

Dual XYZ Motion Controller

820A

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Manual Version 1

AuroraScientific.com

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1 Introduction

The 820A Dual XYZ Motion Controller was designed to control Siskiyou Corporation's 7600-series closed loop 20mm motorized translation stages. These stages are used in Aurora Scientific 1600A and 1700A systems. The 820A controller reads position signals from the encoders built into the stages and controls the motors providing the user with closed loop position feedback with an accuracy of ±20 nm of the desired position.

For familiarity to existing users of these systems, the 820A also supports manual control using the keypad and joystick controllers provided with Siskiyou Corporation's MC-1000e and MC-1000e-J control systems. However, the 820A completely replaces both the MC-1000e and the MC-1000e-J control systems and power supplies. The only part of these control systems that is required for use with the 820A are the hand controls.

For programmatic control, the 820A connects to a PC via Ethernet. The stages can be remotely controlled via a dedicated program that runs on either a Windows or Linux PC, from a platform-independent web interface, or even from your own custom software via a TCP connection.

In addition to simple motion control and position readouts the 820A provides: multiple user defined waypoints, user defined limits, vector position control, virtual 4th axis control which is especially useful for injection type motions, slaving both XYZ stages together for coordinated movement, stretch control, sinusoidal motion control, rotation/alignment along an axes and a series of motion profiles designed to aid in specific experiments such as the attachment of a myocyte to the end of a pipette or the stretching of cultured cells in a Strex chamber.

2 Quick Start Guide

2.1 Instrument Setup

The 820A Dual XYZ Motion Controller should be placed on a stable surface, and kept free from contaminants. Do not expose the instrument to excessive heat, cold, or moisture.

Definition of Axes Directions

The 820A controller controls two Siskiyou 7600 XYZ stages. These stages can be mounted to suit the user's experiment but the following direction conventions should be maintained if possible. The 820A makes the following assumptions:

- a) the positive direction of movement is when the stage moves towards the motor
- b) the X axis is in the left to right direction with respect to a user sitting facing the stages, the positive X direction is to the right (to the right of the user)
- c) the Y axis is in the front to back direction with respect to a user sitting facing the stages, the positive Y direction is to the back (away from the user)
- d) the Z axis is on the up and down direction, the positive Z direction is upwards

Note: an invert axis function is available that inverts the direction of motion of a stage that is aligned in such a manner that the desired positive direction of the stage occurs when the stage moves **away** from the motor. If using Siskiyou 7600L and 7600R XYZ stages, then the X axis of the 7600L (left stage) will need its axis inverted in the software. The inversion will allow the left X stage to move to the right when the + keypad switch is pushed.

To set up the 820A system for use:

- 1. Place the 820A on a stable surface near the experiment.
- 2. Connect appropriate AC power to the back of the instrument.
- 3. Mount the Siskiyou 7600-series stages as required for your experiment. Remember that in order to calibrate them, they will need to be able to travel through their full range of motion. Calibration normally only needs to be done when you first start using the stages. The calibration factors are stored in the controller and will remain in effect until the unit is re-calibrated.
- 4. Connect the DB-25 cables from each stage axis to the connectors on the front panel of the 820A. The wires from each stage are labelled X, Y and Z. Plug the X connector from the left X stage into the mating connector on the front panel of the 820A labelled Left X. In a similar manner connect the left Y and Z stages. Then, in the same manner, connect the right XYZ stages to front panel connectors labelled Right X, Y and Z.
- Place the hand controls (keypads or joysticks) in a convenient location, and connect them to the 820A via their DB-15 cables. Note: The Siskiyou joystick requires a custom cable between the 820A controller and the joystick. Plug the left keypad into the front panel connector labelled L CONTROL and the right keypad into R CONTROL.

- 6. Connect the 820A's Ethernet port to the supplied Router's Port 1.
- 7. Connect your computer's Ethernet port to the Router's Port 2.
- 8. (Optional) Connect an internet connection to the Router's Internet Port (the yellow coloured port).
- 9. Connect power to the Router.
- 10. Install the supplied software on your computer.
- 11. Power on the 820A controller and test the setup by moving the stages using the keypad controls.
- 12. Start the software on the PC and calibrate the stages. The Calibrate function determines each stage's range of motion and places the stage at a known location. If the 820A has already been calibrated then perform a Home function (see Figure 1) instead of the Calibrate function. The Home function moves the stages to one end of the range of motion to set the stages at a known location (this location is the switch position at the end of the range of motion).

Note: The Home function moves the stages to the same location as the Retract function which has both X stages set as far away from each other as possible, the Y stages set as far back as possible and the Z stages set as high as possible. Ensure that your setup is configured so that a Home function can be completed without any of the stages or their probes striking anything.

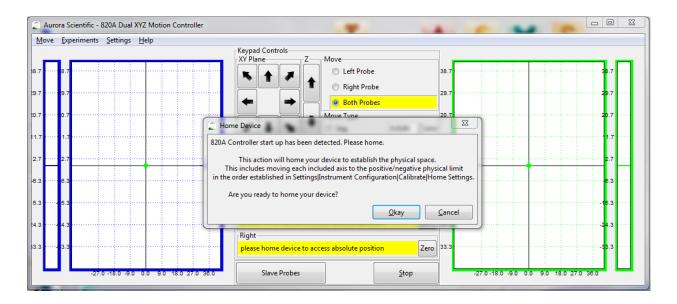


Figure 1 Main Window of ASI820A Control Program with Home Device Prompt Showing

2.2 Sample Experiment – Aligning a Sample in the X-Axis

Problem: Once a sample has been attached it must be aligned precisely in the X axis to allow for a measurement device to function properly (e.g., configuration of an AFM, SL measurement by laser diffraction, or control of the incident angle during photon bombardment).

The following is a step by step procedure to execute this action with the 820A Controller through the Windows/Linux Control Software.

- 1. Start Windows/Linux 820A Control Software which will automatically discover the 820A Controller connected to the network via the supplied Ethernet Router.
- 2. Before physically placing the probes into the experimental apparatus, the instrument should be "Homed". This will run the left and right XYZ stages to their Retract position and set each stage at a known position.
- 3. Place the probes into the experimental apparatus. Try to position the probes so they have maximal available spatial access to the apparatus.
- 4. It is suggested at this point you explore the apparatus with the manual keypads or the control panel provided in the ASI820A control program. This is also a good time to set software limits in the 820A Controller.
- 5. The software limits protect the probes and the experimental apparatus from physical damage by identifying the physical constraints of the system and programmatically limiting the motion of the left and right probes to stay within these limits. To access the programmable limits, select menu Settings > Instrument Configuration and select the Define Limits tab as is shown below. A maximum limit and minimum limit are available for each axis present (12 limits in total). Upon homing the device, the limits will be set to the actual physical limits of the six axes.

Configure Instrument					
Define Limits	Left Set Limits Set to current position Minimum Maximum Minimum Maximum				
Locate Probes	X: -9.0000 @ 9.0000 @ Set Min Max X: -9.0000 @ 9.0000 @ Set Min Max Y: -9.0000 @ 9.0000 @ Set Min Max Y: -9.0000 @ 9.0000 @ Set Min Max				
Define Waypoints	Z: -5.0000 + 9.0000 + Set Min Max Current Limits				
Options	10.1 10.1 10.1 9.00 9.00 Y 10.1 10.2 9.00 9.00 10.2 10.2 9.00 9.00				
Calibrate	Z Image: Constraint of the second s				

Figure 2 Define Limits Window of Configure Instrument Menu

The bottom portion of the screen graphically indicates the current settings of the limits (see Figure 3). For each axis the indicator will show the current physical maximum and minimum limit

settings (numbers located at the ends of each line) and indicate the current limits (red lines and numbers located under each line).



Figure 3 Physical and Software Limits Indicator

The top portion of the screen allows the user to manually set the limits. To set the limits it is suggested the user move the left and right probes to the edges of the spatial boundaries needed to execute the experiment and press the **Min** or **Max** button depending on which limit you are currently at. This step is not necessary for operation but is recommended to prevent physical damage. Of particular importance are the Z minimum limits for the left and right XYZ manipulators. When properly defined the Z minimum limits prevent a probe from colliding with the bottom of a bath and causing physical damage to the probes or the bath.

Note: the limits are active for control via the software and also for control via the keypad or joystick controllers. Once the limits are set the controller will not allow motion past the limit except when a Calibrate, Home or Retract function is executed. These are the only functions that will not respect the software limits.

- 6. To later align the sample, we will need access to the more complex functions the 820A Controller can perform. These functions require that the left and right probes be located in the common space. To do so we establish a frame of reference by identifying a common point. To do so:
 - a. In the ASI820A Control Software open the Locate Probes page by selecting the menu
 Settings > Instrument Configuration and selecting the Locate Probes button.
 - b. Through your microscope identify a single point which both the end points of the left and right probe have access too.
 - c. Using either the control pad on the main window, or the manual keypads, move the end of the left probe to the identified point as accurately as possible.
 - d. Press the **Next** button on the **Locate Probe** page.
 - e. Move the left probe away from the identified point into a position where it will not obstruct the right probe's path.
 - f. Move the right probe to the identified point as accurately as possible.
 - g. Press the **Next** on the **Locate Probe** page.

h. The colocation process is now complete and you should have access to all the functions the AIS820A Control Program has to offer.

💈 Configure Instrument	
Define Limits	
Locate Probes	
Define Waypoints	Place the left probe on an easily identifiable position and press Next.
Locate Probes Define Waypoints Options Calibrate	
Calibrate	Left Position (mm) (0.0000,0.0000,0.0000) (0.0000,0.0000,0.0000) Next

Figure 4 Locate Probes Window of Configure Instrument Menu

Note: The accuracy of some functions (rotation and stretch) depend on the accuracy of collocating the probes. The more accurate you are when locating the probes the better these functions operate.

- 7. Now that the 820A Controller is configured for your particular apparatus, the next step is to find and attach the sample needed for the experiment. Using your microscope identify the required sample and move the probes into position using their manual controls (either the software's control panel or the keypad) and attach the sample.
- 8. Once the sample has been attached it is preferable not to vary the distance between the two probes and risk damaging the sample. The rotation function is used to orient the sample with minimal risk of damage.
 - a. Open the rotation page by selecting the menu **Move > Rotation**.
 - b. Chose a reasonable **Rotate About** option for your particular experiment (left probe, right probe or center point).
 - c. Select the Rotate Mode based on the desired alignment (Continuous sample will continue to rotate as long as you hold the Rotate button, Align in X axis sample will rotate and automatically stop when the sample is aligned along the X axis, Align in Y axis sample will rotate and automatically stop when the sample is aligned along the Y axis).

