INSTRUCTION MANUAL

Models 801B

Small Intact Fiber Test Apparatus

February 20, 2008, Revision 2

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Table of Contents

Table of Contents1
List of Figures
1.0 Introduction
1.1 Specifications
2.0 Apparatus Setup7
2.1 Unpacking7
2.2 Attaching the Motor
2.3 Attaching the Force Transducer
2.4 Attaching the Cooling Water Lines
2.5 Thermoelectric Controller
2.6 Attaching the Perfusion Inlet and Suction Outlet Lines10
2.7 Attaching the 801B to an Inverted Microscope
3.0 Using the 801B11
3.1 Adjusting the Location of the Force Transducer
3.2 Adjusting the Location of the Lever Arm
3.3 Controlling the Flow of Perfusate through the Bath
3.4 Attaching a Muscle Fiber to the 801B
3.5 Adjusting the Resting Tension or Sarcomere Length
3.6 Adjusting the Stimulation Electrodes
3.6 Adjusting the Thermocouple Position
4.0 Warranty
Appendix A Fiber Attachment Methods14
Appendix B Motor and Force Transducer Attachments16
Radnoti Instruction Sheet

List of Figures

Figure 1 801B Apparatus with 825A Thermometer/TEC Controller.	3
Figure 2 825A TEC Controller	4
Figure 3 801B Apparatus mounted on Storage Stand	5
Figure 4 Close-up of 801B showing 322C Length Controller and Force Transducer	8

1.0 Introduction

The 801B small intact fiber test apparatus was designed to enable physiology researchers to easily test small intact muscle tissue with an ASI model 312C, 315C or 322C high-speed length controller and an ASI series 400A force transducer.

The 801B consists of an aluminum bath plate with a single perfusion well with a glass bottom, 2 TEC heater/coolers, water-cooled heat sink plate, model 825A TEC controller, micrometer drive XYZ translation stages for motor and force transducer positioning, and mounts for the high-speed length controller and the 400A series force transducer. The system also comes with a 1.0 liter Radnoti temperature jacketed reservoir for the perfusate. Also included are mounting screws for mounting the force transducer and a set of Imperial Allen keys. All parts are manufactured from corrosion resistant materials (stainless steel, anodized aluminum and Delrin).

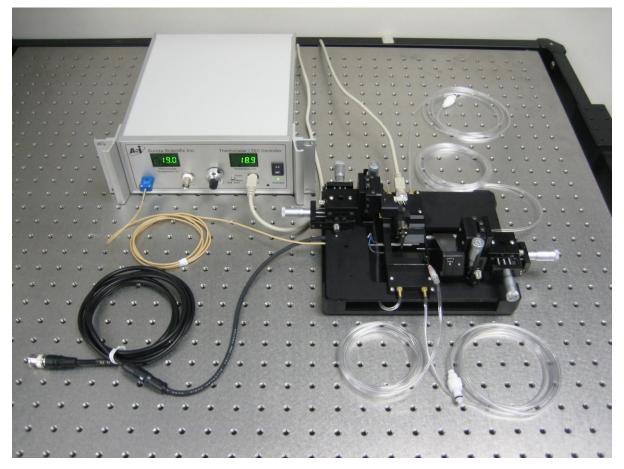


Figure 1 801B Apparatus with 825A Thermometer/TEC Controller.

The bath plate is manufactured from aluminum that is coated with Magnaplate HCR 31. This is a hard anodized coating followed by polymer impregnation and polymer coating. The resultant surface has a Rockwell hardness of about 50 with excellent dielectric properties while maintaining high thermal conductivity. Thus the aluminum is completely sealed from

the tissue and the perfusate. Field stimulation can be performed within the bath with little or no current flowing to the chamber walls because the surface of the bath plate is a dielectric.

The bath plate is temperature controlled using Peltier coolers mounted on either end of the plate. The Peltier coolers are controlled by an 825A Thermometer/TEC Controller. The 825A is capable of controlling the temperature of the bath plate from 4 to 38°C with 0.1°C accuracy. A T-type thermocouple is provided with the 801B apparatus for measuring the liquid temperature in the bath. The thermocouple is plugged into the 825A which provides a digital readout of the bath temperature and an output voltage proportional to the bath temperature.

