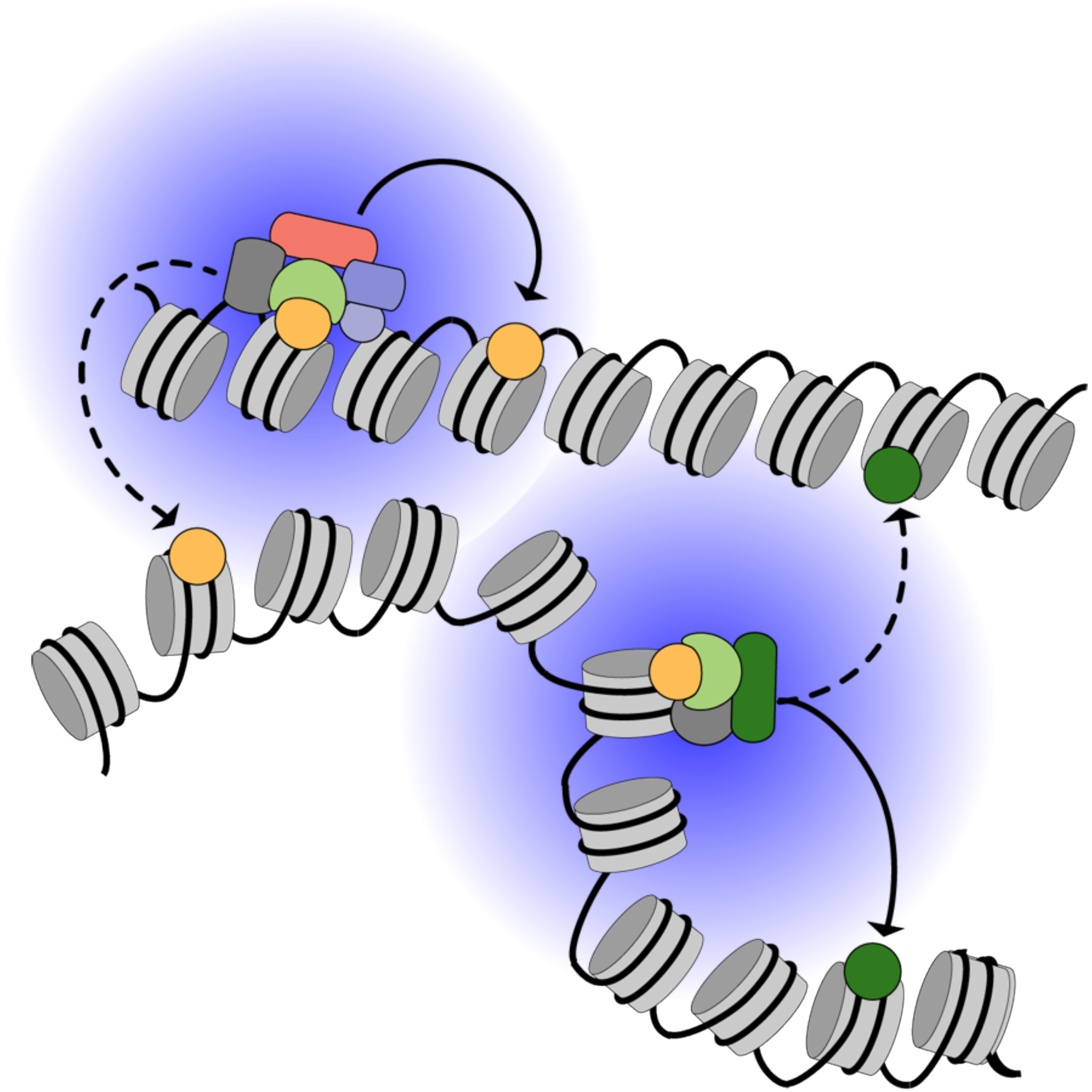
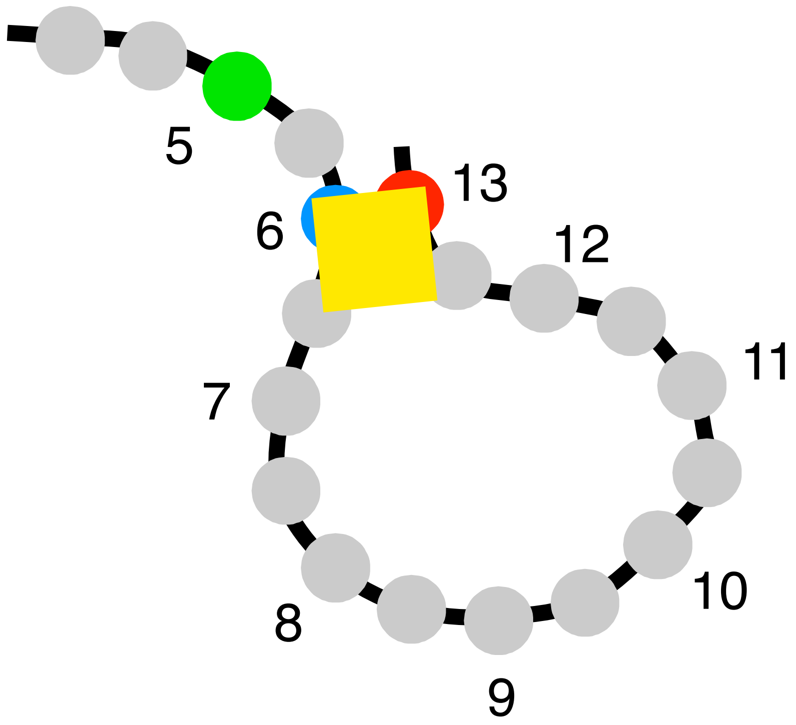
Mapping interactions by chromatin looping

