BE172 Spring 2018

Week VI: Viscoelastic Properties of Biological Tissues

Objective

The goal of this lab is to determine the viscoelastic properties of several biological specimens. Experiments will be performed to look at hysteresis, stress relaxation and creep in each specimen. Each of the exponential responses will be fitted to a mathematical function. The constants found from the fitting will be used to determine the properties of a mechanical model (standard linear solid) of the viscoelastic material. Thus it will be possible to gain insight into the structural features of different biomaterials based on the experimental data and mathematical models.

Background

Most biological tissues are viscoelastic, in other words they have properties of both an elastic solid and a viscous fluid. A simple elastic solid can be thought of as an elastic spring. In a linear spring, force is proportional to displacement, or $F=\mu u$, where F and u are the force and displacement, and μ is the spring constant. Similarly, a simple model for a liquid as a purely viscous material is a dashpot, in which force is proportional to velocity, or $F=\eta\dot{u}$, where F and $\dot{u}$ are the force and velocity, and η is the dashpot/viscous constant. Several mechanical models for a viscoelastic material combine these two types of elements, resulting in a force/displacement relationship which is neither elastic nor viscous, but a combination of both.

One simple mechanical model which can produce results that match experimental data is the standard linear solid (also call the Kelvin model) as shown in Figure 1.

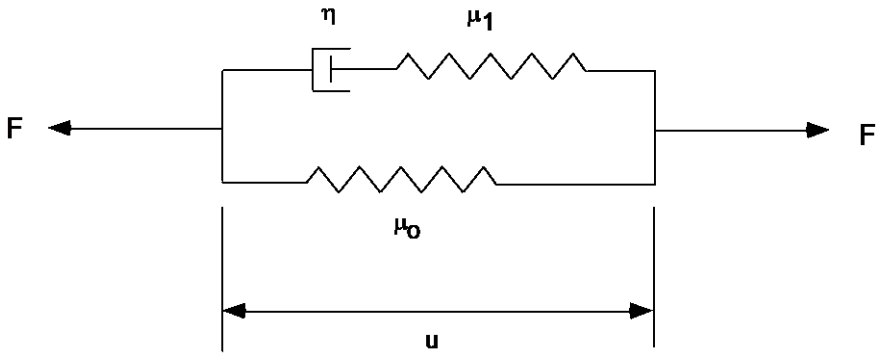

Figure 1: The standard linear solid model

This model combines two springs and one dashpot. By examining the physical combination of elements, one can discern properties of such a model. For example, if a step force is imposed on the solid, it will initially displace to a length dependent on both spring constants. The dashpot acts as a solid connection in response to a step load. After the initial step, the model will continue to stretch due to the dashpot, and eventually reach a steady-state condition in which the dashpot supports no load (why?) and the displacement *u* will only depend on μ_o. This type of response is known as creep, since the length "creeps" even after the initial step change in force/length. A similar analysis can be made if the solid is subjected to a step increase in length. In such a test, the relaxation response shows that

force initially peaks to a value dependent on both springs and the initial displacement, then the force relaxes to a final value determined by μ_o only. Thus the term 'stress relaxation'.

Notice that the governing differential equation for such a system is first order, thus the responses (for both force and displacement) are exponential functions, whose time constants are functions of the spring and dashpot constants. Thus viscoelastic materials will display creep, stress relaxation and hysteresis. Hysteresis is seen in a viscoelastic material as the difference between loading and unloading stress-strain relationships, which may be quantified as the area between the 2 curves (energy dissipation).

Equipment

- Oscilloscope
- Isometric and isotonic force transducers
- Pole/clamp setup
- Computer data acquisition system
- Suture
- Suture-sample interface linkages (bent staples)
- Weights: washers (~0.8 g each), rectangular weights and weight pan
- Rubber band test sample
- Saline in squirt bottle

Tissue

- Rat tissue samples: aorta, esophagus, skin, intestine, trachea

Prelab Questions

- Why do biological tissues in general have viscoelastic properties?
- Write the equations used to convert the measured parameters from the experiment (τ_σ, τ_ε and E_r) to the unknown parameters in the 3-element Kelvin model.
- What does the area between the loading and unloading stress-strain curves represent, in other words, what is the physical cause of hysteresis?
- Explain what happens physically in the Kelvin model when a constant force is applied for a long, long time (i.e. a creep test at t = infinity). What is the value of the creep function (i.e. the displacement) at this time in terms of the Kelvin model parameters?

Experimental Procedure

Set up the experimental apparatus as shown in Figure 2. For calibrations, replace the specimen and 2 sutures by one single suture. Thus the force transducer is coupled directly to the displacement gauge. The metal bar in the displacement gauge should be close to vertical. Notice the two clear elastic stoppers on the bar to prevent the sutures from slipping. All of the components are on the side of the bar facing you. The wiring you will need is shown in Figure 3.

