Slanted spiral microfluidics for the ultra-fast, label-free isolation of circulating tumor cells

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Supplementary information

Figure S1.

Phase contrast microscopy images of T24 cells at different treatment conditions. Morphology of cells remains relatively constant after lysis and processing using the spiral microfluidic chip. Viability of cells was confirmed using PI staining. The scale bar is 10 μm.
Phase contrast micrographs of cultures of control (unsorted) MDA-MB-231 cells and sorted cells by spiral chip. The images indicate no significant differences between the morphology and proliferation rate of the cells suggesting high viability and sterility. Scale bar is 100 μm.
Figure S3.

A library of CTC images displaying morphological variations of CTCs. The scale bar is 10 μm.
Table S1: Quantification of circulating tumor cells per mL of blood among 15 samples from healthy donors (n=5) and patients with advanced breast (n=5) and lung (n=5) cancer.

<table>
<thead>
<tr>
<th>Sample no</th>
<th>Subject initial</th>
<th>CTCs/mL</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Healthy</td>
<td>1</td>
</tr>
<tr>
<td>2</td>
<td>Healthy</td>
<td>2</td>
</tr>
<tr>
<td>3</td>
<td>Healthy</td>
<td>3</td>
</tr>
<tr>
<td>4</td>
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<tr>
<td>5</td>
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<td>2</td>
</tr>
<tr>
<td>1</td>
<td>Breast</td>
<td>57</td>
</tr>
<tr>
<td>2</td>
<td>Breast</td>
<td>33</td>
</tr>
<tr>
<td>3</td>
<td>Breast</td>
<td>43</td>
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<tr>
<td>5</td>
<td>Lung</td>
<td>7</td>
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</tbody>
</table>

Figure S4.

Flow cytometric analysis of isolated CTCs for cleaved caspase-3 protein. Only 9.9% of cells were positive for cleaved caspase-3, confirming that the high flow rates in the microfluidic chip do not...
affect cell viability and integrity.

**Figure S5.**

Viability of captured CTC by trapezoidal cross-section chip. Captured cells are plated onto 2D polylysine coated substrates and allowed to spread overnight. Clustering of platelets with CTCs can be observed *in vitro*. Viability of CTCs was confirmed using PI staining.

**SI Movie Legends**

**Movie S1**
High speed video (6400 fps) illustrating the complete (near 100%) capturing of MDA-MB-231 cells spiked in PBS buffer.

**Movie S2**
High speed video (6400 fps) illustrating the complete isolation of MDA-MB-231 cells from WBCs at the device outlet. Focused MDA-MB-231 cells (near the inner wall (top side)) are clearly distinguished from WBCs based on morphology and phase contrast. Few platelets are observed going into the CTC outlet. However, their presence do not interfere during counting using immunofluorescence staining or downstream molecular assay such as PCR.

**Movie S3**
High speed video (6400 fps) captured at the outlet of spiral biochip showing isolation of few CTCs from peripheral blood of a patient with advanced metastatic lung cancer. This movie clearly demonstrates the performance of our device for efficient enrichment of CTC from blood samples.