Standard Operating Procedures of CellScale Biomechanical Testing

Univert (Uniaxial loading)

Note: Before testing, check the load capacity of the installed load cell.

Connections:

1. Select the appropriate load cell, and connect data cable from the load cell to the controller.

2. Plug the two USB connections (actuator and camera) in the computer.

3. Switch on the controller box, and double click the icon 

on the desktop.

4. If you want to do image tracking, **don't forget to speckle the sample to get contrast pattern on the sample surface.**

Running test:

1. File—Collect New: on the pop up window, select Template (if you are new, select demo)

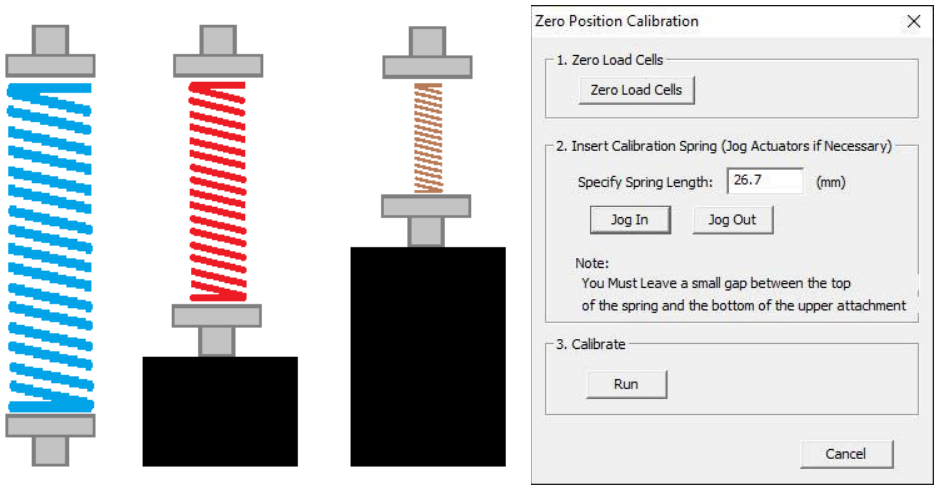
2. Do NOT modify the factory calibration parameters of the load cell. If a new load cell or fixture is installed: Tools → Zero Load Cell Verify the force reading is approximately 0 N before testing.

3. Settings—Test Mode—select expected tension or compression.

4. Use the "Actuator Control" to adjust the position of the actuator. (Recommended Jog Speed: 1–3 for sample alignment; 4–6 for coarse positioning; Avoid speed 8–10 due to oscillation)

5. Actuator Zero Position Calibration: (Perform this step only when changing spring type or when the actuator reference position is lost.)

Tools—Zero position calibration, in the pop up window input the spring length (depend on the selected types of spring), then Jog in the actuator until a small gap. Last, Run.



6. Mount the sample.

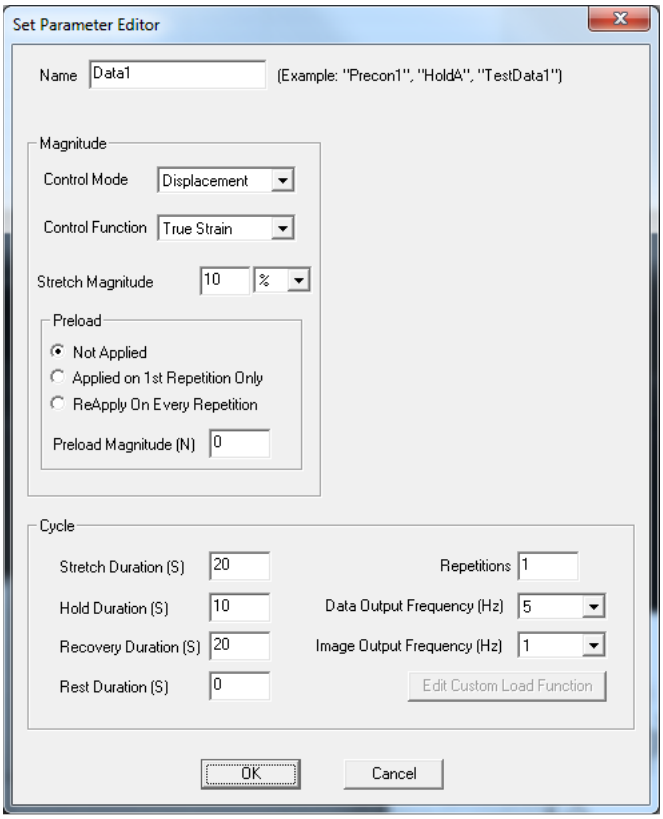
- For tension tests, securely clamp both ends of the specimen.
- For compression tests, place the specimen centrally between the compression platens.
- For bending tests, position the specimen on the bending fixture according to the span length.

