Chromosome Positioning from Activity-based Segregation

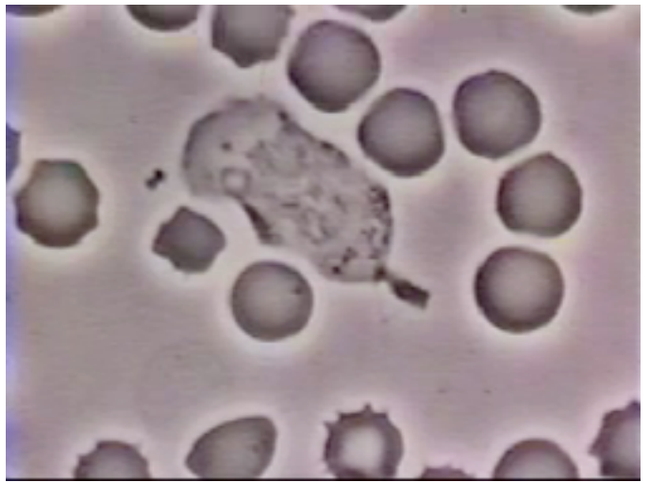
Gautam I. Menon

The Institute of Mathematical Sciences,
Chennai, India

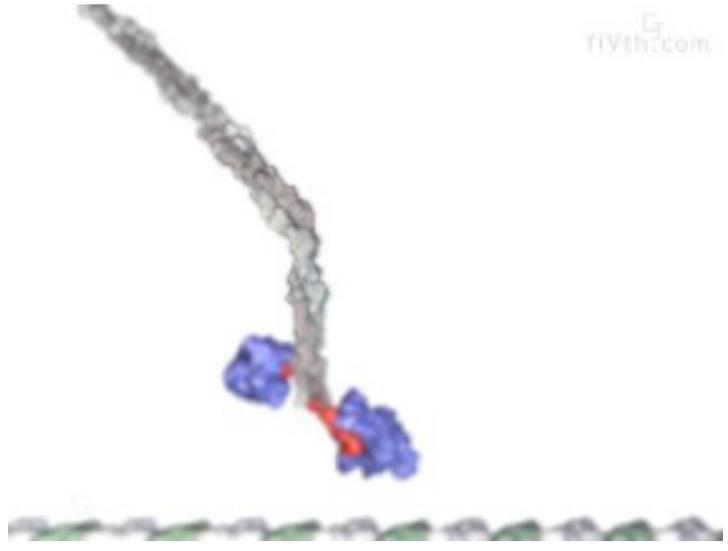
With: Nirmalendu Ganai (Vidyasagar College, India) and Surajit Sengupta (TIFR-H, IACS, India)

Active Matter paradigm for biological systems

Physical description: Matter out of thermal equilibrium, driven internally by ATP consuming processes, non-equilibrium biochemical reactions



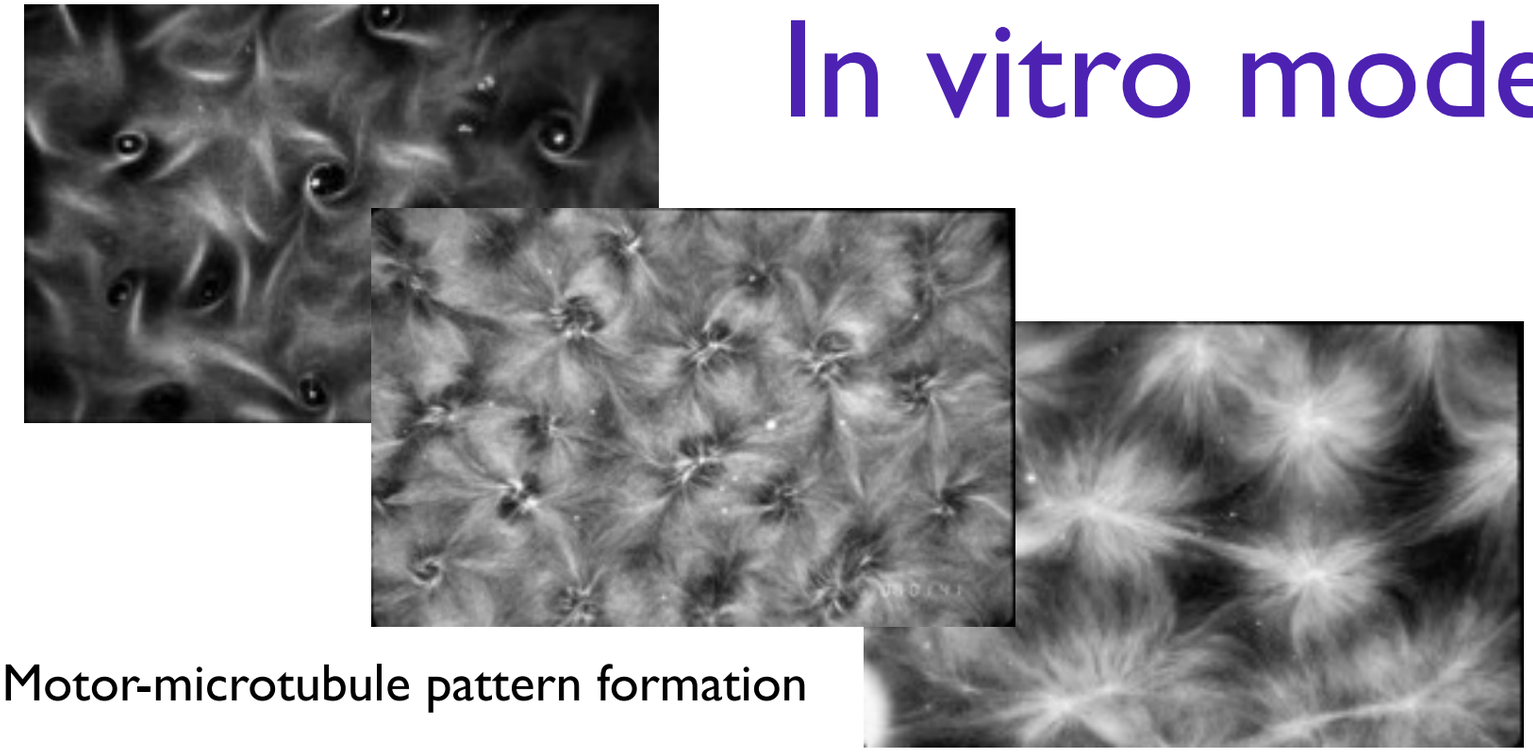
http://www.biochemweb.org/neutrophil.shtml



http://valelab.ucsf.edu/moviepages/movies.html

Specific consequences: Large fluctuations, increased sensitivity, collective behaviour, coupling across very different length and time scales, spatial structuring

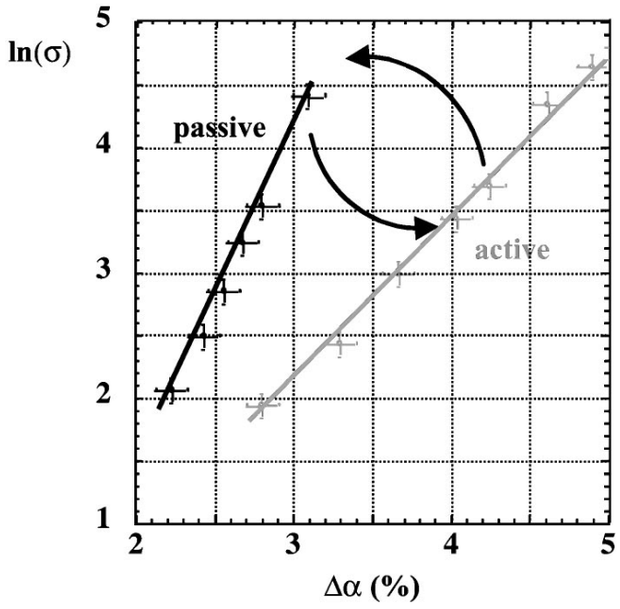
In vitro models



Motor-microtubule pattern formation

Nedelec, Surrey, Maggs and Leibler Nature 1997.

Active membranes



Prost, Bruinsma, Bassereau, Manneville, Ramaswamy ..
EPL '96, PRL '99, PRE '01

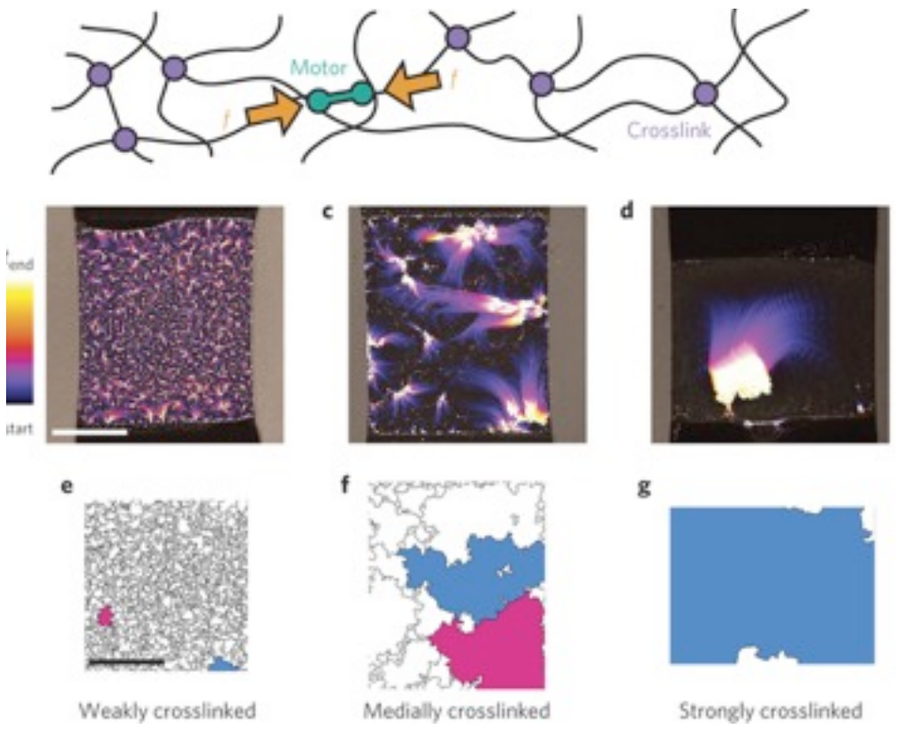
Self-propelled objects *a la* Vicsek

Andrea Cavagna
homepage

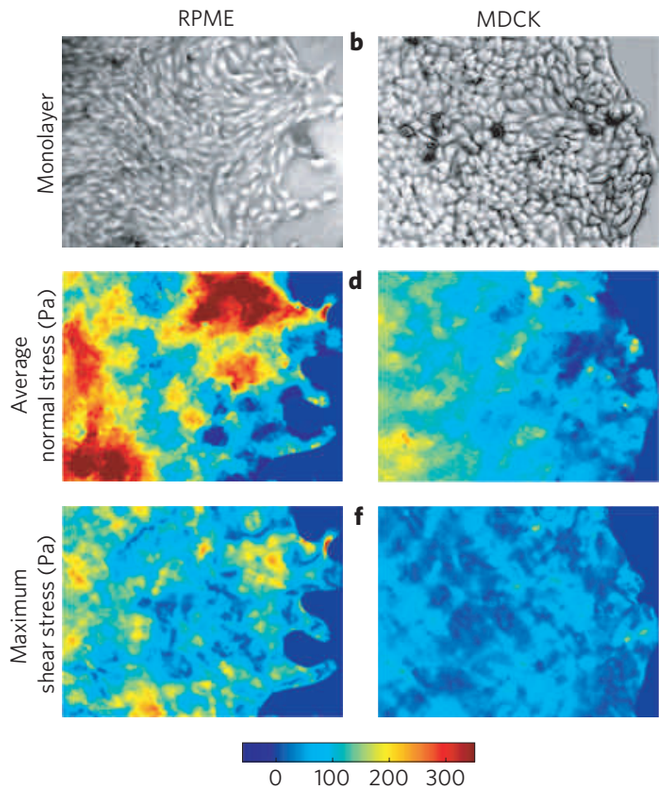
http://cognition.ups-tlse.fr/dynactom/

Active self-organization

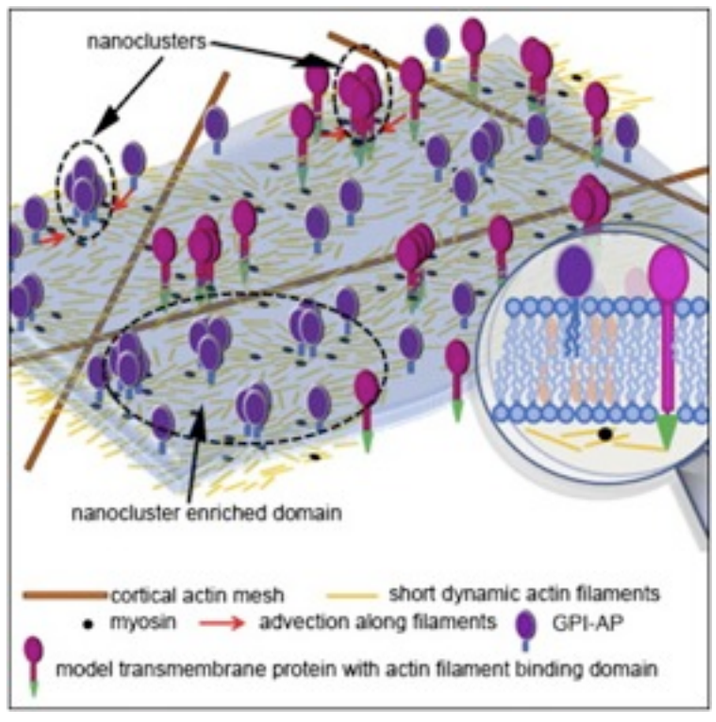
+ acto-myosin gels, cell-substrate interactions, nanoclustered domains on membranes, tissues ..



Alvarado et al, Nat Phys 2013



Tambe et al, Nat Mat 2011

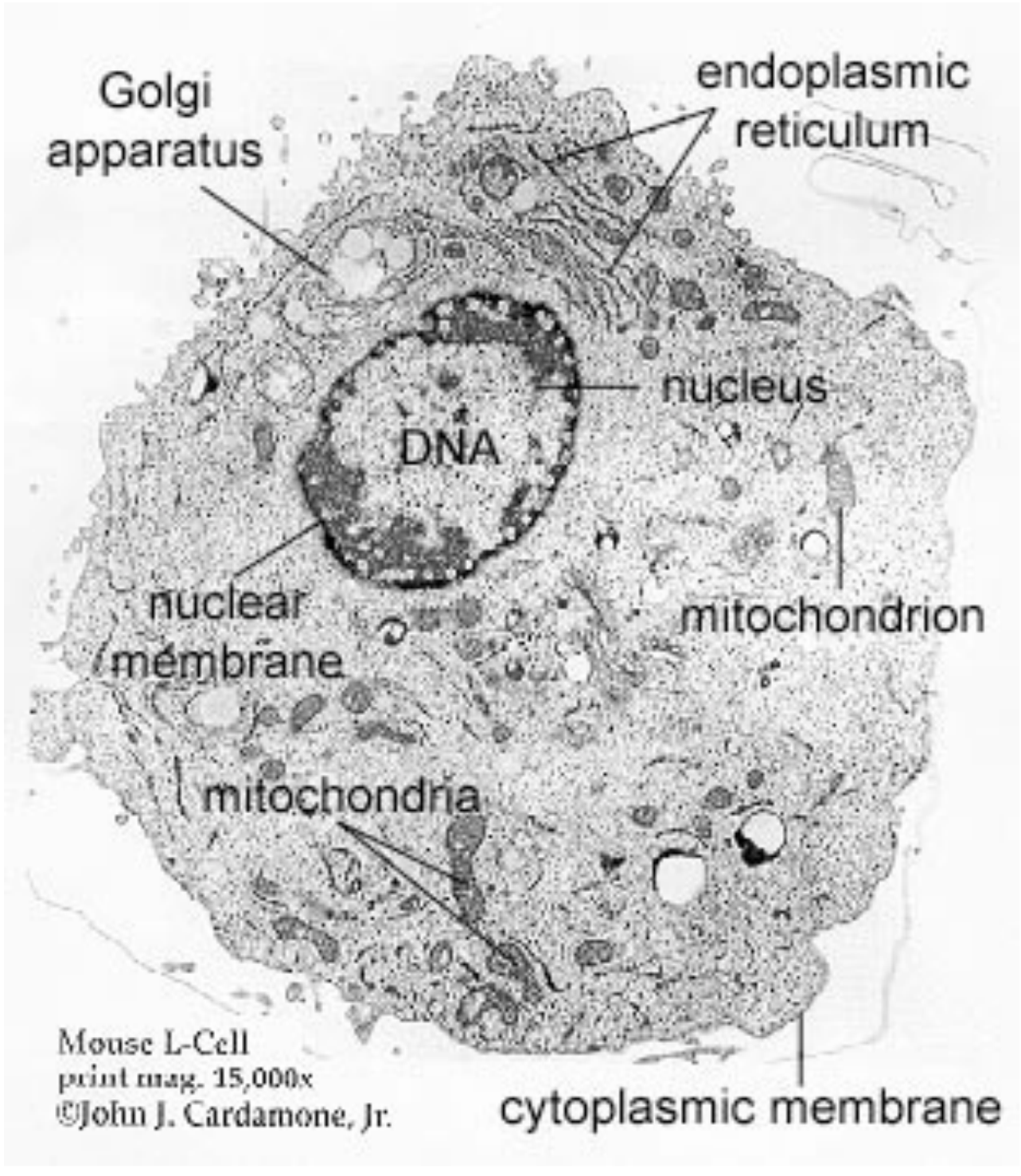


Gowrishankar et al, Cell 2012

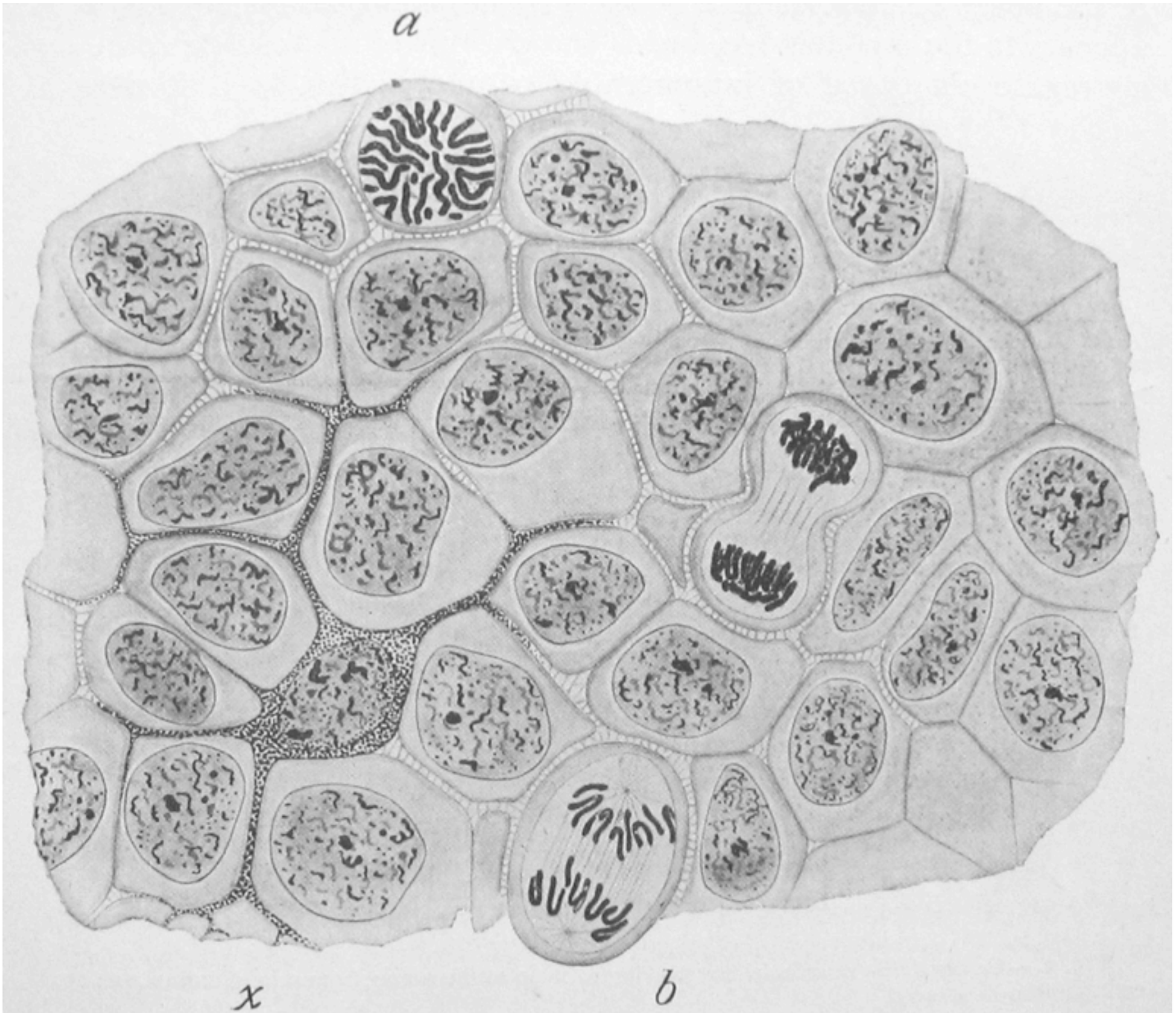
Other "tractable" *in vivo* active matter systems?

