Design of Pinched Flow Fractionation Based MEMS Device for Size and Density Based Cell Separation

P. Mahiz Mathi, D. Lingaraja*, S. Ramya, G. Dinesh Ram
Department of ECE, Saveetha Engineering College, Chennai
***Corresponding author: E-Mail: lingaraja@saveetha.ac.in**

ABSTRACT

The cell separation application is garnering more attention in the areas of clinical diagnosis and medical environment. In this paper, the pinched flow fractionation technique (PFF) based microchannel is designed to separate the biomolecules (RBCs, WBCs and platelets) from the given sample. While comparing with other passive based techniques, PFF based Microchannel helps to separate the biomolecules with respect to its size and density. The design and simulation of microchannel is done by using Comsol Multiphysics. In order to improve the separation efficiency, 2 inlets (liquid with particles and without particles) are used. Blood sample having the biomolecule is introduced continuously in one of the inlet of the microchannel. With the help of liquid flow (without particle) at another inlet, the suspended particles are aligned to one sidewall. The aligned particles are then collected separately based on its size in the broadened segment.

KEY WORDS: Pinched flow fractionation (PFF), blood molecule separation, COMSOL.

1. INTRODUCTION

MEMS technology provides a great boon for the application of medical research and clinical diagnosis. Biomems (Biological Micro Electro Mechanical System) plays a vital role in the modernized medical and surgical applications. The main advantage of biomems is that it requires less amount of sample and produces more accurate results (Jellema, 2008; Wan Shi Low, 2015). Microfluidic technology is highly suitable for size based cell separation. The mechanisms involved in microfluidic separation have two main categories: Active and Passive. If external source is used to induce the separation of biomolecules, then it is said to be active based filter. If there is no external source to induce the biomolecules separation, then it is said to be passive based biofilter. The main drawbacks of active based biofilter are that it alters the properties of bio molecules, very complex to implement and not suitable to make an in-vitro devices. To overcome the drawbacks of active based method, the microfluidic device is designed with passive based biofilter technique (Nicole Pamme, 2007).

Microchannel can be integrated onto a single automated device for performing many processes like sampling, reaction, separation and detection. Dimension of microchannel is in the range of micro or nano scale and it requires less amount of sample. During the separation process, a small aliquot of fluid is handled precisely and manipulated in minimum duration. In passive based approach, the biomolecules are separated based on the fluid flow rate, size of the particles or density. Dimension, fluid flow rate and fluid properties decides the flow type whether it is laminar or turbulent. One of the dimensionless parameter, Reynolds number (Re) is used to decide whether the flow is laminar or turbulent. In microfluidic channel, Laminar flow is the most acceptable flow for cell separation applications (Masumi Yamada, 2004).

The mechanisms involved in passive based filtering technique are hydrodynamic chromatography, pinched flow fractionation, hydrodynamic filtration, sorting by diffusion, deterministic lateral displacement, hydrophoretic separation, secondary flow based separation, etc. (Satoshi Sunahiro, 2008; Sumedh R. Risbud, 2014). Among these, pinched flow fractionation separates the particles based on the density (Nakashima, 2004). In biological and chemical applications, density based separation is a crucial step to prepare the summary. The importance of density based separation is often used to distinguish the dead cells, stem cells and identify the specific bacterium in complex mixture (Jing-Tao Ma, 2016). While comparing with field flow fractionation, PFF can eliminate the need of the complicated outer field (Teknologi, 2010).

In this paper, the microchannel is designed to separate the biomolecules of different sizes. The designed microchannel consists of three sections as follows, two inlet section, one pinched section and four broadened outlet section. In this method, the fluid with biomolecules is introduced continuously at the inlet of the microchannel and liquid without particles or biomolecules (Buffer solution) are continuously introduced through another inlet of the microchannel. At the pinched segment, due to the mixing of buffer solution, the biomolecules are aligned at the one sidewall. Based on size of the biomolecules, spreading flow profile starved to separate the biomolecules perpendicular to the flow direction. The branches are made at the broadened section that helps to collect the separated biomolecules independently.

