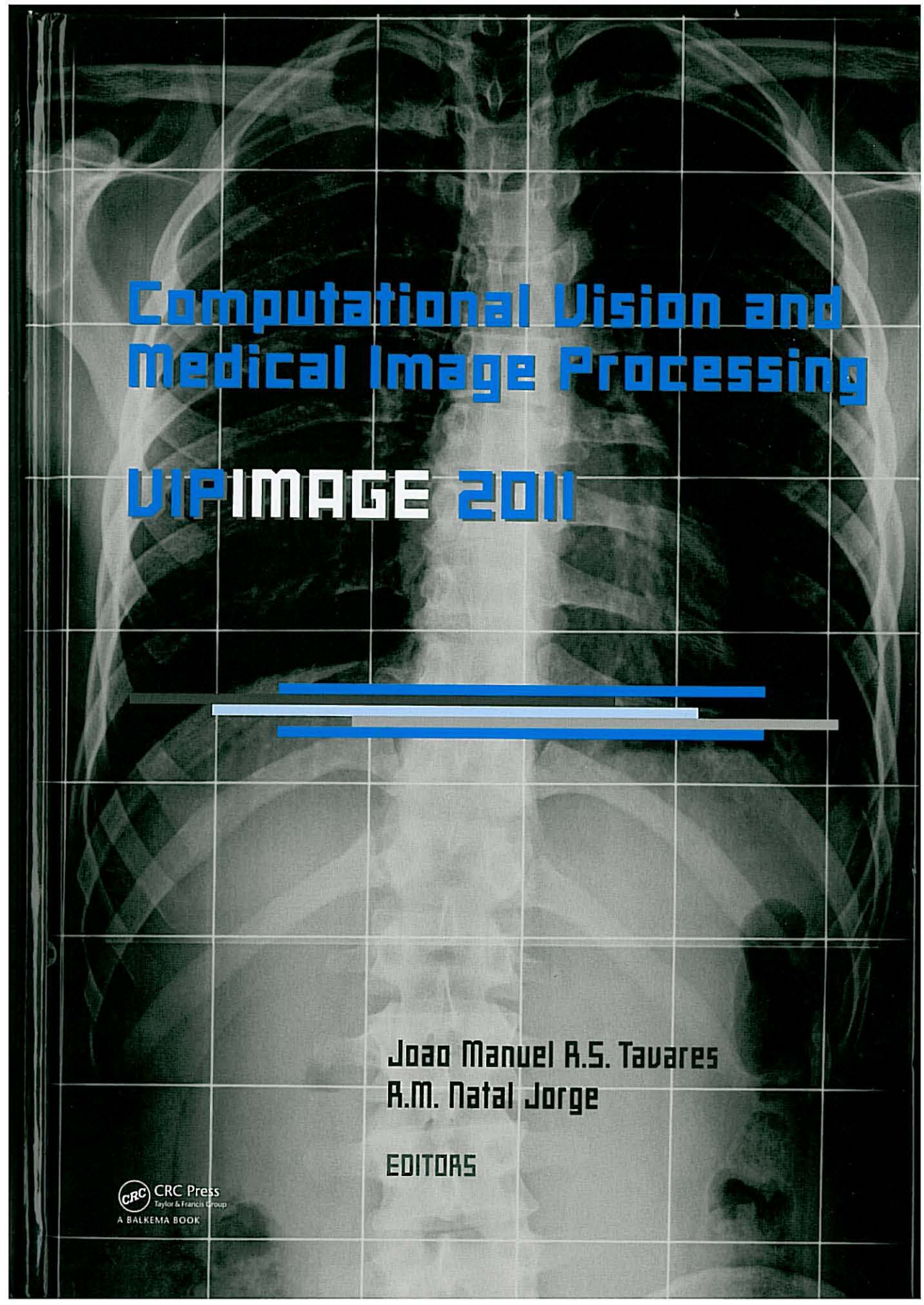

Computational Vision and Medical Image Processing

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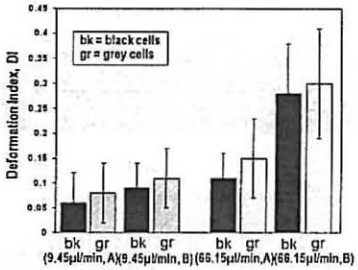

Figure 4. Comparison of deformation index at different flow rates in different regions.

