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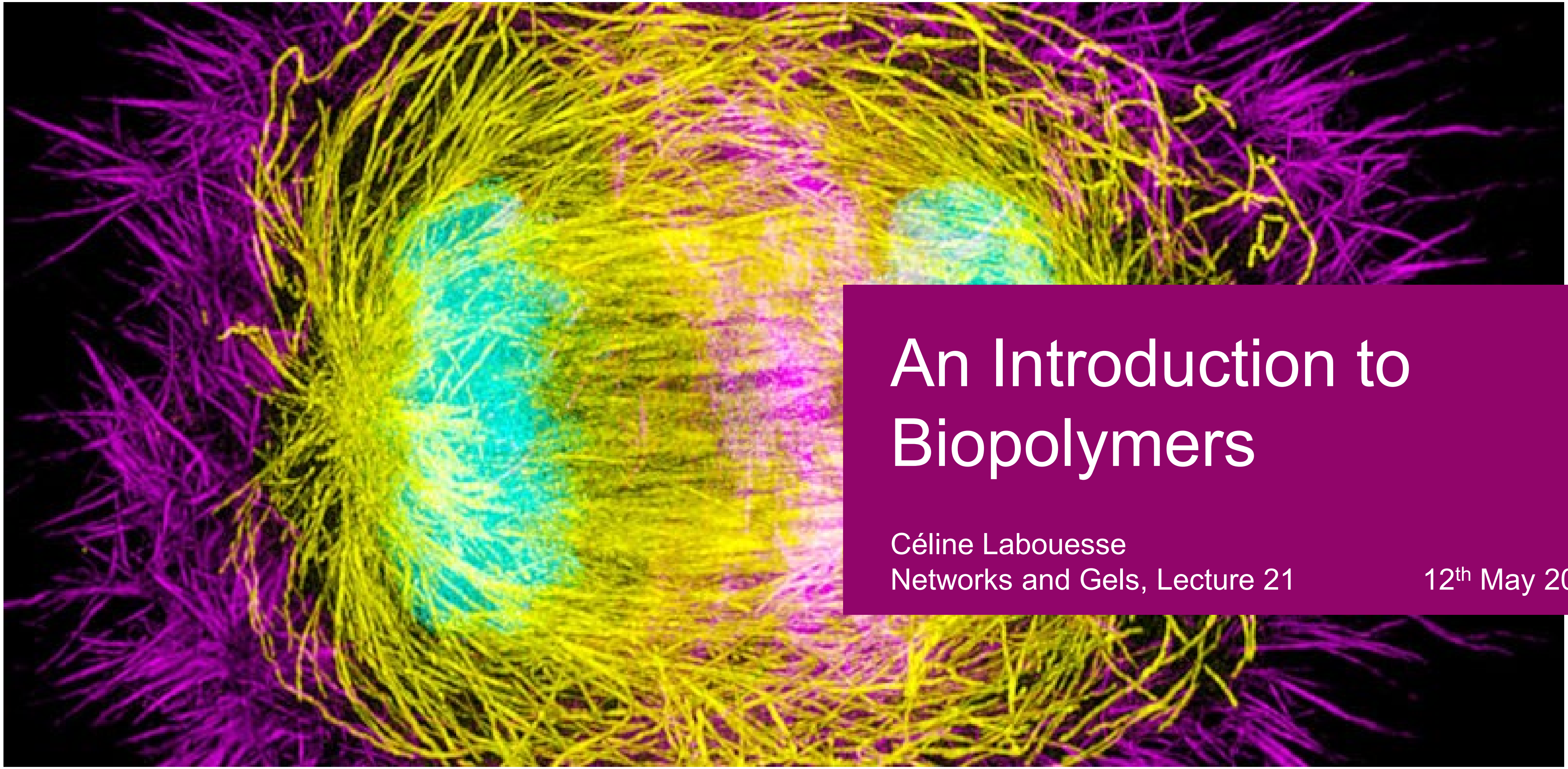

An Introduction to Biopolymers

Céline Labouesse
Networks and Gels, Lecture 21

12th May 2022

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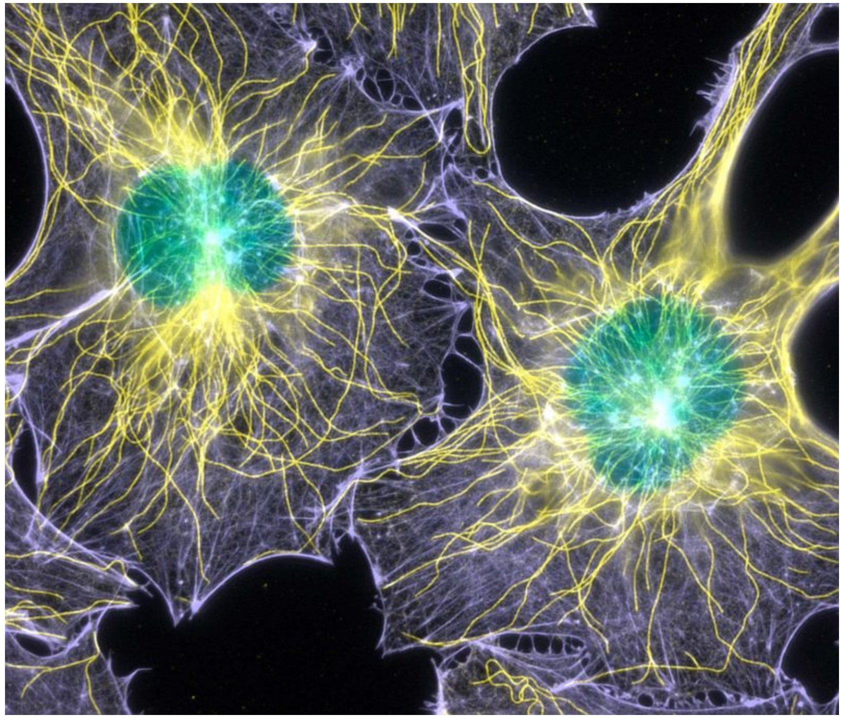
Macromolecular Engineering Laboratory

Learning Objectives

- Identify the different types of biopolymers found in living organisms
- Relate polymer physics concepts to functions in living systems



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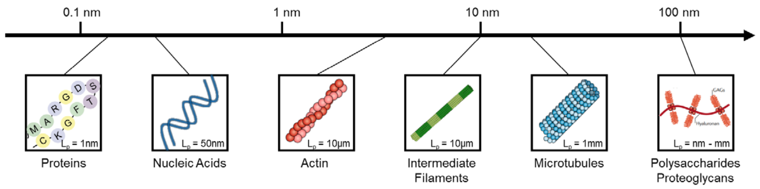
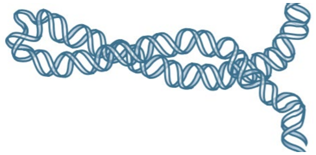
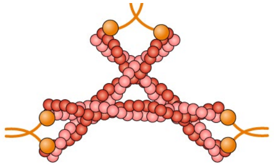
1. Properties of biopolymer filaments and networks



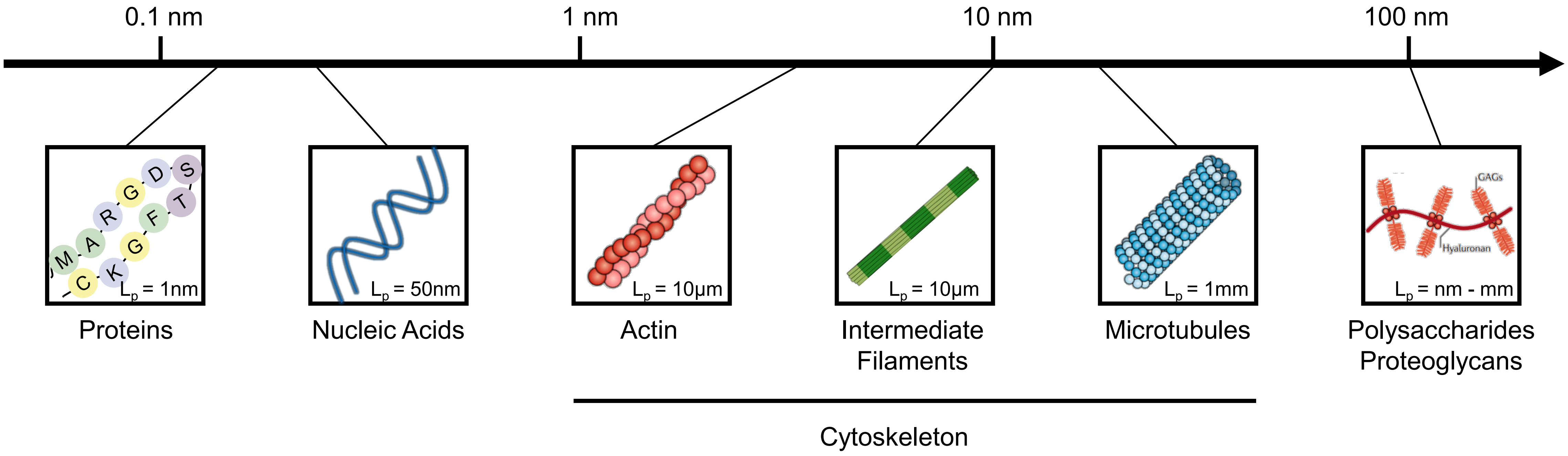
2. Does DNA behave like an ideal chain?



