Lecture 17: Cell Mechanics

We will focus on how the cell functions as a mechanical unit, with all of the membrane and cytoskeletal components acting as an integrated whole to accomplish a mechanical function. We will use two different types of blood cells as case studies.

Short Intro to Blood Cells

White blood cells (leukocytes) are a broad class of cells in the blood that participate in a variety of functions, including the immune response, the inflammatory response, and wound healing. These cells are generally spherical in suspension.

Red blood cells (erythrocytes) are responsible for transporting oxygen throughout the body. These cells assume a biconcave shape in suspension culture.

Both of these cell types have to undergo large deformations to squeeze through the small diameter capillaries of the circulatory system. We will explore how each of these cell types handles this challenge.

White Blood Cells

In a simplified view of a white blood cell squeezing into a small diameter capillary, we see that it changes from spherical cell to a sausage-shaped cell. The volumes of these two shapes are the same, since the cytoplasm of the cell is incompressible. Thus, the surface area of the cell increases dramatically when it enters the capillary. However, we know that the plasma membrane can only tolerate a 4% increase in area before lysis. How does the white cell handle this? By having excess membrane area, in the form of folds and microvilli in the plasma membrane. Osmotic swelling studies show that the apparent surface area of a neutrophil (one type of white blood cell) at lysis is 2.6 times the apparent surface area under isotonic conditions.

How does the white cell maintain a spherical shape with all this excess membrane area? There is a tension in the cortical actin layer that pulls the cell into a spherical shape, similar to surface tension pulling a water drop into a sphere. This cortical tension also plays an important role in many white cell functions, including, for example, phagocytosis.

We can study this cortical tension using a variety of techniques. One common method is micropipette aspiration. In this technique, a small diameter glass pipet is brought into contact with a cell. A known suction pressure is then applied within the pipette, causing an aspiration of the cell into the pipette. By measuring the length of aspiration, we can measure several important cell mechanical properties, including cortical tension.

For white cells, we can analyze this type of experiment using the Law of Laplace. This is a relationship between the surface tension and pressure within a fluid drop that has a membrane with surface tension in it. Here we will derive the Law of Laplace for the simple case of a spherical drop of fluid with an internal pressure (P_c, with units of force per area) and a uniform surface tension (T_c, with units of force per length). A free body diagram for half of the drop, including both the surface and the internal fluid, is shown in Fig. 1. If we examine a summation of forces in the x-direction, we get

$$\Sigma F_x = 0 = P_c \left(\pi R_c^2\right) - T_c \left(2\pi R_c\right) \tag{1}$$

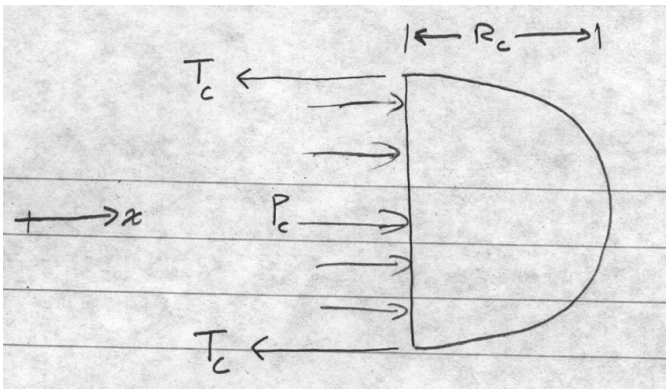

Figure 1:

