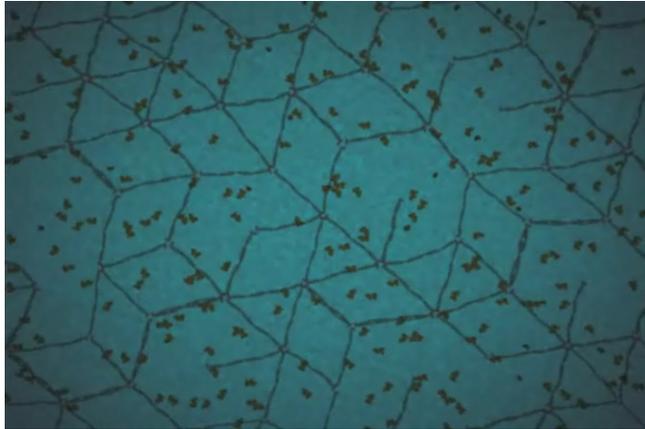
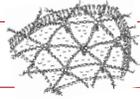


4. the cytoskeleton - network models



the inner life of a cell, viel & lue, harvard [2006]

me239 mechanics of the cell

1

Problem 1 - Filament buckling

In class, we have discussed filament buckling of actin polymers. We have seen that the critical buckling force of filopodia

$$F_{\text{fil}} = \frac{\pi^2 EI}{[2L_{\text{fil}}]^2} = \frac{\pi^2 EI}{4L_{\text{fil}}^2}$$

can be increased by fascin which can create tightly crosslinked filament bundles.

- In class, we have assumed that there are about $n = 30$ actin filaments in one filopodium. Assume we do not know the exact number n . What is the minimum number of actin filaments to obtain a critical filopodia length of $L_{\text{fil}} = 5\mu\text{m}$ assuming that the actin filaments are (i) loosely assembled (4.2.9) and (ii) tightly crosslinked (4.2.12).
- How would the minimum number of required actin filaments n change if you assume a critical filopodia length of $L_{\text{fil}} = 2.5\mu\text{m}$?

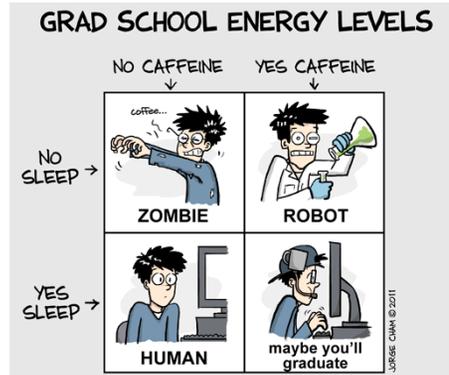
homework 02 - problem 01

3

Homework II - The cytoskeleton

due Thu 02/03/11, 3:15pm, 200-034

You can drop off late homework in a box in front of Durand 217. Please mark clearly with date and time @drop off. We will take off 10 points for each 24 hours late.



homework 02

2

Problem 2 - Forces on the cell membrane

To gain a better feeling for stresses that the cytoskeleton might induce on the cell membrane, this problem deals with the membrane pressure resulting from microtubules. Consider a generic spherical cell of radius $10\mu\text{m}$ with a tubulin heterodimer concentration of $C = n/V = 5\mu\text{M}$.

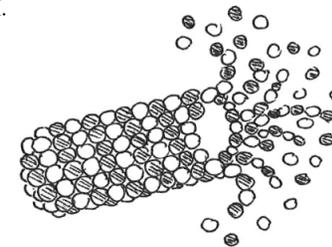


Figure. Microtubules are made up of rings of thirteen tubulin heterodimers which are 8 nm in diameter. The rings of tubulin heterodimers form a cylinder of thirteen protofilaments.

- Calculate the total length of microtubules that could be made from this amount of protein if each heterodimer is approximately 8 nm in diameter. Take into account that microtubules are made up of rings of thirteen tubulin heterodimers. Remember that $1\mu\text{M} = 1\mu\text{mol/liter}$ and that 1 mol contains $6.02 \cdot 10^{23}$ heterodimers.

homework 02 - problem 02

4

Biomechanics: Cell Research and Applications for the Next Decade

DENNIS DISCHER,¹ CHENG DONG,² JEFFREY J. FREDBERG,³ FARSHID GUILAK,⁴ DONALD INGBER,⁵
PAUL JANMEY,¹ ROGER D. KAMM,⁶ GEERT W. SCHMID-SCHÖNBEIN,⁷ and SHELDON WEINBAUM⁸

¹University of Pennsylvania, Philadelphia, PA, USA; ²Pennsylvania State University, University Park, PA, USA; ³Harvard School of Public Health, Boston, MA, USA; ⁴Duke University, Durham, NC, USA; ⁵Harvard Medical School, Boston, MA, USA; ⁶MIT, Boston, MA, USA; ⁷University of California, San Diego, San Diego, CA, USA; and ⁸City College of New York, New York, NY, USA

(Received 10 July 2008; accepted 21 February 2009; published online 4 March 2009)

Abstract—With the recent revolution in Molecular Biology and the deciphering of the Human Genome, understanding of the building blocks that comprise living systems has advanced rapidly. We have yet to understand, however, how the physical forces that animate life affect the synthesis, folding, assembly, and function of these molecular building blocks. We are equally uncertain as to how these building blocks interact dynamically to create coupled regulatory networks from which integrative biological behaviors emerge. Here we review recent advances in the field of biomechanics at the cellular and molecular levels, and set forth challenges confronting the field. Living systems work and move as multi-molecular collectives, and in order to understand key aspects of health and disease we must first be able to explain how physical forces and mechanical structures contribute to the active material properties of living cells and tissues, as well as how these forces impact information processing and cellular decision making. Such insights will no doubt inform basic biology and rational engineering of effective new approaches to clinical therapy.

Keywords—Biomechanics, Cell, Mechanics, Rheology, Signaling, Force, Stress.

emerge through collective interactions within dynamically coupled regulatory networks. Systems Biology presently emphasizes information transfer,¹⁷ but the three-dimensional geometries and physical forces that play so large a role in biological structure and function have yet to be fully taken into account. Indeed, without these biomechanical factors there would be no form, no function, no life.

