

## Post-mortem interval estimation by changes in the optical density of the vitreous humour

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### Abstract

**Introduction:** One of the most urgent tasks of forensic science and practice for many years, both in Ukraine and abroad, remains the exact estimation of time since death (TSD).

**Aim:** To develop a complex of forensic objective criteria to improve the accuracy of TSD estimation at the long-term time intervals on the basis of complex direct and large-scale selective statistical data analysis of the polarization-correlation mapping of microscopic images of polycrystalline films of vitreous body (VB) from a human cadaver.

**Material and methods:** The object of study is polycrystalline films of VB, taken from 52 cadavers of both sexes aged from 35 to 81-year with pre-known time of death coming ranged from 1 to 24 h. Measuring the coordinate allocation (two-dimensional measurements in the plane of the samples of polycrystalline films of VB) meanings of parameters of polarization in the points of microscopic images was carried out at the location of the standard Stokes-polarimeter.

**Results and conclusions:** The analysis of the data obtained on the time dependences of the magnitude of the set of statistical moments of the 1st to 4th orders, which characterize the distributions of phase parameters at different gaps in the TSD, revealed the maximum accuracy of the Stokes polarimetric mapping of the VB layers in the determination of the TSD in the range of 50-51 min.

**Key words:** time since death estimation, vitreous body, polarising microscopy

### Stanovenie času smrti na základe zmien optickej denzity očného sklovca

#### Abstrakt

**Úvod:** Jednou z najnaliehavejších úloh súdnolekárskej vedy a praxe po mnoho rokov, tak na Ukrajine, ako aj v zahraničí, zostáva presné stanovenie času smrti.

**Cieľ:** Vypracovať komplex objektívnych forenzných kritérií na zlepšenie presnosti odhadu času smrti v dlhodobých časových intervaloch na základe komplexnej priamej a rozsiahlej selektívnej štatistickej analýzy údajov polarizačno-korelačného mapovania mikroskopických snímok polykryštalických filmov očného sklovca odobratého pri pitve.

**Materiál a metódy:** Predmetom štúdia sú polykryštalické filmy očného sklovca, odobraté z 52 mŕtvych tiel oboch pohlaví vo veku od 35 do 81 rokov s vopred známym časom úmrtia od 1 do 24 hodín. Meranie alokácie súradníc (dvojmerné merania v rovine vzoriek polykryštalických filmov očného sklovca) významu parametrov polarizácie v bodoch mikroskopických obrazov sa uskutočnilo štandardným Stokesovým polarimetrom.

**Výsledky a závery:** Analýza získaných údajov o časových závislostiach veľkosti súboru štatistických momentov 1. až 4. rádu, ktoré charakterizujú rozdelenie fázových parametrov pri rôznych intervaloch času po smrti, odhalila maximálnu presnosť Stoksovho polarimetrického mapovania vrstiev očného sklovca pri stanovení času smrti v rozsahu 50 - 51 min.

**Kľúčové slová:** stanovenie času smrti, očný sklovec, polarizačná mikroskopia

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## Introduction

One of the most urgent tasks of forensic science and practice for many years, both in Ukraine and abroad, remains the exact establishment of the time since death (TSD) (1-14). The high accuracy of the TSD estimation is an important goal of the medical and legal investigation and a guarantee of the successful disclosure of crimes against the life and health of citizens. However, at present, the most accessible methods for determining the TSD for a forensic expert-practitioner are the visual assessment of posthumous changes, which are a macroscopic reflection of the biochemical and biophysical processes occurring in the human body after death. These include diagnosis of livores mortis, cooling and drying of the body, rigor mortis, assessing of supravital reactions, etc. They are very informative and of great forensic significance, but in some cases they do not allow us to reliably answer the questions posed to the expert. After all, natural biological as well as artefact changes that occur in the human body after death are complex and unpredictable, as these phenomena are affected by a wide range of variables.

