The Two-Phase Microchannel Flow Study of Chicken Blood on Lab-on-a-Chip

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Abstract

In previous study, we introduce problems in assembly the polydimethylsiloxane (PDMS) lab-on-a-chip (LOC) devices from literatures; a manual assembly by using acrylic plates and an adhered assembly by using a plasma cleaner. Since electrodes in the devices had been designed to provide a strong dielectrophoretic force by using sputtering and electroplating techniques, the thick electrodes were used. The PDMS LOC was also aimed to use with biological samples to study the different flow patterns of the samples prepared from chicken blood and to solve a PDMS hydrophobic problem from our previous study where the sample cannot flow through a channel of the adhered assembly LOC. Since the chicken blood contains of whole blood and plasma, the sample can be considered as a combination of particle and fluid parts or the two-phase flow. The channels on the PDMS LOC having dimensions of the order of microns can be considered as microchannels; Knudsen number is used to distinguish flow regimes. From the experiment study, the flow problem has still existed in the adhered method but the flow problem did not exist on the manual method with a new-acrylic-plate set. Slip flow occurred at the leak areas because interactions between the sample and walls became significant. Different flow patterns occurred in the channels of workable LOCs in different channel distances. No slip (continuum) flow occurred at the middle path where pressure drop was decreased. The hydrophobic problem was solved by using Bovine Serum Abumin solution to reduce PDMS hydrophobic behavior and this solution did not affect red blood cells because of none damaged cells.

Keywords*:* LOC, Lab-on-a-Chip, Chicken Blood, Two-Phase Flow, PDMS, DEP Force.

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1. Introduction

Lab-on-a-chip or LOC devices are miniature laboratories built on a thin glass or plastic chip of several micrometers in dimensions. These small devices can duplicate the specialized functions as their room-sized counterparts in clinical diagnoses; the advantages of these devices include significantly reduced reagent consumption, short analysis time, automation, and portability [1]. Researches on LOC systems have been developed by the aim to miniaturize, integrate, and automate biochemical assays. The development of the LOC systems and devices requires the integration of multiple fluidic functions onto a single system or device with the ability of a well defined fabrication process to fabricate cost efficient and portable devices. Since there is the large diversity in microfluidic developments, collaborations, and supply chains of different companies forming new solutions by combining off-the-shelf components are still out of reach [2]. Microfluidic platforms provide a variety of design components and fabrication technologies that are well tuned on to each other [3]. In a microfluidic platform, basic fluidic functions (unit operations), are reagent storage, reagent release, fluid transport, fluid metering, fluid mixing, flow controlling, and separation or concentration of molecules or particles [2].

Many experimental and analytical researches have been performed to understand fluid flow at microscale for the design and the development of micro devices. However, there are no generalized solutions for the determination of flow characteristics in designing micro devices. On the other hand, the predictions of the friction coefficients and velocity profiles in microchannels are important in the application of electronics where such channels formed in the micro devices. Experimental and theoretical results for liquid flows cannot be considered or modified for gaseous flows in the microchannels because the flow regime boundaries are significantly different as well as flow characteristics. Surface to volume ratio in microstructures are greater than it is for macrostructures so surface effects become dominant [4]. This effects result an important pressure drop and greater mass flow rate than that is predicted with the continuum theory. The velocity in microchannels is not very high due to large pressure drops. Because of small dimensions, Reynolds number becomes smaller. In the transition from laminar to turbulent flows viscous effects and friction factors may be higher than those are predicted by conventional theory. All these discrepancies affect both the fluid flow and the convective heat transfer in microchannels [4, 5]. The high pressure gradients in microchannels lead to low flow rates. In small devices, the ability of the fluid stream to carry the heat away for a given temperature rise becomes limited but reducing the flow length of the channels and increasing the liquid flow rate must be