Chromatin in the interphase cell nucleus

Nucleus in eukaryotic cells

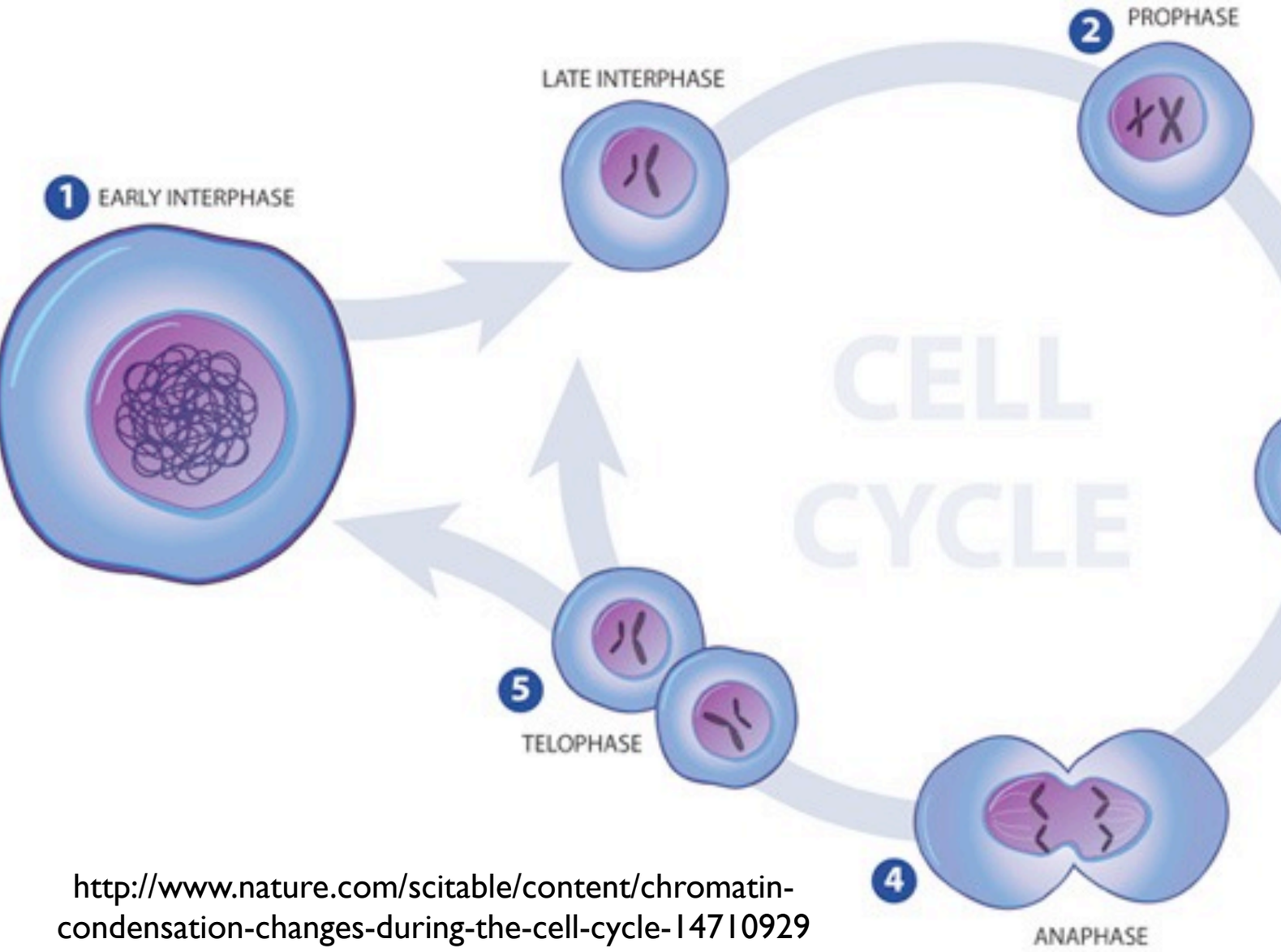


http://student.ccbcmd.edu/~gkaiser/biotutorials/eustruct/u1fig2.html



Wilson, Edmund B. (1900) The cell in Development and Inheritance (second edition). Wikimedia Commons

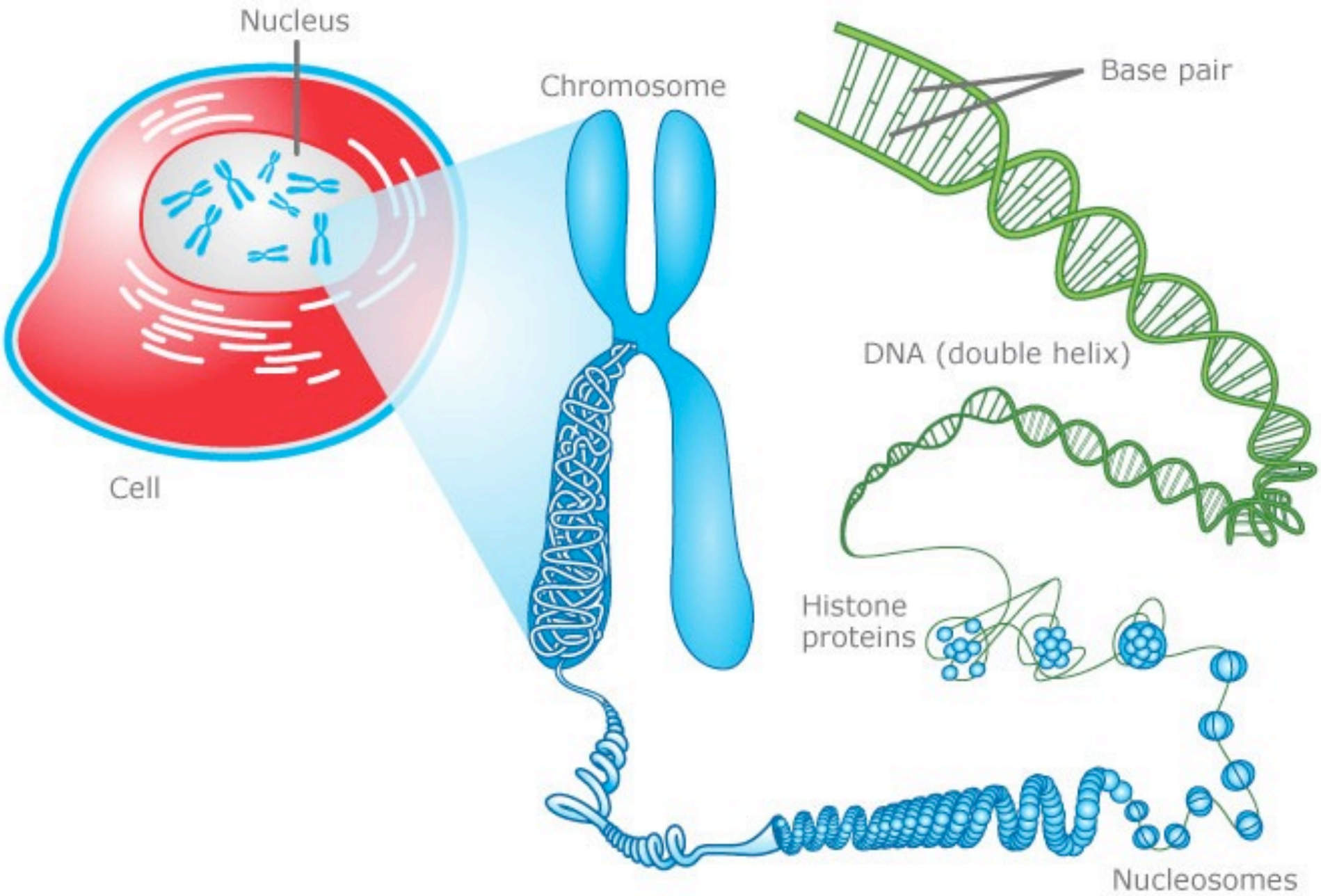
The Cell Cycle



http://www.nature.com/scitable/content/chromatin-condensation-changes-during-the-cell-cycle-14710929

http://www.answers.com/topic/cell-cycle

Chromosomes in interphase nuclei

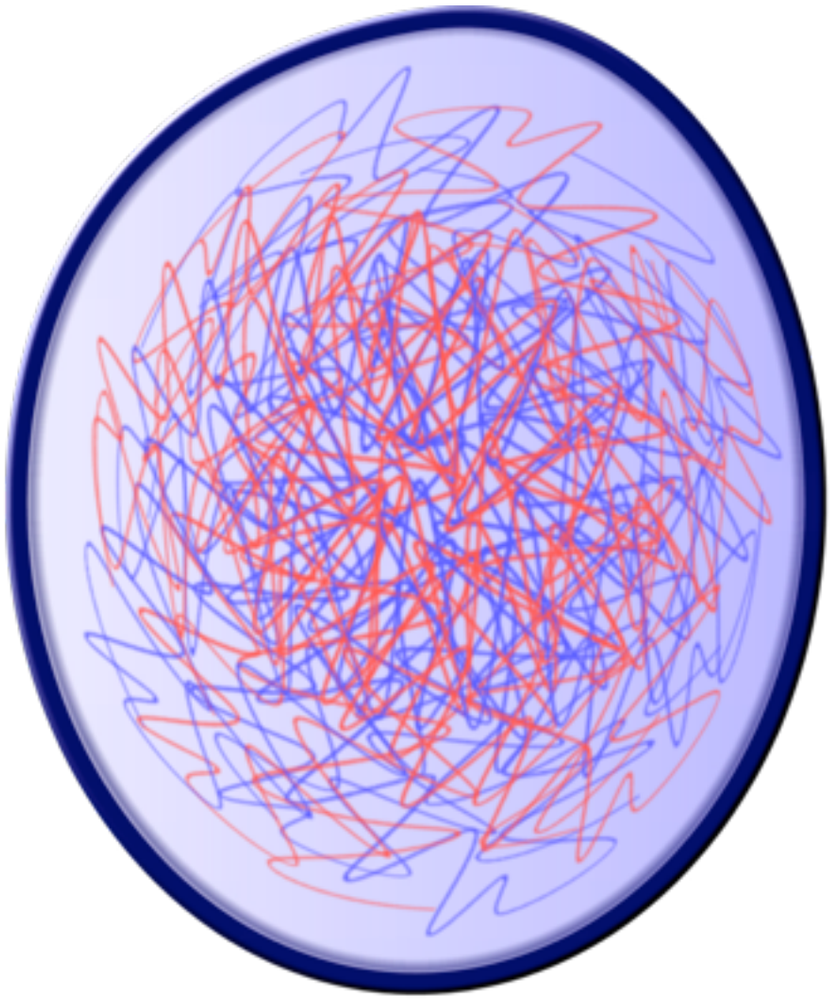


DNA packaged by histones.

Formed protein/DNA complex is chromatin (heterochromatin/euchromatin)

Structural entity of chromatin is the nucleosome

Eukaryotic interphase chromatin was thought for many years to have no obvious pattern of organization



http://www.edupic.net

During the 1970s and 1980s, most researchers seemed content with the assumption that the nucleus is filled with intermingling chromatin fibers and loops like a dish of spaghetti, an assumption widely reflected by textbooks of cell biology .....

T. Cremer and M. Cremer, Cold Spring Harb Perspect Biol (2010)

Ability to fluorescently label each chromosome (FISH), showed that chromosomes

- are **territorial**

- have **nonrandom arrangements**

Rabl 1885, Boveri 1908, Stack 1977, Cremer, Bickmore, Misteli, ..

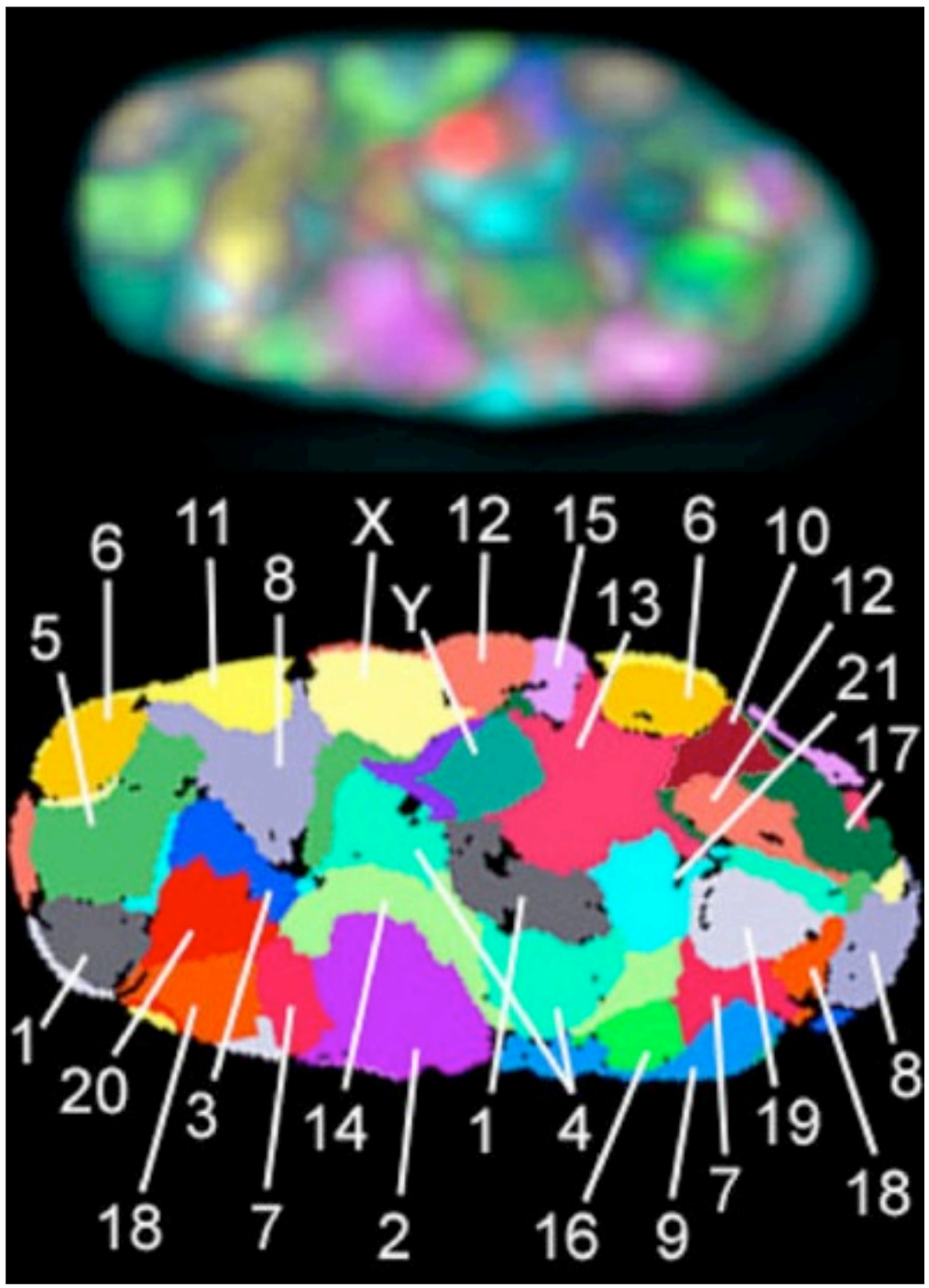
FISH = Fluorescence In-situ Hybridization

What governs the large-scale architecture of chromosome territories?

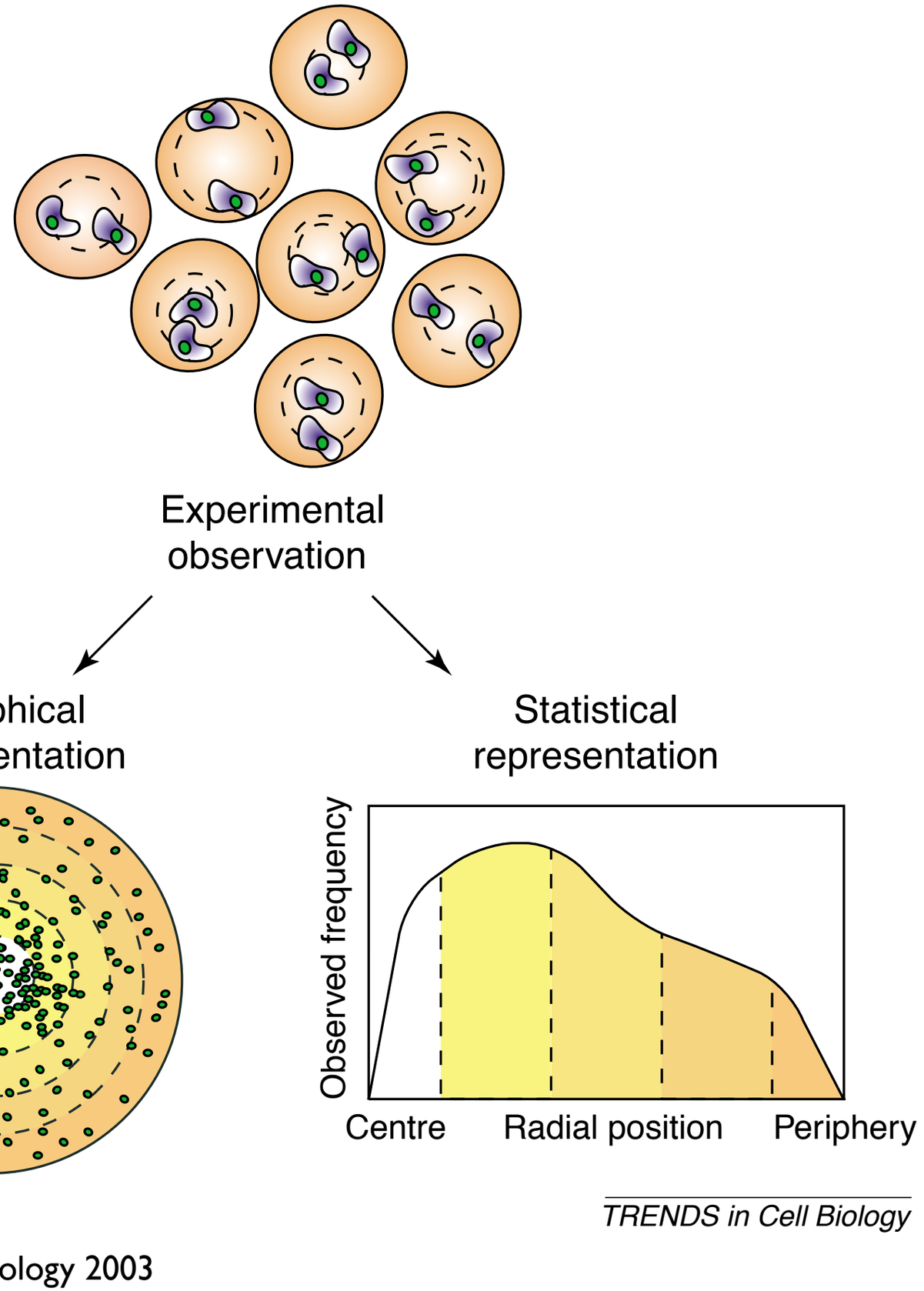
Three-Dimensional Maps of All Chromosomes in Human Male Fibroblast Nuclei and Prometaphase Rosettes

Andreas Bolzer[1¤], Gregor Kreth[2], Irina Solovei[1], Daniela Koehler[1], Kaan Saracoglu[3], Christine Fauth[4,5], Stefan Müller[1], Roland Eils[3], Christoph Cremer[2], Michael R. Speicher[4,5], Thomas Cremer[1*]

Bolzer et al, PloS Biology (2005)



Given that chromosomes occupy non-random locations, what are the 'positioning rules' that govern them?



