

## R230 Focused-ultrasonicator Setup & Instruction Manual

Instrument and Software Developed for High-performance Sample Preparation using Adaptive Focused Acoustics<sup>®</sup> (AFA<sup>®</sup>) Energy

For Research Only

Model Numbers: R230 | Software: SonoLab 10





## **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 and Prevention (CDC), Universal Precautions for Prevention of Transmission of Human Immunodeficiency Virus, Hepatitis B Virus, and Other Bloodborne Pathogens in Healthcare Settings, published 1988, updated 2016
- Clinical and Laboratory Standards Institute (CLSI), GP17-A3 Clinical Laboratory Safety; Approved Guideline Third Edition, published 2012, ISBN 1-56238-798-7 (Electronic)
- Clinical and Laboratory Standards Institute (CLSI), M29-A4 Protection of Laboratory Workers From Occupationally
   Acquired Infections, Fourth Edition, published 2014, ISBN 1-56238-962-9
- Occupational Safety and Health Administration (OSHA), 29 CFR 1910.1030 Bloodborne Pathogens
- International Standards Organization (ISO) 15190:2020, Medical Laboratories Requirements for Safety

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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 to 25 °C (66 to 77 °F).
   DO NOT operate the instrument in a cold room environment. The system is designed to operate with a water bath and recirculating heater/chiller apparatus to control sample temperature.
- **NEVER** run a method without water in the bath since this could damage the transducer. The instrument is equipped with a water level sensor to protect the transducer and degassing pump. The system will not allow the degassing pump to operate or the acoustic treatment to start unless an adequate volume of water is detected.
- Distilled or deionized water should be used to fill the water bath.
- Unless the Water Conditioning System 3 (WCS) is actively employed, empty the water bath and wipe it dry **EVERY DAY** with a lint-free cloth. **DO NOT** leave water in the tank for an extended time as there is no water filtration or water cleaning system within the apparatus (unless WCS is employed).
- **DO NOT** employ isopropyl alcohol, ammonia-based or abrasive cleaners in the system, as these will damage the acrylic surfaces.
- Establish a standard of operation and test the equipment periodically, as described in the maintenance section of this manual.

**CAUTION:** All WCS 3 components have pressure ratings above Covaris' specified maximum operating pressure which have been extensively tested. However, if plant DI water is connected to the WCS, a catastrophic failure may result in substantial plant water spillage. It is the user's responsibility to specify and implement appropriate containment/alarm schema for their facility.



## 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, LLC. 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 (UTC-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/GMT
- E-mail queries to techsupport@covaris.com or applicationsupport@covaris.com

**Parts Replacement** – Replacement of parts (excluding consumables) from normal operation of equipment are provided on a priority basis. All labor and shipping charges are included. Failure due to accident, abuse, or improper operation is not covered.

This manual includes maintenance guidelines and troubleshooting tips.



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## 1. Introduction

## 1.1 Overview of the User Manual

This manual contains the operation and maintenance instructions for the Covaris R230 Focused-ultrasonicators. This document contains information essential to the proper use and care of the 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: Inconvenience if disregarded.

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 on its icon.

## 1.2 The R230 Instrument and the Covaris Process

The R230 Focused-ultrasonicators are based on Covaris Adaptive Focused Acoustics (AFA) technology. These instruments are designed to provide an integrated tool to be used in a high-volume production environment. This instrument is ideal for a variety of applications including DNA shearing, mammalian cell lysis, biomarker extraction from FFPE and tissue, compound screening, bead mixing, and pellet resuspension among other applications.

The instrument is comprised of the:

- Treatment System delivers the acoustic energy to the sample
- Safety System protects users from inadvertently contacting the high intensity acoustic energy
- Computer and Application Software provides an interface to control the device via a standard user interface or API
- Water Management System which consists of a cooler/heater and a UV treatment chamber with particulate filter to maintain water quality in the instrument bath

The Covaris Process uses adaptive focused acoustic energy to precisely control cavitation and acoustic streaming within the sample treatment vessel in a non-contact, temperature-controlled 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. Unpacking and Setup

## 2.1 Unpack and Identify Components & Accessories

The following items are included in the packaging found with the instrument:

• Electronics Box (eBox)



• On-Deck R230 Focused-ultrasonicator (for placement on the liquid handler deck)



• Water Conditioning System 3 (WCS 3)





• Notebook Computer with AC Adapter. Most installations will transfer Sonolab to the dedicated computer that is connected to the Liquid Handler.

**NOTE**: The notebook computer should be available for IQ/OQ, PM and Service. Please DO NOT use this computer for other purposes.

- Cables
  - Computer AC adapter power cord
  - Two instrument power cords
  - Ethernet cable from eBox to Computer
  - Communications and power cables between eBox, WCS 3, and On-Deck AFA
- Accessories
  - The WCS Tubing Kit contains 3 hoses
  - The Reservoir Kit contains 2 carboys and 2 hoses
  - Mount adapter, sample tubes and racks (varies by application)
  - Setup kit

## 2.2 Install the On-Deck Module

**2.2.1** Install the appropriate mounting/locating plate to the liquid handler by following the instructions provided with the mounting plate.

NOTE: A mount plate must be used for all integrations with liquid handlers.

2.2.2 Remove the shipping locks from the On-Deck module.

**CAUTION**: Shipping locks must be removed before powering up the instrument.

There are 2 bolts that lock the On-Deck motion system in place for shipping. The bolts are accessed from the bottom of the base plate.

- Tilt the On-Deck module up to access the bottom of the plate. In the image below, the module is resting on its back edge with the 2 shipping bolts circled. Be careful to remove only these 2 bolts.
- Lower the On-Deck module so that it is again resting flat on the bench.





- **2.2.3** Store the shipping bolts on the On-Deck base plate.
  - Remove the 5 flat head cap screws, 2 from the right, and 3 from the left lower edge of the On-Deck cover. Do not remove the screws in the inverted "U" shaped cut-outs.



Be careful to remove only the circled screws



• Lift the cover off and set aside.





Thread the two shipping bolts into the holes provided at the left rear of the module's base plate, near the cable connectors.



**2.2.4** Level the On-Deck module in the liquid handler.

- Place the On-Deck module, without its cover, into the mounting plate installed previously in the liquid handler.
- Perform any final adjustments required to the mounting plate.
- Level the On-Deck module by adjusting the 4 leveling thumbscrews, as necessary, while monitoring the 2 levels provided on the base. There is a leveling screw at each corner of the module base. Three of the screws and the left- to-right level are shown in the image of the module without the cover above. The front-to-back level is on the left side of the base plate, between the cable and hose connector panels. See image below.



• Remove the On-Deck module from the liquid handler and reinstall the outer cover.



- **2.2.5** Complete installation of the On-Deck module. Because of limited clearance in some liquid handlers, it is easier to connect hoses and cables prior to placing the On-Deck module in the liquid handler.
  - Connect the fittings of the "A", "B", and "DRAIN" hoses to the On-Deck module. The other end of the hoses can be connected after the On-Deck module is in place on the liquid handler. Refer to *Section 2.2.7* below.



- Connect one end of the male 25-pin d-sub cable to the female 25-pin connector on the On-Deck module.
- Connect one end of the male 44-pin d-sub cable to the female 44-pin connector on the On-Deck module.
- Connect the right-angle connector of the coax cable to the BNC connector on the On-Deck module.

**Note:** optional cables with right-angle fittings at one end are available for installations with restricted clearance behind the On-Deck module. Refer to images below.



• Place the On-Deck module with cables and hoses attached into the mounting plate on the liquid handler.



2.2.6 Arrange the eBox and WCS 3 as shown. These may be stacked on the floor or an adjacent table. The eBox mounts on top of the WCS 3 as shown below. Use the two M4 x 25 button head cap screws that come in the accessories box to secure the eBox to the WCS 3.



CAUTION: When positioning the system, please keep fan and panel vents free of obstructions that may block the flow of air.

## 2.2.7 Complete the water connections.

There are two hose kits included with the R230. The WCS Tubing Kit is used to connect the On-Deck to the WCS and to the Waste reservoir. The Reservoir Kit includes the Waste and Supply carboys and the two hoses which connect the WCS to the carboys. From the WCS Tubing Kit:

- Connect one end of the "A" hose to the WCS "A" port. The "A" hose has a yellow tag at each end.
- Connect one end of the "B" hose to the WCS "B" port. The "B" hose has smaller fittings than the "A" hose.

2.2.8



• Connect the straight end of the remaining, clear "SPILL WAY DRAIN" hose to the port on the side of the waste reservoir cap. Refer to the next three images.



• Connect the right angle end of the clear hose to the "SPILL WAY DRAIN" port of the On-Deck.



Waste Carboy with Hose Connections





From the Reservoir Kit:

- Connect one hose from the WCS "Waste" port to the top port of the waste carboy's cap.
- Connect the remaining hose from WCS "Supply" port to the supply reservoir cap, or another source of distilled or deionized laboratory water.

The WCS contains a UV sterilizer and particulate filter to retard biological growth.

- **2.2.9** Complete the electrical connections from the On-Deck Module:
  - Connect the eBox female 25-pin connector, labeled "OD25", to the On-Deck female 25-pin connector.
  - Connect the eBox female 44-pin connector, labeled "OD44", to the On-Deck female 44-pin connector.
  - Connect the eBox BNC connector to On-Deck BNC connector.



2.2.10 Complete the electrical connections from the WCS 3.



- Connect the eBox male 25-pin connector, labeled "WCS25", to the WCS male 25-pin connector.
- Connect the eBox male 15-pin connector, labeled "WCS15", to the WCS male 15-pin connector.
- Plug the WCS 3 into a power source using one of the line cords. The WCS 3 is a "universal input" device and automatically employs any worldwide line voltage or frequency (100 to 240 volts AC, 50 or 60 Hz). Leave power switched OFF at this time.



- **2.2.11** Complete electrical connections from the eBox and laptop computer.
  - Using the enclosed eithernet cable, connect either of the two eBox ethernet ports to the ethernet port on the laptop computer. The R230 uses 4 fixed addresses (5, 7, 10, and 22) on the 192.168.0 subnet.
     Note: When the R230 includes the optional Downholder, there is only one ethernet port on the ebox.
  - Connect the computer to a power source. Connect the mouse or other accessories to the computer.
  - Plug the eBox into a power source using one of the line cords. The eBox is a "universal input" device and automatically employs any worldwide line voltage or frequency (100 to 240 volts AC, 50 or 60 Hz). Leave the instrument power and notebook power switched OFF at this time.





## 3. Getting Started

## 3.1 Start the System

- Power on the eBox using the power switch located on the rear, right side of the eBox.
  - This provides power to the On-Deck module.
- Power on the WCS 3
  - SonoLab software will perform all temperature control.

## 3.2 Start SonoLab

Wait 30 seconds after powering on the WCS 3, to allow the WCS 3 to initialize. Locate the Sonolab 10 icon on the laptop's Desktop.



Double click on the icon. SonoLab will load and the user is prompted to home the R230 motion system.

?	Home Instrument?
The instrument ne	eeds to be homed in order to run methods.
Press YES to home Press NO to conti	e. nue without homing.
l	Yes No

Click "**Yes**", then "**OK**" when prompted to empty the sample holder and the On-Deck module will go through a motion homing process. When homing is complete, SonoLab will either fill the water bath to the Load water level of the current method (if any), turn on temperature control of the water bath, or the user will be asked if the bath should be filled. The action performed is controlled by the Startup Water Control switch on the Admin screen General tab. See **Section 5.5.4**, The General Tab, for details.





The *Method* screen displays the last successful treatment method run and provides access to status information and basic instrument control functions. The green Run button indicates the R230 is ready to run the selected treatment method. See image below.

Cov	aris S	SonoL	ab 10.0 for R2.	30		Logged in a	as: Administrator		EMU	LATION MOD	E 7/13/	2023 1:49 F	, <mark>–</mark> 🗵 >
1	MET	HOD	SCHEDULE	HISTORY	ADMIN	ABOUT							Log Out
N	<b>/lethc</b> Plate:	od	175 bp R230 96 R230_520291 96 Al	AFA-TUBE TPX FA-TUBE TPX Plat	Plate - 1 colu te +0.5 offset	ımn		v	Edit	New		R	un
	Tempe State:	erature:	10.4 (°C) Setpoir Idle	nt: <b>10.0</b> (°C)					Deg	Power: 0.0 (watts) as Time: 00:00:00		٨٩	<b>t</b>
S	teps							R	tun Time: <b>00</b>	04:32 / 00:04:32		Ab	ort
	Proces	ss peat					Current Column:	Columns: 1 Cur	Dithering: Y	=3.0, Speed=20.0 Iterations: 13	$\oslash$	Syste	m Status 📑
		Treatm	ent				Duration: 10.	0 PIP: 280.0	DF: 25 C	PB: 50 AIP: 70.0	$\dot{\leftarrow}^{\uparrow}_{\downarrow}$	Motic	on Control
		Delay								Duration: 10.0	6	Wate	r Control
											錢	Meth	od Options
												È.	On Off
													On Off
												Creatio	n Notes

Click the green Run button to begin treatment method. When the treatment is complete, the Method Ended pane is displayed. If the treatment method is paused for any reason, and then resumed, an informational message is presented in the pane to inform the user that the pause occurred.

i Method Ended	
State: Completed	Notification of Method Pause During Treatment Run
The method was paused during the run	
Time: 11/16/2021 1:54:43 PM	
ок	



### SonoLab Method Screen

The first time SonoLab is started, remove the desiccant bag from the On-Deck module and fill the On-Deck bath with deionized water:

• From the Method screen, click on the Motion Control button at the right side of the screen.



