An Integrated Microfluidic Device Utilizing Dielectrophoresis and Multiplex Array PCR for Point-of-Care Detection of Pathogens

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Supporting Information

Chemicals and Materials. Bovine serum albumin (BSA) was purchased from Sigma-Aldrich. SsoFast EvaGreen Supermix (2X) was purchased from Bio-Rad Laboratories (Beijing, China). Mineral oil (DNase, RNase and Protease free), Dichlorodimethylsilane and Dimethoxydimethylsilane were purchased from TCI (Shanghai, China). Soda-lime glass plates coated with photoresist and chromium were ordered from Telic Company (Valencia, CA). ITO glass was purchased from CSG Company (Shenzhen, China). SU8 2005 was purchased from MicroChem Corp. (Newton, MA).
The PCR mixture for preloading the microwells was prepared by mixing 25 µL of 2× PCR master mixture (SsoFast EvaGreen SuperMix, Bio-rad), 5 µL of 10 mg/mL BSA (Sigma) solution, 15 µL of RNase free water and 2.5µL of each primer (0.5 µM) for different species of pathogens. All primers for PCR amplification were ordered from Sangon biotech (Shanghai, China). Primer sequences were listed in Table S1.

A set of clinical strains isolated from patients’ positive blood cultures from Department of Clinical Laboratory, Peking Union Medical College Hospital (PUMCH, Beijing, China) during January 1, 2013 to June 1, 2013 were used for evaluation of broad-range utility of the methods. The bacterial and fungal names and their serial numbers are:

1. *Escherichia coli* (E. coli): 13B00401, 13XB00121, 13B00357, 13B00431
11. *Candida krusei* (C. krusei): ATCC 6258
Table S1. Names of pathogens and sequences of primer pairs tested by the SlipChip.

<table>
<thead>
<tr>
<th>Name of Pathogen</th>
<th>Primer sequence</th>
<th>Target gene</th>
<th>Amplicon size (bp)</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>E. coli</em> (ATCC 8739)</td>
<td>GAA CAT CAA ACA TTA AA</td>
<td>LacZ&lt;sup&gt;1&lt;/sup&gt;</td>
<td>454</td>
</tr>
<tr>
<td></td>
<td>AAT CAG TCG AAG ATA</td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>E. coli O157:H7</em> (ATCC 35150)</td>
<td>CGG ACA TCC ATG TGA TAT GG</td>
<td>rfb O157&lt;sup&gt;2&lt;/sup&gt;</td>
<td>259</td>
</tr>
<tr>
<td></td>
<td>TTG CCT ATG TAC AGC TAA TCC</td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>P. aeruginosa</em> (ATCC 9027)</td>
<td>GTC AGT GTT ACC TAA</td>
<td>23S rRNA&lt;sup&gt;1&lt;/sup&gt;</td>
<td>377</td>
</tr>
<tr>
<td></td>
<td>GAA AGG ATC TTT GAA</td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>S. aureus</em> (ATCC 6538p)</td>
<td>GGA GGA AGG TGG GGA TGA CG</td>
<td>16s rRNA&lt;sup&gt;3&lt;/sup&gt;</td>
<td>241</td>
</tr>
<tr>
<td></td>
<td>ATG GTG TGA CGG GCG GTG TG</td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>S. mutans</em></td>
<td>AGA TAT GAT TGC AAC AAT TGA A</td>
<td>dlt&lt;sup&gt;4&lt;/sup&gt;</td>
<td>412</td>
</tr>
<tr>
<td></td>
<td>CGC ATG ATT GAT TTG ATA AG</td>
<td></td>
<td></td>
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<tr>
<td><em>C. tropicalis</em></td>
<td>CAA TCC TAC CGC CAG AGG TTA T TGG CCA CTA GCA AAA TAA GCG T</td>
<td>rRNA ITS&lt;sup&gt;5&lt;/sup&gt;</td>
<td>357</td>
</tr>
</tbody>
</table>

REFERENCE:

Figure S1. a) A photograph of the device with zoom-in view; the microchannels and microwells were loaded with dye solutions of different colors; b) Schematic illustration showed that the system was consisted of the top glass plate, bottom plate with 5 μm SU-8 coating and ITO microelectrodes; c) Interdigitated ITO microelectrodes before and after fabricating three capture grooves. The detailed geometry was shown in the zoom-in schematic drawing.
**Figure S2.** a) Schematic illustration of bacterial DEP capture by grooves with a ladder-like layout in five units; b) A fluorescence microphotograph of three grooves in unit 2 with captured RFP-tagged *E. coli*. Scale bar=100 μm; The average fluorescence intensity profile perpendicular to the flow direction was shown on the right; c) The capture ratio of five units with equal groove width of 53.3 μm; d) Capture ratio of the first unit with groove widths in the range of 20-40 μm.