Most diseases present as a complex genetic profile with multiple changes in molecular expression.^{18,19} Nonetheless, a patient goes to the doctor's office often because of a mechanical defect in a tissue or organ: a new swelling or lump, pain due to nerve compression, stiffness that limits movement, edema caused by a leak of tissue bodily fluids, constricted blood flow or lymph flow, or obstructed airflow that restricts breathing. Cures and remedies are often judged successful by the patient only when such mechanical defects are remedied. In order to understand health-related and disease-related aspects of living systems—all of which work and

homework 02 - problem 03

5

Problem 4 - Final project

Inspired by the recent manuscript on major accomplishments and challenges in cell mechanics,

- Identify a title for your final project.
- Identify five key words for your final project.
- Write a tentative abstract of approximately 150-200 words.

Papers in the past have generally been about 4-6 pages long, two column, with about 3-5 figures and 8-12 references. Here are some examples of individual projects.

- Predicting microtubules structure using molecular dynamics
- The primary cilium: A well-designed fluid flow sensor
- The tensegrity paradigm
- Mechanotransduction in hair cells Translating sound waves into neural signals
- Modeling cell membrane dynamics
- Theoretical and experimental study of the penetration of the cell membrane
- Integrin and its role in mechanotransduction
- Finite element analysis of micropipette aspiration

homework 02 - problem 04

7

Problem 3 - Cell mechanics research

The recent manuscript “*Biomechanics: Cell Research and Applications for the Next Decade*” by Discher, Dong, Fredberg, Guilak, Ingber, Janmey, Kamm, Schmid-Schönbein and Weinbaum discusses the challenges for cell mechanics now and in the future.

- Read the manuscript carefully and summarize it with approximately 150 words.
- The authors have identified ten major accomplishments in cell mechanics. List all the ten accomplishments by title.
- Select your favorite past accomplishment and describe it in less than 100 words.
- The authors have identified seven major challenges for the future. List all the seven future challenges by title.
- Select your favorite future challenge and describe it in less than 100 words.

homework 02 - problem 03

6

Final Project ME339, Fall 2008

CS Simmons

ME339 FINAL PROJECT

THE ROLE OF CELL-CELL JUNCTIONS IN CARDIOMYOCYTE CONTRACTILITY AND MODIFICATIONS NEEDED IN MECHANICAL MODELS

Chelsey S. Simmons

Department of Mechanical Engineering, Stanford University
Stanford, California

ABSTRACT

As adult cardiomyocytes (CMs) cannot recover from damage due to pathologies like myocardial infarction, researchers are looking for ways to culture functional cardiomyocytes to repopulate diseased cardiac tissue. However, as damaged tissue may interfere with cell-cell communication, the role of cell-cell junctions in cardiomyocyte contractility must be considered. This paper examines current literature on CM contractility, evaluating first the role of the cytoskeleton in CM contractility and then the hypothesized role of cell-cell junctions. Additionally, the likelihood of adapting computational models to reflect abnormal conditions of cell-cell junctions is considered. Further examination of contractile force generation with modification of certain cytoskeletal and junctional proteins is recommended to facilitate incorporation into tissue-scale models and to understand the contribution of these various proteins to heart disease.

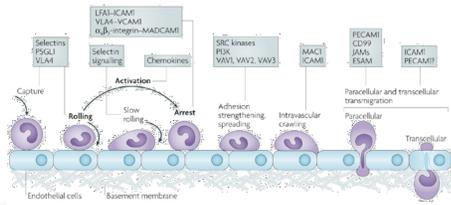
me239 mechanics of the cell - final project

8

MODELS AND MECHANISMS OF LEUKOCYTE EXTRAVASATION

Anusuya M. Ramasubramanian
Department of Biomechanical Engineering, Stanford University
Stanford, CA

ABSTRACT | Leukocyte extravasation is an innate immune system process by which white-blood cells leave the circulatory system and enter the inflamed tissue or site of infection.¹ Although leukocyte migration across the walls of microvessels was observed over 200 years ago, the molecular mechanisms of the migration were not clarified until the early part of the twentieth century.² Today, leukocyte extravasation is thought to occur in a multistep cascade involving the initial capture of the leukocyte by the endothelium, rolling of the leukocyte, slow rolling, arrest, adhesion and spreading of the white blood-cell, intraluminal crawling, and transmigration of the leukocyte across the endothelium.¹ Over the past two decades, a number of molecular and mechanical models have surfaced that view the leukocyte as a shell membrane, a hard sphere defined by Goldman hydrodynamics and liquid droplets. By exploring each leukocyte model in the context of the extravasation process, a greater insight into the importance of integrin and selectin-based mechanotransduction as well as the shear-flow-induced physics of extravasation can be gained. Such understanding can be particularly valuable in the context of drug and cancer development – two radically different processes that mimic leukocyte extravasation.



me239 mechanics of the cell - final project⁹

Mechanisms of Neuron Repair and Stem Cell Neural Differentiation:

Neural Stem Cell Grafts and Current Advances in Spinal Cord Injury Research
José Solomon

Abstract—Neural tissue regeneration and neuronal stem cell differentiation are discussed within the context of spinal cord injuries. The neuron is the fundamental element of the spinal cord and nervous system, and its functioning characteristics as well as the process of synaptic transmission are introduced to understand the impact of spinal cord injury. Three techniques are reviewed that address the issue of neural tissue regrowth from varying perspectives. The methods are interrelated and demonstrate the potential role of neuron stem cell grafting in resolving severe spinal cord injury.

Index Term—Neuron stem cell differentiation, axon ion discharge, synaptic transmission, spinal cord injury.

I. INTRODUCTION

SPINAL cord injuries represent a pervasive and often severe source of neurological trauma. The spinal cord represents the connection bridge between the central nervous system (CNS) and the peripheral nervous system (PNS). The CNS normally refers to the brain and spinal cord itself, where as the PNS is the vast network of nerves that permeates throughout the body. The human spinal cord consists of five main regions, as illustrated in Figure 1(a), each of which has a varying number of vertebrae, shown in Figure 1(b). The vertebrae have a set configuration network of nerves which they protect, and thus the physiological impact of a spinal cord injury is directly correlated to the specific vertebrae affected.

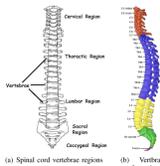


Fig. 1. The spinal cord and the five primary vertebrae regions. [9]

Although there is a certain randomness in the specific effects of a spinal cord injury that stems from an individual's age and

TABLE I
REGIONS OF TRAUMA AND RESULTING NEUROLOGICAL IMPACT

C1- C2	Loss of all motor skills
C4	Minimal upper body control, very weak
C5	Regular shoulder and elbow control, no hand or wrist sensation/control
T2-T9	Good induction control, little to no lower body sensation/control
S1-S2	Some lower body control

the nature of the impact, a general guideline can be arrived at that directly correlates the injured vertebrae with the most likely resulting region of sensory/mobility loss. Some specific correlations are given in Table 1. As shown in the table, injuries in the cervical region normally tend to lead to major mechanical and sensory feedback loss, affecting a larger range of nerve networks than lower spinal regions since it is situated directly at the root of the brain-spinal cord interface. As the region of spinal trauma moves further down the length of the spinal cord, the affected region is reduced in scope.