The liquid enters the 801B chamber through an 18AWG hypodermic needle. The needle is a close fit in the bath plate and thus the temperature of the incoming liquid is brought to the operating temperature of the plate as it flows through the inlet needle. It is important that the liquid entering the bath is close to the operating temperature of the plate to ensure the most effective temperature control of the liquid (see next paragraph for a discussion of temperature control of the liquid reservoir). The liquid is removed through an 18AWG hypodermic needle located at the force transducer end of the bath. The researcher needs to provide suction to remove the liquid. Please see chapter 3.0 for more information about temperature control and liquid flow through the bath.

The perfusion liquid stored in the Radnoti reservoir is maintained at the appropriate temperature by use of a laboratory water circulator that is supplied by the researcher. The water jacketed glass reservoir provides for liquid storage, oxygenation, and temperature control.



Figure 2 825A TEC 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.

An AD590 temperature sensor is mounted to the bath plate and provides temperature feedback to the model 825A TEC controller. Further details of the controller can be found in the 825A Instruction Manual.

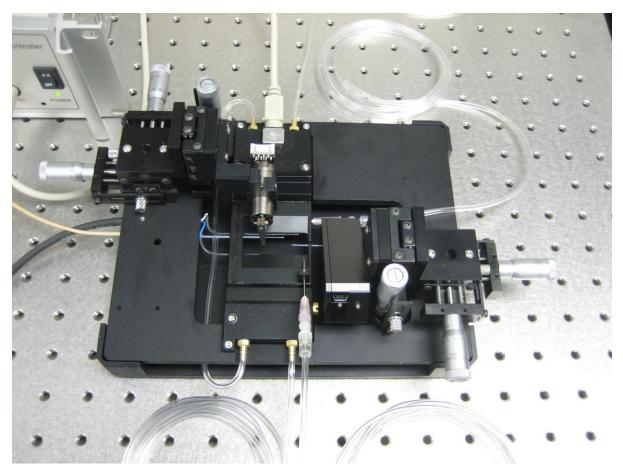


Figure 3 801B Apparatus mounted on Storage Stand

1.1 Specifications

Bath Plate No. of Baths: Bath Plate Material: Bath Dimensions: Bath Volume:	1 6061-T6 aluminum with Magnaplate HCR 31 coating 40mm (1.575") L x 15mm (0.590") W x 3mm (0.118") D 1.8ml
<u>Inlet</u> Vinyl Tube Size: Stainless Tube Size:	1/16" ID x 1/8" OD 18AWG
<u>Outlet</u> Vinyl Tube Size: Stainless Tube Size: Suction Height Range:	1/16" ID x 1/8" OD 18AWG 3.0-6.0mm (measured from bottom of bath)
<u>Thermoelectric Coolers</u> No. of TECs: Power: Voltage: Temperature Sensor:	2 33 W 15 V AD590
<u>Thermocouple</u> Type: Model:	T Physitemp model MT-23/5
<u>Stimulation Electrodes</u> Number: Material: Dimensions:	2 mounted through end wall of bath plate 99.9% pure Platinum 36mm (1.417") L x 26AWG (0.018") Diameter
Equipment Included Reservoir: TEC Controller: Water Cooler:	Radnoti model: 120142-1, 1 litre volume ASI model 825A Thermometer/TEC Controller ASI model 826A Water Cooler

2.0 Apparatus Setup

2.1 Unpacking

Unpack the apparatus from the four shipping boxes. The first box contains the 801B apparatus with the XYZ translation stages attached. The second contains the 825A Thermometer/TEC Controller. The third box contains the 826A Water Cooler, mounting screws for the force transducer, water hoses, perfusate hoses, BNC cable for the stimulation electrodes and Allen keys. The fourth box contains the Radnoti Reservoir with bubbler and mounting clamp.

The 801B apparatus is shipped assembled however the user must attach the motor, force transducer, water lines, perfusate lines, TEC control cable and the stimulation BNC cable.

