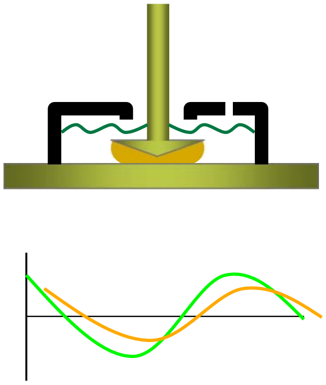
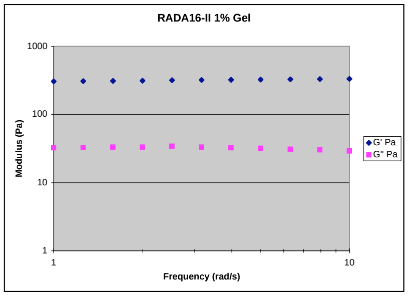
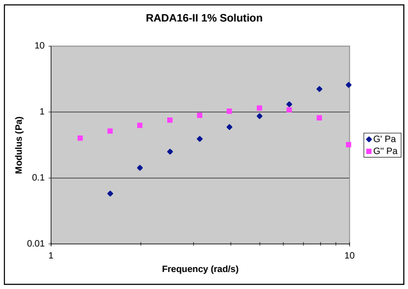
Rheological Measurements: Viscoelastic Properties



- Cone-plate rheometer oscillated over a range of frequencies (ω)
- Imposed: sinusoidal torque (τ)
- Measured: sinusoidal strain (ε)





Self-assembling peptide.

Gel:

•G’ >> G”

•Little frequency dependence

Viscous solution:

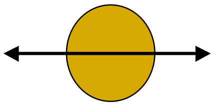
•Lower values of moduli

•Frequency dependent

G. Kim, 2003

Interpretation of displacement (x) of a microsphere (radius a) inside the cytoplasm.

Assumes elastic and viscous contributions are additive as in a Voigt model. ($G'' = \omega\mu$)



$$F(t) = 6\pi a\left(G'x + \mu\frac{dx}{dt}\right)$$

Sphere in an infinite elastic medium

Sphere in an infinite Newtonian fluid

$$G' = \frac{F_0\cos\varphi}{6\pi a x_0} \qquad G'' = \frac{F_0\sin\varphi}{6\pi a x_0}$$

Smooth muscle cells -- with and without activation. Obtained using magnetic twisting cytometry.

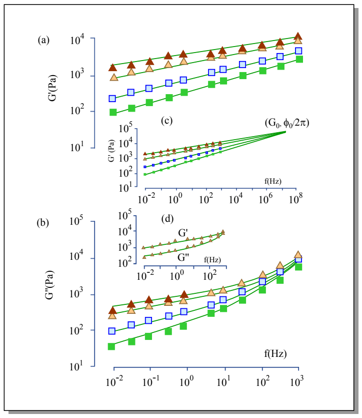


Figure by MIT OCW.

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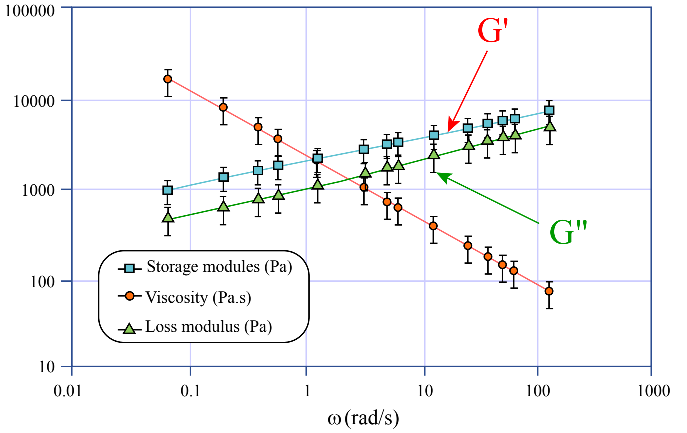
Storage (a) and loss (b) moduli plotted over 5 decades of frequency for smooth muscle cells under control conditions (solid squares), after treatment for 10 minutes with histamine to produce smooth muscle activation (open squares), an agent to eliminate baseline tone, DBcAMP (solid triangles), or cytochalasin D, to disrupt actin filaments (open triangles). (c) shows the extrapolation of the data for illustrating the intersection at high frequency, and (d) directly compares the data from (a) and (b) under control conditions. Solid lines are the fit to the data by eqn. (68) with G_0=53.6 kPa and ω_0=2.5x10^8 rad/s . (Reproduced from Fabry, et al., 2001)

