Covaris®

# ME220 Focused-ultrasonicator
# Setup & Instruction Manual

## Instrument Developed for High-performance
## Sample Preparation using Adaptive Focused Acoustic® (AFA®)

For Research Only

Model Number: ME220 | Software: SonoLab 8

# UNIVERSAL PRECAUTIONS

Universal Precautions should be followed on all specimen samples, regardless of whether a sample is known to contain an infectious agent. Laboratories handling specimen samples are advised to comply with applicable parts of the following government and clinical standards, or their equivalent in the country of use:

- Centers for Disease Control (CDC), Universal Precautions for Prevention of Transmission of HIV and Other Bloodborne Infections, published 1987, updated 1996
- Clinical and Laboratory Standards Institute (CLSI), GP17-A2 Clinical Laboratory Safety; Approved Guideline - Second Edition, published 2004, ISBN 1-56238-530-5
- Clinical and Laboratory Standards Institute (CLSI), M29-A3 Protection of Laboratory Workers from Occupationally Acquired Infections; Approved Guideline, Third Edition, published 2005, ISBN 1-56238-5674
- Occupational Safety and Health Administration (OSHA), 29 CFR 1910.1030 Bloodborne Pathogens
- International Standards Organization (ISO) 15190:2003, Medical Laboratories – Requirements for Safety

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Teflon is a registered trademark of The DuPont Company.

Covaris, Inc.

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## Warnings

**For the Safety of Operating Personnel:**

- Make sure that the equipment is properly grounded.  *DO NOT* operate if it's not properly grounded.

- The unit is equipped with a power plug appropriate for the destination country.  *DO NOT*, under any circumstances, remove the grounding prong from the power cord.

- *DO NOT* run an acoustic treatment with the sample cover in the OPEN position or without a water bath - the acoustic system will not work.

- If there is any indication that the Safety System is not functioning properly, *DO NOT* operate the equipment and contact Covaris immediately.

- If the equipment is used in a manner not specified by the manufacturer, the protection provided by the equipment may be impaired.

**To Prevent Damage to the Equipment:**

- The instruments are designed to operate in ambient laboratory conditions e.g., 19 °C to 25 °C (66 °F to 77 °F).  *DO NOT* operate the instrument in a cold room environment; the system is designed to operate with a water bath and includes a solid state heater/chiller apparatus to control sample temperature.

- *NEVER* run a method without water in the acoustic assembly; this could cause damage to the transducer.  The instrument is equipped with a water level sensor to protect the transducer.  The system will not allow the acoustic wave treatment to start unless an adequate volume of water is detected.

- AFA-grade water should be used to fill the supply water bottle.

- Empty the water bath daily.  *DO NOT* leave water in the water bath for an extended time since there is no water filtration or water cleaning system with the apparatus.

- *DO NOT* employ isopropyl alcohol, ammonia-based or abrasive cleaners on the acoustic assembly.

- The Tube Holder Insert should be stored in a dry place when not in use.

- *DO NOT* load third party hardware, software, or parts onto the system without consulting with Covaris Technical Support.

## Warranty

When used in accordance with written instruction and under normal operating conditions, the Covaris instruments are guaranteed to be free of defects in MATERIAL and WORKMANSHIP for one (1) year from the date of original delivery by an authorized representative.  Any component proven defective during the stated period will be repaired free of charge or replaced at the sole discretion of Covaris, F.O.B., Woburn, Massachusetts, U.S.A. provided that the defective component is returned properly packaged with all transportation charges prepaid.  The customer is expected to perform basic diagnostics and component replacement via telephone support from Covaris personnel.  If Covaris personnel are required to perform on-site repair, all travel related costs are paid by the customer.  A limited warranty as specified may apply to certain components of the equipment.

### Warranty Exceptions

This warranty is void if failure of the software or hardware has resulted from accidents, abuse, improper maintenance or repair, or by misapplication from the customer.  It is also void if damage is caused by any unauthorized attachments or if modifications are made to the equipment.  Removing or tampering with the Safety Enclosure will void the warranty, and the customer will assume all liabilities.

This warranty is limited to the original purchaser and is not transferable.

The software will perform according to the accompanying written materials and the medium on which the software is delivered is free of defects in materials under normal use and service.  The warranty is void if damage has resulted from third party software not intended for use with the system.

The high-power focused transducer is designed to give maximal mechanical energy output in water.  Permanent damage to the transducer and electronic circuits could result if the transducer is operated without water.  Operation of the system without water in the water bath voids the warranty.

*CONTACT COVARIS, INC. SHOULD YOU HAVE QUESTIONS OR CONCERNS PERTAINING TO THE USE OF OUR EQUIPMENT.*

### Warranty Services

The purchased equipment is covered by a twelve (12) month warranty which includes all the necessary services and support necessary so that the customer can successfully operate their equipment.  Extended warranties are available at the end of the original 12 month warranty period.

Services included with the original purchase of the system are:

*Technical Support* – Ongoing assistance with the operation or application of the equipment and/or troubleshooting is provided via:

- Telephone
    - United States: Tel:  +1 781.932.3959 during the hours of 9:00 a.m. to 5:00 p.m., Monday through Friday, Eastern Standard Time (EST), Greenwich Mean Time (GMT-05:00)
    - Europe: Tel:  44 (0) 845 872 0100, during the hours of 9:00 a.m. to 5:00 p.m., Monday through Friday, Greenwich Mean Time
- E-mail queries to techsupport@covaris.com or applicationsupport@covaris.com

*ME220 Instrument Repair*  – Warranty repair of the ME220 is provided on site.  All labor and expense charges are included.  Failure due to accident, abuse, or improper operation is not covered.  An Operator's Manual is provided with the equipment.  This manual includes sections on the operating instructions, maintenance guidelines, and troubleshooting tips.

## Table of Contents

# 1. Introduction

## 1.1 Overview of the User Manual

This manual contains the installation, operation and service instructions for the Covaris ME220 Focused-ultrasonicator. It contains information essential to the proper use and care of this equipment. Should any unforeseen problems occur with the normal operation of the equipment, contact Covaris Technical Support immediately.

The following definitions apply in this manual:

| **NOTE:** Informational. | **WARNING:** Personal injury may occur. | **CAUTION:** Equipment damage may occur. |
|---|---|---|

There is also a PDF version of this manual, typically available on the computer desktop. Open the manual by double clicking the user manual icon.

## 1.2 The ME220 and the Covaris Process

The ME220 Focused-ultrasonicator is based on Covaris Adaptive Focused Acoustics (AFA) technology. This instrument is designed to provide researchers an integrated stand-alone tool to be used in laboratory environment. The instrument is ideal for those who work with DNA shearing and sample preparation.

The system is comprised of the **Treatment System** which delivers the acoustic energy to the sample, the **Safety System** which protects users from inadvertently contacting the high intensity acoustic energy, and the **Computer and Application Software** which provides users an interface to control the device.

**The Covaris Process** uses Adaptive Focused Acoustic energy to precisely control cavitation and acoustic streaming within the sample treatment vessel in a non-contact, isothermal fashion. Focused acoustic energy is generated and delivered to individual samples. The mechanical energy imparted on the sample results in a controlled series of compression and rarefaction events.

# 2. Installation and Setup

## 2.1 Unpacking the System

Follow the included instructions for removing the ME220 instrument and accessories from their shipping materials.

> **NOTE:** To remove the foam shipping insert from the acoustic assembly, the ME220 needs to be powered on with SonoLab software running.  See **Section 2.3**, Starting SonoLab for details.

## 2.2 Assembly

The following items should be found in the package of the Covaris ME220 instrument:

- Notebook Computer with AC Adapter
- Cables
  - 1 USB cable
  - 1 ME220 power cord
  - 1 Computer AC Adaptor power cord (Non US customer only)
- Accessories
  - Wrench to Adjust Leveling Legs
  - 2 Plastic water bottles, one containing 1L AFA-grade water
  - 2 Quick Disconnect caps and dip tubes
  - Drip tray
- ME220 main unit
- Sample Racks and Consumables, depending on order configuration

> **CAUTION:** The ME220 weighs about 55 pounds (25kg).  With the front of the instrument closest to your body, lift the ME220 by gripping from its base plate.  Lifting the unit by its plastic case or doors will damage the instrument.
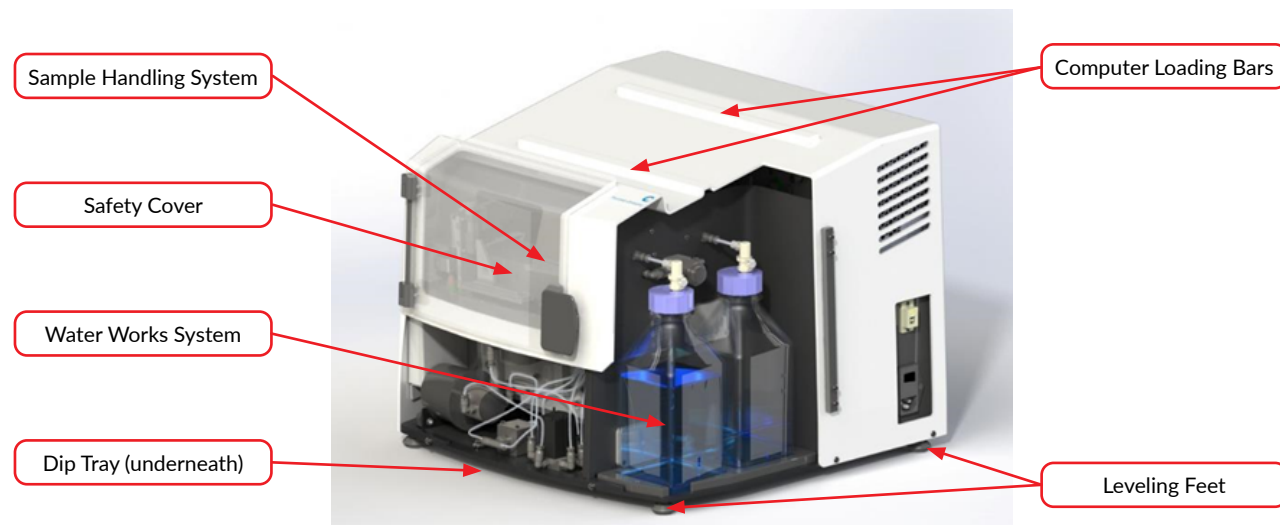


**Figure 2.1** A front view of the ME220 instrument

After the instrument is unpacked, position the ME220 on a flat surface near two power outlets.  Use both sides of the base (*Figure 2.1*) to lift the instrument.  Do not lift by the plastic case or doors.

1. Assemble the Water Works supply and waste bottles.

- Locate the two 1L plastic bottles, one empty, one filled with AFA-grade water.  Remove the shipping caps and replace with the Quick Disconnect Caps.  The cap with the dip tube goes on the supply bottle of AFA grade water.  The cap without the dip tube goes on the empty waste bottle.
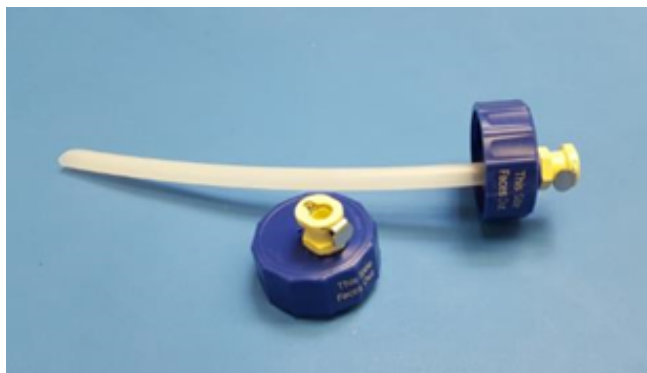


**Figure 2.2** Water Works Bottle Caps

- Open the clear plastic Water Works access door on the right side of the ME220.

- Place the empty Water Works waste bottle in the waste compartment of the Water Works bay, with the label on the cap facing out as shown in *Figure 2.3*.
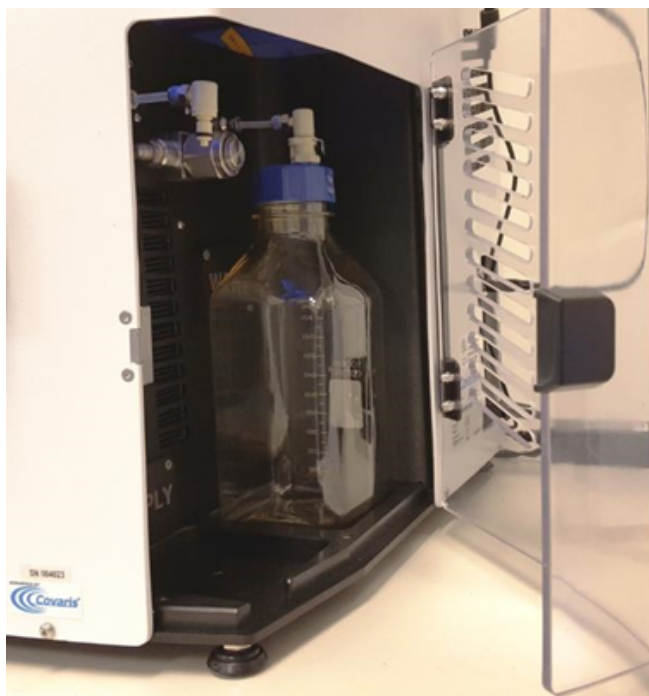


**Figure 2.3** Placing Bottles into Water Works Bay

- Press the waste Quick Disconnect Insert into the Quick Disconnect Cap until it clicks into place.  See *Figure 2.4*.

