A MICROFLUIDIC DEVICE FOR SORTING CANCER CELLS BASED ON CELL MOTILITY

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ABSTRACT

In this paper, we present a microfluidic device that allows for sorting of cancer cells based on their chemotactic migration speeds. We used breast cancer cell line cells to demonstrate cell seeding, migration and releasing in our microfluidic device. The ability of our device to provide subpopulations of chemotactically heterogeneous cancer cells should find useful applications in studying cancer and metastasis.

KEYWORDS: Microfluidics, metastasis, chemotaxis, cancer cell migration, cell sorting

INTRODUCTION

Cancer metastases are the major causes of death in cancer related diseases, and are associated with the migration of cancer cells guided by various environmental factors including extracellular matrix, cytokines and growth factors [1,2]. Understanding the downstream signaling pathways of chemotaxis in heterogeneous cancer cell populations will benefit the screening of inhibitors to block cancer cell migration and serve as a potential therapeutic drug for cancer treatment. Conventional in-vitro assays [3] have been widely used for studying cancer cell migration for decades, and have become indispensable in the field of cancer research. In the past few years, microfluidics technology has emerged as a powerful tool for studying cancer cell chemotaxis based on its ability to precisely control the cellular environment during in vitro experiments [4,5,6,7,8]. Despite these new devices have helped to gain new insights in the biology of cancer cell migration, the cells could only be analyzed on-device with microscopy, and thus further characterization and applications of the cells are limited. To overcome this limitation, we have devised a microfluidic chemotaxis platform that allows for sorting and collecting the migrated cancer cells for applications that require subpopulations of chemotactically heterogeneous cells.

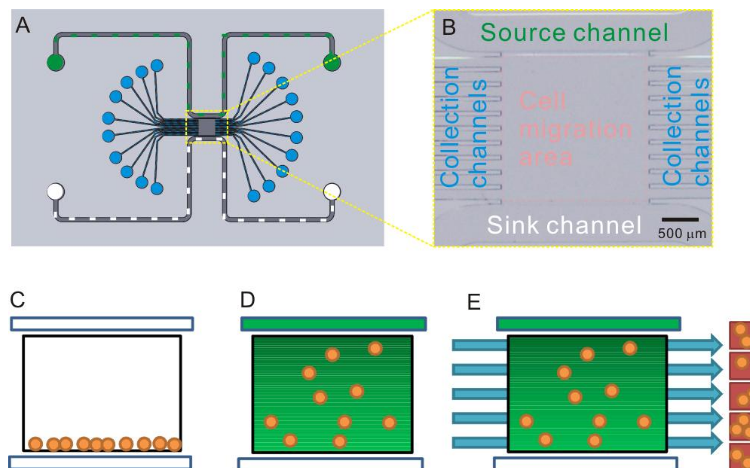

Figure 1: Device design and operation. (A) The schematics of the design of the device consisting of a source (green dashed line), a sink (white dashed line) and cell collection channels (dashed blue lines). There are small channels connecting the source and sink channels to cell migration channel. (B) A picture showing the enlarged area in the middle of the device. (C-E)A cartoon showing the operation of the device: (C) cell loading and aligning, (D) chemoattractant gradient generation and cell migration, and (E) cell release and sorting based on their transverse locations in the microfluidic channel. All the channels are 80 μm high except the small connecting channels that are 2.2 μm high.

RESULTS AND DISCUSSION

The design and operation of the microfluidic device are shown in Figure 1. The operation of the device consists of three major steps: 1) the alignment of cells at one side of the cell migration channel, 2) the formation of a concentration gradient of a soluble chemoattractant across the cell migration channel to induce cell migration, and 3) the release and collection of the cells to different channels according to their lateral positions in the cell migration channel. The cell alignment allows the cells to start their migrations at the same lateral position making it easy to compare their motilities at the end of the experiment. The

concentration gradient of the chemoattractant is generated by diffusion via the small channels between the source/sink channels and the cell migration chamber. Our results show that a stable chemoattractant concentration can be generated (figure 2).

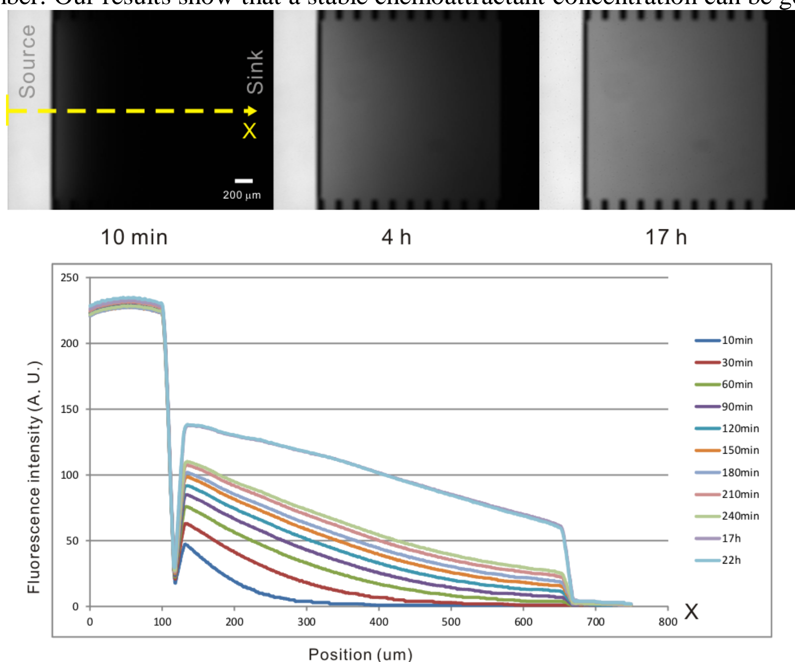

Figure 2: Characterization of the concentration profiles of a fluorescence-tagged dextran (10,000 Dalton). The upper three fluorescence images show the fluorescence intensity profiles at different times after the experiment. Fluorescence intensity profiles of more time points from the same experiment are plotted below.