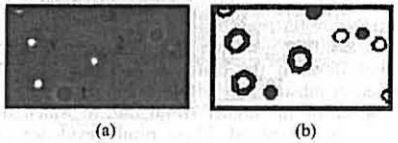

Figure 5. (a) Original image containing RBCs with various intensities: 1. low (black), 2. intermediate (grey) and 3. high (white), and (b) Corresponding binary image.

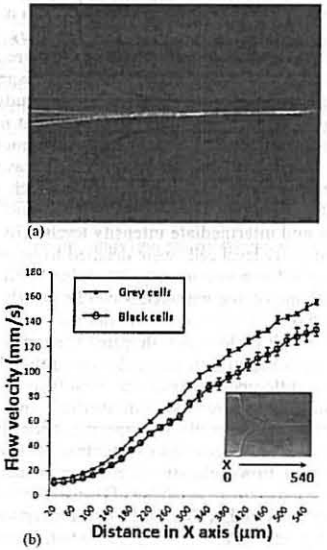

Figure 6. (a) Tracking of RBCs with different intensity levels for velocity measurements. (b) Axial velocity profiles of the low and intermediate intensity RBCs along the centerline at $Q = 9.45$ µl/min.

volume illumination, in which the depth-of-focus is determined by the characteristics of objective used. As a consequence, despite being centered at the mid-plane of the channel where the cell velocity is the highest, cells at different z-planes are also captured. In this case, we believe that the black cells are not truly located at the mid-plane and therefore its velocity is slightly lower than that of the grey cells. Following these preliminary results, further investigation on the cell velocities and deformation index in various regions of the microchannels under different flow conditions will be performed.

ACKNOWLEDGEMENTS

We thank Dr. Matsuki for help with blood sample collection. Additionally, we acknowledge the financial support provided by 2007 GlobalCOE Program "Global Nano-BME Education and Research Network", Japan. We are also thankful to FCT (Portugal) and COMPETE for financial support through projects PTDC/SAU-BEB/108728/2008, PTDC/SAU-BEB/105650/2008 and PTDC/EME-MFE/099109/2008.

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An automatic method to track Red Blood Cells in microchannels

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ABSTRACT: Image analysis is extremely important to obtain crucial information about the blood phenomena in microcirculation. The current study proposes an automatic method for segmentation and tracking Red Blood Cells (RBCs) flowing through a 100 µm Glass capillary. The original images were obtained by means of a confocal system and then processed in Matlab using the Image Processing Toolbox. The automatic measurements obtained with the proposed automatic method are compared with a manual tracking method using a plugin from ImageJ.

1 INTRODUCTION

The study of the red blood cells (RBCs) flowing in microvessels and microchannels is very important to provide a better understanding on the blood rheological properties and disorders in microvessels [1–5]. In this kind of study, the image analysis is an essential part to obtain crucial information about the blood rheology. However, most of the data analysis procedures have been executed manually [1–3] which is an extremely time consuming task especially with a large amount of data. Additionally, manual tracking methods can also introduce user errors into the data. Hence, it is important to develop image analysis methods able to get the data automatically. The main purpose of this work is to develop an approach able to track the RBCs with x and y coordinates automatically. To accomplish it we tested filtering, segmentation and feature extraction functions available in MatLab.

2 MATERIALS AND METHODS

2.1 *Experimental set-up*

The confocal micro-PIV system used in this study consists of an inverted microscope (IX71; Olympus) combined with a Confocal Scanning Unit (CSU22; Yokogawa), a Diode-Pumped Solid-State (DPSS) laser (Laser Quantum) with an excitation wavelength of 532 nm and a high-speed camera (Phantom v7.1; Vision Research) (Fig. 1). The glass capillary was placed on the stage of the inverted microscope and by using a syringe pump (KD Scientific) a pressure-driven flow was kept constant (Re ~ 0.008).

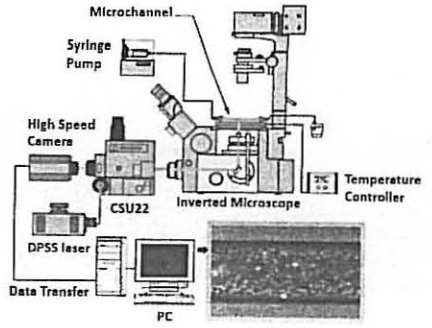

Figure 1. Experimental set-up.

