DIAGNOSTIC POSSIBILITIES OF ANALYSIS OF THE MAP OF LINEAR BIREFRINGENCE OF THE CRYSTAL FRACTION OF VITREOUS BODY FOR ACCURATE DETERMINATION OF THE TIME SINCE DEATH

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Abstract

Introduction: This paper discusses the possibility of polarization microscopic tomography of polycrystalline structure of vitreous body (VB) for use in forensics and in determining the time since death (TSD).

Objectives: The purpose of this study was to develop a new set of forensic criteria to enhance the functionality of the high-precision definition of TSD over a long period of time according to polarization microscopic tomography of the polycrystalline structure of the VB of the human eye by statistical and wavelet analysis.

Results: We obtained the numerical values of the change in the magnitude of the statistical moments of the 1-4 orders, which characterize the coordinate distributions of the magnitude of the linear birefringence (LB) of the polycrystalline component of the VB by the magnitude of the TSD. The results from our research illustrate the differences between the optical anisotropy of fibrillar collagen networks of VB layers with different TSD. The sensitivity range (36 hours) and accuracy (15 minutes) of the method of polarization tomography of the LB distributions of the polycrystalline component of the VB layers in the determined TSD were established.

Conclusion: These results confirm the experimental processes outlined accurately determine the time of death. In turn, will provide scientific evidence, specifications thereof, and objective expert opinion.

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Keywords: Post-mortem interval, vitreous body, polarization.

Introduction

In contemporary forensics, a fundamental issue is the development and modernization of a more advanced method in the diagnosis of post-mortem intervals. Improving accuracy and reliability in determining the TSD would significantly facilitate the operative-search activities and bring further clarity to the circumstances of the case (Bonete et al., 2017; Duan et al., 2017).

As a visually descriptive method for the diagnosis of TSD, frequent studies are linked to the rate of cooling of the corpse, taking into account environmental conditions, assessment of the type and degree of development of post-mortem changes, and supravital reactions. However, the formation of post-mortem changes is affected by a large number of changing external and internal factors. In view of this, the expert judgment on the TSD has an interval character, the value of which is determined on the basis of a comparative analysis of the whole set of data received, and in each case can fluctuate within several hours or days (Garcia et al., 2017; Scrivano et al., 2019). The large degree of variation within existing post mortem assessment methods prompts researchers to find new approaches to solve this problem.

Ushenko et al. (2016); Garazdyuk et al. (2016); Vanchulyak et al. (2019) have shown the possibilities of medico-physical methods of investigating the relationship between necrotic changes in the structure of biological tissues of different morphological structure, as well as causes and prescription of death.

Purpose

The purpose of the study was to develop an objectively reliable forensic methodology to enhance the functionality of the high-precision definition of TSD over a long period of time according to

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polarization microscopic tomography of the polycrystalline structure of the VB of the human eye by statistical and wavelet analysis.

Materials and Methods

The object of the study was native layers of the VB, selected from 79 corpses of both sexes, aged 27 to 73 years of age, with a known time of death that ranged between 1 and 40 hours.

The VB was collected under mixed lighting conditions, varying air temperatures +18-22 °C and a relative humidity of 60-75%. Sampling was performed with an insulin syringe (needle was inserted in the area of the outer corner of the eye) in the amount of 0.25 ml in people who died of cardiovascular pathology with a known time of death (n=76) at different intervals (within the first 6 hours after death, measurements were taken every 15 min, from 6 to 40 hours at 1 hour intervals).

The exclusion criteria were the presence of traumatic brain injury and eyeball trauma and laboratory confirmation of any exogenous intoxication.

Optical Circuit and Method of Experimental

Measurements of the coordinate distributions of the magnitude of LB of VB preparations were performed using the classical Stokes polarimeter (Figure 1) according to Ushenko (2015).

Laser sounding of the VB layers (4) was performed using formed by a collimator (2), parallel (2 mm diameter) beam of a gas He-Ne laser (1) with a wavelength $\lambda=0.63\text{ mcm}$.

The formation of the states of polarization of the laser probe was carried out using phase-shifting (0.25λ) plates (3, 5) (manufacturer - Achromatic True Zero-Order Waveplate) and a linear polarizer (6) (manufacturer - B+W Kaesemann XS-Pro Polarizer MRC Nano).

VB layer (4) was sequentially illuminated by a series of laser beams with different polarization states. The formation of a set of linear polarizations was carried out by orienting the axis of the highest velocity of the phase-shifting element (3) by an angle of + 45° relative to the polarization plane of the laser radiation, followed by rotation of the plane of transmission of the polarizer at angles 0°, 90°, +45°.