**Figure 2.4** Connecting Water Works Bottle to System

> **NOTE:** Label faces out. (See **Figure 2.4**).

- Repeat the above steps, placing the filled supply bottle of AFA-grade water into the supply compartment of the Water Works bay.

2. Slide the drip tray under the front of the ME220.

3. Place the computer onto the loading bars, as shown in **Figure 2.5**, aligning the Velcro strips on the computer with the mating strips on the loading bars.  Gently press the computer to secure it in place.



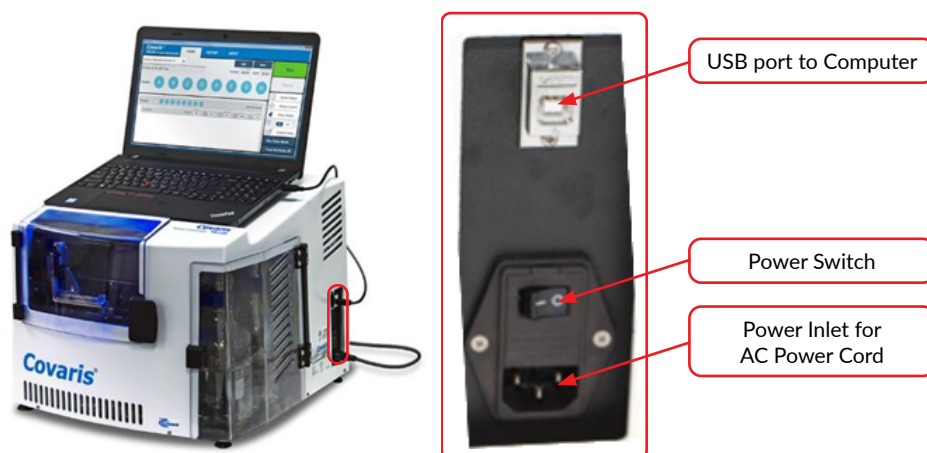**Figure 2.5** Secure the computer on top of the ME220

Figure 2.6 The power panel of the ME220 instrument

4. As shown in **Figure 2.6**, plug the AC power cord into the power inlet located toward the rear of the right side of the instrument. Plug the AC power cord into the power outlet.

5. Plug the AC adaptor into the computer and into the power outlet.

6. Connect the USB cable into the USB port located above the AC inlet of the instrument (**Figure 2.6**) and USB port on the notebook computer.

The entire instrument may be relocated in this assembled configuration by disconnecting power, lifting from the bottom, relocating, and repeating the appropriate assembly steps.

> **NOTE:** Any time the ME220 is moved to a new location, follow the instrument leveling procedure described in *Section 2.5*.

## 2.3 ME220 Software

SonoLab has been pre-installed in the computer provided with the ME220 instrument. No further installation or configuration is required.

SonoLab is the application used to control a Covaris AFA (Adaptive Focused Acoustics) instrument for processing a sample. It is designed to create, edit, and run a sample processing program called a method. It has provisions to run, pause or stop a method and view execution histories which contain a record of key instrument parameter values during a run.

If it is necessary to install or re-install SonoLab or instrument drivers on a computer, please contact Covaris for the latest versions, downloading, and installation instructions.

## 2.4 Starting SonoLab

*Important Note:* The ME220 is shipped with a twist-tie securing the sample weight to the sample tray and a foam shipping insert supporting the sample tray from below. If this is the first time the ME220 has been operated, open the front safety cover and remove the twist-tie securing the sample weight to the sample tray. Close the safety cover and follow the steps below to home the sample handling motion system. Only after the sample tray has stopped moving, remove the foam shipping insert from the water bath.

Turn on the computer. Turn on the ME220 power switch located at the rear, right side of the instrument. Wait at least 10 seconds to allow the instrument to initialize. Double click the SonoLab icon on the desktop to start SonoLab.

SonoLab will automatically connect to the instrument, and a window appears asking whether the instrument is to be homed. Click "Yes". The motion system will perform its initialization sequence, the homing window will close, and the SonoLab Home screen will be displayed, as illustrated in *Figure 2.7*.

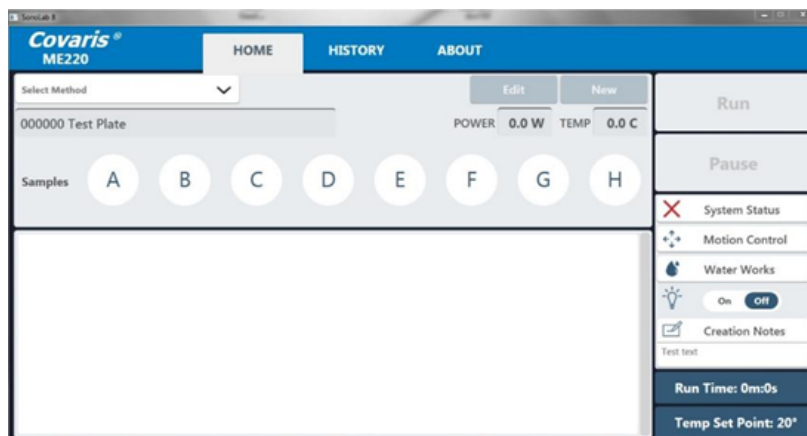> **NOTE:** If foam shipping insert is still in place, remove it at this time.



**Figure 2.7** The SonoLab Home Screen

> **NOTE:** Covaris recommends that the Laptop computer be employed exclusively for instrument control and that operation of additional application software be minimized.

## 2.5 Leveling the ME220 Instrument

This step is very important to ensure proper filling and draining of the water bath and proper instrument operation. Once the ME220 installed in the location in which it will be used:

- Open the Front plastic safety cover.
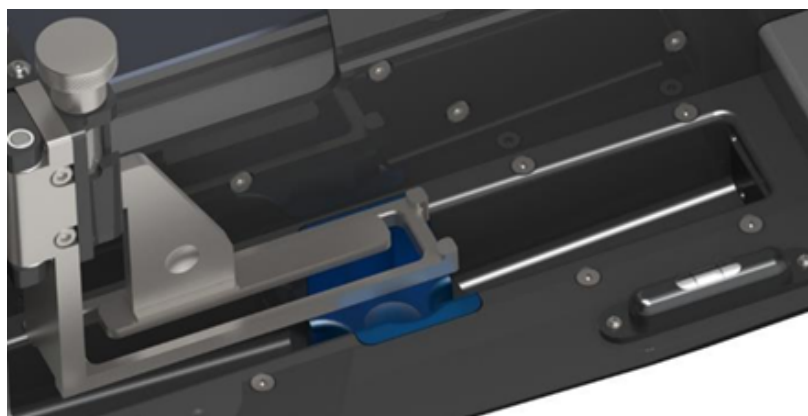- Locate the leveling fixture toward the right end of the water bath. See **Figure 2.8** below.



**Figure 2.8** Leveling Fixture

- Adjust only the feet on the right side.  If the bubble is in the center of the fixture sight glass, no adjustment is required.  If the bubble is to the right side of the center markings of sight glass, the right side is high, and the feet need to be shortened; if the bubble is to the left side, the right side is low, and the legs must be lengthened.  Adjust both front and rear feet an equal number of turns.

- Use the included wrench, if necessary, to loosen the locking nut on each right side leg as shown in **Figure 2.9**.  The lock nuts are shipped finger-tight only.
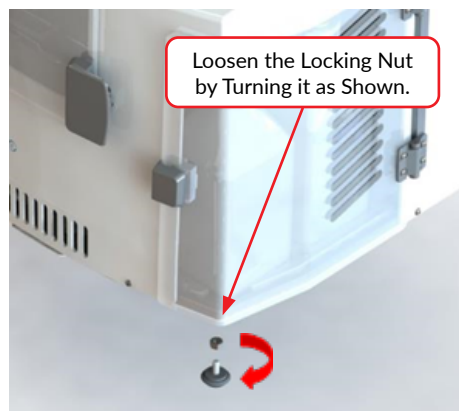


**Figure 2.9** ME220 Leg Locking Nut

- Lengthen or shorten both legs on the right side of the instrument an equal amount, as in **Figure 2.10**, until the bubble is centered in the leveling fixture sight glass.
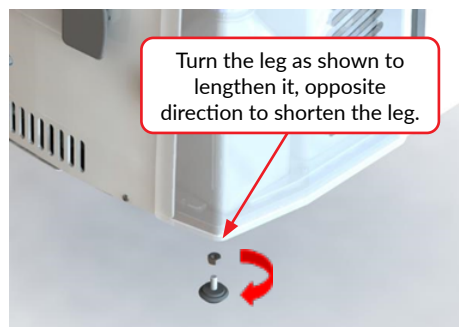


**Figure 2.10** ME220 Leg Height Adjustment

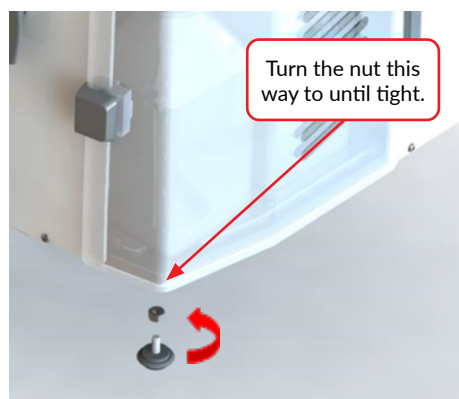- With the bubble centered, use the included wrench to tighten the nuts.  This locks the legs in position.



**Figure 2.11** Locking the ME220 Legs

# 3. Operation

## 3.1 General Overview



Sample Handling System

Electronics Cabinet

Computer Loading Bars

Safety Cover

Chiller Vents

Water Works Supply & Waste Bottles
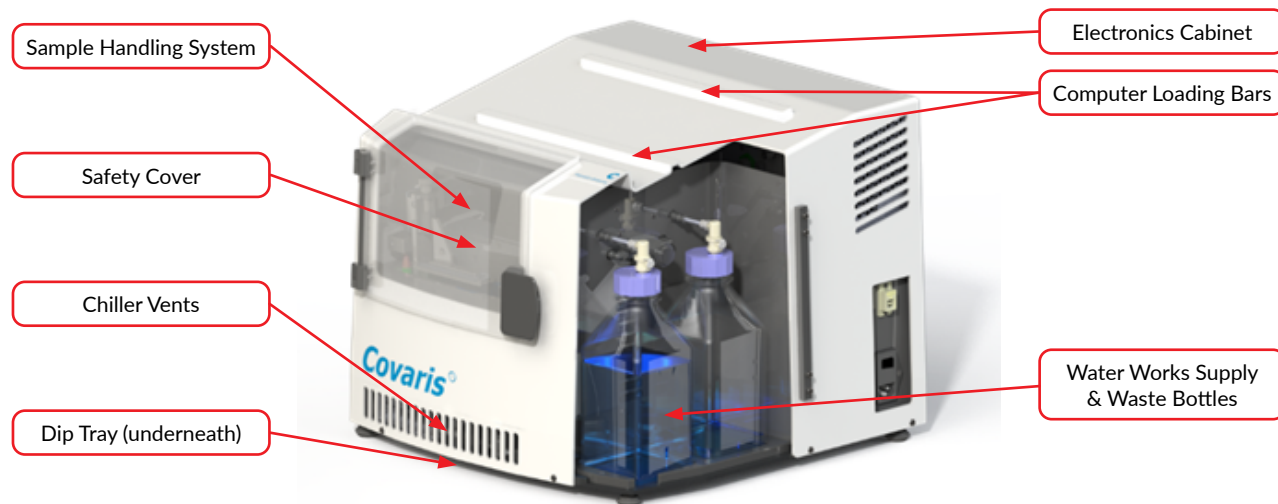
Dip Tray (underneath)

**Figure 3.1** Principal components of the ME220 instrument

The **Safety Cover** is used to protect users from any potential acoustic hazard during the treatment.  The acoustic treatment will not be started if the Safety Cover is open.

The **Acoustic Assembly**, located in the water bath below the sample tray, is used to transmit the acoustic energy from transducer to the sample through the coupling media (AFA-grade water).  The assembly incorporates an Acoustic Transducer which converts electrical signals into ultrasonic acoustic signals.  The assembly also incorporates a Sensor Module with water level and temperature sensors to ensure that these are appropriate during acoustic treatments.

The 2-axis **Motion System** is designed to automatically align the sample tube at the appropriate position and height in the acoustic field for processing.  Tubes are loaded into appropriate Racks, which are then placed on the ME220 sample tray.

The **Computer** provides an interface and software to the operator for controlling the ME220 instrument through SonoLab.

The **Electronics Cabinet** provides the hardware for controlling the acoustic transducer, the temperature, and sample position during processing.

The **Enclosure Light** is normally lit when the instrument is turned on.  The user can disable the light from the SonoLab Home screen.

The **Water Works** consists of a pump, valves, supply and waste bottles, and interconnecting plumbing.  These components are controlled automatically by SonoLab software to fill, drain, and maintain the correct water level in the water bath.  A drip tray under the instrument ensures protection should any water drip from the bath.

The **Thermal Module** controls the water bath temperature.  It is able to chill or heat the acoustic assembly, maintaining the water bath at or near the set point programmed in the method.  No external chiller is required.

> **WARNING:** During treatments, objects immersed in the focal zone above the transducer will be exposed to high acoustic intensity when the system is activated.  Always close the Safety Cover prior to starting a method.  Do not tamper with the door-closed sensor system.