The Motion Control panel is displayed.

Home	Load	Start	Open Cover
RFID	Service	STOP	Close Cover

- Click the Open Cover button. The On-Deck safety covers will open.
- Remove or cut the plastic wrap securing the desiccant bag to the On-Deck sample tray and remove the desiccant bag.
- With the desiccant bag removed, manually pour approximately 1 liter, or a bit more, of deionized water into the water bath to prevent flow errors when the WCS 3 is powered on for the first time. It is only necessary to do this the first time the R230 is started, when the water system is completely empty.
- Click the Close Cover button in the Motion Control panel.

The R230 is now ready to use. The automation computer can connect and assume control of the R230, or treatment methods may be run directly from the Method screen.

See Section 5 below for a more detailed description of SonoLab.

## 3.3 System Shutdown

If the instrument will be idle for an extended period of time, refer to the system shutdown procedure in the User Maintenance section of this manual.



## 4. AFA Operation

AFA uses four parameters to fine tune specific acoustic treatments. These parameters are programmed into **Methods**, provided by Covaris. **Methods** can be executed manually or by automation software, including specifying the treatment to be performed and whether a full or only part of a plate is to be treated. A brief explanation of the treatment parameters is provided below.

## Peak Incident Power (PIP)

This parameter measures the peak electrical power used to drive the ultrasound transducer. In the R230, the range of Peak Incident Power is 1.5 to 450 Watts.

## **Duty Factor**

This parameter defines the ratio between transducer on-time and total operation time. Duty Factor 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 2 to 50% in the R230.

The product of the Peak Power and Duty Factor gives the approximate Average Incident Power (AIP) generated from the 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.

## Cycles per Burst

This parameter indicates the number of electrical signals used in each acoustic "burst" of the transducer. Cycles per Burst (cycles/ burst) range from 50 to 1000 in the R230 for different applications.

## Duration

This parameter defines the amount of time for which the sample is under treatment while in indexed operation, i.e. treating one column at a time. The range of Duration is from 1.0 to 3600.0 seconds. For scanning treatments, the sample tray is passed continuously over the transducer. In this mode, treatment time is a function of scanning speed. Scan speed ranges from 0.1 to 50 millimeters/second in the R230.

## **Treatment Records**

SonoLab History contains a record of parameter information for each treatment method run. An RFID scan is performed on each treated sample tray to ensure that the correct consumables are being used for the treatment and to provide information for LIMS. Prior to SonoLab version 10.0.2, a strip consumable may only be RFID scanned two times, a plate consumable six times.

Beginning with SonoLab release 10.0.2, by default, each *column* can be treated twice for plates that are RFID scanned, either with the built-in or a handheld RFID scanner.

Within a single method run, the usage count for a column is only incremented by one, regardless of how many times the column is treated in the method.

The count is only incremented if the column is actually treated during the method run. The count is not incremented if the column is not included in the "Columns to Treat" list, or if the method is aborted before the first time the column is treated.



## 5. SonoLab Software

## 5.1 The Method Screen

When SonoLab is launched, the **Method** screen is displayed with the last successfully run treatment method.

Covaris	SonoL	ab 10.0 for R23	30		Logged in	as: Administrator		EMULA		E 7/13/202	13 1:49 PM – (	× X
М	ETHOD	SCHEDULE	HISTORY	ADMIN	ABOUT						Log	g Out
Me Pla	thod	175 bp R230 96 A	AFA-TUBE TPX	Plate - 1 colu te +0.5 offset	ımn		v	Edit	New		Run	
Ter Sta	mperature: ite:	10.4 (°C) Setpoin Idle	t: <b>10.0</b> (°C)					Pow Degas 1	ver: 0.0 (watts) Time: 00:00:00		About	
Ste	ps						R	un Time: 00:04:	32 / 00:04:32		Abort	
Pro	Repeat					Current Column:	Columns: 1 Cur	Dithering: Y=3.0	, Speed=20.0 Iterations: 13	$\bigcirc$	System Status	-
	Treatm	ent				Duration: 10.0	PIP: 280.0	DF: 25 CPB: 5	50 AIP: 70.0	¢ <sup>↑</sup> →	Motion Contro	Ы
	Delay							l	Duration: <b>10.0</b>	6	Water Control	
										鐐	Method Optio	ns
										-,	On Off	
											On Off	
										C	reation Notes	

- Method displays the current selected treatment method and its status.
  - Plate: the name of the sample plate specified by the current selected method.
  - Temperature: the actual temperature of the water in the On-Deck bath.
  - Setpoint: the On-Deck bath temperature specified by the current selected method.
  - State: possible states, depending on what the R230 system is doing, includes:
    - ° At initial startup
      - > Idle: waiting for a method launch
    - ° During a treatment method, the possible states include:
      - > **RFID Scanning:** scanning and validating the RFID tag on the current treated sample plate.
      - > Waiting for Water Level: waiting for the water to reach the level specified by the selected method.
      - > Waiting for Water Temperature: waiting for the water temperature of the water bath to be within the range specified by the selected method.
      - > Waiting for Water Degas: waiting for sufficient elapsed time for effective degassing to occur.
      - > **Running:** currently performing the selected treatment method steps.
      - > **Paused:** method is paused by the user or the API.
    - Following a treatment:
      - > Completed: the treatment terminated normally.
      - > Aborted: the treatment was halted by a command from a user or the API.
      - > Failed: the method run was unsuccessfully completed with the displayed reason.
  - **Run Time:** an estimate of the total time to run a method, including treatment duration (acoustic on), delays (acoustics off), and motion. It does not include time waiting for degas, water temperature, or water level.
  - Method: methods saved on the system are selected from the drop-down list.



METHOD	SCHEDULE HISTORY ABOUT
Method	500 base pair -
Plate: Temperature: State:	<<< no method >>> 350 bp 500 base pair Pop Pulsing
Steps	Scan_Full_Plate

• The power delivered during a treatment is displayed just below the 'Edit' and 'New' buttons at the right of the method bar. Remaining time required for degassing is displayed immediately below the power display.



• Additional system control functions are accessed via buttons at the right side of the screen.



System Status displays the System Status panel, which shows whether or not the instrument conditions required to run a treatment method are met. A red X (x) indicates a parameter has not been met, a green checkmark (✓) indicates it has. In the cases of Water Degassed and Temperature within limits, a yellow triangle (▲) indicates that degassing time or temperature targets have not yet been met.

Acoustics Connected:	$\checkmark$
Motion Connected:	~
WCS Connected:	~
Homed:	~
Water Degassed:	~
Temperature within limits:	~
UV Light:	~
Method Valid:	~
API Connected:	No

- Acoustics Connected: a green checkmark ( ✓ ) indicates the eBox has successfully connected to the software.



- Motion Connected: a green checkmark ( </ ) indicates the motion controller board is detected.
- WCS Connected: a green checkmark ( ✓ ) indicates that the chiller contained in the WCS has been recognized by the software.
- Homed: a green checkmark ( 🗸 ) indicates that the On-Deck motion system has been initialized.
- Water Degassed: a green checkmark ( $\checkmark$ ) indicates the water has been degassing for a sufficient length while a yellow triangle ( $\land$ ) indicates that the water has not yet degassed for a sufficient time. Overridden is displayed next to the triangle or check if the degas timer has been overridden in Method Options.
- Temperature within limits: a green checkmark (✓) indicates the water is within the temperature range specified by the currently selected method. A yellow triangle (▲) indicates that the temperature has not reached the method's specified temperature range. Overridden" is displayed next to the triangle or you can check to see if the temperature has been overridden in Method Options.
- UV Light: a green checkmark ( ) indicates that the ultraviolet water sterilizer is on and functioning normally. If the UV sterilizer is not functioning normally, a yellow triangle ( ) is displayed. In addition, a message is displayed informing the user of the issue and stating the instrument can be used, though Covaris service should be notified.
- **API Connected:** "No" is the default condition, indicating that the R230 is not under external automation control. "Yes" indicates that the R230 is under external control, but most functionality continues to be available, unless the external control is executing a treatment method. In this case, a method may not be started, and Motion and Water controls are locked out.
- Motion Control displays the Motion Control panel, which contains buttons for opening and closing the On-Deck safety covers and moving the On-Deck sample tray to predefined positions.



- Home: initializes the motion system and moves the sample tray to its initialized position.
- *Load*: moves the sample tray to the right end of the bath and opens the safety cover to provide access by the liquid handler.
- Start: moves the sample tray to the first treatment position as specified by the current selected method.
- RFID: moves the sample tray to the position in which a plate's RFID tag is read by the RFID reader.
- *Service:* moves the sample tray to the right end of the bath and raises it to its maximum height to provide access for servicing the bath.
- Open Cover: opens the safety cover.
- Close Cover: closes the safety cover.
- Stop: stops all current motion. SonoLab shows the STOP button as disabled when no motion is occurring.



• Water Control displays the Water Control panel, which contains both water system status indicator and user control functions.

Level (mm): Temperature (°C):	Current: <b>10.4</b> Current: <b>20.4</b>	Load: 20.0 Ru Setpoint: 20.0	in: <b>21.0</b> Range: <b>17.0</b> to <b>23</b> .	.0
Degas Time:	00:00:00			
Flow State:	Circulating			
Fill to Load	Fill	Drain	Circulate	Off

### - Water System Indicators

### ° Level

- > Current Level indicates the current bath water level. When acoustics are on, the level displays changes to "-".
- > Load Level is the level to which the method initially fills the bath with the sample tray out of the water bath.
- > Run Level is the level to which the method fills the bath with the sample tray in the bath, immediately prior to the method being run.

### ° Temperature

- > Current Temperature is the current temperature of the water bath.
- > Setpoint Temperature is the water bath temperature specified in the currently selected method.
- Degas Time: the remaining degassing time before a method can be run (unless the degas timer is overridden).
- $^{\circ}$  Flow State
  - > Circulating: the bath water is circulating between the bath and the WCS 3 for temperature control, degassing, and cleaning.
  - > Leveling: filling from the supply reservoir, either to the maximum bath level, or the level specified by the currently selected method, depending on whether the filling was invoked via the Fill button, the Fill to Load button, or directly by the selected method.
  - > Draining: the water bath is draining to the waste reservoir.
  - > **Off:** the pump is off. Degassing and temperature control are disabled.

### - Controls

- ° Off: turns off the circulation pump and control valves.
- Fill to Load: fills the bath to the level specified by the currently selected method.
- ° Fill: fills the bath to its maximum level.
- ° Drain: drains the bath.
- *Circulate:* circulates the water between the bath and the WCS 3.



• Method Options displays the Method Options panel, which contains controls for overriding degassing or temperature parameters, enabling Consumable-based Method Selection and Positions to Treat.

Method Options	
Override Temperature:	On Off
Override Degas:	On Off
Move to Load Position at end of run:	On Off
Consumable-based Method Selection:	On Off
Positions to Treat:	
Close	

- **Override Temp:** when On, the method will run regardless of the bath water temperature; when Off, the bath water temperature must be within the temperature range specified by the method.
- **Override Degas:** when On, the method can be run regardless of the degas timer state; when Off, the system will degas for approximately 20 minutes from startup, depending the water level specified by the current method. After initial startup, if more than 5 millimeters of water are added to the bath, the degas timer is set to 1 minute per millimeter.
- *Move to Load Position at end of run:* when On, the sample tray moves to the Load position following a successfully completed method; when Off, the sample tray remains at the last position treated.
- **Consumable-based Method Selection:** when On, enables Consumable-based Method Selection. See **Section 7.1** for details and instructions.
- **Positions to Treat:** a list of columns to treat when only a subset of a method's columns are to be treated. See **Section 7.2** for details and instructions.
- Other Controls:
  - Light is a control and an indicator. Clicking on the switch toggles the light and light status indicator on and off.



