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Theme 4: Cell Mechanics

Flow of physiological fluids in microchannels: the sedimentation effect

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Abstract— **Microfluidic devices are becoming one of the most promising new tools for diagnostic applications and treatment of several chronic diseases. Hence, it is increasingly important to investigate the rheological behaviour of physiological fluids in microchannels. The main purpose of the present experimental work is to investigate the flow of two different physiological fluids frequently used in microfluidic devices. The working fluids were physiological saline (PS) and dextran 40 (Dx40) containing about 6% of sheep red blood cells (RBCs), respectively. The capillaries were placed horizontally on a slide glass and the flow rate of the working fluids was kept constant by using a syringe pump. By means of a camera the images were taken and transferred to the computer to be analysed. Generally, the results show that PS and Dx40 have different flow behaviour due to the sedimentation of the RBCs.**

Keywords— **Physiological fluids, Red blood cells, microfluidic devices, sedimentation, microcirculation.**

I. Introduction

In microcirculation there are two well known phenomena, i. e., the Faharaeus effect and Faharaeus-Lindqvist effect. These two effects are strongly related to the microtube diameter and other physical and hemorheological factors. An explanation for the Faharaeus-Lindqvist effect is the decrease of the dynamic hematocrit (Hct) with decreasing microtube diameter. This effect can be explained by the formation of a cell-free layer (CFL) near the wall of the microtube. This CFL located between the RBC core and wall contributes to the reduction of apparent blood viscosity. However, the complex formation of the CFL has not yet convincingly demonstrated mainly due to multi-physical and hemorheological factors that affect CFL [1-3].

Currently, biomedical microdevices are becoming one of the most promising tools for the diagnostic and treatment of several diseases, such as diabetes, malaria and cancer. Hence, it is increasingly important to investigate the rheological behaviour of physiological fluids in microchannels in order to make use on the physics of microfluidics to either develop new lab-on-chip devices or to optimize the design of the existent microfluidic chips.

In this paper we investigated the flow behaviour of two different physiological fluids frequently used in biomedical microdevices. The working fluids used in this study were physiological saline (PS) and dextran 40 (Dx40) containing about 6% of sheep red blood cells (RBCs), respectively. By using a syringe pump and a camera it was possible to measure qualitatively flow behaviour within a horizontal capillary.

II. Materials & Methods

Blood sample preparation

Four working fluids were used in this study: physiological saline (PS) containing about 6% (6Hct) of sheep red blood cells (RBCs), dextran 40 (Dx40; Otsuka Medicine) containing about 6% (6Hct) of sheep RBCs, PS with 10% of dextran 40 (Dx40s10; Sigma aldrich, BioChemika)and PS containing 30% of dextran 40 (Dx40s30; Sigma aldrich, BioChemika) containing in both fluids about 2% (3Hct) of sheep RBCs.

The blood was collected from a healthy adult sheep, where heparin was added to prevent coagulation. The RBCs where separated from the bulk blood by centrifugation (3000 RPM for 15 min) and aspiration of the plasma and buffy coat and then washed twice with PS. The washed RBCs where diluted with PS to make up the required RBCs concentration by volume. The hematocrit (Hct) of the RBCs suspension sample was about 6% (6Hct). Note that all the blood samples were stored hermetical at 4ºC until the experiment was performed at room temperature (18 to 20ºC). Figure 1 shows the main steps for the preparation of the blood samples.

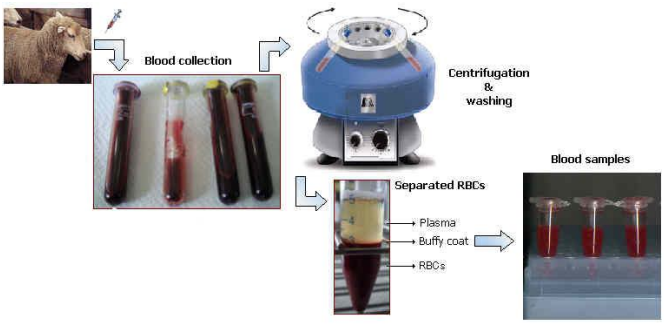

Fig. 1 Main steps for the preparation of the blood samples.