- d. Press the **Rotate Counter Clockwise** button or **Rotate Clockwise** button depending on the current orientation of the probes and hold the button down (in Continuous mode the sample will continue to rotate, in Align mode the sample will stop rotating when aligned). Note: rotation is always in the XY plane.
- e. Confirm the sample is aligned as desired by viewing it through the microscope.

Warning: Do not rotate the probes into each other by selecting the wrong direction of rotation. The rotation will continue as long as the Rotate button is depressed and for the case of aligning in the X or Y axis the rotation will automatically stop when the probes aligned along the X (or Y) axis independent of physical obstructions. If you are not careful you may crash the left and right probes into each other during the rotation.

🖹 Rotate/Align Probes	- - X
Rotate Counter Clockwise Rotate About Rotate Mode Performance Rotate About Rotate Mode Continous Rotate Mode O left probe Rotate Mode O continous Align in X axis Calign in Y axis Angular Speed 10	Rotate Clockwise

Figure 5 Rotate/Align Probes Window

Note: The Rotate/Align Probes function will not do anything if the length between the probes is zero.

9. At this point you should have a sample attached to the end points of the left and right probes and, if you used Align in X (Y) axis the sample will also be aligned along the desired axis. It is possible that the sample will need to be translated to place it in the center of the microscope's objective or for other experimental reasons. To translate the sample while maintaining the distance between the probes press the **Slave Probes** button on the Main window (see Figure 6) and use either the program's control panel or the manual keypad to move the probes. In slave mode the right and left probes will move in unison in all axes as if they are one rigid object. A visual indication of slave mode is presented by the square box outlining the location of the right and left probes.

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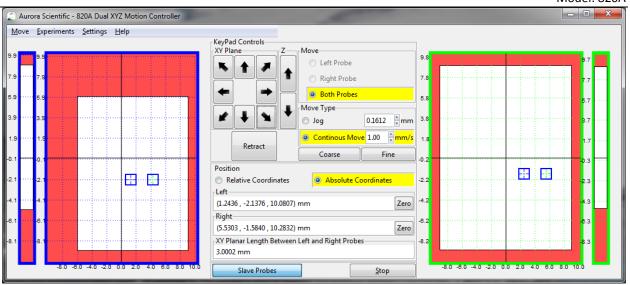


Figure 6 Main window with Slave Probes Enabled

10. The setup should now be complete. Begin the experiment.

3 General Operating Procedure

This chapter provides further hardware details of the 820A Dual XYZ Motion Controller and a full description of the control software.

3.1 Network Setup

In order to access the advanced features of the 820A Dual XYZ Motion Controller the controller must be connected to the control PC (or web device) using a standard Ethernet network. The controller will attempt to automatically connect to a network using DHCP to obtain an IP address. In many cases you can simply plug the 820A into a network connection in the lab and it will receive an IP address from the local network. Any computer on the same network can then communicate with the controller. Because many institutions have restrictions on devices being plugged into their network we supply the 820A with a wireless router to simply the set up the controller. The router provides a DHCP host and allows the user to plug the 820A and their PC into the router to make the connection between the two without touching the institution's network. If you intend to use the router then in most cases, no configuration is required. For the case of a setup with no internet connection, simply connect your computer to router port 1 and the 820A to router port 2. The router's default settings should allow correct operation without further setup. If you wish to connect to the internet, a connection with internet access should be connected to the router's yellow internet port. In most cases, this will be the institution's internal network, and may require authorization from the IT department. The wireless features of the router can also be switched on to allow laptops and mobile devices (phones, tablets, etc.) to connect to, and control the 820A.

3.2 Hand Controller Interfaces

On the front panel of the 820A are two 15-pin control ports labelled L CONTROL and R CONTROL. Connecting controllers to these interfaces allows the user to manually control the 6 axis of their 820A.

3.2.1 Siskiyou Keypad Interface

The 820A supports direct connection of Siskiyou controller keypads. These keypads are included with the Siskiyou Corporation MC1000e control system for 7600-series stages. Simply connect the left keypad to the L CONTROL port and the right keypad to the R CONTROL port on the front panel of the 820A to achieve comparable control on 6 axes as you did using a pair of Siskiyou MC1000e controllers.

Note: The 820A completely replaces the MC1000e controllers and power supplies, the only part that you can reuse are the keypads.

The keypads provide simple manual control, even when using the advanced software features presented by the 820A. The keypads map Axis "1" to the X axis, "2" to the Y axis, "3" to the Z axis and, if configured in the software, axis "4" to the virtual axis. In addition to reading the push buttons the 820A also reads the speed controls found on the Siskiyou keypads and adjusts the motion accordingly.

3.2.2 Siskiyou Joystick Interface

For customers who already use a Siskiyou Corporation MC1000e-J to control their 7600-series stages, Aurora Scientific can provide an adapter cable to connect the 9-pin Joystick to the 15-pin Control connectors. With this adapter cable, you can connect your joystick(s) to the 820A and use them instead of a keypad.

Note: The 820A completely replaces the MC1000e-J controllers and power supplies, the only part that you can reuse are the joysticks.

3.2.3 Aurora Scientific Model 822A Smart Keypad (coming Fall 2017)

For customers who do not already own a Siskiyou Corporation MC1000e-series controller, Aurora Scientific has created a smart keypad (the 822A Smart Handheld Keypad) which also plugs into the same R CONTROL and L CONTROL ports. This smart keypad allows manual control via the pushbuttons and access to the 820A's more advanced features through a touch-screen interface.

3.3 Aurora Scientific ASI820A Software Control Program

The 820A Dual XYZ Motion Controller is supplied with a software package which runs on either Windows or Linux based PC's. This software package allows advanced control tailored to the type of experiment the user is doing. This is the simplest way to achieve advanced control of your 820A. When the software is started, it searches the network and automatically finds any connected 820A's. If it discovers more than one, it will ask the user to select the one they wish to connect to. The software presents on-screen manual controls, and wizard-style interfaces for specific experiment types. If you have a suggestion for experiment additions, please contact us with your feedback!

3.3.1 Primary Controls

The primary controls of the ASI820A software can be found on the main window upon startup of the application. This window is meant to give easy access to all the common functions the user may need during operation. The window can be run in a small footprint without visualization of probe location (see Figure 7) or in a full size footprint that includes probe visualization (see Figure 10).

In the central control portion of the screen is a Keypad control panel with direction arrows that allow the user to move the stages in the XY plane and the Z plane respectively. These buttons are similar in function to the buttons on the Siskiyou keypad but with the added capability of moving the XY stages diagonally. Simply place the mouse pointer on the desired direction button and press the left mouse button to move the probe.

Beneath the control arrows is a large **Retract** button that will retract the probes of both the left and right XYZ stages. The **Retract** button begins a user defined motion of the probes designed to remove them from the apparatus safely. The default defined motion is as follows:

- a) Move the left Z axis all the way up, move the left X axis all the way to the left, move the left Y axis all the way to the back. Once this move is complete,
- b) Move the right Z axis all the way up, move the right X axis all the way to the right, move the right Y axis all the way to the back.

Note: The axes to be included, order in which the axes move and the direction of each axes can all be set for the Retract and Home functions by setting changing the settings parameters under Settings > Instrument Configuration > Calibrate > Home Settings.

The **Retract** button is good for moving the probes out of the way for cleaning, storage or other maintenance. When the **Retract** button is pressed the current location of all 6 axis is saved, the stages start to move to the retracted position and the name of the button changes to **Return**. Pressing the **Return** button returns the probes to the saved position that they were in when **Retract** was pressed.

Note: the saved return position is erased as soon as any of the XYZ stages are moved from the retracted position by any means other than pressing the **Return** button (keypad press, press of the directional arrows, execution of a Move function, etc.).

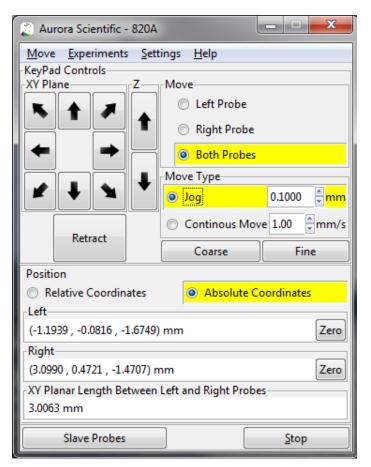


Figure 7 Main Window without Visualization

To the right of the control arrows is the Move section which determines which stages will move when the arrow buttons are pressed. Selecting the **Left Probe** radio button will cause only the left XYZ stages to move. Selecting the **Right Probe** radio button will limit motion to the right XYZ stage. Selecting the **Both Probes** radio button will cause both XYZ stages to move.

There are two types of motions that can be initiated through the control panel, which can be selected in the **Move Type** box (see Figure 8).



Figure 8 Move Type Controls on Main Window

A Jog moves the specified probe(s) by the total distance specified when the desired direction is pressed. Continuous Move moves the probe(s) in the specified direction and at the specified speed as long as the directional button is held down. The **Coarse** and **Fine** buttons load predefined values for the Jog and Continuous Move, which can be changed in **Settings > Instrument Configurations > Options**.

The bottom Portion of the screen (see Figure 9) gives the current coordinates of the two probes. These coordinates are given in reference to the physical bounds of the Siskiyou 7600 translation stages as determined via a **Home** function (see **Movement Controls** for details). Depending on the information available to the software it may also display an XY planar length measurement. (See **Locate Probes** for details).

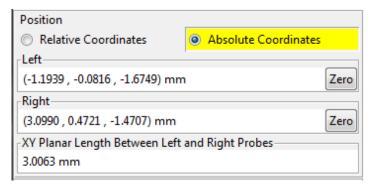


Figure 9 Position Readout

The **Zero** button allows the user to zero the position displays around the current location. When the **Zero** button is pressed the displays are set to zero and the coordinate radio button is changed from Absolute Coordinates to Relative Coordinates. Switching the radio button back to Absolute Coordinates reverts the coordinates to their absolute values as previously described. The coordinate system can also be zeroed, cleared and changed between relative and absolute on the **Settings > Instrument Configurations > Options** window.

At the bottom of the main window is the **Slave Probes** button which links the motion of one probe to the motion of the other. Slave mode can be toggled on and off with the **Slave Probes** button. When activated this button will be applied to all axes of both stages. Slaved motion will occur regardless of how the motion is initiated (program Keypad control, Siskiyou pushbutton control or selected move functions. The particular slave settings can be modified in **Settings > Instrument Configurations > Options > Slave Mode**.

