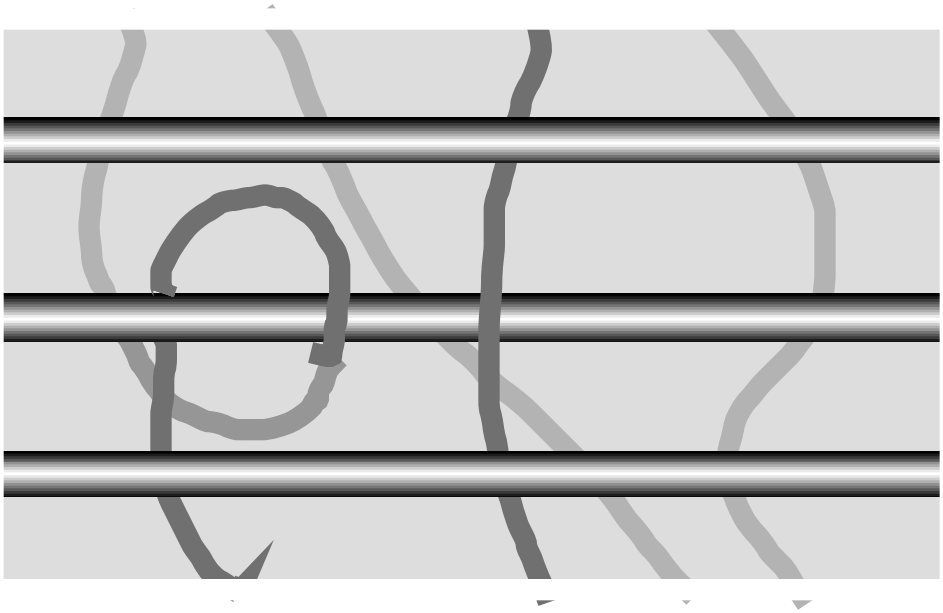
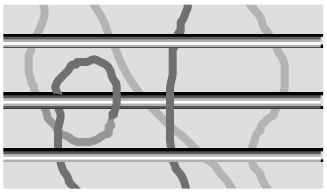


# PART I - RODS AND ROPES



# CHAPTER 2 - POLYMERS

The structural elements of the cell can be broadly classified as filaments or sheets, where by the term *filament*, we mean a string-like object whose length is much greater than its width. Some filaments, such as DNA, function as independent units, but most structural filaments in the cell are linked to form two- or three-dimensional networks. As seen on the cellular length scale of a micron, individual filaments may be relatively straight or highly convoluted, reflecting, in part, their resistance to bending. This opening chapter to Part I concentrates on the mechanical properties of individual filaments, such as their bending or stretching resistance; the two chapters making up the remainder of Part I consider how filaments are knit together to form networks, perhaps closely associated with a membrane as a two-dimensional web (Chap. 3) or perhaps extending though the three-dimensional volume of the cell (Chap. 4).

## 2.1 Filaments in the cell

Our discussion of cellular ropes and rods begins with a look at their molecular composition and linear dimensions. All of the ropes are linear polymers, in the sense that they are constructed from individual monomeric units to form an unbranched chain. The monomers need not be identical, and may themselves be constructed of more elementary chemical units. For example, the monomeric unit of DNA and RNA is a troika of phosphate, sugar and organic base, with the phosphate and sugar units alternating along the backbone of the polymer (see Appendix B). However, the monomers are not completely identical because the base may vary from one monomer to the next. The double helix of DNA contains two sugar-base-phosphate strands, with a length along the helix of 0.34 nm per pair of organic bases, and a corresponding molecular mass per unit length of about 1900 daltons/nm (Saenger, 1984).