2.2 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 (to the 2 mark 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 4-40 socket head cap (SHC) screw. To mount a model 312C or 315C motor first remove the clamp screw completely from the clamp ring. Then line up the arm with the slot in the clamp and insert the motor into the motor clamp ring. Now rotate the motor so that the connector is at the top and then gently tighten the motor mount ring clamp screw to hold the motor in position. To mount a model 322C motor so that the connector is at the top and then gently tighten the 322C motor so that the connector is at the top and then ring clamp. Once the motor is clamped in position you can then attach the lever arm.

The motor can also slide within the mount. 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 the bath. The X and Y stages may also require adjustment to properly locate the motor.

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. Ensure that the Z axis stage is positioned so that the lever arm doesn't hit the glass cover slip at the bottom of the bath.

2.3 Attaching the Force Transducer

Normally a length of fine gauge stainless steel tubing is fastened into the end of the force transducer output tube; see Appendix A for details of various attachment methods. 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 into the slot and if the transducer is located too close to the plate the output tube could be broken when the transducer is moved.

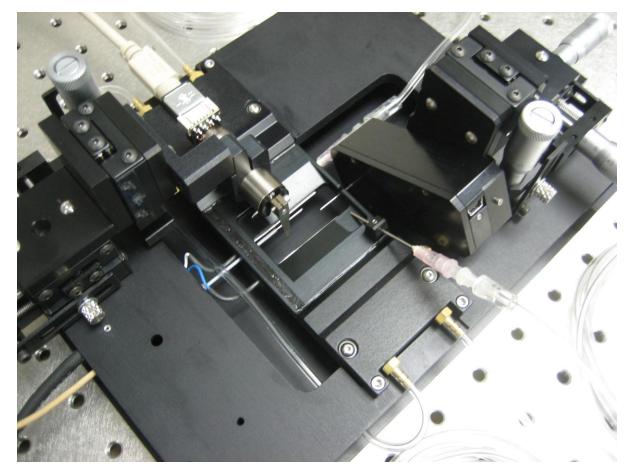


Figure 4 Close-up of 801B showing 322C Length Controller and Force Transducer

Refer to Figure 3 for correct orientation of the force transducer in the 801B apparatus. The transducer is tipped back at a 30-degree angle to the horizontal. This angle provides greater access to the bath. Before mounting the transducer, position the Y-axis translation stage as far from the bath plate 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 (2 on the dial). Remove the transducer mounting plate from the XYZ stage and then use the four 4-40x1/4" button head screws supplied to attach the force transducer to the mounting plate. Now reattach the transducer mounting plate, along with the transducer, to the Delrin mount plate attached to the Z stage using two of the 4-40x3/8" button head screws 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.

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. Use a microscope to observe the needle and slot positions relative to each other. 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 the transducer simply use the XYZ stages to reposition the transducer to the middle of the slot.

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 801B base plate. Cable ties and tape backed cable mounts are provided for this purpose. Cable movement can lead to increased noise on the length and force signals.

2.4 Attaching the Cooling Water Lines

Cooling water lines are attached to the 801B when it is shipped. These lines are terminated with a dry-break quick-connect tube fittings. The shipping box that contains the 826A Water Cooler has two 6 foot long 1/4"x3/8" clear hoses with a mating quick-connect to the 826A on one end and the mating quick connect fitting to match the connectors on the 801B on the other end. The quick-connect fittings include a valve inside them so they can be disconnected with water in the lines without any spillage. The connectors are disconnected by pressing the metal tab on the side of the connector and then pulling the connection apart.

Connect the hoses to the mating fittings on the back of the 826A Water Cooler and the other end to the mating fitting on the 1/8" tubes that lead to the 801B heat sinks. Please refer to the 826A operating manual for instructions on filling and using the water cooler.

Ensure that you have a flow of water through the cooling plates before turning on the 825A TEC Controller. Operating the thermoelectric coolers (TEC) without water flowing will damage the TECs.

2.5 Thermoelectric Controller

Two thermoelectric coolers (TECs) are built into the 801B. These devices are rated at 15 volts and 33 watts. A model 825A Thermometer/TEC Controller is also included with the shipment. A separate manual for the controller has been provided and therefore only a minimum of information will be provided here.

