ENHANCED CELL STIFFNESS EVALUATION BY TWO-PHASE DECOMPOSITION

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ABSTRACT

This work proposes an enhanced method for cell stiffness evaluation by eliminating the effect from cell viscosity. While the passing time of cell through a μ-channel is used for evaluating cell stiffness in the conventional method, the time includes both the effects of cell stiffness and cell viscosity. The key idea is that we separate the cell motion inside a μ-channel into two phases, the phases of deformation and constant velocity. We evaluate the cell stiffness by focusing on the cell motion in the phase of constant velocity. The results of experimental study are presented and discussed.

KEYWORD

cell stiffness, μ-channel, stiffness evaluation

INTRODUCTION

It is documented that there are human diseases associating with stiffening/softening of red blood cells [1,2]. As a result, it is important to develop a fast and accurate method to evaluate cell stiffness for medical practices. There are different approaches for evaluating biomechanical properties of cells [3]. The method of evaluating cell stiffness by a μ-channel [4] is much faster than most of conventional methods, such as micropipette aspiration [5], optical tweezers [6] and atomic force microscope [7]. However, the accuracy of the μ-channel method is sacrificed for its high sensing speed. In this work, we proposed a novel idea for improving the accuracy of the stiffness evaluation with a μ-channel. As far as we know that this is the first work of eliminating viscosity effect for the cell stiffness evaluation.

MODELING AND KEY IDEA

A spring is used for modeling the cell in the conventional method, while a spring and a damper are used for modeling the cell in the proposed model, as shown Figure 1(a). The spring and damper represent the stiffness and internal viscosity of a cell, respectively. The cell is pushed through the μ-channel by the pressure difference between two sides of the channel, and it deforms in order to squeeze into the channel. We defined Phase I as the duration while the spring and damper both react to the deformation, as left two cells illustrated in the lower diagram of Figure 1(a). After the cell fully entered the channel and reached to an equilibrium state, the shape of the cell becomes constant, as well as the velocity. Hence, only the spring is still responding to the deformation, and we defined this duration as Phase II, as right two cells illustrated in the lower diagram of Figure 1(a). According to this idea, the motion of the cell inside a μ-channel can be separated into two phases based on the motion profile. Phase I is the region where the motion profile is nonlinear, as the red curve in Figure 1(b). Phase II is the region where the motion profile is linear, as the green line in Figure 1(b).

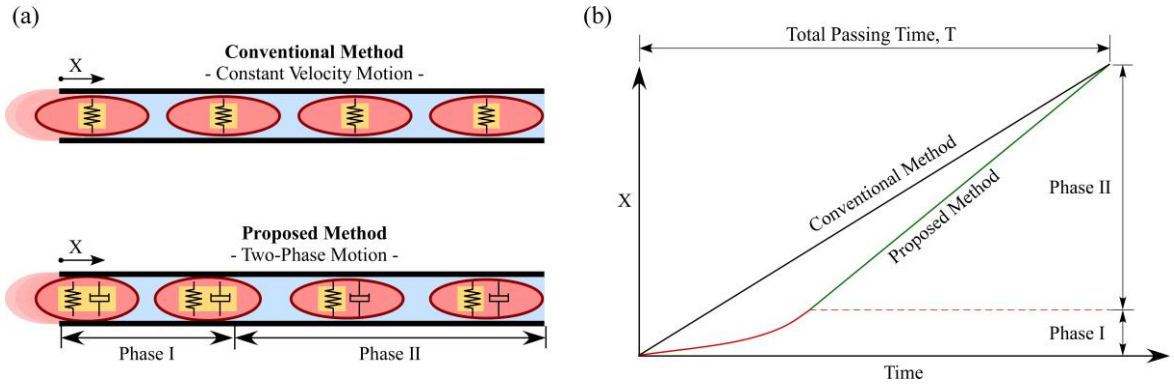

Figure 1: Comparison between the conventional and proposed μ-channel methods. (a) The cell is modeled with a spring in conventional method, but is modeled with a spring and a damper. Phase I is defined as both the spring and damper reacting to the deformation, while only the spring is in effect in Phase II. (b) Comparison between conventional methods and the proposed method.

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In conventional μ-channel method, it is assumed the cell reaches equilibrium immediately, and moves in a constant velocity through the channel, as the black line illustrated in Figure 1(b). From the comparison in Figure 1(b), we can clearly see the difference of equilibrium velocities between the conventional method (black) and the proposed method (green). We believe that the difference is the reason for the inaccuracy of the conventional μ-channel method.