More detailed information about this system can be found elsewhere [1].

2.2 *Image analysis*

The laser beam was illuminated from below the microscope stage through a dry 40× objective lens with a Numerical Aperture (NA) equal to 0.9. The confocal images were captured in middle of the capillary with a resolution of 640 × 480 pixel at a rate of 100 frames/s with an exposure time of 9.4 ms. Two image analyses methods were used in this study: method 1 (manual approach) and method 2 (automatic approach).

2.2.1 *Method 1*

A manual tracking plugin (MTrackJ) [6] of an image analysis software (ImageJ, NIH) [7] was used to track individual RBC. By using MTrackJ plugin, the bright centroid of the selected RBC was automatically computed through successive images for an interval of time of 10 ms. After obtaining x and y positions, the data were exported for the determination of each individual RBC trajectory.

2.2.2 *Method 2*

All frames were loaded and pre-processed using Matlab [8]. The region of interest was then cropped from the images with the function *imcrop*. The median function, *medfilt2*, with one mask 5 × 5 pixel, was applied to eliminate most of the noise and to enhance the flowing object. In Fig. 2 we can see the result of these processing steps. In the next step, the images are subject to a segmentation filter, *Sobel*. With this segmentation it is possible to separate RBCs from the background, i.e. differentiate the area of interest (the RBCs) from the not-interest area (background image). This is possible using a *threshold* method, where a definition of one or more values of separation is enough to divide the image into one or more regions. The function *iterative threshold* was applied for the sequence of all the images.

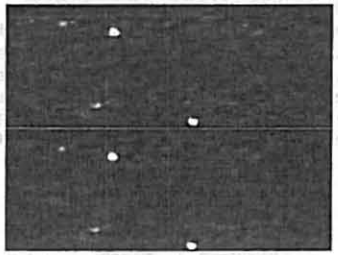

Figure 2. The region of interest (above) and the image filtered by using the median function *medfilt2*.

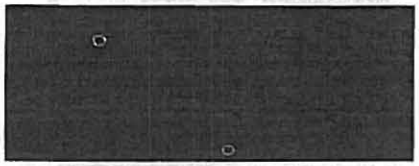

Figure 3. Result of the iterative *threshold* method and the filter *Sobel*.

The objects are defined with the *Sobel* filter (see Fig. 3), which shows only the edge of the objects. The *Sobel* computes an approximation of the gradient of the image intensity. At each pixel point in the image, the result of the *Sobel* operator is either the corresponding gradient vector or the norm of this vector.

3 RESULTS AND DISCUSSION

After the segmentation processing, the RBCs were tracked and sets of data (x and y positions) were obtained with the Matlab function (Fig. 4), stored in the image processing toolbox, *regionprops*. This function measures a set of properties (area, centroid, etc.) for each connected component (RBC) in the binary image.

In the Fig. 5 we can see the tracking of two RBCs, in a sequence of successive images, with an interval of 4 frames.

All of these image processes, presented in this work, are placed in an application, *RBC Data Tracking*, built in MatLab, in which all the steps can be done automatically.

Fig. 6 shows a qualitative comparison between method 1 (manual) and method 2 (automatic). The trajectories obtained from the proposed automatic method looks more smooth when compared with manual method.

Some deviations are observed between both methods. This may be due to the inaccuracy in manual tracking, especially for determination of the center of the RBCs, because the automatic method is more sensitive, even in the presence of small changes in the centroid.

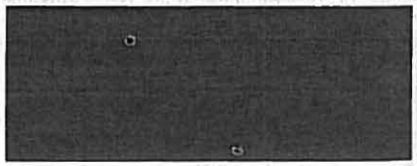

Figure 4. RBCs tracking and data extraction.

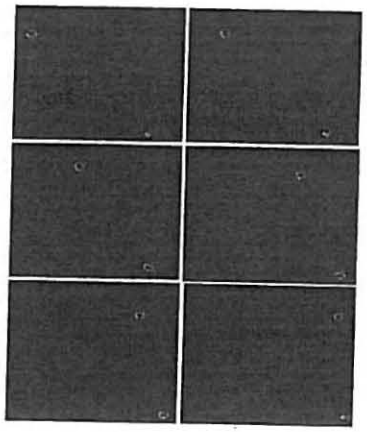

Figure 5. RBCs tracking and data extraction in a sequence of 4 to 4 frames.