- Bar Code: when on, bar code scanning is enabled. See the Advanced Topics section of this manual for additional information.



- Hand-held RFID: when on, Hand-held RFID scanning is enabled. See the Advanced Topics section of this manual for additional information.





### 5.2. The Schedule Screen

The Schedule screen is used to configure bath Periodic Fill and scheduling daily Ready and Off states. In the Ready state, the motion system is homed, the water bath is degassed, the water bath temperature is at the temperature specified in the current selected method, the sample tray moves to the Load position, and the water level is adjusted to the Load level.

In the Off state, Periodic Fill and the WCS temperature control are turned off, but the WCS circulation pump is on to circulate the water through the filter and UV chamber of the WCS, preserving water cleanliness. The selected method is set to none.

Periodic Fill: Off On Periodic Fill Interval: minutes 15 Schedule: 0. Off Off Temperature (°C) Day Ready Method Sunday ------------Monday ---------Tuesday - -- -- ----Wednesday ------------Thursday - -- -- -Friday -----------Saturday ---Select All Select Weekdays Unselect All

The operator must be logged in as Administrator to adjust the parameters on this screen.

## • Periodic Fill

- SonoLab checks and adjusts the R230 water bath level at the start of each treatment method. However, if the R230 will be idle for long periods of time during the day, the Periodic Fill function can be configured to check the bath level periodically while the system is idle. If necessary, the water level will be corrected, degassed, and the temperature adjusted so that the system will be ready when needed. Beginning with SonoLab version 10.0.2, to prevent the sample tray from being out of position for automated robotic sample loading, the sample tray will not move during a periodic fill.
- Click the Periodic Fill On/Off switch so that On is highlighted dark blue.
   Enter frequency desired for checking the water level. The default value is every 15 minutes.
- Schedule
  - Toggle the 'Schedule' button On to create or edit schedules. To configure a specific day, for instance Sunday, click in the Sunday line of the chart, then click the Edit button, or just double-click in the Sunday line of the chart, to display the Schedule Settings screen.

# **Covaris**®

		Days: Monday	
READY:	On Off		
	Ready Time:	8:00 AM 🗸	
	Method:		$\sim$
	Enable Periodic Fill:	On Off	
OFF:	On Off		
	Off Time:	6:00 PM 🗸	

There are two sections of the Schedule Settings screen, READY and OFF. READY is for configuring a time for the system to be ready to run, typically ahead of a daily plant startup in the morning; OFF is for placing the system in an idle state, typically at the end of the day.

## • Ready

- Click on the on/off switch until "On" is highlighted in dark blue.
- Click on the Ready Time drop down list and select the time the R230 will be ready to run treatment methods.
- Click on the Method drop-down list and select the method to be run.
- Water level will be set at the beginning of each method run. But, if the R230 may be idle for long periods of time, click the Enable Periodic Fill on/off switch so that "On" is highlighted in dark blue. SonoLab will automatically maintain the correct water level, degassing, and temperature parameters. Beginning with SonoLab version 10.0.2, to prevent the sample tray from being out of position for automated robotic sample loading, the sample tray will not move during a periodic fill.

## • OFF

- Click on the OFF section on/off switch so that "On" is highlighted dark blue.
- Select the time for the system to switch to standby (6:00 p.m. in this example) from the drop-down list.
- Leave the Schedule On/Off switch "On", highlighted dark blue, to activate the scheduler.

			Periodic Fill: Periodic Fill Interval:	on or 15 minutes
			Schedule:	On Of
Day	Ready	Off	Temperature (°C)	Method
Sunday	7:30 AM	6:00 PM	20.0	Scan_Full_Plate
Monday	833			
Tuesday				
Wednesday	844			342
Thursday				
Friday				- 25
Saturday	244	**		



Click *Select All*, and then *Edit* if all seven days are to use the same scheduling parameters. Click *Select* Weekdays then *Edit* if only the five weekdays are to use the same scheduling parameters.

**NOTE**: If a method is running at the time the scheduler is set to switch the R230 to the OFF state, SonoLab ignores the scheduler and the R230 will not be switched to standby on that day.

## 5.3 The History Screen

The History screen provides graphic and textual display of information for every treatment method run on the R230 system.

Covaris SonoLab 10.0 for I	R230 Logge	ed in as: Administrator	
METHOD SCHEDULE	HISTORY ADMI	N ABOUT	
	Go To:	25 v June 2020 v	Previous 100 Next 100
Method	Run Date	Run By	Status
350_bp_uTUBE-50	6/25/2020 2:44:36 PM		Completed
350_bp_uTUBE-50	6/25/2020 2:44:12 PM		Aborted
350_bp_uTUBE-50	6/25/2020 2:43:04 PM		Aborted
350_bp_uTUBE-50	6/25/2020 2:39:05 PM		Aborted
350_bp_uTUBE-50	6/25/2020 2:37:19 PM		Aborted
350_bp_uTUBE-50	6/25/2020 2:36:52 PM		Aborted
350_bp_uTUBE-50	6/25/2020 2:36:29 PM		Aborted
350_bp_uTUBE-50	6/25/2020 2:35:55 PM		Aborted
350_bp_uTUBE-50	6/25/2020 2:35:20 PM		Aborted
Prompt_example	3/12/2020 11:47:17 AM	Admin Admin (Administrator)	Completed
Logged_In_Test	3/11/2020 12:22:48 PM		Completed
Logged_In_Test	3/11/2020 12:19:43 PM	Admin Admin (Administrator)	Completed
No_login_Test	3/11/2020 12:18:41 PM	0	Completed
Repeat Example 2	3/10/2020 2:54:25 PM		Aborted
GSK E220 X pattern	3/10/2020 10:10:02 AM		Completed
GSK E220 X pattern	3/9/2020 2:01:50 PM	Admin Admin (Administrator)	Completed
GSK E220 X pattern	1/21/2020 1:25:13 PM		Aborted
300_bp_96_microTUBE_Plate	1/6/2020 2:57:14 PM		Aborted

When SonoLab is started, the most recent 100 results are loaded and displayed in the scrolling list, showing the name of the method with the date and time the method ran.

The status bar's Next 100 button loads the next 100 less recent files. The Previous 100 button loads the next 100 more recent files.

The Go To control allows the operator to select files run on a specific date. The files on the specified date will be displayed at the top of the list.



Double-click on one of the listed treatments to see details of the treatment run.



**History** files are found at C:\Users\Public\Public Documents\Covaris\SonoLab 10\History. Each treatment method's history data are contained in a folder whose name is the same as the method with the date and time of the run appended. For example, the folder for a method named "300\_bp" that was run on June 1, 2019 at exactly 10:30AM is named "300\_bp\_06\_01\_2019-10-30-00".

The history of a treatment consists of three or four files found in its folder, the Measurements, Summary, Treatments, and, if used, RFID files.

*Measurements* is a .csv file which can be opened as a spreadsheet, containing information about the methods run, including times, columns treated, acoustic power, bath water temperature and bath water level.

The *Treatments* file is also a .csv file containing treatment parameters and start and end times for the method, along with water level and temperature data. The Treatments file contains the same data as that sent to the LIMS.

The **Summary** is a .xmL file containing textual information which is shown along with the graphics in the History display.

The **RFID** file, with a time-date stamp appended to its name, present if RFID is enabled, is a .csv file containing the RFID information from the consumables used.



## 5.4 The About Screen

The About screen contains information about components contained in the R230 system, such as serial numbers, software, and firmware versions. The information on this screen can be provided to Covaris support personnel assisting with problem resolution.

SonoLab 10 Version:	SonoLab 10.0.2.2049
Instrument Type:	R230
Serial Number:	RevA3
On-Deck Serial Number:	RevA3
eBox Serial Number:	RevA3
WCS Type:	3.1
WCS Serial Number:	RevA3
Controller Board Serial Number:	J04230379
Controller Board Version:	290042-C
Controller Firmware Version:	13035
Transducer Serial Number:	L-472
Transducer Center Frequency (kHz):	500
Transducer Impedance:	42
Acoustic Efficiency:	0.00
Motion Firmware Version:	VGEN80
API IP Address:	127.0.0.1, Port: 8100
Controller Board IP Address:	192.168.0.22
Motion IP Address:	192.168.0.5
RFID IP Address:	192.168.0.7
Open Source Licenses:	Show Licenses

## 5.5 The Admin Screen

The ADMIN tab is only available if an administrative user is logged in. The ADMIN tab is used for managing users, their permission levels, and access to treatment methods. It is also used to set system-wide parameters.

### 5.5.1. User Authentication

SonoLab's default configuration requires no login to create, run, and edit methods. User Authentication may be enabled to limit access to SonoLab functionality. To enable User Authentication:

- Start SonoLab.
- Click the Log In button at the upper right corner of the SonoLab screen.



• The Login pane is displayed.

Covaris	
Welcome to SonoLab 10	
Username	
Password	
Login Cancel	
Set Password	

- Enter the username "Administrator" and password "admin". The password is case sensitive.
- Click *Login* and the ADMIN tab is added to the SonoLab window.

Covarias Sonol	ab 10.0 for R230	Logged in as: Administrator
METHOD	SCHEDULE HISTORY	ADMIN ABOUT
Method	500 base pair	
Plate: Temperature: State:	R230_96_AFA-TUBE_PLATE 20.4 (°C) Setpoint: 10.0 (°C) Idle	
Steps		
Process		Current Colun
Treatment		Duration: 10.0

• Click on the ADMIN tab. The list of default SonoLab users, their roles, and assigned methods is displayed, along with the "USERS" and "GENERAL" tabs at the upper left.

метнор SCH	EDULE HISTORY	ADMIN ABOUT		
USERS GENERAL				
Username	First Name	Last Name	Role	Methods
admin	Bruce	P	Administrator	All
Administrator	Admin	Admin	Administrator	All
Editor	Editor	Editor	Editor	All
Operator	Operator	Operator	Operator	All



• Click on the "GENERAL" tab.

Require Login:	On Off
Reminder to Archive:	$\odot~$ Never $\odot~$ Every 30 days $\odot~$ Every 60 days $@~$ Every 90 days
Startup Water Behavior:	$\odot~$ Control Temperature $\ {\ensuremath{\overline{\bullet}}}$ Ask user $\ {\ensuremath{\overline{\circ}}}$ Fill upon startup
Handheld RFID:	On Off
API IP Address:	192.168.22.106, Port: 8100
Load position:	0.0, -5.0, 35.0 Edit Restore Default
	Require Login: Reminder to Archive: Startup Water Behavior: Handheld RFID: API IP Address: Load position:

Click the On/Off switch to turn on "Require Login". If "Require Login" is not on, the Default user is active when SonoLab starts and no other user has logged in. An administrator can adjust the role of the Default user to allow this user the permissions desired. By default, the Default user has Editor permissions, access to all methods, and Optional Consumable-based Method Selection control. See *Section 5.5.2* below for details on users and their roles. See *Section 5.5.4* below for a complete discussion of the General tab.

### 5.5.2 Users and Roles

There are three permission levels within the user authentication, Administrator, Editor, and Operator.

#### An Operator:

- Can run methods assigned to them by an administrator.
- Can view History screens for all method runs.
- Can view Schedule screen.
- Can view the About screen

#### An Editor:

Can do everything an operator can do plus:

- Can run and edit methods assigned to them by an administrator.
- Can create methods and run the methods they create.

#### An Administrator:

Can do everything an Editor can do plus:

- Can create and edit methods.
- Can change settings on the Schedule screen.
- Can create and edit users, including other Administrator level users.
- Can assign access to specific methods to Editors and Operators.
- Can change settings on the Admin / General screen

#### 5.5.3 Managing Users

To create a new user:

- Log into SonoLab as Administrator and click on the ADMIN tab.
- Click the **New** button at the upper right side of the screen. The New User pane is displayed.

Properties		
rioperties	Username	Role
	First Name	Operator *
Methods		
<ul> <li>Assigned</li> </ul>	Available	Assigned
175 bp R 175 bp Ti 350 bp Ti	230 96 AFA-TUBE TPX Plate - 1 column me course R230 96 AFA-TUBE TPX Plate me course R230 96 AFA-TUBE TPX Plate	♪
Consumable	e-based Method Selection: OAlw	ays On O Always Off I Optional

• Enter a new Username, the user's first and last names.

