1 Introduction, with the biological basis for cell mechanics

Roger D. Kamm and Mohammad R. K. Mofrad

Introduction

All living things, despite their profound diversity, share a common architectural building block: the cell. Cells are the basic functional units of life, yet are themselves comprised of numerous components with distinct mechanical characteristics. To perform their various functions, cells undergo or control a host of intra- and extracellular events, many of which involve mechanical phenomena or that may be guided by the forces experienced by the cell. The subject of *cell mechanics* encompasses a wide range of essential cellular processes, ranging from macroscopic events like the maintenance of cell shape, cell motility, adhesion, and deformation to microscopic events such as how cells sense mechanical signals and transduce them into a cascade of biochemical signals ultimately leading to a host of biological responses. One goal of the study of cell mechanics is to describe and evaluate mechanical properties of cells and cellular structures and the mechanical interactions between cells and their environment.

The field of cell mechanics recently has undergone rapid development with particular attention to the rheology of the cytoskeleton and the reconstituted gels of some of the major cytoskeletal components – actin filaments, intermediate filaments, microtubules, and their cross-linking proteins – that collectively are responsible for the main structural properties and motilities of the cell. Another area of intense investigation is the mechanical interaction of the cell with its surroundings and how this interaction causes changes in cell morphology and biological signaling that ultimately lead to functional adaptation or pathological conditions.

A wide range of computational models exists for cytoskeletal mechanics, ranging from finite element-based continuum models for cell deformation to actin filament-based models for cell motility. Numerous experimental techniques have also been developed to quantify cytoskeletal mechanics, typically involving a mechanical perturbation of the cell in the form of either an imposed deformation or force and observation of the static and dynamic responses of the cell. These experimental measurements, along with new computational approaches, have given rise to several theories for describing the mechanics of living cells, modeling the cytoskeleton as a simple mechanical elastic, viscoelastic, or poro-viscoelastic continuum, a porous gel or

soft glassy material, or a tensegrity (tension integrity) network incorporating discrete structural elements that bear compression. With such remarkable disparity among these models, largely due to the relevant scales and biomechanical issues of interest, it may appear to the uninitiated that various authors are describing entirely different structures. Yet depending on the test conditions or length scale of the measurement, identical cells may be viewed quite differently: as either a continuum or a matrix with fine microstructure; as fluid-like or elastic; as a static structure; or as one with dynamically changing properties. This resembles the old Rumi tale about various people gathered in a dark room touching different parts of an elephant, each coming up with a different theory on what indeed that object was. Light reveals the whole object to prove the unity in diversity.

The objective of this book is to bring together diverse points of view regarding cell mechanics, to contrast and compare these models, and to attempt to offer a unified approach to the cell while addressing apparently irreconcilable differences. As with many rapidly evolving fields, there are conflicting points of view. We have sought in this book to capture the broad spectrum of opinions found in the literature and present them to you, the reader, so that you can draw your own conclusions.

In this Introduction we will lay the groundwork for subsequent chapters by providing some essential background information on the environment surrounding a cell, the molecular building blocks used to impart structural strength to the cell, and the importance of cell mechanics in biological function. As one would expect, diverse cell types exhibit diverse structure and nature has come up with a variety of ways in which to convey structural integrity.

The role of cell mechanics in biological function

This topic could constitute an entire book in itself, so it is necessary to place some constraints on our discussion. In this text, we focus primarily on eukaryotic cells of animals. One exception to this is the red blood cell, or erythrocyte, which contains no nucleus but which has been the prototypical cell for many mechanical studies over the years. Also, while many of the chapters are restricted to issues relating to the mechanics or dynamics of a cell as a material with properties that are time invariant, it is important to recognize that cells are living, changing entities with the capability to alter their mechanical properties in response to external stimuli. Many of the biological functions of cells for which mechanics is central are active processes for which the mechanics and biology are intrinsically linked. This is reflected in many of the examples that follow and it is the specific focus of Chapter 10.

Maintenance of cell shape

In many cases, the ability of a cell to perform its function depends on its shape, and shape is maintained through structural stiffness. In the circulation, erythrocytes exist in the form of biconcave disks that are easily deformed to help facilitate their flow through the microcirculation and have a relatively large surface-to-area ratio to enhance gas exchange. White cells, or leucocytes, are spherical, enabling them to roll

