INSTRUCTION MANUAL

Models 802A

Permeabilized Fiber Test Apparatus

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1.0 Introduction

The 802A permeabilized fiber test apparatus was designed to enable physiology researchers to easily test permeabilized fibers with an ASI model 312B or 322B high-speed length controller and an ASI series 400A force transducer.

The 802A consists of a 6-well stainless steel bath plate that includes right-angle prisms on either side of bath 1 and a glass bottom on all 6 wells, 2 TEC heater/coolers, water-cooled heat sink plate, 2 stepper motors for up/down movement and bath-to-bath indexing, micro-processor based electronic control system, remote control, high-speed length controller motor mount, 400A series force transducer mount and micrometer drive XYZ translation stages for motor and force transducer positioning. Also included are mounting screws and a set of Imperial Allen keys. All parts are manufactured from corrosion resistant materials (anodized aluminum, stainless steel, Delrin and Lexan).

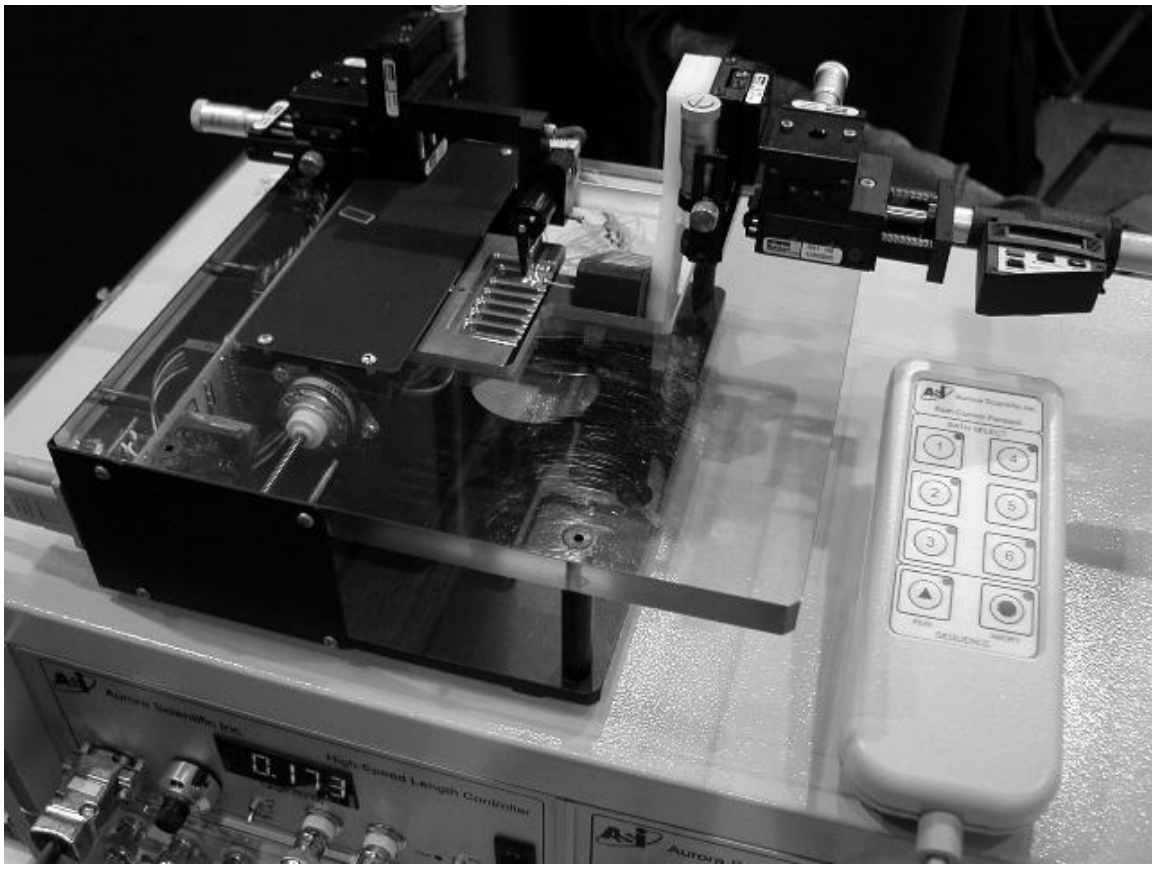

Figure 1 802A Apparatus with Pendant Control.

The 802A works by moving the bath plate relative to the fiber. The fiber is clamped between the output tube of the force transducer and the lever arm of the high-speed length controller. In operation the researcher selects the desired bath by either entering the bath number on the front panel of the controller or by simply pressing the desired bath number button on the remote pendant control. When a new bath is selected the bath plate moves

downwards and then indexes to the desired bath. When it arrives at the new position the plate automatically rises to the operating position. The electronic controller includes a digital display that shows the current bath position. The pendant controller has LEDs next to each bath switch and the LED lights to indicate the current position of the bath plate. The bath-to-bath index time is approximately 1.2 seconds between adjacent baths and approximately 3.5 seconds from one extremity to the other. This time includes the time required to lower the bath, move to the new position and then raise the bath plate to its working position.

The bath plate is manufactured from stainless steel and has six baths machined in its upper surface. This number allows for test protocols involving multiple solutions without the need to empty and re-fill baths. All baths have a glass bottom to allow for microscope observation of the fiber or laser-based sarcomere length measurement. The top of each bath features a machined recess to accept a glass cover slide. Bath 1 is larger than the rest as it was designed to be the bath in which the fiber is attached to the force transducer and length controller. Bath 1 also includes right angle prisms on both sides of the bath to allow the fiber to be viewed from the side as well as from the top.

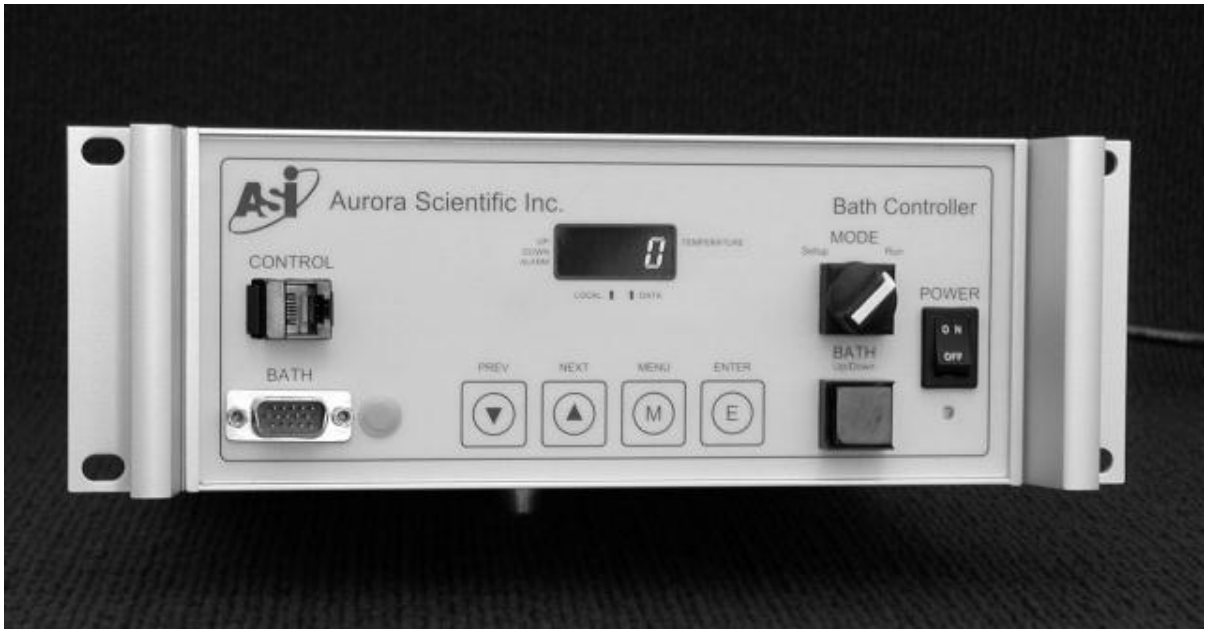

Figure 2 802A Bath Controller

Both the high-speed length controller and the force transducer are mounted on XYZ micrometer translation stages to allow them to be positioned relative to the bath plate. Even though the 802A accurately controls the position of the bath plate with respect the length controller and force transducer the position of the lever arm and force transducer relative to the plate must be adjusted before use. The bath-to-bath positional accuracy is better than +/- 0.05 mm (+/-0.002") in both the x (horizontal) and z (vertical) directions.