	New User
Proper	ties Username Role Fuzzy1 Operator First Name Last Name Generic Operator
Metho ○ Al ● A:	ds I ssigned Available Assigned
1 1 3	75 bp R230 96 AFA-TUBE TPX Plate - 1 column 75 bp Time course R230 96 AFA-TUBE TPX Plate 50 bp Time course R230 96 AFA-TUBE TPX Plate
Con	sumable-based Method Selection: O Always On O Always Off O Optional

By default, SonoLab assigns all available methods to a new user. If the Administrator only wants some methods assigned:

- Click on the Assigned button as shown in the image above.
- Click on the method(s) to be assigned in the list at left. Multiple methods may be selected by clicking "Shift" or "Control" simultaneously.
- Click the *right arrow* button. The methods to be assigned are moved to the box at the right. See the image below.

roperties					
	Username	Role			
	Fuzzy1	Ope	rator <sup>v</sup>		
	First Name		Last Name		
	Generic	Ope	rator		
/lethods ○ All					
<ul> <li>Assigned</li> </ul>					
	Available		Assigned		
175 bp Ti	me course R230 96 AFA-TUBE TPX Plate	⇔	175 bp R230 96 AFA-TUBE TPX Plate - 1 column 350 bp Time course R230 96 AFA-TUBE TPX Plate		
		<del>\</del>			
	-based Method Selection: O Alw	avs On	○ Always Off		

• Click Save to save changes and exit the New User screen. The new user now appears in the list of system users.

METHOD SCH	IEDULE HISTORY	ADMIN ABOUT	r.		
USERS GENERAL					
Username	First Name	Last Name	Role	Methods	
admin	Bruce	Р	Administrator	All	
Administrator	Admin	Admin	Administrator	All	
Editor	Editor	Editor	Editor	All	
Fuzzy1	Generic	Operator	Operator	3	1
Operator	Operator	Operator	Operator	All	

The first time a new user is logs in, the user must click on "Set Password" to display the Set Password pane.

Covaris	Set Password
Welcome to SonoLab 10	Username
Username	New Password
Password	Confirm Password
Login Cancel	
Set Password	Set Password Cance



The user enters their Username, new password, and then must enter it again to verify correct spelling. Clicking "OK" returns the user to the login screen where the user may now enter their new password and log in.

### 5.5.4 The General Tab

METHOD SCH	EDULE HISTORY	ADMIN ABOUT	r	
USERS GENERAL				
Username	First Name	Last Name	Role	Methods
admin	Bruce	P	Administrator	All
Administrator	Admin	Admin	Administrator	All
Editor	Editor	Editor	Editor	All
Operator	Operator	Operator	Operator	All

• Click on the "GENERAL" tab.

METH	OD	SCHE	DULE	HISTORY	ADMIN	ABO	UT		
USERS	GENE	RAL							
				Req	uire Login:		On Off		
				Rem	inder to Archive	e:	○ Never ○ Every 30 days	O Every 60 day	ys 💿 Every 90 days
				Star	tup Water Beha	vior:	○ Control Temperature ●	Ask user $\bigcirc$ Fil	l upon startup
				Han	dheld RFID:		On Off		
				API	IP Address:		192.168.22.106, Port: 8100	Edit	
				Load	d position:		0.0, -5.0, 35.0	Edit	Restore Default

• Require Login: toggle the On/Off switch to turn "Require Login" On or Off. If "Require Login" is not on, the Default user is active when SonoLab starts and no other user has logged in. An administrator can adjust the role of the Default user to allow this user the permissions desired. By default, the Default user has Editor permissions, access to all methods, and Optional Consumable-based Method Selection control. See *Section 5.5.2* below for details on users and their roles.

If "Require Login" is turned on when SonoLab starts, a login screen is displayed immediately following the homing sequence. If the user logs in, they have the SonoLab privileges defined by their predefined role. See *Section 5.5.2* below for details on users and their roles. If the user cancels the login, they have access to minimal SonoLab functionality, for instance, viewing History files or the About tab.

- Reminder to Archive specifies how often a message is displayed, when SonoLab is started, suggesting that History files be archived to preserve disk space.
- Startup Water Behavior controls SonoLab behavior immediately following homing when SonoLab is started. There are three options:
  - Control Temperature adjusts the water bath temperature to that specified by the current method.
  - Fill upon startup fills the water bath to the Load water level of the current method and temperature control is turned on.
  - Ask user presents a message box asking if the bath should be filled. The level and temperature to which the bath will be set is displayed, as shown:

?	Fill to Load
Would you lil	ke to fill the bath to the load water level?
Current temp Load water le	erature setpoint: 10 degrees evel: 21 mm
Please check	that the correct method is selected
	Yes No

- Handheld RFID enables the use of a handheld RFID scanner as described in *Section 7.3*, Handheld Barcode and RFID Scanners.
- API IP Address displays the current IP address and port number of the API server (the SonoLab computer), and an Edit button to modify the IP address. Click the 'Edit' button to open the API IP Address dialog. If the API server is currently connected, the user is instructed to first disconnect the server. The dialog opens initially showing the current IP address and port number. Beginning with SonoLab 10.0.2, the defaults will be 127.0.0.1 and 8100.

Edit API IP Address			
IP Address	192.168.22.106 ~		
Port Number	8100		
Save	Cancel		

Click on the IP Address drop-down to see a list of available IP addresses. In this example, only the original and loopback addresses are available. If the API client is also running on the SonoLab computer, the loopback address should be selected.

Edit API IP Address		
IP Address	192.168.22.106 ~	
Port Number	192.168.22.106	
	127.0.0.1	
Save	Cancel	

The Port number must be an integer between 1 and 65535. The Save button will become active if changes are made and the port number is valid.


• Load Position displays the current X, Y, and Z coordinates for the Load position, 'Edit ' and 'Restore Default' buttons. The Load position may be changed to accommodate a sample handling robot arm.

To adjust the Load position:

- Open the Motion Control panel on the SonoLab Method screen and send the sample tray to the current Load position.
- Return to the Admin General tab and click Load position 'Edit' button. The Edit Load Position dialog displays the current Load position in millimeters from the Home position, in this example, 0.0 for the X axis, -5.0 for the Y axis, and 35.0 for the Z axis.

Edit Load Position											
Load Position	X 0.0	Y -5.0	Z 35.0								
Load coordinate X is a	a number l	petween -117	7.5 and 0.0								
ОК		Cancel									

• Estimate an approximate location from the current Load position the sample tray needs to be. The range of travel for the 3 axes are:

X axis: 0 to -117.5 mm. 0 is at the end of the bath away from the water & electrical connections.

Y axis: 0 to -9.5 mm. 0 is toward right side of the On-Deck when facing the end with the water & electrical connectors.

Z axis: 0 to 50 mm. 0 is as low as the tray will move. All movement is up from 0.

As an example, assume that the Load position must be 10mm toward the end with hoses and cables, 3mm to the left (as viewed from the end of the bath with the cables and hoses), and 5mm lower than relative to the current load position.

The new X position is -10.0. The new Y position is -8. The new Z position is 30.

- o Enter these new positions in X, Y , and Z fields, then click 'OK'.
- Return to the SonoLab 'Method' screen and use the 'Motion Control' panel to Home the motion system, then move to the 'Load Position'. Verify that this new location is acceptable. If not repeat the above steps to adjust it closer to the required position.

The 'Restore Default' button returns the 'Load Position' parameters to the factory default settings.



### 6. Creating & Editing Treatment Methods

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

A method consists of one or more *Process or Scan Steps*. A Process contains at least one Treatment. Most methods will contain a single process, containing a single treatment step.

If more complex methods are required, they can be easily constructed.

### 6.1 Create a New Method Using Templates

SonoLab 10.0.2 introduces predefined templates to assist in creating more complex R230 treatment methods. Available templates are displayed when creating a new method.

1. Click the "New" button on the METHOD screen to display the New Method Template panel.



*Empty*: no predefined operations. Build either a pulsing treatment method as described *Section 6* above or a scanning treatment.

*Multiple Scans*: provides a template to construct a treatment consisting of 1 to 10,000 scans on a contiguous group of from 1 to 12 columns. Refer to **Section 6.4** Using the Repeat Function and **Section 6.5** Scanning Treatments.

*Pulsing for 1 Column:* provides a template to treat and delay one or more columns from 1 to 10,000 times. The treatment and delay is applied to the first specified column the number of times specified, then the next column is treated, and so forth. This is described in more detail below in *Section 6.2*, Creating Methods Manually and *Section 6.4* Using the Repeat Functions.

*Pulsing for Multiple Columns*: provides a template to treat one or more columns from 1 to 10,000 times. The treatment is applied to all selected columns, then repeats on all selected columns for the specified number of repeats. This is described in more detail below in *Section 6.2*, Creating Methods Manually and *Section 6.4* Using the Repeat Functions.

*Time Course – Pulsing for 1 Column:* as described in the panel, 6 columns are treated, each for a different number of repeats. This is useful for determining parameters to use when developing a new treatment protocol.

• Click to select the desired template. For example, clicking "Multiple scans" displays this template:



METH	OD	SCHEDULE	HISTORY	ABOUT								Log	In
Method	l Editor	Ľ											
Method					• New					Sa	ve Cancel	Dele	
Plate					¥			Temp Set	tpoint (	° <b>C)</b> 20.0	Range (°C)	17.0 to a	23.0
Steps								Run ti	me:	ŧ	Selected S	amples C B A	
	lepeat	Iterations	-1					+	ð	×	00000	000	1
	Scar	[Select to edit sa	imples]		Speed (mm/s) 0.0 PIP	0.0 DF 0	CPB 0 AIP	0.0	ð	$\times$			2 3
													4
													5
													6
													8
													9
													10
													11
													12

Enter the desired treatment parameters into the fields outlined in red, adjust the number of columns to be scanned and save the method. Details regarding the specific treatment parameters are described in *Section 6.4* Using the Repeat Function and *Section 6.5* Scanning Treatments.

### 6.2 Create a New Method Manually

1) Click the "New" button on the METHOD screen.

METHOD	SCHEDULE HISTORY ADMIN ABOUT		
Method	175 bp R230 96 AFA-TUBE TPX Plate - 1 column	Edit	New
Plate:	R230_520291 96 AFA-TUBE TPX Plate +0.5 offset		
Temperature:	10.3 (°C) Setpoint: 10.0 (°C)	Powe	er: 0.0 (watts)
State:	Idle	Degas Ti	me: <b>00:00:00</b>
Steps	Run T	Time: 00:04:3	2 / 00:04:32
Process	Current Column: Columns: 1 Dit	hering: <b>Y=3.0</b> ,	Speed=20.0

2) Then click the "Empty" template option to display the Method Editor Window.



- 3) Select the appropriate plate for the consumables used in this method from the "Plate Definition" drop-down list.
- 4) Click the + button. The "Choose a Step Type" pop-up is displayed.

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P	roces	s
f	Repea	t
	Scan	
3	Delay	
F	romp	t

5) Click "Process". The process bar is displayed.

Method	Editor	i												
Method			~	New				s	ave	G	ance			ete
Plate	R230_500723 F	PSU Ho	lder 8 AFA-TUBE TPX Strip +0.5 offset 🗳			Temp Se	tpoint (	° <b>C)</b> 20	.0 F	lange	(°C)	17.0	to	23.0
Steps						Run time: 00	:00:12	÷	н	Sele	ected	Samp	les B A	
E P	rocess	All	[Select to edit samples]			Ð	ð	×	0	00	0 0	0	0 0	1
								_	0		00			2
									0	0 0	0 0	0	0 0	4
									0	0 0		0		5
									0	0 0	0 0	0	0 0	6
									0	0 0	00	0	0 0	7
									0	0 0	00	0	0 0	8
									0	0 0	00	0	0 0	9
									0	0 0	0 0	0	0 0	10
									0	0 0	00	0	0 0	11
									0	00	00	0	0 0	12

For descriptions of **Delay and Prompt steps**, see step 11 of this section below. **Repeat** steps are discussed in **Section 6.4** of this document. **Scanning** is discussed in **Section 6.5**.

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

Repeat
Treatment
Delay
Prompt

7) Choose Treatment to specify the four acoustic parameters: Duration, Peak Incident Power, Duty Factor, and Cycles per Burst.



Method	iditor 🔽	
Method	* New	Save Cancel Delete
Plate	R230_500723 PSU Holder 8 AFA-TUBE TPX Strip +0.5 offset 👋	Temp Setpoint (°C) 20.0 Range (°C) 17.0 to 23.0
Stens		Bun time: 00:00:12 + Selected Samples
oteps		H G F E D C B A
≡ Pr	All [Select to edit samples]	
_		0000002
Ξ	Treatment 0.0 PIP 15.0 DF 20 CPB 20	00 AIP 3.0 🖸 🗵 0000000 3
		00000004
	Duration 10.0 PIP 15.0 DF 20	CPB 200 AIP 3.0

Treatment parameters are chosen by directly editing the value boxes. When a parameter's value box is selected, the valid parameter range is displayed in the blue bar above. If an invalid parameter is entered, the value box is outlined in red and the valid parameter range is displayed in red.