For compression test, position the actuator close to the specimen surface. Leave a small gap (or apply a minimal preload if required) before starting the test.

7. Click Edit set, or double click the row of the set parameters.

In the pop up window, input the control mode, control parameters. For the Cycle, two things you need to consider: a, Data output frequency—number of recorded points per second; b, Image frequency—number of acquired pictures per second. Note: if you need multiple ramps, click insert above, or copy the existed row (ctrl+C), and past it (ctrl+V).

Recommended: Data frequency: 5–20 Hz; Image frequency: 1–5 Hz



8. The last thing you need to watch before testing is: a, click Settings—Hardward—Temperature, in the window, remove the warnings; b, put the Ronchi ruler or another ruler (from Microsquisher) in the clamp position, and then go to Tools—Snap image. DO NOT change: • Zoom • Focus • Camera position after Snap Image. Otherwise, you have to snap another picture for the purpose of getting a picture containing real distance—optic calibration for imaging track)

9. **Start Test**

Post analysis:

1. File—Analyze And Review Images (Refer to Page 28). In the pop out window, find .tst file

Details can be referred to Page 9 in the Manual.

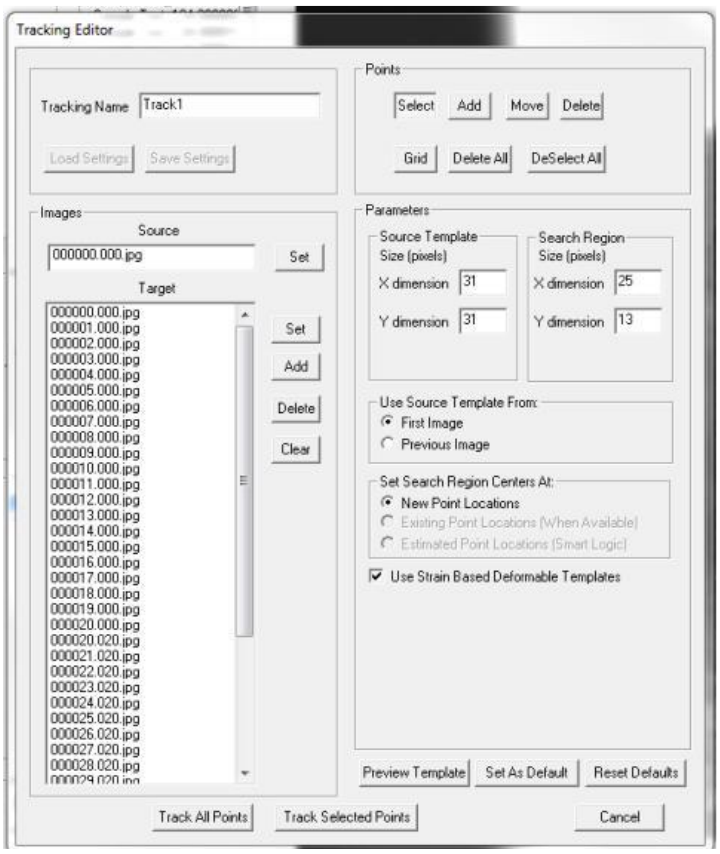
2. Imaging tracking analysis:

A: Under Tracking, click “Create”, then in the pop up window, input Name, in the “Source”, select the first picture as the reference, and click “set”. In the Target, Select all the pictures you want, and click “Set”.

B: Click the first picture. In “Points”, directly draw rectangular using mouse on the sample surface, and then click Grid”.

C: In the “Parameters”, assign the X and Y facet size. If you make a nice pattern using spray, 15 and 12 pixels are recommended for the source template and search region. But, if the pattern is very poor, use the default value.

D: Click Track All points for calculation. Verify tracking quality before accepting results. Points that lose tracking should be removed.