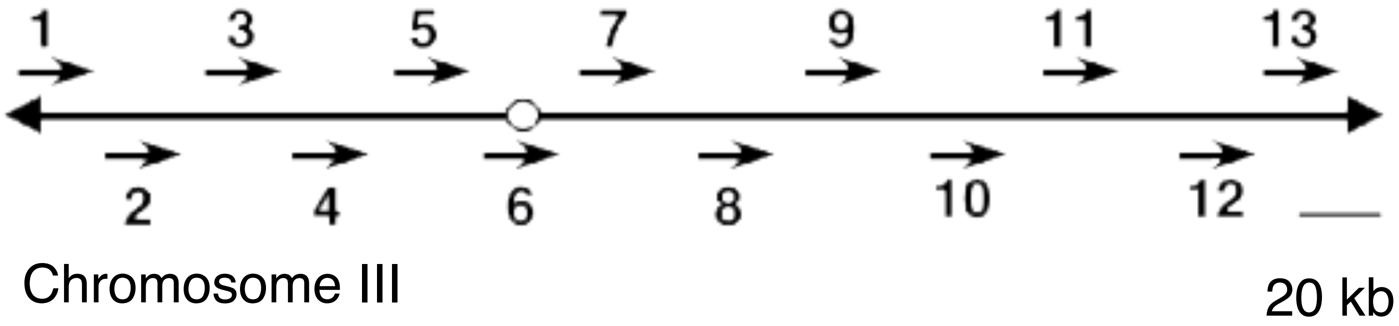


Capturing **C**hromatin **C**onformation (3C assay) by *in vivo* cross-linking of whole cells or isolated nuclei



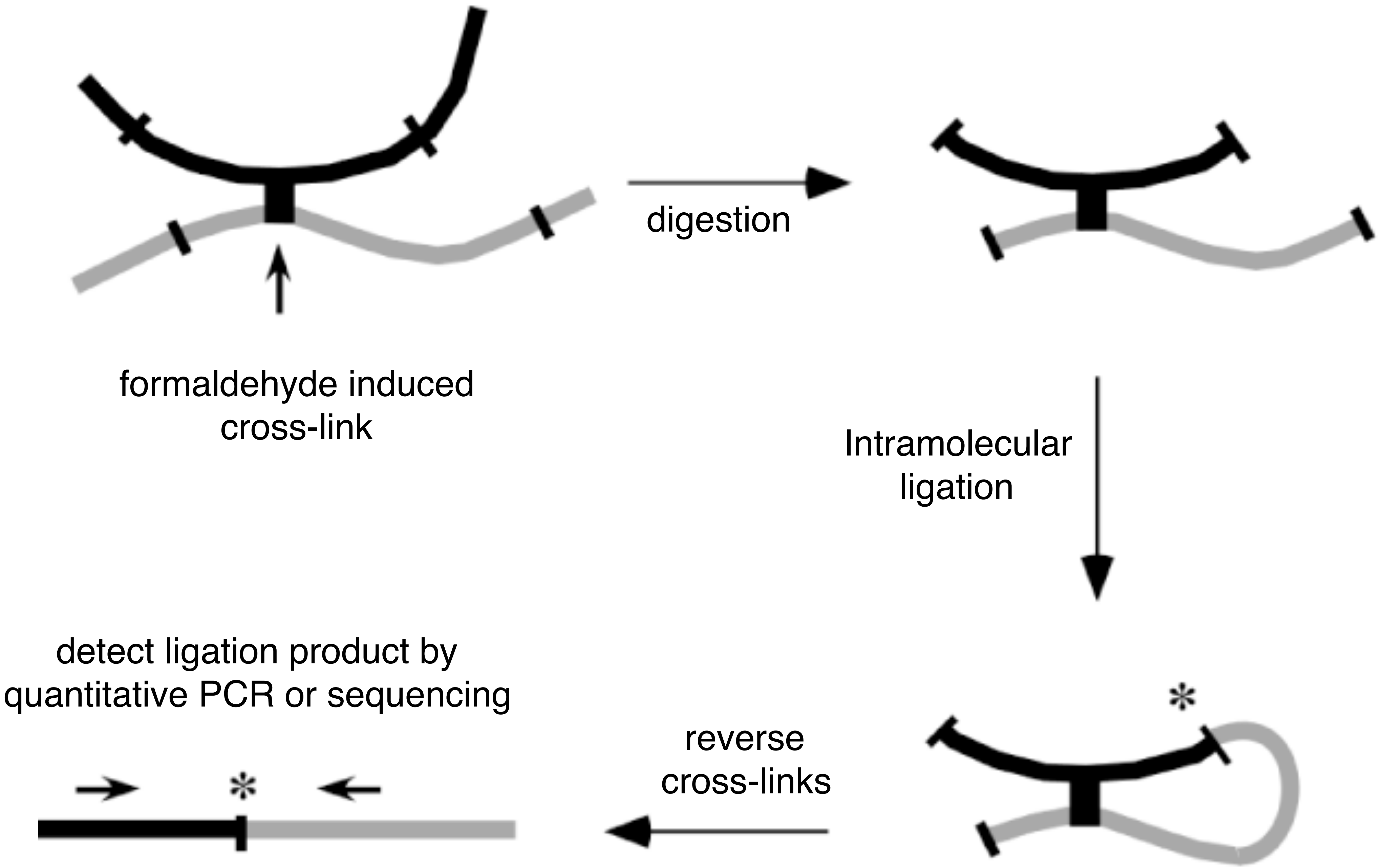
Chemical cross-link two sites on the chromosome

Quantitate cross-link efficiency and relate it to **genomic distance**



Dekker, J., Rippe, K., Dekker, M. & Kleckner, N. (2002) *Science* **295**, 1306-1311.

Capturing **C**hromatin **C**onformation (3C assay) by *in vivo* cross-linking of whole cells or isolated nuclei