$$G^* = G_0\left(\frac{\omega}{\omega_0}\right)^{x-1}(1+i\overline{\eta})\Gamma(2-x)\cos\left[\frac{\pi}{2}(x-1)\right] + i\omega\mu$$

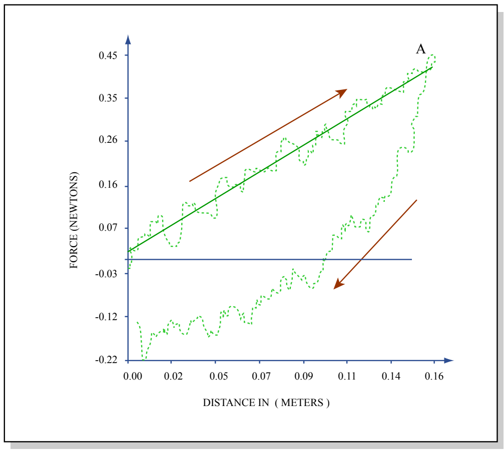
Oscillatory shear tests. Storage and loss moduli (in Pa).



Shear oscillation tests of pig kidney at strain 0.2%, which specifies the components of the dynamic modulus in terms of frequency. The standard deviations are also shown as error bars

Figure by MIT OCW.

Experimental measurements made by cell poking (lymphocyte) (Zahalak, et al., 1990)



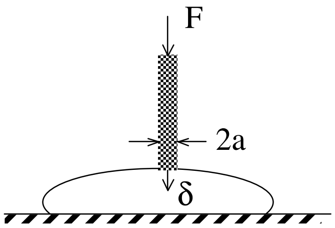


Figure by MIT OCW.

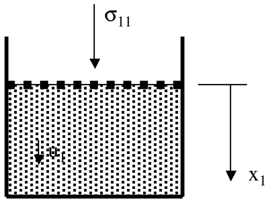
Terminology for poroelasticity

$$H = 2G + \lambda = confined\_compression\_\text{mod}\,ulus$$
$$\vec{v}_f = local\_fluid\_velocity$$
$$\vec{v}_s = local\_solid\_velocity = \frac{\partial \vec{u}}{\partial t}$$
$$\vec{U} = mean\_fluid\_speed\_(relative\_to\_solid\_phase)$$
$$A_f = fluid\_area$$
$$A_s = solid\_area$$
$$\phi = porosity = \frac{A_f}{A_f + A_s}$$
$$k = hydraulic\_permeability$$

Poroelastic materials

Governing equations:

1. Constitutive law
$$\sigma_{ij}^{tot} = 2G\varepsilon_{ij} + \lambda\left(\varepsilon_{11} + \varepsilon_{22} + \varepsilon_{33}\right)\delta_{ij} - p\delta_{ij}$$
2. Fluid-solid viscous interactions (Darcy's Law)
$$\vec{U} = -k\nabla p$$
3. Conservation of mass
$$\vec{U} = \phi\left(\vec{v}_f - \vec{v}_s\right) = \phi\vec{v}_{rel} \qquad \vec{v}_s = \frac{\partial \vec{u}}{\partial t}$$
4. Conservation of momentum
$$\nabla \cdot \bar{\bar{\sigma}}^{tot} = 0$$



