Laser reflectance oximetry and Doppler flowmetry in assessment of complex physiological parameters of cutaneous blood microcirculation

Andrey V. Dunaev*[a], Victor V. Sidorov[b], Neil A. Stewart[a], Sergei G. Sokolovski[a], Edik U. Rafailov[a]
[a]Photonics and Nanoscience Group, Division of Physics, School of Engineering, Physics and Mathematics, University of Dundee, Dundee DD1 4HN, UK; [b]SPE "LAZMA" Ltd., Moscow 125252, RF

ABSTRACT

The integration of multiple optical techniques within a single diagnostic device is used to address the difficulties in standardising measurement of cutaneous blood micro-dynamics caused by high variability. We demonstrate the benefits of simultaneous assessment of blood relative volume (V_b), microcirculation index (I_m) and tissue oxygen saturation (S_tO_2), during long-term examination of healthy volunteers. Consequently, five rhythmic components: endothelial, neurogenic, myogenic, breath and heart pulses were established showing high variability up to 30 – 50% as well as in initial parameters around 16%. All rhythmic components were synchronous with some latency between I_m and S_tO_2 in the myogenic component supports the hypothesis of strong correlation between peripheral hemodynamics and oxygen utilisation in tissues.

Keywords: laser Doppler flowmetry, tissue reflectance oximetry, microcirculation, bloodflow oscillations, variability

1. INTRODUCTION

Optical techniques are one of the promising non-invasive technologies for diagnosis of medical conditions. The integration of various techniques in an instrument and methodological framework that combines them in a single device using integrating algorithms for multi-modal diagnostics is particularly promising. Multi-functional laser non-invasive diagnostic systems (MLNDS), with up to 4 active channels are emerging, for example: laser Doppler flowmetry (LDF), tissue reflectance oximetry (TRO), laser fluorescence diagnostics (LFD), pulse oximetry and other. The most promising methods of optical non-invasive diagnostic methods are LDF and TRO. These methods are widely used in studying the dynamics of processes of blood microcirculation and oxygen transport and utilisation in biological tissues.

The results of LDF measurements, index of blood microcirculation (I_m) or perfusion, assessed in conventional perfusion units (PU), reveal a complex, non-periodic process. This variable component contains information on the modulation of blood flow. Use of spectral signal processing algorithms (LDF-graphs) for decoding and analysis provides information about the condition of vascular tone in terms of its contribution to the different mechanisms of micro-hemodynamic regulation (myogenic, endothelial, etc.) [1].

The TRO method determines relative blood volume (V_b) microcirculation in the surface layers of the soft tissues (skin, mucous membranes of the organs) and tissue oxygen saturation (S_tO_2) of the microvasculature in the inspected area of biological tissue. There are isolated cases of spectral processing algorithms recorded signals (S_tO_2- and V_b-graphs), being used to assess vasomotion and myogenic rhythms, for example[2]. We propose that analysis of oscillation signals recorded by TRO according to the frequency ranges, similar to LDF-graphs, is of practical interest in studying the parameters of microcirculation of blood, as the relationships between LDF and TRO attract the increasing attention of researchers in this field.

*a.v.dunaev@dundee.ac.uk; phone 44 1382 386571; dundee.ac.uk/elecengphysics/research/photonics/photonicsnanoscience

Advanced Biomedical and Clinical Diagnostic Systems XI, edited by Anita Mahadevan-Jansen, Tuan Vo-Dinh, Warren S. Grundfest, Proc. of SPIE Vol. 8572, 857205 · © 2013 SPIE
CCC code: 1605-7422/13/$18 · doi: 10.1117/12.2001797

It is well known that *in vivo* LDF and TRO measurements have high variability, at least in the range of ± 30% in terms of σ (standard deviation, *SD*) of the average measured values for each parameter (I_m, S_tO_2, V_b) [3]. Moreover, these results were obtained during long-term studies in two apparently healthy volunteers to estimate the individual momentary variability in the oscillation of the oxygenation in peripheral tissue blood microflow and also over a long period of observation. It should be emphasised that this work did not set out to evaluate the variability of the oscillations in a statistically defined group (age, sex, pathology, etc.). The aim was to assess long-term individual variability of the oscillations in a subject (a healthy volunteer) over one month, as such data does not exist in the literature.