EXPERIMENT

The experimental setup is as shown in Figure 2(a). A microscope is equipped with a high speed vision system. The spatial resolution of captured image is 0.25 μm, and the temporal resolution is 1 ms. The PDMS μ-chips are fabricated with μ-channels inside, and the dimensions of the μ-channels are shown in Figure 2(b). Figure 2(c) shows a series of images of a red blood cell (RBC) passing through a channel from the experiment.

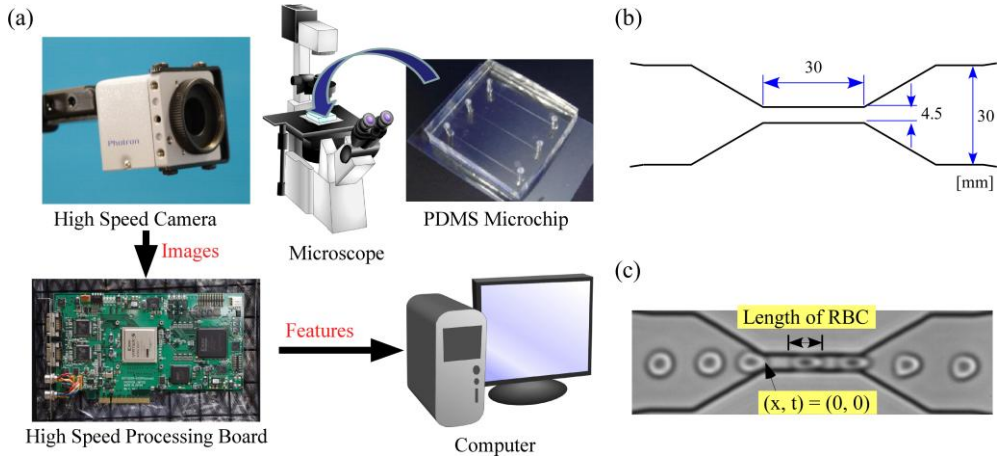

Figure2: (a) The experimental system; (b) The dimensions of the μ-channel; (c) A series of images of a RBC passing through the μ-channel

RESULTS AND DISCUSSION

The motion profiles of tested RBCs in the experiment are tracked by the computer and are plotted in Figure 3(a). We separate each motion profile into two segments, a curve and a straight line, and plot all the analysis results in Figure 3(b). In Figure 3(b), the red marks represent the data points in Phase I, while the green marks represent the data points in Phase II.

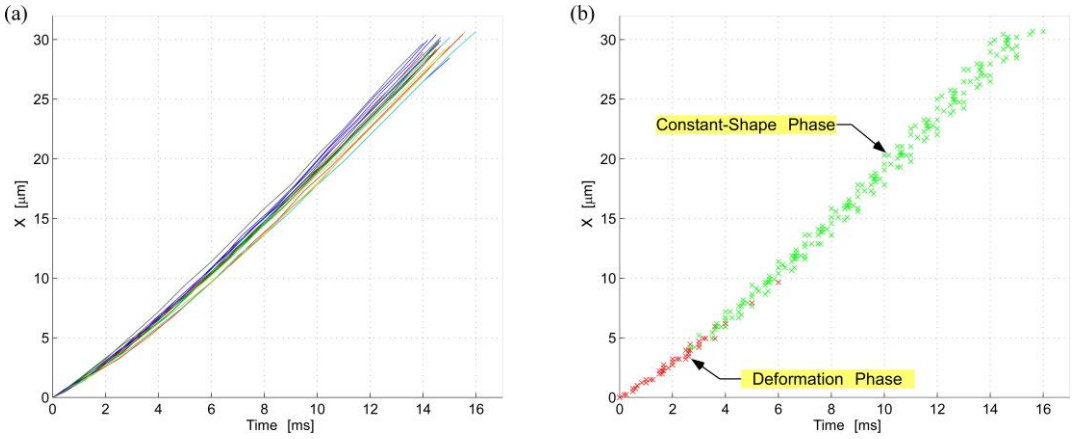

Figure 3: (a) The experimental results of RBCs passing through a μ-channel; (b) The motion profiles are separated into Phase I (red marks) and Phase II (green marks).

