

trophic factor: pharma-
neurology, 1994. 35(2).

Sciences USA, 1995, 92.

ship between epidermal
Journal of Biological

5

Cell and Tissue Mechanics

Since things in motion sooner catch the eye
Than what not stirs.

William Shakespeare, *Troilus and Cressida*

Mechanics is the branch of physics that is concerned with the action of forces on matter. Tissue engineers can encounter mechanics in various settings. Often, the mechanical properties of replacement biological materials must replicate the normal tissue: for example, there is limited use for a tissue-engineered bone that cannot support the load encountered by its natural counterpart. In addition, the mechanical properties of cells and cell-cell adhesions can determine the architecture of a tissue during development. This phenomenon can sometimes be exploited, since the final form of engineered tissues depends on the forces encountered during assembly and maturation. Finally, the mechanics of individual cells—and the molecular interactions that restrain cells—are important determinants of cell growth, movement, and function within an organism.

This chapter introduces the basic elements of mechanics applied to biological systems. Some examples of biomechanical principles that appear to be important for tissue engineering are also provided. For further reading, comprehensive treatments of various aspects of biomechanics are also available [1–3].

5.1 Elementary Solid Mechanics

5.1.1 Elastic Deformation and Young's Modulus

Consider an elongated object—for example, a segment of a biological tissue or a synthetic biomaterial—that is fixed at one end and suddenly exposed to a constant applied load (Figure 5.1). The material will change or deform in response to the load. For some materials, the deformation is instantaneous and, under conditions of low loading, deformation varies linearly with the magnitude of the applied force:

$$\sigma \left[\equiv \frac{F}{A} \right] = E\epsilon \quad (5-1)$$

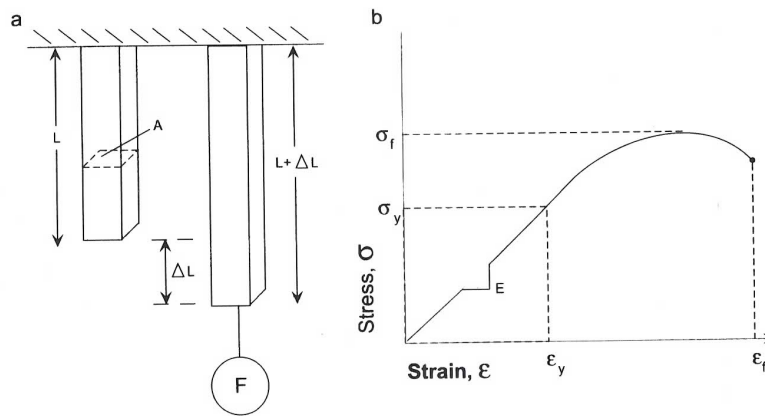


Figure 5.1. Typical characteristics of an elastic material. (a) A uniform material is subjected to an applied load. (b) Typical response of the material to tensile loads of increasing magnitude.

where σ is the applied stress and ϵ is the resulting strain. This relationship is called Hooke's law, after the British physicist Robert Hooke, and it describes the behavior of many elastic materials, such as springs, which deform linearly upon loading and recover their original shape upon removal of the load. The Young's modulus or tensile elastic modulus, E , is a property of the material; some typical values are provided in Table 5.1. Not all elastic materials obey Hooke's law (for example, rubber does not); some materials will recover their original shape, but strain is not linearly related to stress. Fortunately, many interesting materials do follow Equation 5-1, particularly if the deformations are small.

Materials become irreversibly altered if they are deformed beyond a critical yield strain, or elastic limit, ϵ_y . (Many materials obey Hooke's law for all strains less than the elastic limit; other materials obey Hooke's law over a more limited range, called the proportional limit, and continue to deform elastically, but not linearly, up to the yield stress.) This state occurs at a characteristic yield stress σ_y . Further strain of the material results in plastic (rather than elastic) deformation; an irreversible change in the material prevents it from recovering its original state after removal of the applied load. The largest stress that a material can endure without failing (that is, breaking or fracturing) is called the ultimate or failure stress, σ_f .

It is convenient to analyze deformations with respect to excursions on the stress-strain plane (Figure 5.2). If an elastic material is subjected to a load producing strain ϵ_A , which is less than the yield strain ϵ_y , it will return to its original shape (after removal of the load) by following the same locus of stress-strain coordinates that characterized its deformation. If, however, the material is deformed beyond the elastic limit, to strain ϵ_B for example, the material will not recover completely. Generally, the relaxation of the material occurs along a

Table 5.1
Mechanical Properties

Materials

Biological Materials

Bone—long bone

Bone—cancellous

Bone—vertebrae

Bone—skull

Cartilage

Articular cartilage

Human knee joint

Brain

Brain tissue—gray

Brain tissue—white

Tendon

Tendon—Tendon

Ligament

Aorta

Human small intestine

Elastin

Isolated collagen

Formalin-fixed skin

Skin

Fibroblast-populated

Collagen sponge

Polymers

Polyimides

Polyester

Polystyrene

Poly(methyl methacrylate)

Polyethylene—high density

Polytetrafluoroethylene

Polyethylene—low density

Metals

Stainless steel (304)

Titanium

Aluminum

Others

Silicon

Aluminum oxide

Magnesium oxide

Fused silica (SiO₂)

Concrete

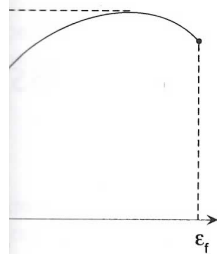
Wood

Compiled from [2]

Table 5.1
Mechanical Properties of Commonly Encountered Biological Materials

Materials	E (MPa)	Strength, Ultimate Stress (MPa)	Poisson's Ratio
<i>Biological Materials</i>			
Bone—long bone	15,000 to 30,000	130 to 220 in compression 80 to 150 in tension 70 in shear	
Bone—cancellous	90 to 500	2 to 5	
Bone—vertebrae	100 to 300		
Bone—skull	6,500		0.22
Cartilage		5	
Articular cartilage	1 to 10 in tension 1 in compression	9 to 40	
Human knee menisci	70 to 150		
Brain	0.067		0.48
Brain tissue—gray matter	0.005		
Brain tissue—white matter	0.014		
Tendon	1,000 to 2,000	50 to 100	
Tendon—Tendo achillis	375		
Ligament		50 to 100	
Aorta		0.3 to 0.8	
Human small artery	0.1 to 4		
Elastin	0.6		
Isolated collagen fibers	1,000	50	
Formalin-fixed myocardium	101		
Skin	0.1 to 2 (phase I)	1 to 20	
Fibroblast-populated matrix	0.08 to 0.8		
Collagen sponge	0.017 to 0.028		
<i>Polymers</i>			
Polyimides	3,000 to 5,000		
Polyester	1,000 to 5,000		
Polystyrene	2,300 to 3,300		
Poly(methyl methacrylate)	2,000 to 3,000		
Polyethylene—high density	1,100		
Polytetrafluoroethylene	400 to 600		
Polyethylene—low density	200 to 500		
<i>Metals</i>			
Stainless steel (316)	210,000	450 in tension	
Titanium	107,000		
Aluminum	69,000		
<i>Others</i>			
Silicon	150,000		
Aluminum oxide (Al_2O_3)	393,000		
Magnesium oxide (MgO)	225,000		
Fused silica (SiO_2)	73,000		
Concrete	2,800	4.5 in compression	
Wood	140	3.6	

Compiled from [21, 24, 29].



A uniform material is
material to tensile loads of

in. This relationship is
Hooke, and it describes
which deform linearly
removal of the load. The
property of the material;
elastic materials obey
materials will recover their
ess. Fortunately, many
only if the deformations

formed beyond a critical
by Hooke's law for all
by Hooke's law over a
and continue to deform
This state occurs at a
material results in plastic
change in the material
removal of the applied
without failing (that is,
stress, σ_f .

et to excursions on the
is subjected to a load
 ϵ_y , it will return to its
the same locus of stress—
however, the material
mple, the material will
material occurs along a

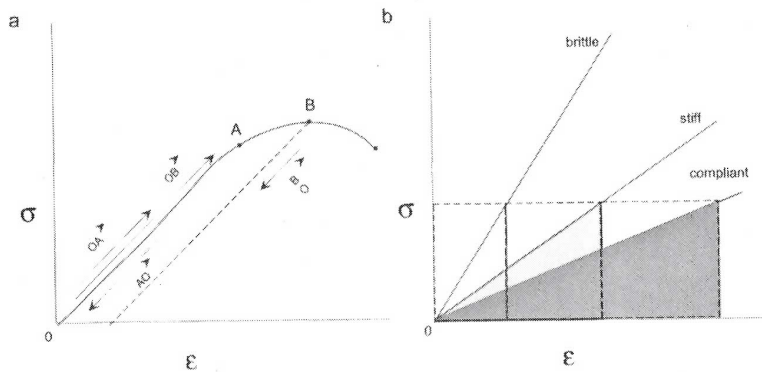


Figure 5.2. Typical characteristics of an elastic material. (a) Deformation in response to applied load for an elastic and a non-elastic material. (b) The energy stored in a material during deformation can be determined by integrating under the stress-strain curve.

line that is parallel to the initial deformation (that is, with slope equal to E , if it is a linear elastic solid).

Energy is added to a material when it is stressed; this mechanical energy, called strain energy, is stored in the material. For an elastic material, the strain energy U_0 , is determined from

$$U_0 = \frac{1}{2} \sigma \epsilon \quad (5-2)$$

which can be determined graphically from the area under the stress-strain curve. Elastic elongation and relaxation has no net energy cost; all of the energy stored in the material during elongation is returned during relaxation. But energy is lost when deformation goes beyond the elastic limit. The net loss of energy can be calculated from the difference between the strain energy required to accomplish the elongation and the energy recovered after removal of the load (net energy loss can be determined graphically, as well). The ability to store energy can be an important material property. The potential of a material for energy storage can be represented by the strain energy at failure. Brittle materials have a low U_0 at fracture, whereas compliant materials, which deform readily, can store substantial amounts of strain energy.

5.1.2 Poisson's Ratio

In the examples developed in the previous section, a stress was applied in a single direction and the response (or strain) in that same direction was observed and analyzed. Most materials will elongate when stress is applied in the axial direction, but the cross-sectional area will also decrease (Figure 5.3). The rela-

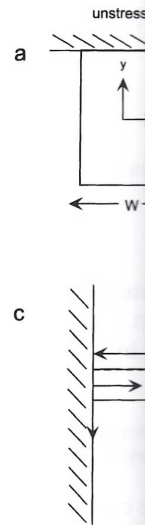


Figure 5.3. Deformation of a material in different directions. (a) Unstressed state. (b) Bending deformation. (c) Bending deformation when subjected to a load.

tive magnitude of the lateral strain, ν_{xy} , which

In the simple example, the cross-sectional area decreases continuously with increasing stress. This discrepancy between the true stress in the point. The true stress decreases in cross-sectional area, decrease in "stress".