2. CLASSIFICATION OF THE BLOOD FLOW OSCILLATIONS AND ITS VARIABILITY

Currently, for diagnostic purposes five rhythmic components (oscillations) were isolated from LDF recordings with help of the wavelet analysis in accordance with the modern interpretation of their genesis [4-7]:

- endothelial rhythms (frequency interval 0.0095-0.02 Hz);
- neurogenic rhythms (0.02-0.06 Hz);
- myogenic rhythms (0.06-0.16 Hz);
- breathing rhythms (0.16-0.4 Hz);
- pulse rhythms (0.4-1.6 Hz).

Possible physiological processes causing fluctuations in blood microflow in the frequency range of endothelial rhythms still are currently under discussion. However one of the possible explanation is a release of vasodilator nitric oxide into the endothelium [6]. It is believed that fluctuations in neurogenic rhythms (about 0.04 Hz) are irrelevant to vasomotion of arterioles. The increase in the amplitude of the oscillations is an indicator of lower neurogenic resistance and the possible increase in blood flow in the arterioles, venular shunt with an increase in myogenic tone [1]. It is believed that the source of myogenic oscillations (approximately 0.1 Hz) is the spontaneous activity of smooth muscle cells in resistance vessels and pre-capillary sphincters, also called vasomotion associated with regulation of blood pressure. Periodic oscillations with a frequency of about 0.3 Hz are synchronized with the breath. The activity of respiratory function poorly represented in the LDF-signal. Periodic oscillations with a frequency of about 1 Hz in the skin are synchronized with the heart rate and represent the variations that reflect changes in the diameter of blood vessels induced by the pulsation of the flow due to cardiac cycle.

It is known that the amplitude of the oscillations in these ranges varies considerably for individuals as well as the high lability of blood flow in the capillaries and other vessels of the microvasculature are a prerequisite for tissue homeostasis. Displaying the average amplitude distribution of blood flow to the rhythms of two healthy age groups 18-20 years (62 subjects) and more than 40 years (40 subjects). As it can be seen from the distribution of the amplitude of the oscillations in the neurogenic range for both age groups predominate. Deviation of the oscillation amplitude at a test area for a homogeneous group of subjects can be up to 15% [8]. However, data on the physiological variation in one individual assessed for a long period of time was impossible to find in the literature.

3. THE METHOD OF RESEARCH

In this study we used a MLNDS "LAKK-M" (SPE "LAZMA", Russia), which besides of LDF and TRO had pulse oximetry and LFD channels (Fig. 1). This system allows simultaneous recording of the I_m, S_tO_2, V_b parameters in a tissue volume [9]. The study was conducted on an adult male, age 35, with no cardiovascular disease history. The measurements were performed on a skin pad (palmar surface) right middle finger. This area was chosen because it is rich in arteriolar-venular anastomoses (AVA) and variability of the LDF signal is less than in tissue with fewer shunts [4]. Consequently, it is ideal for this research aimed at evaluating the proportion of nutritive blood flow (*NB*) from the known formula, and hence the parameters such as myogenic (*MT*), neurogenic (*NT*) and endothelial-dependent component (*ET*) tone [1].

All measurements were performed daily during the month (except holidays) at the same time (around 11:00 am) to avoid circadian rhythms influence on the blood circulation. Length measurements was 3 min, total of 22 measurements (days). An example of screenshot of the registered parameters is presented in Fig. 2a. At the end of the observation time the oscillation rhythms of each measurement were analysed using the built-in module "wavelet analysis" (example screenshot is shown in Fig.2b). The wavelet analysis determined the maximum amplitude (δI_m, δS_tO_2, δV_b) and the corresponding frequency for each of the 5 oscillation types for each of the 3 parameters of I_m, S_tO_2 and V_b.

