Supporting information

Fabrication of the multilayer PDMS microfluidic loading device

The PDMS microfluidic device is composed of two layers. The upper layer was fabricated by casting the PDMS precursor and curing agent (5:1) against a silicon master. The silicon master is patterned using photolithography[21]. The bottom layer was a thin PDMS membrane containing micropores. The bottom layer was fabricated by spin-coating a thin layer of the PDMS precursor (monomer: curing agent = 20:1) onto the surface of a second silicon master. The silicon master contains arrays of microposts. The size of the microposts determines the size of the micropores of the PDMS membrane. Hundreds to thousands of micropores can be easily fit into one 1 cm × 1 cm PDMS membrane.

The bonding of the two PDMS layers was achieved by controlling the different curing rate between the upper and bottom PDMS layers.[22] After 30 min thermo annealing at 80 ℃, the upper layer was totally cured while the bottom layer was half-cured, and the alignment of the two layers can be performed. After another few hours of thermal annealing, the two layers were fully bound.

Contact printing method for microspots fabrication

A PDMS stamp with micropost array was fabricated by photolithography and soft lithography. The diameters of the microposts we tested were 1.00 mm, 500 μm, 200 μm, and 100 μm. The PDMS stamp was first immersed in the fresh dopamine solution (pH=8.5) for 4 hours, washed by distilled water and dried in air for 1 hour. The stamp was then attached onto the clean FEP substrate surface, with an applied force of about 15 G. The attachment between the stamp and the surface was maintained for 15 min, and the result was checked by microscopy.

The characterization of the polydopamine by AFM

To ensure the sample flatness, the substrate used for AFM characterization was cytop (Asahi Glass) coated silicon wafer instead of FEP membrane. Briefly, silicon wafer was first cleaned by piranha solution and dried by air; the wafer was then treated by air plasma for 2 min; finally the diluted cytop solution was spin-coated on the whole surface and baked on the 180℃ hotplate for 1 hour. Polydopamine was coated onto the surface by channel dispensing for 3 hours. The AFM instrument used was Multimode Scanning Probe Microscope (MMSPM) with the model number NanoSope IVa (Bruker).The AFM tip was Tap300E (BudgetSensors) with the force about 40 N/m.

Sequence of the peptides

Pep-2: DYKDDDDK
Pep-3: HYPNELLQEYNWELADQPQNLEEILMHCQT
Pep-N: MNGEEQYYAATQLYKDPCAFQRGPAPEFSA

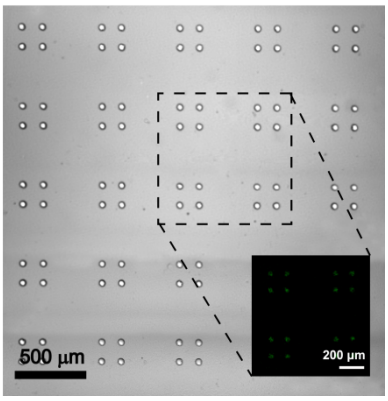

Figure S1 Bright field image of the microarray with water droplets dispensed on the microspots, which were 50 μm in diameter. The inset is the confocal image of the microspots coated with FITC labelled Pep-1.

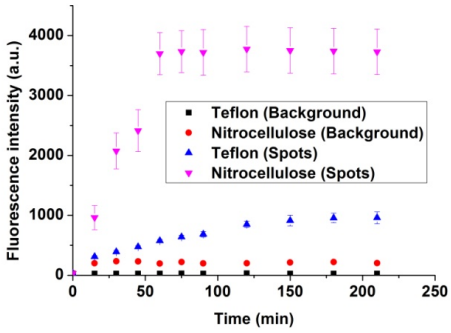

Figure S2 Fluorescence signal from IgG based microarray for the detection of 50 μg/ml FITC labelled anti-IgG based on the FEP substrate and the nitrocellulose substrate.

Table S1 Z-factor of the FEP substrate based protein microarray for cytokine detection

Concentration (pg/ml)	Z-factor		
	IL-1β	IL-6	IL-10
1.95	-∞	-1.55	-1.32
3.9	-∞	-1.55	-0.0896
7.8	-2.90	-1.55	0.0364
15.6	0.0164	-1.55	0.189
31.2	0.364	0.237	0.349
62.5	0.361	0.254	0.257
125	0.262	0.588	0.401
250	0.376	0.431	0.437
500	0.139	0.363	0.316
1000	0.610	0.290	0.452
2000	0.715	0.712	0.804
4000	-	0.996	-

Table S2 Z-factor of the FEP substrate based peptide microarray for antibody detection

Anti-Pep-2		Anti-Pep-3	
Concentration (ng/ml)	Z-factor	Concentration (ng/ml)	Z-factor
0.01	-0.426	0.156	-0.0138
0.02	-0.851	0.312	0.378
0.04	0.140	0.625	0.608
0.08	0.0655	1.25	0.905
0.1	0.463	2.5	0.621
0.2	0.589	5	0.632
0.4	0.464	8	0.951
0.8	0.539	10	0.934
1	0.475		
2	0.813		
4	0.850		
10	0.742		

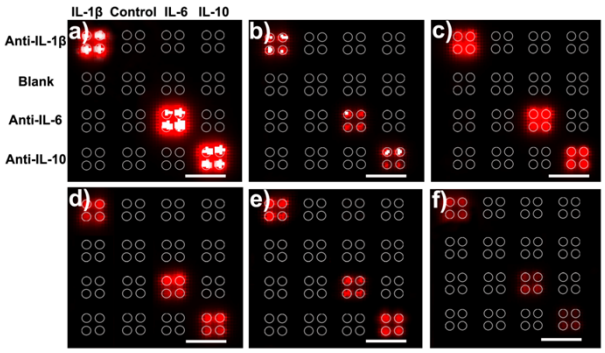

Figure S3 Multiplex protein microarray platform for the quantitative cytokine detection. The solutions were added as shown in (a). The concentrations of the target cytokines (IL-1b, IL-6, IL-10) were 2000 pg/ml, 500 pg/ml, 250 pg/ml, 125 pg/ml, 31.2 pg/ml, 20 pg/ml from (a) to (f), respectively.