5.1.3 Quantifying Stress

Stress that is applied in a single direction (Figure 5.3). Shear stress is applied. Since the

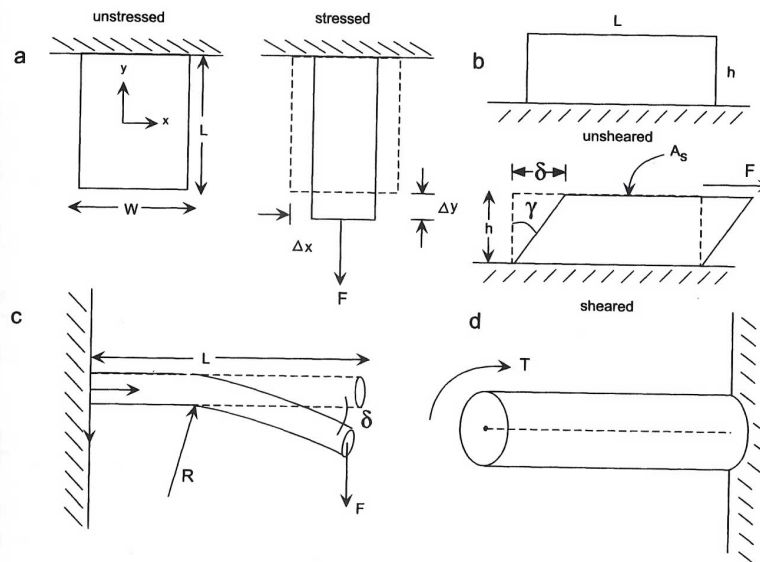


Figure 5.3. Deformation of materials under stress. (a) The extent of deformation of a material in directions other than the direction of force application is determined by Poisson's ratio. (b) Materials also deform when exposed to a shearing stress. (c) Bending deformation occurs for elongated or slender bodies. (d) Materials can deform when subjected to torsional forces.

tive magnitude of strains experienced in two dimensions is given by Poisson's ratio, ν_{xy} , which is also a property of the material:

$$\nu_{xy} = -\frac{\epsilon_y}{\epsilon_x} \quad (5-3)$$

In the simple example shown in Figure 5.1, the cross-sectional area decreases continuously with loading. Stresses are usually defined on the basis of the unstressed area and therefore underestimate the actual stress in the material. This discrepancy between "stress" (which is defined for convenience) and the true stress in the material accounts for the decrease in "stress" near the fracture point. The true stress is increasing up to the point of fracture but the rapid decrease in cross-sectional area that precedes failure leads to the apparent decrease in "stress".

5.1.3 Quantifying Deformations in Other Geometries

Stress that is applied to a material tangentially causes shear deformation (Figure 5.3). Shear stress, τ , is defined per area of the surface on which it is applied. Since the deformation in shear is a function of distance from the plane

on which the shear is applied, shear strain (ϵ_s) is measured by the angular deformation of the material:

$$\epsilon_s = \frac{\delta}{h} = \tan \gamma \quad (5-4)$$

For small deformations, that is, γ near 0 so that $\tan \gamma \simeq \gamma$, a shear modulus (G) can be defined:

$$\tau = G\gamma \quad (5-5)$$

Bending deformations also are important, particularly in elongated objects that bear tangential loads (Figure 5.3). If a force F is applied at the end of a symmetrical beam of length L , and the material is isotropic and linearly elastic, the deflection due to bending is

$$\delta = -\frac{FL^3}{3EI} \quad (5-6)$$

where E is Young's modulus and I is moment of inertia for the cross-sectional area. The stress within the material depends on the distance from the central axis, y :

$$\sigma_x = -\frac{My}{I} \quad (5-7)$$

where M is the bending moment (equal to FL at the end of the beam in Figure 5.3). If $M > 0$, the negative sign indicates that the stress is compressive for $y > 0$ and tensile for $y < 0$. Torsional forces also produce stress within materials. The shear stress due to torsion is

$$\tau = \frac{Tr}{J} \quad (5-8)$$

where J is the radial moment of inertia and T is the torque.

5.2 Elementary Fluid Mechanics

5.2.1 Basics and Definitions

A fluid deforms continuously under the action of a shearing stress. Two classes of fluids can be defined: incompressible fluids, in which the density is constant; and compressible fluids, in which the density (ρ) is a function of pressure. Most liquids are incompressible at pressures near atmospheric; pure water has a density of 999 kg/m^3 at 15°C and 993 kg/m^3 at 37°C ; blood has a density of $1,060 \text{ kg/m}^3$. Gases are much less dense than liquids and cannot always be assumed incompressible; air has a density of 1.22 kg/m^3 at 15°C and 1 atm.

When a fluid is at rest—that is, in the absence of shearing stresses—the fluid is said to be in hydrostatic equilibrium. A force balance on an element of a stationary incompressible fluid (Figure 5.4) yields the following relationship:

Figure 5.4. For the z -direction

where g is the equation can be spatial location expression mu

Viscosity deformation of (Figure 5.5). If the right with fluid within the motion. The force area of plate/t F/A , where A shear stress is distance h :

More precisely derivative of the

where τ_{xy} is the of constant y . in the y -direction

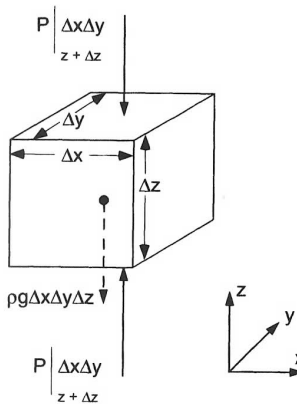


Figure 5.4. Force balance on a volume element in a fluid at rest. Forces acting in the z -direction (parallel to gravitational force) are shown.

$$\frac{dp}{dz} = -\rho g \quad (5-9)$$

where g is the acceleration due to gravity (9.80665 m/s^2). This differential equation can be used to calculate the difference in pressure between any two spatial locations in a stationary fluid. If the fluid is incompressible, the expression must be integrated with density as a function of position.

Viscosity is a material property related to the resistance to “flow” or deformation of a fluid. Consider a fluid entrapped between two parallel plates (Figure 5.5). If the bottom plate is held stationary and the top plate is moved to the right with a constant velocity v_0 by application of a tangential force F , the fluid within the gap will be subjected to a shearing stress that produces fluid motion. The force applied to the plate is uniformly transmitted over the entire area of plate/fluid contact; therefore, the tangential shear stress is equal to F/A , where A is the cross-sectional area of the plate. Experimentally, the shear stress is proportional to the velocity of the upper plate v_0 and the gap distance h :

$$\tau \propto \frac{v_0}{h} \quad (5-10)$$

More precisely, the shear stress is equal to a constant multiplied by the first derivative of the velocity with respect to distance normal to the moving plate:

$$\tau_{xhy} = \eta \frac{dv_x}{dy} \quad (5-11)$$

where τ_{xy} is the viscous shear stress in the x -direction exerted on a fluid surface of constant y . This quantity, τ_{yx} , is also the viscous flux of x -momentum (ρv_x) in the y -direction. The negative sign must be included because viscosity has a

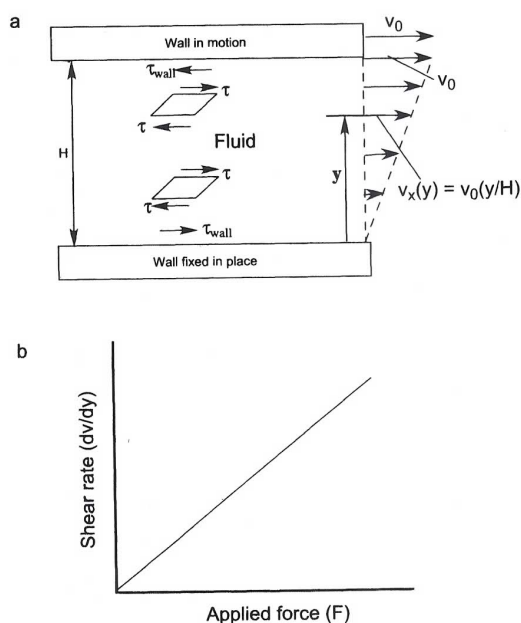


Figure 5.5. Viscosity in a fluid between two parallel plates in relative motion. (a) Schematic diagram showing movement of the upper plate and velocity profile within the fluid. (b) Variation of shear rate with applied force.

positive value ($\eta > 0$) and the momentum flux is positive in the direction of decreasing velocity.

The shear stress and the velocity gradient (also called the shear rate) are proportional, with the constant of proportionality $-\eta$, where η is the viscosity of the fluid. For some fluids, Equation 5-11 holds over a wide range of shear rates with constant η (as in Figure 5.5b); these are called *Newtonian* fluids. The value of the viscosity η is a property of the fluid being a function of fluid phase composition, temperature, and pressure. Water is a Newtonian fluid; at body temperature (37°C) the viscosity of water is 0.75 cp (centipoise; $1 \text{ cp} = 0.01 \text{ g/cm-s}$) and that of plasma is 1.2 cp. Whole blood has a viscosity of 3.0 cp, provided that the shear rate is sufficiently high so that red cell aggregates do not form (see discussion on blood cell aggregation in Chapter 8). At lower shear rates, the viscosity of blood is a function of shear rate and blood composition.

5.2.2 Kinematics of Fluid Flow

One is often interested in knowing the velocity of flow within a fluid subjected to shearing stresses. Fluid motion can be described through the use of mathematical expressions of the basic principles of conservation of mass, momen-

tum, and energy. A system of interest is a particular system of characteristic infinitesimal volume of total mass in a

The rate of change of mass addition to the system through one of the

$$\frac{\partial}{\partial t}(\rho) \Delta x \Delta y$$

$$- \rho v_y|_{y+}$$

Dividing each term

$$\frac{\partial \rho}{\partial t} = \left[\frac{\rho v_x|_x \Delta x}{\Delta x} + \frac{\rho v_z|_z \Delta z}{\Delta z} \right]$$

In the limit, as the volume of the control volume approaches zero, the right-hand-side

which can be written

Figure 5.6. Balance of mass for a control volume. The control volume is a rectangular prism of length Δx and height Δy perpendicular to the

tum, and energy. These conservation equations can be derived for a particular system of interest or, alternately, can be developed generally and then applied to a particular system. The latter approach requires consideration of a characteristic infinitesimal volume element; Figure 5.6 illustrates the conservation of total mass in a rectangular coordinate system.

