

## Lab 3: Stress, strain, and stiffness of biomaterials and simple biological structures

### Bio427 — Biomechanics

This lab explores experimental methods for quantifying the mechanical properties of biological materials (recall that material properties are independent of structural size and shape). In addition we will take our first foray into structural measurements of biological beams and use beam theory to estimate both the material and structural stiffness. We will be using both plant and animal material in this lab.

#### Goals

- Learn how to measure force and displacement so that you can construct a stress-strain curve
- Understand and measure the second moment of Area
- Understand and estimate the flexural stiffness ( $EI$ ) of beams from their deflection
- Explore the concept of anisotropy

#### Conceptual Basis

Last week we explored in lecture the concepts of stress ( $\sigma$ ), strain ( $\epsilon$ ), stiffness ( $E$ , Young's modulus) and strength ( $\sigma_{\max}$ ). These are all material properties and represent biomechanical ways to characterize materials living systems. These physical quantities are summarized here:

$$\sigma = \frac{F}{A} \quad (1)$$

$$\epsilon = \frac{\Delta L}{L_0} \quad (2)$$

$$E = \frac{d\sigma}{d\epsilon} \quad (3)$$

And for a linearly elastic materials (having a constant  $E$ )

$$E = \frac{\Delta\sigma}{\Delta\epsilon} \quad (4)$$

where  $F$  is the applied force,  $A$  is the cross-sectional area over which that force operates,  $\Delta L$  is the change in length in response to that force, and  $L_0$  is the initial length of the specimen. Young's modulus ( $E$ ) is the slope of the stress-strain relationship which is constant for

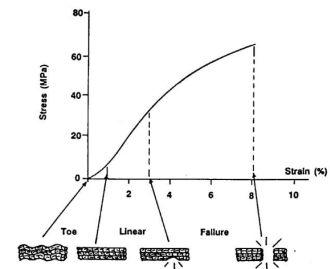


Figure 1: Stress-strain relationship for ligament: from Lee and Hyman, Modeling of failure mode in knee ligaments depending on the strain rate. BMC Musculoskeletal Disord. 2002 doi: 10.1186/1471-2474-3-3

Hookean materials (that is, they obey Hooke's Law). However, as we may learn in this lab, many biological materials are non-Hookean and the relationship between stress and strain is nonlinear. Moreover, biomaterials may also show *anisotropic behavior* – that is, the relationship between the stress and strain depends on the direction in which the load is applied. For example, any biomaterial containing fibers with some average axial orientation will show some level of anisotropy. Examples of biomaterials with containing fibers include skin, intestine, muscle epimysium, wood, plant stems and many others.

In addition to nonlinear and anisotropic behaviors, many biomaterials are viscoelastic (as we have discussed in lecture), showing a time-dependence in their their response to loads. There are a host of methods available for quantifying time-dependent properties including creep tests (constant force: isotonic loads), stress relaxation tests (constant length: isometric loads) as well as cyclic dynamic loads.

### *Beam bending and the second moment of area*

While we can use very simple loading regimes to explain stress, strain, stiffness and strength, in practice many biological structures are often subject to loads that are not purely tensile or compressive, but which entail a variable distribution of tensile and compressive stresses throughout the structure.

A classic example of a loading regime that creates both tensile and compressive stresses within the loaded structure is beam bending, as we see in Fig 2.

If you were to push down on one end of a beam which was fixed at its other end (which we will call a *cantilever beam*) to the wall, that beam would bend downwards under the applied load. The material on the upper side of the beam will be stretched, and the material on the lower side will be compressed. This implies that there is some region within the material that experiences no stress at all, in between the tensile and compressive zones. This region is a plane within the structure perpendicular to the vector of the applied load, and we call this plane the *neutral plane*.

The neutral plane bisects the cross-section of the beam, such that exactly half of the cross-sectional area of the beam lies on either side of the neutral plane. The way in which the material of the beam is distributed on either side of this neutral axis within the beam determines, along with the stiffness of the material, just how difficult it is to bend the beam. The quantity that relates the deflection of a beam to the applied force is termed the flexural stiffness ( $EI$ ): the product of the material stiffness  $E$  and the second moment of area  $I$ . The

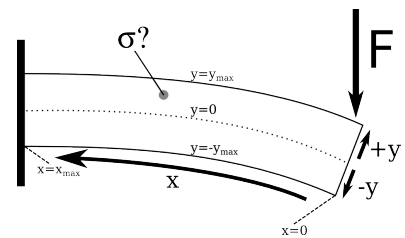


Figure 2: Stress distribution in an end-loaded cantilever beam.