## 3.2 Preparing the ME220 for Use

To run an acoustic treatment, a number of conditions must be met, including:

a) The ME220 motion system must be initialized

b) There must be an acoustic method selected in the Method box

c) The proper rack and consumables must be in place on the ME220 sample tray.  See S*ection 3.5*, **Loading the Samples**

d) The correct waveguide for the specified consumables must be in place in the water bath.  See **Section 3.4**, **Acoustic Waveguide**

e) Water Temperature must be within acceptable min and max limits

f) Water must be at the level required by the consumables specified in the selected method

g) The safety cover must be closed

The Run button will remain gray and inactive until all necessary conditions except water level and temperature are met.  The ME220 automatically corrects these when "Run" is selected.

To achieve an acceptable status on these conditions:

a) The ME220 motion system normally initializes when SonoLab starts, unless the user purposely skipped this step, in which case the motion system can be homed from the Motion Control pane.

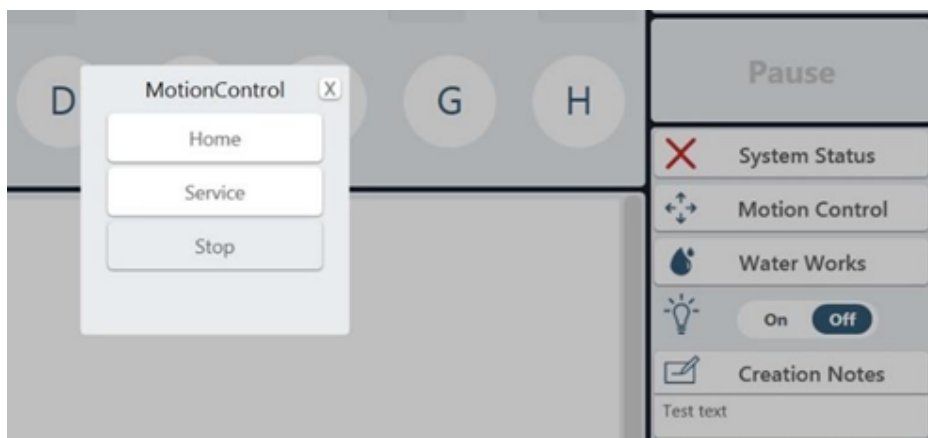Click on the **Motion Control** button to display the Motion Control pane.



**Figure 3.2** Motion Control Pane

Click on the **Motion Control** button to display the Motion Control pane.

b) To select and load an acoustic method, click the "Select Method" pull down menu, as shown in **Figure 3.3**, and a list of existing method names will be displayed.  Navigate through the list with the vertical scroll bar and select a method.

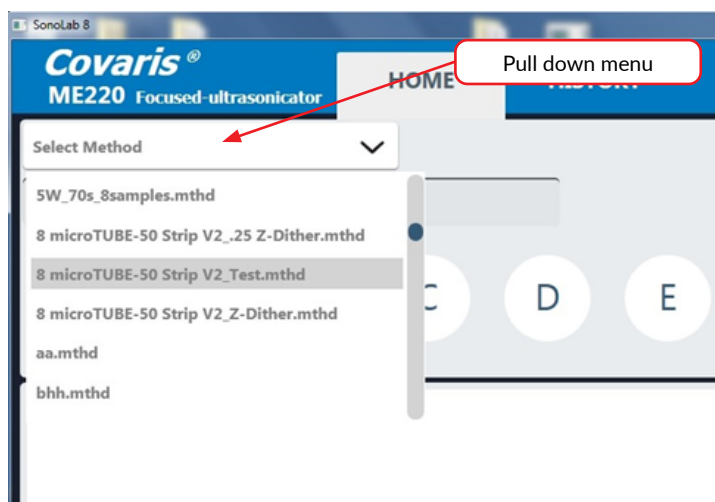**NOTE:** Several acoustic methods will come pre-installed into your system.



**Figure 3.3** Selecting and loading an acoustic method

The method's run time (minutes and seconds) and temperature set point will be displayed in the lower right corner of the main screen.
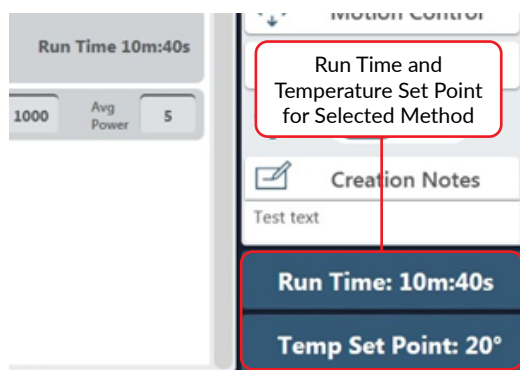


**Figure 3.4** Method run time and set point

c) The ME220 system automatically adjusts the water temperature to match the method temperature set point after Run is selected.  Water Temperature status will turn green when temperature is within the Min and Max limits (typically 2 degrees above or below the set point).  The default set point (in the absence of a selected method) is 20 °C.

d) The ME220 Water Works automatically fills the bath to the appropriate level for the tubes specified in the method, when the Run button is clicked to start a method.  (See **Section 3.3**)

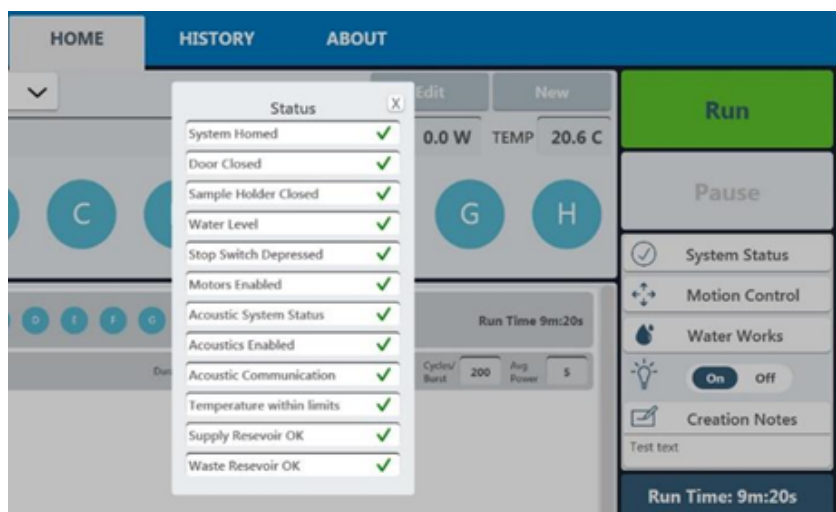e) The Door Closed status will turn green as soon as the Safety Cover is completely closed.

**Figure 3.5** Instrument Status Panel: All OK

The remaining conditions, along with their related status, are displayed in the Instrument Status panel (see **Figure 3.5**). Click the "System Status" button to display the Instrument Status panel. An acceptable condition is indicated by a green check mark (✔) in the Status box. If a condition is not acceptable, the status will show (X).

If any of the items in the Status list display (X), a red X will also be displayed in the System Status button, as shown in **Figure 3.6**.
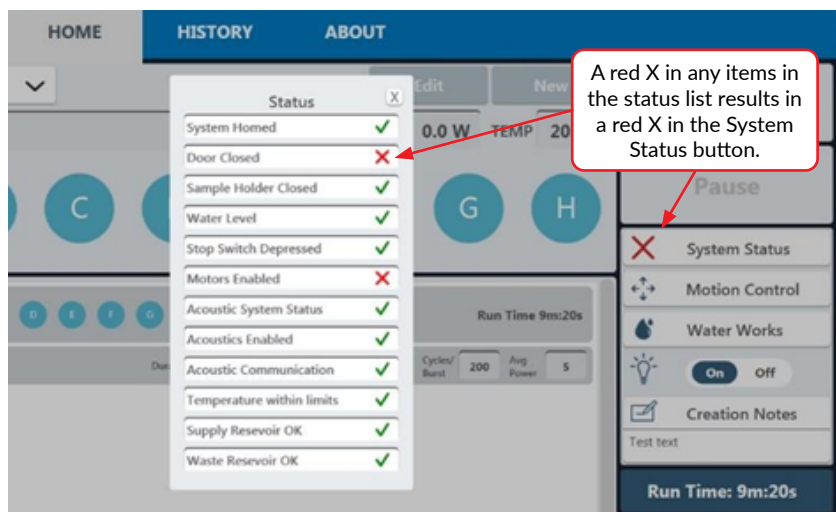


**Figure 3.6** Instrument Status Panel: Door Opened Resulting in Motors Disabled

Once a method has been loaded and all conditions except temperature and water level are acceptable, the Run button will turn green and the selected acoustic Method can be started.

To modify an existing method, see **Create and Edit Methods** in **Section 5**.

## 3.3 ME220 Water Works

To transmit acoustic energy to the sample vessel, the bath surrounding the transducer must be filled with deionized water.  Covaris recommends AFA-grade Water.  You can also use highly purified water (at least ASTM Type III or ISO grade 3).

The ME220 **Water Works** automatically maintains the correct water level in the bath for the sample vessels specified by a method.  This system either adds water from the Water Works supply bottle to raise the level of the bath, or removes water from the bath to the Water Works waste bottle.

Sensors detect if the supply bottle is empty or the waste bottle is full, and these conditions are reported in the Instrument Status panel. To service the bottles, open the access cover on the right of the ME220.  Press the metal thumb latch on the Quick Disconnect Cap, as shown in **Figure 3.7**, to release the Quick Disconnect Insert of the bottle being serviced and lift out the bottle.  Empty the waste bottle or fill the supply bottle, as appropriate, and reverse steps to reinstall the bottle.  The Supply bottle is toward the front of the ME220, the Waste bottle toward the back.  Observe the label on the cap specifying "This Side Faces Out".  This orients the bottle correctly so that the Quick Disconnect Insert and Cap are properly aligned.  See **Figure 3.7** below.
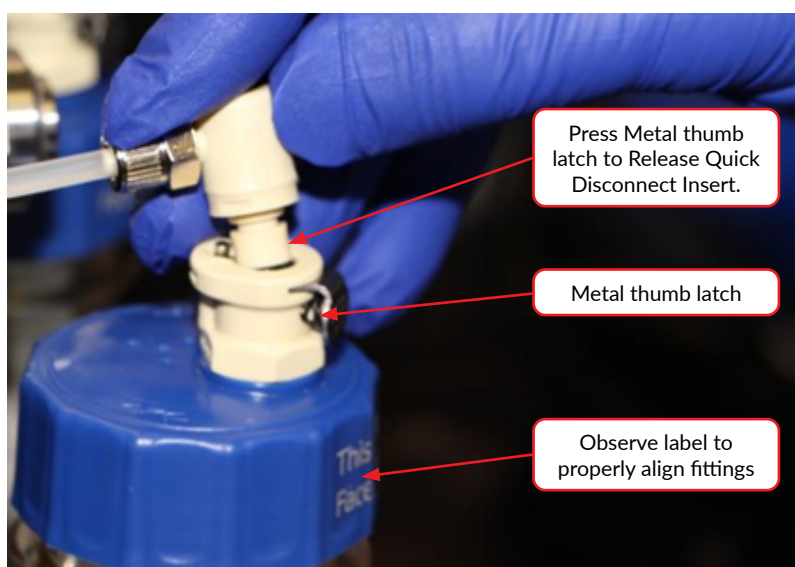


Press Metal thumb latch to Release Quick Disconnect Insert.

Metal thumb latch

Observe label to properly align fittings

**Figure 3.7** Releasing hoses via quick disconnect fittings

## 3.4 Acoustic Waveguide

The **Acoustic Waveguide** focuses the acoustic energy around the sample vessel for the most effective treatment.

Sample racks are color-coded to match the appropriate waveguide.  Always match the waveguide and rack to ensure proper operation and to prevent mechanical interference between the waveguide and rack.

To change waveguides, with the motion system in the "Home" position, rotate the sample tray forward (clockwise).  Then, remove the installed waveguide by lifting it from the water bath.  Install the designated waveguide by lowering it into the bath as shown in **Figure 3.8**.
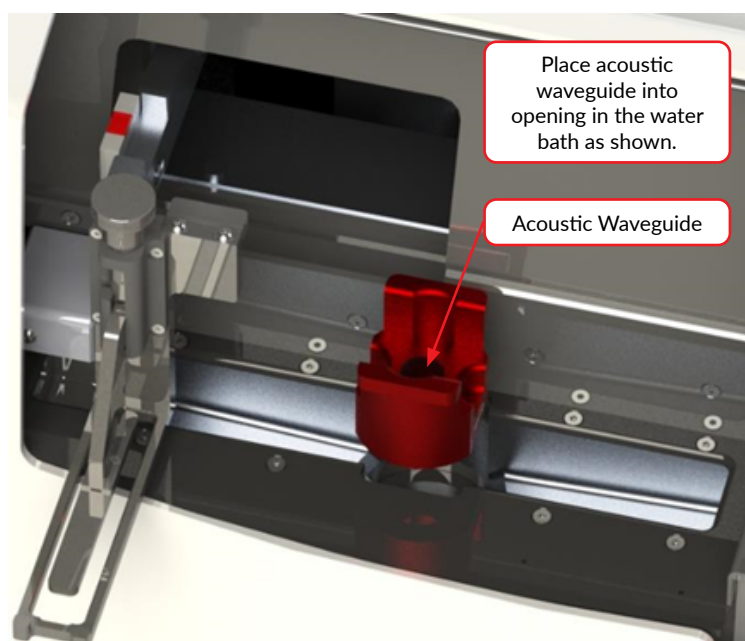
**Place acoustic waveguide into opening in the water bath as shown.**

**Acoustic Waveguide**

**Figure 3.8** Installing the Waveguide

## 3.5 Loading the Samples

A variety of tubes and their associated racks are available for use with the ME220.  In some cases, individual tubes are simply placed into the rack as shown.  The racks may be nested in the ME220 Rack Loading Station to facilitate the loading process.