Spinal cord injuries themselves can be classified into two general categories: complete and incomplete. [9]. In a complete injury, the damage is for the most part permanent and the affected individual usually exhibits little to no recovery of motor or sensory feedback. In incomplete injuries, the effects of the injury tend to be of a temporary nature and the individual may exhibit a full recovery. Two examples of individuals who experienced complete spinal cord injury are presented in Figure 2.



Fig. 2. Examples of complete spinal cord injury

me239 mechanics of the cell - final project¹¹

THE PRIMARY CILIUM: A WELL-DESIGNED FLUID FLOW SENSOR

Bryan C. Petzold
Department of Mechanical Engineering, Stanford University
Stanford, California

The primary cilium is a highly specialized surface projection which extends from the apical surface of almost every vertebrate cell. After its initial discovery over 100 years ago, primary cilia were long overlooked and even purported by some to be extraneous genetic remnants from our evolutionary past. However, in the past decade, a wealth of evidence has begun to accumulate, indicating that cilia in various cell types act not only as mechanical and chemical sensors, but also play important roles in intracellular signaling and cell division¹. Some have even suggested that cilia-related dysfunction may have an important role in modern human epidemics such as obesity, hypertension and diabetes². One such link between cilia-related dysfunction and human disease that has been explored extensively involves the role of the primary cilia of renal epithelial cells as flow sensors. It is believed that a dysfunction in these cilia results in polycystic kidney disease (PKD), the most common inherited disease in the United States, with an estimated 600,000 current cases³. Numerous models have been proposed to explain the mechanotransduction mechanism which allows the primary cilia of renal epithelial cells to detect fluid flow, but many questions remain. Understanding the transduction mechanism and the features of the primary cilium which make it an ideal flow sensor will not only answer many interesting questions in biology and biomechanics, but could aid in the treatment of PKD and other diseases which are caused by cilia-related dysfunction.

INTRODUCTION TO THE PRIMARY CILIUM

The primary cilium is a long, cylindrical, microtubule-based structure which extends from the apical surface of most vertebrate cells, as shown in Figure 1. In general, cells only have a single primary cilium. Referred to as the axoneme, the main structural element of the primary cilium is a collection of nine circumferentially-arranged doublet microtubules encased by membrane continuous with the cell membrane⁴. These doublet microtubules extend from a structure known as a basal body within the cell, which links the base of the primary cilium to the cytoskeleton. The basal body consists of nine triplet microtubules, and two of the microtubules of each triplet form the axoneme of the primary cilium⁵. Further structural support is provided by the transitional fibers (alar shields), which add stability to the complex via attachment to the cell membrane⁶. In conjunction with a terminal plate at the end of the basal body, these transitional fibers also act as a protein filter, only allowing certain proteins to enter and exit the cilium⁷. At the far end of the cilium, the axoneme becomes more variable, but is typically composed of nine single microtubules⁸. Although cilia are not isolated from the cell by a membrane, it seems reasonable to consider them to be organelles due to their unique structure, their extreme location past the cell periphery, and the selectivity to protein movement across their boundaries resulting from the transitional fibers and the terminal plate.

Depending on the species, primary cilia of renal epithelial cells typically vary between 2-20 μm in length *in vivo*⁹. However, lengths up to 30 μm have been observed *in vitro*¹⁰. In addition, studies involving mice renal epithelial cells measured primary cilia 2-3 μm long and 0.2 μm in diameter on average¹¹. Since microtubules have an outer diameter of ~ 30 nm, this relatively small diameter indicates that nearly half the volume of a primary cilium is occupied by the microtubules alone¹².

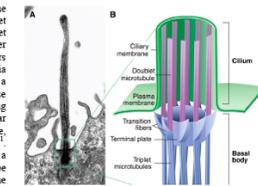
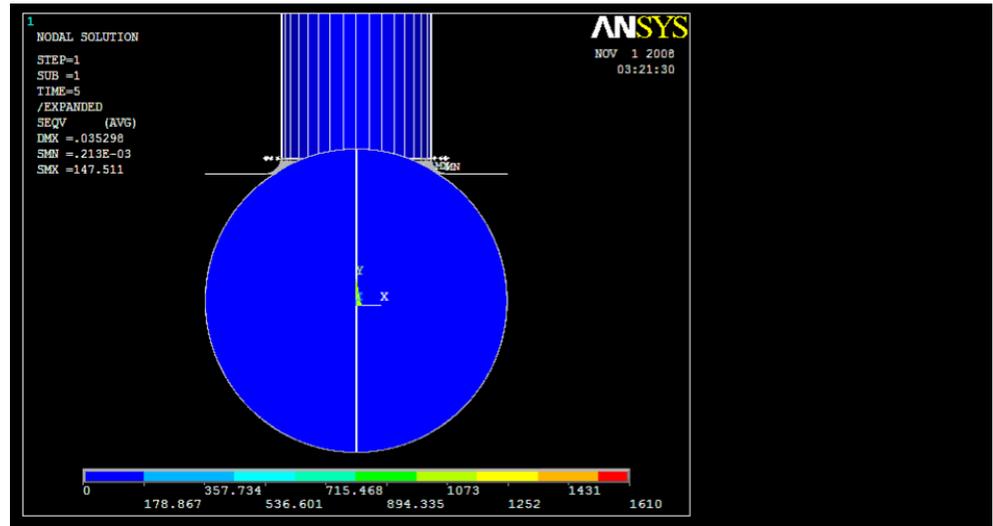


Fig. 1. Primary cilium structure. (A) Electron micrograph of the primary cilium of a canary brain radial glia. (B) Schematic showing structure of the basal body and primary cilium. Adapted from Singla *et al.* (2006)¹³.