The principal components of the cytoskeleton - actin, intermediate filaments and microtubules - are themselves composite structures made from protein subunits, each of which is a linear chain hundreds of amino acids long (Hesketh and Prym, 1996). In addition, some cells contain fine strings of the protein spectrin, whose structure we discuss first before considering the thicker filaments of the cytoskeleton. As organized in the human erythrocyte, two pairs of chains, each pair containing two intertwined and inequivalent strings of spectrin (called α and β), are joined end-to-end to form a filament about 200 nm in contour length. The α and β chains have molecular masses of 230,000 and 220,000 daltons, respectively, giving a mass per unit length along the tetramer of 4500 daltons/nm. An individual chain folds back on itself repeatedly like a Z, so that each monomer is a series of 19 or 20 relatively rigid barrels 106 amino acid residues long, as illustrated in Fig. 2.1(a) (see Rief *et al*., 1999, for spectrin folding).

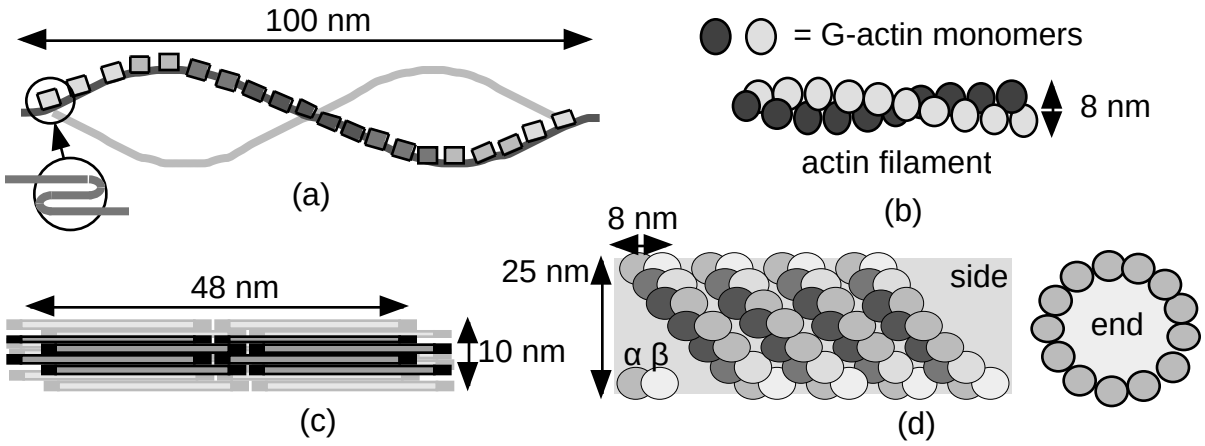



Fig. 2.1. (a) Two spectrin chains intertwine in a filament, where the boxes represent regions in which the protein chain has folded back on itself (as in the inset). The two strings are stretched and separated for clarity. (b) Monomers of G-actin associate to form a filament of F-actin, which superficially appears like two intertwined strands. (c) A hollow intermediate filament is composed of 8 protofilaments, each of which is itself four monomers (see Fig. 2.2). (d) Microtubules usually contain 13 protofilaments, whose elementary unit is an 8 nm long dimer of the proteins α and β tubulin. Both side and end views of the cylinder are shown.

Forming somewhat thicker filaments than spectrin, the protein actin is present in many different cell types and plays a variety of roles in the cytoskeleton. The elementary actin building block is the protein G-actin (*G* for globular), a single chain of approximately 375 amino acids having a molecular mass of 42,000 daltons. G-actin units can assemble into a long string called F-actin (*F* for filamentous), which, as illustrated in Fig. 2.1(b), has the superficial appearance of two strands forming a coil, although the strands are not, in fact, independently stable. The filament has a width of about 8 nm and a mass per unit length of 16,000 daltons/nm. Typical actin monomer concentrations in the cell are 1-5 mg/ml; as a benchmark, a concentration of 1 mg/ml is 24 μM for a molecular mass of 42 kD.

Intermediate filaments are slightly wider than F-actin and have a more complex hierarchical structure, as illustrated by the model in Figs. 2.1(c) and 2.2. The basic building blocks of the filament are two protein chains intertwined as a helix. Pairs of helices lie side-by-side to form a linear protofilament some 2 - 3 nm in width. The intermediate filament itself is a bundle of 8 protofilaments in a roughly cylindrical shape about 10 nm in diameter. Many intermediate-filament monomers have masses in the 40,000 - 70,000 dalton range and lengths of the order 50 nm, such that the mass per unit length of a 4-strand protofilament is about 4,500 D/nm, and that of a 32-strand filament is about 35,000 D/nm, with some variation.