Parada, Roix, Misteli, Trends in Cell Biology 2003

Differences in the Localization and Morphology of Chromosomes in the Human Nucleus

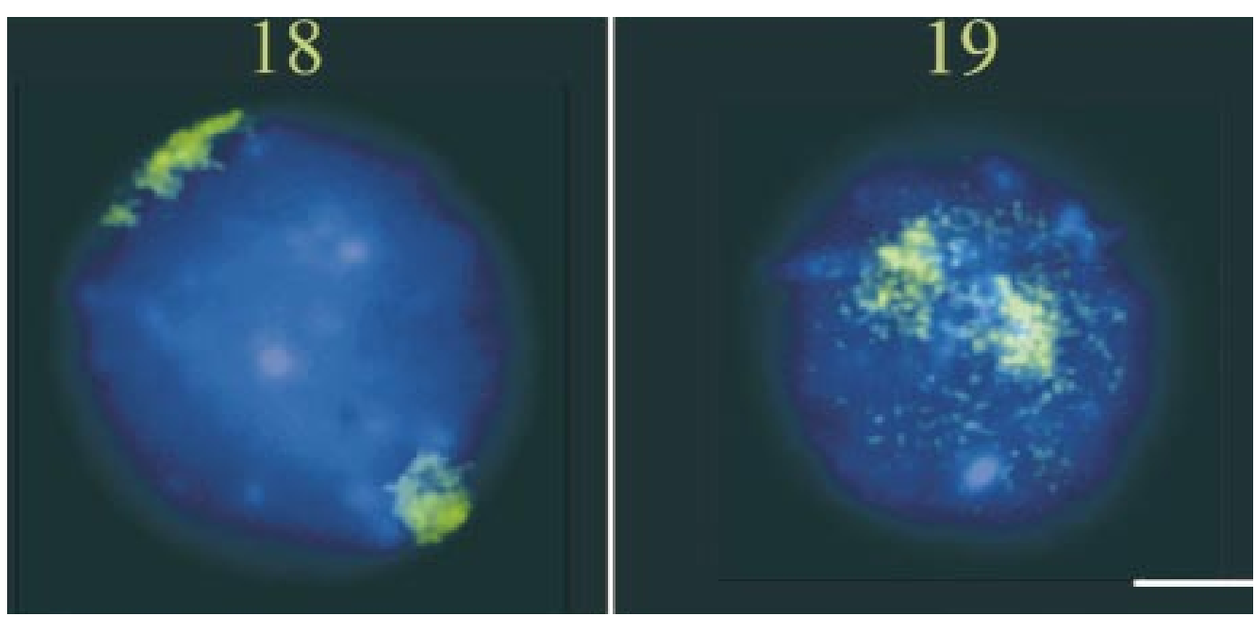
Jenny A. Croft, Joanna M. Bridger, Shelagh Boyle, Paul Perry, Peter Teague, and Wendy A. Bickmore

The Journal of Cell Biology, Volume 145, Number 6, June 14, 1999 1119–1131

Abstract. Using fluorescence in situ hybridization we show striking differences in nuclear position, chromosome morphology, and interactions with nuclear substructure for human chromosomes 18 and 19. Human chromosome 19 is shown to adopt a more internal position in the nucleus than chromosome 18 and to be more extensively associated with the nuclear matrix.

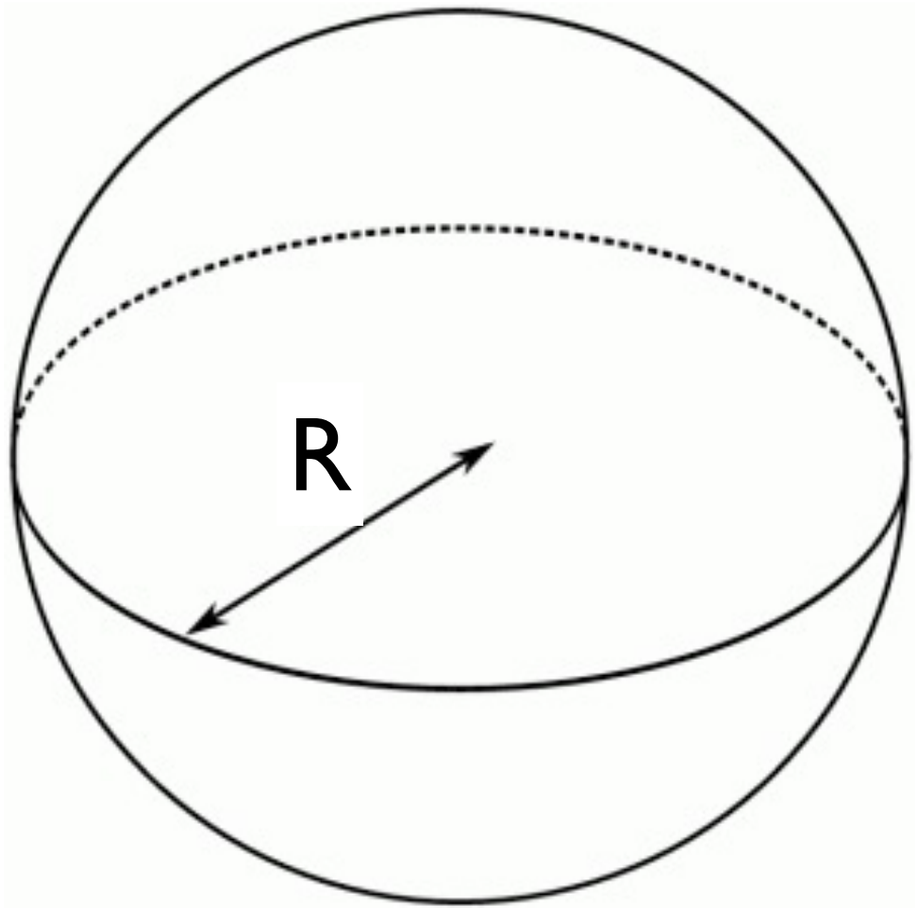
Chromosome 19:
62.03 genes/Mb and
60 Mb size

Chromosome18:
18.64 genes/Mb and
78 Mb size

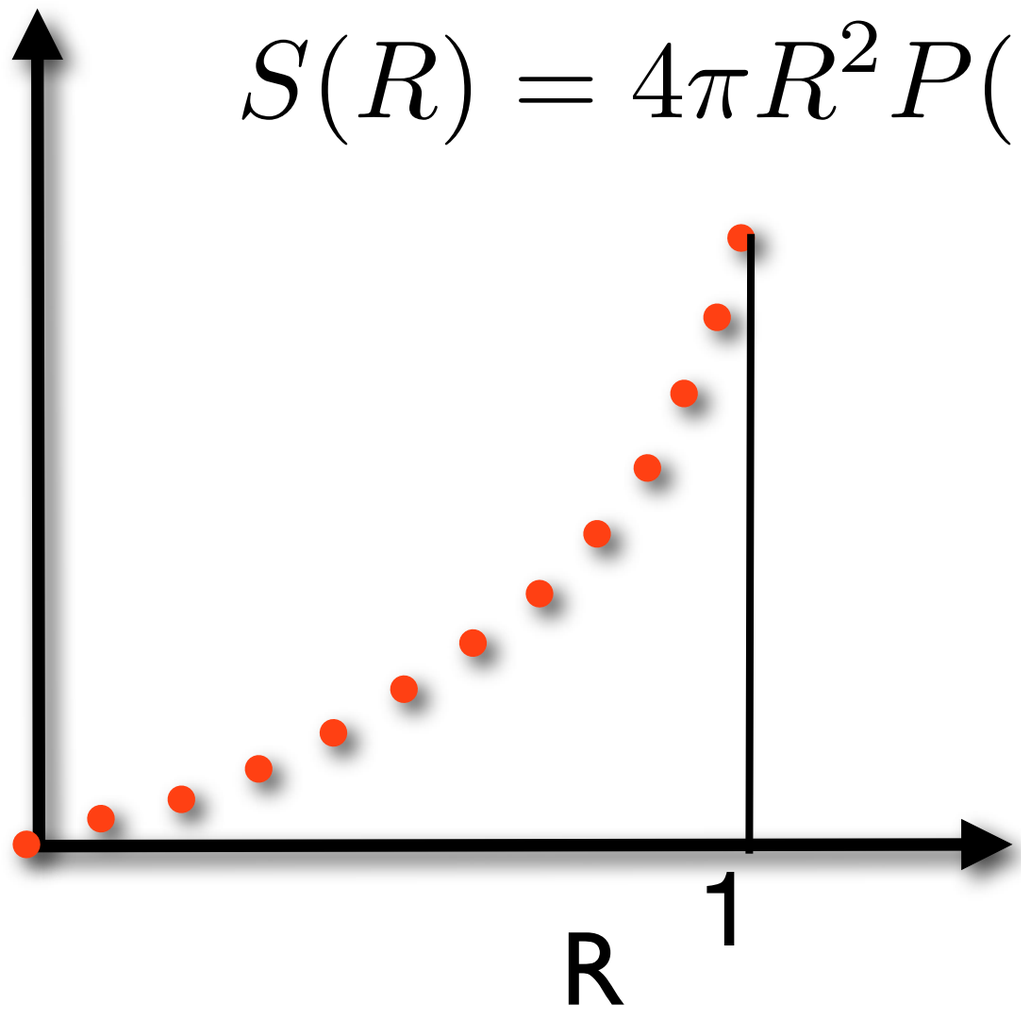


Radial chromosome positioning dependent on gene density?

Radial symmetry



$$S(R) = 4\pi R^2 P(|\vec{R}|)$$



If each chromosome is distributed uniformly throughout the nucleus, then S(R) quadratic, maximum at nuclear envelope

Good diagnostic for non-random placement

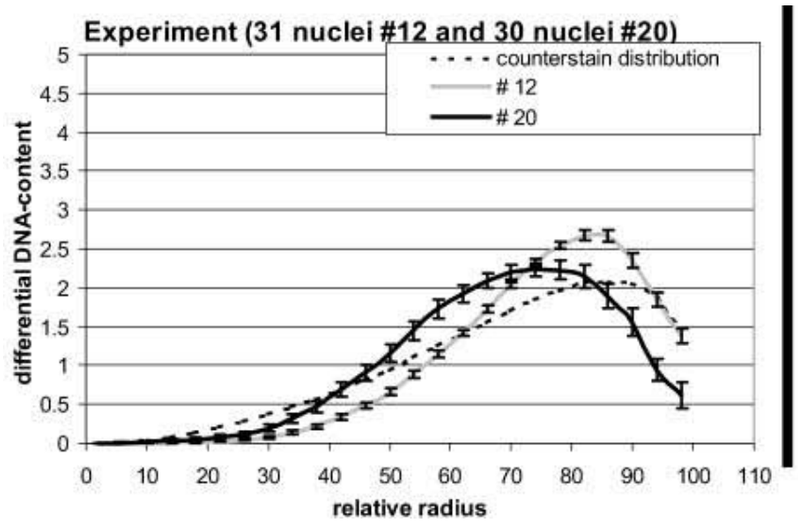
Cremer et al, Chrom. Res. 2001 Weierich et al Chrom Res. 2003, Kreth et al Biophys J 2005

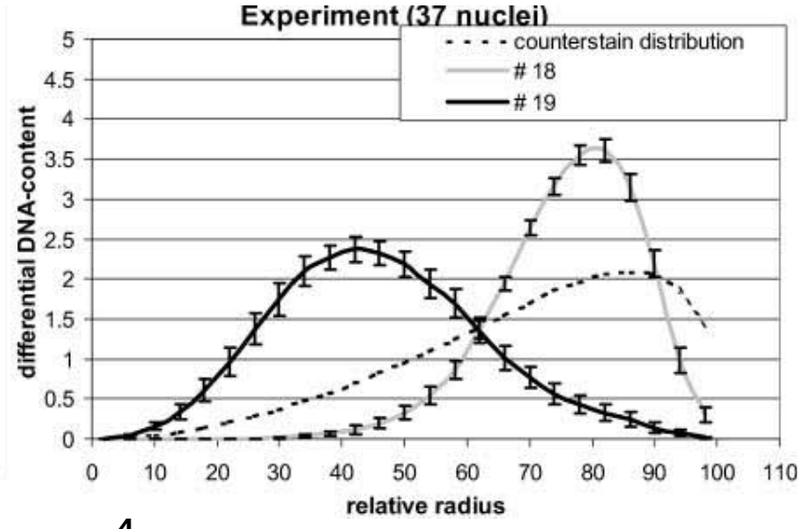
Chromosome 12:
30.92 genes/Mb
and 134 Mb size,

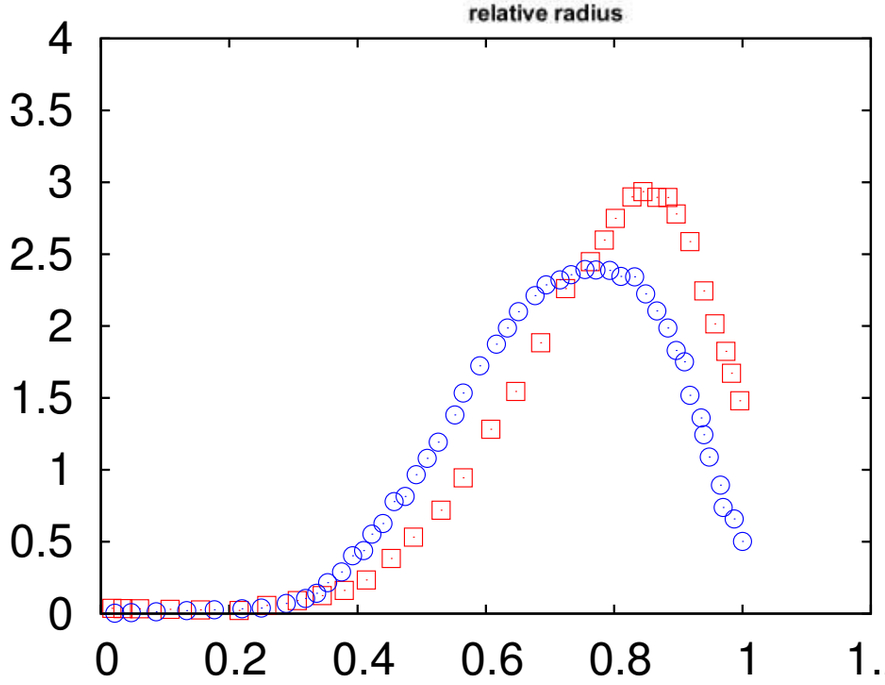
Chromosome 20:
29.71 genes/Mb
and 63 Mb size)

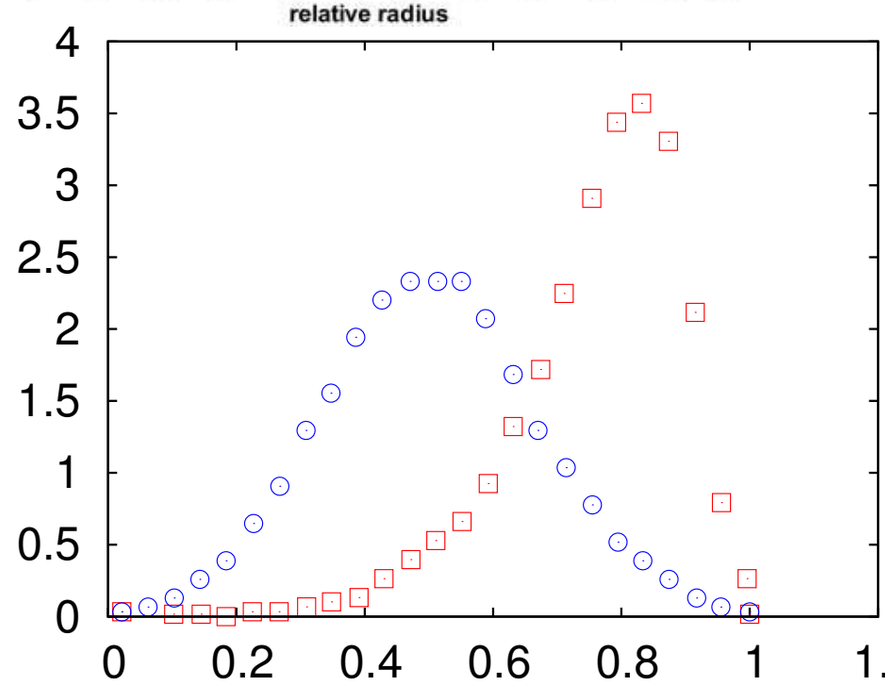
Chromosome 19:
62.03 genes/Mb
and 60 Mb size

Chromosome 18:
(red) 18.64 genes/
Mb and 78 Mb size





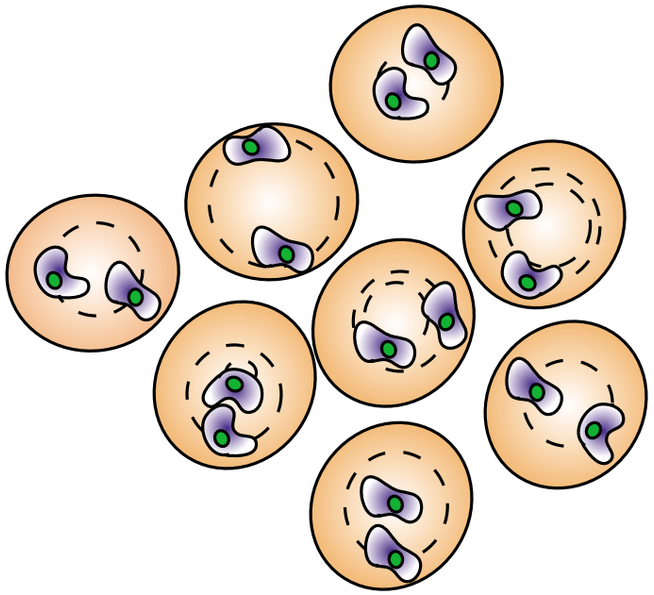




Chromosomes 12 and 20 have different sizes but the same gene density

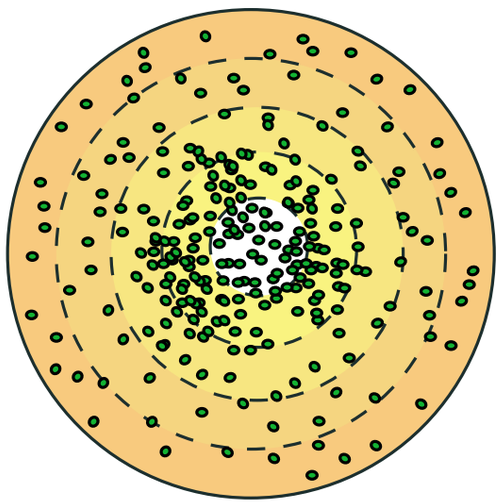
Chromosomes 18 and 19 have roughly the same size but different gene densities

Given that chromosomes are (in many but not all cases) radially ordered by gene density, how do we understand this specific 'positioning rule'?