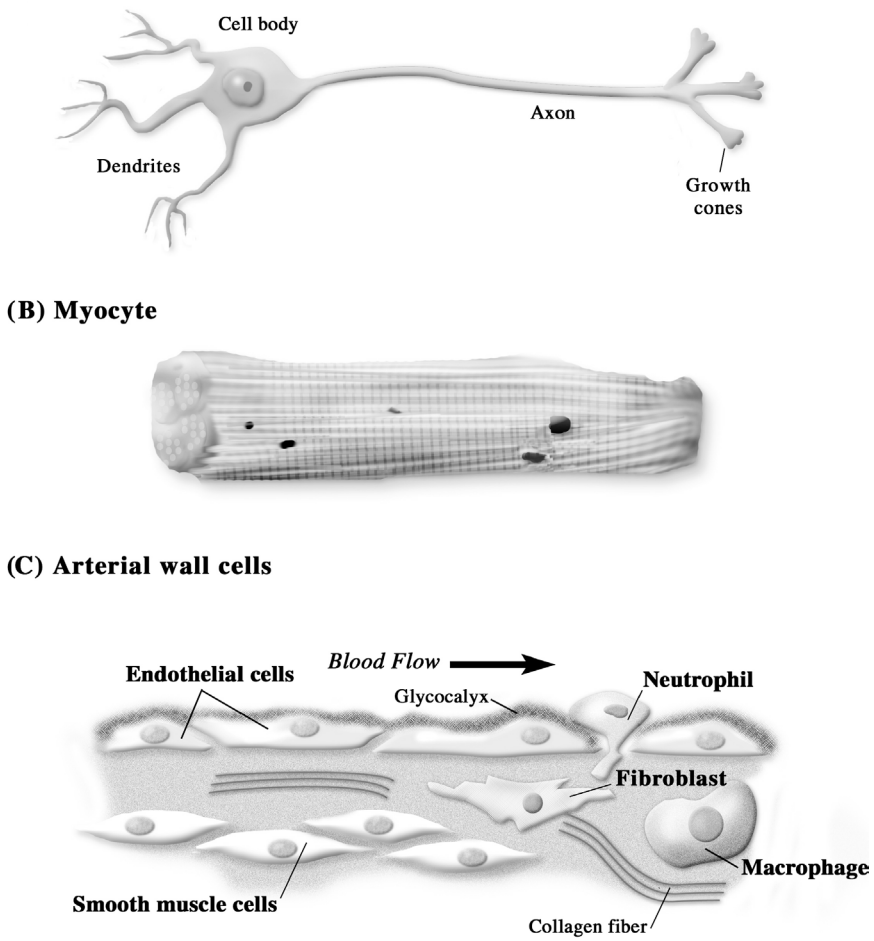

Fig. 1-1. Some selected examples of cell morphology. (A) Neuron, with long projections (dendrites and axons) that can extend a distance of 10 s of centimeters and form connections for communication with other cells. (B) Cardiac myocyte, showing the striations associated with the individual sarcomeres of the contractile apparatus. (C) Various cells found in the arterial wall. Endothelial cells line the vascular system, with a flattened, "pancake-like" morphology; neutrophils circulate in the blood until recruited by chemoattractants to transmigrate into the tissue and convert to macrophages; fibroblasts function as the "factories" for the extracellular matrix; and smooth muscle cells contribute to vessel contractility and flow control.

along the vascular endothelium before adhering and migrating into the tissue. Because their diameter is larger than some of the capillaries they pass through, leucocytes maintain excess membrane in the form of microvilli so they can elongate at constant volume and not obstruct the microcirculation. Neuronal cells extend long processes along which signals are conducted. Airway epithelial cells are covered with a bed of cilia, finger-like cell extensions that propel mucus along the airways of the lung. Some of the varieties of cell type are shown in Fig. 1-1. In each example, the internal

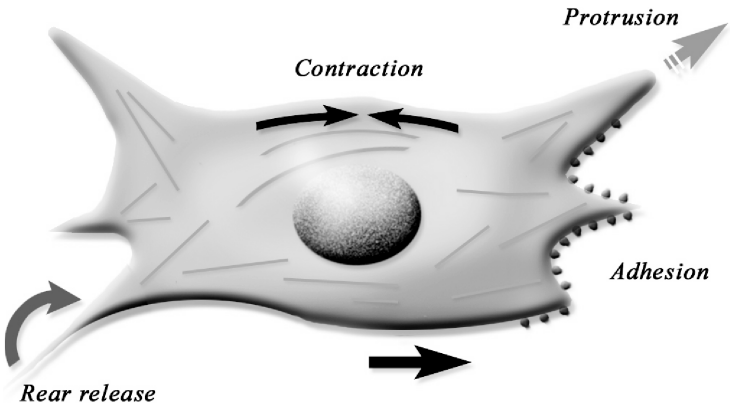

Fig. 1-2. The processes contributing to cell migration: protrusion, adhesion, contraction, and rear release. These steps can proceed in random order or simultaneously, but they all need to be operative for cell migration to take place.

structure of the cell, along with the cell membrane, provides the structural integrity that maintains the particular shape needed by the cell to accomplish its function, although the specific components of the structure are highly variable and diverse.

Cell migration

Many cells migrate, certainly during development (as the organism grows its various parts), but also at maturity for purposes of wound repair (when cells from the surrounding undamaged tissue migrate into the wound and renew the tissues) and in combating infection (when cells of the immune system transmigrate from the vascular system across the vessel wall and into the infected tissues). Migration is also an essential feature in cancer metastasis and during angiogenesis, the generation of new vessels.

Descriptions of cell migration depict a process that occurs in several stages: *protrusion*, the extension of the cell at the leading edge in the direction of movement; *adhesion* of the protrusion to the surrounding substrate or matrix; *contraction* of the cell that transmits a force from these protrusions at the leading edge to the cell body, pulling it forward; and *release* of the attachments at the rear, allowing net forward movement of the cell to occur (see, for example, DiMilla, Barbee et al., 1991; Horwitz and Webb, 2003; Friedl, Hegerfeldt et al., 2004; Christopher and Guan, 2000; and Fig. 1-2). These events might occur sequentially, with the cellular protrusions – called either filopodia ("finger-like") or lamellapodia ("sheet-like") projections – occurring as discrete events: suddenly reaching forward, extending from the main body of the cell, or more gradually and simultaneously, much like the progressive advance of a spreading pool of viscous syrup down an inclined surface. While it is well known that cells sense biochemical cues such as gradients in chemotactic agents, they can also apparently sense their physical environment, because their direction of migration can be influenced by variations in the stiffness of the substrate to which they adhere. Whatever the mode of migration, however, the central role of cell mechanics, both its passive stiffness and its active contractility, is obvious.