Microscopic images of VB layers (using a polarizing micro lens (5) (manufacturer - Nikon CFI Achromat P, focal length – 30mm, numerical aperture – 0.1, magnification – 4x) projected into the plane of a digital camera (7) (The Imaging Source DMK 41AU02.AS, monochrome 1/2" CCD, Sony ICX205AL (progressive scan); resolution – 1280x960; the size of the photosensitive pad – 7600x6200mcm; sensitivity – 0.05 lx; dynamic range – 8 bit, SNR – 9 bit), photosensitive pad which contains $m \times n = 1280 \times 960$ pixels.

Polarization filtering of coordinate intensity distributions of microscopic images of VB layers (4) was carried out by means of a polarizing filter (6) (linear polarizer with angles of the transmission plane 0°, 90°, 45°, 135°) and the quarter-wave plate, the angle of orientation of the axis of the highest velocity was 45°, 135°).

Methods of calculation within each pixel of the photosensitive pad of the digital camera (7) of the set of parameters of LB of VB layers based on traditional Stokes-polarimetry algorithms, which are

superpositions of the intensity value of orthogonally-polarized laser radiation amplitude components for different states of the probe beam polarization. These are described in detail in a series of publications by OG Ushenko et.al (2016 a, b).

The results from these studies gave the temporal dynamics of necrotic changes in the coordinate distributions of the magnitude of the LB of the crystalline fraction of the VB layers of the human eye. (Figure 2-4).

Figure 2: Maps of the LB values of the crystalline fraction of the VB layers at the TSD 3 and 12 hours

Source: Authors

Figure 3: Histograms of the LB values of the crystalline fraction of the VB layers at the TSD 3 and 12 hours

Source: Authors

Figure 4: Distributions of the LB values of the crystalline fraction of the VB layers at the TSD 3 and 12 hours

Source: Authors

The results obtained illustrate the differences between the microscopic polarization tomography data of the LB polycrystalline component of the VB layers at different values of TSD. It is established that the coordinate distributions of the magnitude of the LB the sample of a layer of VB with a larger TSD (12 hours) is characterized by a smaller average value ($SM_1=0.0086$) and spread range ($SM_2=0.0048$) random values versus histograms ($SM_1=0.0068$, $SM_2=0.0034$), which obtained for LB polycrystalline component of the sample layers of VB with TSD 3 hours. (Figure 2).

The associated pattern was associated with the destructive necrotic changes in the structural anisotropy of the polycrystalline component of the VB layers, whose polarization manifestations are accompanied by a decrease in the magnitude of the LB, which is determined by the spatial ordering of the fibrillar collagen networks. As the posthumous interval increases, the integrity of the mesh structure of the CT is disturbed and the magnitude of the LB decreases.

Quantitatively, such necrotic transformation of fibrillary grids of polycrystalline structure of layers of VB of a human eye with different TSD is illustrated by mean values and errors within groups, statistical moments of 1 - 4 orders, which characterize the coordinate distributions of LB (Table 1).

Table 1: Temporal dynamics of changes in the magnitude of the statistical moments of 1–4 orders ($SM_{i=1;2;3;4}$), which characterize the distribution of the magnitude of the LB crystalline fraction of the VB layers with different TSD (T, hours).

$SM_i \times 10^{-2}$	$T=1$	$T=3$	$T=6$	$T=12$	$T=18$	$T=24$
SM_1	0.92± 0.043	0.86± 0.038	0.81± 0.035	0.68± 0.031	0.56± 0.025	0.41± 0.019
p	$p < 0.05$	$p < 0.05$	$p < 0.05$	$p < 0.05$	$p < 0.05$	$p < 0.05$
SM_2	0.53± 0.026	0.48± 0.024	0.43± 0.021	0.34± 0.015	0.24± 0.011	0.14± 0.006
p	$p < 0.05$	$p < 0.05$	$p < 0.05$	$p < 0.05$	$p < 0.05$	$p < 0.05$
SM_3	0.71± 0.036	0.82± 0.041	0.92± 0.045	1.13± 0.055	1.34± 0.059	1.54± 0.068
p	$p < 0.05$	$p < 0.05$	$p < 0.05$	$p < 0.05$	$p < 0.05$	$p < 0.05$
SM_4	1.21± 0.054	1.35± 0.058	1.49± 0.065	1.76± 0.073	2.04± 0.092	2.32± 0.098
p	$p < 0.05$	$p < 0.05$	$p < 0.05$	$p < 0.05$	$p < 0.05$	$p < 0.05$

Source: Authors

The analysis of the given data revealed:

- the linear range of necrotic change in the magnitude of the statistical moments 1 - 4 orders, which characterize the distribution of the magnitude of the LB of fibrillar collagen nets of the VB layers by the magnitude of the TSD is 24 hours;
- value of the statistical moment of the 1st order SM_1 , which characterizes the mean distribution of LB varies from 0.92 to 0.41;
- value of the statistical moment of the 2nd order SM_2 , which characterizes the dispersion of LB distributions varies from 0.53 to 0.14;
- value of the statistical moment of the third order SM_3 , which characterizes the asymmetry of LB distributions varies from 0.71 to 1.54;
- value of the statistical moment of the 4th order SM_4 , which characterizes the excess of LB distributions varies from 1.21 to 2.32.