For the chemotaxis experiment, we used MDA-MB-231 breast cancer cells as our model to test the device. The cells were first introduced into the microfluidic chamber and aligned at one side of the microchamber. The alignment of cells was achieved by applying a gentle suction (using hydrostatic pressure) via the small channels that pined the cells on the sidewall of the microchamber. An epidermal growth factor (EGF) (figure 3) concentration gradient was generated and induced the cells to migrate. After forty hours, the majority of the tested cells had migrated toward the EGF source in the experiment device while most of the tested cells in the control experiment did not (figure 3), demonstrating the chemotatic behavior of the MDA-MB-231 cells in response to EFG gradient.

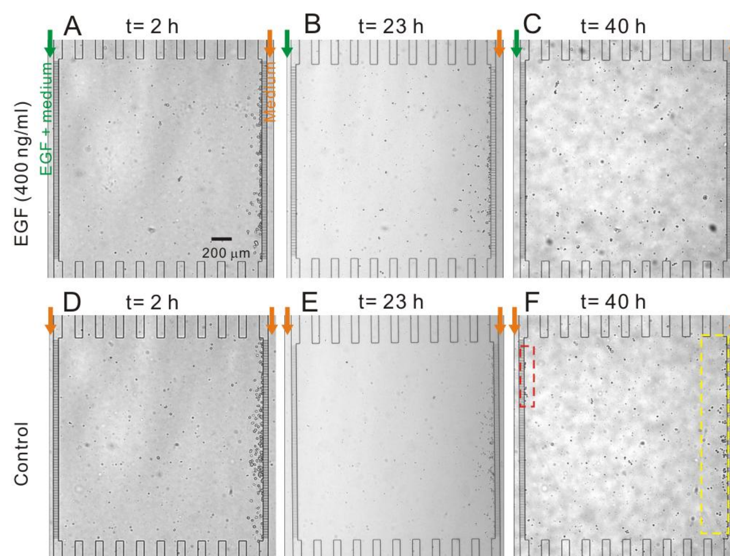

Figure 3: Bright-field images showing the chemotaxis of MDA-MB-231 breast cancer cells in response to EFG in the microfluidic device. In (A) EGF containing and (B) non-EGF containing device, cells were (two hours after cell seeding) located near where they well aligned at the right of the devices. (B) and (C) shows that cells have migrated left toward the

EGF source. (E) and (F) show that in the absence of EGF, the tested cells showed very little directional migration behavior as indicated by many cells were still around their initial locations highlighted in the dashed yellow box. Interestingly, few cells in the non-EGF containing device actually migrated to the left of the channel (highlighted in the dashed red box in (F)).

We have also showed the feasibility of releasing cells from the device (figure4). It is crucial to maintain the balance of the streams used for flowing the released cells into downstream channels to achieve the cell sorting.

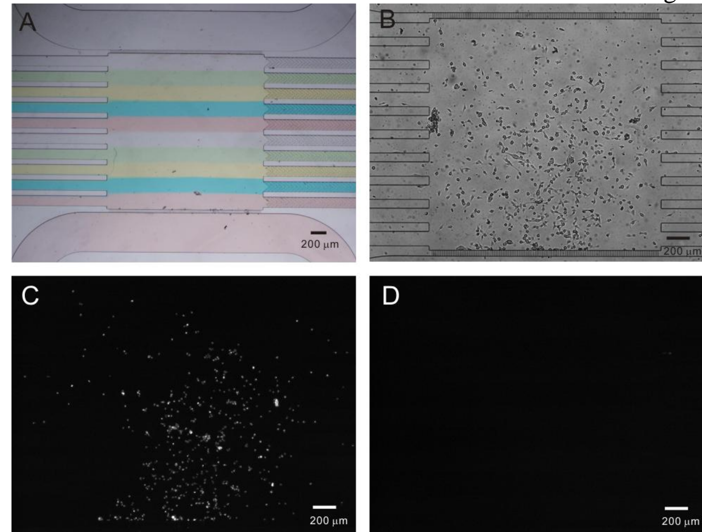

Figure 4: (A) Ten fluid streams (introduced from the left simultaneously) of containing different food dyes were used to visualize the flow paths to ensure that cells could be collected according to their lateral positions after cell release. (B) The MDA-MB-231 cells could be released from the device after the introduction of cell medium containing 0.5% trypsin-EDTA flowing at a 0.5 ml/h flow rate for 4 min (C) and 7 min (D). The cells were pre-labeled with a cell tracker dye. (C) and (D) are taken with fluorescence microscopy

CONCLUSION

In summary, we have designed a fabricated a microfluidic cell-motility-based sorting device and demonstrated the operation of the device with MDA-MB-231 cancer cells. This device may find useful application in studies of cancer cell chemotaxis that require analyzing subpopulations of chemotactically heterogeneous cells.

ACKNOWLEDGEMENTS

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