formaldehyde induced
cross-link

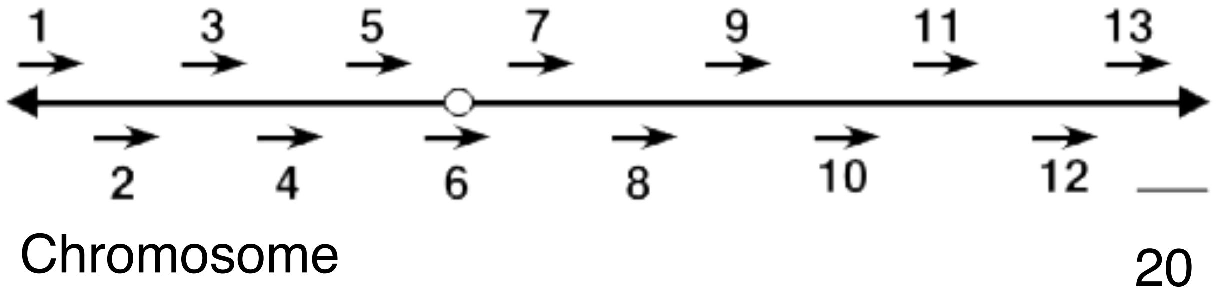
digestion

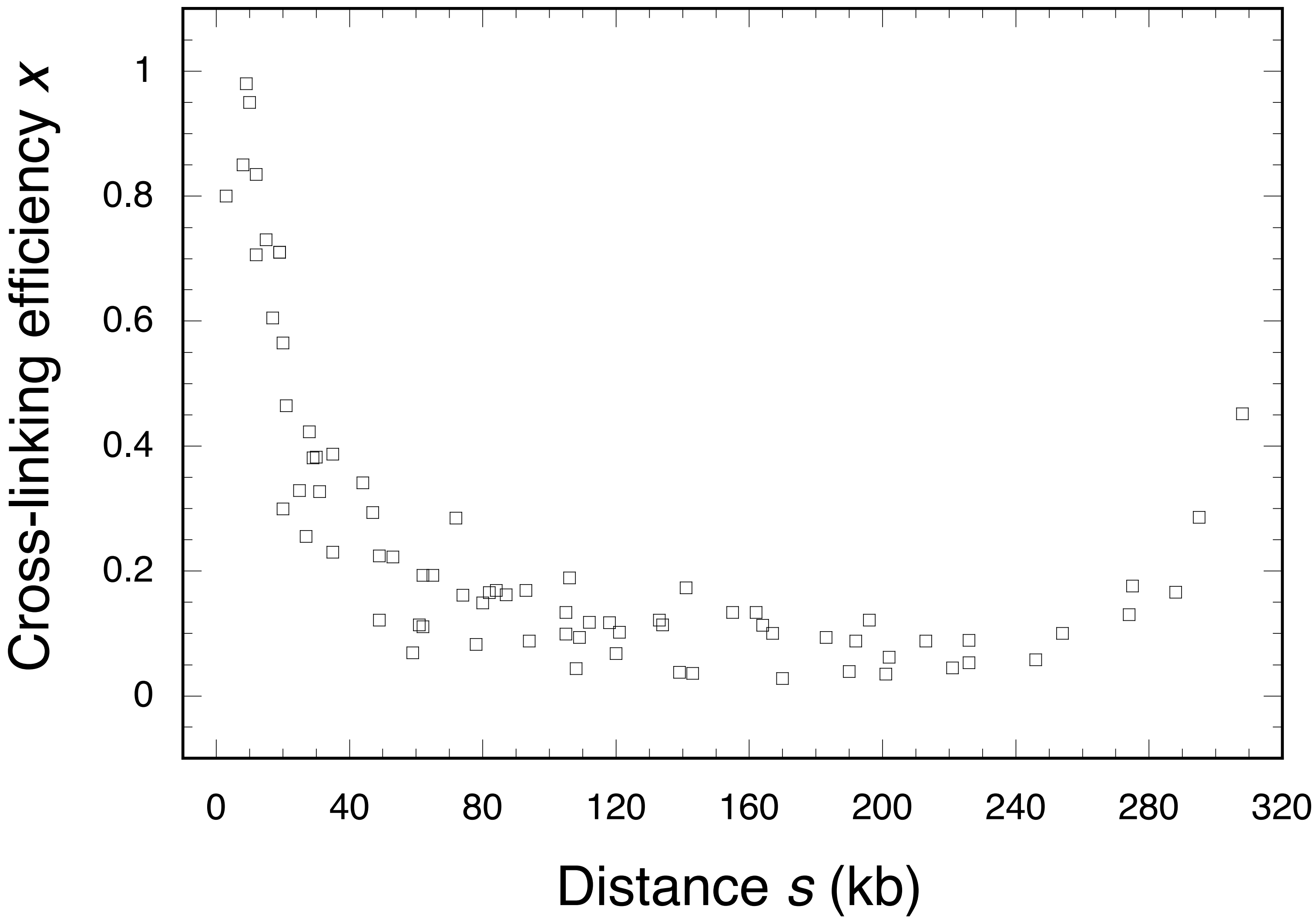
Intramolecular
