Electronic Supplementary Material (ESI) for ChemComm.

Superwetting comonomers reduce bacterial adhesion

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Supporting information

Experimental Section

Materials

Ethyl 4-(dimethylamino)benzoate (EDB), acryloyl chloride, butyl methacrylate (BMA), camphorquinone (CQ), diethyl ether, diethylene glycol diacrylate (DEGDA), hydroquinone monomethyl ether (MEHQ) inhibitor remover, methyl methacrylate (MMA), poly(ethylene glycol) acrylate (Mn ≈ 375) (hPEG), poly(ethylene glycol) monooleate (Mn ≈ 860), and triethylamine were purchased from Sigma Aldrich. EDB, acryloyl chloride, CQ, diethyl ether, DEGDA, MEHQ inhibitor remover, and triethylamine were used as received. MMA, BMA, PEG, poly(ethylene glycol) monooleate had the radical inhibitors removed by passage through a column packed with MEHQ inhibitor remover and stored at 2 °C until used. Silmer ACR A008-UP (ACR) was a gift from Siltech Corporation and was used as received. Photopolymerization was initiated by a blue light source, Kerber Applied Research BlueCure 25, which was graciously provided by Kerber Applied Research Inc.

Methods

Shore Hardness Measurements

Shore hardness measurements were taken using a Type OO Model 1600 Rex® Durometer purchased from Rex Gauge Company, Inc. Three small

discs were punched out from the main polymer body and stacked, before the hardness reading was obtained. By stacking them, this prevents the durometer measured only the polymer and not the metal substrate beneath.

Wettability Measurements

Water contact angles measurements were obtained through manual measurements of digital images depicting the water droplets on the surface of the polymers. The images were obtained through the use of a Krüss Contact Angle Measuring Instrument G10 and the manual measurements were obtained through the use of an angling tool function in GIMP 2.6.8, a GNU image manipulation program. While monitoring the surface using the Krüss instrument, a 3 μL droplet of Milli-Q water was placed onto of the surface of the polymer being examined. A digital image of the water droplet on the surface is captured, and by using the angling tool provided by GIMP, a contact angle was determined by averaging the left and right angles of the droplet.

Soxhlet Extraction

A conventional Soxhlet extractor was used to extract the unreacted material and low molecular weight oligomers from the matrix of the copolymers. The extraction solvent used was 2-propanol and the extraction process ran overnight at 90 °C following a procedure described by de Castro and García-Ayuso.[1]

Surface Analysis

The topographical features of the polymers were obtained using a Tescan Vega II LSU scanning electron microscope (Tescan USA, Pennsylvania, United States) operating at 10 kV. In order to optimize imaging of the pattern, the stage was slightly tilted approximately 28°.

Chemical Structure Analysis

^{1}H NMR data was obtained using a Bruker AVANCE 200 MHz nuclear magnetic resonance spectrometer (Bruker Corp., Milton, Canada); samples were measured in deuterated chloroform.

Mass Determination

The mass spectrum of the oPEG monomer was obtained using a Waters/Micromass Global Q-TOF (Quadrupole-Time of Flight) mass spectrometer. The sample was run in ESI(+ve) mode at 6000 mass resolution.

Bacterial Adhesion Studies

LB agar plates were created using 10 g of tryptone, 5 g of yeast extract, 10 g of NaCl, 15 g of agar and 1 L of distilled water (dH_2O). The dry ingredients were measured into a 2 L Erlenmeyer flask followed by 500 mL of dH_2O and the mixture was stirred to achieve complete solvation of the starting materials. The agar was added along with the rest of the H_2O before the solution was autoclaved. Following autoclaving, approximately 12 mL of the media were transferred into a dish in a laminar flow hood and the process was continued until all the media had been utilized. The media in the dishes was left to solidify for 30 min, after which the capped plates were stacked in their original packaging, sealed and stored at 4 °C until further use. LB media for culturing E. coli in solution was made in the same manner with the exception of agar. The autoclaved solution was sealed and stored at room temperature. The protocol for the adhesion assay was based on published results.[2, 3] Exactly 100 µL of *E. coli* culture broth were streaked on an agar plate that was incubated overnight. Multiple colonies (3-4) were obtained from the resultant lawn using an autoclaved