The 802A Bath Controller features a programmable motion control sequencer. This sequencer allows the researcher to automate the movement of the bath plate. The sequencer program is easy to use and consists of a six-step program that has two variables for each step.

The first variable sets the desired bath and the second sets the amount of time to wait before proceeding to the next step. A repeat command is also provided. Pressing the Run button on the remote pendant control starts the sequencer. An abort button is also provided to stop the sequence program at any time. The bath plate must be positioned at the bath listed in step 1 of the sequence before pressing the Run button. If the bath plate is not located at the bath number that corresponds to the first step in the sequence then the sequence will not run.

A thermocouple holder is attached to the motor mount to allow the researcher to monitor the temperature of the bath in use. A second thermocouple is placed in a wet well in the bath plate for temperature feedback to the TEC controller.

1.1 Specifications

Bath Plate	
No. of Baths:	6
Bath Plate Material:	304 Stainless steel
Right-Angle Prisms:	3x3x6mm, aluminized hypotenuse with incolnel overcoat and black paint overcoat.
Bath Dimensions:	
Bath 1:	18.67mm (0.735”) L x 4.83mm (0.190”) W x 3.30mm (0.130”) D
Baths 2 – 6:	18.67mm (0.735”) L x 3.80mm (0.150”) W x 3.30mm (0.130”) D
Bath Volume:	
Bath 1:	300µl
Baths 2 – 6:	235µl
Centerline Distance X-axis:	
Bath 1 – 2:	8.89 mm (0.350”)
Baths 2 – 6:	6.35 mm (0.250”)
Angular Movement, Z-axis:	3.5°
Excursion, Z-axis:	
Max. (at FT end of Bath):	4.93 mm (0.194”)
Min. (at lever end of Bath):	3.78 mm (0.149”)
Indexing Time:	
Bath 1 – Bath 2:	1.4 seconds
Adjacent Baths in range 2-6:	1.2 seconds
Bath 1 – Bath 6:	3.5 seconds
Thermoelectric Coolers	
No. of TECs:	2
Power:	33 W
Voltage:	15 V
Stepper Motors	
No. of Motors:	2
Voltage:	12 V
Movement:	25µm (0.001”)/step
Warm Up Time:	10 seconds to rated accuracy
Power Requirements:	100–240VAC±10%, 50/60Hz, 2 amps max.

2.0 Apparatus Setup

2.1 Unpacking

Unpack the apparatus from the two shipping boxes. One box contains the controller, pendant control and cables. The other holds the 802A apparatus, XYZ translation stages, Allen keys, mounting screws and the TEC controller cable. The two XYZ translation stages are assembled and boxed separately. Tie wraps are used to hold the stages into the shipping boxes. Open the boxes and cut the tie wraps to remove the stages. The main part of the 802A is shipped assembled however the user must attach the XYZ translation stages, the motor, force transducer, TEC controller and water lines.

2.2 Attaching the 802A to a Parker 4450-DM Translation Stage

The aluminum base plate of the 802A has four drilled and countersunk holes that match the mounting holes on a Parker 4450-DM translation stage. Before mounting the motor or force transducer we recommend that the 802A be attached to the Parker 4450. Four ¼-20 flat head screws are provided for this purpose. The easiest method of attaching the 802A to the 4450 is to remove the aluminum base plate from the 802A, attach the base plate to the 4450 and then reassemble the 802A. To remove the base plate, first remove the seven 4-40 button head screws that hold the side cover to the black aluminum base plate. Then remove the four 8-32 flat head screws that can be found in the top of the Lexan plate. Once these are removed simply lift the 802A off of the aluminum base plate. What you should see is the black aluminum base plate with four aluminum posts mounted to it. Now using the ¼-20 flat head screws provided attach the base plate to the 4450. Place the 802A back on the base plate and reattach the 802A to the base plate using the 8-32 flat head screws. Ensure that the side cover is refastened using the 4-40 button head screws.

2.3 Attaching the XYZ Micrometer Translation Stages to the 802A

In this manual we will define the X-axis to be the horizontal direction in which the bath plate moves, the Y-axis to be the horizontal direction that would shorten or lengthen a fiber and the Z-axis to be vertical direction.

Two XYZ micrometer translation stages are included with the 802A. The one for the force transducer includes a digital micrometer for the Y-axis and a Delrin transducer mount. The XYZ stage for the motor includes the motor mount and an 18G stainless-steel hypodermic tube for holding a Physitemp model IT-18 thermocouple microprobe.

2.3.1 Mounting the Motor Mount XYZ Translation Stage

The motor XYZ stage mounts at the rear left side of the 802A top plate. Two 4-40 tapped holes are provided in the Lexan plate.

The Z-axis stage must be removed in order to gain access to the mounting holes in the Y-axis stage. Remove the right-angle bracket from the Y-axis stage by removing the two 4-40 socket head cap (SHC) screws. Unlock the stage lock screws and turn the Y-axis micrometer counter clockwise to uncover the mounting hole in the base of the stage. Use one of the 4-40UNC x 3/4” SHC screws provided to attach the Y-axis stage to the Lexan base

plate. Push the stage with your finger in the opposite direction to uncover the other mounting hole. Install the other 4-40UNC x 3/4" SHC screw. Align the stage to be perpendicular to the left side of the 802A base plate and then firmly tightened the screws taking care not to over tighten them as you can strip the threads out of the Lexan. Re-attach the Z-axis right-angle bracket complete with the Z-axis stage and the motor mount. Align the bracket with the X-axis stage and tighten the screws. Note: it may be necessary to adjust the micrometers on the X and Y stages in order to properly align and mount the stages.

2.3.2 Mounting the Force Transducer XYZ Translation Stage

The force transducer XYZ stage mounts at the rear right side of the 802A top plate. Two 6-32 tapped holes are provided in the Lexan plate.

Remove the Z-axis right-angle bracket from the Y-axis stage by removing the two 4-40 socket head cap (SHC) screws. Unlock the stage lock screws and turn the Y-axis micrometer counter clockwise to uncover the mounting hole in the base of the stage. Use one of the 6-32UNC x 3/4" SHC screws provided to attach the Y-axis stage to the Lexan base plate. Push the stage with your finger in the opposite direction to uncover the other mounting hole. Install the other 6-32UNC x 3/4" SHC screw. Align the stage to be perpendicular to the right side of the 802A base plate and then firmly tightened the screws taking care not to over tighten them as you can strip the threads out of the Lexan. You can re-attach the Z-axis right-angle bracket complete with the Z-axis stage and the Force Transducer mount at this time or wait and re-attach it after the force transducer has been attached to the mount (see section 2.6 below). Align the bracket with the X-axis stage and tighten the screws. Note: it may be necessary to adjust the micrometers on the Y or X-axis stages to properly align the stages.

