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# Study of the metrological characteristics of a spatial frequency domain imaging system for biological tissues structure visualization

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The work is devoted to the study of the measurement accuracy of the spatial frequency domain imaging system for the detection and objective numerical evaluation of optical inhomogeneities in biological tissues. As a result of the initial stage of experimental studies on a stand simulating a spatial frequency domain imaging system, the operating range of spatial frequencies of illuminating irradiation modulation was determined, which ensures maximum reproducibility of the measurement results, and the possibility of achieving a relative measurement error of optical parameters of no more than 10% was shown.

**Keywords:** spatial frequency domain imaging, optical properties, biological tissues, spectrophotometry, optical imaging, metrological characteristics, quantitative assessment, measurement errors.

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## Introduction

In the last decades, methods for studying the optical properties of biological tissues have become increasingly important in the field of healthcare and biology, in the problems of studying hemodynamics, microcirculation, controlling regeneration processes, and detecting neurovascular structures during surgery [1,2]. The measurement of the optical properties of biological tissues is widely used in fundamental research to gain new knowledge about biological processes, for example, in the study of the properties of brain tissues. [3,4]. To determine the optical parameters of biological tissues, the collimated transmission method, the integrating sphere method, and near-infrared band spectroscopy in the time domain are used [5]. Frequency, spatially resolved, spatially frequency methods for measuring optical parameters are also known [6]. Methods for non-invasive measurement of the optical parameters of biological tissues and imaging, such as thermography, laser speckle contrast imaging (LSI), spectral optical coherence tomography (SOCT), spatial frequency domain imaging (hereinafter referred to as SFDI) and others, are successfully used separately or in combination to assess and classify the severity of burns. Thus, for example, thermography shows an accuracy of 73%, LSI shows an accuracy of 75%, SFDI shows an accuracy of 85%, SOCT shows an accuracy of 76% (up to 90% in case of deep burns) [7,8]. The above-listed methods have their advantages and disadvantages in terms of depth, field of view, spatial resolution and speed (the SOCT method is limited in penetration depth to several hundred microns [8]), so the appropriateness of their use depends on the specific medical problem being solved.

Due to the ability to quickly map the distribution of optical parameters of biological tissues, the spatial frequency domain imaging is increasingly used in medical research [4]. Currently, the SFDI technology is widely used in various fields of healthcare, for example, in dentistry for imaging and numerical assessment of inflammatory process foci, erosive and ulcerative lesions at an early stage, in surgery, for example, for monitoring the state of surgical wounds, as well as in diagnosing the diabetic foot disease [9–11]. In addition, the SFDI allows assessing the state of the subject of study in real time, due to which the method is becoming increasingly relevant.

In addition to the accuracy, the reproducibility of measurements is important for the clinical application of the technology. In [12], a large-scale comparison of 30 instruments that implement various methods for studying the optical properties of biological objects, including the „OxiplexTS“ spectrophotometric complex that implements the phase modulation approach (FD NIRS), and a device for assessment of the burn depth, which implements the SFDI technology. More than 70% of the instruments tested in the study [12] had the coefficient of measurement result variation used as a measure of reproducibility less than 5%, and some instruments had it even less than 1%. The possibility is noted to improve the reproducibility of FD-NIRS results in the case of calibration using measures with known parameters [12]. At the same time, the coefficient of variation of SFDI measurements exceeded 10% [12], which makes it reasonable to study aimed at improving the reproducibility of SFDI measurement results. Another important challenge that needs to be solved to ensure the possibility of clinical application of SFDI technology is the definition of the operating range of parameters where measurements can be made.

To solve these problems and determine the metrological characteristics of the SFDI technology, it is necessary to