pipette tip and a new vial of broth (200 mL) was inoculated. This vial was placed in an incubator from where 1 mL aliquots were taken every 30 min to measure the OD_{600} of the solution. Once the OD_{600} value reached 0.7, 0.5-1 mM, IPTG was added to the vial, which was incubated for 5-6 h. E. coli from the vial was filtered using a cellulose acetate filter 0.45 microns (37 mm diameter) and the filter paper was washed thrice with autoclaved 0.9% PBS into new vial. 100 mL of PBS were added to the vial, which was supplemented with 2% w/v nutrient broth. The solution was agitated to facilitate equal dispersion of E. coli. Copolymer coupons (n = 4 for each type) were placed in a 48- well polystyrene, flat- bottom plate and to each polymer-containing well, 400 µL of the broth-supplemented E. coli mixture were added. The plate was incubated overnight (12 h), after which each coupon was removed from its well using sterile forceps, rinsed thrice with autoclaved PBS and placed in a well of a fresh plate. A microplate reader (Gemini XPS) was used to obtain GFP fluorescence readings using an excitation and emission wavelength of 395 nm and 509 nm, respectively, from the rinsed polymer coupons in the new plate. The procedure was repeated for hydrated coupons (coupons that had been soaked in dH_2O for 30 min prior to incubation with 400 µL of broth-supplemented E. coli in PBS). The fluorescence readings for each set were plotted for comparison after the background fluorescence (reading from a sample of each type incubated with uninduced E. coli) had been subtracted. Readings for the dry set were also plotted against the average percent water uptake and the sessile drop contact angles to determine the correlation, if any, between the three different variables.

Synthesis of oPEG

To a stirring and sealed 500 mL round-bottomed flask, under nitrogen, was added poly(ethylene glycol) monooleate (9.04 g, 0.011 mol, 1.0 eq, Mn ≈ 860) and dry diethyl ether (250 mL). Once the mixture was homogenized, triethylamine (7.33 mL, 0.053 mol, 5.0 eq) was slowly introduced to the reaction. Then, while stirring vigorously, acryloyl chloride (1.70 mL, 0.021 mol, 2.0 eq) was slowly introduced dropwise to the reaction mixture. A white precipitate formed instantaneously when acryloyl chloride was added to the mixture. After stirring overnight, solvents were removed using evaporation under reduced pressure until a thick, viscous slushy residue remained. The residue was diluted with diethyl ether and filtered through a pad of Celite using vacuum filtration to collect the product. The process was repeated 3 times. The ether extracts were dried over magnesium sulfate and, after filter, the solvents were removed obtain the purified oPEG monomer (9.162 g, 91.62%).