ligation

reverse
cross-links

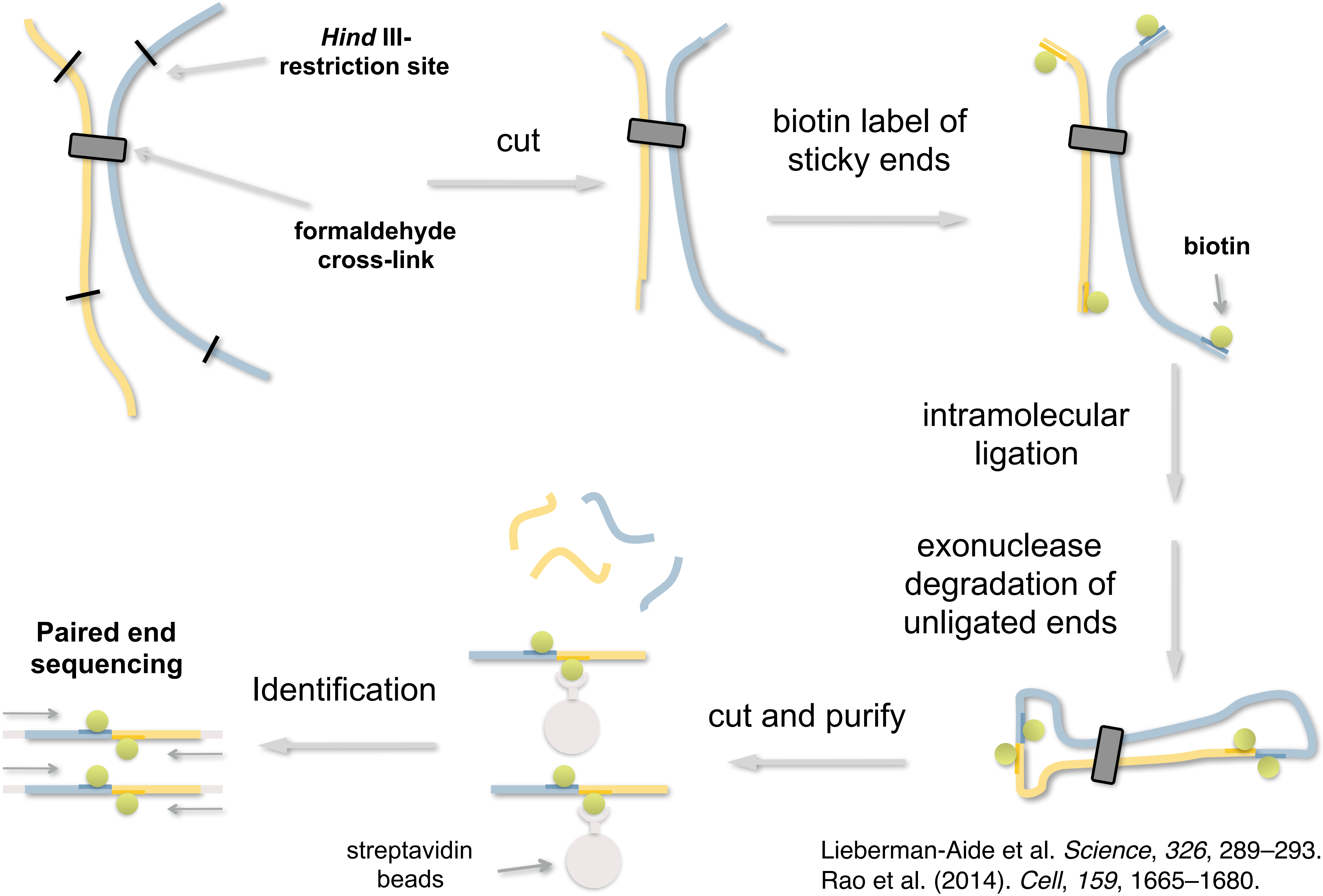
detect ligation product by
quantitative PCR or sequencing

