Mario Feingold – Research summary

During the last few years we were involved in three projects related to Single Cell Optical Tomography (SCOT).

First, we have used a simplified form of tomography to analyze the structure of the Z-ring in *E. coli* cells that have not yet started to septate. Specifically, we have imaged the cells in both horizontal and vertical orientations. The horizontal orientation allows establishing whether a Z-ring has formed but the cell has not yet started to constrict. On the other hand, the vertical orientation allows viewing the Z-ring as it lies in the plane of view leading to a circularly symmetric image that renders itself to quantitative analysis. This approach allows measuring the radial width of the Z-ring. It turns out to be ~120 nm wide much larger than expected (1).

Second, we have extended our structural study of the Z-ring into the time domain to determine its formation time, τ_z , and correlate it with the other timescales of the bacterial cell cycle (2). The combined information will shed new light on the possible mechanisms leading to the signal for the onset of septation. To determine τ_z in single *E. coli* cells, we have used time-lapse microscopy that alternates between phase contrast and fluorescence imaging modes. We find that τ_z is independent of both the time for the onset of septation, τ_c , and the generation time, τ_g . We also find that τ_c is linearly correlated with the period of divisome assembly, $\tau_c - \tau_z$.

Third, we have developed a necessary instrument for the implementation of SCOT, namely, a method to measure the cell angle of rotation from its phase contrast image (3,4). Since the ends of the rotated cell are strongly defocused, angle measurement is difficult. It is achieved using an interesting feature of the phase contrast intensity profiles, namely, the critical point. We found that the longitudinal phase contrast profiles of defocused images all intersect in one point that lies in the vicinity of the cell edge. This point, which we denote as the critical point, is used to locate the cell edge despite defocusing allowing to measure the cell rotation angle (see Fig. 1).



Fig. 1 – A large step θ -scan of an *E. coli* cell (θ is the angle between the long cell axis and the optical axis). Bar = 1 µm.

1. G. Carmon, I. Fishov and M. Feingold, 2011, *Oriented imaging of 3D sub-cellular structures in bacterial cells using Optical Tweezers*, submitted to Opt. Lett.

2. G. Carmon and M. Feingold, 2011, *Rotation of single bacterial cells relative to the* optical *axis using Optical Tweezers*, Opt. Lett. **36**, 40-42. Reprinted in the Virtual Journal for Biomedical Optics, **6** (2011).

3. G. Carmon and M. Feingold, 2011, *Controlled alignment of bacterial cells with oscillating Optical Tweezers*, J. Nanophotonics **5**, 051803-1-9. Reprinted in the Virtual Journal of Biological Physics Research, **21** (2011).

4. R. Tsukanov, G. Reshes, G. Carmon, E. Fischer-Friedrich, N.S. Gov, I. Fishov and M. Feingold, 2011, *Timing of Z-ring localization in Escherichia coli*, Phys. Biol. **8**, 066003.