**Figure 3.9** microTUBE Screw-caps Loaded in Rack

The tubes are then held in place by the ME220 sample retaining weight, as shown in **Figure 3.12** below.

The 8 microTUBE strip is fitted to its rack as a unit.  Open the hinged rack and insert the 8 microTUBE strip.  The strip is keyed to fit the rack in the correct orientation.
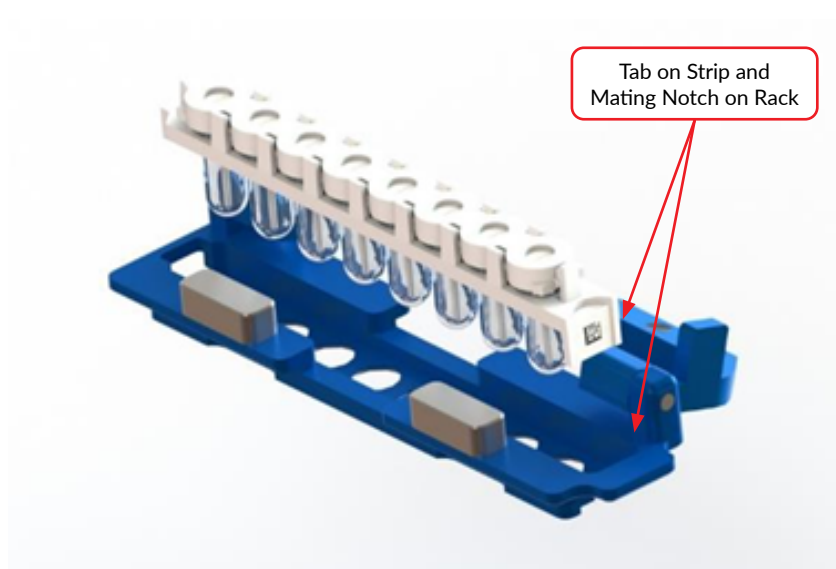
**Figure 3.10** Assembling the 8 microTUBE strip into Rack

Close the hinged rack cover to secure the strip.  Magnets in the cover hold the cover in the closed position.



**Figure 3.11** 8 microTUBE strip assembled in Rack

**NOTE:** Sample racks are color-coded to match the appropriate waveguide. Always match the waveguide and rack to ensure proper operation and to prevent mechanical interference between the waveguide and rack.

Open the ME220 safety cover.  Lift the sample weight and rotate it clockwise (forward) to the loading position, as shown in *Figure 3.12*.
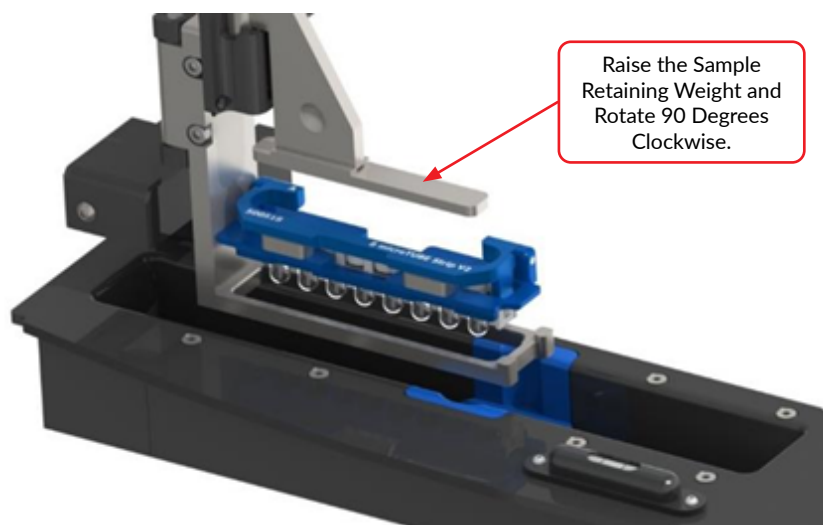
**Figure 3.12** Lifted Sample Tube Retaining Weight for Loading

Place tubes and rack assembly into the ME220 sample tray shown in **Figure 3.12**. Rotate the weight 90° counter-clockwise and lower it onto the tubes as shown in **Figure 3.13** below.

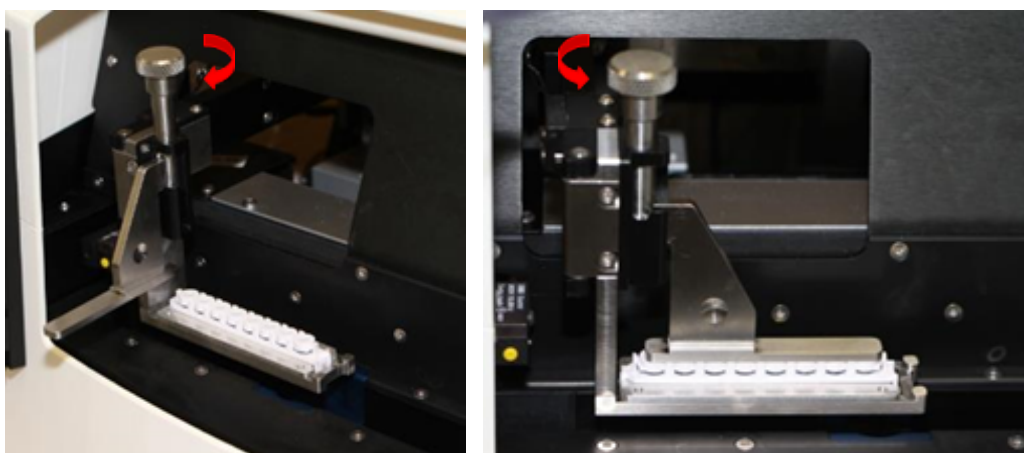The racks are keyed to load into the sample tray in the correct orientation only.



**Figure 3.13** Place rack in sample tray, rotate weight counter-clockwise & lower onto tubes

## 3.6 Running a Method

When the "Run" button is green, the selected method may be started. Click the "Run" button to start. The instrument will automatically adjust the water level and temperature to the appropriate settings, then begin the acoustic treatment.

As the method runs, the graphic for the tube under treatment flashes, and the actual average incident power and temperature data are displayed, as shown in **Figure 3.14**.
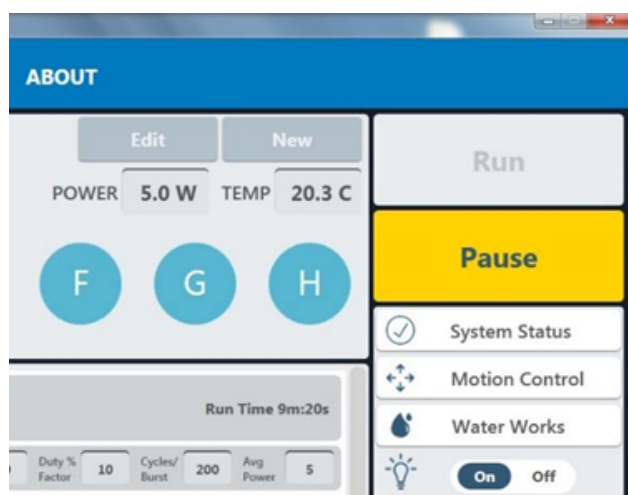
**Figure 3.14** Screen Shot of a Running Method

The user can, at any time while running a method, click "Pause" to pause a running method.

When the method is paused, the user may continue the process by clicking on the "Resume" button or terminate the process by clicking on the "Abort" button.

When the method has finished, the message "Method Complete" is displayed.  Click "OK", then click the "Motion Control" button and select "Home" from the motion menu to raise the sample tray out of the water bath.  When homing is finished, open the safety cover and remove the treated sample rack.

> **NOTE:** Failure to select the "Home" button can result in misalignment of the ME220 under some conditions.  Always use the "Home" position to remove vessels from the ME220.

## 3.7 Instrument Light
The instrument light is located above the acoustic assembly and inside of the **Safety Cover**. By default, the light will be lit once the instrument is turned on.  This light may be turned off by clicking the Light control on the Home screen.
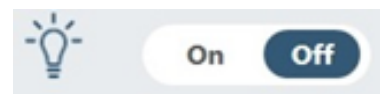
## 3.8 User Maintenance
The water bath should be drained when the ME220 is not in use by clicking the **Water Works** button at the right of the main screen.  The Water Works options will be displayed.
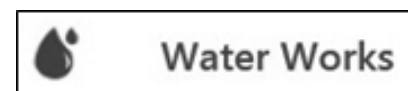
Click on "*Empty*".   Remove the rack from the sample holder and place it on a clean, dry surface. Rotate the sample arm forward and remove the wave guide.  Blot up water remaining on transducer or in bath using a soft, lint-free cloth, wipe, or swabs included with instrument.  Dry the wave guide and store it on a clean, dry surface, or in the ME220 bath.  Close the Safety Cover.

### Cleaning Protocol

A cleaning protocol using a solution of 10% bleach or equivalent (such as NaDCC) should be performed once a month as follows, to prevent biological growth in the water bath or plumbing of the Water Works system.

- Place a solution of 300 ml of water and 30 ml of household bleach, or equivalent, in the Water Works Supply bottle, and verify the Waste bottle is not full.

> **NOTE:** DO NOT use "fresh" or "pleasant smell" bleach products.

- Click on the Water Works button to display the Water Works menu.
- Click "Fill" and wait for bath to fill.
- Click "Circulate" in the Water Works menu.
- Allow the bleach/water solution to circulate for at least 10 minutes.
- Click "Stop", then click "Empty" in the Water Works menu and allow the bath to drain.
- Empty and thoroughly rinse the Supply bottle, fill the Supply botte with deionized water, and reinstall bottle to the ME220 bottle bay.
- Click "Fill" in the Water Works menu, then "Circulate".  Allow the water to circulate for 3 to 5 minutes.
- Click "Stop", then click "Empty".  Allow the bath to drain.
- Repeat the above 2 steps twice more.
- Empty and thoroughly rinse the Waste bottle and reinstall the empty Waste bottle in the ME220 bottle bay.

### Water Works Particulate Filter

The Water Works particulate filter element should be replaced every 12 months.  Replacement filters, PN 500529 can be obtained from Covaris Customer Service.  See **Appendix D** for filter replacement instructions.

### Archiving History Files

SonoLab generates a history file each time a method is run.  This file enables the user to maintain a document of the run, including temperature and power graphics and other pertinent textual information.  Over time, a large number of history files can cause SonoLab performance to slow down, especially when starting the application.  To prevent this slowdown, the history should periodically be archived.

To archive the history, navigate to Documents\Covaris\SonoLab 8\History.  Sort the files by date, then select older files, for instance, files older than 30 days.  Right-click on the selected files and select "Send to Compressed (zip) File".  The zip file may be left in place or moved to a location of the user's preference.

## 3.9 Shutdown Procedure

If the instrument will be idle for an extended time (e.g. overnight or a weekend), shut down the instrument using the following procedure:  Empty the water bath (refer to **Section 3.8**), and store the sample rack in a clean, dry place.

> **CAUTION:** Leaving water in the water bath for an extended time may allow algae growth which will interfere with sample processing.

Exit SonoLab software by clicking on the red "X" in the upper right corner.  If there are any unsaved changes, a message box will be displayed asking if user wants to save the changes; select "Yes" to save the changes and leave SonoLab, or select "No" to leave SonoLab without saving the changes.  The user can also select "Cancel" to continue working with SonoLab.

To shut down the laptop, the user should select "Shut down" from the Start menu on the Windows screen to power down the computer.

The instrument should be powered off after the SonoLab software has closed.  To turn off the instrument, the user needs to move the power switch to the OFF position.  The power switch of the ME220 is located at the rear of the right side of the instrument, above the power cord.

# 4. Using a Pre-installed Method

SonoLab has protocols preinstalled on the computer running SonoLab. These methods have been developed by Covaris scientists and are ready-to-use for most common applications. To select a preinstalled protocol, click on the pull-down menu to display the list of available methods.
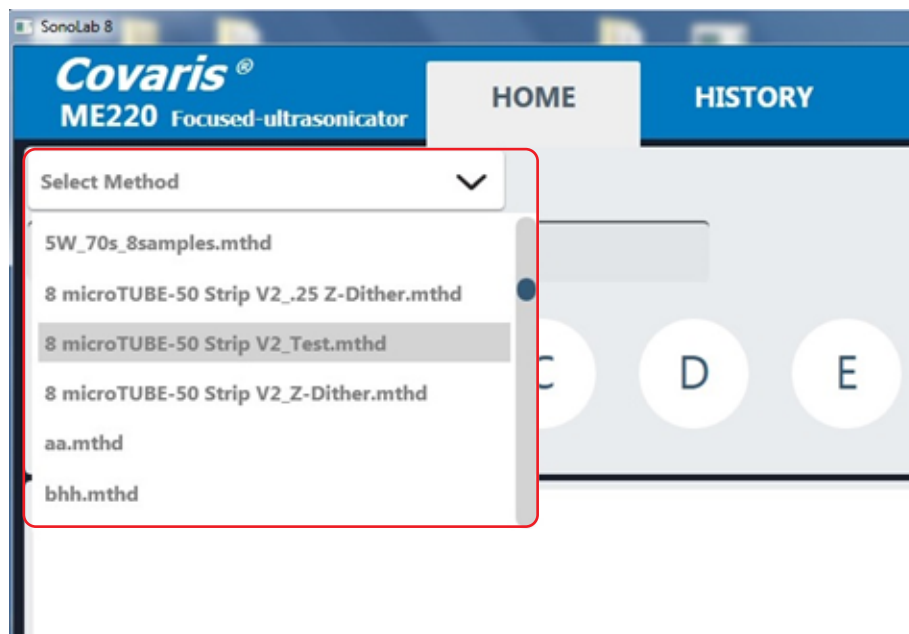


## 4.1 DNA Shearing Protocols

DNA Shearing protocols are optimized for the mean DNA fragments size, sample volume, and sample vessel used. These protocols are stored in a folder called "DNA Shearing for ME220".