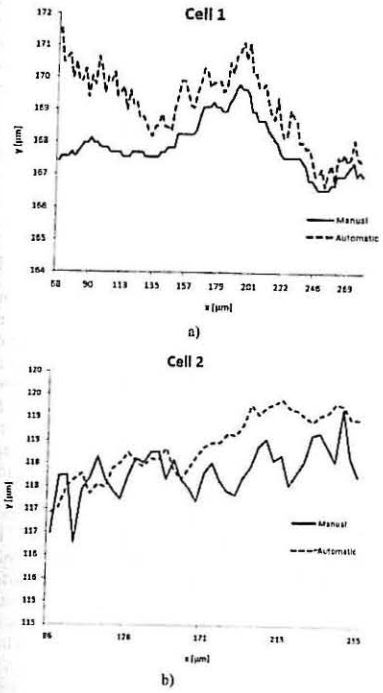

Figure 6. Comparison of the manual (a) and automatic (b) methods.

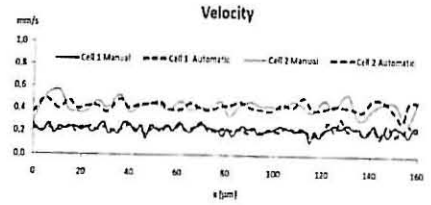

Figure 7. Velocity of two cells by using both methods.

Fig. 7 shows the velocity of cell 1 and cell 2 calculated by data obtained from both methods. The results show good agreement between the two methods.

4 CONCLUSIONS

Although the automatic method presented in this study is a promising way to track the flowing RBCs, additional image analysis needs to be performed. Hence, detailed quantitative measurements of the RBC trajectories are currently under way and will be presented in due time.

In future work we are planning to explore more techniques to obtain quantitative measurements of the RBC trajectories, and more image analysis strategies need to be performed.

ACKNOWLEDGEMENTS

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Speech articulation assessment using dynamic Magnetic Resonance Imaging techniques

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ABSTRACT: Magnetic Resonance Imaging (MRI) has been successfully applied on real-time analysis of the articulators during speech production along the whole vocal tract, with good signal-to-noise ratio and without ionizing effects. Because speech dynamic events need a minimal sampling rate, an improvement on the temporal resolution of MRI systems is demanded. Our aim is to describe a dynamic MRI technique to acquire and assess the main articulatory events during the production of some European Portuguese utterances. Hence, novel perceptions for dynamic MRI technique using a 3.0 Tesla System are presented in order to study the shape of the vocal tract during speech production.

Keywords: image analysis, medical imaging, speech production, dynamic techniques

1 INTRODUCTION

1.1 *Speech production analysis and challenges*

The speech production mechanism is a complex human motor activity that is able to achieve voice modulation and produce speech based mainly in the articulators' movements. The organs involved, mostly formed of soft tissues, such as the tongue, the lips, the velum and the pharynx, assume extremely important roles during speech production. In fact, these organs together with some bones, i.e. the palate and the jaw, modify the resonance cavities and the shape of the vocal tract in order to produce the sounds.

The human vocal tract's shape (Fig. 1) is different among subjects and presents a non-regular contour defined by the air-soft tissues' boundaries. This tube extends from the lips to the glottis, and is formed by four main structures: the oral cavity, the nasal cavity, the velum and the pharynx.

The tongue is the most important articulator, mainly because it is the largest one, and performs a wide range of slow and fast movements during speech production.

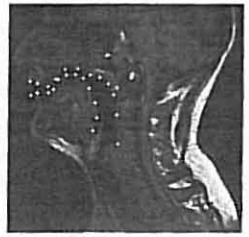

Figure 1. The shape of the vocal tract during the production of [ɛ] vowel in an image acquired by a 3.0 Tesla MR system.

Many approaches have been used to track and observe the movements of the articulators, in particular of the tongue, but most of them employ sensors (e.g. electromagnetic articulography) or the direct contact with the tongue and the palate (e.g. electropalatography).

Magnetic Resonance Imaging (MRI) has been successfully applied on real-time analysis of the