

Mantarray

User Manual

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Platform for Human-Relevant 3D
Engineered Muscle Tissue Analysis



Terms and Conditions

Release of Liability

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The Curi Bio Mantarray is designed to work exclusively with Curi Bio Mantarray Plate Kits. Use of kits that are not supplied by Curi Bio, Inc. may void the warranty. Use of the Mantarray Plate Kits are covered in section 2 and 3 of this manual, which discusses preparation and treatment of the consumables for use.

Conditions of Use

The Curi Bio Mantarray is for life science research use only. You are responsible for understanding and safely performing the protocols described within this guide. Curi Bio, Inc. does not guarantee any results you may achieve. These protocols are provided as Curi Bio, Inc. recommendations to you and does not constitute a guarantee of success.

Technical Support

Contact support@curibio.com for tissue casting protocols and other assistance.

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Section 1: General Information

Thank You

Thank you for purchasing the Curi Bio Mantarray instrument. This device is designed to measure contractility kinetics of 3D engineered muscle tissues.

Handling

Please follow the instructions in this manual carefully. Before using the Curi Bio Mantarray, it is recommended that you sterilize the instrument. We recommend wiping the unit down with 70% ethanol and ultrapure water. Avoid getting liquid into the cable ports. Ensure that the instrument is fully dried before connecting any cables. Note that the electronics are sealed from liquid intrusion but using excessive amounts of liquid, spraying, or immersing parts of the Mantarray can damage the instrument.

There are no user serviceable parts inside the Mantarray instrument. Opening the Mantarray case will immediately void the warranty as it could result in damages to the internal components and will destroy the factory environmental seal. For service-related inquiries, please contact us via email at support@curibio.com or call +1-800-913-4403 x704.

Instrument Overview

The Mantarray instrument is comprised of three primary parts: the Main Body, the Power Brick, and the Cooling Unit. The Mantarray Main Body is designed to be housed and operated inside a standard cell culture incubator while the Power Brick sits outside of the incubator to allow the user to conveniently operate the instrument while saving valuable incubator space.

⚠ Note: In some cases, the Mantarray may generate excess heat that cannot be efficiently dissipated based on the make and the model of the cell culture incubators. The Mantarray Cooling Unit can be used to maintain the temperature of the Mantarray Plate environment within a temperature of 0.7°C from the value set by the user.

This section summarizes the components and functions of each part of the Mantarray instrument.

1. Mantarray Main Body

Plate Recording Area: Place your plate on this area to capture data. Ensure that well A1 is located on the back left (when looking at instrument from angle demonstrated in Figure 1) of the recording area. The casing of the main body is marked with a green triangle for where A1 should be adjacent to.

Barcode Scanner: If your plate is oriented correctly, the plate barcode will scan and populate in your Mantarray Controller software.



Figure 1: Components and functions of each part of the Mantarray instrument.



Figure 2: Components and functions of each part of the Mantarray instrument.

Stimulation Ribbon Cable Connection: Connect the ribbon cable to this port to allow the stimulation of tissues using a Curi Bio Stimulation Lid.

Instrument Status: This green light indicates what state your instrument is in. Interpret the status using the following list:

- Solid Light = your instrument is ready for data capture
- Rapidly Blinking Light = your instrument is actively collecting data
- Slowly Blinking Light = your Mantarray is connected to power, but is not connected to a computer

Stimulation Status: This blue light indicates the state of the stimulation feature. Interpret the status using the following list:

- Rapidly Blinking Light = a stimulation protocol is active
- Solid Light = power is connected, but no stimulation protocol is active

Cooling Unit Connection: Port where the Cooling Unit electrical communication cable connects. This plug can only be oriented in one

way, shown by the notch on the cord's plug and groove on the main body's connection port.

Cooling Unit Tubing Connection Ports: Connect the tubing from the Cooling Unit here to allow the flow of water. ⚠ **Note:** Considerable force is required to connect the water lock mechanism that connects the tubing to its port.

Mantarray to Computer Connection: Micro USB that connects to the Mantarray laptop. This will be supplied with the Mantarray.

Power Brick Connection Port: Mantarray to Power Brick connection cord goes here.

2. Mantarray Power Brick



Figure 3: Components and functions of the Mantarray Power Brick.



Figure 4: Components and functions of the Mantarray Power Brick.

Power Brick: Contains the power source for the Mantarray. The unit must be located outside of the cell culture incubator.