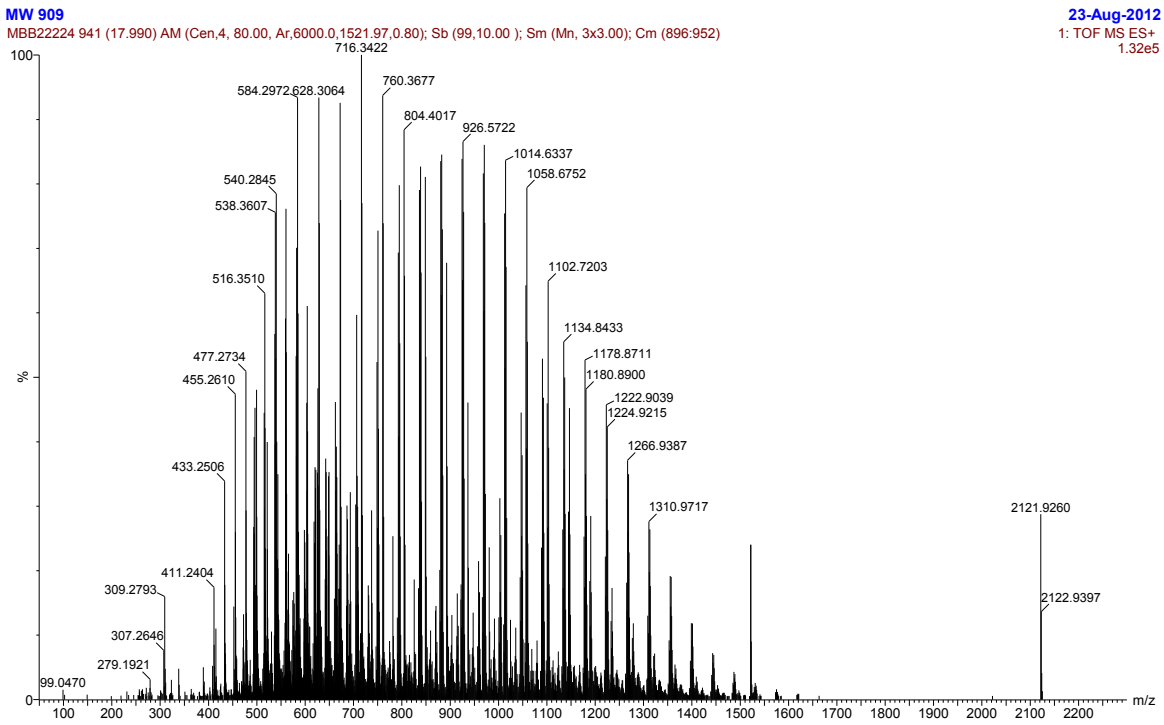

Figure 1: Mass spectrum of oPEG.

