MEASUREMENT AND COMPUTATION OF LYMPHOCYTE DEFORMATION BY USING MICROCHANNEL FLOW AND THE COMPOUND DROP MODEL

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ABSTRACT

The aim of this study was to examine the physical properties of lymphocytes and to develop a numerical model to predict their motion and deformation. Measurements were made using microchannels with a contraction region and numerical computations were carried out using the compound drop model described in this study, which considers the shear-thinning effects of the cytoplasm and the presence of the nucleus. The results of computational and experimental efforts to study the non-linear effects of elongational flow on the lymphocyte deformation showed excellent agreement.

KEYWORDS: Drop model, Lymphocyte characteristics, Deformation, Microchannel

INTRODUCTION

Micro technologies have been used to gain a better understanding of physical aspects such as properties, motion and deformation of single blood cells during flow, with the aim to provide novel biological insights that may have potential medical applications. While red blood cells have been the main subject of these previous studies, little attention has been paid to lymphocytes and other white blood cells. Further, lymphocyte deformability has mostly been measured using the micropipette aspiration, the accuracy of which may be low because of wall friction and limited number of samples.

In this study, microchannel flow was used to directly measure the shape of each lymphocyte, quickly and continuously. The contraction region shown in Figure 1 produced an elongational flow, which deformed the lymphocyte with time, thus allowing us to measure the viscoelastic properties of the cell. On the bases of these results, we present a compound drop model which considers the shear-thinning effects and the presence of the nucleus to precisely match the properties and deformation characteristics.

NUMERICAL MODEL

The present model is based on the drop model which consists of the membrane with zero thickness and the cytoplasm only (the fluid inside). Surface tension is applied to the membrane. The ND model is the model with the Newtonian fluid properties applied to the cytoplasm. The shear-thinning drop (STD) model considers the shear-thinning effect of the cytoplasm as Eq. (1).

$$\mu_c = \max\left[\mu_{c0}\left(1+\frac{\dot{\gamma}}{\dot{\gamma}_c}\right)^{-\beta}, 6.25 \right] \text{ [Pa·s]} \tag{1}$$

μ_{c0} is the zero-shear viscosity. Note that a lower limit 6.25Pa·s is applied to μ_c in order to prevent a significant decrease of the viscosity. In addition to this, the STCD model considers the nucleus placed in the cytoplasm by applying another drop of much higher viscosity.

Table 1 shows the physical properties and parameters of each model. For the coupling the lymphocyte and the flow field, Immersed Boundary method was employed to the boundary.

EXPERIMENTAL SETUP

Figure 1 shows the schematic of the microchannel together with the dimensions. The contraction part is located at the middle. The microchannel was made of PDMS (poly-dimethylsiloxane) with a cover glass attached to the bottom. The motion and shape of the lymphocytes were measured by a high speed video camera (Vision Research Co. Inc., Phantom V7.3). The image recoded by the camera had the size of 800×600pixels with the resolution of 0.217μm/pixel.

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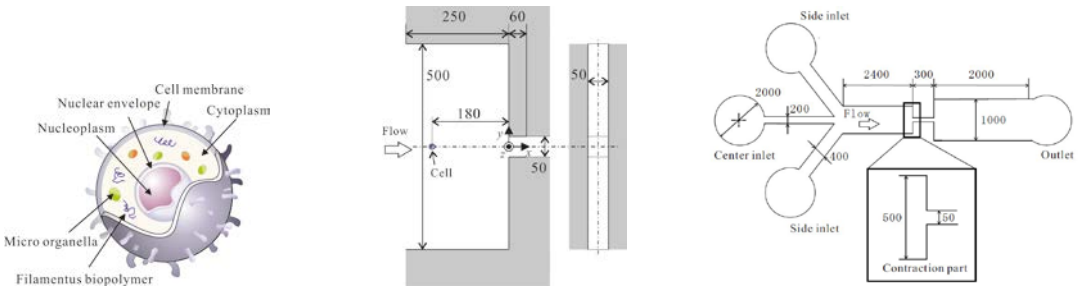


(a) Schematic of lymphocytes *(b) Computational domain* *(c) Microchannel*

Figure 1: Schematic illustrations of the lymphocyte, and contraction region; dimensions are in micrometers

Table 1. Lymphocyte properties of each drop model

Parameters	Models		
	ND	STD	STCD
Cell membrane tension τ_c [μN/m]	30	30	30
Apparent viscosity [Pa·s]	10	–	–
Zero shear viscosity μ_{c0} [Pa·s]	–	250	250
Shear-thinning parameter β [–]	–	0.5	0.5
Characteristic shear rate $\dot{\gamma}_c$ [1/s]	–	–	0.05
Nuclear membrane tension τ_n [Pa·s]	–	–	300
Nucleoplasm viscosity μ_n [μN/m]	–	–	250

Jurkat cells (ATCC, CRL-2570) was used as lymphocytes in the experiment. The collected cell was then suspended in PBS to which PVP (MW=3.6×105, Nakalai tesque, K-90) was mixed. PVP 8wt% and 12wt% solutions were used to obtain solutions of different viscosity.