Cross-linking analysis of the *in vivo* interphase conformation of the complete yeast chromosome III (315 kb) in G1





Hi-C – map all interactions



***Hind* III-restriction site**

formaldehyde cross-link

cut

biotin label of sticky ends

biotin

intramolecular ligation

exonuclease degradation of unligated ends

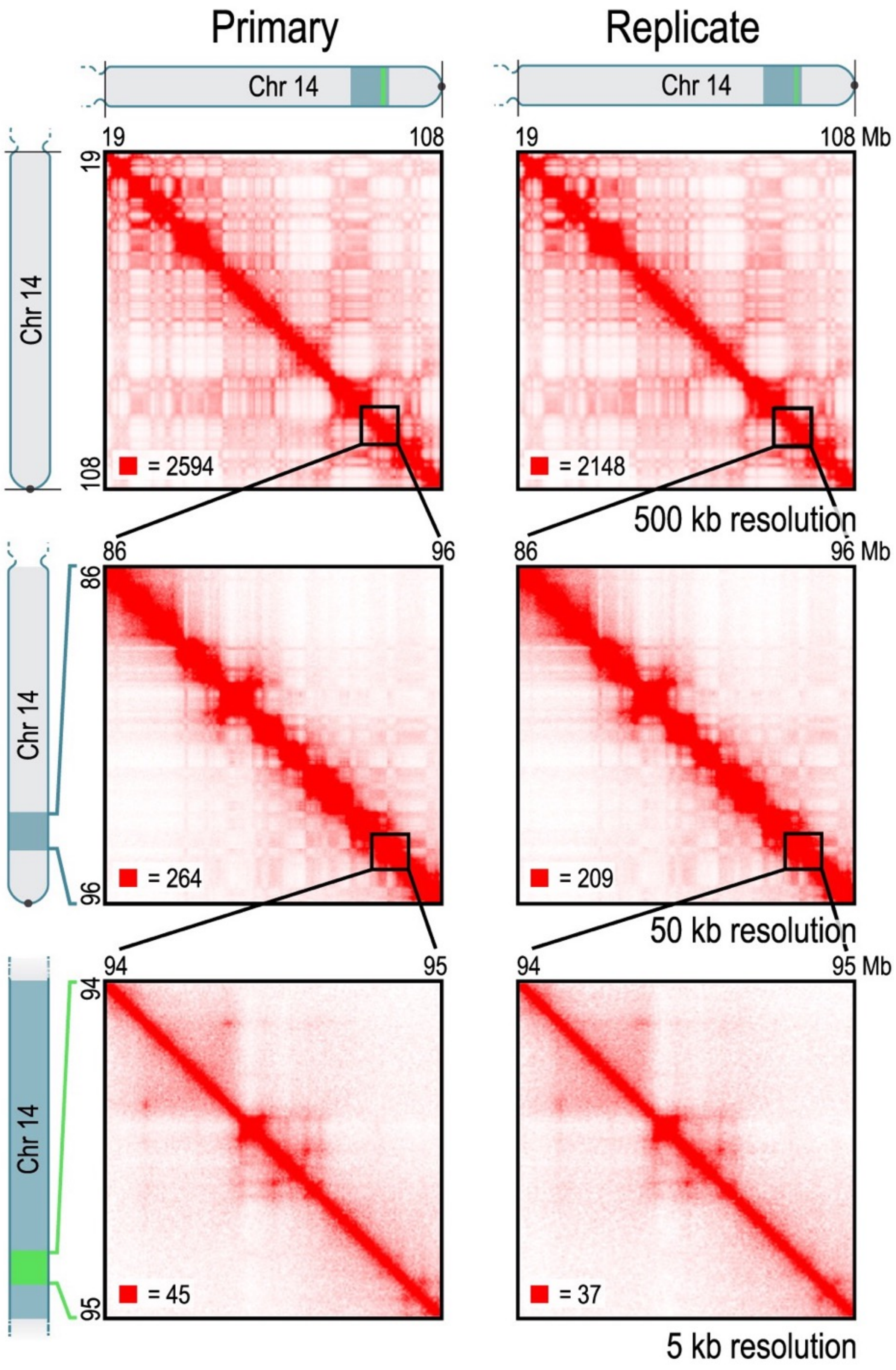
cut and purify

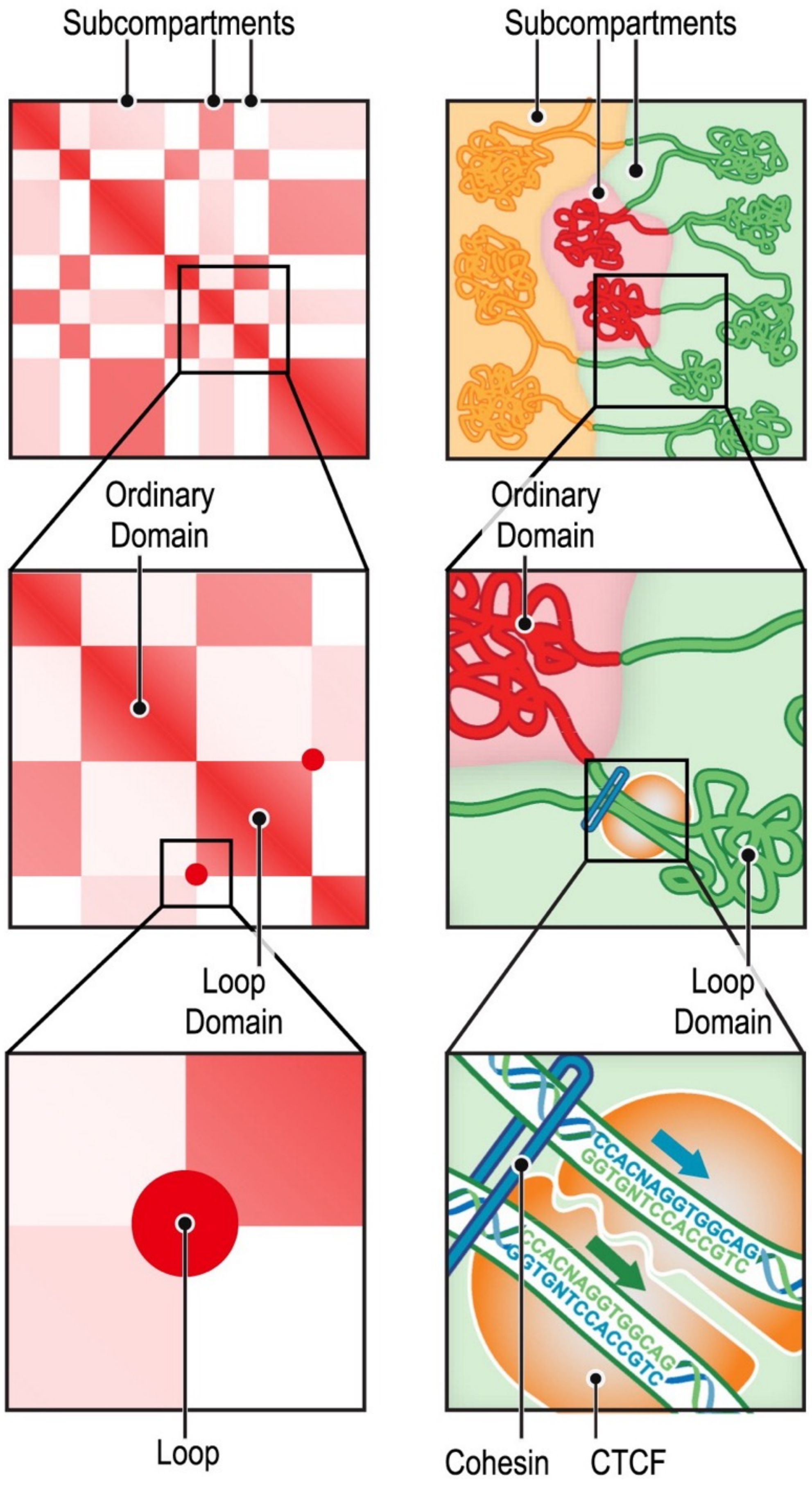
Identification

streptavidin beads

Paired end sequencing

Lieberman-Aide et al. *Science*, *326*, 289–293.
Rao et al. (2014). *Cell*, *159*, 1665–1680.