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matched with their performances. As a result, employing multiple streams with short paths in a microdevice was recommended. One should note that, in the transition and turbulent flow regions, the reduced flow length reduces the pressure drop, multiple inlets enlarging channel area where the heat transfer rate is higher, and turbulent flow can help to increase heat transfer coefficients in these regions [6]. When interactions between fluid molecules and channel walls become as frequent as intermolecular collisions, the boundaries and the molecular structure become more effective on the fluid flow, these effects are called rarefaction effects [4]. The local Knudsen number is used to determine the degree of rarefaction and the degree of deviation from continuum model.

$$Kn = \frac{\lambda}{L} \quad (1)$$

where L is a characteristic flow dimension such as channel hydraulic diameter (D_h) and λ is the mean free molecular path corresponding to the distance travelled by the molecules between the collision. In continuum flow regime, no-slip flow where $Kn \leq 0.001$, continuum assumption which is widely used for macroscopic problems becomes valid. In the slip flow regime where $0.001 < Kn \leq 0.1$, continuum model is applicable except in the layer next to the wall which can be identified as Knudsen layer. For the Knudsen layer, slip boundary conditions should be considered. If the flow is in the transition regime, where $0.1 < Kn \leq 10$ and continues into free molecular flow regime, molecular approach should be used. In other words, Boltzman equation should be considered for atomic level studies of gaseous flow in transition regime [5]. Many experimental investigations on convective heat transfer of single-phase liquid flows in microchannels have been in the continuum regime [4].

Our previous study [7] presented problems and results in fabricating the microfluidic devices for biological sample manipulation at Kasetsart University, Chalermphrakiat Sakon Nakhon Province Campus. Electrodes obtained from electroplating and sputtering techniques were used to produce dielectrophoretic force on the devices. In the work, the electrodes were fabricated with three different current conditions, the higher current in electroplating process, the coarser electrode surfaces. The polydimethylsiloxane (PDMS) lab-on-a-chip (LOC) design from a literature was considered specifically as a microfluidic device prototype. The PDMS LOCs were adhered by using the plasma cleaner at at Nanoelectronics and MEMS Laboratory, National Electronics and Computer Technology Center (NECTEC) and were fabricated manually at Chalermphrakiat Sakon Nakhon Province Campus but all components were prepared at NECTEC. The finished LOC with the coarse surface electrodes showed assembly problems in both assembly methods. Moreover, in the work, the biological sample could not flow through a channel of the adhered assembly LOC showing a hydrophobic problem. The LOC fabricated locally and manually without using plasma cleaners can be reassembled, considered as the main advantage. However, the re-assembly requires skills and techniques to arrange all components to be on exact positions. The completed-workable LOCs were examined with two sets of biological samples, prepared from chickens with poor and good health conditions. The results showed that red blood cells in the sample from the good health condition could flow and be accelerated pass the electric field from the electrodes. But the red blood cells in the sample from the poor health condition could not flow smoothly to pass the electric field and some of them were moved forward to the electrode. Therefore, the last feasibility study showed that there was possibility to develop the LOC technology locally with helps from Nanoelectronics and MEMS Laboratory [7].

In this current study, the leak LOC problems were examined and solved by observing biological samples flow in the LOCs. The best electrode condition from the previous work was implemented in the current work. The biological sample was prepared [7] from healthy chicken blood. Sample flow patterns in the microchannels were observed to investigate flow behaviors, since the biological sample flow can be distinguished as two-phase flow in the microchannel. The hydrophobic problem was investigated and solved.

2. Experimental Study

The LOCs were fabricated according to the literature [7]; a manual assembly by using acrylic plates and an adhered assembly by using a plasma cleaner. The mask for fabricating electrode was designed according to Cetin design [8] by using L-Edit software. In the fabrication process, the silicon wafer was used to produce a master prototyping of the PDMS microstructure and was patterned by using the negative photo-resist (SU-8 25, MicroChem Co., Newton, MA) technique. The dielectrophoretic chamber was made from PDMS prepared by mixing the precursors sylgard with a curing agent at a ratio of 10:1 by volume. The prepolymer mixture was degassed at 20-50 mTorr at room temperature in desiccators pumped with a mechanical vacuum pump for 10 minutes to remove any air bubbles in the mixture. The PDMS mixtures were gradually poured onto the patterned silicon wafer or a mold. After the PDMS was cured at 100^{o}C for 30 minutes on the mold, the molded polymer samples were peeled off and punched holes in order to create chambers. Because electroplating technique can vary electrode surface conditions [9], the chromium electrode array was patterned on glass slides by only DC sputtering technique through microshadow masking. The chromium were sputtering under argon plasma with sputtering pressure, sputtering current, and time of $3x10^{-3}$ mbar, 0.2 A, and 2 minutes, respectively. The sputtering was conducted at room temperature.

