3.4 Entropy

When we talk about biopolymers, it is important to realize that the free energy of a biopolymer in thermal equilibrium is not constant. Unlike solids, biopolymers are characterized through free energy states that fluctuate in time. Just think of the kinetic energy of a gas which might fluctuate due to molecular collisions. This means that polymeric chains are usually characterized through entropy rather than through strain energy meaning that the free energy

$$\psi = W - T\,S \qquad ... \qquad \text{free energy} \tag{3.4.1}$$

as introduced in equation (2.1.1) is actually dominated by the product of temperature T and entropy S rather than by the strain energy W. Cell biologists sometimes characterize entropy dominated phenomena as finite temperature phenomena whereas strain energy dominated phenomena are referred to as zero-temperature phenomena. But how can we quantify thermal fluctuations and finite temperature phenomena? You might remember that the notion of entropy is related to disorder, but what does that actually mean? To describe the entropy of a polymer, we will compare two different models, the freely jointed chain model and the wormlike chain model. Depending on the type of polymer, either of the two models might be more appropriate. Both model are important for biopolymers, they are actually motivated by elastomers and rubber, and ideally suited to characterize soft jiggely matter that cannot be described by stored energy alone. The key equation to describe the entropy state of a polymer is the Boltzmann equation

$$S = k\,\ln(p) \qquad ... \qquad \text{Boltzmann equation} \tag{3.4.2}$$

which relates the entropy S to the probability density p via the Boltzmann constant $k = 1.38 \cdot 10^{-23}$ J/K. In this section, we will assume that the energy of biopolymers is exclusively entropic.

$$\psi = W - T\,S \approx -T\,S = -T\,k\,\ln(p) \tag{3.4.3}$$

The free energy can thus be expressed as the product of the absolut temperature, typically $T = 300K$, the Boltzmann constant $k = 1.38 \cdot 10^{-23}$ J/K, and the natural logarithm of the probability p. For biopolymers, the probability measures the disorder of the chain. To be more precise, it denotes the probability that a chain of a certain stretched out length L takes a configuration in which its ends are r apart.

3.4.1 Uncorrelated chains - Freely jointed chain model

Uncorrelated chains resemble a two-dimensional random walk, or a three-dimensional random flight. The fundamental assumption this type of chain is that two neighboring bonds are completely uncorrelated. Accordingly, the overall chain configuration seems entirely random. Figure 3.8 illustrates the kinematics of an uncorrelated chain. It consists of $N = 20$ segments of equal bond length l. The overall chain length, the so-called contour length L then simply follows as $L = N\,l$.

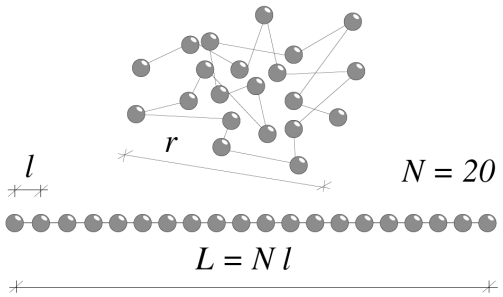

Figure 3.8: Uncorrelated chain. Kinematics of the freely jointed chain model with number of bonds N, bond length l, contour length $L = N\,l$, and end-to-end length r.

Classical Gaussian chain

To get an idea about the notion of entropy, let's consider a chain with N segments in one dimension. Table 3.4 displays the possible configurations and their probability for chains with N bonds. In one dimension, the number of possible configurations is obviously 2^N. The probability $p(r/L)$ is the discrete representation of the binomial coefficients. It is intuitive that the probability tends to zero for the stretched out case as $r = \pm L$ corresponding to the configuration with the highest order, and that it is highest for values close to zero $r = 0$ corresponding to the configuration with the highest disorder. There are many configurations for $r < L$ but only one for $r = \pm L$. More

bonds	configurations	probability $p(r/L)$
N = 1	$2^1 = 2$	$1 - 1$
N = 2	$2^2 = 4$	$1 - 2 - 1$
N = 3	$2^3 = 8$	$1 - 3 - 3 - 1$
N = 4	$2^4 = 16$	$1 - 4 - 6 - 4 - 1$
N = 5	$2^5 = 32$	$1 - 5 - 10 - 10 - 5 - 1$
N = 6	$2^6 = 64$	$1 - 6 - 15 - 20 - 15 - 6 - 1$