2.4 Instrument Checkout and Positioning System at Bath 1

Prior to attaching the motor and force transducer it is recommended that you check the operation of the 802A and ensure that the bath plate is located in the Bath 1 position. Place the 802A on a lab bench in the orientation shown in Figure 1. Attach the 15-pin cable between the controller and the 802A. Tighten the connector screws to ensure a firm connection at both ends. Using the supplied power cord plug the controller into an appropriate AC power outlet. Attach the remote pendant control to the connector on the front panel of the controller. Please note the pendant should only be plugged into the controller under no circumstances should the pendant be plugged into any other type of device (the pendant uses an RJ45 (Ethernet-type) connector but it is not compatible with a computer network). At this point it is not necessary to attach the water lines or the thermoelectric controller. Our aim at this point is to position the bath plate at the bath 1 location prior to attaching the motor and force transducer.

Turn the power on using the power switch located on the front panel of the controller. The display should light and show the current position of the bath plate as well as the up/down status of the plate. A zero on the display indicates that the bath plate is in an unknown position between stations or the cable between the controller and the bath is not attached. Refer to Figure 4 for a detailed drawing showing the front panel controls and location of the status LEDs. Press and hold the bath 1 number on the pendant control. After about 0.5 seconds the bath plate should lower, then move to position 1 and rise. The system

is now in its home (position 1) location and you can proceed to attach the motor and force transducer. You will also note that the controller has an Up/Down switch and a Setup/Run switch. The Up/Down button will move the plate up if it is down and down if it is up. Repeated presses of this button will simply cycle the bath plate up and down. The Setup/Run switch controls the operating mode of the 802A. In the Setup position the bath will not rise at the end of a movement and the manual up/down button located on the side of the 802A will be activated. In the Run position the manual up/down button is disabled and the bath plate will automatically rise at the end of each movement. Try pressing a bath number when the Setup/Run switch is in both positions and observe the difference. When in Setup try pressing the manual up/down button on the side of the 802A. The bath plate will rise slowly as long as the button is pressed. When the button is released the plate will return to the down position. The Up/Down switch on the front panel of the controller is operational in both modes. Further details of the controller are provided in chapter 4.

If desired you can now press any desired bath number to observe the operation of the 802A. Please note that in order to prevent unwanted movement of the bath plate a delay has been built into the pendant controller. The pendant control buttons must be pressed and held for about 0.5 seconds for an action to take place. A button press of shorter duration will be ignored.

The front panel controls on the controller can also be used to control the position of the bath plate. Simply press the up or down arrows until the desired bath number is shown on the display. Then press the E (Enter) button to command the controller to perform the movement.

Before attaching the motor and force transducer it is recommended that you place the controller in Setup mode and then press the Up/Down switch to lower the bath plate.

2.5 Attaching the Motor

Before attaching the motor it is recommended that the X and Y motor mount translation stages be positioned at about their mid travel location (7 on the micrometer). The Z stage should be raised (about 2 on the micrometer).

In some cases it is easiest to attach the motor by first removing the motor mount clamp ring. The clamp ring is held on with a single 6-32 SHC screw. Insert the motor into the motor mount and rotate the motor so that the connector is at the top. It is important to do this so that the motor connector and cable don't interfere with the motion of the bath plate. Gently tighten the motor mount ring clamp screw to hold the motor in position

Re-attach the motor clamp ring complete with the motor to the mount. The clamp ring can be rotated to allow the axis of the motor to be tipped backwards. This moves the motor away from the bath to provide a little more clearance for attaching the fiber and observing the experiment. The motor can also slide within the mount. Attach the arm to the motor at this point and orient the arm so that it is pointing down. Loosen the clamp ring screw if necessary and slide the motor in the clamp so that the tip of the lever arm is located in the center of bath 1. Once the motor is in the desired position tighten both the ring clamp screw and the motor clamp screw. If the motor is not inserted far enough into the clamp the back end of the motor can interfere with the bath plate. Also if the clamp is tipped at too large an angle the motor will interfere with the bath plate.

Once the motor is locked in place the XYZ translation stages can be used to fine-tune the position of the lever arm in the bath.

The controller should be turned on and placed in Setup mode. Press the manual up/down switch on the side of the 802A and observe the motion of the plate. The plate should rise slowly. Ensure that the plate doesn't touch the motor housing and that the cable doesn't interfere with the plate. Also ensure that the lever arm is not located too close to the side of the bath. The lever arm should be located near the centerline of the bath. If at any time you observe interference between the bath plate and anything else simply release the manual up/down switch and the bath plate will automatically lower to its down position. Check that the lever arm doesn't hit the glass bottom of the bath.

Ensure that the motor is not located too close to the back end of the bath (the side farthest from the force transducer). If it is then when the plate lowers the arm will not come clear of the bath and the arm/motor could be damaged. Initially position the motor so that the lever arm is located close to the end of the right angle prism, see Figure 3.

The thermocouple support needle may also need to be positioned so that it doesn't interfere with either the lever arm or the side of the bath. Remember that bath 1 is wider than the rest so it is important to check for clearance in one of the other baths as well.

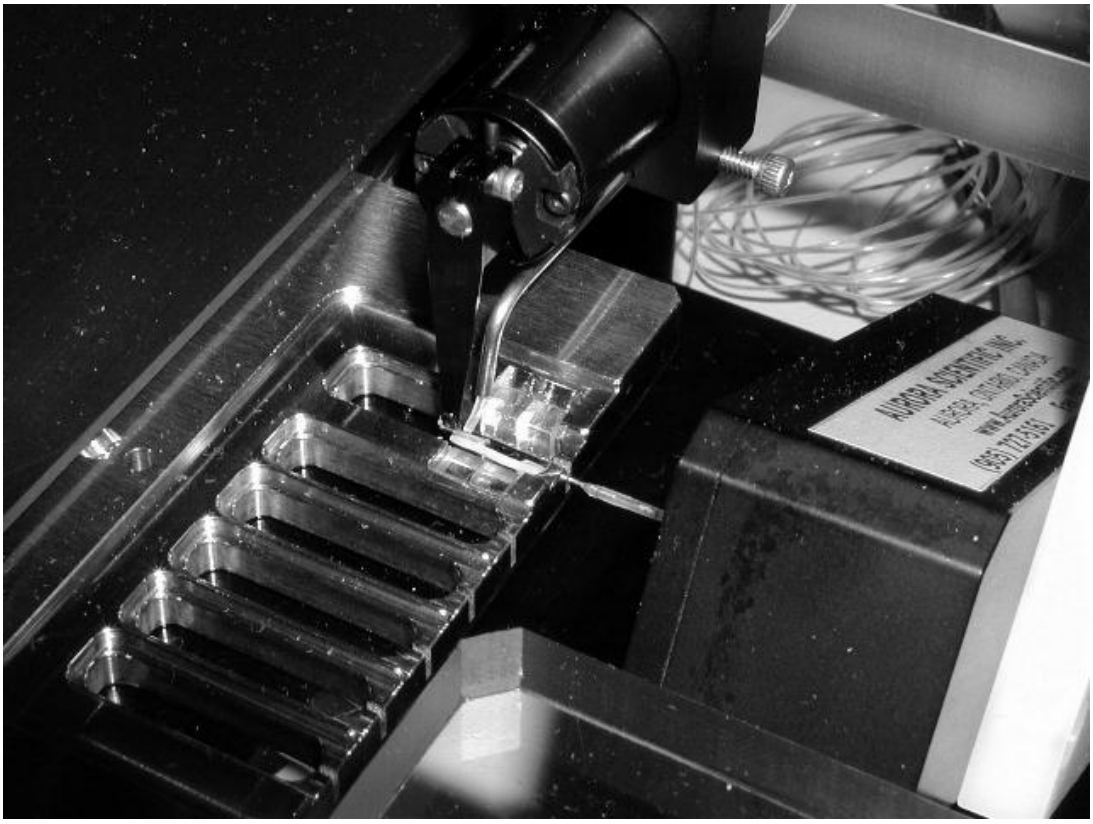

Figure 3 Close-up of Lever and Force Transducer in Bath 1

2.6 Attaching the Force Transducer

Normally a length of fine gauge stainless steel tubing is fastened into the end of the force transducer output tube. This tube then passes through the slot in the end of the bath

plate and the fiber is attached to it. Make sure that the glass output tube on the force transducer is clear of the bath plate. The glass tube is too large to fit in the slot and if the transducer is located too close to the plate the output tube will be broken when the bath is raised. Under no circumstances should the glass tube ever be located above the slot.