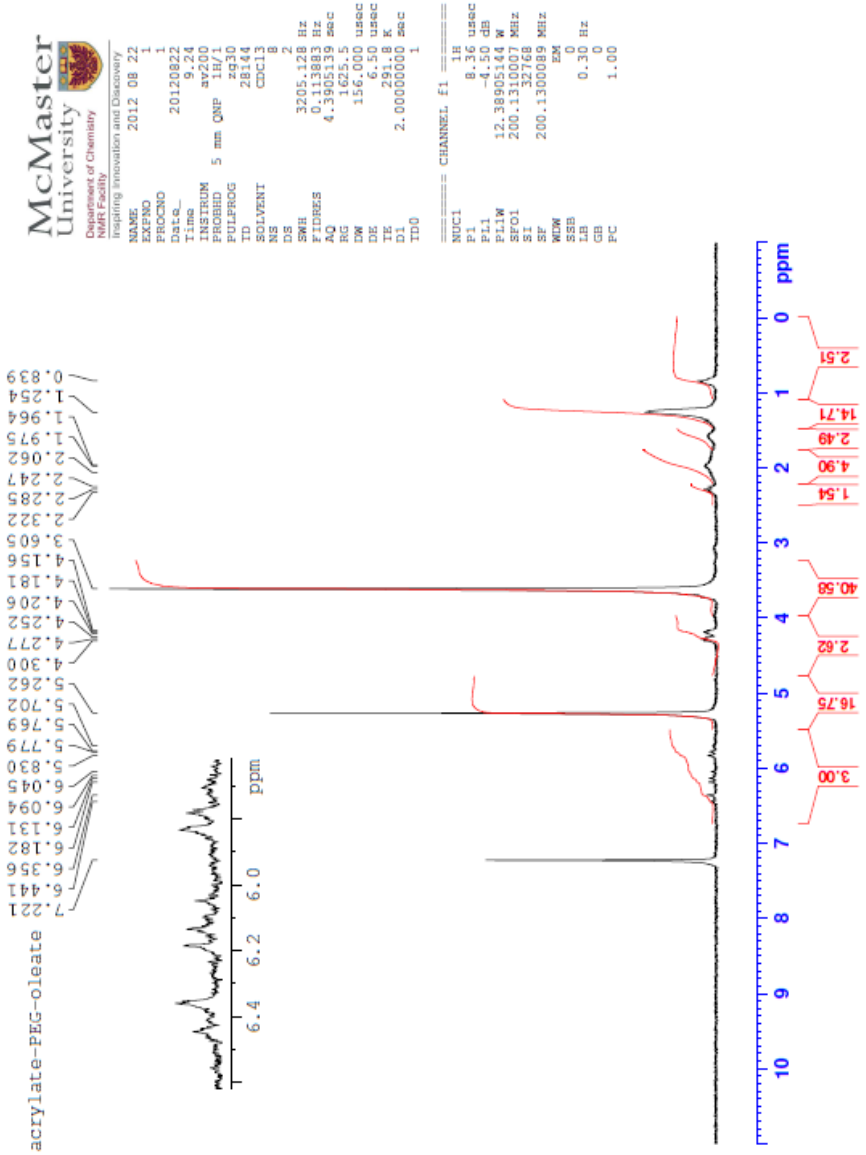

Figure 2: The NMR spectrum of the oPEG monomer.

Polymer Synthesis

As the syntheses of the various polymers are similar, differing only by the natures of the monomers ACR, hPEG, or oPEG, and quantities added (Table 1), a general procedure will be described. All polymers synthesized were formed using a total of 2 g of monomers, contained 1 wt% CQ and 1 wt% EDB as the photoinitiating system, 1 wt% DEGDA as the crosslinker, and all monomers in their respective weight percent ratios.

Synthesis of ACR-MMA-BMA Polymers

CQ (0.02 g, 1 wt%) and EDB (0.02 g, 1 wt%) were weighed into a 10 mL glass test tube. Uninhibited MMA and BMA were added to the test tube followed by the addition of DEGDA (0.02 g, 1 wt%). The reaction mixture was stirred gently to facilitate the dissolution of the solid reagents to give a homogeneous solution. ACR was then added. After the mixture was thoroughly mixed, it was golden yellow in color. The reaction mixture was deoxygenated by bubbling nitrogen gas, through a glass pipette into the solution for 30 s, and then poured into a small Teflon-lined plastic Petri dish and irradiated for 1 h. Solutions with greater percentages of ACR were found to cure more slowly. The solid elastomer was then removed from the Teflon-lined Petri dish and soaked in 2-propanol (40 mL) overnight. The elastomer was removed and dried in a vacuum oven (50 °C, 500 mm Hg) overnight to afford the final product.

Through NMR studies, both the oligomers (from the extracted material) and the polymers contained monomers whose molar ratios reflected the molar ratios of the monomers in the starting material (Table 2, Table 3).

Table 1: Formulation for ACR-MMA-BMA Polymers[1]

