INTEGRATED CENTRIFUGAL PLATFORM FOR POINT-OF-CARE CELL SEPARATION

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ABSTRACT

Most microfluidic cell separation platforms require external pumps, power sources, etc., making them unsuitable for field deployment. Here, we report a portable cell sorting platform based on centrifugal pumping. As a proof-of-concept, we demonstrate size and deformability-based sorting of cancer cells using only 2µl of sample with this platform.

KEYWORDS: Centrifugal pumping, Cell sorting, Point-of-care, RAPID

INTRODUCTION

According to the World Health Organization (WHO), cancer causes ~16% [1] of the total deaths worldwide. To predict the course of cancer, it is important to isolate the cancer cells. Detection of circulating cancer cells (CTCs) in blood is one of the most challenging problems as they are very few in number. Approximately, 1 to 10 circulating cancer cells (size range 20-100µm) are found in 1 billion blood cells (size range 1-30µm). Thus there is a need for a device which can separate cancer cells from blood based on cell size. Most microfluidic cell separation platforms require pumps, power sources, etc., and hence, are not field deployable. We already reported a size-based cell sorting device (radial pillar device or RAPID) [2]. Here we demonstrate a portable cell sorting platform based on RAPID and present initial results on size-based separation of cancer cells.

DEVICE DESIGN

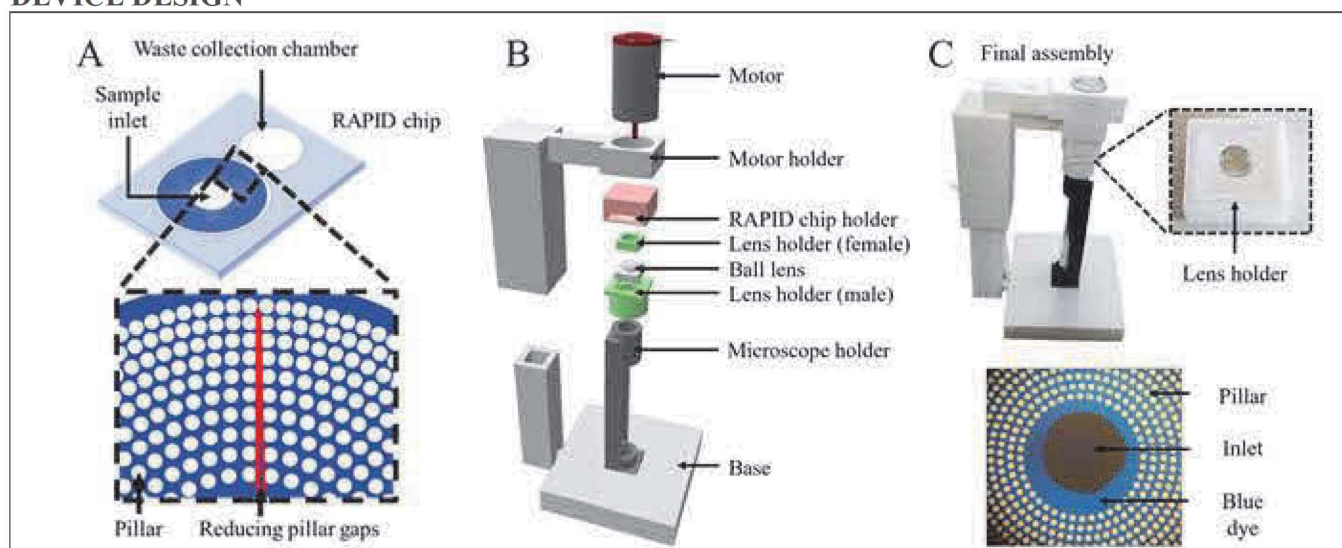

Figure 1: (A) Schematic of the RAPID chip. The device has a central inlet and a waste outlet. The central region holds the captured rare cells. The device has 10 concentrically arranged rows of 100µm diameter pillars. As one moves radially outwards from inlet to outlet, the pillar gap decreases from 30µm to 5µm. (B) Exploded view of the integrated centrifugal cell separation platform. The 100mm x 100mm base minimizes vibrations during centrifugation. The microscope holder can hold a USB microscope. There is another optional lens holder (male-female) for holding a ball lens. The chip holder holds RAPID. The motor holder can hold any standard 25mm DC motor. The DC motor rotates the chip holder (light red), while the other parts (gray and green) remain stationary. (C) Photo of the final assembled prototype, showing the snippet of the lens holder. USB microscope image acquired from the platform shows the RAPID chip filled with blue dye.

