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Supporting Information

Dynamic Fluoroalkyl Polyethylene Glycol Co-Polymers: A New Strategy for Reducing Protein Adhesion in Lab-on-a-Chip Devices

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Supplementary Information for

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NMR Characterization

^{1}H NMR spectra of FPEG-FA were recorded in CDCl$_3$ solution on a Bruker Avance 400 MHz NMR spectrometer. The copolymer composition (Table S1) was calculated from the integrated areas of the ^{1}H NMR signals at 4.5 ppm and 4.0 ppm attributed to the –CH$_2$COO– groups of the FPEG and FA units, respectively.

Automated DMF Device Longevity Analysis

DMF device longevity was evaluated using a test similar to those described in the main text, but using the DropBot automated manipulation system (described in detail in Fobel et al., *Appl. Phys. Lett.* 2013, 102, 193513). The sample tested was RPMI 1640 cell culture medium supplemented with 10% fetal bovine serum and 0.02 % Pluoronic P105. A program was implemented to move unit droplets on device in a circular pattern across six actuation electrodes continuously until failure, applying 10 s voltage steps (at 110 V$_{rms}$) to each electrode. The capacitance was measured repeatedly during each step as a proxy of position to generate instantaneous velocities. Device failure was defined as the first case in which the capacitance measured at the destination electrode after 10 s failed to be greater than 30% of the first capacitance measured at the origin electrode. At least 3 droplets were evaluated on at least 2 different devices formed with each coating. Device lifetime data are shown in Figure S1.

Protein-Fouling Studies

FITC-BSA in PBS (1 mg/ml) with or without pluronic (F68, 0.05% wt/v) was used to evaluate active or passive protein fouling on DMF device-two (see main text) bottom plates. For passive fouling, 15 μL aliquots of FITC-BSA were pipetted onto the substrates, which were

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incubated in a humidified chamber (i.e., a Petri dish partially filled with water) for 1 or 10 h. For active fouling, a top/bottom plate device was assembled and a stationary unit droplet of FITC-BSA was potentiated for 1 or 5 min (110 V_{rms}, 10 KHz; when needed, additional solution was added to compensate for evaporation), after which the device was disassembled. After active or passive fouling, the FTIC-BSA solutions were wicked away with a Kimwipe and non-adsorbed protein was washed away by immersing and gently agitating the substrates first in PBS and then in DI water for 5 min each. The substrates were air dried and stored in the dark until analysis.

Florescence images were collected using a Typhoon Trio variable mode imager (excitation 488 nm; emission 526 nm; PMT driving potential -750 V) from GE Healthcare Biosciences (Piscataway, NJ). ImageJ was used to record the densiometric mean value of fluorescence from the center of the fouled spot in each image. For each unique condition, 6 droplets were evaluated on 4 replicate substrates (passive) or 4 droplets on 3 replicate substrates (active). The data are shown in Figure S2.

Long-Term AFM Analysis

AFM measurements were collected as described in the main text, using activated device two bottom plates coated with 5FPEG-FA. After potentiation and drying, samples were incubated at room temperature for 1h or 30 h prior to analysis. The data are shown in Figure S3.
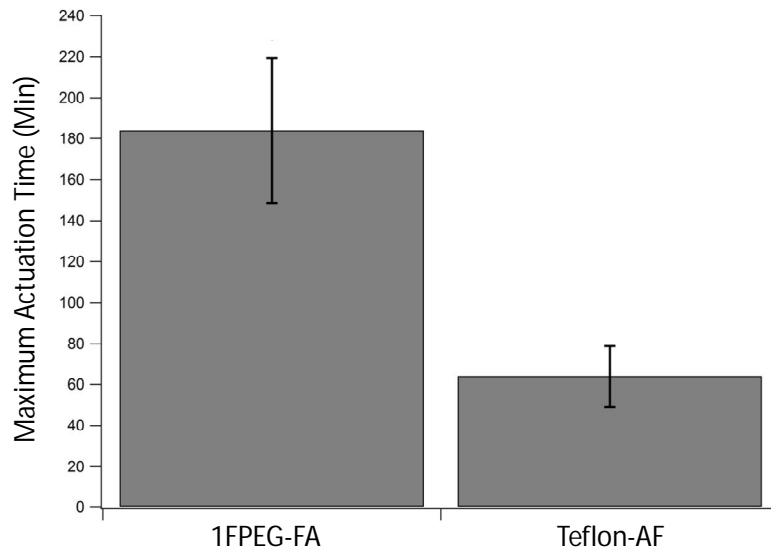
Long-Term Contact Angle Studies