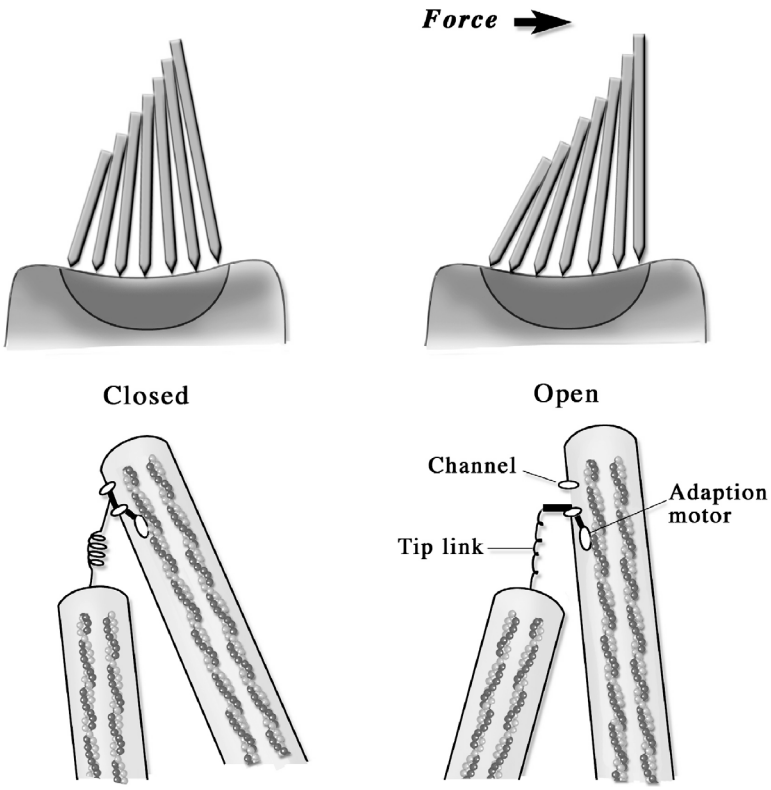

Fig. 1-3. Hair cells found in the inner ear transduce sound via the stereocilia that project from their apical surface. As the stereocilia bundle moves in response to fluid oscillations in the cochlea, tension in the tip link (a fine filament connecting the tip of one stereocilium to the side of another) increases, opening an ion channel to initiate the electrochemical response.

Mechanosensing

Nowhere is the importance of biology in cell mechanics more evident than in the ability of the cell to sense and respond to externally applied forces. Many – perhaps all – cells are able to sense when a physical force is applied to them. They respond through a variety of biological pathways that lead to such diverse consequences as changes in membrane channel activity, up- or down-regulation of gene expression, alterations in protein synthesis, or altered cell morphology. An elegant example of this process can be found in the sensory cells of the inner-ear, called hair cells, which transduce the mechanical vibration of the inner ear fluid into an electrical signal that propagates to the brain (Hamill and Martinac, 2001; Hudspeth, 2001; Hudspeth, Choe et al., 2000). By a remarkably clever design (Fig. 1-3), the stereocilia that extend from the apical surface of the cells form bundles. The individual stereocilia that comprise a bundle are able to slide relative to one another when the bundle is pushed one way or the other, but some are connected through what is termed a "tip link" – nothing more than a fine filament that connects the tip of one stereocilium to the side of another, the tension in which is modulated by an adaption motor that moves along the internal actin filaments and is tethered to the ion channel. As the neighboring filaments slide with respect to

one another, tension is developed in the tip link, generating a force at the point where the filament connects to the side of the stereocilium. This force acts to change the conformation of a transmembrane protein that acts as an ion channel, causing it to open and allowing the transient entry of calcium ions. This flux of positive ions initiates the electrical signal that eventually reaches the brain and is perceived as sound.

Although the details of force transmission to the ion channel in the case of hair-cell excitation are not known, another mechanosensitive ion channel, the *M*echano-*s*ensitive *c*hannel of *L*arge conductance (MscL) has been studied extensively (Chang, Spencer et al., 1998; Hamill and Martinac, 2001), and molecular dynamic simulation has been used to show how stresses in the cell membrane act directly on the channel and cause it to change its conductance (Gullingsrud, Kosztin et al., 2001).

This is but one example of the many ways a cell can physically "feel" its surroundings. Other mechanisms are only now being explored, but include: (1) conformational changes in intracellular proteins due to the transmission of external forces to the cell interior, leading to changes in reaction rates through a change in binding affinity; (2) changes in the viscosity of the cell membrane, altering the rate of diffusion of transmembrane proteins and consequently their reaction rates; and (3) direct transmission of force to the nucleus and to the chromatin contained inside, affecting expression of specific genes. These other mechanisms are less well understood than mechanosensitive channels, and it is likely that other mechanisms exist as well that have not yet been identified (for reviews of this topic, see Bao and Suresh, 2003; Chen, Tan et al., 2004; Huang, Kamm et al., 2004; Davies, 2002; Ingber, 1998; Shyy and Chien, 2002; Janmey and Weitz, 2004).

Although the detailed mechanisms remain ill-defined, the consequences of force applied to cells are well documented (see, for example, Dewey, Bussolari et al., 1981; Lehoux and Tedgui, 2003; Davies, 1995; McCormick, Frye et al., 2003; Gimbrone, Topper et al., 2000). Various forms of force application – whether transmitted via cell membrane adhesion proteins (such as the heterodimeric integrin family) or by the effects of fluid shear stress, transmitted either directly to the cell membrane or via the surface glycocalyx that coats the endothelial surface – elicit a biological response (see Fig. 1-4). Known responses to force can be observed in a matter of seconds, as in the case of channel activation, but can continue for hours after the initiating event, as for example changes in gene expression, protein synthesis, or morphological changes. Various signaling pathways that mediate these cellular responses have been identified and have been extensively reviewed (Davies, 2002; Hamill and Martinac, 2001; Malek and Izumo, 1994; Gimbrone, Topper et al., 2000).