The system has been designed to control the temperature between 0 and 40°C with an accuracy of ± 0.1 °C. The 825A measures the bath plate temperature using an AD590 temperature sensor embedded into the bath plate. The sensor temperature can be viewed on the 825A by setting the display switch to Process. When the switch is in the Setpoint location the LED display shows the Setpoint. A Setpoint adjustment knob allows the Setpoint to be set between 0 and 40°C with an accuracy of 0.1°C. The Process temperature should match the Setpoint temperature within about 3 minutes. However the temperature of the liquid in the bath will take up to 7 minutes to stabilize after a large Setpoint change. Also there is normally a temperature offset between the AD590 temperature and the liquid temperature. This offset is greater the farther the Setpoint is from room temperature. The 825A includes a thermocouple meter and the apparatus includes a T-type stainless steel thermocouple probe that can be used to monitor the temperature of the liquid in the bath.

Ensure that the 826A Water Cooler is operating and that water is flowing through the cooling plates before changing the set point to a temperature other than room temperature. Set the desired temperature on the controller and then monitor the actual temperature to ensure that the temperature is approaching the set point. Adjust the set point as required to achieve the desired bath temperature.

2.6 Attaching the Perfusion Inlet and Suction Outlet Lines

Note the two 18AWG hypodermic needles attached to the apparatus. The needle that exits at the motor end of the bath is the supply. The needle mounted at a 30° angle to the horizontal and located next to the force transducer is the suction. Two 6 foot long 1/8" diameter vinyl tubes are provided for the perfusion liquid inlet and the suction outlet. Both tubes are terminated with a Luer fitting that slides into the hypodermic needle.

Connect the inlet hose to the outlet of the Radnoti reservoir using the stopcock supplied with the reservoir. The suction needle should be connected to a suction source to drain liquid from the bath. The suction needle is mounted on a 30° angle and includes a set screw on the top of needle holder. To adjust the level of the liquid in the bath loosen the set screw, slide the needle up or down in the holder and re-fasten the set screw. A 0.89mm Allen key that fits the set screw has been supplied with the 801B.

The temperature of the reservoir can be controlled using a standard temperature controlled laboratory water circulator. This is not supplied with the 801B. The reservoir includes a clamp ring that mounts to standard retort stand clamps. An oxygenating bubbler is also included to allow the perfusion liquid to have gas bubbled through it. Several fittings and valves are also supplied with the reservoir. Attach the valve supplied to the outlet of the reservoir and then attach the 1/16° tubing to the other side of the valve.

2.7 Attaching the 801B to an Inverted Microscope

The 801B has been designed to minimize the distance between the objective and the muscle tissue. Part of this is accomplished by having the force transducer housing protrude below the level of the bottom of the 801B mounting plate. For this reason care must be taken when mounting the 801B to a microscope stage. Remove any inserts present in the microscope stage. Remove the 801B from its stand; simply lift the apparatus off of its stand. Locate the 801B on the microscope stage with the force transducer protruding downwards into the hole in the stage. Try to position the 801B so that the location of the fiber is in the centre of the stage cutout. The base plate of the 801B has four rubber feet mounted to it. These help to prevent the 801B from sliding when placed on the microscope stage. It is normally not necessary to clamp the 801B to the stage as it doesn't tend to move when in operation. However, if desired, the base plate can be clamped to the stage. Normally the 801B will be used with an XY microscope platform. This allows the entire 801B to be moved with respect to the objective.

3.0 Using the 801B

3.1 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. Lowering the force transducer needle into the bath without first adjusting its position can result in breakage of the force transducer or damage to the bath plate.

3.2 Adjusting the Location of the Lever Arm

The lever system motor and lever arm location should be adjusted prior to mounting tissue. Use a combination of the XYZ translation stage, the angular position of the motor mount clamp ring and the position of motor in the clamp ring to align the lever arm with the bath.

3.3 Controlling the Flow of Perfusate through the Bath

Liquid can be supplied to the bath by a simple gravity feed. Mount the exit of the Radnoti reservoir a minimum of 14" above the bath level to ensure sufficient pressure to drive the liquid into the bath. Use the stopcock supplied with the reservoir to turn on and off and control the flow of liquid.

The suction needle can be connected to a laboratory suction source or to a peristaltic pump if suction is not available. Please note that the pulsing of a peristaltic pump will affect the force readings so it is best to turn off the pump during mechanical testing of the tissue.