Mantarray Connection Port: Plug in your Mantarray to Power Brick connection cord here.

Power Brick Status Light: Indicates the power status. Green light will

illuminate when the instrument is connected to power and the power switch is engaged.

Power Switch: On/Off switch that control power delivery to the Mantarray.

Power Cord Connection: 3-prong receptacle for the power cable.

3. Mantarray Cooling Unit

Temperature Control Unit: Contains a temperature-activated pump and water reservoir for cooling the Mantarray Plate environment. The Temperature Control Unit connects to the Mantarray main body via two silicone tubing water lines (input and output) and a communication cable. When overheating is detected, water is pumped through the cooling pad until the preset temperature is achieved. The unit can be left on indefinitely, but it must be located outside of the cell culture incubator. The bottom of the unit is magnetic, allowing it to mount onto the outside wall of most incubators providing easy access and control to the unit.

Mantarray Plate Composition and Structure

Each well of Mantarray plates is composed of a two-post system, held together by a lattice. One post is rigid and one is flexible. A small magnet is embedded in the end of the flexible post. Deflection of the post as a muscle tissue contracts can then be measured via magnetic sensors in the Mantarray instrument. Tissues are formed in the Tissue Casting Plate pictured below. After a brief 80-minute incubation in the Casting Plate, formed tissues are transferred from the casting plate to a 24-well maintenance plate for long term culture and maintenance. Tissue casting protocols can be found on pages 9-16 in this user manual.

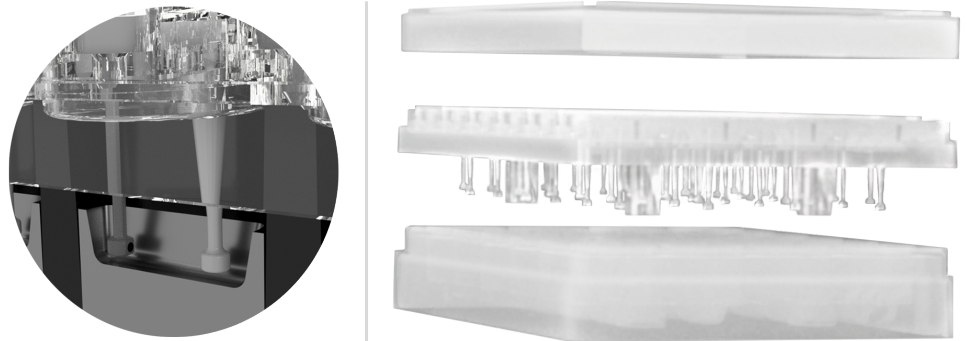


Figure 5: LEFT: Enlarged visualization of the two post system in the casting well. RIGHT: Pieces of the casting plate; (top) plate lid, (middle) holder lattice and posts, (bottom) casting plate.

⚠ **NOTE:** The shelf life for Mantarray Casting Plates is 6 months.

Software

The Mantarray Controller is used to operate the Curi Bio Mantarray. Please email us at support@curibio.com for the latest version. The Mantarray Controller software is designed to exclusively operate on Windows 10 or 11. Operation of the software will be detailed on pages 17-20 in this user manual.

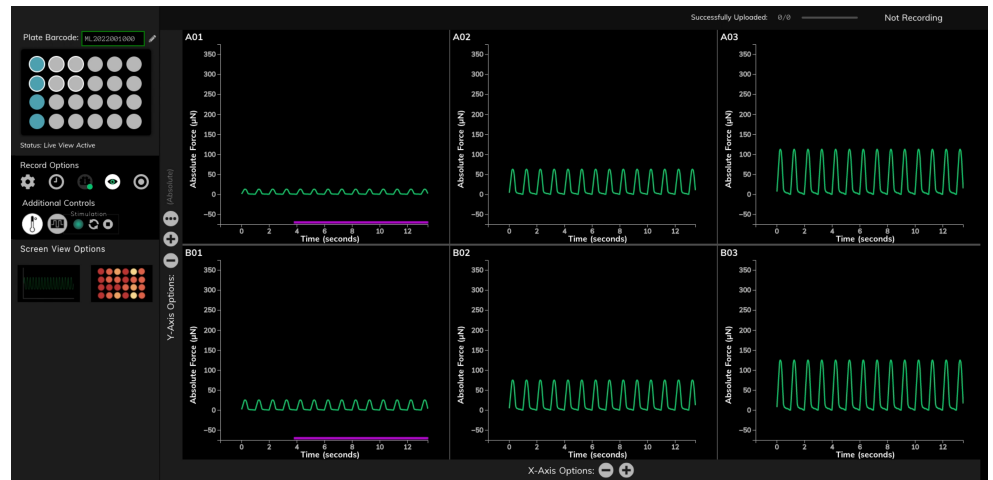


Figure 6: Mantarray Controller software.