Stress responses and the role of mechanical forces in disease

One reason for the strong interest in mechanosensation and the signaling pathways that become activated is that physical forces have been found to be instrumental in the process by which tissues remodel themselves in response to stress. Bone, for example, is known to respond to such changes in internal stress levels as occur following fracture or during prolonged exposure to microgravity. Many cells have shown that they can both sense and respond to a mechanical stimulus. While many of these responses appear designed to help the cell resist large deformations and possible structural

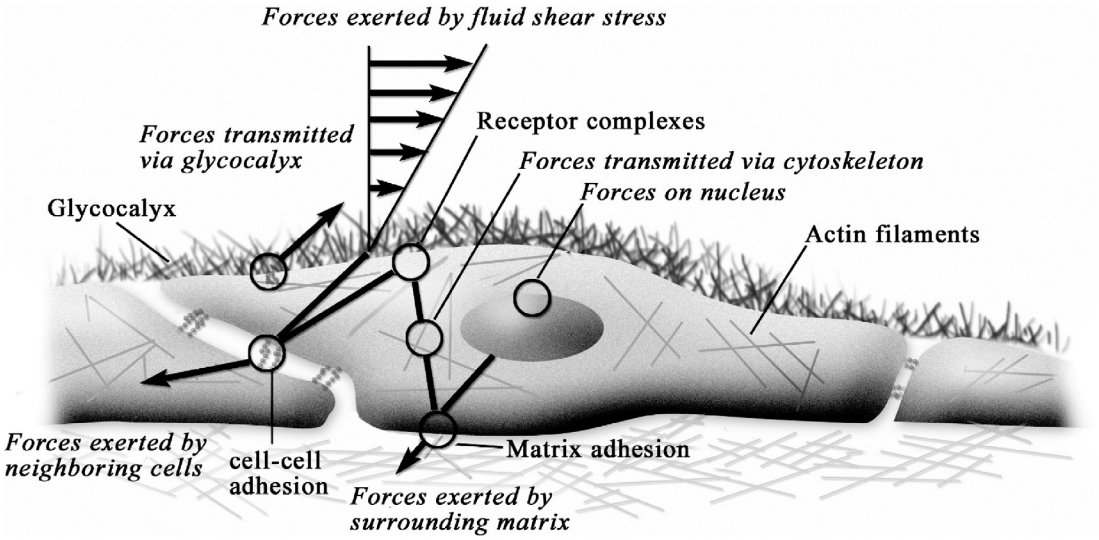

Fig. 1-4. Forces experienced by the endothelial lining of a blood vessel and the various pathways of force transmission, via receptor complexes, the glycocalyx, and the cytoskeleton even reaching the nucleus, cell-cell adhesions, and cell-matrix adhesions. Any of these locations is a potential site at which mechanical force can be transduced into a biochemical signal.

damage, others have an undesirable outcome, including atherosclerosis, arthritis, and pulmonary hypertension; there exists an extensive literature on each of these topics.

Active cell contraction

One important subset of cells primarily exists for the purpose of generating force. Cell types for which this is true include vascular smooth muscle cells, cardiac myocytes, and skeletal muscle cells. While the force-generating structures may differ in detail, the mechanisms of force generation have much in common. All muscle cells use the molecular motor comprised of actin and myosin to produce active contraction. These motor proteins are arranged in a well-defined structure, the sarcomere, and the regularity of the sarcomeres gives rise to the characteristic striated pattern seen clearly in skeletal muscle cells and cardiac myocytes (Fig. 1-5). Even nonmuscle cells contain contractile machinery, however, used for a variety of functions such as maintaining a resting level of cell tension, changing cell shape, or in cell migration. Most cells are capable of migration; in many, this capability only expresses itself when the cell is stimulated. For example, neutrophils are quiescent while in the circulation but become one of the most highly mobile migratory cells in the body when activated by signals emanating from a local infection.

Structural anatomy of a cell

Cells are biologically active, and their structure often reflects or responds to their physical environment. This is perhaps the primary distinction between traditional mechanics and the mechanics of biological materials. This is a fundamental difference

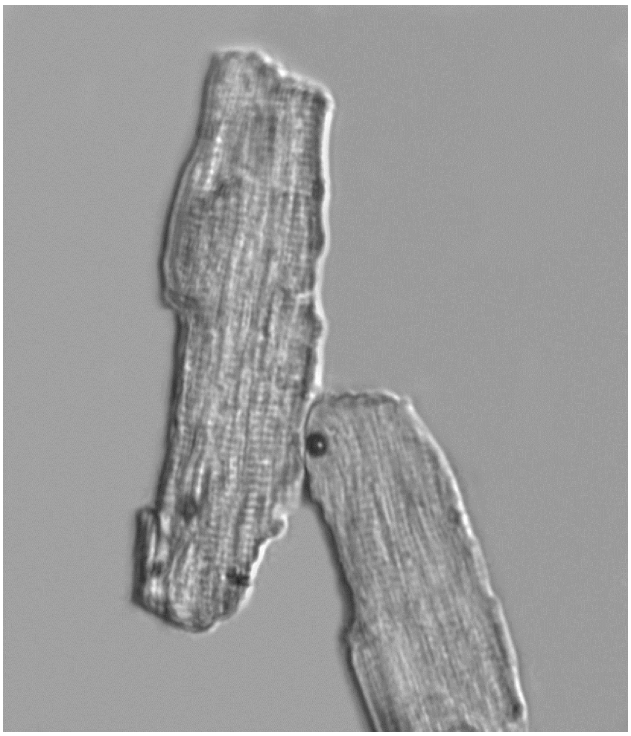

Fig. 1-5. Cardiac myocytes in culture showing the internal striations corresponding to the individual sarcomeres used for contraction. Courtesy of Jan Lammerding.

from inert materials and it must be kept in mind as we progress through the various descriptions found in this book. A second important distinction from most engineering materials is that thermal fluctuations often need to be considered, as these influence both the biochemical processes that lead to intracellular remodeling but also directly influence the elastic characteristics of the membrane and the biological filaments that comprise the cytoskeleton.