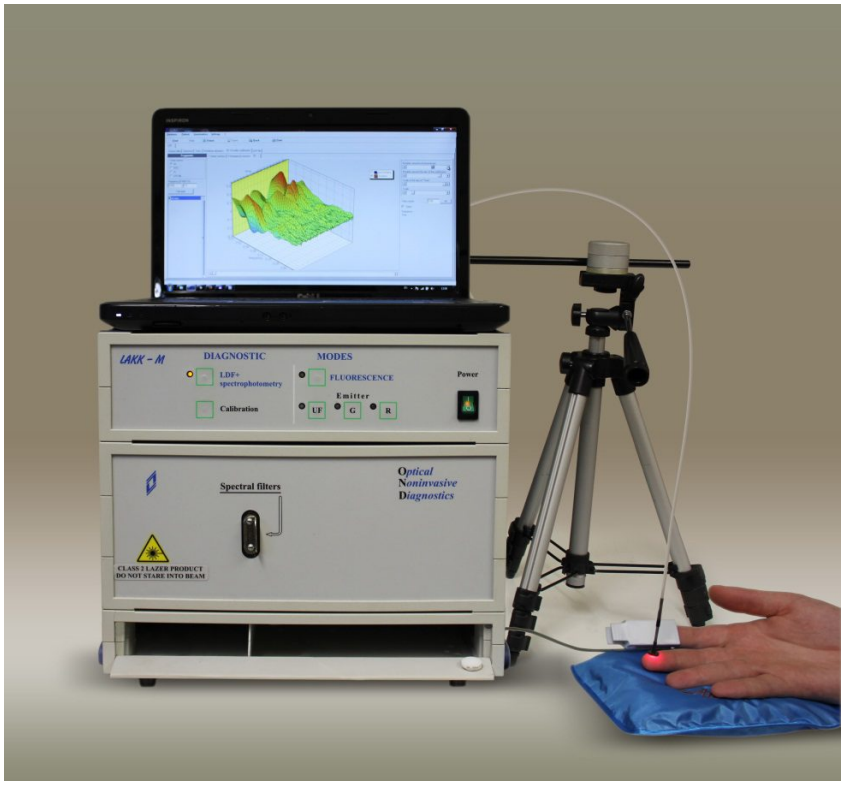
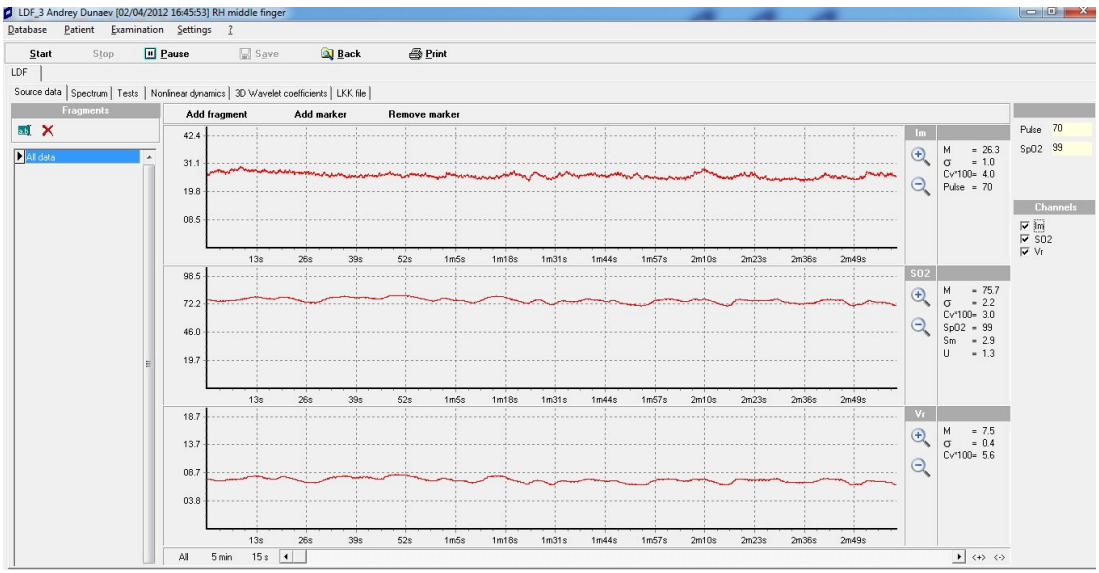