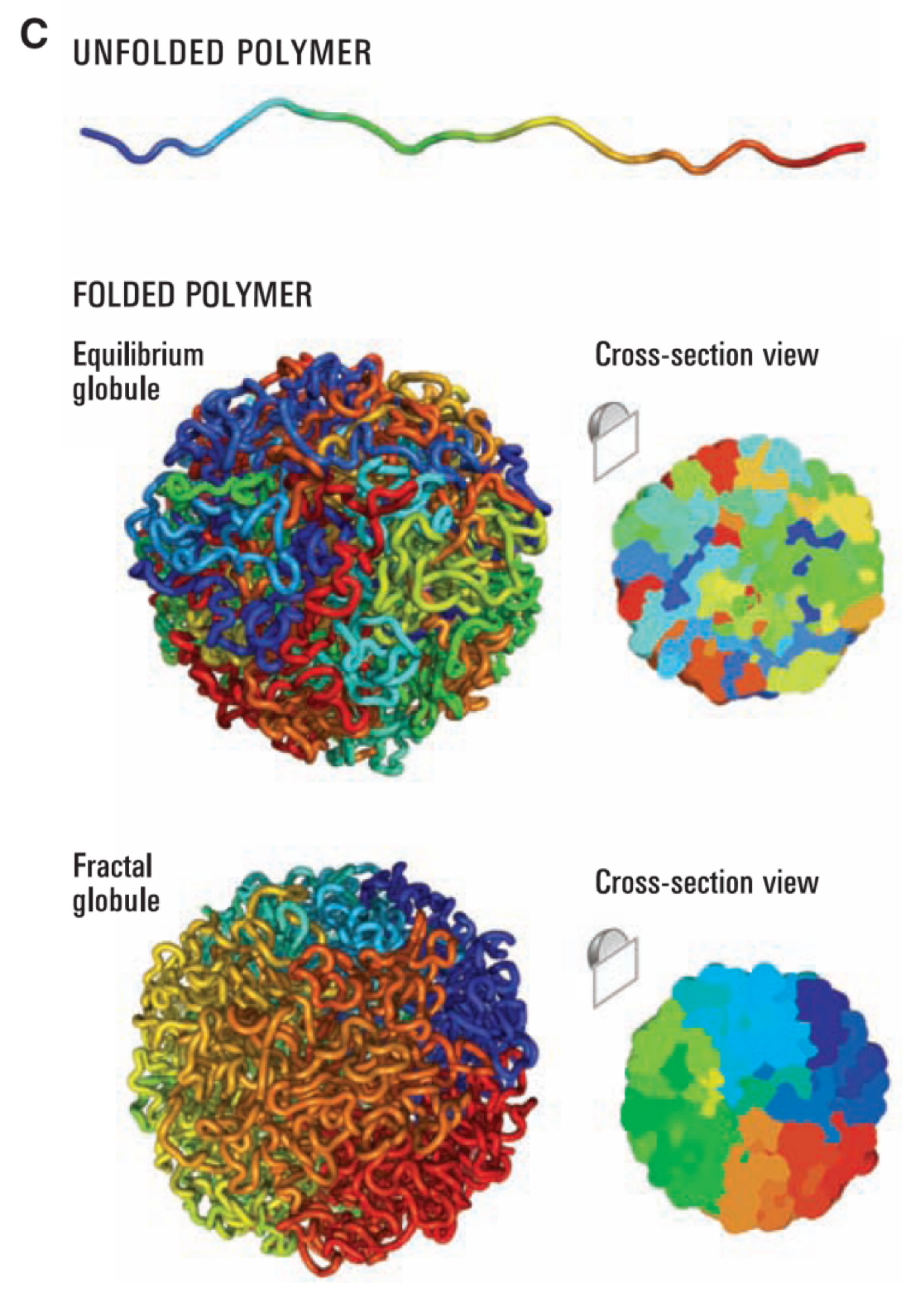

Primary
Replicate
Chr 14
Chr 14
19
108
19
108 Mb
Chr 14
19
108
= 2594
= 2148
500 kb resolution
86
96
86
96 Mb
Chr 14
86
96
= 264
= 209
50 kb resolution
94
95
94
95 Mb
Chr 14
94
95
= 45
= 37
5 kb resolution


Subcompartments
Subcompartments
Ordinary Domain
Ordinary Domain
Loop Domain
Loop Domain
Loop
CCACNAGGTGGCAG
GGTGNTCCACCGTC
CCACNAGGTGGCAG
GGTGNTCCACCGTC
Cohesin
CTCF

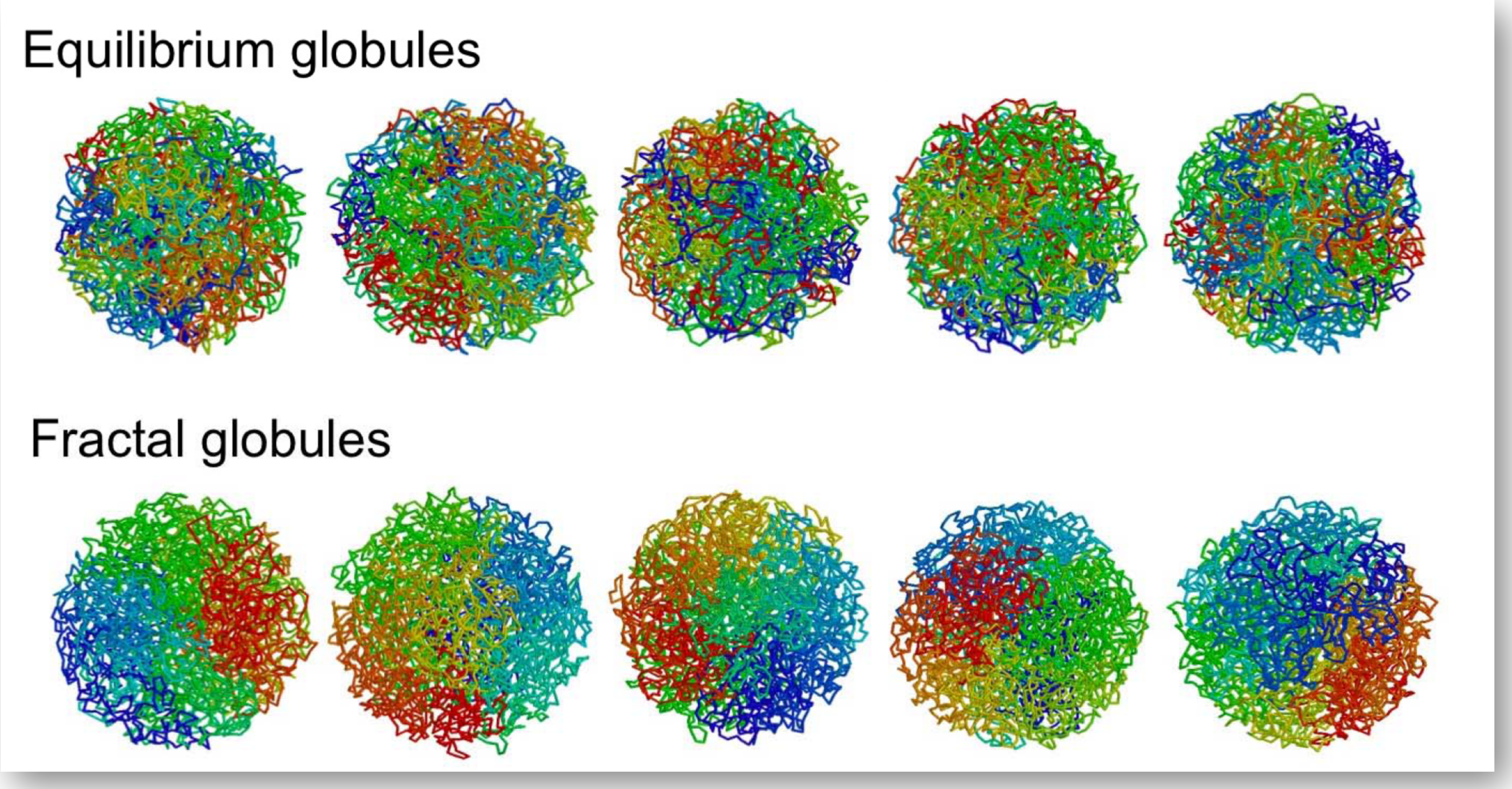
Hi-C – Equilibrium (random coil) ⬄ Fractal Globules

- Equilibrium globule
  - random coil structure
  - adjacent loci on the chain are not necessarily close in 3D
- Fractal globule
  - no knots of the chain
  - adjacent loci on the chain are close in 3D
  - this is visualized by the color coding that leads to blocks with similar coloring in a „hierarchical clusterung“

Lieberman-Aiden et al., Science, 2009.

