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ABSTRACT

A theoretical basis for the method of polarization-interference mapping of optically thin polycrystalline films of human biological fluids is given. The coordinate distributions of the value of the local contrast of the interference distributions of the polarization-inhomogeneous microscopic images of polycrystalline films of the synovial fluid of the human joint are investigated. In the framework of the statistical (statistical moments of the 1st-4th order) approaches, objective criteria for the distribution of the values of local contrast are established. The possibility of differentiation of weak changes in the optical anisotropy of blood films of healthy and patients with breast cancer patients is determined.

Keywords: polarization, interference, anisotropy, cartography.

1. Introduction

Among the many areas of optical diagnostics of biological objects, the original place is occupied by laser polarimetry of optically thin (nondepolarizing) layers of biological tissues [1-15]. The main object of this diagnosis is a variety of tumor or precancerous conditions of tissues of human organs with fibrillar structure of polycrystalline networks. In [16-21], a high sensitivity of the polarimetric differentiation of such samples was demonstrated. It has been established that such pathologies are accompanied by significant changes in phase anisotropy due to the transformation of the optical axes direction distributions and the birefringence of protein fibrils [22-27]. At the same time, obtaining samples of histological sections requires a traumatic biopsy operation. This circumstance complicates the application of laser polarimetry methods of biological tissues in everyday clinical practice. To overcome this disadvantage, it is promising to study the distributions of the polarization-phase parameters of microscopic images of smears of biological fluids. Such objects are easily accessible, which makes it possible to carry out a non-traumatic evaluation of the presence of a pathology at an early stage of its occurrence. This circumstance opens up new possibilities for using methods of polarimetry in screening mass surveys.

In our work we consider the polarization-interference mapping of such layers. The purpose of such studies is to reveal the interrelationships between the local contrast divisions of interference patterns and phases in the plane of images of polycrystalline films and the changes in their birefringence. An applied aspect of the work is the determination of the effectiveness of differentiation of polycrystalline blood films of healthy and patients with breast cancer patients.

2. A brief theory of polarization-interference mapping of polycrystalline films

Let us consider the process of forming an interference distribution in the plane of an interference-inhomogeneous image of an optically thin (nondepolarizing) polycrystalline film of a biological fluid. We use the following approximations:

- irradiating and reference waves are circularly polarized
$$\begin{cases} \Phi_y - \Phi_x = 0.5\pi; \\ \varphi_y - \varphi_x \equiv 0.5\pi; \end{cases} \begin{cases} |B_x| = |W_x| \equiv 1; \\ |B_y| = |w_y| \equiv 1; \end{cases} \tag{1}$$

$$B = W = \begin{pmatrix} 1 \\ i \end{pmatrix};$$ (2)

- phase fluctuations of the polycrystalline layer are small

$$\vartheta \to 0 : \cos \vartheta \approx 1; \sin \vartheta \approx \vartheta.$$ (3)

$$\begin{cases} m_{11} = 1 - i\vartheta \sin^2 \vartheta; \\ m_{12} = d_{21} = i\vartheta \cos \alpha \sin \alpha; \\ m_{22} = 1 - i \cos^2 \alpha. \end{cases}$$ (4)

It is shown that the intensity of the interference field at the point r

$$D(a) = \vartheta^2(a) - 2\vartheta(a)\sin \beta(a) + 2 \cos \beta(a).$$ (5)

From analysis (5) it follows that

$$\begin{cases} \beta(a) = 0 \leftrightarrow D_{\max}(a) = 2 + \vartheta^2(a); \\ \beta(a) = \pi \leftrightarrow I_{\min}(a) = \vartheta^2(a). \end{cases}$$ (6)

The local contrast of the interference distribution is given by

$$V(\beta, \vartheta) = \frac{D_{\max}^{(\beta=0)} - D_{\min}^{(\beta=\pi)}}{D_{\max}^{(\beta=0)} + D_{\min}^{(\beta=\pi)}} = \frac{1}{1 + \vartheta^2}.$$ (7)

Thus, by means of circular polarization-interference mapping of a microscopic image of a polycrystalline biological fluid, it is possible to obtain an azimuthally invariant distribution (within the pixels of a digital camera) of the magnitude of the local contrast $V(m \times n) = W \begin{pmatrix} a_{11} & \dots & a_{1n} \\ . & \dots & . \\ a_{m1} & \dots & a_{mn} \end{pmatrix}$ and to estimate the weak changes in the birefringence of this layer.