Section 2: Installing and Using the Mantarray

Instrument Installation

1. Place the Mantarray into incubator and give the instrument time to heat up to the temperature of the incubator. This takes 6+ hours. During this time, leave all cords unplugged (USB, Power, Cooling Tubes).
2. After 6+ hours, you can plug the Power Brick to Main Unit connection cord, the Cooling Unit's silicon tubing (see below), and the USB connection cord. It is best practice to lead these cords out the back of the incubator.
⚠ Warning: When your instrument is initially put in the incubator, condensation will accumulate on the instrument. Turning on the instrument with large amounts of condensation can break your instrument. Please wait 6 hours for your instrument to warm up to the incubator temperature.
3. Plug the Power Brick's power cord into the wall and leave the instrument off.
4. Attach the Cooling Unit to a flat, vertical, and stable surface, such as an incubator. The unit has magnets that allow it to attach to the side of the incubator hood. Ensure that the unit is upright, with the power button at the top and the silicone tubing port at the bottom.
5. Use the included syringe and filling tube (a small section of tubing with a plastic fitting at the end) to fill the Temperature Control Unit with sterile water.
6. Remove the black silicone plug at the top right of the Cooling Unit and attach the fitting on the end of the fill tube to one of the ports on the bottom of the Cooling Unit. Fill the Cooling Unit to 225 mL of water. Please ensure that the water level stays between the designated lines of the Water Level Window shown in Figure 7.
7. If you need more than one syringe load, remove the filling tube and load the syringe before reattaching and filling.
8. Place the Cooling Unit on the side of your incubator. Remove the filling tube and syringe when the water level reaches the top line of the water level window.
9. Attach the Mantarray to Cooling Unit connection tubes to the ports on the Cooling Unit. It is suggested that you run these tubes through the back port of the incubator. Do not turn the Cooling Unit on.

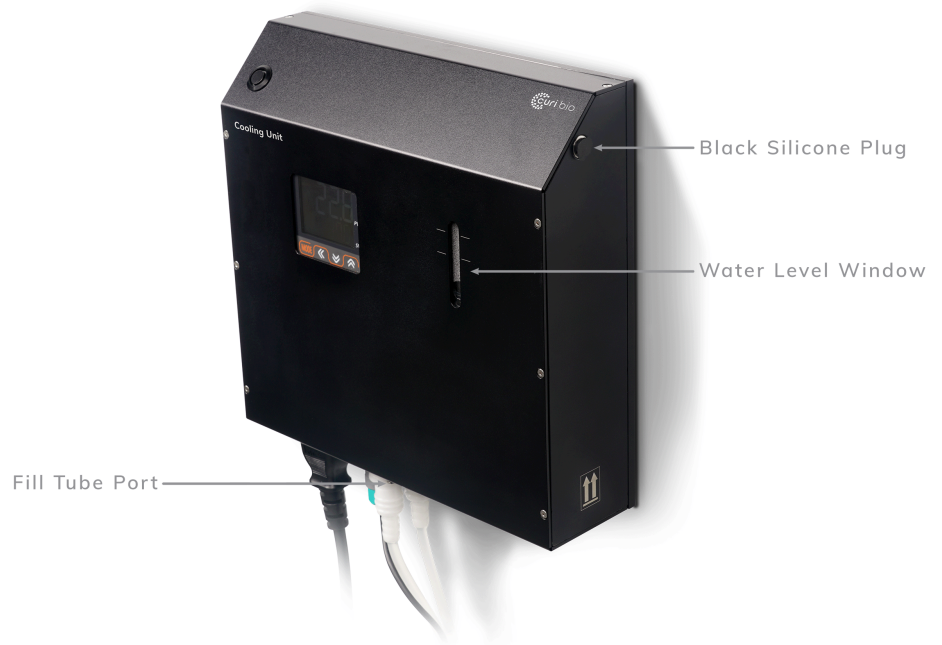


Figure 7: Cooling Unit.