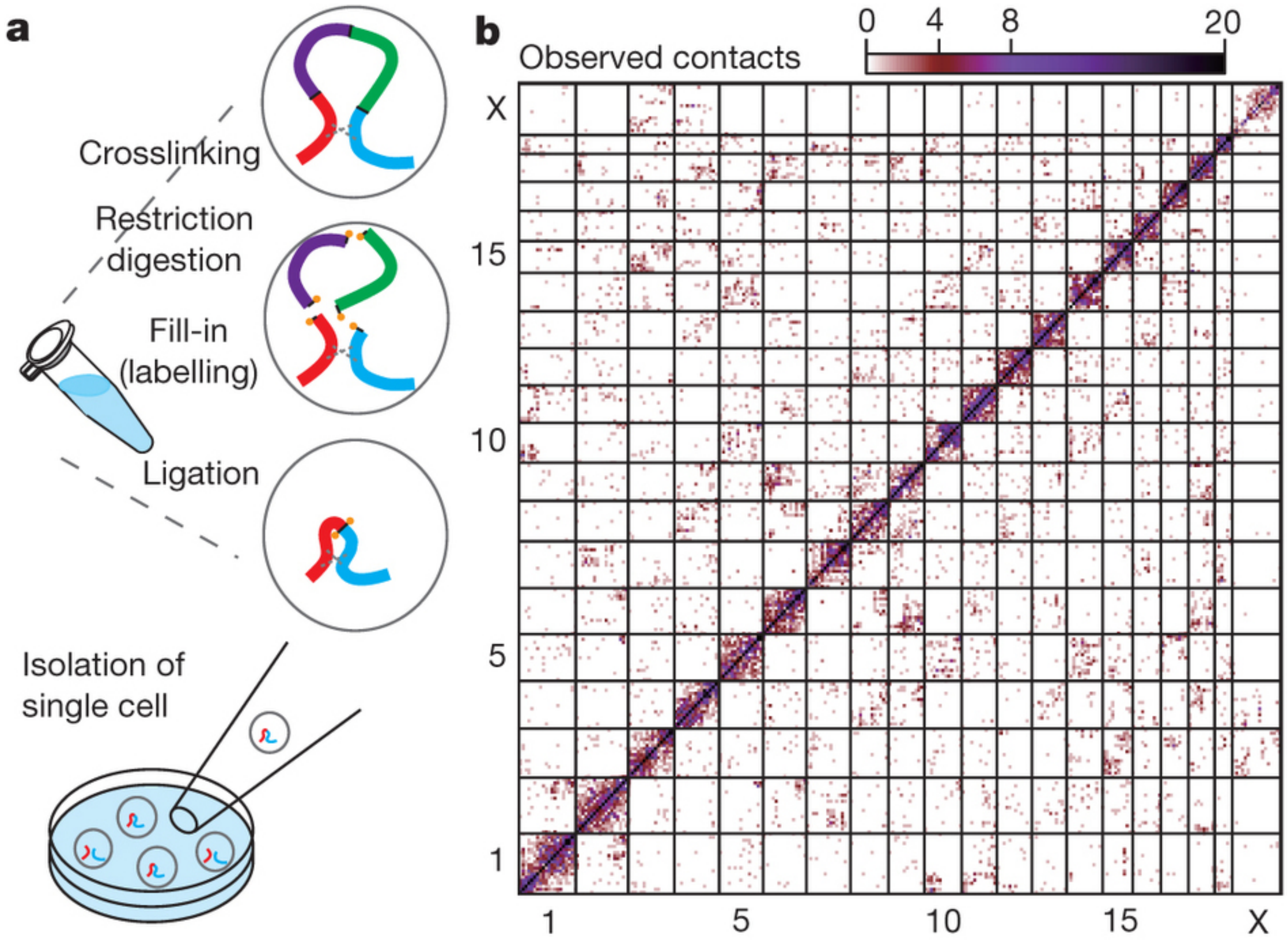


Hi-C – Equilibrium ⬄ Fractal Globules



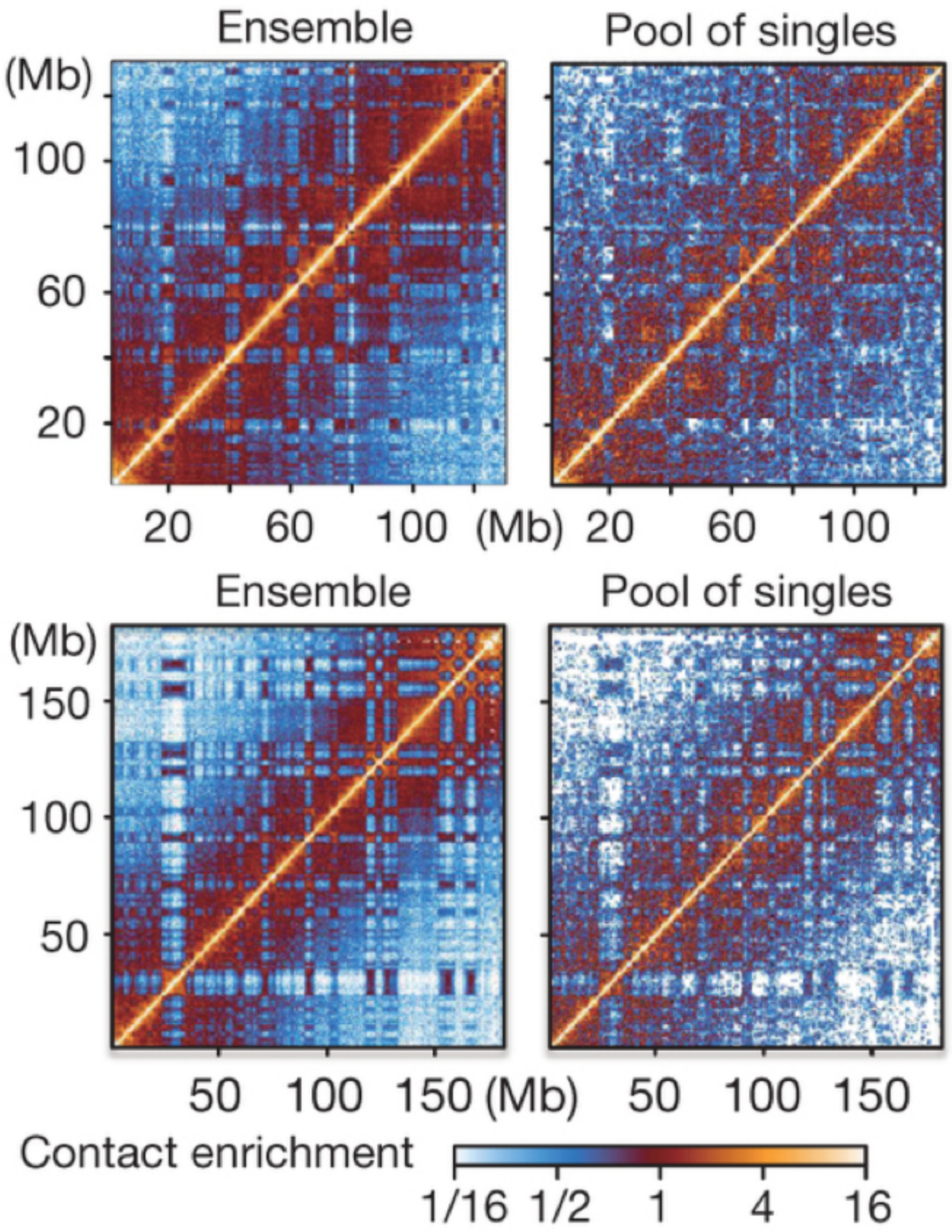
Lieberman-Aiden et al., Science, 2009.

