Characterization of erythrocytes and platelets using precalculated database of light scattering patterns

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Characterization of erythrocytes and platelets is relevant for pathology diagnostics. Measurements of size, shape, volume, and surface area may provide important information about the state of human health. Standard flow cytometers measure light scattering from single particles in two solid angles: forward- and side-scattering. A scanning flow cytometer (SFC) measures light scattering patterns (LSPs) from single particles $I(\theta) = \int_0^{2\pi} \mathrm{d}\varphi [S_{11}(\theta,\varphi) + S_{14}(\theta,\varphi)] \big/ 2\pi$ for polar angles $\theta \in [5°,120°]$ with speed up to 500 particles per second [1] (S_{11} and S_{14} are elements of the Mueller matrix). This work is devoted to solving the inverse light scattering problem for erythrocytes and platelets to extract cell characteristics from its LSP.

The inversion method is based on a precalculated (lookup) database of LSPs and nearest-neighbor interpolation (approximation). In other words, the experimental LSP for any particle in a sample is compared with all theoretical LSPs from the database. Cell parameters corresponding to the nearest (with respect to a certain norm) theoretical LSP are ascribed to the measured particle. The size and structure of the database are determined by an adaptive algorithm, which aims to provide the prescribed accuracy and continuity of the inverse mapping.

The optical model of platelet is a homogenous oblate spheroid with the following parameters: $\alpha = 2\pi R_e n_0/\lambda$ – size parameter, $\rho = 2\alpha[(n/n_0) - 1]$ – phase-shift parameter, ε – axis ratio, β – orientation angle of the particle with respect to the incident beam. Here R_e is radius of the equal-volume sphere, λ – the wavelength of the incident radiation (660 nm), n_0 – the refractive index of the medium (1.337), and n – the refractive index of the particle. The database of LSPs contains about 200 000 records in the following parameters ranges: $\alpha = 6$–26, $\rho = 0.4$–6.4, $\varepsilon = 1$–3, $\beta = 0°$–$90°$. The *T*-matrix method was used for the simulation of light scattering from spheroidal particle. To assess the quality of database-based inversion we simulated 1000 test LSPs with parameters randomly distributed in the same range. Their processing resulted in the following root-mean-square (RMS) errors of cell parameters: $\delta\alpha = 0.26$, $\delta\rho = 0.1$, $\delta\varepsilon = 0.1$, and $\delta\beta = 11°$.

Erythrocyte is modeled by a biconcave disk. Specifying the volume V and surface area S of the erythrocyte, its shape is determined by minimization of the surface-deformation energy. Database of 77 532 LSPs was calculated using the discrete dipole approximation in the following range of parameters: $V = 55$–$220\ \mu m^3$, $S = 65$–$190\ \mu m^2$, refractive index in surrounding media $m = n/n_0 = 1.0355$–1.0596, $\beta = 0°$–$90°$. The quality of the inversion was assessed in the same way as that for platelets. RMS errors of 1000 simulated erythrocytes with randomly chosen parameters were the following: $\delta V = 7.3\ \mu m^3$, $\delta S = 3.4\ \mu m^2$, $\delta m = 0.0018$, and $\delta\beta = 1.2°$.

The application of these inversion methods to a blood sample results in a distribution of the sample over cell parameters. Those results will be presented at the conference.

Reference

[1] V. P. Maltsev and K. A. Semyanov, *Characterization of Bio-Particles from Light Scattering* (VSP, Utrecht, 2004).