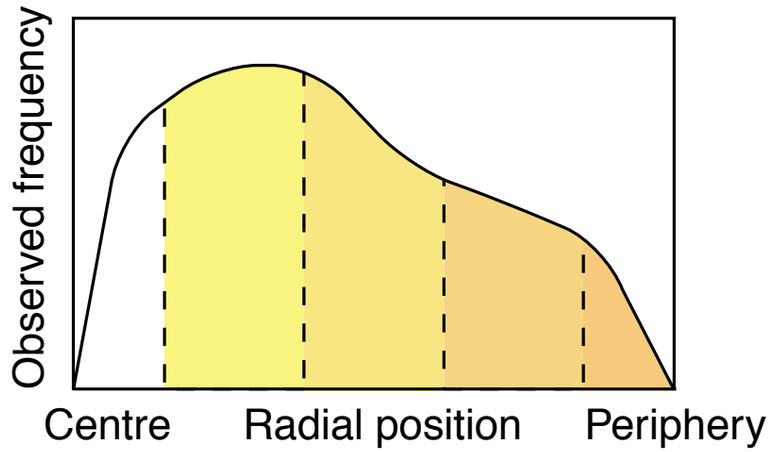


Experimental observation

Graphical representation



Statistical representation

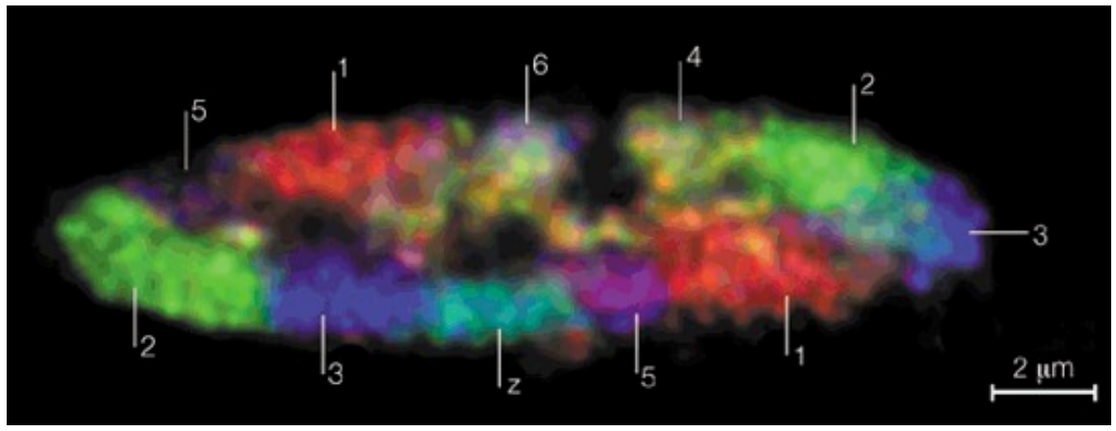


TRENDS in Cell Biology

Parada, Roix, Misteli, Trends in Cell Biology 2003

Hypothesize:

Non-equilibrium mechanical activity (transcription and chromatin remodeling machinery), inhomogeneous across gene-rich and gene-poor regions gives gene-density-based radial segregation



T.Cremer and C. Cremer, Nature Reviews Genetics vol. 2, no. 4, pp. 292-301 (April, 2001)

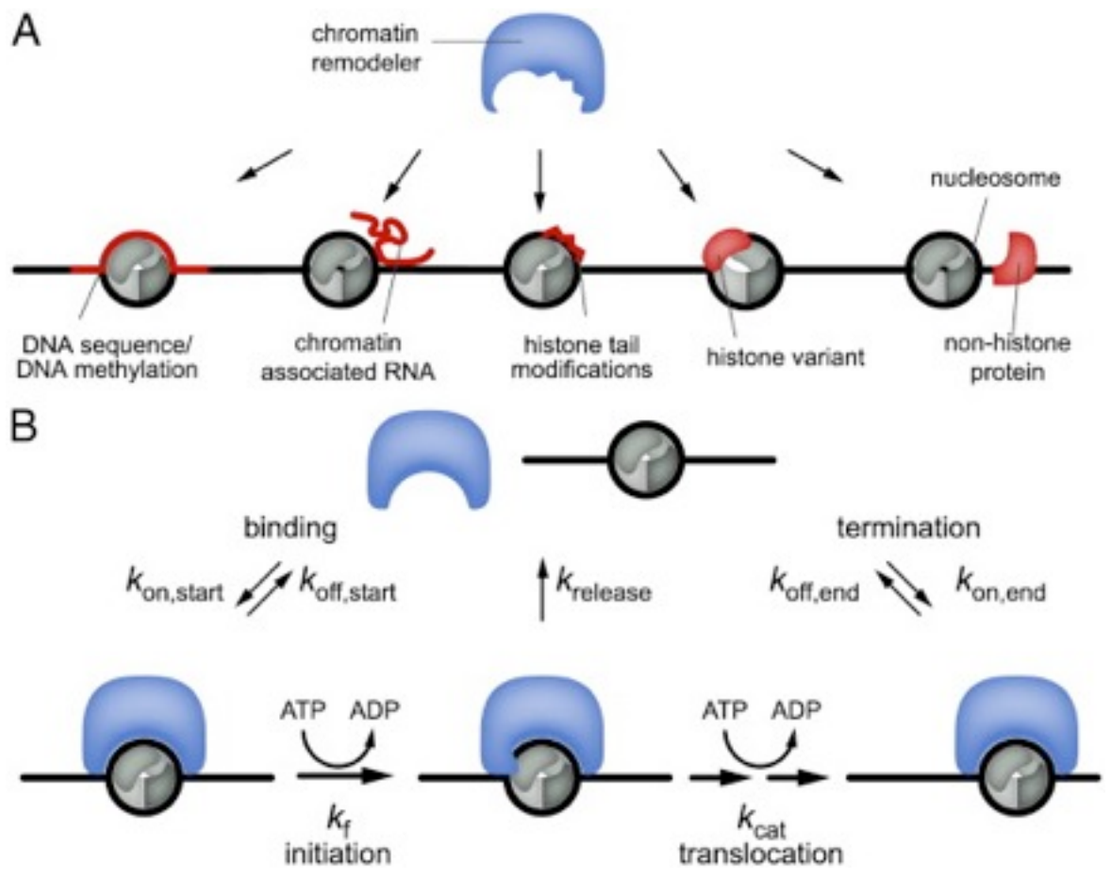
No self-propulsion - unlike in all active matter models studied so far - but inhomogeneous activity, confinement and polymer character all important

Chromosome territories a natural consequence of compact chromosome configurations and activity

Groups of ATPases classified into the Snf2, ISWI, Mi-2, Chd1, Ino80, ERCC6, ALC1, CHD7, Swr1, RAD54, and Lsh subfamilies

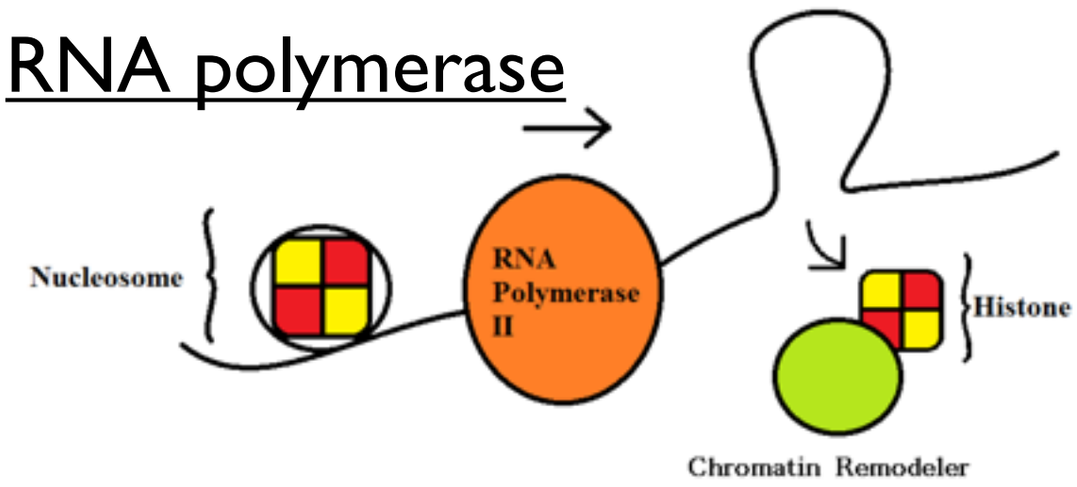
Chromatin Remodelers

ATP-driven molecular machines change chromatin structure, translocate nucleosomes, evict nucleosomes, change nucleosomal histone composition, involved in transcription. Work in concert through remodeler-specific interactions. Highly dynamic. Present at large density



Erdel et al, BBA (2011)

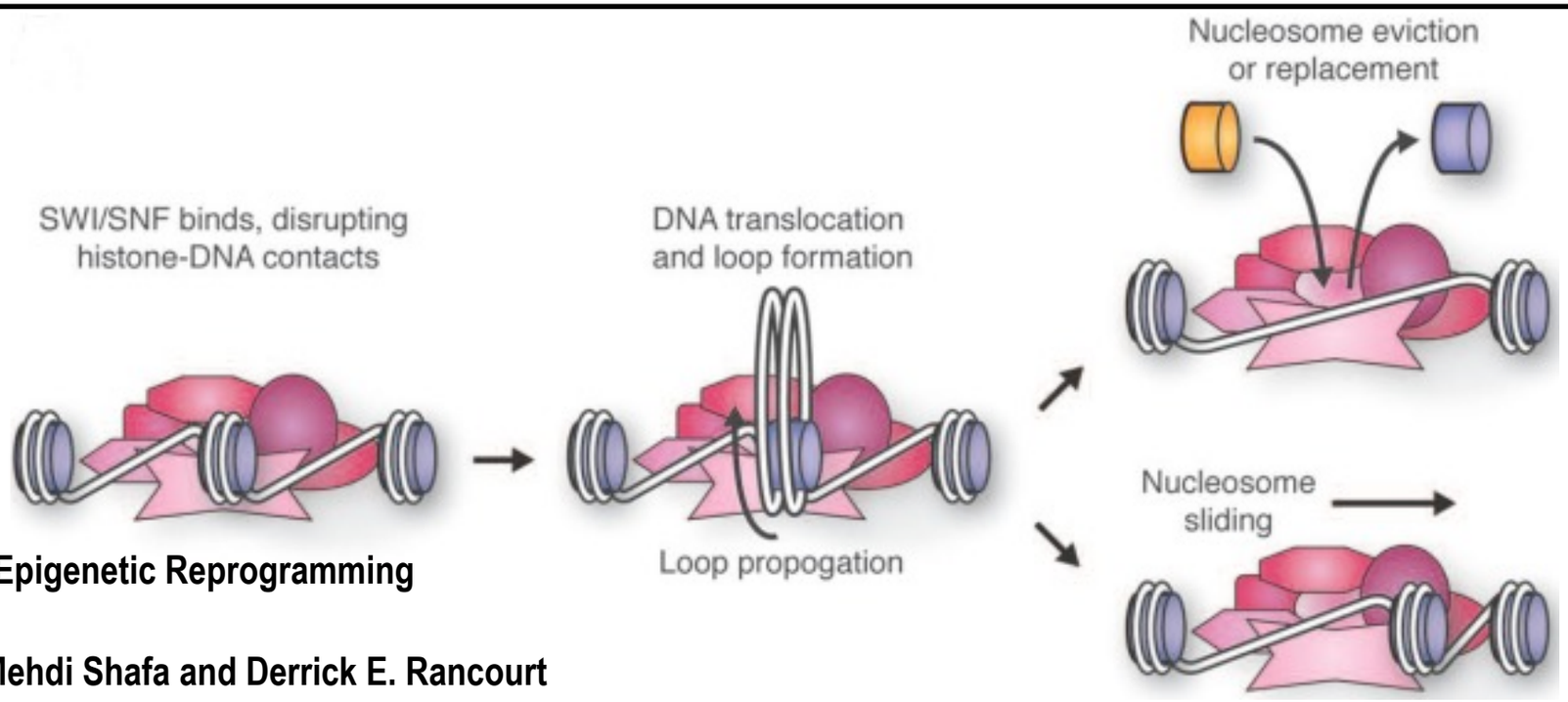
RNA polymerase



Barriers to nucleosome motion: 20-40 kT

http://upload.wikimedia.org/wikipedia/commons/e/e9/RNA_Polymerase_II_Transcription.png

‘Mechanical’ effects of active chromatin remodelers?



Stem Cells and Epigenetic Reprogramming

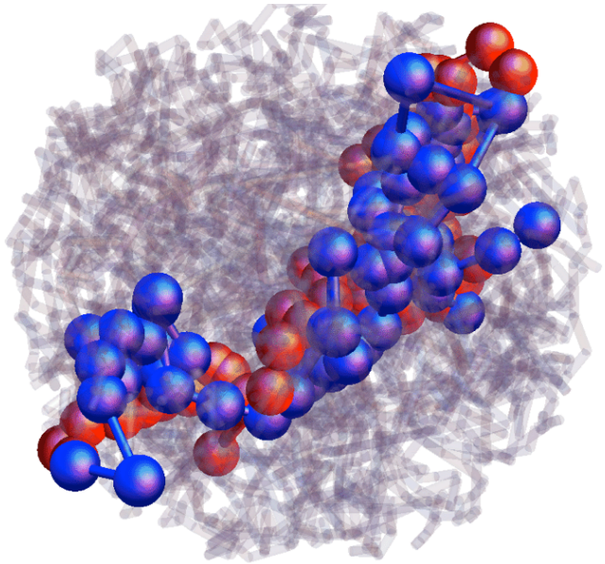
By Perla Cota, Mehdi Shafa and Derrick E. Rancourt

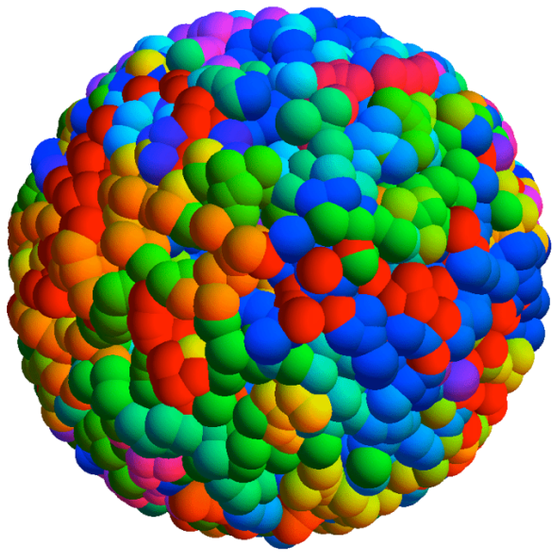
in "Pluripotent Stem Cells", ed. Deepa Bhartiya and Nibedita Lenka, Intech (2013)

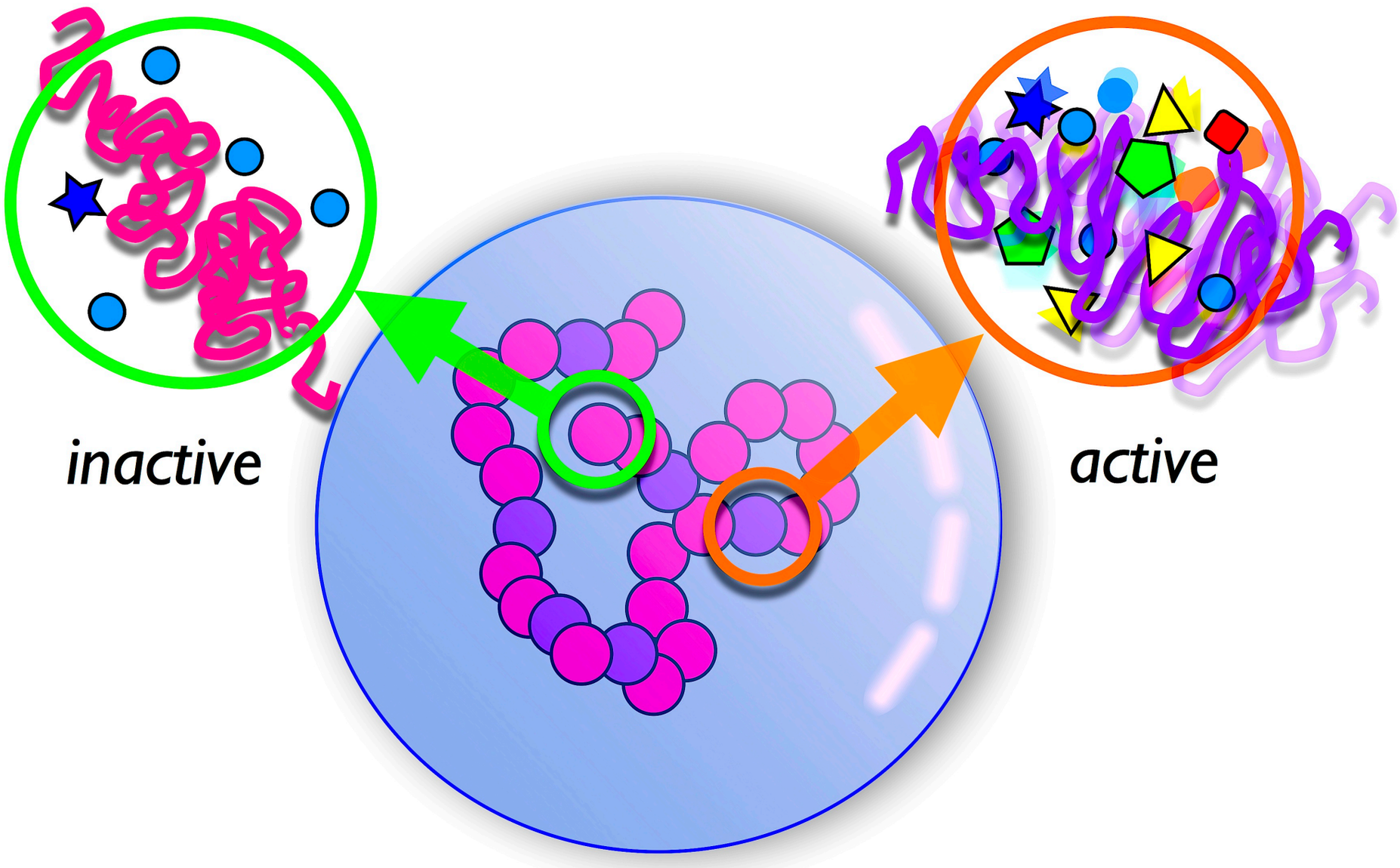
1. Effective local ‘active temperature’ T_{act}, associated with gene density. More transcription implies more ATP-consuming activity from remodeling/transcription-coupled enzymes

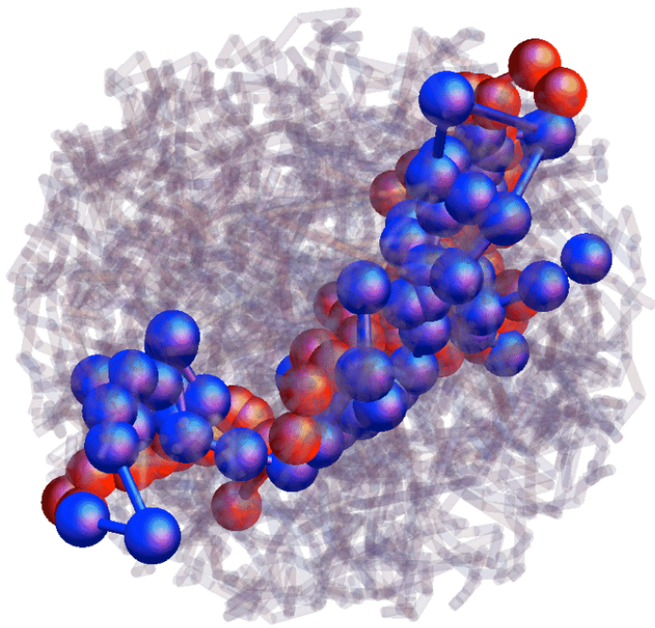
2. Coarse-grain to athermal noise acting on, 1Mb scale monomers (Known to be appropriate to functional units of chromosome territories)

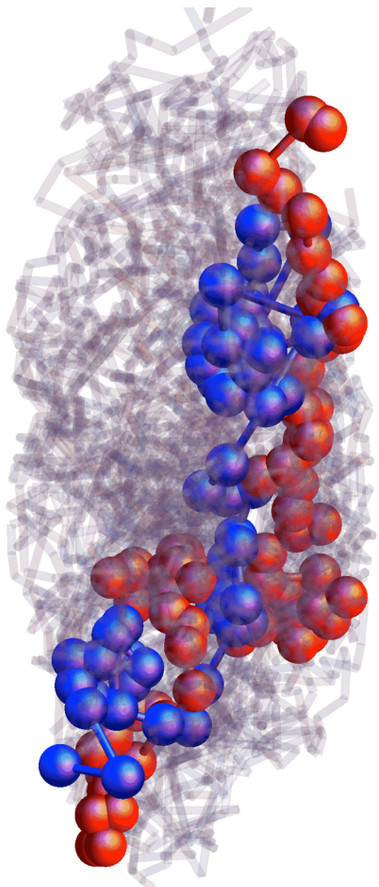
3. Estimate scales of active temperature (barriers to nucleosome motion, etc): $T_{act} = 20 T_{therm}$