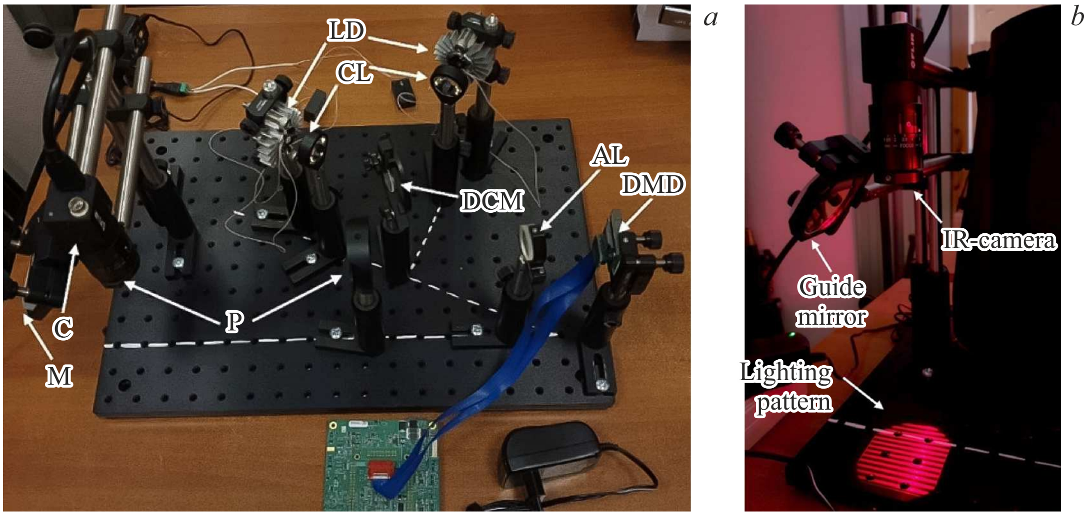


**Figure 1.** Photo of the assembled SFDI stand: *a* — off state (LD — diodes, CL — collimating lenses, DCM — dichroic mirrors, AL — achromatic lens, *P* — linear polarizers, *M* — directional mirror, *C* — camera, DMD — digital micromirror device); *b* — on state.

study the dependence of the SFDI measurement accuracy on various external factors, such as the level of ambient illumination, the spatial modulation frequency, and the geometric parameters of the recording system. The results of studies of the SFDI measurement accuracy dependence for such registration parameters as the distance between the surface of the biological tissue under study to the endoscope camera, the parameters of the profile of the surface under study, the parameters of geometric distortions of the projector have been published [6,13,14].

This study presents preliminary results of studying the dependence of the SFDI measurement accuracy on the spatial frequency of illumination modulation and the level of ambient illumination. Based on the results of a series of measurements of the optical parameters of test objects carried out on the SFDI system simulator and the comparison of the obtained data with the results of measurements performed by an „Oxiplex“ spectrophotometer of ISS, the accuracy of SFDI measurements was evaluated depending on two external factors: the ambient illumination and the spatial frequency of illumination modulation. Based on the evaluation performed, the operating range of the spatial modulation frequencies was preliminarily determined, which ensures maximum reproducibility of SFDI measurement results.

## Materials and methods

In the course of this study, a stand made up of from publicly available components was used for investigations in the spatial frequency domain [15] (hereinafter referred to as the SFDI stand). The SFDI stand contains three main modules: an illumination module that includes laser diodes, collimating lenses, dichroic mirrors; a spatial modulation module, including a digital micromirror device with a control microcontroller and an image recording module, including linear polarizers, a directing mirror, a video camera (Fig. 1).

The object under study must be placed in the area of illumination. To generate illumination patterns and register the resulting images, the stand software on the LabView platform, developed by a research team from the Boston University, was used [16]. The assembled stand makes it possible to record images at wavelengths of 660 and 850 nm at different spatial frequencies of radiation modulation. The processing of experimental results in the general case for the SFDI technology consists of four stages [4]:

– demodulation by three images at the same spatial frequency: with phases of 0, 120 and 240°;

– calibration to separate the response function of the system from the response function of the test sample;

– determination of the diffuse reflection coefficient of the recorded sample;

– determination of the optical characteristics of the recorded sample as a result of solving the inverse problem: known values of the diffuse reflection coefficient at two (or more) spatial frequencies are used to determine the values of optical parameters of the object (absorption and scattering indices) that satisfy the radiation transfer equation.

By measuring the optical properties at several wavelengths, the concentration of tissue chromophores can be determined, which helps to determine the state and metabolic function of the tissue. However, it is necessary to take into account the influence of external factors also to prevent the occurrence of artifacts in the processed images [15]. The result of image processing are maps of the distribution of optical parameters: the absorption index and the transport scattering index of the sample under study. The intensity of each pixel of such a map corresponds to the value of the optical parameter. The images obtained