PIP is a number between 1.5 and 450.0	PIP is a number between 1.5 and 450.0
ect to edit samples]	t to edit samples]
Duration (s) 10.0 Peak \$\$5.0 F	Duration (s) 10.0 Peak 515.0 F

### Peak Incident Power (PIP)

This parameter measures the peak electrical power used to drive the ultrasound transducer. The adjustable range of Peak Incident Power is 1.5 to 450 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 specify a Duty Factor from 2% to 50%.

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.

### Cycles per Burst

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

The range of Cycles per Burst (cycles/burst) is from 50 to 1000 for different applications. Advanced users may use this parameter to fine tune their experiments.

### Duration

This parameter defines the amount of time during which the sample is being treated. 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. The maximum allowed Average Incident Power is 110 Watts.

### Dithering

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

Process Step Parameters													
C Dithering													
X +/- (mm):	0.0												
Y +/- (mm):	3.0												
Speed (mm/s):	20.0												
Pause Duration (s):	0.0												
Apply to all process steps:	On Off												
ОК Са	ancel												

Dither Parameter Pane

The X axis +/- distance range is 0 to 5 millimeters, Y axis is +/- 4.5 millimeters. The Speed range is from 0 to 25 mm/second. The Pause Duration range is from 0 to 60 seconds.

- 8) Enter the desired dithering parameters, if any, and click "OK". If dithering is to be applied to all treatment steps of the method, click the 'Apply to all process steps' switch to toggle it on. Click OK.
- 9) Enter the temperature set point for the protocol and the allowable range into the text entry fields. SonoLab allows a minimum range of plus/minus 2 degrees; the default range is plus/minus 3 degrees.



Once the treatment parameters have been defined, click in the process bar to highlight it, then click in the treatment well graphic to define which plate positions will be treated. The R230 treats an entire column at a time. Therefore, clicking anywhere in a column causes the row to be selected. Any number of columns may be selected for treatment; columns selected for treatment turn from white to yellow. Clicking the All button (circled below) selects all rows for treatment. Clicking on a column (or *All* button) a second time removes it from the list to be treated. Multiple columns may be selected by clicking on one column and dragging to adjacent columns. Holding down the shift key while doing so allows multiple groups of columns to be selected.



Method	Editor	ì								
Method			• New			s	ave	Cancel	Del	lete
Plate	R230_500723	PSU Ho	older 8 AFA-TUBE TPX Strip +0.5 offset 💙	Temp Se	tpoint	(°C) 20.	Rai	nge (°C)	17.0 to	23.0
Steps			Ru	un time: 00	:02:33	÷	НG	Selected S	amples	
E P	rocess	All	[Select to edit samples]	+   Ø	ð	×				1
Ξ	Treatment		Duration 10.0 PIP 15.0 DF 20 CPB 200 A	AIP 3.0	Ð	×				2
										4
										6
										7
			All Columns Selected for Treatr	ment			0 0	000	000	9
				nem	·			000		0 10
										12

When the method is run, the column under treatment is displayed in the 'Process' bar.

Process	Current Column: 1	Columns: 1-2,		
Treatment	Duration: 10.0	PIP: 15.0	DF	

### Adding a Delay Step

10) As an example and if desired, choose a Delay step to introduce a fixed time delay into a method to allow the sample to settle between the two different treatment steps. The delay is specified in seconds.

Metho	d Editor	ľ																
Method						v	New									S	Sav	е
Plate	R230_500	723 PSU Ho	older 8 AF	A-TUBE TPX	Strip +0.5 offs	et v							Tem	ıp Setp	point (	(° <b>C)</b> 20	0.0	Rang
Steps												R	un tin	ne: 00:0	)2:45	+	F	Se I G I
Ξ·	Process	All	[Selec	t to edit sampl	es]								+	Ø	Ð	×		
	Treatn	nent				Du (se	econds) 10.0	PIP 15.	0 DF	20	СРВ	200	AIP	3.0	ð	×		
	<b>Delay</b>		(	Duration 1.0														
	Enter Delay in Seconds																	

### Adding a Prompt Step

- 11) If desired, choose a Prompt step to display a message on the screen and pause execution of the method until the operator clicks OK.
  - Clicking 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.

Method	Editor	Ø																					
Method							~	New									s	ave	Can	icel	Dele		
Plate	R230_50	00723 PS	U Hol	der 8 AFA	-TUBE TPX	Strip +	+0.5 offset ×							Tem	p Setp	ooint (	°C) 20	Ra	ange (°C	<b>()</b> 17.0	to	23.0	
Steps													F	lun tin	ne: 00:0	2:45	ŧ	не	Selecte	ed Sam D C	nples B A	-	
E P	rocess		All	[Select	to edit samp	les]								+	Ø	Ð	$\mathbf{X}$					1	
	Trea	tment					Durat (seco	tion nds) 10.0	PIP	15.0 DI	20	СРВ	200	AIP	3.0	ð	$\times$					2	
	Dela	у											Dura (seco	tion nds)	1.0	Ð	×					4	
	Pron	npt		Message: S	Spin Down	Before	Click to text o	creat f the	te oi pror	r edi npt	t	-	_	_(	Ø	ð	×					6 7 8 9 10	
Promp	t		Me	essage	r: Spin	Dow	n Before Pr	ocessi	ng														



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



12) Clicking the Method Parameters button, 🧖, in the orange Method Editor bar accesses optional Method Parameters.

METHOD SCHEDULE	HISTC	
Method Editor	method i unimeters buttori	
Method 175 bp R230 96 AFA-TU	Method Parameters	
Plate R230_520291 96 AFA-TL		
Steps	Creation Notes Edit	
Process All Repeat Itera	Adjust Level During Method On Off Min Water Adjustment Interval (min) Water Adjustment Level Threshold (mm) OK Cancel	20 5 CPB

*Creation Notes*: clicking the Edit button displays a text entry window. Enter descriptive text regarding the method into the text entry field. This may be left blank.

*Adjust Level During Method*: By default, Adjust Level During Method is off. This means that SonoLab checks and, if necessary, adjusts the water level prior to running a treatment method. Once the method starts, there are no more adjustments. Beginning with SonoLab version 10.0.2, there is an option to check and adjust the water level while a treatment method is running. Each time acoustics are turned off, during the method, the water level is checked and, if necessary, adjusted before acoustics are again turned on. This feature is used to prevent a change in water level over the course of a lengthy treatment method.

To use this feature:

- Click the 'Adjust Level During Method' button to 'On'.
- Enter the minimum adjustment level, in minutes. The default interval is 20 minutes. The actual interval may be longer as the adjustment will not take place if acoustics are on at the end of the specified time interval.
- Enter the 'Adjustment Level Threshold' in millimeters. This is the minimum change in water level to trigger an adjustment. The default value is 1 mm.



13) When all method parameters and steps have been defined, click on the *Save* button to save changes. The user is prompted to name the method.

300 Base Pair

14) Enter a name for the treatment method and click Save. If the method name is the same as an existing method, the 'Save button is disabled and the 'Overwrite' button is enabled. Clicking the 'Overwrite' button will result in the previous version being replaced.

### 6.3 Deleting and Archiving Methods

1) Select the method to be deleted or archived, then click 'Edit'.

METHOD	SCHEDULE	HISTORY	ADMIN	MANFIELD	ENGINEERING	ABOUT		
Method	test					~	Edit	New
Plate:	R230_520291 96 AF	A-TUBE TPX Plat	e +0.5 offset -	Hi-Power				

The Method Editor screen is displayed.

2) Click on the 'Delete' button at the upper right.

METH	OD	SCH	EDULI	HISTORY	ADMIN	MANFIELD	ENGINEERING	ABOUT		Log Out
Method	Edito	or [	ź							
Method	test					• New			Save Cancel	Delete
Plate						v			Temp Setpoint (°C) 20.0 Range (°C) 17	.0 to 23.0

The 'Delete Method' panel is displayed.

?	Delete Method	
Press Delete Press Archive Press Cancel	o delete the method file. to move the method file into the method archive director to cancel this operation.	y.
	Delete Archive Cancel	

3) As instructed on the panel, click 'Delete' to remove the method from the system, click 'Archive to move the method to the Archive folder, or click 'Cancel' exit the operation without making any changes.



### 6.4 Using the Repeat Function

The Repeat function is used when one or more treatment, delay, or prompt steps are to be applied to each column of the plate multiple times. There are two scenarios for how this may need to be performed, and the required scenario determines how the repeat function is implemented.

In scenario A, all treatment (or delay or prompt) steps are applied to the first column, the number of times specified in the Repeat bar, then all steps are applied to the next column. This continues until all columns in the plate are treated.

Scenario A is referred to as "pulsing".

#### 6.4.1 Implement Scenario A:

• Create a method with a Process step, as explained previously.

Method	Editor	5													
Method				New						5ave		Cance	н		
Plate	R230_500723	PSU H	older 8 AFA-TUBE TPX Strip +0.5 offset				Temp	Setpoint	(°C)	0.0	Range	(°C)	17.0	to	23.0
Steps							Run time:	00:00:12	+	н	Sel G F	ected E f	Sam	ples B	A 7
E P	rocess	All	[Select to edit samples]			(	Ð	Z D	×	0	00	00	0 0	0	0 1
										0	0 0	00	0 0	ŏ	0 3
															04
										0	0 0	0 0	0 0	0	0 6
										0	00	00		0	078
										0	0 0	00	0 0	0	9
										0	00	00	00	0	0 11
															O 12

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



- Click on *Repeat*.
- Click the + button inside the repeat bar to display the step type menu and create two or more treatment or delay steps. See image below.



Method Editor					
Method	• New		Sa	ve Cancel D	
Plate R230_500723 PSU Holder 8 AFA-TUBE TPX Strip +0	).5 offset ¥	Temp Setpoint	(°C) 20.0	Range (°C) 17.0 to	o 23.0
Steps	ŗ	Run time: 00:02:45	Ŧ	Selected Sample	s
Process All [Select to edit samples]		• C 0	×	0000000	0 1
Repeat Iteration 2		± D	×		<ul><li>2</li><li>3</li></ul>
Treatment	Duration 10.0 PIP 15.0 DF 20 CPB 200	AIP 3.0	×		0 4
Delay	Dura (seci	ation 1.0	×		0 G
					0 7 0 8
	Repeat these steps; This number	of times			0 9
C C			)		0 10

All listed treatment (or delay, etc.) steps will be applied to the first column selected for treatment, the number of times specified in the Repeat bar. Then all will be applied to the second column, and so forth, until the entire plate is treated.

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

In scenario B, all treatment (or delay or prompt) steps are applied to all selected columns of the plate once. This repeats for the number of times specified in the Repeat bar.

Scenario B is referred to as "cycling". With cycling, a column is treated with an acoustic dose, and is then allowed to rest while the remaining columns are treated. Once the remaining columns are treated, a second acoustic dose is applied to each column.

### 6.4.2 Implement Scenario B:

• Create a method as explained above, but select "Repeat" instead of "Process" for the step type.

Method Editor	
Method	New S
Plate R230_500723 PSU Holder 8 AFA-TUBE TPX Strip +0.5 offset V	Temp Setpoint (°C) 20.
Steps	Run time: 00:00:12
Repeat Iterations 2	+ D ×

• Click the + button inside the Repeat bar and select Process.

C	Choose a step type	:
	Process	
	Scan	
	Delay	
	Prompt	
	Cancel	



Method	Editor									
Method				v	Nev	N				s
Plate	R230_5007	23 PSU Ho	older 8	AFA-TUBE TPX Strip +0.5 offset `				Temp	Setpoint	(° <b>C)</b> 20
Steps								Run time	e: 00:00:12	Ŧ
	epeat	Iteration	is 2						Ð	×
	Process	5	All	[Select to edit samples]				Ŧ	r 1	$\times$

• Click the + button inside the Process bar and create one or more treatment steps. Two treatment steps are created in this example.

Method	l Editor	Ø									
Method					• New						Sa
Plate	R230_500	Select Meth	hod <b>ler 8</b>	AFA-TUBE TPX Strip +0.5 offs	et Y				Temp Setp	oint (	° <b>C)</b> 20.
Steps				PIP is a number betw	een 1.5 and 450.0				Run time: 00:0	0:12	÷
	lepeat	Iteratio	ons 2						+	ð	×
	Proces	s	All	[Select to edit samples]					+ 2	Ð	×
	Ξ Tr	eatmen	t		Duration (seconds) 30.0	PIP 15.0	DF 20	СРВ 200	AIP 3.0	ð	×
	ΞTr	eatmen	t		Duration (seconds) 60.0	PIP 25.0	DF 20	СРВ 200	AIP 5.0	ð	×

- All treatment steps will be applied to all selected columns of the plate once and then repeated for the number of times specified in the Repeat bar. That is, both programmed Treatments will be given to column A, then both Treatments will be given to column B, and so on until the entire plate is treated and then the treatment steps are repeated.
- When all method parameters and steps have been defined, click on *Save* to save changes.