Cells often do not constitute the primary structural elements of the tissue in which they reside. For example, in either bone or cartilage, the mechanical stiffness of the resident cells are inconsequential in terms of their contribution to the modulus of the tissue, and their deformation is dictated almost entirely by that of the surrounding matrix – collagen, and hydroxyapatite in the case of bone, and a mix of collagen and proteoglycans with a high negative charge density in the case of cartilage. The role of cells in these tissues is not structural, yet through the mechanisms discussed above, cells are essential in regulating the composition and organization of the structures contained in the extracellular regions that determine the tissue's elasticity and strength through the cellular response to stress.

In other tissues, the structural role of the resident cells is much more direct and significant. Obviously, in muscle, the contractile force generated and the modulus of the tissue, either in the contracted or the relaxed state, are dominated by cellular activity. In other tissues, such as arterial wall or pulmonary airways, for example, collagen and elastin filaments in the extracellular matrix normally balance the bulk of

Table 1-1. *Major families of adhesion molecules. (E)-extracellular; (I) intracellular*

Family	Location and/or function	Ligands recognized
Integrins	Focal adhesions, hemi-desmosomes, leukocyte ("spreading") adhesion, primarily focal adhesions to matrix but also in some cell-cell adhesions	(E) fibronectin, collagen, laminin, immunoglobulins, (I) actin filaments
Selectins	Circulating cells and endothelial cells, "rolling" adhesion	Carbohydrates
Ig superfamily (immunoglobulin)	Important in immune response	Integrins, homophillic
Cadherens	Adherens junctions, desmosomes	(E) homophillic, (I) actin filaments, intermediate filaments
Transmembrane proteoglycans	Fibroblasts, epithelial cells	(E) collagen, fibronectin (I) actin filaments, heterophillic

the stress. During activation of the smooth muscle, however, stress shifts from these extracellular constituents to the contractile cells, and the vessel constricts to a diameter much smaller than that associated with the passive wall stiffness. In the case of cardiac tissue, the contractile cells, or myocytes (Fig. 1-5), constitute a large fraction of the tissue volume and are primarily responsible for the stresses and deformations of the myocardium that are time varying through the cardiac cycle.

The extracellular matrix and its attachment to cells

Contrary to the situation in most cell mechanics experiments *in vitro*, where forces might be applied directly to the cells via tethered beads, a micropipette, an AFM probe, or fluid shear stress, forces *in vivo* are often transmitted to the cell via the extracellular matrix (ECM), which shares in the load-supporting function. Many cell membrane receptors contain extracellular domains that bind to the various proteins of the ECM. For example, members of the integrin family can bind to fibronectin, vitronectin, collagen, and laminin. Intracellular domains of these same proteins bind directly (or indirectly, through other membrane-associated proteins) to the cytoskeleton. The number and variety of linking proteins is quite remarkable, as described in detail in a recent review (Geiger and Bershadsky, 2002). Other adhesion molecules bind to the ECM, basement membrane, neighboring cells, or cells suspended in flowing blood. Adhesion molecules can be either homophillic (binding to other identical molecules) or heterophillic (Table 1-1). Of these transmembrane molecules (both proteins and proteoglycans) many attach directly to the cytoskeleton, which often exhibits a denser, more rigid structure in the vicinity of an adhesion site.

Transmission of force to the cytoskeleton and the role of the lipid bilayer

Cells are separated from the external environment by a thin lipid bilayer consisting of a rich mix of phospholipids, glycolipids, cholesterol, and a vast array of transmembrane

proteins that constitute about 50 percent of the membrane by weight but only 1 to 2 percent of the total number of molecules residing in the membrane. Phospholipids, which are the most abundant, are amphipathic, having a hydrophilic part residing on the outside surface of the bilayer and a hydrophobic part on the inside. Some of the proteins serve as ion channels, others as a pathway for transmembrane signaling. Still others provide a structural bridge across the membrane, allowing for direct adhesion between the internal cytoskeleton and the extracellular matrix. Together, these are commonly referred to as *integral membrane proteins*. Roughly half of these integral proteins are able to freely diffuse within the membrane, while the rest are anchored to the cytoskeleton.

In addition to its role in communicating stress and biochemical signals into the cell, the membrane also serves a barrier function, isolating the cell interior from its extracellular environment and maintaining the appropriate biochemical conditions within for critical cell functions. By itself, the bilayer generally contributes little to the overall stiffness of the cell, except in situations in which the membrane becomes taut, as might occur due to osmotic swelling. In general, the bilayer can be thought of as a two-dimensional fluid within which the numerous integral membrane proteins diffuse, a concept first introduced in 1972 by Singer and Nicolson as the *fluid mosaic model* (Singer and Nicolson, 1972). The bilayer maintains a nearly constant thickness of about 6 nm under stress, and exhibits an area-expansion modulus, defined as the in-plane tension divided by the fractional area change, of about 0.1–1.0 N/m (for pure lipid bilayers) or 0.45 N/m (for a red blood cell) (Waugh and Evans, 1979). Rupture strength, in terms of the maximum tension that the membrane can withstand, lies in the range of 0.01–0.02 N/m, for a red blood cell and a lipid vesicle, respectively (Mohandas and Evans, 1994). Values for membrane and cortex bending stiffness reported in the literature (for example, $\sim 2 - 4 \times 10^{-19}$ N·m for the red blood cell membrane (Strey, Peterson et al., 1995; Scheffer, Bitler et al., 2001), and $1 - 2 \times 10^{-18}$ N·m for neutrophils (Zhelev, Needham et al., 1994), are not much larger than that for pure lipid bilayers (Evans and Rawicz, 1990), despite the fact that they include the effects of the membrane-associated cortex of cytoskeletal filaments, primarily spectrin in the case of erythrocytes and actin for leukocytes. When subjected to in-plane shear stresses, pure lipid bilayers exhibit a negligible shear modulus, whereas red blood cells have a shear modulus of about 10^{-6} N·s/m (Evans and Rawicz, 1990). Forces can be transmitted to the membrane via transmembrane proteins or proteins that extend only partially through the bilayer. When tethered to an external bead, for example, the latter can transmit normal forces; when forces are applied tangent to the bilayer, the protein can be dragged along, experiencing primarily a viscous resistance unless it is tethered to the cytoskeleton. Many proteins project some distance into the cell, so their motion is impeded even if they are not bound to the cytoskeleton due to steric interactions with the membrane-associated cytoskeleton.

