Objective

Poly(ethylene glycol) diacrylate (PEGDA) hydrogels are widely used in tissue engineering and many other biomedical applications. Their mechanical properties can affect various cellular activities when culturing cells in three-dimensional scaffolds. The objective of this lab is to determine material properties of PEGDA hydrogels including the shear modulus, Poisson’s ratio, density and elastic modulus.

The main experimental objectives are:
1. To test compressive static mechanical properties of hydrogels, in particular determine the Poisson’s ratio for the material.
2. Directly measure the shear modulus of the material.
3. To characterize compressive properties of the material using a computer-controlled compression system.

Introduction

Poly(ethylene glycol) diacrylate (PEGDA) hydrogels are three dimensional and crosslinked networks of polymers that can imbibe large amounts of aqueous solution, such as water and biological fluids. Their network is formed by UV photopolymerization and their acrylate groups function as a crosslinkable moiety, which can control the mechanical properties.¹ These hydrogels have been widely used in cartilage tissue engineering as cell scaffolds due to their biocompatibility, hydrophilicity, and ease of controlling their mechanical properties. Understanding the mechanical properties of PEGDA hydrogels can provide basic background knowledge for tissue engineering investigations when considering their effects on the various cellular activities of encapsulated cells in three-dimensional scaffolds in vitro.² The tests used here can be applied to a variety of materials in order to quantify material properties.

Equipment

- USB flash drive for data transfer (supplied by you!)
- Bose-Enduratec computer-controlled compression system
- Single-edge razor blades and biopsy punch device
- Saline in squirt bottle
- Glass microscope slides, Teflon spacers
- Binder clips
- Shear blocks (with attached friction surfaces)
- Suture
- Video camera and acquisition system

Samples

- Hydrogel samples will be supplied. There will be thin disks of the material about 1mm thick (for Poisson’s ratio and Enduratec compression tests). There will also be a thicker block, about 5-6 mm thick, to be used for the shear test. The material is fragile, do not bend or pull on the sample or it will tear! For compression & Poisson’s ratio tests, 6 different PEGDA hydrogels (labels A1-A3, B1-B3) will be supplied, and for the shear test, 3 different hydrogels (A1-A3) will also be supplied. Their compositions & nomenclatures are tabulated below.
Make sure to note the molecular weight and weight/volume (w/v) concentration of your samples and include this information in your write up.

**Compression & Poisson’s ratio tests**

<table>
<thead>
<tr>
<th>Concentration (w/v)</th>
<th>PEGDA Molecular weight=508</th>
<th>Molecular weight=1000</th>
</tr>
</thead>
<tbody>
<tr>
<td>10% w/v</td>
<td>A1</td>
<td>B1</td>
</tr>
<tr>
<td>15% w/v</td>
<td>A2</td>
<td>B2</td>
</tr>
<tr>
<td>20% w/v</td>
<td>A3</td>
<td>B3</td>
</tr>
</tbody>
</table>

**Shear ratio tests**

<table>
<thead>
<tr>
<th>Concentration (w/v)</th>
<th>PEGDA Molecular weight=508</th>
</tr>
</thead>
<tbody>
<tr>
<td>10% w/v</td>
<td>C1</td>
</tr>
<tr>
<td>15% w/v</td>
<td>C2</td>
</tr>
<tr>
<td>20% w/v</td>
<td>C3</td>
</tr>
</tbody>
</table>

**Prelab**

1. Illustrate and briefly describe a typical tensile stress-strain curve, including the elastic and plastic region.
2. Assume you have a rod-like specimen subjected to uniaxial tension. It will exhibit some shrinkage in the lateral direction. The ratio of lateral strain and axial strain is defined as Poisson's ratio. Independently of the material in the specimen, what are the theoretical limits for the Poisson's ratio?
3. Find some standard values from references and calculate the normal tissue Poisson's ratio.
4. Define in your own term shear modulus and bulk modulus.
5. Since the change in potential energy of an object between two positions is equal to the work required that must be done to move the object from one point to the other, what are the differences on potential energy between a confined and unconfined compression test (see fig)?

**Lab Overview**

This lab will consist of 3 experimental parts:
1. Compressive tests to determine Poisson’s ratio of the material
2. Shear test to estimate the shear modulus of the material; material density and bulk modulus
3. Compressive tests using the Enduratec loading apparatus in the back room
Since there is only one Enduratec testing machine, each group will be assigned a time slot to use that machine, for 20 minutes only. You will need to start your procedures and finish them during that 20 minute time slot! When you are not using the Enduratec machine, you should work on setup and testing for the other 2 parts of the experiment.