Modern physico-optical methods of diagnostics of the TSD are very perspective, since they are based on the use of hardware computer technology, which allows to obtain objective data of the dynamics of posthumous changes of the studied biological tissues and human environments and, accordingly, more precisely determine the TSD (1-3). Distinctive feature of biophysical methods of research is the high sensitivity and the possibility of accurate, objective recording of the obtained results.

## Aim

To develop a complex of forensic objective criteria to improve the accuracy of TSD estimation at the long-term time intervals on the basis of complex direct and large-scale selective statistical data analysis of the polarization-correlation mapping of microscopic images of polycrystalline films of vitreous body (VB) from a human cadaver.

## Material and methods

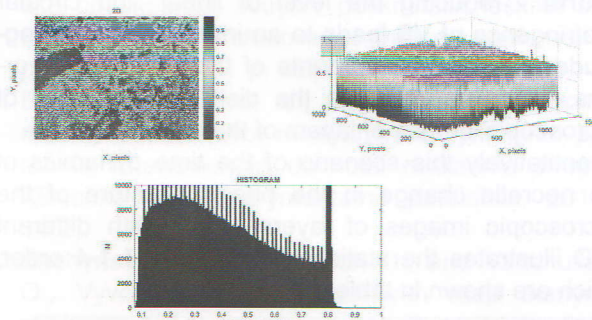
The object of study is polycrystalline films of VB, taken from 52 cadavers of both sexes aged from 35 to 81-year with pre-known time of death coming ranged from 1 to 24 h. Concerning the time that passed after death, the selection is as follows: 1h - 5 cases (9.6 %), 3h - 8 cases (15.4 %), 6h - 10 cases (19.2 %), 12h - 8 cases (15.4 %), 18h - 10 cases (19.2 %), 24h - 11 cases (21.2 %).

Measuring the coordinate allocation (two-dimensional measurements in the plane of the samples of polycrystalline films of liquor) meanings of parameters of polarization in the points of microscopic images was carried out at the location of the standard Stokes-polarimeter (6,7).

The results of the statistical analysis of the time distribution dynamics of the fourth parameter of the Stokes vector (hereinafter "phase parameter" - PP (4)) of the microscopic images of VB layers of the deceased persons with the severity TSD interval are presented. Experimental measurements of Stokes-parametric images of biological layers were carried out according to the method presented in (5-7).

On the Fig. 1 the maps (1) and distributions ((2), (3)) of the magnitudes of the PP of the Stokes vector of the microscopic images of VB layers with TSD 3 hours are shown.

The obtained results illustrate the existence of differences between the data of azimuthally invariant polarization microscopy of layers of VB of the deceased with different TSD. Distributions of the PP values (Fig. 1, fragments (2), (3)) of the sample of the deceased with a larger TSD (3 hours) are characterized by lower average values (the statistical moment of the 1st order) and the range of distribution (statistical moment of the 2nd order) of random values.



**Fig. 1** Map (1) and distributions ((2), (3)) of the magnitude of the PP of the Stokes vector of the microscopic images of VB layers with TSD 3 hours

This fact can be related to the results of polarization mapping of coordinate distributions of the magnitude of Stokes vector parameters of microscopic images of histological sections of biological tissues of different organs of a person (8-10). Here it is shown that the main factor in the formation of the coordinate structure of the distribution of the Stokes vector parameters at the points of polarization microscopic images is the optical anisotropy of biological objects. This mechanism leads to the appearance of phase shifts between the orthogonal components of the amplitude of laser radiation at the points of the microscopic image (distribution of the magnitudes of PP - Fig. 1, fragments (1), (2)) of the VB layer. Such phase distributions in the microscopic image are formed due to the influence of the following factors:

- structural anisotropy of a spatially ordered network of collagen fibrils - linear birefringence;
- "islet" anisotropy of molecular protein complexes of protein fraction of VB - circular birefringence;
- multiple scattering of laser radiation in the volume of optical anisotropic layer of VB.