- Naming convention is as follow: DNA_[size]_bp_[volume]_ul_ [type of vessel]

## 4.2 Chromatin Shearing Protocols

Chromatin shearing protocols allow the user to run a time course at a level of power optimized for efficient chromatin fragmentation when used in conjunction with Covaris truChIP® kits.

- Naming convention is as follow: ChIP [duty factor]_[time]

# 5. Create and Edits Methods

The SonoLab Method Editor supports the creation of new methods and editing of existing ones.

A method consists of one or more Processes.  A Process contains at least one Treatment.  Most methods will contain a single process, containing a single treatment step.  But more complex methods are easily constructed.

A Process, and its included treatment(s), may be repeated as many times as necessary.  A subset of treatments within a process may also be repeated.  If all the steps of the Process are to be repeated, a global Repeat step is placed outside the process.  See **Section 5.1.1** below.  If only some treatment steps are to be repeated, select the Repeat step from within the Process, as explained in **Section 5.1.6** below.

## 5.1 Create a New Method

To create a new method, click the "New" button on the main screen (see **Figure 5.1**).
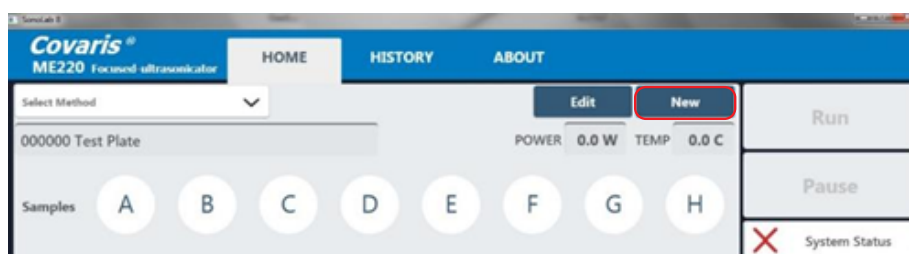


**Figure 5.1** New Method Button on Main Screen

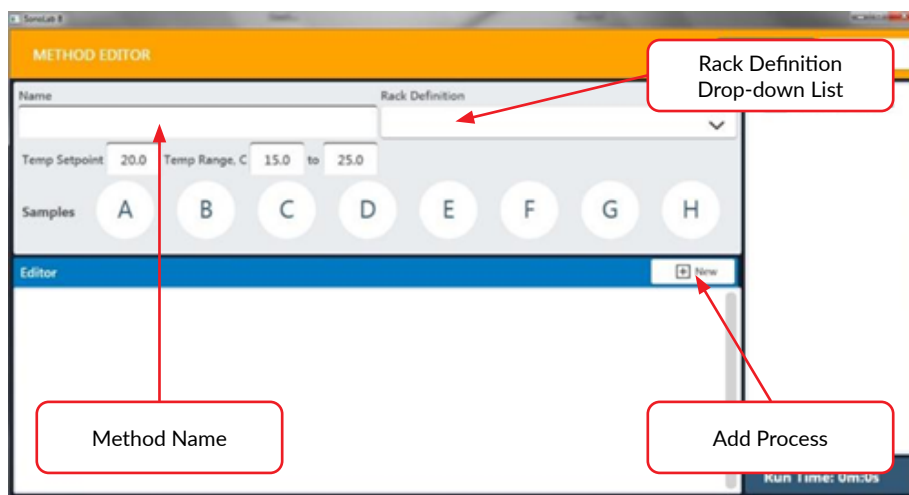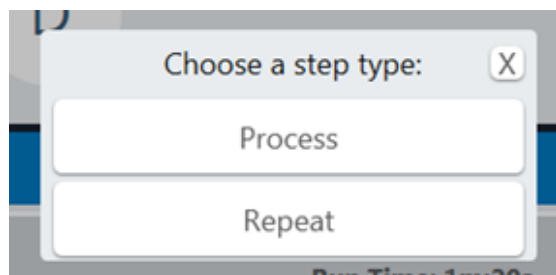The Method Editor window will appear (See **Figure 5.2**).



**Figure 5.2** Method Editor Window

1) Enter the name of the new method in "Name" box as shown.

2) Select the appropriate rack for the tubes used in this method from the "Rack Definition" drop-down list.
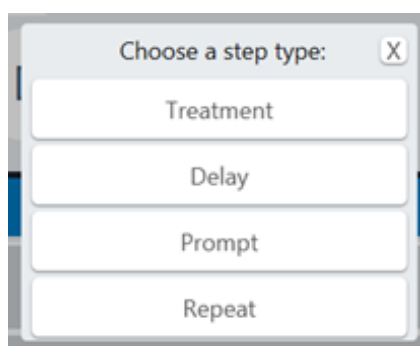
### 5.1.1 Add a Step in the Method

Click the **+** New button.  The "Choose a Step Type" pop-up is displayed.



Select "Repeat" if all steps are to be repeated one or more times.  Otherwise click "Process".  The process bar is displayed.  If only some steps are to be repeated, see using the repeat function in *Section 5.1.6* below.
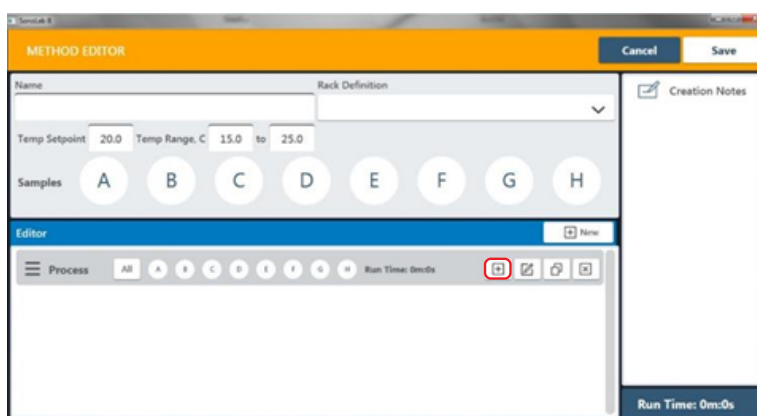


Click the **+** button inside the process bar to display the step type menu.

### 5.1.2 Choosing a Treatment

As an example, choosing a Delay step introduces a fixed time delay into a method which allows the sample to settle between two different treatment steps.  The delay is specified in seconds.
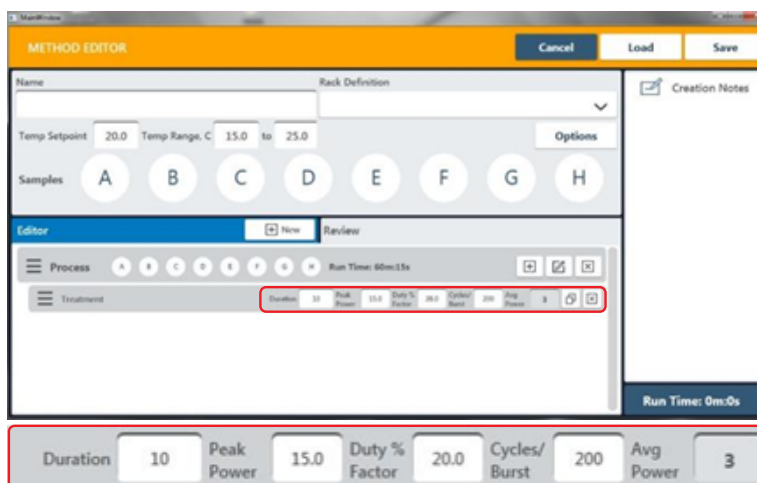


**Figure 5.3** Add a treatment

Treatment parameters are chosen by directly editing the Value box.  The treatment parameters are:

*Peak Incident Power (PIP)*

This parameter measures the peak electrical power used to drive the ultrasound transducer.  In the ME220 instrument, the adjustable range of Peak Incident Power is 0.1 to 75 Watts.

*Duty Factor*

This parameter defines the ratio between the on-time of the transducer and total operation time, and is expressed as a percentage.  For a given Peak Incident Power, a larger Duty Factor results in more acoustic energy generated from the transducer.  Users can define the Duty Factor from 0.1 to 50% in the ME220 instrument, depending on the PIP chosen.

The product of the Peak Power and Duty Factor gives the approximate Average Incident Power (AIP) generated from transducer.  For example, if Peak Incident Power is set to 10 Watts and Duty Factor is set to 15%, the Average Incident Power will be approximately 1.5 Watts.  However, the real average incident power from the transducer may vary due to factors such as sample tube type, water level and water temperature.

SonoLab will not allow a combination of Duty Factor and PIP which results in an Average Incident Power exceeding the ME220 limit of 20 watts.

*Cycles per Burst*

This parameter indicates the number of electrical signals used in each acoustic "burst" of the transducer.

One may define Cycles per Burst (cycles/burst) from 50 to 1000 in the ME220 for different applications.  Advanced users may use this parameter to fine tune their experiments.
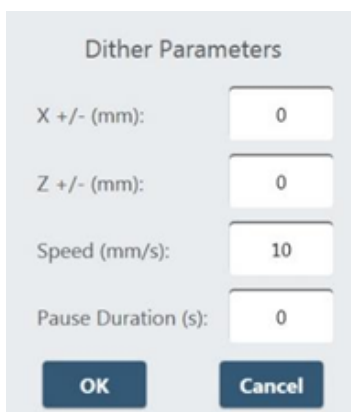
### Duration

This parameter defines the amount of time for which the sample is under a treatment. The measurement of the Duration is in seconds.  The maximum time for any treatment is 3600 seconds.  Should a longer treatment be necessary, "Duplicate Steps" can be used to extend the treatment.

SonoLab displays the Average Incident Power, calculated from the Peak Incident Power and Duty Factor, adjacent to the treatment parameters in the treatment step bar.  In the ME220 instrument, the maximum Average Incident Power is 20 Watts (for example, 50 watts Peak Incident Power and 40% Duty Factor are selected).  SonoLab does not allow the user to select a combination of Peak Incident Power and Duty Factor that results in more than 20 Watts average incident power.

### Dithering

The ME220 motion system provides dithering motion capability in the X axis (left-to-right) and Z axis (up-and-down).  If a treatment specifies dithering, click on the "Pen-&-Paper" icon on the right end of the Process bar to display the Dithering Parameters pane.
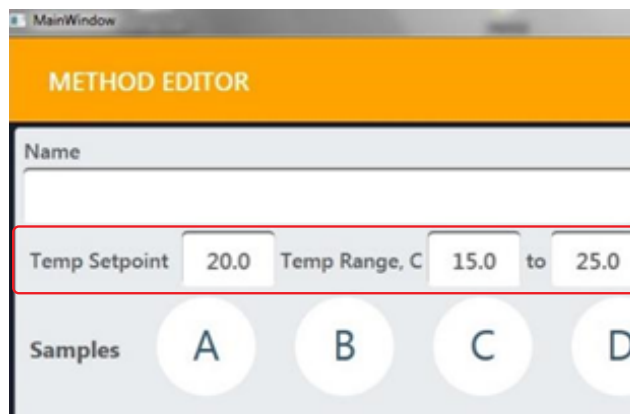


**Figure 5.4** Dither Parameter Pane

Enter the specified dithering parameters and click "OK".

### Temperature Set Point and Range

Enter the temperature set point for the protocol and the allowable range into the text entry fields.  SonoLab allows a minimum range of +/- 3 degrees.