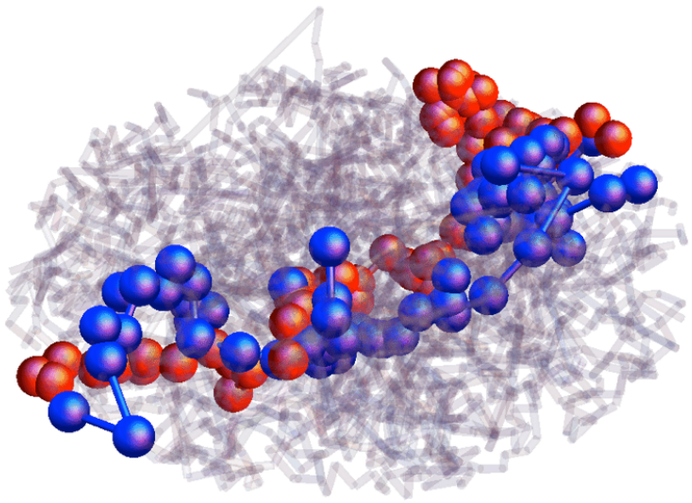





inactive
active







Simulations using the SCD model (spherical chromatin domain)

$$\zeta \frac{d\mathbf{r}_i}{dt} = \mathbf{F}_i + \eta_i$$

$$\langle \eta_i^\alpha(t)\eta_j^\alpha(t')\rangle = 2k_B T_i \zeta \delta_{ij}\delta(t-t').$$

Brownian dynamics, 3-d: 6098 monomers

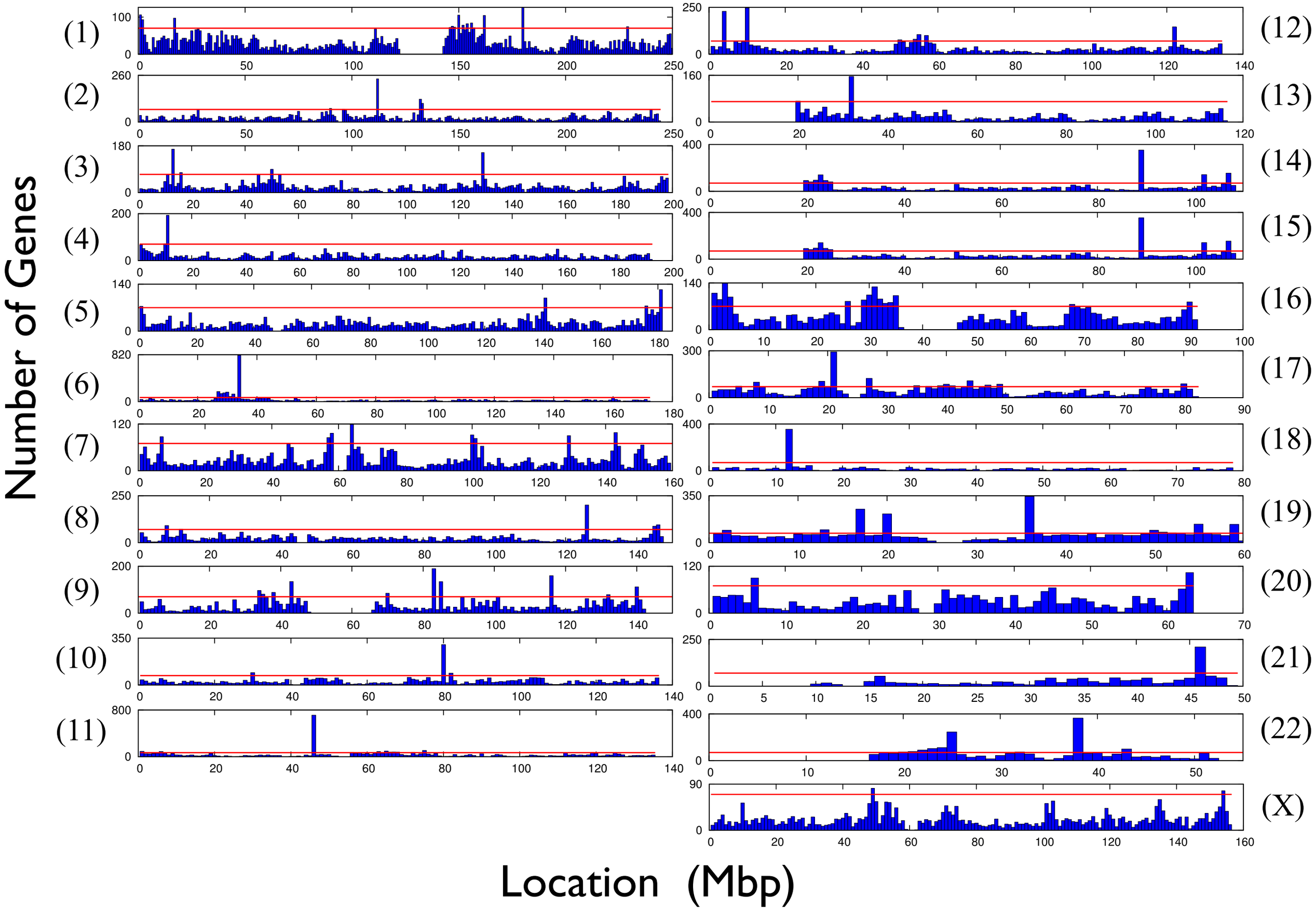
Different effective temperature for each monomer

Assign this depending on gene density (very nonlinearly)

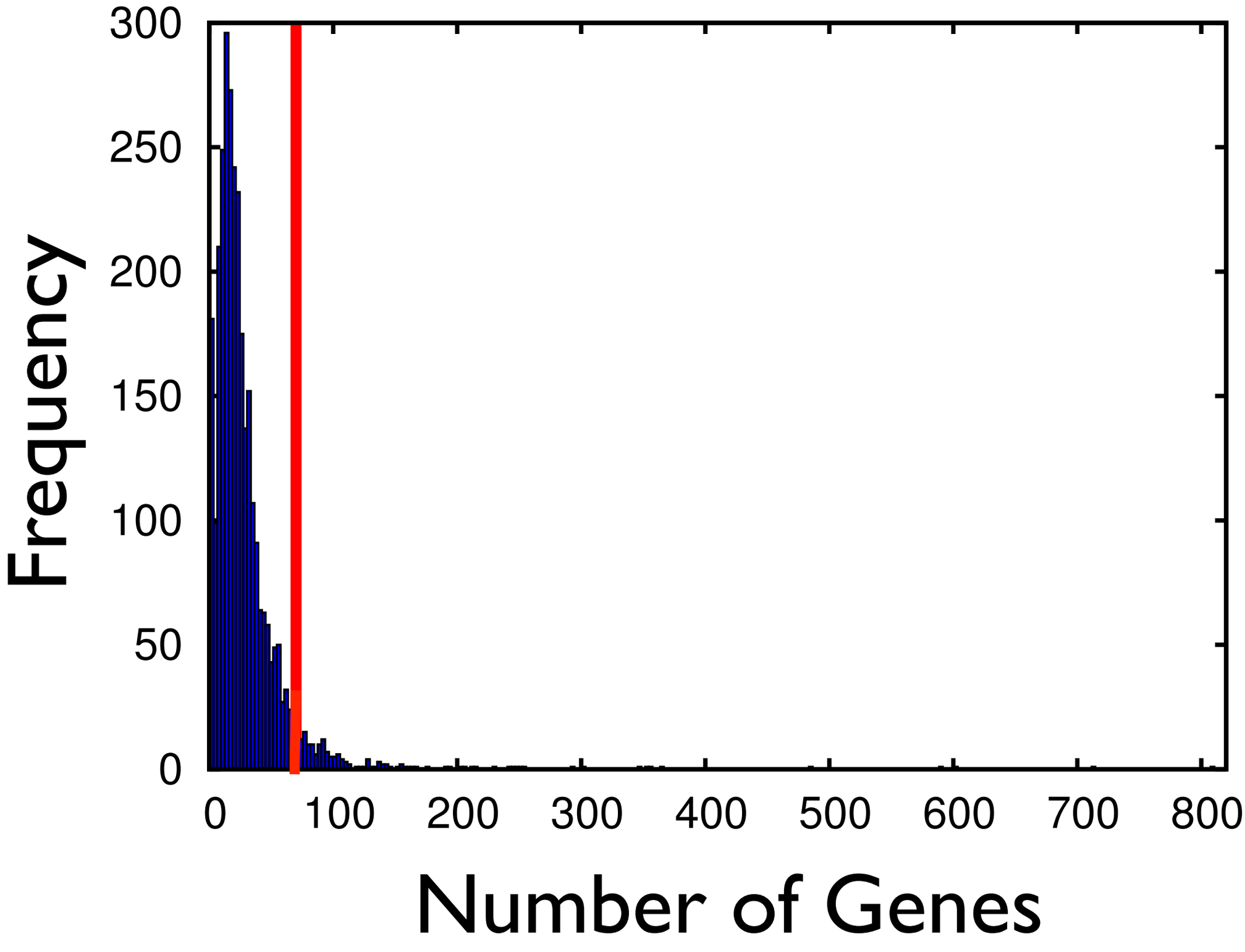
Varied nuclear shape: spherical, ellipsoidal

Passive and active confinement

GeneCards: Rebhan,M., Chalifa-Caspi,V., Prilusky,J. and Lancet,D. (1998) GeneCards: a novel functional genomics compendium with automated data mining and query reformulation support. Bioinformatics, 14, 656–664.

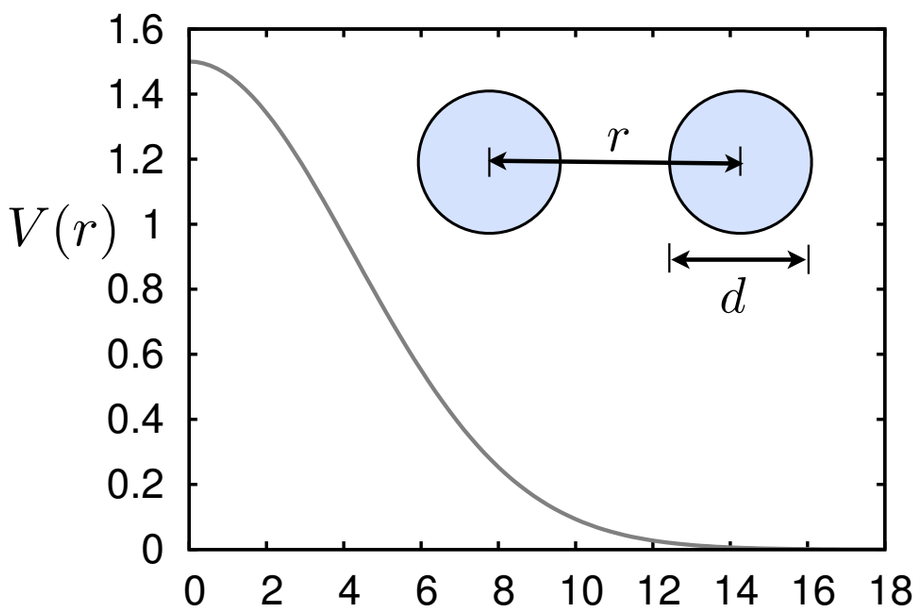


Assigning activity to each monomer, from gene densities



$$V_{neighbour\ monomers}(\mathbf{r}_i, \mathbf{r}_{i+1}) = \frac{1}{2}k(|\mathbf{r}_i - \mathbf{r}_{i+1}|)^2$$

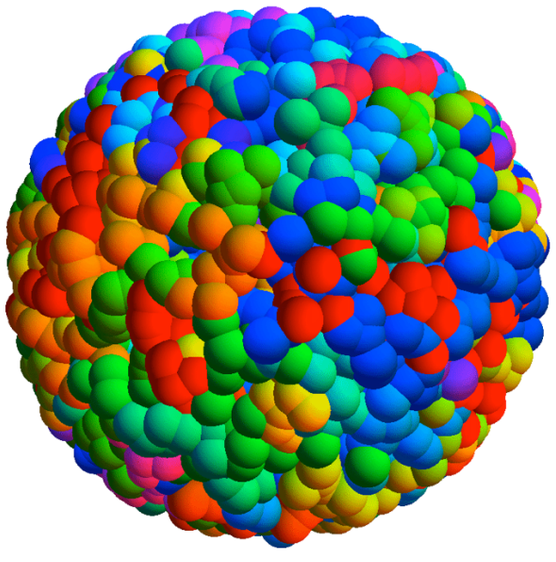
Monomer-Monomer Interactions



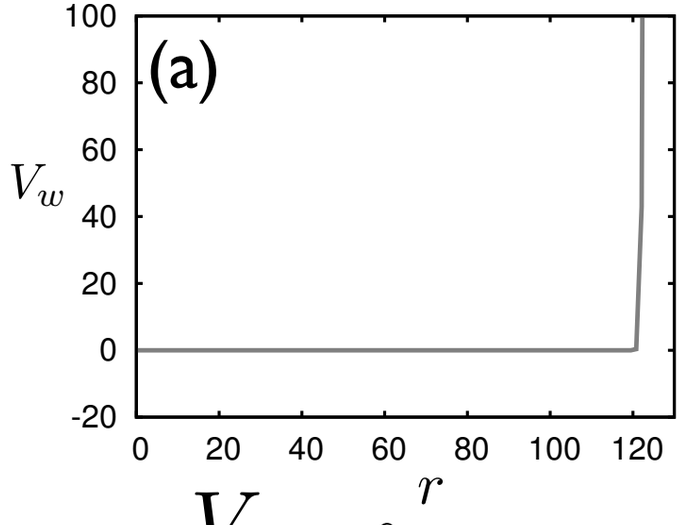
$$d \simeq 500nm,$$
$$R_0 \simeq 6.7\mu m$$
$$\ell_0 \simeq 600nm$$
$$V_0 \simeq 1.5 k_B T_{eq}$$

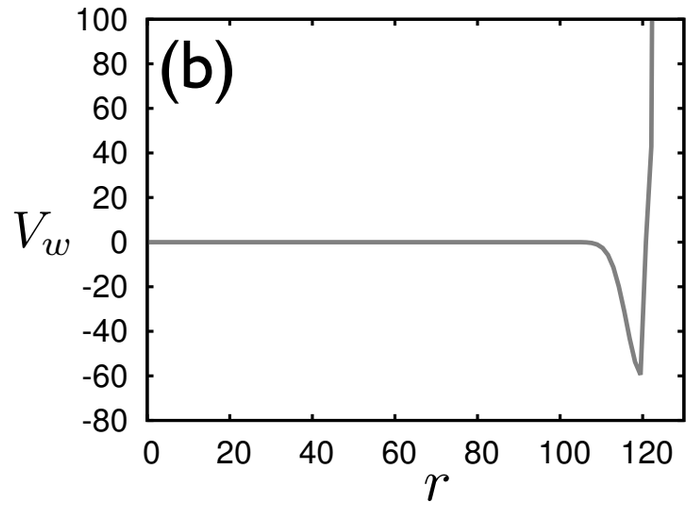
Self-repelling but not self-avoiding

$$V_{monomer-monomer}(\mathbf{r}_i, \mathbf{r}_j) = V_0 \exp\left(-|\mathbf{r}_i - \mathbf{r}_j|^2/\sigma^2\right)$$



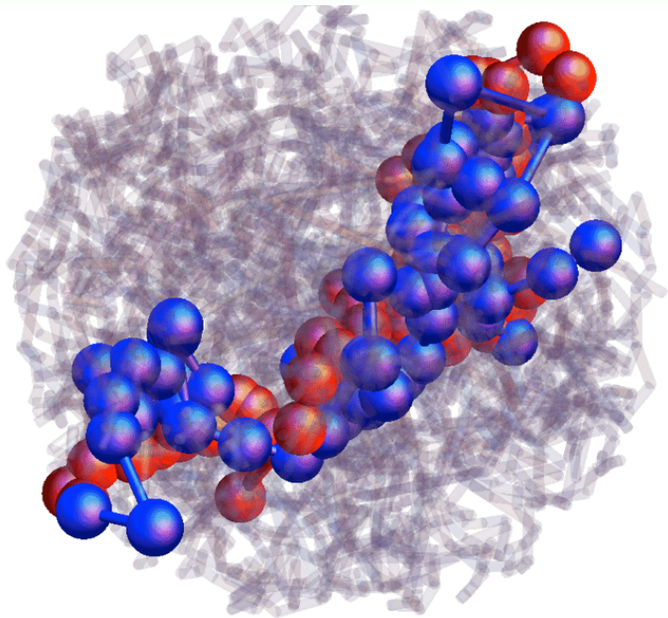
Monomer-Wall Interactions



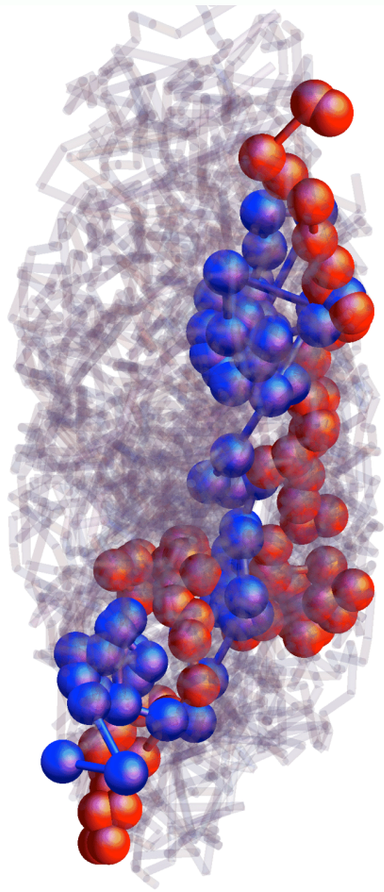


$$V_{wall}(\mathbf{r}_i) = \frac{V_{conf}}{a^5}(|\mathbf{r}_i| - R_0)^5, \qquad |\mathbf{r_i}| > R_0$$

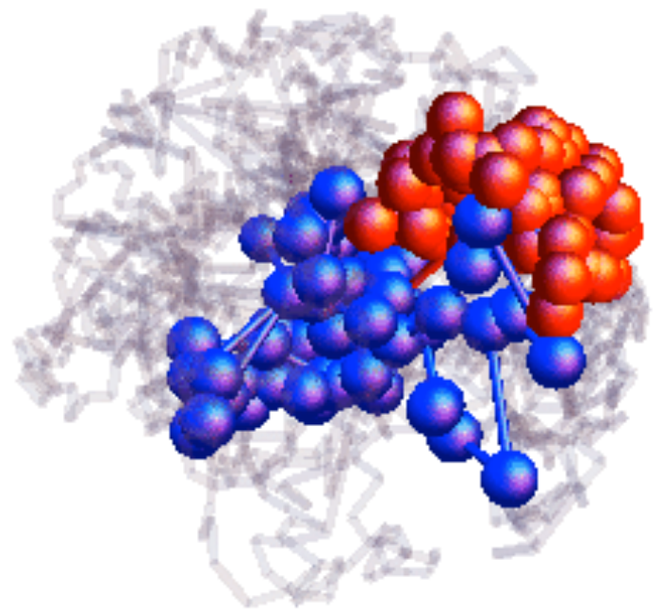
Simulation Configurations



Spherical Confinement, Equilibrium



Ellipsoidal Confinement, Equilibrium



Spherical Confinement, non-Equilibrium

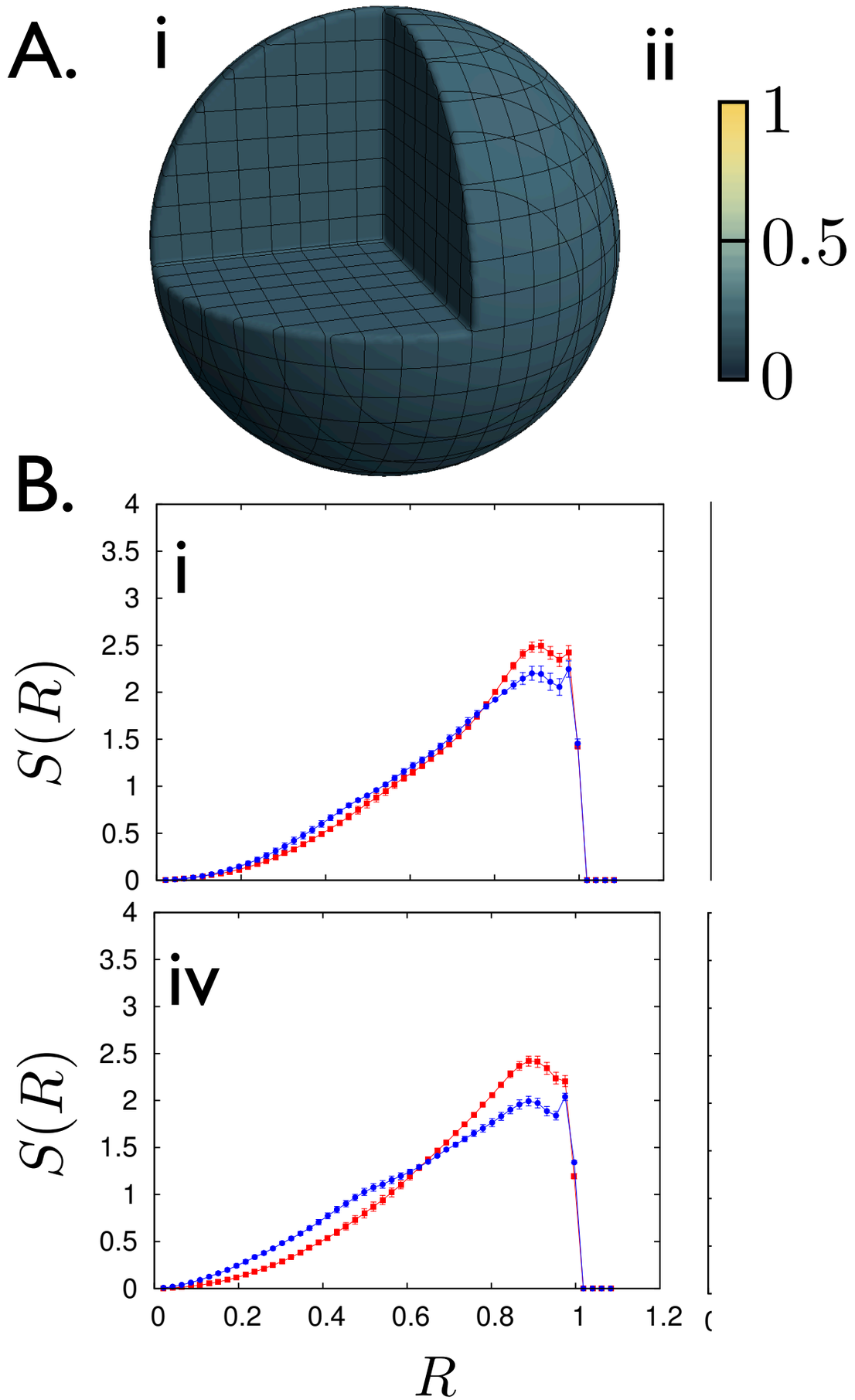
Cut-away sphere representation of local activity