Due to the latter factor, averaging of the distribution of the magnitude of the polarization parameters of

the Stokes vector and the reduction of phase modulation in the plane of the microscopic image of the sample of the VB are present. Proceeding from this, we can formulate the following scenario of the dynamics of the transformation of polarization manifestations of necrotic changes in the polycrystalline structure of layers in case of different TSD interval.

**First** - with an increase of the TSD, the level of optical anisotropy decreases. The disordered collagen network is reduced and the concentration of protein complexes decreases.

**Secondly** - the optically process of necrotic changes is manifested in reducing the depth of phase modulation of laser radiation by polycrystalline structures of the VB.

**Thirdly** - the statistically increasing degree of necrotic degradation of the polycrystalline component of VB is accompanied by a decrease in the mean and dispersion of the coordinate distributions of the size of the PP (Fig. 1, fragments (2), (3)) of microscopic images of biological preparations.

**Fourth** - reducing the level of linear and circular birefringence of VB leads to an increase in the magnitude of statistical moments of the 3rd and 4th orders, which characterize the distributions of PP of microscopic images of layers of the deceased.

Quantitatively this scenario of the time dynamics of the necrotic change in the phase structure of the microscopic images of layers of VB with different TSD illustrates the statistical moments of 1-4 order, which are shown in table 1:

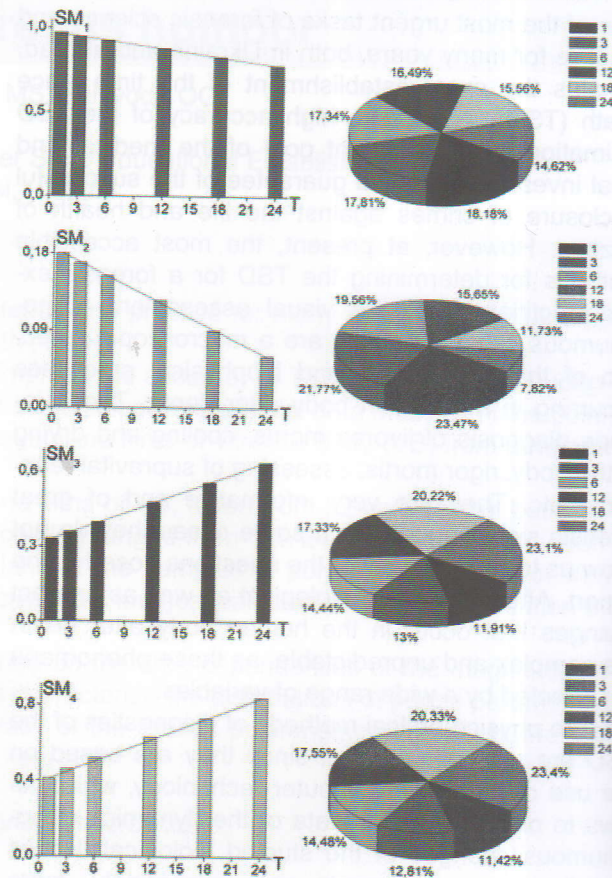
SM <sub>i</sub>	T=1	T=3	T=6	T=12	T=18	T=24
SM <sub>1</sub>	0.98 ± 0.045	0.93 ± 0.043	0.88 ± 0.041	0.83 ± 0.037	0.81 ± 0.036	0.78 ± 0.035
p	p π 0,05	p π 0,05	p π 0,05	p π 0,05	p π 0,05	p π 0,05
SM <sub>2</sub>	0.18 ± 0.008	0.165 ± 0.007	0.15 ± 0.006	0.12 ± 0.005	0.09 ± 0.004	0.06 ± 0.003
p	p π 0,05	p π 0,05	p π 0,05	p π 0,05	p π 0,05	p π 0,05
SM <sub>3</sub>	0.33 ± 0.014	0.37 ± 0.016	0.41 ± 0.018	0.49 ± 0.021	0.57 ± 0.023	0.64 ± 0.028
p	p π 0,05	p π 0,05	p π 0,05	p π 0,05	p π 0,05	p π 0,05
SM <sub>4</sub>	0.41 ± 0.018	0.46 ± 0.021	0.52 ± 0.024	0.63 ± 0.028	0.73 ± 0.034	0.84 ± 0.038
p	p π 0,05	p π 0,05	p π 0,05	p π 0,05	p π 0,05	p π 0,05

