

# Evaluation of blood microcirculation parameters by combined use of laser Doppler flowmetry and videocapillaroscopy methods

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## ABSTRACT

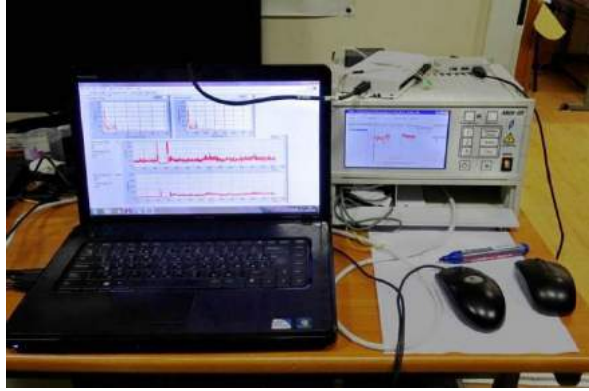
Laser Doppler flowmetry (LDF) is widely used for diagnosing blood microcirculation diseases. It is well known that the Doppler shift of laser radiation scattered by moving red blood cells (RBC) can be assessed through analyzing photocurrent produced by a photodetector. LDF signal contains information about regulating blood flow rhythms: myogenic, cardiac, nervous and endothelial. The method of videocapillaroscopy (VCS) allows local capillary blood flow velocity evaluation and, using video data processing algorithms, is able to assess RBC velocity changes into capillary. We present the results of simultaneous investigations of changes in tissue perfusion of the distal phalanx of human finger by the LDF as well as changes in capillary blood flow velocity in the nail bed evaluated by the VCS method during arterial occlusion test. The experimental results confirmed the correspondence between blood perfusion and blood flow velocity.

**Keywords:** videocapillaroscopy, laser Doppler flowmetry, capillary blood flow, microcirculation.

## 1. INTRODUCTION

Non-invasive laser Doppler flowmetry (LDF)<sup>1,2</sup> and video capillaroscopy (VCS)<sup>3,4</sup> are well-known methods to study the blood microcirculation parameters for early diagnosis of various diseases and to monitor the effectiveness of therapeutic impacts<sup>5</sup>. The LDF method (Fig. 1a) allows to assess the dynamics of a tissue structure perfusion by the index of microcirculation. This is an integrated estimate characterizing general functional state of a tissue, that is a complex disordered biological structure<sup>6</sup>. Spectral analysis of the microcirculation index fluctuations allows to extract information about modulation of peripheral blood flow with high speed within the frequency of cardiac rhythm (~1-1.5 Hz), respiratory rhythm (~ 0.4 Hz), and rhythm within the frequency range <0.2 Hz<sup>7,8</sup>. This frequency range is associated with endothelial, myogenic and neurogenic vascular tone. The effect of pulsatile transmural pressure of the arteries compresses/decompresses the density of capillaries in the dermis<sup>9</sup>, thus modulating the blood volume in the capillary bed.

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(a)



(b)

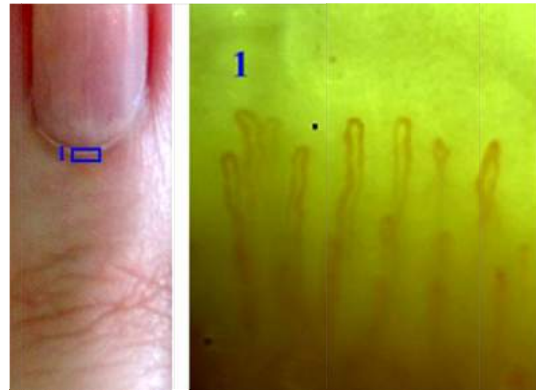
Figure 1. (a) Laser Doppler flowmetry setup based on “LAKK-OP” device and custom electronic board; (b) LDF-probe placed at middle finger of hand.

The VCS method relates to digital microscopy technique. It is based on registration and analysis of a video frames series representing capillary blood flow of the nail bed (Fig. 2b). The method requires significant computational power, but it allows extracting the parameters of capillary blood flow of an individual capillary.

This method can be used for verification of data obtained by the LDF-devices, as well as for interpretation of identified features of LDF-grams. Thus, the aim of this research is simultaneously investigate the dynamics index of microcirculation of finger distal phalanx and capillary blood flow using LDF and VCS methods, respectively.



(a)



(b)

Figure 2. (a) Videocapillaroscopy setup; (b) area of interest at nailfold of finger.

