Characterization of E. coli morphology by scanning flow cytometry

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Light scattering is a powerful physical method for identification and characterization of bacteria. Light scattering by a particle is determined by its overall morphology, including shape and internal distribution of the refractive index. Therefore, angle-resolved light scattering contains valuable information on morphological properties of the particle. In addition to identifying or distinguishing microorganisms by its morphology, light-scattering can also potentially provide real-time monitoring of bacterial growth in order to study cell cycle kinetics or analyze growth rate for antibiotic sensitivity testing. Scanning Flow Cytometer (SFC) is a technique capable of measuring angle-resolved intensity light scattering patterns (LSPs) of individual particles in flow. Characterization of particles morphology from measured LSP requires the solution of the inverse light-scattering (ILS) problem. This abstract describes a method for characterization of individual *E. coli* cells using SFC.

E. coli cell was modeled as a cylinder capped with hemispheres of the same radius. This model is described by three morphological parameters (length, diameter, and refractive index) and an auxiliary parameter (orientation angle of cell in the flow of the SFC). The light scattering by a single *E. coli* cell was simulated by the discrete dipole approximation (DDA). In particular, open-source code ADDA v.1.0 was used. Thus the solution of ILS problem is reduced to a fit of an experimental LSP by theoretical ones. To accelerate the fit we used a precalculated database of 80 000 LSPs in a wide range of model parameters and performed the nearest-neighbor interpolation on it. This allowed us to determine length and diameter of individual bacterium including errors of these estimates.

This method allows characterization of population of any rod-shaped bacteria cells by their length and diameter distributions. The only additional effort may be needed for extension of the database to larger or smaller bacteria sizes.

The developed method was tested by two strains of *E. coli,* showing 135 and 15 nm median precision in determination of length and diameter of single cells, respectively, which is very good for optical methods. Obtained length and diameter distributions showed a good agreement with microscopic measurements of the same samples. A detailed account of these results can be found in the paper [1].

[1] Konokhova AI, Yurkin MA, Gelash AA, Chernyshev AV, Maltsev VP. High-precision characterization of individual E. coli cell morphology by scanning flow cytometry. *Submitted to Cytometry A.*