This data from the statistical analysis of polarized reproducible LB distributions shows that the magnitudes of the mean, dispersion, asymmetry and excess, which characterize the maps of the fibrillar collagen nets of the polycrystalline component of the VB layers of the dead with different TSD changed linearly within 24 hours.

The most sensitive to necrotic changes in the polycrystalline structure of the layers of CT were the temporal changes of SM_3 and SM_4 , which characterize the polarized coordinate distributions of the magnitude of the LB. Quantitatively, this is witnessed by the increase of the angles of inclination of the corresponding linear time dependences of necrotic changes in the magnitude of the indicated statistical moments of higher orders.

We have obtained the following results of the accuracy of determination of TSD by the method of microscopic polarization tomography of LB collagen grids of VB layers with different TSD, – table 2

The analysis of the results in Table 2 revealed that for the excess SM_3 of LB distributions, the maximum level (highlighted in gray) of the method of microscopic polarization tomography of the LB collagen mesh layers of the VB layers in determining TSD within 19 - 21 min was reached.

In our previous studies, diagnostic efficacy (extension of the time interval of determination of TSD) and improvement of accuracy by using a scale-selective wavelet analysis were found.

Table 2: Accuracy ($\pm\Delta T$) for the determination of TSD by the method of microscopic polarization tomography of LB collagen mesh layers

SM_1	$T=1$	$T=3$	$T=6$	$T=12$	$T=18$	$T=24$
SM_2	40 min	41 min	41 min	40 min	42 min	43 min
SM_3	36 min	38 min	37 min	38 min	38 min	38 min
SM_4	24 min	25 min	24 min	25 min	25 min	25 min
SM_4	19 min	20 min	20 min	20 min	21 min	21 min

Source: Authors

Figure 5: Maps $W_{a,b}$ of the wavelet coefficient amplitude $C_{15,b}$ LB values of the crystalline fraction of the VB layers at the TSD 3 hours

Source: Authors

Figure 6: Distributions the magnitude of the wavelet coefficient amplitude $C_{15,b}$ LB values of the crystalline fraction of the VB layers at the TSD 3 hours

Source: Authors

Figures 5 - 8 show the results of analytic calculations of the wavelet coefficient amplitude maps $K_{a,b}$, that characterize the large-scale structure ($a=15$; $a=55$) of optically anisotropic LB of collagen grids of layers of VB layers with different TSD (3 and 12 hours).

Comparative analysis of the results of the study of the dependencies of the amplitudes of the wavelet coefficients $W_{a,b}(x,y)$, that characterize the different scales of structural elements of LB maps of collagen grids of VB layers with different TSD, revealed maximum differences for small scales a_{min} wavelet functions. This is due to the fact that necrotic changes in the fibrillar collagen networks mainly occur on a relatively small scale.

Quantitatively, necrotic transformations of the fibrillar structure of samples of VB layers with different TSD illustrate the variation of the statistical moments of the 1–4 th order characterizing the amplitude distribution of the wavelet coefficients $C_{a=15,b}$ of the maps of LB (Table 3).

Figure 7: Maps $W_{a,b}$ of the wavelet coefficient amplitude $C_{15,b}$ LB values of the crystalline fraction of the VB layers at the TSD 12 hours

Source: Authors

Figure 8: Distributions of the magnitude of the wavelet coefficient amplitude $C_{15,b}$ LB values of the crystalline fraction of the VB layers at the TSD 12 hours

Source: Authors

Table 3: Temporal dynamics of change of magnitude of statistical moments of 1 - 4 orders ($SM_{i=1;2;3;4}$), which characterize the distributions of the linear section $C_{15,b}$ maps of wavelet coefficients $W_{a,b}$ the magnitude of the LD of the crystalline fraction of the layers of VB with different TSD (T, hours)