## 2. MATERIALS AND METHODS

Index of microcirculation was first obtained by the LDF method. As a laser source, one-mode laser with 1064 nm wavelength was utilized. Optical fibers were used to deliver radiation to the skin and to collect backscattered light. Si-

photodiodes were used to convert detected radiation into photocurrent. Then the signal was amplified by a custom electronic board. Analog-to-digital conversion was performed by the data acquisition board model NI USB 6211.

Study of nail bed capillary net was performed using the experimental VCS setup (Fig. 2a) for direct evaluation of blood velocity. Nail bed of left hand ring finger was illuminated by a LED. A microscope objective with 0.12 aperture and long-focus lens formed capillary image at a photo sensitive matrix. During the experiments registration of video data was being conducted by a high-speed camera with 135 fps. Thus, the method allows evaluating capillary blood flow within velocity range up to 5 mm/s. Stabilization and drift compensation of frame sequence were carried out by the advanced processing method with several reference frames<sup>10</sup>. Figures 3-5 illustrate main stages of processing of video information for stabilized sequence of frames. Result of capillary image background elimination is demonstrated in Fig. 3.

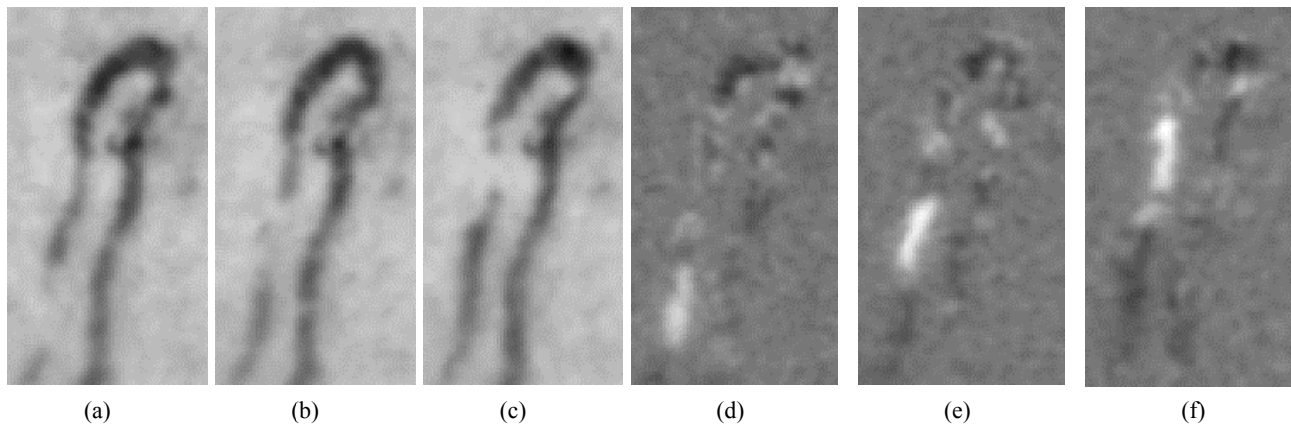


Figure 3. (a), (b), (c) Example of original frames set; (d), (e), (f) resulting frames after capillary image background removal.

Result of further processing is illustrated in Fig. 4. After background removal, calculation of average intensity through whole sequence of frames is accomplished (Fig. 4a). At the next stage, algorithm performs capillary image boundary detection (Fig. 4b). Onwards, blood flow velocity profile is calculated involving small 2D areas ( $dx, dy$ ) of neighbor video frames. The velocity estimates are illustrated by greyscale representation in Fig. 4c, d. Then capillary central line is extracted using information about changes of velocity profile (Fig. 4e). Finally, local diameter of the capillary is determined (Fig. 4f).