3. Actin filaments & actin networks: dynamic cell scaffolding

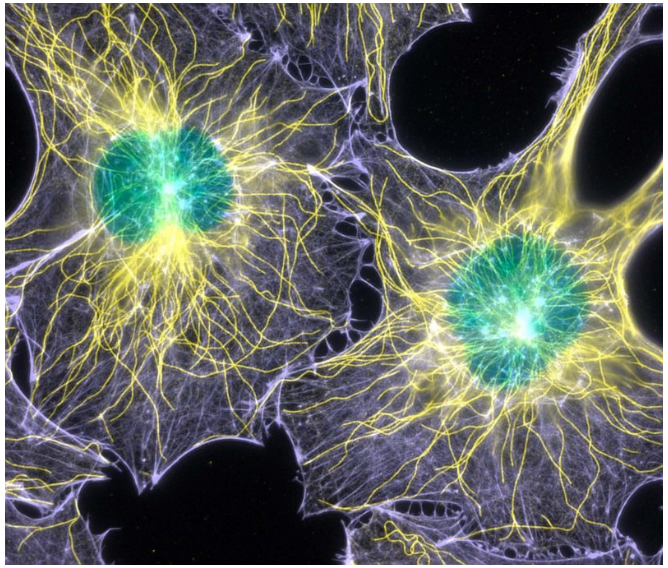


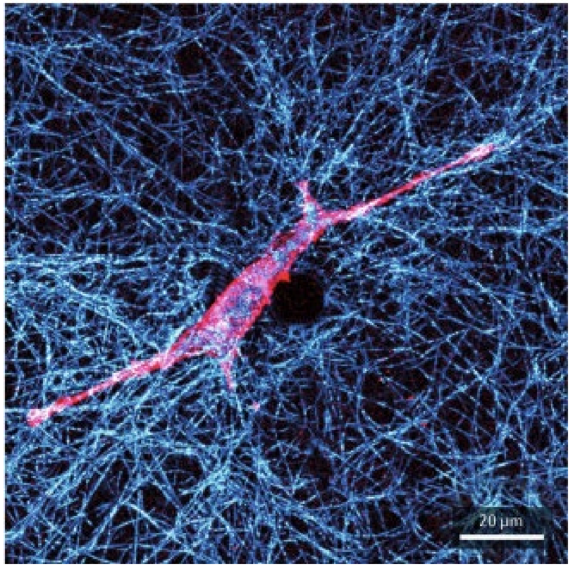
1.1 Biopolymer filaments and networks



<u>Intracellular</u>

- Actin
- Microtubules
- DNA





<u>Extracellular</u>

- collagen
- PS / PG

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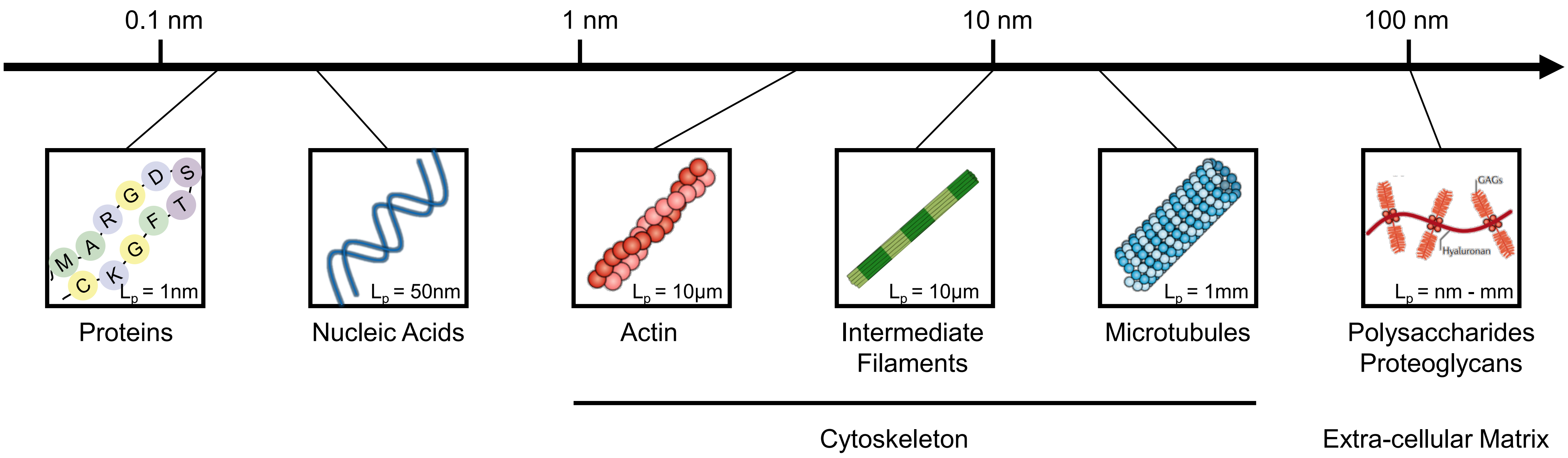
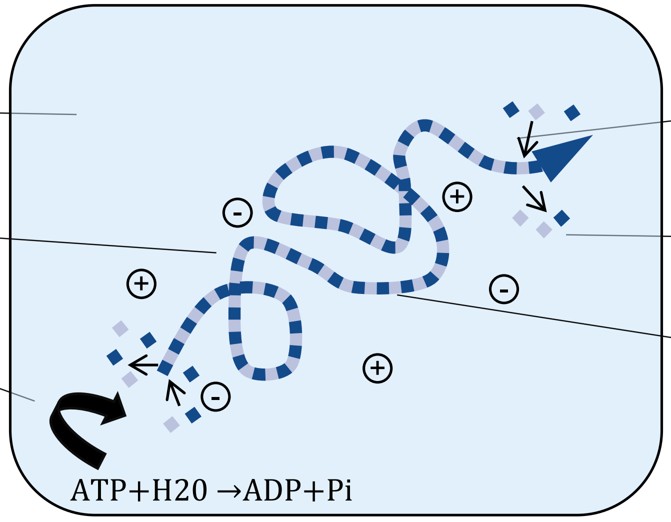
1.1 Biopolymer filaments and networks



solvent = H_2O (+ salts)

either non-covalent bonds or covalent bonds

Polymerization driven by chemical reaction (ATP hydrolysis , $\Delta G = -31$ kJ/mol)

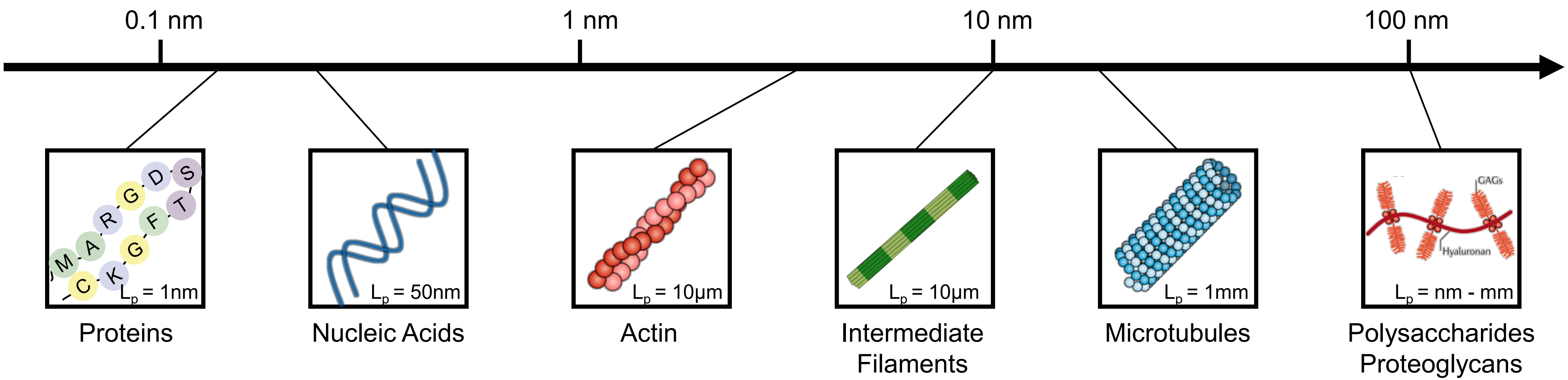


most filaments have directional polarity

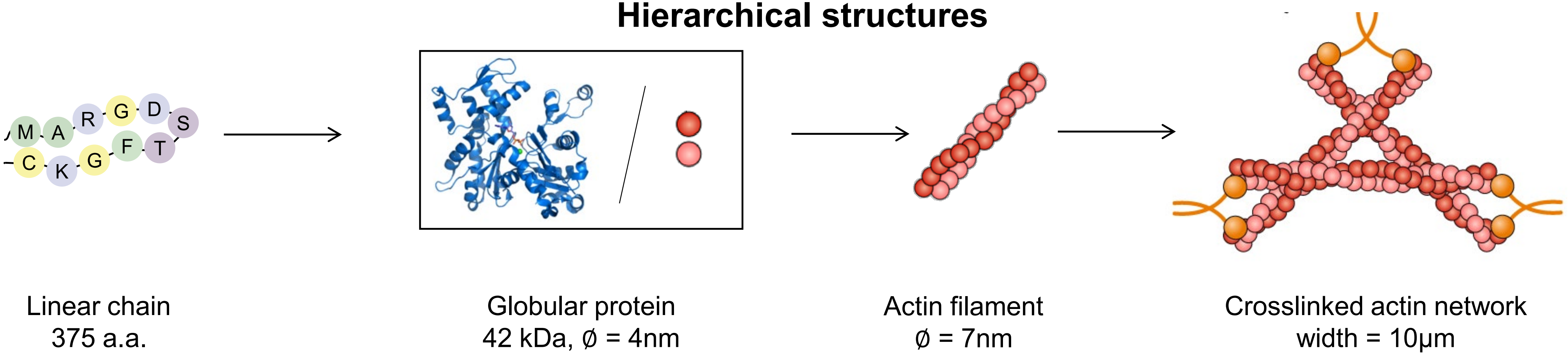
polymerization / depolymerization is dynamic

heterogeneous building blocks

1.1 Biopolymer filaments and networks



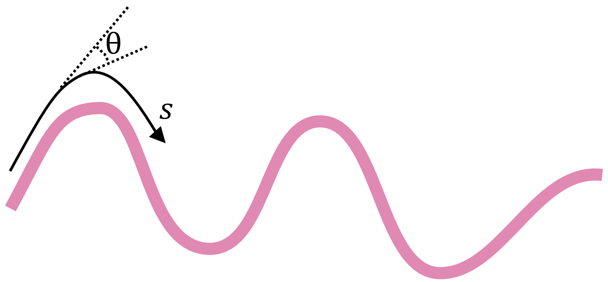
Hierarchical structures



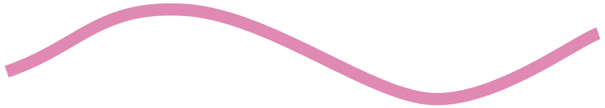
1.2 Elasticity of biopolymer filaments

Filament stiffness quantified by the *persistence length*

- related to filament bending rigidity κ $\qquad L_p = \kappa / k_B T$
- decay length of the angular correlation along contour $\qquad < \cos\theta > = e^{-s/L_p}$