The rate of change in mass of the volume element is equal to the net rate of mass addition to the volume element, which can only occur by fluid flow through one of the six boundaries:

$$\begin{aligned} \frac{\partial}{\partial t}(\rho)\Delta x\Delta y\Delta z = & \rho v_x|_x\Delta y\Delta z - \rho v_x|_{x+\Delta x}\Delta y\Delta z + \rho v_y|_y\Delta x\Delta z \\ & - \rho v_y|_{y+\Delta y}\Delta x\Delta z + \rho v_z|_z\Delta x\Delta y - \rho v_z|_{z+\Delta z}\Delta x\Delta y \end{aligned} \quad (5-12)$$

Dividing each term by the volume yields

$$\begin{aligned} \frac{\partial \rho}{\partial t} = & \left[\frac{\rho v_x|_x\Delta y\Delta z - \rho v_x|_{x+\Delta x}\Delta y\Delta z}{\Delta x\Delta y\Delta z} \right] + \left[\frac{\rho v_y|_y\Delta x\Delta z - \rho v_y|_{y+\Delta y}\Delta x\Delta z}{\Delta x\Delta y\Delta z} \right] \\ & + \left[\frac{\rho v_z|_z\Delta x\Delta y - \rho v_z|_{z+\Delta z}\Delta x\Delta y}{\Delta x\Delta y\Delta z} \right] \end{aligned} \quad (5-13)$$

In the limit, as the volume element becomes infinitesimal, the three terms on the right-hand-side become partial derivatives:

$$\frac{\partial \rho}{\partial t} = \left[\frac{\partial(\rho v_x)}{\partial x} \right] + \left[\frac{\partial(\rho v_y)}{\partial y} \right] + \left[\frac{\partial(\rho v_z)}{\partial z} \right] \quad (5-14)$$

which can be written in more compact notation as

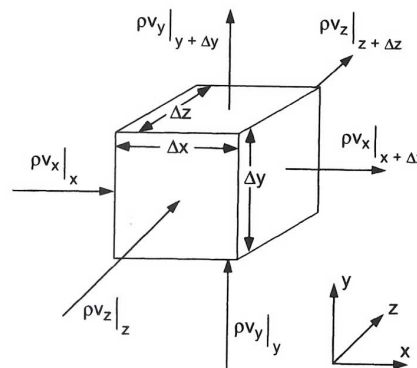


Figure 5.6. Balance of total mass on a volume element within a fluid in motion, illustrated for a rectangular coordinate system. The rate of mass flow through each boundary of the volume element is determined from the local density and velocity perpendicular to the boundary.

$$\frac{\partial \rho}{\partial t} = -\nabla \cdot (\rho \bar{v}) \quad (5-15)$$

or expanded to allow inclusion of the substantial derivative of density:

$$\frac{D\rho}{Dt} \left[= \frac{\partial \rho}{\partial t} + \rho \bar{v} \right] = -\rho \nabla \cdot \bar{v} \quad (5-16)$$

In the special case of an incompressible fluid, the density is not a function of time or position and the mass balance equation can be simplified:

$$0 = \nabla \cdot \bar{v} \quad (5-17)$$

The conservation equation for momentum is somewhat more difficult to obtain, but can be derived by a similar procedure (for more details, see [4, 5]):

$$\frac{\partial(\rho v_x)}{\partial t} = - \left(\frac{\partial(\rho v_x v_x)}{\partial x} + \frac{\partial(\rho v_y v_x)}{\partial y} + \frac{\partial(\rho v_z v_x)}{\partial z} \right) - \left(\frac{\partial \tau_{xx}}{\partial x} + \frac{\partial \tau_{yx}}{\partial y} + \frac{\partial \tau_{zx}}{\partial z} \right) - \frac{\partial p}{\partial x} + \rho g_x \quad (5-18)$$

Equation 5-18 gives the expression for conservation of momentum in the x -direction; similar expressions are obtained in the y - and z -directions [4,5]. These three component expressions can be written as a single, more compact vector expression:

$$\frac{\partial(\rho \bar{v})}{\partial t} = -\nabla p + \rho \bar{g} - \bar{\nabla} \cdot \bar{\tau} - \nabla \cdot \rho \bar{v} \bar{v} \quad (5-19)$$

To use this differential equation, we need a constitutive equation that relates the individual elements of the stress tensor ($\bar{\tau}$) to gradients in the velocity field. This constitutive equation relates local rates of movement in the fluid (that is, velocity) to local stress and therefore depends on the physical properties of the fluid of interest. For a Newtonian fluid, the constitutive equation is

$$\left. \begin{aligned} \tau_{xx} &= -2\eta \frac{\partial v_x}{\partial x} + \frac{2}{3}\eta \left[\frac{\partial v_x}{\partial x} + \frac{\partial v_y}{\partial y} + \frac{\partial v_z}{\partial z} \right] \\ \tau_{yy} &= -2\eta \frac{\partial v_y}{\partial y} + \frac{2}{3}\eta \left[\frac{\partial v_x}{\partial x} + \frac{\partial v_y}{\partial y} + \frac{\partial v_z}{\partial z} \right] \\ \tau_{zz} &= -2\eta \frac{\partial v_z}{\partial z} + \frac{2}{3}\eta \left[\frac{\partial v_x}{\partial x} + \frac{\partial v_y}{\partial y} + \frac{\partial v_z}{\partial z} \right] \\ \tau_{xy} &= \tau_{yx} = -\eta \left(\frac{\partial v_x}{\partial y} + \frac{\partial v_y}{\partial x} \right) \\ \tau_{yz} &= \tau_{zy} = -\eta \left(\frac{\partial v_y}{\partial z} + \frac{\partial v_z}{\partial y} \right) \\ \tau_{zx} &= \tau_{xz} = -\eta \left(\frac{\partial v_x}{\partial z} + \frac{\partial v_z}{\partial x} \right) \end{aligned} \right\} \quad (5-20)$$

For a Newtonian fluid (Equations 5-15 to 5-19)

The velocity field \bar{v} in Eq. 5-21 in certain cases varies with position, v_x , v_y , v_z . Therefore, the useful information in the x -component of the momentum balance equation is

In most cases, the velocity field is relative to the parallel plates. The gravitational side of Equation 5-21 is

But there is no gravity, which leads to

The values for $v_x = 0$, $v_y = 0$ are evaluated:

This linear profile is shown in Figure 5-22.

5.2.3 Example

Consider an example of the formation of a biofilm. The formation of a biofilm is a problem for oxygen transport in tissue. The velocity through the biofilm is

(5-15)

of density:

(5-16)

not a function of
lified:

(5-17)

more difficult to
details, see [4, 5]:

$$\left(\frac{\partial \tau_{zx}}{\partial z} \right) - \frac{\partial p}{\partial x} + \rho g_x$$

(5-18)

momentum in the x -
directions [4,5].
e, more compact

(5-19)

ation that relates
the velocity field.
the fluid (that is,
properties of the
tion is

(5-20)

For a Newtonian fluid, the equation for conservation of momentum (Equations 5-19 and 5-20) reduces to

$$\rho \frac{D\vec{v}}{Dt} = -\nabla p + \eta \nabla^2 \vec{v} + \rho \vec{g} \quad (5-21)$$

The velocity field can be determined by solution of Equations 5-17 through 5-21 in certain situations. In the simple system illustrated in Figure 5.5, velocity varies with position in the fluid, but velocity only occurs in the tangential direction, v_x , and is only a function of distance normal to the moving plate y . Therefore, the conservation of mass equation (Equation 5-14) provides no useful information in this system. Conservation of momentum does provide interesting information; the equation for conservation of x -momentum (that is the x -component of Equation 5-21) reduces to

$$0 = -\frac{dp}{dx} + \eta \frac{d^2 v_x}{dy^2} + \rho g_x \quad (5-22)$$

In most cases of interest for tissue engineering, the effect of gravity is small relative to the pressure drop (alternately, depending on the orientation of the parallel plates with respect to the gravitational field, the x -component of the gravitational vector may be equal to zero); the third term on the right-hand side of Equation 5-22 can therefore be neglected. Dropping the third term in Equation 5-22 requires that the other two terms exactly balance:

$$\frac{dp}{dx} = \eta \frac{d^2 v_x}{dy^2} \quad (5-23)$$

But there is no hydrostatic pressure drop in the x -direction in this situation, which leads to a simple solution for the functional form of the velocity profile:

$$v_x = Ay + B \quad (5-24)$$

The values for the velocity in the x -direction are known at the boundaries v_0 at $y = 0$, $v_x = 0$ and at $y = H$, $v_x = v_0$. Therefore, the constants A and B can be evaluated:

$$v_x = v_0 \left(\frac{y}{H} \right) \quad (5-25)$$

This linear profile for velocity as a function of distance from the stationary plate is shown in Figure 5.5.

5.2.3 Example: Flow Through a Cylindrical Tube

Consider an additional example of relevance to tissue engineering (Figure 5.7). The formation of functional blood vessels within tissues is one of the fundamental problems of tissue engineering; blood flow through tissues is essential for oxygen transport to cells within the tissue mass. Mechanically, blood vessels in tissue serve as conduits for the flow of blood. Blood moves at varying velocity through individual tissues within the body via an interconnected and

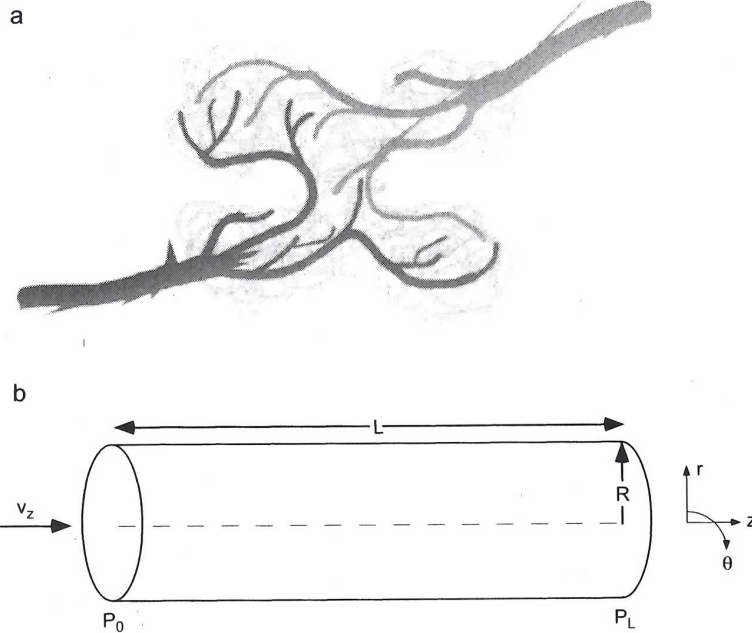


Figure 5.7. Fluid flow through a blood vessel. (a) The development of vasculature within a growing tissue is a key problem in tissue engineering. Adapted from a drawing on the Carnegie Mellon University Bone Tissue Engineering Initiative website (<http://www-2.cs.cmu.edu/People/tissue/tutorial.html>). (b) The flow of blood through a capillary, vein, or artery can be modeled as an incompressible fluid flowing through a cylindrical tube.

highly branched network of cylindrical vessels. Therefore, the flow of an incompressible fluid within a cylindrical vessel is of central importance to the biophysics of the circulatory system and to our understanding of vascular tissue engineering.