**Figure 5.5** Entering Temperature Set Point and Range from lower to upper limits

### 5.1.3 Treatment Parameters

Once the treatment parameters have been defined, click in the treatment well graphic to define which rack positions will be treated. Any number of wells may be selected for treatment; positions selected for treatment turn from white to blue. When the method is run, the graphic for the position being treated will flash, indicating which position is under treatment.
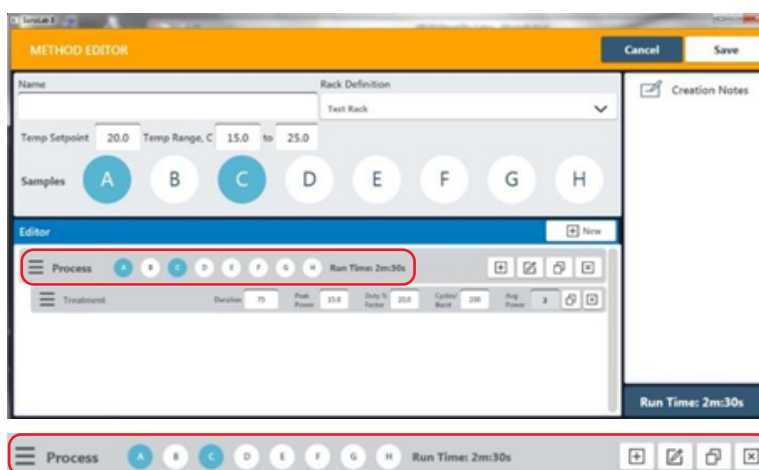


**Figure 5.6** Wells A and C Selected for Treatment

### 5.1.4 Choosing a Delay Step

Choosing a Delay step introduces a fixed time delay into a method, allowing the sample to settle between two different treatment steps, for example. The delay is specified in seconds. See **Figure 5.7**.
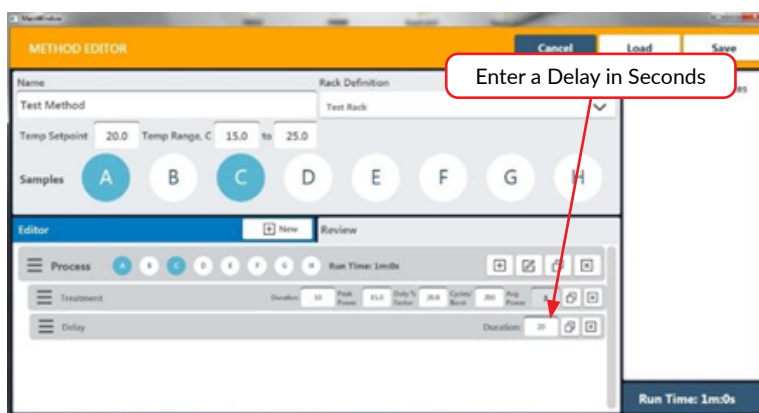


**Figure 5.7** Add a Delay Step

### 5.1.5 Choosing a Prompt Step

Choosing a Prompt step displays a specified message on the screen and pauses execution of the method until the operator clicks OK.

Define the prompt message by clicking on the "pen & paper" button of the prompt bar to display the text entry field for the prompt. Enter the text of the prompt, and click OK. See **Figure 5.8**.
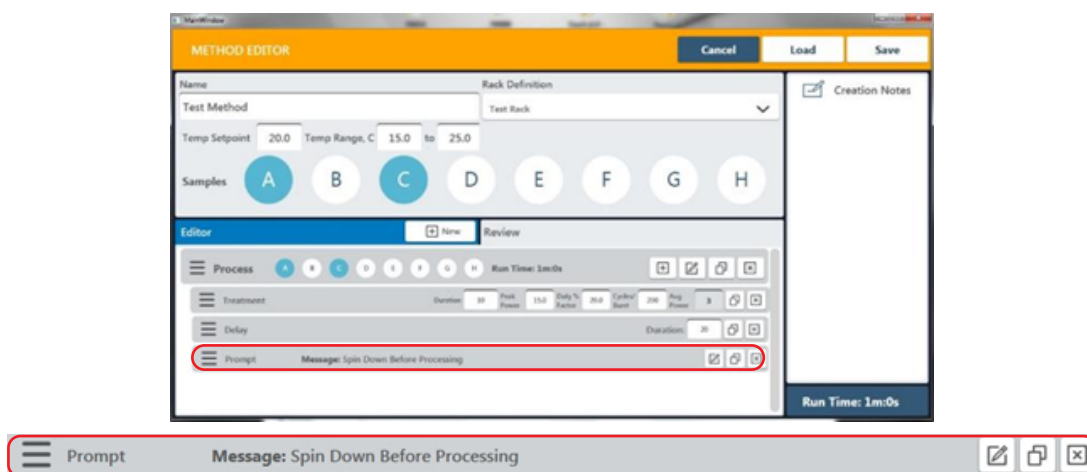
Figure 5.8 Add a Prompt Step

When the prompt step is reached during the treatment, the method pauses, and the prompt message will be displayed as shown in **Figure 5.9**.
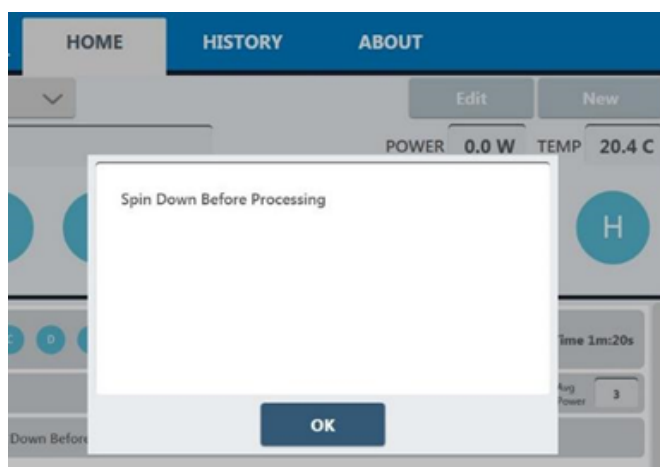


Figure 5.9 Example of a prompt message displayed

The "Creation Notes" section is a text entry box at the right side of the Method Editor screen in which the user may enter, for example, descriptive text relating to the method being created.

### 5.1.6 Choosing a Repeat Step

Choose a Repeat Step when one or more Treatment, Delay, or Prompt step needs to be repeated one or more times.
See **Figure 5.10**.

> **NOTE:** The repeat functionality is only available on SonoLab version 8.0.1 and up.

**Figure 5.10** Repeat function programming for sequential sample treatment

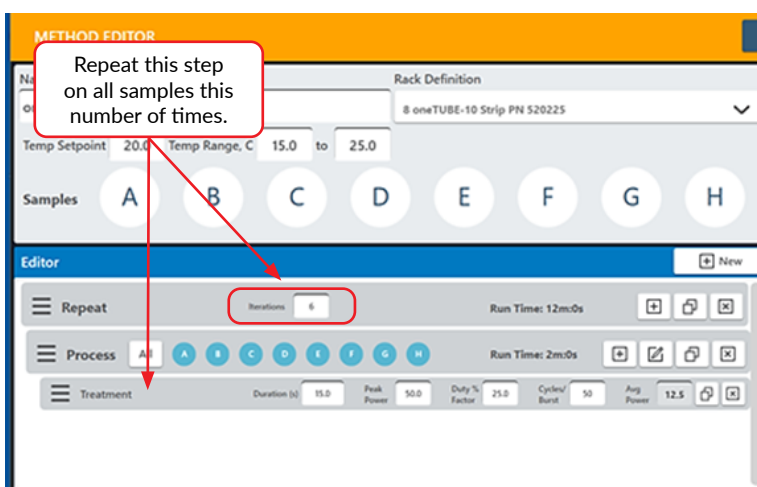Above: The Repeat function selected prior to the Process and Treatment Steps will provide one pulsing step to each selected sample before repeating the next pulsing step in the same manner. In other words, samples A through H will be treated one time with the programmed process and then return to sample A to repeat the process.  This method design is optimal for applications such as DNA shearing.

### 5.1.7 Protocol Arrangement

If desired, protocols can be arranged to treat one sample completely before moving on to the next sample.  In other words, sample A will be treated with the entire repeated process before moving onto sample B.  Simply select a Process step first, then select a Repeat step with Treatment and Delay steps following, as needed (see **Figure 5.11**).
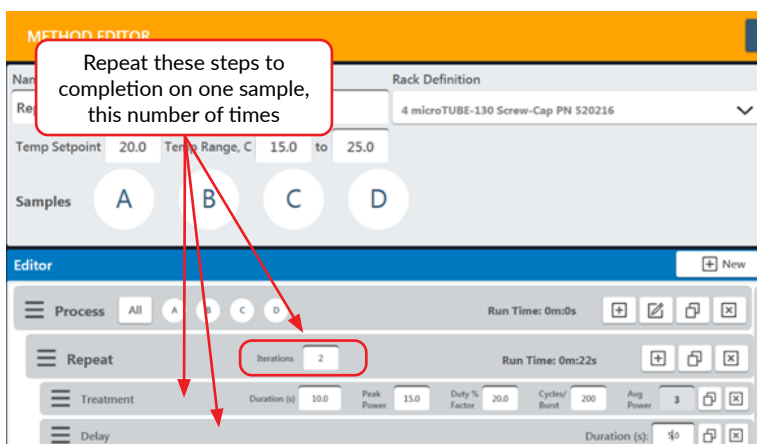


**Figure 5.11** Repeat function programming for one sample at a time

Please contact ApplicationSupport@covaris.com for inquiries regarding the Repeat pulsing feature and method setup.

### 5.1.8 Save Parameters

When all method parameters and steps have been defined, click to save changes.

## 5.2 Modify Existing Method

To modify an existing method, select the method as described above, then click the "Edit" button.  The method editor window will be displayed.



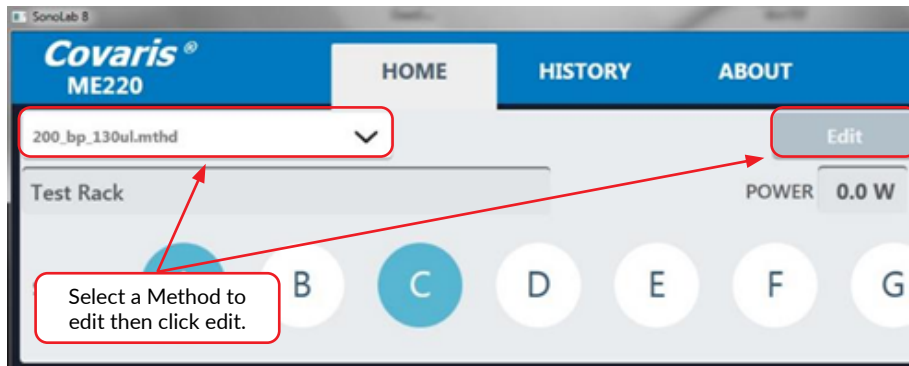**Figure 5.11** Repeat function programming for one sample at a time

# 6. SonoLab Application Software

The main components of SonoLab software include:

- **Home** – compose or select methods to run
- **History** – view system usage history
- A**bout** – information on system configuration

## 6.1 Home Screen

The Home Screen (**Figure 6.1**) is the principal user screen for the system.  See **Section 3.2** for details on operating the instrument through the Home Screen.



**Figure 6.1** The Home Screen

## 6.2 History Tab

The History tab (**Figure 6.2**) provides a scrolling list containing the history of every method performed on the instrument.  This list can be sorted by method name, date the method was run, the operator who ran the method, or the run status (complete, aborted, failed).



**Figure 6.2** The History Screen

Double-click on a line to display its details.  The left side of the screen will display textual information concerning the method.  A pane on the right side contains graphs of the temperature and power levels generated for each sample treated by the method.  This pane scrolls as necessary to display the graphs of all samples treated by the method.  See *Figure 6.3*.
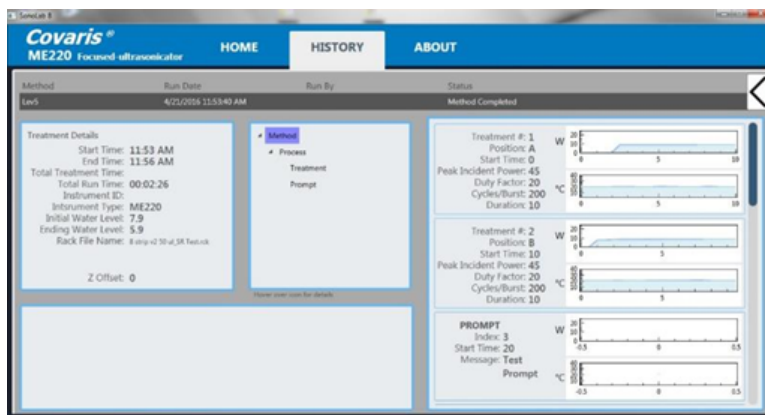


**Figure 6.3** Details of a Method History

Double-click on one of the graphics on the right side.  The graphic will be outlined in orange, indicating it has been selected, and additional information pertaining to the part of the method which generated the specific graphic is displayed at the lower left.  See *Figure 6.4*.
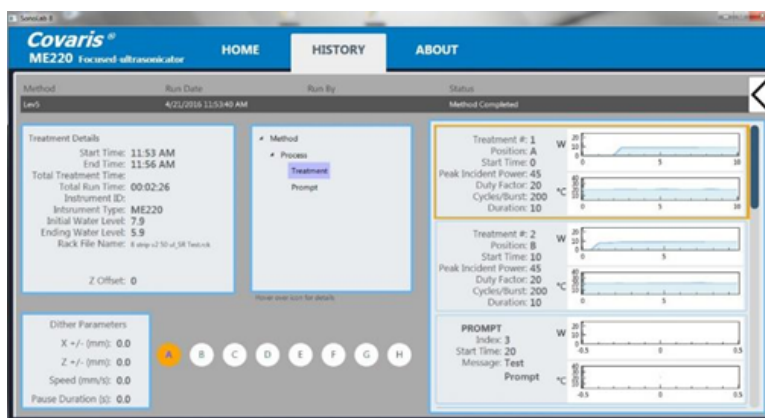


**Figure 6.4** Additional History Details

**NOTE:** Occasionally, a history file may be corrupted.  If this occurs, clicking on the "History" tab will result in an error message similar *Figure 6.5*.
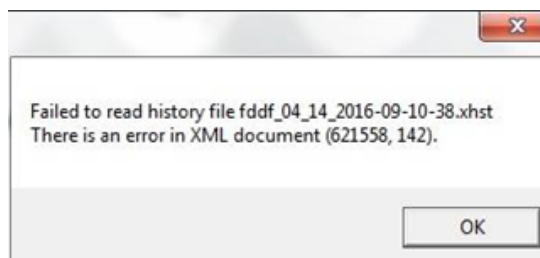


**Figure 6.5** Message Indicating a Problematic History File

If this occurs, click the "OK" button to continue to the "History" screen.  However, the error will be presented each time the "History" tab is selected, until the offending file is removed.