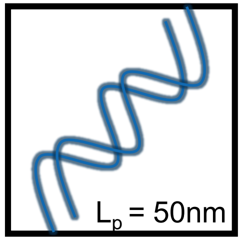


flexible (freely jointed chain)
$L_p \ll L$

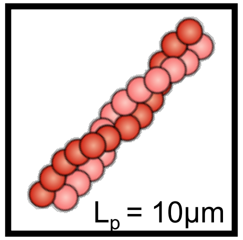


semiflexible
$L_p \cong L$

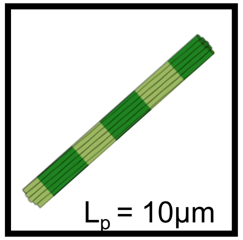
stiff (rod-like)
$L_p \gg L$



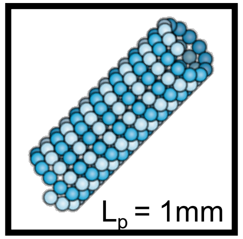
Nucleic Acids



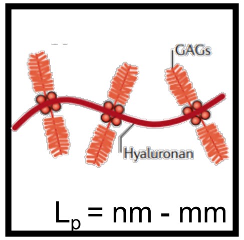
Actin



Intermediate Filaments



Microtubules

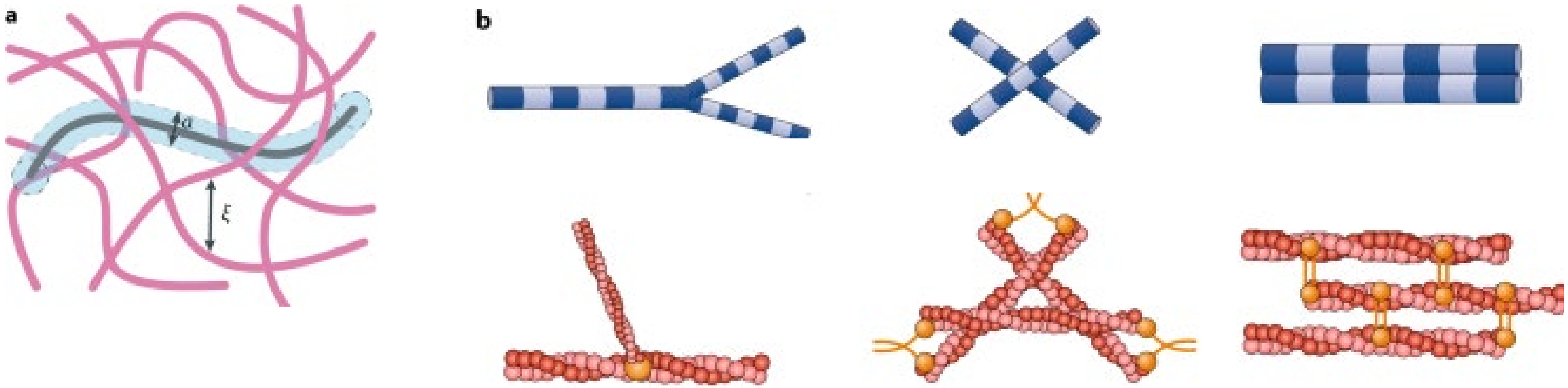


Polysaccharides Proteoglycans

1.2 Elasticity of biopolymer networks

Network properties depend on the nature of the interactions between filaments

intermolecular interactions

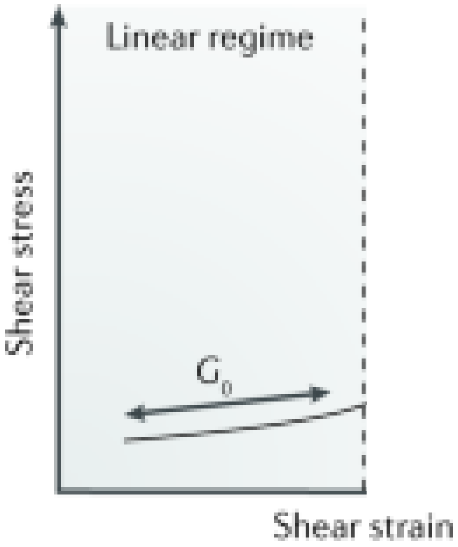


entanglement

crosslinking protein

1.2 Elasticity of biopolymer networks

At small strains & in linear regime $\tau = G\gamma$ (Hooke's Law)



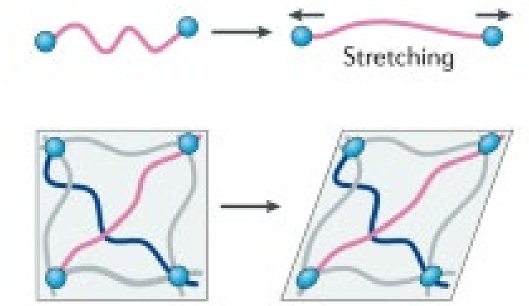
1.2 Elasticity of biopolymer networks

At small strains & in linear regime $\tau = G\gamma$ (Hooke's Law)

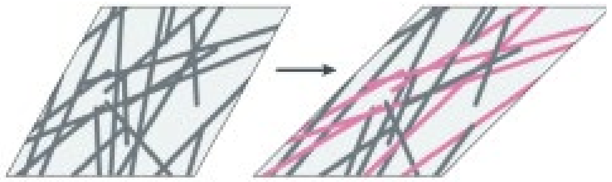
Strain-stiffening at high strain

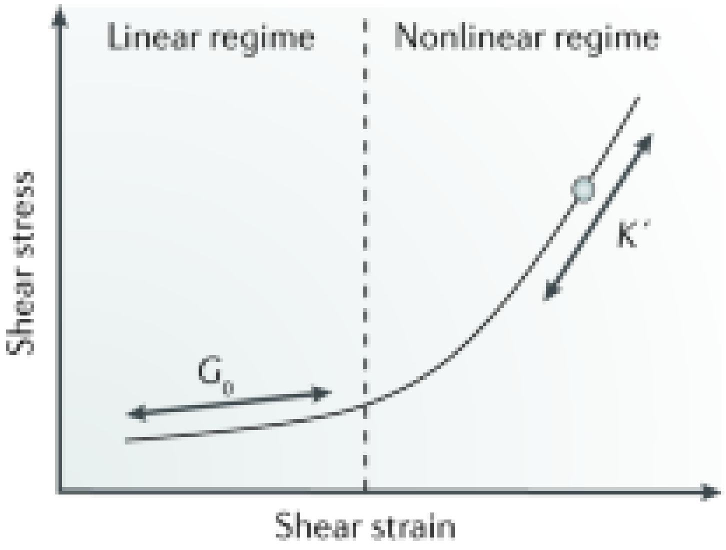
- entropic elasticity
- reorientation of load-bearing elements

entropic elasticity



reorientation of elements along direction of strain



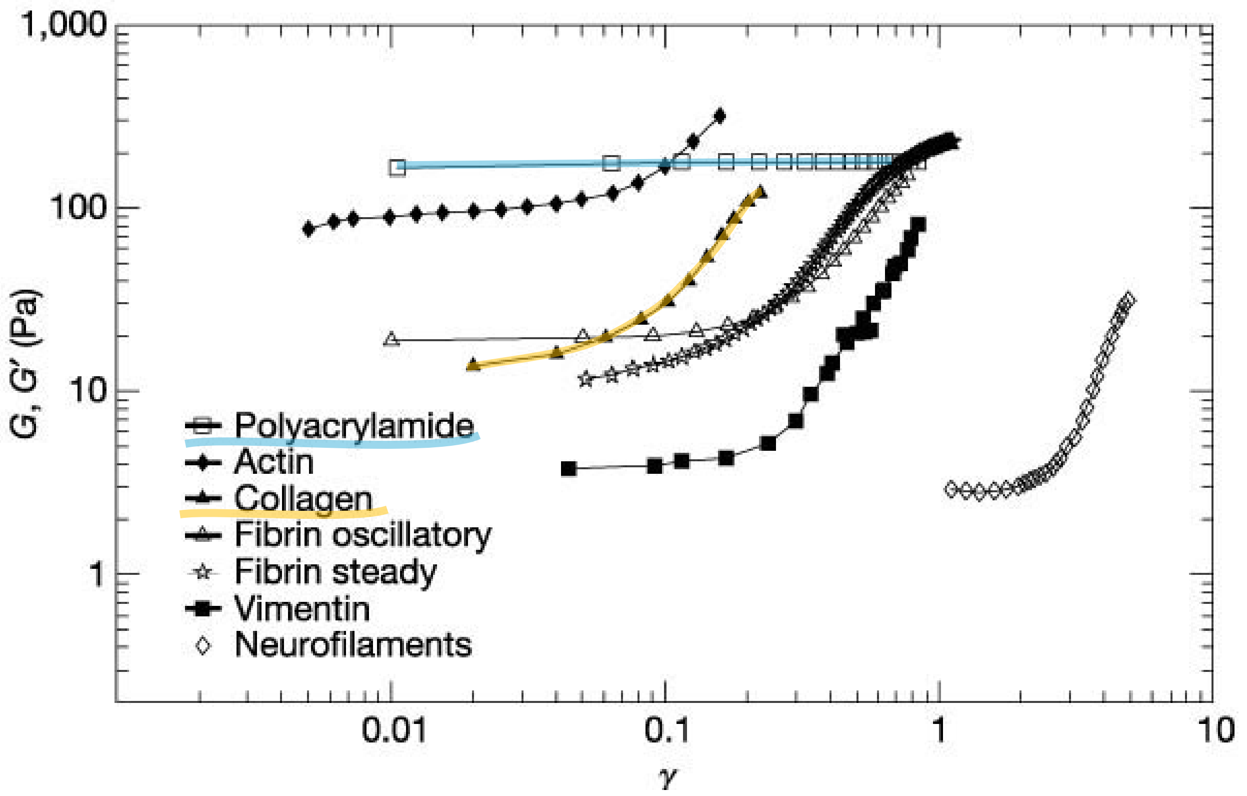


1.2 Elasticity of biopolymer networks

Strain-stiffening under tensile & shear loading

→ compliance at very low strain allows dynamic conformational changes & movement

→ stiffening at larger strain allows mechanical protection of cells & tissues to prevent large deformation and tissue rupture