For best results we recommend filling the bath with fresh liquid from the reservoir then stopping the flow before performing the mechanical tests on the tissue. We make this suggestion because any flow through the bath will introduce noise in the force readings. A simple method of determining the extent of this noise is to record force data with the liquid flowing and with it stopped. Comparison of the signals will provide a measure of the increased noise due to the flow.

In most cases it is fine to stop the flow during testing since the pH, oxygen content and other characteristics of the perfusion liquid will remain relatively constant for 5 to 10 minutes. Since most testing takes less than this length of time it is reasonable to introduce fresh solution, stop the flow, do the mechanical testing and then introduce more fresh solution before continuing with testing. Having said this it is the responsibility of the researcher to ensure that this method of stopping the flow will work in their circumstances.

If a continuous flow is required then it is important that the flowrate be sufficient to prevent surging in the inlet to the bath. Due to the coating used on the bath plate the liquid contact angle is quite high and this can result in a flow regime where a large pool of liquid grows at the inlet needle until it is large enough to drain into the bath and then another large pool forms. This results in a surging type flow which adds far more noise to the force transducer readings. Raising the flow rate by a small amount will usually prevent this surging from happening. Our tests indicate a flowrate of about 0.25ml/sec (14ml/minute) is sufficient to prevent surging. If the stopped flow method is used then the surging is not an issue as no readings are taken while the flow is turned on.

3.4 Attaching a Muscle Fiber to the 801B

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. Refer to Appendix A for more details of attaching fibers.

The 801B was designed for the lever arm to be in the bath along with the fiber. The 312C and 315C lever arms are made from titanium to minimize corrosion and the possibility of contaminating the bath. The 322C lever arm is made from aluminum that is coated with the same material as the bath plate. This electrically insulates the arm while sealing the aluminum from contacting the liquid in the bath. If desired, the lever arm may be coated with a thin layer of epoxy if it is felt that the arm requires additional sealing from the solutions in the bath.

The 801B 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 is attached to the force transducer enter the slot. It will not fit and the transducer will be broken.

3.5 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. To use the lever system to set resting tension simply turn the front panel Length Offset control to change the position of the lever arm. The digital panel meter on the front of the 312C, 315C or 322C controller can be used to monitor the amount of movement. For the 312C and 315C a 1 micron movement corresponds to a voltage change of 0.007 volts. For the 322C a 1 micron movement corresponds to a voltage change of 0.003 volts.

3.6 Adjusting the Stimulation Electrodes

The position of the stimulation electrodes may need to be adjusted for optimum tissue stimulation. The electrodes are made from 26AWG Platinum wire and can easily be bent to a new position. In most cases it is advantageous to locate the electrodes within a millimeter of the tissue.

If you elect to bend the electrodes then ensure that neither of the electrodes contact the metal portion of the thermocouple probe. The thermocouple probe has a Teflon jacket on most of its length so it is important that if the electrode has to cross over the probe then the cross-over occurs where the Teflon is present. Likewise the electrodes should not touch the lever arm or muscle tissue attachments.

3.6 Adjusting the Thermocouple Position

The position of the thermocouple can be controlled by simply sliding the thermocouple in or out of the bath. The thermocouple is located in the tip of the needle so the tip should be placed close to the centre of the tissue.

4.0 Warranty

The 801B Small Intact Fiber Apparatus is warranted to be free of defects in materials and workmanship for three years from the date of shipment. Aurora Scientific Inc. will repair or replace, at our option, any part of the system that upon our examination is found to be defective while under warranty. Obligations under this warranty are limited to repair or replacement of the instrument. Aurora Scientific Inc. shall not be liable for any other damages of any kind, including consequential damages, personal injury, or the like. Disassembly of the unit will void this warranty. Damage to the system through misuse will void this warranty. Aurora Scientific Inc. pursues a policy of continual product development and improvement therefore we reserve the right to change published specifications without prior notice.