Design Principle: The principle involved in pinched flow fractionation is shown in Figure.1. Liquid containing particles (Blood) and without particles (Buffer) are introduced at both inlets of the microchannel simultaneously. Inside the pinched segment, the liquid containing particles experience centrifugal force and the particles are highly focused to one of the side wall (Teknologi, 2010). By controlling the flow rates of both the liquids, the particles are

aligned on one sidewall. While passing the liquid with particles from pinched segment to the broadened segment, based on the size, the particles are separated.

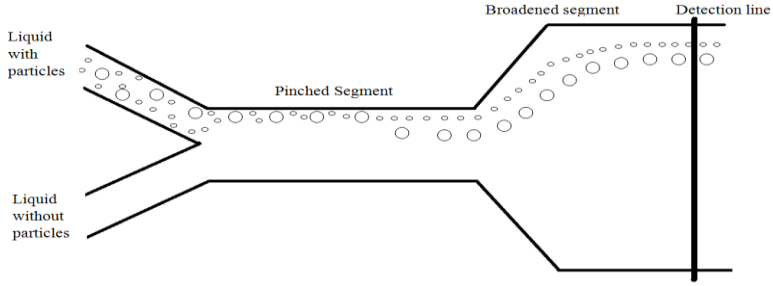

Figure.1. Principle of pinched flow fractionation

If the size of the particles is larger, then the exerted force helps the particles to move towards the center. If the particles are smaller in size, then the exerted force helps the particles to move towards the wall (Junya Takagi, 2005).

The main advantage of the broadened section is to amplify the slightly different position of the particles which is obtained in the pinched segment into large one. The performance of the separation can be affected by various factors such as flow rate, device geometry and boundary angle (Junya Takagi, 2005). In the pinched segment, the area of the liquid with particles is determined by the ratio of the inlet flow rates. Variation in the inlet flow rates may alter the alignment of the particles at the pinched segment. Additionally, the movement of the particles can be affected by the dimension of the pinched segment and the boundary angle between the pinched and broadened segment (Kao-Feng Yarn, 2012). When the speed of the particles is increased, the initial movement of the particles are also getting increased (Asger Vig Larsen, 2008).

Design of Microchannel: For cell separation applications, microchannel mostly works under the conditions of laminar flow profile. To ensure the laminar flow condition, the dimensionless number called Reynolds number (Re) is calculated. The criterion to achieve the laminar flow condition is that the Reynolds number must be less than 2100. The formula to calculate the Reynolds number is given by,

$$Re = \frac{\rho D_h}{\eta} v$$

Where, ρ is the fluid density, V is the fluid velocity, η is the viscosity, D_h is the hydraulic diameter.

The hydraulic diameter is calculated as follows;

$$D_h = \frac{2wh}{(w+h)}$$

Where w and h represents the width and height of the microchannel.

The structure of the PFF based microchannel is shown in figure.3. The designed microchannel consists of two inlets and four outlets. Figure.3(a), represents the 2D view of the designed microchannel with proper dimensions to separate the particles effectively and figure.3(b), represents the 3D view of the microchannel. Two inlets are having the same dimensions as 35µm and all the four outlets are having the same dimension as 15µm. The length and width of the pinched segment are 45µm and 30µm respectively. The liquids containing and not containing particles are continuously introduced into the microchannel via the two inlets and the separated particles are collected based on the size of the designed four outlets.

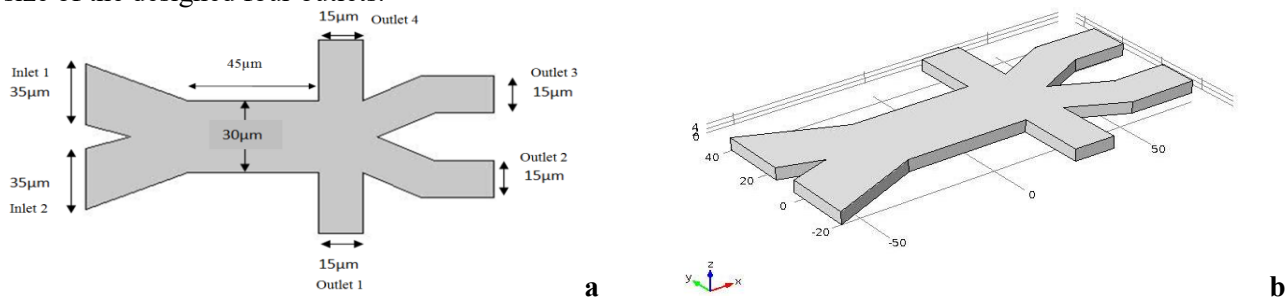

Figure.3. Basic structure of PFF based microchannel (a) 2D view (b) 3D view

Simulation of PFF Based Microchannel: PFF based microchannel is designed and simulated using the COMSOL multiphysics software. In order to analyze the laminar flow profile, simple straight microchannel is designed and analyzed. The laminar flow profile for straight microchannel is represented in figure.4.