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example: finite element simulation of pipette aspiration



zubin huang [2007]

me239 mechanics of the cell - final project¹²

EXPLORING CELLULAR TENSEGRITY: PHYSICAL MODELING AND COMPUTATIONAL SIMULATION



Chun hua Zheng¹, Joseph Doll², Emily Gu³, Elizabeth Hager-Barnard⁴, Zubin Huang¹, AmirAli Kia¹, Monica Ortiz², Bryan Petzold², Takane Usui¹, Ronald Kwon¹, Christopher Jacobs^{1*}, Ellen Kuhl^{1*}
¹Mechanical Engineering, ²Materials Science and Engineering, ³Bioengineering – Stanford University, Stanford, CA

Introduction

Tensegrity

The word **tensegrity** was coined by Buckminster Fuller to describe structures in which continuous tension in their members forms the basis for structural integrity. This structural integrity is created through the dynamic distribution of tensile and compressive forces amongst members.

Fuller most famously demonstrated the concept of tensegrity in architecture through the design of geodesic domes while his student Kenneth Snelson applied the concept of tensegrity to sculpture (Fig. 1). The structural efficiency and dynamic force balance properties of tensegrity have inspired its adoption as a paradigm for analyzing cell structure and mechanics.



Figure 1. Tensegrity structures: Fuller's geodesic dome, Snelson's sculpture "Maze", steel-wood model, and fibroblast cytoskeleton

Cellular Tensegrity Model

The cellular tensegrity model aims to explain intracellular and extracellular processes via a biomechanics viewpoint. The model uses three distinct biopolymers to describe cell cytoskeletal structure. These three biopolymers work in conjunction to provide structure and support for the cell and its internal organelles (Fig. 2).



Figure 2. Structural biopolymers found within the cell cytoskeleton

1. Microfilaments - Thin tension members (5-9 nm diameter), observed as straight and bent in vivo
2. Microtubules - Hollow tubular compression members that are the largest of the three biopolymers (25 nm diameter) and mechanically the stiffest
3. Intermediate filaments - Highly flexible and extensible members that act like guy wires (10 nm diameter), keeping individual microtubules from buckling

Purpose

Motivated by the simple mechanical elegance of the tensegrity model, this study investigates cellular structure by creating physical models and computational models that are analyzed for structural integrity and design efficiency.

The goal of this study is to gain a preliminary understanding of how tensegrity structures physically respond to external loading, use this learning to analyze the response characteristics of different tensegrity forms and to draw parallels between these observations and cell mechanics.

References

[1] Ingher SA 1996. [2] Ingher JCS 2003. [3] Ingher JCS 2003. [4] Chem O&C 1999.

Models

Physical Models

Physical tensegrity models were built using wooden struts and elastic bands (Fig. 3). Varying numbers of compression and tension members were used to achieve different structures with unique mechanical properties.



Figure 3. Physical tensegrity cell models built using compressive wooden struts and tensile elastic bands.

Computational Models

Computational models of were created in Matlab (Fig. 4). Tensile, compressive and shear stiffness, as well as structural efficiency were calculated using nonlinear finite element based analysis (Fig. 5).

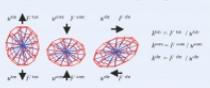
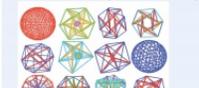


Figure 4. Cell membrane and subcellular structural models.

Results

- A minimum number of filaments is required to establish structural integrity; failure of a non-redundant member results in structural collapse.
- Properly reinforced structures intrinsically recover from large deformations without irreversible damage.
- Altering prestress, compliance and cross-linking significantly impacts cell shape, stiffness and response to load.
- Distinct locations on the surface of the tensegrity cell are more mechano-transductive than others (analogous to cell membrane adhesion receptors known as integrins).

	A	B	C	D
# Microtubules	8	8	9	95
# Filaments	36	36	12	60
Tensile stiffness	0.165 N/mm	1.177 N/mm	1.527 N/mm	0 N/mm
Shear stiffness	0.050 N/mm	0.010 N/mm	0.080 N/mm	0 N/mm

Table 1. Normalized tensile and shear stiffness for tensegrity cells.

Conclusions

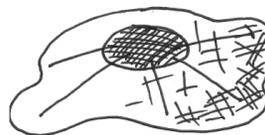
To gain an understanding of the response of tensegrity cell structures, physical and computational models were designed and elaborated in this study. The tensegrity structures varied in stiffness depending on the magnitude of prestress and the geometric interconnections. Observations from the models revealed characteristics that are analogous to those observed in biological cells such as dynamic response to load, the ability to sustain large deformations without failure and existence of mechano-sensitive locales / receptors. Computational simulations enabled a quantitative analysis of the highly nonlinear force network generated within the cell.

from molecular level to cellular level



assuming we know the mechanical properties of the individual filaments, what does that actually tell us about the assembly of filaments that we find in the cell?

- could we then predict the **stiffness of the overall assembly**?
- how does the filament microstructure affect **cytoskeletal properties**?
- how can we calculate the **macroscopic network properties** from the individual microscopic filament properties?



elements of the cytoskeleton
 microtubules
 intermediate filaments
 actin filaments

Figure 4.1: The cytoskeleton provides structural stability and is responsible for forces during cell locomotion. Microtubules are thick hollow cylinders reaching out from the nucleus to the membrane, intermediate filaments can be found anywhere in the cytosol, and actin filaments are usually concentrated close to the cell membrane.

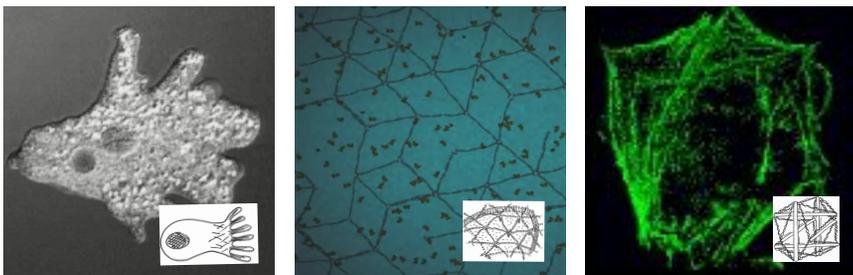
me239 mechanics of the cell - final project¹³

from molecular level to cellular level



three examples

- **fiber bundle model** for filopodia
- **network model** for red blood cell membranes
- **tensegrity model** for generic cell structures



microstructural arrangement of actin

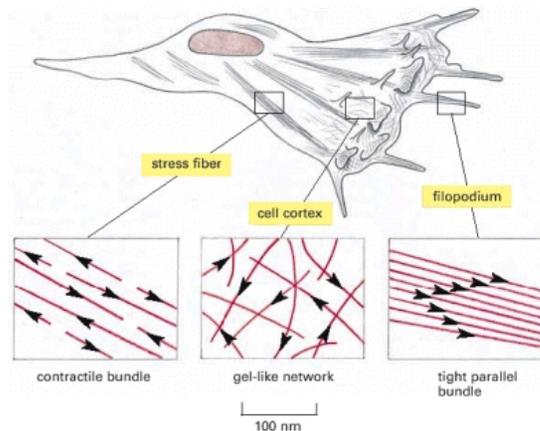


Figure 4.2.1. A crawling cell, drawn to scale, is shown with three areas enlarged to show the arrangement of actin filaments. The actin filaments are shown in red, with arrowheads pointing toward the plus end. Stress fibers are contractile and exert tension. The cell cortex underlies the plasma membrane. Filopodia are spike-like projections of the plasma membrane that allow a cell to explore its environments.