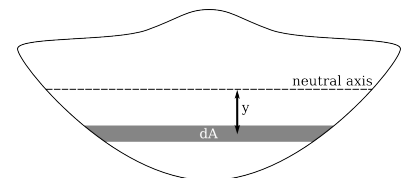


Figure 3: Cross-section of a palm petiole, showing the neutral axis of the beam under an applied vertical load, and a thin region of cross section  $dA$  parallel to the neutral axis displaced by a distance  $y$

second moment of area is defined as

$$I = \int y^2 dA \quad (5)$$

which, in practical terms, means that you take your structure and slice it up parallel to the neutral plane and *add up the sum of all of the areas multiplied by the square of their distance from the neutral plane.*

So, for a given amount of material,  $I$  will be maximized if you can distribute as much of that material as far from the neutral axis as possible. This is why, when we build structures, we often use tubes instead of solid rods: for the same amount of material, with the same stiffness  $E$ , we can vastly increase the flexural stiffness of our structure by using tubes because tubes have a much greater  $I$  than a rod with the same cross-sectional area.

The deflection of a cantilever beam under a load applied at the free end of the beam is given by

$$d = \frac{Fl^3}{3EI} \quad (6)$$

where  $d$  is the deflection,  $F$  is the applied force,  $l$  is the length of the beam, and  $EI$  is the flexural stiffness. Using Eqn. 6, if we measure the deflection of a beam of length  $l$  under a known load  $F$ , we can derive an estimate for the flexural stiffness of the beam,  $EI$ .

### *Methods*

There are two parts to this lab: stress-strain relationships and static bending loads applied to whole structures. In the former you will use force and length measurement to plot the stress strain relationship for some biomaterials and use that relationship to examine the stiffness and the extent to which that material has an anisotropic behavior. In the latter part, you will measure  $I$  and use biological beams subject to bending loads.

### *It's a stretch*

For this part of the lab you will use cyanoacrylate glue (superglue) to mount a piece of animal tissue on a force transducer. The animal tissue we will be using is pig intestine, which is a wonderfully thin and stretchy material typically used by humans as a sausage casing. We will measure the stress-strain relationship of pig intestine by cutting a small strip of the material and stretching it by applying a force to one end. This force will create stress within the material and cause it to elongate (strain). As you modulate the amount of stress applied to the material, you will measure its deformation and

thereby derive its stress-strain curve. You'll repeat this both along the long axis and along the circumference of this tube-tissue to examine whether its stretchiness depends on the direction of loading and is therefore anisotropic.

*Preparing your sample* The pig intestine is obtained from a local butcher and arrives sleeved around a strip of plastic. The material is difficult to work with once it is removed from the plastic core sheet, so you will want to do as much preparation of the sample as possible before cutting away the plastic.

First, determine whether you want to test the material along the axial or circumferential direction – you will test the gut in both directions for this lab, so the order doesn't matter. Next, find a clean section of the intestine and glue two pieces of stiff plastic to the material with a  $\sim 5$  cm wide section of material between them. Try not to glue your fingers to the intestine, or to each other. Once you have glued the plastic tabs to the gut, cut away excess gut beyond the glued section so that you are left with a thin strip of gut glued at either end to the tabs.

*Measuring stress and strain* In this section, you will obtain a stress-strain relationship for your sample by stretching your sample with known weights. Hang the sample from a hole punched into the plastic strip. Now you will hang weights from the free end of the sample to apply a stress. Our weights in this case are metal nuts, which each weigh 12.6 g, and you will suspend these from your sample using a hooked paperclip weighing 0.2 g.

As you add more mass to the free end of the sample, measure the extension of the sample with calipers or a ruler. Plot your results on the worksheet for both an axially-loaded sample and a circumferentially-loaded sample.

### Measuring I

We have provided you with a number of images showing the cross section of various biological structures. Your job will be to determine the second moment of area for these cross sections.

Begin by selecting one of the cross-section printouts that we have provided for you, and choose a direction for the applied load – the direction you choose will affect the location of the neutral axis for your beam, as well as its second moment of area and therefore flexural stiffness, so choose a loading direction you think will be interesting!

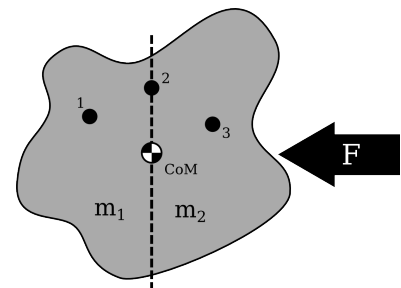


Figure 4: Finding the neutral axis: if a paper clip is inserted at point 1, the cross-section will spin clockwise, because  $m_1 < m_2$ , where  $m_1$  and  $m_2$  represent the mass of the cross section on either side of the pin. If a paper clip is inserted at point 3, the cross section will spin counterclockwise. At point 2, however,  $m_1 = m_2$ , and the cross section will not rotate – you've found the neutral axis!

*Finding the neutral axis* Unlike the exercise we did in class, the neutral axis is not marked on the cross-sections we have given you (partly to give you some flexibility in choosing the direction of loading). Your first task in determining the second moment of area for your cross section, then, will be to find the neutral axis for the direction of loading that you have chosen.

For the thin paper cross-sections we have given you, finding the neutral axis is equivalent to finding the line perpendicular to the direction of the applied load which contains the center of mass of the cross section — see Fig 4.