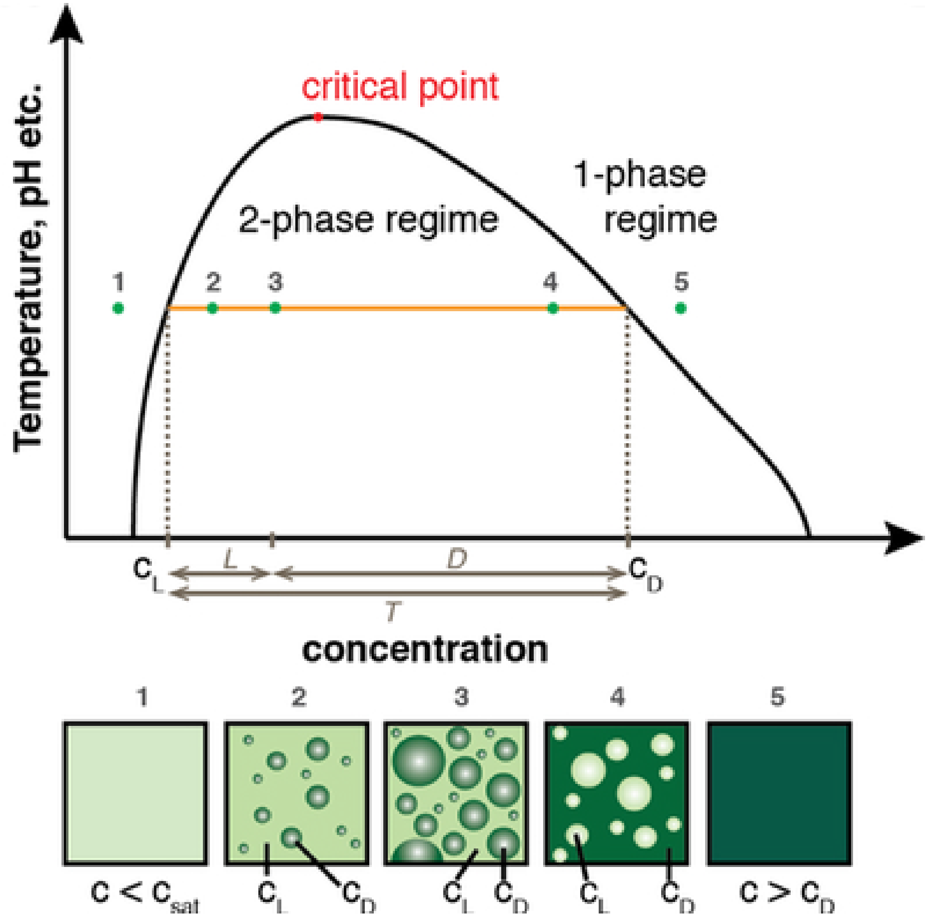


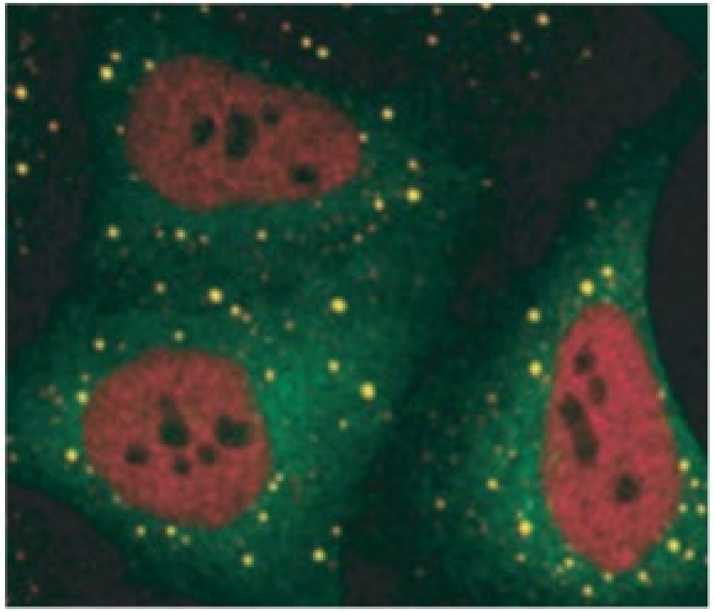
Biopolymer networks often display strain-stiffening at high strains

1.3 Liquid-liquid phase separation

Proteins & nucleic acids can exist in **dilute** phase or **dense** phase – with coexistence of the two phases

analogous to Flory-Huggins theory

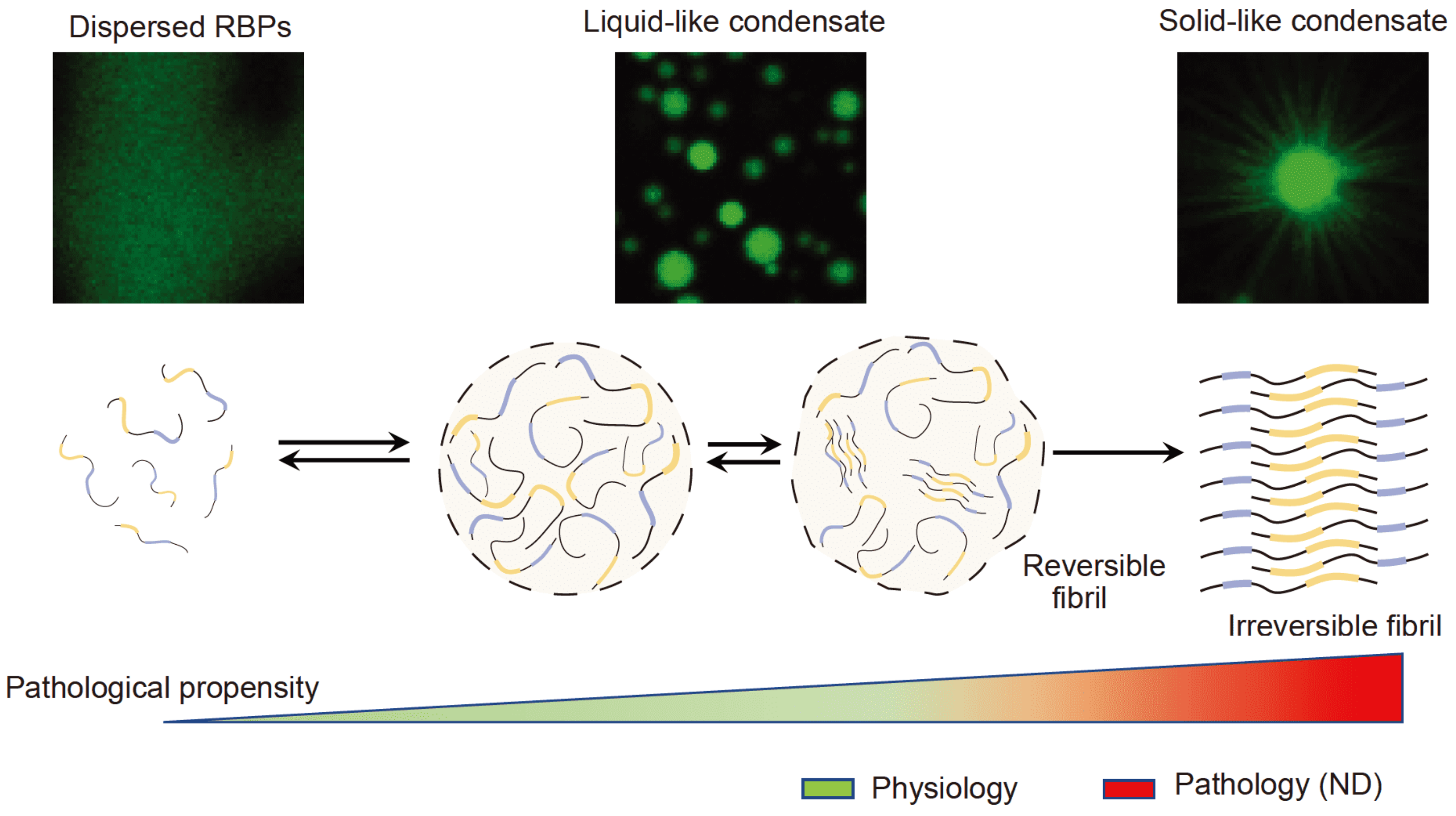




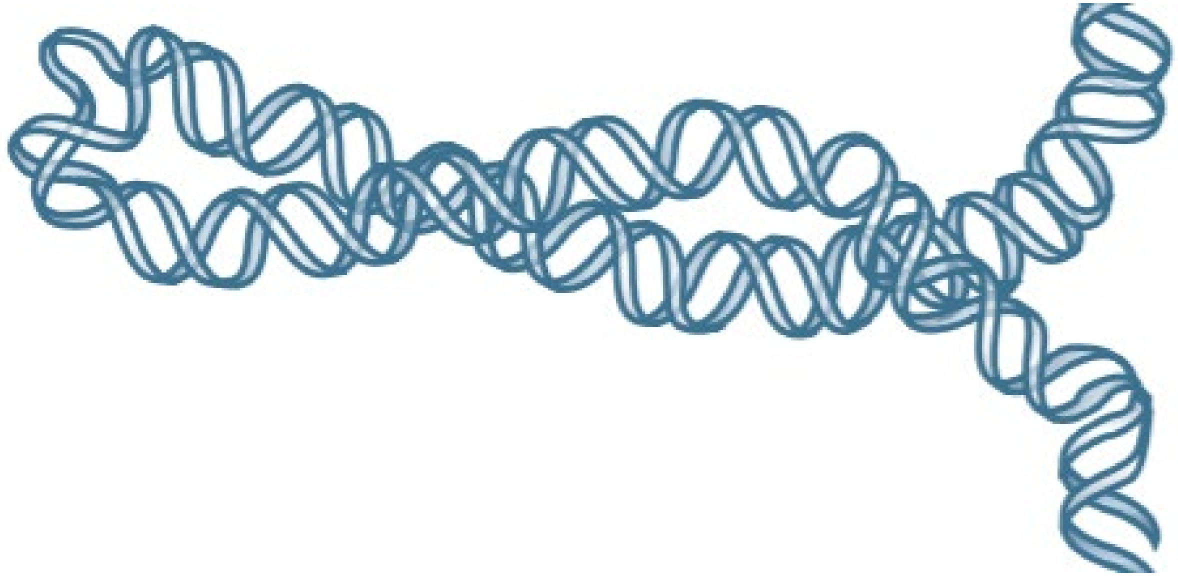
1.3 Liquid-liquid phase separation

Proteins & nucleic acids can exist in **dilute** phase or **dense** phase – with coexistence of the two phases

→ **Phase separation** occurs in normal biological function (reversible condensates) and in pathological contexts (irreversible condensates)



2. DNA elasticity

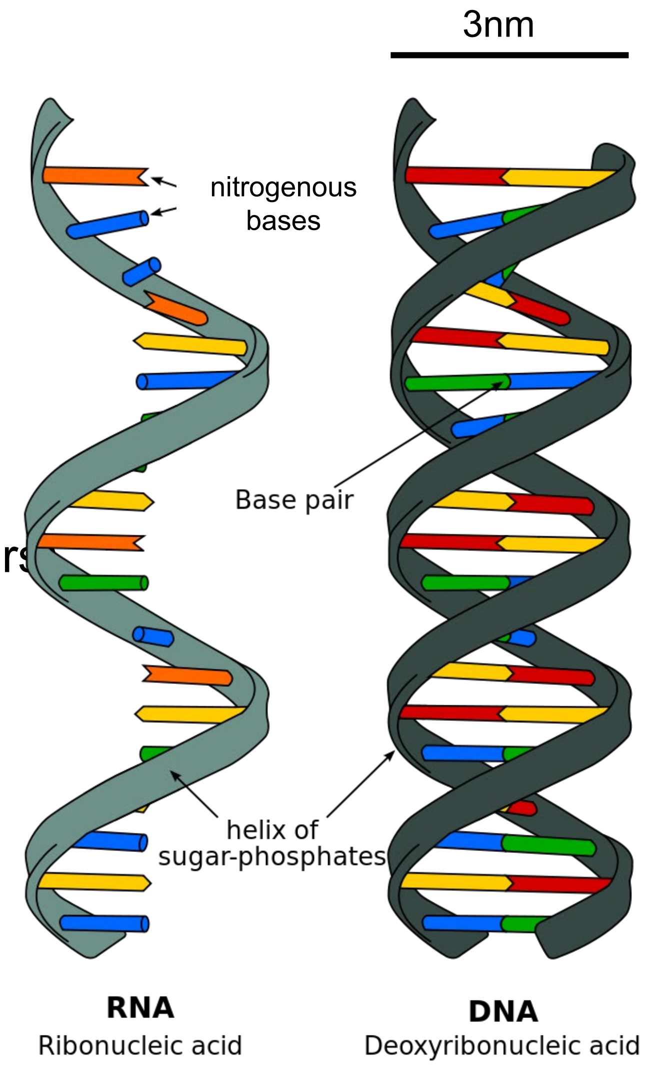


2.1 Nucleic Acids

- sugar-phosphate backbone + nitrogenous bases
  - deoxyribonucleic acid (DNA) or ribonucleic acid (RNA)
  - phosphate group, charged PO_4^-
  - 4 nitrogenous bases form the sequence