As in section 2.5 ensure that the 802A controller is powered and that the bath plate is located on bath 1 and is in the down position before attaching the transducer. A mount for an ASI series 400A force transducer is included with the 802A. Refer to Figure 1 for correct orientation of the force transducer in the 802A apparatus. The force transducer is clamped to an XYZ translation stage located on the right side of the 802A. The transducer is tipped back at a 30-degree angle to the horizontal. This angle provides greater access to the bath.

Position the Y-axis translation stage as far to the right as possible. Note the stage movement is limited by the Z-axis stage. Position the X-axis stage at about the 7 mark on the micrometer dial. Raise the Z-axis stage to near the top. Slide the transducer under the Lexan mounting plate and fasten it to the Delrin mounting plate with the four 4-40 socket head cap screws that are provided. Take care when attaching the transducer that the output tube doesn't strike the bath plate as this could break the transducer. An alternate method of attaching the transducer is to first remove the Z-axis right-angle bracket along with the Z-axis stage from the X-axis stage. Attach the transducer to the disassembled Z-axis stage and then re-attach the stage.

Once the transducer is attached use the X, Y and Z stages to orient the needle attached to the transducer output tube on the centerline of the slot that is at the end of bath 1. Use a microscope to observe the needle and slot positions relative to each other. The controller should be powered and in Setup mode. Press the manual up/down switch on the side of the 802A and observe the motion of the plate. The plate should rise slowly. Ensure that the needle is located in the center of the slot and that it doesn't touch the bath plate. Use the Z-axis stage to set the depth of the output tube, and thus the fiber, in the bath. If at any time you observe interference between the bath plate and anything else simply release the manual up/down switch and the bath plate will automatically lower to its down position.

Ensure that the transducer cable is routed clear of the bath plate. It is best to strain relief the transducer and motor cables by attaching them to the 802A base plate. Cable movement can lead to increased noise on the length and force signals.

2.7 Attaching the Cooling Water Lines

On the left side of the 802A you will see two quick connect tube fittings. The quick connect fittings can be disconnected by turning the outer half of the connector in a counter clockwise direction and then pulling the connector apart. These fittings have a hose barb designed for 1/8" ID vinyl tubing. Connect a source of cool water to one of the quick connects and attach the other to the drain. Ensure that you have a steady flow of water through the cooling plate before turning on the thermoelectric controller. Operating the thermoelectric coolers (TEC) without water flowing can damage the TECs.

2.8 Attaching the Thermoelectric Controller

Two thermoelectric coolers (TECs) are included with the 802A. These devices are rated at 15 volts and 33 watts. An external TEC controller is required. When using an

Alpha Omega series 800 TEC controller the bath plate should reach the set point temperature in about one minute.

Connect the thermoelectric controller to the 3-pin connector located on the left side of the 802A. The mating connector has been supplied with a 6-foot long wire that should be attached to the appropriate connector on the thermoelectric controller. The white wire should be connected to the positive connection and the black wire to the negative.

A wet well is located in the top of the bath plate near the center of the plate. Fill the hole with water and then place the control thermocouple in the hole. Note: the thermocouple can be inserted into the wet well through the angled hole that has been machined into the side of the well. The thermocouple wire can then be taped to the surface of the bath plate. It may be more convenient to first remove the black aluminum cover plate to gain better access to the wet well. Ensure that the cover plate is re-attached prior to using the 802A.

Ensure that water is flowing through the cooling plate and then turn on the thermoelectric controller. Set the desired temperature on the controller and then monitor the actual temperature to ensure that the temperature is approaching the set point. If the temperature is moving in the wrong direction then it is likely that the TECs are connected with the wrong polarity. Turn off the thermoelectric controller and reverse the white and black wires at the controller. Turn the controller back on and observe for the correct control action.

3.0 Bath Controller

The controller includes a microprocessor that monitors and controls the position of the bath plate and a power supply. The switches on the front panel include a Mode switch (1), up/down switch (2), four menu buttons (3,4,5,6) and a power switch (7). See Figure 4 for a diagram of the front panel. The controller comes with a remote pendant control that can be placed up to 6 feet (2 m) from the controller. The pendant has eight buttons on it including a button for each bath plus Run and Abort buttons for the sequencer (see Figure 5).

Motion control can be initiated by using the menu keys on the front panel or by pressing the desired station button on the remote pendant control. To use the front panel select the desired station using the up or down arrow keys (3 or 4) and then press the Enter Key (E, 6) to initiate the movement. The station ID number shown on the front panel will blink during movement and then turn steady when the new position is reached. To use the pendant control simply press and hold the button corresponding to the desired station. The pendant control buttons have been programmed to not respond to short pushes. This ensures that accidental touching of the keys will not initiate movement. The button needs to be held for about 0.5 seconds to initiate movement.

In addition to the normal point-to-point motion control the controller also features a sequencer that allows a series of movements to be pre-preprogrammed and then executed by a single push of the Run button. Each step in a sequence is defined by a desired bath location and a delay time at that bath. The sequencer also includes a repeat function. Details of programming the sequencer are given later in this chapter. To initiate a sequence the bath plate must be located in the bath well that corresponds to the first step in the sequencer program. Simply press the Run button in the Sequencer section of the pendant control to start a sequence. A sequence can be aborted by pressing the Sequence Abort button on the pendant control. As with the bath buttons on the pendant the Run and Abort buttons must also be held for about 0.5 seconds before the command will be processed.

The behaviour of the 802A depends on the location of the Mode switch (1). When in the Setup position the bath plate will not rise at the end of a movement and the manual up/down switch located on the side of the 802A will be active. In the Run position the 802A will make complete movements consisting of lowering the plate (if it is up) indexing to the new location and then raising the plate to the operating position. In Run mode the manual up/down switch on the 802A is not active.

The controller also features an Up/Down switch (2). The Up/Down switch changes the vertical location of the bath plate regardless of the Mode switch setting. If down the plate will move up, if up it will move down.

The accuracy of all movements is ensured by the use of a series of very accurate position sensors, one for each bath location plus one each for up and down control. By using position sensors at each location there are no cumulative positioning errors that occur because of lost steps when controlling the stepper motors that drive the bath plate. The position sensors change state from a logic HI to LO within 150 microns (0.006") of travel. This very short actuation distance coupled with the method that the microprocessor uses to determine position results in the 802A being able to relocate itself on any bath within ±25 microns (±0.001") of the correct position. To further aid the positioning accuracy an anti-backlash control is built into the micro code. When moving from a lower bath number to a higher

number the plate is indexed past the correct location and then moved back to the correct location. This removes any backlash present in the mechanical drive and increases accuracy.

The movement profile consists of high speed (2500 steps per second) and low speed (300 steps per second) sections. The high-speed section of the movement profile occurs from the current position to the location where the position sensor of the desired bath is tripped. Once the position sensor has been located the motion profile switches to low speed drive that steps the motor a small number of steps to the final position. The low speed section of the profile improves the overall accuracy and repeatability of the positioning. Both the high and low speed values can be modified through the menu section of the controller (see the section on menu controls). The distance traveled during the low speed portion of the movement profile can also be controlled through the menu section. These low speed movements are referred to as the bath offset values and are typically different for each bath location. The offset values compensate for tolerances on the machining of the bath plate and for sensor location tolerances. The offset values are set at the factory and shouldn't require adjustment.

