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Electrophoretic Light Scattering Based on Total Internal Reflection as a Promising Diagnostic Method

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Abstract : The development of pathological processes, such as cardiovascular and oncological diseases, are accompanied by changes in molecular parameters in cells, tissues, and serum. The study of the behavior of protein molecules in solutions is of primarily importance for diagnosis of such diseases. Various physical and chemical methods are used to study molecular systems. With the advent of the laser and advances in electronics, optical methods, such as scanning electron microscopy, sedimentation analysis, nephelometry, static and dynamic light scattering, have become the most universal, informative and accurate tools for estimating the parameters of nanoscale objects. The electrophoretic light scattering is the most effective technique. It has a high potential in the study of biological solutions and their properties. This technique allows one to investigate the processes of aggregation and dissociation of different macromolecules and obtain information on their shapes, sizes and molecular weights. Electrophoretic light scattering is an analytical method for registration of the motion of microscopic particles under the influence of an electric field by means of quasi-elastic light scattering in a homogeneous solution with a subsequent registration of the spectral or correlation characteristics of the light scattered from a moving object. We modified the technique by using the regime of total internal reflection with the aim of increasing its sensitivity and reducing the volume of the sample to be investigated, which opens the prospects of automating simultaneous multiparameter measurements. In addition, the method of total internal reflection allows one to study biological fluids on the level of single molecules, which also makes it possible to increase the sensitivity and the informativeness of the results because the data obtained from an individual molecule is not averaged over an ensemble, which is important in the study of bimolecular fluids. To our best knowledge the study of electrophoretic light scattering in the regime of total internal reflection is proposed for the first time, latex microspheres 1 μm in size were used as test objects. In this study, the total internal reflection regime was realized on a quartz prism where the free electrophoresis regime was set. A semiconductor laser with a wavelength of 655 nm was used as a radiation source, and the light scattering signal was registered by a pin-diode. Then the signal from a photodetector was transmitted to a digital oscilloscope and to a computer. The autocorrelation functions and the fast Fourier transform in the regime of Brownian motion and under the action of the field were calculated to obtain the parameters of the object investigated. The main result of the study was the dependence of the autocorrelation function on the concentration of microspheres and the applied field magnitude. The effect of heating became more pronounced with increasing sample concentrations and electric field. The results obtained in our study demonstrated the applicability of the method for the examination of liquid solutions, including biological fluids.

Keywords : light scattering, electrophoretic light scattering, electrophoresis, total internal reflection
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