Experimental setup

To investigate the RBC sedimentation we have decided to use two kinds of experimental methods. In the first method the capillaries were placed horizontally on a slide glass and by using a syringe pump (New Era Pump Systems, USA) a pressure-driven flow was kept constant at 50 µl/min which corresponds to a Reynolds ~0.9(PS) ~0.3(Dx40) (see Fig.2). For the second method we decided to fill 4 tubes with different kinds of physiological fluids containing sheep RBCs, i.e., PS, Dx40, Dx40s10, Dx40s30 (see Fig.3). Additionally the visualization of the flow in microchannels was possible by means of a high-speed video microscopy system (see Fig.4). Detailed information about this latter microvisualization system can be found elsewhere [4-6].

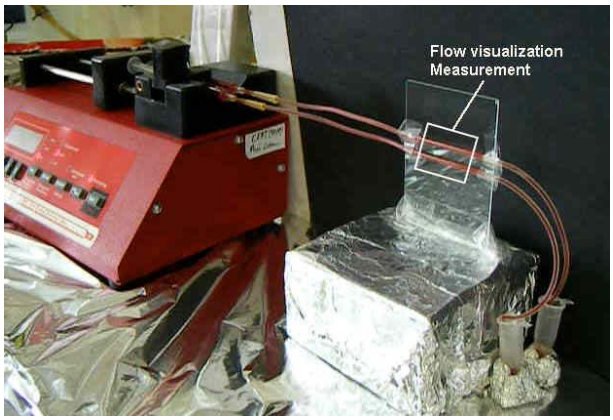

Fig. 2 Experimental setup for the dynamic sedimentation measurements.

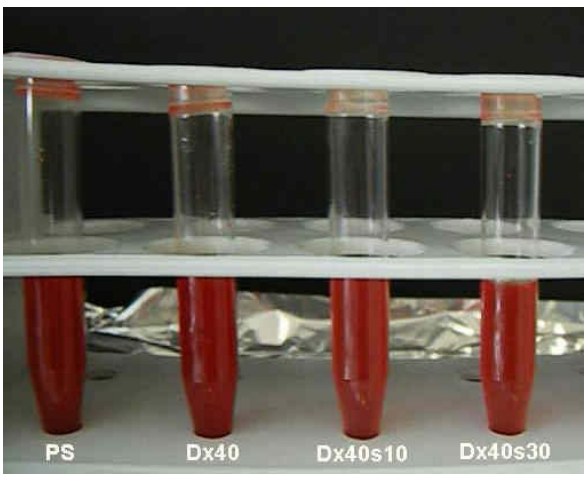

Fig. 3 Experimental setup for the static sedimentation measurements.

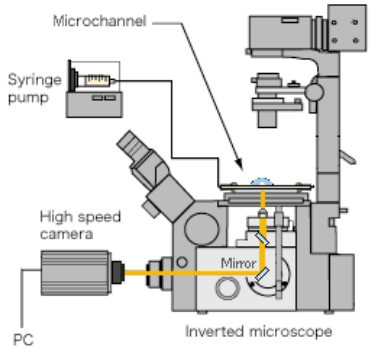

Fig. 4 Experimental setup for the visualization of the in vitro blood flow in glass microchannels (adapted from 4, 6).

III. Results & Discussion

To analyse the dynamic sedimentation of PS and Dx40 containing RBCs we decided to use flow rates close to the one observed in vivo, i.e., 10 µl/min. During the experiment we made flow qualitative visualizations measurements in glass capillaries with diameters of about 1.2 mm. The visualizations were captured by a camera for about 15 minutes. Figure 5 shows the flow qualitative measurements for 0 minutes and 15 minutes. This image shows clearly that for a period of 15 minutes the RBC tend to settle down in the fluid with PS whereas using Dx40 we did not observe any RBC sedimentation. Although not shown in Figure 5, for the case of PS fluid we did not observe any RBC sedimentation for the first 10 minutes. According to our visualization the RBCs tend to settle down for period of time superior to 10 minutes.