3. Materials and methods

In fig. 1 shows the optical scheme of polarization interferometry of laser microscopic images of polycrystalline films of biological fluids.
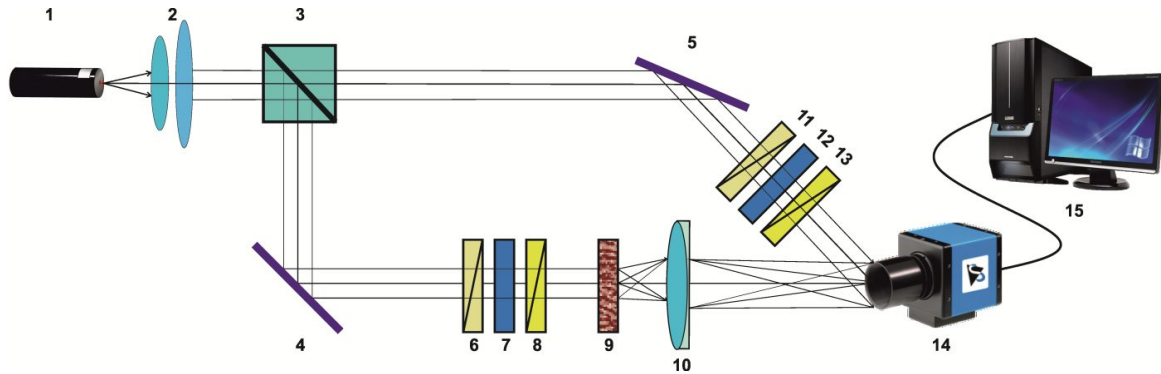
Fig. 1. Optical scheme of polarization interferometry of microscopic images of optically anisotropic biological layers. Explanations in the text

The parallel ($\varnothing = 2 \times 10^3 \, \mu m$) beam He-Ne ($\lambda = 0,6328 \, \mu m$) of laser 1, formed with the help of a collimator 2, the beam splitter 3 is divided into two - "irradiating" and "reference".

The "irradiating" beam with the help of the rotary mirror 4 is guided through the polarization filter 6 - 8 in the direction of the sample of the biological layer 9. The polarization-inhomogeneous image of the object 9 by the objective 10 (Nikon CFI Achromat P, focal length 30 mm, numerical aperture 0.1, magnification - 4x) is projected into the plane of the photosensitive pad ($m \times n = 1280 \times 960$ pixels) of the digital camera 14.

The "reference" beam by the mirror 5 is guided through the polarization filter 11-13 into the plane of the polarization-inhomogeneous image of the object 9. As a result, an interference pattern is formed, the coordinate intensity distribution is recorded by the digital camera 14 (The Imaging Source DMK 41AU02.AS, monochrome 1/2 "CCD , Sony ICX205AL (progressive scan); the resolution is 1280x960; the size of the light-sensitive pad is 7600x6200μm; the sensitivity is 0,05 lx; the dynamic range is 8 bit; SNR is 9 bit).

3.1. Method for measuring the coordinate distributions of the local contrast value of interference patterns of polarization-inhomogeneous images

- polarizers 8 and 13 "are output" from the path of the irradiating beam (Fig. 1);

- the axes of the highest velocity of quarter-wave plates 7 and 12 are oriented at an angle $\Omega = 45^0$ with respect to the transmission axis of the polarizers 6 and 11;

- the digital camera 14 registers a discrete ($m \times n$) image of the interference intensity distribution in the plane of the microscopic image of the polycrystalline film;

- the registered distribution $I(m \times n)$ is scanned by the aggregate ($k = 1,...m$) of the rows ($k1,...kn$) in increments $\Delta r = 1 \, pix$;

