



Deposited via The University of York.

White Rose Research Online URL for this paper:

<https://eprints.whiterose.ac.uk/id/eprint/100143/>

Version: Accepted Version

Book Section:

Leake, Mark Christian (2016) New advances in chromosome architecture. In: Leake, Mark C., (ed.) Chromosome Architecture. Methods in Molecular Biology. Springer.

Reuse

Items deposited in White Rose Research Online are protected by copyright, with all rights reserved unless indicated otherwise. They may be downloaded and/or printed for private study, or other acts as permitted by national copyright laws. The publisher or other rights holders may allow further reproduction and re-use of the full text version. This is indicated by the licence information on the White Rose Research Online record for the item.

Takedown

If you consider content in White Rose Research Online to be in breach of UK law, please notify us by emailing eprints@whiterose.ac.uk including the URL of the record and the reason for the withdrawal request.

New advances in chromosome architecture

*Mark C. Leake*¹

¹ Biological Physical Sciences Institute (BPSI)

University of York

York

YO10 5DD

United Kingdom

¹ Corresponding author

e-mail: mark.leake@york.ac.uk

Tel: +44 (0)1904 322697

Running head: Chromosome architecture advances.

Abstract

Our knowledge of the ‘architecture’ of chromosomes has grown enormously in the past decade. This new insight has been enabled largely through advances in interdisciplinary research methods at the cutting-edge interface of the life and physical sciences. Importantly this has involved several state-of-the-art biophysical tools used in conjunction with molecular biology approaches which enable investigation of chromosome structure and function in living cells. Also, there are new and emerging interfacial science tools which enable significant improvements to the spatial and temporal resolution of quantitative measurements, such as *in vivo* super-resolution and powerful new single-molecule biophysics methods, which facilitate probing of dynamic chromosome processes hitherto impossible. And there are also important advances in the methods of theoretical biophysics which have enabled advances in predictive modelling of this high quality experimental data from molecular and physical biology to generate new understanding of the modes of operation of chromosomes, both in eukaryotic and prokaryotic cells. Here, I discuss these advances, and take stock on the current state of our knowledge of chromosome architecture and speculate where future advances may lead.

Key words: Single-molecule biophysics, super-resolution, DNA, nucleus

1. Introduction

This volume of Springer’s Methods in Molecular Biology series consists of a collection of truly cutting-edge laboratory protocols, techniques and applications in use today by some of the leading international experts in the broad field of ‘Chromosome Architecture’. A key difference in emphasis, compared with previous collections of review articles published in

this area over the past 5 years, is on the emphasis on the development and application of complex techniques and protocols which increase the physiological relevance of chromosome architecture investigation compared to methods utilized previously – these developments are manifest both through application of far more complex bottom-up assays *in vitro*, as well as in striving to maintain the native physiological context through investigation of living, functional cells. **(1)** In particular, experimental methods which have used advances in light microscopy, **(2)** especially the use of fluorescence microscopy methods to probe functional, living cells, especially so using prokaryotic systems as model organisms. **(3-12)** The length scale of precision of experimental protocols in this area has improved dramatically over recent years and many cutting-edge methods now utilize state-of-the-art single-molecule approaches, **(13)** both for imaging the DNA content of chromosome and proteins that bind to DNA, as well as using methods that can controllably manipulate single DNA molecules and can image its structure to a precision better the standard optical resolution limit. **(14)** This volume also includes more complex, physiologically representative methods to investigate chromosome architecture through the use of advanced computational methods and mathematical analysis.

What is clear is that the combination of pioneering molecular biology, biochemistry and genetics methods with emerging, exciting tools from biophysics, bioengineering, computer science and biomathematics are transforming our knowledge of functional chromosome architecture. Improvements in these fields are likely to add yet more insight over the next few years into the complex interactions between multiple key molecular players inside chromosomes.

Acknowledgments

MCL was assisted by a Royal Society URF and research funds from the Biological Physical Sciences Institute (BPSI) of the University of York, UK.

References

1. A.J.M. Wollman, H. Miller, Z. Zhou, et al. (2015) Probing DNA interactions with proteins using a single-molecule toolbox: inside the cell, in a test tube and in a computer, *Biochemical Society Transactions*. 43, 139–145.
2. A.J.M. Wollman, R. Nudd, E.G. Hedlund, et al. (2015) From Animaculum to single molecules: 300 years of the light microscope, *Open Biology*. 5, 150019–150019.
3. T. Lenn, M.C. Leake, and C.W. Mullineaux (2008) Are Escherichia coli OXPHOS complexes concentrated in specialized zones within the plasma membrane?, *Biochemical Society transactions*. 36, 1032–6.
4. M. Plank, G.H. Wadhams, and M.C. Leake (2009) Millisecond timescale slimfield imaging and automated quantification of single fluorescent protein molecules for use in probing complex biological processes., *Integrative biology : quantitative biosciences from nano to macro*. 1, 602–12.
5. S.-W. Chiu and M.C. Leake (2011) Functioning nanomachines seen in real-time in living bacteria using single-molecule and super-resolution fluorescence imaging., *International journal of molecular sciences*. 12, 2518–42.
6. A. Robson, K. Burrage, and M.C. Leake (2013) Inferring diffusion in single live cells at the single-molecule level., *Philosophical transactions of the Royal Society of London. Series B, Biological sciences*. 368, 20120029.

7. S.J. Bryan, N.J. Burroughs, D. Shevela, et al. (2014) Localisation and interactions of the Vipp1 protein in cyanobacteria., *Molecular microbiology*.
8. I. Llorente-Garcia, T. Lenn, H. Erhardt, et al. (2014) Single-molecule in vivo imaging of bacterial respiratory complexes indicates delocalized oxidative phosphorylation., *Biochimica et biophysica acta*. 1837, 811–24.
9. R. Reyes-Lamothe, D.J. Sherratt, and M.C. Leake (2010) Stoichiometry and architecture of active DNA replication machinery in *Escherichia coli*., *Science*. 328, 498–501.
10. A. Badrinarayanan, R. Reyes-Lamothe, S. Uphoff, et al. (2012) In vivo architecture and action of bacterial structural maintenance of chromosome proteins., *Science*. 338, 528–31.
11. A. Wollman and M.C. Leake (2015) FD2015-Single Molecule Microscopy: Millisecond single-molecule localization microscopy combined with convolution analysis and automated image segmentation to determine protein concentrations in complexly structured, functional cells, one cell at a time, *Faraday Discuss*.
12. T. Lenn and M.C. Leake (2015) Single-molecule studies of the dynamics and interactions of bacterial OXPHOS complexes. *Biochim Biophys Acta* 2015 Oct 20. pii: S0005-2728(15)00215-7. doi: 10.1016/j.bbabi.2015.10.008.
13. M.C. Leake. (2013) The physics of life: one molecule at a time. *Philos Trans R Soc Lond B Biol Sci*. 368(1611):20120248.
14. H. Miller, Z. Zhaokun, A.J.M. Wollman, et al. (2015) Superresolution imaging of single DNA molecules using stochastic photoblinking of minor groove and intercalating dyes, *Methods*.