Thermal equilibrium

Chromosome 18/19

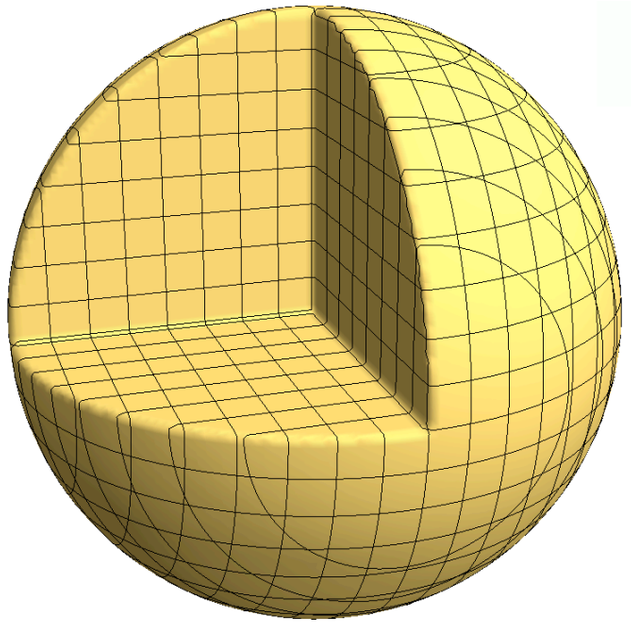
$T = T_{therm}$

Chromosome 12/20

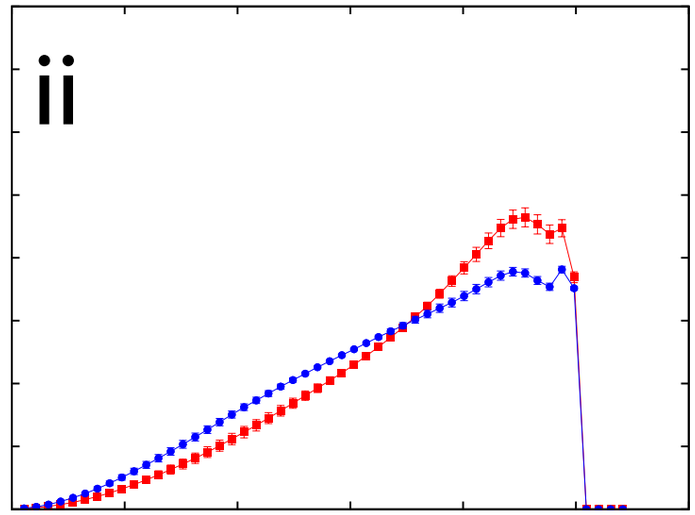


Cut-away sphere representation of local activity

All monomers at uniform high active temperature

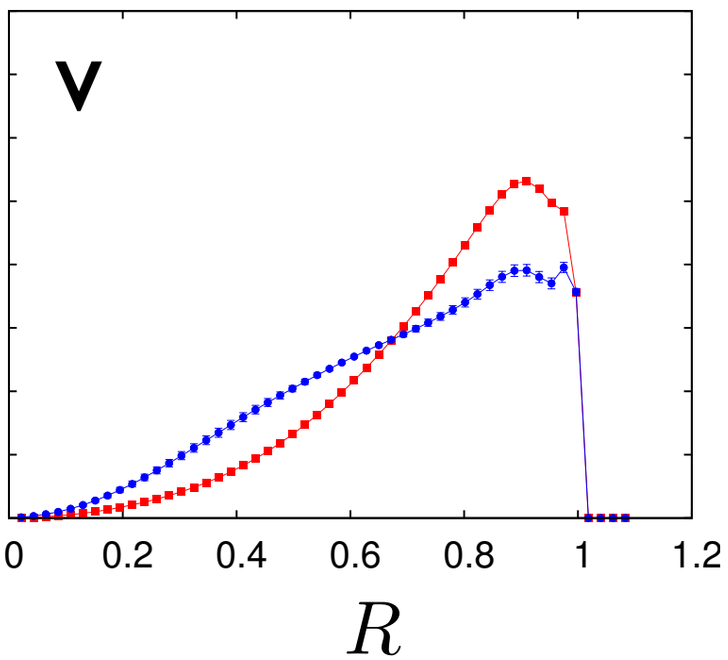


Chromosome 18/19

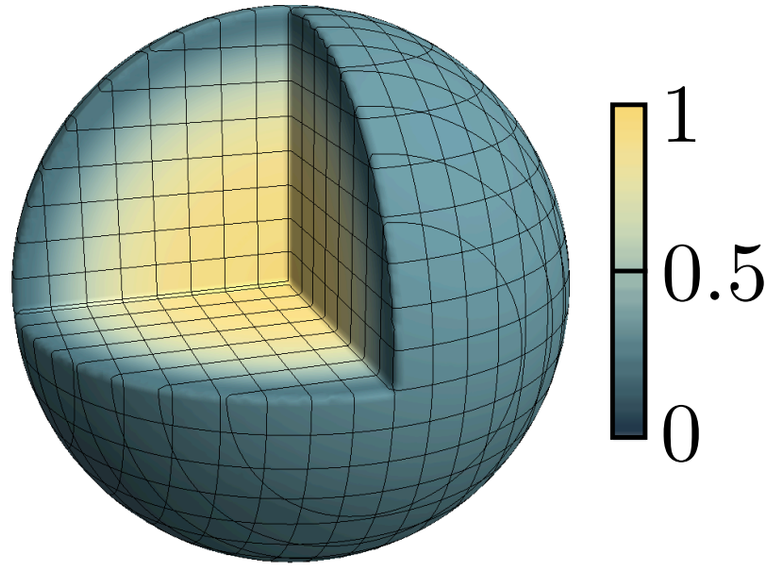


$T = 20\ T_{therm}$

Chromosome 12/20



Cut-away sphere
representation of local activity

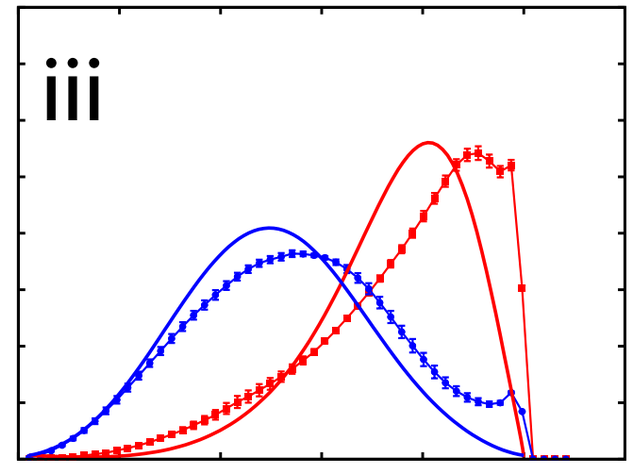


Mixture of monomers at different
active temperatures

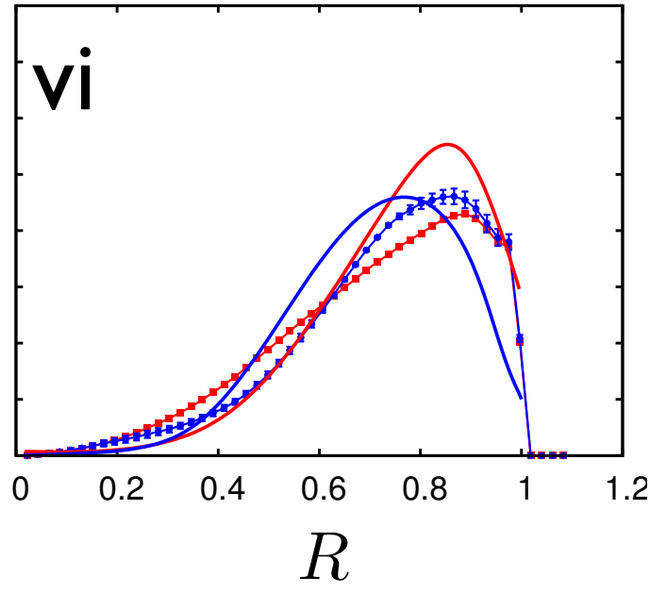
95% at $T = T_{therm}$

5% at $T = 20\ T_{therm}$

Chromosome 18/19

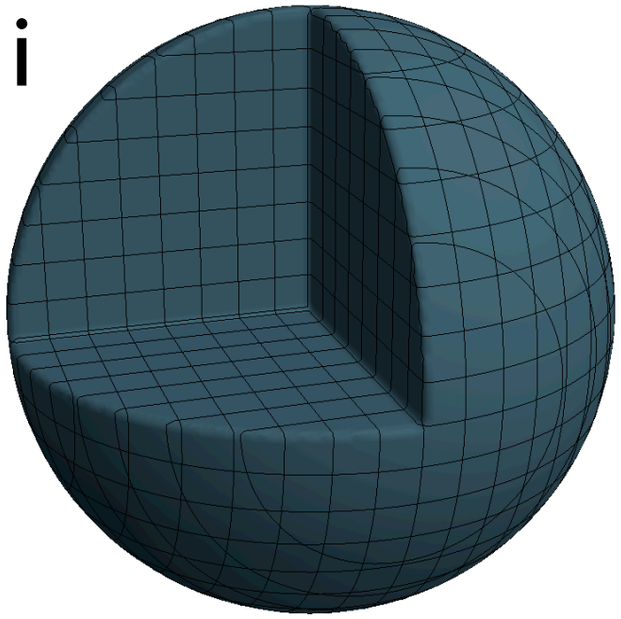


Chromosome 12/20

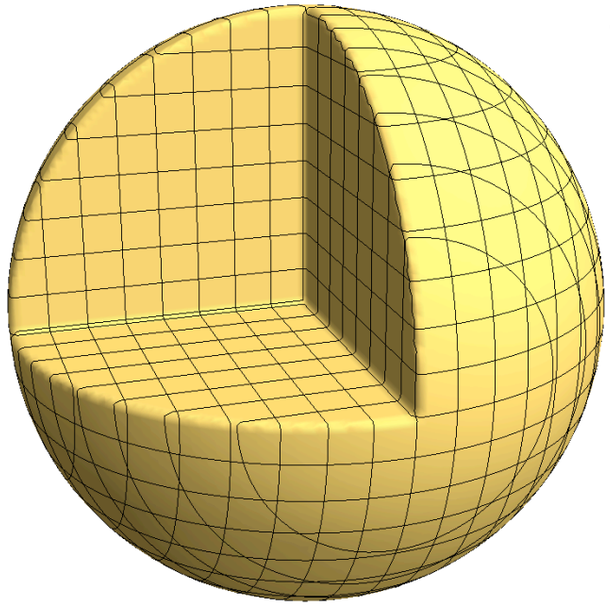


A.

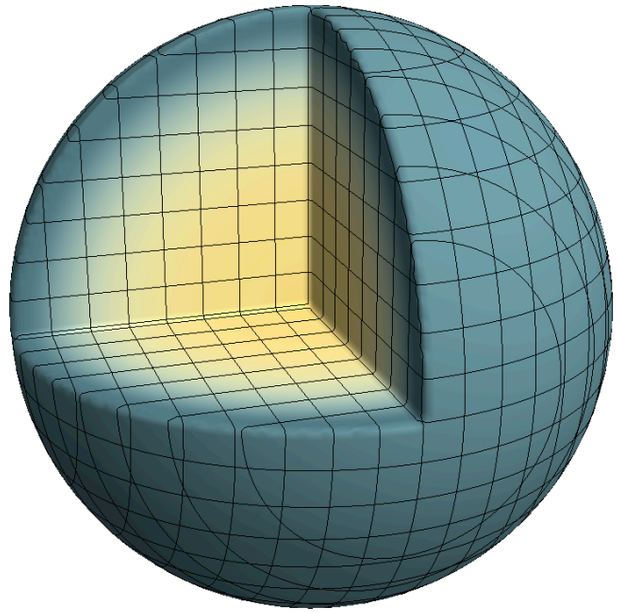
i

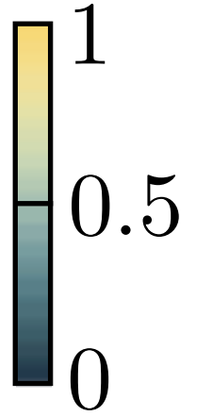


ii

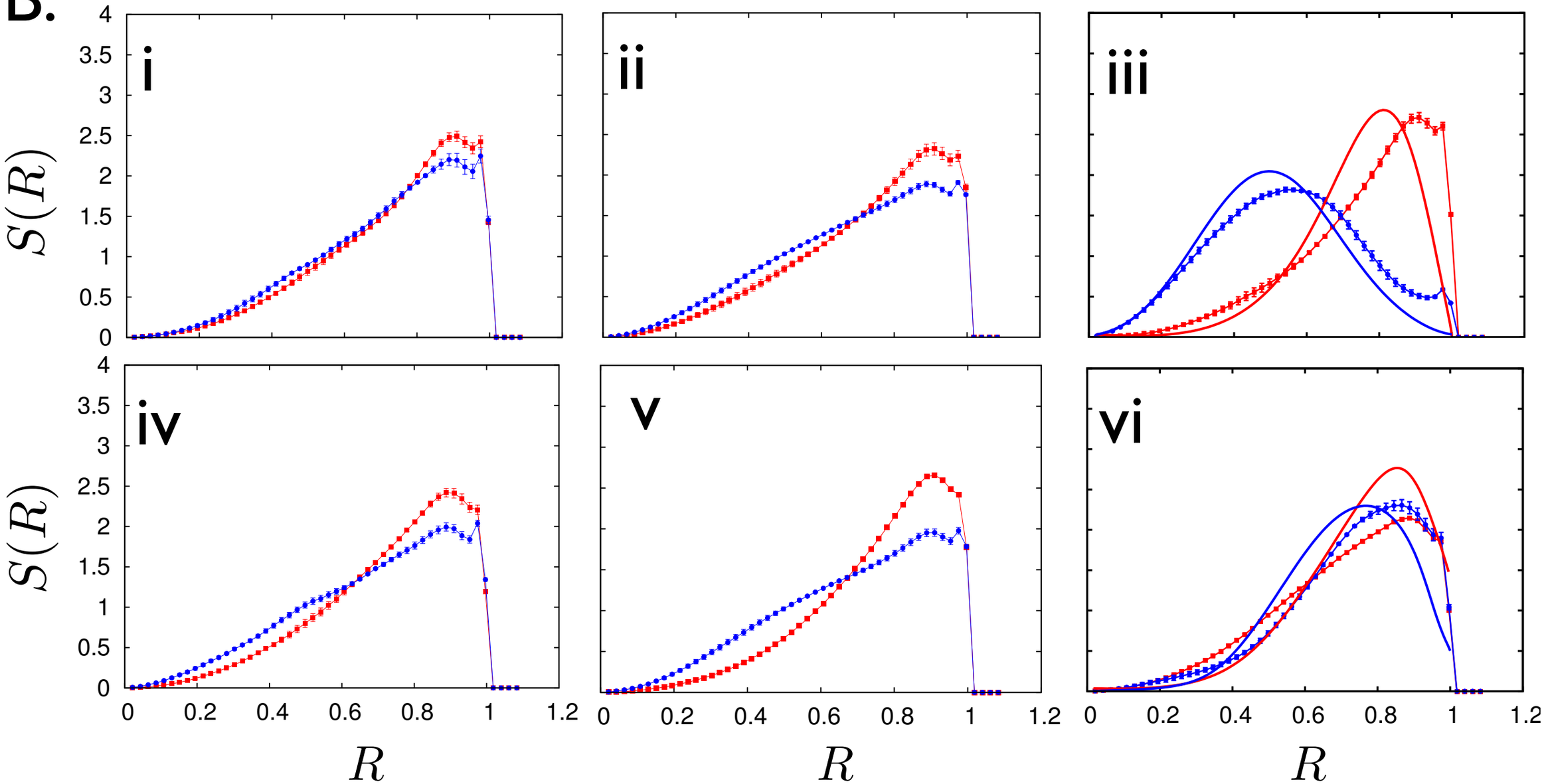


iii



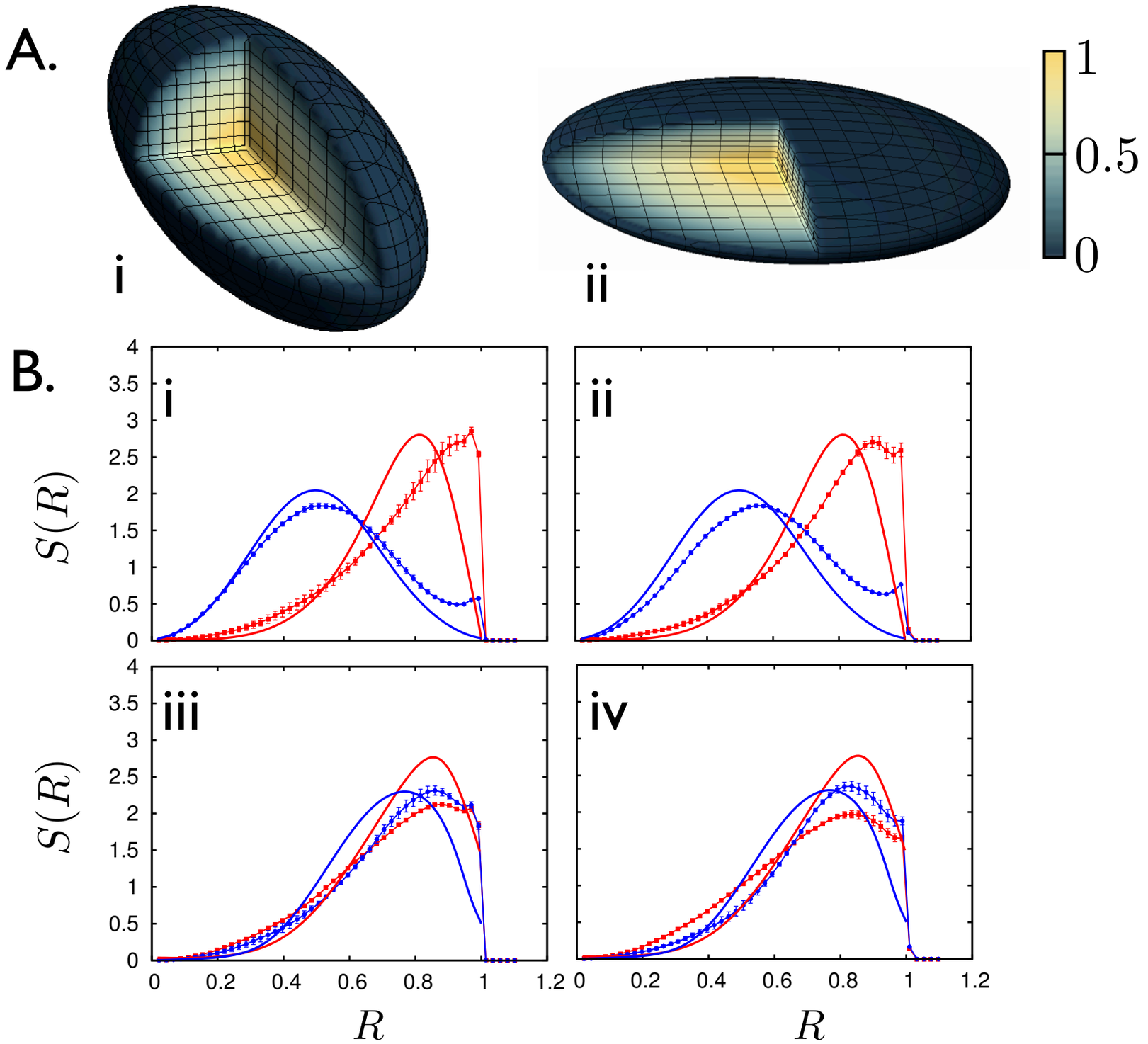


B.