alberts, johnson, lewis, raff, roberts, walter [2002]

4.1 mechanics of the cytoskeleton

4.2 fiber bundle model for filopodia

assembly of crosslinked actin filaments



cell with filopodia



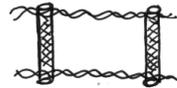
10 μm

filopodia as bundles of actin filaments



2 μm

actin cross-linking through fascin



0.1 μm

Figure 4.3. Bundles of actin filaments tightly crosslinked through fascin are known as filopodia. The mechanical properties of filopodia play an essential role in various different physiological processes including hearing, cell migration, and growth.

filopodia are very thin structures approximately 0.2 μm in diameter. they can easily extend up to 1.5 μm. they typically polymerize and depolymerize at rates of approximately 10 μm/min. the mechanical properties of filopodia play an essential role in various different physiological processes, including hearing, cell migration, and growth. despite their importance to cell function, the structural architecture responsible for their overall mechanical behavior remains largely unknown.

4.2 fiber bundle model for filopodia

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pushing the envelope - critical length

Newton's third law: actio = reactio

$$F_{\text{fil}} \doteq F_{\text{mem}}$$

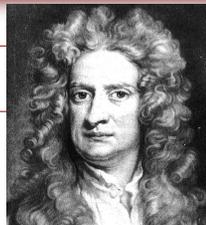
$$F_{\text{fil}} = \frac{\pi^2 EI}{[2L]^2} = \frac{\pi^2 EI}{4L}$$

$$F_{\text{mem}} \approx 5 \sqrt{n} r_{\text{act}} \text{ pN/nm}$$

$$\frac{\pi^2 EI}{4L_{\text{crit}}^2} = 5 \sqrt{n} r_{\text{act}} \frac{\text{pN}}{\text{nm}} \quad \text{thus} \quad L_{\text{crit}} = \frac{\pi}{2} \sqrt{\frac{EI}{5 \sqrt{n} r_{\text{act}} \text{ pN/nm}}}$$

$$E = 1.9 \cdot 10^9 \text{ N/m}^2 = 1.9 \text{ GPa} \quad r_{\text{act}} = 2.5 \quad \text{moment of inertia } I$$

- loose assembly
- tightly crosslinked



4.2 fiber bundle model for filopodia

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pushing the envelope



simplified model for cell locomotion

- protrusion ... polymerization at the leading edge of the cell
- attachment ... formation of focal adhesions to link the cell to the surface
- retraction ... contraction of stress fibers to retract the rear of the cell

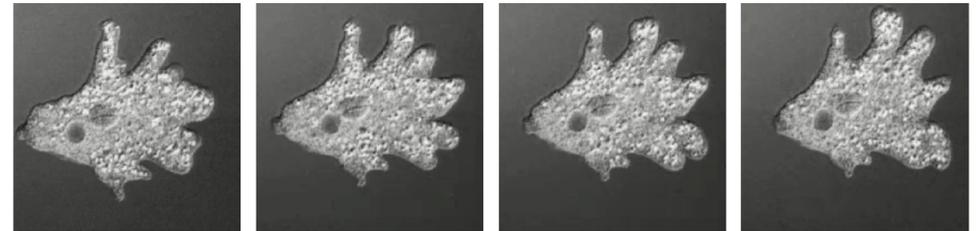


Figure 4.4: Single-celled amoeba crawling around by using actin polymerization to push out pseudopods to explore new territory. Organelles move in complex patterns within the cell, alberts, johnson, lewis, raff, roberts, walter [2002]

4.2 fiber bundle model for filopodia

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case I - loosely assembled actin filaments

$$L_{\text{crit}} = \frac{\pi}{2} \sqrt{\frac{EI}{5 \sqrt{n} r_{\text{act}} \text{ pN/nm}}}$$

moment of inertia I

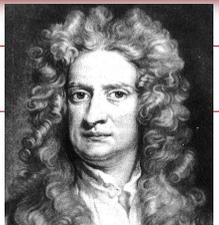
$$I = n I_{\text{act}} \quad \text{with} \quad I_{\text{act}} = \frac{\pi r_{\text{act}}^4}{4}$$

$$E = 1.9 \cdot 10^9 \text{ N/m}^2 = 1.9 \text{ GPa} \quad r_{\text{act}} = 2.5$$

$$L_{\text{crit}} = \frac{\pi}{2} \sqrt{\frac{1.9 \cdot 10^9 \text{ N/m}^2 \cdot n \pi / 4 [3.5 \cdot 10^{-9}]^4 \text{ m}^4}{5 \sqrt{n} 3.5 \cdot 10^{-12} \text{ N}}} \approx 0.17769 \mu\text{m} n^{1/4}$$

$$n = 30 \text{ filaments} \quad L_{\text{crit}} = 0.416 \mu\text{m}$$

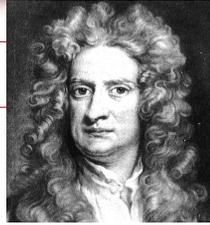
much too low - disagrees with observations of 2 μm



4.2 fiber bundle model for filopodia

20

case II - tightly crosslinked actin filaments



$$L_{\text{crit}} = \frac{\pi}{2} \sqrt{\frac{EI}{5\sqrt{n}r_{\text{act}}pN/\text{nm}}}$$

moment of inertia I

$$I = \frac{\pi r_{\text{fil}}^4}{4} = n^2 \frac{\pi r_{\text{act}}^4}{4} \quad \text{with } r_{\text{fil}} = \sqrt{n} r_{\text{act}}$$

$$E = 1.9 \cdot 10^9 \text{ N/m}^2 = 1.9 \text{ GPa} \quad r_{\text{act}} = 2.5$$

$$L_{\text{crit}} = \frac{\pi}{2} \sqrt{\frac{1.9 \cdot 10^9 \text{ N/m}^2 \cdot n^2 \pi / 4 [3.5 \cdot 10^{-9}]^4 \text{m}^4}{5\sqrt{n} 3.5 \cdot 10^{-12} \text{ N}}} \approx 0.17769 \mu\text{m } n^{3/4}$$