Well-behaved flow through a cylindrical tube (Figure 5.7) occurs only in the axial direction ($\vec{v} = v_z(r)\hat{k}$), where v_z is a function of radial distance r from the tube centerline. The z -component of the conservation of momentum equation, Equation 5-21, in cylindrical coordinates, is

$$\begin{aligned} \rho \left(\frac{\partial v_z}{\partial t} + v_r \frac{\partial v_z}{\partial r} + \frac{v_\theta}{r} \frac{\partial v_z}{\partial \theta} + v_z \frac{\partial v_z}{\partial z} \right) \\ = - \frac{\partial p}{\partial z} + \eta \left[\frac{1}{r} \frac{\partial}{\partial r} \left(r \frac{\partial v_z}{\partial r} \right) + \frac{1}{r^2} \frac{\partial^2 v_z}{\partial^2 r} + \frac{\partial^2 v_z}{\partial z^2} \right] + \rho g_z \end{aligned} \quad (5-26)$$

This equation can be simplified by the following assumptions, which apply reasonably well to blood flows in most vessels in humans:

- steady flow
- flow only in the z -direction
- gravitational effects are negligible

These assumptions

The term on the left is only a function of z , the second derivative is only a function of r , and the term on the right is a constant, which can be separated into

If the pressure drop is constant K corresponds to a pressure drop Δp , for example, if the pressure drop is

$$p = p_0 - Kz$$

then the solution to the equation is

Therefore, the value of K in Equation 5-28 can be determined from measurements of pressure drop on velocity:

$$v_z = 0$$

to yield

This parabolic dependence of velocity on radial distance is characteristic of Hagen-Poiseuille flow (after Jean Marie Poiseuille and Johann Heinrich Hagen (physicist) and Hagen (physicist) measurements of pressure drop in 1840).

- steady flow ($\partial v_z / \partial t = 0$);
- flow only in axial direction ($v_r = v_\theta = 0$);
- gravitational forces are negligible ($g_z \approx 0$).

These assumptions can be used to simplify Equation 5-26:

$$\frac{\partial p}{\partial z} = \eta \frac{1}{r} \frac{\partial}{\partial r} \left(r \frac{\partial v_z}{\partial r} \right) \quad (5-27)$$

The term on the left-hand side of Equation 5-27 (that is, the pressure gradient) is only a function of z , whereas the term on the right-hand side (that is, the second derivative of the velocity component in the z -direction with respect to r) is only a function of r . These two terms can be equal only if they are both equal to a constant, which we will temporarily call K . Therefore, Equation 5-27 can be separated into two ordinary differential equations:

$$\left. \begin{aligned} \frac{dp}{dz} &= K \\ \eta \frac{1}{r} \frac{d}{dr} \left(r \frac{dv_z}{dr} \right) &= K \end{aligned} \right\} \quad (5-28)$$

If the pressure drop over the length of the vessel is known, the value of the constant K corresponding to this physical situation can be determined. For example, if the pressures at the inlet and outlet of the vessel are known,

$$p = p_0 \quad \text{at} \quad z = 0; \quad p = p_L \quad \text{at} \quad z = L \quad (5-29)$$

then the solution to the first part of Equation 5-28 is

$$p = p_0 + \left(\frac{P_L - P_0}{L} \right) z \quad (5-30)$$

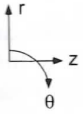
Therefore, the value of K in this case is $(P_L - P_0)/L$. The second part of Equation 5-28 can be solved, subject to the following boundary conditions on velocity:

$$v_z = 0 \quad \text{at} \quad r = R; \quad \frac{\partial v_z}{\partial r} = 0 \quad \text{at} \quad r = 0 \quad (5-31)$$

to yield

$$v_z = \frac{(P_0 - P_L)R^2}{4\eta L} \left(1 - \frac{r^2}{R^2} \right) \quad (5-32)$$

This parabolic dependence of local velocity on radial position (Figure 5.8) is characteristic of Hagen-Poiseuille flow (named in honor of Jean Leonard Marie Poiseuille and Gotthilf Heinrich Ludwig Hagen). Poiseuille (a physiologist) and Hagen (an engineer) independently published the first systematic measurements of pressure drop within flowing fluids in simple tubes in 1839 and 1840.



vasculature
from a
initative
ow of blood
e fluid

e flow of an
ortance to the
g of vascular

occurs only
ial distance r
of momentum

(5-26)

which apply

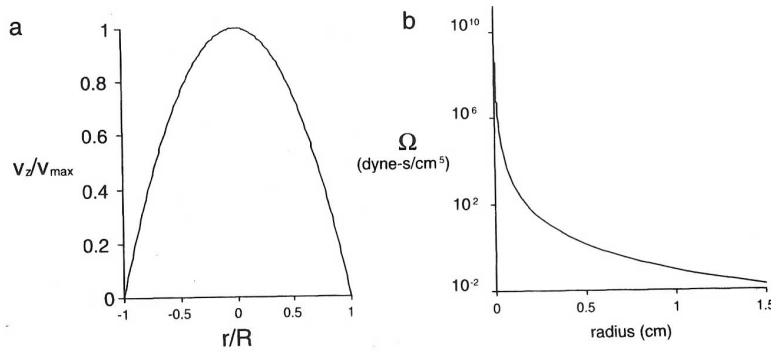


Figure 5.8. Velocity profile through a cylindrical vessel. (a) The velocity varies with the radial distance from the vessel centerline, as in Equation 5-37. (b) The overall resistance of the vessel to flow is a strong function of the vessel radius.

Equation 5-32 can be integrated over the vessel cross-section to obtain an overall rate of blood flow:

$$Q = \int_0^{2\pi} \int_0^R v_z(r) r dr d\theta = \frac{(P_0 - P_L) \pi R^4}{8\eta L} \quad (5-33)$$

which provides—by analogy to the resistance of electrical circuits ($\Delta V = i\Omega$)—the overall resistance of a cylindrical vessel to flow:

$$\Omega = \frac{\Delta p}{Q} = \frac{8\eta L}{\pi R^4} \quad (5-34)$$

The resistance to flow is a strong function of vessel radius: $\Omega \propto R^{-4}$. As blood vessels become smaller the resistance to flow increases dramatically (Figure 5.8). One consequence of the branching pattern of blood vessels is that the majority of the overall resistance to blood flow resides in the smallest vessels; in the human circulatory system, the majority of the pressure drop (~80%) occurs in arterioles and capillaries. This natural consequence of the physics of fluid flows is exploited in regulation of blood flow to organs of the body. Local blood flow to a tissue is controlled by constriction and dilation of the arterioles delivering blood to that tissue. Since the greatest overall resistance is provided by arterioles, and because individual arterioles have muscular walls which permit them to adjust their diameter, and therefore their resistance (Equation 5-34), the proportion of blood flow arriving at the tissue served by an arteriole can be regulated with precision.

5.3 Mechanical Properties of Biological Fluids and Gels

The preceding two sections describe the mechanical behavior of idealized elastic materials and incompressible fluids, respectively. Real materials some-

times behave like one logical materials, which complex behaviors than an incompressible fluid models. Materials that viscoelastic.

5.3.1 Models of Viscoelasticity

Consider, first, the behavior of elastic materials. Although these materials are often deformed, they also have characteristic shapes without a constant force. Although they may deform, they often continue to deform under a constant force, called creep (Figure 5.9). The force required to maintain a constant deformation is called stress relaxation.

Ideal elastic materials are predicted by Hooke's law, which states that the elongation of a material is proportional to the force that occurs during creep.

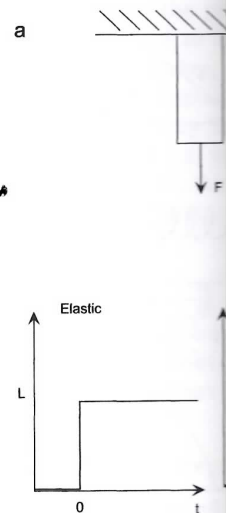


Figure 5.9. Comparison of elastic and viscoelastic behavior upon application of a constant force.

times behave like one of these idealized models. On the other hand, biological materials, which are frequently complex in composition, can exhibit complex behaviors that resemble aspects of both the elastic material and an incompressible fluid, but are also unlike either of these idealized models. Materials that exhibit both viscous and elastic natures are called viscoelastic.

5.3.1 Models of Viscoelastic Materials

Consider, first, the behavior of biological tissues such as skin and muscle. Although these materials are rich in water, which is a fluid at body temperature, they also have characteristics of an elastic solid: for example, they retain their shape without a containing vessel. Ideal elastic materials—that is, materials that are deformed to less than the elastic limit—deform instantaneously. Although they may deform very rapidly after loading, biological materials often continue to deform slowly after the initial period, exhibiting a behavior called creep (Figure 5.9). When this same material is rapidly deformed, the force required to maintain this deformation decreases gradually; this process is called stress relaxation.

Ideal elastic materials are modeled as springs; their behavior can be predicted by Hooke's law (Equation 5-1). Another element—one that slowly elongates upon application of a force—must be added to model the changes that occur during creep and stress relaxation. Continuous deformation after

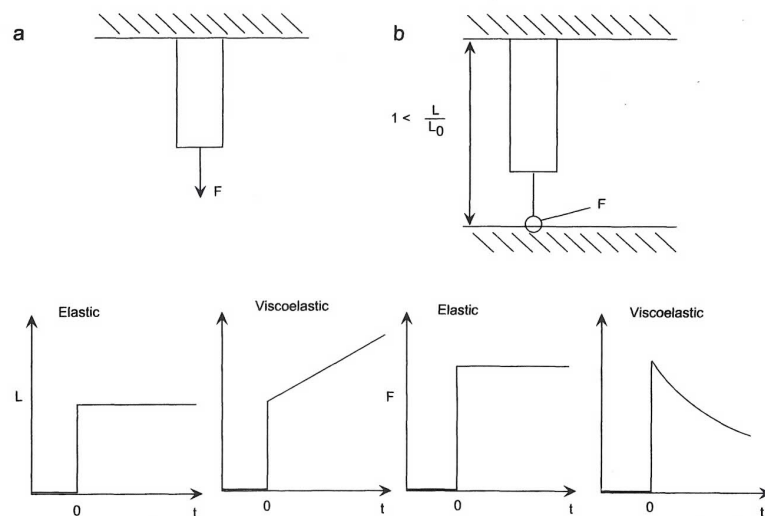


Figure 5.9. Comparison of elastic and viscoelastic behavior. Typical responses of elastic and viscoelastic materials to (a) application of a constant load and (b) application of a constant elongation.

loading is a characteristic of fluids; the rate of deformation is determined by the viscosity of the fluid, as in Equation 5-11. A dashpot, or piston within a cylinder is the mechanical analog for viscosity; the piston slowly moves through the cylinder at a rate that is determined by friction between the surfaces, in response to an applied load. By combining elastic (that is, spring) and viscous (that is, dashpot) elements, models that predict aspects of the behavior of real viscoelastic materials can be developed (Figure 5.10).