- for every k-th row, a set of local contrast values is determined from the following relationship

$$V_k = \frac{\left| D(a_{i=1,...n}) - D(a_{i=1,...n} + \Delta a) \right|}{D(a_{i=1,...n}) + D(a_{i=1,...n} + \Delta a)} \qquad (8)$$

- for the whole set of series, a coordinate distribution (map) of the value of the local contrast of the interference distribution in the plane of the microscopic image of a polycrystalline biological film

$$V(m \times n) = \begin{pmatrix} V_{11} & \cdots & V_{1n} \\ . & \cdots & . \\ V_{m1} & \cdots & V_{mn} \end{pmatrix} \qquad (9)$$

4. Brief description of the objects of research.

The following optically thin (attenuation coefficient $\tau \prec 0.01$) samples of polycrystalline films (geometric thickness $l = 15 \mu m$ $0.0089 \leq \tau \leq 0.0097$) of blood were studied:

- norm (25 samples), - group 1;
- breast cancer (25 samples), - group 2.

In Fig. 2 presents microscopic images of polycrystalline group 1 (fragment (1)) and group 2 (fragment (2)).
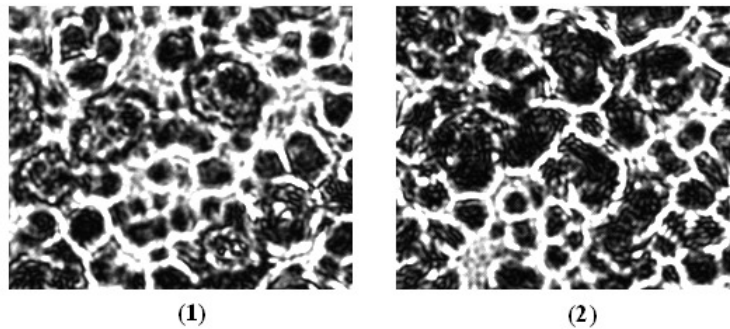

(1)   **(2)**

Fig. 2. Microscopic images of polycrystalline blood films healthy (fragment (1)) and patients with breast cancer (fragment (2)) of patients.

A comparative analysis of the reduced microscopic images revealed a polycrystalline structure of the blood films similar in orientation to the structure. The main optically anisotropic components of such a liquid are birefringent albumin, globulin and uniform elements. Due to linear birefringence, a coordinate distribution of the phase shifts between the linearly polarized orthogonal components of the laser radiation amplitude is formed.

5. Experimental results and their discussion

In fig. 3. polarization-interference images of polycrystalline blood films of healthy (fragment (1)) and a patient with cancer of the blood (fragment (2)) of patients are presented.
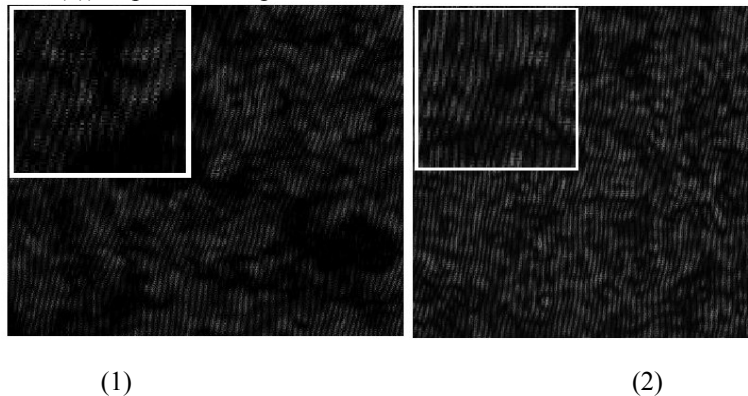

(1)   (2)

Fig. 3. Polarization-interference images of polycrystalline blood films of patients from group 1 (fragment (1)) and group 2 (fragment (2)).