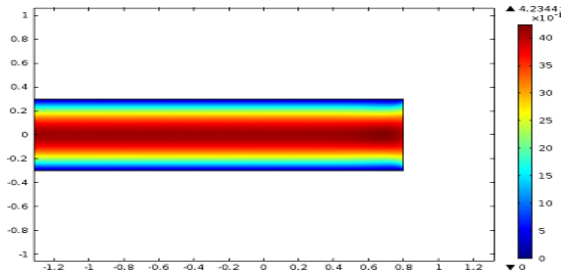

Figure.4. 2D simulation of laminar flow

In Figure.4, the red colour indicates the maximum velocity flow and the blue colour indicates the minimum velocity flow. As per the parabolic profile of laminar flow, the particles having the larger diameter attain the maximum velocity and it travels along the midway of the channel. Whereas the biomolecules that are having the minimum diameter attain the minimum velocity and it travel just below the walls. The height and width of the straight channel are 80μm and 140μm respectively. Density, Viscosity and flow rate of the fluid (blood) are 1025 kg/m^3, 3.5e^{-3}m^2/s and 10μl/min respectively. With this defined dimensions, the obtained Reynolds number is 0.27x10^{-8}. Reynolds number obtained is very small which indicates the laminar flow profile of fluid inside the microchannel.

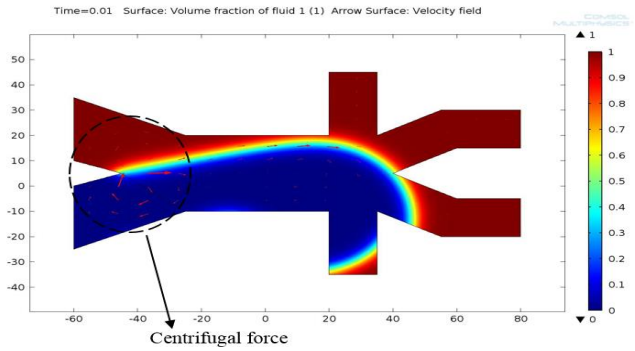

Figure.5. Two liquids flow through the microchannel

The blood sample (liquid with particles) is introduced into the microfluidic channel continuously via the inlet 1. At the same time the buffer solution (liquid without particles) is also introduced via the inlet 2. Both the liquids were introduced with different fluid flow velocity rates. The blood sample is introduced with 0.0015m/s velocity rate and the buffer solution is introduced with 0.001m/s. The different fluid flow rate is used to align the particles effectively at the pinched segment. At the beginning stage in the pinched segment, the two liquids are mixed with surface tension 0.07 (which is the surface tension of blood). Due to the centrifugal force that exists at this point as shown in figure.5, the particles in the liquid with higher density starts moving towards one of the side wall. The buffer solution helps in aligning the particles based on the size. According to laminar flow profile, smaller molecules attain minimum velocity and larger molecules attain maximum velocity. Due to this laminar profile, the smaller molecules travel just below the walls and larger molecules travel along the midway of the channel. The entire length of the pinched segment is used to align RBC, WBC and Platelets at one side wall.

At the exit of the pinched segment, based on the size, the particles are aligned. From the pinched segment, the particles enter the broadened segment. The broadened segment is divided into four outlets of equal width and this helps collecting the WBC, RBC, Platelets and other biomolecules individually and effectively. The separation of different biomolecules can be illustrated with the help of streamlines that is created in the microchannel. The particles when flows through the microchannel follow the streamline path. The streamlines as shown in figure.6, clearly shows that the particles with smaller size (< 1μm) flows nearer to the wall and it enters the outlet 4. The particles with size 2-8μm enters flows the streamline path and it enters the outlet 2 and 3. The particles with larger size (>8μm) enters the outlet 1.

