Supporting Information

Blood Cell Repellence and Tumor Cell Capture Surface for High-Purity Enrichment of Circulating Tumor Cells

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Fig. S1 Schematic illustration of the step by step construction of cell membrane mimetic surface (CMMS) and FA and RGD ligands decorated CMMS (CMMS-FA-RGD) surface on a material-independent substrate.

Fig. S2 Chemical structure and $^1$H NMR spectrum of PMEN copolymer used in this work.
Fig. S3 High resolution N1s and P2p XPS spectra of the step by step coated and the bare glass surfaces.
Fig. S4 SPR response curves of each the coating procedures on-line. (a) PDA, (b) PDA/PEG, (c) PDA/PEG/PMEN (CMMS), (d) CMMS-FA, (e) CMMS-RGD and (f) CMMS-FA+RGD.
Table S1. Protein interaction parameters measured by SPR on FA ligand (15.2 ng/cm²) decorated CMMS.

<table>
<thead>
<tr>
<th>Protein (concentration)</th>
<th>FRα: 0.50 µg/mL (1.92×10⁻⁸ mol/L)</th>
<th>BSA: 1.00 mg/mL (1.47×10⁻⁵ mol/L)</th>
<th>Fg: 1.00 mg/mL (2.94×10⁻⁶ mol/L)</th>
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<tbody>
<tr>
<td>Adsorbed Amount (ng/cm²)</td>
<td>85.6±1.6</td>
<td>0.8±0.2</td>
<td>11.3±1.7</td>
</tr>
<tr>
<td>Binding Rate $k_a$ (M⁻¹ s⁻¹)</td>
<td>1.66×10⁵</td>
<td>100</td>
<td>350.1</td>
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<tr>
<td>Dissociation Rate $k_d$ (s⁻¹)</td>
<td>4.68×10⁻⁵</td>
<td>8.00×10⁻³</td>
<td>1.66×10⁻³</td>
</tr>
<tr>
<td>Equilibrium Constant $K_A$ (M⁻¹)</td>
<td>3.54×10⁹</td>
<td>1.25×10⁴</td>
<td>2.11×10⁵</td>
</tr>
<tr>
<td>Dissociation Constant $K_D$ (M)</td>
<td>2.82×10⁻¹⁰</td>
<td>8.00×10⁻⁵</td>
<td>4.75×10⁻⁶</td>
</tr>
</tbody>
</table>
Fig. S5 Cell density of L929, 3T3, MCF-7 and HeLa adhered for 20 min, 40 min and 60 min on differently decorated surfaces. (a) CMMS, (b) CMMS-FA, (c) CMMS-RGD, (e) CMMS-FA-RGD. The Bars represent means ± SD (n=6).

Fig. S6 (a) HeLa and (b) MCF-7 cell capture efficiency curves on differently decorated CMMS surfaces incubated for 20, 40, and 60 min, respectively. Significant synergistic effect of the ligands (23 ng/cm² FA and 11 ng/cm² RGD) on the CMMS-23FA-11RGD surface is observed for capturing both HeLa and MCF-7 cells when compared the measured capture efficiencies with those of calculated values (open circles linked by dash line).
Fig. S7 Fluorescence images of adhered HeLa cells from mixed cell suspensions (L929: HeLa = 100:1) onto CMMS-FA-RGD surface for (a) 20 min, (b) 40 min, (c) 60 min.
Fig. S8 Fluorescence microscopic images of HeLa and MCF-7 cells stained with PI and calcein AM. (A), (C) cultured cells; (B), (D) captured cells. (E) Quantitative viability analysis of the captured cells and cultured tumor cells.

Fig. S9 Optical microscopic images of adhered cells on different surfaces cultured for 40 min. All the scale bars are 30 μm. HeLa and MCF-7 cells show pseudopods on all the ligand decorated surfaces, suggesting enhanced interaction with the surfaces.
Figure S10. Microscopic images of HeLa cell capture on CMMS-FA-RGD surface from 9400 HeLa cells (stained green) spiked in 1.0 mL whole blood (WBC:HeLa = 1060:1) incubated for 60 min. After washing with PBS buffer solution under controlled conditions, blood cells could be removed completely and did not reduce the strongly captured tumor cells. Thus, the purity of captured tumor cells could be enhanced up to 90–100% on CMMS-FA-RGD surface.

Figure S11. Microscopic images of intentionally adhered whole blood cells (a) and selectively captured HeLa cells on CMMS-FA-RGD surface after rinsing with PBS (b). almost no detectable blood cells (WBC and RBC) adhered on the PBS rinsed CMMS-FA-RGD surface (b) by comparing with those adhered in image (a).