The thickest individual filaments are microtubules of the protein tubulin, present as a heterodimer of α-tubulin and β-tubulin, each with a molecular mass of about 50,000 D. Pairs of α- and β-tubulin form a unit 8 nm in length, and these units can

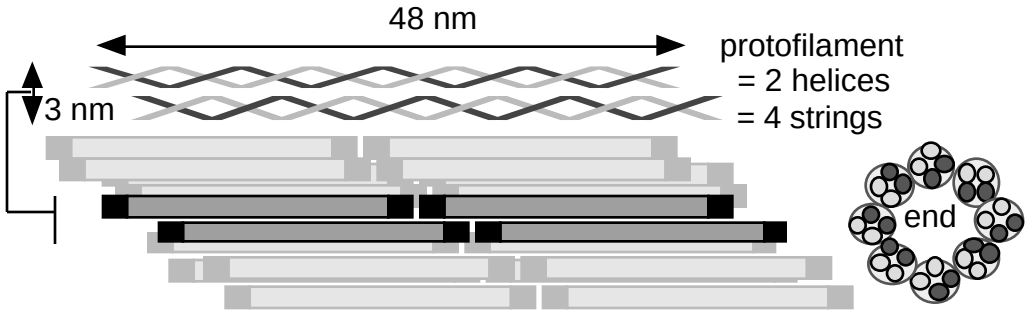



Fig. 2.2. Model of an intermediate filament, consisting of 8 protofilaments, each containing 4 protein strings, intertwined as two helical doublets.

assemble α to β successively into a hollow microtubule consisting of 13 linear protofilaments (in almost all cells), as shown in Fig. 2.1(d). The overall molecular mass per unit length of a microtubule is about 160,000 D/nm, ten times that of actin. Tubulin is present at a few mg/ml in a common cell; given a molecular mass of 100 kD for a tubulin dimer, a concentration of 1 mg/ml corresponds to 10 μM.

Biological filaments can also be found outside of the cell. Frequently present in connective tissue such as tendon, the many types of collagen are fibrous proteins that are organized into hierarchical structures such as ropes and mats. The string of amino acids in type I collagen, for example, is arranged in a helix with three amino acid residues per turn, and three such strands associate to form a larger molecule called tropocollagen. Displayed in Fig. 2.3, tropocollagen is about 300 nm long and 1.5 nm in diameter, with a mass per unit length of about 1000 D/nm. Many threads of tropocollagen are organized into a collagen fibril, which may have a diameter of 10-300 nm. The collagen fibrils themselves can assemble in parallel formation into a collagen fiber.

As a last example of cellular filaments, we mention cellulose, which, as the principal tension-bearing component of the plant cell, is a linear polysaccharide made

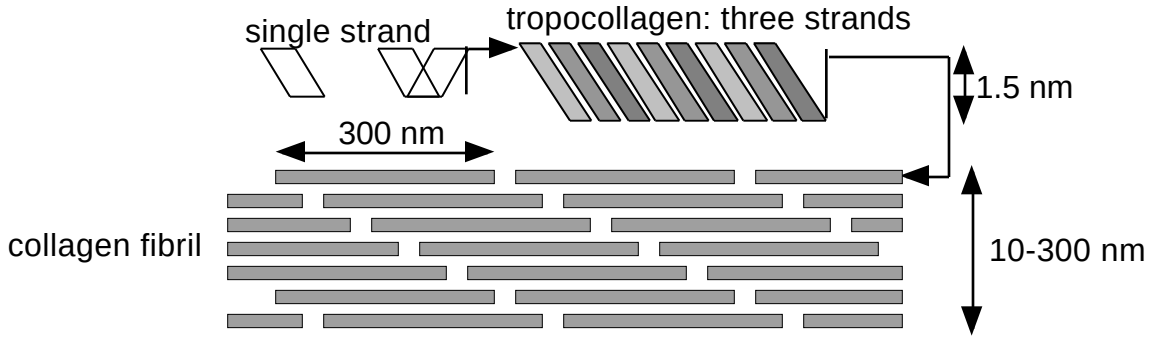



Fig. 2.3. Schematic representation of the hierarchical structure of type I collagen. A single collagen molecule is a left-handed helix; three of these strands form a right-handed helix of tropocollagen, many threads of which form a collagen fibril.