1D forms

$$\sigma_{11}^{tot} = \left(2G + \lambda\right)\varepsilon_{11} - p$$
$$U_1 = -k\frac{\partial p}{\partial x_1}$$
$$U_1 = -\frac{\partial u_1}{\partial t} + U_0$$
$$\frac{\partial \sigma_{11}^{tot}}{\partial x_1} = 0$$

$$\boxed{\frac{\partial u_1}{\partial t} - U_0 = Hk\frac{\partial^2 u_1}{\partial x_1^2}}$$

Poroelasticity -- confined compression

Impose displacements at boundaries:

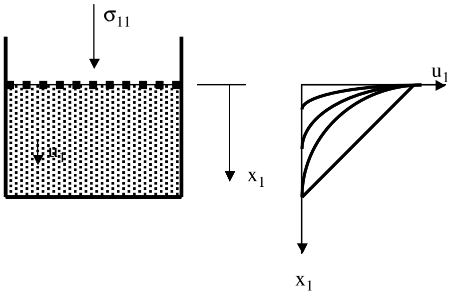
$u_1(x_1, t=0) = 0$

$u_1(x_1=L, t>0) = 0$

$u_1(x_1=0, t>0) = u_0$

$U_0 = 0$

$$\frac{\partial u_1}{\partial t} = Hk \frac{\partial^2 u_1}{\partial x_1}$$



Characteristic time ~ L^2/Hk

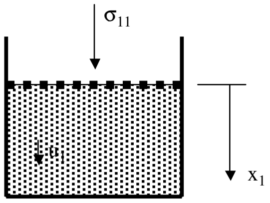
Solution (Fourier series)

$$u_1(x_1,t) = u_0\left(1-\frac{x_1}{L}\right) - \sum_n A_n \sin\left(\frac{n\pi x_1}{L}\right)\exp\left(-\frac{t}{\tau_n}\right)$$

$$\tau_n = \frac{L^2}{n^2\pi^2 Hk}$$

Stress relaxation results for 4 tumor types

Confined compression experiments

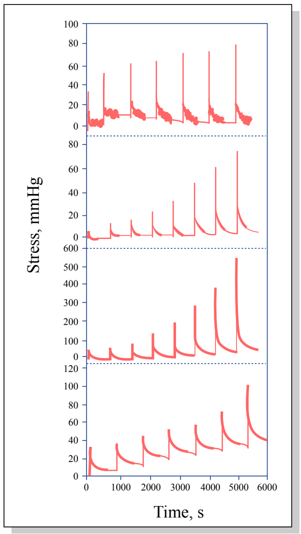


[CANCER RESEARCH 60, 2497–2503, May 1, 2000]

Role of Extracellular Matrix Assembly in Interstitial Transport in Solid Tumors[1]

Paolo A. Netti,[2] David A. Berk,[3] Melody A. Swartz,[4] Alan J. Grodzinsky, and Rakesh K. Jain[5]

Steele Laboratory for Tumor Biology, Department of Radiation Oncology, Massachusetts General Hospital and Harvard Medical School, Boston, Massachusetts 02114 [P. A. N., D. A. B., M. A. S., R. K. J.], and Department of Electrical Engineering, Massachusetts Institute of Technology, Cambridge, Massachusetts 02139 [A. J. G.]



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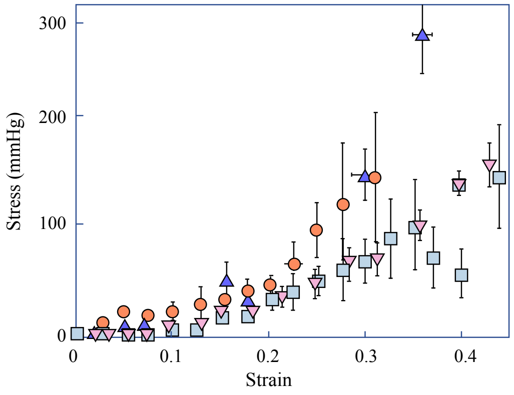
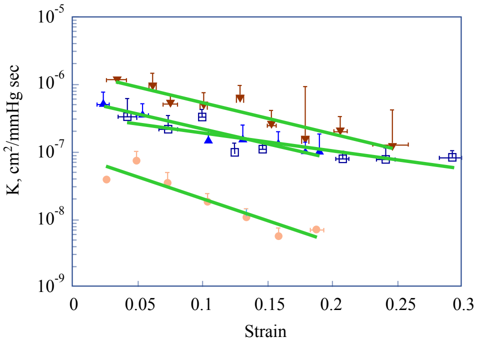
Equilibrium stress-strain curves for 4 tumor types



Used to calculate confined compression modulus, H. Assumed linear for strains < 0.1.