Contact angles for water and hexadecane were measured as described in the main text. The substrates were DMF device-two bottom plates (with no top plate), and were immersed in DI water for 0, 24, 48, or 72 h. After immersion, the excess water was wicked away with a

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Kimwipe. Data are shown in Figure S4, and were recorded from 3 devices (6 readings each) for both of the interrogating liquids.
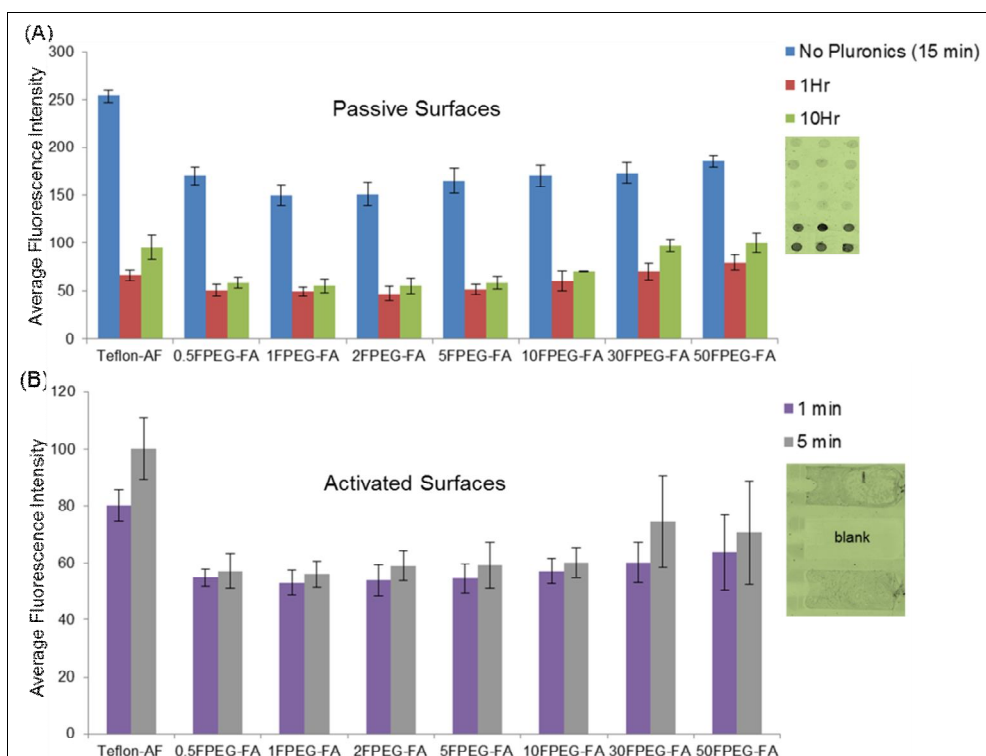
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Figure S1: Automated device longevity assay. Plots of maximum actuation time for continuous actuation of droplets of cell culture medium supplemented with 10% serum and 0.02% P105 on devices coated with 1FPEG-FA (left) or Teflon-AF (right). Each condition was repeated three times on two different devices, and error bars represent ± 1 S.D. Instantaneous velocities on 1FPEG-FA coated devices were observed to remain consistent at ~1.2 mm/s throughout the test (until sudden failure), while instantaneous velocities on Teflon-AF coated devices were observed to begin at ~8 mm/s and to decrease steadily to < 1 mm/s (until gradual failure).
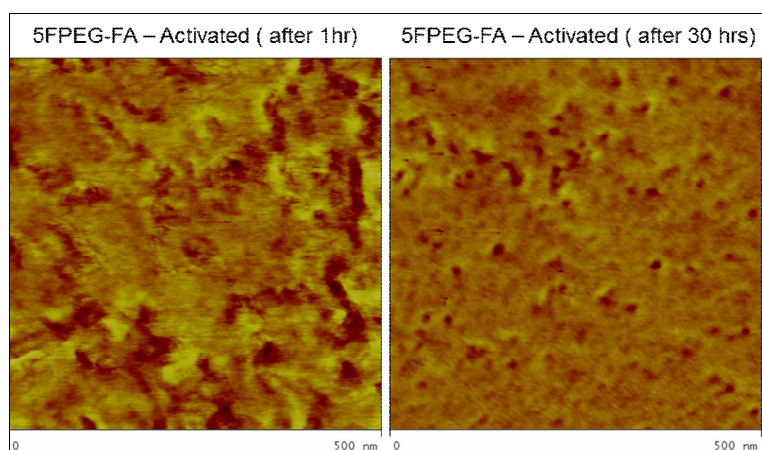
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Figure S2: Protein fouling. Fluorescent intensity from FITC-BSA on (A) passive surfaces (fouling with no potential applied) for 1 h (red) and 10 h (green) durations (and for comparison, no pluronics for 15 min, blue), and (B) activated surfaces (fouling with DMF driving potential applied) for 1 min (purple) and 5 min (grey) durations. Each condition was repeated six time were evaluated on four replicate substrates (passive) or four times on three replicate substrates (active), and error bars represent ± 1 S.D. Representative fluorescent images of the substrates shown in the insets. For active fouling, a blank electrode is shown in the centre with two fouled electrodes at the top and bottom.
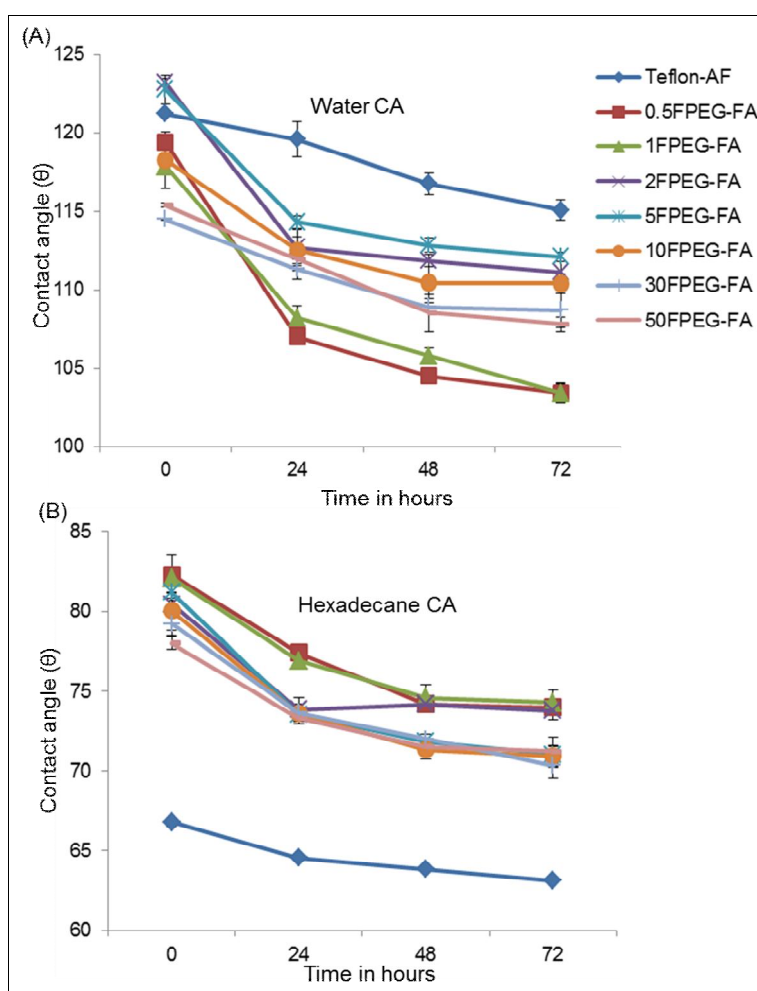
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Figure S3: Surface recovery. AFM phase images of an activated 5PEG-FA surface over time. Images generated 1 hr after activation (left) and 30 hr after activation (right) show evidence of gradual recovery of the activated surface, particularly when compared to images collected immediately after activation (e.g., Fig. 3C in the main text).