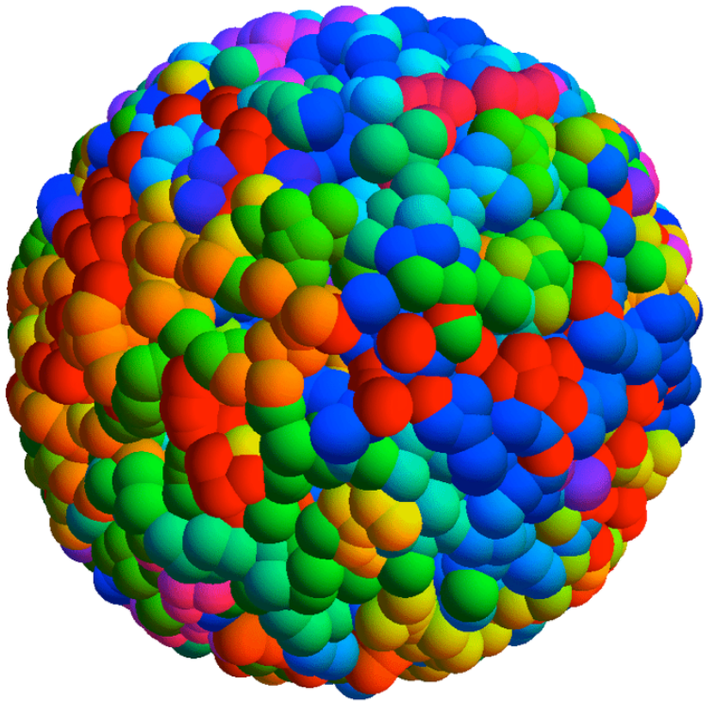
Can simulate prolate and oblate ellipsoidal nuclei

Only a marginal influence of nuclear shape



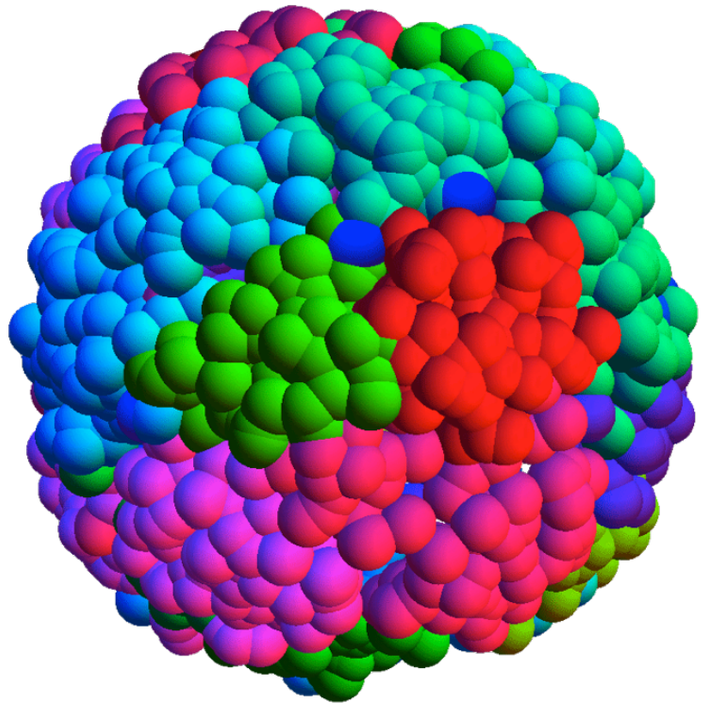
But, no chromosome territories ...

We get



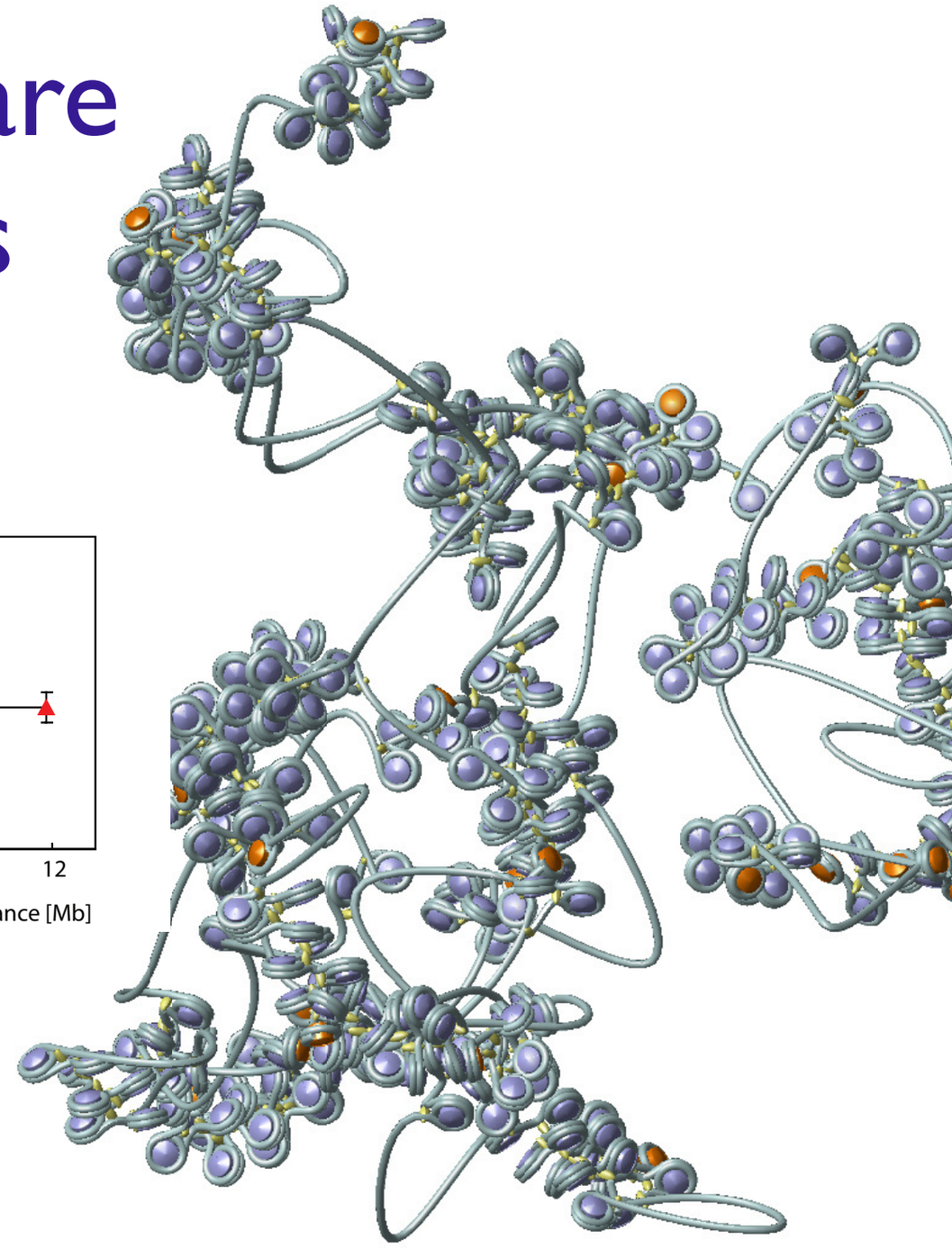
Why?

Whereas we should have got



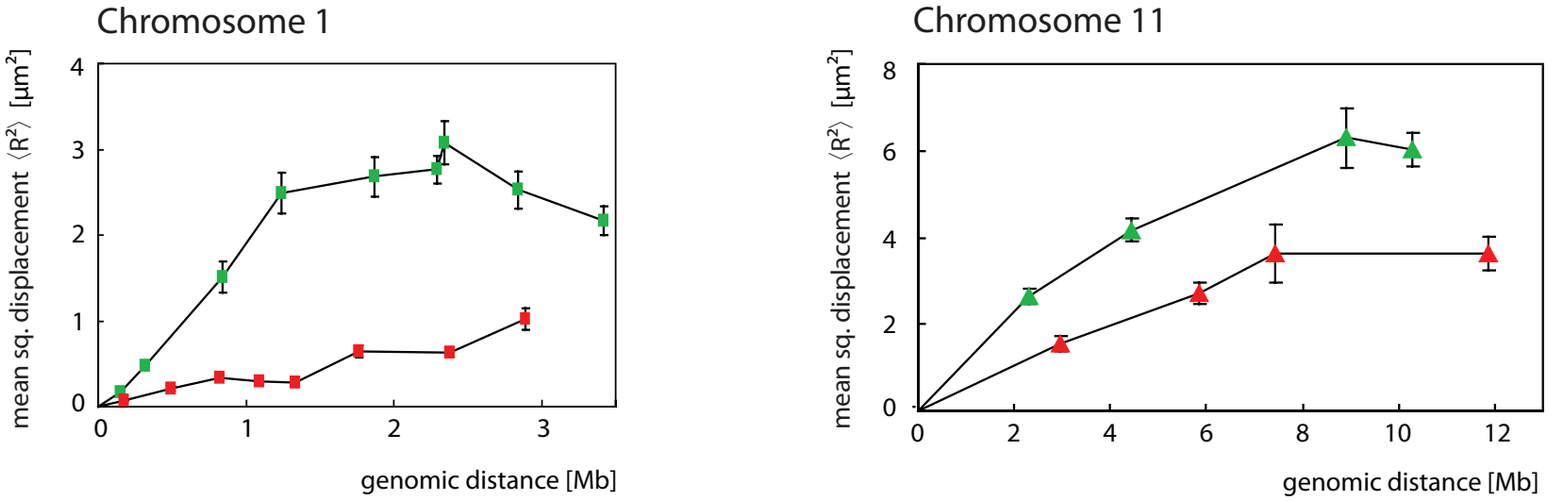
Our model for individual chromosomes assumes no structuring

Individual chromosomes are compact on large scales



B

Mateos-Langerak et al, PNAS 2009



Models for large-scale chromosome structure

Rosette model

Helical fold model

Network model

Random loop model

Fractal globule model + ...

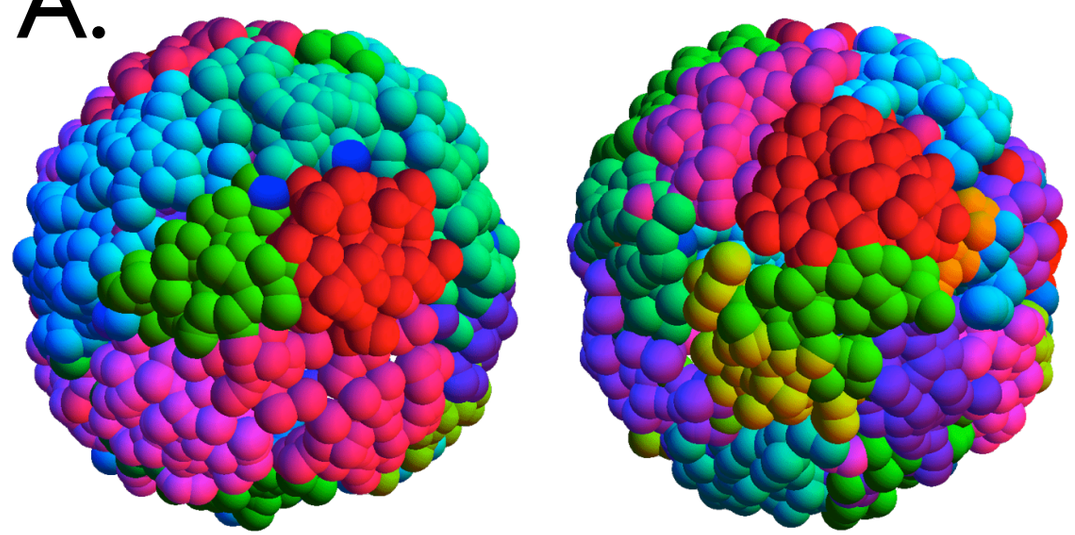
P.M. Diesinger and D.W. Heermann, Biophys. J. (2009)

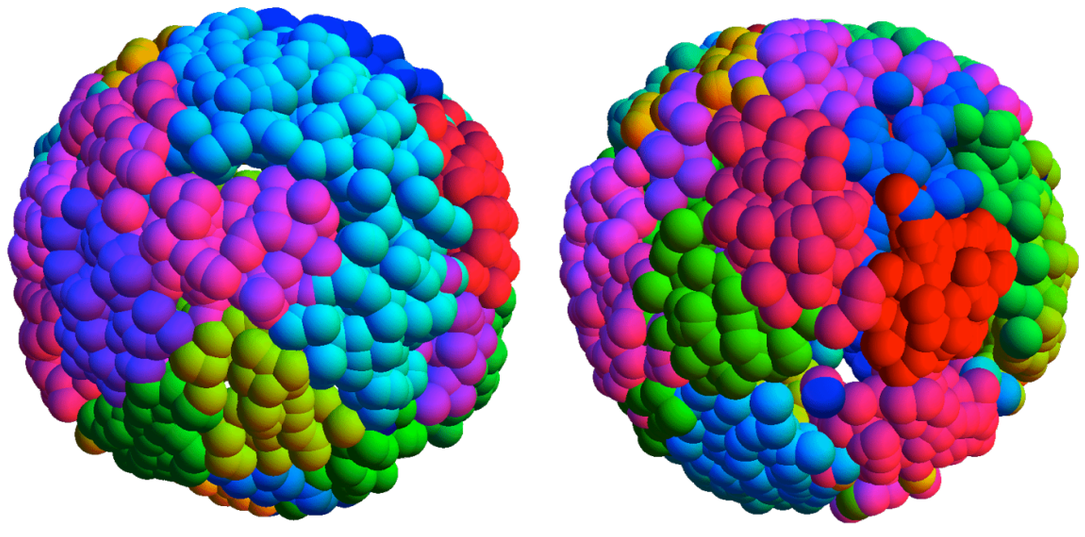
We implement the random loop model

Our model chromosomes are now compact on large scales

Can compactness and inhomogenous activity generate “chromosome territories”?

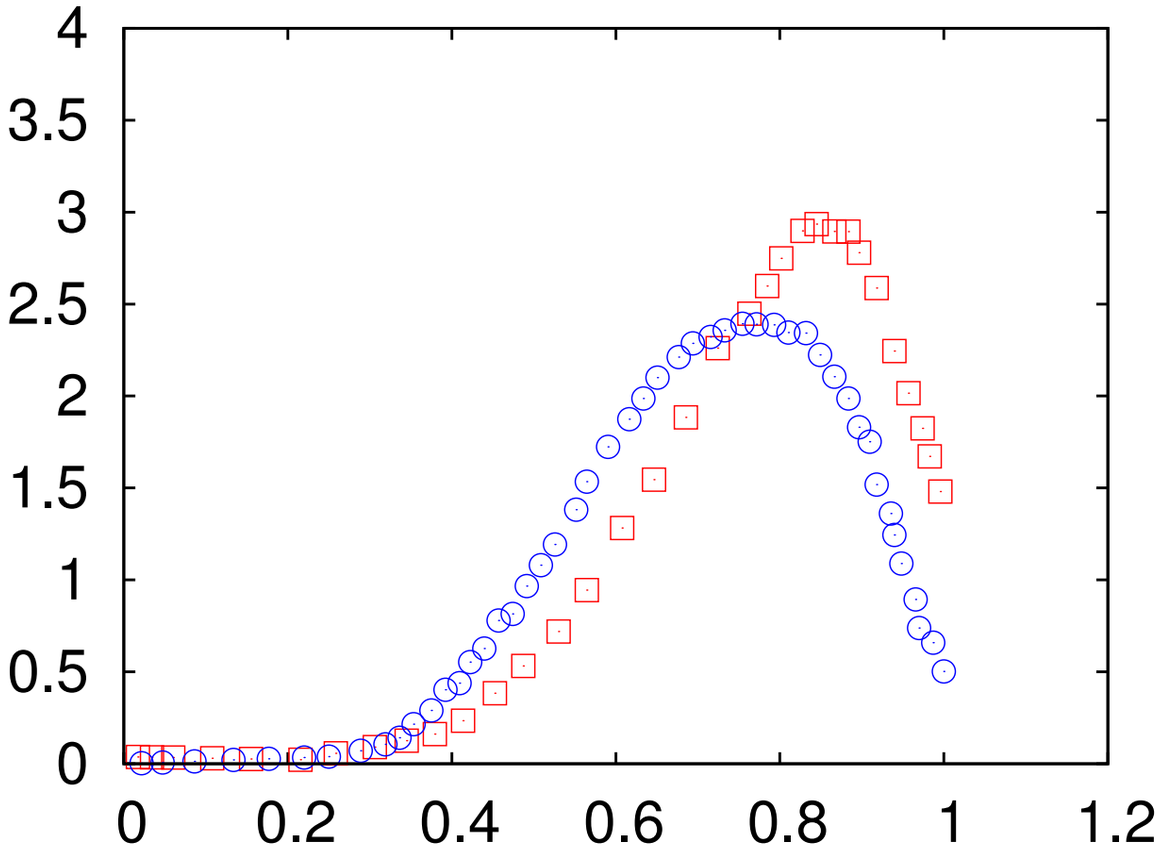
A.