Experimental Methods

(1) Calibrate the force transducer. With the force gauge connected directly to the metal bar of the displacement gauge, balance the force transducer box to read zero output on the oscilloscope with zero load (use the 50g setting on the transducer box). Then hang a known weight on the end of the suture that goes over the pulley, and obtain a calibration factor for force in terms of grams/volt. Remember to make equal lever arms on the metal bar so that the force transmitted to the sample is the same as that hanging on the pulley. Once this calibration is done, make sure the sutures do not move along the metal bar.

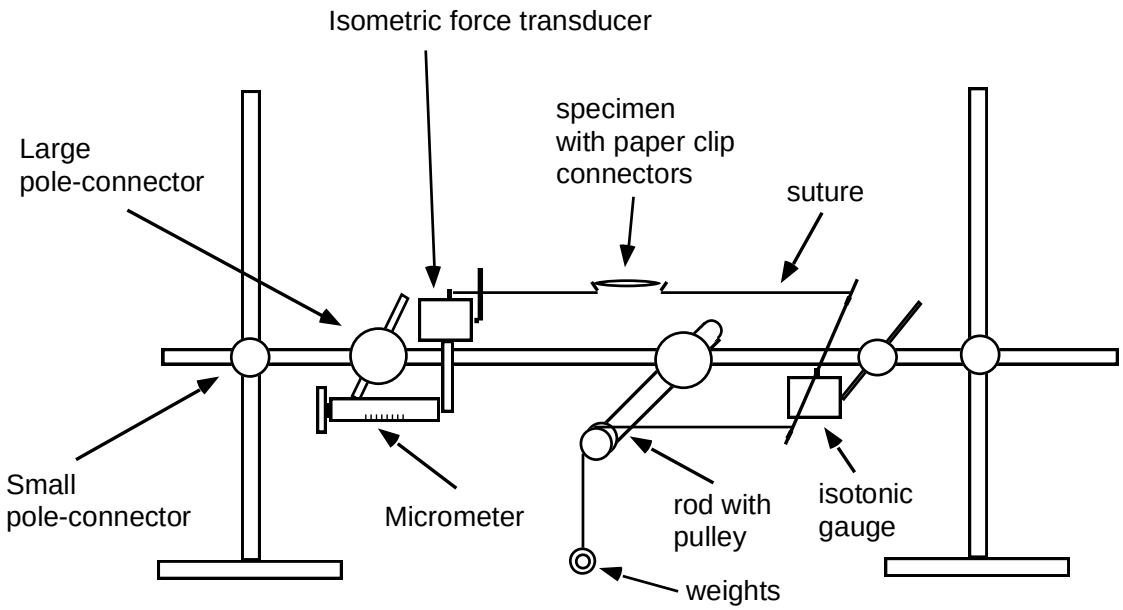

Figure 2: Experimental setup

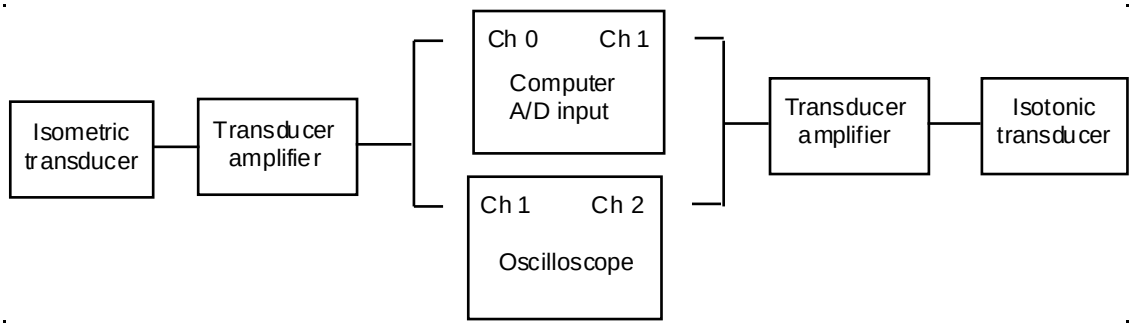

Figure 3: Wiring Block Diagram

(2) Calibrate the length transducer. To calibrate the length (isotonic) transducer, move the micrometer and obtain the voltage change/mm movement. Remember you are interested in displacements, so only relative changes are needed (negative calibration is OK). Obtain a calibration factor for displacement in terms of mm/volt. You only need to displace the specimen 2-3mm at most. Once you have obtained both length and force calibration factors, enter them into the data acquisition program on the computer.