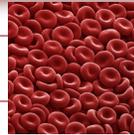
$n = 30$ filaments $L_{\text{crit}} = 2.278 \mu\text{m}$

better model - agrees with observations of $2\mu\text{m}$

4.2 fiber bundle model for filopodia

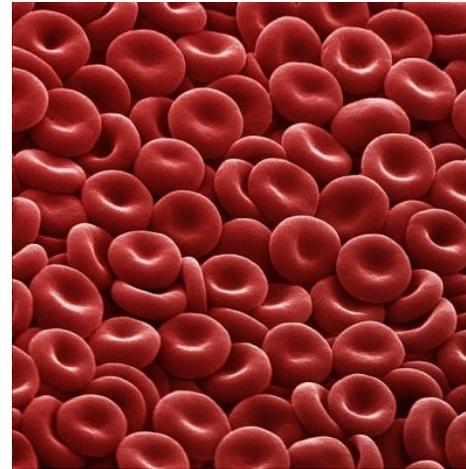
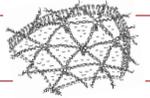
21

red blood cells



human mature red blood cells are **flexible biconcave disks** that lack a cell nucleus and most organelles. typical human erythrocytes have a disk diameter of $6\text{--}8\mu\text{m}$, a thickness of $2\mu\text{m}$, a volume of 90fL , and a surface of $136\mu\text{m}^2$. they can swell to spherical shape of 150fL , without membrane distension. the membrane of the red blood cell plays a key role in regulating **surface deformability, flexibility**, and adhesion to other cells. these functions are highly dependent on its composition. the red blood cell membrane is composed of 3 layers: the glycocalyx on the exterior, which is rich in carbohydrates; the lipid bilayer consisting of lipidic main constituents and transmembrane proteins; and the membrane skeleton, a **structural network of proteins** located **on the inner surface** of the lipid bilayer.

red blood cells



erythrocytes, red blood cells are essential to deliver oxygen to the body via the blood flow through the circulatory system. they take up oxygen in the lungs and release it while squeezing through the body's capillaries. adult humans have about $2\text{--}3 \cdot 10^{13}$, 20-30 trillion, red blood cells comprising about a quarter of the total amount of cells in the human body.

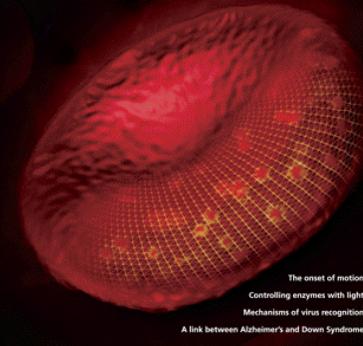
4.3 network model for red blood cells

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red blood cells



Remarkable deformability of red blood cells



Metabolic remodeling of the human red blood cell membrane

YongKeun Park^{1*}, Catherine A. Best¹, Thorsten Auth^{2*}, Nir S. Gov³, Samuel A. Safran¹, Gabriel Popescu⁴, Subra Suresh^{5,6,7}, and Michael S. Feld¹

¹Harvard Spectroscopy Laboratory and ²School of Engineering, Massachusetts Institute of Technology, Cambridge, MA 02139; ³College of Medicine and ⁴Quantitative Light Imaging Laboratory, Department of Electrical and Computer Engineering, Beckman Institute for Advanced Science & Technology, University of Illinois at Urbana-Champaign, Urbana, IL 61801; ⁵Forschungszentrum Jülich, Institute for Solid State Research, 52425 Jülich, Germany, and ⁶Department of Materials and Interfaces and ⁷Department of Chemical Physics, Weizmann Institute of Science, P.O. Box 26, Rehovot 76100, Israel

Edited by Zdeněk P. Bažant, Northwestern University, Evanston, IL, and approved November 25, 2009 (received for review September 18, 2009)

The remarkable deformability of the human red blood cell (RBC) results from the coupled dynamic response of the phospholipid bilayer and the spectrin molecular network. Here we present quantitative connections between spectrin morphology and membrane fluctuations of human RBCs by using dynamic full-field laser interferometry techniques. We present conclusive evidence that the presence of adenosine 5'-triphosphate (ATP) facilitates non-equilibrium dynamic fluctuations in the RBC membrane that are highly correlated with the biconcave shape of RBCs. Spatial analysis of the fluctuations reveals that these non-equilibrium membrane vibrations are enhanced at the scale of spectrin mesh size. Our results indicate that the dynamic remodeling of the coupled membranes powered by ATP results in non-equilibrium membrane fluctuations manifesting from both metabolic and thermal energies and also maintains the biconcave shape of RBCs.

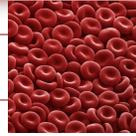
4.3 network model for red blood cells

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4.3 network model for red blood cells

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red blood cells



Metabolic remodeling of the human red blood cell membrane

YongKeun Park^{1,2}, Catherine A. Best¹, Thorsten Auth^{1,2}, Nir S. Gov¹, Samuel A. Safran¹, Gabriel Popescu¹, Subra Suresh^{3,4,1}, and Michael S. Feld¹

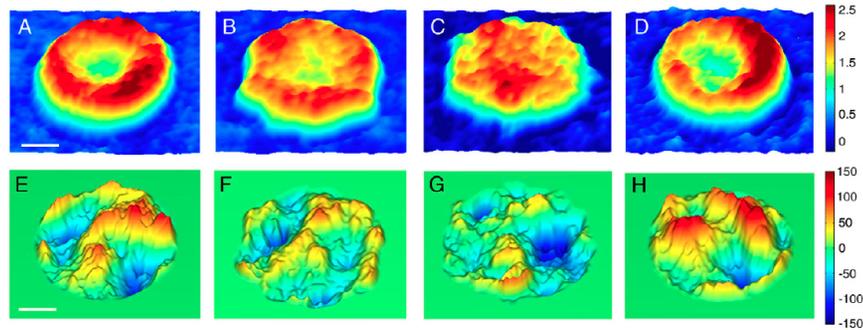
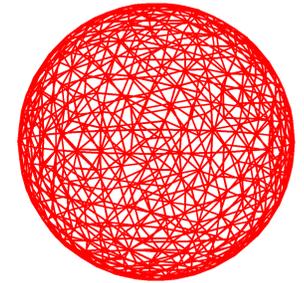
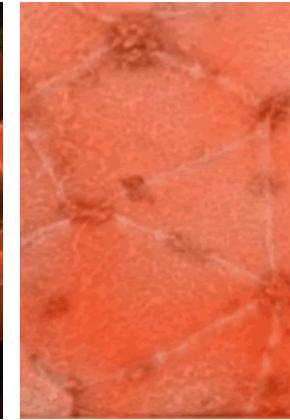
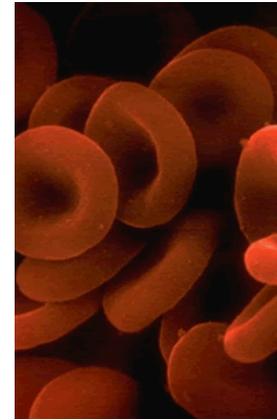
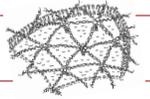


