Characterization of *E. Coli* cells by the scanning flow cytometry

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Dynamical studies of bacterial growth have direct medical applications, e.g., for antibiotic sensitivity tests. Current implementations of the latter require up to a few days because many cell divisions have to occur before the detection. The time for such analysis can be shortened to less than one cell cycle (on the order of an hour) if one is able to measure morphological characteristics of bacteria, based on the correlation of the latter with bacterial growth. Scanning flow cytometry (SFC) is suitable for such measurements, since it allows analysis of individual cells with high throughput and large amount of information collected from each cell (angle-resolved light scattering pattern – LSP). The remaining issue is the solution of the inverse light scattering problem, i.e., the characterization of a bacterium from its LSP. This abstract addresses this issue for rod-shaped *E. Coli* bacteria.

As an optical model of *E. Coli* we have chosen a cylinder capped with hemispheres, based on a microscopic analysis of *E. Coli* and literature data. We pursue two different approaches to the inverse light scattering problem. The first is based on a precalculated (look-up) 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. To calculate theoretical LSPs we used the discrete dipole approximation [1] and the null-field method with discrete sources [2]. The following ranges of bacteria sizes were sampled, covering the biological variability of almost all *E. Coli* strains: lengths from 2 to 10 μm and diameters from 0.7 to 1.3 μm. We also varied the refractive index and orientation relative to the incident light.

The second approach, the so-called parametric one, is based on compressing the LSP into several parameters and finding easy-to-invert relations between them and particle parameters. We observed the correlation of both first minima location and the distance between the first and second minima with bacteria size. We are currently working on an inversion scheme, exploiting this feature.

Using the SFC we measured LSPs of *E. Coli* in two different growth phases. In the stationary phase the lengths of bacteria approximately ranged from 1.5 to 2.5 μm and diameters – from 0.8 to 1 μm, while in logarithmic phase they were from 4 to 8 μm and from 1 to 1.2 μm, respectively. There was a good agreement between the theoretical and experimental LSPs. Moreover, we applied the database-based method to experimental data. Obtained distributions of samples over bacteria lengths and diameters showed a good agreement with both literature data on these strains and microscopic measurements of the same samples.

References

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