CQ (g)	EDB (g)	ACR (g)	MMA (g)	MMA (uL)	BMA (g)	BMA (uL)	DEGDA (g)	DEGDA (µL)
0.02	0.02	0.8	1.2	1282.1	0	0.0	0.02	13.7
0.02	0.02	0.9	1.1	1175.2	0	0.0	0.02	13.7
0.02	0.02	1	1	1068.4	0	0.0	0.02	13.7
0.02	0.02	1.1	0.9	961.5	0	0.0	0.02	13.7
0.02	0.02	1.2	0.8	854.7	0	0.0	0.02	13.7
0.02	0.02	1.3	0.7	747.9	0	0.0	0.02	13.7
0.02	0.02	1.4	0.6	641.0	0	0.0	0.02	13.7
0.02	0.02	1.5	0.5	534.2	0	0.0	0.02	13.7
0.02	0.02	1.6	0.4	427.4	0	0.0	0.02	13.7
0.02	0.02	1.7	0.3	320.5	0	0.0	0.02	13.7
0.02	0.02	1.8	0.2	213.7	0	0.0	0.02	13.7
0.02	0.02	1.9	0.1	106.8	0	0.0	0.02	13.7
0.02	0.02	0.8	0	0.0	1.2	1345.3	0.02	13.7
0.02	0.02	0.9	0	0.0	1.1	1233.2	0.02	13.7
0.02	0.02	1	0	0.0	1	1121.1	0.02	13.7
0.02	0.02	1.1	0	0.0	0.9	1009.0	0.02	13.7
0.02	0.02	1.2	0	0.0	0.8	896.9	0.02	13.7
0.02	0.02	1.3	0	0.0	0.7	784.8	0.02	13.7
0.02	0.02	1.4	0	0.0	0.6	672.6	0.02	13.7
0.02	0.02	1.5	0	0.0	0.5	560.5	0.02	13.7
0.02	0.02	1.6	0	0.0	0.4	448.4	0.02	13.7
0.02	0.02	1.7	0	0.0	0.3	336.3	0.02	13.7
0.02	0.02	1.8	0	0.0	0.2	224.2	0.02	13.7
0.02	0.02	1.9	0	0.0	0.1	112.1	0.02	13.7
0.02	0.02	0.8	0.6	641.0	0.6	672.6	0.02	13.7
0.02	0.02	0.9	0.55	587.6	0.55	616.6	0.02	13.7
0.02	0.02	1	0.5	534.2	0.5	560.5	0.02	13.7
0.02	0.02	1.1	0.45	480.8	0.45	504.5	0.02	13.7
0.02	0.02	1.2	0.4	427.4	0.4	448.4	0.02	13.7
0.02	0.02	1.3	0.35	373.9	0.35	392.4	0.02	13.7
0.02	0.02	1.4	0.3	320.5	0.3	336.3	0.02	13.7
0.02	0.02	1.5	0.25	267.1	0.25	280.3	0.02	13.7
0.02	0.02	1.6	0.2	213.7	0.2	224.2	0.02	13.7
0.02	0.02	1.7	0.15	160.3	0.15	168.2	0.02	13.7
0.02	0.02	1.8	0.1	106.8	0.1	112.1	0.02	13.7
0.02	0.02	1.9	0.05	53.4	0.05	56.1	0.02	13.7

[1] Note: the 1wt% DEGDA used in the formulation is not included in this Table.

Table 2: Ratio of Monomers Incorporated into Oligomers of the Extracted Material[2]

Weight Ratio of Monomers		**Theoretical Ratio of Monomers Incorporated into Polymer**		**Measured Ratio of Monomers Incorporated into Oligomer**		**Relative Integrations**	
%wt ACR	%wt BMA	ACR	BMA	ACR	BMA	ACR	BMA
60	40	1.00	3.63	1.00	1.88	51.80	18.87
80	20	1.00	1.36	1.00	2.27	64.67	27.94

[2] To see the constitution of the crosslinked polymer, please see Table 3. Note: the 1wt% DEGDA used in the formulation is not included in this Table.

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Table 3: Ratio of Monomers Incorporated into Polymers[3]

Weight Ratio of Monomers			**Theoretical Ratio of Monomers Incorporated into Polymer**			**Measured Ratio of Monomers Incorporated into Polymer**		
%wt ACR	%wt MMA	%wt BMA	ACR	MMA	BMA	ACR	MMA	BMA
40	60	0	1.00	11.61	0.00	1.00	13.39	0.00
40	30	30	1.00	5.81	4.09	1.00	7.42	8.09
40	0	60	1.00	0.00	8.18	1.00	0.00	6.84
60	40	0	1.00	5.16	0.00	1.00	4.56	0.00
60	20	20	1.00	2.58	1.82	1.00	4.53	3.02
60	0	40	1.00	0.00	3.63	1.00	0.00	5.64
80	20	0	1.00	1.94	0.00	1.00	3.17	0.00
80	10	10	1.00	0.97	0.68	1.00	2.33	1.00
80	0	20	1.00	0.00	1.36	1.00	0.00	2.29