The Slave Mode settings include: which controller will be the master (Left Master or Right Master) and whether any of the axes need to be inverted in order to achieve slaved motion (the default setup has the X axes slave inverted). The easiest method of checking whether any of the axes need to be inverted is to place the controller in slave mode by pressing the Slave Probes button and then move each axes separately. This is best done before a sample is attached to the probes. If both probes move together in the same direction then the setting of the Axis Slave Inversion is correct. If the probes move in opposite directions then change the check box of that axis.

For emergency purposes a **Stop** button is located at the bottom of the main window. Pressing the button stops all current motion being executed by the 820A Dual XYZ Motion Controller.

A visual representation of the position of the probes is also available by switching the main window to show visualization (see Figure 10). To activate the coordinate map visualization, go to **Settings > Instrument Configurations > Options** and select **Show Coordinate Maps.** The left probe is visualized on the left side of the window and the right probe on the right. This visualization will display the location of the probe in reference to the physical limits, the programmable limits set by the user (shown in red), the way points defined by the user (colored dots) and the relative position of the other probe (blue and green crosses). See the corresponding sections for details on each of these functions. A click zoom is available on these representations and a right click will return it to the largest view point.

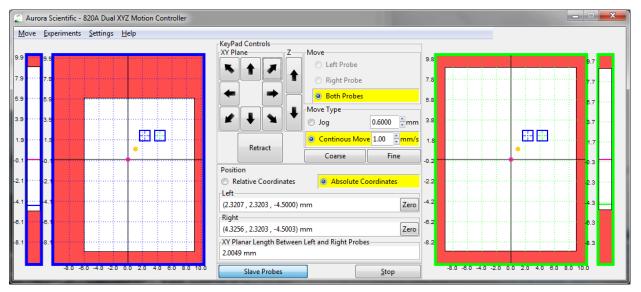


Figure 10 Main Window with Visualization

3.3.2 Movement Controls

The ASI820A software contains a number of motion functions allowing the experimenter to perform actions that would otherwise be cumbersome or difficult to achieve manually with a pushbutton keypad or a joystick. These movement controls are available via the **Move** menu at the top of the main window.

3.3.2.1 Move>Home

Warning: To prevent collisions and possible damage, ensure that the stages and attachments are free to travel their entire range before running the Home function.

The Home function is used to establish the location of the Siskiyou stages in physical space. It is recommended to Home the 820A every time it has been power cycled. During the **Home** function the six translations stages will move to their maximum position. At the end of the Home function the physical location of the stages will be set. During a Home function all coordinates and limits will be reset, however Waypoints will not be reset. A Clear Data function is included at the bottom of the Move menu that will reset all values.

Note: The Home function does not erase software limits, waypoints or the Locate Probes function. However, because the physical location is being set by the switches built into the stages there is the possibility that software limits, waypoints and co-location information may be slightly in error due to that fact that switch actuation is not always at the same location. Therefore, we recommend that after a Home function you check critical limits, waypoints and co-location before performing experiments.

3.3.2.2 Move>Waypoints

Waypoints are used to save particular probe positions the user may want to return to at a later time. To add a waypoint simply go to the menu **Move > Way Points > Add Current Position** (see Figure 11). This will spawn a dialog which will allow you to edit the properties of the waypoint (see Figure 12). The left and right coordinates of the current location of the probes will automatically be entered and pressing the Save button will save these coordinates as the named waypoint. You also can edit the coordinates directly on the Add Waypoint window and then press Save and the edited coordinates will be what is saved as that waypoint.

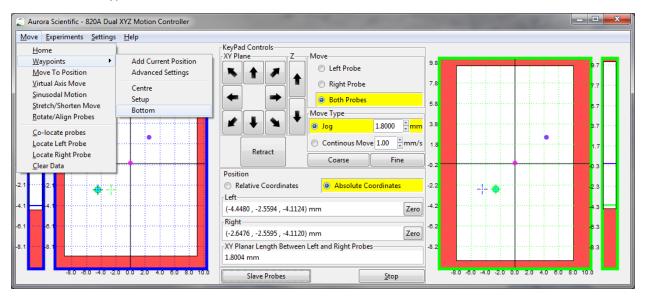


Figure 11 Waypoints Menu Dropdown – Slave Probes Off

If you want to edit or create a several waypoints it is often easier to click on Move > Waypoints > Advanced Settings which will open a waypoint window in Settings > Configure Instrument > Define Waypoints.

🙇 Add Waypoint	
-Name	Color
Center	
Left	7
X	Z 0 ★ 0.0000 ↓
-Right	
X 0.0000	0 - Z 0.0000
Save	Cancel

Figure 12 Add Waypoint Window

If Waypoints have been defined then the Waypoints drop down menu changes based on whether the probes are slaved or not. When not slaved the Waypoints dropdown menu appears as in Figure 11. At the top of the dropdown are **Add Current Position** and **Advanced Settings**, under this are a list of the currently defined Waypoints (in this example they are named: Centre, Bottom and location #3). Clicking on any of the waypoint names will cause both the left and right probes to move to their coordinates defined by that waypoint.

When the Slave Probes button is pressed then the drop down menu appears as is shown in Figure 13. In this case the user can select a named waypoint based on having the **Left As Master** or the **Right As Master**. Since the waypoints may have been defined prior to slaving the probes both probes can't be moved to a particular waypoint as this could cause the probes to move with respect to each other (i.e., change sample length which is not allowed under slave mode). Therefore for any given waypoint the user needs to specify whether they want the left or the right probe to move to that waypoint, the other probe will stay slaved and simply translate alongside the master probe.

To move the probes to a waypoint simply choose the desired waypoint from the Way Points menu or click on the waypoint on the coordinate map to bring that probe to the way point's position. When in slaved

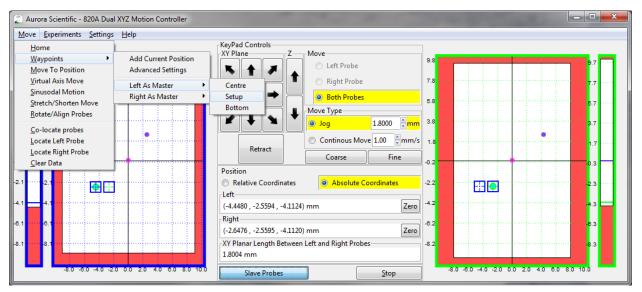


Figure 13 Waypoint Menu Dropdown – Slave Probes On

mode clicking on a waypoint shown on the left visualization screen will move the left probe to that waypoint and the right probe will stay slaved to the left. If you want to move the right probe to a particular waypoint then clicking on that waypoint on the right visualization screen will cause the right probe to move to the selected waypoint and the left probe will stay slaved to the right.

Note: Only the probes indicated in the Move selection shown on the main window will move to the selected waypoint. In other words, if you want to have both probes move to a selected waypoint then you must have the Both Probes radio button selected under the Move section of the main window.

3.3.2.3 Move>Move To Position

The **Move To Position** function is used to move the probes to coordinates entered in the text boxes shown in Figure 15. Simply enter the desired coordinates and press the move button on the right side of the window.

🙇 Move To I	Position		
Left			
X (mm)—	Y (mm)	Z (mm)—	
0.0000	0.0000	0.0000	▲ move
Right			
X (mm)—	Y (mm)	Z (mm)	
0.0000	0.0000	0.0000	move
Close			

Figure 14 Move To Position Window

Note: The Move To Position function respects the Slave Probes mode. Therefore, when in Slave Probes mode, you can't move both probes to arbitrary locations that you specify. If you press the Left move button then the left probe will move to the desired location and the right probe will stay slaved to it. If you move the Right probe then it will move to the desired position and the left probe will remain slaved to it.

3.3.2.4 Move>Virtual Axis Move

Warning: Virtual Axis Move is a function that is independent of the Slave Probes mode. Whenever this function is used the probes will not stay slaved together.

The **Virtual Axis Move** function is designed to allow the user to move the Left or Right Probe along a desired vector. There are two ways to define this vector, the first is meant for injection motions where the experimenter may choose the angle of injection (**Theta**) and the desired piercing speed (|V|), see the left side of Figure 16. This will define a vector motion in the XZ plane as shown by the graphic shown at the top of the window. To perform the motion simply press **Inject** for a forward motion or **Retract** for a backward motion. The two stages will move in unison to create a vector move at the given speed as long as the button is held down. The Siskiyou 4th axis buttons can also be used to perform the Inject and Retract functions, press the 4 (+) button to inject and press the 4 (-) button will retract the probes.

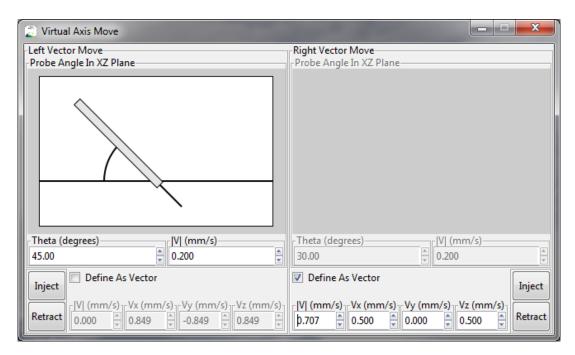


Figure 15 Virtual Axis Move Window

The second method (shown on the right side of Figure 16) is used when the experimenter wishes to define a more general vector. To do so simple check the **Define As Vector** box and then the input boxes allow the user to define the velocity magnitudes of all three axis components. The **Inject** button will perform a move of this vector while the **Retract** button will perform the negative of the defined vector.

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3.3.2.5 Move>Sinusoidal Motion

Warning: Sinusoidal Motion is a function that is independent of the Slave Probes mode. Whenever this function is used the probes will not stay slaved together.

The **Sinusoidal Motion** function allows the experimenter to oscillate the probes in a sinusoidal manner with a desired amplitude and period. There are two modes for this function **Stretch/Shorten** and **Manual**. The **Stretch/Shorten** mode allows the user to define the stretch axis, the magnitude of the oscillation, the Period, Phase and number of Cycles. The **Manual** mode is similar to the **Virtual Axis Move** function in that the user can specify an amplitude for each individual axis component. The controls seen at the bottom of the window vary based on whether the defined oscillation has been set to **Stretch/Shorten** (see Figure 17) or to **Manual** (see Figure 18).