Troubleshooting

Error	Possible Cause
Force not zero	Forgot Zero Load Cell
Large noise	Load cell overloaded
Oscillation	Jog speed too high
Tracking failure	Poor speckle pattern
Target not achieved	Sample too stiff / displacement too large / actuator limit reached
Wrong strain	Camera moved after Snap Image

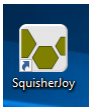
Microsquisher

Notes: Optics Calibration is only required when the camera magnification, zoom, focus, or optical setup has changed.

Connections and Testing Preparation:

1. Plug the three USB connections in the computer.

2. Switch on the Temperature controller and the power.

3. Turn on software, 

then File—collect new to open the interface.

Please verify:

✓ Camera image visible

✓ Temperature controller detected

✓ Piezo controller detected

✓ No error message in the log window

4. Install the sample temporarily and adjust focus.

5. Remove the sample and perform optics calibration.

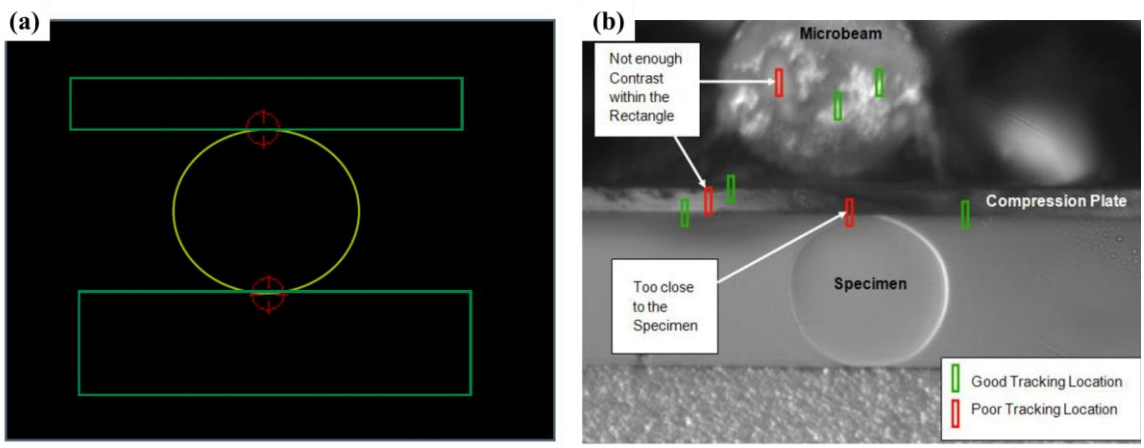
6 Do NOT change zoom or focus after optics calibration. 6. Click Tools—Advanced—Optics Calibration. (Here, the operation of 'focus' is recommended) Place the two red cross markers on black-white boundaries. The two markers must:

- Be aligned vertically (or horizontally)
- Span multiple black-white pairs
- Remain on clear contrast edges

Count the number of black-white pairs between the two markers. Example: 5 pairs = 1000 μm Then input the value in the pop up window (optics calibration), then remove the target and click 'Forward' to move back the compression plate to the position where the optic calibration is conducted.

WARNING: After optics calibration: Do NOT change
• Camera zoom ring
• Software zoom value
• Camera focus
Otherwise repeat optics calibration.

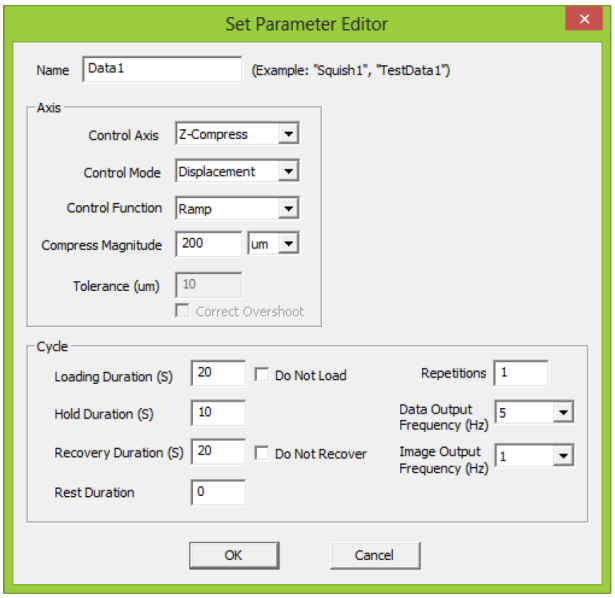
Routine testing does not require optics calibration before every experiment.