Figure by MIT OCW.

Hydraulic permeability as a function of tissue deformation



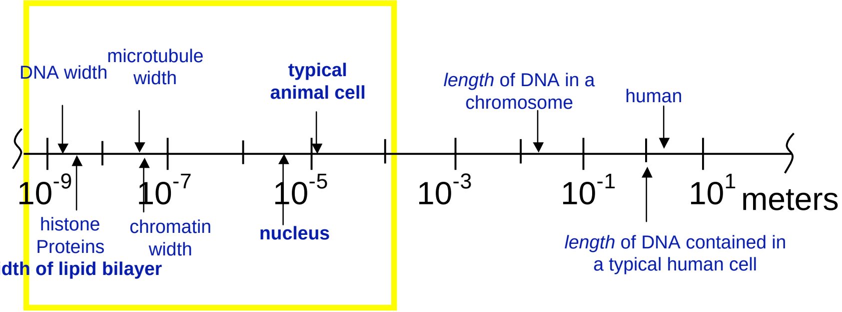
Four different tumor tissues.

Obtained from stress relaxation experiments in confined compression.

10^{-7} cm^2 / (mmHg · s) = $1.4\text{x}10^{-13}m^4$ / (N · s)

Figure by MIT OCW.

Typical Length Scales in Biology



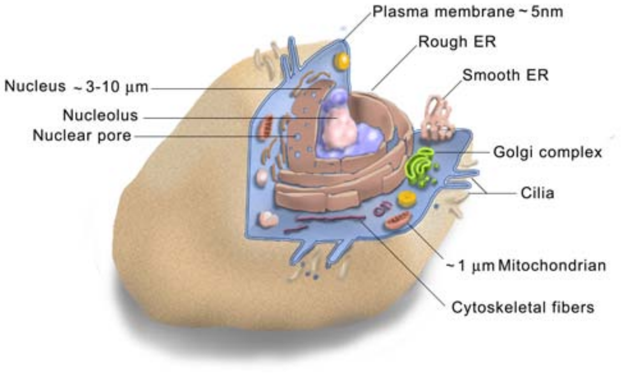
DNA width
microtubule width
typical animal cell
length of DNA in a chromosome
human

10^{-9} 10^{-7} 10^{-5} 10^{-3} 10^{-1} 10^{1} meters

histone Proteins
chromatin width
nucleus
Width of lipid bilayer
length of DNA contained in a typical human cell

Similar spectra exist in time scales or energy scales.

Typical Eukaryotic Cell



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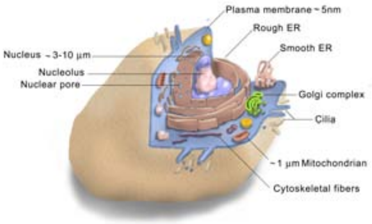
$1\ \mu m = 10^{-6}$ m
1 nm $= 10^{-9}$ m
1 Å $= 10^{-10}$m

Plasma Membrane

Plasma Membrane



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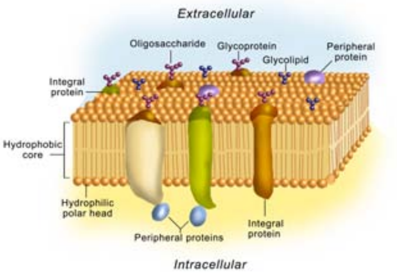
2-D Elastic Plate