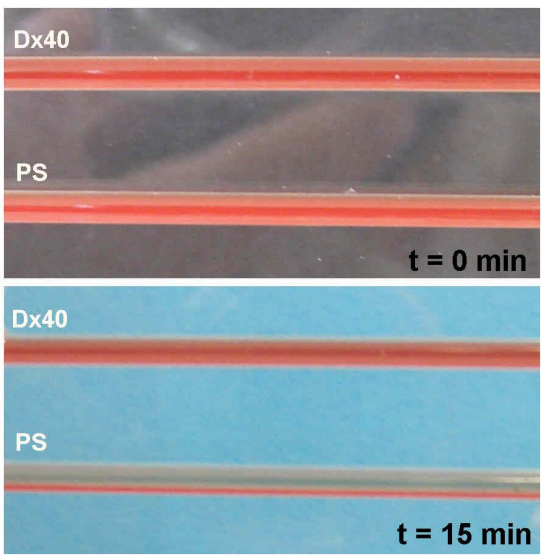

Fig. 5 Dynamic sedimentation measurements for two different time periods of PS and Dx40 containing RBCs (Flow rate = 50 µl/min) .

To study the static sedimentation of the sheep RBCs in PS and Dx40 we decided to fill 4 tubes containing identical fluids to the ones used in the previous experiments and also additional fluids containing PS with 10% of Dextran 40 (Dx40s10) and PS with 30% of Dextran 40 (Dx40s30). The visualizations were captured by a camera for a period of about 2 hours. Figure 6 shows a summary of the most relevant visualization results. This image shows that for a period of 30 minutes the RBC tend to settle down in the fluid with PS. The sedimentation tends to increase with the time. For a period of 1 hour and 30 minutes it is very clear the interface between the high concentration of cells and the cell-free fluid. For a period superior to 1 hour little sedimentation was observed in the sample Dx40s10. For the case of Dx40 and Dx40s30 we did not observe any significant RBC sedimentation.

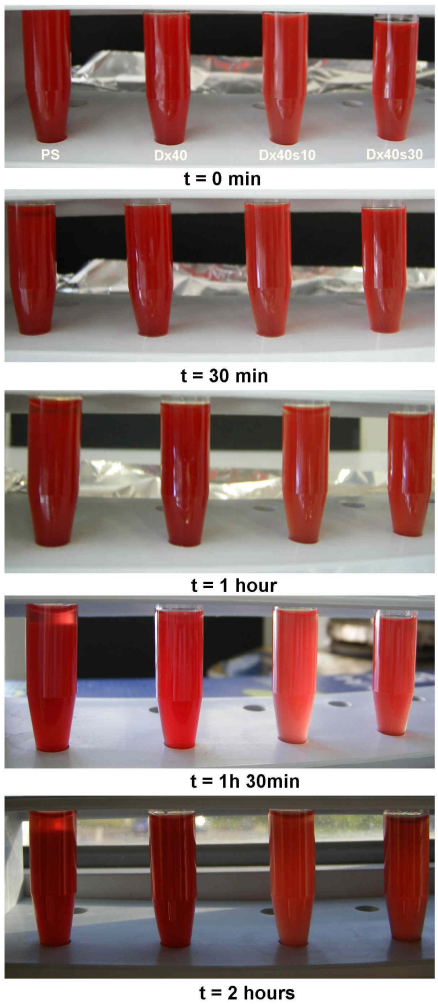

Fig. 6 Static sedimentation measurements for a period of 2 hours of PS, Dx40, Dx40s10 and Dx40s10 containing RBCs .

Additionally flow visualization measurements were also performed in glass microchannels (see Fig.7) and compared with *in vivo* blood flow (see Fig.8). Figure 7 shows that for the case of Dx40 there is a clear formation of cell-free layer adjacent to the walls of microchannels. However, in the fluid with PS the RBCs do not exhibit a clear tendency to migrate into the microtube axis. The *in vivo* visualization measurements (Fig. 8) have shown a clear tendency for the formation of a plasma layer in microvessels [6, 7]. These results indicate that *in vitro* blood containing Dx40 has a flow behaviour closer to the one observed in vivo microvessels.