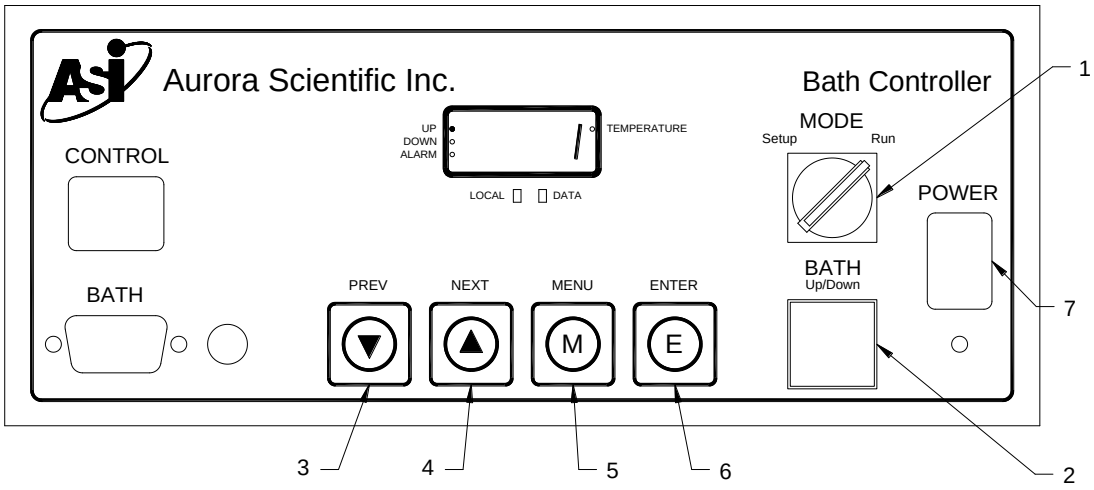

Figure 4 Diagram of Bath Controller Front Panel

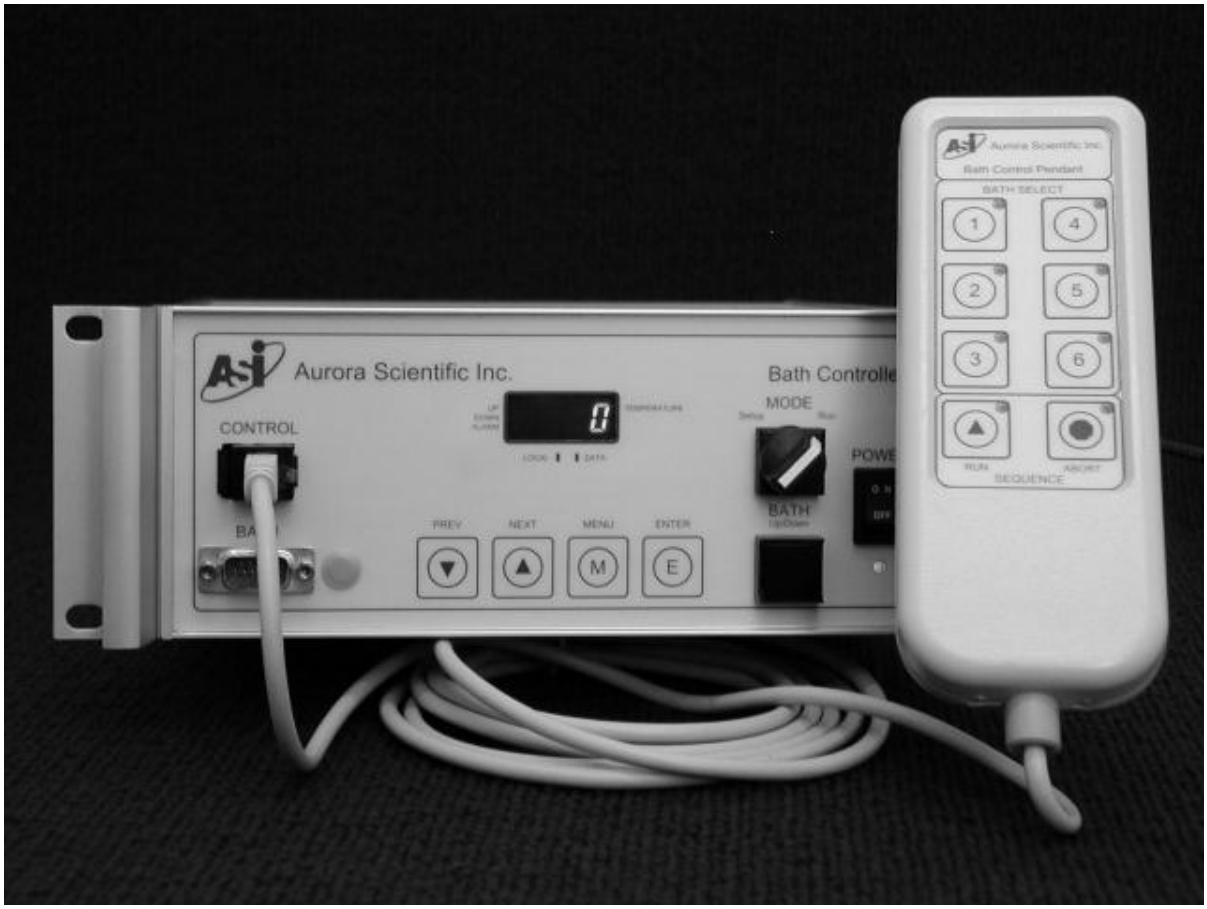

Figure 5 Bath Controller with Pendant

3.1 Operating Modes

The Mode switch (1 in Figure 4) has two positions, Setup and Run. Setup is used during initial equipment setup or anytime the researcher doesn't want the plate to automatically rise at the end of the motion cycle. In Run mode the controller does a complete movement including lowering the plate, indexing to the desired bath well and then raising the plate.

3.1.1 Setup Mode

In Setup Mode the plate does not move up after an index. The setup manual Up/Down button, located on the side of the 802A near the water inlet/outlet, is active and will move the platform up at a slow rate as long as the button is depressed. As soon as the switch is released the bath lowers. The switch only functions when the plate is at a bath location. If the manual Up/Down switch is held long enough for the plate to reach the top then the plate will remain at the top. If the button is held long enough for the plate to reach the upper position sensor then the Z-axis offset will be performed.

The sequencer is disabled in Setup mode. If a sequence is started and the Mode switch is changed from Run to Setup then the sequence will abort after completing the current movement.

3.1.2 Run Mode

In Run Mode the plate completes a full index cycle including raising the plate at the end of the movement. The manual Up/Down button does not work and Z-axis offset is automatically performed for both the up and down movements.

The sequencer operates in Run mode and can be programmed through the menu section and started using the Run button on the pendant control.

3.2 Menu Section

The menu section allows the user to program sequences and access and modify control parameters. The top level of the menu system is indicated by the front panel display showing the supervisor prompt (SUP). The menu section contains three menus, sequencer (SEU), speed (SPd) and offset (OS). Each of these is described in detail in sections 3.2.3 through 3.2.5.

3.2.1 Accessing the Menu

In order to prevent unwanted alteration of the controller's setup parameters the Menu key (M, 5) is normally locked. In order to unlock the menu section the Menu key must be held while turning the power on. Hold the Menu key and turn on the power, when SUP is displayed on the front panel display you can release the Menu key and you will be in the menu section. Once the menu has been unlocked the user can enter and exit the menu system as often as necessary. Simply press the Menu key to enter the menu section. When the power is cycled, or the controller resets, the menu key will be locked again.