Fig. 1. Effects of ATP on morphology and dynamic fluctuation in RBC membrane. Topography of a healthy RBC, (A) of an ATP-depleted RBC (irreversible-ATP group), (B) of an ATP-depleted RBC (metabolic-ATP group), (C), and of a RBC with recovered ATP level (+ATP group), (D) resp. (E-H) Instantaneous displacement maps of membrane fluctuation in the Fig. 1A-D, resp. The scale bar is 2 μm . The colorbar scales are in μm and nm, resp.

red blood cells



the human red blood cell membrane skeleton is a network of roughly 33,000 protein hexagons that looks like a microscopic geodesic dome

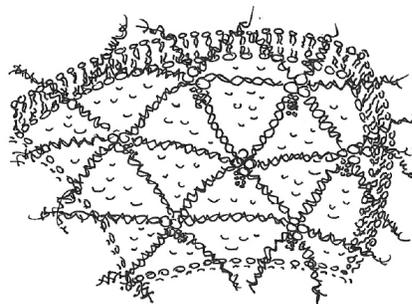
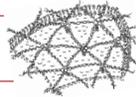
4.3 network model for red blood cells

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4.3 network model for red blood cells

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network model for red blood cells



outer membrane surface
phospholipid bilayer
inner membrane surface

network of spectrin tetramers
crosslinked through actin

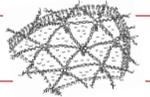
inner membrane surface

Figure 4.6: Microstructural architecture of the cell membrane of a red blood cell. A six-fold connected network of spectrin tetramers which are crosslinked through short actin filaments, anchored to the phospholipid bilayer, provides structural support to the inner cell membrane.

4.3 network model for red blood cells

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homogenization - hill-mandel condition



aim. to determine the overall material properties κ and μ of the network of spectrin chains in terms of the spectrin chain stiffness k

ENERGY APPROACH

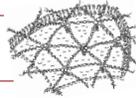
$$W^{\text{mac}} \doteq W^{\text{mic}}$$

It has been shown how the central problem is reducible to the calculation of average stress or strain in one or other phase. A more versatile approach stems directly from classical theorems in elasticity and focusses attention on strain energies.

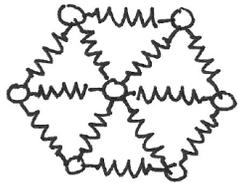
hill, r. elastic properties of reinforced solids: some theoretical principles, journal of the mechanics and physics of solids, 1963, 11:357-372.

4.3 network model for red blood cells

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six-fold connected network



four-fold connected network

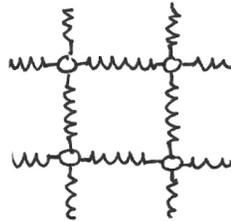
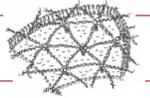


Figure 4.8: Microstructural architecture of a six-fold and four-fold connected network. The theory of homogenization helps to explain why nature prefers a six-fold connected network geometry.



free energy W^{spr} of a single spring

$$W^{\text{spr}} = \frac{1}{2} k \delta^2 = \frac{1}{2} k [l - l_0]^2 \quad \text{where} \quad \delta = l - l_0$$

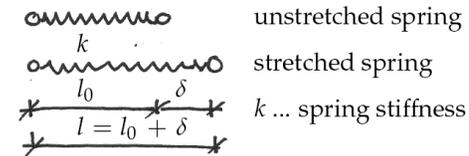
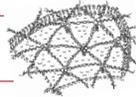


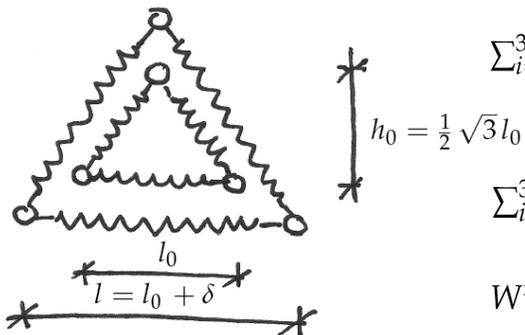
Figure 4.7: Spectrin can be modeled as Gaussian chain which we can conceptually replace by an equivalent linear entropic spring with a spring stiffness of $k = 3 k T N / L$. The strain energy of this spring can then be expressed as $W^{\text{spr}} = \frac{1}{2} k \delta^2$.

4.3 network model for red blood cells

4.3 network model for red blood cells



extension

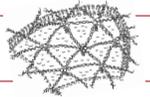


$$W^{\text{mic}} = \frac{\sum_{i=1}^3 W_i^{\text{spr}}}{\sum_{i=1}^3 A_i^{\text{spr}}}$$