As shown in figure 1A, the RAPID chip has 800 pillars, each with a diameter of 100 µm. The pillar gap of consecutive rows reduces from 30 µm (innermost row) to 5 µm (outermost row) as one travels radially outwards from the central inlet. The device has a central inlet through which the sample is loaded, along with outlet for waste

removal. After the experiment, the captured cells are recovered from the inlet itself. The device footprint is 3 mm x 4.5 mm and the chamber height is 25 µm. The pillar arrangement in RAPID is symmetrical about the central inlet, thereby making it suitable for use with centrifugal pumping. Figure 1B shows the schematic diagram of the centrifugal pumping system integrated with a low-cost imaging system. 3D printed poly-lactic acid (PLA) was used for fabricating the system (figure 1C). It consists of a DC motor holder which can hold any motor with 25 mm diameter (14,000 RPM / 12V). The prototype has a detachable microfluidic chip holder and a holder for a commercial USB microscope.

EXPERIMENTAL

The sample was prepared by diluting blood 10,000 times in 1X phosphate buffered saline (PBS). Cultured MDA-MB-231 cells were trypsinized and suspended in PBS. These cells were then spiked in the dilute blood with final concentrations of 80 cancer cells/µl and 150 blood cells/µl. A sample volume of 2µl was then loaded in to the RAPID chip and centrifuged at 3500 RPM for 3 minutes. The large MDA-MB-231 cells were captured by the pillar network. Finally, an image processing algorithm was used to count the captured cells from images acquired.

RESULTS AND DISCUSSIONS

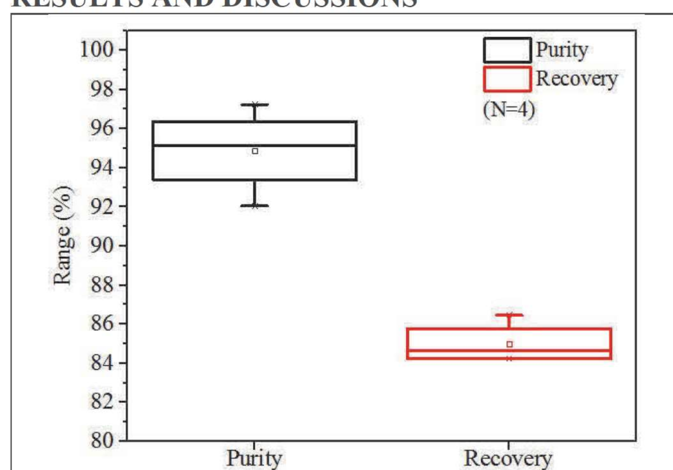

Figure 2: Results of the sorting performed with dilute blood spiked with MDA-MB-231 cells. The average purity of the cancer cells trapped at the inlet was 95%. The recovery was ~ 85%, as a few of cancer cells were able to squeeze through the pillar gaps or got lysed during separation.

The device performance was measured in terms of purity and recovery of isolated cancer cells. The purity was defined as the ratio of desired cells to total number of cells. The recovery was defined as the ratio of total number of cells at outlet to total number of cells at inlet.

Based on four runs, the average purity of the captured MDA-MB-231 cells was 95% along with an 85% recovery of the cancer cells (figure 2). We assumed that the remaining cells had passed through the pillar gaps into the waste outlet.

Our system can use a variable speed motor, unlike the traditional centrifugal systems that use CD/DVD drives with fixed speed of 7200 RPM. The centrally positioned USB microscope obviates the need for a high speed camera or mounting it on the moving disk. The RAPID platform requires very low sample volume (~2µl), can be fabricated at low cost, and is completely portable.

CONCLUSION

We have demonstrated the portable prototype of a centrifugal cell sorting platform that separates cells based on size and deformability. Due to the low sample volume and in-built imaging facility, this platform has a high potential for field deployment.

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