Figure 1. MLNDS "LAKK-M".

In addition to variability, the data was assessed for parameters such as: bypass index (*BI*), index of tissue oxygen use (taking into account nutritional blood flow) (*I*), index of perfusion oxygen saturation in microvascular blood (S_m), index of oxygen consumption rate in tissue (U_1 and U_2), calculated according to [10] and [11] respectively.



a)

b)

Figure 2. Screenshots of LDF- and TRO-recordings (a) and wavelet analysis (b).

4. RESULTS AND DISCUSSION

These results have been presented in Table 1 and Fig. 3-6.

Table 1 shows the statistical evaluation (*M*, is mean value, *SD is* standard deviation, k_v is a coefficient of variation) of the measured LDF and TRO methods for parameters of I_m, S_tO_2, V_b and calculated on the basis of their indicators. Fig. 3 show a diagram of changes in the parameters of I_m, S_tO_2 and V_b for a month in one subject.

Table 1. The statistical evaluation of the LDF and TRO parameters.

№	Statistic parameters	Measured parameters			Calculated parameters								
		I_m, PU	S_tO_2, %	V_b, %	*NT*	*MT*	*ET*	*BI*	*NB*, PU	*I*	S_m	U_1	U_2
1	*M*	25.6	62.5	8.7	1.76	2.06	2.00	1.23	11.90	19.36	2.5	1.6	3.9
2	*SD*	3.5	10.2	1.4	0.49	0.54	0.54	0.40	3.10	6.01	0.5	0.3	1.5
3	k_v, %	13.5	16.4	16.3	27.59	26.47	27.17	32.57	25.80	31.02	20.5	16.8	37.7

Analysis of the data shows the greatest variability in the parameters obtained by the TRO (16%), whereas the variability of the microcirculation index (I_m) in our study turned out not more than 14%, more than 2 times less than previously reported data [3], obtained for 10 days. Perhaps this is due to the specific devices used by these authors with different diagnostic volumes (due to different measurement bases) and the design of the optical probes. We conducted longer-term studies in order to contrast with the published short-term study data previously used to study variation.