- DNA
  - as **double stranded helix** (hydrogen bonds between base pairs
  - as **single strand**
  - structure independent of sequence

- RNA
  - as single strand
  - forming “hairpin” loops (sequence-dependent)



2.2 Does DNA behave like an ideal chain?

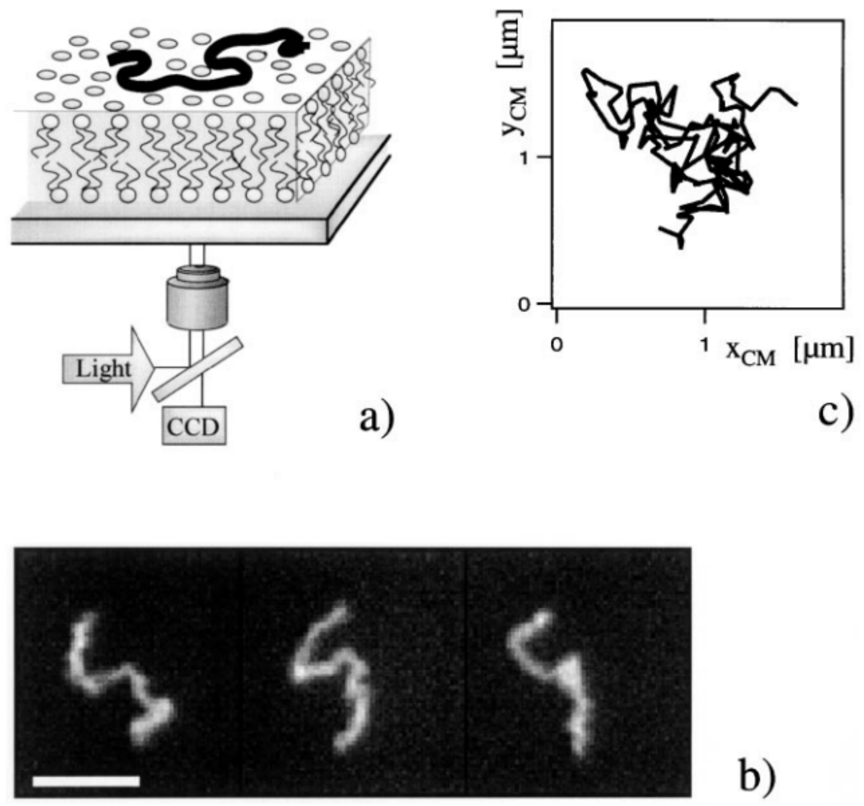
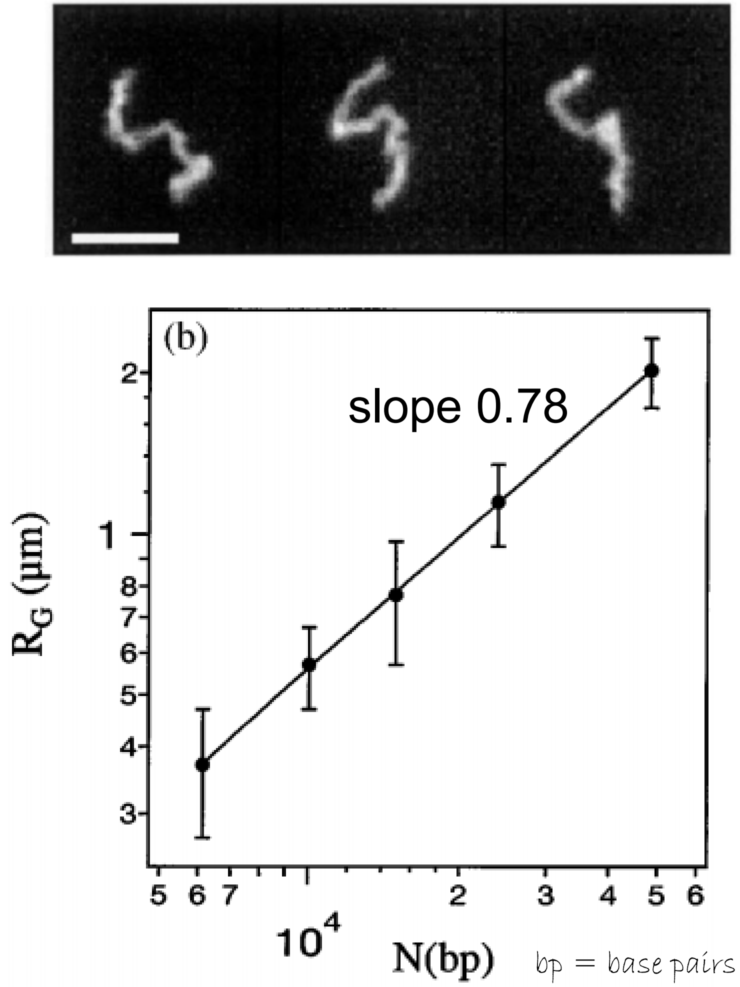


FIG. 1. Single λ-phage DNA molecule adsorbed onto a cationic lipid bilayer supported on a glass substrate. (a) Schematic (not to scale) sketch of the experimental setup. (b) Time series of fluorescence images at 2 sec intervals (bar represents 10 µm). (c) The center-of-mass motion of a 10.090 bp λ-DNA fragment following diffusive behavior.

- Measuring 2D fluctuations
  - confining chain on a surface
  - image chain conformations, measure $< R_G >$

2.2 Does DNA behave like an ideal chain?



bp = base pairs

- Measuring 2D fluctuations
  - confining chain on a surface
  - image chain conformations, measure $< R_G >$

- scaling of $< R_G >$ with chain length: $< R_G > \propto N^{3/4}$

NB: Flory exponent for a real chain in a good solvent

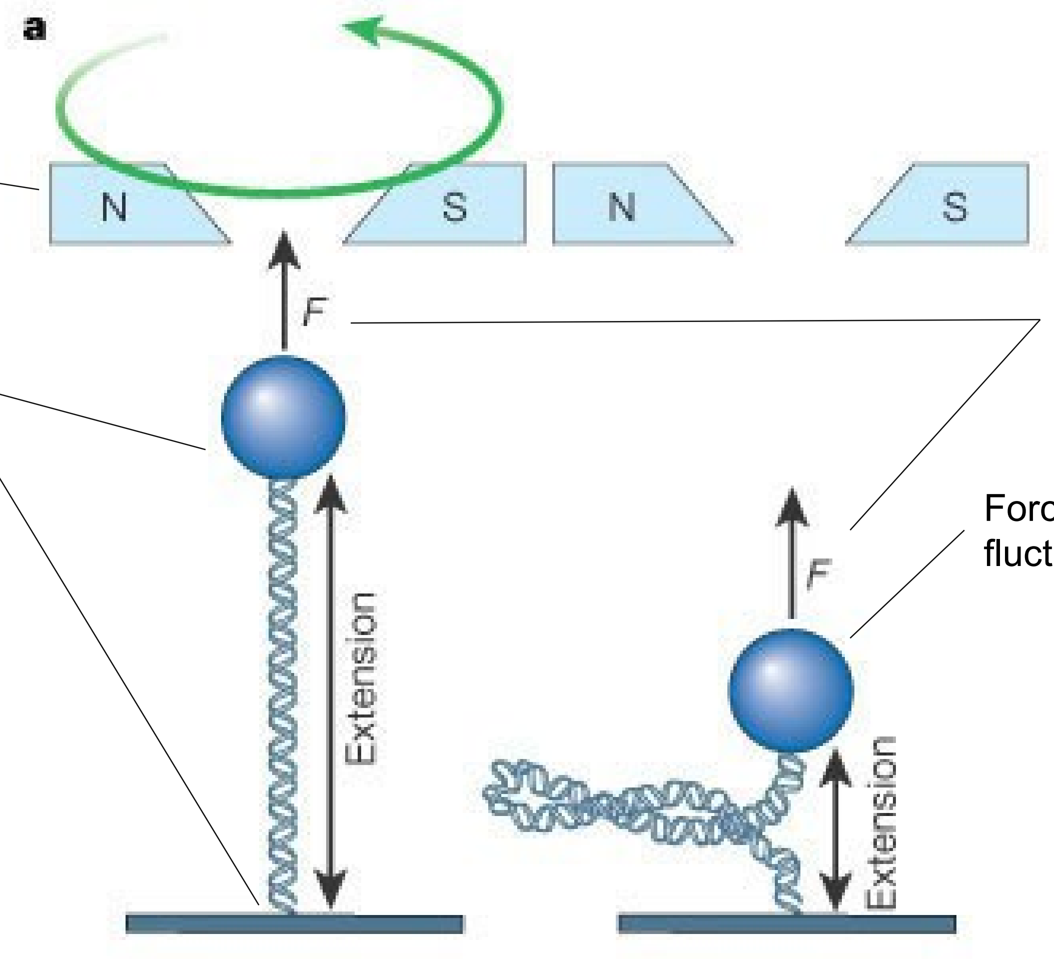
in 2D $\nu = \frac{3}{4}$ in 3D $\nu = \frac{3}{5}$

At zero-force, DNA conformation modeled by a self-avoiding chain

2.3 Stretching a single DNA molecule using magnetic tweezers

small magnets are used to apply stretch / torque on DNA

DNA is bound to glass slide on one end, and to a microbead (∅ 1-4.5µm)