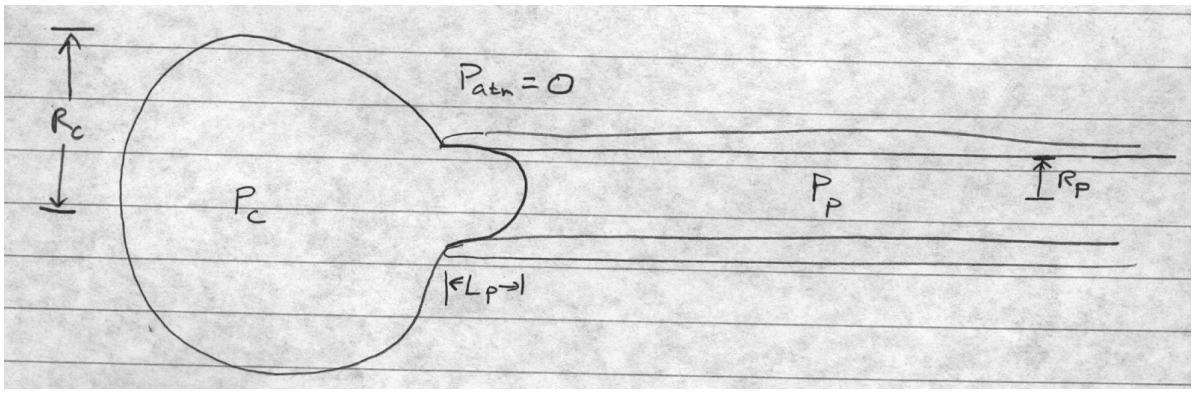

Figure 2:

which yields the relationship

$$P_c = \frac{2T_c}{R_c} \tag{2}$$

This is the Law of Laplace.

Now we will apply the Law of Laplace to an analysis of micropipette aspiration. We will assume that the cell has been aspirated such that the aspiration length (L_p) is equal to the pipet radius (R_p), as shown in Fig. 2. First, we examine a free body diagram of the back half of the cell (Fig. 3). This is the same situation as before, and so we have

$$P_c = \frac{2T_c}{R_c}. \tag{3}$$

However, P_c is unknown. Next, examine the aspirated region of the cell. Because $L_p = R_p$, the aspirated region is a hemisphere. The free body diagram is given in Fig. 4. As I will show in class, the summation of forces in this case is given by

$$\Sigma F_x = 0 = P_c \left(\pi R_p^2\right) + P_p \left(\pi R_p^2\right) - T_c \left(2\pi R_p\right). \tag{4}$$

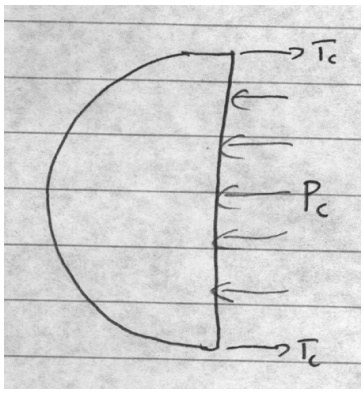

Figure 3:

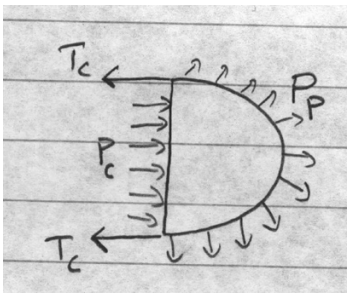

Figure 4:

Using this result and eq. 3, we obtain the relationship

$$P_p = 2T_c \left(\frac{1}{R_p} - \frac{1}{R_c} \right). \tag{5}$$

We can measure P_p, R_p, and R_c, and so we can use this equation to calculate the cortical tension from a micropipette aspiration test.

What happens if the cell is aspirated further into the pipet? Examine what happens to the terms in eq. 5:

- P_p is increased (to draw the cell further into the pipet).
- T_c is constant.
- R_p is constant.
- R_c decreases as more of the cell is drawn into the pipet. Thus, $1/R_c$ increases and $(1/R_p - 1/R_c)$ decreases.

So the left hand side of eq. 5 increases, but the right hand side decreases. This means the cell is no longer in equilibrium, and the cell will be drawn completely into the pipet. This is indeed what is observed experimentally, indicating that the passive white cell behaves as a liquid drop.