Single-cell Hi-C maps of CD4+ mouse T cells



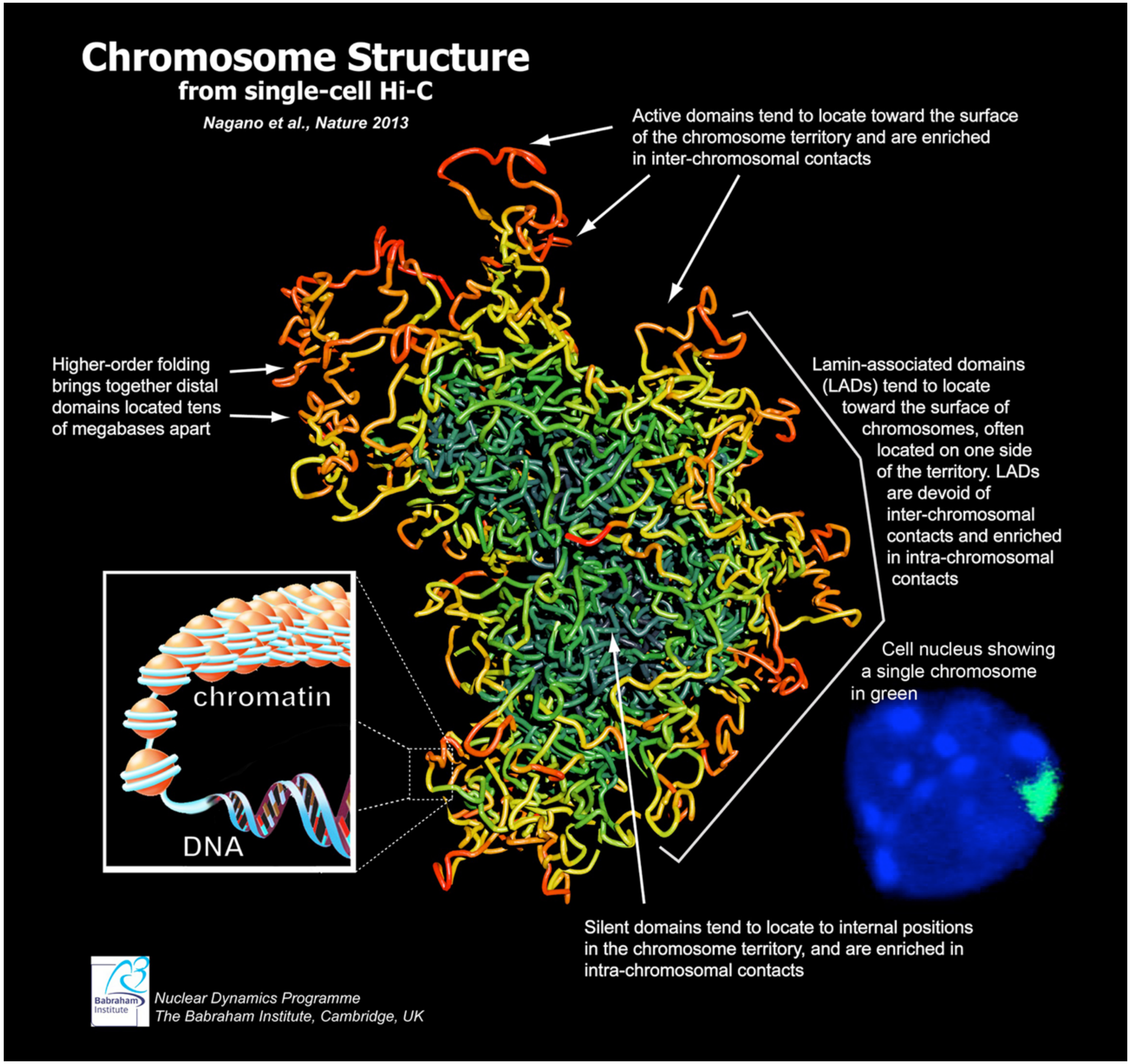
T Nagano *et al.* *Nature* **000**, 1-6 (2013) doi:10.1038/nature12593

Single-cell and ensemble Hi-C



Nagano et al. 2013. Single-cell Hi-C reveals cell-to-cell variability in chromosome structure. Nature *502*, 59–64.

Single cell genome folding maps from Hi-C



Chromosome Structure
from single-cell Hi-C
Nagano et al., Nature 2013

Active domains tend to locate toward the surface of the chromosome territory and are enriched in inter-chromosomal contacts

Higher-order folding brings together distal domains located tens of megabases apart

Lamin-associated domains (LADs) tend to locate toward the surface of chromosomes, often located on one side of the territory. LADs are devoid of inter-chromosomal contacts and enriched in intra-chromosomal contacts

Cell nucleus showing a single chromosome in green

Silent domains tend to locate to internal positions in the chromosome territory, and are enriched in intra-chromosomal contacts

Nuclear Dynamics Programme
The Babraham Institute, Cambridge, UK

Nagano et al. 2013. Single-cell Hi-C reveals cell-to-cell variability in chromosome structure. Nature *502*, 59–64.
http://www.babraham.ac.uk/our-research/nuclear-dynamics/peter-fraser