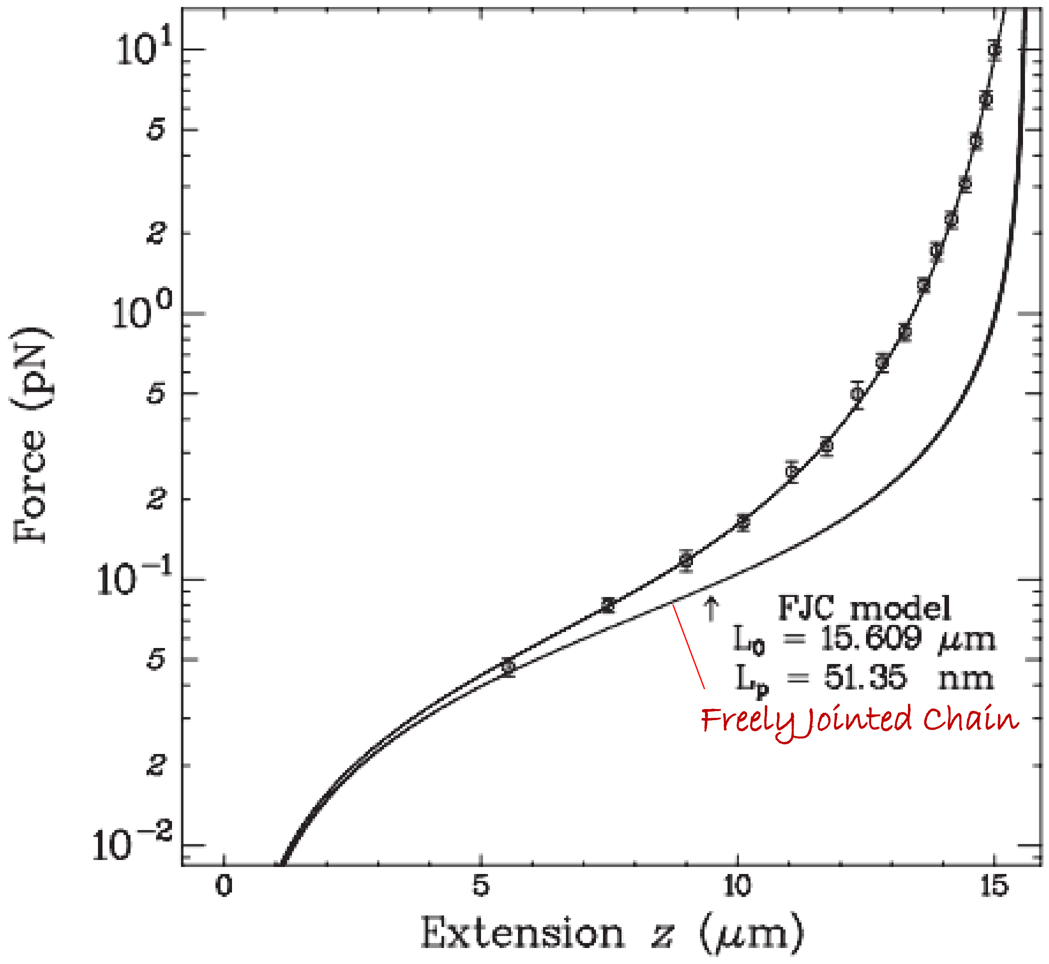
Forces: 10 fN (10^{-11} N) to 100 pN (10^{-7}N)

Force measurement based on the Brownian fluctuations of the bead $\delta x^2 \propto 1/F$

Magnetic tweezers enable to measure force-extension relationship of nucleic acids

2.3 Elasticity of DNA under force (entropic regime)

Force-extension measured using magnetic tweezers



- At forces, freely-jointed chain model is sufficient:

$$\frac{L}{L_0} \cong \frac{1}{3}\frac{Fb}{k_B T}$$

L/L_0: relative chain extension
b: Kuhn length
F: force applied

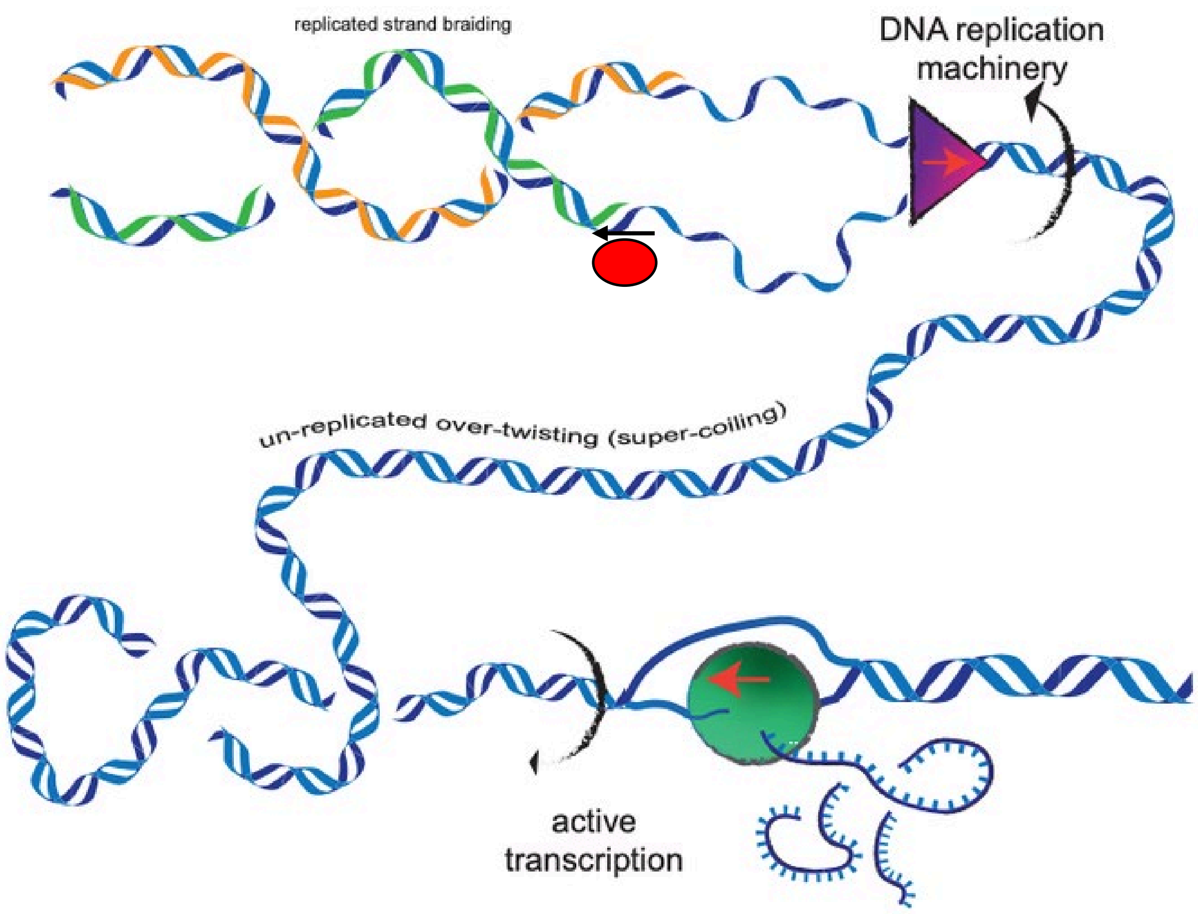
- At intermediate forces (< 70pN), the **Worm Like Chain** model fits better

WLC: with angular correlation between consecutive segments

- Persistence length $L_p = 50\,nm$ (dsDNA)
- Persistence length $L_p \leq 1\,nm$ (ssDNA)

DNA elasticity under forces < 70pN is entropic
→ Freely-jointed chain only valid at small forces

2.4 Biological relevance of DNA elasticity



S. Sevier, PRR, **2**, 023280 (2020)

DNA stretch / bending / twist important

→ protein-DNA binding depends on curvature

→ enzyme activity (molecular motors)

unwind (helicase)

read / transcribe (RNA polymerases)

copy / paste (DNA polymerases)

cut (nuclease, “molecular scissors”, *e.g.* Crispr-Cas9)

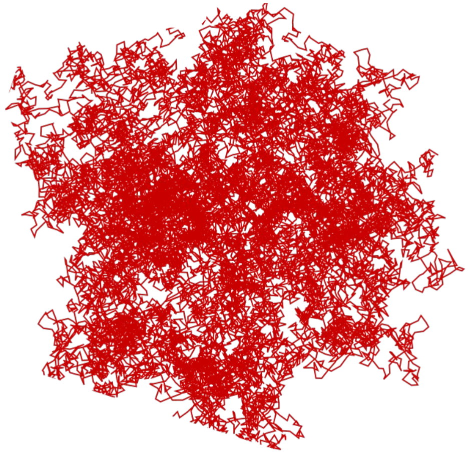
→ rate of these motors depends on extension of DNA, motor (polymerase) rate peaks at 4pN

Extensional and rotational conformation of DNA critical for biological function.

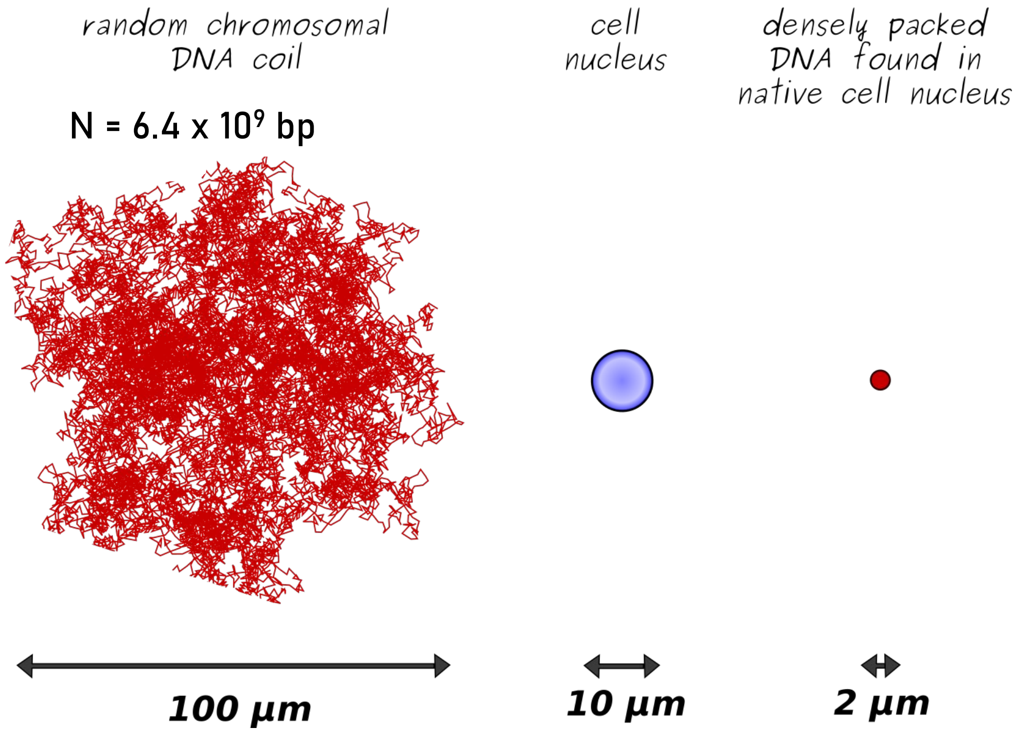
2.4 Beyond the ideal chain: DNA packing in the nucleus