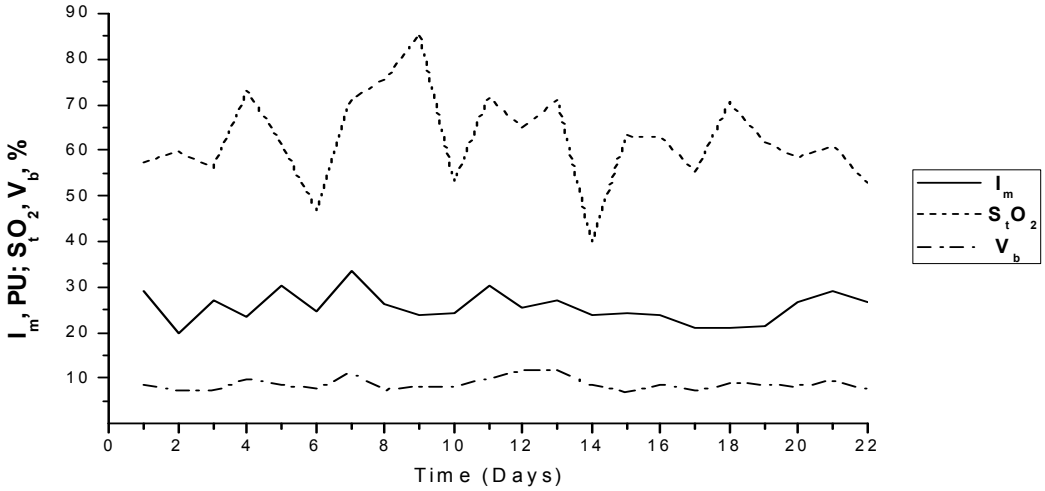
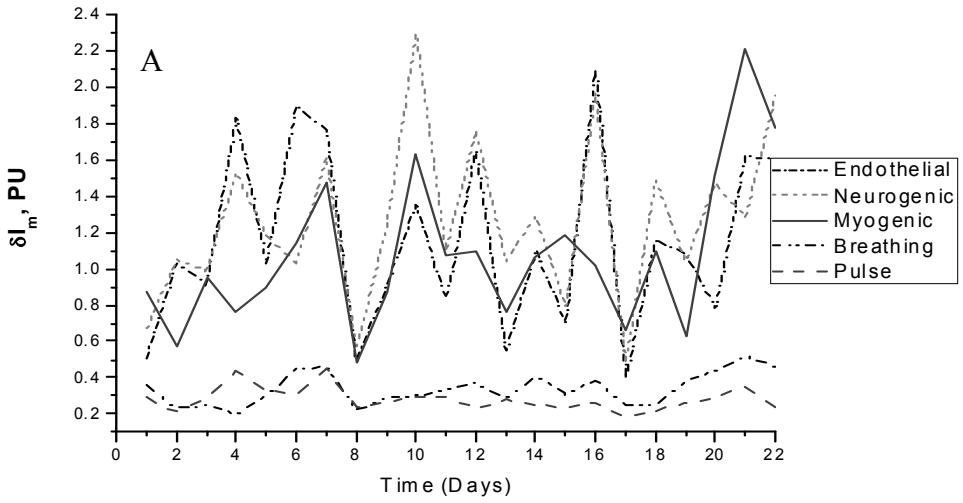


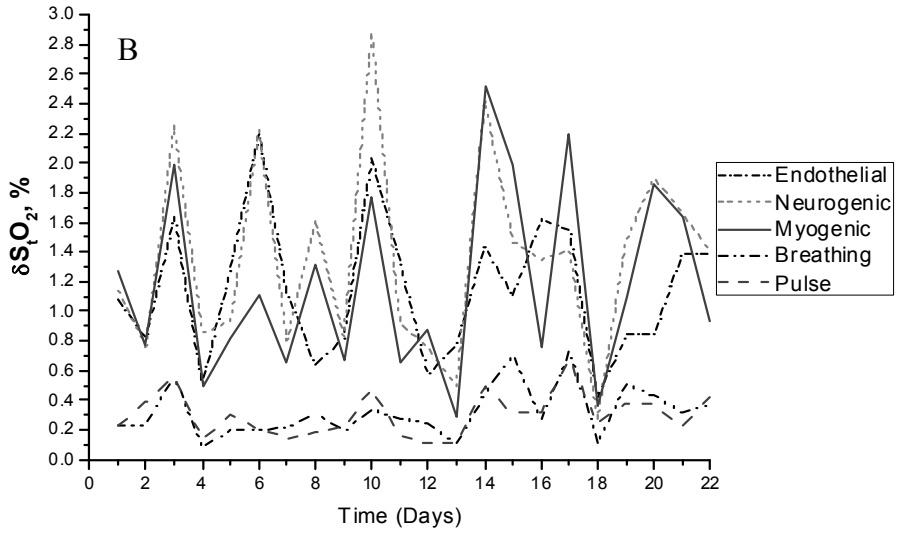
Figure 3.Representitive I_m, S_tO_2 and V_b parameters from one individual.