Stretch/Shorten Mode (see Figure 17)

Alignment allows the user to determine whether the sinusoidal motion will be along the X axis, the Y axis or along the sample axis.

Stretch Magnitude provides a simple method of entering the total motion that the sample will have applied to it. The total motion is $4 \times |A|$.

Period (s) sets the duration of the stretch in seconds.

Cycles sets the number of sinusoidal stretches that are to be performed.

Phase allows the user to determine the initial point of the oscillation. This is where the oscillation will start and stop in reference to a sine function as shown in the top visualization of Figure 17. A numeric phase can be entered in the **Phase (Degrees)** text entry box. As an alternative to entering phase numerically there are three buttons that allow the user to quickly set the phase: **Stretch** (the initial position will be the minimum length between the probes, phase of 270°), **Shorten** (the initial point is a maximum length between the probes, phase of 90°) and **Symmetric** (the initial point is half way between the maximum and minimum length of the sample, phase of 0°).

Execute starts the defined sinusoidal motion.

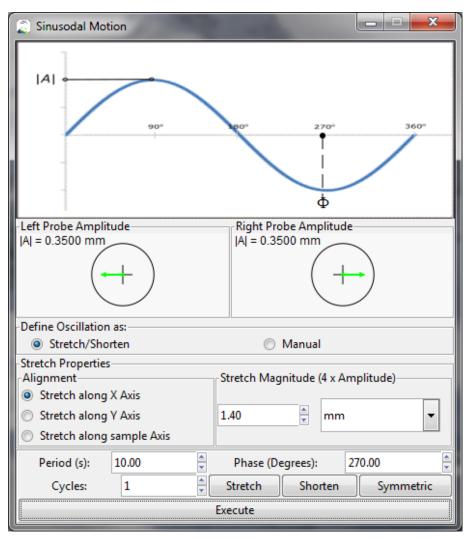


Figure 16 Sinusoidal Motion Window showing Stretch/Shorten Parameters

Manual Mode (see Figure 18)

Left/Right Probe Amplitudes sets the maximal point on the sine function. A unique amplitude can be chosen for each axis (**Ax, Ay, Az**) to allow for motion along any desired vector. The vector is plotted in the XY plane in the circular graphic area in the middle portion of the window.

Frequency, Cycles and Phase are set the same as with the Stretch/Shorten Mode.

Once the parameters are set pressing the **Execute** button will start the sinusoidal motion.

Warning: Care must be taken when defining sinusoidal motion that will cause a shortening motion or a symmetric motion. If the amplitude of the motion is specified larger than the sample length then the probes can strike each other resulting in damage to your experimental apparatus.

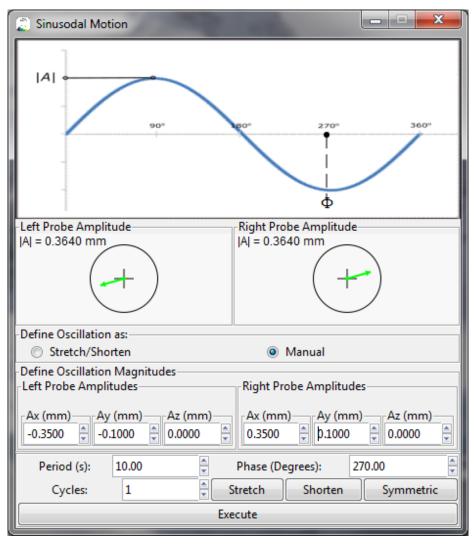


Figure 17 Sinusoidal Motion Window showing Manual Oscillation Definition

3.3.2.6 Move>Stretch/Shorten Move

Warning: Stretch/Shorten Move is a function that is independent of the Slave Probes mode. Whenever this function is used the probes will not stay slaved together.

Note: before the Stretch/Shorten Move can be executed the Locate Probes action must be completed. Please refer to the Locate Probes section for more details.

The **Stretch/Shorten Move** is designed to move the probes along an imaginary line drawn through the end points of the two probes. The user must complete the Locate Probes section in **Settings > Instrument Configuration > Locate Probes** before this move function can be used.

Stretch/Shorten By determines the magnitude of the move to be executed. This can be defined by an absolute quantity (mm or μ m) or by a percentage of the current length between the probes or as a percentage of a Lo distance set by the user.

🖉 Probe Stretch/Shorten Motion 📃 🖃 💻 🗙					
	Stretch/Shorten By Stretch From				
Stretch	10.000 🌻 🗞 Length 💌 🔿 right probe				
	O left probe				
Shorten	✓ Use Lo 2.0001 ↓ mm Set Lo				
Length					
2.0001	mm				

Figure 18 Stretch/Shorten Move Window

Use Lo checkbox forces the program to calculate the percentage length change from the defined Lo value and not the current length value.

Lo data entry box. The user can enter a desired Lo value here or use the Set Lo button to set the Lo value.

Set Lo button sets Lo to the current length.

Stretch From allows the user to choose the stationary point when executing a stretch or shorten move. The stationary point can be defined as the center of the sample (the midpoint between the two probes) or the left or right probe.

Stretch button executes the current settings for a stretch move.

Shorten button executes the current settings for a shorten move.

Length is the current length between the probes (tissue length).

3.3.2.7 Move>Rotate/Align Probes

Note: before the Rotation function can be executed the Locate Probes action must be completed. Please refer to the Locate Probes section for more details.

A rotation move allows for the user to align samples attached to the probes along an axis without varying the length between the probes. This rotation is done exclusively in the XY plane and about the imaginary line drawn through the end points of both probes.

Rotate About determines the center of rotation. The rotation can occur around either of the probes or around the center point between the two probes.

Rotate Mode determines the rotation mode. In Continuous mode the probes will rotate as long as either of the rotate buttons are pressed. In the Align in X axis or Align in Y axis modes the rotation will automatically stop when the two probes form a line parallel to the chosen axis.

Angular Speed determines the angular speed of the rotation when executed. The tangential speed of the probes will be a result of this value, the distance between the two probes and the Rotate About location.

The motion itself is executed by the **Rotate Counter-Clockwise** or **Rotate Clockwise** buttons which continuously execute the current rotation settings in the given rotational directions while the button is pressed down.

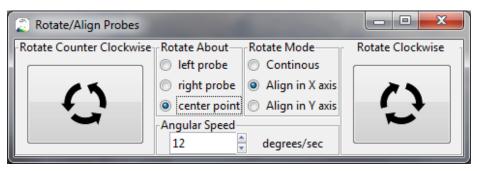


Figure 19 Rotate/Align Probes Window

Warning: be mindful of the relative location of each of the two probes when choosing the direction of rotation. Depending on the probe locations a rotation movement could result in the probes striking each other.

3.3.2.8 Move>Co-locate Probes

The **Co-locate Probes** function presented on the Move dropdown menu is a quick version of the **Locate Probes** function found in **Settings > Instrument Configuration > Locate Probes**. This version assumes that you have located the two probes so that they are physically touching each other and are aligned correctly in the X, Y and Z directions. Pressing the **Co-locate Probes** button will store the current physical locations of each stage and the fact that these locations are coincident.

3.3.2.9 Move>Locate Left Probe

The **Locate Left Probe** function presented on the Move dropdown menu is a quick method of co-locating the probes after a change has been made to the left probe. This function is especially useful when you need to change a probe. Move the left probe to a location appropriate for making a probe change. Change the probe. Now move the left probe back until it touches the right probe and press the **Locate Left Probe** button. The locations of the left stages will be updated based on the current location of the right probe.

3.3.2.10 Move>Locate Right Probe

The **Locate Right Probe** function presented on the Move dropdown menu is a quick method of co-locating the probes after a change has been made to the right probe. This function is especially useful when you need to change a probe. Move the right probe to a location appropriate for making a probe change. Change the probe. Now move the right probe back until it touches the left probe and press the **Locate**

Right Probe button. The locations of the right stages will be updated based on the current location of the left probe.

3.3.2.11 Move>Clear Data

The **Clear Data** function presented on the Move dropdown menu is used to clear all limits, waypoints and co-location information. After a Home function this data is retained but you may want to clear it, the **Clear Data** button will do this.

3.3.3 Settings

Settings and advanced functions can be found in the **Settings > Instrument Configuration** menu.

3.3.3.1 Define Limits

Software limits are used to restrict the range of motion of all axes in each of the left and right stages. The limits apply to all movement commands (except Calibrate, Home and Retract) issued by the software and they also apply to movements initiated with the keypad controllers. After software limits have been set there is no way the user can drive a stage beyond these limits. Setting limits should be done to avoid physical damage to the probes or to any other equipment due to the physical layout of the instruments. One of the most important limits is the minimum Z limit as once this is set the user can be assured that the probes cannot crash into the bottom of the test chamber.

A maximum limit and minimum limit are available for each axis (12 limits in total).

Note: The Home function does not erase the limits previously set. However, the actual location of the limits may be slightly different due to differences in the precise position where the switches inside the stages make contact. Therefore it is suggested that critical software limits be checked after a Home is executed.

Note: After calibrating the 820A, all software limits, waypoints and co-location information is erased. Therefore these values must be reset after calibrating.

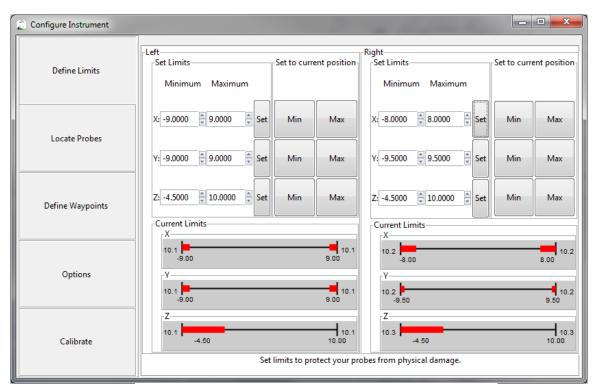


Figure 20 Configure Instrument > Define Limits Window

The top portion of the screen allows the user to manually set the limits (see Figure 21). Limits can be set two ways, the first is by entering position coordinates for the minimum and maximum of each axis and then pressing the **Set** button corresponding to each axis. The second method is to position the probe where you want the limit to be and then press the **Min** or **Max** button on the appropriate axis. Once the limit has been set using the Min/Max button the user can edit the limit in the text entry box and then press **Set**. This is especially useful when you are trying to set the limit a fixed distance from a physical boundary. For example, you want to set the limit 10 µm above the bottom of the tissue bath. Manually move the Z axis down until you see your probe contact the glass. Set the Z axis minimum by pressing the **Min** button. The position coordinates for the Minimum Z setting will then update to the current position. Now simply add 0.010 to the number in the text box and press **Set** which will update the limit to the desired location.

