AMERICAN SOCIETY FOR MICROBIOLOGY | Journal of Bacteriology

EDITORIAL

Classic Spotlight: Dynamics of the Bacterial Cytoplasm

Conrad W. Mullineaux

School of Biological and Chemical Sciences, Queen Mary University of London, London, United Kingdom

The bacterial cytoplasm is complex and crowded with macromolecules, providing an environment very different from the dilute solutions used in traditional *in vitro* biochemistry (1). The ability of different molecules to diffuse within this complex environment must influence almost every aspect of bacterial biochemistry, yet the sheer complexity of the cytoplasm makes prediction of diffusion rates extremely uncertain.

Until 1999, no information was available on molecular diffusion kinetics in the bacterial cytoplasm. Diffusion kinetics can be quantified by fluorescence recovery after photobleaching (FRAP) and other fluorescence-based techniques such as fluorescence correlation spectroscopy (FCS) and single-particle tracking (SPT), but the methods traditionally applied to eukaryotic cells were technically challenging in bacteria due to their much smaller cell size and the impracticability of microinjection of fluorescent tracer molecules. Then, in a groundbreaking paper in the *Journal of Bacteriology*, Elowitz et al. reported the first direct measurements of protein diffusion in the cytoplasm of *Escherichia coli* (2). These authors solved the microinjection problem by using as their fluorescent tracer green fluorescent protein (GFP) endogenously expressed, and they were able to perform accurate FRAP measurements by a combination of state-of-the-art microscopy, careful data analysis, and the simple but ingenious trick of making the *E. coli* cells longer by treatment with the septation inhibitor cephalexin (2).

The GFP diffusion coefficient (7.7 $\mu m^2 s^{-1}$) reported by Elowitz et al. (2) became a standard reference point for modeling the dynamics of the bacterial cytoplasm (see, e.g., reference 3). Furthermore, by showing that a short tag of 6 histidine residues significantly slowed diffusion, Elowitz et al. demonstrated an influence of electrostatic interactions on protein mobility. Their results have been used to aid in quantitative understanding of specific dynamic processes from chemotactic signal transduction (4, 5) to chromosome segregation (6) and the spatial organization of translation (7).

REFERENCES

1. **Ellis RJ.** 2001. Macromolecular crowding: an important but neglected aspect of the intracellular environment. Curr Opin Struct Biol **11:**114–119. http://dx.doi.org/10.1016/S0959-440X(00)00172-X.
2. **Elowitz MB, Surette MG, Wolf P-E, Stock JB, Leibler S.** 1999. Protein mobility in the cytoplasm of *Escherichia coli*. J Bacteriol **181:**197–203.
3. **McGuffee SR, Elcock AH.** 2010. Diffusion, crowding and protein stability in a dynamic molecular model of the bacterial cytoplasm. PLoS Comp Biol **6:**e1000694. http://dx.doi.org/10.1371/journal.pcbi.1000694.
4. **Maki N, Gestwicki JE, Lake EM, Kiessling LL, Adler J.** 2000. Motility and chemotaxis of filamentous cells of *Escherichia coli*. J Bacteriol **182:**4337–4342. http://dx.doi.org/10.1128/JB.182.15.4337-4342.2000.
5. **Lipkow K, Andrews SS, Bray D.** 2005. Simulated diffusion of phosphorylated CheY through the cytoplasm of *Escherichia coli*. J Bacteriol **187:**45–53. http://dx.doi.org/10.1128/JB.187.1.45-53.2005.
6. **Woldringh CL.** 2002. The role of co-transcriptional translation and protein translocation (transertion) in bacterial chromosome segregation. Mol Microbiol **45:**17–29. http://dx.doi.org/10.1046/j.1365-2958.2002.02993.x.
7. **Montero Llopis P, Jackson AF, Sliusarenko O, Surovtsev I, Heinritz J, Emonet T, Jacobs-Wagner C.** 2010. Spatial organization of the flow of genetic information in bacteria. Nature **466:**77–81. http://dx.doi.org/10.1038/nature09152.

Citation Mullineaux CW. 2016. Classic spotlight: dynamics of the bacterial cytoplasm. J Bacteriol 198:1183. doi:10.1128/JB.00010-16.

Address correspondence to c.mullineaux@qmul.ac.uk.

Copyright © 2016, American Society for Microbiology. All Rights Reserved.

The views expressed in this Editorial do not necessarily reflect the views of the journal or of ASM.

Downloaded from http://jb.asm.org/ on April 22, 2016 by Queen Mary, University of London