**Laboratory 2: 2D Strain Measurement**
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BIOEN 5201 – Introduction to Biomechanics
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**Lab Quiz:** A 10 point lab quiz will be given at the beginning of lab (10% of the lab report grade). Be familiar with the entire protocol. Please contact Heath if you have ANY questions.

**Background:**
Strain can be defined as a deformation that occurs due to stresses. In bioengineering, coupled with force data, strain provides valuable information that can be used in defining the function of tissue, understanding pathological impacts and assessing treatments.

Several non-optical instruments exist to measure strain in biological materials (i.e. DVRTs, strain gauges), however they are invasive and will likely affect material behavior. Optical techniques provide a non-contact alternative to measuring local strains by tracking displacements of fixed landmarks. In vivo optical methods include using contrast MRIs, while in-vitro optical methods include spray painting speckle patterns upon tissue or affixing fiducial markers. Measuring local strain is important in assessing material behavior, due to the heterogeneous characteristics of biological tissues. In addition, in controlled experiments that employ clamps, uniform strain is best measured at the center of the material (Fig. 1) due to a clamping effect that occurs. Measuring strain in the uniform center is easily accommodated by optical means. Strain determined from clamp displacement will likely incorrectly predict the strain tensor.

**Objective:**
The objective of this laboratory is to use the DMAS motion analysis system and mini-materials test machine to determine the shear response of thin rubber samples, and to compare strains determined from the motion analysis system to those determined using measurements of clamp displacement. Stress-strain graphs will also be generated using load cell force response data and clamp displacement data.

NOTE – there are 2 digital cameras and 4 framegrabbers installed in two separate computers. Thus, two groups will perform this experiment at one time.

**Equipment required (each test station):**
- 1 mini materials test machine
- 1 Pulnix TM 1040 digital camera, tripod and incandescent light, lens and extension tube
- 1 PC with NI A/D card and NI motor controller card, NI Labview software
- 1 PC with Bitflow Roadrunner Framegrabber and DMAS motion analysis software
• 2D calibration object
• 1 load cell (Sensotec, 1000 gram, limit-stop protected, waterproof)
• 1 set tissue clamps and associated mounting hardware
• 1 set Allen wrenches
• Digital calipers
• Hard, disposable surface for use with rectangular punch
• Hammer
• Rectangular punch, forceps, scalpel handle and blade

Supplies required:
CD-R or USB drive for data backup

Experimental procedure:

NOTE – DO NOT MOVE THE CAMERAS DURING TESTING AFTER CALIBRATION! THEY ARE CALIBRATED BASED ON THEIR CURRENT LOCATIONS!

NOTE – BE EXTREMELY CAREFUL WITH THE LOAD CELL! IT CAN EASILY BE DESTROYED VIA OVERLOADING.

1. Load cell calibration – enter the load cell calibration factor (Newtons per volt) in the Labview VI based on the calibration page provided with the load cell from the manufacturer (amplifier excitation is 2.5 volts).

Calculating the Load Calibration

A. The certificate of calibration from the Load Cell company (Sensotec) is required.
B. Find what the load cell output is at maximum load (Fig. 3)

Example: Calibrated at: 1000 g
Excitation: 5VDC
Calibration factor 37.7171 mV/ V

This means: excitation of 37.7171 mV/V at 1000g.

C. Load cell amplifier is set to give 2.5V excitation, multiply the load cell output by 2.5V (37.7171 mV/V * 2.5 V = 94.29 mV)
D. Next, multiply by the gain. (Gain Fixed at 100X, so 94.29 mV * 100 = 9429 mV = 9.429 V)
E. Since the Load is proportional to Voltage, the Force will be proportional to Voltage as well (F α V) and therefore F=k*V. So, we need to find the constant (k). (k=F/V=m*g/V) Calculate k from this equation. (1000g = 1kg; k=(1 kg * 9.81 m/s^2)/ 9.429V = 1.040.

This value is put into the Cal Load box, to convert the voltage into a Force (Newton)

2. Check the load cell calibration using the pre-test_load.vi by hanging weights from the load cell to determine how well the load is predicted on the PC.

NOTE: if your amplifier does not allow you to zero the starting load, subtract a known baseline “zero” from the data prior to reporting the results.
3. Now use the “Max Automation Motion controller window” to position the load cell-clamp in such a way that it is almost parallel to the other clamp fastened to the base.