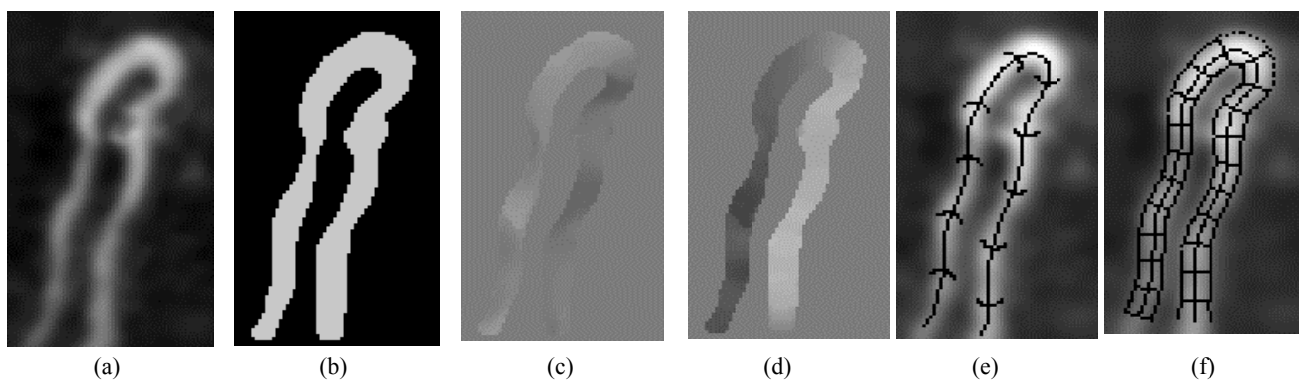


Figure 4. (a) Inverted average image for whole sequence of frames; (b) result of capillary image boundary detection; (c),(d) components of 2D-velocity distribution within the neighbour frames; (e) result of automatic evaluation of capillary central line; (f) evaluation of diameter of the capillary.

To evaluate blood flow velocity, a set of points along the capillary central line was selected. These points marked by pieces of normal line in Fig. 5a. Transformation of capillary images along straightened capillary central line is shown in Fig. 5b. The blood flow velocity is calculated using the shift of RBCs in time as it is illustrated in Fig. 5b.

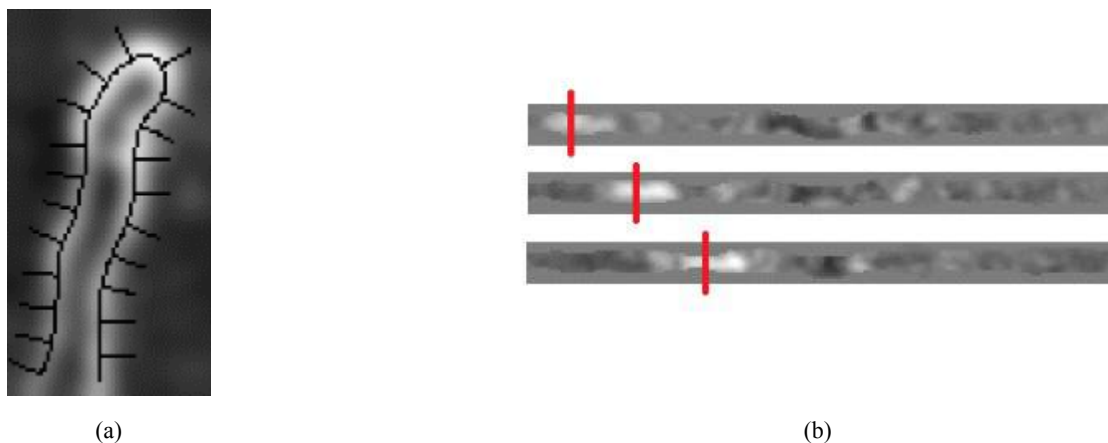


Figure 5. (a) Capillary image with the set of points marked by pieces of normal line; (b) images of RBCs ensemble corresponding to its positions along capillary central line at different moments of time.

The algorithm described above performs evaluation of local velocity of capillary blood flow to obtain a time graph of average velocity through all capillary line points. Experiments included the following procedures: recording of background level of perfusion and velocity (0.25-0.5 min), occlusion test (1-1.5 min), post-occlusion recording (0.1-1 min). Occlusion pressure was set about 220 mmHg.

### 3. EXPERIMENTAL RESULTS AND DISCUSSION

An example of graph with LDF-gram and capillary blood flow velocity is presented in Fig. 6.

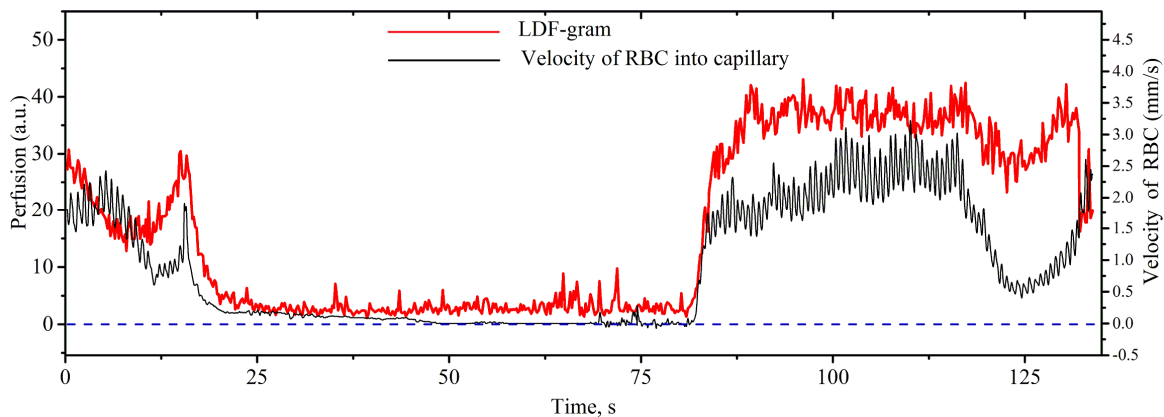


Figure 6. Experimental records of LDF-gram and blood flow velocity.