Appendix A Fiber Attachment Methods

Three main attachment methods exist. These are:

- 1) Tying
- 2) T-clips
- 3) Glue

1. Tying

Tying requires adding a short length of fine gauge hypodermic needle tubing to the force transducer and length controller. The fiber is then tied to these two needles using fine suture (10-0 monofilament nylon suture). See the attached photo of a small intact muscle tied to a force transducer and lever. Neither the force transducer output tube nor the lever arm are visible in this photo but the researcher was using a model 400A force transducer and a model 318B length controller. In the photo the wire at the bottom is a platinum stimulation electrode and the needle at the top is a thermocouple. The force generated by the fiber will determine the number of suture loops to use. This photo shows four on each end which was required to prevent slippage.

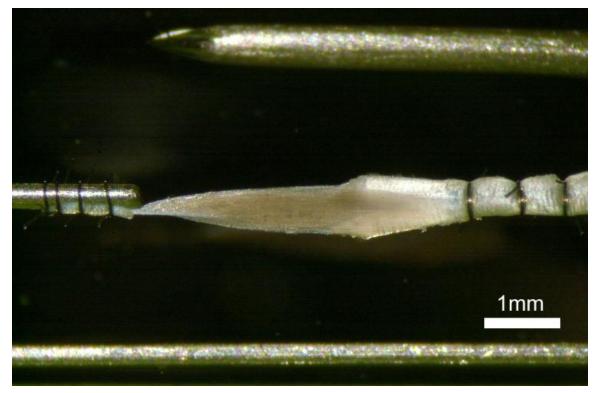


Photo 1 Lumbrical Muscle Tied to 29 AWG Tubes

With permeabilized fibers, or even fiber bundles it is more likely that you would only require 2 or 3 sutures per end. Our force transducer has a hollow glass tube that the load is attached to. Stainless steel tubing, or a 29AWG hypodermic needle, can be attached to the transducer by sliding the needle 5-10mm down inside the glass tube and then applying a small amount of

melted paraffin wax to the joint between the needle and tube. The overall length of the tube will be dependent on the bath arrangement you are using but I would think the tube would project out of the force transducer tube about 8-10mm, overall length 13 to 20mm. The wax will "wick" up the glass tube and, when hard, it will easily be able to withstand the forces generated by fibers. Using wax has the added advantage that all you need to do is warm up the wax in order to remove the needle should it require changing. If you are expecting very high forces 10g or more then you may need to epoxy the tube into the glass tube of the transducer as wax may start to slip at these higher forces.

To attach the needle to the lever arm we suggest bending the needle into an "L" shape and then using epoxy to glue the needle to the arm. It is easiest to first "tack" the needle to the arm using superglue and then apply the epoxy. We suggest a 2 hour epoxy as the 5 minute kind often breaks down in water. The vertical part of the L should lie parallel to the length of the arm with the bottom of the L facing perpendicular to the long axis of the arm. The bottom part of the L should be kept as short as practical and the least amount of glue that will do the job should be used. This is because the added mass of the needle and the arm will affect the tuning of the lever system. The L should be about 5mm x 4 mm long. 29AWG stainless tubing works well for this task and if you can't find any then you can cut up a hypodermic needle. Please note that the ends of the tubing should be carefully tapered to prevent metal burrs from cutting into the fiber. A short, abrupt taper seems to work best as the fiber is less likely to slip. Two research groups using the tying method include Prof. John Faulkner's group at the University of Michigan and Prof. Rick Moss's group at the University of Wisconsin-Madison.

2. T-Clips

T-clips are small clips shaped in a "T" shape and cut from aluminum foil. The bottom of the T has a small hole in it. The top of the T is wrapped around the fiber and squashed down onto the fiber. This leaves the vertical part of the T sticking out the end of the fiber. A small hook is created in a similar manner to the tube used in the tying method but this time the tube is bent to form a hook. In operation the hole in the T clip is slipped over the hook and the fiber is then attached. T clips seem to work fine for low force applications, single permeabilized fibers. If you intend to test higher force fibers, for example frog fibers or bundles or strips you may find that the tissue slips inside the T clip. Pieter de Tombe's group in Chicago and Frank Brozovich, at the Mayo, use T clips.

3. Glue

Depending on the size of the tissue preparation I have seen researchers also attach tissue with glue. Typically this seems to be used with very small preps including skinned myocytes. In this method a small amount of glue is placed on the tip of a tube attached to the force transducer and another to the lever arm. The tissue is then brought into contact with the glue and attached. Two main types of glue seem to be used, the first is expanding foam called Great Stuff that is available in the USA. The other is silicone glue. Pieter de Tombe from Chicago and G. Stienen from the VU University Medical Centre in Amsterdam are two researchers using glue.