Deformation of viscoelastic materials depends on the total history of applied force. If $F(t)$ is a function describing the history of applied force on a material, then, over a small time interval $d\tau$, the change in applied force is given by $(dF/dt)|_{\tau}d\tau$. The change in deformation at time t , $dU(t)$, can be expressed in terms of a history, or creep, function, $c(t - \tau)$:

$$dU(t) = c(t - \tau) \frac{dF}{dt} \Big|_{\tau} d\tau \quad (5-35)$$

This expression can be integrated to obtain the total deformation as a function of time t :

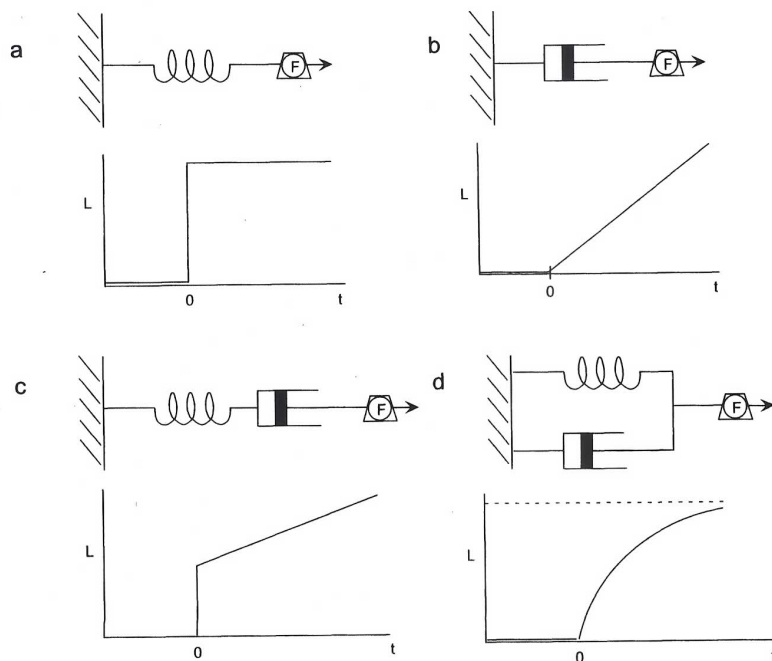


Figure 5.10. Behavior of spring-dashpot models. The elongation after application of a constant load to (a) spring, (b) dashpot, (c) spring and dashpot in series (Maxwell solid), and (d) spring and dashpot in parallel (Voigt solid).

The relaxation function defined in a similar way

The relaxation function applied to produce a $c(\cdot)$ can be interpreted in applied force. It depends only on the $k(0) = 0$. The challenge a viscoelastic material

It is often useful idealized models. A simple perfectly elastic spring elastic material is provided in form

where F is the total force displacement of the material the behavior of the spring which is identical to applied to the spring. When a force is applied length prescribed by Equation 5.10a). Similarly, a simple mechanical object, the simple viscous liquid:

which is similar to Equation behaves like a viscous material will continue

5.3.2 Example: Creep

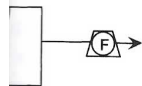
Many materials do not load is applied, the material some ultimate deformation materials can be considered

etermined by the
piston within a
n slowly moves
between the sur-
at is, spring) and
s of the behavior

total history of
applied force on
applied force is
t, $dU(t)$, can be

(5-35)

on as a function



er application of
series (Maxwell

$$U(t) = \int_0^t c(t - \tau) \frac{dF}{d\tau} \bigg|_{\tau} d\tau \quad (5-36)$$

The relaxation function $k(t - \tau)$, the inverse of the creep function, can be defined in a similar way:

$$dF(t) = k(t - \tau) \frac{dU}{d\tau} \bigg|_{\tau} d\tau \quad (5-37)$$

The relaxation function $k(\cdot)$ can be interpreted as the force which must be applied to produce a unit step increase in deformation; the creep function $c(\cdot)$ can be interpreted as the elongation which results from a unit step increase in applied force. In general, the state of the material at the present time depends only on the previous history, so that one may assume $c(0) = 0$ and $k(0) = 0$. The challenge is to find the creep or relaxation function that describes a viscoelastic material of interest.

It is often useful to compare the behavior of real materials with that of idealized models. A simple elastic material, for example, can be compared to a perfectly elastic spring, as shown in Figure 5.10a. The behavior of the perfectly elastic material is provided by Equation 5-1, which can also be written in the form

$$F = \mu U \quad (5-38)$$

where F is the total force applied to the material and U is the instantaneous displacement of the material, as defined in Equations 5-35 through 5-37. All of the behavior of the perfectly elastic material is contained in Equation 5-38, which is identical to the equation describing a spring (where F is the force applied to the spring, U is the displacement, and μ is the spring constant). When a force is applied to a spring, the spring instantaneously deforms to the length prescribed by Equation 5-38 (this deformation is illustrated in Figure 5.10a). Similarly, a viscous liquid can be compared to another idealized mechanical object, the dashpot (Figure 5.10b). The dashpot behaves like a simple viscous liquid:

$$F = \eta \frac{dU}{dt} \quad (5-39)$$

which is similar to Equation 5-11. When a force is applied to a material that behaves like a viscous liquid, it deforms continuously; the deformation of the material will continue for as long as the force is applied.

5.3.2 Example: Creep Function for a Maxwell Solid

Many materials do not behave like perfect springs or dashpots. When a fixed load is applied, the material might deform with time (like a dashpot), reaching some ultimate deformation that is not exceeded (like a spring). Models of materials can be constructed by combining springs and dashpots in different

combinations; the Maxwell model and the Voigt model are illustrated in Figures 5.10c and 5.10d respectively. A Maxwell solid behaves like a spring and a dashpot in series; it will deform instantaneously, like a spring, but the deformation will continue at some steady rate, like a dashpot. The total rate of deformation at any time, therefore, depends on the characteristics of both the spring and the dashpot:

$$\frac{dU}{dt} = \frac{1}{\mu} \frac{dF}{dt} + \frac{1}{\eta} F \quad (5-40)$$

where the initial elongation depends only on the force applied at time 0: $U(0) = F(0)/\eta$. Recall that the creep function can be interpreted as the deformation that results from a step change in force; the unit step change can be described mathematically as

$$F(t) = H(t), \quad \text{where} \quad H(t) = \begin{cases} 1 & \text{when } t \geq 0 \\ 0 & \text{when } t < 0 \end{cases} \quad (5-41)$$

The total deformation at time t can be obtained by integration of Equation 5-40, giving

$$U(t) = \int_{-\infty}^t \left(\frac{1}{\mu} \frac{dF}{dt} + \frac{F}{\eta} \right) dt \quad (5-42)$$

which, in the case where the force is a unit step change, becomes

$$U(t) = \int_{-\infty}^t \left(\frac{1}{\mu} \delta(t) + \frac{H(t)}{\eta} \right) dt = \frac{1}{\mu} + \frac{t}{\eta} \quad (5-43)$$

where the delta function, $\delta(t)$, is the first derivative of the step function:

$$\delta(t) = \begin{cases} 1 & \text{when } t = 0 \\ 0 & \text{elsewhere} \end{cases} \quad (5-44)$$

The creep function is defined by Equation 5-35. When the applied force is a unit step change, so that the first derivative of F is equal to the delta function, Equation 5-35 reduces to

$$U(t) = \int_0^t c(t - \tau) \delta(\tau) d\tau = c(t) \quad (5-45)$$

Comparison of Equations 5-43 and 5-45 yields a functional form for the creep function of a Maxwell model:

$$c(t) = \frac{1}{\mu} + \frac{t}{\eta} \quad (5-46)$$

or, more generally, given that the value of the creep function $c(0)$ is known at $t = 0$:

Similar approach
model or the
Exercises 5.3 a

5.3.3 Rheology

The deformation
its function. In
tance to flow t
work that must
system. A num
(that is, the de
developed. One
forced by press
the fluid is ther

Another co
is confined betw
plate is rotated
The torque requ
the viscosity of
Newtonian, the

where α is the a
shear stress τ is

where R is the

This steady
of many fluids,
blood is placed
as a function of
increasing shear
blood form aggr
shear rate incre
ing a decrease in
appears to be du
individual cells,

In some bio
irreversibly alte
might be interes
tions. Consider,

$$c(t - \tau) = \left(\frac{1}{\mu} + \frac{t - \tau}{\eta} \right) H(t - \tau) \quad (5-47)$$

Similar approaches can be used to find the relaxation function for a Maxwell model or the creep/relaxation functions for other model materials (see Exercises 5.3 and 5.4).

5.3.3 Rheological Properties of Biological Fluids and Gels

The deformation character of a biological fluid is often a critical determinant of its function. In blood, for example, the viscosity determines the overall resistance to flow through a vessel (Equation 5-34) and, therefore, the amount of work that must be supplied by the heart to move blood through the circulatory system. A number of methods for determining rheological properties of a fluid (that is, the deformation response of a fluid to applied stresses) have been developed. One common method is capillary viscometry, in which the fluid is forced by pressure through a capillary of known dimensions; the viscosity of the fluid is then calculated from Equation 5-34.

Another common method is cone-and-plate viscometry, in which the fluid is confined between two surfaces as shown in Figure 5.11. In this apparatus the plate is rotated with a constant angular velocity Ω and the cone held stationary. The torque required to hold the cone, T , is measured; this torque is related to the viscosity of the fluid that fills the gap between the surfaces. If the fluid is Newtonian, the shear rate imposed on the fluid is given by

$$\dot{\gamma} = \frac{\Omega}{\tan \alpha} \quad (5-48)$$

where α is the angle between the cone and the plate and $\dot{\gamma}$ is the shear rate. The shear stress τ is calculated from the torque T :

$$\tau = \frac{3T}{2\pi R^3} \quad (5-49)$$

where R is the radius of the cone.

This steady flow approach works for measuring the rheological properties of many fluids, even fluids that are structurally complex, such as blood. When blood is placed between the cone and the plate, the viscosity can be measured as a function of shear rate (Figure 5.11b). The viscosity of blood decreases with increasing shear rate. At low shear rates, $\dot{\gamma} < 0.1 \text{ s}^{-1}$, red blood cells within the blood form aggregates called rouleaux (see Figure 8.2) that resist flow. As the shear rate increases, the flow breaks the aggregates into individual cells, causing a decrease in the bulk viscosity. The complex rheological behavior of blood appears to be due to the aggregation of red cells as well as the deformability of individual cells, which is also a function of shear rate.