The bottom portion of the screen provides a visualization of the physical limits and the current software limits. For each axis the indicator will show the current maximum and minimum limit settings and indicate the amount of usable space. The numbers at the ends of each line (see Figure 22) indicate the physical limits of the stage while the numbers beneath the line indicate the current software limit settings. The red lines provide a visualization of where the software limits are within the physical range.

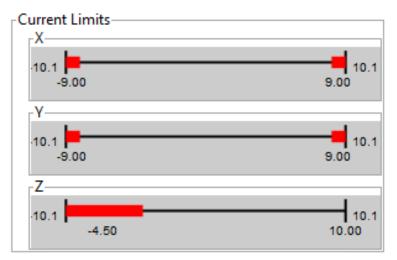


Figure 21 Limit Visualization

3.3.3.2 Locate Probes

The locate probes function allows the experimenter to identify the physical location of the tip of each probe with relation to the other probe. Locating the probes allows the 820A to perform advanced movement functions (Rotate/Align Probes, Stretch/Shorten Move) and also allows the length between the probes to be calculated, recorded and manipulated. When using the locate probes function the user should ensure they are locating the tips of each probe and not some point elsewhere on the probe.

To perform the Locate Probes function the user must be able to move both probes to the same point in space in a sequential manner. Once a probe is at the common point the 820A program will record the coordinates. From this information the program can then calculate the distance between the probes at any time. Normally the common point is identified using a microscope and either a recognizable feature in the field of view is used as the common point or the microscope's cross hairs (reticule lines) can be used.

Press the **Locate Probes** button (see Figure 23) to start the function. The program will guide you through the correct procedure with an onscreen tutorial. The procedure involves: moving the left probe to the common point and pressing the **Next** button, move the left probe away from the common point and then move the right probe to the common point and press the **Next** button. The probes are now located in physical space and the program can use this information.

Warning: Distance calculations and move functions that depend on the Locate Probes function are only as accurate as the initial measurement! Please take care to accurately identify a common point if you wish to use these functions.

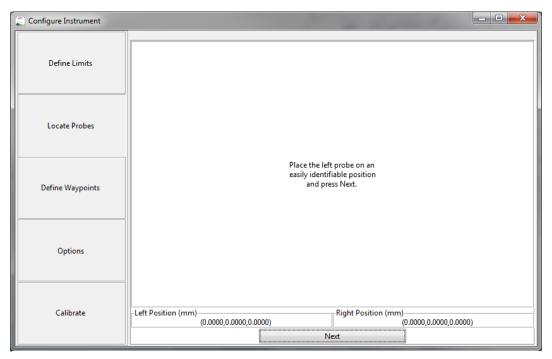


Figure 22 Locate Probes - Set Left Probe

3.3.3.3 Define Waypoints

The Define Waypoints function allows the experimenter to add, edit and delete waypoints manually. In normal operation the user will create waypoints using the **Move > Waypoints** dropdown menu. However, the user can also create and edit waypoints through the **Configure Instrument > Define Waypoints** function (see Figure 24). Waypoints can be added by either entering the position coordinates manually or moving the stages to the desired location and then pressing the **New** button. Waypoints are automatically named with location and *#* but the names can be changed to reflect locations in the experiment (e.g., bottom, center, pre-attach, etc.). Existing waypoints can be overwritten by editing the waypoint name or by changing the location and then pressing the **Save** button. Waypoints can be deleted by selecting the waypoint and pressing the **Delete** button.

Waypoints have a colour associated with them and this colour is displayed on the coordinate maps found on the main screen. Probes can be moved to any waypoint by simply clicking on the coloured dot on the coordinate map.

2 Configure Instrument					
Define Limits	Waypoints Centre Setup Bottom	Name Bottom Left	Color		
Locate Probes		-4.4480 v -2.5594 Right X -2.6471 v -2.5594	A.1124 ↓ ▼ -4.1124 ↓ ▼ -4.1091 ↓ Delete		
Define Waypoints		New Save	Delete		
Options					
Calibrate					

Figure 23 Define Waypoints Window

3.3.3.4 Options

Selecting **Configure Instrument > Options** opens the Options window (see Figure 25). This window allows the user to set all of the miscellaneous settings for the ASI820A software. The window is divided into six sections: Display Options, Jog Defaults, Cont. Move Defaults, Slave Mode, Available 820A Devices, and Coordinate System.

Display Options

Show Coordinate Maps checkbox controls whether the coordinate maps are shown on the main window or not. Check the box to show the coordinate maps.

Jog Defaults

Allows the user to set the Coarse and Fine jog distances that are available on the main control page when the Coarse and Fine buttons are selected. Either enter the desired value in the text box or press the up/down arrows to scroll the values.

Cont. Move Defaults

Allows the user to set the Coarse and Fine continuous move speeds that are available on the main control page when the Coarse and Fine buttons are selected. Either enter the desired value in the text box or press the up/down arrows to scroll the values.

🖉 Configure Instrument							
Define Limits	-Display Options	Jog Defaults Coarse Step: 1.00 mm Fine Step: 0.0100 mm	Axis Slave Inversions	Available 820A Device Device List 192.168.175.109	es		
Locate Probes	Show Coordinate Maps	Cont. Move Defaults			Set Device		
Define Waypoints	Coordinate System Coordinate System Zero L Absolute Relative	Fine Speed: 0.0200 mm/s					
Options		LL ALL					
Calibrate							

Figure 24 Options Window

Slave Mode

Determines which probe will become the master control when slave mode is turned on. Click the radio button next to Left Master to make the left keypad the master control. Under this situation the left keypad will control both the left and right XYZ stages and the right control will not function.

Beneath this section is the **Axis Slave Inversions** section which allows the user to invert the direction of motion of each axis of the slave XYZ manipulator. An example of when axis inversion is required is when the X motor on the left side and the X motor on the right side have their orientation opposite each other, i.e., the left X motor is located to the left side of the stage and the right X motor is located to the right side of the stage.

Note: if during slave operation the distance between the probes changes then one, or more, of the stages needs to be inverted.

Available 820A Devices

If the experimenter has multiple 820A controllers on a single network, then the controlled device can be changed by simply selecting the appropriate device IP address and pressing the **Set Device** button.

Coordinate System

These settings provide a more advanced version of the zero function which is available on the main page. The **Absolute** radio button presents the coordinate positions of all stages in absolute coordinates that are determined during a **Home** function. This makes the zero coordinates exactly in the center of the range of motion of each axis. The **Relative** radio button allows any, or all, axes to be zeroed at any location within the physical space. Thus the user can make a location of interest (glue location, test location, retracted location, etc.) have zero coordinates. Press the **ALL** button to zero all axes at the current location of the stages. Press either the **X**, **Y**, or **Z** button to zero just the selected axis. The **Clear** button makes the Relative coordinates equal to the Absolute coordinates.

3.3.3.5 Calibrate

Selecting **Configure Instrument > Calibrate** opens the Calibrate window (see Figure 26). This window allows the user to calibrate the 820A to the Siskiyou stages that are being controlled. The window is divided into two sections: Home Settings and 820A Controller Settings. The Calibrate Space button at the bottom of the screen starts the calibrate function.

🖉 Configure Instrument		x
Define Limits	Home Settings Axis Include Order Inverse X Z Z V Z V 1 V	
Locate Probes	$ \begin{array}{c c c c c c c c c c c c c c c c c c c $	
Define Waypoints		
Options		
Locate Probes Define Waypoints Options Calibrate	Calibrate Space	

Figure 25 Calibrate Window

Calibration

The **Calibrate Space** button initiates the calibration function. Calibration is used to determine the range of motion of each stage. During the calibration all stages are run to their minimum physical limit and then

to their maximum limit. The number of encoder counts between the two extremes are recorded and then, using the encoder resolution, the range of motion of the stages are determined in mm units. The calibration factor for each stage is recorded in non-volatile memory in the 820A and these values are retrieved each time the 820A is powered on. Normally calibration is only required when you first purchase the 820A. However, if you change the stages you are using with the 820A, or plug the stages into the 820A differently, then you must re-calibrate.

Warning: Calibrating the device will move the stages from one end of the range of motion to the opposite. It is imperative no physical obstacles be in this range while the procedure is active.

Warning: limits, waypoints and co-location information are all deleted during the Calibrate function. Ensure you re-set the software limits before proceeding with an experiment.

Home Settings

The **Home Settings** section controls the order in which the axes move when performing a Home or Retract function. The Home Settings panel is split into two sections, one for the left XYZ stages and the other for the right XYZ stages. Each section contains the same controls. These are: Include checkbox, Order of movement and Inverse movement.

The include checkbox is used to include an axis in the Home and Retract functions. If the checkbox is not checked then this axis will not be moved during a Home or a Retract function. In most cases all axes should be checked.

The Order boxes allow the user to specify the order in which the stages move. In many cases the probes may be located in a bath where maximum X and Y movement is restricted by the walls of the bath. If this is the case then it is preferable to move the Z axes first to get the probes above the bath. At this point the X and Y axes can then be moved to their extreme positions without risk of striking the probe on the bath. The default order of movement is left Z, left X, left Y, right Z, right X and finally right Y but the user can define the order.

The Inverse checkboxes allow the user to define which end of the range of motion of each axis will be the Home and Retract position. Depending on the physical arrangement of the stages and the experimental apparatus the user may decide to have the Home and Retract locations inverted. The default direction (checkbox not checked) is to move the stage towards the end with the motor. Placing a check mark in any of the Inverse checkboxes will cause that axis to move to the opposite end.

820A Controller Settings

The **Axis Inversions** section allows the user to invert the direction of motion of any individual axis. These inversions are used whenever the user wants to change the direction of motion that corresponds to a + button or – button keypad press. Having said this the 820A was designed so that a positive movement will drive the stage towards the motor end of the Siskiyou 7600 stage. Further to this if the stages are used with an inverted microscope then the 820A defines the X direction as the direction across the

microscope, Y is the direction from front to back and Z is vertical. Depending on how the user has configured the stages some of the axis will need to be inverted. In most setups the Z axis is attached so that the motor is vertically above the stage and therefore the positive Z direction is vertically upwards and no axis inversion is required. Likewise, in most setups the left and right Y stages have the same orientation (either the motor is at the back or at the front but it isn't normal that one is at the front and the other at the back). For the case when the Y stages are oriented so that the motor is at the back then no axis inversion is required. In most cases the left and right X stages will be opposite each other (on the left side the motor is on the left side of the stage and on the right side the motor is on the right side of the stage). Under this circumstance one of the X axes will need to be inverted. Since the 820A defines the positive direction of all axes using the right hand rule the positive X direction is when the stage moves towards the right side of the motor. Since the left X stage is actually moving away from the motor then this is the stage that needs to have its axis inverted.