**Tab. 1** The time dynamics of the change in the magnitude of the statistical moments of the 1st to 4th orders (SM<sub>i</sub>), that characterize the distributions of the PP of the Stokes vector of the microscopic images of the VB layers of the deceased with different TSD (T, h)

Installed:

- the linear range of the change in the magnitude of the statistical moments of the 1st to 4th orders, which characterizes the distributions of the PP of the microscopic images of the layers of the deceased by the size of the TSD, is 24 hours;
- the value SM<sub>1</sub> varies within the range of the average values set for all groups from 0.98 to 0.78;
- the value SM<sub>2</sub> varies within the range of the average values set for all groups from 0.06 to 0.18;
- the value SM<sub>3</sub> varies within the range of the average values set for all groups from 0.33 to 0.64;
- the value SM<sub>4</sub> varies within the range of the average values set for all groups from 0.41 to 0.84.

Fig. 2 illustrates linear and circular diagrams for changing the value of a set of statistical moments :



**Fig. 2** Time charts of the change in the magnitude of the statistical moments of the 1st-4th order (SM<sub>i</sub>), that characterize the distributions of the PP of the Stokes vector of the microscopic images of VB layers of the deceased with different TSD (T, h.)

From the data obtained (Fig. 2) can be seen that the mean (1), dispersion (2), asymmetric (3) and kurtosis (4), describing Stokes-polarimetric maps of microscopic images of VB layers with a different TSD demonstrate linear change in within 24 hours. The most sensitive to necrotic changes in the polycrystalline structure of such samples were time changes in asymmetry and kurtosis .

Quantitatively this is manifested in the growth of the angles of inclination of such linear dependencies of statistical moments of higher orders.

We obtained the following results for the method of azimuthally invariant polarization microscopy of layers of decayed with different TSD – see table 2:

SM <sub>i</sub>	T=1	T=3	T=6	T=12	T=18	T=24
SM <sub>1</sub>	58 min	60 min	59 min	63 min	62 min	66 min
SM <sub>2</sub>	56 min	58 min	57 min	58 min	60 min	62 min
SM <sub>3</sub>	53 min	54 min	55 min	54 min	55 min	55 min
SM <sub>4</sub>	50 min	51 min	51 min	52 min	51 min	51 min

**Tab. 2** Accuracy (T) of TSD estimation by the method of Stokes polarimetric mapping of VB layer

The analysis of the data obtained on the time dependences of the magnitude of the set of statistical moments of the 1st to 4th orders that characterize the

distributions of the PP at different intervals of the TSD revealed the maximum level (highlighted by grey colour) of the accuracy of the Stokes polarimetric mapping of the VB layers in the estimation of the TSD in the range of 50-51 min. This level corresponds to the temporal parameters of the TSD estimation, which were received by the scientists of the prof. Bachinsky V.T. school from the Bukovinian State Medical University (11-14).

### Conclusions

1. The method of azimuthally-invariant Stokes-polarimetric mapping of a set of polarization maps and histograms of distributions of random values of the phase parameter of microscopic images of layers of deceased persons with different time since death intervals was investigated.
2. The temporal dynamics of the change in the magnitude of the statistical moments of the 1st-4th order, which characterizes the distributions of the phase parameter of the polarization microscopic images of the layers of the vitreous body in case of different time since death intervals was investigated.
3. The sensitivity range (24 h) and the accuracy (50 min) of the Stokes polarimetry method of the microscopic images of the vitreous body layers in case of different time since death intervals were established.

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