

Innovation on Microfluidics-based Device for Single Cell Analysis, A Canine Cutaneous Mast Cell Tumor Model

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INTRODUCTION

Recently, our laboratory, Companion Animal Cancer Research Unit, CAC-RU is interested in cancer stem cell (CSC) analysis both at the single cell and the tissue-based levels. However, cellular heterogeneity is still the major hassle for our comprehension in CSC biology. Therefore, to overcome and eradicate this big obstacles, a single cell analysis method must be established. Our laboratory has finally setup and integrated the microfluidics-based single cell analysis into our CSC researches under the association with Department of Mechanical Engineering, Faculty of Engineering, Chulalongkorn University and Thai Micro-electronic Centre, NECTEC, Ministry of Science and Technology, Thailand and Faculty of Medical Technic, Mahidol University since 2013 till present.

BACKGROUND

Cancers are one of the best-known lethally-vicious diseases in all organisms. The diseases have the last long history since the first evidence of cancers has been recognized in the bone of Egyptian ancient fossil which proposed to be the putative osteosarcoma. Cancers are also thought to be the aberrantly proliferative disease of cells owing to genetic instabilities.

Cancer stem cell (CSC) hypothesis is deemed to be the state of art item in oncologic research field both in human medicine and veterinary medicine. The cellular heterogeneity is the major hassle in cell biology. To overcome the problem, the single cell analysis is indispensably recommended. Fortunately, microfluidics has been introduced as a research gear to optimize the researchers to perform single cell analyses in the recent decade.

MICROFLUIDICS DEVICE IN CANINE ROUND CELL TUMORS

The on-going researches using microfluidics applied to the canine round cell tumors, canine cutaneous mast cell tumors, CMCT, which are;

- Sized-based cancer stem cell sorting with Archimedean spiral microchannel based on Dean drag forces and micro-vortices which focus cells into the responsible streamlines depending on their individual sizes; this project has been aimed to separate putative cancer stem cells of various animal neoplasms from the other cell components in a given specimen by which putative cancer stem cells have been speculated to be bigger than non-cancer stem cells. The viability of the sorted cells has been observed.

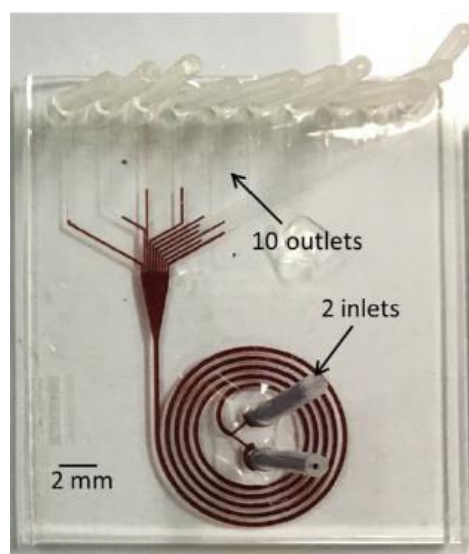


Fig 1. Microfluidics-based spiral-like device for isolating and sorting cells by size (Photographed by: Pimpin, A)

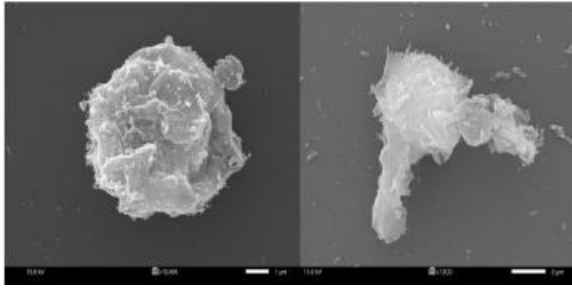


Fig 2. Normal white blood cell and white blood cell damaged from flow force (Photographed by: Suwannaphan, T.)

- b. Microfluidics-single CSC trapping; the objectives of this project are to design and devise the microfluidics-based device which can trap putative cancer stem cells in to the triangular microwell array at the ratio of 1 cell per microwell using recirculation flow generated by microwells. This isolation system is so indispensable because it is the prototype for cultivating any putative cancer stem cells could in microwell array and can be merged the releasing system into the chip for liberating our targeted putative CSC from the microwell for further biological and viability investigation.

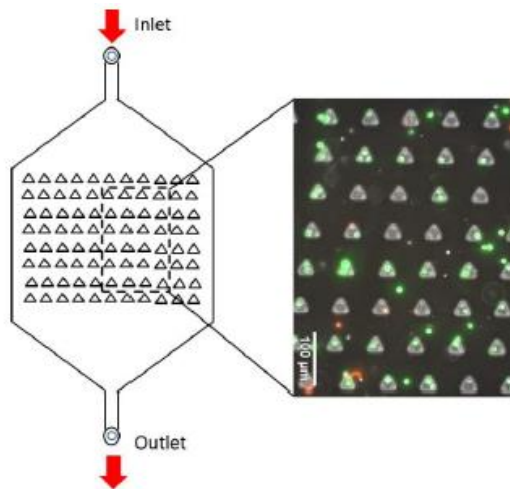


Fig 3: Microscopic triangular trapping system for cancer cells and fluorescent imaging of viable and dead cells in the microscopic reservoirs (Source: Panpattanakul, S and Wongpakham, T)

- c. Microfluidics-single CSC culture and release; this project is aimed to establish the microfluidics-device which can sustain putative CSC cultivation. Moreover, an additional system, releasing system, is also integrated into the device for targeting and releasing the putative CSC of interest.

CONCLUSION

These three phases of the researches are ongoing. We could demonstrated how to use the fundamental of microfluidic devices to manipulate

the behavior of fluid in microchannels resulting in the separation of different-sized cancer cells and trapping in the single cell in the wells with satisfactory viability results.

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