7. Install sample between the anvil and the compression plate. Verify that the zoom setting on the camera and software matches the value used during optics calibration.

8. Before editing the control parameters in the 'Edit Set', Right click to create a tracking box. Green box: Good tracking region; Red box: Poor tracking region

Select regions with strong contrast and sharp features. Avoid smooth surfaces. (Refer to the picture, in the above).



9. Edit the control parameters. Click 'Edit set'. In the 'set Parameter Editor', under 'Cycle': Loading Duration means the times reaching the compress magnitude;
The Hold duration means the time holding the final compress magnitude constant;
The Recovery Duration means the times recovering back from the compress magnitude to the initial position;
The repetitions means the number of cycles of the event (loading-hold-recovery).

Contact Mode: Displacement

Compress Magnitude: Compression distance (μm)

Data Frequency: 5 Hz Image Frequency: 1-5 Hz

Others: After installing a new micro-beam, measure and record the distance between the wall and the front edge of the compression plate. This value is required for accurate displacement calculations.

Troubleshooting

Target Not Achieved
Possible causes:
• Sample too stiff
• Compression magnitude too large
• Beam too flexible

Solution:
Use a stiffer beam
Reduce displacement
Increase loading duration

Compression Magnitude Table

Sample	Recommended
Cell	1–10 μm
Hydrogel	5–50 μm
PDMS	10–100 μm
Rubber	50–500 μm

After testing:

1. Save data
2. Export images if needed
3. Remove specimen
4. Move actuator backward
5. Close software
6. Turn off power

Common Errors

Target Not Achieved:→ Sample too stiff or beam too flexible

Maximum Ramp Speed Exceeded→ Loading duration too short

Poor Tracking→ Select a higher contrast region

Incorrect Strain→ Zoom or focus changed after optics calibration

No Image Tracking→ Tracking box not created before test

Biotester (Biaxial Extension)

Note:

A: For the load cell, when installing tap adapter, the shoulder side will be threaded into the load cell.

B: Install the load cell on the “L” frame firstly, then, install the “L” frame on the actuator.

C: Use 'Snap Image' to capture an image containing a known distance reference for image calibration. A flat object with a known distance marker on the surface is recommended.

D: Image tracking, don’t forget to set the first image as the reference.

E: Center Position Calibration is only required after changing BioRakes, load cells, or actuator configurations. Routine testing does not require recalibration every time.

Testing Preparation:

1. Switch on ‘Main’ on the machine. If no need heating, don’t switch on ‘Heater’. Then the LED is on.

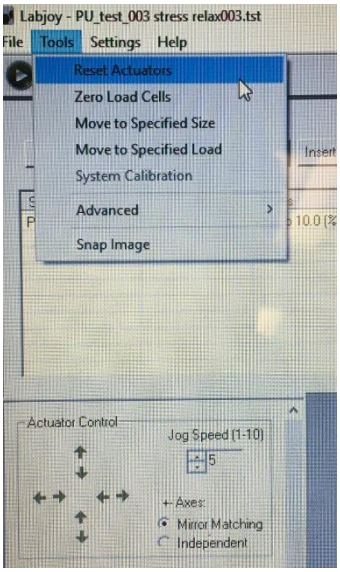
2. Connect the 2 USB on the laptop. (Only two USB)

3. Turn on software, 

then File—collect new to open the interface.

4. In the software, click File-Collect New, and in the pop up window, select the template. If first time, select ‘demo’, when finishing experiment, save as the current file as a template.

5. **Center Position Calibration and BioRake Alignment:** First of all, look at the picture and learn the functions of the actuators (The explanation in this step is mainly using this function a lot):



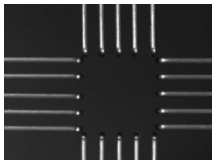
A: ‘Reset Actuators’ means withdraw all the arms back to ‘zero’ position (away from the center)

B: For the actuator control, adjust Jog speed to select the moving speed when clicking the ‘eight’ -direction arrows. ‘Mirror matching’ means a pair of the arms moving together; ‘Independent’ means individual arm moves independently.

5.1: Click Tools-Reset actuators; or click the logo: 
Warning: Never reset actuators with a specimen mounted. Doing so may damage the sample, BioRakes, and load cells.

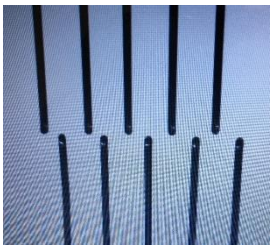
5.2: Put all the four BioRakes on the arms. Tricks: make sure the BioRakes can be adjustable in its magnet position.