Table 3.4: Probability distribution for configurations of one-dimensional chain with N bonds

possible configurations are related to a higher entropy level since $S = k\,\ln(p)$. Stretching a polymer reduces the number of potential configurations, it lowers the entropy. Heating a polymer makes it more jiggly, it increases the entropic contribution to the free energy. For an infinitely long chain $N \to \infty$, the probability density converges to the classical Gauss distribution in one dimension, i.e., $p^{1\text{dim}} = p_0\,\exp(-\frac{1}{2}\,N\,r^2/L^2)$. If we generalize the motivation to a three-dimensional setting, we obtain the following probability density distribution, $p^{3\text{dim}} = p_0\,\exp(-\frac{3}{2}\,N\,r^2/L^2)$. For the classical Gaussian chain, the probability that the two ends of a chain of contour length L are a distance of r apart can thus be expressed as follows.

$$p = p_0\,\exp(-\tfrac{3}{2}\,N\,r^2/L^2) \tag{3.4.4}$$

Combining this probability with the expression for the free energy (3.4.3) and the Boltzmann equation (3.4.2), we obtain the following free energy for one individual biopolymer modeled as a freely jointed Gaussian chain.

$$\psi^{\text{fjc}} = \psi_0^{\text{fjc}} + k\,T\,N\,\tfrac{3}{2}\,\frac{r^2}{L^2} \tag{3.4.5}$$

Here, ψ_0^{fjc} is the energy of the unperturbed state. To determine the chain force f^{fjc}, we take the derivative of the energy with respect to the chain kinematics represented through (r/L).

$$f^{\text{fjc}} = \frac{\partial \psi^{\text{fjc}}}{\partial (r/L)} = k\,T\,N\,3\,\frac{r}{L} \tag{3.4.6}$$

From this force expression, we concluce that for Gaussian chains, the force f^{fjc} increases linearly with the chain stretch (r/L). This seems to be a rather crucial assumption. Imagine you stretch a rubber band! Our experience tells us that the initial resistance to stretching is rather low, but the resistance increases as we keep stretching. In fact, most polymers cannot be stretched beyond a certain stretch or they would rupture. In summary, the model of the classical Gaussian chain is only valid for small stretches (r/L). Once the chain stretch is beyond approximately $(r/L) < 0.2$, the response becomes highly nonlinear. The inverse Langevin model which we will discuss in the next section is able to describe these characteristics more appropriately.

Entropic spring Do you remember Hooke's law for a linear elastic spring? For that simple model, the spring stiffness k could be calculated as the second derivative of the spring energy $\psi = \frac{1}{2}\,k^{\text{spr}}\,u^2$ such that $\partial^2\psi/\partial u^2 = k^{\text{spr}}$. We can do the same thing for the entropic polymer. The second derivative of its energy $\psi^{\text{fjc}} = \psi_0^{\text{fjc}} + k\,T\,N\,\frac{3}{2}\,r^2/L^2$ with respect to r gives us $\partial^2\psi/\partial r^2 = 3\,k\,T\,N/L^2$. This is the equivalent stiffness of a spring that had the same stretch resistance as the biopolymer modeled with as an uncorrelated Gaussian chain. The biopolymer can thus be understood as an entropic spring with the spring stiffness $3\,k\,T\,N/L^2$.

Langevin chain

For the Langevin chain, the probability that a chain of contour length L takes a configuration such that its two ends are a distance r apart takes the following form.

$$p = p_0\,\exp\left(-N\,\mathcal{L}^{-1}\,r/L\,-N\,\ln(\mathcal{L}^{-1}/\sinh(\mathcal{L}^{-1}))\right) \tag{3.4.7}$$

Here $\mathcal{L}^{-1}$ is the inverse Langevin function which is defined as $\mathcal{L}(r/L) = \coth(r/L) - L/r$. Again, we combine this probability with the expression for the free energy (3.4.3) and the Boltzmann equation (3.4.2), and obtain the free energy for one individual biopolymer modeled as a freely jointed Langevin chain.