(1) Compressive tests to determine Poisson’s ratio

Poisson’s ratio determines the amount of lateral deformation that occurs when a material is stretched (or in this case compressed) in one direction. Experimentally we can estimate Poisson’s ratio by measuring the lateral deflection with a known axial deformation applied. In this case, we will apply a stress (strain) to the disk-shaped sample to its flat faces, and measure the change in diameter of the sample.

To do this experimentally, use 2 microscope slides and binder clips to squeeze the sample down to a known thickness set by the white Teflon plastic spacer, as shown below. 4 binder clips (one on each edge) is optimal.

If you can measure the change in distance between the slides (based on the sample thickness and stopper thickness), and then the change in diameter of the sample under load, the Poisson’s ratio can be determined. For finite deformations, use the following for Poisson’s ratio:

$$\Delta D = -D(1 - (1 + \frac{\Delta L}{L})^{-\nu})$$

Where $D$ is the initial diameter, $\Delta D$ is the change in diameter, $L$ is the initial thickness, $\Delta L$ is the change in thickness and $\nu$ is the Poisson’s ratio. Note that $\Delta D$ and $\Delta L$ are opposite signs since the diameter gets bigger as the length (thickness) gets smaller. Use the video camera and Scion acquisition software to record images of the sample before and after loading. Do this for 3 spacer thicknesses. After the lab, use the images to calculate the needed deformations in order to estimate the Poisson’s ratio from both tests. Also estimate the shear modulus $G$ of the material (you will need an estimate of Young’s modulus to do this from Part 3 below). Compare this shear modulus to that measured directly in Part 2 below.

Include a video calibration measurement (using a ruler), and estimate the resolution of the video imaging system for the magnification used in your diameter image. Report this resolution (in mm) and comment on the implications for your actual measurement.
(2) Shear modulus of the material

A shear modulus can be determined by measuring the relationship between an applied shear stress and the shearing deformation. You may assume small (infinitesimal) strains for this analysis. Set up a system to apply incremental shear stress to the sample, and measure the shear strain at each load. A possible set up is shown below.

Use the ‘shear plate’ and ‘shear block” with sandpaper attached to hold the sample. Use the thick sample, cutting a block about 1.5 cm on each side with a razor blade. Typically, during shear tests, the sample is compressed; in this case the compressive loading will help stabilize the sample. You can load the sample (apply shear stress), by moving the micrometer is small increments. Do this test for 2 different applied vertical weights. Measure the shear strain with the video camera. Determine the shear modulus (include units of course), and note the differences in shear modulus as a function of compressive load. Discuss limitations, assumptions and deviations from theoretical predictions.

Material Density: determine the density of the material with simple measurements. Report this and all of your other parameters in consistent units.

Bulk Modulus: calculate the bulk modulus of the material from an equation using your measured parameters. Make sure the reference the equation in your write-up.

(3) Compressive tests using the Enduratec loading device

The goal of this test is to determine the relaxed Young’s modulus of the specimen, and examine the relaxation parameters during compression.
Methods

**Specimen Maintenance:** Hydrogel sheets will be provided in a phosphate-buffered saline solution. Keep the sheet submerged in the solution. Record the identification code.

**Ramp Compression Testing**

i. From one corner of the hydrogel sheet, prepare an 8 mm diameter disk using a dermal punch.

ii. Make measurements of the generated sample and plan the displacements to be applied. Measure thickness at selected sites (12, 3, 6, 9 o’clock positions), record values, and determine average.

   a. Thickness: __________ __________ __________ __________ Average: __________

   Calculate the displacements needed to achieve 10% and 20% compression:

   b. Displacement for 10% compression: \( b = a \times 0.1 \)

   c. Displacement for 20% compression: \( c = a \times 0.2 \)

   Assuming “zero” displacement is set at a thickness of 1.500 mm, determine positions in the actuator frame-of-reference.

   d. Zero position, platen contacting sample dish: __________

   Relative positions for

   e. 0% Compression, platen contacting top of sample \( e = d + a \)

   f. 10% Compression \( f = e - b \):

   g. 20% Compression \( g = e - c \):

![Figure 1](image.png)

**Figure 1.** Schematic of experimental setup. (A) Zero position set at -1.500 mm. (B) Platen is raised to +1.500 mm to create space to insert sample. (C) Platen is moved to position calculated in **2.B.i.e.**, and then compressed by (D) 10% and (E) 20%.

References