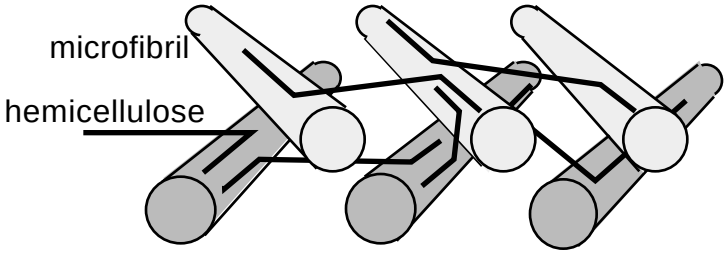



Fig. 2.4. In the plant cell wall, 60-70 individual cellulose polymers are bundled together to form a microfibril. Hemicellulose links the microfibrils at 20-40 nm intervals to form oriented sheets. The pectin network of the plant cell wall is not displayed. All elements have been separated in the drawing for clarity.

from the sugar glucose (see Appendix B for more details). A bundle of 60-70 molecular strands of cellulose lie parallel to each other to form a linear microfibril that typically has a diameter of about 10 microns. In the plant cell wall, microfibrils are themselves linked by hemicellulose molecules to form a sheetlike structure in which the fibrils are approximately aligned with each other. The cell wall then consists of multiple sheets of microfibrils, with successive sheets having different orientations of the fibrils, as in Fig. 2.4.

The design of cellular filaments has been presented in some detail in order to illustrate both their similarities and differences. Most of the filaments possess a hierarchical organization of threads wound into strings, which then may be wound into ropes. The filaments within a cell are, to an order of magnitude, about 10 nm across, which is less than 1% of the diameters of the cells themselves. As one might expect, the visual appearance of the cytoskeletal filaments on cellular length scales varies with their thickness. The thickest filaments, microtubules, are stiff on the length scale of a micron, such that isolated filaments are only gently curved. In contrast, intertwined strings of spectrin are relatively flexible: at ambient temperatures, a 200 nm filament of spectrin adopts such convoluted shapes that the distance between its end-points is only 75 nm on average (for spectrin filaments that are part of a network).

The biological rods and ropes of a cell may undergo a variety of deformations, depending upon the nature of the applied forces and the mechanical properties of the filament. Analogous to the tension and compression experienced by the rigging and masts of a sailing ship, some forces lie along the length of the filament, causing it to stretch, shorten or perhaps buckle. In other cases, the forces are transverse to the filament, causing it to bend or twist. Whatever the deformation mode, energy is required to distort the filament from its "natural" shape, by which we mean its shape at zero temperature and zero stress. Consider, for example, a uniform straight rod of length $L$ bent into an arc of a circle of radius $R$, as illustrated in Fig. 2.5(a). Within a simple model for the bending of rods introduced in Sec. 2.2, the energy $E_{arc}$ required to perform this deformation is given by

$$E_{arc} = \kappa_f L / 2R^2 = Y\mathcal{O}L / 2R^2, \qquad (2.1)$$

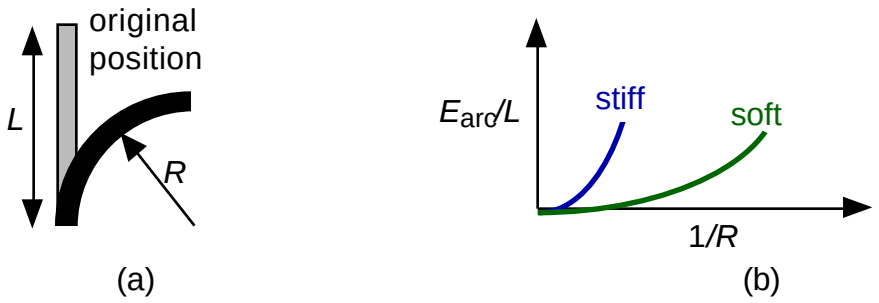



Fig. 2.5. Bending a rod of length $L$ into the shape of an arc of radius $R$ [in part (a)] requires an input of energy $E_{arc}$ whose magnitude depends upon the severity of the deformation and the stiffness of the rod (b).