$$\psi^{\text{fjc}} = \psi_0^{\text{fjc}} + k\,T\,N\,(\mathcal{L}^{-1}\,r/L\,+\ln(\mathcal{L}^{-1}/\sinh(\mathcal{L}^{-1}))) \tag{3.4.8}$$

The corresponding chain force f^{fjc} follows from the derivative of the energy with respect to the chain kinematics (r/L).

$$f^{\text{fjc}} = \frac{\partial \psi^{\text{fjc}}}{\partial (r/L)} = k\,T\,N\,\mathcal{L}^{-1} \tag{3.4.9}$$

So, now, how does this relate to the Gaussian chain? We can approximate the inverse Langevin function by the following Padé approximation

$$\mathcal{L}^{-1} \approx \frac{3 - r^2/L^2}{1 - r^2/L^2}\,\frac{r}{L} \qquad \ldots \qquad \text{Padé approximation of Langevin function} \tag{3.4.10}$$

to get a better understanding about what these equations mean. For small stretches, we can neglect the quadratic terms $r^2/L^2 \approx 0$ such that $\mathcal{L}^{-1} \approx [3-0]/[1-0]\,r/L = 3\,r/L$. Does this make sense? In the limit of small strains, the chain force of the Langevin chain $f^{\text{fjc}} = k\,T\,N\,\mathcal{L}^{-1} = k\,T\,N\,3\,r/L$ corresponds to the chain force of the classical linear Gaussian chain. At larger strains, however, the Langevin chain is capable of reproducing the experimentally observed increase in resistance as the chain gets stretched out, see figure 3.10 for a comparison of both chain models.

3.4.2 Correlated chains - Wormlike chain model

So now, if you think of the biopolymers we have talked about in class, do you really think they look like the chain in figure 3.8? Microtubules are rather stiff, almost beam like. We would expect them to have a certain initial stiffness, even at low stretches. How can we account for biopolymers, that are not entirely jiggly, but are somewhat in between soft like an uncorrelated chain and stiff like a beam? We can use a model that has originally been developed for elastomers [24] and has then be adopted to model double-stranded DNA, unstructured RNA, unstructured proteins, and the collagen

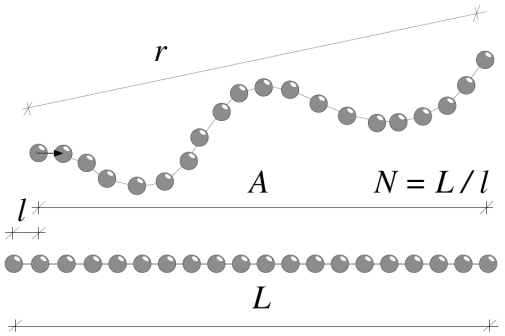

Figure 3.9: Correlated chain. Kinematics of the wormlike chain model with number of bonds N, bond length l, contour length $L = N\,l$, end-to-end length r, and persistence length A.

triple helix [8, 28]. For these biopolymers, two neighboring segments are not completely uncorrelated. In essence, typical biopolymers do not randomly change their orientation. Instead, they have a wormlike appearance which coined the name of the wormlike chain model. Figure 3.9 illustrates the kinematics of the wormlike chain

model characterized through the number of bonds N, the bond length l the contour length $L = N\,l$ and the end-to-end length r. In contrast to the freely jointed chain, however, there is another important kinematic parameter that characterized the configuration of the chain: the persistence length A which can take values between the bond length and the contour length, $l \leq A \leq L$. This persistence length is a measure of the initial chain stiffness. In its limits $A = l$ and $A = L$ the chain corresponds to the uncorrelated chain of section 3.4.1 and to the simple elastic beam of section 3.3.1, respectively. The free energy of a single wormlike chain polymer can be approximated by the following equation.

$$\psi^{\text{wlc}} = \psi_0^{\text{wlc}} + \frac{k\,T}{4\,A\,L}\left[2\,\frac{r^2}{L^2} + \frac{1}{1-r/L} - \frac{r}{L}\right] \tag{3.4.11}$$