- At this point, the Mantarray is fully installed. Before the instrument can be used, the Cooling Unit must be calibrated.

Note: Please contact support@curibio.com for calibration assistance.

Data Recording

- Flip on Power Brick switch to turn on the Mantarray.
- Open up the Mantarray Controller Software on your computer, ensuring that the USB connection has been made with the computer. If there is a connection issue, the software will indicate that it is in "Simulation Mode", and you must reconnect the instrument.
- Once the software is open, run the calibration procedure by clicking the Calibration icon, as shown in the figure below by a yellow box. The tissue plate should NOT be loaded into the device at this point.

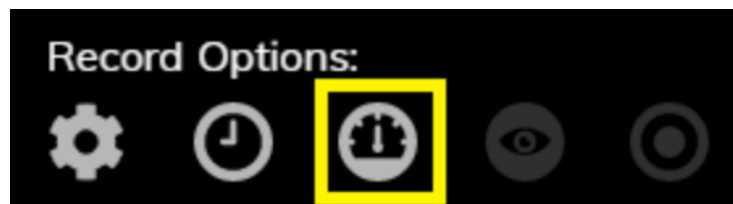


Figure 8

- When calibration is complete, this will be indicated by a green dot over the Calibration button, as seen in Figure 9. At this stage, you may place your tissue plate in the instrument.

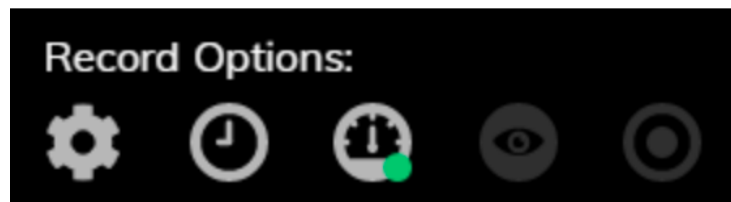


Figure 9

5. Make sure the A1 well is located adjacent to the green marking next to the plate loading area. A barcode should automatically populate if the plate is oriented correctly. If it is not oriented correctly, there will be a red box around the Plate Barcode section. Rotate the plate and wait a few moments until the Plate Barcode section on the Mantarray Controller Software shows green.
6. Clicking the "Live View" button, shown in figure 10, will make Mantarray waveform traces visible in the 6 on-screen graphs. If no trace is visible (or only some), zoom out on the Y-axis until all traces are visible.



Figure 10

7. The Mantarray is now ready to record. This will be indicated by the shade of the "Record" button, shown below in the yellow box in figure 11. Click this button to begin recording. Data from all 24 wells will be recorded, regardless of which 6 wells you are currently viewing.

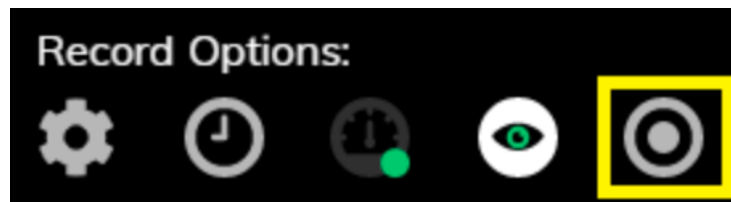


Figure 11

8. The "Record" button will change shape/color to indicate that a recording is in progress. This is shown in figure 12.

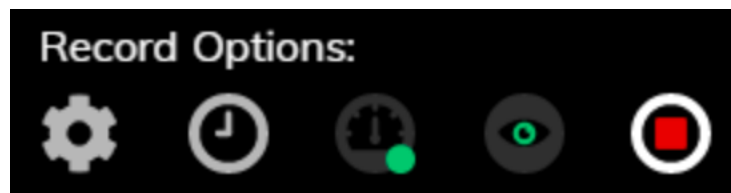


Figure 12

9. Once you are done with your desired recording, press "Stop Recording" and a pop-up to save the recording will appear.
10. After finishing your recording, you will be given the option to see a "snapshot feature". Curi Bio suggests that you look at this snapshot feature as it will give you an indication of the level of magnetic noise in your recording. If there is a large amount of noise, you should consider recording your tissues again.
11. Recordings will be found in your user folder:
C:\Users\YourName\AppData\Roaming\MantarrayController\recordings
12. Logs (to send to Curi Bio for troubleshooting) are in a similar folder:
C:\Users\YourName\AppData\Roaming\MantarrayController\logs_flask

Stimulation

To apply a Stimulation protocol, you will need to use a Stimulation Lid. Please consult the "Stimulation Lid Use & Maintenance" on how to use and clean Stimulation Lids.