RESULTS AND DISCUSSION

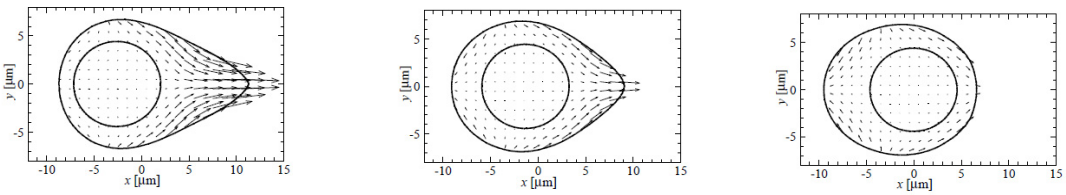
Figure 4 (a) shows the relationship between deformation index (*DI*) and flow rate (*Q*). *DI* is the value measured at the inlet of the contraction region, *x*=0. In the case of ND model, the *DI* remained constant or gradually decreased with *Q*. This is due to the fact that under simple elongational flow condition, the deformation rate becomes linear to time when the cell property is viscous dominant. Therefore, although the enlongational force increases with the flow rate, the *DI* at *x*=0, which is the integrated value, remained constant against *Q*.

On the other hand, the *DI* of the STD and STCD models increased, corresponding with the experimental measurements. Figure 3 shows the influence of the nucleus and its position on deformation characteristics. The results of STCD model ((a)-(c)) shows that the rigid nucleus suppressed cell deformation and that a slight deviation in its position markedly affected the deformation rate. Further, the shape of the lymphocyte predicted by the computation were in close agreement with the experimental results. As shown in Fig. 4 (a), the results of STCD model show a better agreement with the experiment than the STD model particularly in the small *Q* region. These results indicate the validity of the model and how important it is to include the nucleus in the computation.

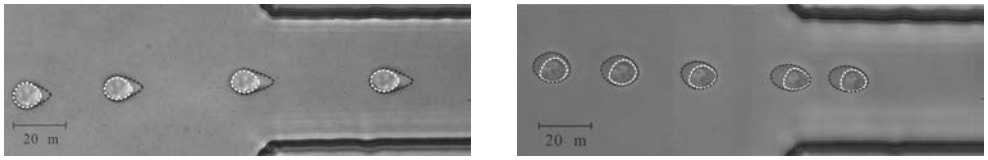
Further analysis of deformation characteristics allowed us to predict the fluid properties of the cytoplasm. Figure 4 shows the relationship between shear rate and viscosity, as measured by the present model. μ_c is the viscosity considering the shear-thinning effect, and is defined as Eq. (2).

$$\mu_c = \frac{0.74\sigma_{ext0}}{DI} f\left(\dot{\gamma}_0\right) \tag{2}$$

f is an adjusting function obtained by a preliminary computation for elongational flow, and σ_{exp0} is the stress applied to the cell by the surrounding fluid. Although the present measurement of the viscosity is lower than that predicted by the empirically derived formula of Drury et al., the results show a reasonable match with the distribution.

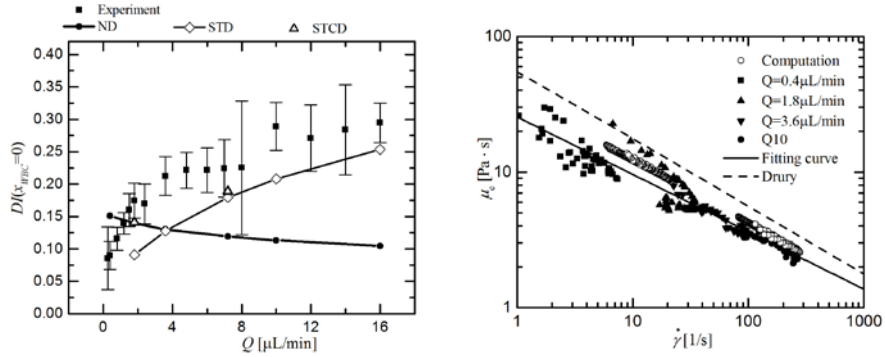


(a) $\Delta x_n = -1.5$ μm (b) $\Delta x_n = 0$ μm (c) $\Delta x_n = 1.5$ μm



(d)Nucleus located at the trailing end (e)Nucleus located at the leading end

Figure 3: (a)~(c)Effects of the nucleus position on the lymphocyte deformation and flow pattern of the cytoplasm in the computation: the position is moved ±1.5 μm in the streamwise direction. (d) & (e) snapshots of the measurements showing identical effects of nucleus.



(a) *DI* and *Q* (b) shear rate and visocity

*Figure 4: **(a)** Relationship between deformation index (DI) measured at the contraction region, and flow rate (Q): comparison of the results obtained from the experiment (scattering symbols) and computation using different models (lines), and (b) Relationship between the shear rate and viscosity predicted from the computation (filled symbols) and experiments (outlined symbol), and the results obtained by Dury et al.*

CONCLUSION

The nucleus showed significant effects on the deformation characteristic of the lymphocyte. STCD model considering this effect showed a good agreement with the measurement result in the flow rate and *DI* relationship. Further, by comparing the experimental results with the simulation, the shear-thinning effect of the lymphocyte could be estimated with a simplified model.

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