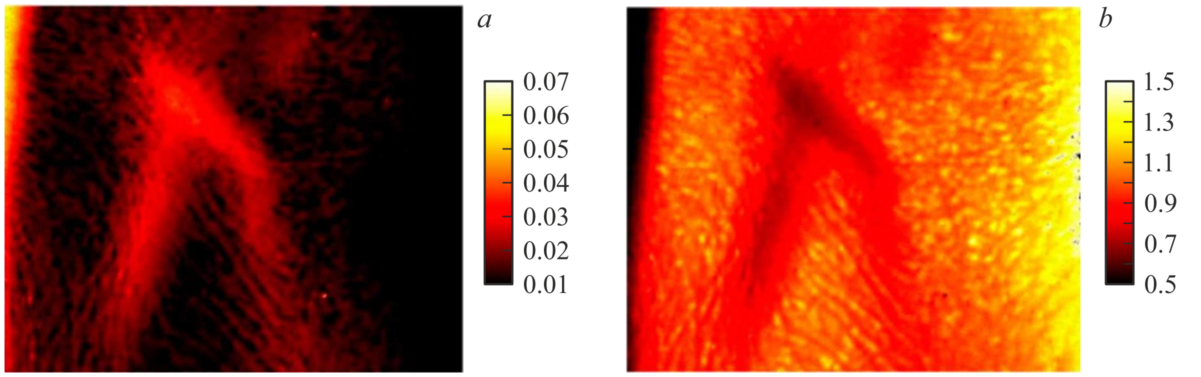


**Figure 2.** SFDI — images of veins on the back of the hand, recorded using the stand: *a* — absorption index distribution, $\mu_a$ (mm$^{-1}$); *b* — distribution of the transport scattering index, $\mu_s$ (mm$^{-1}$).

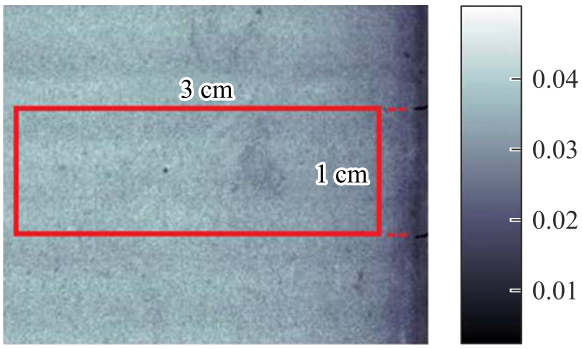


**Figure 3.** Selection of the image area to calculate the mean value and standard deviation.

after calibration contain a unique combination of optical characteristics in each of its pixels, which can be extracted by solving the inverse problem, for example, by fitting. The result of processing will be distribution maps of the optical parameters of the image, the pixel intensity of which corresponds to the value of the optical parameter. For the convenience of visual perception, the maps are adapted using pseudo-colors.

Fig. 2 shows SFDI images of veins on the back side recorded on the stand (wavelength is $\lambda = 660$ nm, spatial modulation frequency is $f_x = 0.1$ lp/mm, ambient illumination is $L = 40$ lx). Metrological characteristics of the SFDI system in this study were investigated on test objects based on acetoxysilicone with the addition of graphite and titanium oxide that simulate the optical properties of biological tissues with local inhomogeneities at different depths, made in accordance with a proven procedure [17].

Results for the optical parameters of the test object obtained using a „OxiplexTS" spectrophotometer (ISS, Inc., USA) with a phase modulation approach (FD NIRS) [18] in accordance with the proven procedure [19] were used as reference values of the optical parameters. Biotissue probing was carried out with two wavelengths of 692 and 850 nm from laser diodes with a power of 1 mW. The radiation intensity was modulated in amplitude with a frequency of 110 MHz and a modulation depth of 0.5. On the distribution maps of optical parameters obtained by the SFDI method, areas corresponding to the coverage of the „Oxiplex" sensor were identified where the mean value and standard deviation of the optical parameters were calculated (Fig. 3).

The relative error of SFDI measurements $\delta$ was determined by the following formula:

$$\delta = \frac{\mu - \mu_{\text{REF}}}{\mu_{\text{REF}}} 100\%, \tag{1}$$

where $\mu$ is the result of the SFDI measurement of the optical parameter, $\mu_{\text{REF}}$is the result of the optical parameter measured by the „OxiplexTS" spectrophotometer.

## Results

Fig. 4 shows the experimental dependence of the relative error of SFDI measurements of the test object on the spatial frequency of illumination modulation at different levels of ambient illumination obtained on the SFDI bench at wavelengths of 660 and 850 nm.