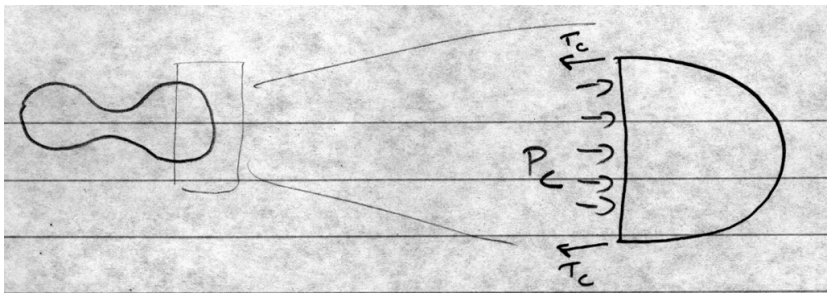

Figure 5:

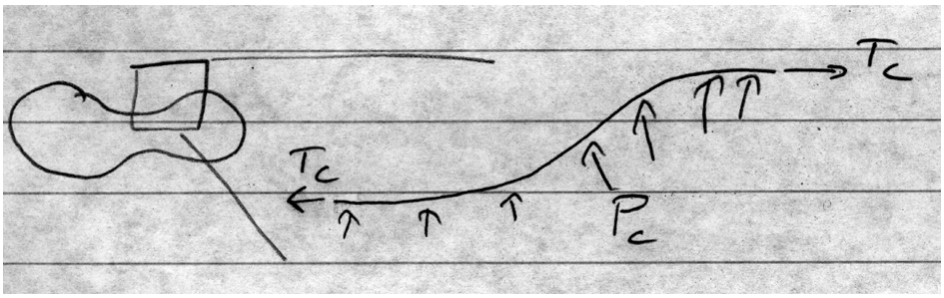

Figure 6:

Red Cells

For the red cell, we return to the initial question of how the cell handles the large deformations experienced during blood flow. Since the red cell does not have any membrane folds, it must use a different strategy than the white cell. The answer lies in the biconcave shape of the red cell.

We now examine the red cell using an analysis based on the Law of Laplace. If we examine one of the ends, we get a free body diagram similar to those seen above for the white cell (Fig 5). However, if we examine a free body diagram of the membrane only for a region near the concavity (Fig 6), we get an interesting result. In this case, there is no force to balance the vertical component of the internal pressure (P_c). Therefore, $P_c = 0$ and consequently $T_c = 0$. So at rest, the red cell is in a stress-free state, while the white cell exhibits a cortical tension at rest.

To address the question of how the red cell handles the deformations of capillary flow, we list two requirements for deformation of red cells during blood flow:

1. There is no expansion of the apparent membrane area. This is because there is no excess membrane area, and so an increase in membrane area of 4% would lead to cell lysis. It is important to note, however, that the membrane can sustain large deformations in bending without increases in area.

2. There is no change in volume, since the cytoplasm within the cell is incompressible.

For a spherical cell, there are no deformations that satisfy both of these criteria. However, for the biconcave red cell, there are an infinite number of deformations. Fig. 7 shows one

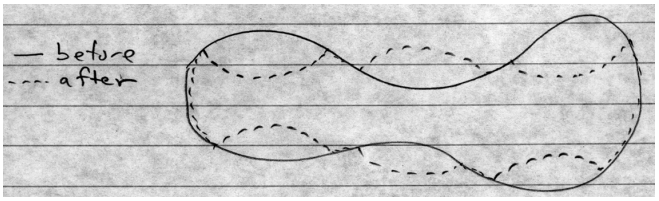

Figure 7:

(somewhat goofy) example. Thus, the shape of the red cell allows it to undergo large and complex deformations without sustaining large strains or generating large membrane stresses.

Cytoskeleton

The fact that the red cell membrane is in a stress-free state at rest indicates that the biconcave shape of the membrane is its natural resting shape. This resting shape is determined by the underlying membrane cytoskeleton. In the red cell, there is a network of long spectrin filaments crosslinked by short actin filaments. Along with many other proteins, this is the network that gives the red cell membrane its resting shape and unique mechanical properties. In contrast, the white cell has a cortical actin network similar to many other cell types, in part because it needs to activate the motility machinery on short notice a a site of inflammation or infection.