SM_i	$T=1$	$T=6$	$T=12$	$T=18$	$T=24$	$T=36$
SM_1	0.59 ± 0.023	0.45 ± 0.019	0.31 ± 0.014	0.16 ± 0.007	0.013 ± 0.006	0.004 ± 0.002
p	$p < 0.05$	$p < 0.05$	$p < 0.05$	$p < 0.05$	$p < 0.05$	$p < 0.05$
SM_2	0.45 ± 0.021	0.38 ± 0.016	0.32 ± 0.014	0.25 ± 0.011	0.19 ± 0.009	0.054 ± 0.002
p	$p < 0.05$	$p < 0.05$	$p < 0.05$	$p < 0.05$	$p < 0.05$	$p < 0.05$
SM_3	0.31 ± 0.014	0.54 ± 0.024	0.78 ± 0.033	1.01 ± 0.044	1.25 ± 0.054	1.71 ± 0.077
p	$p < 0.05$	$p < 0.05$	$p < 0.05$	$p < 0.05$	$p < 0.05$	$p < 0.05$
SM_4	0.38 ± 0.016	0.72 ± 0.031	1.05 ± 0.044	1.39 ± 0.054	1.72 ± 0.065	2.39 ± 0.108
p	$p < 0.05$	$p < 0.05$	$p < 0.05$	$p < 0.05$	$p < 0.05$	$p < 0.05$

Source: Authors

Analysis of the above data increased for a period up to 36 hours, of the linear range of determination of TSD - changes in the magnitude of the set of all statistical moments of 1 - 4 orders, which characterize the maps of wavelet coefficients $C_{a=15,b}$ distribution of LB collagen mesh layers of CT.

The following statistical parameters are defined:

- average value SM_1 , which characterizes the amplitude distributions of the wavelet coefficients $C_{a=15,b}$ map of LB, varying within the range of average values from 0.59 to 0.004 defined for all groups of samples;
- the magnitude of the variance SM_2 , which characterizes the amplitude distributions of the wavelet coefficients $C_{a=15,b}$ map of LB, varies within the range of sample values defined for all groups from 0.45 to 0.054;
- asymmetry SM_3 , which characterizes the amplitude distributions of the wavelet coefficients $C_{a=15,b}$ map of LB, varies within the range of sample values defined for all groups from 0.31 to 1.71;

excess SM_4 map $\Pi\Pi$, which characterizes the amplitude distributions of the wavelet coefficients $C_{a=15,b}$, varies within the range of sample values defined for all groups from 0.38 to 2.39.

The obtained data show that the size of all the statistical moments characterizing the magnitude distributions of the wavelet coefficients of the maps of the polarized reproduced LB of the collagen networks of the polycrystalline component of VB layers of biomannequins with different TSD vary linearly within a 36 hour timeframe. At the same time, the most sensitive to necrotic changes in the fibrillar structure of collagen fibers of such samples, such as in the case of statistical analysis of microscopic polarization tomography data, were only temporary changes in the magnitude of the statistical moments of the 3-4th orders (SM_3 , SM_4). Quantitatively, this is witnessed by the increase of the angles of inclination of linear dependences of the magnitude of higher-order statistical moments, which characterize the magnitudal distributions of the wavelet coefficients of maps of polarized reproducible LB of collagen networks in comparison with similar parameters of map of polarization manifestations of collagen structures.

This increases the sensitivity of the wavelet analysis method and improves by 5 minutes. The accuracy of determination of TSD (Table 4).

Table 4: Accuracy ($\pm\Delta T$) of determination of TSD by wavelet analysis of polarization reproduction of LB distributions of polycrystalline component of VB layers

SM_1	$T=1$	$T=6$	$T=12$	$T=18$	$T=24$	$T=36$
SM_2	35 min	36 min	36 min	36 min	37 min	37 min
SM_3	31 min	32 min	32 min	32 min	33 min	33 min
SM_4	18 min	19 min	19 min	20 min	20 min	21 min
SM_4	14 min	15 min	15 min	15 min	15 min	16 min

Source: Authors

Analysis of the research data on the time dependencies of necrotic changes in the magnitude of the set of statistical moments of 1-4 orders, which characterize the magnitude distributions of the wavelet coefficients of the small-scale structure of the coordinate distributions of the magnitude of the LB collagen networks of layers of the VB layers at different TSD intervals, found the maximum level (highlighted in gray) of accuracy in the determination of TSD within a period of 14 to 16 minutes.

Conclusion

The range of sensitivity (24 hours) and accuracy (20 minutes) of the method of polarization tomography of the LB distributions of the polycrystalline constituent of the VB layers in the determining TSD were proven.

It has been clearly demonstrated that scale-selective wavelet analysis provides, inter alia, an increased sensitivity range to 36 hours and an increased accuracy of the determination of TSD by the method of polarization tomography of the distributions of LB of the polycrystalline component of the VB layers in the determined TSD up to 15 minutes.

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