The calculated values of *MT*, *NT* and *ET*, as well as *BI* have a high variability (up to30%) due to use in their calculations, as it will be shown below, the oscillation amplitudes with high variability. The nutritional bloodflow (*NB*) component was 11.9±3.1 PU, which is about two times lower than the mean value of the index of blood microcirculation 25.6±3.5 PU. The S_m index has a variability of about 20%, while the index of oxygen rate of use in tissue (U_2) has a variability factor twice that of (U_1), since its calculation is based on two parameters at once with variability of 16%. In aiming to reducing the influence of the variation in the measured parameters it is preferable to use the integrated index calculation U_1.

Fig. 4 shows the changes in the study of oscillations during the month for the LDF-, S_tO_2- and V_b-graphs respectively (Fig. 4a, 4b, 4c). In Fig. 5 shows corresponding histogram showing the distributions of mean value and *SD* for these oscillations.

Analysis of the data in general shows a high variability of the investigated oscillations in all 3 graphs. The smallest variation is the V_b in endothelial (30%), neurogenic (35%) and myogenic components (40%). While pulse rhythms variability turned out about 33%, which is greater than 23% of data variability rhythms in blood perfusion. The greatest variable component in all the 5 rhythms was S_tO_2-parameter (50%) while the greatest variation is observed in endothelial (44%) and myogenic rhythms (40%) LDF-graphs, in myogenic (54%) and breathing rhythms (55%) S_tO_2-graphs and in myogenic (38%) and pulse rhythms (33%) V_b-graphs.





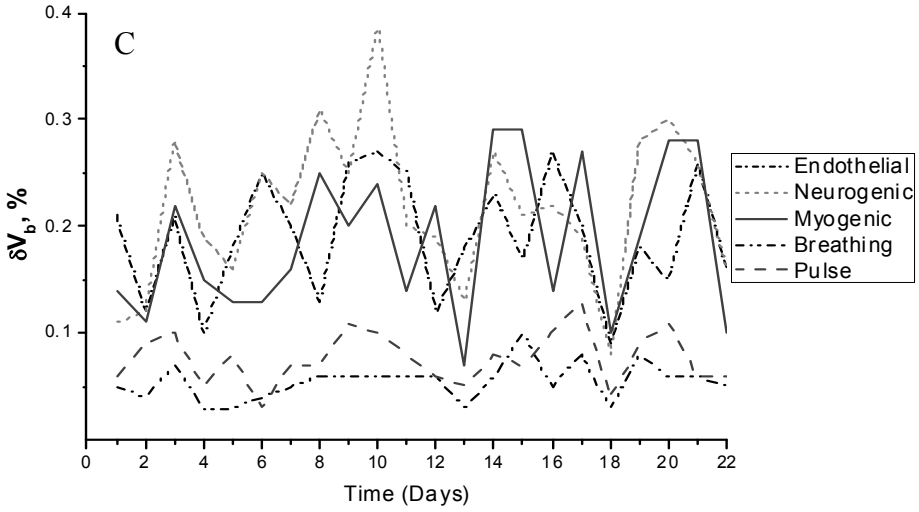


Figure 4. Representative changes in blood perfusion (A), S_tO_2 (B) and V_b (C) oscillations recorded from one individual.