In some biological materials, however, the structure of the material is irreversibly altered by the simple shear flow process illustrated above; we might be interested, however, in the material's behavior with small deformations. Consider, for example, a polymer solution in which the concentration of

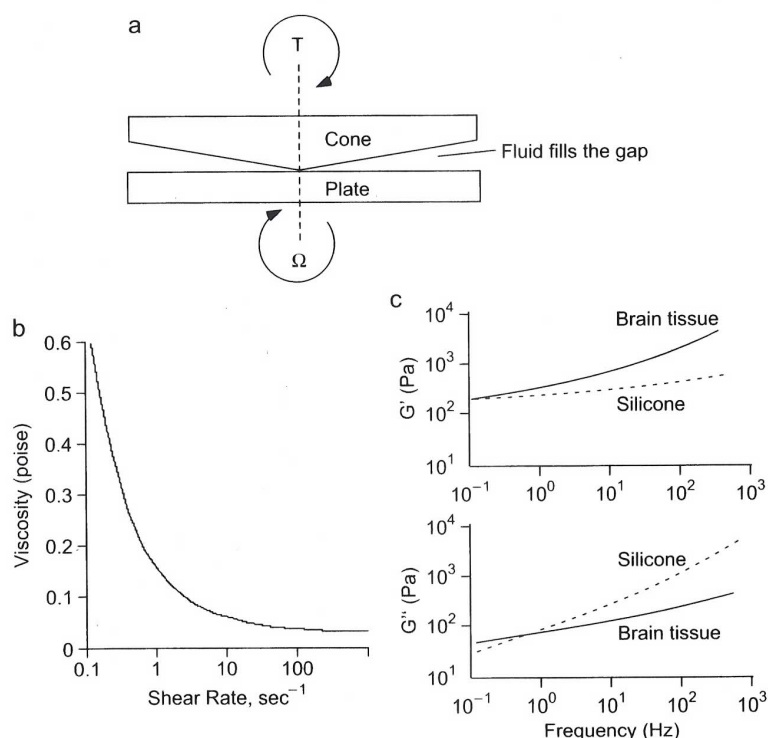


Figure 5.11. Cone-and-plate viscometry. The viscometer (a) can be used to measure the rheological behavior of blood (b) and of gels or tissues (c). Panel c was redrawn from [28].

polymer is above the gel point; that is, the concentration of polymer is high enough that polymer chains within the solution are connected (either chemically by crosslinks or physically by entanglements) into a large unit that spans the entire sample. A continuous shear flow, such as the one created by rotating the plate at a constant angular speed, would alter the structure of the gel phase by disrupting connections between the polymer chains. In biological gels, such as those formed from collagen, it is these polymer-polymer interactions that create the biologically important behavior of the material; therefore, measurements of viscosity in simple shear flow would not be relevant to the biological function. In these cases, the cone-and-plate rheometer can be operated in an oscillatory shear mode by oscillating the plate over small angles; the material between the plates is therefore exposed to a limited range of strains. The shear dependence of the response of the material can be measured by oscillating the plate at different frequencies. For example, a collagen gel sample might be subjected to oscillatory shear by rotating the plate at 2% strain with a frequency ranging from 0.1 to 100 rad/s [6].

In oscillatory shear, the angular frequency ω is in rad/s. The resulting torque τ is oscillatory but sinusoidal.

where τ_{\max} is the maximum torque, which can be decomposed into

$$\tau = -\eta' \gamma_{\max} \omega \sin(\omega t - \phi)$$

where γ_{\max} is the maximum shear strain, η' is the storage modulus, and η'' is the loss modulus. The value of G' , relative to G'' , is related to the phase shift of the response of the material about the elastic response. As an example of the use of this technique, we consider a model material.

5.4 Mechanical Properties

5.4.1 Mechanical Properties of Cells

Cells are complex structures. The smallest capillary diameter in circulation (Table 5.2) is smaller than red cells; the diameter of a single cell is smaller than one would predict from the size of the cell.

Table 5.2
Characteristics of Blood Cells

Cell type
Red cell (erythrocyte)
Platelet
Neutrophil
Lymphocyte
Monocyte

Data obtained from [8].

In oscillatory shear measurements, a sinusoidally varying shear field of frequency ω is imposed on the material by oscillating the plate; the amplitude of the resulting torque and the phase angle between the imposed shear and resulting torque on the cone are then measured. The stress response (τ) is also oscillatory but shifted from the imposed shear by an angle ϕ :

$$\tau = \tau_{\max} \cos(\omega t - \phi) \quad (5-50)$$

where τ_{\max} is the maximum value of the shear response. This stress can be decomposed into in-phase and out-of-phase terms:

$$\tau = -\eta' \gamma_{\max} \omega \cos(\omega t) - \eta'' \gamma_{\max} \omega \sin(\omega t) = -G'' \gamma_{\max} \cos(\omega t) - G' \gamma_{\max} \sin(\omega t) \quad (5-51)$$

where γ_{\max} is maximum strain, η' and η'' are the two dynamic viscosity coefficients, and G' and G'' are the storage and loss moduli, respectively. The value of G' , related to the stress in phase with strain, provides information about the elasticity of the gel. The loss modulus, G'' , related to the stress out-of-phase with the strain, is a measure of the dissipated energy in the system [7]. As an example of results obtained by this method, the response, measured from a model material (silicone) and brain tissue, is shown in Figure 5.11c.

5.4 Mechanical Properties of Cells

5.4.1 Mechanical Behavior of Blood Cells

Cells are complex, deformable objects. Red blood cells are slightly larger than the smallest capillaries and therefore must deform in order to move through the circulation (Table 5.2). White cells are substantially larger and less deformable than red cells; therefore, they can have a larger impact on blood flow properties than one would predict from their abundance. As will be discussed in Chapter

Table 5.2
Characteristics of Blood Cells

Cell type	Shape	Characteristic dimensions (μm)	Volume (μm^3)	Cell fraction	Cortical tension (mN/m)
Red cell (erythrocyte)	Biconcave disc	$7.7 \times (1.4 \text{ to } 2.8)$	96	0.997	
Platelet	Biconcave disc	2×0.2	5 to 10	0.007	
Neutrophil	Spherical	8.2 to 8.4	300 to 310	< 0.002	0.024 to 0.035
Lymphocyte	Spherical	7.5	220	< 0.001	0.06
Monocyte	Spherical	9.1	400	< 0.0005	0.035

Data obtained from [8].

10, the mechanical properties of white cells have a profound influence on their fate in the circulation after infusion. A reduction in the deformability of white cells may also play a role in certain diseases, such as leukemia and diabetes, in which the microcirculation can be impaired. The mechanical properties of red and white blood cells have been well studied and are reviewed in [8].

The mechanical properties of many cells—including blood cells—have been directly measured by aspiration into a micropipette (Figure 5.12). The properties of the cell are deduced from the deformations observed as the cell is pulled into the pipette under gentle pressure [9, 10]. For example, in one method, the suction pressure is gradually increased to a critical pressure, $\Delta p_{\text{critical}}$, at which a small hemispherical section of the cell is pulled into the pipette. Laplace's law permits calculation of cell cortical tension, T , from the critical pressure:

$$T = \frac{\Delta p_{\text{critical}} R_p R_c}{2(R_c - R_p)} \quad (5-52)$$

where R_p is the radius of the micropipette and R_c is the radius of the cell [11]. Similarly, the overall viscosity of a cell can be measured by aspirating the whole cell into a larger micropipette ($\sim 4 \mu\text{m}$ radius for neutrophils) and comparing the time course for cell movement through the pipette lumen to numerical models of individual cell deformation [11].

Other techniques have been used to measure the mechanical properties of individual cells, by deforming them between parallel plates, for example, or by applying smaller mechanical probes such as microneedles [12] and atomic force microscopes [13].

5.4.2 Mechanical Properties of the Cytoskeleton

The intracellular fluid, or cytoplasm, is a much more complicated fluid than plasma. Plasma, or the acellular fraction of blood, is a concentrated protein solution that behaves as a Newtonian fluid with a viscosity of 1.2 cP (or 1.2 mPa-s) at 37°C. The simplest cytoplasm is probably found in red blood cells, which have no nucleus or internal organelles. Red cell cytoplasm is a

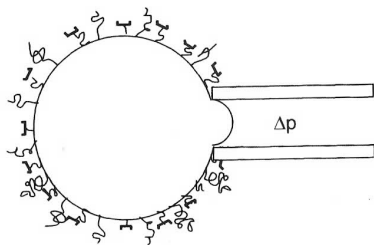


Figure 5.12. Micropipette aspiration to determine cellular mechanics. Gentle suction can be used to deform cells under controlled conditions for evaluation of mechanical properties.

concentrated he
390 g/L of hemo

The basic el
which the shape
plasm. The thre
3.12: actin mic
filaments have i
a persistence len
In some cases,
measured direct
of filament mec
filaments differ
flexible but una
actin filaments
more flexible th

Within the
ling greatly enh
the ability to bu
properties of the
filaments. Many
process; actin fi
agents (α -actinin
of tools for dyn
behavior is obser
teins; in the case
is observed in r
general, solutio
characterized us

New metho
tial to study the

5.4.3 Influence

Cells respond to
been known th

fluence on their
nability of white
and diabetes, in
properties of red
d in [8].
cells—have been
) The properties
cell is pulled into
thod, the suction
at which a small
ace's law permits
e:

(5-52)

s of the cell [11].
rating the whole
and comparing
en to numerical
cal properties of
r example, or by
and atomic force

ated fluid than
entrated protein
y of 1.2 cP (or
d in red blood
cytoplasm is a

concentrated hemoglobin solution; individual cells contain between 290 and 390 g/L of hemoglobin, which has a viscosity of between 4.2 to 17.2 cP [8].

The basic elements of the cytoskeleton were introduced in Section 3.3.1, in which the shape of a cell was associated with protein filaments in the cytoplasm. The three most important protein filaments are illustrated in Figure 3.12: actin microfilaments, microtubules, and intermediate filaments. These filaments have intrinsic mechanical properties. They can be characterized by a persistence length, which is related to the stiffness of the filament (Table 5.3). In some cases, the mechanical properties of individual filaments have been measured directly, although the measurements are difficult and a clear picture of filament mechanics is not yet available. But it is clear that the three main filaments differ greatly in mechanical properties: actin microfilaments are flexible but unable to withstand tensile forces; microtubules are stiffer than actin filaments but still have poor tensile strength; intermediate filaments are more flexible than actin and stronger under tension [14].

Within the cytoplasm, individual filaments aggregate into bundles; bundling greatly enhances the mechanical strength of the system. In addition, once the ability to bundle and unbundle filaments is regulated, the local mechanical properties of the cytoplasm can be controlled via assembly and disassembly of filaments. Many cytoplasmic constituents participate in the filament assembly process; actin filaments, for example, are crosslinked by at least four different agents (α -actinin, spectrin, fimbrin, and villin), providing the cell with a variety of tools for dynamically regulating filament assembly. Complex mechanical behavior is observed in simple systems of filaments and filament-binding proteins; in the case of actin filaments and α -actinin, complex rheological behavior is observed in reconstituted samples of purified cytoskeletal elements [15]. In general, solutions of cytoskeletal filaments are viscoelastic and have been characterized using the rheological methods described in Section 5.3.3.

New methods, such as laser tracking microrheometry [16], offer the potential to study the mechanical behavior of cytoplasm within living cells.

5.4.3 Influence of Mechanical Forces on Cell Structure and Function

Cells respond to forces by deformation, as described above. It has also long been known that cell behavior (such as growth control) is dependent on cell

Table 5.3
Characteristics of Cytoskeletal
Filaments

Filament	$L_p(\mu\text{m})$
Actin microfilament	18
Intermediate filament	2
Microtubule	6,000

Data obtained from [14].

Gentle suction
n of mechanical

shape [17], which suggests that the mechanical forces that act to deform cells (and therefore alter cell shape) can also influence the biochemical function of cells.