1. Navigate to the "stimulation studio" drop down menu to stimulate tissues.
2. Acquire a Mantarray Stimulation Lid from Curi Bio. Place the lid directly on top of the Holder Lattice on the plate.
3. If oriented properly, the "Stim Lid Barcode" section will automatically populate and appear green, if it remains red, rotate the Stim lid until it turns green.
4. Next click on the "Stimulation Studio" button to set up a protocol. The studio is shown in figure 13.
5. Once the protocol has been set, perform the "Configuration Check" to ensure the Stimulation Lid is functioning properly. Please contact Curi Bio if there is an error in your configuration check.
6. Once the check is complete, run the stimulation protocol using the "Run" button and stop as desired using the "Stop" button.
7. When all recordings and stimulation protocols are complete, run a data analysis by clicking on the "Data Analysis" drop down menu.
8. Click on the "Select Recordings" button and select which recordings to analyze. Select "Run".
9. After everything is completed, turn off the Mantarray device using the Power Brick switch.
10. Recordings will be found in the user folder:
C:\Users\YourUsername\AppData\Roaming\MantarrayController\recordings

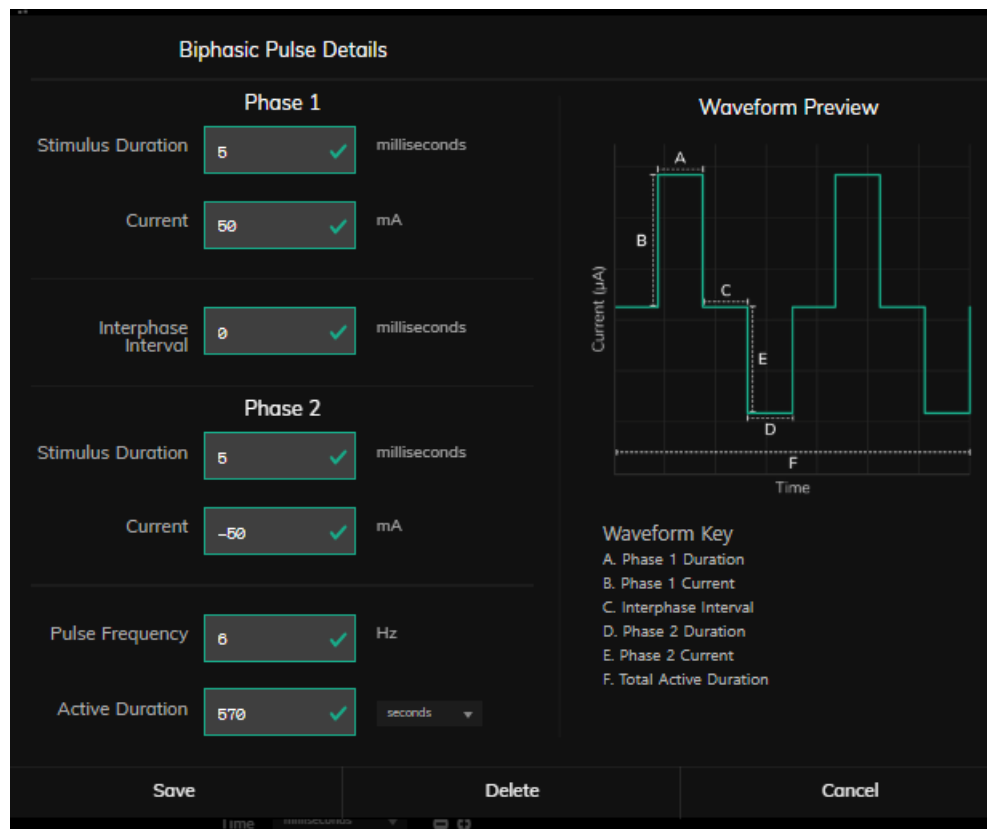


Figure 13

11. Logs (to send to Curi Bio for troubleshooting) are in a similar folder:
 C:\Users\YourUsername\AppData\Roaming\MantarrayController
 \logs_flask

Instrument Maintenance

1. Always leave the Mantarray and Cooling Unit switched OFF while not in use.
2. Please confirm on a monthly basis that the water level in the Cooling Unit stays within the range marked on the Water Level Window. If the water level drops below the marked range, please refill or contact Curi Bio at support@curibio.com for further assistance.