References

I used the following two references extensively in preparing this lecture:

1. Fung YC: *Biomechanics: Mechanical Properties of Living Tissues.* New York, Springer-Verlag, 1993
2. Hochmuth RM: Micropipette aspiration of living cells. *J Biomech* 33:15-22, 2000

Problems

1. A white cell with an initial diameter of 8 μm in resting suspension culture is completely drawn into a pipet with an inner diameter of 2.5 μm.

   (a) What is the change in the apparent surface area of the cell before and after aspiration?

   (b) How can this technique be used to measure the amount of excess membrane area in the cell?

2. Under micropipet aspiration, red cells do not flow fully into the pipet like white cells. Instead, red cells membranes exhibit a *shear elasticity* that allows it to resist aspirations where $L_p > R_p$. In this way, red cells, like most biological materials, exhibit characteristics of both solid and fluid materials. This deformation has been analyzed

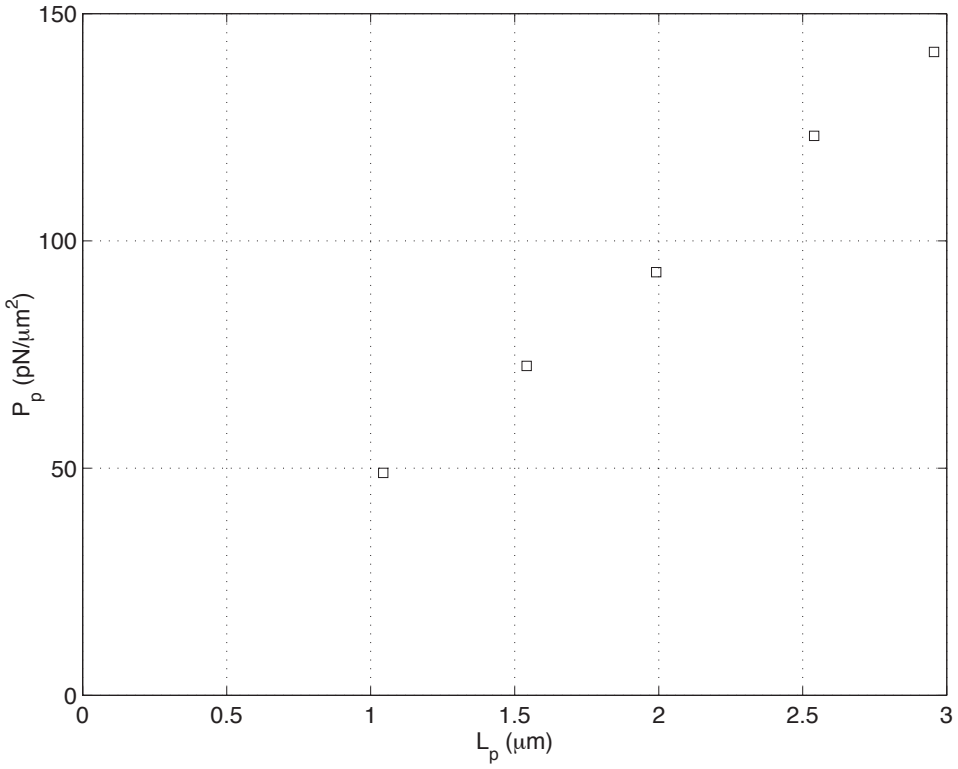

Figure 8:

under the assumption of constant membrane area; Chien et al. (*Biophys J*, 24:463, 1978) linearized to the result to give:

$$\frac{P_p R_p}{\mu} = 2.45 \frac{L_p}{R_p}, \quad \text{for } L_p > R_p \tag{6}$$

where μ is the shear elastic modulus of the membrane.

(a) Use eq. 6 to determine the shear elastic modulus of a red cell from the fictitious experimental data shown in fig. 8. Assume that the inner pipet diameter is 1.4 μm.

(b) Based on eq. 6, what is likely to be the largest source of error in measuring μ?