4. Be careful as you move down the crosshead with the load cell, **DO NOT CRASH THE LOAD CELL INTO THE BASE OF THE FIXTURE.**

5. On the Motor positioning window, select the appropriate controller card, select ‘Relative position’ and enter a low velocity value for the crosshead. (About 10 is a safe value). Move the motor by about 1000 encoder counts at a time for fine positioning. **Entering a positive value moves the clamp down and a negative value moves it up.**
   
   a. Pull up on the Emergency Stop button to release the safety interlock
   
   b. Initialize the motor in the Max Automation window
      
      i. Highlight PCI-7344 and click “Initialize”
   
   c. Enter a velocity, target position, click “Reset Position” and click “Apply”
   
   d. Click “Start” to initiate motion
   
   e. **Always keep a hand on the Emergency Stop button and press it should unexpected motion occur.**

6. Align the base clamp with the load cell clamp by moving the X-Y table plates.

7. Punch out sample, measure dimensions using calipers (width, thickness, height).

8. Mount the sample first in the clamp that attaches to the load cell. The clamp that attaches to the load cell will have a longer stump, while the other clamp will be fastened to the base of the test fixture.

9. Mount the sample in such a way that its edges are nearly perpendicular to the clamp sides. Screw it on tight, without damaging the load cell. Always watch the Labview VI when this is being done.
10. Mount sample in tissue clamps so that the region in the x-y plane between the clamps will be nearly square. Adjust the x-y table so that there is minimal force in the x-direction and so that the sample resides in the x-y plane.

11. Now identify a small square area at the center of the rubber piece to attach markers.

12. You will punch out white marker discs and affix them to the rubber specimen. Secure contrast markers (4) to the center of the sample using cyanoacrylate to form the corners of a quadrilateral (see Figure 1). Note that the angles do not have to be 90 degrees.

13. Now make sure all the clamps and XY table pieces are well screwed down and there is no relative motion between them.

14. Now position the camera and calibrate it as described below.

15. Measure the distance between the clamp edges (the sample width to be used in calculating the needed clamp displacement) using digital calipers.

16. Determine the amount of actuator displacement necessary to generate a shear angle of 30 degrees. This corresponds to the \( \tan(\theta) = 0.4 \) in the shear.vi

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**Figure 1:** Schematic of experimental
Camera Setup and Calibration:

Position the camera in front of the test machine so that the imaging plane of the CCD is nearly parallel to the x-y plane of the test sample. Adjust the magnification and focus so that the sample occupies as much of the field of view as possible but will not move out of the field of view during the test. A 20 mm extension tube is mounted between the camera and the lens. The camera will be extremely close (almost in contact) with the air table. **DO NOT MOVE THE TABLE ONCE THE CAMERA IS CALIBRATED.** Calibrate the camera using the DMAS software and 2D calibration object:

a) Open DMAS software program  
b) Click on the “Main” menu on the left side, then click “Create New Data Set”.  
c) Choose “One Pulnix TM1040” from the drop down menu and click “Select” in the lower left of the window.  
d) Click “Start Preview” in the “Acquisition Main Control” window.  
e) If the contrast is poor, adjust the aperture on the camera lens or re-position the lights and click on “Refresh” to get a new image (NOTE: try rapid clicking of “Refresh” before making adjustments). Focus the camera on the markers of the rubber sample in the clamps.  
f) Position the 2D calibration object in the camera view. Make sure that the markers are in focus and there is good contrast by adjusting the focusing ring and the aperture on the lens. You will position the calibration frame exactly where your markers will be positioned for testing.  
g) When you are satisfied with the placement of the 2D calibration object, click on “Stop Preview” and then on “Start Calibration.”  
h) Once you are satisfied with the image in the calibration window, click on each marker while holding down the “Ctrl” button. The order in which the markers are identified is important. Follow the names of the markers for the correct order (i.e. Bottom Left is the first marker name (Bottom row, Left column), so start with that marker). NOTE: A 2D calibration model and an appropriate spatial model for the camera will need to be selected (TA).  
   a. One camera runs at 30FPS while the other runs at 5FPS. In this lab, the camera near the door is 30FPS. Please note this for your processing code.  
i) Your volume error should be less than 0.1%. This value is found in the lower left of the calibration window. See the TA for assistance is this is not the case. Once all markers have been identified, click on “Save Calibration” in the “Acquisition Main Control.” Then click, “Stop Calibration.”  
j) Remove the calibration sample prior to proceeding  
k) **The camera is now calibrated. The camera, test machine and lighting must not be moved after this point.**

9. Adjust the load cell balance so that the output is 0 volts. This is done in the amplifier by turning the potentiometer labeled “X”. Check this using the “pre-test-load.vi”. Remember to press “Stop” in the VI after zeroing the load as only 1 VI can run at once.

