DISPERSION OF RED BLOOD CELLS IN MICROCHANNELS: A CONFOCAL MICRO-PTV ASSESSMENT

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1. Introduction

Blood in large arteries may be treated as a homogenous fluid from a macroscopic prospective. However, in reality blood is a suspension of deformable cells in viscous fluid plasma. In microcirculation, which comprises the smallest arteries and veins, the flow behavior of individual blood cells and their interactions provide the microrheological basis of flow properties of blood at a macroscopic level. Hence, in microcirculation it is fundamental to study the flow behavior of blood at cellular level. Several studies on both individual and concentrated RBCs have already been performed in the past [1, 2]. However, all studies used conventional microscopes and also ghost cells to obtain visible trace RBCs through the microchannel. The present study is concerned in providing further insights into the microscale blood flow behavior through microchannels by applying an emerging optical technique known as confocal micro-PIV/PTV [3, 4, 5]. The technique consists of a spinning disk confocal microscope, high speed camera and a diode-pumped solid state (DPSS) laser combined with a single particle tracking (SPT) software (MtrackJ). Detailed measurements on the motions of RBCs were measured at different haematocrits (Hct) and the correspondent radial dispersion coefficient was determined.

2. Materials and methods

2.1. Working fluids and microchannel

Three working fluids were used in this study: dextran 40 (Dx40) containing about 3% (3Hct) 15% (15Hct) and 35% (35Hct) of red blood cells (RBCs). The RBCs were collected from a healthy adult volunteer, centrifuged and then washed with physiological saline (PS). The washed RBCs were labeled with a fluorescent cell tracker (CM-Dil, C-7000, Molecular Probes) and then diluted with Dx40 to make up the required RBCs concentration by volume. All blood samples were stored hermetical at 4ºC until the experiment was performed at controlled temperature of about 37ºC.

2.2. Confocal micro-PTV experimental set-up

The confocal micro-PTV system used in our experiment consists of an inverted microscope (IX71, Olympus) combined with a confocal scanning unit (CSU22, Yokogawa), a diode-pumped solid state (DPSS) laser (Laser Quantum Ltd) and a high-speed camera (Phantom v7.1) was connected into the outlet port of the CSU22. The microchannel was placed on the stage of the inverted microscope where the flow rate of the working fluids was kept constant by means of a syringe pump (KD Scientific Inc.). A thermo plate controller (Tokai Hit) was set to 37ºC. All the confocal images were captured with a resolution of 640×480 pixels, at a rate of 100 frames/s with an exposure time of 9.4 ms. The recorded images were transfered to the computer and then evaluated in the Image J (NIH) [6] by using the manual tracking MTrackJ [7] plugin. As a result it was possible to track single RBCs through the middle plane of the microchannel. Deatailed information about the experimental set-up, used in the present study, has already been described previously [3].

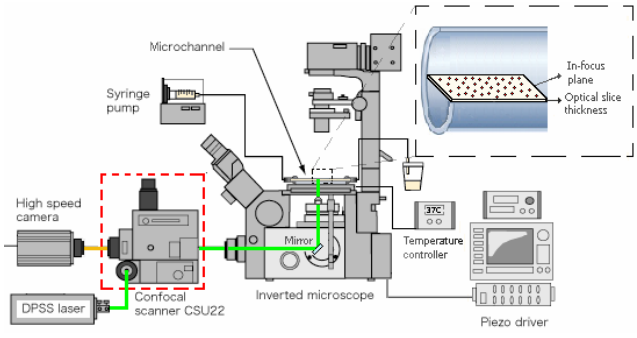


Figure 1. Experimental set-up.

2.3. RBC dispersion coefficient

A RBC radial dispersion coefficient (D_{yy}) to quantify the radial displacement of the tracer RBCs was defined as :

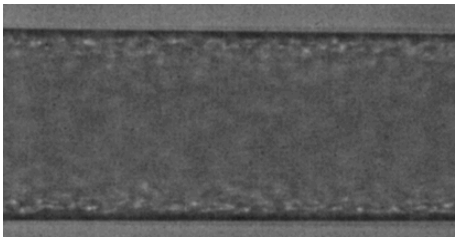
$$D_{yy}(t) = \frac{1}{N}\sum_{i=1}^{N} \frac{\left\langle (R_{i,y}(t) - R_{i,y}(0))^2 \right\rangle}{2t} \qquad (1)$$

where $\langle (R_{i,y}(t)-R_{i,y}(0))^2 \rangle$, t and N are the mean square displacement, time interval and number of measured RBCs respectively.

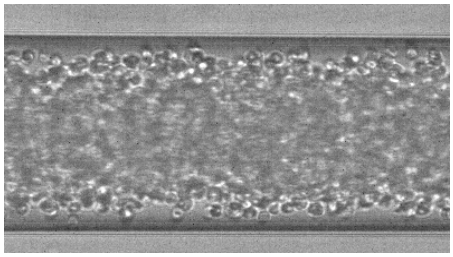
3. Results and discussion

3.1. Tracking displacement of RBCs at different Hcts

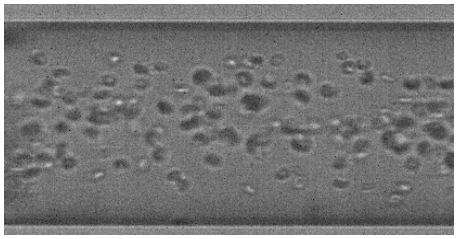
By using our confocal system we were able to track single RBCs in the middle plane with Hct up to 35% (see Fig.2). Furthermore it also is possible to observe that at 3% Hct the RBC paths are almost parallel to the flow direction without any appreciable fluctuations on the radial direction. By contrast, at 15% and 35% Hct the RBC paths exhibit erratic radial displacements due the high-concentration of RBCs on the adjacent streamlines.



a) 35Hct



b) 15 Hct



c) 3Hct

Figure 2. Halogen images in the middle plane of a 100μm glass capillary with (a) 35% Hct, (b) 15% Hct, (c) 3% Hct.

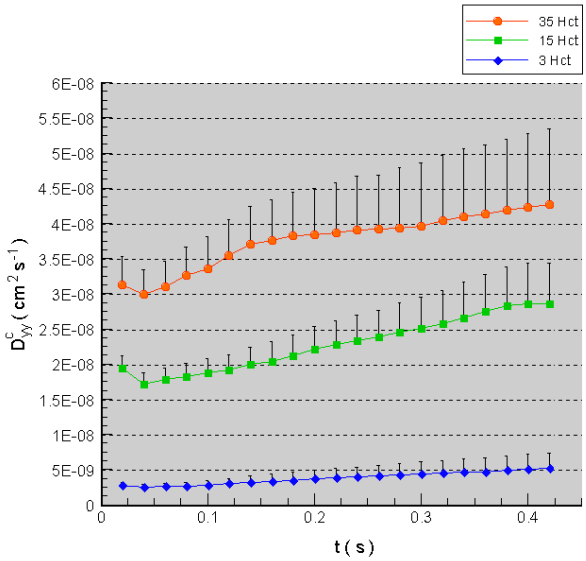


Figure 3. RBC radial dispersion coefficient (D_{yy}) at the centre plane for several Hcts (Re ~ 0.005).

Figure 3 shows clearly that RBC radial dispersion coefficient (D_{yy}) are strongly dependent on the Hct as they tend to increase with it.

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