where $\kappa_f$ is called the flexural rigidity of the rod: large $\kappa_f$ corresponds to stiff rods. The flexural rigidity of uniform rods can be written as $\kappa_f = Y\mathcal{I}$, where $Y$ and $\mathcal{I}$ are terms reflecting the composition and geometry of the rod, respectively, as will be explained momentarily. Fig. 2.5(b) displays how the bending energy behaves according to Eq. (2.1): a straight rod has $R = \infty$, and hence $E_{arc} = 0$, while a strongly curved rod might have $L/R$ near unity, and hence $E_{arc} >> 0$, depending on the magnitude of $\kappa_f$.

The Young's modulus $Y$ is a measure of the bulk elasticity of the material from which the rod is fabricated. Stiff materials, such as steel, have $Y \sim 2 \times 10^{11}$ J/m$^3$, while softer materials, such as plastics, have $Y \sim 10^9$ J/m$^3$. The moment of inertia of the cross section, $\mathcal{I}$ (not to be confused with the moment of inertia of the mass, familiar from rotational motion), depends upon the geometry of the rod; for instance, a cylindrical rod of constant density has $\mathcal{I} = \pi R^4/4$. Owing to its power law dependence on filament radius, the flexural rigidity of polymers in the cell spans nearly five orders of magnitude.

Now, the energy of an object in thermal equilibrium is not constant, but fluctuates with time. The reader is familiar with this phenomenon in the context of the kinetic theory of gases, wherein the kinetic energy of an individual gas molecule changes through collisions, such that the average kinetic energy per molecule is 3/2 $k_B T$, where $T$ is the absolute temperature and $k_B$ is Boltzmann's constant ($1.38 \times 10^{-23}$ J/K). Thus, the thermal energy scale is set by $k_B T$, equal to $4 \times 10^{-21}$ J at room temperature. At finite temperature, then, an otherwise straight rod bends as it exchanges energy with its environment, as suggested by Fig. 2.6. One way of quantifying the amplitude of the shape fluctuations at finite temperature is finding the typical distance along the rod over which it undergoes a significant change in direction: flexible rods change direction over shorter distances than stiff rods. This length scale must be directly proportional to the flexural rigidity $\kappa_f$ (stiffer filaments are straighter) and inversely proportional to temperature $k_B T$ (colder filaments are straighter). In fact, the combination $\kappa_f / k_B T$ has the units of length, and is defined as the persistence length $\xi_p$ of the filament:

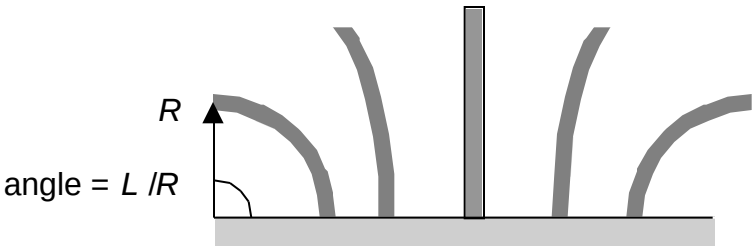



Fig. 2.6. Sample configurations of a very flexible rod at non-zero temperature as it exchanges energy with its surroundings. The base of the filament in the diagram is fixed. The configuration at the far left is an arc of a circle subtending an angle of $L/R$ radians.

$$\xi_p = \kappa_f / k_B T = Y\mathcal{I} / k_B T, \qquad (2.2)$$

an expression arising naturally in the treatment of shape fluctuations in Sec. 2.2. Some intuition about the meaning of the persistence length can be found by considering a filament with a length $L = \xi_p$. From Eq. (2.1), the energy $E_{arc}$ rises as a filament bends, reaching the thermal energy scale $k_B T$ when $R = \xi_p / \sqrt{2}$ for $L = \xi_p$. This corresponds to a highly curved configuration, where the arc subtends an angle $L/R = \sqrt{2}$ radians = 81 degrees, according to Fig. 2.6. In other words, a filament displays significant bending due to thermal fluctuations at length scales of the order of $\xi_p$.