Again, the force f^{wlc} that a polymer sustains upon stretching follows from the derivative of the energy with respect to the chain kinematics (r/L).

$$f^{\text{wlc}} = \frac{\partial \psi^{\text{wlc}}}{\partial (r/L)} = \frac{k\,T}{4\,A\,L}\left[4\,\frac{r}{L} + \frac{1}{[1-r/L]^2} - 1\right] \tag{3.4.12}$$

In figure 3.10 we can see how the choice of the persistence length A influences the force stretch response of the polymer. It is important to remember that for the wormlike chain model based on the particular equation for the chain force (3.4.12), stretches are limited to $r < L$ in the model since the denominator of one of the terms then tends to zero. Depending on the particular problem, this might limit the application of the model. In reality polymers can, of course, be stretched beyond their contour length L.

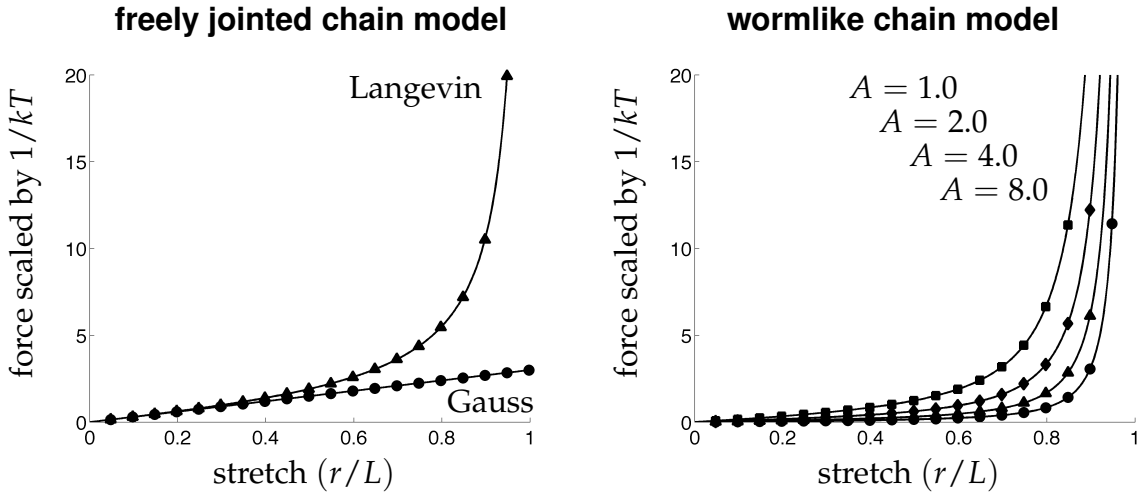

Figure 3.10: Force stretch relation of freely jointed chain models, left, and wormlike chain models, right. In the small strain limit, the Gaussian chain and the Langevin chain display an identical behavior. The Gaussian chain is linear throughout the entire regime whereas the Langevin chain stiffens significantly close to the locking stretch at $r/L = 1$. Different persistence length produce a different force stretch response of the wormlike chain model.

3.4.3 Concept of persistence length

The persistence length of biopolymers can be measured experimentally with the help of atomic force microscopy and optical tweezers. Table 3.6 summarizes some experimentally measured persistence lengths for different biological filaments.

biopolymer	configuration	A [nm]
spectrin	double-stranded filament	$10 - 20$
DNA	double helix	$51 - 55$
F-actin	filament	$10 - 20 \cdot 10^3$
microtubules	13 protofilaments	$1 - 6 \cdot 10^6$

Table 3.5: Experimentally measured persistence lengths of different biopolymers

When we explore the thermal fluctuations of typical biopolymers, we will observe two phenomena: (i) stiffer filaments are straighter, and (ii) colder filaments are straighter. But how we quantify straight? In our common understanding, straight is related to the length scale over which a polymer changes directions. From observations, we know that this length scale: (i) should be proportional to the bending stiffness EI and (ii) inversely proportional to the temperature weighted by the Boltzmann constant kT. This length scale defines the persistence length A.