For the adhered assembly by using a plasma cleaner [10-20], the chamber and electrodes were treated under oxygen plasma (Harrick scientific corp. model PDC –32 G) before being attached to each others. The microelectrodes were inserted into the PDMS electrode chambers manually under the microscope. In a manual assembly [9], acrylic plates were investigated to improve a leak problem and searched for a new design.

On the LOCs, channel width and height were 50 μm and 25 μm, respectively. With these dimensions, the channels can be considered as the microchannels. In the workable LOCs, the sample flow patterns were observed to investigate flow behaviors, since two-phase flow in the microchannels can be different from liquid flow in the microchannels.

With the PDMS hydrophobic problem, Bovine Serum Abumin solution as a new solution was mixed with the sample and the PDMS sheets were kept under water to solve the hydrophobic behavior. The Bovine

Serum Abumin solution was inspected for cell damaging while it flowed with the sample.

3. Results and Discussion

The thick electrodes affected both ways of LOC assembly, the thinner electrodes from only applying the sputtering technique were used in the LOCs. The sample flowing through the adhered LOC with the thinner electrode showed a problem, Fig.1, collapse of channel ceiling. In the manual assembly, the former (simple) acrylic plates (Fig. 2) were replaced by a new-acrylic-plate set as shown in Fig. 3. The LOCs from both assembly methods were workable.

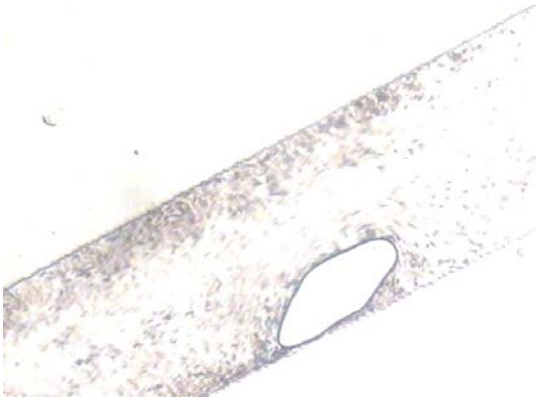

Fig. 1 Collapse of channel ceiling in the adhered LOC.

Fig. 2 The first acrylic plates used in the previous work [7].

Different flow patterns were observed in the channels for both workable LOCs and leak LOCs. From the observations, slip flow occurred at the leak areas as in Fig. 4–5 because friction between the flow and the surfaces was increased, interactions between the sample and the surfaces became significant and higher pressure drop occurred because higher friction force at boundaries. In the workable LOCs, the LOCs were inclined, before the sample loaded, to let the gravitational force drove the sample flow. No slip (continuum) flow occurred in the workable LOC at the middle path (Fig. 6) because of less friction at the boundaries which implied that lower pressure drop and smaller mean free molecular path (less friction to pull molecules apart), hence, lower Knudsen number took place.

In this study, we found the sample flowed in the microchannels pass the electrode area, in the workable LOC, with the velocity of 25.28×10^{-6} m/s. At low velocity, the particles flowed smoothly along the liquid part with no-slip condition between the particles and the liquid. From the flow patterns and the velocity, the sample including particle and liquid parts flowed in the same patterns as single-phase liquid.

Next, the hydrophobic problem was investigated and solved by choosing Bovine Serum Abumin to reduce the PDMS hydrophobic behavior as shown in Fig. 7. From Fig. 7(b), the Bovine Serum Abumin did not affect red blood cells as it did not damage any red blood cells. Another experiment to reduce the PDMS hydrophobic behavior was keeping the PDMS sheets under water, then the sheets were dried by air blasting before they were assembled with other components. Since the sample flowed inside the LOCs with the submerged PDMS as shown in Fig. 8, keeping the sheets under the water can be another solution to solve the hydrophobic behavior.