The VCS method allowed to register and quantify such features as reverse blood flow during occlusion test (see Fig. 7). This effect points out that occlusion leads to simultaneous reduction of blood flow in a single capillary from approximately 4 mm/s to small negative values, and index of microcirculation in the distal phalanx of the finger varies from 30 to 2 perfusion units. Subsequent gradual increase in the index of microcirculation to 5 perfusion units with continued occlusion is caused by detectable reverse blood flow in the capillaries.

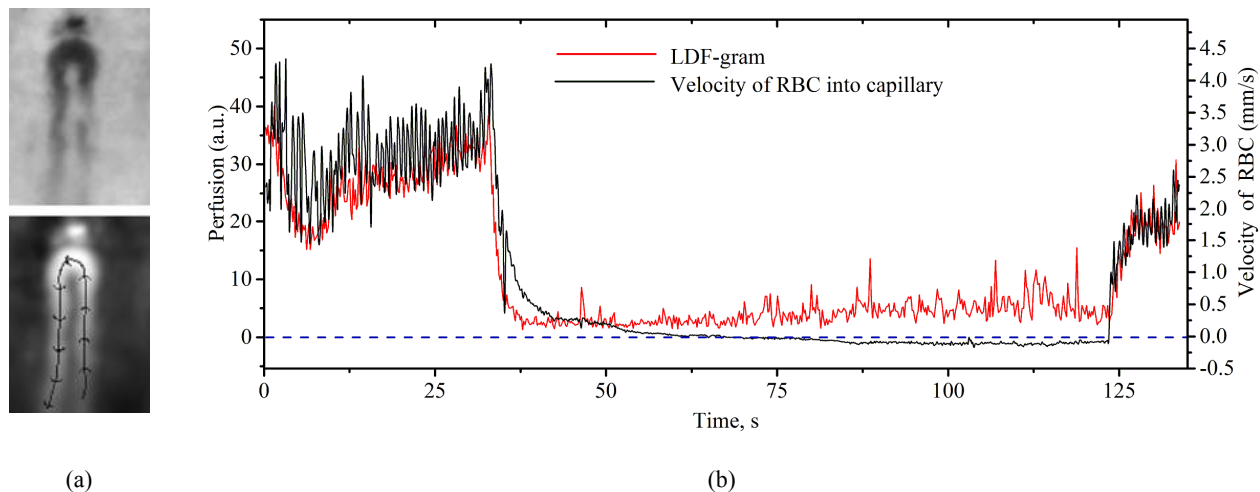


Figure 7. (a) Central line of a capillary; (b) simultaneous experimental record of LDF-gram with reverse blood flow.

The absolute blood velocity in capillaries increases gradually from 0 to 0.2 mm/s during the time of registration of the effect (from 75 s to 125 s of experiment duration). Occlusion finishing is characterized by a sharp increase in both the index of microcirculation and blood flow velocity. The curves reflect high degree of correspondence between the local variations of blood perfusion and blood flow velocity before and after occlusion (Fig. 6, 7). However, this correlation is absent at occlusion period. It can be explained by absence of influence of cardiac activity to blood microcirculation during occlusion.

#### 4. CONCLUSION

The obtained results show that the combined use of the LDF and VCS methods gives additional useful information to improve the reliability of data interpretation. It becomes possible to compare the changes in the perfusion in certain tissue volume with local tissue blood flow velocity and its direction in a single capillary. Effect of significant LDF-signal during occlusion can be caused by reverse blood flow. Besides, detected effect of reverse blood flow requires explanation from the point of view of human physiology. As a result, this study substantiates the approach of empirical confirmation of the relationship between blood flow velocity into capillaries and blood perfusion evaluated by the LDF method. Additional experiments with other provocative factors are assumed to be conducted for further investigation of blood flow regulation system.

#### ACKNOWLEDGMENTS

This work was partially supported by Russian Foundation for Basic Research (grant №16-32-50144-mol\_nr).

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