3.2.2 Menu Navigation

The top level of the menu section is indicated by the front panel display showing SUP (supervisor). Press the Menu key (M, 5) to enter the menu system. Once SUP is displayed press the Enter key (E, 6). The display will now change to SEU. Pressing the enter key again will enter the sequencer menu and allow a sequence to be programmed or modified. If instead the Menu key is pressed again then the display will change to the speed menu (SPd). The Enter key will now cause the speed setup section to be entered. If the Menu key is pressed again then the display will change to the offset menu (OS). As with the previous menus the Enter key allows the menu to be accessed and the Menu key cycles between menus. To exit the menu sections press the Next/up arrow key (4), once the display shows SUP press the Menu key to exit the menu section. Figure 6 is a diagram of the top-level menu navigation with the key presses indicated in parentheses.

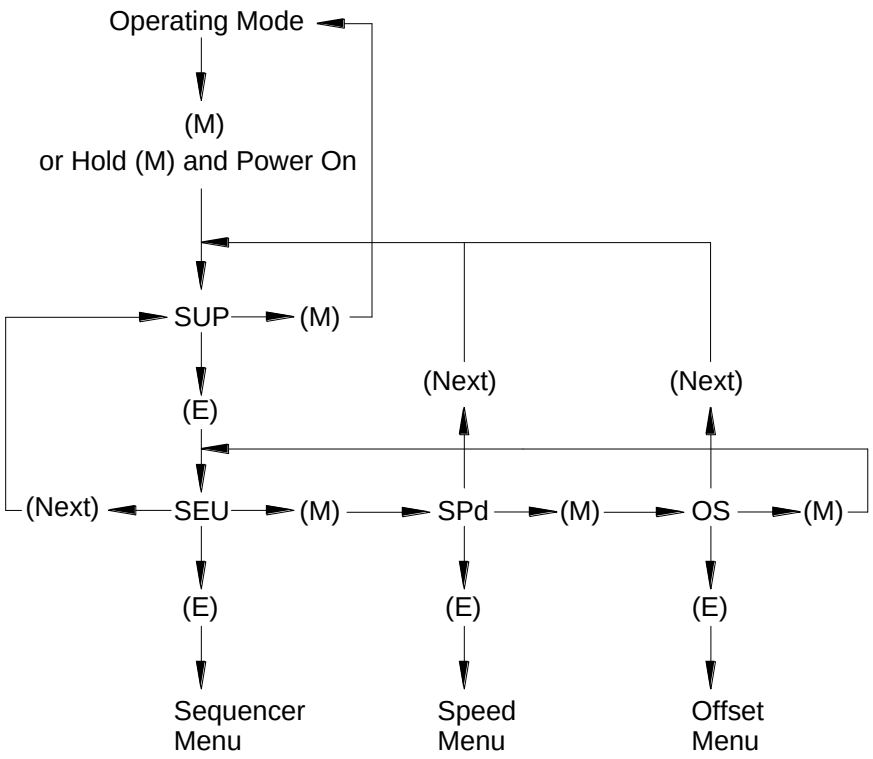

Figure 6 Top-Level Menu Navigation

3.2.3 Sequencer Menu

The sequencer menu (SEU) can be used to program up to 6 steps with a delay time specified for each step. A loop parameter is also provided that allows the sequence to run between 1 and 255 times. The loop parameter can also be set to run continuously by setting the loop value to 0.

The sequencer menu operates like all other menus in that the Menu key cycles between menu items (SS1, dEL1, SS2, dEL2, … LP, SS1, …). The Enter key accesses the menu item and then stores values. Values can vary from a single digit (for example the bath number ranges from 0 to 6) to up to four digits (for example the delay time in seconds can range from 0 to 9999 seconds). The first press of the Enter key will show the current value of the selected menu item, for example assume that you are on menu item dEL2 then pressing the Enter key will show 30 on the display (assuming that you had previously programmed this value to 30 seconds). Once 30 is shown on the display then the PREV/down arrow and the NEXT/up arrow can be used to change the units' digit. The NEXT key increases the value and the PREV key decreases it. A brief press of the Enter key cycles between the 1's, 10's, 100's and 1000's digits. The digit that will be modified flashes while it is selected. By pressing and holding the Enter key for about 1 second the value shown on the display will be written to memory and then pressing the Menu key will return the display to the current menu item display. For example, assume that you want to program 102 seconds into the delay for sequence step 4 (dEL4). Proceed as shown in Table 1. In the table under Display an underline indicates a flashing digit.

Table 1 Example of Programming 102 seconds into dEL4

Step	Key Press	Display
1		dEL4
2	E	0
3	E	0000
4	NEXT	0001
5	NEXT	0002
6	E	0002
7	E	0002
8	NEXT	0102
9	E (hold)	102
10	M	dEL4

Example of Programming a Complete Sequence

Assume that the experimental protocol requires starting in bath 1 then indexing to bath 2, staying there for 30 seconds and then indexing to bath 3 where the sequence would end. The protocol also calls for this sequence to only execute once. This simple sequence would be programmed as follows.

SS1 = 1
dEL1 = 0
SS2 = 2
dEL2 = 30
SS3 = 3
dEL3 = 0
SS4 = 0
It is not necessary to program values into SS5, 6 and dEL4, 5 and 6. Since SS4 = 0 the sequence stops at this point and doesn't look at the rest of the entries except for the Loop value.
LP = 1

Once the sequence is programmed into the controller the menu system must be exited before the sequence can be run. In addition the bath system must be located at the bath that corresponds to the value in SS1 (in our above case this is bath 1). The only way to start a sequence is by using the Run key in the sequence section of the pendant control. Sequences can be aborted at any time by pressing the Abort key. If the indexer is in the middle of a move then the move will finish before the sequence is aborted.

Figure 7 is a diagram that shows the navigation through the sequence menu. In the figure key presses are indicated in parentheses.

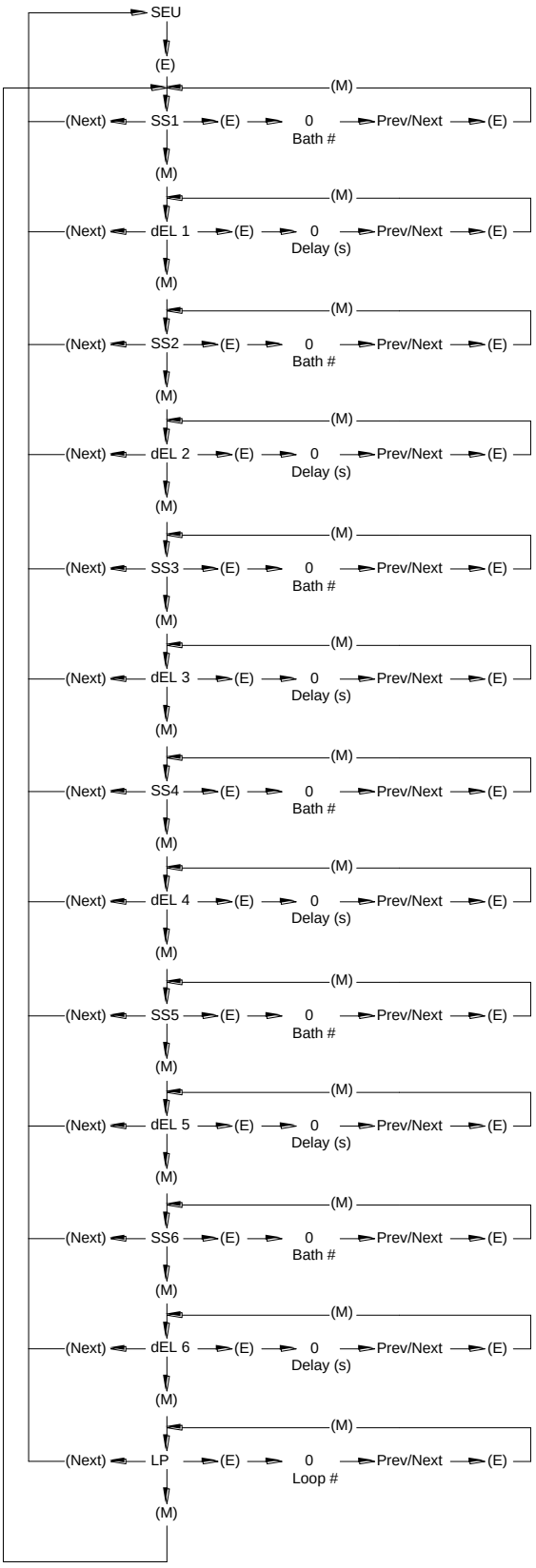

Figure 7 Sequence Menu Navigation

Table 2 presents a list of all items in the sequence menu along with the range of values for all variables.