Continued from Above

Weight Ratio of Monomers			**Relative Integrations[4]**		
%wt ACR	%wt MMA	%wt BMA	ACR	MMA	BMA
40	60	0	21.00	40.18	-
40	30	30	21.00	22.25	32.34
40	0	60	21.00	-	27.36
60	40	0	21.00	13.67	-
60	20	20	21.00	9.05	18.10
60	0	40	21.00	-	22.54
80	20	0	21.00	9.52	-
80	10	10	21.00	7.49	3.83
80	0	20	21.00	-	9.14

[3] To see the constitution of the oligomers from extracted from select samples, please see Table 2. Note: the 1wt% DEGDA used in the formulation is not included in this Table.

Synthesis of hPEG-MMA-BMA Polymers

The synthesis of hPEG-MMA-BMA polymers were similar to the general procedure given above except that hPEG was used instead of ACR.

Table 4: Formulation for hPEG-MMA-BMA Polymers[5]

CQ (g)	EDB (g)	hPEG (g)	MMA (g)	MMA (uL)	BMA (g)	BMA (uL)	DEGDA (g)	DEGDA (µL)
0.02	0.02	1.6	0.4	427.4	0	0.0	0.02	13.7
0.02	0.02	1.2	0.8	854.7	0	0.0	0.02	13.7
0.02	0.02	0.8	1.2	1282.1	0	0.0	0.02	13.7
0.02	0.02	0.4	1.6	1709.4	0	0.0	0.02	13.7
0.02	0.02	0	2	2136.8	0	0.0	0.02	13.7
0.02	0.02	1.6	0	0.0	0.4	448.4	0.02	13.7
0.02	0.02	1.2	0	0.0	0.8	896.9	0.02	13.7
0.02	0.02	0.8	0	0.0	1.2	1345.3	0.02	13.7
0.02	0.02	0.4	0	0.0	1.6	1793.7	0.02	13.7
0.02	0.02	0	0	0.0	4	4484.3	0.02	13.7
0.02	0.02	1.6	0.2	213.7	0.2	224.2	0.02	13.7
0.02	0.02	1.2	0.4	427.4	0.4	448.4	0.02	13.7
0.02	0.02	0.8	0.6	641.0	0.6	672.6	0.02	13.7
0.02	0.02	0.4	0.8	854.7	0.8	896.9	0.02	13.7
0.02	0.02	0	1	1068.4	1	1121.1	0.02	13.7

[5] Note: the 1wt% DEGDA used in the formulation is not included in this Table.

Synthesis of oPEG-MMA-BMA Polymers

The synthesis of oPEG-MMA-BMA polymers were essentially identical as described in Section 5.3. The main difference was oPEG was used instead of ACR.

Table 5: Formulation for oPEG-MMA-BMA Polymers[6]

CQ (g)	EDB (g)	oPEG (g)	MMA (g)	MMA (uL)	BMA (g)	BMA (uL)	DEGDA (g)	DEGDA (µL)
0.02	0.02	2	0	0.0	0	0.0	0.02	13.7
0.02	0.02	1.2	0.8	854.7	0	0.0	0.02	13.7
0.02	0.02	0.6	1.4	1495.7	0	0.0	0.02	13.7
0.02	0.02	0	2	2136.8	0	0.0	0.02	13.7
0.02	0.02	2	0	0.0	0	0.0	0.02	13.7
0.02	0.02	1.2	0	0.0	0.8	896.9	0.02	13.7
0.02	0.02	0.6	0	0.0	1.4	1569.5	0.02	13.7
0.02	0.02	0	0	0.0	2	2242.2	0.02	13.7
0.02	0.02	2	0	0.0	0	0.0	0.02	13.7
0.02	0.02	1.2	0.4	427.4	0.4	448.4	0.02	13.7
0.02	0.02	0.6	0.7	747.9	0.7	784.8	0.02	13.7
0.02	0.02	0	1	1068.4	1	1121.1	0.02	13.7

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[6] Note: the 1wt% DEGDA used in the formulation is not included in this Table.