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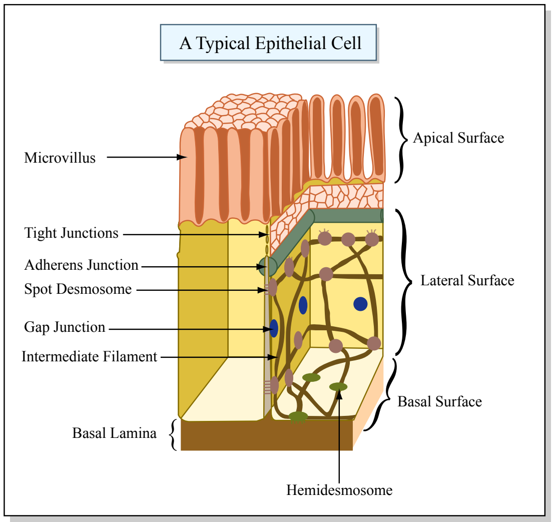

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Cell-cell junctions

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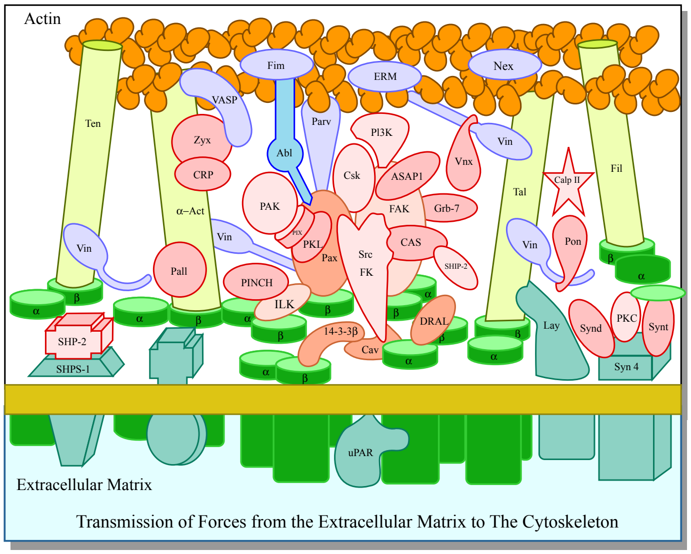
Fig. 2.1.10. Schematic showing the different types of cell junctions present in an epithelial cell as found in the small intestine. Tight junctions near the apical surface essentially prevent the passage of all molecules. The spot desmosomes and adherens junctions provide for cell-cell anchoring, and the hemidesmosomes for anchoring to the basal lamina. Gap junctions provide a means for communication between neighboring cells. [Reproduced from Lodish et al., Molecular Cell Biology, 2000.]

Detailed structure of a focal adhesion



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Staining of actin and nuclei in fibroblast cells

(J. Lammerding)

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The cytoskeleton as a homogeneous, isotropic, elastic material.

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The cytoskeleton of a macrophage lamellipodium as seen by electron microscopy. The fibrous structure is mainly comprised of actin filaments. (John Hartiwick, http://expmed.bwh.harvard.edu)

Forces and deformations are transmitted throughout the cell by the cytoskeleton

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Fibroblast with fluorescent mitochondria forced by a magnetic bead