Cells of the vascular system are constantly exposed to the forces provided by blood flow and, therefore, it has been logical to study these cells for the impact of fluid shear forces on cell function. It is now well established that shear stress can 1) influence the morphology and orientation of cultured vascular cells; 2) induce changes in signal transduction systems and secondary messengers; 3) influence metabolite secretion; and 4) regulate gene expression (see review [18]). Similarly, mechanical forces that are provided through mechanisms other than from fluid flow—such as stretching of a deformable substrate on which cells are attached—can also influence cell function. Molecular mechanisms for mechanotransduction—that is, conversion of the mechanical force into a biological signal—are not clearly established, but it is likely that multiple mechanisms are involved, since cells encounter mechanical forces from many different sources. Mechanical forces can influence cellular function within tissues, as well; for example, the expression of genes in cells within brain slices is differentially regulated by controlled mechanical strains [19].

5.5 Mechanical Properties of Tissues

The mechanical properties of engineered tissues are often critical for their function (recall Figure 2.1 and related discussion). This section briefly reviews the mechanical behavior of tissues by focusing on the mechanical properties of bone, soft tissues, and tissue-engineered materials. The role of biomechanics in the evaluation of tissue-engineered materials has been reviewed [20].

5.5.1 Bone

Bone is a hard (that is, mechanically strong) tissue that is composed of a mineral phase (60%), a collagen-rich matrix (30%), and water (10%) [21]. The mineral phase appears to provide the stiffness to bone, while the matrix phase provides other properties that are still not well defined [21]. Bone has two typical architectures, cortical (or compact) bone and cancellous (or trabecular) bone, which differ in microscopic structure and mechanical properties (Table 5.1). The dense, rigid cortical architecture is found in long bones whereas the cancellous architecture, which is more porous and oriented, is found in the ribs, spine, and epiphysis. A comprehensive review of those biomechanical properties of bone that are relevant for tissue engineering is available [21].

5.5.2 Soft Connective Tissues

Soft connective tissues surround our organs, provide structural integrity, and protect them from damage. In most soft connective tissues—such as articular cartilage, tendons, ligaments, dermis of skin, and blood vessels—cells are sparse-

ly distributed. The mechanical properties of these tissues are discussed in Chapter 6.

Soft connective tissues are composed of a material that is often called the extracellular matrix [22]. Figure 5.1 illustrates that connective tissues are composed of parallel bundles of fibers with large changes in stiffness or mechanical properties when the fibers are normally aligned. When forces are applied, the tissue architecture is also influenced.

5.5.3 Tissues

Many tissues are composed of a three-dimensional network of fibers.

Figure 5.1 illustrates that connective tissues are composed of parallel bundles of fibers with large changes in stiffness or mechanical properties when the fibers are normally aligned. When forces are applied, the tissue architecture is also influenced.

ly distributed within an extracellular matrix that provides the mechanical property of the tissue. The molecular constituents of extracellular matrix are reviewed in Chapter 6 (see Section 6.4). In contrast to bone, which is a rigid material, soft connective tissues are typically flexible and deformable (Table 5.1).

Soft tissues are often viscoelastic, behaving like a reinforced composite material in which the structure and orientation of the fiber-reinforcing phase (often collagen and elastin fibers) determines the bulk mechanical behavior [22]. Figure 5.13 shows schematically the behavior of skin—a typical soft tissue that consists predominantly of connective tissue and shows an orientation parallel to the skin surface. With small tensile deformations (phase I in Figure 5.13), the tissue behaves as an elastic material; microscopically, collagen fibers within the tissue are deforming without stretching or bringing about large changes in structure. As the strain increases (phase II), collagen fibers become deformed, straightening in the direction of the strain and increasing the stiffness of the skin. With increased loads, in deformations that are just less than the ultimate tensile strength (phase III), the collagen fibers are individually aligned in the direction of the applied load, and stretched. This illustration demonstrates some of the complexity of the interplay between mechanical forces, tissue structure, and tissue function. Figure 5.13 shows that molecular architecture is influenced by strain; the functional properties of the tissues are also influenced by these changes.

5.5.3 Tissue-Engineered Materials

Many tissue-engineered materials consist of a population of cells embedded into a three-dimensional extracellular matrix. Chapter 1 illustrated this concept

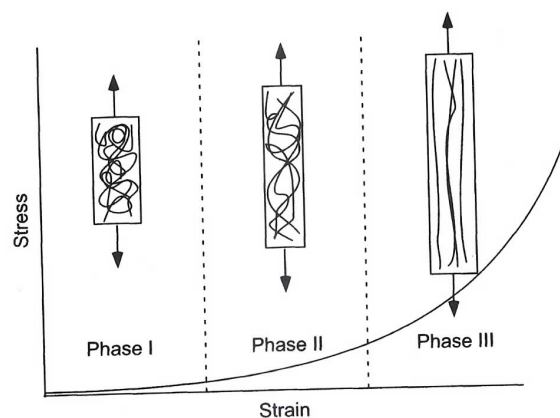


Figure 5.13. Mechanical behavior of soft tissues. The mechanical behavior of soft tissues is illustrated schematically for skin, in which fibers that are oriented parallel to the skin surface provide a nonlinear relationship between stress in the material and deformation. Redrawn from [22].

as a natural extension of conventional cell culture, in which cells are maintained in a three-dimensional environment that permits interaction of cells with the matrix material and with other cells (Figure 1.1).

In Chapter 2, the use of hybrid materials—containing both cells and matrix components, which might be natural or synthetic polymers—for tissue repair was also introduced (Figure 2.3). When cells are embedded into a matrix material, they can interact with the matrix. This interaction may change the functional state of the cells, but they may also alter the physical properties of the matrix by changing the amount or orientation of the matrix materials or by exerting forces on components of the matrix that serve to orient or disrupt the overall architecture.

This phenomenon has been studied in model cell culture systems, sometimes called tissue equivalents [23] or reconstituted model tissues [24], in which cells are suspended within a tissue-like extracellular matrix. When fibroblasts are cultured within a collagen matrix, for example, the cells will attach to and then deform collagen fibers; this action can be observed macroscopically because the mechanical action of the cells causes the gel material to change shape and, in the particular case of fibroblasts in collagen, become more compact. The dynamics of the compaction process reveal the action of cells within the bulk material (Figure 5.14). Usually a lag period is observed first, in which the matrix is forming around the cells. During the early phase of compaction, which usually lasts for the first 24 hours of culture, the cells form attachments to the matrix material and begin to contract it slowly; strains during this phase are typically small, $\sim 10\%$ [25]. Over the first week or two in culture, the cells will steadily compact the matrix materials by a process of adhesion to fibers and mechanical contraction by the cells;

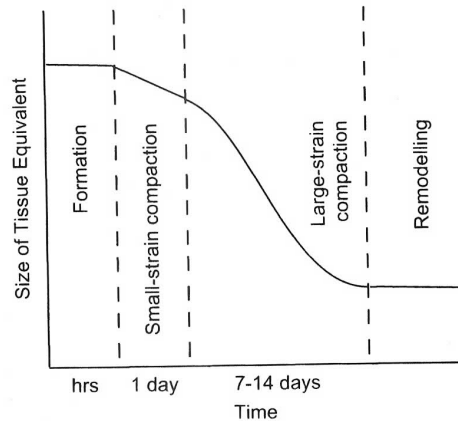


Figure 5.14. Dynamic compaction of a tissue equivalent. Cells can be suspended within a matrix material to produce a three-dimensional tissue equivalent, which will undergo mechanical and geometric changes during maintenance in culture.

are main-
f cells with

cells and
for tissue
to a matrix
change the
properties of
materials or by
disrupt the

ms, some-
es [24], in
rix. When
e cells will
ed macro-
material to
n, become
e action of
s observed
early phase
e, the cells
it slowly;
r the first
c materials
y the cells;

strains during this phase can be 50 to 90% [26]. In the final phase, there is no further compaction of the material, but it can become substantially stiffer due to the action of the cells, which may secrete additional matrix proteins or cause crosslinking of existing matrix materials. The extent and the time course of compaction depend on the density of cells in the material, the type of cells that are suspended, and the nature of the material and extracellular environment. In all stages of this process, the cells may be modifying the matrix (by stiffening it, for example) and, at the same time, the matrix may be inducing changes in the structure and function of the entrapped cells (as described in Section 5.4.3).

5.5.4 Example: Compression of Cellular Aggregates

Some of these mechanical principles can be illustrated with a practical example of importance to tissue engineering. (The importance of cellular aggregates in tissue engineering will be elaborated in Chapter 8.) As described in Chapter 3, cellular aggregates and tissue fragments have been powerful models for developmental biology. Force relaxation experiments on embryonic tissue fragments, which are allowed to round-up into spherical aggregates in culture, show a characteristic behavior (Figure 5.15a); in response to a fixed deformation, the aggregate exerts a resisting force which declines with a rapid initial rate, followed by a slow, more persistent relaxation [27]. This relaxation is similar to the behaviors illustrated in the spring and dashpot models of Figure 5.10—especially the behavior in panel c (rapid initial response and persistent continuing response) and panel d (slow approach to some limiting response). But the behavior here is more complicated in that two rates of relaxation, or characteristic time constants, are observed.

The simplest model that can incorporate all of the behaviors observed in the dynamics response of living tissue segments to compression is the Kelvin model (Figure 5.15b). In this model, a third element—the slide wire, indicated in the figure with its parameter σ —is used to indicate the equilibrium shape of the aggregate under the compressive force. A force balance on the elements shown in Figure 5.15b yields

$$F + \left(\frac{\eta_1}{\mu_1} + \frac{\eta_2}{\mu_2} \right) \frac{dF}{dt} + \frac{\eta_1 \eta_2}{\mu_1 \mu_2} \frac{d^2 F}{dt^2} = \sigma U_0 \quad (5-53)$$

which can be solved to obtain

$$F = (\sigma + \mu_1 e^{-\mu_1 t / \eta_1} + \mu_2 e^{-\mu_2 t / \eta_2}) U_0 \quad (5-54)$$

This model was compared to force relaxation behavior observed in tissues taken from different regions of the embryo; the mechanical behavior of the living tissue depends on composition (Table 5.4).

ended
which will

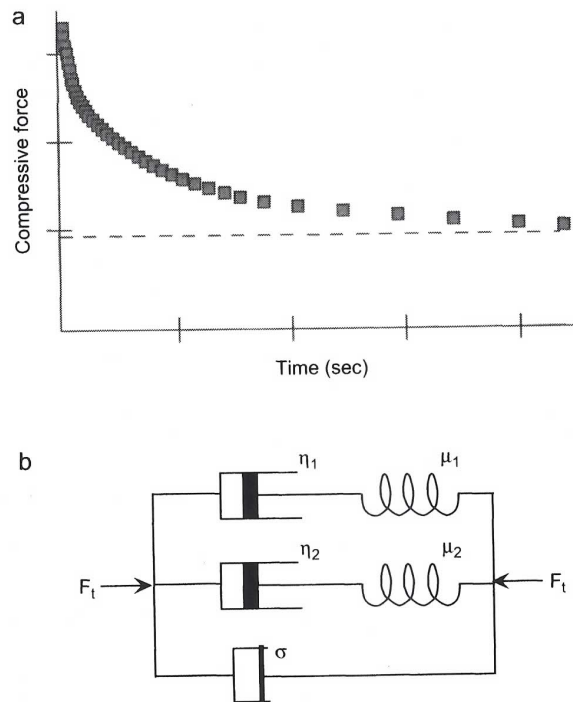


Figure 5.15. Compression of cellular aggregates formed from embryonic tissues. Stress relaxation during compression of the cellular aggregate: experimental data (a) and mechanical model (b). Adapted from [27].