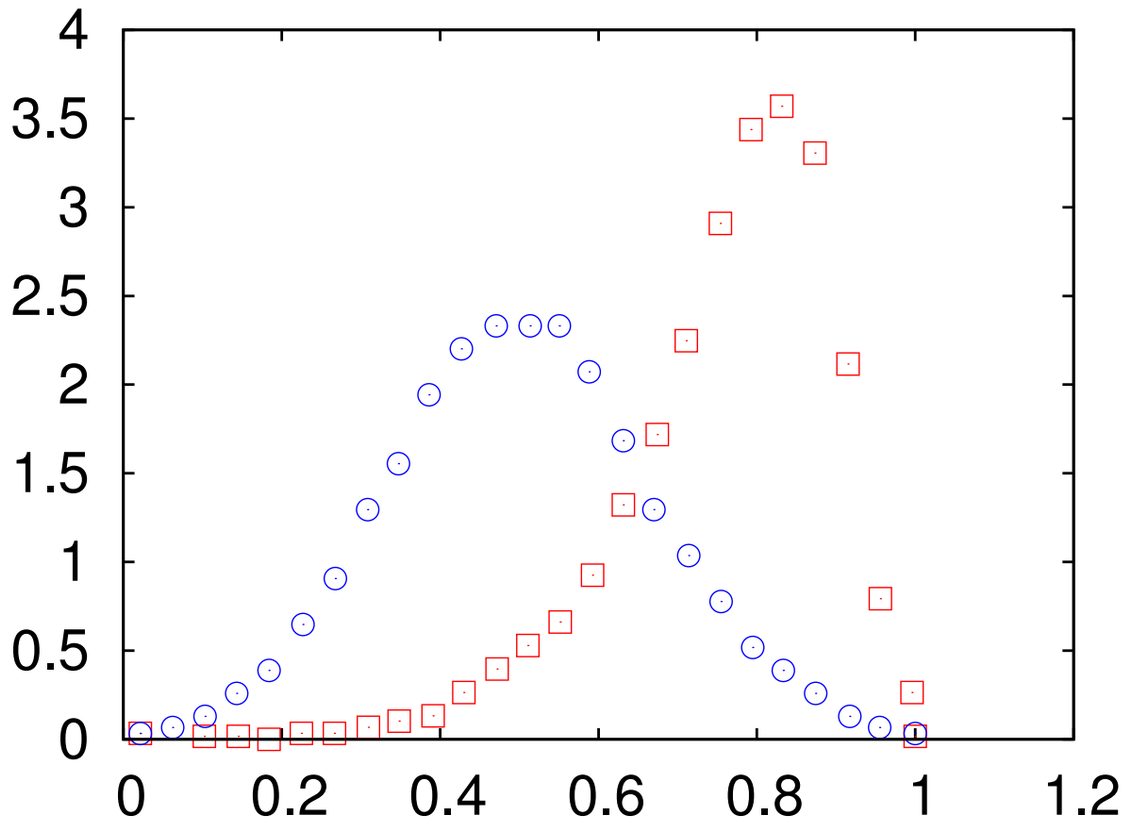




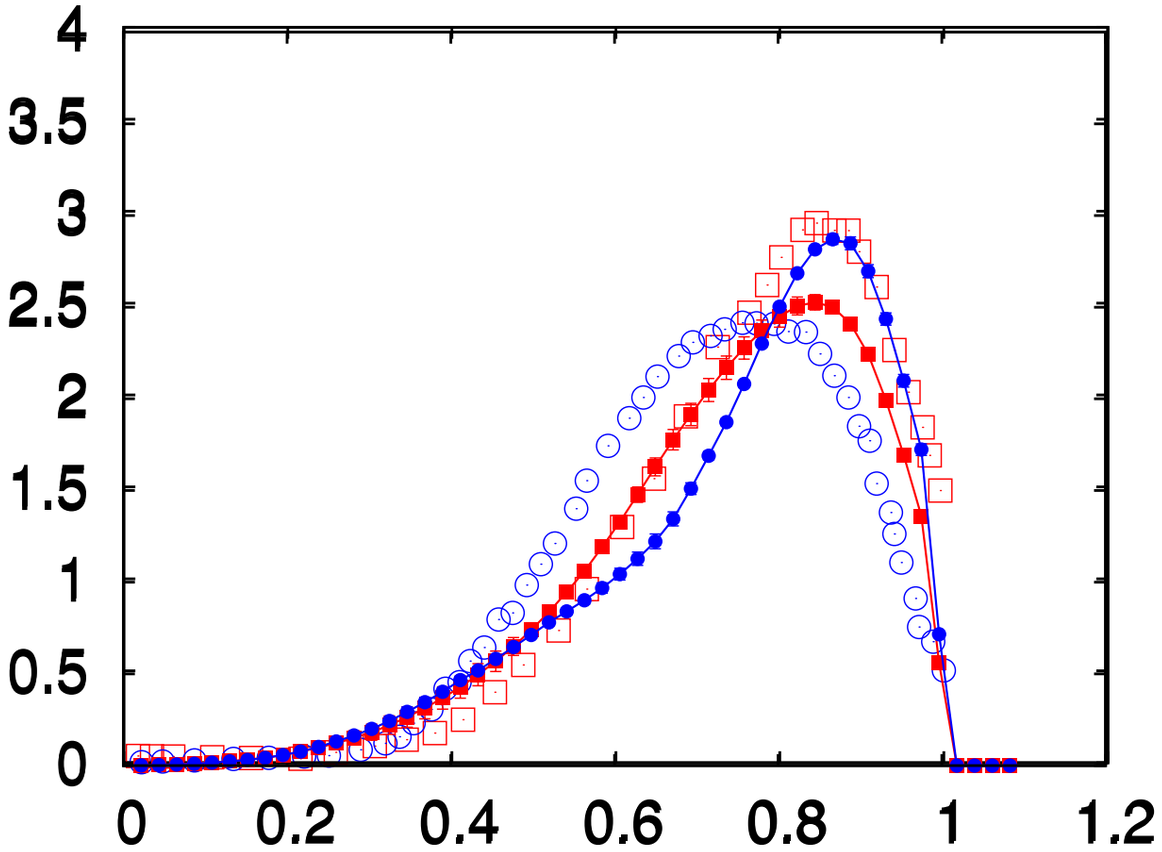
Yes

With inhomogeneous activity and compactness at large scales, how does our model compare to data?



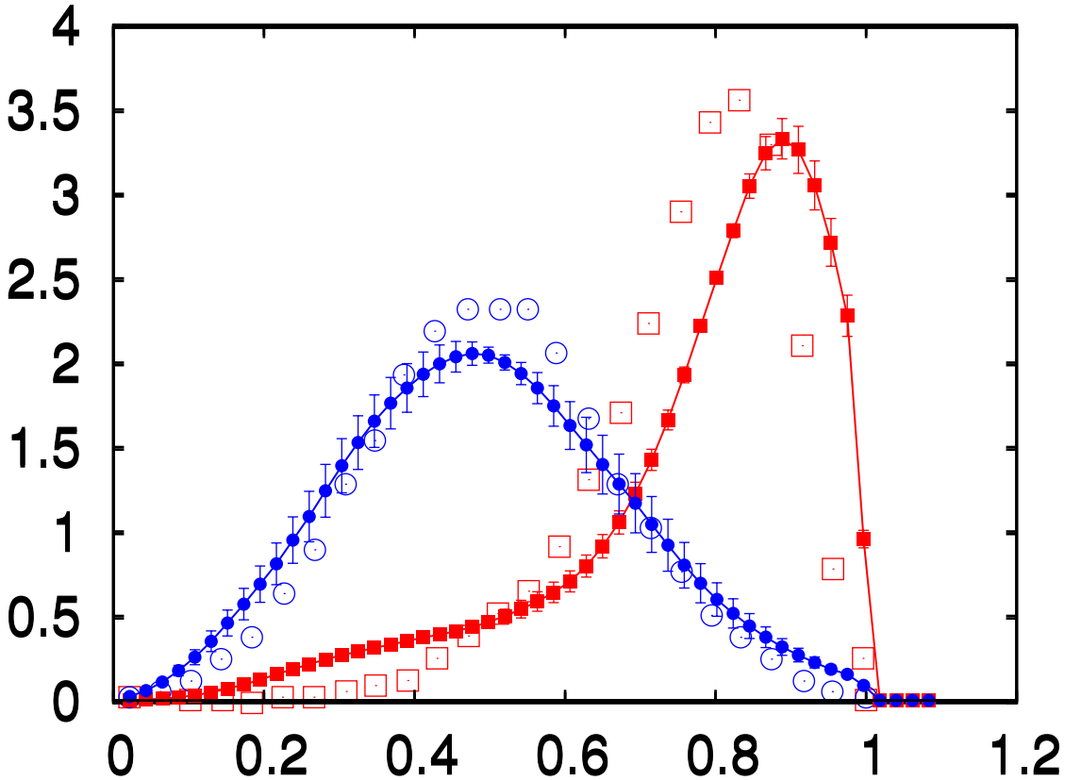


With inhomogeneous activity and compactness at large scales, how does our model compare to data?

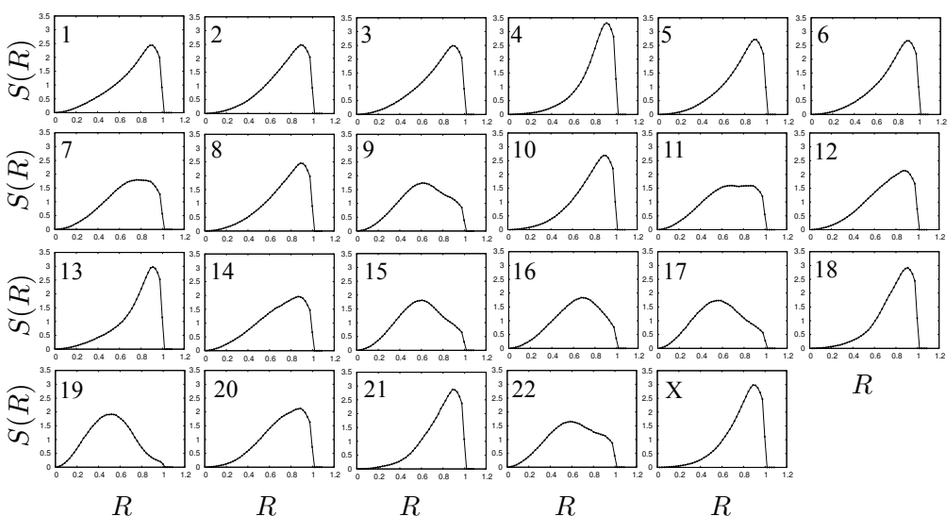


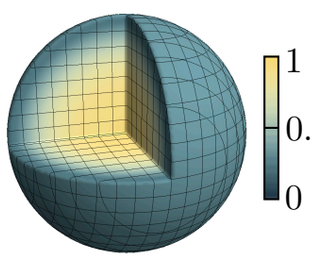
Reasonably well, despite the utter simplicity of the model

Chromosome 18/19

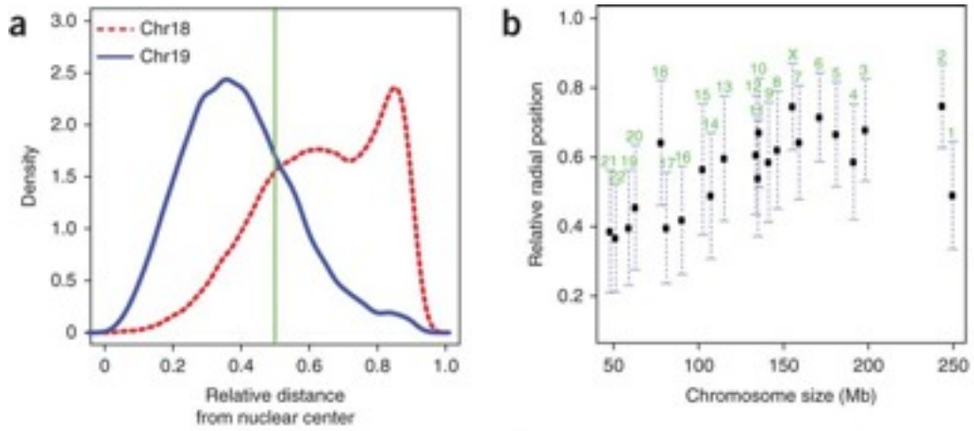


Chromosome 12/20





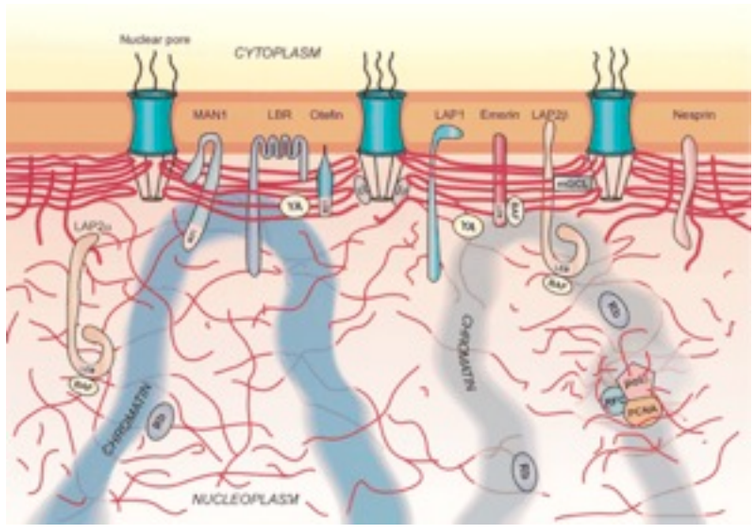
Positioning correlates to chromosome size? Nuclear shape?



Kalhor et al, Nat Biotech (2012)

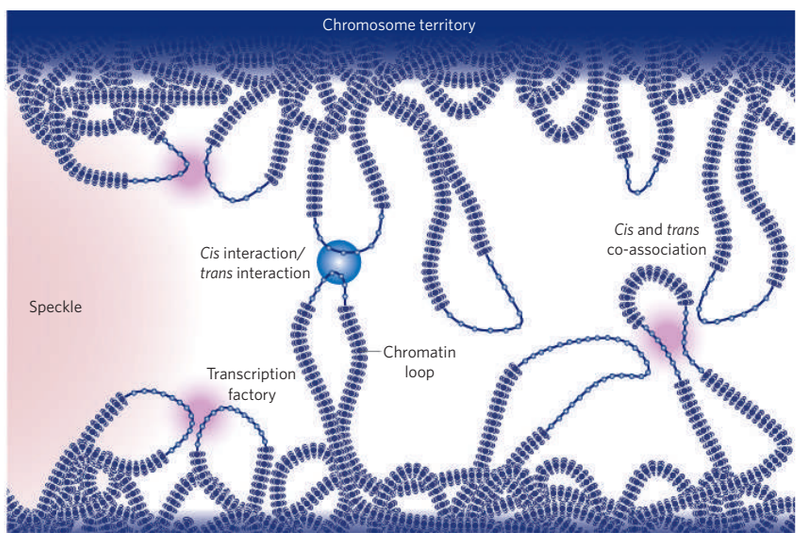
Caution: Not so fast ...

Lamins and nuclear architecture/ chromatin structuring?



Goldman et al, Genes Dev (2002)

Importance of relative positioning, transcription factories, looping ... ?

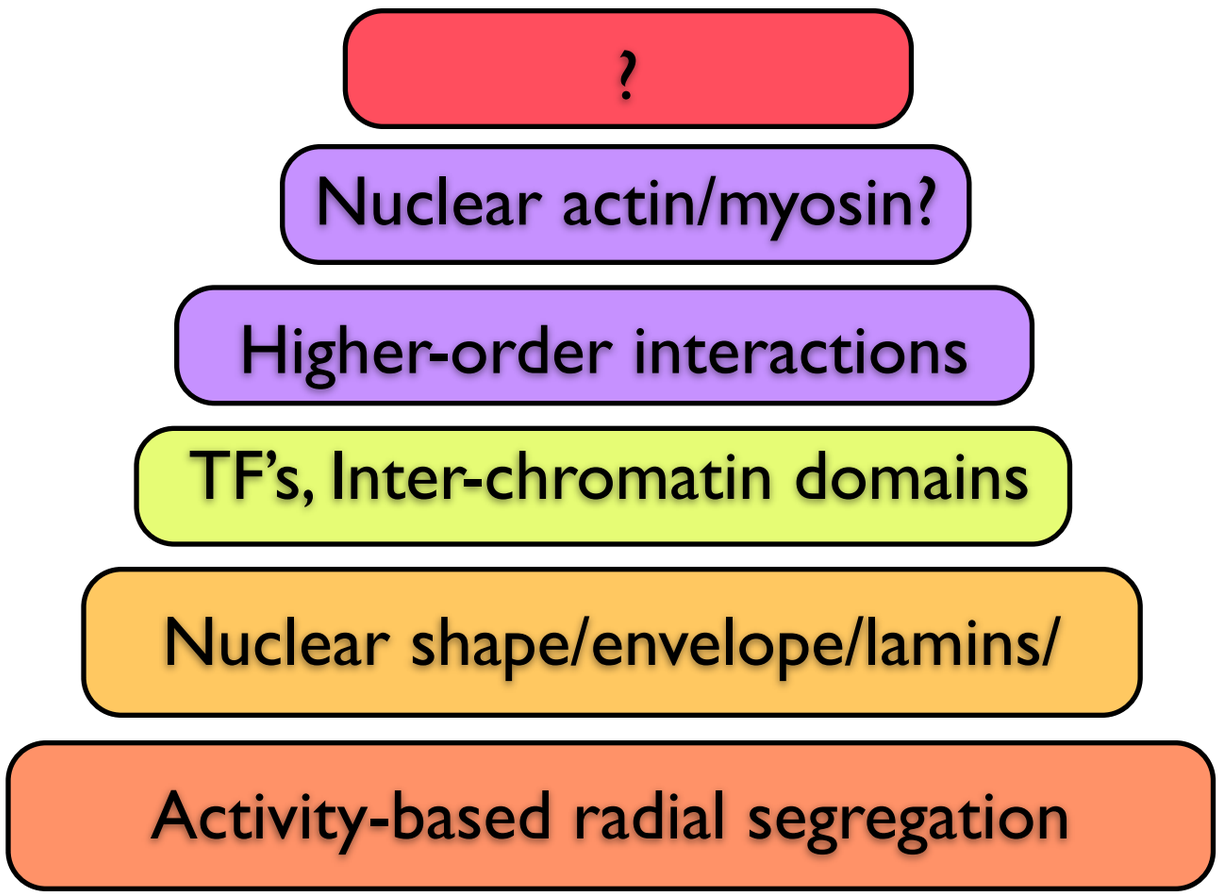


Fraser and Bickmore, Nature (2007)

Very far from these ..

A hierarchy of positioning drivers ... ?

Increasing complexity



Activity- based radial segregation might provide a generic initial template for local physical and biochemical events acting to further stabilize and optimize positioning

Conclusions

1. Chromatin as a model system for active matter

2. Suggest: Segregation by gene density and the formation of chromosome territories have a common origin in "activity-based segregation"

3. Inhomogeneous activity, confinement and polymeric nature of chromosomes are central

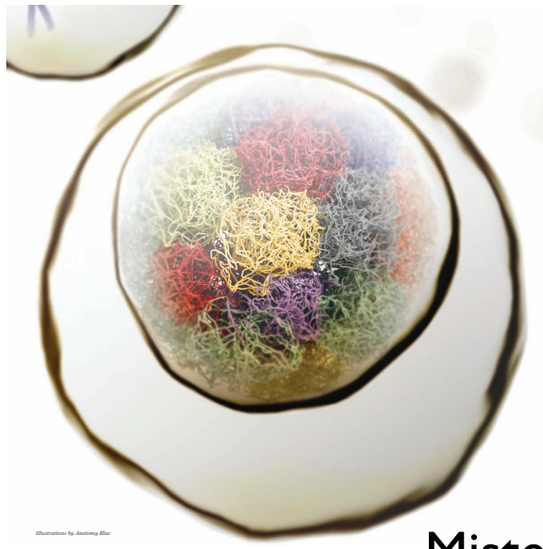
Nucleic Acids Research Advance Access published January 22, 2014

Nucleic Acids Research, 2014, 1–15
doi:10.1093/nar/gkt1417

Chromosome positioning from activity-based segregation

Nirmalendu Ganai[1], Surajit Sengupta[2,3] and Gautam I. Menon[4,5,6,*]

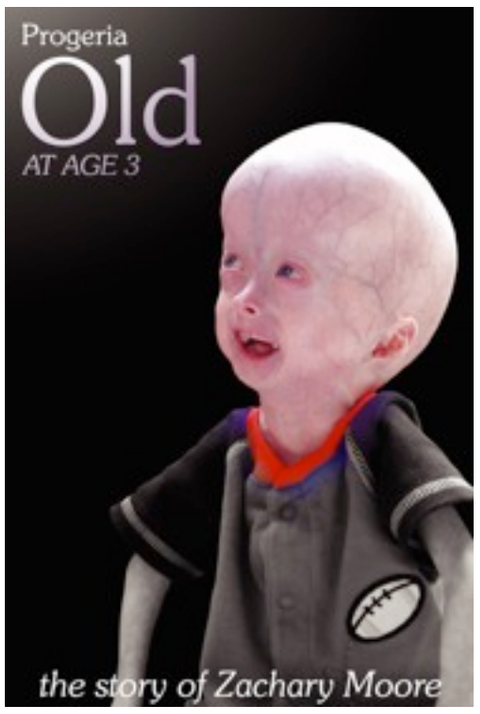
The discovery of distinct radial positions of chromosomes and genes has changed the way we think about genome organization. It has highlighted the non-randomness of higher-order genome organization and it has inspired the pursuit of how spatial genome organization contributes to function.



Misteli, Scientific American (2011)

"Meaning of gene positioning", Takizawa, Meaburn & Misteli, Cell (2008)

Further Work



http://realitypod.com/2010/07

Other directions (biological): positioning in specific cell types, effects of nuclear envelope, repositioning on DNA damage, stem cell chromatin ... ?

Other directions (statistical-mechanical): What determines activity-based segregation, confined active matter

Hutchinson–Gilford progeria syndrome