1. Begin by cutting out your cross section with scissors.
2. Choose your direction of loading, and draw a straight line across your cutout indicating that this is the loading direction (label your line).
3. Hold the cross-section up vertically so that the direction of loading is perpendicular to gravity.
4. Using an unbent paper clip, poke holes through the paper and let go of the cross section, allowing it to rotate around the axle created by the paper clip. Make sure to poke those holes on the upper part of the section to be above the estimated center of mass.
5. When the cross section does not rotate when let go, you have found the vertical line containing the center of mass of the cross section – mark the neutral axis on your sample!
6. Draw lines on the sample at 1 cm intervals, parallel to the neutral axis.
7. Measure the area of each slice – if the edge is irregular, choose the length of the slice using the line that bisects the slice parallel to the neutral axis – and mark this area on your cross section.
8. Add up the areas of each slice multiplied by the square of the distance separating the neutral axis and the middle of each slice to get  $I$ .

### *Measuring $EI$*

In the next part of the lab, we will be applying loads to biological beams (kindly donated by the UW greenhouse!) to measure their deflection under load and hence their flexural stiffness.

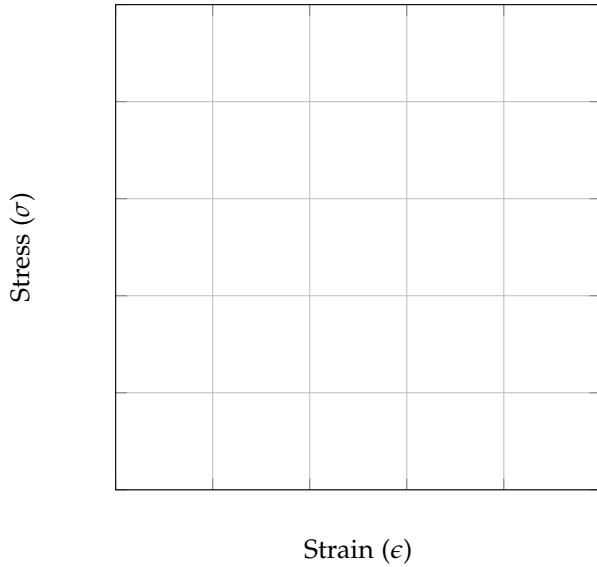
We will have a bunch of cool plant stems to play with; you should measure flexural stiffness for at least two stems.

To measure  $EI$ , take a stem and clamp it against the end of the table with your hand. Use the various machine screw nuts to make weights, and hang these from the free end of your beam to measure the deflection of the end of the beam. Plot your results on the worksheet. Use your measured deflections to estimate the flexural stiffness of your structures. If your structure is not symmetric, try loading your stem in different directions!

*Lab 3: Stress, strain, stiffness, and time-dependent properties*

Lab Section: \_\_\_\_\_

Your Name: \_\_\_\_\_ Partner's Name: \_\_\_\_\_



In the graph above, plot the stress strain curve for your samples. Use the same graph for the stress strain curves for both axial and circumferential loading. Remember to label your axis ticks!

*Stress-strain data*

		Longitudinal	Circumferential
1	Sample length (m)	_____	_____
2	Sample thickness (m)	_____	_____
3	Sample width (m)	_____	_____
4	Cross-sectional area (m <sup>2</sup> )	_____	_____

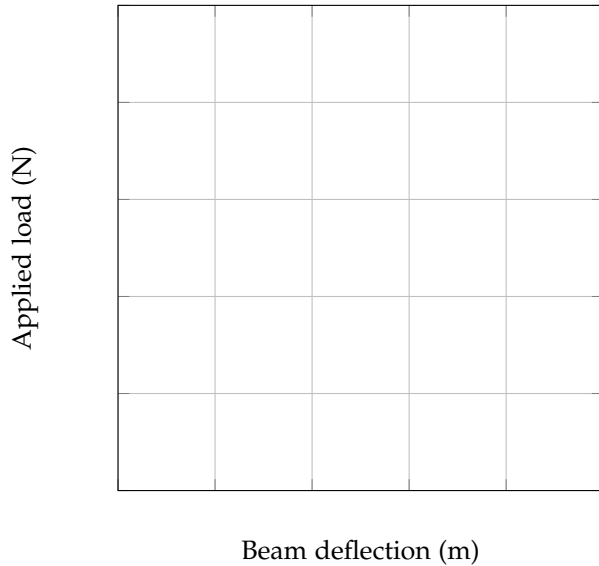
*Second moment of area*

Tape your cross-section to the back of this worksheet, and fill in the following table:

1	Area of cross-section	_____
2	Second moment of area	_____

*Flexural stiffness of plant stems*

For two different stems, measure the deflection of the stem under an applied point load. Use at least 4 different loads, and plot your results for both stems on the axes below. Label each line appropriately.



		Stem 1	Stem 2
1	Name of plant	_____	_____
2	Flexural stiffness	_____	_____

*Questions*

1. What evidence do you have that intestinal lining is anisotropic? What do you think the function of anisotropy is in the function of an intestine?

2. For the beam you measured, provide an estimate of its Young's Modulus. If the beam consisted of an anisotropic material, explain a method by which you could directly measure that anisotropy.