Intracellular structures

In this text we primarily address the properties of a generic cell, without explicitly recognizing the distinctions, often quite marked, between different cell types. It is important, however, to recognize several different intracellular structures that

influence the material properties of the cell that may, at times, need to be taken into account in modeling. Many cells (leucocytes, erythrocytes, and epithelial cells, for example) contain a relatively dense structure adjacent to the cell membrane called the *cortex*, with little by way of an internal network. In erythrocytes, this cortex contains another filamentous protein, spectrin, and largely accounts for the shape rigidity of the cell. Many epithelial cells, such as those found in the intestine or lining the pulmonary airways, also contain projections (called microvili in the intestine and cilia in the lung) that extend from their apical surface. Cilia, in particular, are instrumental in the transport of mucus along the airway tree and have a well-defined internal structure, primarily due to microtubules, that imparts considerable rigidity.

Of the various internal structures, the nucleus is perhaps the most significant, from both a biological and a structural perspective. We know relatively little about the mechanical properties of the nucleus, but some recent studies have begun to probe nuclear mechanics, considering the separate contributions of the nuclear envelope, consisting of two lipid bilayers and a nuclear lamina, and the nucleoplasm, consisting largely of chromatin (Dahl, Kahn et al., 2004; Dahl, Engler et al., 2005).

Migrating cells have a rather unique structure, but again are quite variable from cell type to cell type. In general, the leading edge of the cell sends out protrusions, either lamellipodia or filopodia, that are rich in actin and highly cross-linked. The dynamics of actin polymerization and depolymerization is critical to migration and is the focus of much recent investigation (see, for example Chapter 9 and Bindschadler, Dewey, and McGrath, 2004). Active contraction of the network due to actin-myosin interactions also plays a central role and provides the necessary propulsive force.

Actin filaments form by polymerization of globular, monomeric actin (G-actin) into a twisted strand of filamentous actin (F-actin) 7–9 nm in diameter with structural polarity having a barbed end and a pointed end. Monomers consist of 375 amino acids with a molecular weight of 43 kDa. ATP can bind to the barbed end, which allows for monomer addition and filament growth, while depolymerization occurs preferentially at the pointed end (Fig. 1-6A). Filament growth and organization is regulated by many factors, including ionic concentrations and a variety of capping, binding, branching, and severing proteins. From actin filaments, tertiary structures such as fiber bundles, termed "stress fibers," or a three-dimensional lattice-like network can be formed through the action of various actin-binding proteins (ABPs). Some examples of ABPs are fimbrin and α-actinin, both instrumental in the formation of *stress fibers* or bundles of actin filaments, and filamin, which connects filaments into a three-dimensional space-filling matrix or gel with filaments joined at nearly a right angle. Recent rheological studies of reconstituted actin gels containing various concentrations of ABPs (see Chapter 2, or Tseng, An et al., 2004) have illustrated the rich complexities of even such simple systems and have also provided new insights into the nature of such matrices.

The importance of actin filaments is reflected in the fact that actin constitutes from 1 to 10 percent of all the protein in most cells, and is present at even higher concentrations in muscle cells. Actin is thought to be the primary structural component of most cells; it responds rapidly and dramatically to external forces and is also instrumental in the formation of leading-edge protrusions during cell migration. As the data in Table 1-2 illustrate, actin filaments measured by a variety of techniques (Yasuda,