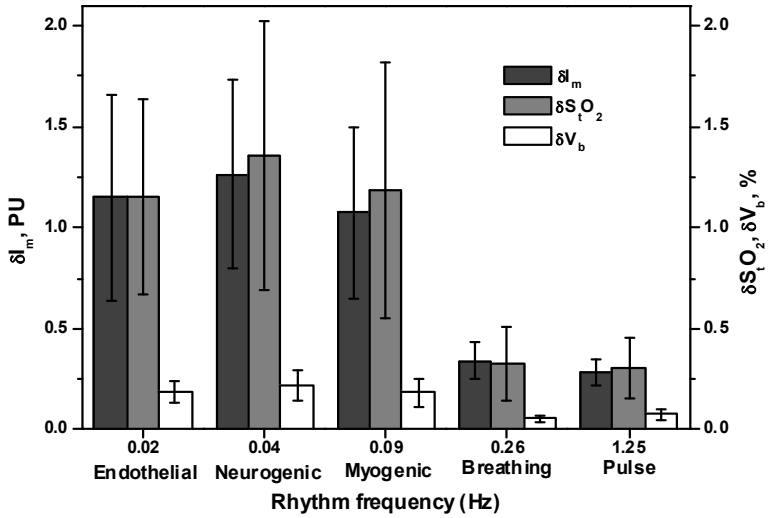


Figure 5. Blood perfusion, S_tO_2, and V_b parameters regularly taken from one individual form a month.

The synchronicity of all 5 components of the fluctuations during the whole period of studies can be seen in the Fig. 4 For example, analysis of Fig. 3 and Fig. 4 shows rising neurogenic rhythm of oscillations with a periodicity of about 8 days (3-, 10-, 16- and 22- days), which corresponds precisely to an increase in perfusion (I_m) on the same days. This may be considered quite natural due to the more intense work of myocytes of arterioles. At this time a subject indicated an increase in tissue oxygen saturation (S_tO_2).

It is interesting to note the ability the results provide to assess the relationships between blood perfusion and S_tO_2. For example, one can trace a pattern manifested three times during the time trials (7-, 16- and 18- days) where we observe a decrease in tissue oxygen saturation with increasing amplitude of myogenic oscillations. Thus, the increase in hydrostatic pressure is accompanied by an increase in the diffusion of oxygen in tissues and, consequently, a decrease in tissue oxygen saturation, which corresponds to the data given in [2].

Fig. 5, presents the histograms of oscillations in the LDF-, S_tO_2-and V_b-data of the amplitudes of repeated known data [8] for the LDF-graphs, except for the amplitudes of pulse rhythms to V_b-graphs, in which the pulse oscillations predominate over breathing. This is most likely due to the physiological meaning of the parameter V_b being more dependent on the pulse fluctuations. In addition, these data allow us to estimate the long-term variability of 5 rhythms for all 3 parameters.

Fig. 6 shows the related oscillations of myogenic vasomotion for the blood perfusion, S_tO_2 and V_b. This approach extends the assessment of the contribution of a particular component in each of the 3 graphs.

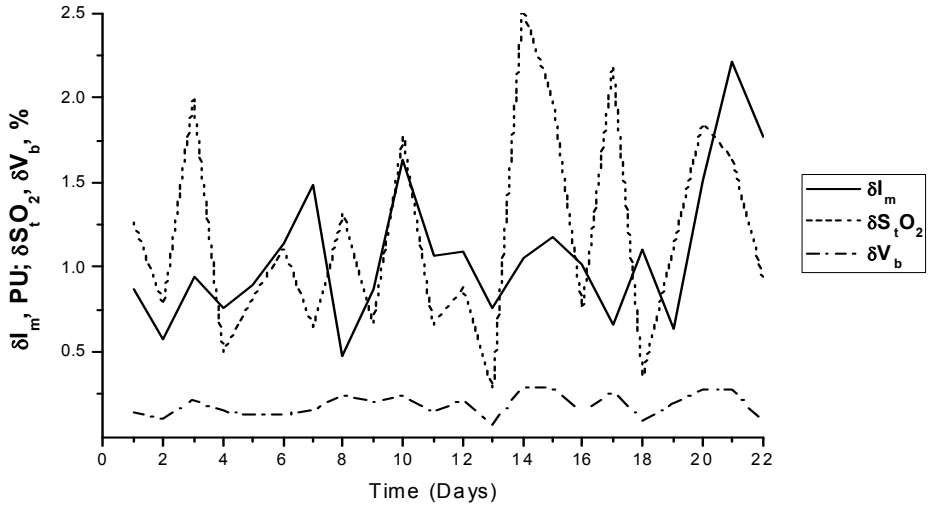


Figure 6.Representitive myogenic vasomotion oscillations of blood perfusion, S_tO_2 and V_b. recorded from one individual.