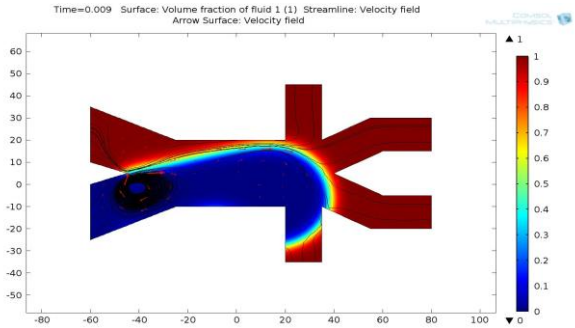

Figure.6. Streamlines regime of PFF based microchannel

2. CONCLUSION

The designed microchannel works based on the mechanism of pinched flow fractionation. Due to the presence of the pinched flow segment, inside the bifurcated microfluidic channel, the biomolecules attains the centrifugal force which helps to separate the biomolecules with respect to their size and density. The critical flow rate helps to separate the larger and smaller biomolecules. The width of the designed pinched segment is greater than targeted biomolecules. Hence the obtained critical flow rate is sufficient to collect the separated platelets, RBCs and WBCs at the outlets 1, 2, 3 & 4 accordingly.

REFERENCES

Asger Vig Larsen, Lena Poulsen, Henrik Birgens, Martin Dufvaa, Anders Kristensen, Pinched flow fractionation devices for detection of single nucleotide polymorphisms, Lab Chip, 8, 2008, 818–821.

Jellema L.C, Markesteijn A.P, Westerweel J, Verpoorte E.M.J, Size Separation of Particles Using Flowing induced Electrokinetic Trapping, International Conference on Miniaturized Systems for Chemistry and Life Sciences, 2008, 170-172.

Jing-Tao Ma, Yuan-Qing Xu, Xiao-Ying Tang, A Numerical Simulation of Cell Separation by Simplified Asymmetric Pinched Flow Fractionation, Computational and Mathematical Methods in Medicine, 2016.

Junya Takagi, Masumi Yamada, Masahiro Yasudaa, Minoru Seki, Continuous particle separation in a microchannel having asymmetrically arranged multiple branches, Lab Chip, 5, 2005, 778–784.

Kao-Feng Yarn, Yu-Jen Pan, Win-Jet Luo, Chun-Nan Chen, Separation And Trapping of Microparticles Utilizing AC Electroosmotic flow in a Y-Shaped Microchannel With Asymmetric Electrode Pairs, Digest Journal of Nanomaterials and Biostructures, 7 (4), 2012, 1735-1745.

Masumi Yamada, Megumi Nakashima, Minoru Seki, Pinched Flow Fractionation: Continuous Size Separation of Particles Utilizing a Laminar Flow Profile in a Pinched Microchannel, Anal. Chem., 76 (18), 2004, 5465-5471.

Masumi Yamada, Vivi Kasim, Megumi Nakashima, Junichi Edahiro, Minoru Seki, Continuous cell partitioning using an aqueous two-phase flow system in microfluidic devices, Article in Biotechnology and Bioengineering, 2004.

Nakashima M, Yamada M, Seki M, Pinched Flow Fractionation (PFF) for Continuous Particle Separation in a Microfluidic device, IEEE, 2004, 33-36.

Nicole Pamme, Continuous flow separations in microfluidic devices, Lab Chip, 7, 2007, 1644–1659.

Satoshi Sunahiro, Masashi Senaha, Masumi Yamada, Minoru Seki, Pinched Flow Fractionation Device For Size- and Density-Dependent Separation of Particles Utilizing Centrifugal Pumping, International Conference on Miniaturized Systems for Chemistry and Life Sciences, 2008, 164-166.

Sumedh R. Risbud, German Drazer, Mechanism governing separation in microfluidic pinched flow fractionation Devices, Microfluidic and Nanofluidics, Springer, 17, 2014, 1003-1009.

Teknologi and Applikation Vig, Asger Laurberg, Kristensen, Anders, Pinched Flow Fractionation, Department of Micro- and Nanotechnology, Technical University of Denmark, 2010.

Wan Shi Low and Wan Abu Bakar Wan Abas, Benchtop Technologies for Circulating Tumor Cells Separation Based on Biophysical Properties, BioMed Research International, Hindawi Publishing Corporation, 2015.