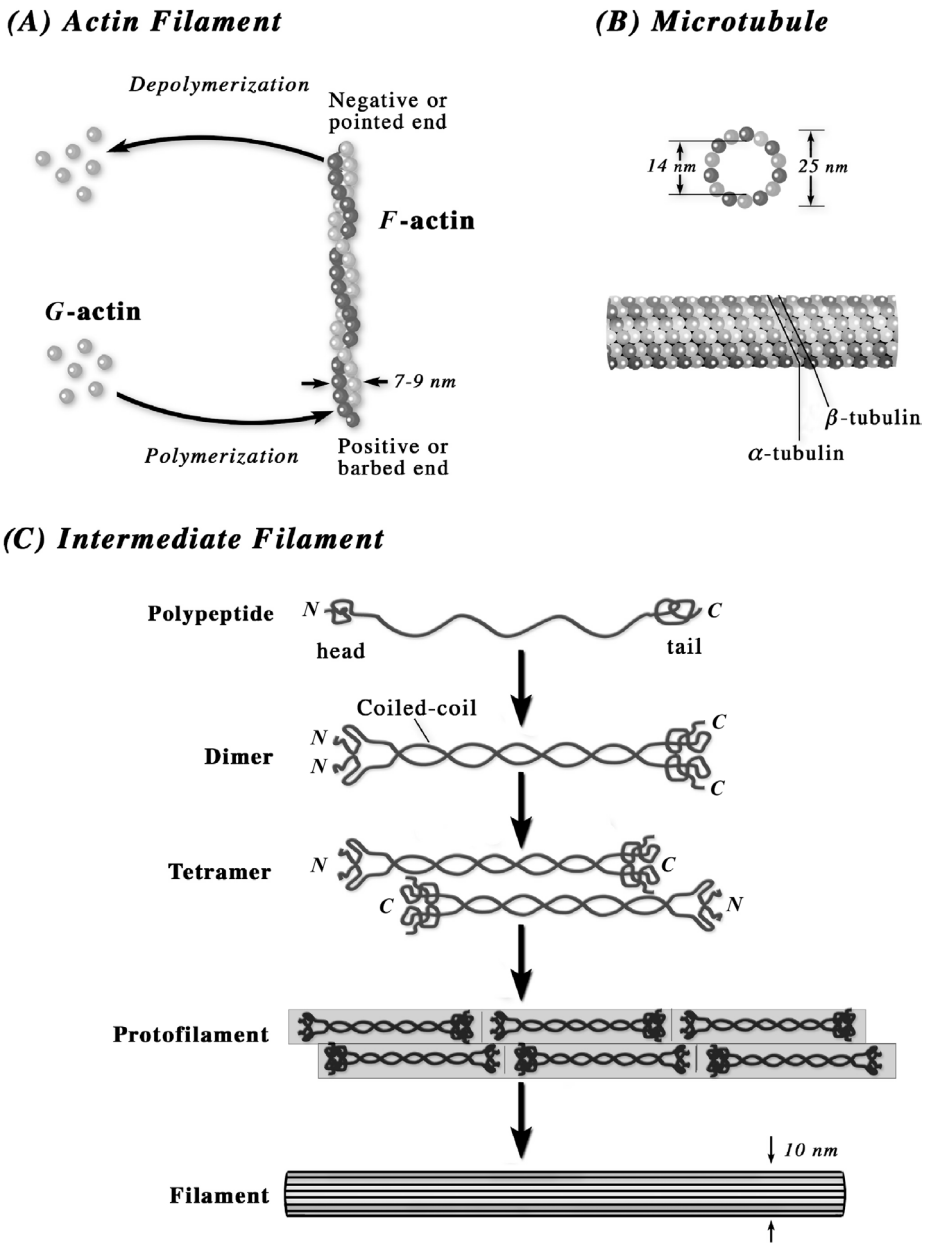

Fig. 1-6. Filaments that constitute the cytoskeleton. (A) Actin filaments. (B) Microtubules. (C) Intermediate filaments.

Miyata et al., 1996; Tsuda, Yasutake et al., 1996; Higuchi and Goldman, 1995) are stiff, having a persistence length of several microns, and an effective Young's modulus, determined from its bending stiffness and radius of $1 - 3 \times 10^9$ Pa, comparable to that of polystyrene (3×10^9 Pa) and nearly equal to that of bone (9×10^9 Pa).

Microtubules constitute a second major constituent of the cytoskeleton. These are polymerized filaments constructed from monomers of α- and β-tubulin in a helical

Table 1-2. *Elastic properties of cytoskeletal filaments*

	Diameter, $2a$ (nm)	Persistence length, l_p (μm)	Bending stiffness, K_B (Nm^2)	Young's modulus, E (Pa)
Actin filament	6–8	15	7×10^{-26}	$1.3–2.5 \times 10^9$
Microtubule	25	6000	2.6×10^{-23}	1.9×10^9
Intermediate filament	10	∼1	4×10^{-27}	1×10^9

The elastic properties of actin filaments and microtubules are approximately consistent with a prediction based on the force of van der Waals attraction between two surfaces (J. Howard, 2001). Persistence length (l_p) and bending stiffness (K_B) are related through the expression $l_p = K_B / k_B T$. Bending stiffness and Young's modulus (E) are related through the expression $K_B = EI = \frac{\pi}{4} a^4 E$ for a solid rod of circular cross-section with radius a, and $I = \frac{\pi}{4}(a_o^4 - a_i^4)$ for a hollow cylinder with inside and outside radii a_i and a_o, respectively.

arrangement, both 55 kDa polypeptides, that organize into a small, hollow cylinder (Fig. 1-6B). The filaments have an outer diameter of about 25 nm and exhibit a high bending stiffness, even greater than that of an actin filament (Table 1-2) with a persistence length of about 6 mm (Gittes, Mickey et al., 1993). Tubular structures tend to be more resistant to bending than solid cylinders with the same amount of material per unit length, and this combined with the larger radius accounts for the high bending stiffness of microtubules despite having an effective Young's modulus similar to that of actin. Because of their high bending stiffness, they are especially useful in the formation of long slender structures such as cilia and flagella. They also provide the network along which chromosomes are transported during cell division.

Microtubules are highly dynamic, even more so than actin, undergoing constant polymerization and depolymerization, so that the half-life of a microtubule is typically only a few minutes. (Mitchison and Kirschner, 1984). Growth is asymmetric, as with actin, with polymerization typically occurring rapidly at one end and more slowly at the other, and turnover is generally quite rapid; the half-life of a microtubule is typically on the order of minutes.

Intermediate filaments (IFs) constitute a superfamily of proteins containing more than fifty different members. They have in common a structure consisting of a central α-helical domain of over 300 residues that forms a coiled coil. The dimers then assemble into a staggered array to form tetramers that connect end-to-end, forming protofilaments (Fig. 1-6C). These in turn bundle into ropelike structures, each containing about eight protofilaments with a persistence length of about 1 μm (Mucke, Kreplak et al., 2004). Aside from these differences in structure, intermediate filaments differ from microfilaments and microtubules in terms of their long-term stability and high resistance to solubility in salts. Also, unlike polymerization of other cytoskeletal filaments, intermediate filaments form without the need for GTP or ATP hydrolysis.