Since the RBCs are from the same subject, we assume the cell stiffness are similar. Therefore, the equilibrium velocity would be smaller with a bigger-size cell because a greater resisting force caused by greater amount of deformation. Figure 4 shows the comparison of size effect between the conventional method and the proposed method. The y-axis indicates the time needed for the cell to move 1 μm, while the x-axis is the length of the compressed cell inside the μ-channel. Figures 4(a) and 4(b) are the analysis results from the same set of experimental results by difference evaluation methods.

Figure 4(a) shows the analysis results by the conventional method, which assumes the cell moves in a constant velocity throughout the μ-channel. Figure 4(b) shows the analysis results processed by the proposed method which only focuses on the part where the motion profile is linear. We find that the correlation by using the proposed model (r = 0.66) is greater than it by the conventional model (r = 0.38). It proves that the newly proposed method is more sensitive to the cell size due to the elimination of viscosity effect.

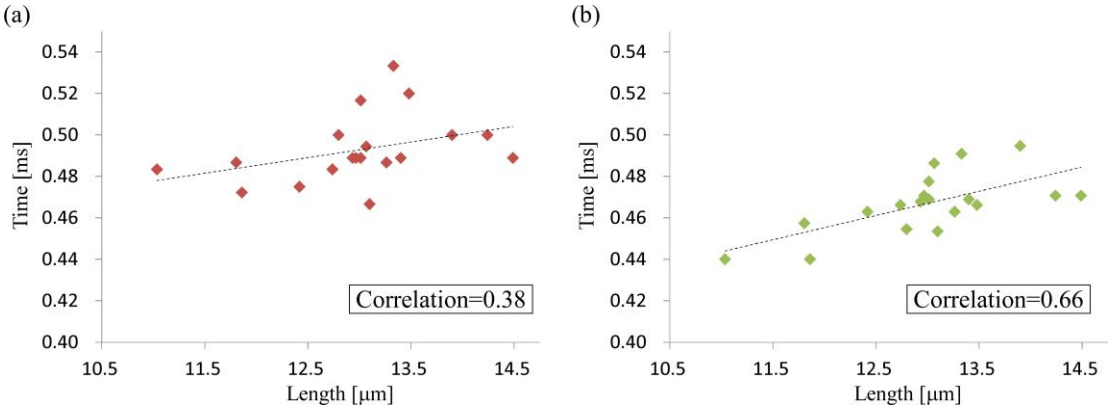

Figure 4: The plots of analysis results in the chart of passing time per μm v.s. the length of RBCs. (a) The analysis results by the conventional method; (b) the analysis results by the proposed method.

CONCLUSION

In conclusion, an enhanced method for stiffness evaluation is proposed in this paper. Two phases of cell motion inside a μ-channel is found with significantly physical meaning. The analysis results support that the proposed method is more sensitive to the effect of cell size, that is, the stiffness evaluation by the proposed method is more reliable than the conventional one.

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REFERENCES:

1. G. Y. H. Lee and C. T. Lim, *Biomechanics approaches to studying human diseases*, TRENDS in Biotechnology, 25(3):112-118, 2007
2. F. Glenister, R. Coppel, A. Cowman, N. Mohandas and B. Cooke, *Contribution of parasite proteins to altered mechanical properties of malaria-infected red blood cells*, Blood, 99(3): 1060-1063, 2002
3. Y. C. Fung, *Biomechanics: Mechanical Properties of Living Tissues*, Springer-Verlag, 1993.
4. Y. Hirose, K. Tadakuma, M. Higashimori, T. Arai, M. Kaneko, R. Iitsuka, Y. Yamanishi and F. Arai, *A new stiffness evaluation toward high speed cell sorter*, Proc. of the IEEE Int. Conf. on Robotics and Automation (ICRA): 4113–4118, Anchorage, USA, May 2010.
5. W. R. Tricky, G. M. Lee and F. Guilak, *Viscoelastic properties of chondrocytes from normal and osteoarthritic human cartilage*, J. Orthop. Res. 18(6), 891-898, 2000
6. M. M. Brandão, A. Fontes, M. L. Barjas-Castro, L. C. Barbosa, F. F. Costa, C. L. Cesar and S. T. O. Saad, *Optical tweezers for measuring red blood cell elasticity: application to the study of drug response in sickle cell disease*, Eur. J. Hematol. 70, 207-211, 2003
7. G. Binnig, C. F. Quate, and C. Gerber. *Atomic force microscope*, Physical Review Letters, 56(9):930–933, 1986.

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