5.3: Control the ‘actuator control’ to move the BioRakes in the center position (**not exactly**). This step needs ‘mirror matching’ and ‘independent’. **Then click Tools-Advanced-Center position calibration**, and then click ‘Yes’.



5.4: Click ‘Reset actuators’.

5.5: Click **Tools-Advanced-Move to center. Before clicking ‘Yes’, remove all the four Rakes.**



5.6: Place one pair of Rakes on the arms in Y directions. Then in ‘Mirror matching’ mode, click Y direction to get the overlap position. Tricks: Adjust one Rakes in on direction a little bit for avoiding hitting. **Note: When this step is finishing, don’t run the Y-direction actuator.**

5.7: Remove the Rakes in the Y direction, and place the other pair of Rakes on the arms in X-direction. Repeat step 5.6.

5.8: Click **tools-advanced-Center position calibration**. Then click ‘Yes’.

5.9: Reset actuators

5.10: This is the last step: Place all the Rakes on the arms. Depending on the types of the Rakes, the value under the ‘Specified Size’ is determined by the equation:

Specified size = Tine spacing×5+Tine diameter×(5-1).

Enter the calculated value, and click Tools-‘Move to Specified size’. Now, the square size is: 8020×8020 um.

5.11 Zero Load Cells (Recommended)

Tools → Zero Load Cells

Note: Perform at the beginning of each testing session. Do NOT zero the load cells after a sample has been mounted.

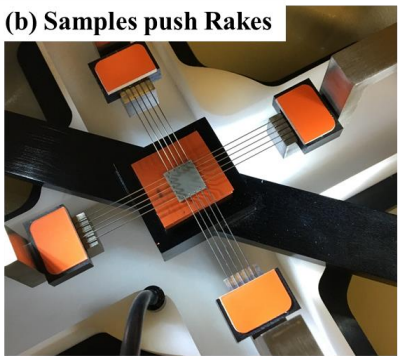
6. The next step is to install sample. Mount the specimen using the BioRakes.

1). Position the specimen in the center.
2). Raise the sample stage until the specimen lightly touches the BioRakes.
3). Press the BioRakes into the specimen.
4). Lower the stage and remove the support material.
5). Raise the stage again if testing in fluid.

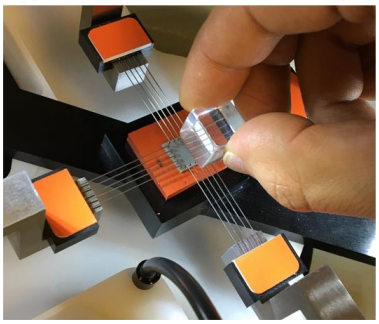
Tricks: when adjusting the knob, adjust the height of the sample, lift up the sample until the sample surface touches the Rakes a little bit to **allow the BioRakes to penetrate the specimen**.

(a) Position sample

(b) Samples push Rakes



(c) Compress Rakes into sample



Knob for adjusting, usage: lift up firstly and then pull out. Once in the expected position, lift down for locking.

7. 'Edit set' to set up control parameters. Typical parameters:

Control Mode: Displacement Control
Displacement Control is recommended for most tissue and hydrogel testing.
Force Control may be used for creep or stress-relaxation studies.

Function: Ramp

Stretch: 5–20 %

Hold: 0–10 s

Recovery: Same as Stretch

Repetition: 5–10 cycles

8. Before executing, click Settings → Hardware → Temperature

If temperature control is not used,
disable the temperature warning message.

9. Execute Test

Click Execute (green arrow).

Monitor:

• Force
• Displacement
• Sample deformation

Stop immediately if:

• BioRakes slip
• Sample tears
• Force exceeds expected range

10. Save Template
File → Save As Template

Save the optimized parameters for future experiments.

Common Mistakes

Never:

❌ Reset Actuator with sample mounted

❌ Zero Load Cell with sample loaded

❌ Move actuators beyond BioRake collision limit

❌ Start test without checking specified size

❌ Ignore force overload

❌ Reset Actuator without removing BioRakes during center calibration

Always:

✅ Reset Actuator before new session

✅ Zero Load Cell before new session

✅ Verify specified size

✅ Save template after optimization

✅ Check force-displacement curve after test