$$A = \frac{EI}{kT} \qquad ... \qquad \text{persistence length} \tag{3.4.13}$$

It is easy to show that A has indeed the unit of a length, i.e., $[A] = [EI]/[kT]$ =[N/m^2 · m^4] / [J/K · K] =[Nm^2/J] = m. The analytical results for the persistence lengths of the cytoskeletal filaments are given in table 3.5.

	r	E	EI	$A = [EI]/[kT]$
microtubule	12.5 nm	$1.9{\cdot}10^9$N/m^2	$364{\cdot}10^{-25}$Nm^2	8.800 mm
intermediate filament	5.0 nm	$2{\cdot}10^9$N/m^2	$10{\cdot}10^{-25}$Nm^2	0.240 mm
actin filament	3.5 nm	$1.9{\cdot}10^9$N/m^2	$2{\cdot}10^{-25}$Nm^2	0.048 mm

Table 3.6: Persistence lengths of major consituents of cytoskeleton at room temperature: microtubules, intermediate filaments and actin filaments

Persistence length of spaghetti Try to guess the persistence length A of spaghetti at room temperature. Would it be smaller than the spaghetti length, approximately the same or larger? Assume spaghetti have a diameter of $d = 2\text{mm}$ and a Young's modulus of $E = 1 \cdot 10^8 \text{J/m}^3 = 1 \cdot 10^8 \text{N/m}^2$. The temperature and the Boltzmann constant are $T = 300\text{K}$ and $k = 1.38 \cdot 10^{-23}\text{J/K}$. The persistence length of spaghetti is $A = [EI] \,/\, [kT]$, with the moment of inertia $I = [\pi r^4] \,/\, 4$ with r=1mm. Accordingly, $A = [1 \cdot 10^8\,\text{N/m}^2\,\pi\,\text{mm}^4] \,/\, [4 \cdot 1.38 \cdot 10^{-23}\,\text{J/K} \cdot 300\,\text{K}] = 1.8 \cdot 10^{18}\text{m}$. An uncooked spaghetti changes its direction at length scales of the order of $A = 1.8 \cdot 10^{15}\text{km}$. Is that a lot? Well, yes, that's quite stiff if you consider that the distance from the earth to the moon is about $3.8 \cdot 10^5\text{km}$!

Persistence length of flagella Flagella are tail-like structures that project from the cell body of certain prokaryotic and eukaryotic cells. Flagella are hollow cylinders, of the order of 10μm long, used for locomotion. Calculate the persistence length A of flagella at room temperature $T = 300\text{K}$. Assume an inner and outer radius of $r^{\text{int}} = 0.07\,\mu\text{m}$ and $r^{\text{out}} = 0.10\,\mu\text{m}$, respectively, and a Young's modulus of $E = 1 \cdot 10^8\,\text{J/m}^3 = 1 \cdot 10^8\,\text{N/m}^2$. For hollow cylinders, $I = \pi\,[\,r^{\text{out}\,4} - r^{\text{int}\,4}\,] \,/\, 4$. Accordinlgy, $A = [EI] \,/\, [kT] = [1 \cdot 10^8\,\text{J/m}^3\,\pi\,[0.10^4 - 0.07^4]\,\mu\text{m}^4] \,/\, [4 \cdot 1.38 \cdot 10^{-23} \cdot \text{J/K}\,300\,\text{K}] = 1.44$ m. The persistence length of flagella is $A = 1.44\text{m}$. As expected, they are relatively stiff to support cell locomotion.

3.5 Summary

3.6 Problems

Problem 3.1 - Polymerization kinetics

A polymer starts to grows in a monomer solution of initial concentration C_0. Assume the rate equation for the number of monomers in the filament is governed by the kinetics of assembly as discussed in class.

$$\frac{\mathrm{d}n}{\mathrm{d}t} = k_{on}\,C - k_{off}$$

In class, we have assumed that the concentration, the number of free monomers in the volume, does not change in time, $C = \text{const}$. Assume now, that no new monomer is added to the solution as the filament grows and therefore $C = C(t)$.

- Sketch the evolution of the free monomer concentration C as a function of time t.
- Determine the tangent to the curve at $t \to 0$.
- Determine the asymptotic value for the concentration C as $t \to \infty$.