random chromosomal
DNA coil

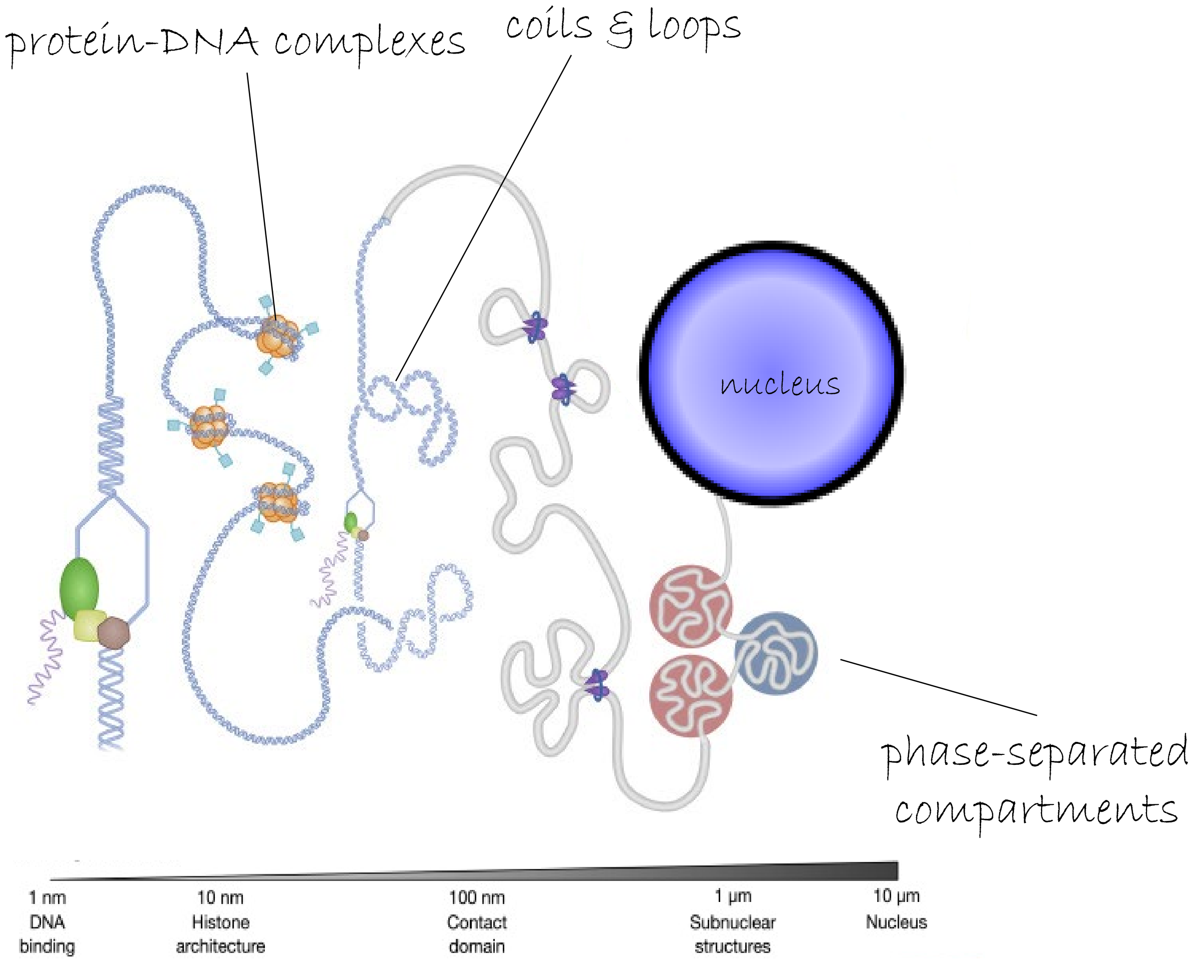
N = 6.4 x 10^9 bp



2.4 Beyond the ideal chain: DNA packing in the nucleus



Compacted DNA no longer a random coil



Chromatin conformation & rheology is scale dependent

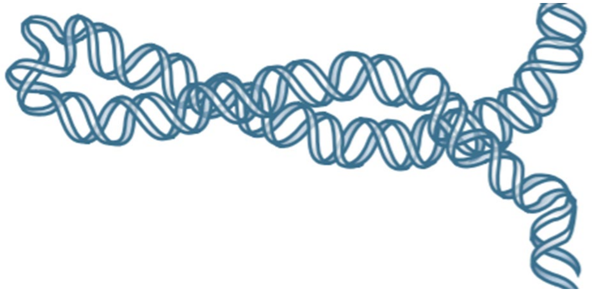
2. Summary

- DNA can be in double stranded (stiffer) or single strand (softer) configuration

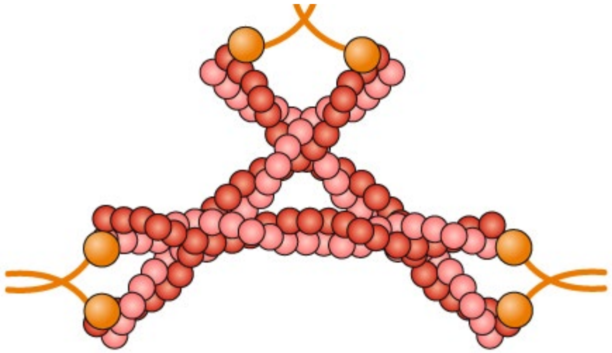
- DNA elasticity: entropic origin
  - self-avoiding chain at rest (2D)
  - freely-jointed chain (low forces) / worm-like chain under higher forces

- Mechanical work on DNA by molecular motors allow to stretch / twist / uncoil it

- In the cell, DNA is highly compacted into higher order “polymer-like” called chromatin



3. Actin filaments and actin networks: dynamic cell scaffolding



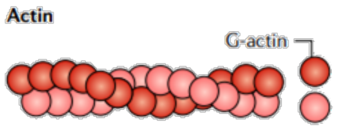
3.1 Actin filaments and actin networks

Filaments:

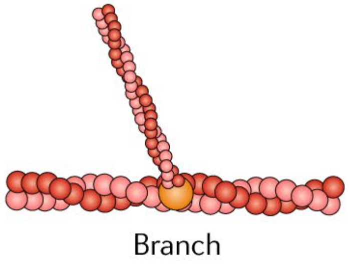
- monomers assemble via non-covalent bonds: dynamic
- helical assembly of two polar strands, ∅ ~ 7nm
- persistence length: *Lp ~ 10µm* (semi-flexible filament)

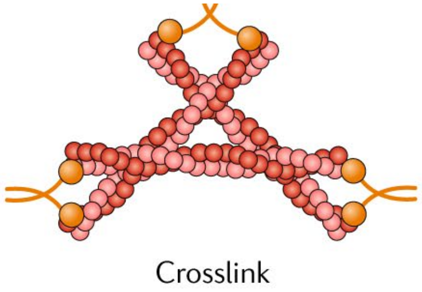
Networks

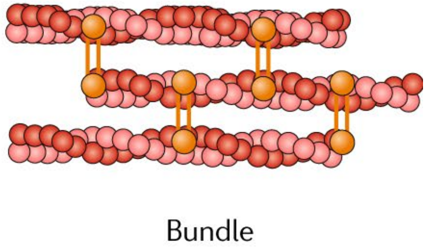
- large diversity of architectures
- many crosslinking elements (proteins, motors)



double helical strand





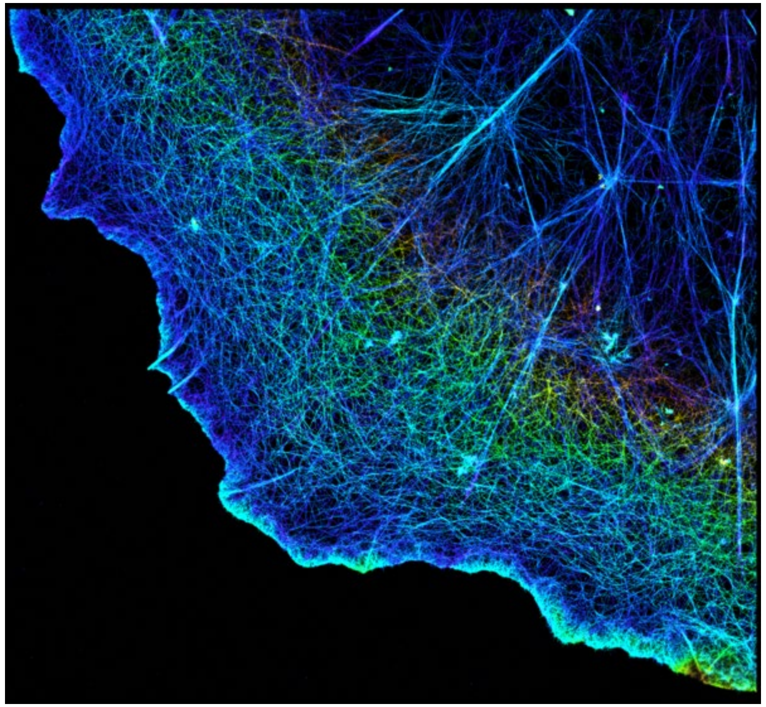


Network architecture & mechanics is determined by crosslinker type and concentration

3.2 Actin scaffolding in cell function

Cell “skeleton”

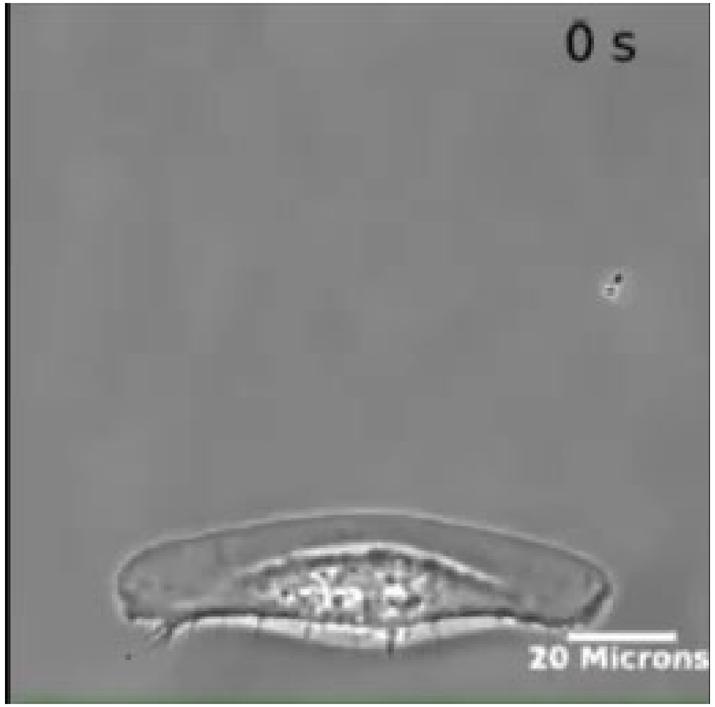
shape, resistance to tensile load, surface tension



Superresolution image of actin in a cell.
The z-dimension information is color-coded

