ANALYSIS OF SPECTROSCOPIC METHODS OF DETECTION OF STRUCTURAL AND FUNCTIONAL STATES OF HEMOGLOBIN IN RBCS

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Spectroscopic methods of detection of structural and functional states of hemoglobin in RBCs are considered. It should be noted that in recent years the SERS method has been actively developing. Using this method, can detect various forms of Hb including in single live erythrocytes; selective detection of Hb bound to erythrocyte membranes.

Keywords: erythrocytes, hemoglobin, methods of detection of structural and functional states.

Red blood cells (RBCs), also known as erythrocytes, are normally biconcave disks of diameter 7–9 μm. An erythrocyte is the thickest (1,7–2,4 μm) at the edges and the thinnest (0,9–1,2 μm) at the center. RBCs are able to aggregate, stick to each other by the surface of the disks, thus, forming a rouleau. The number of RBCs in the aggregate can vary from a few to hundreds of pieces. The change of the degree of aggregation, size and shape of RBCs can indicate blood disorders. Structure and functional state of RBCs and quantity of various forms of hemoglobin (Hb) in them are the most important indicators reflecting the state of the body. The main function of RBCs is the transport oxygen (O_2). This function depends on the functional state (conformation) of Hb, i.e. affinity of Hb for O_2. The concentration of hydroxy-Hb, deoxy-Hb, carboxy-Hb and meth-Hb is determined in the blood. Methods of microscopy, optical, microwave, and Mossbauer spectroscopy are widely used to solve the mentioned and other specific analytical problems.

The purpose of this work was a comparative study of the unique analytical capabilities of various methods for the detection of structural and functional states of RBCs and various forms of Hb in them. The results of the studies are tabulated. It should be noted that in recent years the SERS method has been actively developped.

Method name	Physical principle	Analytical capabilities
Optical microscopy	The phenomena of refraction and reflection of light at the interface between media are used.	Measurement of the shape and size of erythrocytes.
Optical spectrophotometry	Resonance absorption of light due to electronic transitions in molecules.	Determination of the concentration of various forms of Hb in the concentration range $>10^{-6}$ M
Raman scattering (RS)	Displacement of frequency of light scattered by molecules as a result of the interaction of laser radiation with intramolecular vibrations.	Determination of the concentration of various forms of Hb in the concentration range $>10^{-3}$ M
Surface enhanced Raman scattering (SERS)	Enhance of Raman scattering as a result electromagnetic and chemical interaction of molecules with a nanostructured surface of noble metals.	Detection of various forms of Hb including in single live erythrocytes; selective detection of Hb bound to erythrocyte membranes.
Nuclear magnetic resonance (NMR)	Resonant absorption of electromagnetic radiation by atomic nuclei in magnetic fields.	Study and integral control of the structure of erythrocyte membranes.
Mossbauer Spectroscopy	Resonant absorption of the gamma radiation of a moving source by atomic nuclei of the sample.	Determination of the concentration of various forms of Hb on the basis of the determination of the quantum state of the iron ion.