Table 5.4
Viscoelastic Properties of Embryonic Tissues

Tissue Type	μ_1 (dyne/cm)	η_1 (dyne-s/cm)	η_1/μ_1 (s)	μ_2 (dyne/cm)	η_2 (dyne-s/cm)	η_1/μ_1 (s)	σ (dyne/cm)
Neural retina	3.4	7.3	2.2	2.1	55	27	1.6
Liver	4.0	7.6	1.9	2.3	50	23	4.6
Heart	5.1	9.5	1.9	2.5	66	26	8.5
Limb	8.6	24	2.7	7.7	340	45	20

Values for the parameters of the Kelvin model were obtained by measuring force relaxation in aggregates from embryonic tissues. Original data reported in [30].

Summary

- For small deformations, the mechanical behavior of cells and tissues can be described by linear viscoelastic models.
- Fluids are modeled as Newtonian or non-Newtonian, depending on whether the response is a constant or time-dependent.
- The physical basis for the mechanical behavior of cells and tissues is an important topic for future research and development.
- Many biological processes, such as cell migration, are purely mechanical in nature and can be predicted using mechanical models.
- Rheology is the study of the flow and deformation of materials, and it is an important tool for analyzing the mechanical behavior of cells and tissues.
- Cells exhibit a wide range of mechanical properties, and these properties can be used to study the mechanical behavior of cells and tissues.
- The mechanical behavior of cells and tissues is a complex phenomenon that involves many biological and mechanical factors.

Exercises

Exercise 5.1

In a radial flow viscometer, a fluid is sheared between two concentric cylinders. The inner cylinder is fixed, and the outer cylinder rotates. Assuming laminar flow, show that the shear stress τ at the center is $\tau = 3\eta\dot{\gamma}$, where η is the viscosity.

Exercise 5.2

Consider the following:

- Read F

Summary

- For small strains, many solid materials deform elastically; the mechanical properties of these are similar to those of an ideal spring and can be described by an elastic modulus E .
- Fluids deform continuously under the action of a shearing force; for Newtonian fluids, the viscosity, or ratio of shear force to shear rate, is a constant.
- The physics of fluid flow can be quantified for certain geometries. In the case of steady laminar flow through a cylindrical tube—which is an important flow in biological systems—the behavior of the flow is well described and predictable.
- Many biological materials exhibit both elastic and viscous properties; these materials are called viscoelastic. Mechanical models that combine purely elastic and purely viscous components can sometimes be used to predict their deformation behavior.
- Rheology, or the study of deformations in materials, can be used to analyze the mechanical behavior of biological fluids and gels. Examination of the rheological properties often gives insight into the molecular properties of the material.
- Cells exhibit complex mechanical behaviors and respond in complicated ways to mechanical forces.
- The mechanical properties of many tissues are important for their biological function; the development of methods for biomechanical analysis is an important part of tissue engineering.

Exercises

Exercise 5.1

In a radial flow detachment assay (a device for measuring cell adhesion to a surface, which is discussed in more detail in Chapter 6), cells attached to a planar, cylindrical surface are exposed to a steady fluid flow which originates at the center of the surface ($r = 0$) and flows toward the outer edge ($r = R$). Assuming laminar flow through the gap between two stationary surfaces, one containing the adherent cells and the other a uniform height (h) above it, show that the shear stress at the surface depends on radial position from the center: $S = 3Q\mu/\pi rh^2$, where Q is the volumetric flow rate and μ is the fluid viscosity.

Exercise 5.2 (provided by Song Li)

Consider the optimum design of vascular network.

- Read Fung, Y. C. *Biodynamics: Circulation*, Sections 3.1–3.4 [3].

of tissues.
Experimental data (a)

μ_1 (s)	σ (dyne/cm)
27	1.6
23	4.6
26	8.5
45	20

of relaxation in

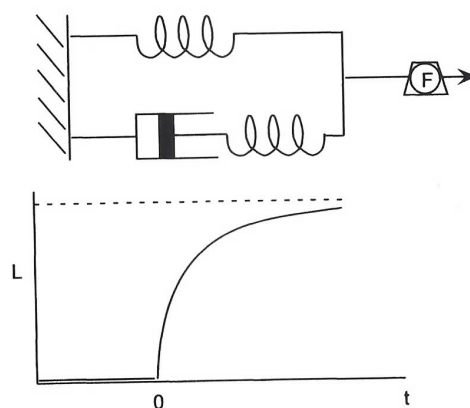


Figure 5.16. Kelvin model of viscoelastic materials.

References

1. Fung, Y.-C., *Biomechanics: Mechanical Properties of Living Tissues*, 2nd ed. New York: Springer-Verlag, 1993.
2. Ozkaya, N. and M. Nordin, *Fundamentals of Biomechanics: Equilibrium, Motion, and Deformation*, 2nd ed. New York: Springer-Verlag, 1999.
3. Fung, Y.C., *Biodynamics: Circulation*, 2nd ed. New York: Springer-Verlag, 1997, 571 pp.
4. Bird, R.B., W.E. Stewart, and E.N. Lightfoot, *Transport Phenomena*. New York: John Wiley & Sons, 1960, 780 pp.
5. Welty, J.R., C.E. Wicks, and R.E. Wilson, *Fundamentals of Momentum, Heat, and Mass Transfer*, 3rd ed. New York: John Wiley & Sons, 1984.
6. Kuntz, R.M. and W.M. Saltzman, Neutrophil motility in extracellular matrix gels: mesh size and adhesion affect speed of migration. *Biophysical Journal*, 1997, **72**, 1472–1480.
7. Prud'homme, R.K., Rheological characterization, in *Electronic Materials Handbook*. Ohio: ASM International, 1989.
8. Waugh, R.E. and R.M. Hochmuth, Mechanics and deformability of hematocytes, in J. D. Bronzino (ed.), *The Biomedical Engineering Handbook*. Boca Raton, FL: CRC Press, 2000, pp. 32-1–32-13.
9. Evans, E. and P. La Celle, Intrinsic material properties of the erythrocyte membrane indicated by mechanical analysis of deformation. *Blood*, 1975, **45**, 29–43.
10. Evans, E. and R. Hochmuth, A solid-liquid composite model of the red cell membrane. *Journal of Membrane Biology*, 1977, **30**, 351–362.
11. Tsai, M.A., R.E. Waugh, and P.C. Keng, Passive mechanical behavior of human neutrophils: effects of colchicine and paclitaxel. *Biophysical Journal*, 1998, **74**, 3282–3291.
12. Felder, S. and E.L. Elson, Mechanics of fibroblast locomotion: quantitative analysis of forces and motions at the leading lamellas of fibroblasts. *Journal of Cell Biology*, 1990, **111**, 2513–2526.

13. Rotsch, C., K. Jacobson, and M. Radmacher, Dimensional and mechanical dynamics of active and stable edges in motile fibroblasts investigated by using atomic force microscopy. *Proceedings of the National Academy of Sciences, USA*, 1999, **96**, 921–926.
14. Bray, D., *Cell Movement*, 2nd ed. New York: Garland Publishing, 2001, 372 pp.
15. Sato, M., W. Schwarz, and T. Pollard, Dependence of the mechanical properties of actin/a-actinin gels on deformation rate. *Nature*, 1987, **325**, 828–830.
16. Yamada, S., D. Wirtz, and S.C. Kuo, Mechanics of living cells measured by laser tracking microrheology. *Biophysical Journal*, 2000, **78**, 1736–1747.
17. Folkman, J. and A. Moscona, Role of cell shape in growth control. *Nature*, 1978, **273**, 345–349.
18. Patrick, C.W., R. Sampath, and L.V. McIntire, Fluid shear stress effects on cellular function, in J.D. Bronzino (ed.), *The Biomedical Engineering Handbook*. Boca Raton, FL: CRC Press, 2000, pp. 114-1–114-20.
19. Morrison, B., et al., Dynamic mechanical stretch of organotypic brain slice cultures induces differential genomic expression: relationship to mechanical parameters. *Journal of Biomechanical Engineering*, 2000, **122**, 224–230.
20. Butler, D.L., S.A. Goldstein, and F. Guilak, Functional tissue engineering: the role of biomechanics. *Journal of Biomechanical Engineering*, 2000, **122**, 570–575.
21. Athanasiou, K.A., et al., Fundamentals of biomechanics in tissue engineering of bone. *Tissue Engineering*, 2000, **6**(4), 361–381.
22. Holzapfel, G.A., Biomechanics of soft tissue, in *Handbook of Material Behavior*. J. Lemaitre (ed.), San Diego, CA: Academic Press, 2000.
23. Barocas, V.H. and R.T. Tranquillo, Biphasic theory and in vitro assays of cell–fibril mechanical interactions in tissue-equivalent gels, in V.C. Mow (ed.), *Cell Mechanics and Cellular Engineering*. New York: Springer-Verlag, 1994, pp. 1–25.
24. Wakatsuki, T., et al., Cell mechanics studied by a reconstituted model tissue. *Biophysical Journal*, 2000, **79**, 2353–2368.
25. Barocas, V.H., A.G. Moon, and R.T. Tranquillo, The fibroblast-populated collagen microsphere assay of cell traction force. Part 2. Measurement of the cell traction coefficient. *Journal of Biomechanical Engineering*, 1995, **117**(2), 161–170.
26. Moon, A. G. and R. T. Tranquillo, Fibroblast-populated collagen microsphere assay of cell traction force: Part 1. Continuum model. *AIChE Journal*, 1993, **39**, 163–177.
27. Beysens, D.A., G. Forgacs, and J. A. Glazier, Embryonic tissues are viscoelastic materials. *Canadian Journal of Physics*, 2000, **78**, 243–251.
28. Brands, D.W.A., Predicting brain mechanics during closed head impact. Eindhoven: Technische Universiteit Eindhoven, 2002.
29. Ratner, B., et al. (eds.), *Biomaterials Science: an Introduction to Materials in Medicine*. San Diego, CA: Academic Press, 1996.
30. Forgacs, G., et al., Viscoelastic properties of living embryonic tissues: a quantitative study. *Biophysical Journal*, 1998, **74**, 2227–2234.

Cell Ad

I am a k

The external
a carbohydra
within the gl
a net negativ
form the gly
these compo
brane-associ
abundant. P
carbohydrat
with the ext

Cells ca
geometricall
fiber, or an
a cell and a
are consider
and Chapt
consequenc
(Cell Migra

Since tl
ment and s
are fundam
important
aggregation
port. Cell
methods a
tion.

The n
specific as
mentary n
exist free
cellular m