To remove the bad file, navigate to Documents\Covaris\SonoLab\History, then locate the offending file, in this example, "fddf_04_14_2016-09-10-38.xhst".  Drag this file to the Recycle Bin.

> **CAUTION:** The history list can become long and slow the system. Periodically, move older files from Documents\Covaris\ SonoLab\History to a convenient storage location.  This will prevent slowing of the system while allowing access to older history as necessary.
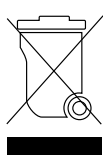
## 6.3 About Tab

The About Tab provides user basic information about the instrument such as serial number, software version, board serial numbers etc. See *Figure 6.6*.



**Figure 6.6** About Tab

# 7. System Specifications

| | |
|---|---|
| **Treatment System** | Bench-top; high intensity acoustic transducer, 1 to 8 sample batch process, temperature monitoring and controlling device, water bath with sample holder and safety enclosure |
| **Treatment Power** | 75 Watts Peak Incident Power, 20 Watts Average Incident Power |
| **Dimensions** | 18.75" W x 18.75" D x 14.75" H (47.75 cm x 47.75 cm x 37.5 cm) |
| **Weight** | Approximately 55 lbs. (25 Kg) (without computer) |
| **Power Requirements** | 100-240 VAC 500 VA, 50-60Hz, 5 Amp Slo-Blo Fuse |
| **Ambient Temperature Range** | 19 °C to 25 °C (66 °F to 77 °F) |
| **Ambient Humidity Range** | 30 to 70% |
| **Regulatory Labeling** | CE, ETL Mark (for Product Safety), WEEE |
| **Safety** | Certified to IEC/EN/ANSI/UL61010-1 and CAN/CSA C22.2 No. 61010-1 "Safety Requirements for Electrical Equipment for Measurement, Control, and Laboratory Use, Part 1: General Requirements" |
| **EMC** | Certified to EN 61326-1 "Electrical equipment for measurement, control and laboratory use – EMC requirements – Part 1: General requirements". Also certified to FCC 47CFR Part 15 – Radio Frequency Devices and Part 18 – Industrial, Scientific and Medical Equipment, and ICES-003 Class A for Industry Canada |
| **Water Bath** | AFA-grade Water or Highly Purified Water, at least ASTM Type III or ISO grade 3 |
| **Method Temperature Set Point** | Programmable +6.0 °C to +40.0 °C |
| **Computer** | Notebook computer supplied by Covaris |
| **Operating System** | Microsoft Windows 7 or Higher |
| **Application Software** | Covaris SonoLab |
| **Data Input** | Keyboard, mouse |
| **Chiller** | Solid state chiller for heating and cooling (built-in) |
| **Power** | 0 to 160 Watts, 24 volts |

# 8. Troubleshooting

| Problem | Corrective Actions |
|---|---|
| Unable to HOME | - Ensure the Instrument is turned on and the USB cable is connected.<br>- Ensure the door is closed.<br>- The SW will not function properly if multiple instances of SonoLab 8 are running simultaneously.  Close out of all instances of SonoLab 8 and restart a single instance of the Software.<br>- You can HOME the system by going to the Motion Control button and clicking on HOME. |
| Interference between the Waveguide and Consumable rack when the arm moves into position for treatment | - Ensure the correct Waveguide is in place for the chosen consumable.<br>- Ensure you are using the correct rack and consumable.<br>- Ensure the correct Method (and rack definition) has been picked for the consumable chosen.  The Method sets the arm motion. |
| RUN button disabled | - A Method must be chosen to enable RUN.  Ensure a Method has been chosen.<br>- System status conditions have not been met.  Click on the System Status button.  Identify the conditions that are marked with a red X and address the indicated conditions. |
| Drain Taking Too Long or Water Pressure Too High message | - Ensure the Waste bottle is properly connected to the Quick Disconnect Cap (see **Section 3**).<br>- There could be a blockage in the drain lines.  Ensure the bath is clean and free of debris.  This may be resolved with a bath cleaning operation (See Section 3). The filter may need to be changed (See **Appendix D**). |
| Water Level Failure Error / Method Failure Due to Water Level Adjustment Timing Out | - Rerun / Restart the method |
| Water in bath will not completely drain | - If the water level is below 1, as seen under the Water Works status panel, the bath will not drain further.  Use the Fill button to fill the bath above level 1 then use the Empty button to start the drain operation. |
| Shearing not as expected or inconsistent across a rack | - Verify the ME220 is level (see **Section 2**).<br>- Ensure the rack is properly seated into the sample arm and the weight is in place. |
| "Failed to read history file…" error message | - Refer to **Section 6.2** for handling damaged history files. |
| Additional Comments | - The SW will not function properly if multiple instances of SonoLab 8 are running simultaneously.  Close out of all instances of SonoLab 8 and restart a single instance of the Software.<br>- The system can be reset by turning off both the SW and the instrument and restarting.  It is recommended that the instrument be powered up first followed by the SW. |

# 9. Appendix

## Appendix A: M-Series Site or Transport Preparation

**Placement of Equipment**

The area required for the instrument is approximately 18.75" wide by 18.75" deep by 14.75" high (47.75 cm x 47.75 cm x 37.5 cm). Allow sufficient space in front of the bench to access each component of the installation. *DO NOT* cover the air intake at the front panel.

Allow sufficient space to the right of the ME220 to allow access to the Water Works supply and waste bottle compartment.

**Power Requirements**

Provide at least two standard outlets, one for the laptop computer, and one for the ME220. The ME220 draws a maximum of 500 VA. The Covaris M-series instruments are configured with a universal power supply.

> **WARNING:** To prevent the possibility of electrical shock, always plug the system into a grounded power source.

**Water Requirements**

The ME220 system water bath requires about 85 milliliters of AFA-grade Water. The water bath should be drained daily, or when the ME220 is not in use for a significant period of time. Water is added to the bath automatically as necessary by the *Water Works* system. Sensors notify the user if the Water Works supply bottle is empty or if the waste bottle is full. Covaris recommends AFA -grade Water. It is also possible to use highly purified water (Highly Purified Water at least ASTM Type III or ISO grade 3)

**Moving or Packing the Instrument for Transport**

When moving the instrument within a laboratory, or between nearby laboratories, please employ the following guidelines:

- Drain the water bath by clicking on the Water Works button and selecting "Empty".
- Gently support the sample tray using foam or bubble-pac to prevent loss of alignment from vibration while transporting the ME220.
- Turn the instrument off. Disconnect line cords and USB cable.
- Remove the laptop from the top of the instrument (it is held in place by Velcro strips)
- Remove remaining water from the acoustic assembly
- Lift the unit by its base-plate (not by the plastic case or doors). Transport by cart.

When packing the unit for shipment, please follow the guidelines above. Additionally, if feasible, obtain a packing box from Covaris and employ the foam end-cap inserts to support the instrument. If this is not feasible, wrap the main instrument in ample bubble wrap or similar packing. Wrap and package accessories (laptop, safety cover, sample holder, and cords) separately to avoid damage.

Please contact our Covaris Customer Support Department (customerservice@covaris.com) for guidance and assistance.

## Appendix B: Updating SonoLab Software

The latest version of SonoLab can be found in the resource section of Covaris website, https://www.covaris.com/m-series/

You will need to create an account to access this area of the website.

**Un-install Existing SonoLab**

1. Turn off ME220 series device and close SonoLab software application.

2. Navigate to the Control Panel>Programs>Uninstall a program.

3. Select the program named SonoLab and click uninstall.

4. "Are you sure you want to uninstall SonoLab?" Click Yes.

5. The desktop shortcut titled "SonoLab" will remove itself.

**Install updated SonoLab Application**

1. Download the latest SonoLab version and save it on your desktop.

2. Open the SonoLab.x.zip file.

3. Right click on the setup.exe application. Run as an Administrator.

4. You must Agree to the Program License Agreement, click Next to put SonoLab.x in C:\Program Files, and then click Next to begin the installation.

5. Click Yes to enable the installer to install SonoLab.x if Windows User Access Controls have not been disabled.

6. Turn on the instrument and plug the USB cable into a USB port on the computer.

7. Navigate to C:\Program Files\Covaris\SonoLab on the computer.

8. Right Click on Sonolab.exe and select "Run as Administrator". SonoLab will open its' splash screen.

Depending of your previous SonoLab version, you may have to update your instrument Firmware.  Contact Covaris Technical Service (techsupport@covaris.com) for additional information.

![Covaris logo]

## Appendix C: Installing SonoLab on a New Computer and Installing Windows Instrument Drivers

The latest version of SonoLab can be found in the resource section of Covaris website, http://covarisinc.com/resources/registered-users-login/m-series/

You will need to create an account to access this area of the website.

**Install SonoLab Application**

1. Download the latest SonoLab version and save it on your desktop.

2. Open the SonoLab.zip file.

3. Right click on the setup.exe application. Run as an Administrator.

4. You must Agree to the Program License Agreement, click next to put SonoLab.x in C:\Program Files, and then click Next to begin the installation.

5. Click Yes to enable the installer to install SonoLab if Windows User Access Controls have not been disabled.

6. Turn on the instrument and plug the USB cable into a USB port on the computer.

7. Navigate to C:\Program Files\Covaris\SonoLab on the computer.

8. Right Click on Sonolab.exe and select "Run as Administrator". SonoLab will open its' splash screen.

Depending of your previous SonoLab version, you may have to update your instrument Firmware.  Contact Covaris Technical Service (techsupport@covaris.com) for additional information.

**Install Windows Instrument Driver**

When an ME220 (with power ON) is connected by USB (Universal Serial Bus) for the first time, Windows will identify the ME220 by its Vendor and Product ID numbers and install specific device drivers (software files required for communications with a USB device).

Windows will attempt to install these drivers in the following order:

***Automatic Driver Installation:*** If any ME220 has ever been connected to the computer, then a copy of the appropriate drivers will be available to Windows locally, and Windows will automatically install these.

If Windows does not find a local copy of the necessary drivers, it will automatically search Windows Update (assuming internet access is available via wireless link or Ethernet cable and Windows is configured for automatic updates).  In either case, upon locating them, Windows will automatically install the drivers and pop up the following message onto its desktop:
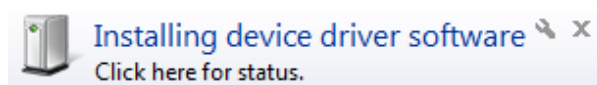


Drivers for several devices within the ME220 will be installed sequentially.  This may take several minutes.  If you click on the pop-up message, the status of these installations may be viewed.  Please allow all of these drivers to install and for Windows to confirm successful installation of all drivers with the following message:
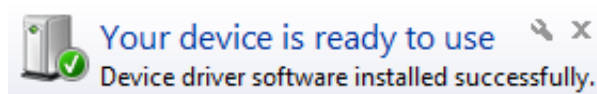


Drivers are now installed and SonoLab will automatically connect with the ME220 once started.

***Manaul Driver Installation:*** If the automatic installation fails to occur, Windows will post the following message:



If restoring internet connection or automatic Windows Update fails to start the automated installation process, then the following manual process will be required.