In addition to the aforementioned procedures, the Mantarray instrument is designed to remain operational without the need for additional maintenance. Should you have any concerns regarding the instrument's functionality, please reach out to support@curibio.com for assistance.

Stimulation Lid Use & Maintenance

Stimulation Lid Use

⚠ Warning: Consider carefully the use of a single Stimulation Lid across multiple plates to prevent transfer of small quantities of such molecules.

It is advised to clean and sterilize the Stimulation lid before transferring to a new culture plate to avoid cross-contamination. Please consult the "Stimulation Lid Cleaning & Sterilization" below.

Setting up for stimulation

1. Remove the Stimulation Lid from its sterile packaging inside a biological safety cabinet. When handling the unpacked Stimulation Lid, apply standard sterile techniques, hold the Stim Lid from its circumference and avoid touching the electrodes.
2. Before attempting to place the Stimulation Lid in your culture plate, take note of the alignment marks on the Stimulation Lid and rotate as needed. The Stimulation Lid must be aligned to the culture plate. Attempting to insert a misaligned Stimulation Lid can damage the Lid and/or the culture plate.
3. Remove the regular lid from your culture plate and insert the stimulation lid in the plate. If you meet resistance when sliding the electrodes through the tissue lattice, please evaluate if your Stimulation Lid is misaligned.
4. Place the regular lid on the Stimulation Lid. The regular lid should align and fit tightly around the alignment pattern of the Stimulation Lid to ensure a sterile enclosure.
5. Transfer the culture plate at the location where the stimulation will be performed.
6. Plug one end of the ribbon cable into the Mantarray instrument that will deliver the stimulation. Apply a sufficient amount of pressure to ensure proper connection.
7. Plug the second end of the ribbon cable into the Stim Lid. To securely connect the cable, you will need to apply sufficient pressure, as poor connection will lead to inability to deliver stimulation. As you press, please ensure to apply the right amount of counter pressure to avoid dislodging the Stimulation Lid from the plate.
8. If stimulation is conducted in the incubator, you may now close the incubator.
9. Proceed to define the stimulation settings and perform your stimulation experiment following the "Stimulation" protocol.

After running stimulation

1. Once your stimulation protocol is complete, remove the ribbon cable from the Stimulation Lid. Press firmly on one of the tabs on the sides of the connector to lower the tab. Once the tab has been lowered, lower the second tab. Lowering both tabs should release the ribbon cable. Ensure to apply a sufficient amount of counter-pressure throughout to avoid dislodging the stimulation lid.
2. Move the plate to the biological safety cabinet.

3. In the cabinet, hold the tissue lattice and the culture plate down in position and in parallel lift the Stimulation Lid. The Stimulation Lid should slide out of the plate.
4. Remove the regular lid from the Stimulation Lid and position it back on your culture plate.
5. Store or discard your culture plate.
6. Returning to your instrument, you may leave the ribbon cable connected to the instrument. If you are concerned about sterility, you may apply 70% ethanol or isopropanol on a wipe and gently wipe the ribbon cable and the connectors of the cable. Stay clear of the electrical ports of the connectors.

Stimulation Lid Cleaning & Sterilization

The Stimulation Lid features carbon electrodes that will absorb byproducts from your tissue culture wells. It is recommended to clean them every 24-48 hours of use. You are advised to follow the full protocol below. Resorting to a plain rinse will remove excess media from electrodes, but might not be sufficient to remove small molecules such as drug, fluorescent dyes, or viral particles.

Please be sure to clean the Stimulation Lid before autoclaving, following the protocol outlined below.

1. Fill a clean, waterproof container only with distilled water. Add a stir bar in the container and place the container on a stir plate. Do NOT add detergents in the water.
2. Place the Stimulation Lid in the waterproof container, ensuring that ONLY the carbon electrodes are submerged. The water level should remain below the electrode socket by several millimeters. The stir bar should be either below the electrodes or adjacent to the electrodes.
3. Set the stir plate to a low rotation setting to avoid damaging the stimulation lid. Begin stirring.
4. Wash for at least 48 hours. The water should be changed at least every 24 hours. More frequent water changes (every 12 hours) are recommended.