The smallest relative error corresponds to a spatial frequency of 0.2lp/mm. The standard deviation increases with increasing spatial frequency. The section with spatial frequencies from 0.2 to 0.4 lp/mm can be linearly approximated for wavelengths of 660 and 850 nm (Fig. 5). Thus, the spatial frequency range from 0.2 to 0.4 lp/mm is the operating range where maximum reproducibility of the SFDI measurement results is possible. In this case, in the range of spatial frequencies from 0.2 to 0.25 lp/mm, the relative measurement error not higher than 10% can be achieved.

The preliminary results obtained confirm the possibility of determining the values of optical parameters using the SFDI technology in the spatial frequency range from 0.2 to 0.25 lp/mm at wavelengths of 660 and 850 nm with a relative error not exceeding 10% at an ambient illumination level of up to 540 lx. In the course of further studies, it is reasonable to verify the working

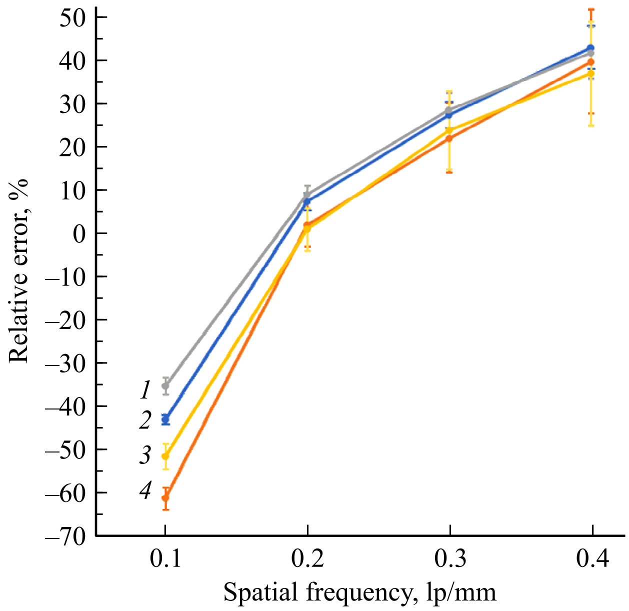


**Figure 4.** Experimental dependence of the relative error of SFDI measurements of the test object on the spatial frequency of illumination modulation at 60 (*1, 2*) and 540 lx (*3, 4*) at wavelengths of 660 (*2, 4*) and 850 nm (*1, 3*).

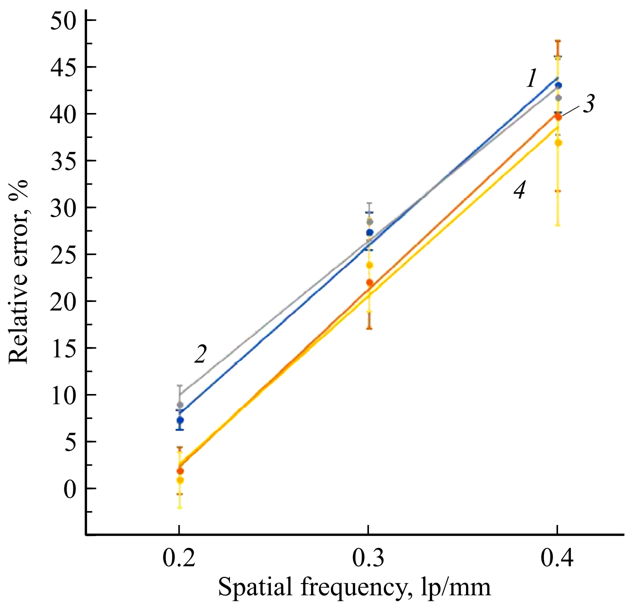


**Figure 5.** Linear approximation of the relative error at 60 (*1, 2*) and 540 lx (*3, 4*) at a wavelength of 660 (*1, 3*) and 850 nm (*2, 4*).

range of spatial modulation frequencies and the values of the relative measurement error on a line of test objects with different values of optical parameters. Also, in the course of further research, it is planned to find a physical interpretation of the experimental dependence of the SFDI measurement relative error on the spatial modulation frequency.

## Conclusion

As a result of the initial stage of experimental studies on a stand simulating an imaging system in the spatial frequency domain, the operating range of spatial frequencies of illumination modulation was determined, which ensures maximum reproducibility of the measurements of the optical parameters of biological tissues, and the possibility is shown to achieve a relative error of SFDI measurements of optical parameters not more than 10%.

### Conflict of interest

The authors declare that they have no conflict of interest.

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