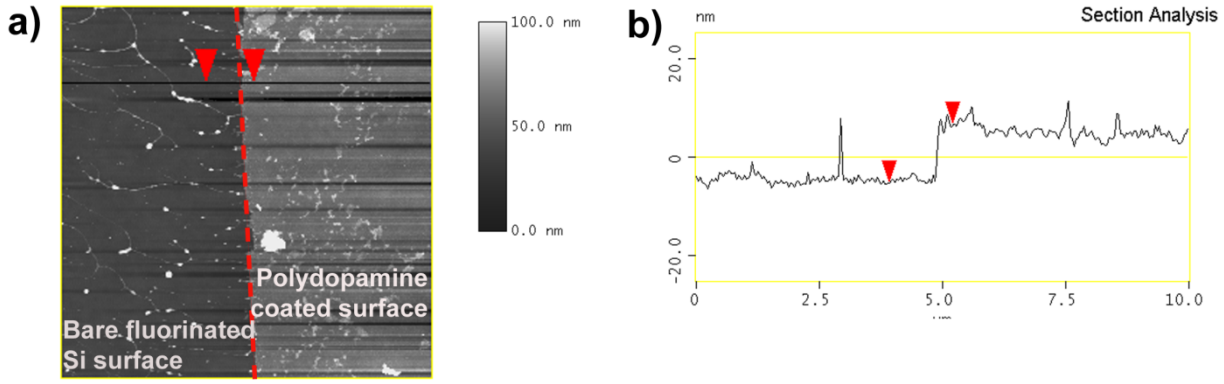

Figure S4 a) AFM image of the cytop coated Si surface. The red dash line indicates the borderline of the non-polydopamine deposited area (left) and polydopamine deposited area (right); b) The height profile for the AFM images in a).

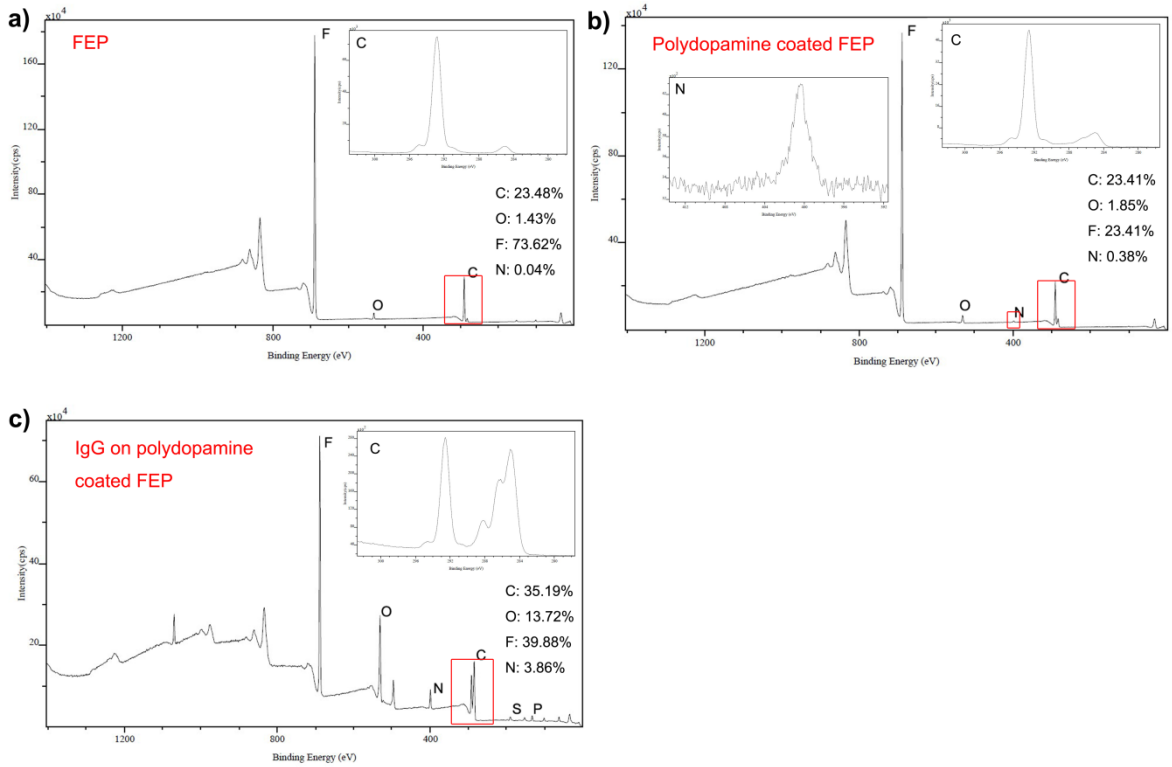

Figure S5 XPS results of (a) FEP (b) polydopamine coated FEP (c) IgG on polydopamine coated FEP. In (b), the amount of N increased, indicating the formation of polydopamine layer on the FEP surface. In (c), the amount of O, N, S, P increased, indicating the IgG being successfully immobilized onto the polydopamine surface.