Table 2 Sequence Menu

Item	Value	Description
SEU	None	Sequence Menu
SS1	0-6	Step 1, Value=0 indicates no sequence or end of sequence
dEL1	0-9999	Delay 1, units are seconds
SS2	0-6	Step 2
dEL2	0-9999	Delay 2, units are seconds
SS3	0-6	Step 3
dEL3	0-9999	Delay 3, units are seconds
SS4	0-6	Step 4
dEL4	0-9999	Delay 4, units are seconds
SS5	0-6	Step 5
dEL5	0-9999	Delay 5, units are seconds
SS6	0-6	Step 6
dEL6	0-9999	Delay 6, units are seconds
LP	0-255	Loop, Value=0 for infinite repeats, otherwise indicates number of cycles

3.2.4 Speed Menu

The speed menu (SPd) is used to set the various operating speeds for the controller. These parameters have been factory set and it is not recommended that the user change the factory values. The menu consists of control parameters for the high and low speed motion in both the X and Z directions and the speed for the manual up/down button when in Setup mode. There are also two period parameters that control a timeout that triggers error conditions.

Figure 8 is a diagram that shows the navigation through the speed menu. In the figure key presses are indicated in parentheses.

Table 3 presents a list of all items in the speed menu along with the range of values for all variables.

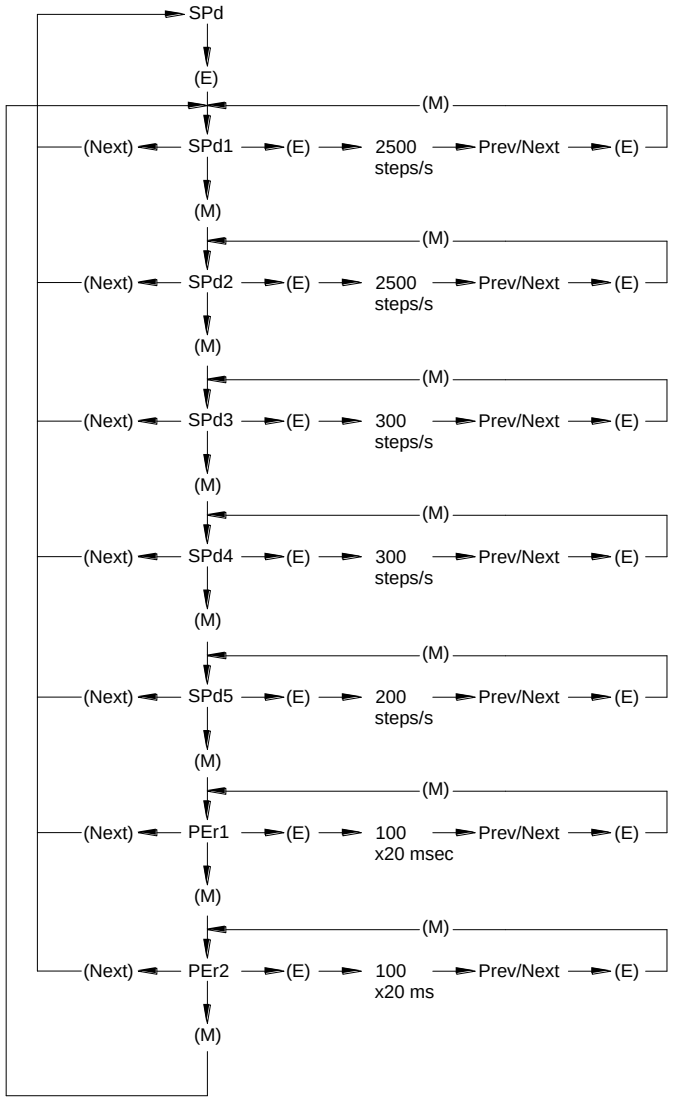

Figure 8 Speed Menu Navigation

Table 3 Speed Menu

Item	Value	Description
SPd	None	Speed Menu
SPd1	1000-3000	High Speed X-axis, Default Value=2500, units steps/sec
SPd2	1000-3000	High Speed Z-axis, Default Value=2500, units steps/sec
SPd3	100-1000	Low Speed X-axis, Default Value=300, units steps/sec
SPd4	100-1000	Low Speed X-axis, Default Value=300, units steps/sec
SPd5	100-500	Low Speed Setup manual up, Default Value=150, units steps/sec
PEr1	0-255	Period of X-axis index from bath to bath, Default Value=150, units x20 msec
PEr2	0-255	Period of Z-axis index in up/down direction, Default Value=150, units x20 msec

3.2.5 Offset Menu

The offset menu (OS) is used to apply a position offset at the end of each move. This offset is used to compensate for small dimensional errors in the machining and sensor location. A complete motion cycle of the 802A consists of a high-speed movement until the appropriate sensor is located followed by a low-speed final indexing that adjusts the final position of the plate. The distance moved during the low speed portion of the movement is controlled by the offset value associated with the particular sensor.

The offset parameters have been factory set and it is not recommended that the user change the factory values. However if the position sensor flag is moved, if the 802A is disassembled for maintenance, or through normal wear, it may be necessary to adjust the offset values. If it is noticed that the plate is off of the correct position one or more baths then the offset values for those baths should be adjusted. Note: if all baths are out of position by the same amount then the motor and force transducer should be moved relative to the bath plate. Don't use the offset parameters to compensate for incorrect location of the force transducer and lever system.

The menu consists of eight offset parameters one for each bath location and one each for up and down. Each offset step is equivalent to a linear movement of 25 microns (0.001"). The offset parameters can be checked by first positioning the bath plate at bath 1. Set the controller mode to Setup. Position the force transducer (using the XYZ translation stages) such that the muscle attachment tube/wire is located exactly in the center of the bath 1 slot. Index the bath plate to each of the other baths and use the manual up/down switch to raise the bath plate. Note if the force transducer attachment wire appears to be in the center of each slot. This checking must be done using a microscope. The slots are 700 microns (0.028") wide and typically the attachment tube/wire is about 350 microns (0.014") diameter. This leaves about 175 microns (0.007") clearance on either side of the attachment tube/wire.

If attachment tube is out of position for some of the baths then adjust the offset parameter for those particular baths. Note: if all of baths 2 through 6 seem to be offset by the same amount then it is better to simply change the offset of bath 1, readjust the XYZ stage and test the position of the other baths. One method to check the new offset value is to index to an adjacent bath and then back again. Alternatively press the button for the current bath position and if the bath plate is down then the position will be re-indexed.

The offset can only be applied in one direction. For the X-axis, increasing the offset moves the bath plate towards the bath 1 end. This has the effect of moving the attachment tube/wire the opposite direction (toward bath 6). Therefore if the attachment tube is too close to the bath 1 side of the slot, increase the offset. If the attachment tube is too close to the bath 6 side of the slot, decrease the offset. For the Z-axis, increasing the offset for the up sensor moves the plate farther up. Increasing the offset for the down sensor moves the plate farther down.

If it turns out that one of the baths requires a negative offset then there are two solutions. The first would be to add more offset to all of the other baths and then reposition the force transducer and lever system using the XYZ stages. The other solution is to readjust the position of the sensor flag. This is a very involved process that requires disassembly of a large part of the 802A and if it is determined that this is required then we recommend the unit be returned to the factory for repair.