The **Encoder Resolution** shows the defaults for the Siskiyou 7600 stages. These values should not be changed. However, if you are using a different Siskiyou stage that has a different encoder resolution then contact Siskiyou for the encoder resolution of your model and enter it here. In most cases all axes will use the same motor/encoder and therefore the same resolution numbers will be in all of the text entry boxes.

3.3.4 Experiments

3.3.4.1 Intact Cell Attachment

The Intact Cell Attachment module is designed to simplify and streamline the attachment process by identifying important waypoints and automating the required motions.

The module gives quick access to the three positions shown in red, blue and green on Figure 27Figure 28.

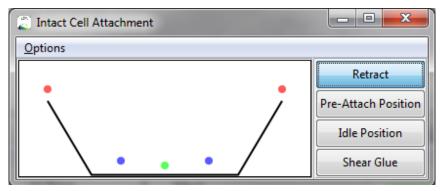


Figure 26 Intact Cell Attachment Control Window

Retract, shown in red, moves the probes to the retract position of the apparatus. This is the same function as is found on the main control page.

Pre-Attach Position, shown in green, is defined to be at the center of the field of view of the microscope at a predetermined height from the bottom of the sample area. This location is used to bring the probes to a convenient location in preparation for other manipulations (attach glue, attach cell, etc.).

Idle Position, shown in blue, is a predefined location a small distance up and away from Pre-Attach location but outside of the view of the microscope's objective. This is an out of the way location where the left and right probes can be placed which will not obstruct the view through the microscope.

Shear Glue Is an automated move command. When performed this command moves the left and right probes up and away in the XZ plane at a specified angle for a specified distance from its current position. This command is used to extract the probes from a glue blob in a controlled manner.

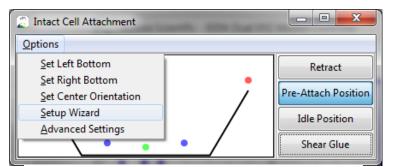
Note: this move is performed on **both** probes when the **Shear Glue** button is pressed.

Options Menu

To use the **Intact Cell Attachment** module, the experimenter will have to use the **Options** menu (see Figure 28) to characterize the physical parameters of the experimentation area. This characterization includes identifying the bottom of the bath for both the left and right probe. Once identified the 820A controller will be able to avoid physical damage by limiting the motion of the probes to above this floor. Additionally, the module uses this bottom as a reference point to configure the **Pre-Attachment Position** as well as the **Idle Position**.

Set Bottom

To identify the bottom of the apparatus the experimenter must manually bring each probe to the bottom of the apparatus, watching for the probe to touch against the glass (caution: be careful not to push the probe into the glass and damage the probe or the glass). Once the probe has been brought to this position simply open the **Options** menu and choose **Set Left Bottom** or **Set Right Bottom** depending on which probe you are setting.



Note: the bottom of the bath must be defined for both probes before using the Intact Cell Attachment module.

Figure 27 Options Menu for Intact Cell Attachment Experiment

Set Center Orientation

The pre-attach position for the left and right probes must be set before every experiment. To do this start by aligning the left and right probes in the XY plane of the microscope's field of view (see Figure 29). Once they are aligned, press **Options > Set Center Orientation** to save the XY- settings of the left and right probe for the **Pre-Attach Position.** Note that the depth of the left and right probes will be determined by the bottom setting done previously and also by the parameters found in **Advanced Settings**.



Figure 28 XY Orientation of the left and right probes in the field of view of the microscope

Advanced Settings

The Advanced Settings menu (see Figure 30) is used to set the parameters that drive the Shear Glue function. The bottom and center must be defined first and then all of the movements are done in reference to these positions. These parameters typically don't need changing to use the function, instead they are changed when the experiment changes or when the experimental apparatus is changed.

🖉 Intact Cell Attachment - Advanc	ed Settings		-	_ D X
	Lef	t Probe	Rig	ht Probe
Bottom Depth (z-axis):	-5.0000	mm	-5.0000	🔹 mm
X Center Position:	0.0000	mm	0.0000	nm
Y Center Position:	0.0000	mm	0.0000	🚔 mm
Idle Position Shear Move				
Z Distance From Center Position:		0.0000	mm Glue Shear Exit And	le: 60.00 🌻 Degrees
X Distance From Center Position:		1.5000	mm	
Space from bottom for Pre-Attach	position (z-axis):	0.2500	mm Shear Move Lengt	h: 0.50000 🛓 mm

Figure 29 Advanced Settings Window of the Intact Cell Attachment Experiment

The top portion of the window shows the currently defined coordinates for the left and right bottom locations as well as the XY coordinates of the center position. The user can modify these values by changing the numbers in the text boxes.

Idle Position section allows the user to define the distance the left and right probes will move from the center position in order to remove them from the field of view. The user must enter a Z move and an X move from the center position. These distances should be large enough to remove the probes from the field of view but small enough to keep them in the bath.

Space from Bottom for Pre-Attach position (Z-axis) defines the distance above the bottom that the probes will be placed at when in the Pre-Attach position. Many researchers apply glue to a small piece of cover slip and then drop the cover slip into the bath so that it sits on the bottom. For this case a good Space from Bottom for Pre-Attach position is the thickness of the cover slip plus a little extra. This allows the glue and cover slip to be brought to the probe location while the probes are in the Pre-Attach position without fear of damaging the probes by having them strike the edge of the cover slip.

Shear Move section allows the user to define the motion that will be used to remove the probes from the glue. It has been found that a shear like motion provides optimum glue coverage on the probe. The shear move consists of a retraction from the current position at an angle specified by **Glue Shear Exit Angle** over a distance specified by **Shear Move Length**. The angle is defined in the XZ plane along the X Axis. The shear move length is a vector length along the specified angle.

Note: Setting the Bottom and the Center must be done for every experiment. Advanced settings can be set once and then reused as long as none of the parameters need adjustment.

3.3.4.2 Assay Stretch

The Assay Stretch module allows the user to repeat a number of different stretch oscillation cycles on an attachment chamber while keeping the cells in the chamber stationary in the field of view of the microscope. A practical example of this is when using a Strex cultured cell chamber. When the chamber is stretched the cells translate out of the field of view of the microscope because it is impossible to position the cells of interest exactly at the centre of the chamber (see Figure 31). The Assay Stretch function compensates for this translation and allows continuous observation of the sample, even under high magnification, during the entire stretch cycle.

The Assay Stretch function is a sinusoidal movement which starts at the minimum of the sine wave. This means the movement will stretch the sample from the starting length and then return the sample to the starting length (this is equivalent to a sine wave with an initial phase of 270 degrees which makes Lo the minimum length in the stretch motion).

Setup

The stretch chamber should be attached to the left and right XYZ stages, aligned in the X, Y and Z axes and then stretched along the X axis to the desired initial length. Once you have identified the cells of interest move them into the center of the field of view of the microscope. This can all be accomplished with the normal functions of the 820A Dual XYZ Motion Controller. If you don't want to cause any length changes to the chamber during this move, then make sure you Slave the right and left stages together. When the cells of interest are positioned correctly press the **Set Lo** button on the Assay Stretch window (see Figure 32) to save the position of the left and right XYZ stages before setup procedures are done. This will allow you to return to the initial state afterwards.

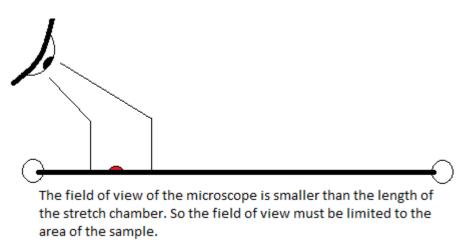


Figure 30 Field of View Limitations when Stretching

To keep the sample in the field of view of the microscope, while a stretch motion is executed, a measurement of the amount of translation must be made. Enter a small length into the **Length Change** (Delta L) entry box, this length should be large enough to result in a noticeable translation of the sample but small enough that the sample stays within the field of view. Now mark the position of a distinguishable point of the sample, this point will be used as a reference for measurement. Press the Perform Delta L Move button. At this point the left and right probe will move to complete a stretch of the given magnitude (Length Change (Delta L)) along the X axis. Mark the new position of the distinguishable point and measure the length change between the two points. Enter this value into the Position Change of Followed Point (Delta P) text entry box (sign matters + is a movement to the right, - is a movement to the left). This measurement allows the 820A Dual XYZ Motion Controller to calculate the required translation that must be added to the sinusoidal stretch to keep the cell stationary in the field of view of the microscope. Return to the initial state by pressing the Return to Lo button. If during the calibration the cell does not move a measurable amount, then increase the Length Change (Delta L) value. If during the calibration the cell moves outside of the field of view, then reduce the Length Change (Delta L) value.

Assay Stretch	
Settings	
Length Change (Delta L):	25.00 erform Delta L Move
Position Change of Followed Point (Delta P):	: 7.40 💂 um
Set Lo	Return to Lo
Cycles Stretc	ch Magnitude
4	75 um
Execu	ute

Figure 31 Assay Stretch Window

Note: The addition of the translation movement to the stretch movement may cause one of the probes to move a larger distance during the stretch move than otherwise expected. Please position the probes so that a movement equal to the **Stretch Magnitude** length of either probe will not cause physical damage.

Now that the translation has been measured you are ready to define the stretch motion and then execute it.

Cycles defines the number of stretch oscillations you wish to perform

Execute button runs the Assay Stretch function using the values set on the Assay Stretch Settings window.

Settings > Assay Stretch Settings

The Assay Stretch Settings window (see Figure 33) allows the user to specify the stretch mode and the magnitude and frequency of the stretch.

Delta L Stretch Mode sets the origin point from which the stretch is performed during the **Length Change** (Delta L) move.

Note: The stretch itself must have a unique origin point to preserve the sample's position inside the microscopes field of view.

Stretch Magnitude sets the magnitude of the length change of the stretch chamber when the stretch oscillation is executed. Both the magnitude and the units can be set by this control.