### 6.5 Scanning Treatments

In a scanning treatment, rather than the sample tray indexing to place one column of the sample tray at a time over the transducer, the sample tray moves continuously over the transducer. In this mode, treatment time is a function of the speed at which the sample tray moves. One or more columns may be treated, but the columns must be adjacent.

### 6.5.1. Create a Scanning Treatment:

• Click the + button.

METH	OD SCHEDULE	HISTORY	ADMIN	ABOUT	г						
Method	Editor 🛛 🖉										
Method				v	New				Save	9	Can
Plate	R230_500723 PSU Hold	er 8 AFA-TUBE TF	X Strip +0.5 o	ffset ×			Temp	Setpoint (°C)	20.0	Rar	nge (°(
Steps							Run time	: 00:00:12			Select
							Click +	ſ			
									C	0	0 0

• Select Repeat from the step type list. It is not necessary to use a Repeat step if the method will contain only one scan iteration.

(	Choose a step type	:
	Process	
	Repeat	
	Scan	
	Delay	
	Prompt	
	Cancel	

• Enter the desired number of scans in the Iterations field. One scan passes the sample tray over the transducer one time.

Steps		Run time: 00:00:12
E Repeat	Iterations 4	of Scans Click to Add Scan Step

• Click the add process button on the Repeat bar.



- Select Scan from the Step Type list.
- Enter the desired treatment parameters as for a normal treatment step.

Method		• Nev	w							-	Save	Cancel	Dele	
Plate	R230_500723 PSU Holder 8 AFA-TUBE TPX Strip +0.5 offset	v						Temp S	Setpoir	nt (°C) 2	0.0 Ra	nge (°C)	7.0 to	23.0
Steps							I	Run time:	00:01:10	; +	нg	Selected Sa	mples	
	epeat Iterations 4							6	Ð	×				1
	Scan [Select to edit samples] Spe	eed m/s) 10.0	PIP 15.	D DF	20	СРВ 200	AIP	3.0	2 ] 6	PX				2
														4
														6
														8
														9 10
														11

Note that the first parameter is "Speed", not "Duration" as in non-scanning treatments. The total length of a treatment dose in a scan treatment is a function of the scan speed and the number of scans.

• With the Scan step outlined in orange, click and drag on the columns to be treated. Entire or partial plates may be treated. When treating a partial plate, the columns being scanned must be contiguous. SonoLab will not accept a non-contiguous selection. For 384-well plates only contiguous pairs wells may be selected.



	А	B	C	D	Е	F	G	н
1	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0
з								
4								
5								
6								
7								
8								
9	0	0	0	0	0	0	0	Ø
10	0	0	0	0	0	0	0	0
1	0	0	0	0	0	0	0	0
12	0	0	0	0	0	0	0	0

Full Plate Selected for Scanning

• Click the Step Edit button to display additional Scanning step options.

The 'Scan Step Parameters' pane provides controls to enable dithering and Bidirectional scanning.



A standard scan steps treats the sample only as the tray is moved forward over the transducer. Bidirectional scanning increases throughput by applying the acoustic dose as the sample tray moves over the transducer in both directions. Specifying 1 iteration in the Repeat step results in a single, normal scan moving toward the front of the instrument. If more than 1 iteration is specified, odd numbered iterations will move from the rear toward the front, even numbers from front to rear.

Scan Step Parameters
Dithering Y +/- (mm): 0.0 Speed (mm/s): 0.0
Scan Direction Bidirectional: On Off Bidirectional scanning requires that the scan step must be within a repeat step. Odd numbered scans move in the forward direction, while even numbered
Apply to all scan steps: On Off OK Cancel

• Click **Save** and give the method a name as with normal treatment methods.

Contact Covaris Applications Support for assistance in specifying an appropriate scanning speed and treatment parameters.

### 6.6 Using Drag and Drop with Treatment Steps

The order of treatment steps may be rearranged and Treatment steps may be moved in and out of Process steps and Repeat steps to optimize treatment or expedite experimenting with different orders of operations. In the example below, there are initially a single process and three treatment steps as shown.

Steps									Treat	ment ti	me: 00:0	0:00	ŧ
E Pro	cess	All	[Select to edit samples]							Ð		ත	×
Ξ	Treatment	t	Duration (s) 30.0	Peak Power	15.0	Duty Factor	20	Cycles per Burst	200	Avg Pow	er 1.0	ð	×
Ξ	Treatment	E	Duration (s) 60.0	Peak Power	300.0	Duty Factor	20	Cycles per Burst	200	Avg	er 60.0	ð	×
Ξ	Treatment	£	Duration (s) 10.0	Peak Power	100.0	Duty Factor	20	Cycles per Burst	200	Avg Pow	20.0	ð	×
E	,		Drag-and-Drop handles	rower		Prector		PIP valu	<b>es t</b> 30(	op to D. and	bottc	om:	10

To swap the positions of the second and third steps:

- Click and hold on the three bars at the left of the third step. A 'Move Treatment' box will hover over the mouse.
- Drag the 'Move Treatment' box over the first treatment step until a blue line shows between the first and second steps.



• Release the mouse button when the blue line appears. The former third step will be inserted below the first step ahead of the former second step.

Process	A!!	[Select to edit samples]		Move	Treatment Bo
Move Treatr	nent	•	Durat		
Treatment			Durat	Blue Li and	ne between f second steps
<b>Treatment</b>			Durat		
Process	All	[Select to edit samples]			
Process	All	[Select to edit samples]	Duration (s	s) 30.0	Peak Power 15.0
Process  Process  Treatment  Treatment	All	[Select to edit samples]	Duration (s	s) 30.0 s) 10.0	Peak Power 15.0 Peak Power 100.0

To move treatment steps into a repeat step:

- Click the '+' button in the process bar to display the step types list as explained previously.
- Select 'Repeat'. The new Repeat step displays below the three treatment steps.

E Pr	ocess	Select Sample	15								
E	Treatment	í,		Duration (s)	30.0	Peak Power	15.0	Duty Factor	20	Cycles per Burst	200
Ξ	Treatment	l.		Duration (s)	10.0	Peak Power	300.0	Duty Factor	20	Cycles per Burst	200
Ξ	Treatment	ŧ.		Duration (s)	60.0	Peak Power	300.0	Duty Factor	20	Cycles per Burst	200
	Repeat	Iterations	2								

- Click on the three bars at the left of the first Treatment step.
- Drag it over the Repeat step until the blue line shows below the Repeat step.
- Release the mouse button.
- Do the same for the two remaining Treatment steps. The screen is now configured as shown below.



	ocess	All	[Select to edit	samples]						
Ξ	Repeat	Ite	ration 2							
	∃ Treat	ment		Duration (s)	60.0	Peak Power	300.0	Duty Factor	20	C) pr
	∃ Treat	ment		Duration (s)	10.0	Peak Power	100.0	Duty Factor	20	C) pr
	= Treat	ment		Duration (s)	10.0	Peak Power	15.0	Duty	20	9

When finished, specify the wells to be treated and save the method as described in *Section 6.2* above. Behavior of Repeat steps is described in detail in *Section 6.4* above.



### 7. Advanced Topics

### 7.1 Using SonoLab Consumable-based Method Selection

SonoLab can be configured with Consumable-based Method Selection to limit the displayed list of methods to only those using a specified consumable. For individual SonoLab users, Consumable-based Method Selection is set to either Always On, Always Off, or Optional, as described in *Section 5.5.3*, Managing Users. If Consumable-based Method Selection is set to Optional for a user, the user controls whether to enable it via the switch in the Method Options pane, described in *Section 5.1*.

Method Options	
Override Temperature:	On Off
Override Degas:	On Off
Move to Load Position at end of run:	On Off
Consumable-based Method Selection:	On Off
Positions to Treat:	
Close	

If a user's Consumable-based Method Selection option is configured Always On or Always Off, these setting take precedence and the switch in the Method Options pane is disabled. See 'Managing Users' in *Section 5.5.3* for details on configuring users for Consumable-based Method Selection.

Using Consumable-based Method Selection

• Enable Consumable-based Method Selection; the currently selected method will be cleared, and the Method dropdown box will look like this:

<<< no method >>>	v	
-------------------	---	--

• Click the Method dropdown box; there will be only one option:

<<< no method >>>	
<<< select method >>>	

Click on "<<< select method >>>". The system performs an RFID scan of the consumable currently in the sample holder, and then displays a list of methods compatible with that consumable, as shown:

Select Method						
Selected:	175 bp Time course R230 96 AFA-TUBE TPX Pla	ate				
175 bp R	230 96 AFA-TUBE TPX Plate - 1 column					
175 bp R	230 96 AFA-TUBE TPX Plate - more than 1 column					
175 bp T	me course R230 96 AFA-TUBE TPX Plate					
350 bp R	230 96 AFA-TUBE TPX Plate - 1 column					
350 bp R	230 96 AFA-TUBE TPX Plate - more than 1 column					
350 bp T	me course R230 96 AFA-TUBE TPX Plate					
	OK Cancel					



• Select the desired method and then click "OK". The method dropdown now looks like this:

175 bp Time course R230 96 AFA-TUBE TPX Plate

- Click the "Run" button and the method executes normally. This includes performing RFID validation of the consumable currently in the sample holder. If the consumable is replaced with a different consumable which is not compatible with the selected method, the method will fail and the user will be notified of the incompatibility. A strip consumable may only be RFID scanned two times, a plate consumable six times.
- To select a different method, click the Method dropdown box, which will now look like this:

175 bp Time course R230 96 AFA-TUBE TPX Plate
<<< select method >>>

• Click on "<<< select method >>>", and repeat the process described above.

### 7.2 Positions to Treat

It is sometimes convenient to run a method on a plate that is only partially populated with samples. For example, consider a method that treats all 12 columns in a 96-well plate. But now you have a plate that has samples only in columns 4 to 6. Using SonoLab Positions to Treat allows treating a subset of a method's specified positions.

Positions to Treat is a list of the positions that you want to treat when a method runs. These positions are used to filter the set of positions that are treated by a method. Only positions that appear both in the method and the Positions to Treat list will be treated. Positions to Treat does not affect the order in which positions are treated. Order of treatment is determined by the method.

For Positions to Treat on the R230 instrument, the following column specifications can be made:

- Column number, for example. 4, 5, 6
- Column number range, for example. 4-6

When treating 384-well plates, columns are specified in a range of 1 to 12, not 1 to 24. Two contiguous columns are treated simultaneously.

For methods without scan steps, Positions to Treat can be a comma-separated list of any combination of column numbers or ranges of column numbers. For example, the following position restrictions: 1, 2, 4-6, 8-10 will treat columns 1, 2, 4, 5, 6, 8, 9, 10.

If a method has a scan step, then Positions to Treat must be either a single column number or a single range of column numbers. All the numbers in Positions to Treat must be integers within the valid range of columns for the plate being used (1 to 12 for a 96- well plate, for example).



When specifying a range, the order of columns does not matter, 5-2 is treated the same as 2-5. Duplicate column numbers are ignored. For example, the Positions to Treat "4-6, 5, 4, 6" is equivalent to "4-6".

Positions to Treat are entered into a text box in the Method Options panel as shown:

Method Options	
Override Temperature:	On Off
Override Degas:	On Off
Move to Load Position at end of run:	On Off
Consumable-based Method Selection:	On Off
Positions to Treat:	
Close	

When Positions to Treat is active, a yellow Untreated Positions button is displayed next to the Run button.



Clicking the yellow button displays the Untreated Positions dialog box.



Clicking the Edit Positions to Treat button opens the Method Options window, if it is not already open, allowing the user to change the positions to be treated.

### 7.3 Handheld Barcode and RFID Readers

Handheld Barcode Reader

SonoLab for the R230 supports the following Barcode Scan levels:

1. None: barcoding is not supported.

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- 2. Single: One barcode for the entire consumable (plate or strip).
- 3. Column: One barcode for each strip in a 2D rack.
- 4. Position: One barcode for each tube in a 1D or 2D rack.

The plate definition will specify the barcode level to use when running a method.

### To use barcoding:

- Select a treatment method from the drop down list on the Method screen.
- Turn on the Barcode On/Off switch located on the Method screen, below the light switch.



The green Run button becomes a yellow Scan Barcodes button.