5FPEG-FA – Activated (after 1hr)   5FPEG-FA – Activated (after 30 hrs)

0   500 nm   0   500 nm

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Figure S4: Long-term surface wettability. Static contact angles measured for (A) water (CA_W) and (B) n-Hexadecane (CA_H) for devices after submersion in water for 0, 24, 48, or 72 h. Devices were coated with Teflon-AF (blue diamonds), 0.5FPEG-FA (red squares), 1FPEG-FA (green triangles), 2FPEG-FA (purple Xs), 5FPEG-FA (blue asterisks), 10FPEG-FA (orange circles), 30FPEG-FA (blue lines), and 50FPEG-FA (pink without markers). Each condition was repeated six times on three different devices, and error bars represent ± 1 S.D.

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Table S1: FPEG-FA copolymer composition. The composition was calculated from the integrating areas of the ^{1}H NMR signals originating from the –CH$_2$COO– groups of the FPEG and FA units are shown in the table.

Copolymer	Observed FPEG composition in % (by NMR)	Copolymer	Observed FPEG composition in % (by NMR)
50FPEG-FA	49.27	2FPEG-FA	2.24
30FPEG-FA	29.32	1FPEG-FA	1.14
10FPEG-FA	10.61	0.5FPEG-FA	0.54
5FPEG-FA	4.56		