Avoid using an offset of 0 steps. Values between 5 and 20 seem to provide the most positioning accuracy. Also avoid values greater than 30 since large offset values increase the indexing time.

Figure 9 is a diagram that shows the navigation through the offset menu. In the figure key presses are indicated in parentheses.

Table 4 presents a list of all items in the offset menu along with the range of values for all variables.

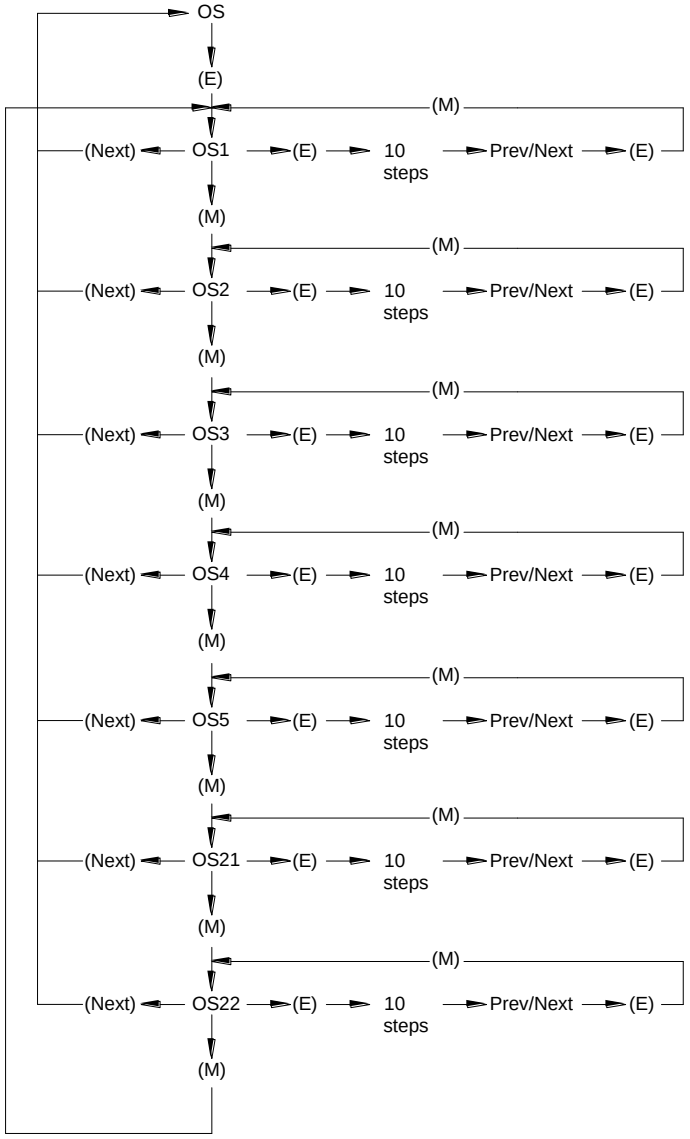

Figure 9 Offset Menu Navigation

Table 4 Offset Menu

Item	Value	Description
OS	None	Offset Menu
OS1	0-255	Offset for bath 1, Default value=10, units steps
OS2	0-255	Offset for bath 2, Default value=10, units steps
OS3	0-255	Offset for bath 3, Default value=10, units steps
OS4	0-255	Offset for bath 4, Default value=10, units steps
OS5	0-255	Offset for bath 5, Default value=10, units steps
OS6	0-255	Offset for bath 6, Default value=10, units steps
OS21	0-255	Offset for the up sensor, Default value=10, units steps
OS22	0-255	Offset for the down sensor, Default value=10, units steps

3.3 Error Codes

The controller monitors five possible errors. These include: X-direction movement, Z-direction movement, X-direction offset, Z-direction offset and sequence errors. All errors will sound the buzzer and will flash the ALARM indicator in the front panel display.

Table 5 Error Codes

Error Code	Display	Description
ERR1	Bath digit flashes	X-direction index error, correct bath sensor was not detected before PEr1 interval expired.
ERR2	Up or Down indicator flashes	Z-direction index error, Up or Down sensor was not detected before PEr2 interval expired.
ERR3	ERR3	X offset problem.
ERR4	ERR4	Z offset problem.
ERR5	ERR5	Sequencer problem.

4.0 Using the 802A

4.1 Adjusting the Location of the Force Transducer

The force transducer location can be adjusted using the XYZ translation stages. It is strongly recommended that the position of the force transducer be adjusted with the aid of a microscope prior to setting the controller in Run mode and raising the bath plate. Raising the bath plate without first adjusting the force transducer position can result in breakage of the force transducer or damage to the bath plate.

4.2 Adjusting the Location of the Lever Arm

The lever system motor and lever arm location must be adjusted prior to moving the bath plate. Use a combination of the XYZ translation stage, the angular position of the motor mount clamp ring, the position of motor in the clamp ring and the position of the arm on the motor shaft to align the lever arm with the bath. Raising the bath plate or indexing it without first adjusting the lever arm position can result in damage to the lever arm, motor or bath plate.

4.3 Attaching a Muscle Fiber to the 802A

The 802A was designed with the intent that the fiber would be attached in bath 1. For this reason bath 1 is wider than the rest of the baths and also contains right angle prisms that allow the side of the fiber to be viewed.

Researchers have various methods for attaching muscle fiber to a lever arm and force transducer. Most methods involve attachment of a short length of fine gauge hypodermic tubing to the force transducer and the lever arm. The fiber is then attached to the tubing by tying, clips, glue or a similar technique.

The 802A was designed for the lever arm to be in the bath along with the fiber. It may be advantageous to first coat the portion of the lever arm that will be submerged with a thin coating of epoxy. This seals the aluminum arm and minimizes corrosion and the possibility of contaminating the bath.

The 802A design calls for the force transducer to be outside of the bath with the fiber mount tubing entering the bath chamber through a narrow vertical slot (700 microns (0.028”) wide). In most cases the surface tension of the bath liquid will be high enough to easily retain the liquid in the bath. Under no circumstances should you attempt to have the glass tube that exits the force transducer enter the slot. It will not fit and the transducer will be broken.

4.4 Adjusting the Resting Tension or Sarcomere Length

Once the fiber is attached the resting tension or initial sarcomere length can be adjusted by several methods. The Y-axis translation stages for either the force transducer or the motor mount can be used to adjust the fiber length or resting tension. The lever system can also be used to adjust the fiber length/force. Use the front panel Length Offset control to change the position of the lever arm.

4.5 Measuring the Fiber Length and Cross-section

Fiber length and cross-section can be measured using an optical microscope equipped with a measuring reticule. The right angle prisms located in bath 1 provide a convenient method of obtaining a cross-section measurement in the vertical plane.

4.6 Point-to-Point Movement

In normal operation the Mode switch should be set to Run this causes the controller to automatically raise the bath at the end of the movement. Pressing the bath button of the desired bath on the pendant control will cause point-to-point movement. The bath plate will automatically lower move to the commanded bath location and rise. The controls on the front panel can also achieve this same movement by using the PREV/down arrow or NEXT/up arrow controls to choose the desired bath and then pressing ENTER to cause the action to occur. For a complete motion

4.7 Sequencer Controlled Movement

For sequences of movement that involve up to six different movements with varying delay times at each movement location the sequencer should be programmed. Details of sequencer programming can be found in chapter 3.

To initiate a sequence ensure that the current location corresponds to the location of the first step in the sequence. Press the Run button located in the Sequence Control section of the pendant control and the sequence will begin. A green light on the Run button indicates that a sequence is running.

To stop a sequence press the Abort button on the pendant control. A red light on the Abort button indicates an aborted sequence or a sequence that has not started.