				Log Out
5	Edit	New	Scan Barcode	
	Power: <b>2.1</b> (watts) Degas Time: <b>00:00:00</b>			
Run Time: 00:04:32 / 00:04:32				Abort
urrent	teration:	Iterations: 13	$\bigcirc$	System Status
.0 DI	F: 25 CPB: 5	0 AIP: 70.0 Duration: 10.0	$\overset{\uparrow}{\underset{\downarrow}{\overset{\downarrow}{}}}$	Motion Control

• Click on the Scan Barcodes button. The Scan/Enter Barcode pane is displayed.

Scan/Enter Barcode						
Use the handheld barcode scanner to	o scan the barcode on the consumable					
Barcode						
Done	Cancel					

- Scan (or type) the barcode(s) on the consumables to be treated.
- Click "Done" when all labels' barcodes have been scanned. The Scan/Enter Barcode pane closes and the Scan Barcodes button reverts to the green Run button.
- Click the Run button to begin the treatment.

### Handheld RFID Scanning

SonoLab for the R230 supports using multiple RFID-enabled consumables in a single method. The RFID tag in each consumable is read via a handheld RFID scanner. Each RFID tag scanned is validated, and the serial and lot numbers of the tags are accessible via



history files and the API.

An RFID scan is performed on each treated sample tray to ensure that the correct consumables are being used for the treatment and to provide information for LIMS. Prior to SonoLab version 10.0.2, a strip consumable may only be RFID scanned two times, a plate consumable six times.

Beginning with SonoLab release 10.0.2, by default, each *column* can be RFID scanned twice, either built-in or handheld RFID scanning, or a combination of both.

SonoLab 10 will support the following RFID Scan levels:

- 1. Single: One RFID tag for the entire consumable being processed (plate or strip).
- 2. Column: One RFID tag for each strip in a 2D rack.

The plate definition will specify the RFID level to use when running a method.

### Using Handheld RFID Scanning

Turn on Handheld RFID in the "General" tab of the Admin screen.

метно	DD SCH	EDULE	HISTORY	ADMIN	ABOUT	
USERS	GENERAL					
				Require Login:		On Off
				Reminder to A	rchive:	$\odot$ Never $\odot$ Every 30 days $\odot$ Every 60 days $\odot$ Every 90 days
				Startup Water	Behavior:	○ Control Temperature
				Handheld RFIE	):	on off

The lower right corner of the Method screen, below the light and barcode switches, now contains a switch to turn on Handheld RFID scanning.



When the switch is turned on, the green Run button becomes a yellow Scan RFIDs button.

Click the Scan RFIDs button to display the Scan RFID pane.

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Scan RFID							
Use the handheld RFID scanner to scan the RFID tag on the consumable at:							
	Column 1						
• Skipped treatment	positions will not be treated.						
	Skip Cancel						

Scan the first column to be treated. In this example, column 1 is first to be scanned. The next column to be scanned is shown in the pane.

Scan the remaining columns to be scanned as above. As the columns are scanned, the display in the pane increments to the next column and the number of columns scanned is shown in the RFID button at the lower right of the Method screen.



In this example, column 10 is next to be scanned and 3 of 4 columns to be scanned in the method have been scanned. When all columns to be treated have been scanned, the counter at the lower right changes from yellow to green.



When all the columns have been scanned, the yellow Scan RFIDs button reverts to the green Run button and the method runs.

For both Bar Code scanning and RFID scanning positions specified in the treatment method which are not scanned, will not be treated. In these cases, the yellow Untreated Positions button is displayed next to the Run button.

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Clicking the Untreated Positions button displays the Untreated Positions dialog box.



Clicking the Re-scan RFIDs button returns to the Method screen displaying the Scan RFIDs button.

Handheld RFID Scanning can be combined with Positions to Treat. In that case, the Untreated Positions dialog displays both skipped scan positions and the Positions to Treat selections.



In this example, the original method called for treating all 12 columns, but only columns 1 through 8 were selected for treatment, as indicated at the top of the box. Then, the first 3 columns were skipped during the handheld RFID scanning, as indicated in the second row in the dialog box. The net result is that only columns 4 through 8 will be treated.



### 8. System Specifications

Treatment System	High intensity acoustic transducer, temperature monitoring device, circulation pump, water bath with safety enclosure	
Maximum Treatment Power	450 Watts Peak Incident Power, 110 Watts Average Power	
Dimensions Electronics	410.5 mm x 372.4 mm x 119.3 mm (16.2" x 14.7" x 4.7")	
Weight Electronic Unit	5.8 kg (12.8 lbs.)	
Dimensions Acoustic Unit	362.3 mm W x 286.8 mm D x 151.2 mm H (14.25" x 11.3" x 6.0")	
Weight Acoustic Unit	8.7 kg (19.2 lbs.)	
Dimensions Chiller WCS	406.4 mm x 368.3 mm x 368.3 mm (15.9" x 14.5" x 14.5")	
Weight Chiller WCS	25.9 kg (57 lbs.)	
Power Requirements	100-240 VAC 500 VA maximum, 50-60Hz, Electronic unit	
Power Requirements	100-240 VAC, 50-60Hz, 9-4A, Chiller WCS unit	
Ambient Temperature Range	19°C to 25°C (66°F to 77°C)	
Ambient Humidity Range	30% to 70%	
Regulatory Labeling	CE, ETL Mark (for Product Safety), WEEE	
Safety	Complies with Low Voltage Directive 2006/95/EC. Certified to IEC/EN/ANSI/UL 61010-1:2004 and CAN/CSA C22.2 No. 61010-1:2004, 2nd Edition "Safety Requirements for Electrical Equipment for Measurement, Control and Laboratory Use, Part 1: General Requirements"	
EMC	Complies with Class A Industrial/Scientific/Medical (ISM) equipment under EN 61326-1:2005, EN 61000-3-2:2004 and EN 61000-3-3:1995 for EU EMC Directive 2004/108/EC. Also FCC Part 15 Class A radio emissions requirements for the USA and ICES-003 Class A for Industry Canada.	
Water Bath	Distilled or deionized water only	
Temperature Alarms:	Can be programmed from +6.0 °C to +40.0 °C	
Computer	Notebook computer	
Operating System	Microsoft Windows 10 or 11	
Application Software	Covaris SonoLab 10	
Data Input:	Keyboard, mouse	







### 9. Maintenance

### 9.1 Cooling Air Intake

Unplug the unit periodically and remove any obstruction to the fans and air vents to ensure that objects are not restricting the flow of air.

### 9.2 Water Bath

### 9.2.1. Recommendations for Daily Maintenance

Only distilled or deionized water should be used to fill the water bath. To avoid algae growth, the water bath should be emptied, the degas lines purged, and the tank dried prior to longer term (more than a week) idle time.

### 9.2.2. Recommendations for Monthly Maintenance

The R230 NaDCC Cleaning Protocol should be performed monthly as follows:

Start SonoLab and open the 'Water Control' panel by clicking the 'Water Control' button on the right of the 'Method' screen.

				Power		Kun
	W	ater Conti	rol		o Display	Abort
Level (mm): Temperature (°C): Degas Time: Flow State:	Current: 15.3 Current: 20.6 00:15:07 Off	Load: Setpoint:	Run: Range: to		Column Re	estrictions:
Fill to Load	Fill	Drain	Circulate	Off		Motion Control
		Close	Click H	lere	> 6	Water Control
					203	Method Options

Use the 'Fill', 'Drain' and 'Circulate' buttons on the 'Water Control' pane as instructed below.

- 1. Empty 'Waste' reservoir and dissolve 1 NaDCC 13.1 gram tablet in 4 liters of deionized water in the 'Supply' reservoir.
- 2. Drain the R230, if not already empty.
- 3. Fill the R230.
- 4. Circulate the NaDCC/water solution for 5 minutes.
- 5. Drain the R230.
- Disconnect and empty the 'Supply' reservoir and 'Waste' reservoir. Rinse out the Supply reservoir thoroughly with deionized water. Fill the 'Supply' reservoir with 9.5L of deionized water. Reconnect 'Supply' and 'Waste' reservoirs.
- 7. Fill the R230.
- 8. Circulate the water for 3 minutes.
- 9. Drain the R230.
- 10. Repeat steps 7 through 9 twice more.
- 11. Empty 'Waste' reservoir and refill 'Supply' reservoir with deionized water.



### 9.3 Transducer

Use care when handling the transducer. Permanent damage could result if anything is dropped onto the face of the transducer or if the face of the transducer is damaged.

If drying or cleaning the transducer face, use a soft lint-free cloth. If broken glass must be removed from the transducer surface, carefully pick any shards off the surface with tweezers. Do not rub them away, as this may scratch the transducer surface.

### 9.4 WCS 3

The particulate filter should be changed annually. The filter part number is 500175 and may be purchased from Covaris Customer Service. Instructions for changing the filter are in Appendix A of this document.

### 9.5 Safety System

Contact the Covaris Technical Support Department for more information.

**WARNING**: Report any failure of the Safety System immediately to Covaris. DO NOT attempt to operate if the Safety System is malfunctioning.

### 9.6 Degassing System

The degassing system is incorporated into the WCS 3 and operates automatically.

### 9.7 Cleaning the System

Clean the external surfaces of the equipment as necessary with a damp lint-free cloth.

**CAUTION**: Do not employ isopropyl alcohol, ammonia-based, or abrasive cleaners on the water tank, as these will damage the acrylic surfaces.

Leaving water in the water bath or degassing lines for an extended time may promote algal growth in the tank and lines. This growth can interfere with water quality and sample processing.

### 9.8 Long Term Shutdown Procedure

If the R230 will not be used for a period of two weeks or more, prepare the R230 as described below.

1) Click on the 'Water Control' button. The Water Control pane is displayed.



2) Click on the 'Drain' button on the Water Control pane.



3) Wait for the On-Deck water bath to drain and Flow State Status on the Water Control pane to change to 'Off'.

Water Control					
Level (mm): Temperature (°C): Degas Time: Flow State:	Current: 0.4 Current: 20.4 00:00:00 Off	Load: <b>21.0</b> RL Setpoint: <b>20.0</b>	ın: 22.0 Range: 17.0 to 23	3.0	
Fill to Load	Fill	Drain	Circulate	Off	
		Close			

4) Close the Water Control pane and click on the 'Motion Control' button.

Motion Control					
Home	Load	Start	Open Cover		
RFID	Service	STOP	Close Cover		
Close					

- 5) Click the 'Load' button. The cover opens and the sample tray will rise.
- 6) Dry the on-deck water bath with a soft, lint-free cloth.
- 7) Click the 'Service' button, then the 'Close Cover' button, if necessary.
- 8) Exit the SonoLab application.
- 9) Power off the WCS, the E-box, and the computer.
- 10) Empty and thoroughly rinse both the 'Supply' and 'Waste' bottles. Store the bottles with the covers on to prevent dust from collecting inside.

When turning the system back on:

1) Fill the water bath and drain twice before initiating a run method.



### 9.9 The Utility Menu

The Utility Menu provides the user a simple interface to import method and plate files from, or export to, other instruments; to generate data to help Covaris personnel troubleshoot service issues; and to test the optional Down Holder (if present).

Clicking on the Covaris logo at the upper left corner of the screen displays the Utilities Menu.

Covaris SonoLo	1b 10.0 for R230	
METHOD	SCHEDULE HISTORY	ABOUT
Method Plate:	Utilities	íffset - Hi-Power
Temperature: State:	Collect Diagnostic Data	
Steps	Import Files	ind plate definition file
Process		Currei
Treatment	Export Files	Duration
	Downholder Control	Ĵ
	Close	

9.9.1 Collect Diagnostic Data: Clicking this button displays the Export Diagnostic Data dialog.

© A∥				
	test			
Log and H	listory Files			
	last Week Last More	ath		
÷				
Export To				
<insert< td=""><td>thumb drive&gt;</td><td></td></insert<>	thumb drive>			
	Export Cancel			
	Export Curreer			

In the Method Files section, click the 'All' button to export all treatment methods and their associated plate definitions. Click the 'Selected Method' radio button and select a specific method from the drop-down list to export a specific method and its associated plate definition file.

Exp	oort Diagnostic Data	Power:			
Method Files —		Degas Time			
○ All ●	Selected Method	me: 00:31:25			
	175 bp R230 96 AFA-TUBE TPX Plate - $\degree$	eration: Ite			
	175 bp R230 96 AFA-TUBE TPX Plate - 1 co	olumn			
Log and History	175 bp R230 96 AFA-TUBE TPX Plate - more than 1 column				
○ All ●	175 bp Time course R230 96 AFA-TUBE TPX Plate				
	350 bp R230 96 AFA-TUBE TPX Plate - 1				
Export Io	350 bp R230 96 AFA-TUBE TPX Plate - mo	ore than 1 column			
<insert th="" thum<=""><td colspan="3">350 bp Time course R230 96 AFA-TUBE TPX Plate</td></insert>	350 bp Time course R230 96 AFA-TUBE TPX Plate				
	test				
	Export Cancel				



In the 'Log and History Files' section, use the radio buttons to export either all logs and history files, logs and history files from the last week (the default option), or all log and history files from the last month.