D. Ingber, P. LeDuc

A variety of methods have been used to probe cell mechanics

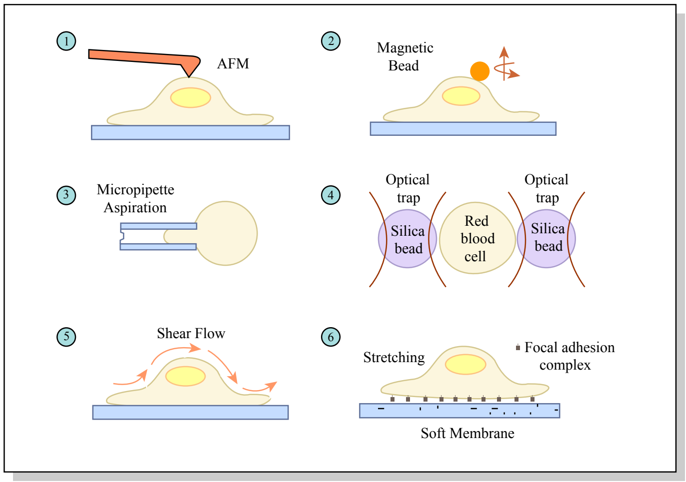
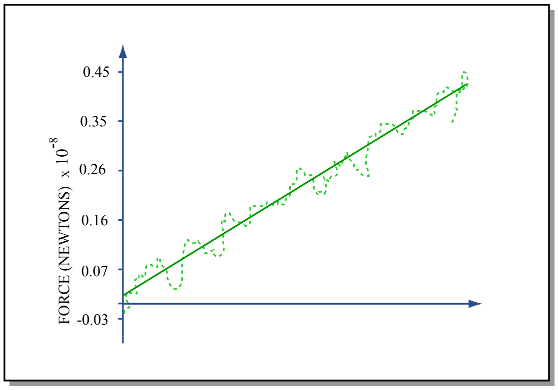


Figure by MIT OCW.

Bao & Suresh, 2003

Are cells linear elastic materials?

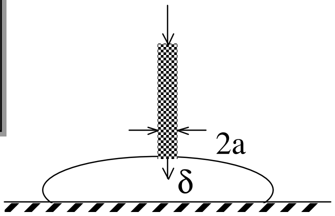


Neutrophils

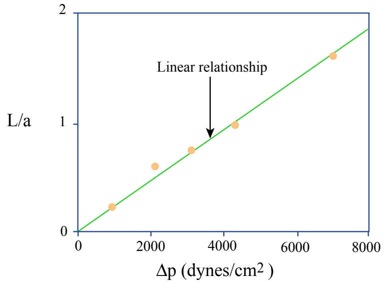
Figure by MIT OCW.

(Zahalak et al., 1990)

One example:
Indentation
Experiments



Micropipette aspiration



Experimental result obtained on a from an endothelial cell micropipette experiment.
Data from Theret et al., 1988.

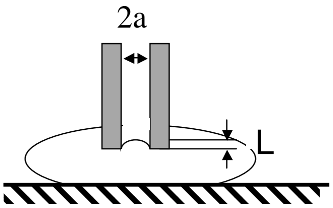


Figure by MIT OCW.

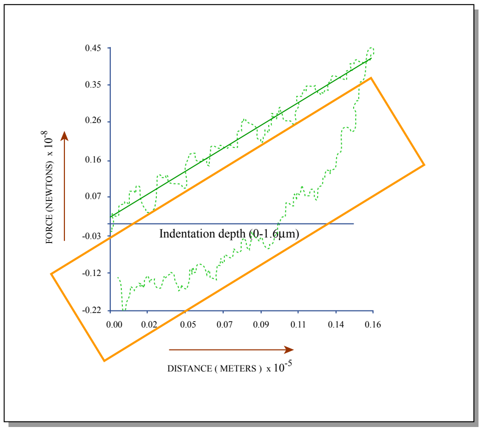
Measurements of cell shear modulus found in the literature.

Values range over 3-4 orders of magnitude!

Cell type	*Measurement method*	*Shear modulus (Pa)*	*Reference*
lymphocyte	poking	300	Zahalak et al.
lymphocyte (activated)	poking	700 - 1100	Zahalak et al.
neutrophil (activated)	poking		Zahalak et al.
neutrophil	poking	110	Zahalak et al.
NIH 3T3 fibrobalst	magnetic tweezers	20,000-40,000	Bausch et al.
NIH 3T3 fibroblast	AFM	4,000-100,000	Haga et al.
J774 mouse macrophage	magnetic tweezers	343	Bausch et al.
3T3 and NRK fibroblast	AFM	1,000-10,000	Rotsch & Rademacher
mouse fibroblast	poking	1600 (E)	Peterson et al.
endothelial cell	aspiration	40-50	Theret et al.
bovine endothelial cell	indentation	400-600	Sato et al.
porcine endothelial cell	aspiration	75 (E)	Sato et al.
endothelial cell	magnetic twisting cytometry (MTC)	2.2 (round) (E) 4.5 (spread) (E)	Wang et al.
bovine endothelial cell	MTC	3-7.5	Wang & Stamenovic
human chondrocytes	aspiration	330	Trickey et al.
smooth muscle cell	MTC	11.5 (E)	Stamenovic & Coughlin
COS7 (kidney epithelial)	laser tracking microrheology	33-82 ($\lvert G^* \rvert$)	Yamada, et al.