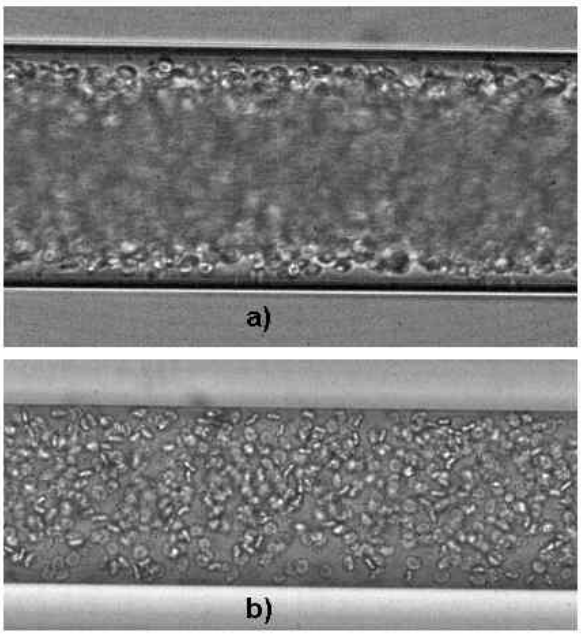

Fig. 7 *In vitro* flow visualization in glass microchannels for a period time bigger than 10 minutes a) Dx40 containing RBCs; b) PS containing RBCs.

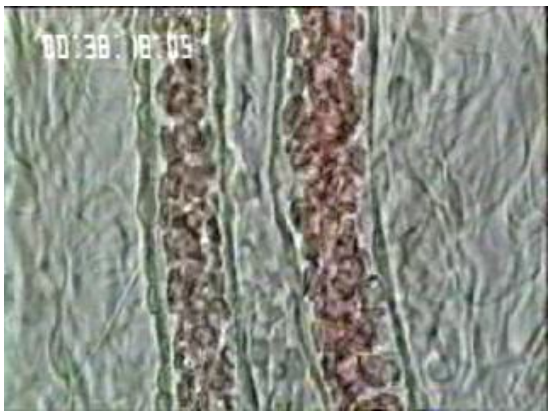

Fig. 8. I*n vivo* flow visualization in a microvessel. [7].

IV. Conclusion & future directions

The flow behaviour of two different physiological fluids frequently used in biomedical microdevices was investigated. Our preliminary results indicate that *in vitro* blood containing Dx40 has a flow behaviour closer to the one observed *in vivo* microvesseles. The *in vitro* blood containing PS did not show a clear formation of cell-free layer which might be due to the fast sedimentation of the RBCs. In the near future we plan to vary the flow rate and diameter to study the influence of these effects on the RBC sedimentation.

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References

1. Lima R, Ishikawa T, et al. (2010) Blood flow behavior in microchannels: advances and future trends. Single and two-Phase Flows on Chemical and Biomedical Engineering. Bentham (in press).
2. Goldsmith H, Turitto V (1986) Rheological aspects of thrombosis and haemostasis: basic principles and applications. ICTH-Report-Subcommittee on Rheology of the International Committee on Thrombosis and Haemostasis. Thromb Haemost. 55(3): 415–435
3. Maeda N (1996) Erythrocyte rheology in microcirculation. Japanese Journal of Physiology 46, 1-14
4. Lima R, Wada S, Tsubota K, Yamaguchi T (2006) Confocal micro-PIV measurements of three dimensional profiles of cell suspension flow in a square microchannel. Measurement Science and Technology 17: 797-808
5. Lima R, et al. (2009) Measurement of individual red blood cell motions under high hematocrit conditions using a confocal micro-PTV system, Annals of Biomedical Engineering, 37, 1546-1559
6. Kim S, Kong, R L, et al (2006) A computer-based method for determination of the cell-free layer with in microcirculation. Microcirculation 13: 199-207.
7. Minamiyama M (2000) In Vivo Microcirculatory Studies at http://www.ne.jp/asahi/minamiya/medicine/

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