Combination of laser correlation and dielectric spectroscopy in albumin investigations

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Abstract. Joint use of laser correlation and dielectric spectroscopies for studies of biomolecular properties of albumin in water solution is considered. The conditions and parameters of the experiments are discussed. Similar behaviors of albumin molecular sizes and maximum frequency of peak of dielectric dissipation factor with increasing acidity were revealed. Using the suggested approach, biomolecular aggregation dynamics and changes in electrophysical properties on transition from one molecular structure to another may be investigated.

1. Introduction

Biomolecular objects attract an ever increasing attention of researches in different areas [1,2]. The interest is caused by a wide range of possible application of biomolecular technologies — from diagnostics and drug manufacturing in medicine to development of new elements and devices in molecular electronics [3–6].

Biomolecular technologies are invaluable in modern chemical biology and medicine, helping to advance fundamental studies of biophysical interactions and recognition, drug discovery, and the translation of new insights into clinical application [7]. For goals of molecular electronics biomolecules have the advantages of functionality, high-density, low-consumption and specificity [3]. Biomolecular electronics is considered to be an alternative or supplement to microelectronics in continuing miniaturization. A number of researches believe that biomolecular materials have great promise for potential applications in electronics, optoelectronics, nonlinear optics; and conductive materials for electronics [3, 8]. Amino acids and proteins are considered to be the most promising materials and investigation of their electrophysical properties is the actual task.

One of the objects of research in our study is human serum albumin — the globular protein Albumin is the most abundant protein in human blood plasma which transports hormones, fatty acids, and other compounds, buffers pH, and maintains osmotic pressure, among other functions.

It is known that shift in pH cause conformational changes in protein molecules and their dysfunction. So, investigation of changes in molecular structure and dielectric features under the influence of different factors including pH can give valuable information. There are a lot of different methods of investigation of molecular structure and properties but electrophysical properties of proteins are understudied.
The goal of our study was to estimate molecular aggregation dynamics and to analyze the changes of electrophysical properties of albumin depending on solution acidity by joint use of laser correlation and dielectric spectroscopies.

2. Methods and objects
We used combination of two methods — laser correlation spectroscopy (LCS) and dielectric spectroscopy in frequency range 20 Hz – 1 MHz.

2.1. Samples
In our experiments we used the solutions of human serum albumin in distilled water. The concentration of albumin was 0.01. The acidity of the solution was changed by addition of acid and alkaline buffers in the range pH = 1.4 – 8.2.

2.2. Laser correlation spectroscopy
The measure procedure of laser correlation spectroscopy was described in [9]. This method was used for evaluation of molecular size and dynamics of molecular aggregation in solution.

The experimental setup is presented in figure 1.

![LCS setup](image)

**Figure 1.** LCS setup. 1 — junction laser, 2 — optical cell with water suspension of biomolecules, 3 — filter, 4 — photomultiplier, 5 — rotating table, 6 — oscilloscope, 7 — computer with software for correlation and statistical analysis.

In the setup, a coherent light beam from a junction laser (1) (KLM-G650-13-5) was passed through the optical cell with a water suspension of particles to be studied (2). The scattered light at a scattering angle from 20 to 160 degrees (in figure 1, the angle is 90 degrees) was detected by a photomultiplier (4) (H11706-01). The signal from the photomultiplier was transmitted to the oscilloscope (6) and computer (7) for further processing.
2.3. Dielectric spectroscopy method

The dielectric spectroscopy (DS) method occupies a special place among the numerous modern methods used for physical and chemical analysis of material, because it enables investigation of dielectric relaxation processes. Intermolecular interactions and cooperative processes may be monitored by help of dielectric spectroscopy.

The experimental setup consisted of a special liquid cell with two pole plates and LCR meter “MNIP E7-20” (figure 2).

![Figure 2. Experimental setup for DS measurements.](image)

1 — LCR-meter, 2 — optical cell with water suspension of biomolecules, 3 — micrometer, 4 — coaxial wires, 5 — moving and motionless electrodes.

The albumin solution was placed in the optical cell (2). The distance between two flat electrodes was fixed and controlled by micrometer (3). The impedance $Z$ and phase angle $\phi$ were detected by LCR-meter. For further analysis the dielectric dissipation factor was calculated:

$$tg\delta = \frac{1}{\omega R C},$$

where

$$R = \frac{Z}{\cos(\phi)}, \quad C = \frac{\sin(\phi)}{\omega Z}.$$ 

The dielectric dissipation factor dependences on frequency in different conditions were studied. It was revealed that albumin solution has peaks of dielectric dissipation factor, and the peak is corresponds to different frequencies depending on pH of solution.

3. Results and discussion

The results of our study revealed similar behaviors of two characteristics: the size of the molecules in the solution (measured by LCS) and frequencies of maximum dielectric dissipation factor peaks of albumin (measured by DS) depending on acidity of solution. The obtained results are presented in figures 3 and 4.
Figure 3. Dependence of molecular sizes of albumin on solution acidity

Figure 4. Dependence of frequencies of maximal dielectric loss angle tangent on acidity of albumin solution.

The maximal sizes of albumin molecules and maximal frequencies of dielectric loss angle tangent peaks were observed near the isoelectric point of albumin (pH = 4.8). This may be concerned with forming of special molecular structures in isoelectric solution.
4. Conclusion
Joint use of laser correlation and dielectric spectroscopies for studies of biomolecular properties is presented. Using the suggested approach, biomolecular aggregation dynamics and changes in electrophysical properties on transition from one molecular structure to another may be investigated.

References
[1] www.scopus.com/results/results.url?sort=plf-f&src=s&st1=biomolecular+electronics 2015
[2] Liao C.-D., Tsai T.C. et al. 2015 Device Architecture and Biosensing Applications for Attractive One- and Two-Dimensional Nanostructures, ed. M.C. Vestergaard Nanobiosensors and nanobioanalyses (Japan: Springer) p.41
[3] Phadke R.S. 2001 Applied Biochemistry and Biotechnology 96 279
[4] Cingolani R, Rinaldi R., Maruccio G., Biasco A. 2002 Phisica E 13 1229
[5] Maruccio G. et al 2004 Electroanalysis 16 (22)
[6] Jiang W. et al 2011 J. Am. Chem. Soc. 133 1
[7] Kindt J.T, Bailey R.C 2013 Current Opinion in Chemical Biology 17 (5) 818
[8] Velichko E., Tsybin O. 2011 Biomolecular electronics (SPb, SPbSPU) 256 p.
[9] Nepomnyashchaya E, Velichko E, Aksenov E and Bogomaz T 2014 IOP Journal of Physics: Conference Series 541 012039