So long as its persistence length is large compared to its contour length, *i.e.* $\xi_p >> L$, a filament appears relatively straight, and recognizable as a rod. However, if $\xi_p << L$, the filament adopts more convoluted shapes, such as that in Fig. 2.7(a). What is the likelihood that a flexible filament will be observed in a convoluted shape, as opposed to a straight one? Using the end-to-end displacement $r_{ee}$ as an observable of a configuration, there are many convoluted shapes with $r_{ee}$ close to zero, but very few extended configurations with $r_{ee} \sim L$, as shown in the example in Fig. 1.13. Recall that entropy is proportional to the logarithm of the number of configurations (see Appendix C), so that $r_{ee}/L$ near zero corresponds to large entropy and $r_{ee}/L$ near unity

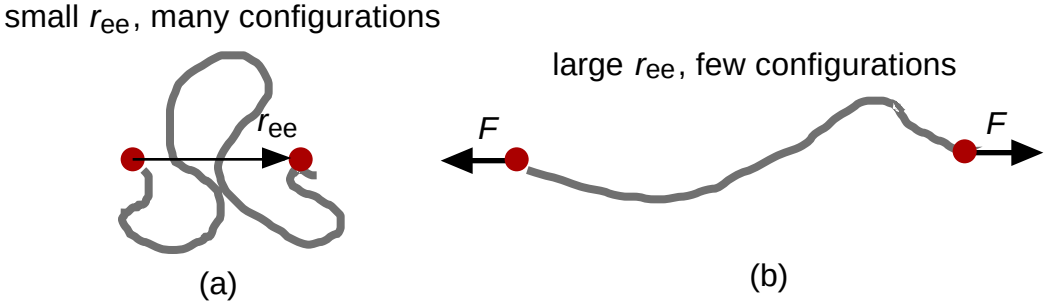



Fig. 2.7. Two samples from the set of configurations available to a highly flexible filament. The end-to-end displacement vector $r_{ee}$ is indicated by the arrow in part (a). The number of configurations available at a given end-to-end distance is reduced as a force $F$ is applied to the ends of the filament in (a) to stretch it out like that in (b).

corresponds to low entropy. Thus, entropy favors convoluted configurations, and consequently, $r_{ee}/L$ is more likely to be near 0 than 1. Further, it will be shown in Sec. 2.3 that the average value of $r_{ee}$ does not grow linearly with the contour length, but rather as a fractional power like the square root.

Lastly, what happens as we stretch a flexible filament by pulling on its ends, as in Fig. 2.7(b)? Stretching the filament reduces the number of configurations available to it, thus lowering its entropy; thermodynamics tells us that this is not a desirable situation - systems do not spontaneously lower their entropy, all other things being equal. Because of this, a force must be applied to the ends of the filament to pull it straight and the filament is elastic by virtue of its entropy, as explained in Sec. 1.3. For small extensions, this force is proportional to the change in $r_{ee}$ from its equilibrium value, just like Hooke's law for springs. In fact, the elastic behavior of convoluted filaments can be represented by an effective spring constant $k_{sp}$ given by

$$k_{sp} = 3k_B T / 2L\xi_p, \qquad (2.3)$$

which is valid for a chain in three dimensions near equilibrium (see Sec. 2.4).

Thus, we see that filaments exhibit elastic behavior with differing microscopic origins. At low temperatures, a filament may resist stretching and bending for purely energetic reasons associated with displacing atoms from their most energetically favored positions. On the other hand, at high temperatures, the shape of a very flexible filament may fluctuate strongly, and entropy discourages such filaments from straightening out. In Secs. 2.2 - 2.4, we investigate these situations using the formalism of statistical mechanics, before returning in Sec. 2.5 to apply the formal results to biological polymers.