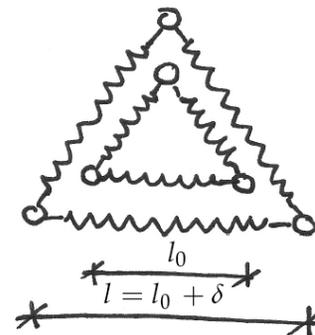
$$\sum_{i=1}^3 W_i^{\text{spr}} = 3 W^{\text{spr}} = 3 \left[\frac{1}{2} k \delta^2 \right]$$

$$\sum_{i=1}^3 A_i^{\text{spr}} = 3 A^{\text{spr}} = \frac{1}{2} \sqrt{3} l_0^2$$

$$W^{\text{mic}} = \frac{3 \frac{1}{2} k \delta^2}{\frac{1}{2} \sqrt{3} l_0^2} = \sqrt{3} k \left[\frac{\delta}{l_0} \right]^2$$



extension



$$W^{\text{mac}} = \frac{1}{2} \kappa [\epsilon_{xx} + \epsilon_{yy}]^2 + \frac{1}{2} \mu [\epsilon_{xx} - \epsilon_{yy}]^2 + 2 \mu \epsilon_{xy}^2$$

micro-to-macro kinematics
 $\epsilon_{xx} = \epsilon_{yy} = \delta / l_0 \quad \epsilon_{xy} = 0$

$$W^{\text{mac}} \doteq W^{\text{mic}}$$

$$\frac{1}{2} \kappa \left[\frac{\delta}{l_0} + \frac{\delta}{l_0} \right]^2 = \sqrt{3} k \left[\frac{\delta}{l_0} \right]^2$$

$$\underline{\underline{\kappa = \frac{1}{2} \sqrt{3} k}}$$

4.3 network model for red blood cells

4.3 network model for red blood cells

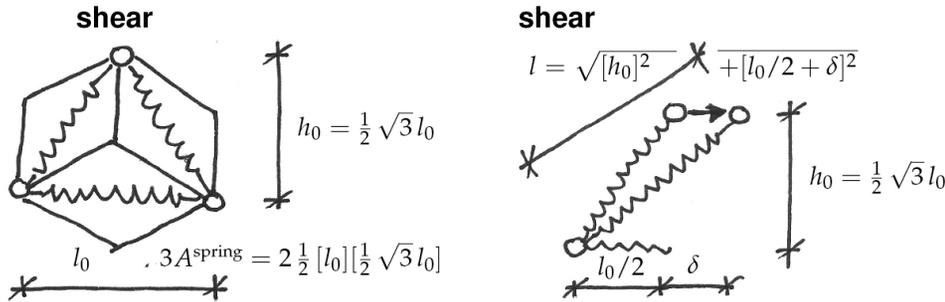
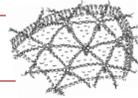
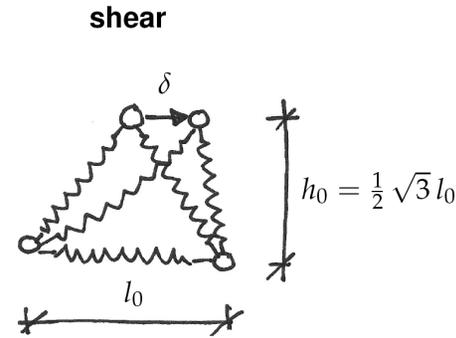
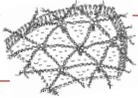


Figure 4.10: Illustration of representative area of three chains $3A^{\text{spring}} = \sqrt{3}/2 l_0^2$ in six-fold connected network model, left. Illustration of deformed spring length $l = l_0 + \delta/2$ in six-fold connected network model subjected to shear, right.

4.3 network model for red blood cells



$$W^{\text{mic}} = \frac{\sum_{i=1}^3 W_i^{\text{spr}}}{\sum_{i=1}^3 A_i^{\text{spr}}}$$

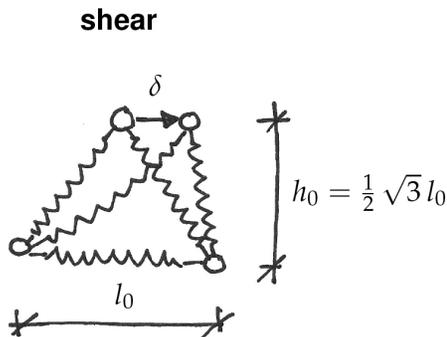
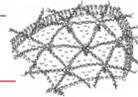
$$\sum_{i=1}^3 W_i^{\text{spr}} = \frac{1}{2} k \left[+\frac{1}{2} \delta \right]^2 + \frac{1}{2} k \left[-\frac{1}{2} \delta \right]^2 + \underbrace{0}_{\text{lower spring}} = \frac{1}{4} k \delta^2$$

right spring left spring

$$\sum_{i=1}^3 A_i^{\text{spr}} = 3 A^{\text{spr}} = \frac{1}{2} \sqrt{3} l_0^2$$

$$W^{\text{mic}} = \frac{\frac{1}{4} k \delta^2}{\frac{1}{2} \sqrt{3} l_0^2} = \frac{\sqrt{3}}{6} k \left[\frac{\delta}{l_0} \right]^2$$

4.3 network model for red blood cells



$$W^{\text{mac}} = \frac{1}{2} \kappa [\epsilon_{xx} + \epsilon_{yy}]^2 + \frac{1}{2} \mu [\epsilon_{xx} - \epsilon_{yy}]^2 + 2 \mu \epsilon_{xy}^2$$

micro-to-macro kinematics

$$\epsilon_{xx} = 0 \quad \epsilon_{yy} = 0$$

$$\epsilon_{xy} = \frac{1}{2} \left[\frac{\delta}{\frac{1}{2} \sqrt{3} l_0} + 0 \right] = \frac{1}{\sqrt{3}} \frac{\delta}{l_0}$$

$$W^{\text{mac}} \doteq W^{\text{mic}}$$

$$2 \mu \left[\frac{1}{\sqrt{3}} \frac{\delta}{l_0} \right]^2 = \frac{\sqrt{3}}{6} k \left[\frac{\delta}{l_0} \right]^2$$

$$\underline{\underline{\mu = \frac{1}{4} \sqrt{3} k}}$$

4.3 network model for red blood cells

heterogeneous fluctuating rod models for unfolded proteins and application to fibrin networks

biofilaments, such as, actin and DNA, have for long been modeled as thermally fluctuating elastic rods with homogeneous material properties. such models are adequate if the length scale of the filaments being studied is much larger than the scale of the heterogeneity. however, advanced single molecule experimental techniques have now made it possible to probe the properties of biomolecules at the scale of a few nanometers. the data emerging from these experiments ought to be greeted with appropriately detailed models. in this presentation we study the mechanics of a thermally fluctuating elastic rod whose moduli are a function of position. such a rod can be used as a model for DNA whose sequence specific properties are known or for a protein oligomer in an AFM where some of the monomers might be unfolded. the mechanics of these rods is understood by first evaluating a partition function through path integral techniques. similar methods can also be applied to heterogeneous networks of filaments. In this presentation we will show how protein unfolding at the level of a single filament can determine the macroscopic mechanical behavior of a network.

mechanics & computation, me395, thu 02/03/11, 4:15-5:25, 320-105

heterogeneous fluctuating rod models