Cell dynamics

shape changes, migration

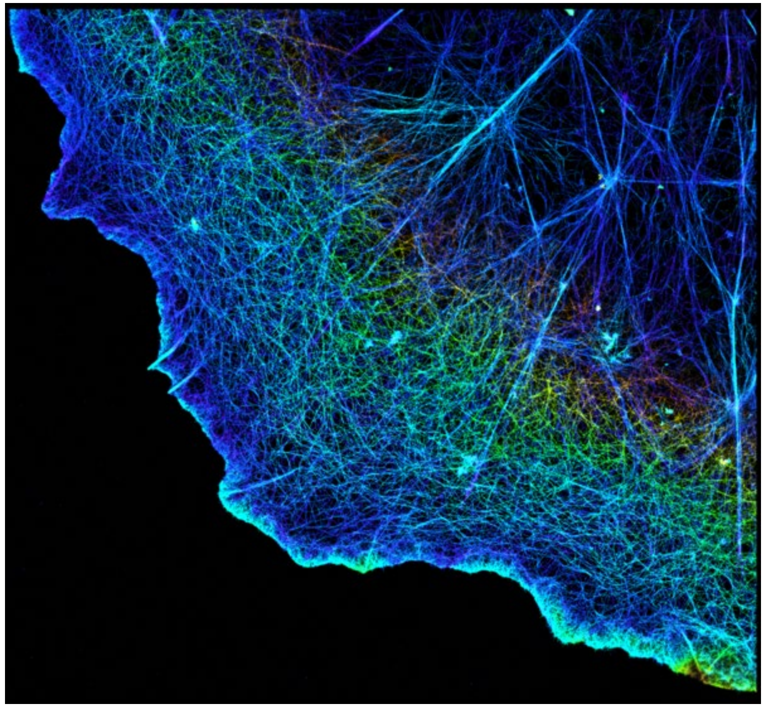


fish scale keratocyte

3.2 Actin scaffolding in cell function

Cell "skeleton"

shape, resistance to tensile load, surface tension



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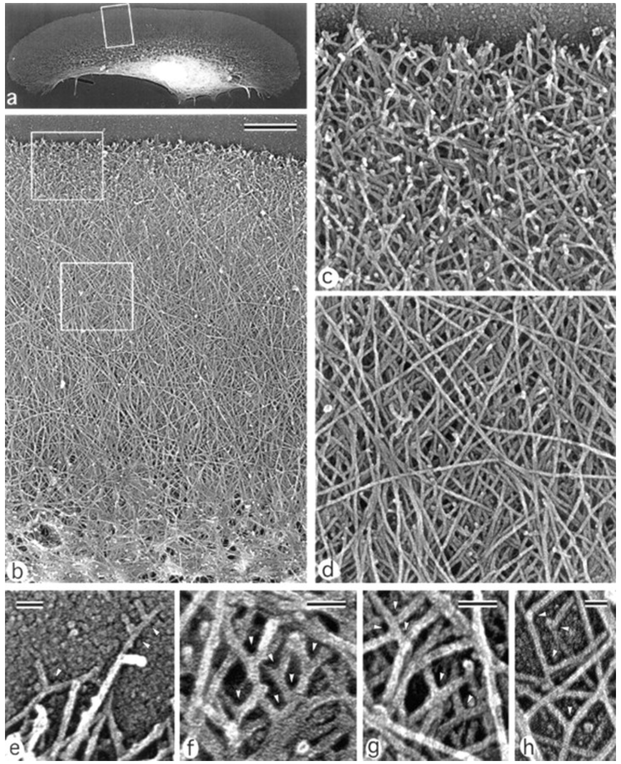
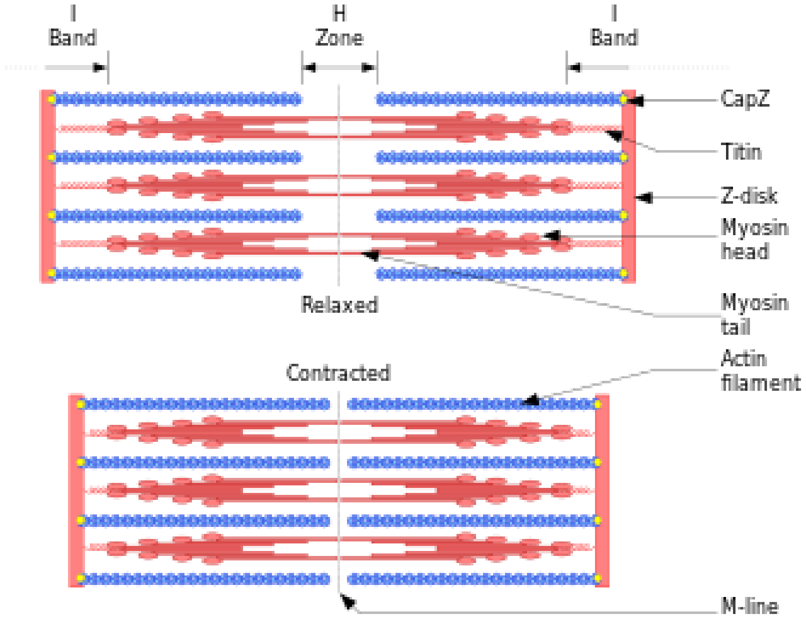


Electron Microscopy, scale bar 1µm (a-d), 50nm (e-h)

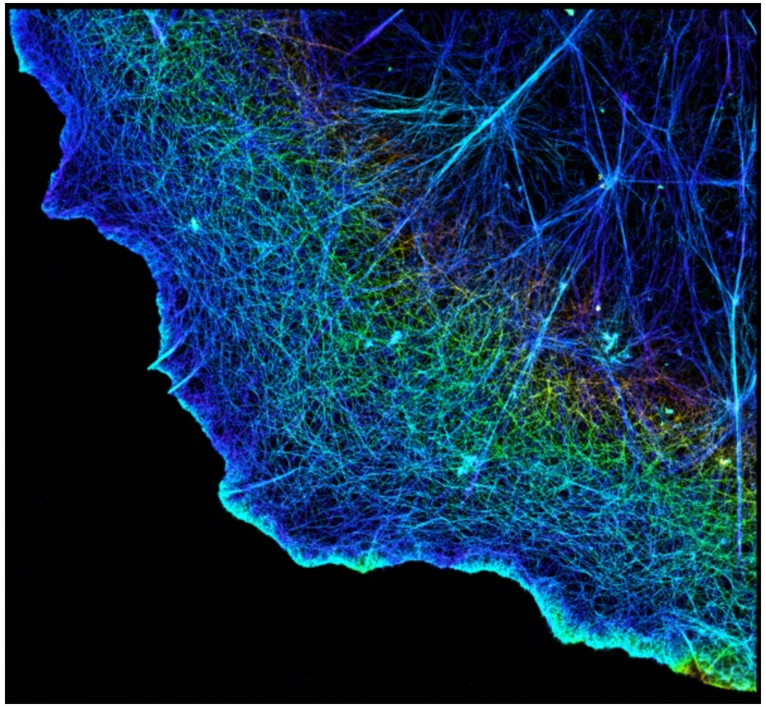
Active Matter

actin + motors = contraction

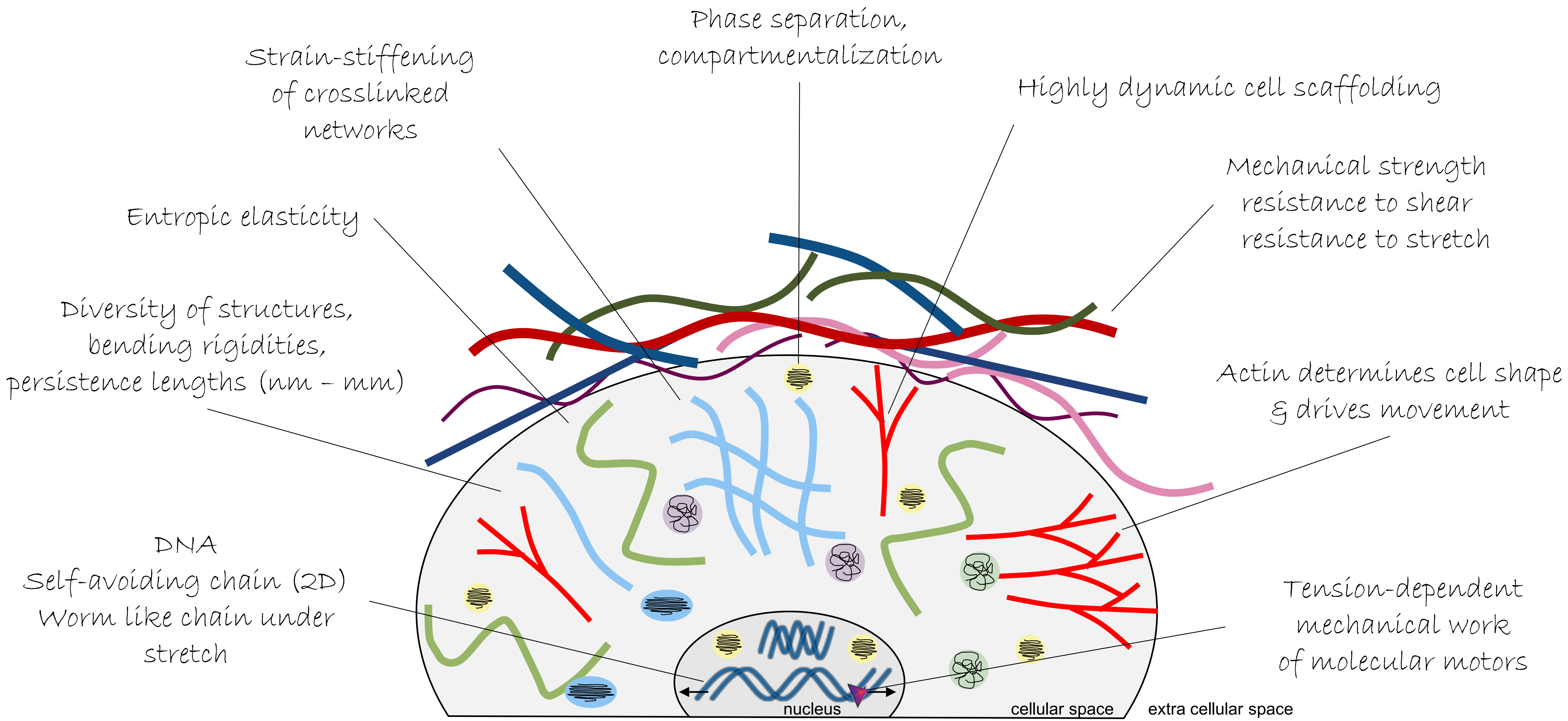


3. Summary

- actin is semiflexible polymer
- actin polymerization is dynamic
- actin is organized in networks
  branched / parallel bundles / contractile bundles
- actin drives cell movement at the macro-scale



Conclusion



Strain-stiffening of crosslinked networks
Phase separation, compartmentalization
Highly dynamic cell scaffolding
Mechanical strength resistance to shear resistance to stretch
Entropic elasticity
Diversity of structures, bending rigidities, persistence lengths (nm – mm)
Actin determines cell shape & drives movement
DNA Self-avoiding chain (2D) Worm like chain under stretch
Tension-dependent mechanical work of molecular motors
nucleus
cellular space
extra cellular space

References and further reading

Mechanical properties of biopolymer networks

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Reviews on phase separation

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Source of figures

Introduction: biopolymer filaments and networks

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