In recent experiments, intermediate filaments have been labeled with a fluorescent marker and used to map the strain field within the cell (Helmke, Thakker et al., 2001). This is facilitated by the tendency for IFs to be present throughout the entire cell at a sufficiently high concentration that they can serve as fiducial markers.

Of course these are but a few of the numerous proteins that contribute to the mechanical properties of a cell. The ones mentioned above – actin filaments, microtubules,

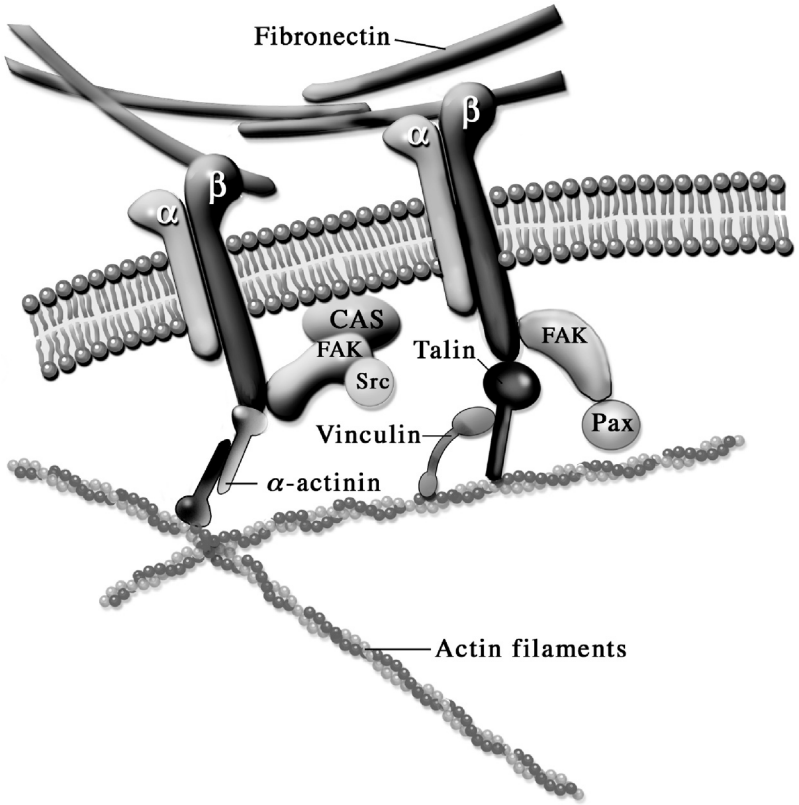

Fig. 1-7. A small sampling of the proteins found in a focal adhesion complex (FAC). Forces are typically transmitted from the extracellular matrix (for example, fibronectin), via the integral membrane adhesion receptors ($\alpha-$ and $\beta-$integrins), various membrane-associated proteins (focal adhesion kinase (FAK), paxillin (Pax), talin, Crk-associated substrate (CAS)), to actin-binding proteins (α-actinin) that link the FAC to the cytoskeleton. Adapted from Geiger and Bershadsky 2002.

and intermediate filaments – are primarily associated with the cytoskeleton, but even within the cytoskeletal network are found numerous linking proteins (ABPs constituting one family) that influence the strength and integrity of the resulting matrix. In addition to these are the molecular constituents of the cell membrane, nuclear membrane, and all the organelles and other intracellular bodies that influence the overall mechanical response of a cell. In fact, intracellular structure should be noted for its complexity, as can be seen in Fig. 1-7, which shows just a small subset of the numerous proteins that link the extracellular matrix and the cytoskeleton. Any of these constitutes a pathway for transmitting force across the cell membrane, between the proteins found in the adhesion complexes, and through the cytoskeletal network. To the extent that a particular protein is located along the force transmission pathway, not only does it play a role in transmitting stress, but it also represents a candidate for mechanosensing due to the conformational changes that arise from the transmission of force.

Active contraction is another fundamental feature of the cytoskeleton that influences its structural properties. While this is an obvious characteristic of the various types of muscle cell, most cells contain contractile machinery, and even in their resting

state can exert a force on their surroundings. Forces have been measured in resting fibroblasts, for example, where intracellular tension gives rise to stresses in the focal adhesions of the cell adherent to a flexible two-dimensional substrate of about 5 nN/μm^2, or 5 kPa (Balaban, Schwarz et al., 2001). In experiments with various cell types grown in a three-dimensional gel such as collagen, the cells actively contract the matrix by more than 50 percent (Sieminski, Hebbel et al., 2004). These contractile forces are associated with intracellular molecular motors such as those in the myosin family.

Overview

This book presents a full spectrum of views on current approaches to modeling cell mechanics. In part, this diversity of opinion stems from the different backgrounds of contributors to the field. Indeed, the authors of this book come from the biophysics, bioengineering, and physical chemistry communities, and each joins the discussion with a unique perspective on biological systems. Consequently, the approaches range from finite element methods commonly used in continuum mechanics to models of the cytoskeleton as a cross-linked polymer network to models of soft glassy materials and gels. Studies reflect both the static, instantaneous nature of the structure as well as its dynamic nature due to polymerization and the full array of biological processes. It is unlikely that a single unifying approach will evolve from this diversity, in part because of the complexity of the phenomena underlying the mechanical properties of the cell. It is our hope, however, that a better appreciation of the various perspectives will lead to a more highly coordinated approach to the essential problems and might facilitate discussions among investigators with differing views.

Perhaps the most important purpose of this monograph is to stimulate new ideas and approaches. Because no single method has emerged as clearly superior, this might reflect the need for approaches not yet envisaged. That much of the work presented here derives from publications over the past several years reinforces the notion that cell mechanics is a rapidly evolving field. The next decade will likely yield further advances not yet foreseen.

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