10. Enter the necessary parameters in the Labview VI (lab2_shear.vi) to begin the cyclic test. These include:
   a. Cal Load, File path, Sample width clamp to clamp, scan rate (10), and strain rate (0.02)
11. Change the file path and file name by typing it into the path bar in the middle of the window. Be sure to label the filename with “.txt” and click “New File, Yes”.

12. Perform the shear test, cycling the sample for 10 cycles with a triangular displacement waveform.
   a. Click the arrow in the upper right of the lab2_shear.vi to initialize the program
      i. A green light in the program window will reset
   b. Start the camera recording a second or two before pressing the Start button in the VI
      i. Press “Stop Preview” in the camera Acquisition window
      ii. Press the red record icon (circle)
      iii. Click “Continue” on the first window and wait for the memory to allocate
      iv. Once allocated, clicking “Start” initiates data collection
   c. Press the “Start” button in the middle of the VI
   d. Record at least 3 full cycles of motion with the camera before stopping recording.
      Be sure to let the VI cycle through all 10 cycles of motion even though the camera
      will not record all of them.
   e. Click “Save” in the camera Acquisition window and name your video file. Click
      OK on the window that brings up a series of blank fields.

13. Determine the 2D marker coordinates by using the DMAS software to track the markers.
   a. Close the Acquisition window after saving the video
   b. Click Main/Open – select your *.dds video file
      i. Click “>” and it will drop down
      ii. Click “> Pulnix” and it will drop down
      iii. Double click on “Tracker” to bring up the tracking window
   c. Get the TA to assist you with the following steps:
      i. Track/select all, Display, Tracking/Set Threshold, Label markers, Go back
to 1st frame, >, confirm no marker jumping, File/Save, Reporter, Data,
Export all markers, TXT – name your data file, Horizontal/Tabs, OK

14. Back up the VI data file (txt) and the marker motion data (txt) onto CD-R or USB before
leaving the laboratory.
**Data analysis:** The objective of the data analysis is to determine the in-plane components of the Green-Lagrange strain tensor based on the marker coordinates for the central region of the sample and based on the actuator displacement for the whole sample.

**Strain tensor from actuator displacement:** Assuming that the test configuration corresponded to finite simple shear, determine the nonzero components of the Green-Lagrange strain tensor based on the actuator displacement. Plot the components as a function of test time for the entire test duration.

**Stress-strain curve:** Plot the average 1st Piola-Kirchoff shear stress component $P_{12}$ (this is the current force over the reference cross-sectional area, the “engineering stress”) as a function of Green-Lagrange shear strain $E_{12}$ determined based on actuator displacement for all ten cycles of the test. Explain any variation in the curve between cycles.

**Strain tensor from marker coordinates:** Determine the components of the Green-Lagrange strain tensor within the region defined by the markers, assuming that the strain is homogenous within that region, using the equation:

$$\text{Assume that the out-of-plane shear strains } (E_{13}, E_{23}) \text{ are zero. You have four markers, so you can construct six different } \text{“} dX \text{“ vectors. This will yield six equations for the three unknown strains } (E_{11}, E_{22}, E_{12}). \text{ You can determine these unknowns for every time that you have marker data during the test by solving the normal equations. Alternatively, you could use singular value decomposition. Matlab can be used for this analysis – see “nnls”, “lsqnonneg” and “pinv” functions.}$$

There are several other approaches that can be used to determine the Green-Lagrange strain tensor from the marker components. For instance, instead of determining the $E_{ij}$ components directly, you could determine the components of the deformation gradient $F_{ij}$ within the region. Feel free to use any other approach in your analysis.

Generate three additional plots using the data from the above analysis

- $E_{11}^{\text{marker}}$ and $E_{11}^{\text{actuator}}$ versus test time
- $E_{22}^{\text{marker}}$ and $E_{22}^{\text{actuator}}$ versus test time
- $E_{12}^{\text{marker}}$ and $E_{12}^{\text{actuator}}$ versus test time

Provide a detailed explanation for any differences between the strains determined from the marker coordinates versus those determined from the actuator displacement.

Assuming incompressibility, calculate and plot the out-of-plane normal strain $E_{33}$ based on the values for the in-plane strain components determined from the marker data as a function of test time.