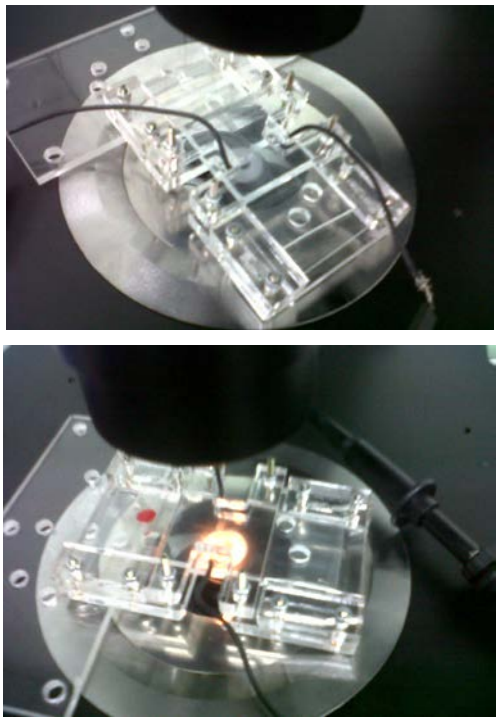

Fig. 3 A new-acrylic-plate set used in the current work.

Fig. 4 Slip flow profile occurred at entrance of a leak LOC because of high friction force at boundaries.

Fig. 5 Slip flow profile occurred at the middle path of the channel because of leaking next to an electrode

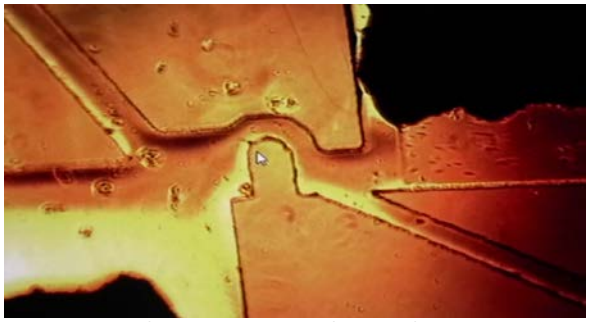

Fig. 6 No slip flow or continuum flow profile occurred at the middle path of the channel because of less pressure drop.

(a)

(b)

Fig. 7 Biological sample flowing with Bovine Serum Abumin (black particles) (a) overview picture and (b) zoomed in picture.

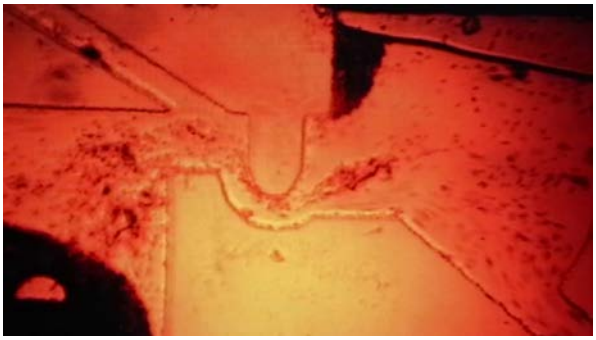

Fig. 8 The workable LOC with a submerged PDMSsheet.

4. Conclusion

From the experiment study, the problem in the adhered LOC has still existed. The sample flow in the manual LOC was improved with a new-acrylic-plate set. Slip flow occurred at the leak areas because higher friction. The sample flow in our study was a two-phase flow with low velocity, so no-slip condition between particles and fluid. Therefore, the sample flow in the microchannels behaved the same trend as the liquid flow in the microchannels, a generalized solution for the determination of flow characteristics as mentioned in a literature [21]. The hydrophobic problem was solved by keeping the PDMS sheets in water and using Bovine Serum Abumin solution to reduce PDMS hydrophobic behavior. The latter solution did not affect red blood cells because of none damaged cells.

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