5. CONCLUSION

The obtained results confirm that peripheral blood flow and tissue oxygenation is characterised by high variability (lability), which is essential for adaptation of the organism to stresses. The observed high variability of the oscillations (about 30-50%) also may be partially explained by the peculiarities of non-invasive optical diagnostic techniques, primarily associated with low diagnostic volume (in this case about 1 mm^3). Obviously, the blood flow and oxygenation in a small volume will vary significant due to spatial heterogeneity in biological tissues.

However, the long-term observation of subject(s) and analysis of the individual variability of her/his blood rhythm (LDF- and S_tO_2-parameters) identified a new correlation between the peripheral hemodynamics and oxygen utilisation in tissues. This confirmed increase in the information content of the data also confirms the requirement to factor these variables into any models or algorithms used for non-invasive optical diagnostic techniques.

This work was supported by FP7 EU IAPP project (MEDILASE, #806026).

REFERENCES

[1] Krupatkin, A. I. and Sidorov, V. V., [Laser Doppler flowmetry of blood], Meditcina-Press, Moscow(2005).
[2] Thorn, C. E., Kyte, H., Slaff, D. W. and Shore, A. C., "An association between vasomotion and oxygen extraction," American Journal of Physiology-Heart and Circulatory Physiology, 301(2), H442-H449 (2011).
[3] Makarov, D. S. and Rogatkin, D. A., "Physiological deviation of individual blood microcirculation parameters as a source of errors in noninvasive medical spectrophotometry," Proc. "Physics and radioelectronics in medicine and ecology PREME'2010". 82-86 (2010).
[4] Salerud, E. G., Tenland, T., Nilsson, G. E. and Oberg, P. A., "Rhythmical variations in human-skin blood-flow," International Journal of Microcirculation-Clinical and Experimental, 2(2), 91-102 (1983).

[5] Bracic, M. and Stefanovska, A., “Wavelet-based analysis of human blood-flow dynamics,” Bulletin of Mathematical Biology, 60(5), 919-935 (1998).
[6] Stefanovska, A., Bracic, M. and Kvernmo, H. D., “Wavelet analysis of oscillations in the peripheral blood circulation measured by laser Doppler technique,” IEEE Transactions on Biomedical Engineering, 46(10), 1230-1239 (1999).
[7] Kvandal, P., Landsverk, S. A., Bernjak, A., Stefanovska, A., Kvernmo, H. D. and Kirkeboen, K. A., “Low-frequency oscillations of the laser Doppler perfusion signal in human skin,” Microvascular Research, 72(3), 120-127 (2006).
[8] Krasnikov, G. V., Matrusov, S. G., Piskunova, G. M., Sidorov, V. V. and Chemeris, N. K., "Age characteristics of peripheral blood flow oscillations in the skin," Proc. "Application of laser Doppler flowmetry in medical practice". 32-34 (2000).
[9] Rogatkin, D. A., Sokolovski, S. G., Fedorova, K. A., Sidorov, V. V., Stewart, N. Z. and Rafailov, E. U., "Basic principles of design and functioning of multifunctional laser diagnostic system for non-invasive medical spectrophotometry," Proc. SPIE. 7890, 78901H1 (2011).
[10] Stewart, N. A., Dunaev, A. V., Sokolovski, S. G., Sidorov, V. V. and Rafailov, E. U., "Multi-parameter analysis in blood circulation and and perfusion based diagnostics," Proc. "Laser Optics 2012". (2012).
[11] Krupatkin, A. I., Rogatkin, D. A. and Sidorov, V. V., "Clinical-diagnostic parameters for complex investigation of microhaemodynamics and oxygen transport in the system of microcirculation," Proc. "Hemorheology and microcirculation". 106 (2007).