Microfluidic Devices for Tracking Cell Lineage and Differentiation

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Motivation

Genetically identical cells within a given population can exhibit heritable genetic variation across several generations. When observing and studying these cells as an ensemble, we might be in a situation where the genetic dependencies are tight. This is particularly true in the case of cancer stem cells, which exhibit asymmetric cell division, giving rise to stem cell renewal or differentiation into particular subtypes based on cell generation.

Multichannel Device

Multichannel Device Design:

- CAD Design:
- Master Preparation:
  - Resin Application: P-20 3k rpm, 1k rpm/s, 30s, S1827 3k rpm, 1k rpm/s, 30s
  - Exposure: SU-8 contact aligner, hard contact mode, 2.6 exposure developer
  - Develop: MIF720 120s
- Bonding:
  - UV Bonding: UV Bond A 10min light
  - Etch:
    -SU-8 750 Beach Etcher
  - Metrology:
    - The bonded array was further confirmed with a stylus profilometer
  - Post-processing:
    - FOTS monolayer deposited using MVD100
  - Device:
    - Dow Corning Sylgard 184 PDMS (10:1), UV bonded to glass slide using a thermal oxygen plasma chamber

Gradient Device

Gradient Device Design:

- CAD Design:
- Master Preparation:
  - Resin Application: P-20 3k rpm, 1k rpm/s, 30s, S1827 3k rpm, 1k rpm/s, 30s
  - Exposure: SU-8 contact aligner, hard contact mode, 2.6 exposure developer
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Gradient Device:

Buffer solution was introduced into the culture chamber at a flow rate of 10uL/min. After filling, the inlet and outlet ports of the culture chamber were capped with plugs. Fluorescein at a concentration of 40uM was flowed into the source channel while a blank buffer solution (PBS) was flowed into the sink channel at rates of 1uL/min, 2uL/min, 5uL/min, 10uL/min, and 20uL/min. The device was permitted to reach equilibrium in between changes in flow rate. This was done for both gradient designs.

Procedure

Materials and Fabrication

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Discussion

Cells were successfully loaded into the trapping array, and it appears that several divisions resulted. The devices were fabricated inside of this unique geometric confinement system.

Future: To discriminate between genuine cell division and possible multiple loading events: time lapse imaging, implementation of dual inlet version of device, possible incorporation of valve system, use of colon cancer stem cells.

Gradient device design with increased spacing between transverse channels yielded more reliable gradients. For future: Culture cells in culture chamber, replace fluorescein with Wnt3.

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References