Appendix B Motor and Force Transducer Attachments

For the motor (312C, 315C or 322C) start with a piece of 29 gauge stainless steel tubing that's about 10mm long and then "dress up" the ends by inserting the tubing into a "pin vise" and using a very fine sharpening stone. Aim for a very smooth end with a fairly abrupt taper (long taper encourages slipping of prep). Next put a 90degree bend in the tubing by chucking it in the pin vise 1/2 way down its length and then, starting with the pin vise at a very shallow angle with respect to a flat surface, slowly raise the pin vise while keeping the half of the tubing that extends from the vise in full-length-contact with the flat surface. Continue to raise the vise until it is normal to the flat surface (at which time the tubing ought to have a 90deg bend). Too much bend and the tubing will "crimp" and you will have to start over again. Once you have an L-shaped piece of tubing that you are happy with (the legs of the "L" will be on the order of 4.5mm each), you will epoxy it in such a way that one leg is in line with the long axis of the motor arm and the other leg comes off the end of the motor arm at a 90deg angle. Use 2 hour epoxy (5min isn't typically waterproof). First "tack" the "L" to the arm using a tiny amount of superglue - this is only to hold the "L" in place while the epoxy is curing. The 312C motor is very sensitive to added mass. You may need to make the "L" about 3mm x 3mm when used with a 312C. Also use the minimum amount of epoxy that will get the job done. Even the added mass of the epoxy can have a detrimental effect on motor tuning.

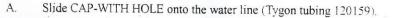
For the transducer, the length of tubing will depend in part on bath configuration. As a rule of thumb use the shortest tube that will do the job (less mass, higher frequency response). Reasonable dimensions are approx. 13mm, 5mm of which are inside the glass capillary "coupling tube" with the remaining 8 outside. Dress the ends of the tubing as described above and then epoxy one end to the coupling tube. Don't try to get it concentric with the coupling tube, this can be very difficult. Instead let its outside surface rest against the inner surface of the coupling tube, this ensures that the stainless steel tubing and the glass output tube are lined up. Getting epoxy in the coupling tube is a bit of a trick. Dip the end of the stainless steel tubing in the epoxy pool and then insert it. It takes very little glue to hold it. You will want to keep an eye on it until the epoxy is cured, making constant minor adjustments to maintain a straight stainless steel tube extension.

Use 29 gauge stainless steel tubing (0.013 inch outside diameter) that is readily available from McMaster-Carr or you can make the attachments from a 29 gauge hypodermic needle. The pin vise and sharpening stones are also available from McMaster-Carr.

Hypodermic Needle Tubing:	McMaster-Carr Catalogue #8988K429
Pin Vise:	McMaster-Carr Catalogue #8455A14
Sharpening stones:	McMaster-Carr Catalogue #4456A17



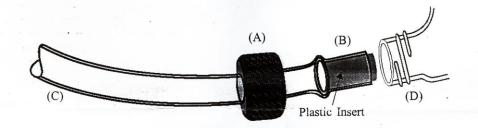
RADNOTI "QUICK DISCONNECT" INSTRUCTIONS



B. Push the Plastic Sleeve Insert into the end of the Tygon tubing with the flared end first. (Catalog No. 120160).

C. Be sure to use 5/16 x 3/16 inch Tygon tubing (Catalog No.120159).

D. <u>Moisten tip of Tygon tubing</u> before insertion into the threaded glass. CAUTION: To Avoid Breakage, Do Not Over-Tighten The Screw Cap.



Replacement	Part List:	
Catalog No.	Description	

120160	Plastic Sleeve Insert, 25/Pkg
120159	Tygon Tubing 5/16 x 3/16 ID, 50 Ft/Min.
160196	Drilled caps for Water Jacketed QD Connections 12/Pkg

RADNOTI GLASS TECHNOLOGY, INC. 227 W. Maple Ave, Monrovia CA 91016 USA Tel: 1-626-357-8827 (1-800-428-1416) FAX 1-626-303-2998 www.radnoti.com