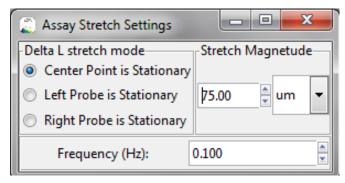


Figure 32 Assay Stretch Settings Window

Frequency sets the frequency of the oscillation, this frequency is limited by the maximal velocity of the left and right XYZ stages. This maximum velocity cannot be determined until both a **Length Change** has been executed and a **Position Change** measured. Upon Execution the program will determine whether the frequency of the stretch can be achieved with the given Stretch Magnitude.

3.4 Web Browser Interface

The 820A's web interface provides access to the more advanced functions through a platformindependent web-page interface. No specialized software is required to connect to this interface, a modern web browser (such as Google Chrome) is all that is required. Thus you can control the 820A with any device that supports a web browser such as a smart phone or tablet.

Note: not all functions are available through the web browser interface. The web browser does not allow either the Cell Attachment module or the Assay Stretch module.

Open a browser and enter HTTP://IPaddress where IPaddress is the IP address is in the form xxx.xxx.xxx, where each x is a number. Go to **Settings > Instrument Configuration > Options > Available 820A Devices** to look up the IP address. The main page should open and look like Figure 34.

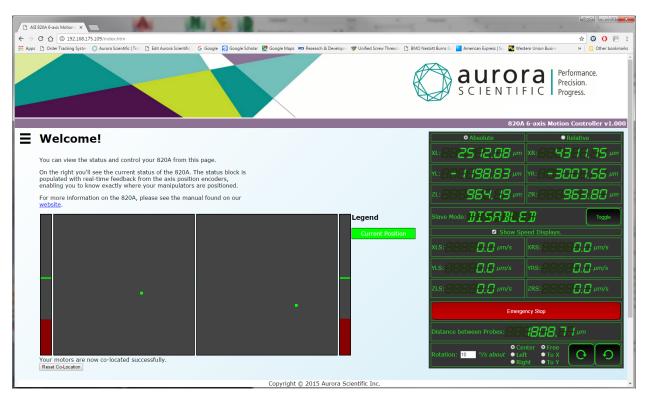


Figure 33 820A Web Interface

3.5 Advanced TCP Interfaces

The 820A exposes a number of interfaces on several TCP sockets. These interfaces allow the advanced user to develop their own custom software to control the 820A. These interfaces are described fully in Appendix 7.1.

4 Calibration, Instrument Tuning and Maintenance

4.1 Calibration

The calibration function determines the overall distance that each stage can travel between its physical limits. Calibration is only required when you first receive the 820A and anytime you change the stages you are using or change which port a particular stage is plugged into on the 820A front panel. The calibration values are stored in non-volatile memory inside the 820A and are read by the 820A each time it is powered on. The calibrate function is available via the Aurora Scientific Software, the web interface, or through the custom software TCP channels. When a calibrate function is commanded, the 820A runs all six axes to the minimum and maximum physical limits. Before issuing this command ensure that the stages can safely move to their physical limits without colliding with your bath or any other delicate instrumentation.

4.2 Home

On each power cycle, the 820A requires that it is 'homed' prior to the usage of the advanced functions. The home operation is available via the Aurora Scientific Software, the web interface, or through the custom software TCP channels. When a Home operation is commanded, the 820A runs all six axes to the maximum physical limits (the same location as is specified by the Retract function). Before issuing this command ensure that the stages can safely move to their physical limits without colliding with your bath or any other delicate instrumentation. This operation sets the physical location of the stages.

4.3 Software Limits

Once a home calibration has been run, the user can set more restrictive software limits to prevent accidental collisions between the probe tips and the bath. Similar to the home calibration, this operation is available via the Aurora Scientific Software, the web interface, or the custom software TCP channels. We recommend that you set these limits to prevent accidental damage to your instrumentation.

4.4 Maintenance

No maintenance is required for the 820A Dual XYZ Controller. However, please ensure that you keep your Siskiyou Corporation 7600-series stages free from debris and contaminants, and exercise great caution during cleaning operations. The precision drive components are sensitive to moisture and dirt.

5 Performance Guarantee, Technical Support, Warranty and Repair Information

Aurora Scientific is dedicated to providing you with products that allow you to meet your research goals. For this reason we offer a performance guarantee, technical support and a new product warranty. Our performance guarantee ensures you purchase the correct instrument for your research. Technical assistance is always free and will be available for the life of your product. If you do have a problem with a product then please know that all Aurora Scientific products are covered by a three-year warranty covering both parts and labour. If you need to return a product to us for repair then consult the final section of this chapter for returns information.

5.1 Performance Guarantee

Our performance guarantee states: if for any reason a new product does not meet your research needs then you can return it to Aurora Scientific for exchange or a full refund. The performance guarantee only applies to new products and must be exercised within 60 days of receipt of the instrument.

5.2 Technical Support

Technical assistance is always free and will be available for the life of your product. Please don't hesitate to contact us if you have any technical support issues. Contact us by telephone, email, fax, or regular mail.

5.3 Technical Support Contact Information and Return Shipping Addresses

Canada, USA, South America, Middle East, Africa

Aurora Scientific 25 Industry St. Aurora, Ontario, CANADA L4G 1X6 Attn: RMA Returns Tel: +1-905-727-5161 Fax: +1-905-713-6882 Email (all Aurora Scientific Offices): <u>techsupport@aurorascientific.com</u> Web Site (all Aurora Scientific Offices): <u>www.AuroraScientific.com</u>

Europe

Aurora Scientific Europe Hilton House 3 Ardee Road Rathmines, Dublin 6, Ireland Attn: RMA Returns Tel: +353-1-525-3300 Fax: +353-1-443-0784

Asia, Australia, New Zealand

Aurora Scientific Asia Unit B, 10/F Charmhill Centre 50 Hillwood Road Tsimshatsui, Kowloon, Hong Kong Attn: RMA Returns Tel: +852-3188-9946 Fax: +852-2724-2633

Distributors Japan

Kantoh Electronics Co., Ltd. 1-25-14, Nakacho, Meguro-ku Tokyo, 153-0065, Japan Tel: +81-03-5773-5028 Fax: +81-03-5773-5029 Email: <u>info@Kantoh-elec.co.jp</u> Web site: <u>http://www.kantoh-elec.co.jp</u>

5.4 Warranty

Products manufactured by Aurora Scientific Inc. are guaranteed to the original purchaser for a period of three (3) years. Under this warranty, the liability of Aurora Scientific is limited to servicing, adjusting and replacing any defective parts that are of Aurora Scientific manufacture. Aurora Scientific is not liable to the customer for consequential or other damages, labour losses or expenses in connection with or by reason of the use or inability to use the products manufactured by Aurora Scientific.

Guarantee of parts and components not manufactured by Aurora Scientific shall be the same as the guarantee extended by the manufacturer of such components or parts. Where possible such parts returned to Aurora Scientific will be sent to the manufacturer for credit or replacement. Ultimate disposition of these items will depend upon the manufacturer's decision.

All shortages must be reported within ten (10) days after receipt of shipment.

Except where deviations are specified in literature describing particular products, the limited warranty above is applicable to all Aurora Scientific products, provided the products are returned to Aurora Scientific and are demonstrated to the satisfaction of Aurora Scientific to be defective.

Transportation costs of all products returned to Aurora Scientific must be borne by the customer and products must be returned to Aurora Scientific within three years after delivery to the original purchaser. Aurora Scientific cannot assume responsibility for repairs or changes not authorized by Aurora Scientific or damage resulting from abnormal or misuse or lack of proper maintenance.

Repair or service work not covered under the limited warranty will be billed at current service rates.

NO EXPRESS WARRANTIES AND NO IMPLIED WARRANTIES WHETHER FOR MERCHANTABILITY OR FITNESS FOR ANY PARTICULAR USE, OR OTHERWISE OTHER THAN THOSE EXPRESSLY SET FORTH ABOVE WHICH ARE MADE EXPRESSLY IN LIEU OF ALL OTHER WARRANTIES, SHALL APPLY TO PRODUCTS SOLD BY AURORA SCIENTIFIC INC, AND NO WAIVER, ALTERATION OR MODIFICATION OF THE FOREGOING CONDITIONS SHALL BE VALID UNLESS MADE IN WRITING AND SIGNED BY AN EXECUTIVE OFFICER OF AURORA SCIENTIFIC INC.

5.5 Returning Products to Aurora Scientific for Repair

There are a few simple steps that must be completed before returning your product to Aurora Scientific.

1. Obtain a Return Material Authorization number (RMA#).

<u>Go to our website</u> or contact our technical support department to obtain a RMA #. We require the serial number of the product along with your contact information, i.e., your name, institution, phone number and email address.

2. Package your instrument.

Use the original packaging materials if available. If you do not have original packaging, then ensure that the product is wrapped in bubble pack and placed in a sturdy corrugated cardboard box.

3. Prepare Customs documents.

Canadian Clients: no customs documents are required, skip to step 4.

European Clients: no customs documents are required, skip to step 4 and ship to Aurora Scientific Europe.

Asia, Australia and New Zealand Clients: no customs documents are required, skip to step 4 and ship to Aurora Scientific Asia.

USA and Rest of the World Clients: We will supply you with a Commercial Invoice (CI) that must be included with the shipment. Please look for the CI in our return email message. You must sign and date the commercial invoice.

Place three (3) copies of the CI in an envelope and mark the outside CUSTOMS PAPERS ENCLOSED. Attach the envelope to the outside of the box.

4. Choose a shipper and prepare the waybill.

European Clients: ship your instrument to Aurora Scientific Europe in Dublin, Ireland.

Asia, Australia and New Zealand Clients: ship your instrument to Aurora Scientific Asia in Hong Kong.

Canadian, USA and all other Clients: ship your instrument to Aurora Scientific in Ontario, Canada.

You may ship your instrument back to us via the courier of your choice or via parcel post. If possible we prefer that you ship via FedEx. You are responsible for both the shipping and brokerage charges so please mark the waybill accordingly. Please don't ship freight collect. Shipments sent freight collect will be received but you will be invoiced for the shipping charges when your instrument is returned.

5. Prepare and send a purchase order.

After we receive the instrument we will evaluate it and contact you with the estimated repair cost. We require a purchase order before we can repair and return your instrument. Please fax or email us the purchase order at your earliest convenience.