⚠ Warning: At the end of this washing step, the water should be clear and no residual dyes (e.g. phenol red) should be observed in the water. If the water is not clear, repeat the washing step in increments of 24 hours until clear.

⚠ Warning: After multiple use cycles of the Stimulation Lid, residue might accumulate at the top of the electrodes, close to the socket with the main board. To remove this residue, dip a q-tip in distilled water and gently rub the electrodes. Rinse the q-tip and repeat until the residue is sufficiently removed. Ensure to minimize the exposure of the main board to water.

5. Allow the electrodes to dry over an absorbent surface, such as a lab-grade wipe. Avoid rubbing the electrodes dry.
6. Place the dry Stimulation Lid in a autoclave pouch.
7. Autoclave the Stimulation Lid. We recommend using a steam ("Gravity") cycle with 20 minutes of sterilization at 120°C/248°F. If your autoclave has a cooldown ("Dry") cycle setting, we recommend 15 minutes of cooldown.
8. Store the autoclaved Stimulation Lid at room temperature and humidity until next use.

⚠Warning: Remove the stimulation lid from the autoclave with caution. The stimulation lid will be hot.

Data Analysis

There are three methods of analyzing data to understand the kinetics of your Muscle Tissues

1. The first method is Local analysis using the Mantarray controller app: This analysis will only provide the raw data for Force vs Time. These data are saved for any recording locally on the Mantarray Laptop. This method generates a downloadable excel file that can be used to analyze the raw data.
2. The second method is an automatic data upload for analysis on the cloud. Enter the user login information on the Mantarray Controller, which will allow the software to automatically upload and analyze the data. Login to the cloud to access the data. This method requires an internet connection on the laptop you are using.
3. The third method is a manual data upload for cloud analysis. This method is the same as step 2 on this list, but the data is manually uploaded. To use this feature, navigate to the Curi Bio Dashboard. You will first need to create a user account.

Contact support@curibio.com for any assistance.

Section 3: Warranty and Technical Specifications

Warranty

The Curi Bio Mantarray is sold with a one year warranty, effective from the date of purchase, for any defects or failures in parts or workmanship. Additional warranty coverage may be purchased on a per-year basis by contacting sales@curibio.com. If the Mantarray fails or breaks within one year of purchase, Curi Bio will repair it at no charge to the customer. The customer is responsible for the cost of shipping the Mantarray to Curi Bio's headquarters in Seattle, WA, USA. Curi Bio will pay for the return shipment of the repaired equipment. All warranty returns must be accompanied by an RMA number. For any questions, please contact us at support@curibio.com or +1 (800) 913-4403.

This warranty does not cover cosmetic damage or wear. Damage due to mishandling, abuse, misuse, or through failure to adhere to the guidelines in this manual will not be covered.

Specifications

Instrument Footprint: 312 x 200 x 49 mm

Instrument Mass: 5.2 kg

Power Brick Footprint: 125 x 186 x 52 mm

Power Brick Mass: 1.1 kg

Connectivity: USB and onboard memory for stored protocols

Power: Product consumes <1 Amp, Single Phase, 50-60Hz and requires 100-240 VAC input voltage. The plug type provided by default is American standard. If requested, we can provide plug types for Europe, Asia, Middle East, Oceania, and additional country or region types. Alternatively, a locally sourced adapter can be used on the standard issue power cord to convert the plug and meet requirements.

FAQs

How is your Mantarray plate oriented in the instrument?

The Mantarray plate should be oriented such that well A1 is adjacent to the green triangle mark next to the plate loading area.

Am I able to electrically stimulate my tissues in culture?

Yes you can. The Mantarray has a built in stimulation system that allows for individual well stimulation.

Do I need to use a Curi Bio Stimulation Lid to stimulate my tissues?

Yes, you must use the Curi Bio Stimulation Lid with the Mantarray plate and instrument.

How can I analyze my data?

Mantarray users can use both the desktop and the Pulse3D cloud analysis options.

Do I need to use the Pulse 3D cloud services to operate the instrument?

No, you do not need to use Curi Bio's cloud services, however it is highly recommended.

What types of cells can I use in my tissues? What cell types are suggested?

We have extensive experience with in-house and commercial lines for both cardiac and skeletal muscle applications, and we are always interested in expanding to new applications. Please contact our support team for suggestions specific to your cell line of interest.

More Questions?

If you have additional questions or concerns, please contact support@curibio.com for assistance.