(3) Test the system for both stress relaxation and creep. Use the rubber-band sample "specimen" to perform both a stress relaxation and creep test. Use 2 shorter sutures to replace the one used for calibration. You do not need to permanently tie any sutures with knots! Follow the applicable instructions in steps (4-6) below with the test specimen. You do not need to save any of this data, only to familiarize yourself with the experimental protocol, and try to produce "good" curves (i.e. small oscillations, smooth measurements). At this time you might think about how to perform a hysteresis test with the existing set-up (hint: a continuous displacement loading will be best).

(4) Insert biological specimen. Replace the rubber-band with an actual specimen. Again use 2 shorter sutures and 2 staples to secure the sample. Remember not to permanently tie any sutures with knots to the equipment. Periodically spray or drip saline over the specimen to keep it moist. Do not overstretch the sample. **When you are not testing the sample, make sure the weight is not hanging on the specimen!** Adjust the reference state of the specimen so that there is just enough tension in the sutures to take up the slack (assume this is the zero-force and zero-displacement reference state).

(5) Stress relaxation test. Perform a stress relaxation study on the specimen, recording your data on the computer. To do this you will need to impose a known step increase in length, and measure the ensuing force, which should exponentially decay over time. Adjust the screws on the isotonic transducer so that at one extreme the specimen is in the reference state, and at the other extreme the sample is at a given stretched length (and make sure it stays there during the test). The screws should only be about 1-2mm apart. The micrometer is helpful for making minor length adjustments. Save your computer files for post-acquisition analysis.

(6) Creep test. A creep test can be performed with minor modifications to the setup for stress relaxation. To impose a constant force to measure creep, release the screw on the transducer so that the specimen carries the full load of weights when stretched. Remember not to let the specimen carry the full load for too long, or you will use up all the creeping ability! Now if you hold the rod at the reference length then release, the specimen will carry a constant load, and the length should "creep" from the initial value to a slightly longer value. Save these computer files also. Approximate weights for the specimens for a creep test might be: esophagus, intestine = 5g; trachea = 10g; aorta = 20g; skin = 30 g.

(6) Hysteresis test. This test will be performed separately **after** the creep and stress relaxation tests for each specimen. Determine how, using the set up as-is, you can obtain data to quantify hysteresis in each sample. Measure the hysteresis of the specimen as a difference between the loading and unloading curves. Remember that an instantaneous change in length or force will not be very useful for this type of test. Try to use the same procedure for every sample, i.e. the same strain rate (why?).

Repeat creep, stress relaxation and hysteresis tests for a total of 4 different types of specimens. Assume the calibration factors are the same for all the specimens.

Data Analysis

After you have recorded force and length data for all the specimens, and have cleaned up your entire lab station, you may start post-acquisition analysis (this may be on the next working day!)

(1) Determine the constants in the creep and relaxation functions for each specimen. Assume each specimen can be modeled as a standard linear solid. The creep and relaxation functions [c(t) and k(t)]

for such a model are give by equations (12) on Page 43 and (16) on Page 44 of Fung. Determine τ_σ, τ_ε and E_r from your experimental results. Notice the theoretical c(t) and k(t) are given in the book, and also that in your experiments, the unit step function $\mathbf{1}(t)$ in the book needs to be multiplied by a constant since you probably did not apply a unit force or displacement. Find the 2 time constants (one from each type of experiment) either with manual techniques, or you may use Sigmaplot on the PC's to fit your experimental data to the proper exponential curves (hence find τ_σ and τ_ε). See the course Web page for details on Sigmaplot.

(2) Determine the theoretical constants in the Kelvin model for each specimen. After you have determined the time constants and relaxed elastic moduli, find the spring and dashpot constants in the Kelvin model for each specimen.

(3) Determine the relative hysteresis for each biological specimen. For each sample, first normalize the amount of hysteresis to the maximum displacement for that specimen. In this way you can compare the amount of hysteresis for all the specimens irrespective of the absolute amount of stretch. One way to quantify the hysteresis is to find the area under the loading and unloading curves numerically, i.e. numerical integration (try using a spread sheet with a simple integration scheme).

Write-up notes:

In your write-up, give a sample plot of 1 stress relaxation curve and 1 creep curve. These would come from the computer acquisition data files. Tabulate all of the time constants and relaxed moduli, and Kelvin model constants. Give any equations you used to derive your model constants. Comment on the differences between the tissues and how the different constants in the models relate to possible structural differences in the tissues. Explain your hysteresis test and results, and why these tests should be done at the same strain rate. Also discuss limitations of the experimental setup, for example the mechanical link between the tissue and the transducer, the suture, etc. In the methods, briefly describe how you did the curve fitting, noting any limitations. Describe the experimental methods for the hysteresis curves, and how you quantified the amount of hysteresis. Comment on the structural components of the different tissues and how they may have affected the measured hysteresis.