6 Specifications

820A Dual XYZ Motion Controller

Positioning Accuracy [µm]:	±0.02
Maximum Speed [mm/s]:	1.7
Minimum Speed [µm/s]:	2.0
Encoder Resolution [µm/tick]:	0.005
Motor Drive Voltage [VDC]:	±15
Stage Connectors:	DB-25
Controller Connectors:	DB-15
Hand Controllers:	Siskiyou push button (MC-1000e), Siskiyou Joystick (MC-1000e-J)
Stages:	Siskiyou 7600
Communication Port:	Ethernet (RJ-45)
Communication Speed [Mbps]:	10/100

General Specifications

Operating Temperature [°C]:	0 - 40
Power Required:	100-240 VAC, 50/60 Hz.
Power Consumption [W]:	50
CAUTION	Replace Fuse with same Type and Rating
Fuse Type:	IEC 127-III: 5x20mm, 250V Time Delay
Fuse Rating:	110/125VAC – T 2.0A
	220/240VAC – T 1.0A
Weight [kg]:	3.1
Dimensions [cm]:	21W (1/2 rack mount) x 25D x 9H (2U)

820A Software

Recommended PC Specific	ations
Processor:	Quad-core Intel or AMD
RAM:	4 GB
Hard Disk:	500+ GB
Operating System:	Windows 7/8/10, Linux
Software Functions	
Move Controls:	On-screen arrows up/down, X/Y +/-, XY diagonal +/-
Stage Control:	Left XYZ, Right XYZ, Both, Slaved (one controller controls both left and right stages allowing the sample to be translated without any length change)

	allowing the sample to be translated without any length change)
Move Type:	Jog or Continuous
Speed Controls:	Fast/slow user defined pre-sets, speed entry box 2μ m/s – 1.7 mm/s
Position Readout:	Digital indicators of position of all 6 stages (can be zeroed at any point)
Position Visualization:	Graphical representation of position of all 6 stages
Software Limits:	User settable +/- limits on all 6 axes
Locate Probes:	Allows locating probes with respect to each other (required for length output
	and must be performed prior to Stretch/Shorten Move, Rotate/Align Probes)

Movement Functions:	Home – determines physical limits of all 6 stages
	Retract -Moves all stages up, out and back to their physical limits
	Waypoints – up to 100 user defined points
	Move to Position – vector move to user entered position coordinates
	Virtual Axis – user defined injection/retract motion in XZ plane or general vector movement
	Sinusoidal Motion – user defined sinusoidal motion of probes, specify frequency and amplitude
	Stretch/Shorten Move - moves the probes along an imaginary line drawn
	through the end points of the two probes
	Rotate/Align Probes - allows for the user to align samples attached to the probes along an axis without varying the length between the probes
Experiments:	Intact Cell Attachment – provides user defined waypoints and shear movement required when applying glue to a probe prior to attaching an intact cell
	Assay Stretch – provides sinusoidal stretch of a sample with translation, this allows the center of the sample to remain stationary in the field of view of the microscope

7 Appendices

7.1 Appendix A: 820A TCP Communications Structure

To communicate with an 820A, you must connect it to your PC via Ethernet. The 820A will connect to the network via DHCP. It can be found by software on a networked computer by sending a UDP Broadcast message with the word "Discovery" on port **30303** to the broadcast IP address **255.255.255.255.255**. The 820A will send a response to this message detailing its IP address and serial number.

The 820A exposes multiple TCP communications ports. Firstly, you can access its webpage at http://<IP Address>/ on the network. This page will give you some debugging information about the current position as well as allowing some simple control.

On TCP port **820** the 820A exposes information about the current position of all encoders as 24 bit unsigned integers from zero to 16,777,215. (0x000000 to 0xFFFFF). Values greater than 0x800000 are representative of a position nearer to the negative limit than the starting point.

Port 820 Responses look like this:

WL: 0 XL: 513208 YL: 16424191 ZL: 1928098 WR: 0 XR: 0 YR: 0 ZR: 0

On TCP port **821**, the 820A exposes information about the status of the Siskiyou MC1000e handheld keypads plugged into the L Control and R Control ports on the 820A. For each of the 4 axes on the keypad, you get a +, 0, - marker for whether the user is pushing the button, and the analog value from the axis which denotes the desired speed, as set by the knobs on the keypad.

Port **821** Responses look like this:

L1: 0 L1A: 61 L2: 0 L2A: 61 L3: 0 L3A: 61 L4: 0 L4A: 59 R1: 0 R1A: 132 R2: 0 R2A: 136 R3: 0 R3A: 134 R4: 0 R4A: 136

On TCP port **822**, the 820A listens for incoming commands. All commands are formatted as: ***?hh#ss**, where ***** is the start of frame character, **?** is the upper case command character being sent, hh are parameter or data values (note: depending on the command there may be more or less h parameters than listed here), **#** is the end of frame character, and ss are 2 lower case hex digits which are the check sum for the characters from ***** through **#** inclusive. The commands available are described later in this document.

Checksums are computed by summing the integer values of the characters from * through # inclusive, modulus divided by 256 and returning the result as a 2 digit lower-case hexadecimal value.

On TCP Port **823**, the 820A Echoes back commands and their results. Most commands are simply echoed back as they were received to acknowledge their receipt. If a problem is encountered, a descriptive error message is echoed back instead. A few commands return information about what they did instead of echoing the inbound command back. For example, the Go To Position command returns information about the component vectors calculated.

On TCP Port **824**, the 820A exposes information about the current position of the software limits. These can be set with a command from the user, and they will be automatically set at the physical limits if one is hit.

Port **824** responses look like this:

WLP: 8388607 WLN: 8388608 WRP: 8388607 WRN: 8388608 XLP: 8388607 XLN: 8388608 XRP: 8388607 XRN: 8388608 YLP: 8388607 YLN: 8388608 YRP: 8388607 YRN: 8388608 ZLP: 8388607 ZLN: 8388608 ZRP: 8388607 ZRN: 8388608

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Commands

The following is a list of the valid commands, and examples of their usage and the replies returned from the 802D. In all cases the instrument acknowledges a command as soon as it is received by either a successful reply or by an unsuccessful reply.

Go At <u>S</u>peed

The Go At Speed command is used to set the 820A's motors to a desired speed. This is typically used for manual control to an unknown point. Each motor is given a 16 bit hexadecimal speed, where 0xFFFF is full speed forward, 0x0000 is full speed reverse, and 0x8000 is stopped. All hex values should be transmitted in lower case.

Comm	nand	* S XLhhhhYLhhhhZLhhhhXRhhhhYRhhhhZRhhhh#ss
eg.	StopAll	*SXL8000YL8000ZL8000XR8000YR8000ZR8000#29

Go To Position

The Go To Position command is used to send the 820A to a specified position on all 6 axes, with each XYZ stack moving at a specified vector speed to the destination. Each axis is given a 24bit hex encoder count to go to, and each XYZ stack is given a 16bit hex speed to travel along the vector at, where 0x7FFF is max speed.

Command *PXLhhhhhhYLhhhhhhZLhhhhhhXRhhhhhhYRhhhhhhZRhhhhhhLShhhhhRShhhh#ss eg. GoToZeroMaxSpeed *PXL000000YL000000ZL000000XR000000YR000000ZR000000LS7fffRS7fff#29

Go To <u>H</u>ome

The Go To Home command is used to command the 820A to execute a home routine, to go and find the physical limits of the travel.

Command	* H #ss
eg.	*H#29

Set <u>L</u>imit

The Set Limit command is used to set a software limit on the instrument's position. Once a software limit is set, the manual control options will not allow the position of the instrument to exceed the set limit by more than 15nm. In this command aa is the axis in upper-case letters, d is the direction (P or N) and then the 24 bit hex encoder position to limit at. Alternatively, instead of an axis, sending Q queries for physical limits.

Command	*Laadhhhhhh#ss
eg.	*LXLP01fcd4#f6
Query	*LQ#ss

<u>I</u>dent

This command is used to Identify the 820A.

Command *I#ss eg. *I#2f Successful reply:ASI 820A 6 Axis Motion Controller ©2016 Aurora Scientific Inc.

Sla<u>V</u>ing

This command is used to set or query the current Slave mode. Master is 0 for off, 1 for Left, 2 for Right. X,Y,Zinvert is 0 for no invert, 1 for invert.

Query Command	* VQ #ss
Reply eg.	Master: 0 Xinv: 0 Yinv: 0 Zinv: 0
Set Command	*VSM0X0Y0Z0#ss

<u>Co-Location</u>

These commands are used to set or query the current Co-Location Space. L sets the left origin to the current position. R sets the right origin to the current position. Q queries the current origins.

Query Command	*CQ#ss
Reply eg.	CLSpace Left Origin: X: n Y: n Z: n Right Origin: X: n Y: n Z: n Configured: n
Set Command	*CL#ss or *CR#ss

<u>R</u>otate

This command is used to rotate the probes. You must set a direction (0 = STOP, 1 = CCW, 2 = CW), a rotational speed (in deg/s), your rotation center (0 = CENTER, 1 = LEFT, 2 = RIGHT), and your rotation limit (0 = FREE, 1 = TO X, 2 = TO Y). Like the GoAtSpeed command, this command times out and stops rotation if not sent again.

Rotate Command ***R1S10A2L0**#ss (rotate CCW at 10 deg/s about the Right probe freely).

Si<u>N</u>e

This command is used to execute a sinusoidal stretch. You must set amplitudes in each axis (24 bit hex encoder counts), a sine frequency (in base 10 millihertz), and a number of cycles (also in base 10) to move through.

Sine Command *NXLhhhhhhYLhhhhhhZLhhhhhhXRhhhhhhYRhhhhhhZRhhhhhhFnnnnnCnnnnAnnn#ss

S<u>T</u>retch

This command is used to execute a linear stretch or relax. You must set the stretch amount (in nm), a direction (P = positive, N = negative), a stationary point (0 = CENTER, 1 = LEFT, 2 = RIGHT) and a speed (16 bit hex, 0-7FFF) to move at.

Stretch Command, eg. ***T00001000DPA0S7fff**#ss (stretch 1 um in the positive direction keeping the center stationary at a speed of 7fff).

AXis Setup

These commands are used to set or query the current Axis Configuration. S sets the setup, encoder resolution in hex, invert = 1, no invert = 0, Q queries the current setup.

 Query Command
 *XQ#ss

 Reply eg.
 Axis Setup: XL: 198472 I: 1 YL: 198472 I: 0 ZL: 198472 I: 0 XR: 198472 I: 0 YR:

 198472 I: 0 ZR: 198472 I: 0
 Set Command

 *XSXLhhhhhhl1YLhhhhhl0ZLhhhhhhl0YRhhhhhhl0ZRhhhhhhl0#ss