Elastic or viscoelastic??

Cells are viscoelastic



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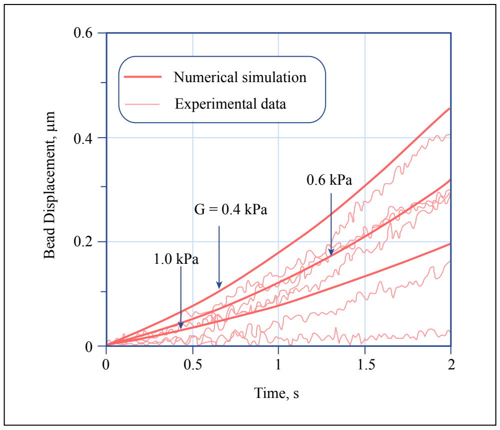
Micropipette Aspiration

Figure by MIT OCW.

Indentation
(Zahalak et al., 1990)

Comparison between experiments and Maxwell fluid model: Ramped force application



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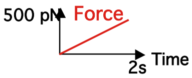
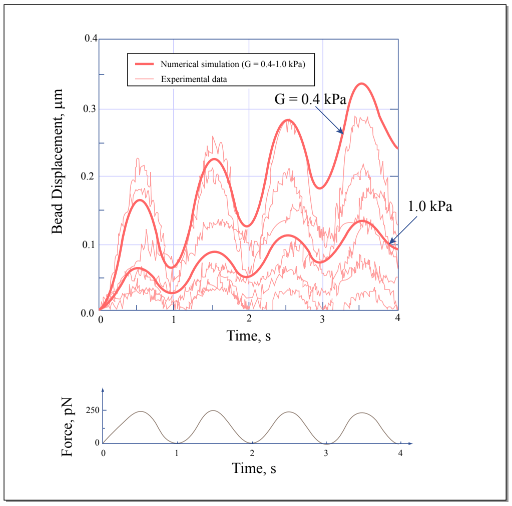


- Experiments suggest a shear modulus of about 1.0 kPa for NIH T3T fibroblasts

Sinusoidal Forcing



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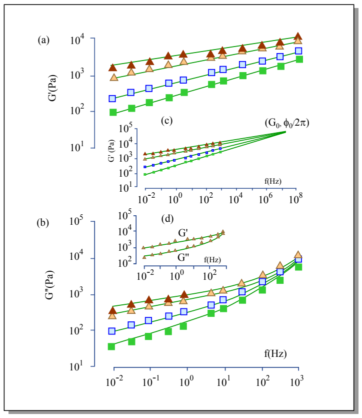
Cells appear to behave as a Maxwell viscoelastic material with characteristic time constant of ~1s

Magnetic Twisting Cytometry



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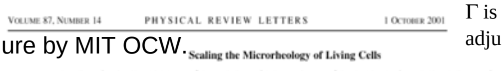


VOLUME 87, NUMBER 14 PHYSICAL REVIEW LETTERS 1 OCTOBER 2001

Scaling the Microrheology of Living Cells

Ben Fabry,[1,*] Geoffrey N. Maksym,[2] James P. Butler,[1] Michael Glogauer,[3] Daniel Navajas,[4] and Jeffrey J. Fredberg[1]

Testing over a wider range of frequencies illustrates a more complex, but still viscoelastic, behavior.