Use the drop-down list in the 'Export To' section to export the data either to the user's Documents folder or to a USB thumb drive. When a USB drive is inserted into the computer, the <insert thumb drive > label changes to the thumb drive letter designation, and the 'Export' button is enabled.

Export To -			
D:\	Ŷ		
		·	
	Export	Cancel	
	Export	Cancer	

Click the 'Export' button to export the data to a .zip.txt file to allow more reliable emailing. The recipient will remove the .txt extension and access the data as from a normal zip file.

**9.9.2** Import Files: Clicking the 'Import Files' button launches the Import Files wizard.

↑ Go Up			Open
Apps			
Dell			
Drivers			
Intel			
PerfLogs			
SiLabs			
SQLServe	r2016Media		
📄 temp			
Users			
VXIPNP			
lo mothod	files found		
io method	mes round		

The first time 'Import Files' is selected, it defaults to searching for Plates and Methods in folders on the C:\ drive, as shown above. Click on to select a subfolder, then click Open, double-click the folder, as in a normal Windows Explorer. Click on the  $\frown Go Up$  button, then click Open, or just double-click to display all available drives on the computer, including removeable thumb drives.

4

应 C:\		
🔗 D:\	REMOVABLE	
<u>)</u> H:\		
应 L:\		
应 R:\		
应 S:\		
😥 U:\		
🖸 V:\		



The wizard remembers the last file location visited and defaults to this last location the next time 'Import Files' is selected.

Navigate to the location from which the methods and plates are to be imported. In this example, these files are in a .zip file.

Import Method & Plate	e Definition Files
Select Import	Folder
Folder: C:\Users\bpettingell\Desktop\tmp\sci	rathpad_for_Import-Export_files\me
↑ Go Up	Open
Methods	
Plate Definitions	
👛 method-n-plates.zip	
😕 Methods.zip	
Plate Definitions.zip	

Click on the folder or, as in this example, the zip file to select it. Then Click the 'Open' button.

The contents of the .zip file (or folder) are displayed. The number of plate and methods files in the folder are shown at the bottom of the Select Import panel.



Click the 'Next' button at the bottom of the screen. A list of available methods is displayed. All methods may be selected (or unselected) or individual methods may be selected as shown below.



Click 'Next'. A list of available plate definition files are displayed. The display defaults to selecting only plate definitions used by the methods selected in the previous step, in this case, the "R230\_520291 96 AFA-Tube PS Plate +0.5 offset.plt" file.

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### Import Method & Plate Definition Files

#### Select Plate Definitions to Import

Select All	Unselect All	# selected: 1
R230_5007	08 PSU Rack 384 A	AFA-TUBE 20 PP Plate +0.5 offset.plt
R230_5007	08 PSU Rack 384 A	AFA-TUBE 5 PP Plate +0.5 offset.plt
R230_5007	23 Holder_520292	Strip_23.5WL_0.5OS.plt
R230_5007	23 Holder_520312	Strip_23.5WL_0.5OS.plt
R230_5007	23 PSU Holder 8 A	FA-TUBE TPX Strip +0.5 offset.plt
R230_5007	50 PSU Rack 96 m	icroTUBE Plate +0.5 offset.plt
R230_5007	50 Rack_520291 P	ate_23.5WL_0.5OS.plt
R230_5007	50 Rack_520310 P	ate_23.5WL_0.5OS.plt
R230_5202	91 96 AFA-TUBE T	PX Plate +0.5 offset.plt
R230_5203	11 96 AFA-TUBE P	S Plate +0.5 offset.plt

Click 'Next'. The confirmation screen is displayed.



Use the back button at the bottom of the window if changes are desired. Otherwise, click the 'Finish' button.

A message is displayed confirming the import was successful.

1	File Import
	File import succeeded
	ок

**9.9.3** Export Files: Clicking the Export Files button launches the Export Files wizard, which operates much like the Import Files wizard discussed above.





A window with a choice of file type to be exported is displayed, with "< Back", "Next >", "Finish", and "Cancel" buttons (not shown in above image) at the bottom of the window. Use the radio buttons to select the type of files, Methods & Plates, History, of Logs, to be exported. Then click Next.

If Methods & Plates are being exported, a method file selection dialog is provided, exactly like the one used in the Method & Plate import function discussed above. Three methods are chosen in this example. Click Next.

Export Methods & Plate Definitions				
Select Methods to Export				
Select All Unselect All	# selected: 3			
☑ 175 bp R230 96 AFA-TUBE TPX Plate - 1 column.mthd				
□ 175 bp Time course R230 96 AFA-TUBE TPX Plate.mthd				
☑ 350 bp Time course R230 96 AFA-TUBE TPX Plate.mthd				
6C Chiller Setpoint_LE220plus.mthd				
Creation Notes Test.mthd				
LE220-plus_QCTest_200bp_96 CrimpCap_uTUBE.mthd				
- IF220 alus OCTast 200ha OF CrimaCan UTURE withd				

As with the import function, a list of available plate definitions is displayed, but only the plate definition(s) needed for the selected methods being exported selected for export. Any or all other definitions may be selected for export. Click Next >. Export Options are displayed.

	Specify Export Options
Export Folder: C:\Users\bpettingell\Docu	ments
Export Filename	R230_001126_MethodsAndPlateDefs_06_14_2023-02-00-06.zip
Delete Files after Export	On Off

Click Next < to display the Confirm Export Choices window. Click the < Back button if corrections are required or click Finish. An Export Succeeded confirmation is displayed.

File Export	
File export succeeded	
ок	

**9.9.4** Downholder Control: The Downholder is an optional fixture which holds a plate for automated filling or emptying so that the automated pipette which pierces the plates foil cover does not carry the plate away when the pipette is withdrawn. Clicking the Downholder Control button displays a panel for testing the functionality of the Downholder, if installed.

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Downholder Control	
Open/Close Status: Plate Presence Status:	Open No Plate Detected
Refresh Status	
Open Downholder	Close Downholder
Close	

The Open/Close and Plate Presence status reflect the condition of the plate holding clamp (Open or Closed) and whether a sample plate is detected on the Downholder. Clicking the 'Refresh Status' button updates these to the actual current state.

The 'Open Downholder' and 'Close Downholder' buttons, respectively, open and close the plate holding clamp for visual confirmation that the Downholder is functioning.

### 10. Troubleshooting

*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 (UTC-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/GMT
- E-mail queries to techsupport@covaris.com or applicationsupport@covaris.com



### Appendix A: Replacing the WCS 3 Particulate Filter

The WCS particulate filter is the only user serviceable component and should be changed annually at minimum. The part number of the particulate filter is 500175 and can be purchased from Covaris Customer Service.

### Changing the Filter

- Power off the WCS 3 and disconnect the power cord from the power source.
- Note which hoses are connected to which ports on the WCS and remove the hoses.
- Remove the cover at the hose side of the WCS by removing the 4 button head cap screws, 2 on each side, and slide the cover off. Removing the hoses is not necessary but gets them out of the way when the cover is removed.



• Locate the Particulate Filter and note the direction of flow arrow.





- Disconnect the hoses at each end of the filter by depressing the release clip on the quick-disconnect fittings.
- With both fittings removed, grasp filter and gently pull it from single retaining clip near the center of the filter.



Reverse the steps to install the replacement filter.

• Note the direction of flow arrow, and place filter into retaining clip. Be careful not to pinch fingers behind filter as the filter snaps firmly into place.


- Press the hose connections back into each end of the replacement filter until they click into place.
- Reconnect the hoses and power source. Power on the WCS and check for leaks.
- Slide the side panel on (it is not necessary to remove the hoses to reinstall the side cover) and secure with the four button head screws.

# Appendix B: R230 Site Preparation

## Placement of Equipment

The R230 electronics unit (ebox) mounts on top of the WCS/Chiller unit. The area required for this assembly (see drawing below) is approximately 14.75" wide by 16.25" deep by 19.75" high (375 mm x 415 mm x 500 mm). Leave sufficient space around this assembly for cooling fan exhaust. Do not block panel vents or fans. There should be a minimum of 12 inches clearance for the ventilation fans on both sides of the WCS/ebox assembly.

The R230 acoustic unit (on-deck unit) is designed to be mounted into a liquid handler or may be placed on a bench. The approximate area required is 11.5" wide by 14.25" deep by 6" high (292 mm x 362 mm x 153 mm).

The on-deck unit has two D-Connectors that must be within 5 feet (1525 mm) of the connectors on the ebox.





## Water Requirements

The R230 water bath requires either deionized or distilled water.

Water is provided by a Supply bottle and drained into a Waste bottle, typically placed on the floor adjacent to the Water Conditioning System (WCS) unit. Each bottle is  $7\frac{1}{2}$ " (190 mm) square and 18" (460 mm) tall to the top of the connections on the caps. Together, the pair of bottles are 15" (380 mm) wide by  $7\frac{1}{2}$ " (190 mm) deep. The body of the bottles, not including the caps and connections, are 13" (330 mm) tall.





# Appendix C: Installing SonoLab 10 Software

These instructions are only for installing SonoLab 10 and other required software packages on a new computer. Do not use these instructions for an incremental upgrade from one version of SonoLab 10 to a newer version. For an upgrade, simply install the new version by double-clicking its "SonoLab 10.msi" file. There is no need to uninstall the existing version prior to upgrading.

#### Install SonoLab 10 on a New Computer

- 1. Download SonoLab for the R230 and the R230 Plate Definitions Installer from the Covaris website <u>https://www.covaris.com/r-series/</u>.
- 2. Unzip the two downloaded files on the R230 computer desktop.
- 3. Install SonoLab 10: Double-click the "SonoLab 10.msi" file. Accept the defaults when prompted.
- 4. Double-click the SonoLab icon on the computer desktop. The "SonoLab is not enabled" panel is displayed:

SonoLab is not enabled
Click Emulate to launch SonoLab in Emulation Mode, or
Click Exit to exit SonoLab, or
Click Enable to enable SonoLab
Emulate Exit Enable

5. Click "Enable". The "Enable SonoLab?" panel is displayed:



- 6. Click "Enable". SonoLab launches. SonoLab is now enabled and will start normally on subsequent startups.
- 7. Copy the R230 Plate definitions from the unzipped Plate Definitions file to C:\Users\Public\Public Documents\Covaris\SonoLab 10\Plate Definitions, or use the 'Import Files' function described in *Section 9.9.2* of this document.
- 8. If desired, copy the R230 Starter Methods from the unzipped file to C:\Users\Public\Public Documents\Covaris\SonoLab 10\Methods, or use the 'Import Files' function described in *Section 9.9.2* of this document.



#### Configuring the Network Environment for the R230

The R230 uses Ethernet to communicate with SonoLab. Unfortunately, it is possible that software running on the SonoLab computer may inadvertently try to communicate with one of the three devices inside the R230. Some of these devices can get confused if they receive network packets other than those sent by SonoLab. When this happens, it may be necessary to restart SonoLab and/or power cycle the instrument to recover.

When the computer on which SonoLab runs is networked, then various network protocols (e.g., LDAP, NetBIOS, DNS) may cause this issue. Some anti-virus software also does this.

We've created Windows firewall rules to prevent this issue from occurring. When SonoLab 10 is installed, it places two PowerShell scripts in this folder:

C:\Users\Public\Documents\Covaris\SonoLab 10\Scripts

The two scripts work in different ways and are incompatible with each other. Only one of the scripts should be run.

These are the scripts:

#### 1. CreateAllowSonoLab10FirewallRule.ps1

This script works by setting the profile of the network adapter used to communicate with the R230 to block all outbound traffic. It then creates a firewall rule that grants SonoLab permission to communicate with the R230.

#### 2. CreateFirewallBlockingRules.ps1

This script works by setting the profile of the network adapter used to communicate with the R230 to allow all outbound traffic. It then creates several firewall rules that block all communication on the IP addresses and port numbers that SonoLab does not use to communicate with the R230.

Running the first script is preferred, as it ensures that only SonoLab can communicate with the R230.

With the second script, there is a very small possibility that another application could try to communicate with the R230 over one of the specific IP address/port number combinations that are permitted.

We have not seen this happen at any R230 installations.

Both scripts are provided because your IT department may prefer one script over the other.

These scripts must be run from a Windows account with Administrator access. In addition, PowerShell must be enabled to run on the computer.

If the computer on which SonoLab is running is connected to your corporate network then these scripts should be reviewed and run by an IT professional, to ensure compliance with your company's IT policies.