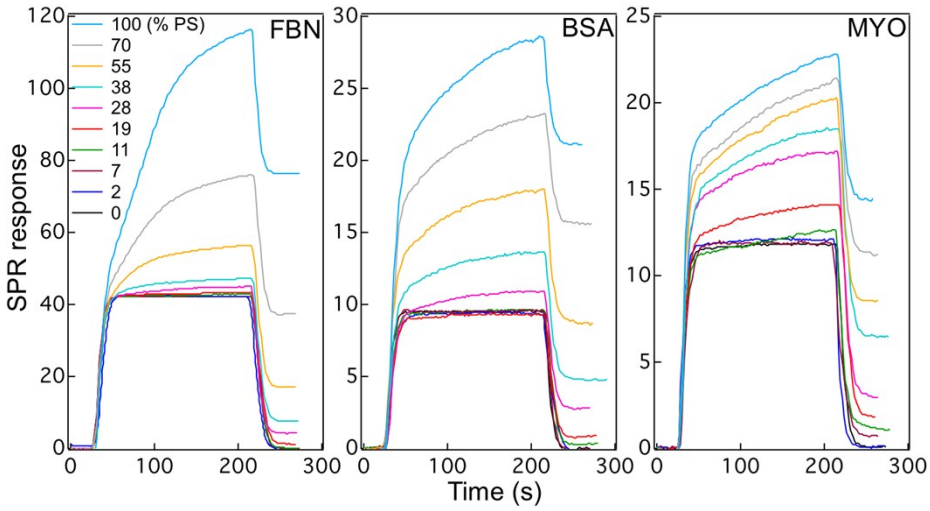


Figure S2. The SPR responses for fibrinogen (FBN), bovine serum albumin (BSA), and myoglobin (MYO) (50 μg/mL) adsorption on PS_{60}-*b*-$PHEMA_{150}$/PS nanostructured surfaces with various surface ratio of PS homopolymer (%).

References

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[2] A. S. Brill, J. S. Olin, B. M. Siegel, *J. Cell Biol.* **1962**, *13*, 249.