$$G^* = G' + iG'' = \alpha \frac{\tilde{T}(t)}{\tilde{\delta}(t)}$$

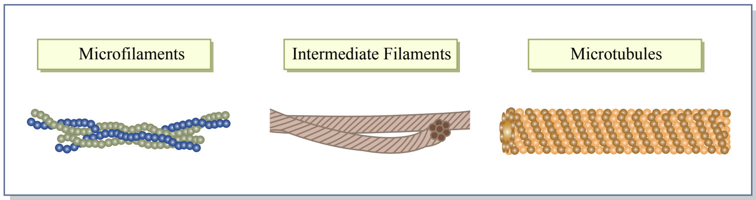
α is a geometry-dependent prefactor determined from finite element analysis

$$G^* = G_0 \left(\frac{\omega}{\omega_0} \right)^{x-1} (1 + i\overline{\eta}) \Gamma(2-x) \cos\left[\frac{\pi}{2}(x-1) \right] + i\omega\mu$$

Γ is the Gamma-function; G_0, Φ_0 and x are adjustable parameters

Consistent with models for soft, glassy materials

Viscoelastic or Poroelastic??



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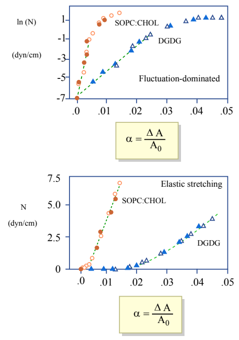
Cells have a porous, fluid-filled matrix.

Are thermal (Brownian) effects important?

Lipid vesicles exhibit a "fluctuation-dominated" regime and an "elastic" regime when inflated from zero tension. (Evans and Rawicz, PRL, 1990)



Measurements of membrane tension and fractional area dilation for two vesicles with different lipid compositions.
(a) A fluctuation-dominated regime appears at low tensions:
a slope of $8\pi k_c/kT$.
(b) Crossover to direct expansion at high tension:
a slope of $\bar{K}$.
(open symbols, ascending pressure; solid symbols, descending pressure.)

Molecular, Cell and Tissue Biomechanics
Fall Term, 2006

Figure by MIT OCW.

Homogeneous, isotropic??

Mapping cell surface elasticity using AFM

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Elasticity mapping in a fibroblast (NIH3T3) cell using atomic force microscopy. The elasticity map (A) shows gross differences with the lowest values corresponding to the nucleus (N), and a small pocket (arrow) low in actin content. The height of the cell from the substrate is shown in (B). The two lower images are stained for actin (C) and microtubules (D) from the same cell. (Reproduced from Haga, et al., 2000)

Ultramicroscopy 82 (2000) 253–258

www.elsevier.nl/locate/ultramic

Elasticity mapping of living fibroblasts by AFM and immunofluorescence observation of the cytoskeleton

Hisashi Haga[a,*], Shigeo Sasaki[a], Kazushige Kawabata[a], Etsuro Ito[b], Tatsuo Ushiki[c], Takashi Sambongi[a]

Cells are inhomogeneous and anisotropic.

Molecular, Cell and Tissue Biomechanics
Fall Term, 2006

What else needs to be considered??

Cells are dynamic.
Properties are constantly changing.

Cell Motility

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Fluorescently tagged actin

- Actin is a polymer
- The cytoskeleton is active
- Coordinated processes:

adhesion, (de-) polymerization

Active Cell Contraction

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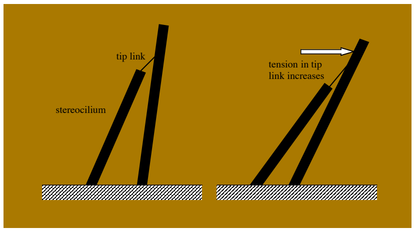
Cardiac myocyte (Jan Lammerding)

Cells can sense and respond to mechanical stimuli

Mechanotransduction: Hair cell stimulation



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SEM of the stereocilia
on the surface of a
single hair cell
(Hudspeth)

Tension in the tip
link activates a
stretch-activated
ion channel, leading
to intracellular
calcium ion
fluctuations.