Click on the Windows Start button.  Either via Control Panel, or by right-clicking on Computer and choosing Properties, locate Device Manager.  Click on this link to open the Device Manager window.  In the list of devices, locate Other devices and entries for RF Controller Board:
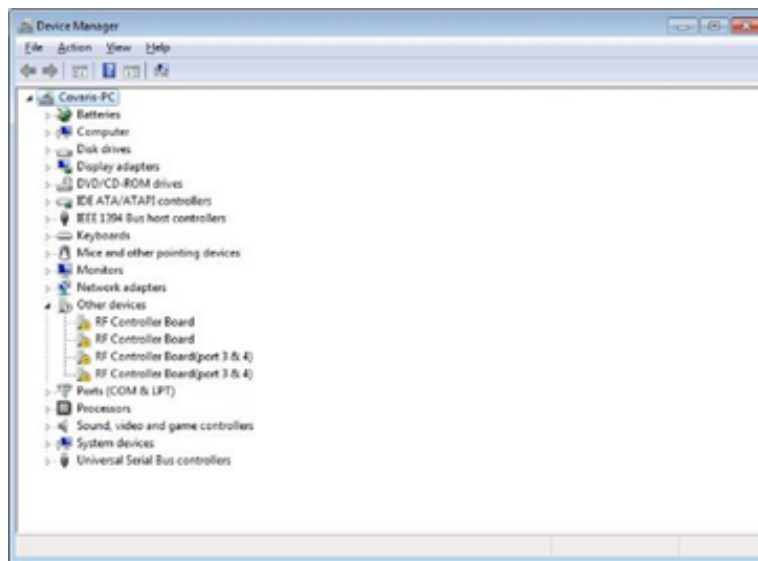


Under Other devices, right click first device RF Controller Board and select Update Driver Software:
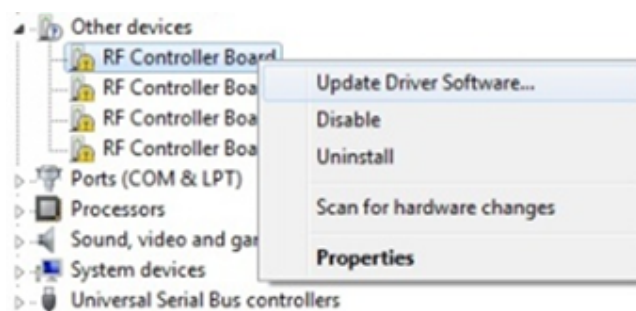


When the driver software update dialog opens, select the option to Browse my computer for driver software:
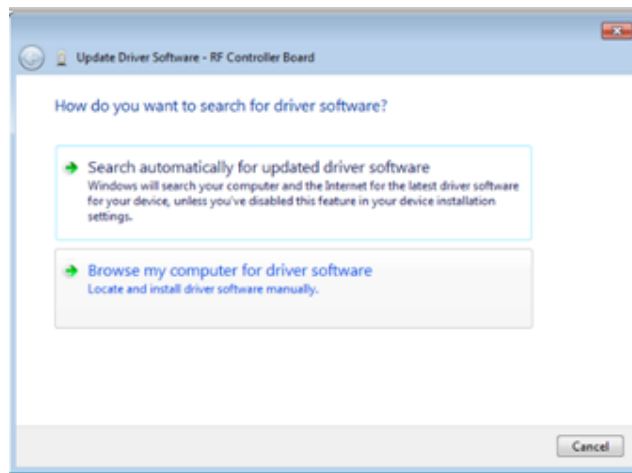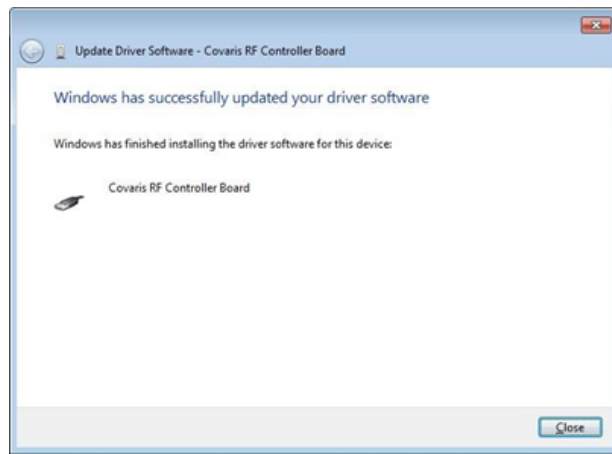
Point the file open dialog to "C:\Program Files\Covaris\Driver" and verify that "Include subfolders" is checked:
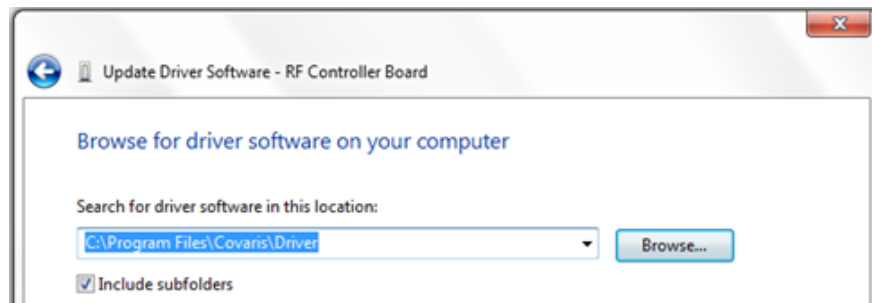


Click next. You may be notified that "Windows can't verify the publisher of this driver software". Click on "Install the driver software anyway":
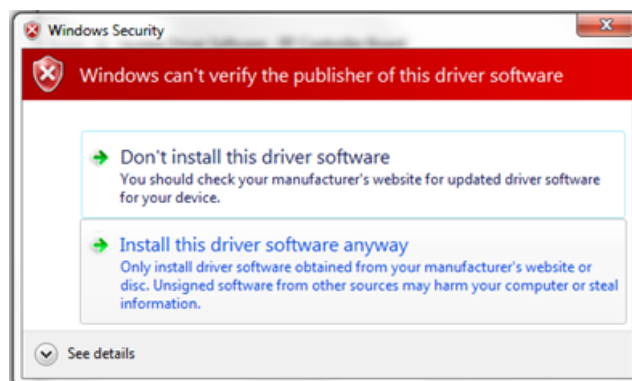


Windows will install the driver and post the following dialog box with a message, "Windows has successfully updated your driver software".

Click the Close button.  The Device Manager screen will refresh.  Return to Other devices.  Note that there is now a 'USB Serial Port' listed under the three 'RF Controller' lines.  Right-click on the 'USB Serial Port' and select 'Update Driver Software…', as shown in the image below.  Repeat the installation steps shown above.
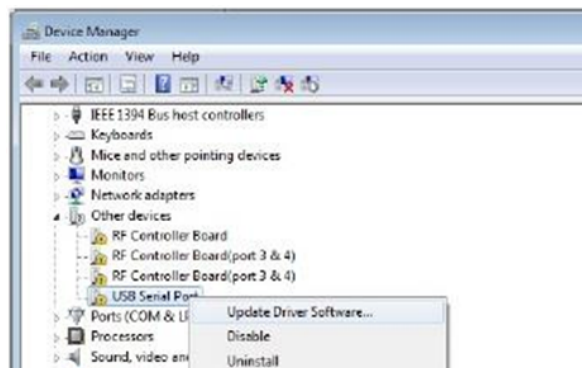


Return to Other devices and repeat this process in order (top to bottom), for the 3 remaining entries, until all Covaris device entries have been assigned a COM number under Ports (COM & LPT).  The 'Device Manager' window should now show:
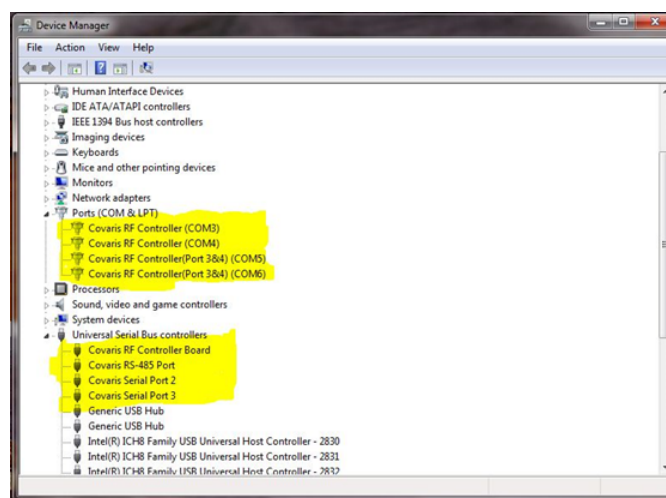


> **NOTE:** The 'COM' numbers will vary from computer to computer but should occupy four consecutive ports.

## Appendix D: Replacing the ME220 Particle Filter Element and Gasket (PN 500529)

The Particle Filter removes particulate that could otherwise damage the pump or negatively affect the acoustic performance of the instrument. The Particle Filter Element and Gasket should be replaced annually.

1. Wear gloves when replacing the Particle Filter Element and Gasket.

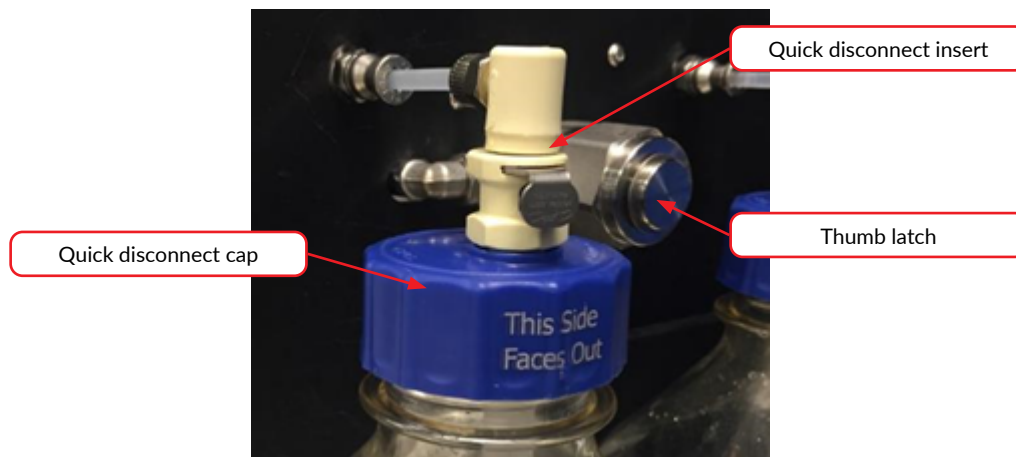2. Disconnect the bottle by pressing the thumb latch on the Quick Disconnect Cap. See *Figure D.1*.



**Figure D.1** Quick Disconnect Components

3. Use the Particle Filter Wrench, which was provided with the instrument, to remove the filter cap.



**Figure D.2** Quick Disconnect Components

4. Remove the loosened filter cap; then remove the Particle Filter Element and Gasket behind it. Discard the used filter element and gasket.
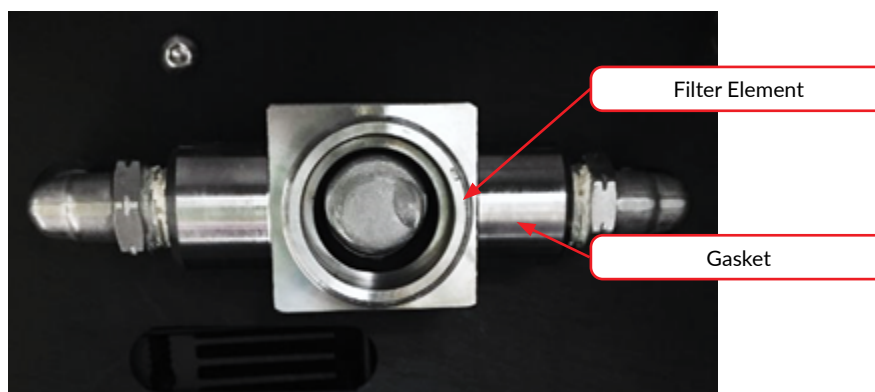
**Filter Element**

**Gasket**

**Figure D.3** Particle Filter Components

5. Rinse the gasket surface with a squirt water bottle and wipe with a Kim-wipe or similar lint-free towel.

6. Place the replacement Particle Filter Gasket against the flat surface in the filter housing.



**Figure D.4** Particle Filter Gasket

7. Position the replacement Particle Filter Element so that the closed end of the cup faces out. G ently, but firmly, press the Particle Filter Element into the filter housing until it remains centered without being held.

8. Thread the filter cap onto the housing until it is finger-tight.  There should be no threads showing behind the filter cap (see **Figure D.5**).  If there are, the Particle Filter Element may not be properly aligned.  Remove the filter cap, straighten the Particle Filter Element, and reinstall the filter cap.
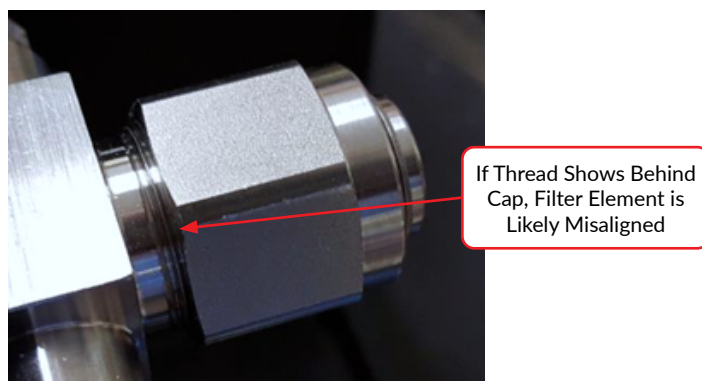


**If Thread Shows Behind Cap, Filter Element is Likely Misaligned**

**Figure D.5** Particle Filter Element Misaligned

9. Using the Particle Filter Wrench, tighten the filter cap approximately 1/8 turn beyond finger-tight.

10. Orient the bottle in the Water Works Bay with the label on the Quick Disconnect Cap facing out, as shown in **Figure D.6**.
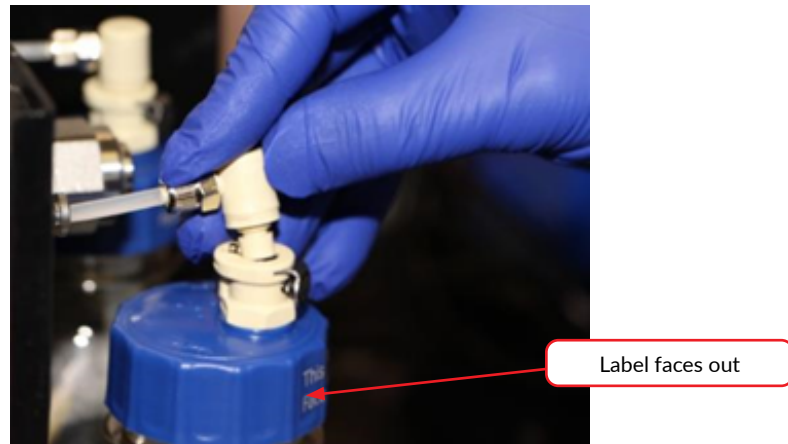


Label faces out

**Figure D.6** Connecting the Bottle to the Water Works System

11. Reconnect the bottle by pressing the Quick Disconnect Insert into the coupling on the Quick Disconnect Cap until it clicks into place.