In fig. 4 presents the results of the determination of the coordinate distributions of the local contrast value of polycrystalline blood films of healthy (fragments (1), (3)) and patients with breast cancer (fragment (2), (4)) of patients.
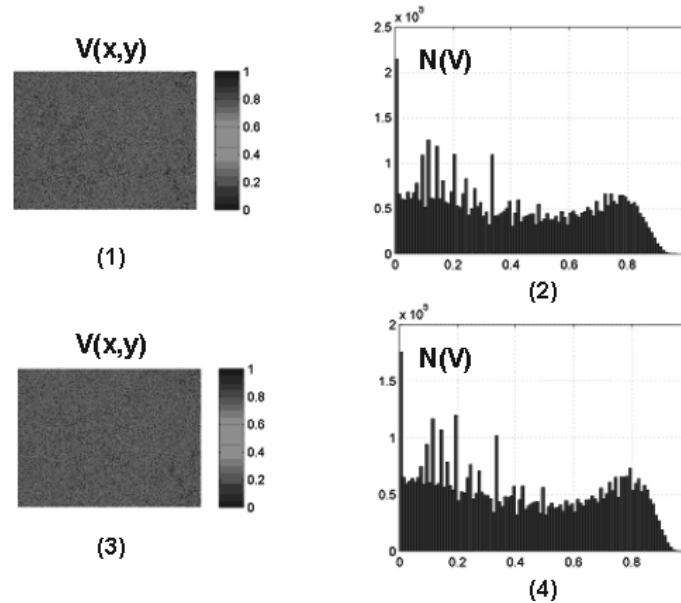

Fig. 4. Maps and histograms of the distribution of local contrast values of polarization-interference distributions in the plane of images of polycrystalline blood films of healthy (fragments (1), (3)) and patients with breast cancer (fragment (2), (4)) of patients.

A comparative analysis of the obtained maps and histograms of distributions of local contrast values of polarization-interference patterns of polycrystalline films of blood of both groups of patients revealed:

- practically the maximum range of changes in the value of local contrast $0 \leq V \leq 0.9$ (fig. 4, fragments (2), (4));
- histograms $N(V)$ of distributions of random values of local contrast V for both interference patterns have a complex asymmetric structure with the presence of a series of individual extrema that are in different parts of the interval of change V - $N_{max}(V) \approx 0$; $0.2 \leq N_{max}(V) \leq 0.35$; $N_{max}(V) \approx 0.8$ (Fig. 4, fragments (2), (4));
- for the histogram $N(V)$ calculated for the polarization-interference distribution in the plane of the microscopic image of the polycrystalline film of blood from group 2, there is a high probability of high local contrast values - the extremes of the minimum and maximum values V vary according to the scenario - $N_{max}(V) \approx 0 \downarrow$ and $N_{max}(V) \approx 0.8 \uparrow$ (Fig. 4, fragment (4)).

Table 1 shows the average values and mean errors of statistical ($Z_{i=1;2;3;4}$), which characterize the distributions of the local contrast W value of polarization-interference distributions (Fig. 4, fragments (2), (4)) of images of polycrystalline blood films of both groups of patients.

Table 1 Statistical parameters of local contrast distributions of polycrystalline blood films

Samples	Norm	Cancer	$Ac(W),\%$
Z_1	$0,092 \pm 0,0049$	$0,13 \pm 0,0074$	86
Z_2	$0,14 \pm 0,074$	$0,19 \pm 0,0089$	77
Z_3	$1,36 \pm 0,081$	$2,35 \pm 0,14$	92
Z_4	$0,87 \pm 0,051$	$0,51 \pm 0,029$	91

The conducted cycle of studies of the polarization-interference structure of microscopic images of polycrystalline films of blood was found to be excellent ($90\% \leq Ac(W) \leq 92\%$) the quality of the diagnostic test [28-30]).

Conclusion

A new method of polarization-interference mapping of polarization-inhomogeneous images of polycrystalline films of biological fluids with objective statistical analysis of local contrast maps is proposed and analytically justified.

Objective criteria characterizing maps of local contrast of interference distributions of images of polycrystalline blood films of healthy and patients with breast cancer patients are determined.

An analysis of the results of the diagnostic efficiency of the method of polarization-interference mapping of microscopic images of the investigated samples of polycrystalline films demonstrated an excellent ($Ac \succ 90\%$) accuracy of differential diagnostics of changes in the optical anisotropy of blood films.

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